World Journal of Advanced Research and Reviews
eISSN: 2581-9615 CODEN (USA): WJARAI
Cross Ref DOI: 10.30574/wjarr
Journal homepage: https://wjarr.com/

(Research Article)

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Prevalence of bacterial isolates of post caesarean wounds from hospitals in Otukpo, Benue state, Nigeria

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World Journal of Advanced Research and Reviews, 2024, 23(01), 1136–1146

Publication history: Received on 28 May 2024; revised on 10 July 2024; accepted on 12 July 2024

Article DOI: https://doi.org/10.30574/wjarr.2024.23.1.2049

Abstract

Sepsis after caesarean section (CS) is still a major problem in our hospitals. A large percentage of caesarean section wound infections arise from preventable causes which if addressed could significantly reduce rate of wound sepsis. The aim of this study was to identify the prevalence of bacterial pathogens associated with post caesarean wound infections and determine their relationship with some risk factors. A prospective descriptive study was conducted in randomly selected hospitals in Otukpo which included 278 women (mean age; 26.94) who underwent surgical procedure for delivery. Data were collected from patients following wound examination and compared in terms of surgical site infection and study variables (age, parity, type of CS, type of incision, duration of hospital stay and duration of Labour. Out of the 278 caesarean sections, 187(67.3%) were emergency procedures and 91 (32.7%) were elective procedures. Twenty three of the cases developed surgical site infections (SSI). *Staphylococcus aureus* (n=11; 47.83%) was the most commonly isolated organism followed by *E. coli* (n=5; 21.74). Caesarean section infection was found to be common in women who had emergency CS ($p<0.0001$), who had longer duration of Labour and hospital stay ($p<0.0001$) and had sub-umblical type of skin incision ($p<0.0001$). Development of infection following caesarean section is multifactorial and can be minimized or prevented if health care professionals develop and implement strict protocol for performing caesarean sections.

Keywords: Caesarean section; Surgical site; Infection; Prevalence; Frequency; Bacteria; Wounds

1. Introduction

Caesarean section (CS) operation is common management of delivery which can be done either as emergency or elective. There are two priorities that must be achieved; the baby and the mother, including good wound care [1].

For every skin incision, the skin barrier is disrupted and microbial contamination becomes inevitable despite the best skin preparation [2]

Infections caused by an invasive surgical procedure that occurs in the wound are commonly referred to as surgical site infections (SSIs). It is clinically characterized as an infection that occurs within 30 days of surgery (or within a year if an implant is left in place after the procedure) and affects either the incision or deep tissue at the site of the surgery [3]. These infections can be superficial or deep incisional infections, or infections affecting organs or body spaces. SSIs are the most common infections associated with health care settings. They are associated with significant morbidity and over one-third of postoperative deaths have been reported to be linked to SSI. SSI will double the duration of a patient's hospital stay and therefore increase the cost of health care [3]. Depending on the type of surgery and the severity of the infection, extra costs due to SSI have been recorded.

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Contamination of wound site and pathogenicity of microorganisms, balanced against the host's immune response can determine the occurrence of SSI [4]. The organism which causes SSI are usually derived from the endogenous environment, that is the patient skin or opened viscus. Surgical instrument or theatre environments which contaminate the site during operation can lead to exogenous causes of SSI [5]. Hematogenous spread of organisms from distant sources of infection can rarely cause SSI by attachment to the prosthesis or other implant left in the operative site. The infection prevention and control practices of SSI are therefore aimed at minimizing the number of pathogens at surgical site [6].

The most common microorganism cultured from SSIs is *Staphylococcus aureus* [5]. When a viscus, such as the large bowel, is opened, tissues are likely to be contaminated by numerous organisms. For example, *Enterobacteriaceae* and anaerobes can cause SSI after colorectal surgery [4]. The presence of a foreign body from prosthetic surgery reduces the number of pathogenic organisms required to cause SSI [3]. Microorganisms, which are non-pathogenic such as *Staphylococcus epidermidis*, may also cause SSI in such environment. The type of wound also dictates the presence of microorganisms at surgical sites. For instance, operations on sterile sites have less than 2%, whereas, SSI will occur more than 10% after operations in "contaminated" or "dirty" sites [7].

These infections can involve apparent or deep tissue infections, organ or cavity infections. Surgical site infections are the severe issues in the hospital population problems for patients undergoing surgeries, with the third most commonly identified hospital-acquired infection. [8].

Pathogens that infect surgical wounds can be part of the patient's normal flora (endogenous source) or acquired from hospital environment (nosocomial infections) or other infected patients (exogenous source). Advances in control of infections have not completely eliminated the problem because of development of resistance [9].

Antimicrobial resistance can increase complications and costs associated with procedures and treatment. The extensive uses of antibiotics, together with the length of time over which they have been available, have led to major problems of resistant organisms, contributing to morbidity and mortality. Identification of microorganisms recovered from clinical specimen is beneficial to the patient and assists in selection of chemotherapy [10]. Wound care is an expensive area of treatment for health care services [11]. Whenever a person has a caesarean section there is a risk that there will be a problem with wound healing. The most common type of wound healing problem is separation, opening of the skin and fatty tissue just beneath the skin [12].

Although appropriate use of caesarean section (CS) greatly reduces perinatal morbidity and mortality, associated complications, especially infection, have been correlated with the procedure thereby impacting negatively on the finances of surgical patients due to cost of treatments in prolonged hospital stay [13]. SSIs are the most common and most costly of all hospital acquired infections [14].

Globally, surgical site infections are the most reported healthcare-associated infection and common surgical complication [15]. In developing countries, there is a scantiness of published reports on the microbiologic profile and resistance patterns of isolates [9].

Resistance pattern of SSI-associated bacteria vary globally, depending on the region, local epidemiology reports, and susceptibility testing methodology [16].

This study gives an estimate of the SSI rate in Otukpo following both labouring and non-laboring Caesarean sections. It gives evidence that independent risk factors for SSI differ depending upon what diagnostic and procedure codes are included in the SSI definition. The study discusses the methods that are used in the isolation and identification of pathogens associated in CS wounds. It begins by discussing study design, study population, sample size, ethical considerations, and the database used. Next the outcome and exploratory variables that were examined are discussed followed by how each of the objectives was analyzed. This is followed by the results of each objective and then a discussion of these results.

Findings from a study conducted in Calabar General Hospital, Cross river state Nigeria by [17] showed that Cesarean section rate ranged from 3.85% in 2009 to 7.38% in2013 with an average cesarean section rate of 5.39%. Based on available data, elective cesarean section progressed substantially from 17.2% in2009 and slightly declined to 15.3% in 2010, then increased again from 22.5% in 2011, 29.6% in 2012 to 31.0% in 2013. Emergency cesarean section on the other hand increased progressively from 82.8% in 2009 to 84.7% in 2010 then declined from 77.5% in 2011, 70.4% in 2012 to 69.0% in 2013.

From a study conducted in the Benue state teaching hospital, Makurdi, a tertiary hospital between September 2002 and August 2014, the rate of caesarean section was 19.3% and the commonest indication for CS was cephalopelvic disproportion (28%), followed by previous CS (14-4%) and fetal distress (12.3%). [18].

The causes of caesarean wound infection are globally similar, however the relative contribution differ from region to the region and even from center to center [19]. Many risk factors have been documented as correlating with post caesarean wound infection such as duration of labor prior to the caesarean section, prolonged rupture of membranes, post operation anaemia, competence of the surgeon, duration of operation and multiple vaginal examinations [20].

Factors that promote SSIs include length of hospital stay, obesity, diabetes mellitus, smoking, etc. The development of a postoperative wound infection depends on the complex interplay of many factors, [4]. For most postoperative wounds, the source of infection is endogenous. Exogenous infections are mainly acquired from the nose or skin flora of the operating team and transmitted through the hands of the surgeon or improper operation theater sterilization, which includes preoperative, intraoperative, and postoperative care [19].

Pathogens that infect surgical wounds can be part of the patient's normal flora (endogenous source) or acquired from hospital environment (nosocomial infections) or other infected patients (exogenous source). Common offending organisms include *Staphylococcus* species*, Enterococcus faecalis, Escherichia coli, Proteus mirabilis,* and *Pseudomonas* species. *Staphylococcus aureus* is the most commonly isolated bacteria in wound infections following cesarean section [21]. These organisms cause serious infections and have shown resistance to commonly available and affordable antibiotics like the penicillin [22].

In as much as some scholars reported *S. aureus* as the single organism most frequently isolated organism [23], others observed more Gram negative organism like *E.coli*, *Proteus mirabilis, Pseudomonas* spp*.* and *Klebsiella* spp. in surgical site infection [24], [25].

[26] reported that there were 42% *Staphylococcus aureus*, 27.7% *Escherichia coli*, 20.5% *Klebsiella* spp., 5.3% Pseudomonas spp., *Enterococcus* spp. and 1.8% anaerobes from a study in a Jordanian hospital.

Staphylococcus aureus is a gram-positive coccal bacterium that is a member of the Firm cutes, and is frequently found in the nose, respiratory tract, and on the skin. It is often positive for catalase and nitrate reduction and is a facultative aerobe that can grow without the need for oxygen. Although *S. aureus* is not always pathogenic, it is a common cause of skin infections such as abscesses, respiratory infections such as sinusitis, and food poisoning. Pathogenic strains often promote infections by producing potent protein toxins, and expressing cell-surface proteins that bind and inactivate antibodies. The emergence of antibiotic-resistant strains of *S. aureus* such as methicillin-resistant *S. aureus* (MRSA) is a worldwide problem in clinical medicine [27].

Klebsiella pneumoniae is a Gram-negative, non-motile, encapsulated, lactose-fermenting, facultative anaerobic, rod-shaped bacterium. It appears as a mucoid lactose fermenter on MacConkey agar. Although found in the normal flora of the mouth, skin, and intestines, it can cause destructive changes to human and animal lungs if aspirated (inhaled), specifically to the alveoli (in the lungs) resulting in bloody sputum. In recent years, *Klebsiella* species have become important pathogens in nosocomial infections.

Proteus mirabilis is a gram negative, facultative anaerobic, rod shaped bacterium which shows swarming motility and urease activity. It shows resistance to first generation cephalosporins and ampicillin [27].

In general, effective wound management depends on understanding a number of different factors such as the type of wound being treated, the healing process, patient conditions in terms of health (e.g. diabetes), environment and social setting, and the physical chemical properties of the available dressings [5].

In order to control and prevent post caesarean wound infection in our locality, there is the need to monitor the prevailing type of bacterial pathogens and antibacterial resistance testing with a view to using appropriate antibiotic for prophylactic purposes [5]. The Centers for Disease Control and Prevention (CDC) and the American College of Obstetricians and Gynecologists agree that the percentage of CSs should be reduced, particularly in low-risk women, because it has been widely shown that, unless otherwise indicated and well documented, spontaneous vaginal delivery is safer for the both the mother and child alike [28].

[29] Have highlighted the importance of adequate patient information type as well as a number of other key interventions as a means of reducing the risk of surgical site infections. Proper hand washing prior to procedures and

pre-operatively is important in decreasing the risk of infection despite use of gloves. Glove puncture occurs in as many as 60% of all surgical procedures [30].

2. Material and methods

2.1. Sample Size and Selection Criteria

A total of 278 samples using creative research system sample size calculator were collected from patients including those that developed surgical site wound infection and those without clinical manifestation of wound infection following caesarean section from randomly selected hospitals in Otukpo town. Patients were enrolled after obtaining informed consent from them. Those that developed infection after hospital discharge were not included in the study.

2.2. Ethical Considerations

Ethical clearance was obtained from the Benue State Ministry of Health and Human Services before commencement of the study.

2.3. Sample Collection

Swab samples were collected following aseptic techniques as described by (28) as given below:

- Sterile normal saline was applied to moisten the head of the swab to increase the adherence of bacteria.
- The swab was rubbed over the surgical wound area in a zigzag motion while twisting the swab so that the entire head of the swab comes into contact with the wound surface.
- The swab was moved from the centre of the wound outward to the edge of the wound while being pressed firmly enough so that fluid is expressed from the wound tissue.
- The swabs were inserted back into its sterile case, covered, labeled with patients' information and taken to Obivine laboratory Otukpo, for analysis.
- Information such as culture result and antibiotic susceptibility pattern were documented.

2.4. Bacteriological Test

This was carried out to verify the morphology of the pathogens. Procedure was adopted from [31] as follows:

- The samples were inoculated onto blood and MacConkey agars by rubbing the swabs on an edge of the media and forming a pool or primary inoculum.
- A sterile wire loop was used to streak out the bacteria, drawing parallel lines on the media. MacConkey and blood agars were incubated aerobically at 370C for 24-48 hours.

2.5. Isolation of Bacteria

At the end of the incubation period, the inoculated plates were examined for characteristics such as colonial morphology, colour and growth pattern.

A discrete colony of each bacterial species was re-inoculated into fresh MacConkey and blood agars for isolation of pure cultures of bacteria species for identification.

2.6. Biochemical Tests

This was carried out to further confirm and identify the presence of individual bacterial pathogens.

The reaction of the bacteria species to gram stain was used to classify the bacterial species into Gram -positive and Gram -negative bacteria due to their staining reactions.

2.7. Catalase Test

This is to differentiate *Staphylococcus* from *Streptococcus*.

Procedure of catalase test

- A drop of 3% H_2O_2 was placed on a slide and a colony of bacteria on Blood agar was transferred onto it with a sterile wire loop and mixed.
- A positive result is the rapid evolution of oxygen gas evidenced by bubbling was considered to be *Staphylococci* [31].

The *Staphylococcus* was further subjected to coagulase test

2.8. Coagulase Test

This was done to differentiate *S. aureus* from other *Staphylococci.*

- A drop of distilled water was placed on two separate slides
- A colony of the test organism was emulsified in each of the drops to make two thick suspensions.
- A loopful of plasma was added to one of the suspensions and mixed gently.
- Positive result shown by formation of clot/clumps within 10 seconds confirms *Staphylococcus aureus* [31].

2.9. Oxidase Test

- Filter paper soaked with the substrate tetramethyl-p phenylenediaminedihydrochloride was moistened with sterile distilled water. A colony of the bacteria was picked using glass rod and smeared on the filter paper using a wire loop.
- A change in colour in the inoculated area to deep blue-purple after 10-20 seconds indicated the presence of oxidase - positive organism.

2.10. Indole Test

This was done to differentiate gram-negative rods particularly *E. coli* from others. It was achieved as follows:

- The test organism was inoculated in a bijou bottle containing 3 ml of sterile tryptone water.
- It was incubated at 35–37°C for up to 48 h.
- Test for indole was done by adding 0.5 ml of Kovac's reagent and shaken gently. This was examined for a red colour in the surface layer within 10 minutes.

2.11. Citrate method using Simmon's citrate agar

- Slopes of the medium was prepared in bijou bottles as recommended by the manufacturer and stored at 2–8°C.
- Using a sterile straight wire, the slope was streaked with a light inoculum picked from the center of a well isolated colony of the test organism and then stabbed at the butt.
- This was incubated at 35°C for 48 hours.
- A bright blue colour in the medium shows a positive result.

2.12. Urease test

Testing for urease enzyme activity was done to differentiate enterobacteria especially *Proteus spp.*

- Test organism was inoculated heavily in a bijou bottle containing 3 ml sterile Christensen's modified urea broth
- This was incubated at 37°C for 3–12hrs and examined for the development of a pink colour in the medium.

2.13. Antimicrobial Susceptibility Test

- Kirby-Bauer disc diffusion susceptibility technique as documented by [32] was adopted for the susceptibility assay.
- Each of the various isolates obtained were used for this assay.
- In this technique, a well dried nutrient agar plate was seeded with appropriate inoculum.
- Filter paper discs impregnated with various antibiotics were placed at specific locations on the seeded agar plate.
- The plates were incubated at 37 °C for 18 hours after which sensitivity to antimicrobial agents was measured as zones of inhibition, in millimeter around the antibiotic discs.

2.14. Questionnaire Administration

A structured questionnaire was administered for each participant. Data were collected on demographics of participants such as age and parity. The questionnaire was also designed to capture some clinical characteristics of the patients such as the indication for CS, type of CS, duration of hospital stay, type of incision and duration of labour. The questionnaire was administered to the medical personnel in the hospital.

2.15. Statistical Analysis

The data obtained were analyzed using IBM SPSS ver.20. Categorical variables were compared using chi-square test. P values ≤ 0.05 was considered significant

3. Results

3.1. Prevalence of Post Caesarean Wound Infections in Patients

Table 1 shows the prevalence of post caesarean wound infections in patients. From the 278 patients examined, 23(8.27) wounds were infected.

Table 1 Prevalence of Post Caesarean Wound Infections

Variables	Frequency	Percentage
Non infected	255	91.73
Infected	23	8.27
Total	278	100

3.2. Frequencies of occurrence of bacteria isolates from post caesarean wounds of patients.

Table 2 shows the frequencies of occurrence of bacterial isolates from the 23 cases with surgical site infection. *Staphylococcus aureus* had the highest frequency of occurrence of 11(47.83%) followed by *Escherichia coli*, 5 (21.74%) while *Pseudomonas aeruginosa*1 (4.35%) was the least isolated.

Table 2 Frequencies of occurrence of Bacterial Isolates from wounds of Post caesarean patients

Bacterial Isolates	Frequency of occurrence (%)
Staphylococcus aureus	11 (47.83)
Escherichia coli	5 (21.74)
Proteus spp	3 (13.04)
Klebsiella pneumonia	3 (13.04)
Pseudomonas aeruginosa	1 (4.35)

3.3. Prevalence of Bacterial Isolates in Relation to Age

Table 3 shows the level of bacterial isolates in respect to age. The highest prevalence 21.05 was recorded within age group 15-19 years. Low prevalence of 5.81 and 3.70 was seen amongst age groups 25-29 years and 30-34 years respectively. There was no statistical significance in the prevalence of post caesarean section wound infection in respect to age.

Table 3 Age Distribution of Prevalence of Bacterial Isolates from Wounds of Post Caesarean Patients

Age	No examined	No infected	Prevalence
15-19	38	8	21.05
20-24	62	5	8.06
25-29	86	5	5.81
30-34	54	2	3.70
35-40	38	3	7.89
Total	278	23	8.27

3.4. Prevalence of Bacterial Isolates in Relation to Parity

The prevalence of bacterial isolates from post caesarean wounds of patients in relation to parity is presented in table 4 with patients in category 2 having a prevalence of 11.94 closely followed by those in category 1 with 11.86. The least prevalence of 4.00 was reported in those that have had more than five children. There was no significant difference ($p>0.05$) in wound infection and parity.

Table 4 Prevalence of Bacterial Isolates of Post Caesarean wounds in Relation to Parity

Parity	No examined	No infected	Prevalence
1	59	7	11.86
2	67	8	11.94
3	43	3	6.97
4	46	2	4.34
5	38	2	5.26
>5	25	1	4.00
Total	278	23	8.27

3.5. Prevalence of Bacterial Isolates from Post Caesarean Wounds of Patients in Relation to Type of CS

The prevalence of bacterial isolates from post caesarean wounds of patients in relation to the type of CS is captured in table 5 where patients who underwent emergency CS had the highest prevalence of 10.69. This is statistically significant ($p<0.05$)

Table 5 Prevalence of Bacterial Isolates from Post Caesarean Wounds of Patients in Relation to Type of CS

Type of CS	No examined	No Infected	Prevalence
Elective	91	3	3.30
Emergency	187	20	10.69
Total	278	23	8.27

3.6. Prevalence of Bacterial Isolates in Post Caesarean Wounds of Patients in Relation to Type of Incision

Table 6 shows the prevalence of bacterial isolates from post caesarean wounds in respect to the type of incision. Bacterial isolates was more prevalent 20.45 in patients who had the subumblical type of skin incision. This was statistically significant ($p=0.000$).

Table 6 Prevalence of Bacterial Isolates from Post Caesarean Wounds of Patients in Relation to Type of Incision

Type of Incision	No examined	No infected	Prevalence
Sub-umblical	88	18	20.45
Pfanenstiel	190	5	2.63
Total	278	23	8.27

3.7. Prevalence of Post Caesarean Wound Infection in Respect to Duration of Hospital Stay

Table 7 shows the prevalence of wound infections in relation to duration of hospital stay. Patients who had stayed in the hospital for more than seven days before sampling recorded higher prevalence 23.33 compared to those patients that had stayed for the period of less than seven days 4.13. This was statistically significant (p=0.000).

Table 7 Prevalence of Bacterial Isolates from Post Caesarean Wounds of Patients in Relation to Duration of Hospital Stay

Duration of hospital stay	No examined	No Infected	Prevalence
<=7days	218	9	4.13
>7days	60	14	23.33
Total	278	23	8.27

3.8. Prevalence of Post Caesarean Wound Infections in Respect to Duration of Labour

The prevalence of post caesarean wound infections in relation to duration of labour is shown in table 8. Higher rate of infection was recorded amongst patients who had a longer duration of labour of more than 12hours 15(16.67%) compared to those whose labour was less than 12hours 8(4.25%) before caesarean section. This was statistically significant (p=0.000).

Table 8 Prevalence of Post Caesarean Wound Infections in Relation to Duration of Labour

Duration of labour	No examined	No Infected	Prevalence
<12hours	188	8	4.25
>=12hours	90	15	16.67
Total	278	23	8.27

4. Discussion

A large percentage of caesarean section wound infections arise from preventable causes which if addressed could significantly reduce the rate of wound sepsis. This study was carried out to identify the bacterial pathogens associated with post caesarean wound infections and determine their relationship with some risk factors. A prospective descriptive study was conducted in randomly selected hospitals in Otukpo which included 278 women (mean age; 26.94) who underwent surgical procedure for delivery. From the result, twenty three of the cases developed surgical site infections. This agrees with the work of [15] who reported that surgical site infections are the most reported healthcare-associated infections and common surgical complications globally. It also tallies with the findings of [33] who reported that surgical site infections are the most common infections associated with health care settings. They are associated with significant morbidity and over one-third of postoperative deaths have been reported to be linked to SSI. The result is slightly different with the result from a study in China by [20] who reported that only few incidence rate of surgical site infections exist in China. The difference may be instigated by socio-economic variation, poor dietary behaviors, poor personal hygiene, inadequate infection control practices implemented in healthcare, such as limited hygienic practice, inadequate antibiotic prophylaxis, lack of aseptic wound care, and non-adherence with prescribed treatments in our study area. The other differences may be the sampling method of the study population and the study design. The variation may be due to patient-related characteristics and inconsistent infection prevention strategies implemented across the countries. The incongruity could be attributed to factors relating to study participants, such as the patients' own microbial flora, educational level, nutritional status, comorbidities, and sample size. From the result of this study, *Staphylococcus aureus* was the most accounted isolated organism and next to that were *Escherichia coli.* This result is partially in consonance with the report of [34] who opined that *Escherichia coli* was the most common isolate accounted for and *Staphylococcus aureus* had a fewer implication. Caesarean section infection was significantly found to be common in women who had emergency caesarean section, who had longer duration of labour and hospital stay and had sub-umblical type of skin incision. These strongly agree with the report of [19] that reported same and added that the number of women who experience postpartum infection is anticipated to rise, mainly due to the continuing increase in rates of caesarean section deliveries in recent years globally. Development of infection following caesarean section is multifactorial and can be minimized or prevented if health care professionals develop and implement strict protocol for performing caesarean sections.

5. Conclusion

Conclusively, a soaring and extensive burden from post-CS SSIs was identified, principally in low-income countries. Additional research, higher level of awareness and the development of effective prevention and management strategies are necessary to reduce post-CS SSIs. Surgical site infection (SSI) is a major contributor to postoperative morbidity and mortality in developing countries. It is important to determine the magnitude of surgical site infection and its associated risk factors before, during, and after cesarean delivery in order to design preventive strategies in Nigeria.

Recommendation

- Since a higher infection rate was associated with emergency caesarean surgery when compared to elective surgery, the group of antibiotics observed to be effective on invitro antibiotic sensitivity pattern will likely reduce the infection rate when applied as prophylaxis.
- Further studies should be extended to include cultures under anaerobic conditions to establish presence of other organisms that require such environment for growth.
- It is also very important for Proper hygiene to be observed at all times especially where wounds are concerned as the high prevalence of *S. aureus* (a normal body flora of humans) as wound contaminant could be avoided by this practice.
- Finally, there is need to develop national surveillance of antibiotic- resistant organisms.

Compliance with ethical standards

Acknowledgments

I am grateful to the ethical committee of the Benue State Ministry of Health and Human Services for their support. To Prof. G. M. Gberikon and Dr. A. Aondoackaa of the Department of Microbiology for their mentorship, I say thank you. I specially appreciate Dr. Michael Ijiko for assisting in the sample collection. I appreciate my dad, Mr Emmanuel Okpe, my husband, Mr John Danduwa for their encouragement and support.

Disclosure of conflict of interest

No conflict of interest to be disclosed.

Statement of ethical approval

Ethical approval was duly obtained from the Benue State Ministry of Health and Human Services with an issuance of an ethical clearance certificate from the ethics committee.

Statement of informed consent

All participants were informed of the details of the study before samples were collected.

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