Cite as: Kobiałka M, Wrotek A, Jackowska T: Usefulness of RSV rapid diagnostic tests in hospitalized children.
Pediatr Med Rodz 2024; 20 (2): 196–204

Received: 19.11.2023
Accepted: 13.02.2024
Published: 27.09.2024

Małgorzata Kobiałka[1], August Wrotek[1,2], Teresa Jackowska[1,2]

Usefulness of RSV rapid diagnostic tests in hospitalised children

Przydatność szybkich testów diagnostycznych RSV u dzieci hospitalizowanych

[1] Department of Paediatrics, Bielanski Hospital, Warsaw, Poland
[2] Department of Paediatrics, Centre of Postgraduate Medical Education, Warsaw, Poland
Correspondence: Teresa Jackowska, Department of Paediatrics, Centre of Postgraduate Medical Education, Marymoncka 99–103, 01-813 Warsaw, Poland, e-mail: tjackowska@cmkp.edu.pl

https://doi.org/10.15557/PiMR.2024.0028

ORCID iDs
1. Małgorzata Kobiałka https://orcid.org/0000-0002-4763-0558
2. August Wrotek https://orcid.org/0000-0002-3167-3975
3. Teresa Jackowska https://orcid.org/0000-0003-0131-7854

Abstract

Introduction and objective: Respiratory syncytial virus is a major cause of lower respiratory tract infections, particularly in children under two years of age. Diagnostic methods include rapid antigen diagnostic tests, which have shown high specificity and variable sensitivity. The aims of this study were to verify the performance of these tests in hospitalised children, and to identify factors influencing the results. **Materials and methods:** The study enrolled children under two years old who were tested for respiratory syncytial virus using both rapid antigen diagnostic test and a cartridge-based nucleic acid amplification test. The sensitivity, specificity, and positive and negative predictive values of the tests were calculated. The influence of symptoms and fever duration, socioeconomic conditions, presence of siblings, and feeding method were analysed. **Results:** A total of 164 patients aged 10 days to 24 months (median 2.5 months) were included. Sensitivity reached 75% (95% confidence interval: 67.3–81.7%), specificity – 100% (73.5–100%), positive predictive value – 100%, and negative predictive value – 24% (19.3–29.4%). Lower sensitivity was associated with longer duration of signs/symptoms, with the lowest value obtained in the group with 6–7 days of signs/symptoms – 47.4% (24.5–71.1%), which differed from the 2–3 days group ($p = 0.005$) and the 4–5 days group ($p = 0.016$). No association was found between sensitivity and patient age, socioeconomic conditions, presence of siblings, clinical course including fever, or feeding method. None of these factors affected specificity, positive or negative predictive value. **Conclusions:** The reliability of the rapid antigen diagnostic tests in cases of positive results appears to be high, though negative results should be interpreted with caution. Prolonged duration of signs/symptoms before testing might reduce the level of sensitivity.

Keywords: children, sensitivity, specificity, respiratory syncytial virus, rapid antigen diagnostic test

Streszczenie

Wprowadzenie i cel: Syncytialny wirus oddechowy jest główną przyczyną infekcji dolnych dróg oddechowych, szczególnie u dzieci poniżej 2. roku życia. Metody diagnostyczne obejmują szybkie antygenowe testy diagnostyczne, które wykazały wysoką swoistość i zmienną czułość. Celem pracy była weryfikacja ich skuteczności u hospitalizowanych dzieci oraz identyfikacja czynników wpływających na wynik. **Materiał i metody:** Do badania włączono dzieci w wieku poniżej 2 lat, u których wykryto syncytialnego wirusa oddechowego za pomocą szybkiego antygenowego testu diagnostycznego i kasetowego testu amplifikacji kwasu nukleinowego. Obliczono czułość, swoistość, dodatnie i ujemne wartości predykcyjne. Przeanalizowano wpływ objawów i czasu trwania gorączki, warunków socjoekonomicznych, obecności rodzeństwa i sposobu karmienia. **Wyniki:** Do badania włączono 164 pacjentów w wieku od 10 dni do 24 miesięcy (mediana 2,5 miesiąca). Czułość wyniosła 75% (95% przedział ufności: 67,3–81,7%), swoistość – 100% (73,5–100%), dodatnia wartość predykcyjna – 100%, ujemna wartość predykcyjna – 24% (19,3–29,4%). Niższa czułość była związana z dłuższym czasem trwania objawów, z najniższą wartością w grupie z 6–7 dniami objawów – 47,4% (24,5–71,1%), co różniło się od grupy z 2–3 dniami objawów ($p = 0,005$) i grupy z objawami 4–5 dni ($p = 0,016$). Nie stwierdzono związku między czułością a wiekiem pacjenta, warunkami socjoekonomicznymi, obecnością rodzeństwa, przebiegiem klinicznym, w tym gorączką, ani sposobem karmienia. Żaden z tych czynników nie wpływał na swoistość, dodatnią lub ujemną wartość predykcyjną. **Wnioski:** Wiarygodność szybkich antygenowych testów diagnostycznych w przypadku wyniku dodatniego wydaje się wysoka, chociaż wynik ujemny należy interpretować z ostrożnością. Dłuższy czas trwania objawów przed badaniem może zmniejszyć czułość testu.

Słowa kluczowe: dzieci, czułość, swoistość, syncytialny wirus oddechowy, szybkie antygenowe testy diagnostyczne

BACKGROUND

Respiratory syncytial virus (RSV) is one of the most common causes of respiratory infections, often affecting the lower respiratory tract and resulting in bronchiolitis, pneumonia or bronchitis, which are collectively classified as lower respiratory tract infections (LRTI)[1,2]. The RSV attack rate is the highest in the first two years of life, and approximately 60–70% of children are infected with RSV in their first year of life[3–5]. It is estimated that RSV is responsible for approximately 33 million LRTI cases in children under five years of age each year, including 3.6 million hospitalisations[6]. RSV contributes significantly to paediatric hospitalisations with an incidence of 4.37/1,000 children under five years of age, with the highest incidence observed in children under one year of age (19.19/1,000) and especially in children under six months of age (20.01/1,000)[4]. The Polish data, based on the Polish National Health Fund (Narodowy Fundusz Zdrowia, NFZ) registry, show an average hospitalisation rate of 2.67/1,000 in the paediatric group of patients and 11.32/1,000 in infants[7]. The course of disease can vary from mild upper respiratory tract infection to severe LRTI with significant respiratory failure or even death; worldwide, about 2% of paediatric deaths under the age of five are estimated to be due to RSV infection, and the highest mortality is observed in infants under six months of age (about 3.6% of deaths in this age group)[6]. Complications of RSV infection are common and can affect up to 60% of hospitalised children, with otitis media and pneumonia being the most common (risks can be as high as 50% and 33%, respectively)[8]. At present, there is no causal treatment, and only a few candidate molecules for therapy are under investigation; a much broader spectrum of perspectives is seen in the field of prophylaxis, which includes the use of monoclonal antibodies and vaccine studies[9,10].

A detailed knowledge of the viral aetiology of infection is crucial both from the perspective of patients and for epidemiological reasons, as it reduces unnecessary antibiotic use on the one hand, and helps prevent nosocomial spread of infection by cohorting or isolating patients on the other; in addition, accurate epidemiological data are needed for socioeconomic analyses[11–13]. RSV testing includes, in addition to molecular methods and serological studies (which become useful after a certain time from the onset of signs/symptoms), rapid antigen diagnostic tests (RADT), which target RSV antigens in a sample obtained from the upper respiratory tract, such as nasal lavage or nasopharyngeal swabs[14,15]. RADT is characterised by lower costs (compared to molecular diagnostics) and a shorter time to implement infection control measures. An inverse correlation between the number of RADTs performed and nosocomial infections has been reported[16,17]. Previous studies have shown a high specificity of RADT (approximately 90–100%), but a suboptimal sensitivity was observed in the majority of studies, reaching 60–70%[18]. However, while high specificity was repeatedly observed, sensitivity varied from a low level of 8% in infants[19] and 27% in adults[20] up to 90%[15]. Studies on factors influencing RADT performance are rather scarce, but an association with patient age[19,20], longer duration of symptoms before testing[21,22], or low viral load[21,23] has been observed.

The goals of this study were to assess the utility of RADT in hospitalised children under two years of age, and to identify factors relevant to RADT performance.

MATERIALS AND METHODS

The study was approved by the local Ethics Committee at the Centre of Postgraduate Medical Education, Warsaw (permission number 32/PB/2019, issued on 13 March 2019).

A retrospective analysis of RSV RADT performance in comparison to reverse transcription-polymerase chain reaction (RT-PCR) as a reference method was conducted.

Study group

The electronic database of patients admitted to the Department of Paediatrics, Bielanski Hospital, Warsaw, Poland, between 1 January 2017 and 31 January 2019 was analysed, and all records of patients who underwent RADT were retrieved. Children under two years of age were eligible for the study, and only patients who also underwent RT-PCR were included. In order to exclude individuals with nosocomial infections, only patients with signs/symptoms of respiratory infection (i.e. coryza, cough, dyspnoea, apnoea, foamy mucous in the mouth, fever) prior to hospital admission were included in the analysis. Exclusion criteria also comprised the lack of informed consent, completion of only one of the tests, or absence of medical history data on factors affecting RADT performance (e.g. duration of signs/symptoms).

Diagnostic procedures

Patients admitted to hospital with suspected RSV infection were tested by RADT and/or RT-PCR. Testing was performed on a sample from a nasopharyngeal swab taken immediately after hospital admission. The Alere BinaxNOW RSV Card (Alere Scarborough Inc.; Scarborough, Maine, USA) was used as the RADT, while the molecular test was a cartridge-based nucleic acid amplification test (CBNAAT) using the Cepheid GeneXpert (Sunnyvale, California, USA) which, in addition to testing for RSV, also detects influenza A and B viruses, providing important information for further patient management. All diagnostic tests were performed after obtaining informed consent from every patient's legal guardian. All test procedures were done in accordance with the manufacturer's instructions.

Study endpoints

The primary endpoints included sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) of the tests. Secondary outcomes were the

 DOI: 10.15557/PiMR.2024.0028

assessment of factors that might influence RADT results: patient age, feeding method (breastfeeding, mixed feeding, or formula feeding only), socioeconomic conditions (assessed by the number of persons per room in the household), presence or absence of siblings in the household, duration of symptoms before hospitalisation, and duration of fever (before, during, and total).

Statistical analysis

Data are presented as numbers (*n*) and percentages for nominal variables and as means ± standard deviations (*SD*) or medians with interquartile ranges (IQR) for continuous variables, depending on data distribution. Data distribution was tested using the Shapiro–Wilk test, including both visual histogram and skewness assessment. RADT sensitivity, specificity, PPV and NPV with 95% confidence interval (95% CI) were calculated. The Fisher exact test or chi-squared test was used for subgroup comparisons, and the Fisher exact test with and without Bonferroni correction was used for pairwise comparisons. Analyses were performed using R 4.0.5 statistical software [R Core Team (2021). R: language and environment for statistical computing from the R Foundation for Statistical Computing, Vienna, Austria]. A *p*-value below 0.05 was considered statistically significant.

RESULTS

In the analysed period (January 2017 – January 2019), there were 1,852 hospitalisations of children under two years of age, of whom 170 (9.2%) children were tested for RSV by both RADT and RT-PCR. Six patients were excluded because of suspected nosocomial infection, leaving a study group of 164 patients, of whom 104/164 (63.4%) were boys. The age ranged from 10 days to 24 months, with a median of 2.5 months; the majority of children were less than six months old (143/164; 87.2%), including 104 (63.4%) patients ≤3 months old, while there were only 10 patients (6.1%) aged one year and older (Fig. 1). There were 24 premature infants (14.6%), the majority of patients were breastfed at the time of admission (60.4%, 99/164), the median density (persons/room) reached 1.6, and the majority of children had one sibling (62.3%). The most common sign/symptom was cough (98.8%), followed by coryza (77.4%), while apnoea was reported in 3.7% of cases; the median duration of signs/symptoms was four days. The majority of patients had no fever (60.1%), the median respiratory rate was 51/min, the median pulse oximetry oxygen saturation was 96%, and the median heart rate was 140/min (Tab. 1).

There were 114 positive RADTs results, which accounted for 69.5% (114/164) of the study group, while the remainder (30.5%, 50/164) were negative. Compared to RT-PCR, true positives were observed in all 100% of positive RADT cases with no false positives, while 24% (12/50) were true negatives, and the remaining 76% (38/50) were false negatives (Tab. 2).

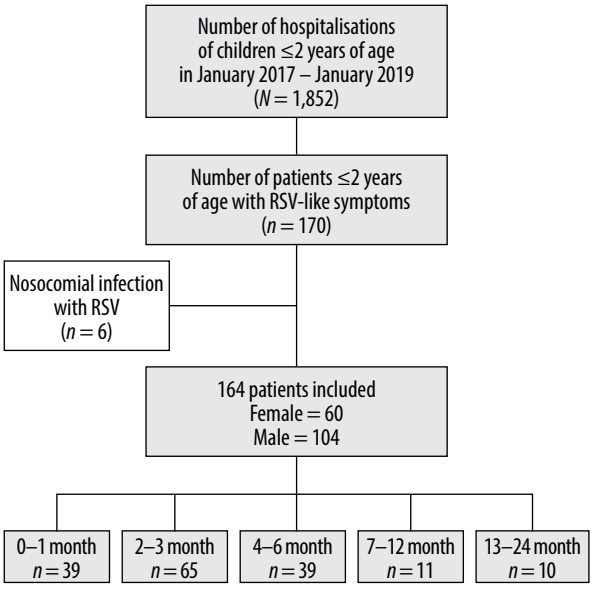

Fig. 1. Flowchart of the patients in the study

Compared with RT-PCR, RADT showed a sensitivity of 75.0% (95% CI: 67.3–81.7) and a specificity of 100.0% (95% CI: 73.5–100.0), with a PPV of 100% and a NPV of 24% (95% CI: 19.3–29.4).

An analysis of confounders showed a statistically significant lower RADT sensitivity (47.4%) in patients with signs/symptoms lasting 6–7 days (95% CI 24.5–71.1; $p = 0.031$). In a direct comparison of the subgroups, children with signs/symptoms lasting 6–7 days showed statistically significant differences from those with signs/symptoms lasting 2–3 days ($p = 0.005$) and 4–5 days ($p = 0.016$); after applying the Bonferroni correction, the *p*-value reached 0.047 and 0.162, respectively, and was significant only for the comparison of 6–7 days *vs.* 2–3 days. In children with signs/symptoms persisting for eight days or more, increased sensitivity was observed again, reaching 75% (95% CI 34.9–96.8) (Fig. 2). An additional analysis compared the duration of signs/symptoms up to 3 days vs. ≥4 days, but found no statistically significant differences.

Differences in sensitivity between the age groups were not statistically significant ($p = 0.688$). The highest sensitivity was observed in children aged 4–6 months (80.6%; 95% CI 63.9–91.8) and the lowest in children aged 7–12 months (63.6%; 95% CI 30.8–89.1). Also, an analysis comparing age ≤3 months with age >3 months showed no statistical significance ($p > 0.999$). There were no statistically significant differences regarding the feeding method, socioeconomic conditions or the presence of siblings (Tab. 3). Neither the presence nor the duration of fever influenced RADT sensitivity.

There were no statistically significant differences in RADT specificity, PPV or NPV (Tab. 4).

DISCUSSION

Our study showed 100% specificity with lower (75%) sensitivity of RSV rapid antigen detection tests. Special

DOI: 10.15557/PiMR.2024.0028

Characteristic	Statistics	Range
Sex, *n* (%):		
Female	60 (36.6)	
Male	104 (63.4)	
Age [months], median (Q1;Q3)	2.54 (1.54;4.40)	0.33–24.0
Hbd [weeks], median (Q1;Q3)	39.00 (38.00;40.00)	26–41
Preterm, *n* (%)	24 (14.6)	
Feeding method, *n* (%):		
Breastfeeding only	99 (60.4)	
Breastfeeding + formula	18 (11.0)	
Formula only	40 (24.4)	
Number of persons in household, median (Q1;Q3)	4.00 (4.00;4.00)	0–8
Number of rooms in household, median (Q1;Q3)	3.00 (2.00;3.00)	1–6
Number of persons in household/number of rooms, mean ± *SD*	1.59 ± 0.67	0–5
Number of siblings, *n* (%):		
0	39 (24.1)	
1	101 (62.3)	
2	17 (10.5)	
3	5 (3.1)	
Median (Q1;Q3)	1.00 (1.00;1.00)	0–3
Rhinitis, *n* (%)	127 (77.4)	
Cough, *n* (%)	162 (98.8)	
Dyspnoea, *n* (%)	90 (54.9)	
Apnoea, *n* (%)	6 (3.7)	
Foamy mucous, *n* (%)	58 (35.4)	
Duration of symptoms [days]	4.04 ± 2.10	0–10
Fever [°C], *n* (%)		
No fever	98 (60.1)	
37.7–38	32 (19.6)	
38–39	26 (16.0)	
39–40	7 (4.3)	
Mean ± *SD*	37.25 ± 0.89	36.6–39.8
Duration of fever before hospitalisation [days], median (Q1;Q3)	0.00 (0.00;1.00)	0–7
Duration of fever in hospital [days], median (Q1;Q3)	0.00 (0.00;0.00)	0–3
Duration of fever (total) [days], median (Q1;Q3)	0.00 (0.00;1.00)	0–7
Respiratory rate on admission to the hospital [/min], mean ± *SD*	51.21 ± 9.97	24–72
Saturation on admission to the hospital [%], mean ± *SD*	95.84 ± 2.07	88–99
Heart rate on admission to the hospital [/min], mean ± *SD*	140.37 ± 16.06	100–190
pH in capillary blood, mean ± *SD*	7.42 ± 0.04	7.12–7.53
pCO_2 in capillary blood [mm Hg], mean ± *SD*	36.22 ± 6.31	20.50–70.90
Saturation in capillary blood [%], mean ± *SD*	90.70 ± 5.15	63.60–99.60
Procalcitonin (PCT) [ng/mL], median (Q1;Q3)	0.09 (0.07;0.13)	0.02–3.10
C-reactive protein (CRP) [mg/L], median (Q1;Q3)	1.70 (0.72;6.33)	0.10–69.17
White blood cells (WBC) [1×10^3/µL], mean ± *SD*	11.25 ± 3.63	4.21–25.79
Neutrophil [%], mean ± *SD*	25.56 ± 14.58	4.60–88.30
Absolute neutrophil count (ANC) [1×10^3/µL], median (Q1;Q3)	2.40 (1.25;3.99)	0.37–15.20
Hbd – weeks of pregnancy; **pCO_2** – partial pressure of carbon dioxide.		

Tab. 1. Baseline characteristics of the study group

attention should be paid to the high PPV combined with the low NPV, which in practice means a high probability of confirmation of RSV infection in case of a positive result. However, negative results should be treated with caution, as they do not provide sufficient certainty to exclude infection. Yet the factors that increase the risk of false negative results are difficult to identify.

The observed specificity of 100% (95% CI: 73.5–100.0) is in line with previously published analyses of RADT performance; Jung et al.[18] analysed four different RADTs

 DOI: 10.15557/PiMR.2024.0028

		PCR method		
		Positive	Negative	Total
Rapid RSV test	Positive	114 (69.5)	0 (0.0)	114 (69.5)
	Negative	38 (23.2)	12 (7.3)	50 (30.5)
	Total	152 (92.7)	12 (7.3)	164 (100.0)
Data presented as *n* (%) of total group.				

Tab. 2. Comparison of the results of rapid RSV tests vs. PCR method

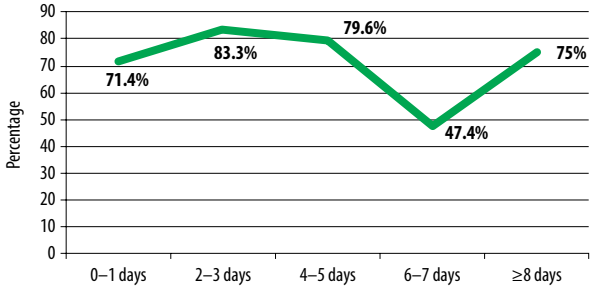

Fig. 2. Sensitivity of RSV RADT regarding symptom duration

(including the BinaxNow test used in our study) and obtained a specificity of 95.8–100%, while other studies evaluating the performance of BinaxNow showed a specificity of 93% to 100%[24]. Similarly, high specificities have been reported for other RADTs[14,15]. The PPV observed in our patient series reached 100%, which is in alignment with the literature[15,19]; thus, a positive test result can be treated with high probability as a true positive and lead to further therapeutic or epidemiological actions[16].

On the other hand, the need to cohort or isolate patients cannot be excluded based on a negative RADT result alone, and the interpretation of negative results may be complicated. In our series of patients, sensitivity reached 75% (95% CI: 67.3–81.7), which is close to the paediatric patient group in the study by Larsson et al.[20], who reported 67% and 70% sensitivity for ImmuViewRSV and BinaxNow, respectively. Slightly lower sensitivities were published by Jung et al.[18], ranging from 62.5% (for BinaxNow) to 67.5% for RSV A and from 61.3% (for BinaxNow) to 67.5% for RSV B. Our study did not differentiate between the RSV groups (A vs. B), but in light of the above results, the virus group does not seem to be a crucial factor for test sensitivity[15,18]. However, in the study by Franck et al.[24], a slightly higher sensitivity was observed for RSV A. Generally, higher results than in our study were observed by Mesquita et al.[15], who analysed the QuickVue® RSV test and reported a sensitivity of 90%; conversely, Franck et al.[24] included 500 children in a retrospective study and obtained a sensitivity below 50% for both analysed tests, including about 30% in the case of BinaxNow. The lowest sensitivity of BinaxNow was reported in the group of infants in the study by Zuurbier et al.[19], with a result of only 7.6% (95% CI: 3.3%–16.5%). These findings could be significant, as the highest number of RSV hospitalisations is

	Result				Sensitivity		Specificity	PPV	NPV	
	TP	TN	FP	FN	(95% CI)	*p*	(95% CI)	(95% CI)	(95% CI)	*p*
Total group	114	12	0	38	75.0 (67.3–81.7)		100.0 (73.5–100.0)	100.0	24.0 (19.3–29.4)	
Age [months]:										
0–1	29	2	0	8	78.4 (61.8–90.2)	0.688[1]	100.0 (15.8–100.0)	100.0	20.0 (11.9–31.6)	0.929
2–3	43	6	0	16	72.9 (59.5–83.6)		100.0 (54.1–100.0)	100.0	27.3 (19.8–36.3)	
4–6	29	3	0	7	80.6 (63.9–91.8)		100.0 (29.2–100.0)	100.0	30.0 (18.1–45.5)	
7–12	7	0	0	4	63.6 (30.8–89.1)		---	---	---	
13–24	6	1	0	3	66.7 (29.9–92.5)		100.0 (2.50–100.0)	100.0	25.0 (11.7–45.6)	
Up to 3 months	72	8	0	24	75.0 (65.1–83.3)	>0.999	100.0 (63.1–100.0)	100.0	25.0 (19.1–32.0)	>0.999
Above 3 months	42	4	0	14	75.0 (61.6–85.6)		100.0 (39.8–100.0)	100.0	22.2 (15.4–31.0)	
Feeding method:										
Breastfeeding only	75	5	0	19	79.8 (70.3–87.4)	0.123	100.0 (47.8–100.0)	100.0	20.8 (14.9–28.2)	0.863[1]
Breastfeeding + formula	10	4	0	4	71.4 (41.9–91.6)	0.750[1]	100.0 (39.8–100.0)	100.0	50.0 (30.4–69.6)	0.082
Formula only	26	2	0	12	68.4 (51.4–82.5)	0.387	100.0 (15.8–100.0)	100.0	14.3 (9.45–21.0)	0.468
Persons/rooms in household:										
0–1	16	2	0	4	80.0 (56.3–94.3)	0.710[1]	100.0 (15.8–100.0)	100.0	33.3 (17.2–54.6)	0.167
2	42	4	0	16	72.4 (59.1–83.3)		100.0 (39.8–100.0)	100.0	20.0 (14.2–27.5)	
>2	4	3	0	2	66.8 (22.3–95.7)		100.0 (29.2–100.0)	100.0	60.0 (32.6–82.3)	
Siblings:										
No	28	3	0	8	77.8 (60.9–89.9)	0.806	100.0 (29.2–100.0)	100.0	27.3 (16.9–40.9)	0.692
Yes	85	8	0	30	73.9 (64.9–81.7)		100.0 (63.6–100.0)	100.0	21.1 (16.4–26.6)	

95% CI – confidence interval; **FN** – false-negative; **FP** – false-positive; **NPV** – negative predictive value; ***p*** – *p*-value; **PPV** – positive predictive value; **TN** – true-negative; **TP** – true-positive.
Sensitivity and NPV % values compared between groups with Fisher's exact test[1] or chi2 test.

Tab. 3. Validity analysis of RSV RADT vs. PCR method

DOI: 10.15557/PiMR.2024.0028

	Result				Sensitivity		Specificity	PPV	NPV	
	TP	TN	FP	FN	(95% CI)	*p*	(95% CI)	(95% CI)	(95% CI)	*p*
Duration of symptoms [days]:										
0–1	5	1	0	2	71.4 (29.0–96.3)	0.031[1]*	100.0 (2.50–100.0)	100.0	33.3 (13.4–61.7)	0.418[1]
2–3	50	3	0	10	83.3 (71.5–91.7)		100.0 (29.2–100.0)	100.0	23.1 (72.7–92.1)	
4–5	39	5	0	10	79.6 (65.7–89.8)		100.0 (47.8–100.0)	100.0	33.3 (22.3–46.5)	
6–7	9	1	0	10	47.4 (24.5–71.1)		100.0 (2.5–100.0)	100.0	9.1 (6.1–13.2)	
≥8	6	2	0	2	75.0 (34.9–96.8)		100.0 (15.8–100.0)	100.0	50.0 (23.1–76.9)	
0–3	55	4	0	12	82.1 (70.8–90.4)	0.177	100.0 (39.8–100.0)	100.0	25.0 (16.6–35.8)	>0.999
≥4	54	8	0	22	71.1 (59.5–80.9)		100.0 (63.1–100.0)	100.0	26.7 (20.4–34.1)	
Fever [°C]:										
37.7–38	94	8	0	28	77.1 (68.6–84.2)	0.335[1]	100.0 (63.1–100.0)	100.0	22.2 (17.1–28.3)	0.469
38–39	16	2	0	8	66.7 (44.7–84.4)		100.0 (15.8–100.0)	100.0	20.0 (12.4–30.6)	
39–40	3	2	0	2	60.0 (14.7–94.7)		100.0 (15.8–100.0)	100.0	50.0 (25.5–74.5)	
<38	94	8	0	28	77.1 (68.6–84.2)	0.295	100.0 (63.1–100.0)	100.0	22.2 (17.1–28.3)	0.718
≥38	19	4	0	10	65.5 (45.7–82.1)		100.0 (39.8–100.0)	100.0	28.6 (19.5–39.8)	
Duration of fever before hospitalisation [days]:										
0	87	5	0	26	77.0 (68.1–84.4)	0.315[1]	100.0 (47.8–100.0)	100.0	16.1 (12.1–21.2)	0.144
1	12	4	0	8	60.0 (36.1–80.9)		100.0 (39.8–100.0)	100.0	33.3 (22.6–46.1)	
2	7	1	0	3	70.0 (34.8–93.3)		100.0 (2.5–100.0)	100.0	25.0 (11.5–46.2)	
≥3	8	2	0	1	98.7 (93.2–99.9)		100.0 (15.8–100.0)	100.0	66.7 (22.2–93.3)	
0	87	5	0	26	77.0 (68.1–84.4)	0.453	100.0 (47.8–100.0)	100.0	16.1 (12.1–21.2)	0.171[1]
≥1	27	7	0	12	69.2 (52.4–82.9)		100.0 (59.0–100.0)	100.0	36.8 (26.7–48.3)	
Duration of fever during hospitalisation [days]:										
0	89	9	0	34	72.4 (63.6–80.0)	0.416[1]	100.0 (66.4–100.0)	100.0	20.9 (16.6–26.0)	0.258
1	14	2	0	2	87.5 (61.7–98.5)		100.0 (15.8–100.0)	100.0	50.0 (21.5–78.5)	
2–3	9	1	0	2	81.8 (48.2–97.7)		100.0 (2.5–100.0)	100.0	33.3 (12.5–63.7)	
0	89	9	0	34	72.4 (63.6–80.0)	0.223[1]	100.0 (66.4–100.0)	100.0	20.9 (16.6–26.0)	0.337
≥1	23	3	0	4	85.2 (66.3–95.8)		100.0 (29.2–100.0)	100.0	42.9 (23.3–64.9)	
Total duration of fever [days]:										
0	74	6	0	25	74.8 (65.0–82.9)	0.899[1]	100.0 (54.1–100.0)	100.0	19.4 (14.6–25.2)	0.659
1	14	2	0	5	73.7 (48.8–90.9)		100.0 (15.8–100.0)	100.0	28.6 (15.9–45.9)	
2–3	14	3	0	6	70.0 (45.7–88.1)		100.0 (29.2–100.0)	100.0	33.3 (20.4–49.4)	
≥4	10	1	0	2	83.3 (51.6–97.9)		100.0 (2.5–100.0)	100.0	33.3 (12.4–63.9)	
0	74	6	0	25	74.8 (65.0–82.9)	>0.999	100.0 (54.1–100.0)	100.0	19.4 (14.6–25.2)	0.496[1]
≥1	38	6	0	13	74.5 (60.4–85.7)		100.0 (54.1–100.0)	100.0	31.6 (22.4–42.5)	

95% CI – confidence interval; **FN** – false-negative; **FP** – false-positive; **NPV** – negative predictive value; ***p*** – *p*-value; **PPV** – positive predictive value; **TN** – true-negative; **TP** – true-positive.
Sensitivity and NPV % values compared between groups with Fisher's exact test[1] or chi2 test.
* In a direct comparison, the sensitivity in the 6–7 days group was significantly different from the 2–3 days group ($p = 0.005$) and from 4–5 days ($p = 0.016$), while after Bonferroni correction the values reached 0.047 and $p = 0.162$, respectively.

Tab. 4. Validity analysis of RSV RADT vs. PCR method with regard to duration of symptoms, height of fever, duration of fever (prior to, during hospitalisation, and total)

observed in the youngest group of patients; moreover, these patients are at the highest risk of a severe course of the disease, and the focus should be placed on preventing nosocomial spread of infection, which might be facilitated in the case of false negative antigen test results[4,5,8]. Conversely, Franck et al.[24] observed a better sensitivity in children under six years of age compared to adults, in contrast to the study by Larsson et al.[20], who showed a significantly superior RADT performance in a small control group of children compared to adults (67–70% vs. 27%). However, our study only included children under the age of two, and a detailed age analysis showed no statistically significant differences between age groups. This finding has a huge practical impact by facilitating the approach to RADT interpretation in children; on the other hand, it is not possible to point to an age group that would be at increased risk of false results and would additionally benefit from molecular testing instead of RADT.

While the positive predictive value in our group is high and in line with previous publications[15,25,26], more obstacles are

 DOI: 10.15557/PiMR.2024.0028

caused by low NPV. The present study revealed much lower values than previously reported for BinaxNow; Mesquita et al. found an NPV of 94.6%[15], while Sanbonmatsu-Gámez et al.[25] showed the value of 95.4% (90.9–97.8), and Moesker et al.[26] – 88% (84–91). Jang et al.[27] reported slightly lower values of 81.1%–81.5% for RSV A and 80.2%–83.3% for RSV B, while Jonckheere et al.[28] found an NPV of 74%–74.2% in children under six years of age, emphasising that a negative result does not exclude RSV infection. A similar analysis to our study was presented by Papenburg et al.[22], who reviewed the performance of BinaxNow and found an NPV of 72.9%, concluding that older age (i.e. 24–35 months), longer duration of signs/symptoms (at least five days), diagnosis of pneumonia, and RSV B infection are more often associated with false negative test results. Although our findings cannot be discussed with regard to age (the study only included children under 24 months), we did not verify the final diagnosis (pneumonia versus bronchiolitis or bronchitis) or the RSV group (A vs. B) of infection, but we did find an association with regard to the duration of signs/symptoms.

The time between the onset of signs/symptoms and viral testing appears to be critical. The highest sensitivity was observed in patients with signs/symptoms present for 2–3 days (83.3%). After this period, test sensitivity decreased to the lowest value in those with signs/symptoms present for 6–7 days (47.4%). This may be related to lower viral load, which can lead to lower sensitivity of the method; Miernyk et al.[21] conducted a study evaluating the BinaxNow test and observed lower viral load in patients with false negative results; also, as in our study, test sensitivity decreased with increasing duration of signs/symptoms. However, Mesquita et al.[15] found no association between test sensitivity and viral load, although the overall sensitivity of the test in this study group was very high and, therefore, less dependent on viral load. It must be emphasised that in our series of patients, an increase in sensitivity was observed in those who presented with signs/symptoms for eight days or more, but the low number of patients in this group might have affected the results and influenced the reverse tendency. Yet another source of bias could be expected, namely a patient could acquire an RSV co-infection (e.g. nosocomial) during an ongoing infection, in which case the true duration of RSV-related signs/symptoms would actually be shorter. However, it cannot be concluded or expected that a longer duration of signs/syndromes would result in better RSV detection.

Other possible factors influencing test results that could also affect viral load include the presence or absence of siblings and socioeconomic conditions. Age structure, larger household size, higher population density, and type of contacts (including higher viral load or presence of symptoms in RSV-infected household members) may influence viral transmission[29–31]. However, no statistically significant associations of the above factors with test sensitivity were found. The feeding method (breastfeeding, mixed feeding or formula feeding) was not associated with test sensitivity, although breastfeeding has been shown to be a protective factor against RSV infection, which would presumably reduce viral load and consequently RADT sensitivity[32,33]. However, it must be underlined that only hospitalised patients were enrolled in the study, i.e. only those with a more severe disease course, and the role of breastfeeding may not have been noticed in this pre-selected group; a comparative analysis between children requiring and not requiring hospital treatment might highlight the role of breastfeeding in RADT susceptibility. Fever in our patient group had no effect on test sensitivity, either in terms of its duration or the highest body temperature observed. Liu et al.[34] found that fever was less common in patients with higher viral loads, and lower viral loads would be expected to cause lower test sensitivity. Interestingly, Liu et al.[34] suggested that less frequent fever in individuals with higher viral loads might be associated with disease progression, with signs/symptoms still present despite the majority of the virus being cleared, as they depend on the host's immunological response.

There are some limitations to the study that need to be highlighted: it was a single-centre study and any generalisations should be made with caution, as the number of patients was not large enough to guarantee the generalisability of findings with certainty. The study protocol included children under two years of age, which is a limitation, but on the other hand this is the most affected age group, with the highest incidence of RSV disease. The retrospective nature of the study is yet another limitation, as a proportion of RSV-infected patients may have been missed, though the retrospective nature of the study did not lead to unnecessary diagnostic procedures, including in oligosymptomatic and asymptomatic patients, and addresses the most common dilemma in everyday hospital practice. Two major virologic limitations include the lack of differentiation between RSV A and RSV B infections and the lack of viral load measurements. Differentiation of RSV groups is not only necessary from an epidemiological perspective, but may also influence test performance, although the risk appears to be quite low in light of previously published data[27]; nevertheless, viral load may be more important, especially in terms of sensitivity variation. In the discussion section, studies evaluating different RADTs are presented. The differences between tests from multiple manufacturers are to be expected, nevertheless the issue needs to be generalised first to facilitate understanding and to disseminate knowledge on the use of RADTs and their limitations. The decision on specific RADT manufacturer could be made locally, also considering product availability and pharmacoeconomic issues. The rapid development of new diagnostic methods makes it difficult to remember the exact performance characteristics of the tests in relation to specific manufacturers, but at the same time guarantees further progress in the field of RSV diagnostics.

CONCLUSION

Rapid RSV antigen tests are a readily available diagnostic method, and a direct comparison with molecular studies

DOI: 10.15557/PiMR.2024.0028

shows their suboptimal sensitivity and high specificity. High positive predictive values with low negative predictive values confirm high reliability of positive results and suggest caution in interpreting negative results. Longer duration of signs/symptoms was found to be associated with a higher risk of false-negative results, while the other factors analysed (patient age, feeding method, socioeconomic conditions, number of siblings, and duration of fever) were not linked to test performance.

Conflict of interest

The authors do not report any financial or personal connections with other persons or organisations which might negatively affect the content of this publication and/or claim authorship rights to this publication.

Funding/Support and role of the sponsor

The study was supported by Centre of Postgraduate Medical Education, grant number 501-1-020-19-23.

Ethics approval

The study was conducted in accordance with the Helsinki Declaration with later amendments and approved by the local Ethics Committee at the Centre of Postgraduate Medical Education in Warsaw (permission number 91/PB/2020, issued on 15 June 2020). The consent of patients and parents/tutors consent was waived due to the retrospective character of the study.

Informed consent statement

The consent of patients and parents/tutors was waived due to the retrospective character of the study.

Consent to participate

Informed consent was obtained from the legal guardians of all participants included in the study.

Author contribution

Original concept of study: MK, AW, TJ. Collection, recording and/or compilation of data: MK. Analysis and interpretation of data: MK, AW. Writing of manuscript: MK. Critical review of manuscript: AW, TJ. Final approval of manuscript: MK, AW, TJ.

References

1. Hacımustafaoğlu M, Celebi S, Bozdemir SE et al.: RSV frequency in children below 2 years hospitalized for lower respiratory tract infections. Turk J Pediatr 2013; 55: 130–139.
2. Svensson C, Berg K, Sigurs N et al.: Incidence, risk factors and hospital burden in children under five years of age hospitalised with respiratory syncytial virus infections. Acta Paediatr 2015; 104: 922–926.
3. Hall CB, Weinberg GA, Blumkin AK et al.: Respiratory syncytial virus-associated hospitalizations among children less than 24 months of age. Pediatrics 2013; 132: e341–e348.
4. Stein RT, Bont LJ, Zar H et al.: Respiratory syncytial virus hospitalization and mortality: systematic review and meta-analysis. Pediatr Pulmonol 2017; 52: 556–569.
5. Obando-Pacheco P, Justicia-Grande AJ, Rivero-Calle I et al.: Respiratory syncytial virus seasonality: a global overview. J Infect Dis 2018; 217: 1356–1364.
6. Li Y, Wang X, Blau DM et al.: Global, regional, and national disease burden estimates of acute lower respiratory infections due to respiratory syncytial virus in children younger than 5 years in 2019: a systematic analysis. Lancet 2022; 399: 2047–2064.
7. Rząd M, Kanecki K, Lewtak K et al.: Human respiratory syncytial virus infections among hospitalized children in Poland during 2010–2020: study based on the National Hospital Registry. J Clin Med 2022; 11: 6451.
8. Wrotek A, Kobiałka M, Grochowski B et al.: Respiratory complications in children hospitalized with respiratory syncytial virus infection. Adv Exp Med Biol 2020; 1279: 113–120.
9. Li X, Hodgson D, Flaig J et al.: Cost-effectiveness of respiratory syncytial virus preventive interventions in children: a model comparison study. Value Health 2023; 26: 508–518.
10. Mazur NI, Terstappen J, Baral R et al.: Respiratory syncytia virus prevention within reach: the vaccine and monoclonal antibody landscape. Lancet Infect Dis 2023; 23: e2–e21.
11. Díez-Domingo J, Pérez-Yarza EG, Melero JA et al.: Social, economic, and health impact of the respiratory syncytial virus: a systematic search. BMC Infect Dis 2014; 14: 544.
12. Young M, Smitherman L: Socioeconomic impact of RSV hospitalization. Infect Dis Ther 2021; 10 (Suppl 1): 35–45.
13. Zheng Z, Warren JL, Shapiro ED et al.: Estimated incidence of respiratory hospitalizations attributable to RSV infections across age and socioeconomic groups. Pneumonia (Nathan) 2022; 14: 6.
14. Peters RM, Schnee SV, Tabatabai J et al.: Evaluation of Alere i RSV for rapid detection of respiratory syncytial virus in children hospitalized with acute respiratory tract infection. J Clin Microbiol 2017; 55: 1032–1036.
15. Mesquita FDS, Oliveira DBL, Crema D et al.: Rapid antigen detection test for respiratory syncytial virus diagnosis as a diagnostic tool. J Pediatr (Rio J) 2017; 93: 246–252.
16. Mills JM, Harper J, Broomfield D et al.: Rapid testing for respiratory syncytial virus in a paediatric emergency department: benefits for infection control and bed management. J Hosp Infect 2011; 77: 248–251.
17. Wrotek A, Czajkowska M, Jackowska T: Nosocomial infections in patients hospitalized with respiratory syncytial virus: a practice review. Adv Exp Med Biol 2020; 1271: 1–10.
18. Jung BK, Choi SH, Lee JH et al.: Performance evaluation of four rapid antigen tests for the detection of respiratory syncytial virus. J Med Virol 2016; 88: 1720–1724.
19. Zuurbier RP, Bont LJ, Langedijk AC et al.: Low sensitivity of BinaxNOW RSV in infants. J Infect Dis 2020; 222 (Suppl 7): S640–S647.
20. Larsson E, Johansson S, Frøbert O et al.: Evaluation of the ImmuView RSV test for rapid detection of respiratory syncytial virus in adult patients with influenza-like symptoms. Microbiol Spectr 2021; 9: e0093721.
21. Miernyk K, Bulkow L, DeByle C et al.: Performance of a rapid antigen test (Binax NOW® RSV) for diagnosis of respiratory syn-

cytial virus compared with real-time polymerase chain reaction in a pediatric population. J Clin Virol 2011; 50: 240–243.
22. Papenburg J, Buckeridge DL, De Serres G et al.: Host and viral factors affecting clinical performance of a rapid diagnostic test for respiratory syncytial virus in hospitalized children. J Pediatr 2013; 163: 911–913.
23. Schnee SV, Pfeil J, Ihling CM et al.: Performance of the Alere i RSV assay for point-of-care detection of respiratory syncytial virus in children. BMC Infect Dis 2017; 17: 767.
24. Franck KT, Schneider UV, Ma CMG et al.: Evaluation of immuview RSV antigen test (SSI siagnostica) and BinaxNOW RSV card (alere) for rapid detection of respiratory syncytial virus in retrospectively and prospectively collected respiratory samples. J Med Virol 2020; 92: 2992–2998.
25. Sanbonmatsu-Gámez S, Pérez-Ruiz M, Lara-Oya A et al.: Analytical performance of the automated multianalyte point-of-care mariPOC® for the detection of respiratory viruses. Diagn Microbiol Infect Dis 2015; 83: 252–256.
26. Moesker FM, van Kampen JJA, Aron G et al.: Diagnostic performance of influenza viruses and RSV rapid antigen detection tests in children in tertiary care. J Clin Virol 2016; 79: 12–17.
27. Jang JW, Cho CH, Nam MH et al.: Clinical performance evaluation of the Sofia RSV FIA rapid antigen test for diagnosis of respiratory syncytial virus infection. J Clin Microbiol 2015; 53: 684–686.
28. Jonckheere S, Verfaillie C, Boel A et al.: Multicenter evaluation of BD Veritor System and RSV K-SeT for rapid detection of respiratory syncytial virus in a diagnostic laboratory setting. Diagn Microbiol Infect Dis 2015; 83: 37–40.
29. Kombe IK, Munywoki PK, Baguelin M et al.: Model-based estimates of transmission of respiratory syncytial virus within households. Epidemics 2019; 27: 1–11.
30. Otomaru H, Sornillo JBT, Kamigaki T et al.: Risk of transmission and viral shedding from the time of infection for respiratory syncytial virus in households. Am J Epidemiol 2021; 190: 2536–2543.
31. Zheng Z, Pitzer VE, Warren JL et al.: Community factors associated with local epidemic timing of respiratory syncytial virus: a spatiotemporal modeling study. Sci Adv 2021; 7: eabd6421.
32. Li R, Dee D, Li CM et al.: Breastfeeding and risk of infections at 6 years. Pediatrics 2014; 134 (Suppl 1): S13–S20.
33. Mineva GM, Purtill H, Dunne CP et al.: Impact of breastfeeding on the incidence and severity of respiratory syncytial virus (RSV)-associated acute lower respiratory infections in infants: a systematic review highlighting the global relevance of primary prevention. BMJ Glob Health 2023; 8: e009693.
34. Liu W, Chen D, Tan W et al.: Epidemiology and clinical presentations of respiratory syncytial virus subgroups A and B detected with multiplex real-time PCR. PLoS One 2016; 11: e0165108.

DOI: 10.15557/PiMR.2024.0028