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Original Article **Open Access**

Prevalence and antibiotic susceptibility of acute bacterial meningitis isolates among children with fever and convulsion in Federal Medical Centre, Bida, Nigeria

[1]Oyeleye, A. E., [1]Afeez, B. O., [1]Ayanbeku, T. S., and *[2]Ubesie, A. C.

[1]Department of Paediatrics, Federal Medical Centre, Bida, Niger State, Nigeria
[2]Department of Paediatrics, College of Medicine, University of Nigeria, Enugu State, Nigeria
*Correspondence to: agozie.ubesie@unn.edu.ng

Abstract:

Background: Acute bacterial meningitis (ABM) is a life-threatening illness associated with high morbidity and mortality as well as debilitating consequences among survivors. The objective of this study is to determine the prevalence of bacterial isolates and antibiotic susceptibility pattern of ABM among children presenting with fever and convulsion at the Federal Medical Centre, Bida, Nigeria.
Methodology: This was a descriptive cross-sectional study of children who presented with fever and convulsion at the Federal Medical Centre, Bida, Nigeria. The study participants were consecutively enrolled between December 2022 and November 2023. Ethical approval was obtained from the Health Research Ethics Committee (HREC) of the hospital, and informed consent from caregivers of the children was obtained. Demographic and clinical data of the participants were collected using a proforma. Samples of cerebrospinal fluid (CSF) were obtained by lumbar puncture for chemistry, microscopy and culture, while blood samples were obtained by venipuncture for random blood glucose. Data were analysed using IBM SPSS version 23. Associations between variables were determined using Chi-square (χ^2) or Fisher Exact test and $p<0.05$ was considered significant.
Results: Of the total 110 children aged 1 month - 14 years recruited, 69 (62.7%) were males, giving a male to female ratio of 1:0.6, and 60 (54.5%) were between age 1-5 years. Seven (6.4%) participants had acute bacterial meningitis (ABM), 5 (71.4%) of which were caused by *Streptococcus pneumoniae*. Age and vaccination status were significantly associated with ABM, with significantly higher prevalence of ABM in children 1 month-1 year old (20.8%) compared to children 1-<5 years old (1.7%), children 5-<10 years old (5.0%) and children 10-14 years old (0%) (p=0.011), and significantly higher prevalence in unvaccinated children (24.9%) compared to vaccinated children (1.2%) (p=0.000). All the *S. pneumoniae* isolates were susceptible to cefotaxime and rifampicin, while 80.0% were susceptible to ceftriaxone, penicillin, meropenem, clindamycin and vancomycin.
Conclusion: The prevalent cause of post-neonatal ABM in Bida from this current study is *S. pneumoniae*, which are sensitive to most commonly available antibiotics in this environment, especially the third generation cephalosporins.

Keywords: Acute bacterial meningitis, children, prevalence, antibiotics, susceptibility

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Prévalence et sensibilité aux antibiotiques des isolats de méningite bactérienne aiguë chez les enfants présentant de la fièvre et des convulsions au Centre Médical Fédéral de Bida, au Nigéria

[1]Oyeleye, A. E., [1]Afeez, B. O., [1]Ayanbeku, T. S., et *[2]Ubesie, A. C.

[1]Département de Pédiatrie, Centre Médical Fédéral de Bida, État du Niger, Nigéria
[2]Département de Pédiatrie, Faculté de Médecine, Université du Nigéria, État d'Enugu, Nigéria
*Correspondance : agozie.ubesie@unn.edu.ng

Résumé:

Contexte: La méningite bactérienne aiguë (MBA) est une maladie potentiellement mortelle associée à une morbidité et une mortalité élevées, ainsi qu'à des conséquences invalidantes chez les survivants. L'objectif de cette étude est de déterminer la prévalence des isolats bactériens et le profil de sensibilité aux antibiotiques de la MBA chez les enfants présentant de la fièvre et des convulsions au Centre Médical Fédéral de Bida, au Nigéria.
Méthodologie: Il s'agissait d'une étude transversale descriptive portant sur des enfants présentant de la fièvre et des convulsions au Centre Médical Fédéral de Bida, au Nigéria. Les participants à l'étude ont été recrutés

consécutivement entre décembre 2022 et novembre 2023. L'approbation éthique a été obtenue auprès du Comité d'éthique de la recherche en santé (HREC) de l'hôpital, et le consentement éclairé des soignants des enfants a été obtenu. Les données démographiques et cliniques des participants ont été recueillies à l'aide d'un formulaire. Des échantillons de liquide céphalorachidien (LCR) ont été obtenus par ponction lombaire pour la chimie, la microscopie et la culture, tandis que des échantillons de sang ont été obtenus par ponction veineuse pour une glycémie aléatoire. Les données ont été analysées à l'aide d'IBM SPSS version 23. Les associations entre les variables ont été déterminées à l'aide du test du Chi carré (χ^2) ou du test exact de Fisher et une valeur de $p<0,05$ a été considérée comme significative.
Résultats: Sur un total de 110 enfants âgés de 1 mois - 14 ans recrutés, 69 (62,7%) étaient de sexe masculin, soit un ratio homme/femme de 1:0,6, et 60 (54,5%) étaient âgés de 1-5 ans. Sept (6,4%) participants souffraient de méningite bactérienne aiguë (MBA), dont 5 (71,4%) étaient causées par *Streptococcus pneumoniae*. L'âge et le statut vaccinal étaient significativement associés à la BMA, avec une prévalence significativement plus élevée de la BMA chez les enfants de 1 mois - 1 an (20,8%) par rapport aux enfants de 1 -< 5 ans (1,7%), aux enfants de 5 -< 10 ans (5,0%) et aux enfants de 10 - 14 ans (0%) (p=0,011), et une prévalence significativement plus élevée chez les enfants non vaccinés (24,9%) par rapport aux enfants vaccinés (1,2%) (p=0,000). Tous les isolats de *S. pneumoniae* étaient sensibles à la céfotaxime et à la rifampicine, tandis que 80.0% étaient sensibles à la ceftriaxone, à la pénicilline, au méropénème, à la clindamycine et à la vancomycine.
Conclusion: La cause prévalente d'ABM post-néonatale à Bida dans cette étude actuelle est *S. pneumoniae*, qui est sensible à la plupart des antibiotiques couramment disponibles dans cet environnement, en particulier les céphalosporines de troisième génération.

Mots-clés: méningite bactérienne aiguë, enfants, prévalence, antibiotiques, sensibilité

Introduction:

Acute bacterial meningitis (ABM) is a life-threatening infection associated with high mortality and morbidity globally, especially among neonates and children under 5 years of age (1). The disease can occur sporadically or in epidemics. Africa bears a large portion of the burden with large part of sub-Saharan Africa (SSA) being referred to as the meningitis belt (1). Nigeria has her 19 northern States and the Federal Capital Territory (FCT) situated within the meningitis belt (2). The prevalence rates in the worst hit areas of Nigeria have ranged between 11.2% and 20.4% with mortalities as high as 7.6% to 11% (1,3–6). Meningitis epidemics in Nigeria, usually occur during the dry, hot and dusty season, from January to March and disappears when the rainy season begins (7). While many cases end fatally, survivors have suffered severe sequelae such as hearing loss, visual impairment, limb loss, and cognitive impairment, with lasting effects on individuals and their families (8, 9).

Globally*, Haemophilus influenzae* type b, *Neisseria meningitidis* and *Streptococcus pneumoniae* are the predominant agents of ABM in post-neonatal children under 5 years of age (10,11,12). Unlike the developed countries where group B streptococcus (GBS), *Listeria monocytogenes* and *Escherichia coli* are implicated in neonatal bacterial meningitis (13,14), agents reported in Nigerian neonates include *E. coli*, *Staphylococcus aureus*, other Gram negatives and *H. influenzae* (15). Odedina and Emumwen (16) in Bida, Nigeria evaluated the CSF samples of children aged 0 to 12 years for causative organisms of acute bacterial meningitis as well as clinical indicators of the disease over a year period (January to December 2001). The authors reported a prevalence of 16.7%. The major isolates in that study were *N. meningiditis* (50.0%)*, E. coli* (26.9%), and *S. pneumoniae* (15.4%). All the isolates were susceptible to chloramphenicol and ofloxacin.

Over the years, the epidemiology and aetiology of bacterial meningitis have been changing globally especially with the introduction of vaccines such as *H. influenzae* type B (Hib), meningococcal A (MEN A) and pneumococcal vaccines (PCV) (12,17). The study by Odedina and Emumwen (16) was done before the introduction of these vaccines to Bida which makes the current study compelling. Furthermore, Iregbu and Abdullahi (18) in a study at Abuja in 2015, identified *S. aureus* (32.2%) and *Klebsiella pneumoniae* (21.5%) as the most common causes of post-neonatal ABM with *S. pneumoniae* (17.6%) a distant third.

If the World Health Organisation (WHO) 2030 global goal of defeating meningitis will be attained, it is imperative to understand the current burden of ABM in Bida, the predominant aetiological agents and their antibiotic sensitivity pattern (7,10) This study thus sought to determine the prevalence and the antibiotic susceptibility of ABM isolates among post-neonatal children presenting with fever and convulsion in Bida, northcentral Nigeria.

Materials and method:

Study setting and design:

This was a descriptive cross-sectional study carried out between December 2022 and November 2023 at the Emergency Paediatric Unit of the Federal Medical Centre Bida, (FMCB), Niger State in the northcentral region of Nigeria.

Ethical approval was obtained from the Health Research Ethics Committee of Federal Medical Centre, Bida (FMCB/HCS/HR EC/APPR/15/2020).

Sample size, sampling and data collection:

The sample size formula for population proportion was used for the sample size calculation. A minimum sample size of 110 was calculated using a prevalence rate of 6.9% from a previous study in Zaria (6) and adjusting for non-response (20–22). Sampling was consecutive with infants and children aged 1 month to 14 years, who presented with fever and convulsion; whose parent/guardian gave informed consent and children above 7 years who gave assent, were recruited consecutively throughout the study.

Children with bleeding diathesis, infection/ulcer around lumbar spine, spinal defects and evidence of raised intracranial pressure (ICP) other than bulging fontanelle were excluded from the study. Information on participants' age, gender ethnic background and use of antibiotics prior to presentation were collected using a study proforma.

CSF sample collection and microscopy

Lumbar puncture was performed following strict aseptic procedures on each participant to obtain 1 ml each of cerebrospinal fluid (CSF) into sterile universal bottle and a fluoride oxalate bottle. The CSF samples were processed and analysed within an hour after collection, to ensure speed, accuracy and appropriate yield (15,23).

CSF samples were centrifuged at 1000 x g for 10 to 15 minutes after which a drop of the sediment was smeared on a glass slide. Each stained smear was examined under a compound light microscope (100x magnification) for bacteria and reported as Gram positive or Gram-negative cocci, diplococci or bacilli accordingly. The immediate CSF reports were used for treatment of participants while CSF culture results were awaited.

For CSF cell count, a drop of sediments from the centrifuged CSF sample was mixed with a drop of toluidine blue diluting fluid and charged into the improved Neubauer counting chamber. The charged Neubauer chamber was viewed under the 40x objective lens microscope. The white blood cells were counted and noted.

CSF culture for bacteria isolation:

For CSF culture, the sediments of the centrifuged CSF were inoculated on Chocolate agar, Blood agar and MacConkey agar aseptically. The inoculated Chocolate and Blood agar plates were incubated in CO_2 enriched atmosphere (using a candle Jar) at 35–37°C. After 48 hours of incubation, plates were inspected. Culture plates with growth had the bacteria colonies identified according to conventional microbiological techniques (24-26). In this study, confirmed bacterial meningitis was defined as presence of bacteria in CSF identified by Gram stain and/or by culture growth (24,27).

Antibiotic sensitivity testing:

The antibiotic susceptibility testing was done on positive cultures. The modified Kirby-Bauer disc diffusion technique was performed for *S. aureus* and *L. monocytogenes* isolates while the E-test was performed for *S. pneumoniae*. Antibiotic discs (Oxoid Ltd, Hampshire, UK) used were cefoxitin, ciprofloxacin, chloramphenicol, penicillin, clindamycin, gentamicin, vancomycin, and rifampicin for *S. aureus,* and penicillin, meropenem, ampicillin and sulphamethoxazole/trimethoprim for *L. monocytogenes.* E-test antibiotics strips (Bio-Mérieux) used were ceftriaxone, cefotaxime, chloramphenicol, penicillin, meropenem, clindamycin, vancomycin, and ciprofloxacin for *S. pneumoniae*.

Standardized inoculum (equivalent of 0.5 McFarland turbidity standard) prepared from colonies of *S. aureus* and *L. monocytegenes* were inoculated on Mueller Hinton (MH) agar containing 5% sheep blood using a sterile cotton swab. The antibiotic discs were firmly applied on the inoculated agar with a sterile forcep and incubated at 35±2°C in an atmosphere of 5% CO_2 for 24 hours. Zones of inhibition were measured with a calibrated ruler and interpreted as sensitive or resistant in line with the Clinical and Laboratory Standards Institute (CLSI) guideline (26).

For *S. pneumoniae,* standardized inoculum prepared from colonies were similarly inoculated uniformly on MH agar plate using sterile cotton swab. The E-test strips were applied on the inoculated agar using sterile forceps and the plates were incubated at 35±2°C in an atmosphere of 5% CO_2 for 24 hours. The ellipse of inhibition for each isolate against the antibiotic strips was read to determine the minimum inhibitory concentration (MICs), and the results interpreted as sensitive or resistant in line with the CLSI guideline (26).

Data analysis:

Data analysis was done using the IBM SPSS for Windows, Version 23.0 (Armonk, NY: IBM Corp) (28). Categorical variables (gender, and ethnicity) were expressed as frequencies and proportions. Age was expressed as mean. Prevalence of meningitis was expressed as a percentage of total number of participants.

The Chi-square test (χ^2) was used to determine significant associations between the prevalence of ABM and categorical variables of the participants. Fisher's Exact probability test was used where values were too small for Chi-squared test. Statistical significance was set at $p<0.05$ at 95% confidence interval.

Results:

Socio-demographics of the study participants:

The mean age of the study participants was 2.74±3.03 years. Fig 1 shows the distribution of participants by age group, with 60 (54.5%) aged 1-5 years. Sixty-nine study participants (62.7%) were males, giving a male to female ratio of 1:0.6. Ninety-five participants (86.4%) were of Nupe ethnic background as shown in Table 1.

Prevalence and bacterial agents of ABM:

Of the 110 participants, 7 had acute bacterial meningitis, giving a prevalence of 6.4% as shown in Fig 2. Six (85.7%) of the 7 participants with acute bacterial meningitis had CSF white cell counts of greater than 100 x 10^6 cells/litre. The CSF samples of all the 7 participants with acute bacterial meningitis stained positive on Gram staining (Table 2).

Of the bacterial isolates, *S. pneumoniae* accounted for 5 (71.4%) of the 7 isolates while *S. aureus* and *L. monocytogenes* accounted for 2 isolates (14.3% each) (Table 2).

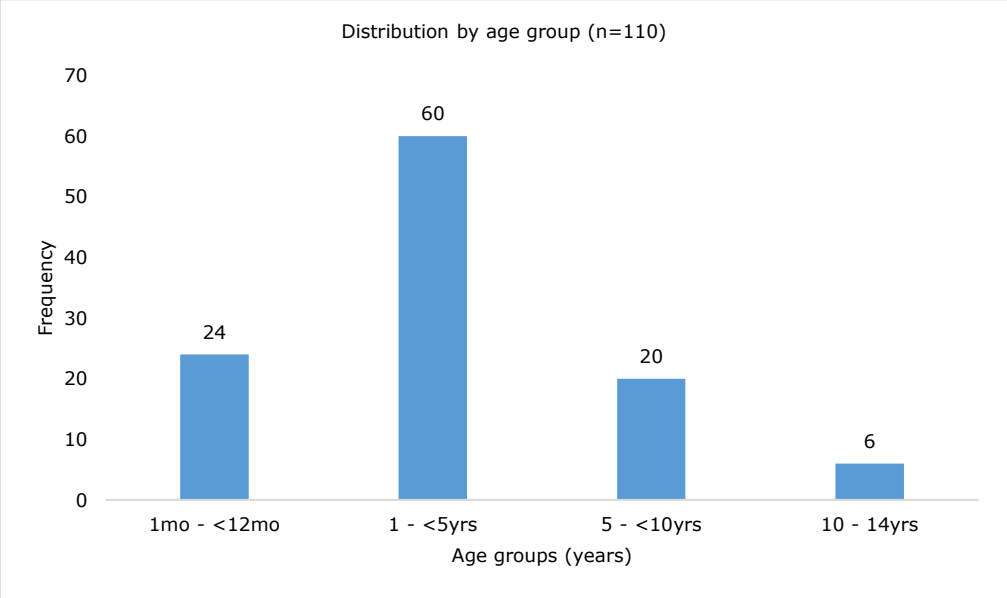

Fig 1: Distribution of the study participants by age groups

Table 1: Distribution of the study participants by gender, ethnicity and vaccination status

Variables	Frequency	Percentages (%)
Gender		
Male	69	62.7
Female	41	37.3
Total	110	100.0
Ethnicity		
Nupe	95	86.4
Hausa	5	4.5
Yoruba	4	3.6
Gbagyi	2	1.8
Fulani	2	1.8
Others	2	1.8
Total	110	100.0
Vaccination status		
Fully vaccinated	64	58.2
Incompletely vaccinated	21	19.1
Not vaccinated	25	22.7
Total	110	100.0

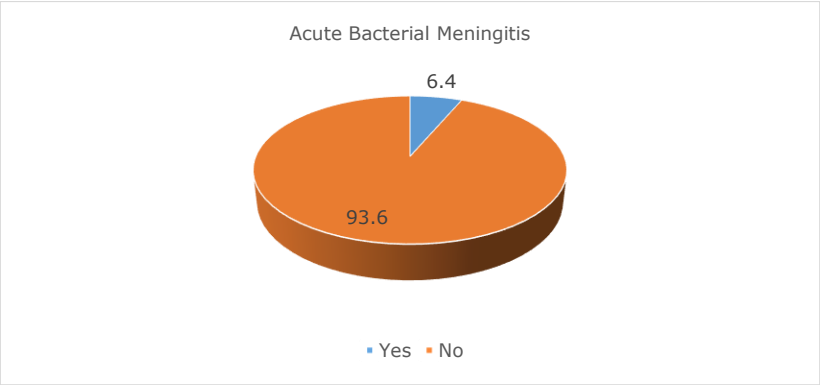

Fig 2: Prevalence of acute bacterial meningitis among the study participants

Table 2: Distribution of study participants by cerebrospinal fluid analysis findings

Variables	Bacterial Meningitis (%)	No Meningitis (%)	Total (%)	Fishers' Exact	*p*-value
White blood cells (cells/L)					
$<5x10^6$	0	103 (100.0)	103 (93.6)	110.000	0.000
5-100 $x10^6$	1 (14.3)	0	1 (0.9)		
101-500 $x10^6$	6 (85.7)	0	6 (5.5)		
Gram stain reaction					
Gram positive	7 (100.0)	0	7 (6.4)	110.000	0.000
None	0	103 (100.0)	103 (93.6)		
Growth on culture					
Growth	7 (100.0)	0	7 (6.4)	110.000	0.000
No growth	0	103 (100.0)	103 (93.6)		

Antibiotic susceptibility of isolates:

All the *S. pneumoniae* isolates were sensitive to cefotaxime and rifampicin while 4 (80%) were sensitive to ceftriaxone, penicillin, meropenem, clindamycin and vancomycin. The only *S. aureus* isolate was sensitive to cefoxitin, ciprofloxacin, penicillin, clindamycin and vancomycin but resistant to chloramphenicol, gentamicin and rifampicin. The only *L. monocytogenes* isolate was sensitive to penicillin, meropenem, ampicillin and sulphamethoxazole/trimethoprim as shown in Table 3.

Table 3: Antibiotic sensitivity pattern of bacterial isolates from the study participants

Antibiotics	***Streptococcus pneumoniae*** **n=5 (%)**	***Staphylococcus aureus*** **n=1 (%)**	***Listeria monocytogenes*** **n=1 (%)**
Ceftriaxone	4 (80.0)	NT	NT
Cefotaxime	5 (100.0)	NT	NT
Cefoxitin	NT	1 (100.0)	NT
Ciprofloxacin	NT	1 (100.0)	NT
Chloramphenicol	2 (40.0)	0	NT
Penicillin	4 (80.0)	1 (100.0)	1 (100.0)
Meropenem	4 (80.0)	NT	1 (100.0)
Clindamycin	4 (80.0)	1 (100.0)	NT
Gentamicin	NT	0	NT
Vancomycin	4 (80.0)	1 (100.0)	NT
Rifampicin	5 (100.0)	0	NT
Ampicillin	NT	NT	1 (100.0)
Sulphamethoxazole/ Trimethoprim	NT	NT	1 (100.0)

NT- not tested

Table 4: Association of acute bacterial meningitis with age, gender, and prior exposure to antibiotics

Variables	Bacterial Meningitis (n=7, 6.4%)	No Meningitis (n=103, 93.6%)	Total (n=110)	Fisher's Exact	*p*-value
Age group (years)					
1 mo-<1	5 (20.8)	19 (79.2)	24	11.125	0.011
1 - < 5	1 (1.7)	59 (98.3)	60		
5 - <10	1 (5.0)	19 (95.0)	20		
10 – 14	0	6 (100.0)	6		
Gender					
Male	3 (4.3)	66 (95.7)	69	1.262	0.261
Female	4 (9.8)	37 (90.2)	41		
Use of antibiotics					
Yes	3 (5.9)	48 (94.1)	51	0.281	0.869
No	4 (7.1)	52 (92.9)	56		
Unknown	0	3 (100.0)	3		
Vaccination status					
Vaccinated (full/partial)	1 (1.2)	84 (98.8)	85	17.490	0.000
Not vaccinated	6 (24.9)	19 (76.0)	25		

Factors associated with ABM:

Five of the 7 participants with ABM were infants while 3 of the 7 had exposure to antibiotics prior presentation. Six of the 7 with ABM had received no vaccination. Age and vaccination status were significantly associated with ABM as shown in Table 4, with significantly higher prevalence of ABM in children 1 month-1 year old (20.8%) compared to children 1-<5 years old (1.7%), children 5-<10 years old (5.0%) and children 10-14 years old (0%) (p=0.011), and also significantly higher prevalence in unvaccinated children (24.9%) compared to vaccinated children (1.2%) (p= 0.000). No significant association of gender (p=0.261) and antibiotic use prior presentation (p=0.869) with prevalence of ABM.

Discussion:

The prevalence of acute bacterial meningitis among children presenting with fever and convulsions in this study was 6.4%. This compared favorably with a prevalence of 6.9% reported by Mado et al*.,* (6) in Zaria in 2012 and 4.2% reported by Akpede et al*.,* (29) in Benin in 1992. It also compared favorably with the prevalence of 5.2% reported by Sargazi et al*.,* (30) in Iran in 2022 and 5.9% in Multan, Pakistan by Urooj et al*.,* (31) in 2021. It was however significantly lower than the prevalence of 15.2% reported by Akpede and Sykes (32) in Benin in 1993, 10.2% reported by Owusu-Ofori et al., (33) in Kumasi, Ghana in 2004, and 16.7% reported by Odedina and Emumwen in Bida (16) in 2008. The lower prevalence in this study compared to the previous Bida study by Odedina and Emumwen (16) over 20 years ago, may be explained by the introduction of vaccines against some common bacterial agents causing ABM.

Furthermore, the prevalence of ABM by age group was highest among the younger age group of 1–11 months. This finding is similar to the reports by Mado et al., (6) and Akpede et al., (29). Owusu-Ofori et al., (33) in Ghana also had majority of ABM diagnosed among those under 12 months of age with decreasing prevalence of ABM as age increased. The higher prevalence of ABM with younger age group and lower prevalence with older age group recorded in this study may affirm the role of increasing maturity of the immune system with age (34).

Streptococcus pneumoniae was the predominant cause of ABM in this study, and agreed with findings by Mado et al*.,* (6) and Johnson et al*.,* (35). *Streptococcus pneumoniae* was also the predominant isolate in a multi-centre paediatric bacterial meningitis surveillance study conducted between 2010 and 2016, involving five Nigerian (Lagos, Enugu, Bauchi, Kwara and Edo) States (10). Contrary to established knowledge of agents causing ABM in the post-neonatal age group, *N. meningitidis* and *H. influenzae* type b were not isolated in this study. Over the years, previous studies conducted in Nigeria (3,5,6, 16,29,32,35) and sub-Saharan Africa (33,36, 37) reported *S. pneumoniae , N. meningitidis* and *H. influenzae* type B as the most common causes of ABM after the neonatal age.

The introduction of robust vaccination programs against these organisms have been proffered as the reason for the declining trend. It is therefore not surprising that the current study recorded neither *H. influenzae* type B nor *N. meningitidis* as causes of ABM in Bida. The earlier study by Odedina and Emumwen (16) in Bida in 2008 that reported *N. meningitidis* and *H. influenzae* type B, was done before the introduction of these vaccines. Seventy-seven percent of the participants (n=85) in the current study received vaccines either fully or partially, through the National program on Immunization (NPI) further buttressing the significant role vaccine may be

playing in reducing the prevalence of the usual isolates.

Other bacterial agents identified as causing ABM in this study were *S. aureus* and *L. monocytogenes*, although the latter is not usually found to cause ABM in children outside the neonatal age group. Reports of *L. monocytogenes* causing meningitis among Nigerian children have been few. Alao et al., (38) in Ogbomoso reported *L. meningitidis* meningitis in a 3-year old boy. *Listeria monocytogenes* is usually associated with immunodeficiency states and it is basically food-borne (39).

In our study, *S. pneumoniae* isolates were almost entirely (80-100%) sensitive to third generation cephalosporins, ciprofloxacin, penicillin, meropenem, clindamycin, rifampicin and vancomycin but 60% of the isolates were resistant to chloramphenicol. Previously in Bida, Odedina and Emumwen (16) in 2008, reported sensitivity of *S. pneumoniae* to chloramphenicol to be 87.5%, but totally resistant to penicillin. This current study utilized the epsilon test (E-test) for antibiotic sensitivity of *S. pneumoniae* which is recommended by CLSI (26), rather than the disc diffusion method used by Odedina and Emumwen (16). The antibiotic sensitivity pattern in our study is similar to that reported by Ogeneh et al., (40) in 2015 in Enugu, where *S. pneumoniae* were totally sensitive to cephalosporins and penicillin. A possible explanation could be that cephalosporins have not been as abused due to high cost compared with the cheaper chloramphenicol.

Our study was limited by the use of only phenotypic method for bacterial detection. The use of newer molecular techniques such as polymerase chain reaction (PCR) or antigen detection test such as the latex agglutination test would have been very helpful to improve yield from cerebrospinal fluid samples especially with the use of antibiotics among some participants prior to hospital presentation.

Conclusion:

There is a shift in the aetiologic pattern of ABM among children in the post-neonatal age groups presenting with fever and convulsions in Bida, Nigeria. *Streptococcus pneumoniae* is the major causative bacterial agent. Most of the bacterial agents causing meningitis in Bida are susceptible to the third generation cephalosporins.

There should be a high index of suspicion for ABM in infants presenting with fever and convulsion. Furthermore, third generation cephalosporins, should be considered for empirical treatment in suspected cases of ABM.

Contributions of authors:

AEO, ACU, BOA and TSA conceptualized and designed the study. AEO, ACU, BOA and TSA were involved in data collection/ acquisition and statistical analysis. AEO, ACU and BOA were involved in the writing and revising the manuscript for intellectual content. All authors read and approved the final manuscript and agreed to be accountable for all aspects of the work.

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Conflict of interest:

Authors declare no conflict of interest.

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