Meningitis due to ampicillin- and chloramphenicol-resistant *Haemophilus influenzae* type b in Canada. Case report and review

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ABSTRACT: The first report of a case of ampicillin- and chloramphenicol-resistant *Haemophilus influenzae* type b invasive infection in Canada is described in a four-month-old male with meningitis. He was treated with cefotaxime 200 mg/kg/day divided every 6 h and dexamethasone 0.6 mg/kg/day divided every 6 h, eventually recovering after a complicated course. Follow-up at 21 months showed mild to moderate global developmental delay. While chloramphenicol resistance is rare

in North America, a case of meningitis initially unresponsive to ampicillin and chloramphenicol must be considered suspect for resistance. Third generation cephalosporins should be used for resistant cases. **Can J Infect Dis 1990;1(3):92-96**

Key Words: Ampicillin, Chloramphenicol-resistant, *Haemophilus influenzae*, Meningitis

HAEMOPHILUS INFLUENZAE TYPE B (HIB) IS A SIGNIFICANT pathogen causing invasive disease in children worldwide (1). It is the leading cause of bacterial meningitis in children, both in Canada and the United States (1,2). Although Hib infections can occur in all age groups, invasive disease is primarily a disease of children below the age of five years (1,3-5). It is estimated that one in 200 children have some form of invasive disease due

to Hib by five years of age (6,7). The incidence of ampicillin-resistant Hib has been increasing since 1974, both in North America and worldwide (8-11). Chloramphenicol resistance has been reported in Spain (12,13), Europe (14,15), rarely in the United States (9,11,16) and Canada (10,17,18), and elsewhere (19,20). Ampicillin- and chloramphenicol-resistant invasive Hib disease in Canada has never been reported (10). The authors report the first case of ampicillin- and chloramphenicol-resistant Hib disease in Canada in a four-month-old male infant with meningitis.

CASE PRESENTATION

A previously well four-month-old Oriental male born in Canada was admitted to hospital in June 1988 with a five day history of fever and irritability. He was seen by his family physician and

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TABLE 1
Antibiotic susceptibility of an *Haemophilus influenzae* type b isolate

Antimicrobial agent	MIC ($\mu\text{g/mL}$)	Kirby Bauer zone sizes (mm)
Ampicillin	>1.0	0
Chloramphenicol	>8.0	11
Tetracycline	>4.0	No data
Cefotaxime	≤ 0.12	30
Rifampin	≤ 1.0	No data
Cotrimoxazole	No data	23
Cefuroxime	No data	30
Ceftriaxone	No data	36

MIC Minimal inhibitory concentration

commenced on oral cefaclor 125 mg tid. His fever and irritability continued and three days later he saw a pediatrician who continued with the same management. On the day of admission, he had a 10 min tonic clonic generalized seizure at home followed by another in the emergency room. He had no history of cough, rhinorrhea, diarrhea or vomiting. There was no history of travel outside of Canada, or contact with any visitors from outside Canada.

Examination at that point revealed a temperature of 38.5°C (axilla), pulse of 150 beats/min, respiratory rate of 30/min and blood pressure of 90/50 mmHg. He was lethargic, irritable, pale and looked toxic. He had shallow respirations and poor peripheral circulation with a capillary refill of 4 s. His head circumference was 42 cm (80th percentile) and his anterior fontanelle was firm and full. He also demonstrated nuchal rigidity and positive Kernig's and Brudzinski's signs. The tympanic membranes were normal, as was the rest of the physical examination.

His lumbar puncture revealed cloudy cerebrospinal fluid with a leukocyte count of $1106 \times 10^6/\text{L}$ (50% neutrophils, 14% lymphocytes, 36% monocytes); red blood cells $9 \times 10^6/\text{L}$; protein 0.55 g/L; and glucose 0.6 mmol/L (serum glucose 4.7 mmol/L). Gram stain showed many Gram-negative coccobacilli resembling *H influenzae* and the cerebrospinal fluid latex agglutination (Wellcogen *H influenzae* b; Wellcome Foundation Ltd, Dartford, United Kingdom) was positive for *H influenzae* type b antigen. The peripheral white blood cell count was $17.1 \times 10^9/\text{L}$; hematocrit 0.300; and platelets $352 \times 10^9/\text{L}$. Treatment with cefotaxime 200 mg/kg/day divided every 6 h, and dexamethasone 0.6 mg/kg/day divided every 6 h was started.

The patient's subsequent course was prolonged and complicated, including persistent fever, focal seizures, bilateral subdural empyemas requiring surgical evacuation, and progressive communi-

cating hydrocephalus treated with ventriculoperitoneal shunt on day 23. Visual evoked potentials prior to discharge were abnormal. Auditory brainstem evoked responses were normal. Follow-up examination at 21 months showed mild to moderate global developmental delay with normal hearing and vision and normal auditory brainstem evoked responses.

MICROBIOLOGICAL DATA

Hib was isolated from both the blood and cerebrospinal fluid. The blood cultures were collected in NR6a and NR7a vials (Becton Dickinson, Cockeysville, Maryland). These vials showed a positive growth value of greater than 20 in under 20 h. Growth of the organism from both the blood and cerebrospinal fluid provided identical biochemical and serological results. The beta-lactamase produced was detected using a Cefinase disk (BBL, Maryland). Production of chloramphenicol acetyltransferase was detected using a reagent impregnated disk test (Remel, Kansas) (21). Appropriate controls were done. Both the agar dilution and modified Kirby Bauer susceptibility methods were performed according to NCCLS M7-a and M2-A3 (22). The results are indicated in Table 1.

DISCUSSION

In Canada, invasive Hib infection became a reportable disease in 1986, while Hib meningitis has been reportable since 1979 (5). In 1986, total invasive Hib cases reported were 2.1 per 100,000 (5). The annual incidence of Hib meningitis has ranged from 0.94 to 1.65 per 100,000 from 1979 to 1985 (5). Most of these cases occur in children below the age of five years (86%) with the highest incidence in infants – 52 per 100,000, and the lowest incidence in 20 to 24 year olds. In 1986, in Canada, 14 deaths from Hib meningitis were reported – 29% were infants and 57% were one to four years old – giving a case fatality rate of 2.6% (5). Similarly, in the United States, *H influenzae* is the single most common cause of bacterial meningitis, with 8000 to 10,000 cases each year (1.24 per 100,000) and 400 to 500 deaths each year (23). The peak attack rate occurs at seven months of age, with 80% less than two years old, and nearly two-thirds less than 12 months of age (23). Thus, Hib disease in children is not rare and has a significant mortality, demanding adequate therapy.

Prior to 1974, *H influenzae* was considered uniformly sensitive to ampicillin. Since the first case reports of ampicillin resistance in 1974 (24-26), the incidence of resistance has been steadily increasing. In the United States, prior to 1978,

most laboratories reported less than 5% isolates producing beta-lactamase (11,27). Beta-lactamase positive isolates from blood and cerebrospinal fluid in children from Houston, Texas increased from 12% in 1977 to 29% in 1979 (28). In Colorado, cerebrospinal fluid isolates producing beta-lactamase increased from 4% in 1977 to 31% in 1981 (29). In the most recent American survey in 1986, the overall beta-lactamase production in both typeable and nontypeable *H influenzae* combined was 20%, and in Hib strains 32% (9). In Canada, in 1986, the beta-lactamase production rate was 18% in the 1232 strains from 10 hospitals across Canada (30). In 1987, in Ontario, from a total of 1139 nonhospital isolates, beta-lactamase positive strains were found in 24% (10). Recently a Toronto study found 20% beta-lactamase-producing strains from a private laboratory setting (31). Although non-beta-lactamase-mediated ampicillin resistance has been reported, it is rare in the United States and Canada, and its actual prevalence is unknown (9,11,32,33).

Although chloramphenicol-resistant *H influenzae* was first reported in 1972 in Houston, Texas (34), and sporadic case reports followed, the rate of increase in resistance has not paralleled that of ampicillin; chloramphenicol resistance remains rare in North America (10,11,16,17). In some parts of Europe, chloramphenicol resistance has become an increasing problem (12). In Switzerland, chloramphenicol resistance has increased from 0.6% in 1983 to 2.0% in 1986 (14,35); in the United Kingdom, from 0.2% in 1977 to 1.7% in 1986 (14,15); and even more strikingly in Spain from 18% in 1983 to 25% in 1986 (12,13). The European Cooperative Study showed, in 1961 isolates, 6% resistance in Hib (19% of isolates) and 5% in nontypeable *H influenzae* (81% of isolates) (14). The recent data from Spain are most alarming because in invasive disease caused by Hib, resistance was even more prevalent; chloramphenicol resistance in cerebrospinal fluid isolates was 66%, while ampicillin and chloramphenicol resistance was 57%. In addition, a case of epiglottitis and three cases of pneumonia were reported that were ampicillin- and chloramphenicol-resistant (12,13).

In the United States, chloramphenicol resistance has not increased based on two national surveillance studies by Doern *et al* (8,9), who reported incidences in 1984 of 0.6% and in 1986 of 0.5% (70% of these were also beta-lactamase producers). The only six cases of invasive ampicillin- and chloramphenicol-resistant *H influenzae* isolates in the United States had meningitis, and these were recently reviewed by Givner *et al* (16).

In Canada, the first report of chloramphenicol-resistant *H influenzae* occurred in 1978 from a sputum sample in an asymptomatic nine-year-old boy (18). Later, a national survey of 921 isolates of *H influenzae* in Canada from 1979 to 1981 found four additional isolates resistant to chloramphenicol: three from throat cultures and one from a bacteremic child (17). Two further surveys in eastern Canada up to 1985 did not report any further chloramphenicol-resistant strains (10,36). The most recently reported survey from Ontario in 1987 found two of 252 isolates resistant to ampicillin and chloramphenicol; both nontypeable, chloramphenicol acetyltransferase positive, beta-lactamase positive sputum isolates (10). A report published in 1988 from Toronto found none of 250 isolates to be chloramphenicol-resistant (31). Thus in Canada and the United States, chloramphenicol resistance continues to be rare. In Canada, no systemic isolate of Hib had been resistant to both chloramphenicol and ampicillin (10,17).

The mechanism of resistance to ampicillin is usually by plasmid-mediated beta-lactamase production (11). Chloramphenicol resistance is, in greater than 90% of cases, also plasmid-mediated via the enzyme chloramphenicol acetyltransferase, which catalyzes diacetylation of chloramphenicol to acetyl coenzyme A (11,37,38). Although chloramphenicol acetyltransferase is the predominant mechanism for chloramphenicol resistance, a permeability barrier mechanism has also been described (39). That not all chloramphenicol resistance is chloramphenicol acetyltransferase-mediated emphasizes the need for confirmatory susceptibility testing by recent NCCLS guidelines, which are supported by several recent studies (40-44).

Most chloramphenicol-resistant strains are also multiply resistant, almost always to tetracycline, and often to ampicillin, kanamycin, streptomycin and sulfamethoxazole (although these resistance patterns do not arise from acquisition of a single R plasmid) (11,45). In Spain, the most frequent pattern for multiple resistance (including ampicillin and chloramphenicol) was ampicillin + chloramphenicol + tetracycline + sulpha + trimethoprim (94.8%) followed by ampicillin + chloramphenicol + tetracycline (3.9%) (12,13). There has not been a report of *H influenzae* resistance to third generation cephalosporins (11,12,13,36).

CONCLUSION

Reported here is a case of ampicillin and chloramphenicol multiply resistant *H influenzae* type b invasive infection in Canada. This occurred in a four-month-old boy with meningitis who had a

very complicated course. This is the first such report in Canada.

Therapy for meningitis with ampicillin and chloramphenicol is considered appropriate in view of the rarity of chloramphenicol resistance in North America at the present time. However, in a case initially unresponsive to this regimen, resistance must be strongly considered, and third generation cephalosporins used for resistant cases. In fact, the American Academy of Pediatrics and the Canadian Pediatric Society recommend third generation cephalosporins as equally acceptable as ampicillin and chloramphenicol for initial therapy of meningitis (46,47). There is a concern

that incidence of resistance could increase in North America, as it has in areas of Europe, especially since this case was, as in the European isolates, chloramphenicol acetyltransferase-mediated and multiply resistant. There is also widespread antibiotic use in North America which is known to increase the incidence of ampicillin resistance (48); however, this has not yet been shown to increase the incidence of chloramphenicol resistance (1,2,13). Finally, the importance of routine chloramphenicol susceptibility testing in all invasive isolates should be stressed, and all chloramphenicol-resistant strains reported in order to allow surveillance for possible emerging resistance patterns.

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