Comparison between Nasal Swabs and Nasopharyngeal Aspirates for, and Effect of Time in Transit on, Isolation of *Streptococcus pneumoniae*, *Staphylococcus aureus*, *Haemophilus influenzae*, and *Moraxella catarrhalis*

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We assessed the impact of the use of nasal swabs or nasopharyngeal aspirates and the time from specimen collection to storage at −70°C on bacterial isolation. *Haemophilus influenzae* was isolated significantly less often from swabs than from nasopharyngeal aspirates. Samples in transit for >3 days were half as likely to grow *Streptococcus pneumoniae* and *H. influenzae* as those in transit for ≤3 days. There was no statistically significant difference for either *Moraxella catarrhalis* or *Staphylococcus aureus*.

*Streptococcus pneumoniae*, *Haemophilus influenzae*, and *Moraxella catarrhalis* are the most important bacterial pathogens associated with otitis media (OM) (1, 3). In 1999, the Kalgoorlie OM Research Project was established to investigate risk factors for OM in Aboriginal and non-Aboriginal children in a semiarid region of Western Australia (10). Study participants were enrolled in the town of Kalgoorlie, some 350 miles east of the capital of Western Australia, Perth, where samples were to be processed. Variations in sampling technique and delays in culturing may affect the isolation rates of various OM pathogens (8), and the OM study afforded an opportunity to examine the potential impact of sampling method and processing delays.

We assessed whether the use of nasal swabs (NSs) instead of nasopharyngeal aspirates (NPAs) or the time interval between specimen collection and storage at −70°C affected isolation rates of *S. pneumoniae*, *H. influenzae*, *M. catarrhalis*, or *Staphylococcus aureus*. Ethical approval to conduct the Kalgoorlie OM Research Project was given by the Western Australian Aboriginal Health and Information Ethics Committee, the Northern Goldfields Health Service and Nursing Education Ethics Committee in Kalgoorlie, the Princess Margaret Hospital Ethics Committee, and the Confidentiality of Health Information Committee of the Health Department of Western Australia.

NPAs were to be collected from young children seven times before age 2 years, and 1 ml of saline was then added to each specimen. A 0.5-ml volume of mucus plug, or if no visible plug the gently mixed specimen, was pipetted into 1 ml of skim milk-trypotone-glucose-glycerol broth, which was placed immediately at −20°C. While a World Health Organization working party recommended the use of nasopharyngeal swabs for studies of upper respiratory tract bacterial carriage (5), NPAs have been successfully used in upper respiratory tract carriage studies in central Australia (2). If a guardian was unwilling to allow collection of an NPA from their child, we requested permission to collect a sample using the less-invasive NS. When this was agreed to, a specimen was collected by inserting a sterile cotton swab (Interpath L8208) into the nostril as far as possible. This method had been used successfully in studies in Papua New Guinea for 20 years (4).

A sample of children (*n* = 41) had both an NS and an NPA taken at the same time, with the NS always being collected first. All samples were cultured using standard techniques as described previously (10). Comparisons were made with the SPSS software package (version 11.5; SPSS Inc., Chicago, IL), using the test for paired proportions (McNemar’s test).

Specimens were either placed immediately at −20°C or transported on ice to −20°C storage within 1 h, where they remained until being transported on dry ice to a central laboratory in the capital city, Perth, for storage at −70°C and culture. The total transit time from Kalgoorlie to the central laboratory was 4 to 5 h, during which time specimens remained frozen.

For three of the pathogens of interest, little difference was seen in isolation rates for the 41 paired samples. *S. aureus* was isolated from 12 NPAs (29%) and 10 NSs (24%) (*P* = 0.63), *M. catarrhalis* from 7 NPAs (17%) and 8 NSs (20%) (*P* = 1.00), and *S. pneumoniae* from 9 NPAs (22%) and 6...
NSs (15%) \((P = 0.25)\). However, \(H.\, influenzae\) was isolated only from 2 NSs (5%) but from 8 NPAs (20%) \((P = 0.03)\). Assessment of the effect of time between specimen collection and long-term storage at \(-70^\circ\)C on bacterial growth was adjusted for age (median, 4 months; range, 1 week to 2 years), gender (60% male), and Aboriginality (38% Aboriginal, \(n = 496\)). NPAs that were in transit for >3 days were half as likely to grow \(S.\, pneumoniae\) and \(H.\, influenzae\) as those that were in transit for \(\leq 3\) days (Table 1). There was no effect of transit time on the isolation of \(M.\, catarrhalis\) or \(S.\, aureus\). When a transit time of >2 days was examined, there was no longer a significant effect on isolation of \(H.\, influenzae\) (data not shown).

Our results indicate that \(H.\, influenzae\) is isolated less frequently from NSs than from NPAs; however, the use of an NS does not appear to affect the isolation of \(S.\, pneumoniae\), \(M.\, catarrhalis\), or \(S.\, aureus\). This corroborates and extends previous work in which \(H.\, influenzae\) was isolated significantly less often from NSs than from NPAs in children but \(S.\, pneumoniae\) was not (8).

Over the past decade, there have been significant changes worldwide in health care delivery, with a focus on reducing expenditure. As a result, there has been a shift to centralized or consolidated laboratory services that has occurred on both a regional and a national scale (7). Although the issue of transit times for clinical specimens to reach a central laboratory has become much more important, there have been very few investigations of the impact of delays (6). Bacteria that are particularly sensitive to ambient conditions include \(Shigella\) spp., \(Neisseria\, gonorrhoeae, \, Neisseria\, meningitidis, \, S.\, pneumoniae, \, H.\, influenzae,\) and anaerobes (9). In our study, long transit times from the rural center, with the consequence of prolonged storage at \(-20^\circ\)C, appeared to result in a substantial decline in isolation of \(S.\, pneumoniae\) and \(H.\, influenzae\) but not of \(M.\, catarrhalis\) or \(S.\, aureus\). Researchers and laboratory staff who may encounter similar conditions should consider these findings when planning for specimen transport and analysis.

<table>
<thead>
<tr>
<th>Organism</th>
<th>No. of isolates (% of NPAs) after transit time of</th>
<th>Crude ORa</th>
<th>Crude CIf</th>
<th>Adjusted ORc</th>
<th>Adjusted CIf</th>
<th>(P) valuec</th>
</tr>
</thead>
<tbody>
<tr>
<td>(S., pneumoniae)</td>
<td>153 (39.9) 29 (25.7)</td>
<td>0.54</td>
<td>0.35–0.85</td>
<td>0.49</td>
<td>0.30–0.78</td>
<td>0.003</td>
</tr>
<tr>
<td>(H., influenzae)</td>
<td>101 (26.4) 18 (15.9)</td>
<td>0.59</td>
<td>0.34–1.40</td>
<td>0.50</td>
<td>0.27–0.93</td>
<td>0.029</td>
</tr>
<tr>
<td>(M., catarrhalis)</td>
<td>148 (38.6) 39 (34.5)</td>
<td>0.89</td>
<td>0.59–1.35</td>
<td>0.85</td>
<td>0.55–1.32</td>
<td>0.475</td>
</tr>
<tr>
<td>(S., aureus)</td>
<td>169 (44.1) 55 (47.7)</td>
<td>1.20</td>
<td>0.82–1.18</td>
<td>1.30</td>
<td>0.76–2.23</td>
<td>0.330</td>
</tr>
</tbody>
</table>

Total no. of NPAs 383 (100) 113 (100)

\(a\) OR, odds ratio.

\(b\) CI, 95% confidence interval.

\(c\) Adjusted for age (continuous variable), gender, and Aboriginality.

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**REFERENCES**


