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**Original Research Article**

# Study of association of respiratory viruses in the etiology of acute lower respiratory tract infections in children and correlation with clinical and laboratory features: role of emerging new viruses

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## ABSTRACT

**Background:** Viruses are one of the major causes of childhood pneumonia with the respiratory syncytial virus getting great attention as an important organism for pneumonia.
**Methods:** This cross-sectional study was conducted in the Department of Paediatrics, Sree Gokulam Medical College and Research Foundation, Thiruvananthapuram, Kerala from May 2022 TO November 2022. 119 children admitted with lower respiratory tract infections were included in this study. Symptoms, signs, and investigation reports including PCR and clinical course in the hospital were recorded.
**Results:** 25% of children were in the age group less than 1 year, 52% were between 1-5 years and 23% were above 5 years. Viruses were isolated in 82 patients (68.9%). The main viruses were respiratory syncytial virus (23.5%), boca virus (16.14%), influenza A (7.5%), influenza B (4.2%), meta pneumonia virus (3.3%), and para influenza virus (2.5%). Patients with boca virus infections had a more severe clinical course.
**Conclusions:** Molecular testing with PCR along with clinical and lab parameters will help us to have more insights into the etiology and clinical presentation of respiratory infections in children and help us to do optimum management avoiding unnecessary antibiotic usage.

**Keywords:** Association, Lower respiratory tract infections, Polymerase chain reaction, Viruses

## INTRODUCTION

Acute respiratory infections are the most common causes of both illness and mortality in children under five, with an average of three to six episodes occurring annually.[1] Recent studies suggest that viruses are one of the major causes of childhood pneumonia with respiratory syncytial virus getting great attention as an important organism for pneumonia.[2] There has been a reduction in bacterial pneumonia due to enhanced vaccination coverage against pneumococci and *H. influenza*. Lack of exclusive breastfeeding during the first six months, improper timing and content of complementary feeding, anemia, undernutrition, indoor pollution due to tobacco smoking, use of coal and wood for cooking food, and lack of vaccinations are important risk factors. Respiratory viruses account for 40 to 50 percent of infection in hospitalized infants and children in developing countries as per a study by Hortal et al, 1990; John et al; Tupasi and others in 1990 with a case-fatality rate ranging from 1.0 to 7.3 percent.[3] Bronchiolitis in children occurs predominantly in the first year of life and with decreasing frequency in the second and third years.[4] RSVs are the main cause of bronchiolitis worldwide and can cause up

to 70 or 80 percent of LRIs during the season (Simoes 1999; Stensballe, Devasundaram, and Simoes 2003).[5] Similar findings were noted by Mathew et al.[6] Human metapneumovirus also causes bronchiolitis (Van den Hoogen and others) that is indistinguishable from RSV disease.[7] Other viruses that cause bronchiolitis include parainfluenza virus type 3 and influenza. The clinical features are rapid breathing and lower chest wall indrawing, fever in one-third of cases, and wheezing (Cherian et al 1990).[8] Inflammatory obstruction of the small airways, which leads to hyperinflation of the lungs, and collapse of segments of the lung occur. Newer viruses like boca virus, and human metapneumoviruses are also being identified. The application of molecular diagnostic methods has improved our understanding of viral epidemiology. A better understanding of the epidemiology and immunopathogenesis of respiratory viral infections will provide opportunities for early identification effective management, and avoidance of unnecessary antibiotics thereby reducing morbidity and mortality. Study aim was to study the frequency of viral etiology in the causation of acute lower respiratory tract infections in children and to correlate the type of virus isolated with clinical and laboratory features.

### *Operational definitions*

WHO definition: The upper respiratory tract consists of the airways from the nostrils to the vocal cords in the larynx, including the paranasal sinuses and the middle ear. The lower respiratory tract covers the continuation of the airways from the trachea and bronchi to the bronchioles and the alveoli. Acute lower respiratory tract infection is diagnosed in a patient with fever, cough, breathlessness of less than 28 days duration with findings of any of the following- tachypnoea, chest indrawing, increased work of breathing, repetitions, reduced breath sounds, wheeze. WHO has categorized pneumonia in under-five children into 2 categories. Tachypnoea with or without retractions is called pneumonia whereas tachypnoea with any danger signs like unable to feed or drink, hypothermia, unconsciousness, convulsions, cyanosis, grunting, head nodding is categorized as severe pneumonia. WHO has defined tachypnea as > 60/minute in infants less than 2 months, > 50/minute in infants 2 months to 12 months, and > 40/minute in the age group 1-5 years.

## METHODS

This was a cross-sectional study conducted from May 2022 to November 2022 at Department of Paediatrics, Sree Gokulam Medical College and Research Foundation, Venjarammoodu, Thiruvananthapuram.

### *Inclusion criteria*

Children admitted with acute lower respiratory tract infections whose parents have given consent for the study were included.

### *Exclusion criteria*

Children with reactive airway disease, children with congenital heart disease, children with chronic lung disease, children with congenital anomalies, children with covid 19 infection confirmed by RT PCR were excluded.

### *Sample size calculation*

According to the literature, viruses are implicated in 60% of cases of acute respiratory tract infections in children. Population size (for finite population correction factor, N) was 100. Hypothesized % frequency of outcome factor in the population (p) was 60%+/-5. Confidence limit as % of 100 was 95% with design effect of 1. Taking a 95% confidence level, a sample size of 79. Consecutive sampling method was used as sampling technique.

Sample size n = [DEFF * Np(1-p)]/[d2/Z2 * (N-1)+p*(1-p)]

### *Study procedure*

Parents of children admitted to the pediatric inpatient department were interviewed and explained about the study, in their language and those who fit into the inclusion/exclusion criteria were included. 119 children were included in the study. After taking the informed consent, relevant data was collected on a predesigned questionnaire regarding biodata, age, sex, birth weight, birth history, past medical history, contact with the infection, clinical features, and clinical examination findings. Clinical examination of the child was done including general examination, anthropometry, and vital signs including heart rate, respiratory rate, blood pressure, temperature, $SPO_2$, work of breathing, added sounds, and air entry was recorded. The severity of illness is classified as mild, moderate, and severe based on the scoring system. Growth parameters including height, weight, and mid-arm circumference were checked to rule out malnutrition. Data on management and outcome of the cases were collected from the medical records. The severity of the disease was defined according to the IMCI guidelines developed by the WHO. Complete blood count, CRP, and chest X-ray were taken. Nasopharyngeal swabs, blood, and urine samples were sent for viral study.

### *Statistical analysis*

Data was analyzed using SPSS software. Qualitative data is expressed in percentages and quantitative data was expressed as mean±SD. The chi-square test and independent sample “t” test were used to establish an association.

## RESULTS

Of the total 119 cases, 64 were males and 55 were females with a male: female ratio of 1.16:1 (Figure 1).

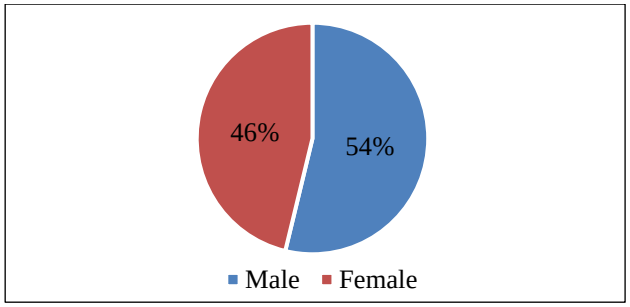


**Figure 1: Gender distribution of the cases.**

Total 25% of children were in the age group less than 1 year, 52% were between 1-5 years and 23% were above 5 years (Figure 2). 80.67% were term babies and 19.33% were preterm babies at birth. 20.16% were born as low birth weight babies and 79.84% were babies with normal birth weight. Contact with respiratory infection was seen in 77.2% of children.

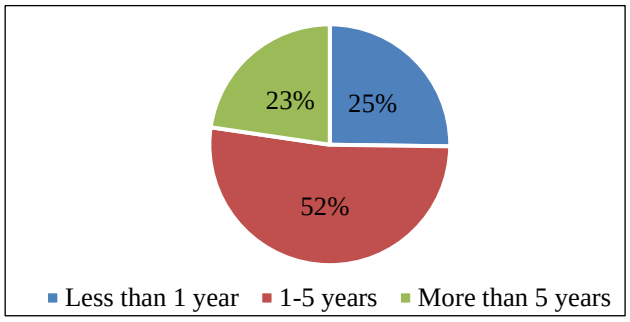


**Figure 2: Age distribution of the cases.**

The presenting complaint was a cough in all the cases. Fever was present in 92 patients (77.3%). There was no fever in 27 patients (22.7%). Duration of fever was 1-3 days in 62 patients (50.4%), 4-7 days in 22 patients (18.4%), and 7-14 days in 6 patients (5%). Coryza was present in 86 (72.2%) cases. Breathlessness was present in 48 cases (40.33 %). Diarrhoea was seen in 6 patients. Vomiting was seen in 18 patients (15.1%). Abdominal pain was seen in 8 patients (Figure 3). GI symptoms were more in patients with influenza infection. Tachypnoea was present in 31% of total cases. Increased work of breathing was seen in 31 (26.1%) of cases. One patient with combined boca and norovirus infection had diarrhea with severe dehydration along with breathlessness.

PICU admission was seen in 22 (26.8%) of cases. The mean duration of PICU stay was 3.5 days. Maximum PICU admission was for bocavirus infections. NIV was required for 15 (12.6%) of cases of which 7 had boca virus infection. Invasive ventilation was not required for any of the patients. The mean duration of hospital stay was 6.5 days for patients with the boca virus.

Viruses were isolated in 82 patients (68.9%) of the sample studied. No viral organism was isolated in 37 (31.09%) of cases. The viruses isolated were respiratory syncytial virus in 28 (23.5%) patients, boca virus in 21 (16.14%), influenza A in 9 (7.5%), influenza B in 5 (4.2%), meta pneumonia virus in 4 (3.3%), para influenza virus in 3 (2.5%), rhinovirus in 3 (2.5%) patients (Figure 4).

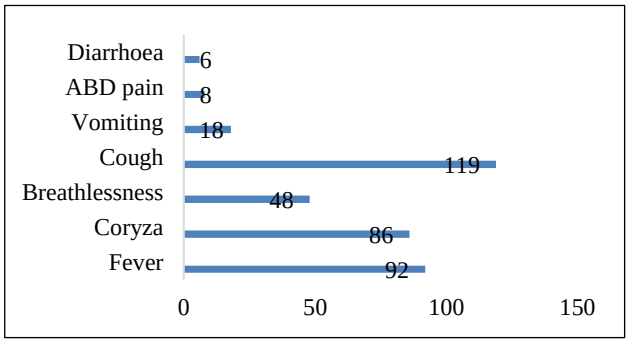


**Figure 3: Distribution of clinical features.**

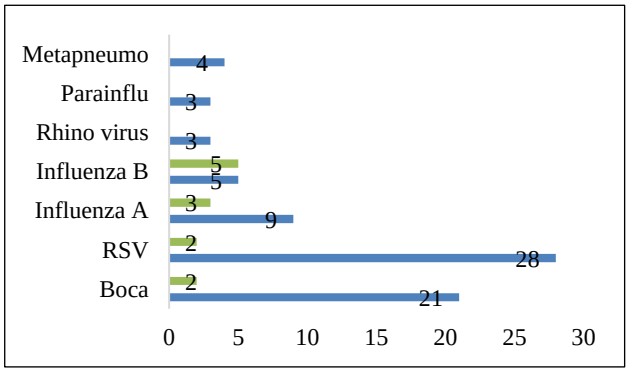


**Figure 4: Pattern of virus isolation from the study population.**

Of the total 82 cases with virus isolation, 77 (93.9%) had a single infection and 5 cases (6.09%) had coinfections. In 5 cases, two viruses were detected. None of the cases had detected more than 2 viruses.

All the babies with boca virus infection were below 5 years of age, 11 were below 1 year (52.3%), and 10 were between 1 and 5 years (47.6%). 3 patients had no fever, but only cough and breathlessness. 12 patients with boca virus infection required PICU admission (57.1%). 7 patients (58.3%) out of this required CPAP while the rest were on supplemental oxygen with prongs.

Total 16 patients with RSV infection were below 1 year of age, 8 patients were between 1-5 years and 4 were above 5 years. All patients with RSV had fever and cough as presenting symptoms. Five patients with RSV infection needed ICU admission.

Of the total sample studied, 47 cases (39.4%) had haziness and infiltrates, 25 cases had increased bronchovascular markings (21.2%) and 24 cases had hyperinflation (20.1%), consistent with the WHO criteria. Chest x-ray was normal in 23 patients.

A total count above 17000 was seen in 3 patients with bocavirus infection. A total count of more than 15000 was seen in 5 patients with RSV infection. Total WBC count was between 5000-10000 in 41 patients (34.45%), below 5000 in 8 patients (6.7%), between 10000-15000 in 26 patients (21.8%), 15000-19,999 in 19 patients (15.9%), above 20000 in 12 (13.45%) patients studied. None of the patients with virus isolation had a count above 20000. Contrary to the usual belief, a WBC count of more than 15000 was seen in patients with boca virus and RSV infection.

C-reactive protein (CRP) analyses were ordered in all 119 cases. CRP was less than 30 in all patients with bocavirus infection. The mean value of CRP for RSV patients was 6.46 and for boca virus patients was 5.02. Only 1 patient with RSV infection had a CRP of more than 30 and an ESR of more than 30. None of the Boca virus cases had an ESR of more than 30.

In cases where antibiotic treatment was started initially, it was stopped when the virus was isolated and there was improvement in clinical symptoms. There was no mortality in the study.

## DISCUSSION

Worldwide, respiratory infections are the most common cause of hospitalization in children. Community-acquired pneumonia is most likely of viral etiology, with respiratory syncytial virus being the most common pathogen, especially in children younger than two years. Using blood or NPA samples, multiplex PCR platforms can distinguish between viral and bacterial etiology and can identify more than one organism at the same time.[9] PCR can also serotype the microorganisms. As per a study by Smith et al, RSV is the most common cause of bronchiolitis and is transmitted through respiratory droplets either directly from an infected person or self-inoculation by contaminated secretions on surfaces.[10] RSV-related hospitalization and deaths were more common among children below 6 months of age with 93% happening in developing countries.

In our study also, the most common organism isolated was RSV (23.5%). The second most common virus isolated was boca virus (16.14%) pointing to the role of emerging viruses. Also, in our study, maximum RSV isolation was in infancy. A study Choi et al found that viruses were identified in 60.6% of patients.[11] RSV-23.7%, HBoV-11.3%, adenovirus-6.8%, parainfluenza virus-3 -6.2%, rhinovirus in 5.8%, hMPV 4.7%, influenza A-4.7%, PIV-1-1.7%, influenza B 1.7%, and HCoV-1.6%). Coinfections with viruses were observed in 11.5% as per this study. In our study, 93.9% had a single infection and 6.09% had coinfections. A study by Mathew et al found that the majority of CAP had multiple pathogens (bacteria and viruses).[9] Adenoviruses were recovered from nasal aspirate specimens of 5.9% of children with LRTI as per Hong et al found that 75% of children had respiratory viruses (RSV only-29%, RSV and non-RSV virus coinfection -6% and non-RSV viruses -40%).[12] Adenoviruses were recovered in 5.9% of children as per this study. Hindupur et al found that infection with influenza A virus was more than influenza B virus.[13] A study Priya et al carried out in Chennai, showed that 12.5% of children with ARI were positive for influenza viruses.[14] A study by Rhedin et al found that out of a total of 121 cases, of which 93 cases met the WHO criteria for radiological pneumonia, viruses were detected in 81% of the cases.[15] Influenza virus, metapneumovirus, and respiratory syncytial virus were detected in 60% of cases and were significantly associated with CAP.

A study by Farshad et al in 202 hospitalized children ages between 1mo-5 year showed that viral agents were identified in 54% of cases with 9 cases having a dual infection (parainfluenza virus-15.8%, RSV-12.9%, influenza A-7.4%, influenza B-3.5%, and adenovirus-5.9%.[16] Emhofer et al, found that influenza accounts for LRI in children of all ages.[17] Yun et al found that the pathogens identified were RSV (27.2%), parainfluenza virus type 3 (7.8%), influenza A (3.9%), adenovirus (3.9%), parainfluenza virus type 1 (1.7%), influenza B (1.4%), parainfluenza type 2 (0.5%), measles (0.1%) and others (0.9%).[18]

A study by Kelly et al found that out of the enrolled 310 children, 55% were male with 34% children presenting with severe pneumonia.[19] In our study male: female ratio was 1.16:1.

A study by Cherian et al in South India showed that 76% of bronchiolitis cases were less than 1 year of age and 94% were less than 2 years of age.[8] In our study, 25% of children were in the age group less than 1 year, 52% were between 1-5 years and 23% were above 5 years.

Children with fast breathing or indrawing were classified as having pneumonia.[20] The study by Cherian et al showed that in CAP, 95% had tachypnea and had 93%subcostal retraction.[8] As per Dustin et al, typical presenting signs and symptoms were tachypnea, cough, fever, and anorexia.[10] In our study, the symptoms were fever (77.4%), coryza (72.2%), and breathlessness (40.33%). Signs were tachypnoea (31%), and increased work of breathing (26.1%). A study by Priya et al showed that fever and cough were the predominant symptoms among positive influenza A and B cases.[14]

Findings most strongly associated with an infiltrate on chest radiography in children with clinical pneumonia are grunting, history of fever, retractions, crackles, tachypnea, and the overall clinical impression. Chest radiography should be ordered if the diagnosis is uncertain, if patients have hypoxemia or significant respiratory distress, or if patients fail to show clinical improvement within 48 to 72 hours after initiation of antibiotic therapy. Of 88 children of whom

roentgenographs were taken, 30 (34%) had evidence of pneumonia as per a study by Cherian et al.[8]

As per Emhofer et al, infants with RSV had a serious clinical course, with pneumonia, bronchiolitis, or obstructive bronchitis, but with no characteristic pattern in laboratory tests.[17] Pulmonary X-rays of the RSV-infected infants revealed interstitial pneumonia with or without pulmonary infiltration with hyperinflation. According to Boivin G, human metapneumovirus infection caused pneumonitis (66.7%) and bronchiolitis (58.3%) and 40% were infants aged <15 months.[21]

In our study, PICU admission was seen in 22 (26.8%) of cases. The mean duration of PICU stay was 3.5 days. Maximum PICU admission was for bocavirus infections. NIV was required for 15 (12.6%) of cases of which 7 had boca virus infection. The study by Kelly et al showed that 34% of children failed treatment at 48 hours and 5.8% died.[19] Median [IQR] length of stay was 3.8 [1.9, 7.9] days with 60% of children receiving supplemental oxygen, 10% requiring CPAP, and 2% requiring mechanical ventilation. The study by Hall et al found that strains of subgroups A2 and B4 of RSV were associated with the highest rates of intensive care admissions.[22] Papadopoulos et al found a more severe clinical score index in RSV-A infection based on respiratory rate, wheezing, heart rate, difficulty in feeding, and oxygen saturation.[23] However, no statistically significant difference was found in duration of hospitalization or need of intensive care as per this study. Jefferson et al found that RSV was detected in 46.3% of patients with an average hospital stay of 5.9 days with 57.5% developing severe hypoxemia.[24] In our study, higher incidence of boca virus infection (19.03%) was seen as against 7.2% in the Bharaj study.[25] Brieu et al found human bocavirus was in 10.8% of children tested.[26] Linstow et al found that BoV was present mainly during the winter months and an additional respiratory virus was identified in 47.4% of HBoV-positive samples.[27] Margaret P and Nelson et al found HBov in 5.0% of cases and the percentage was highest in the 25-36 months group (12%).[28] As per Farshad et al, there were no demographic, clinical, radiologic, or laboratory parameters except for recurrent wheeze and fever which could differentiate between patients with or without viral etiology.[16]

Improved antibiotic stewardship and avoidance of resistance development are important possible effects of molecular testing with multiplex PCR and can reduce the number of antibiotic prescriptions.[29] Sometimes, patients may be given an antibiotic even if the clinician is fairly certain that the illness is caused by a virus. To reduce such prescriptions, a combination of individual M-PCR test results with repeated feedback of aggregated results is probably required.

The study was conducted in a single centre. Limited sample size was another limitation.

## CONCLUSION

Overall, viruses were isolated in 68.9% of the sample studied. The main viruses isolated were respiratory syncytial virus in 23.5% patients, boca virus in 16.14%, influenza A in 7.5%. Advanced technologies will help us to have early virus isolation and can avoid unnecessary use of antibiotics. Studies like this will be an eye opener for clinicians to practice optimum antibiotic usage. Better understanding of clinical presentations and complications of viral pneumonias will help us to provide good care to the patients.

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