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Yao, KK, Navoa-Ng, JA. Comparison of Respiratory Pathogens in Hospitalized Children During and After the COVID-19 Peak in a Philippine Tertiary Hospital
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ORIGINAL ARTICLE

COMPARISON OF RESPIRATORY PATHOGENS IN HOSPITALIZED CHILDREN DURING AND AFTER THE COVID-19 PEAK IN A PHILIPPINE TERTIARY HOSPITAL

Kezzia Kim Yao, RPh, MD, DPPS, Josephine Anne Navoa-Ng, MD, FPPS, FPIDSP
Institute of Pediatrics and Child Health, St. Luke's Medical Center-Global City
Taguig City, Philippines

ABSTRACT

Background: The impact of the COVID-19 pandemic on respiratory virus activity in children has been studied globally, but no published study in the Philippines has provided viral profiling and epidemiological data on children during and after the pandemic's peak.

Objectives: To identify respiratory pathogens detected using a multiplex RT-PCR assay (BioFire® Respiratory 2.1 Panel) among pediatric patients with respiratory symptoms admitted to St. Luke's Medical Center–Global City during (March 2020–February 2022) and after (March 2022–March 2023) the peak of the COVID-19 pandemic; to compare the prevalence of these pathogens between the two periods; and to assess the clinico-demographic characteristics, diagnostic test results, and clinical outcomes of patients with single-pathogen infections versus co-infections.

Methods: A single-center, cross-sectional study was conducted through a retrospective chart review of pediatric patients with respiratory symptoms tested using a multiplex RT-PCR assay (BioFire® Respiratory 2.1 Panel) at St. Luke's Medical Center–Global City from March 2020 to March 2023.

Results: Of 739 children, 92.02% were positive for at least one respiratory pathogen, mostly viruses. Rhinovirus/enterovirus (50.59%), RSV (19.71%), and COVID-19 (12.50%) were the most common. COVID-19 and Influenza A were more prevalent during the peak, while rhinovirus/enterovirus and adenovirus were higher post-peak. Most patients were male, aged 1–5 years, and cough (89.99%) was the most common symptom. Normal leukocyte, CRP, and procalcitonin levels were observed in 70.09%, 47.31%, and 68.25%, respectively. No significant differences were noted in diagnostic test results based on pathogen detection. Antibiotics were given to 53.31% of patients, and 99.86% were discharged. The average length of stay was 3.69 days.

Conclusion: The prevalence of respiratory pathogens among children admitted to our institution during and after the peak of the COVID-19 pandemic predominantly consisted of viruses, showing statistically significant differences. Rhinovirus/enterovirus and RSV were the leading respiratory pathogens in both periods. The peak group showed a higher prevalence of COVID-19 and Influenza A, whereas the post-peak group exhibited a higher prevalence of rhinovirus/enterovirus and adenovirus. Single viral infections were more prevalent compared to co-infections.

KEYWORDS: *Respiratory Panel, Pandemics, Prevalence, Philippines*

Correspondence:
Dr. Kezzia Kim Yao
kezzia.yao@gmail.com

The authors declare that the data presented are original material and have not been previously published, accepted or considered for publication elsewhere; that the manuscript has been approved by all authors, and that the authors have met the requirements for authorship.

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INTRODUCTION

Over the course of the COVID-19 pandemic, testing practices and the prevalence of SARS-CoV-2 and other respiratory pathogens shifted considerably.[1] Respiratory viral co-detections were expected, with studies reporting differences in clinical features and outcomes between children with isolated SARS-CoV-2 infections and those with multiple pathogens.[2] Reports from China, Thailand, and Indonesia showed decreased respiratory pathogen detection during the pandemic, likely due to public health interventions, while co-infections in younger children were often linked to greater clinical severity.[2-9] However, these findings are region-specific, underscoring the need for continued surveillance and further research to clarify the long-term impact of the pandemic on respiratory pathogens and to guide future outbreak management.[7,10] Despite global investigations, no published study in the Philippines has provided viral profiling and epidemiological descriptions during and after the pandemic peak. Locally, the first case of community transmission was confirmed in March 2020, with subsequent surges in September 2021 and January 2022 corresponding to the Delta and Omicron variants, followed by a marked decline in cases by February 2022.[11,12] Based on this timeline, the present study divided children into two groups: the peak period (March 2020–February 2022) and the post-peak period (March 2022–March 2023). The study aimed to identify respiratory pathogens among pediatric patients with respiratory symptoms using multiplex RT-PCR, compare their prevalence between the peak and post-peak periods, and assess associated clinico-demographic features, diagnostic findings, and clinical outcomes.

MATERIALS AND METHODS

Study Design and Participants

This was a single-center cross-sectional analytical study, using retrospective chart review of all pediatric patients with respiratory symptoms who were tested with the BioFire® Respiratory 2.1 Panel and were admitted at St. Luke's Medical Center–Global City from March 2020 to March 2023. It should be noted that in the early phase of the pandemic, the hospital initially relied on RT-PCR testing solely for SARS-CoV-2. The BioFire® Respiratory 2.1 Panel, which detects both SARS-CoV-2 and a broad range of respiratory pathogens, was implemented only later in the study period, restricting the availability of pathogen data during the early phase.

Inclusion and Exclusion Criteria for Subject Selection

This study enrolled pediatric patients aged 0–18 years who presented with respiratory symptoms at the time of consultation and were admitted between March 2020 and March 2023. Only those with a nasopharyngeal swab result using the BioFire® Respiratory 2.1 Panel were included. Patients who were tested only for screening, procedures, or surgery, as well as those with incomplete records were excluded from this study.

Description of Study Procedure

Data management, archiving and confidentiality

Upon approval of the St Luke's Medical Center-Global City Institutional Ethics Review Committee, a letter of approval from the head of the medical record section was secured to access the charts of patients qualified based on the inclusion criteria. Patients' data were protected with a code for confidentiality. A spreadsheet using Microsoft Excel was used to record data collection with no patient identifiers.

Data gathered

The list of all subjects who fulfilled the inclusion criteria was retrieved. This list was generated from the database of all pediatric patients with respiratory symptoms seen at St Luke's Medical Center-Global City from March 2020 to March 2023. The following data were collected and entered into a data collection form: date of admission and discharge, age, sex, clinical symptoms, presence of comorbidities, diagnostics (CBC, CRP, procalcitonin, chest X-ray), respiratory panel result, clinical diagnosis, management, and outcome.

Sample Size Estimation

Sample size was calculated based on the test of hypothesis for the difference in the prevalence of human rhinovirus during and after the peak of the COVID-19 pandemic, as it yielded the largest required sample size among all respiratory microorganisms studied. Assuming that the prevalence of human rhinovirus during the peak was 21.1% and after the peak at 12.97% (Xu et al, 2022), with an alpha error of 5%, power of 90%, and a 1-tailed alternative hypothesis, the sample size calculated is 363 per group, for a total of 726 for two groups.

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Data Analysis

Data was processed and encoded using Microsoft Excel. Statistical analysis was done using Statistical Package for Social Sciences (SPSS) v27. Determination of the clinico-demographic characteristics and the distribution of respiratory pathogens between the two groups was analyzed using descriptive statistics. Mean and standard deviation were used for continuous data, and frequency and percentage for categorical data. Comparison of the clinical characteristics and outcomes between the two groups was analyzed using a chi-square test for categorical variables and an independent t-test for continuous variables. The level of significance was set at α=0.05.

Ethical Considerations

The study abided by the Principles of the Declaration of Helsinki (2013) and was conducted in accordance with the Guidelines of the International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) Good Clinical Practice (GCP), E6 (R2) and other ICH-GCP guidelines (as amended); National Ethical Guidelines For Health and Health-Related Research (NEG HHRR), 2017. The clinical protocol and all relevant documents included in the study were reviewed and approved by the Institutional Ethics Review Committee of St. Luke's Medical Center–Global City. Since this was a retrospective chart review, informed consent was waived. Patient confidentiality was respected by ensuring the anonymity of patient records. Names of patients were concealed and anonymized by using their respective hospital identification numbers. All data were recorded, and investigators were responsible for the integrity of the data. The manner of disseminating and communicating the study results ensured the confidentiality of patient data. All study-related documents, including all versions of the protocol, ethical clearance, data collection forms, and hard copies of source documents were kept and stored by the principal investigator in strict confidentiality and shredded thereafter.

RESULTS

A total of 739 admitted children were tested over the study period, including 116 in the peak group and 623 in the post-peak group. The majority of the patients in both groups were male and belonged to the 1–5-year-old age group. There were no significant differences observed in terms of demographic characteristics between the two groups (Table 1).

Table 1. Demographic characteristics of children with respiratory panel testing during the two periods

Characteristic	Period		*p-value*
	Peak	Post-peak	
	n=116 (%)	n=623 (%)	
Age (Mean±SD)	3.76±3.70	3.66±3.25	
Age group (in years)			0.766
Less than 1	14 (12.1)	89 (14.3)	
1 to 5	76 (65.5)	415 (66.6)	
6 to 11	19 (16.4)	92 (14.8)	
12 to 18	7 (6.0)	27 (4.3)	
Sex			0.694
Male	73 (62.9)	380 (61.0)	
Female	43 (37.1)	243 (39.0)	

Among the children admitted, 92.02% tested positive for at least one respiratory pathogen, with the majority being viruses. Rhinovirus/enterovirus (50.59%) was the most frequently identified pathogen, followed by RSV (19.71%) and COVID-19 (12.50%). A significant difference in pathogen prevalence was found between periods, with COVID-19 and Influenza A being more common during the peak, and rhinovirus/enterovirus and adenovirus being higher post-peak. Furthermore, although not statistically significant, RSV was noted to be more prevalent during the peak period, while the human coronavirus variants (229E, HKU1, OC43, and NL63) were only detected during the post-peak period. *Chlamydophila pneumoniae* was the only bacterium identified among the four bacterial targets included in the panel (Table 2).

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Table 2. Distribution of respiratory pathogens during and after the peak of the pandemic

Pathogen	Total n(%)	Period		*p-value*
		Peak	Post-peak	
		n(%)	n(%)	
Adenovirus	63 (9.26)	4 (3.4)	59 (9.5)	0.033*
Coronavirus 229E	5 (0.74)	0 (0)	5 (0.8)	0.333[b,c]
Coronavirus HKU1	8 (1.18)	0 (0)	8 (1.3)	0.220[b]
Coronavirus OC43	6 (0.88)	0 (0)	6 (1.0)	0.289[b,c]
Coronavirus NL63	10 (1.47)	0 (0)	10 (1.6)	0.169[b]
MERS-CoV	0 (0)	0 (0)	0 (0)	-
COVID-19	85 (12.50)	30 (25.9)	55 (8.8)	<0.001*
Metapneumovirus	41 (6.03)	5 (4.3)	36 (5.8)	0.526
Rhinovirus/ Enterovirus	344 (50.59)	32 (27.6)	312 (50.1)	<0.001*
Influenza A	56 (8.24)	14 (12.1)	42 (6.7)	0.047*
Influenza A/H1	0 (0)	0 (0)	0 (0)	-
Influenza A/H1-2009	0 (0)	0 (0)	0 (0)	-
Influenza A/H3	0 (0)	0 (0)	0 (0)	-
Influenza B	13 (1.91)	3 (2.6)	10 (1.6)	0.461[b]
Parainfluenza 1	12 (1.76)	2 (1.7)	10 (1.6)	0.926[b]
Parainfluenza 2	17 (2.5)	0 (0)	17 (2.7)	0.072[b]
Parainfluenza 3	37 (5.44)	3 (2.6)	44 (7.1)	0.07
Parainfluenza 4	8 (1.18)	0 (0)	8 (1.3)	0.220[b]
Respiratory Syncytial Virus	134 (19.71)	26 (22.4)	108 (17.3)	0.192
Bordetella pertussis	0 (0)	0 (0)	0 (0)	-
Bordetella parapertussis	0 (0)	0 (0)	0 (0)	-
Chlamydophila pneumoniae	3 (0.44)	1 (0.9)	2 (0.3)	0.400[b,c]
Mycoplasma pneumoniae	0 (0)	0 (0)	0 (0)	-
Total	680 (92.02)	107 (92.24)	573 (91.97)	

*. The Chi-square statistic is significant at the .05 level.
b. More than 20% of cells in this subtable have expected cell counts less than 5. Chi-square results may be invalid.
c. The minimum expected cell count in this subtable is less than one. Chi-square

Children aged 1–5 years had the highest rate of detection for respiratory pathogens. The respiratory pathogen predominantly identified in children aged <1 (34.95%), 1 to 5 (48.47%), and 6 to 11 (54.96%) was rhinovirus/enterovirus, while COVID-19 (26.47%) and rhinovirus/enterovirus (26.47%) were the most frequent in children aged 12-18. Furthermore, RSV was more commonly detected in children under 1 year old, while the prevalence of Influenza A and B viruses increased with age (Table 3).

Respiratory panel results showed that the majority of samples yielded a single pathogen. Bacterial infections with *Chlamydophila pneumoniae* were identified in only three patients, two of whom were co-infected with respiratory viruses, including RSV, rhinovirus/enterovirus, and adenovirus (Table 4). Other viruses frequently observed in co-infections included rhinovirus/enterovirus, adenovirus, RSV, parainfluenza virus, Influenza A and B, human coronavirus, metapneumovirus, and COVID-19.

Table 3. Respiratory pathogens identified in children per age group

Pathogen	Age in years, n(%)			
	<1	1-5	6-11	12-18
None detected	8 (7.77)	30 (6.11)	16 (14.41)	5 (14.71)
Adenovirus	7 (6.79)	50 (10.18)	6 (5.41)	0 (0)
Coronavirus 229E	1 (0.97)	3 (0.61)	1 (0.90)	0 (0)
Coronavirus HKU1	0 (0)	6 (1.22)	1 (0.90)	1 (2.94)
Coronavirus OC43	0 (0)	6 (1.22)	0 (0)	0 (0)
Coronavirus NL63	3 (2.91)	6 (1.22)	1 (0.90)	0 (0)
MERS-CoV	0 (0)	0 (0)	0 (0)	0 (0)
COVID-19	17 (16.5)	48 (9.76)	11 (9.91)	9 (26.47)
Metapneumovirus	7 (6.79)	27(5.49)	5 (4.50)	2 (5.88)
Rhinovirus/ Enterovirus	36 (34.95)	238 (48.47)	61 (54.96)	9 (26.47)
Influenza A	4 (3.88)	38 (7.74)	10 (9.01)	4 (11.76)
Influenza A/H1	0 (0)	0 (0)	0 (0)	0 (0)
Influenza A/H1-2009	0 (0)	0 (0)	0 (0)	0 (0)
Influenza A/H3	0 (0)	0 (0)	0 (0)	0 (0)
Influenza B	0 (0)	9 (1.83)	3 (2.70)	1 (2.94)
Parainfluenza 1	1 (0.97)	8 (1.63)	2 (1.80)	1 (2.94)
Parainfluenza 2	2 (1.94)	13 (2.65)	2 (1.80)	0 (0)
Parainfluenza 3	6 (5.83)	40 (8.15)	1 (0.90)	0 (0)
Parainfluenza 4	0 (0)	6 (1.22)	1 (0.90)	1 (2.94)
RSV	34 (33.01)	92 (18.74)	7 (6.30)	1 (2.94)
Bordetella pertussis	0 (0)	0 (0)	0 (0)	0 (0)
Bordetella parapertussis	0 (0)	0 (0)	0 (0)	0 (0)
Chlamydophila pneumoniae	0 (0)	2 (0.41)	0 (0)	1 (2.94)
Mycoplasma pneumoniae	0 (0)	0 (0)	0 (0)	0 (0)
Total	103 (100)	491 (100)	111 (100)	34 (100)

Table 4. Respiratory panel results of children as to the type of infection

Type of infection	n(%)
Single infection	558 (75.5)
Other Viruses	505 (90.5)
COVID-19	52 (9.32)
Bacterial	1 (0.18)
Co-infection	122 (16.51)
Virus + virus	111 (90.98)
COVID-19 + other pathogen/s	9 (7.38)
Virus + bacteria	2 (1.64)
None detected	59 (7.98)
Total	739 (100)

The most common respiratory symptom reported among all patients was cough (89.99%), followed by colds/nasal congestion (71.72%) and fever (66.98%). Bronchial asthma (12.72%) was the most frequently identified underlying condition among patients with comorbidities (Table 5).

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Table 5. Clinical presentation and comorbidities of children with respiratory panel testing

Symptom/s	n(%)
Cough	665 (89.99)
Colds/ Nasal congestion	530 (71.72)
Fever	495 (66.98)
SOB or DOB	249 (33.69)
Sore throat	34 (4.60)
Others	11 (1.49)
Comorbidity	**n=214 (%)**
Bronchial asthma	94 (12.72)
Allergic rhinitis	22 (2.98)
Neurologic	14 (1.89)
Hematology	9 (1.22)
Other pulmonary	7 (0.95)
Cardiovascular	7 (0.95)
Gastroenterology	5 (0.68)
Tuberculosis	4 (0.54)
Malignancy	3 (0.41)
Others	49 (6.63)

When comparing according to the type of infection, a significant difference was observed in all clinical symptoms, except for cough (*p=0.99*). Single infections were more likely to be detected when patients present with cough and difficulty of breathing/shortness of breath, while co-infections are more likely to be detected when patients present with colds/nasal congestion and fever (Table 6).

Table 6. Comparison of signs and symptoms according to the type of infection

Sign/Symptom	Single infection n(%)	Co-infection n(%)	None detected n(%)	*p-value*
Cough	481 (72.33)	131 (19.70)	53 (7.97)	0.99
Colds/ Nasal congestion	382 (72.08)	119 (22.45)	29 (5.47)	0.0001*
Sore throat	22 (64.71)	5 (14.71)	7 (20.59)	0.02*
Fever	332 (67.07)	122 (24.65)	41 (8.28)	0.0001*
SOB/DOB	193 (77.51)	36 (14.46)	20 (8.03)	0.03*
Others	7 (63.64)	1 (9.09)	3 (27.27)	0.05*

In the 702 patients who underwent complete blood count, the majority had leukocyte counts within the normal range. Among the 649 patients who had chest X-rays, 50.23% had normal results. Those with abnormal findings were predominantly observed to have single infections identified on the respiratory panel. Overall, the diagnostic test results of the patients with respiratory symptoms did not show significant differences, regardless of whether no pathogen, a single pathogen, or multiple pathogens were detected in their respiratory panel (Table 7).

Table 7. Laboratory and radiographic findings of children with respiratory symptoms

Diagnostic Test	Total n(%)	Single infection n(%)	Co-infection n(%)	None detected n(%)	*p-value*
Leukocyte count	*n=702*	*n=508*	*n=137*	*n=57*	
Normal	492 (70.09)	346 (70.32)	105 (21.34)	41 (8.33)	
Leukocytosis	122 (17.38)	91 (74.59)	21 (17.21)	10 (8.20)	0.3
Leukopenia	88 (12.54)	71 (80.68)	11 (12.50)	6 (6.82)	
Neutrophils (%)		59.27±(21.27)	58.04±(19.51)	62.40±(19.00)	0.06
Lymphocytes (%)		31.66±(19.95)	33.33±(18.76)	28.81±(17.17)	0.23
Platelet count (×10^3/µl)		342,512±(117141.08)	355,786±(132217.66)	376,903±(140272.99)	0.24
C-Reactive Protein	*n=186*	*n=132*	*n=30*	*n=24*	
Normal	88 (47.31)	66 (75.00)	13 (14.77)	9 (10.23)	0.47
Elevated	98 (52.69)	66 (67.35)	17 (17.35)	15 (15.31)	
Procalcitonin	*n=126*	*n=87*	*n=20*	*n=19*	
Normal	86 (68.25)	64 (74.42)	13 (15.12)	9 (10.47)	
Equivocal	19 (15.08)	9 (47.37)	3 (15.79)	7 (36.84)	0.06
Elevated	21 (16.67)	14 (66.67)	4 (19.05)	3 (14.29)	
Chest X-ray	*n=649*	*n=473*	*n=124*	*n=52*	
Normal	326 (50.23)	238 (73.01)	61 (18.71)	27 (8.28)	0.94
Abnormal	323 (49.77)	235 (72.76)	63 (19.50)	25 (7.74)	
Chest X-ray Findings	*n=323*				
Hazy opacities	31 (9.60)	20 (64.52)	9 (29.03)	2 (6.45)	
Streaky/linear densities	23 (7.12)	19 (82.61)	4 (17.39)	0 (0)	
Hazy and streaky opacities	30 (9.29)	22 (73.33)	7 (23.33)	1 (3.33)	
Increased peribronchial cuffing	14 (4.33)	13 (92.86)	0 (0)	1 (7.14)	0.3
Consolidation	10 (3.10)	5 (50)	1 (10)	4 (40)	
2 or more findings	171 (52.94)	120 (70.17)	37 (21.64)	14 (8.19)	
Others	41 (12.69)	35 (85.37)	4 (9.76)	2 (4.88)	

Pneumonia was the most frequent clinical diagnosis among all the admitted patients during both periods, with a notably higher proportion in the post-peak period. On the other hand, a significant reduction in the number of COVID-19 cases was observed in the post-peak period (Table 8).

Table 8. Clinical diagnosis of children admitted to SLMC-GC during the peak and post-peak period

Clinical Diagnosis	Total n(%)	Period		*p-value*
		Peak n(%)	Post-peak n(%)	
Pneumonia	304 (41.1)	37 (31.9)	267 (42.9)	0.028
Bronchial Asthma	81 (10.9)	10 (8.6)	71 (11.4)	0.38
COVID-19	76 (10.3)	25 (21.6)	51 (8.2)	<0.001
Nasopharyngitis/URTI	69 (9.3)	15 (12.9)	54 (8.7)	0.147
Influenza	61 (8.3)	13 (11.2)	48 (7.7)	0.208
Hyperactive/Reactive Airway Disease	51 (6.9)	6 (5.2)	45 (7.2)	0.424
Bronchiolitis	49 (6.6)	7 (6.0)	42 (6.7)	0.779
Croup	21 (2.8)	6 (5.2)	15 (2.4)	0.1
Bronchitis	20 (2.7)	1 (0.9)	19 (3.0)	0.182
Tonsillitis/ Pharyngitis	10 (1.4)	0 (0.0)	10 (1.6)	0.169
Others	81 (10.9)	12 (10.3)	69 (11.1)	0.817

The management of patients in terms of medications did not show significant differences. Antibiotics were administered to 53.31% regardless of the identified pathogen type. Antivirals were given to 10.1% (75/739) of the patients; all individuals who tested positive for the Influenza virus received Oseltamivir,

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while those diagnosed with severe COVID-19 were treated with Remdesivir. On the other hand, a significant difference was observed in terms of the need for oxygen support, with 66.30% of patients not requiring oxygen. Among the patients who needed oxygen, only patients with single-pathogen detections required invasive oxygen support (Table 9).

Table 9. Management of children with respiratory symptoms admitted to SLMC-GC

Management	Total n(%)	Single infection n(%)	Co-infection n(%)	None detected n(%)	*p-value*
Supportive only	274 (37.08)	202 (73.72)	49 (17.88)	23 (8.39)	0.07
Given antibacterial	394 (53.31)	284 (72.08)	75 (19.04)	35 (8.88)	
Given antiviral	49 (6.63)	32 (65.31)	17 (34.69)	0 (0)	
Given both	22 (2.98)	16 (72.26)	5 (22.73)	1 (4.55)	
Oxygen support					
Without	490 (66.30)	339 (69.18)	107 (21.84)	44 (8.98)	0.03*
With	249 (33.69)	195 (78.31)	39 (15.66)	15 (6.02)	
Invasive	8 (3.21)	8 (100)	0 (0)	0 (0)	0.32
Non-invasive	241 (95.79)	187 (77.59)	39 (16.18)	15 (6.22)	

In terms of patient disposition, the majority were admitted to the ward, with no significant difference observed. However, there was a significant difference in patient discharge outcomes, with 99.86% of patients being discharged. Lastly, the average length of hospital stay was 3.69 days (±4.06), with significantly longer stays observed in patients with no pathogen detected. (Table 10).

Table 10. Disposition and outcomes of children with respiratory symptoms admitted to SLMC-GC

Outcome	Total	Single infection n(%)	Co-infection n(%)	None detected n(%)	*p-value*
Admitted to Ward	686 (92.83)	494 (72.01)	140 (20.41)	52 (7.58)	0.13
Admitted to ICU	53 (7.17)	40 (75.47)	6 (11.32)	7 (13.21)	
Discharge Status					
Discharged	738 (99.86)	534 (72.36)	146 (19.78)	58 (7.86)	0.003*
Expired	1 (0.14)	0 (0)	0 (0)	1 (100)	
Hospital stay (days)	3.69±(4.06)	3.57±(4.37)	3.31±(2.05)	4.63±(4.78)	0.04

DISCUSSION

Respiratory Pathogen Prevalence

During the study period, 680 (92.02%) children tested positive for at least one respiratory pathogen. This percentage was notably higher than the detection rates reported in earlier studies using conventional or multiplex PCR, which ranged from 65.6% to 76.6%.[14-17] The respiratory pathogens identified in this study were predominantly viruses (98.84%), consistent with the findings from a local study on children with acute respiratory infections where viruses were prevalent at a rate of 76.6%.[15] This pattern was also observed in a study in Morocco among hospitalized children with severe respiratory infections before and during the COVID-19 pandemic, where viruses were similarly identified as the most prevalent pathogens.[17] The high percentage of these cases being viral confirms the predominance of viral etiology in respiratory tract infections among children.[17,18]

Predominant Viral Pathogens

Human Rhinovirus/Enterovirus

Human rhinovirus has been recognized as a major virus causing respiratory tract infections in children.[21,22] Our study also consistently found rhinovirus/enterovirus to be the most frequently detected pathogen during the peak (27.6%) and post-peak periods (50.1%) across all age groups. Several studies have reported either a high incidence or persistence of rhinovirus and respiratory enterovirus infections during the COVID-19 pandemic.[16,23-25] Recognized for its resistance to disinfectants containing ethanol, its ability to survive in the environment for long periods[26], and its frequent transmission among family members[27], characteristics of these non-enveloped viruses may suggest that epidemic prevention policies may have less of an impact on its infection rates.[16] However, a higher prevalence of rhinovirus/enterovirus was observed during the post-peak period, possibly due to the relaxation of non-pharmaceutical interventions (NPIs) implemented to combat the pandemic.

Respiratory Syncytial Virus

RSV has consistently emerged as one of the leading pathogens causing respiratory tract infections in children.[6,7,17] Despite the COVID-19 pandemic leading to a decline in cases and deviation from the usual seasonal pattern of RSV, studies have shown no significant difference in case numbers compared to previous years.[17] Our study also found RSV to be among the most frequently detected pathogens, with no significant difference noted during (22.4%) and after (17.3%) the peak of the pandemic. This trend highlights the enduring presence of RSV as a significant pathogen, despite fluctuations related to the pandemic.

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Adenovirus and Other Viral Pathogens

Similar patterns of persistence were also observed for other non-enveloped viruses, including adenovirus, which displayed persistent but fluctuating frequencies as the COVID-19 pandemic progressed.[25] In our study, adenoviruses were still present during the peak period but showed a significant increase during the post-peak period. In contrast, during the peak period, cases of Influenza A were higher, with no documented cases of seasonal human coronavirus and a lower proportion of human metapneumovirus and human parainfluenza viruses were observed. Hospital surveillance data from South Korea also indicated declines in the monthly detection rates of enveloped viruses, including parainfluenza virus, seasonal human coronavirus, and human metapneumovirus.[25] Conversely, in a US pediatric hospital, no cases of hospitalization associated with influenza virus, RSV, human metapneumovirus, human parainfluenza, or seasonal human coronavirus infections were reported until the end of 2020.[24]

Co-infections and Symptom Profiles

Respiratory panels provide the advantage of detecting multiple pathogens in a single sample. Although a higher percentage of patients in this study showed single-pathogen detection (75.5%), co-infections are also common in respiratory tract infections.[16] Rhinovirus/enterovirus, adenovirus, RSV, parainfluenza virus, Influenza A & B, human coronavirus, metapneumovirus, and COVID-19 were the most frequently involved co-infection pathogens, findings comparable to other studies.[28-30] Despite the variety of pathogens, these viral infections can be clinically unpredictable and present symptoms that are difficult to differentiate from those caused by bacterial infections.[31,32] Our findings indicated that cough can be present regardless of the type of infection. Difficulty breathing or shortness of breath was more commonly associated with single infections, while fever and colds/nasal congestion were more likely to occur in cases of co-infections. In a study, fever and cough were found to be the two primary significant predictors for virus co-detection, while fever alone was identified as a significant predictor for bacteria co-detection. Moreover, cases where both virus and bacteria were detected showed a notably higher prevalence of fever, cough, and sputum compared to cases where only one pathogen was identified.[28] However, even children without detectable microorganisms on their swab tests also exhibited symptoms such as upper airway obstruction, shortness of breath, cough, fever, and pharyngitis.[31] The precise role of pathogens in causing respiratory symptoms versus merely colonizing the respiratory tract during symptomatic episodes remains unclear. It can be speculated that the presence of a pathogen does not always lead to respiratory symptoms, and the severity of illness might be influenced by various factors related to the host or environmental conditions.[33]

Diagnostic and Laboratory Findings

Diagnosing respiratory tract infections is typically confirmed based on clinical signs and symptoms, along with diagnostic tests. However, in the present study, no significant differences were observed in terms of blood counts, C-reactive protein, procalcitonin, and chest X-ray findings based on the respiratory panel results, a finding similar to other studies, which reported no significant disparities in blood counts and C-reactive protein levels based on RT-PCR assay results.[31,32] A local study noted an increased probability of a positive respiratory panel result when a chest X-ray is positive. However, 4.3% of patients tested negative on the respiratory panel which could potentially be attributed to other bacterial pathogens that are not included in the panel.[15] This study also found that 72.76% of patients with abnormal chest X-ray findings were likely positive for a single pathogen, whereas 7.74% also had negative respiratory panel results. Additionally, 52.94% exhibited multiple abnormalities on chest X-ray, and 3.10% showed signs of consolidation, suggesting the presence of potentially severe bacterial lung infections. These observations may suggest that while respiratory panel testing can be a reliable method for diagnosing specific infections, it may not be able to exclude concurrent lung infections caused by other bacterial pathogens. Therefore, the identification of concurrent viral and bacterial infections primarily relies on clinical assessment and general laboratory tests,[34] or potentially on the use of an expanded multiplex RT-PCR respiratory panel that includes a broader range of bacterial pathogens. The use of inflammatory markers such as CRP and procalcitonin has been frequently evaluated to aid in distinguishing between viral and bacterial infections, as their levels tend to be higher in bacterial infections.[35] Our study found that 52.69% of patients tested had elevated CRP levels, while 16.67% had elevated procalcitonin levels. Given the

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complexity of children with respiratory infections requiring hospital admission and the concern for potential viral or bacterial co-infections, physicians frequently opt to initiate empirical treatment for possible pneumonia or other serious bacterial infections rather than risk the patient's clinical status worsening.[34] Moreover, healthcare providers' decisions regarding antibiotic use in the emergency department or ambulatory setting may be influenced more by clinical factors such as physical examination findings or patients' medical histories, rather than solely relying on mPCR test results.[36] This likely explains why a significant proportion (53.31%) of our patients received antibiotics despite the majority having tested positive for viruses.

Hospitalization and Clinical Outcomes

All patients who required invasive oxygen support were found to have single-pathogen infections. This contrasts with a U.S.-based study, which reported that 21% of hospitalized children had co-infections, and these were associated with an increased likelihood of requiring oxygen support and ICU admission.[2] Our study likewise found no significant differences in the length of hospital stay or rates of ward and ICU admission between patients with single infections and those with co-infections, except in terms of discharge status. This aligns with previous studies, which also reported no significant differences in hospitalization duration, ICU stay, or in-hospital mortality when comparing patients with negative detections, single infections, and co-infections.[32,37] Notably, the sole mortality recorded in our study involved a patient burdened with several comorbidities, including cerebral palsy, bronchomalacia, and chronic lung disease, who experienced recurrent systemic infections. Interestingly, this patient had a negative respiratory panel result. This implies that the specific type of infection detected does not directly correlate with the duration or outcome of hospital stays. Instead, factors such as complications necessitating prolonged medical intervention or the presence of multiple underlying health conditions appeared to contribute to prolonging hospital stays. This highlights the significance of considering the broader health context of patients, beyond just the specific infectious agents involved, when evaluating and managing hospitalization outcomes.

LIMITATIONS AND RECOMMENDATIONS

This study was retrospective in nature and included only hospitalized pediatric patients, thus potentially limiting its representation to those with more severe or complex medical conditions. As a result, the findings may not fully reflect the broader population of children with respiratory infections.

Another limitation was the unequal distribution of samples between the two study periods, with a ratio of 1:5.4. The sample size, however, was calculated based on a minimum detectable effect size of 8.13% (21.1% vs. 12.9%) with 90% power. Some analyses have likely had limited statistical power due to smaller group sizes, and estimates for rare events may have been associated with wide confidence intervals and should therefore be interpreted with caution.

Furthermore, not all patients underwent the full set of diagnostic tests included in the study variables, which may have affected the completeness and representativeness of the respiratory panel results. It should also be noted that the respiratory panel used in this study included only four bacterial targets (*Bordetella pertussis, Chlamydophila pneumoniae, Mycoplasma pneumoniae, and Bordetella parapertussis*). Consequently, the high proportion of viral detections observed may, in part, reflect this limited bacterial coverage and cannot exclude the possibility of undetected bacterial co-infections.

Lastly, the observational nature of this study precludes conclusions about causality, and the results should be viewed as descriptive rather than confirmatory.

To address these limitations, future studies may consider including a wider range of patient populations, such as outpatients and individuals in community settings, to provide a more comprehensive understanding of the epidemiology and impact of respiratory pathogens. Collaborations across multiple healthcare institutions may also enhance the generalizability and external validity of findings, ensuring their applicability across diverse pediatric populations and healthcare settings nationwide. Moreover, including all patients who underwent respiratory panel testing, regardless of symptom presentation, could provide deeper insights into pathogen prevalence and their potential role in various clinical outcomes beyond respiratory illness alone.

CONCLUSION

This study identified a range of respiratory pathogens among pediatric patients using the BioFire® Respiratory 2.1 Panel, with notable differences in pathogen prevalence observed between the peak and post-peak periods of the COVID-19 pandemic. While viral infections remained predominant in both periods, certain pathogens, such as COVID-19 and Influenza A, were more common during the peak, whereas rhinovirus/enterovirus and adenovirus were more frequently detected post-peak. Children aged 1–5 years had the highest detection rates, with age-related trends noted for RSV and Influenza. While clinical symptoms varied by infection type, cough was consistently the most common. No significant differences were observed in diagnostic results, ward or ICU admissions, or treatment approach, although only single-pathogen infection cases required invasive oxygen support. Patients without detected pathogens had longer hospital stays, and a significant difference was noted in discharge outcomes. These results reflect the dynamic nature of pediatric respiratory infections and reinforce the value of continuous pathogen monitoring throughout and beyond the pandemic.

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CONFLICT OF INTEREST

None declared.

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