Phenotypic detection, antimicrobial susceptibility and virulence profile of staphylococci in the pig production setting, Makurdi, Nigeria

Levi M. Mamfe, Chinedu A. Akwuobu and Emmanuel O. Ngbede*

Abstract

Livestock, particularly pigs, have increasingly been recognized as important reservoirs for zoonotic transmission of pathogenic bacteria, including staphylococci. Livestock production systems in developing countries of sub-Saharan Africa, including Nigeria, are characterized by high misuse/abuse of antimicrobials and a close association between humans and these animals, which promotes the emergence and transmission of resistant and potentially virulent bacteria. In the present study, we investigated the occurrence and characteristics (species distribution, virulence and resistance profile) of staphylococci from small-holder backyard pig farms, slaughter slabs and pig handlers in Makurdi, Nigeria. A total of 330 nasal swabs originating from 300 pigs and 30 in-contact humans were collected and processed. One hundred and thirteen samples (34.2%; 95% confidence interval (CI): 29.1–39.6) comprising 103 (34.3%; 95% CI: 29.0–40.0) and 10 (33.3%; 95% CI: 17.3–52.8%) samples from pigs and humans, respectively, were positive for staphylococci, yielding 120 isolates (pigs n = 110, humans n = 10). The 120 isolates were distributed into 15 species with Staphylococcus aureus (n = 25) followed by Staphylococcus cohnii (n = 19) and Staphylococcus sciuri (n = 14) occurring more frequently. All isolates were resistant to β-lactam (100%) antibiotics. Resistance to some critical antimicrobials, including linezolid (22%), vancomycin (19.2%), gentamicin (7.5%) and the fluoroquinolones ciprofloxacin (75.8%) and enrofloxacin (66.7%), was also observed. Majority (99.2%) of the isolates displayed a multidrug resistance phenotype with the AMP-C-CIP-E-ENR-FOX-OX-P-S-SXT-TE phenotype being predominant. Overall, 70% of the isolates expressed the methicillin resistance phenotype, out of which 20% (n = 17) were MRSA. Resistance to serum bactericidal activity and biofilm production were respectively observed in 45 (100%) and 5 (11.3%) of the coagulase-positive staphylococci. Our findings demonstrated the occurrence of a high diversity of staphylococci expressing multidrug resistance and potentially virulent phenotypes among healthy swine and pig handlers in small-scale backyard farms in North-Central Nigeria. These findings underscore the potential role of pig production settings in the emergence and dissemination of potentially virulent staphylococci and the importance of the development of antimicrobial resistance monitoring systems/implementation of control measures in developing countries. Proper hygienic practices and control of indiscriminate use and misuse of antibiotics are recommended.

INTRODUCTION

Livestock production settings represent an important hotspot for the emergence of virulent and resistant bacteria and may serve as a pool for their spread to other animals, farm workers and inhabitants of the immediate/surrounding community, and contamination of the environment [1]. The risk of acquiring these pathogens is further increased in developing countries, including Nigeria, where animals are increasingly being raised within or in close proximity to human dwellings, biosecurity practices are poor or nonexistent, and wastes from farms are discharged without treatment into the surrounding community [2, 3].

Staphylococci, although commonly regarded as a commensal of the mucosal surfaces of healthy humans and animals, are...
also important opportunistic pathogens implicated in a vast array of infections [4]. Many studies have revealed the occurrence and exchange of indistinguishable staphylococci clones, particularly methicillin-resistant *Staphylococcus aureus*, between animals and humans, suggesting zoonoses. There is, however, limited information – particularly in Nigeria – on the staphylococcal populations inhabiting the nares of pigs, how they relate to those in their handlers and the pathogenic potential of these isolates. Current knowledge on staphylococci from pigs and humans in Nigeria is largely focused on *S. aureus* and its methicillin-resistant strains (MRSA) [5–7], neglecting the role of other staphylococcal species. Although *S. aureus* is the most important pathogen among all staphylococcal species, other staphylococci have in recent times been increasingly recognized and associated with several human and animal diseases and the spread of resistant determinants, including the methicillin resistance genes [8, 9]. More significantly, methicillin-resistant staphylococci (MRS) other than *S. aureus* have been reported in food animals and animal products [10].

The North-Central States, including Benue, are among the hubs for pig production, with pigs playing an essential role in the food security and socio-cultural lives of the inhabitants of this area. Pork is among the most frequently consumed meat products in the study area and pigs are predominantly raised in backyard farms (within or in close proximity to human dwellings) rather than commercial farms. Similar to other resource-limited countries in sub-Saharan Africa, antimicrobial use and abuse is widespread in the pig production settings in this region, and the farming system characterized by poor or non-existent biosecurity practices and no established antimicrobial surveillance system. The characteristics of this predominant pig production system and its practices promote the emergence of resistant bacteria and facilitate potential transmission between humans and animals as well as contamination of the environment. However, there is a knowledge gap concerning how this production system may influence the emergence and dissemination of antimicrobial resistance (AMR) in zoonotic foodborne bacteria in the country. We hypothesize a high circulation rate for potentially virulent and antimicrobial-resistant staphylococci among pigs in this area and that the resistogram will be a reflection of the common antimicrobials used in livestock production in the study area. The purpose of the present study was to investigate staphylococcal carriage and species diversity among apparently healthy pigs and their human handlers and to further

---

**Fig. 1.** Prevalence and species distribution of staphylococci from nasal swabs of pig and human handlers in the pig production setting, Makurdi, Nigeria.
investigate the antimicrobial resistance and virulence profile of the isolates phenotypically.

**METHODS**

**Study site and design**
A cross-sectional survey was conducted between April and December 2019 across pig farms/holdings and pig slaughter slabs in Makurdi, North-Central, Nigeria, one of the major pig-producing areas in the country. Participating farms and individuals were selected based on convenience, i.e. willingness of the farm owner to allow his farm to be included and handlers/workers to participate in the study.

**Ethical approval**
The study was approved by the Research and Ethics Committee of Benue State Ministry of Health and Human Services (MOH/STA/204/VOL.1/128). Informed consent was also obtained from all human participants before their inclusion in the study.

**Sampling and sample size**
The minimum number of pigs to be sampled (sample size) was estimated to be 78 using a 95% confidence level, an expected prevalence of 5.3% [11] and desired precision of 5%. Nasal swabs were collected from at most 30 different pigs in farms with more than 30 pigs, while in farms with small herds, i.e. less than 30 pigs, all the pigs were sampled. For humans, at least one individual working with the pigs was sampled. A total of 330 nasal swabs were collected randomly from 300 pigs and 30 humans in 19 pig farms and 3 slaughter slabs. Nasal swabs were collected using moistened sterile swabs sticks, which were inserted into the nares and rolled. All the nasal swabs collected were inoculated directly into an enrichment broth [tryptone soya broth (Oxoid, UK) supplemented with 6.5% NaCl] and placed in a cold box for onwards transport to the laboratory within 2–4 h for processing.

**Isolation and identification of staphylococci**
The nasal swabs were processed for isolation and identification of staphylococci using standard bacteriological techniques. Briefly, on arrival in the laboratory all the samples (enrichment broth inoculated with the swabs on the field) were incubated at 37°C for 18–24 h followed by inoculation of a loopful of the enrichment broth onto mannitol salt agar (MSA) (Oxoid, UK) for selective isolation of staphylococci. The MSA plates were inoculated for 18–24 h and colonies with staphylococcal morphology (pink or yellow) were picked and sub-cultured to purity on nutrient agar. Presumptive staphylococcal colonies were identified using Gram staining and the catalase test and speciated using a combination of recommended biochemical tests [12], including DNase tests, slide (clumping factor) and tube coagulase tests, sensitivity to novobiocin (5 µg), ornithine decarboxylation, acetoin production, urease activity, and aerobic fermentation of mannitol, mannose, sucrose, xylose, trehalose and lactose. Previously published simplified flow charts for speciation of staphylococci based on the result of these listed tests were also used to ease the speciation for coagulase-positive and -negative isolates [12, 13] and the chart by Goyal et al. [14] for the coagulase-negative isolates.

| Antimicrobial Category | Agent             | Pigs (n=110) | Humans (n=10) | Total (n=120) |
|------------------------|-------------------|-------------|--------------|--------------|
| Aminoglycosides        | Gentamicin        | 8 (7.3)     | 1 (10)       | 9 (7.5)      |
|                        | Streptomycin      | 65 (59.1)   | 8 (80)       | 73 (60.8)    |
| Cephalosporin          | Cefoxitin         | 82 (74.5)   | 9 (90)       | 91 (75.8)    |
| Fluoroquinolones       | Ciprofloxacin     | 84 (76.4)   | 7 (70)       | 91 (75.8)    |
|                        | Enrofloxacin      | 71 (64.5)   | 9 (90)       | 80 (66.7)    |
| Glycopeptides          | Vancomycin        | 22 (20)     | 1 (10)       | 23 (19.2)    |
| Macrolides             | Erythromycin      | 97 (81.2)   | 10 (100)     | 107 (82.2)   |
| Oxazolidone            | Linezolid         | 22 (20)     | 4 (40)       | 26 (21.6)    |
| Penicillins            | Ampicillin        | 110 (100)   | 10 (100)     | 120 (100)    |
|                        | Penicillin        | 89 (80.9)   | 9 (90)       | 98 (81.7)    |
|                        | Oxacillin         | 110 (100)   | 10 (100)     | 120 (100)    |
| Phenicolos             | Chloramphenicol   | 73 (66.4)   | 9 (90)       | 82 (68.3)    |
| Folate                 | Supha-methoxazole | 60 (54.4)   | 9 (90)       | 69 (57.5)    |
| Tetracycline           | Tetracycline      | 89 (80.9)   | 9 (90)       | 98 (81.7)    |
Table 2. Distribution of methicillin-resistant staphylococci (MRS) isolated from pigs and their handlers in Makurdi, Benue State, Nigeria

| Staphylococcus species       | No. of methicillin-resistant |
|------------------------------|------------------------------|
|                              | Pigs | Humans | Total |
| S. aureus                    | 13   | 4      | 17    |
| S. intermedius               | 7    | 1      | 8     |
| S. schleiferi coagulans      | 1    | 0      | 1     |
| S. hominis                   | –    | 1      | 1     |
| S. schleiferi schleiferi     | 3    | 1      | 4     |
| S. haemolyticus              | 2    | –      | 2     |
| S. warneri                   | 3    | –      | 3     |
| S. cohnii                    | 17   | –      | 17    |
| S. epidermidis               | 2    | –      | 2     |
| S. simulans                  | 5    | –      | 5     |
| S. xylosus                   | 7    | –      | 7     |
| S. sciuri                    | 12   | –      | 12    |
| S. lungdensis                | 1    | –      | 1     |
| S. lentus                    | 3    | –      | 3     |
| S. hominis                   | –    | 1      | 1     |

only. The S. aureus species were further confirmed using the Staphytec plus latex agglutination kit (Oxoid, UK).

Antimicrobial susceptibility testing
Susceptibility of the isolates to antibiotics was assessed by the Kirby–Bauer disc diffusion method according to the Clinical and Laboratory standards Institute (CLSI) guidelines [15, 16] using Mueller–Hinton agar and commercial antimicrobial discs (Oxoid, UK). The antimicrobial agents used include: ampicillin (10µg), cefoxitin (30µg), chloramphenicol (30µg), ciprofloxacin (5µg), enrofloxacin (5µg), erythromycin (15µg), linezolid (30µg), oxacillin (1µg), penicillin (10µg), streptomycin (30µg), sulphamethoxazole–trimethoprim (25µg), tetracycline (30µg) and gentamicin (30µg). Vancomycin susceptibility was determined by the minimum inhibitory concentration using the agar dilution method. The susceptibility result was determined by measurement of the zone of inhibition and the result was interpreted as susceptible, intermediate or resistant for each antibiotic according to the CLSI guidelines for veterinary isolates [15] and human isolates [16]. Intermediate isolates were classified as resistant. Cefoxitin- or oxacillin-resistant isolates were categorized as methicillin-resistant staphylococci (MRS in general) depending on the species [15, 16]. Isolates resistant to ≥3 different classes of antimicrobials were classified as multidrug-resistant (MDR) [17].

Virulence profile
The coagulase-positive staphylococcal isolates were tested for expression of biofilm and resistance to serum bactericidal activity. Biofilm production was assayed using the Congo red dye method as previously described by Kaiser et al. [18], while resistance to serum bactericidal activity was assayed using the method described by King et al. [19].

RESULTS
Occurrence and species distribution of staphylococci
Of the 330 samples collected from pigs (n=300) and humans (n=30), staphylococci were isolated from 113 samples comprising 103 (34.3%; CI: 31.2–42.4%) from pigs and 10 (33.3% 95%CI: 17.3–52.8%) from humans. A total of 120 staphylococci were recovered from the 113 positive samples. Coagulase-negative staphylococci (CoNS) were more predominant, particularly among the pig (n=72) isolates, compared to coagulase-positive (CoPS) species (n=38). The 120 staphylococci isolated were distributed into 15 species while the species identity of 7 coagulase-negative isolates could not be completely verified, i.e. could not be assigned species (Fig. 1). S. aureus was the most common species in both pigs (n=21; 21/110) and humans (n=4; 4/10), followed by the coagulase-negative Staphylococcus cohnii (n=19; 19/110) and Staphylococcus sciuri (n=14; 14/110) in pigs, respectively (Fig. 1).

Antimicrobial susceptibility
The antimicrobial susceptibility result is presented in Table 1. All the isolates (100%) tested were resistant to oxacillin and ampicillin. Varying resistance rates ranging from as low as 7.5% for gentamicin to 82.2% for erythromycin, were recorded for the other antimicrobials. Resistance to the high priority critical antimicrobials, including vancomycin (19.2%) and linezolid (21.6%), were also recorded (Table 1). The result for each species is presented in the Tables S1–S3 (available in the online version of this article).

Multidrug resistance
Majority (99.2%) of the isolates were multidrug-resistant (resistant to ≥3 different classes of antimicrobials), with the most resistant being to all 11 antibiotic classes. One S. sciuri isolate from pigs was resistant to all 15 antimicrobial agents tested. AMP-CIP-E-ENR-FOX-OX-P-S-TE was the most predominant (n=6) phenotype among the pig isolates compared to AMP-C- CIP-E-ENR-FOX-LZD-OX-P-S-SXT-TE (n=2) and AMP-C-CIP-E-ENR-FOX-OX-P-S-SXT-TE (n=2) in the human isolates (Tables S4a–c and S5).

All the isolates had a multiple antibiotic resistance (MAR) index ≥0.2. The values of the MAR index for the Staphylococcus isolates ranged from 0.2 to 1 with a mean value of 0.65. (Tables S4a–c and S5).
Methicillin resistance

Of the 120 isolates, 84 (70%) distributed across the 15 species were methicillin-resistant staphylococci (MRS) (Table 2). Although both coagulase-positive and -negative staphylococci expressed the methicillin resistance phenotype, overall coagulase-negative staphylococci species expressed the phenotype more frequently than coagulase-positive species, particularly in pigs. The coagulase-negative S. cohnii (n=17) followed by the coagulase-positive S. aureus (MRSA) were the predominant methicillin-resistant species in pigs, while S. aureus (n=4) was the predominant one in humans.

Virulence profile

The frequency of expression of virulence traits by the coagulase-positive staphylococci species is presented in Table 3. All (45/45) of the coagulase-positive staphylococci were resistant to the bactericidal action of serum, while only 11.1% (5/45) produced biofilm.

| Staphylococcus species | Serum bactericidal resistance | Biofilm production |
|------------------------|-----------------------------|-------------------|
|                        | Pigs | Humans | Total | Pigs | Humans | Total |
| S. aureus              | 21   | 4      | 25    | 3    | 0      | 3     |
| S. intermedius         | 11   | 1      | 12    | 2    | 0      | 2     |
| S. schleiferi coagulans | 3    | 1      | 4     | 0    | 0      | 0     |
| S. hyicus              | 3    | 1      | 4     | 0    | 0      | 0     |

**DISCUSSION**

The emergence and spread of multidrug-resistant bacteria, particularly in livestock production settings, is increasingly a source of public health concern globally. Swine husbandry has been recognized as an important reservoir for the emergence and dissemination of resistant and virulent Staphylococcus species. In the present study, we observed the occurrence of diverse species of staphylococci, the majority of which were multidrug-resistant and possess the potential to cause clinical infections. These findings suggest that the pig production setting in this area could serve as an important source for the dissemination of antimicrobial-resistant and potentially virulent staphylococci. Similar species distributions have previously been reported in pigs, pork and in-contact humans in sub-Saharan Africa [10, 20–22].

All the isolates phenotypically multidrug-resistant (MDR), had an MAR index of ≥0.2, which suggests that the isolates originated from a source where antibiotics are used frequently [23]. The high rates of resistance observed towards the β-lactams, penicillins, tetracyclines and macrolides are consistent with the findings from previous studies in animals and animal products, including pigs and pork [10, 21, 22, 24], poultry [25], cattle [26], camels [27] and humans [11, 21, 25] in Nigeria. This high rate of resistance may not be coincidental, as we hypothesize that it is linked to the well-documented widespread, heavy and unregulated use of these agents in pig production across the country for prophylaxis and therapy [28]. In agreement with a previous report by Ogunleye and Okunlade [29], a high rate (68%) of resistance to chloramphenicol, a drug whose use in veterinary medicine has been banned worldwide, including in Nigeria, was observed. This observation may also be attributed to the widespread use of the drug in animal production in Nigeria in spite of the ban [28, 30–32]. This further supports the need for the roll out of an appropriate programme and enforcement of regulations on drug use and access in the country. Selection pressure from the abusive use of antimicrobials has been shown to facilitate the emergence of resistant strains [32]. The multidrug resistance phenotype expressed by the isolates is particularly worrying, as it may limit the available therapeutic armamentarium in the event of infection by these pathogens.

Although the categorization of Staphylococcus aureus as methicillin-resistant (MRSA) in the present study was purely based on the phenotypic characterization, the findings agree with other studies that reported MRSA from pigs and pig handlers in Nigeria [5–7].

Surprisingly, some of the isolates were resistant to two ‘last resort’ antibiotics, vancomycin and linezolid, which are not commonly used in livestock production or human medicine in Nigeria. There is a recent increase in the frequency of detection of linezolid- and vancomycin-resistant Gram-positive bacteria in livestock in sub-Saharan Africa. The increase in linezolid-resistant isolates may likely be a result of the recent increase in use of florfenicol in many farms across Nigeria [33]. Florfenicol has been shown to provide selective pressure for the emergence of linezolid-resistant strains [34]. Some of the genes mediating resistance to these last-resort drugs have been reported to be located/carried on mobile genetic elements, which may facilitate their rapid dissemination [35].

Notably, all the coagulase-positive species from both humans and pigs displayed resistance to serum bactericidal activity and a few produced biofilm. Biofilm formation is known to confer a fitness advantage on bacteria by enhancing their survivability and resistance to antibiotics and facilitating their ability to acquire virulence and antibiotic resistance genes during horizontal gene transmission due to their high microbial density [36].
Collectively, the MDR, resistance to serum bactericidal activity and biofilm production ability of the isolates could make them difficult to control in swine farms and, once introduced into the community and hospital environments, increase their capacity to survive and proliferate in these environments. Additionally, species with these properties could be disseminated into the environment via manure and other wastes originating from the farms and slaughter facilities.

The detection of resistant and potentially virulent staphylococci in pigs and exposed workers in the farms and slaughter facilities is of great public health significance and highlights a serious food safety threat, as these facilities, particularly the slaughter slabs, serve as the major source of pork for the community. These findings therefore further suggest that pig farms, slaughter facilities and workers in the study location may constitute a potential reservoir and source for spread of foodborne staphylococcal infections, highlighting the importance of the implementation of food safety measures and regulations concerning antimicrobial stewardship in livestock production settings in the study area.

This study, however, has some limitations, particularly the inclusion criteria for sampling, with the willingness of the farm owner or worker to participate being required, which may have introduced some bias. Further, our inability to carry out molecular characterization of the isolates limited our inferences.

CONCLUSION

This study demonstrates that the pig production settings in the study area may constitute a reservoir for the emergence and dissemination of virulent and antibiotic-resistant staphylococci. Further research using high-resolution genomics is required to understand the molecular epidemiology, likelihood of exchange or spillover from the pig population to humans. The findings from our study also suggest the need for increased antimicrobial surveillance in the swine production environment in the area to mitigate the increased emergence and dissemination of resistant strains.

References
1. Zhao Y, Yang QE, Zhou X, Wang F-H, Muurinen J, et al. Antibiotic resistome in the livestock and aquaculture industries: Status and solutions. Crit Rev Environ Sci Technol 2020;51:2159–2196.
2. Founou LL, Founou RC, Essack SY. Antibiotic resistance in the food chain: a developing country-perspective. Front Microbiol 2016;7:1881.
3. Magnusson U, Moodley A, Osbjer K. Antimicrobial resistance at the livestock-human interface: Implications for veterinary services. Rev - Off Int Epizoot 2021;40:2.
4. Parlet CP, Brown MM, Horswill AR. Commensal staphylococci influence Staphylococcus aureus skin colonization and disease. Trends Microbiol 2019;27:497–507.
5. Otalu OJ, Kwaga JKP, Okofoocha EC, Islam MZ, Moodley A. High genetic similarity of MRSA ST88 isolated from pigs and humans in Kogi State, Nigeria. Front Microbiol 2018;9:3098.
6. Okorie-Kanu OJ, Anyanwu MU, Ezenduka EV, Mgbeahuruuike AC, Thapaliya D, et al. Molecular epidemiology, genetic diversity and antimicrobial resistance of Staphylococcus aureus isolated from chicken and pig carcasses, and carcass handlers. PloS One 2020;15:e0232913.
7. Gaddafi MS, Yusuf Y, Abdulkadir J, Bashir MB, Bashiru G, et al. Nasal colonization of pigs and farm attendants by Staphylococcus aureus and methicillin-resistant Staphylococcus aureus (MRSA) in Kebbi, Northwestern Nigeria. The Thai Veterinary Medicine 2020;51:119–124.
8. Moore JE, Millar BC, Crowe M, Buchanan J, Watabe M, et al. Molecular determination of carriage of the mecA locus in coagulase negative staphylococci in screening swabs from patients in an intensive care unit. Mol Pathol 2003;56:63.
9. Otto M. Coagulase-negative staphylococci as reservoirs of genes facilitating MRSA infection: Staphylococcal commensal species such as Staphylococcus epidermidis are being recognized as important sources of genes promoting MRSA colonization and virulence. Bioessays 2013;35:4–11.
10. Igunbiso EO, Beshiru A, Akporhehe LU, Ovisagie FE, Igunbiso OO. Prevalence of methicillin-resistant Staphylococcus aureus and other Staphylococcus species in raw meat samples intended for human consumption in Benin City, Nigeria: Implications for public health. Int J Environ Res Public Health 2016;13:E949.
11. Momoh AH, Kwaga JKP, Bello M, Sackey AKB, Larsen AR. Antibiotic resistance and molecular characteristics of Staphylococcus aureus isolated from backyard-raised pigs and pig workers. Trop Anim Health Prod 2018;50:1565–1571.
12. Barrow GI, Feltham RKA. Cowan and Steel’s Manual for the Identification of Medical Bacteria. 3rd edn. Cambridge University Press; 2009. p. 331.
13. Becker K, von Eiff C. Staphylococcus, Micrococcus, and other catalase-positive cocci. In: Versalovic J, Carroll KC, Funke G, Jorgensen JH and Landry ML (eds). Manual of Clinical Microbiology. ASM press; 2011. p. 310.
14. Goyal R, Singh NP, Kumar A, Kaur I, Singh M, et al. Simple and economical method for speciation and resistotyping of clinically significant coagulase negative staphylococci. Indian J Med Microbiol 2006;24:201–204.
15. Clinical and Laboratory Standards Institute (CLSI). Performance Standards for Antimicrobial Susceptibility Testing. 4th ed. VET08-ED:2018. Wayne, Pennsylvania 19087 USA: Clinical and Laboratory Standards Institute, 950 West Valley Road, Suite 2500; 2018.
16. Clinical and Laboratory Standards Institute (CLSI). Performance Standards for Antimicrobial Susceptibility Testing. 31th ed. M100-ED30:2020. Wayne, Pennsylvania 19087 USA: Clinical and Laboratory Standards Institute, 950 West Valley Road, Suite 2500; 2020.
17. Magiorakos AP, Srinivasan A, Carey RB, Carmeli Y, Falagas ME, et al. Multidrug-resistant, extensively drug-resistant and pandrug-resistant bacteria: an international expert proposal for interim standard definitions for acquired resistance. Clinical Microbiology and Infection 2012;18:268–281.

Funding information
This work received no specific grant from any funding agency.

Author contributions
Conceptualization: L.M.M., C.A.A., E.O.N. Methodology: L.M.M., C.A.A., E.O.N. Validation: L.M.M., C.A.A., E.O.N. Formal analysis: L.M.M. Investigation: L.M.M. Resources: C.A.A., E.O.N. Data curation: L.M.M. Writing – original draft: L.M.M. and E.O.N. Writing – review and editing: L.M.M., C.A.A., E.O.N. Supervision: C.A.A. and E.O.N.

Conflicts of interest
The authors declare that there are no conflicts of interest.

Ethical statement
The study was approved by the Research and Ethics Committee of Benue State Ministry of Health and Human Services (MOH/STA/204/ VOL.1/128). Informed consent was also obtained from all human participants before their inclusion in the study.
Kaiser TDL, Pereira EM, Dos Santos KRN, Maciel ELN, Schuenck RP, et al. Modification of the Congo red agar method to detect biofilm production by *Staphylococcus epidermidis*. *Diagn Microbiol Infect Dis* 2013;75:235–239.

King LB, Swiatlo E, Swiatlo A, McDaniel LS. Serum resistance and biofilm formation in clinical isolates of Acinetobacter baumannii. *FEMS Immunol Med Microbiol* 2009;55:414–421.

Ugwu CC, Gomez-Sanz E, Agbo IC, Torres C, Chah KF. Characterization of mannitol-fermenting methicillin-resistant staphylococci isolated from pigs in Nigeria. *Braz J Microbiol* 2015;46:885–892.

Momoh AH, Kwaga JKP, Bello M, Sackey AKB. Prevalence and antimicrobial resistance pattern of coagulase negative staphylococci isolated from pigs and in-contact humans in Jos metropolis, Nigeria. *Nigerian Veterinary Journal* 2016;37:140–147.

Founou LL, Founou RC, Essack SY, Djoko CF. Mannitol-fermenting methicillin-resistant staphylococci (MRS) in pig abattoirs in Cameroon and South Africa: A serious food safety threat. *Int J Food Microbiol* 2018;285:50–60.

Davis R, Brown PD. Multiple antibiotic resistance index, fitness and virulence potential in respiratory *Pseudomonas aeruginosa* from Jamaica. *J Med Microbiol* 2016;65:261–271.

Adikwu AA, Okolocha EC, Luga II, Ngbede EO. Microbial hazards associated with pig carcasses and molecular detection of enterotoxigenic *Staphylococcus aureus* at different stages of the slaughter process. *Sokoto J Vet Sc* 2019;17:27.

Ogundipe FO, Ojo OE, Feßler AT, Hanke D, Awoyomi OJ, et al. Antimicrobial Resistance and Virulence of Methicillin-Resistant *Staphylococcus aureus* from Human, Chicken and Environmental Samples within Live Bird Markets in Three Nigerian Cities. *Antibiotics* 2020;9:588.

Mamza SA, Geidam YA, Mshelia GD, Egwu GO. Identification and Enumeration of Staphylococcus species isolated from Livestock in North-eastern Nigeria. *Global Advanced Research Journal of Microbiology* 2020;9:040–047.

Yusuf ST, Kwaga JKP, Okolocha EC, Bello M. Phenotypic occurrence of methicillin-resistant *Staphylococcus aureus* in camels slaughtered at Kano abattoir, Kano, Nigeria. *Sokoto J Vet Sc* 2017;15:29.

Adebowale OO, Adeyemo FA, Bankole N, Olasoju M, Adesokan HK, et al. Farmers’ Perceptions and drivers of antimicrobial use and abuse in commercial pig production, Ogun State, Nigeria. *Int J Environ Res Public Health* 2020;17:10.

Ogunleye AO, Okunlaide OA. Antibiotic resistance status of *Escherichia coli* isolated from health pigs from some piggery farms in Ibadan, Nigeria. *Tropical Veterinarian* 2015;33:3–4.

Schwarz S, Kehrenberg C, Doublet B, Cloeckaert A. Molecular basis of bacterial resistance to chloramphenicol and florfenicol. *FEMS Microbiol Rev* 2004;28:519–542.

Davies J, Davies D. Origins and evolution of antibiotic resistance. *Microbiol Mol Biol Rev* 2010;74:417–433.

Skalet AH, Cevallos V, Ayele B, Gebre T, Zhou Z, et al. Antibiotic selection pressure and macrolide resistance in nasopharyngeal *Streptococcus pneumoniae*: a cluster-randomized clinical trial. *PLoS Med* 2010;7:7.

Jibril AH, Okeke IN, Dalsgaard A, Olsen JE. Association between antimicrobial usage and resistance in Salmonella from poultry farms in Nigeria. *BMC Vet Res* 2021;17:234.

Freitas AR, Tedim AP, Duarte B, Elghaieb H, Abbassi MS, et al. Linezolid-resistant (Tn6246::fexB-poxtA) *Enterococcus faecium* strains colonizing humans and bovines on different continents: similarity without epidemiological link. *J Antimicrob Chemother* 2020;75:2416–2423.

Elghaieb H, Freitas AR, Abbassi MS, Novais C, Zouari M, et al. Dispersal of linezolid-resistant enterococci carrying poxtA or oprA in retail meat and food-producing animals from Tunisia. *J Antimicrob Chemother* 2019;74:2865–2869.

Donlan RM, Costerton JW. Biofilms: survival mechanisms of clinically relevant microorganisms. *Clin Microbiol Rev* 2002;15:167–193.

---

**Five reasons to publish your next article with a Microbiology Society journal**

1. The Microbiology Society is a not-for-profit organization.
2. We offer fast and rigorous peer review – average time to first decision is 4–6 weeks.
3. Our journals have a global readership with subscriptions held in research institutions around the world.
4. 80% of our authors rate our submission process as ‘excellent’ or ‘very good’.
5. Your article will be published on an interactive journal platform with advanced metrics.

Find out more and submit your article at microbiologyresearch.org.