Microbial profile of Ear infections and the need for Ear examination before collection of swabs for investigation

Abstract

Acute ear infection is a common childhood ailment. Diagnosis is often difficult mostly because available clinical guidelines in developing countries like Nigeria do not often include ear examinations. Over a twenty-eight month period, records of children 15 years and below sent for ear swab investigations were analysed.

Positive cultures were obtained from 60.8% of the patients. The incidence was greatest in the zero to twelve months and one year to three years age groups. There was a male preponderance of subjects with ear infections. Only 47 (23.6%) of the subjects had full ear examinations by the Otolaryngologist, with 17 of these having perforations of the tympanic membrane. Thus, only these 17 patients (8.5%) can be said to have otitis media. Gram negative organisms were preponderant (62.0%), with *Staphylococcus aureus* and *Pseudomonas aeruginosa* being the most frequently isolated organisms, accounting for 32.5% and 30.2% of all isolates respectively. *Candida albicans* was isolated from six subjects and *Serratia marcescens* from one patient’s specimen. *Staphylococcus aureus*, *Pseudomonas aeruginosa* and Proteus species exhibited multi-drug resistance in vitro, being resistant to three or more classes of antibiotics. Generally, most of the isolates were susceptible in vitro to the quinolones, but resistant to the cephalosporins and nitrofurans. Most of the subjects
did not return for follow-up treatment, with only 3 subjects continually being seen by the ENT specialists.

Conclusion: Ear infections are common in children, but differentiating the types of ear infections is not usually done due to treatment guidelines being vague and not well developed. There is a need for collaborative multi-disciplinary approach in the management of childhood ear infections.

Keywords: Ear infection, children, bacteria, antibiotics, susceptibility

Introduction

Ear infections (otitis) are common in childhood. Otitis externa is the inflammation or infection of the external ear or the external auditory canal [1], while otitis media is that of the middle ear cleft. They can both present with or without ear discharge. Predisposing factors for otitis externa include swimming, humidity, maceration, local trauma, dermatitides, external devices and auditory canal obstruction [2]. Acute Otitis Media (AOM), an acute bacterial infection of the middle ear fluid [3], is one of the most common infectious diseases affecting children. It is estimated that 60 to 80% of children would have experienced at least one episode of acute otitis media within the first 3 years of life [4, 5]. AOM is important as it affects millions of people, with the incidence of AOM reported as 10.85%, with 51% occurring under age of 5 years and about 21000 people dying yearly from its complications [6]. The symptoms of AOM in children include otalgia, ear-rubbing, fever, irritability, restless sleep, diminished appetite and excessive crying, most of which are not peculiar to middle ear infections [7, 8]. These infections are often not obvious initially because of the absence of ear pain or discharge. Also many of the affected children have not reached the age where they can complain about specific symptoms and so ear infections may remain undetected. Clinical
diagnosis is difficult because of overlap of signs and symptoms with those of other respiratory infections [9]. Diagnosis is also hampered by difficulty in examining the ears of children and difficulty in obtaining cultures, so empiric treatment of AOM is therefore common [10]. AOM can also lead to spontaneous perforation of the tympanic membrane and is capable of progressing to chronic suppurative otitis media (CSOM) [11], which is the persistent inflammation of the middle ear or mastoid cavity. It is thus a preventable cause of hearing loss [12].

Risk factors for otitis media in children include: daycare (large group), tobacco smoke, air pollution, bottle feeding, family history, underlying disease (cleft palate, Down syndrome, immunity problems) [3, 13]. Low socio-economic status has been suggested to contribute to acquiring AOM [14], and can contribute to progression to CSOM because of unaffordable treatment or lack of treatment [15].

There has been an increase in the incidence of otitis media in preschool children [16], and also an increase in the prescription of broad-spectrum antibiotics for AOM in recent times [17]. There have also been increasing reports of antibiotic resistance of pathogens responsible for Otitis media [8, 18-20].

This study therefore sought to determine the common microbial causes of ear infections among children sent for ear swab investigations, and the susceptibility patterns of the isolated pathogens to antibiotics. This would hopefully help in determining antibiotics used in the empiric treatment of ear infections in Bayelsa State.

**Materials and Methods**

This retrospective cross-sectional study was carried out between October 2011 and December 2013, at the Niger Delta University Teaching Hospital (NDUTH), Okolobiri, Bayelsa State, Nigeria. It is a 178-bed health facility providing services for
patients across three states in the Niger Delta region of Nigeria (Bayelsa, Delta and
Rivers).

The records of patients at the Paediatrics Outpatient, Ear, Nose and Throat clinic
and the Microbiology Department were examined. Children up to the age of 15 years
suspected of having ear infections (with or without discharging ears) and who had
been sent for collection of ear swabs, were recruited into the study. Children who
had been recently treated for otitis media or had been on any form of antibiotic within
the past four weeks were excluded from the study.

**Ethical consideration**

Ethical approval for the study was obtained from the Ethics Review Board of the
NDUTH.

**Specimen collection**

Records showed that specimen collection was done following the approved Standard
Operating Procedures of the NDUTH. Commercially available sterile swab sticks
were used to gently swab the aural canal following the laid down hospital rules for
the procedure. Two swabs were collected per subject (left and right ears).

Swabs were inoculated onto suitable media: Chocolate agar, Blood agar and
McConkey agar and incubated aerobically for at least 48 hours. Isolates were Gram
stained, and microbial isolates were further identified and classified using standard
biochemical tests. Isolates that could not be further differentiated were described
either as species or Coliforms.

Antibiotic susceptibility patterns were determined using the Kirby-Bauer disk diffusion
method and interpreted using the standards of the Clinical Laboratory Standards
Institute [21]. Sensitivity to the antibiotics was classified as susceptible, intermediate
and resistant. For this study intermediate and resistant were put together as resistant.

The antibiotic discs used were: Amoxicillin 25µg; Amoxicillin-Clavulanic acid 30µg; Chloramphenicol 30µg; Cefixime 5µg; Ceftazidime 30µg; Cefuroxime 30µg; Ciprofloxacin 5µg; Co-trimoxazole 25µg; Gatifloxacin 5µg; Gentamicin 10µg; Nalidixic acid 30µg; Nitrofurantoin 30µg; Ofloxacin 5µg; Streptomycin 10µg; Tetracycline 30µg.

Resistance values were computed by simply dividing the number of isolates of a species resistant to a particular antibiotic tested, by the total number of that particular isolate and expressed as percentage resistance. A bacterial species was considered resistant to a particular antibiotic when more than 50% of the isolates show in vitro resistance to it, and multi-drug resistance when more than 50% of the isolates were resistant to three or more classes of antibiotics.

In line with existing Clinical guidelines of the hospital, most subjects (especially those with discharging ears) were commenced on broad-spectrum antibiotics after the ear swabs were collected, while waiting review with the test results.

**Statistical analysis**

Differences between variables were analysed using the Graphpad Prism version 4® (Graphpad software, San Diego, CA). Significant difference was observed if the probability of the results observed due to chance was less than 5% (p<0.05).

**Results**

Over a period of twenty-seven months, 199 patients aged 2 weeks to 15 years had ear swabs taken in the course of investigation. There were 113 males and 86 females, a male:female ratio of 1.3:1. More than half of the patients were in the age
groups zero to 12 months and 1 year to 3 years (Table 1). The number of patients declined with increasing age.

Table 1. Age, sex distribution of the study subjects

<table>
<thead>
<tr>
<th>Age Group (years)</th>
<th>Age distribution of patients</th>
<th>Patients with positive cultures</th>
<th>% patients with ear infection</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Male</td>
<td>Female</td>
<td>Total</td>
</tr>
<tr>
<td>0 to 12 months</td>
<td>37</td>
<td>32</td>
<td>69</td>
</tr>
<tr>
<td>1 yr - 3 yrs</td>
<td>29</td>
<td>23</td>
<td>52</td>
</tr>
<tr>
<td>4 yrs - 6 yrs</td>
<td>22</td>
<td>18</td>
<td>40</td>
</tr>
<tr>
<td>7 yrs - 9 yrs</td>
<td>9</td>
<td>5</td>
<td>14</td>
</tr>
<tr>
<td>10 yrs - 12 yrs</td>
<td>11</td>
<td>4</td>
<td>15</td>
</tr>
<tr>
<td>13 yrs - 15 yrs</td>
<td>5</td>
<td>4</td>
<td>9</td>
</tr>
<tr>
<td>Total</td>
<td>113</td>
<td>86</td>
<td>199</td>
</tr>
</tbody>
</table>

The prevalence of ear infections was 60.8%, as micro-organisms were isolated from specimens obtained from 121 out of the 199 patients (72 males, 49 females). No microorganisms were isolated from 78 patients: 41 male, 37 female (39.2%).

Patients in the age groups 3 years and below, accounted for 58.7% of microbial isolates. The incidence of ear infection declined with increasing age (Table 1).

There were no statistically significant differences observed in the incidence of ear infection between male and female sexes.

From the 121 patients with positive cultures, 126 microbial isolates were obtained.

Five patients (4.1%) had more than one microorganism isolated from their specimens, three (2.5%) had two different bacterial species in each aural canal, while two (1.7%) had two different bacterial species isolated from the same middle ear. Six patients had fungal infection of the middle ear.
Microbial isolates

Gram negative bacilli made up 62% of the isolates. *Staphylococcus aureus* and *Pseudomonas aeruginosa* were the most common isolates accounting for about two-thirds of the microbial isolates (Table 2.). Three isolates, Gram negative bacilli, could not be further differentiated and were classified as coliforms. One specimen grew *Serratia marcescens*.

Table 2. Frequency of isolated micro-organisms

<table>
<thead>
<tr>
<th>Organism isolated</th>
<th>Sex</th>
<th>Male</th>
<th>Female</th>
<th>Total</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Staphylococcus aureus</em></td>
<td>Male</td>
<td>29</td>
<td>12</td>
<td>41</td>
<td>32.5</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>12</td>
<td>12</td>
<td>24</td>
<td>19</td>
</tr>
<tr>
<td><em>Pseudomonas aeruginosa</em></td>
<td>Male</td>
<td>15</td>
<td>23</td>
<td>38</td>
<td>30.2</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>23</td>
<td>23</td>
<td>46</td>
<td>36.3</td>
</tr>
<tr>
<td>Proteus species</td>
<td>Male</td>
<td>16</td>
<td>6</td>
<td>22</td>
<td>17.5</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>6</td>
<td>6</td>
<td>12</td>
<td>9.6</td>
</tr>
<tr>
<td><em>Klebsiella pneumoniae</em></td>
<td>Male</td>
<td>4</td>
<td>2</td>
<td>6</td>
<td>4.8</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>2</td>
<td>2</td>
<td>4</td>
<td>3.2</td>
</tr>
<tr>
<td><em>Escherichia coli</em></td>
<td>Male</td>
<td>3</td>
<td>3</td>
<td>6</td>
<td>4.8</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>3</td>
<td>3</td>
<td>6</td>
<td>4.8</td>
</tr>
<tr>
<td><em>Candida albicans</em></td>
<td>Male</td>
<td>4</td>
<td>2</td>
<td>6</td>
<td>4.8</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>2</td>
<td>2</td>
<td>4</td>
<td>3.2</td>
</tr>
<tr>
<td>Coliform</td>
<td>Male</td>
<td>3</td>
<td>0</td>
<td>3</td>
<td>2.4</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0.0</td>
</tr>
<tr>
<td><em>Streptococcus pneumoniae</em></td>
<td>Male</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>2.4</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>2</td>
<td>2</td>
<td>4</td>
<td>3.2</td>
</tr>
<tr>
<td><em>Serratia marcescens</em></td>
<td>Male</td>
<td>1</td>
<td>0</td>
<td>1</td>
<td>0.8</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0.0</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td></td>
<td>126</td>
<td>100</td>
<td>226</td>
<td>100</td>
</tr>
</tbody>
</table>

Susceptibility to antibiotics

*Staphylococcus aureus* isolates were mostly resistant in vitro to the aminoglycosides, nitrofurans and one of the cephalosporins (ceftazidime) (Table 3). They were however sensitive to the actions of ofloxacin. *Pseudomonas aeruginosa* was resistant in vitro to four classes of antibiotics - aminoglycosides, cephalosporins, nitrofurans, penicillin combinations and one of the quinolones (nalidixic acid) (Fig. 1), but was sensitive to the other quinolones (ciprofloxacin and ofloxacin). Proteus species were resistant to aminoglycosides, cephalosporins, nitrofurans and penicillin combinations, but were sensitive to the quinolones. *Klebsiella pneumoniae* isolates were resistant to only the cephalosporins and nitrofurans. They were sensitive in
vitro to the actions of aminoglycosides, quinolones and penicillin combinations.

*Escherichia coli* isolates were resistant to Amoxycillin, Cefuroxime, Gentamicin and Ciprofloxacin, but sensitive to Nitrofurantoin. *Staphylococcus aureus, Pseudomonas aeruginosa* and proteus species isolates therefore exhibited multi-drug resistance. Generally, most of the isolates were susceptible in vitro to the quinolones, and resistant to the cephalosporins and nitrofurans.

**Table 3. Susceptibility of the bacterial isolates to antibiotics expressed in percentage resistance**

<table>
<thead>
<tr>
<th>Antibiotic class</th>
<th>Antibiotic</th>
<th>S. aureus</th>
<th>P. aeruginosa</th>
<th>Proteus</th>
<th>K. pneumoniae</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aminoglycoside</td>
<td>Gentamicin</td>
<td>51.2</td>
<td>57.9</td>
<td>55</td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Streptomycin</td>
<td>100</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Quinolones</td>
<td>Ciprofloxacin</td>
<td>-</td>
<td>24.1</td>
<td>29.4</td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Nalidixic acid</td>
<td>-</td>
<td>85.7</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Ofloxacin</td>
<td>25.2</td>
<td>27.9</td>
<td>19.4</td>
<td>20</td>
</tr>
<tr>
<td>Cephalosporins</td>
<td>Cefuroxime</td>
<td>40.6</td>
<td>96.3</td>
<td>73.3</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>Ceftazidime</td>
<td>71</td>
<td>60.7</td>
<td>46.7</td>
<td>60</td>
</tr>
<tr>
<td></td>
<td>Cefixime</td>
<td>-</td>
<td>94.7</td>
<td>86.7</td>
<td>100</td>
</tr>
<tr>
<td>Penicillin</td>
<td>Amoxicillin-Clavulanic acid</td>
<td>45.9</td>
<td>100</td>
<td>85</td>
<td>40</td>
</tr>
<tr>
<td>combination</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nitrofurans</td>
<td>Nitrofurantoin</td>
<td>66.7</td>
<td>85.2</td>
<td>83.3</td>
<td>80</td>
</tr>
</tbody>
</table>
Fig. 1. Resistance pattern of Pseudomonas aeruginosa isolates to four classes of antibiotics

Key: GEN-gentamycin; CPR-ciprofloxacin; NAL-Nalidixic acid; OFL-ofloxacin; CXM-cefuroxime; CAZ-ceftazidime; CFM-cefixime; AMC-amoxicillin-clavulanic acid; NIT-nitrofurantoin

Clinical outcome

There were no clearly defined clinical guidelines for the management of children with ear infections in the Paediatrics and Ear, Nose and Throat Departments. Out of the 199 patients, only 47 had been referred to the Ear, Nose and Throat (ENT) Department and had full examination of their ears. One hundred and fifty two patients had provisional diagnosis of acute otitis media without examination of the ears. Seventeen (36.2%) of these 47 patients had various degrees of perforation of the tympanic membrane. All seventeen had specimens yielding bacterial growth thus confirming otitis media. However, only three of them continued to be seen at the
ENT Department. Twenty-three (11.6%) patients did not return back to the hospital to be reviewed with the results of their ear swab tests.

**Discussion**

Ear infections continue to be important causes of illness in children. The present study observed that 60.8% of the subjects had acute ear infection, which is lower than the 80.4% reported in Ethiopia [22] and less than the earlier observation that 70% of middle ear cultures yielded growth [23]. An earlier study done in the same hospital had yielded only 7.9% growth from ear cultures in children with febrile illnesses [24]. Otitis was noted to occur more in children less than 3 years of age, which is similar to results from other Nigerian studies [25]. There was a male preponderance of children with otitis, similar to previous studies [26]. Our results did not show any difference in the causative organisms between age groups and between the sexes just as in previous reports [23].

*Streptococcus pneumoniae* has previously been reported to be the most common pathogen responsible for acute otitis [27, 28] and accounts for 30-50% of cases [29]. However, the present study observed *Staphylococcus aureus* closely followed by *Pseudomonas aeruginosa* as the commonest causes of acute otitis, similar to findings in Iran, Jordan and India [19, 22, 30, 31]. Other studies in Nigeria found *Pseudomonas aeruginosa* as the commonest bacterial cause of AOM [20, 32]. *Haemophilus influenzae* and *Moraxella catarrhalis* commonly reported as pathogens causing AOM in Western countries were not identified in the present study as well as other studies from Africa, Middle East and Asia, except for South Africa [27]. This suggests the existence of regional/geographical differences in the aetiology of AOM.
and is in keeping with earlier reports [33], however another study posited that the pathogens were similar across countries [34].

Antibiotic treatment of AOM has benefits as long term treatment has been shown to prevent tympanic membrane perforation [35], but the use and overuse of antibiotics has been associated with the increase in antibiotic resistance [10]. This is why determining the antibiotic susceptibility of microbial isolates is important so as to avoid using ineffectual antibiotics.

Ciprofloxacin was the most effective antibiotic against the bacterial isolates in the present study. This was also the observation in some other studies [19, 32]. *Pseudomonas aeruginosa* in this study exhibited very high resistance (100%) in vitro to the actions of amoxicillin-clavulanic acid, similar to reports from Ethiopia [22], but different from others which show amoxicillin-clavulanic acid to be effective [36] (Hoberman. Most of the isolates, apart from Klebsiella pneumoniae, were resistant to gentamicin. This may be linked to the wide usage of gentamicin ear drops for self-medication and also empirical treatment by healthcare workers.

With the development of multi-resistant bacterial strains [37], vaccines to prevent AOM are being advocated and used in some countries [38, 39]. While attention has mostly been paid to bacterial causes of AOM, the present study and several others have isolated fungi, notably *Candida albicans* [19, 20]. More studies are needed to evaluate the consequences of fungal infections of the middle ear.

This study revealed the near absence of ear examination in the management of children with ear complaints. The assumption that any inflammation or infection of the ear is acute otitis media is worrisome. There is a need for collaborative patient [40] the management of childhood otitis. This will ensure proper examination and will
effectively reduce the number of patients not completing treatment. Thus, complete ear examinations will be incorporated into the clinical guidelines for children, because otoscopic findings have been shown to be critical for accurate AOM diagnosis [40]. Findings in the present study will hopefully contribute to the development of evidence-based practice guidelines for the management of AOM in the paediatric population and in the planning of interventions to improve quality healthcare and reduce costs.

**Conclusion**

Acute otitis is common in the NDUTH and its importance is presently underestimated. There is a need for the development of robust clinical guidelines and multidisciplinary approach to the management of childhood otitis media.

**References**


