

Bacteriological pattern of *Otitis media* among children attending the ENT unit of university of Benin teaching hospital

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Abstract

Studies have reported that the types of pathogen involved in *Otitis media* (OM) are dependent on geographical location and respiratory infections. This has resulted in contraction between studies on the prevalent causative pathogen. The prevalent causative bacterial of *Otitis media* (OM) was investigated among 16 children who presented positive at the Ear Nose and Throat (ENT) unit of University of Benin Teaching Hospital between 1st August and 31st October 2011, using standard microbiological methods. The results revealed that of the 16 positive samples collected, 37.5% yielded the growth of *Klebsiella aerogenes*; 25.0% *Proteus spp*; 18.75% *Pseudomonas aeruginosa*; 12.5% *Staphylococcus aureus*, and 6.25% *Escherichia coli*. Base on this results, the causative bacterial of OM can varies and hence, the needs for a microbiological screening before treatment with antibiotic to minimise multi-drug resistant.

Keywords

Otitis media, Children, Causative Organism, Bacterial

1. Introduction

Otitis media (OM) is the infection (an inflammation) of the middle ear caused by pathogenic microorganisms (Bacteria, fungi and virus) resulting in inflammation of mucosal lining (Abera and Biadeglenn, 2008). It is a common infants and young children infection with a peak incidence between 4-7 years of age [Bluestone and Klein, 1995]. Tortora and Funke (1992) added that it is caused by the entrance of bacteria through the eustachian tube. On the other hand, Kumurya et al. (2010), reported it is caused by the build-up of fluid behind the ear drum as a result of a blockage to the Eustachian tube. It is one of the two categories of ear inflammation that underlay what is commonly called an earache, the other being otitis external (Owen et al., 2003).

Worldwide, sixty five to three hundred and thirty million people suffer this infection, of which about 60% experience significant hearing impairment (Woodfield and Dugdale, 2008). Several risk factors; such as previous acute otitis

media, hereditary and family history of middle ear disease, cultural, socio-economic status, parental smoking, age factors, attending day care centers and seasonality, have been associated with it [Rotowa et al., 1999; Alho et al., 1995; Daly, 1991]. However, children below the age of seven years are much more susceptible due to their shorter Eustachian tube that is more of a horizontal angle, as well as lower immunity and or resistance to microbes as found in adults [Chan et al., 1993]. The incidence rate is said to be higher in male than female [Ali et al., 2013, Paradise et al., 1997]. While wide variations exist in the prevalence rate of OM in both rural and urban dwellers, Okoewo (1985) reported a prevalence of 0.6% and 3.6% among urban and rural school children respectively in Lagos. After a decade and half, Olusanya (2000) reported a prevalence of 18.7% in Lagos. On the other hand, Ogisi (1988) working in Benin City reported a prevalence of 1.2%-6%. Shamsuddeen et al. (2010) working on pediatric patients attending Aminu Kano Teaching Hospital, Kano revealed 94.0% of the samples

yielded growth. Nwogwugwu *et al.*, (2014) working on ear swab of 152 children with ear discharge in some parts of Owerri, Imo State – Nigeria revealed that 84.2% were positive bacterial growth.

According to Kilpi *et al.* (2001) and Block (1997), *Streptococcus pneumoniae*, *Haemophilus influenzae*, and *Moraxella catarrhalis* are the most common bacterial pathogens in OM. The most predominant isolate in the study of Shamsuddeen *et al.* (2010) working on pediatric patients attending Aminu Kano Teaching Hospital, was *Staphylococcus aureus*, with a total occurrence of 26 (55.32%) followed by *Proteus mirabilis*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Streptococcus pneumoniae*, with total occurrences of 12 (25.53%), 7 (14.89%), 1(2/13%) and 1 (2.13%) respectively. The predominant organisms isolated in Nwogwugwu *et al.*, (2014) study included *Pseudomonas aeruginosa* (37.5%), *Staphylococcus aureus* (23.4%), *Proteus* species (12.5%), *Klebsiella pneumoniae* (7.8%), *Escherichia coli* (7.8%) and *Streptococcus pneumoniae* (7.8%). Nwankwo and Okeke (2014) working on 50 ear swab samples from patients presenting with otitis media at Federal Medical Centre Umuahia, Abia State, Nigeria reported *Staphylococcus aureus* as the most predominant isolates with a total occurrence of 12 (24.5%) followed by *Haemophilus influenza* 9(18.4%), *Streptococcus* sp 8 (16.3%), *Pseudomonas aeruginosa* 7(14.3%), *Proteus* sp and *Escherichia coli* 5 (10.2%) and *Klebsiella* sp 3 (6.1%). Other pathogens responsible for OM are *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella spp.*, *Pseudomonas aeruginosa*, and *Proteus* spp [Klein, 1994] which Kilpi *et al.* (2001), classified as less commonly. Occasionally, OM may be caused by fungi (*Aspergillus* or *Candida*) or other pathogens such as herpes virus (Ettehad *et al.*, 2006; Mongkolrattanothai *et al.*, 2003), in this situation usually, either there is a problem with immune function or there is a hole (perforation) in the ear drum (Mongkolrattanothai *et al.*, 2003). In this regards, studies have reported that the types of pathogen involved in OM are dependent on geographical location and respiratory infections (Ettehad *et al.*, 2006; Jacobs *et al.*, 1998).

Despite the fact that OM is still a relatively common condition in children in developing countries compared to advanced countries (Lasisi, 2008) as well as the fact that it is an important cause of functional disability, couple with the contradictions in the prevalence causative bacterial as seen in the literature, in Nigeria the significance of OM has not been fully established. Worrisome, OM is the most common reason for antibiotic treatment in childhood (Froom *et al.*, 1997), and increasing antibiotic consumption has been shown to be related to the emerging phenomenon of antimicrobial resistance (Cristino, 1999). This study therefore was carried out to investigate the prevalence causative bacterial of OM among children who presented positive at the Ear Nose and Throat (ENT) unit of University of Benin Teaching Hospital at as the time of the study.

2. Materials and Methods

2.1. Material/ Equipment

The materials used in this study includes weight balance, petridishes, bursen burner, inoculating wireloop, Bijoux bottles, aluminium foil, test tubes, conical flask, measuring cylinder, spatula, Pasteur pipette, hot air oven, incubator, cotton wool, autoclave, test tubes rack, sterile distilled water and disinfectant.

2.2. Media

The media include MacConkey agar, blood agar, Mueller-Hinton agar, Nutrient agar and peptone water. Media were prepared according to the manufacturers' instructions.

2.3. Samples Source and Storage of Isolates

16 positive samples received from 1st of August to 30th of October 2011 from ENT (Ear, Nose and Throat) unit in University of Benin Teaching Hospital (UBTH) were used for this study. Test isolates were kept on nutrient agar slope and kept in the refrigerator at 4⁰C before use.

2.4. Cleaning and Sterilization of Equipment Used

The glass wares were washed with detergent and rinsed in distilled water. All glass wares were sterilized using hot air oven at 160⁰C for 1 hour. The wire loops were also sterilized by passing them through a bursen burner until red hot before use.

2.5. Identification of Test Isolates

The plates were dried briefly in the incubator at 45⁰C for 15 minutes before use. Each specimen was streak inoculated on MacConkey agar plates and blood agar plates and incubated at 37⁰C for 18-48 hours after which it was examined for growth.

The isolates were provisionally identified in the medical microbiology laboratory of University of Benin Teaching Hospital (UBTH) and confirmatory identification was done in microbiology laboratory at Faculty of Natural Sciences, Ambrose Alli University, Ekpoma. The following criteria were used:

2.6. Identification Criteria (Orhue, 2004) Gram Staining

This technique helps to group organism into Gram-positive and Gram-negative. It shows the shape of the organism (rod, spiral, cocci) and arrangement of the organism (i.e. in chains, clusters, singly).

Colonies of the organism on culture media were aseptically picked with sterile wire loop and it was then emulsified in a drop of normal saline on a glass slide. The smear was allowed to air dry and then fixed with heat by passing through heat for about three (3) times. The slide was placed on a staining rack and the fixed smear was covered

with crystal violet for thirty (30) seconds. This was washed off with clean distilled water. This was followed by the application of Lugol iodine for one (1) minute after which clean distilled water was used to rinse.

The smear was decolourized with acetone for ten (10) seconds and was washed immediately with clean distilled water before counter staining with Safranin for one (1) minute. This was then washed off with distilled water and allowed to air dry. After which a drop of immersion oil was dropped on the stained portion of the slide and then viewed under immersion objective lens. All Gram-positive organisms took the colour of the primary dye which is dark purple, while the Gram-negative organisms took the colour of the secondary dye i.e. pink.

3. Results

Table 1 shows the bacteriological pattern of the sample *Otitis media* positive children attending the ENT unit of University of Benin Teaching Hospital. The results revealed that of the 16 positive samples collected, 6(37.5%) yielded the growth of *Klebsiella aerogenes*; 4(25%) *Proteus spp*; 3(18.75%) *Pseudomonas aeruginosa*; 2(12.5%) *Staphylococcus aureus*, and 1(6.25%) *Escherichia coli*. *Klebsiella aerogenes* was the most prevalent bacterial causing OM in children in the study area.

Table 1. Bacteriological pattern of *Otitis media* among children attending University of Benin Teaching Hospital

Bacterial isolate	Frequency	Percentages
<i>Klebsiella aerogenes</i>	6	37.5%
<i>Proteus spp</i>	4	25.0%
<i>Pseudomonas aeruginosa</i>	3	18.75%
<i>Staphylococcus aureus</i>	2	12.5%
<i>Escherichia coli</i>	1	6.25%
	16	100.0

4. Discussion

The prevalence rate of OM obtained from studies in Nigeria has varies widely ranging from as low as 1.2 to as high as 94%. Okoewo (1985) reported a prevalence rate of 0.6 and 3.6 among urban and rural school children respectively in Lagos while Ogisi (1988) reported a 1.2 to 6 prevalence rate in Benin City (Ogisi, 1988) and Olusanya *et al.* (2000) reported a prevalence rate of 18.7% in Lagos. The very high prevalence rates were 94.0% reported by Shamsuddeen *et al.* (2010) working on paediatric patients in Kano and 84.2% reported by Nwogwugwu *et al.*, (2014) working on children with ear discharge in Owerri, Imo State.

According to this study, the most prevalent causative bacterial of OM was *Klebsiella aerogenes* followed by *Proteus spp*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Escherichia coli* in this order. Although previous studies have reported similar groups of bacteria from patients with otitis media and have established the significance of bacterial species as causative agents of otitis media. However, the reported most prevalent bacterial pathogen is

contradicting. Our finding disagrees with the result by Shamsuddeen *et al.* (2010) who reported *Staphylococcus aureus* as the most prevalence bacterial isolates followed by *Proteus mirabilis*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Streptococcus pneumonia* among pediatric patients attending Aminu Kano Teaching Hospital. Also, the finding of this study disagrees with the study of Nwogwugwu *et al.*, (2014) who reported *Pseudomonas aeruginosa* as the most prevalence causative bacterion of OM. According to Nwogwugwu *et al.*, (2014), this was followed by *Staphylococcus aureus*, *Proteus* species, *Klebsiella pneumoniae*, *Escherichia coli* and *Streptococcus pneumoniae*. Our finding also disagrees with the study by Nwankwo and Okeke (2014) who was working in Abia State, Nigeria and reported *Staphylococcus aureus* as the most predominant isolates followed by *Haemophilus influenza*, *Streptococcus sp.*, *Pseudomonas aeruginosa*, *Proteus sp.*, *Escherichia coli*, and lastly *Klebsiella sp.*, which was the most prevalence in our study. Although the organisms in most studies are similar, however, they differ in the most prevalent causative organism. Hence, justifying the fact that OM pathogenic pattern may be dependent on geographical location. Indeed, it has previously been reported by studies that the types of pathogen involved in OM are dependent on geographical location and respiratory infections (Ettehad *et al.*, 2006; Jacobs *et al.*, 1998] and this has been confirmed by the findings in our study considering the variation in the most prevalent causative bacterial observed in our study and other studies in Nigeria.

Although *Streptococcus pneumoniae*, *Haemophilus influenzae*, and *Moraxella catarrhalis* are the most common bacterial pathogens in OM (Kilpi *et al.*, 2001; Block (1997), our finding contradict this common pathogen of OM. Bacterial classified as the less commonly pathogens responsible for OM (Kilpi *et al.*, 2001) were among the most causative pathogens responsible for OM in the present study. This may be the cause of the increasing antibiotic consumption that has been related to the emerging phenomenon of antimicrobial resistance reported by Cristino (1999)

In another line of thought, the most prevalent causative bacterial organism; *Klebsiella aerogenes*, which is one of the most common Gram-negative bacteria seen by physicians worldwide is reported to be difficult to control and has resulted in mortality rates as high as 50% after antibiotic treatment (Straus 1986). Moreover, *K. pneumoniae* genome has been shown to be slightly more divergent from the *E. coli* genome (McClelland *et al.* 2000) and by implication requires different antibiotic and hence, the needs for a microbiological screening before treatment with antibiotic to minimise multi-drug resistant.

5. Conclusion

Conclusively, *Klebsiella aerogenes* was the most prevalent bacterial causing OM in children in the study area. According to our results, this was followed by *Proteus spp* and then

Pseudomonas aeruginosa. The popular most prevalence reported bacterial; *Staphylococcus aureus* and *Escherichia coli*, reported and known by many studies were the least causative bacterial of *MO* in our study. Although our study showed similar group of bacterial to be causative organisms of *OM*, however, the preponderance of *Klebsiella aerogenes* in this study differs from what had been reported. This finding indicates the need for change in drug of choice for the treatment of *OM*. It is therefore recommended that microbiological screening before treatment with antibiotic to minimise multi-drug resistant be advocated.

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