

Viral Aetiology and Epidemiology of Acute Respiratory Infections in Hospitalized Saudi Children

by T. M. F.¹Bakir,* M. Halawani,** and S. Ramia*

Departments of *Pathology and **Medicine, College of Medicine, King Saud University, Riyadh 11461, Saudi Arabia

Summary

A total of 1429 Saudi children of either sex and under 5 years of age who were admitted to King Khalid University Hospital, Riyadh during a three year period (April 1993–March 1996) with complaints suggestive of acute respiratory tract infections (ARTI) were investigated for viral aetiology of the infection. Viruses could be detected in 522 (37 per cent) cases with respiratory syncytial virus (RSV) the most commonly detected (79 per cent) followed by parainfluenza type 3 (8 per cent). Detection of influenza A, B and adenoviruses accounted for 6 per cent, 3 per cent and 2 per cent respectively. Except for parainfluenza virus type 3 the peak of activity of the respiratory viruses was during the winter months (October–February). Parainfluenza virus type 3 could be detected all year round but epidemics can occur in the hottest months of the year (June–August) when the temperature can exceed 40°C. Association between clinical manifestation, type of causative agent, and age was evaluated in 137 infected Saudi children in the first year of the study (April 1993–March 1994). The majority of our cases presented with bronchiolitis (58 per cent) while only 26 per cent had bronchopneumonia. There was a significant association between bronchiolitis and lower age groups (0–6 months), with RSV as the major causative agent of bronchiolitis cases (88 per cent).

Introduction

Acute respiratory tract infection (ARTI) is the major cause of morbidity and mortality in young children worldwide.¹ It is estimated that ARTI can cause four to five million deaths among children every year,² the overwhelming majority occurring in developing countries.^{3,4} Viral and bacterial agents are responsible for most cases of ARTI.^{5,6} Although the case–fatality rate in ARTI caused by bacteria is much higher than that caused by viruses, most cases of ARTI are of viral aetiology.⁶ Geographic location, the types of groups studied, and the laboratory techniques used were important factors in determining the prevalence of these viruses.^{7,8} Evidence of the role of viruses as causative agents of ARTI in Saudi children has been scanty,^{9,10} and hence it was the aim of this study to find the viral aetiology of ARTI in hospitalized Saudi children and to determine the distribution of the responsible agents and their seasonal pattern of activity over a long period, as this might help towards better management and treatment of infected children.

Material and Methods

The study population

The study population comprised 1429 Saudi children of either sex and under 5 years of age who were admitted to King Khalid University Hospital (KKUH), Riyadh with

complaints suggestive of ARTI. The study was conducted from April 1993 to March 1996. A child was considered as having ARTI if he or she had at least three of the following signs and symptoms: cough, fever, difficulty in breathing (manifested by nasal flaring and thoracic retraction), presence of grunting, crackles, and/or wheezing, stridor. Bronchiolitis was differentiated from bronchopneumonia by the absence of bronchial breathing. Chest X-ray was performed in all cases with signs and symptoms suggestive of ARTI. Diagnosis of bronchiolitis was considered if the chest X-ray showed mainly hyperinflation, in contrast to the infiltrates seen in cases of bronchopneumonia.

Specimen collection and laboratory methods

Nasopharyngeal aspirates (NPA) were collected by mechanical suction into 2 ml of virus transport medium (VTM). The sample was divided into two parts: one for antigen detection by an immunofluorescent antibody (IFA) technique and one for virus culture. All specimens were collected within 24 h of admission. During the first year of the study, April 1993–March 1994, both IFA and virus culture were performed but in subsequent years, only the IFA technique was used.

IFA. The sample for IFA was diluted in 10 ml of phosphate buffered saline (PBS) and centrifuged for 10 min at 1000g. The pellet was resuspended and washed three times in PBS. Finally, the pellet was resuspended in PBS, adjusted to an optimum concentration (5–8 cells/high power field), deposited on Teflon-coated slides and

Correspondence: T. M. F. Bakir, address as above.

fixed in cold acetone. Staining and identification of respiratory syncytial virus (RSV); parainfluenza viruses 1, 2, and 3; influenza A and B; and adenoviruses were carried out according to standard procedures of the direct immunofluorescent technique¹¹ using fluorescein-conjugated anti-virus antibodies (DAKO Diagnostics Ltd, Denmark).

Virus culture. Two cell lines obtained from Flow Laboratories (UK) were used for isolation: Hep-2 (a continuous epithelial cell line derived from a human carcinoma of the larynx) and MDCK (a continuous epithelial cell line derived from dog kidney). MDCK was used because of its susceptibility to influenza viruses, especially in the presence of trypsin.¹¹ Virus isolation was carried out according to a previously described method.¹² Cultures were checked for up to 2 weeks for cytopathic effect (CPE). Identification of isolates was confirmed by IFA using cell scrapings from the suspected culture tubes. Early detection of influenza A and B was also attempted blindly in MDCK cells using the shell vial technique.¹³ A specimen was considered positive if a virus was detected by IFA alone or in combination with virus culture.

Statistical analysis

The significance of the association between clinical diagnosis and age among the 137 Saudi children was investigated using the chi-square (χ^2) test. The linear trends of the prevalence of each clinical entity with age were also investigated using the χ^2 test for trends in proportions.

Results

The types of viruses identified in the first year of the study (April 1993–March 1994) and their seasonal pattern is shown in Table 1. RSV was the most commonly detected virus (74 per cent) followed by parainfluenza type 3 (12 per cent). RSV was highly prevalent during the cold months of the year (November–March) with a January–February peak, in contrast to parainfluenza virus type 3 which could be detected all year round. Influenza A and influenza B viruses accounted for 6 per cent and 4 per cent, respectively, of all viruses isolated and cases were also confined to the cold months of the year.

Viruses identified in 1429 Saudi children with ARTI during the three year period (April 1993–March 1996)

TABLE 1
Viruses identified and their seasonal distribution in 137 Saudi children with acute respiratory tract infection during a one year period (April 1993–March 1994)

	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec	Jan	Feb	March	Total (%)
No. of children	22	5	5	20	18	9	26	38	58	74	78	50	403
No. positive (%)	3	1	–	3	–	1	6	9	17	39	37	21	137 (34.0)
RSV	–	–	–	–	–	–	–	3	9	36	34	19	101 (73.7)
Parainfluenza 1	–	–	–	1	–	–	2	–	–	–	–	–	3 (2.2)
Parainfluenza 2	–	–	–	–	–	–	–	–	–	–	–	–	–
Parainfluenza 3	3	1	–	1	–	1	3	1	1	1	3	1	16 (11.6)
Influenza A	–	–	–	1	–	–	–	2	4	1	–	–	8 (5.8)
Influenza B	–	–	–	–	–	–	1	3	2	–	–	–	6 (4.4)
Adenovirus	–	–	–	–	–	–	–	–	1	1	–	1	3 (2.2)
Total no of viruses detected													137

TABLE 2
Viruses identified by IFA in acute respiratory tract infection in 1429 Saudi children in Riyadh from April 1993 to March 1996

	April 1993–March 1994	1994–1995	1995–1996	Total (%)
No of children	403	357	669	1429
No positive (%)	137 (34.0)	127 (35.6)	258 (38.6)	522 (36.5)
RSV	101 (73.7)	102 (80.3)	210 (81.4)	413 (79.1)
Parainfluenza 1	3 (2.2)	0	4 (1.6)	7 (1.3)
Parainfluenza 2	0	0	6 (2.3)	6 (1.2)
Parainfluenza 3	16 (11.6)	7 (5.5)	16 (6.2)	39 (7.5)
Influenza A	8 (5.8)	15 (11.8)	9 (3.5)	32 (6.1)
Influenza B	6 (4.4)	1 (0.8)	6 (2.3)	13 (2.5)
Adenovirus	3 (2.2)	2 (1.6)	7 (2.7)	12 (2.3)

TABLE 3

Distribution of clinical cases in 137 Saudi children with ARTI (April 1993–March 1994) with respect to various age groups

Clinical diagnosis	No of cases	Age (months)				Significance
		0–6 No (%)	>6–12 No (%)	>12–24 No (%)	>24–60 No (%)	
Bronchiolitis	80 ^a	63 (78.6)	10 (12.5)	5 (6.3)	2 (2.5)	$\chi^2 = 39.1; p < 0.00001$
Bronchopneumonia	35 ^b	8 (22.9)	12 (34.3)	7 (20.0)	8 (22.8)	$\chi^2 = 6.99; p < 0.01$
Other respiratory infections (bronchial asthma, croup, upper respiratory infections)	22	3 (13.6)	3 (13.6)	6 (27.3)	10 (45.4)	$\chi^2 = 27.6; p < 0.00001$
Total	137	74 (54.0)	25 (18.2)	18 (13.1)	20 (14.6)	

^a 70 out of 80 were due to RSV (87.5%).

^b 22 out of 35 were due to RSV (63.0%).

are shown in Table 2. RSV was the most commonly identified agent (79 per cent) followed by parainfluenza 3 (8 per cent) and influenza A (6 per cent). The frequency of isolation of the different viruses and their seasonal pattern was similar every year. During June to August of 1996, out of 26 specimens from Saudi children with ARTI submitted to our laboratory for virus detection, 20 of them were identified as positive for parainfluenza virus type 3 by IFA.

Association between clinical symptoms, age, and causative agent was done on infected children in the first year of the study (April 1993–March 1994). Distribution of 137 clinical cases with respect to the various age groups is shown in Table 3. Statistical analysis showed that clinical diagnosis was highly associated with age ($\chi^2 = 60.12, p < 0.0001$). There was a significant association between bronchiolitis and lower age groups, 0–6 months ($p < 0.00001, \chi^2 = 39.1$). Bronchopneumonia was diagnosed more often after the age of 6 months ($p < 0.01, \chi^2 = 6.99$). RSV was the major causative agent of bronchiolitis (88 per cent) in the various age groups and of bronchopneumonia in infants less than 1 year old (63 per cent) (Table 3). Parainfluenza type 3 was the major causative agent of bronchopneumonia in children over 6 months of age while influenza A and B accounted for most of the cases with upper respiratory tract infections.

Discussion

In the present study, recognized viral pathogens (RSV, parainfluenza, influenza, and adenoviruses) were identified in 522 (37 per cent) of ARTI in Saudi children. Routine bacterial cultures were not performed on respiratory secretions because the yield of such cultures has been low in our previous experience. Furthermore, bacterial blood cultures were negative and there was no convincing improvement with antibiotic treatment in severe cases, suggesting that superimposed bacterial infections were rare.

RSV infection occurs in yearly epidemics and can

cause serious bronchiolitis.¹⁴ In our study RSV was the most frequently demonstrated virus of established pathogenic significance in this study and it accounted for 79.1 per cent of all viruses isolated. Our results are similar to those from other countries^{15–17} and emphasize the role of RSV as the major cause of ARTI in infants and young children. Also, our finding of RSV in a much higher percentage of cases diagnosed as bronchiolitis (especially in infants under 6 months of age) compared to bronchopneumonia is similar to results from other countries on hospitalized children with these two clinical entities.^{17,18}

Parainfluenza virus type 3 followed RSV as the next most frequently detected viral agent in our patients (8 per cent) which occurred throughout the year and accounted for the majority of parainfluenza viruses detected. Although parainfluenza virus types 1 and 2 occur in epidemics^{19,20} and can be a major cause of respiratory illness in young children,²¹ only seven cases of parainfluenza virus type 1 and six cases of parainfluenza virus type 2 were detected in our study. Out of 39 parainfluenza virus type 3 cases, 28 (72 per cent) were in children older than one year of age.

Influenza viruses have a worldwide distribution and occur in an epidemic form.²² The peak of incidence of influenza A in our study was in the period 1994–1995. While these viruses are isolated more frequently from older children and adults, studies have shown that these viruses can produce ARTI in children less than 5 years of age.²³ Out of the 32 influenza A cases in our study only five were in children over three years of age, while 10 out of 13 influenza B cases were in children over three years of age.

Adenoviruses have been shown to occur in epidemics and can cause ARTI. Only 12 cases of adenovirus infection were detected in our study. Based on the number of adenovirus cases it is not clear whether this low incidence was due to annual variation or because adenoviruses play a relatively minor role in the causation of ARTI in Saudi children.²⁴ Longer term studies and typing of viruses are needed to explain the

importance of adenoviruses and the other viruses as causative agents of ARTI in our pediatric population. We did not look for rhinoviruses or coronavirus in our study although both viruses can cause severe lower respiratory tract illness in some individuals.²⁵

During the first year of this study both IFT and tissue culture were used as diagnostic tests. The study showed a very close correlation between the two tests which is in agreement with others,²⁶ therefore only IFT was used in subsequent years.

The result of this investigation showed that the recognized viral pathogens, RSV, parainfluenza, influenza, and adenoviruses, account for 37 per cent of hospitalized Saudi children with ARTI, with RSV being the major aetiological agent followed by parainfluenza virus type 3. It must be mentioned, however, that studies on hospitalized children represent only a partial view of the impact of respiratory viruses in the community and hence wider studies covering other parts of the Kingdom are needed.

In the children in whom a virus could be demonstrated, the highest incidence of viral infections occurred during the winter months (October–February) except parainfluenza virus type 3 where the virus could be detected all the year round. It was surprising that epidemics due to parainfluenza 3 can occur in the hottest months of the year in Saudi Arabia where the temperature can exceed 40°C. The finding that most respiratory infections peak during the winter months is similar to that reported from tropical and subtropical countries.^{9–13} The predominance of infants younger than 6 months old in our study could possibly be due to the natural susceptibility of those young infants to primary infections leading to serious disease.

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