

HAEMOPHILUS INFLUENZAE ASSOCIATED WITH ACUTE RESPIRATORY ILLNESSES IN YOUNG CHILDREN

ML THONG MBBS, DIPBACT* AND SB ONG MSc**

Summary

A total of 303 young children with acute respiratory tract illnesses were examined for viral and bacterial agents. The isolation of *Haemophilus influenzae* from 58 (19.1%) of the children was described in relation to respiratory viruses and other bacterial agents isolated at the same time. *H. influenzae* was isolated alone in 24 instances. *Streptococcus pneumoniae* and respiratory syncytial virus were the most common bacterial and viral agents associated with *H. influenzae*. Only 7.5% of the 40 laboratory confirmed cases of influenza yielded *H. influenzae* in their respiratory secretions.

INTRODUCTION

Haemophilus influenzae is one of the important bacterial species associated with acute infection of the respiratory tract.¹ Several investigators have found a high incidence of *H. influenzae* predominating in throat flora coinciding with symptoms of acute respiratory tract disease.²⁻⁵ A possible causal role of *H. influenzae* in these infections was suggested by Dick *et al*³ and Bridger⁴ when they observed the disappearance of this organism coinciding with the recovery of the patients after antibiotic therapy.

Several studies have shown that infections by *H. influenzae* and other bacterial pathogens combined with or superimposed on viral infections are not infrequently found in children with acute respiratory illnesses.⁵⁻⁸ Such mixed infections may cause more severe illness than that of the primary viral disease.¹

This report describes the isolation of *H. influenzae* and other bacterial species from children with acute respiratory tract infections together with isolation of respiratory viruses.

MATERIALS AND METHODS

From March 1974 to November 1977, respiratory secretions from 303 young children with acute respiratory tract infections were cultured for viral and bacterial agents. The children were included in the study if they had symptoms and/or signs of an acute respiratory tract infection and were cooperative during specimen collection. The ages of these children ranged from one month to 10 years of age. Many of the children were less than 2 years old.

Throat and/or nasopharyngeal swabs or nasopharyngeal mucus were collected from each child for cultural examination. The nasopharyngeal mucus was collected in sterile plastic disposable mucus extractors.

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Isolation of Bacteria

Specimens were cultured within two hours of collection onto blood agar and chocolate agar plates, prepared with 10% defibrinated ox blood in blood agar base (Oxoid) as well as charcoal blood agar (Difco) containing 0.25 units of penicillin. The blood agar and chocolate agar plates were incubated at 36°C in a candle jar for one to two days and examined for the presence of *H. influenzae* and other likely pathogens, such as *Streptococcus pneumoniae*, B-haemolytic streptococci and *Staphylococcus aureus*. The charcoal blood agar plates were incubated aerobically at 36°C and examined after 2, 4 and 7 days for colonies of *Bordetella pertussis*. The bacterial pathogens were identified as follows: *H. influenzae* by its requirement for X and V factors, *Strep. pneumoniae* by its sensitivity to optochin, *Staph. aureus* by a positive coagulase test and *B. pertussis* by slide agglutination with specific antiserum.

Isolation of Viruses

Respiratory virus isolations were carried out in parallel with bacterial isolations using separate throat and/or nasopharyngeal swabs but with the same sample of nasopharyngeal mucus. Specimens were sent to the virology laboratory as soon as possible usually on wet ice. In the laboratory the specimens were inoculated into

*Lecturer, Department of Medical Microbiology, Faculty of Medicine, University of Malaya, Kuala Lumpur, Malaysia (Address for reprint requests).

**Lecturer, Department of Medical Microbiology, Faculty of Medicine, University of Malaya, Kuala Lumpur.

appropriate tissue cultures or frozen at -70°C until tissue cultures were available. The methods used for isolation and identification of respiratory viruses have been described previously.⁹

No attempt was made to isolate coronavirus and cytomegalovirus as well as *Mycoplasma pneumoniae* and other fastidious bacteria.

RESULTS

Viral and/or bacterial pathogens were recovered from 165 (54.4%) of the children. Viral agents only were recovered from 79 children (26.1%), bacterial agents only from 61 children (20.1%) and both viral and bacterial agents from 25 (8.2%) of the children.

H. influenzae was isolated from 58 children, an isolation rate of 19.1%. Six of the isolates (10.3%) were capsulated; three were type a, two type b and one type f. *H. influenzae* was the sole likely pathogen isolated from 24 children. In the other 34 children, *H. influenzae* was isolated simultaneously with other bacterial agents in 19 cases, with viral agents in 7 cases and with both viral and other bacterial agents in 8 cases, as shown in Table 1.

Strep. pneumoniae was isolated with *H. influenzae* from 24 cases, including 7 cases of laboratory confirmed virus infections. Respiratory syncytial virus was recovered together with *H. influenzae* from 6 cases including 2 cases with rhinovirus isolates. There was also one

TABLE 1
HAEMOPHILUS INFLUENZAE ISOLATED FROM CHILDREN WITH ACUTE RESPIRATORY ILLNESSES

	Number of Children
<i>H. influenzae</i> isolated as sole agent:	24
<i>H. influenzae</i> isolated with other bacterial agents:	19
<i>Streptococcus pneumoniae</i>	17
<i>Staphylococcus aureus</i>	2
<i>H. influenzae</i> isolated with viral agents:	7
Respiratory syncytial virus (RSV)	3
Parainfluenza virus	2
Rhinovirus	1
RSV and Rhinovirus	1
<i>H. influenzae</i> isolated with other bacterial agents:	8
RSV and <i>Strep. pneumoniae</i>	1
Influenza A or B virus and <i>Strep. pneumoniae</i>	3
RSV, Rhinovirus and <i>Strep. pneumoniae</i>	1
Parainfluenza virus and <i>Strep. pneumoniae</i>	1
Parainfluenza virus and <i>bordetella pertussis</i>	1
Adenovirus and <i>Strep. pneumoniae</i>	1
Total	58

other case of infection with rhinovirus and 4 cases with parainfluenza viruses. In one of these cases, parainfluenza type 2 virus was isolated together with *Bordetella pertussis* and *H. influenzae* in a 11-month old child with bronchitis.

Among the respirator viruses isolated from 104 children in this study, there were 25 influenza type A and 15 influenza type B viruses. *H. influenzae* was associated with only three of these viruses (one type A and two type B viruses).

DISCUSSION

It has been noted in previous studies that when *H. influenzae* was isolated from acute respiratory tract infections, it was often found in association with at least one other potential pathogen, either bacterial, mycoplasmal or viral agents.⁸ In the present study, over half the *H. influenzae* isolates were associated with viral and other bacterial agents. It is possible that in the remaining cases where *H. influenzae* was recovered alone, other agents of respiratory infections like *Mycoplasma pneumoniae*, coronavirus and cytomegalovirus, could have been present but no isolations were done for these agents.

Previous investigators have also noted that *H. influenzae* and *Strep. pneumoniae* were frequently isolated together from respiratory secretions in patients with viral infections.^{6,8} The biologic significance of this association is not clear. There is no clear evidence of synergism in the production of disease.⁸ However, the severity of respiratory illness may be related to infection by multiple agents.⁷ With respiratory viral infections there may be a change in the growth of pathogenic bacteria and in some cases this change may contribute to prolonged and more severe disease.

In the present study *H. influenzae* was associated with only 7.5% of the 40 laboratory confirmed cases of influenza. This finding is in keeping with the observation that *H. influenzae* did not play a significant role in recent outbreaks of influenza.¹

The isolation rate of 19.1% of *H. influenzae* from these 303 symptomatic children is higher than the isolation rate of 13 to 15% of *H. influenzae* in the pharynx of asymptomatic children in the Kuala Lumpur and Petaling Jaya area.² Since both capsulated and non-

capsulated strains of *H. influenzae* was not isolated substantially more frequently in the children with respiratory illnesses, their isolates of *H. influenzae* may merely represent normal throat flora. However, many of the sick children with heavy growth of *H. influenzae* improved clinically after antibiotics were given to eradicate the organisms.

It was unfortunate that paired serum specimens could not be obtained from these children with acute respiratory illnesses to demonstrate any change in titre of antibodies to *H. influenzae*. Sell *et al*^{1,3} were able to demonstrate rising titres of specific *H. influenzae* antibodies in many young children with acute respiratory infections who have profuse growth of *H. influenzae* in their nasopharyngeal cultures.

ACKNOWLEDGEMENTS

We are grateful for the cooperation of the medical and nursing staff of the Department of Paediatrics, University Hospital, in the collection of clinical specimens.

This study was supported in part by a grant (No. 73-306) from the China Medical Board of New York.

REFERENCES

1. Alexander HE. The *Hemophilus* group. In: Dubos RJ, Hirsch JG, eds. Bacteria and mycotic infections of man. Philadelphia: JB Lippincott Company, 1965: 724-41.
2. Masters PL, Brumfitt W, Mendez RL, Likar M. Bacterial flora of the upper respiratory tract in Paddington families, 1952-4. Br Med J 1958;1: 1200-5.
3. Dick EC, Carr DL. *Haemophilus influenzae*. Association with acute respiratory illness in adults. Arch Environ Health 1966;13: 450-3.
4. Bridger RC. *Haemophilus influenzae*: the relationship to upper respiratory tract infection. NZ Med J 1974;80: 19-22.
5. Horn MEC, Yealland SJ. Significance of respiratory virus isolations. A study in primary schoolchildren. Arch Dis Child 1974;49: 516-9.
6. Cherry JD, Diddams JA, Dick EC. Rhinovirus infections in hospitalized children. Provocative bacterial interrelationships. Arch Environ Health 1967; 14: 390-6.

7. Nichol KP, Cherry JD. Bacterial-viral interrelations in respiratory infections of children. *N Engl J Med* 1967; 277: 667–72.
8. Lepow ML, Balassanian N, Emmerich J, Roberts RB, Rosenthal MS, Wolinsky E. Interrelationships of viral, mycoplasmal, and bacterial agents in uncomplicated pneumonia. *Am Rev Respir Dis* 1968; 97: 533–45.
9. Ong SB, Thong ML, Tay LK. Viruses and bacteria associated with acute respiratory illnesses in young children in general practice. *Southeast Asian J Trop Med Public Health* 1978; 9: 98–102.
10. Giles C, Shuttleworth EM. Post-mortem findings in 46 influenza deaths. *Lancet* 1957; 2: 1224–5.
11. Martin CM, Kunin CM, Gottlieb LS, Barnes MW, Liu C, Finland M. Asian influenza A in Boston, 1957–1958. I. Observations in thirty-two influenza-associated fatal cases. *Arch Intern Med* 1959; 103: 515–31.
12. Thong ML. *Haemophilus influenzae* infections in Malaysia. MD thesis, University of Malaya, Kuala Lumpur, 1979.
13. Sell SHW, Sanders RS, Cheatham WJ. *Haemophilus influenzae* in respiratory infections. II. Specific serological antibodies identified by agglutination and immunofluorescent techniques. *Am J Dis Child* 1963; 105: 470–4.