Ampicillin-Resistant *Haemophilus influenzae* in Johannesburg

C. S. BLOCK, LORRAINE ARNTZEN, LYNN GARDNER

SUMMARY

Surveys carried out using a chromogenic cephalosporin test for β-lactamase production (ampicillin resistance) among isolates of *Haemophilus influenzae* in Johannesburg have indicated an appreciable prevalence, especially among children seen at the new Johannesburg Hospital. Of type b strains recovered from these children, 10.9% were ampicillin-resistant. Three of the last 10 cases of serious systemic *H. influenzae* infections encountered at the Johannesburg Hospital were caused by β-lactamase-producing strains, all of these having been acquired in the community rather than in hospital. These findings suggest that the optimal initial starting treatment for serious systemic or life-threatening *H. influenzae* infections should include chloramphenicol, either alone or at least in combination with ampicillin or a similar compound.


Ampicillin-resistant *Haemophilus influenzae* has become a global problem in recent years,1,2 posing serious chemotherapeutic problems, especially in infections of infancy and childhood. To our knowledge, the first ampicillin-insensitive isolate found in Johannesburg was recovered from the cerebrospinal fluid of a Coloured child in 1976. This report presents the results of surveys of β-lactamase-producing strains of *H. influenzae* carried out at the South African Institute for Medical Research (SAIMR) during 1978 and 1979. These studies were undertaken after the development of a simple and economical method for demonstrating β-lactamase using a chromogenic cephalosporin.

MATERIALS AND METHODS

Organisms

All isolates of *H. influenzae* recovered from clinical specimens at the central laboratories of the SAIMR were studied for a 9-month period from March through November 1978. Care was taken to exclude replicate isolates from single patients. From August 1978, a similar study was carried out on organisms isolated from paediatric patients at the Johannesburg Hospital branch of the SAIMR, which had just opened. Organisms were identified by routine laboratory methods3 and tested for β-lactamase production. Depending on the availability of antisera, as many strains as possible were serotyped by a slide agglutination method (Wellcome Laboratories). Since completion of the survey β-lactamase production has routinely been sought in all blood and cerebrospinal fluid isolates of *H. influenzae*.

Test for β-Lactamase

Lyophilized nitrocefin (Glaxo) was reconstituted according to the manufacturer's recommendations and kept in the dark at 4°C. Small amounts of the solution were drawn up into haematocrit tubes which were then stabbed into the surface of agar plates through pure cultures of *H. influenzae*. The tubes were then inverted on modelling clay stands and read at 5 minutes and 30 minutes. A colour change from yellow to pink or red in the nitrocefin indicated the presence of β-lactamase (Fig. 1). Nearly all tests were positive within 5 minutes. Positive control organisms used were β-lactamase-producing strains of *H. influenzae*, *Staphylococcus aureus*, *Bacillus cereus* and *Escherichia coli*. Ampicillin-sensitive controls used were *H. influenzae* and *Esch. coli*. Tube dilution susceptibility tests were carried out on a few initial β-lactamase-producing isolates of *H. influenzae* to confirm their

![Fig. 1. Schematic representation of the test for β-lactamase production.](image-url)
resistance to ampicillin. These tests were carried out in Mueller-Hinton broth using inocula of approximately 100 000 organisms. All isolates tested had minimum inhibitory concentrations of ampicillin of 8 mg/l or more.

**RESULTS**

Table I indicates the proportion of ampicillin-resistant *H. influenzae* according to serotype. The majority of *β*-lactamase producers were of type b, the serotype responsible for most *H. influenzae* infections. It is clear that resistant organisms were more prevalent among isolates from the paediatric populations seen at the new Johannesburg Hospital than among those from the more mixed population studied at the central SAIMR. The age distributions of these groups are given in Table II. The much higher proportion of *β*-lactamase producers recovered from 1-10-year-old children at the Johannesburg Hospital is unexplained by this study. It is noteworthy that no ampicillin-resistant strains were isolated from any of the 45 children less than 1 year old.

The anatomical origins of the organisms are displayed in Table III and their distribution among inpatients and outpatients in Table IV. Examination of all strains of *H. influenzae* isolated from patients with serious systemic infections at the new Johannesburg Hospital during the 17 months from August 1978 to December 1979 yielded the disturbing results set out in Table V. All these organisms proved to be type b.
TABLE III. AMPICILLIN-RESISTANT *H. INFLUENZAE* BY SPECIMEN TYPE

<table>
<thead>
<tr>
<th>Study location</th>
<th>Specimen</th>
<th>b</th>
<th>Non-b</th>
<th>Untypable*</th>
<th>Not typed</th>
<th>All</th>
<th>Total</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>SAIMR (central)</td>
<td>Sputum</td>
<td>13</td>
<td>1</td>
<td>5</td>
<td>6</td>
<td>25</td>
<td>805</td>
<td>3.1</td>
</tr>
<tr>
<td></td>
<td>Throat swab</td>
<td>4</td>
<td></td>
<td></td>
<td></td>
<td>4</td>
<td>362</td>
<td>1.1</td>
</tr>
<tr>
<td></td>
<td>CSF</td>
<td>1</td>
<td></td>
<td>1</td>
<td>1</td>
<td>12</td>
<td>51</td>
<td>8.3</td>
</tr>
<tr>
<td></td>
<td>Blood culture</td>
<td>3</td>
<td></td>
<td>1</td>
<td>4</td>
<td>8</td>
<td>65</td>
<td>6.2</td>
</tr>
<tr>
<td></td>
<td>Other</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>20</td>
<td>5</td>
<td>8</td>
<td>34</td>
<td>1252</td>
<td>4.7%</td>
<td></td>
</tr>
</tbody>
</table>

- Auto-agglutination in normal saline.
- Same patient.
- Organisms isolated from both blood and CSF in 2 patients.

TABLE IV. AMPICILLIN-RESISTANT *H. INFLUENZAE* AMONG INPATIENTS AND OUTPATIENTS

<table>
<thead>
<tr>
<th>Study location</th>
<th>Location</th>
<th>b</th>
<th>Non-b</th>
<th>Untypable*</th>
<th>Not typed</th>
<th>All</th>
<th>Total</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>SAIMR (central)</td>
<td>Inpatients</td>
<td>13</td>
<td>1</td>
<td>5</td>
<td>6</td>
<td>25</td>
<td>701</td>
<td>3.6</td>
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<tr>
<td></td>
<td>Outpatients</td>
<td>7</td>
<td>1</td>
<td>2</td>
<td></td>
<td>9</td>
<td>418</td>
<td>2.2</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>20</td>
<td>1</td>
<td>5</td>
<td>8</td>
<td>34</td>
<td>1119</td>
<td>3.0</td>
</tr>
</tbody>
</table>

- Auto-agglutination in normal saline.

TABLE V. AMPICILLIN-RESISTANT SYSTEMIC *H. INFLUENZAE* TYPE b ISOLATES FROM PAEDIATRIC PATIENTS, AUGUST 1978 - DECEMBER 1979

<table>
<thead>
<tr>
<th>Sources</th>
<th>No. of isolates (β-lactamase-positive/total)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood culture only*</td>
<td>2/4</td>
</tr>
<tr>
<td>CSF only</td>
<td>0/3</td>
</tr>
<tr>
<td>Blood and CSF</td>
<td>1/3</td>
</tr>
</tbody>
</table>

- 3/10

- Includes a patient with epiglottitis and a 3-month-old child with osteomyelitis of the humerus.

DISCUSSION

One of the important features of this study is the much greater prevalence of β-lactamase-producing *H. influenzae* among children seen at the Johannesburg Hospital compared with those seen elsewhere and in adults. Gilliet et al. from the Children’s Hospital in Birmingham, UK, published data suggesting a higher prevalence of β-lactamase-positive isolates in children as opposed to adults. Furthermore, an independent laboratory in Johannesburg finds the prevalence among isolates from children to be approximately three times that among strains from adults (Dr A. C. Mauff — personal communication).

This study unfortunately cannot explain the differences between isolates from children studied at the Johannesburg Hospital or the central SAIMR (Table II). Several factors may have been important, especially the socioeconomic status and race of the populations from which these children were drawn. It is conceivable that the prevalence rates of β-lactamase-positive strains of *H. influenzae* may vary with the frequency of use of ampicillin or related compounds in different communities.

While it is commonly accepted that drug resistance is a feature of hospital-associated bacteria, it would appear that this is not necessarily the case with ampicillin-insensitive *H. influenzae* (Table V). Indeed, all the severe systemic infections due to these organisms were acquired in the community. Two particularly interesting patients were encountered in this group. The first was a 3-month-old male with acute osteomyelitis of the humerus. He had
initially been treated unsuccessfully with penicillin and tobramycin. The organism isolated from blood cultures and pus drained from the arm was found to be \( \beta \)-lactamase-positive and successful therapy with chloramphenicol was instituted. The other patient had bacteraemic epiglottitis due to ampicillin-resistant \( H. \) influenzae. He was intubated and treated with dexamethasone and ampicillin and made a rapid and uneventful recovery, despite the apparently inappropriate antibiotic. This case raises again the question of what constitutes the definitive component of epiglottitis treatment.

The fact that 3 of the last 10 serious cases of \( H. \) influenzae infection seen at the Johannesburg Hospital were caused by \( \beta \)-lactamase-producing strains indicates that primary therapy with ampicillin or similar compounds is likely to be very risky. Studies currently in progress suggest that chloramphenicol resistance has not yet been encountered in locally isolated strains of \( H. \) influenzae. We feel, therefore, that it would be wiser to use chloramphenicol as the initial drug in serious systemic \( H. \) influenzae disease, either alone or at least in addition to ampicillin-type drugs.

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REFERENCES

The Influence of Cardiopulmonary Bypass and Autotransfusion on Platelet Count

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SUMMARY
A pilot study was carried out to determine whether autotransfusion increased the platelet count. Although only a small series was involved significant increases were shown.


In the H. F. Verwoerd Hospital blood for autotransfusion is collected routinely from the venous line of the cardio-pulmonary bypass (CPBP) pump after heparinization. This blood is then reinfused after CPBP. A pilot study was undertaken to prepare a protocol for a more extensive study of the clinical value of autotransfusion as practised here.

METHOD
Nine non-cyanotic patients scheduled for heart valve replacement were anaesthetized according to a standard technique — induction with fentanyl 300 \( \mu \)g followed after 3 minutes by a sleeping dose of diazepam, which varied between 10 and 20 mg. The relaxant used was pancuronium and the anaesthesia was maintained with nitrous oxide in 50% oxygen, supplemented with morphine 1.5 mg/kg body weight given in divided doses over a period of 15 minutes. The central venous pressure, radial arterial pressure, ECG, blood gases, serum electrolyte level, urine output and nasopharyngeal temperature were monitored. Moderate hypothermia of 26 - 28°C was employed.