



Anaerobic Bacteriology Of Middle Ear Aspirate culture in the Developing World: Possible role of Immuno-compromise in its Etio-Pathogenesis?

***Adebola SO, MBBS
Consultant Otorhinolaryngologist.**

**Department of Otorhinolaryngology, Head and Neck surgery,
Ladoke Akintola University of Technology Teaching Hospital, Ogbomosho, Nigeria.**

Disclosures

- ❖ The Author wish to state that some aspects of the data presented here have been presented at the IFOS meeting, Korea, 2013 and also published in the Anaerobe journal.
- ❖ Otherwise the Author declares no conflicts of interest

Introduction

- ❖ In Chronic Suppurative Otitis Media (CSOM) the aetiological agents often involve bacteria but superimposed fungal infections may also occur, hence mixed aetiology has often been reported.¹
- ❖ While epidemiology is said to be global, it tends to involve the paediatric age group and the burden in sub-Saharan Africa is enormous.²⁻⁴
- ❖ Amongst other risk factors, immunocompromised states such as malnutrition have been found to constitute significant risk factors in the aetiology of chronic otitis media.⁵

Introduction I

- ❖ Diabetes mellitus is another immunocompromised condition, for which several immune function factors have been attributed to increased risks of the condition.
- ❖ Often reversed substantially by normalization of the pH and blood glucose levels.⁶
- ❖ Despite the importance of anaerobic bacteria agents in the aetiology of CSOM, there is dearth in literature and also limited knowledge of these agents in our environment.
- ❖ Hence, the study intends to report the profiles of anaerobic bacteria isolated and attempts to evaluate the impact of immunocompromised status of patients on the disease.

Material and Methods

- ❖ This was a prospective hospital-based study of 104 consecutive consenting participants with clinical diagnosis of CSOM attending the ENT clinic at the University Of Ilorin Teaching Hospital, Ilorin.
- ❖ Duration of study was over a 7 month period (September 2010 to March 2011).
- ❖ Ethical approval was sought from the UITH Ethics committee and obtained before the commencement of the study.

Material and Methods I

Inclusion criterion

- was a documented purulent otorrhea through a perforated tympanic membrane for at least 12 weeks.

Exclusion criteria

- i. Those who failed to give a written informed consent to participate in the study,
- ii. Uncooperative patients.
- iii. Patients with inactive ear disease (dry middle ear).
- iv. Patients on topical / systemic antimicrobial therapy whose time of last use of therapy was less than 14 days (< 2 weeks) prior to carrying out the aspirate.
- v. Presence of tympanostomy tubes at the time of carrying out the aspirate.
- vi. Previous surgical intervention done on account of the ear disease.

Material and Methods II

- ❖ External auditory canal of each patient cleaned and middle ear exudates of participants aspirated aseptically by the author.
- ❖ Each ear was done separately in cases of bilateral infection.
- ❖ All specimens were inoculated into the Robertson's Cooked Meat (RCM) enrichment medium (for anaerobic study) while the second portion was inoculated into the Stuart transport medium (for aerobic study).
- ❖ All the microscopy, culture and sensitivity were carried out at the Medical Microbiology department of University of Ilorin Teaching Hospital, Ilorin supervised by a Consultant Microbiologist.

Material and Methods II

- ❖ Antibiotic susceptibility testing was also done using disc diffusion method. *Bacteroides* ATCC 25285 and *Peptostreptococcus* ATCC 29328 were used as control organisms.¹²
- ❖ Data collected was collated using SPSS version 18 (SPSS Inc, Chicago, IL, USA) software package and used for data analysis.
- ❖ Results were then presented in descriptive tables. A p-value of <0.05 was statistically significant.

Results

- ❖ A total of 11 patients with chronically discharging ears, of the 104 studied had anaerobic bacteria cultured from their middle ear aspirates.
- ❖ Age ranged from 4 to 50 years, Male to Female ratio was 1:1.8. The mean age and standard deviation of the patients was 16.3 (SD = 18.8) (Table I).
- ❖ Patients from a low socioeconomic status (SES) constituted about half of the patients studied (45.5%), while the middle SES was 36.4% followed by the high SES with 18.2%.
- ❖ The age-range of the patients who had anaerobic bacteria cultured occurred at the extreme of ages, <10 years (8) and 40 – 50 years (3)

Results I

	Participants	
	Percentages	Frequency
Age range (years)	(n=11)	%
1 - 5	5	45.5
6 - 10	3	27.3
40 - 45	2	18.2
46 - 50	1	9.1
Mean + SD	16.3 (SD = 18.8)	
Gender of participants		
Male	4	36.4
Female	7	63.6
Socioeconomic Status		
High	2	18.2
Middle	4	36.4
Low	5	45.5

Table I: Socio-demographic characteristics of patients that had anaerobic bacteria cultured from their middle ear aspirates

Results II

	Bacteria isolated	Frequency	Percentage
Anaerobic Bacteria			
Gram Positive cocci	Peptostreptococcus	8	25
Gram Negative bacilli	Bacteroides species	3	9.4
Aerobic Bacteria			
Gram Positive cocci	Streptococcus pneumoniae		6.2
Gram Negative bacilli	Staphylococcus aureus		9.4
	Klebsiella spp		6.2
	Escherichia coli		9.4
	Pseudomonas aeruginosa		28.2
	Proteus mirabilis		6.2
Total		32	100.0

Table II: Anaerobic bacteria isolates from patients with chronic suppurative otitis media in Ilorin during the study period

Results III

Anaerobic bacteria isolates from chronic suppurative otitis media specimen	In vitro antibiotic sensitivity [n (%)]					
	Metronidazole	Penicillin	Gentamycin	Chloramphenicol	Clindamycin	Cefoxitin
	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)
Peptostreptococcus	5 (62.5)	4 (50)	5 (62.5)	3 (37.5)	3 (37.5)	4 (50)
Bacteroides species	3 (100)	-	1 (33.3)	2 (66.7)	1 (33.3)	1 (33.3)

Table III: Antibiotic sensitivity pattern of anaerobic bacteria cultured from the aspirates of patients with chronic suppurative otitis media during the study period

Results IV

Immunocompromised Conditions	Group A (N=11)	Group B (N=93)	P-value (power)
Malnutrition	2 (40%)	5 (38.4%)	<0.05 (70%)
HIV/AIDS	2 (40%)	4 (30.8%)	
Diabetes Mellitus	1 (20%)	4 (30.8%)	
Total	5	13	

Table IV: Comparism of the immunocompromised patients with (Group A) and without (Group B) anaerobic bacteria organisms cultured in the middle ear aspirates

Discussion

- ❖ The study noted a female preponderance (male : female ratio, 1 : 1.8) which was opposed to the male preponderance reported in the earlier study done in Ilorin and similar studies done in the West African sub-region.⁷⁻⁹
- ❖ Although some other works have reported a near equal distribution,^{10, 11} this finding may not be unconnected with the improved female awareness and gender sensitivity which might have prompted earlier presentation than previously noted.¹⁸
- ❖ In Nepal, a male preponderance of 1.3 : 1 was reported in the study done by Srivastava *et al.*¹²
- ❖ In that study, the predominance of the disease in males were attributed to their outdoor working habit which exposes them to contamination. This finding was corroborated by other studies done in the Asian continent.^{13, 14}

Discussion I

- ❖ The finding of almost half of the population studied (45.5%), belonging to the low socioeconomic class is in keeping with some other studies done in the West African sub-region^{8, 15, 16,17} and even those involving American Indians.²⁴
- ❖ Lasisi *et al*⁵ found a 7 – 8 times odds of developing otitis media in patients from low socioeconomic class in their own study
- ❖ Some other studies showed higher prevalence in infants from high socioeconomic status which was attributed to infant feeding practices, cigarette smoke exposure and day care attendance.¹⁸
- ❖ Poverty, malnutrition, overcrowding and inability to use the insufficient health facilities available might be responsible for the chronicity of the patients' conditions

Discussion II

- ❖ The anaerobic bacteria organisms seen in the study were *Peptostreptococcus* (25%) and *Bacteroides species* (9.4%) and were all associated with mixed infections.
- ❖ All anaerobic gram negative organisms and 5 (62.5%) gram positive agents were sensitive to metronidazole, while gentamycin had a low coverage against anaerobic gram negative organisms 1 (33.5%), but good coverage over gram positive organisms.
- ❖ The finding is similar to the work carried out by Rotimi et al,¹⁹ in which *Bacteroides fragilis* (42%) was the commonest anaerobic organism cultured, with metronidazole and gentamycin being the most effective therapies.

Discussion III

- ❖ The relatively low prevalence of anaerobic bacteria reported from this study (34.4%) may be due to the high incidence of the tubotympanic (central) type of perforation recorded (89.9%), as anaerobes are often associated with cholesteatoma and granulations which are commoner with the attic-antral (marginal) perforations.
- ❖ This finding is corroborated by the works of Ibekwe et al (Enugu)²⁰, Maji et al(India)²¹ and Srivastava et al (Nepal),¹² whose researches mainly involved patients with tubotympanic disease

Discussion IV

- ❖ The relatively low anaerobic organisms isolated, 11 (34.4%) of the 32 isolates from the 11 patients might have been the duration of incubation of 48 hours used in this study.
- ❖ Other studies in which larger anaerobic yields were reported, such as Indonesia,²² Finland²³ and Sweden,²⁴ used periods of incubation which ranged from 48 hours to 2 weeks (14 days).
- ❖ Thus longer duration of incubation might have resulted in a larger amount of anaerobic organisms being isolated.

Discussion V

- ❖ It was noted from the study that the anaerobic organisms appeared to occur in the extremes of ages studied (< 10 years, 72.8% and 40 – 50 years, 27.2%)
- ❖ Hence the immunity of the patients might be a factor which contributed in its aetiology.
- ❖ While the work of Brook²² was mainly amongst a paediatric population (average age of 12 years and 6 months), Lars et al²⁴ did not make any such age distinction.

Discussion VI

- ❖ The presence of co-morbidities which include malnutrition, diabetes mellitus and retroviral infection (Human Immunodeficiency Virus, HIV) were noted in 5 of the 11 patients with anaerobic bacteria (group A), as opposed to the 13 in the other 93 patients studied (group B), who had documented co-morbidities.
- ❖ Proportion in group A was higher ($p < 0.05$) than in group B.
- ❖ The power of the test was determined to be about 70%.
- ❖ The presence of these immunocompromised states might have played a part in the chronicity of their condition especially malnutrition, a finding corroborated by the study carried out by Lasisi et al.⁵

Discussion VII

- ❖ In a longitudinal review work done by Bernaldez²⁵ (Buenos Aires, Argentina), which was to determine the clinical features of HIV positive children with CSOM done over a 10 year period 1988 - 1998, the prevalence amongst them was 13.24% and severe immunosuppression was reported to be statistically significant.
- ❖ The study of Taipale et al in Luanda, Angola, revealed that paediatrics patients with background HIV and superimposed CSOM had a 64% disease rate as opposed to the controls which were 0% ($p < 0.0001$), suggesting that persistent otorrhea is strongly associated with immunocompromised states.²⁶

Conclusion

- The study reported a female preponderance with a significant percentage of patients (45.5%) being from a low socioeconomic class.
- The study also reported a strong association between patients with anaerobic bacteria cultured in their middle ear aspirates and presence of co-morbidities, power of which was about 70%.
- There is an association of this group of patients with extreme of ages.
- Hence, immunocompromised status and extremes of age of the patients played a key role to play in the patients that had anaerobic bacteria isolated from their middle ear.

Conclusion I

- ❖ However, given the small sample size of the patients in the study and the hospital setting of the study, there might be a need to carry out a larger, community-based study that might actually give more insight into some of the peculiar findings reported in this study.

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