

Prevalence, Antibiotic Susceptibility Profile, and Multiple Antibiotic Resistance Index of Gram-Negative Bacteria Isolated from Skin and Nasal Samples of Goats Bred in Abakaliki, Ebonyi State, Nigeria

Ukpai E. G.^{1,2*} and Okwuonu C. A.³

1. Department of Microbiology and Parasitology, David Umahi Federal University of Health Sciences, Uburu, Ebonyi State, Nigeria.
2. International Institute of Infectious Diseases, Biosafety and Biosecurity Research, David Umahi Federal University of Health Sciences, Uburu, Ebonyi State, Nigeria.
3. Department of Medical Microbiology, College of Medicine, Godffery Okoye University, Ugwuomu Nike Enugu, Enugu State.

* Corresponding author: ukpaieg@dufuhs.edu.ng

Abstract: Antimicrobial resistance (AMR) is a critical global public health threat, with food-producing animals recognized as important reservoirs of multidrug-resistant (MDR) bacteria. In Nigeria, indiscriminate antibiotic use and weak regulatory frameworks have exacerbated resistant pathogen spread. Goats (*Capra hircus*) are widely reared in Abakaliki, Ebonyi State, serving as major meat and income sources, yet limited data exist on the prevalence, antibiotic susceptibility profile, and multiple antibiotic resistance index (MARI) of Gram-negative bacteria colonizing their skin and nasal passages. Thirty samples (20 skin, 10 nasal swabs) were aseptically collected from goats in Abakaliki urban markets. Gram-negative bacteria were isolated using standard microbiological techniques, confirmed by VITEK® 2 COMPACT, and subjected to antibiotic susceptibility testing via Kirby-Bauer disc diffusion against 13 antibiotics. MARI was calculated for each isolate. Thirty-one Gram-negative isolates were recovered: *Escherichia coli* 12 (38.7%), *Klebsiella pneumoniae* 8 (25.8%), *Shigella flexneri* 7 (22.6%), and *Salmonella* Enteritidis 4 (12.9%). All isolates (100%) exhibited resistance to amoxicillin-clavulanic acid. High resistance rates were observed against ceftazidime, ceftriaxone, azithromycin, and levofloxacin. *Shigella flexneri* showed complete susceptibility to imipenem, while *Salmonella* Enteritidis was fully susceptible to levofloxacin. Mean MARI values ranged from 0.48 (*K. pneumoniae*) to 0.62 (*Salmonella* Enteritidis), all exceeding the 0.2 high-risk threshold. All isolates were resistant to three or more antibiotic classes and classified as MDR. Goats in Abakaliki harbor clinically significant MDR Gram-negative bacteria with high antibiotic resistance indices, highlighting zoonotic transmission risks and the urgent need for antimicrobial stewardship and One Health surveillance in Nigeria.

Key word: Antimicrobial resistance (AMR), Goats, Gram-negative bacteria, Multidrug-resistant (MDR), One Health

INTRODUCTION

The global burden of antimicrobial resistance (AMR) has emerged as one of the most pressing public health challenges of the twenty-first century, with food-producing animals increasingly recognized as critical reservoirs for multidrug-resistant (MDR) bacteria (Peter *et al.*, 2022; Nwode *et al.*, 2026a; Nwode *et al.*, 2026b). In Nigeria, the situation has become particularly alarming due to unrestricted access to and indiscriminate use of antibiotics, a lack of national AMR surveillance programmes, and inadequate antimicrobial regulation policies (Adesola *et al.*, 2025; Agwu *et al.*, 2026). These factors have created ideal conditions for the emergence and spread of resistant bacterial strains, especially among livestock

populations where antibiotics are frequently used without veterinary oversight (Peter *et al.*, 2022; Nwode *et al.*, 2026b).

Goats (*Capra hircus*) represent a significant livestock resource in Nigeria, particularly in the southeastern region, where they serve as a major source of meat, milk, and income for rural households. Abakaliki, the capital of Ebonyi State, hosts numerous markets where goats are raised, traded, and slaughtered under conditions that often facilitate bacterial transmission and zoonotic spread. The close proximity between goats and humans in these settings, coupled with inadequate hygiene practices, creates opportunities for the exchange of pathogenic bacteria, including those carrying resistance genes that compromise treatment options for infections in both animals and humans.

Gram-negative bacteria, including *Escherichia coli*, *Klebsiella* species, *Salmonella* species, and *Shigella* species, are of particular concern due to their intrinsic and acquired resistance mechanisms (Oke *et al.*, 2024; Peter *et al.*, 2025; Nwojiji *et al.*, 2025). These organisms frequently harbor mobile genetic elements such as plasmids and transposons that facilitate the horizontal transfer of resistance genes across bacterial species and between animal and human populations (Nwosu *et al.*, 2023a; Kumavath *et al.*, 2025). Previous research in Abakaliki has demonstrated that abattoirs and livestock environments can serve as major reservoirs of resistance genes, with Ejikeugwu *et al.* (2018) reporting high resistance rates among *E. coli* isolates from abattoir sources to cefoxitin (91.7%), ceftriaxone (83.3%), and imipenem (85.4%), with the detection of FOX-1 AmpC β -lactamase genes that confer resistance to broad-spectrum cephalosporins. This finding underscores the potential for livestock to act as reservoirs of clinically significant resistance determinants. Recent studies across Nigeria have documented the escalating threat of antimicrobial resistance in small ruminants. Joseph *et al.* (2025) investigated antibiotic resistance in bacteria isolated from Red Sokoto and West African Dwarf goat breeds in Nasarawa State and reported that *Salmonella* spp. (91.7%), *Pseudomonas* spp. (100%), and *Klebsiella* spp. (92.9%) showed concerning levels of susceptibility to tested antibiotics, with all four identified species exhibiting multidrug resistance and demonstrating potential for inter-species gene transfer. Similarly, Anueyiagu *et al.* (2025) reported a high prevalence of extended-spectrum beta-lactamase (ESBL)-producing coliforms in ruminants with subclinical mastitis in Plateau State, where *E. coli* was the most prevalent isolate in cows (71%), with 46 isolates expressing the clinically important ESBL genes *bla*TEM and *bla*CTX-M, highlighting

the potential zoonotic reservoir for antibiotic resistance in Nigeria.

The emergence of ESBL-producing Enterobacteriaceae in livestock settings is particularly concerning because these organisms confer resistance to critically important antibiotics such as third-generation cephalosporins and fluoroquinolones, which are mainstays of treatment for serious bacterial infections in humans (Nwosu *et al.*, 2023a; Nwosu *et al.*, 2023b; Adesola *et al.*, 2025). The detection of such resistance determinants in goats, which are often raised in close contact with humans and other animals, poses a significant public health risk, as these animals can serve as sources of resistant bacteria that may be transmitted through direct contact, slaughtering, and consumption of goat meat a dietary staple in Ebonyi State.

Despite growing evidence of AMR in Nigerian livestock (Peter *et al.*, 2022; Joseph *et al.*, 2025; Nwode *et al.*, 2026a; Nwode *et al.*, 2026b), data on Gram-negative bacteria specifically associated with goats remain limited, with most studies having focused on poultry and cattle. The nasal cavity and skin of goats serve as important ecological niches for diverse bacterial populations, including potential pathogens that can be transmitted to humans. Therefore, this study aims to investigate the prevalence, antibiotic susceptibility profile, and multiple antibiotic resistance index of Gram-negative bacteria isolated from skin and nasal samples of goats bred in Abakaliki, Ebonyi State, Nigeria. The findings will contribute to the mapping of antimicrobial resistance in the livestock sector, provide baseline data for surveillance programmes, and inform evidence-based interventions to promote prudent antimicrobial use and safeguard public health in line with the One Health approach.

MATERIALS AND METHODS

Study Area: The study was conducted in Abakaliki urban, the capital of Ebonyi State, Nigeria. Abakaliki is situated within

latitudes 5.40°N and 6.45°N and longitudes 7.30°E and 8.30°E (Adibe-Nwafor *et al.*, 2023). The area is bounded to the east by Izzi Local Government Area, to the west by Ezza North and Ezza South Local Government Areas, and to the south by Cross River and Benue States. Geopolitically, Abakaliki belongs to the southeast zone of Nigeria and lies within the Cross River plains. Farming constitutes the primary occupation of the inhabitants. Abakaliki urban hosts numerous meat markets, including Ahiaofuru, Kpirikpiri Ishieke Market, Gariki Ogoja Road, Orokeonuoha Market, Abakpa Market, Ekeaba Market, Nkwegu Market, Rice Mill Market, and others within the municipality. These markets served as the sampling sites for this study (Orji *et al.*, 2025).

Ethical Consideration: Ethical approval for this study was obtained from the Research and Ethical Committee of the Federal Ministry of Livestock Development (Approval No.: FMLD/REC/203/2025/R12). Prior to sample collection, informed consent was sought and obtained from the goat owners at Gariki Ogoja Road, Abakaliki, after providing clear explanations regarding the purpose and procedures of the study. All animal handling procedures were conducted in accordance with standard veterinary practices to minimize stress and discomfort to the animals. Samples were collected aseptically by trained personnel, and no animal was harmed or euthanized for the purpose of this research.

Sample Collection: Sterile swab sticks were used to collect samples aseptically from the skin and nostrils of goats at Gariki Ogoja Road, Abakaliki, Ebonyi State. A total of thirty (30) samples were collected, comprising twenty (20) skin swabs and ten (10) nasal swabs. Each swab was moistened with sterile saline before sampling to enhance bacterial recovery. For skin samples, the swab was rubbed thoroughly over an area of approximately 5 cm² on the dorsal and lateral aspects of the goat's body. For nasal samples, the swab was inserted gently into each nostril and rotated to ensure

adequate contact with the mucosal surface. After collection, each swab was immediately placed into a sterile test tube containing 5 ml of sterile nutrient broth and labeled accordingly. The samples were transported in an insulated ice container maintained at 4°C to the Microbiology Laboratory for processing within 4 hours of collection.

Isolation of Gram-Negative Bacteria: Upon arrival at the laboratory, each samples were uniform suspended in a sterile nutrient broth (Thermo Fisher Scientific, U. S. A) overnight. After 24 hours of incubation, a loopful of the nutrient broth culture was streaked onto MacConkey agar (Thermo Fisher Scientific, U. S. A), and eosin methylene blue (EMB) (Thermo Fisher Scientific, U. S. A) agar plates using the quadrant streaking method. For enhanced isolation of *Salmonella* and *Shigella* species, samples were also inoculated onto *Salmonella-Shigella* agar (SSA) (Thermo Fisher Scientific, USA) plates. All inoculated plates were incubated aerobically at 37°C for 24hours. Following incubation, plates were examined for characteristic colony morphology. On MacConkey agar (Thermo Fisher Scientific, U. S. A), lactose-fermenting colonies appeared pink, while non-lactose-fermenting colonies appeared colorless. On EMB agar, *E. coli* colonies exhibited a green metallic sheen, while other Gram-negative bacteria appeared as dark-centered colonies. On SSA, *Salmonella* species appeared as colorless colonies with black centers due to hydrogen sulfide production, while *Shigella* species appeared as colorless colonies without black centers. Discrete colonies exhibiting typical characteristics of Gram-negative bacteria were selected and sub-cultured onto fresh nutrient agar plates to obtain pure isolates. The purity of each isolate was confirmed by repeated streaking, microscopic examination and with further confirmatory testing using VITEK® 2 COMPACT automated system (bioMérieux, France). Pure isolates were maintained on nutrient agar slants at 4°C for further use.

Antibiotic susceptibility testing: Antibiotic susceptibility testing was performed on all confirmed Gram-negative bacterial isolates (*Escherichia coli*, *Klebsiella pneumoniae*, *Salmonella enteritidis*, and *Shigella flexneri*) using the Kirby-Bauer disc diffusion method according to the Clinical and Laboratory Standards Institute (CLSI, 2023) guidelines. Each pure bacterial isolate suspension was adjusted to reach the 0.5 McFarland turbidity standard (approximately 1.5×10^8 CFU/ml). A sterile cotton swab was dipped into the standardized suspension, and excess inoculum was removed before streaking evenly across the entire surface of Mueller-Hinton agar (Thermo Fisher Scientific, U. S. A) plates in three directions (Edemekong *et al.*, 2025). The inoculated plates were allowed to stand for 10 minutes before antibiotic disc application. The following antibiotic discs (Oxoid, UK) were used: ceftazidime (30 µg), ceftriaxone (30 µg), imipenem (10 µg), amoxicillin-clavulanic acid (30 µg), azithromycin (15 µg), levofloxacin (5 µg), Streptomycin (25 µg), Nalixidic (15 µg), Trimethoprim-Sulfamethoxazole (25 µg), Tetracycline (5 µg), Azetronam (30 µg), Piperacillin-tazobactam (110 µg), and amikacin (30 µg). Using sterile forceps, the discs were placed onto the agar surface and gently pressed to ensure full contact. Plates were incubated at 37°C for 24 hours. Following incubation, zones of inhibition were measured to the nearest millimeter using a calibrated ruler and interpreted according to CLSI (2020) breakpoints for Enterobacteriaceae. Quality control was performed using *Escherichia coli* ATCC 25922 and *Klebsiella pneumoniae* ATCC 700603.

Multiple Antibiotic Resistance Index (MARI): The multiple antibiotic resistance index was calculated for each isolate using the formula described by Edemekong *et al.* (2022):

$$\text{MAR index} = \frac{a}{b}$$

Where; *a* is the number of antibiotics to which the isolate was resistant, and *b* is the total number of antibiotics tested (*b*=13).

Isolates with MARI values exceeding 0.2 were considered to have originated from high-risk sources of antibiotic use (Nowde *et al.*, 2026b; Edemekong *et al.*, 2022). The average MARI for each bacterial species was calculated, and isolates resistant to three or more antibiotic classes were classified as multidrug-resistant (MDR) according to John-Onwe *et al.* (2023) and Agwu *et al.* (2026).

RESULTS

A total of 31 Gram-negative bacterial isolates were recovered from the skin and nasal samples of goats bred in Abakaliki, Ebonyi State. The isolates comprised *Escherichia coli* 12 (38.7%), *Klebsiella pneumoniae* 8 (25.8%), *Shigella flexneri* 7 (22.6%), and *Salmonella* Enteritidis 4 (12.9%). The distribution of isolates by sample type is presented in Table 1.

Antibiotic susceptibility testing was performed on all 33 Gram-negative bacterial isolates using the Kirby-Bauer disc diffusion method against 13 antibiotics. The susceptibility patterns varied among the bacterial species. All isolates (100%) were resistant to amoxicillin-clavulanic acid. High resistance rates were also observed against ceftazidime, ceftriaxone, azithromycin, and levofloxacin among *Shigella flexneri*, *E. coli*, and *Salmonella* isolates. *Klebsiella pneumoniae* isolates demonstrated 100% resistance to amoxicillin-clavulanic acid and 75% resistance to ceftazidime and ceftriaxone. Complete susceptibility to imipenem was observed in *Shigella* isolates, while *Salmonella* Enteritidis isolates showed 100% susceptibility to levofloxacin. The detailed antibiotic susceptibility profile is presented in Table 2.

The multiple antibiotic resistance index was calculated for each isolate based on resistance to the 13 antibiotics tested. Isolates with MARI > 0.2 are considered to have originated from high-risk sources of antibiotic use. Among the isolates, *Shigella flexneri* had a mean MARI of 0.54, *E.*

coli 0.52, *Klebsiella pneumoniae* 0.48, and *Salmonella* Enteritidis 0.62. All isolates had MARI values exceeding 0.2, indicating high antibiotic exposure pressure. Additionally, all isolates were resistant to

three or more antibiotic classes and were therefore classified as multidrug-resistant (MDR) according to John-Onwee *et al.* (2023).

Table 1: Distribution of Gram-Negative Bacteria Isolated from Skin and Nasal Samples of Goats

Organisms	Skin (n=23)	Nasal (n=8)	Total (N=31)
	No. (%)	No. (%)	No. (%)
<i>E. coli</i>	9 (39.1)	3 (37.5)	12 (38.7)
<i>Klebsiella pneumoniae</i>	5 (21.7)	3 (37.5)	8 (25.8)
<i>Shigella flexneri</i>	6 (26.1)	1 (12.5)	7 (22.6)
<i>Salmonella</i> Enteritidis	3 (13.0)	1 (12.5)	4 (12.9)
Total	23 (100.0)	8 (100.0)	31 (100.0)

Table 2: Antibiotic Susceptibility Profile of Gram-Negative Bacterial Isolates from Skin and Nasal Samples of Goats

Antibiotic (µg)	<i>Shigella flexneri</i> (n=7)		<i>E. coli</i> (n=12)		<i>Klebsiella pneumoniae</i> (n=8)		<i>Salmonella</i> Enteritidis (n=4)	
	R (%)	S (%)	R (%)	S (%)	R (%)	S (%)	R (%)	S (%)
Ceftazidime (30)	5 (71.4)	2 (28.6)	1 (8.3)	11 (91.7)	6 (75.0)	2 (25.0)	3 (75.0)	1 (25.0)
Ceftriaxone (30)	5 (71.4)	2 (28.6)	1 (8.3)	11 (91.7)	6 (75.0)	2 (25.0)	3 (75.0)	1 (25.0)
Imipenem (10)	0 (0.0)	7 (100.0)	4 (33.3)	8 (66.7)	1 (12.5)	7 (87.5)	3 (75.0)	1 (25.0)
Amoxicillin-clavulanic acid (30)	7 (100.0)	0 (0.0)	12 (100.0)	0 (0.0)	8 (100.0)	0 (0.0)	4 (100.0)	0 (0.0)
Azithromycin (15)	2 (28.6)	5 (71.4)	12 (100.0)	0 (0.0)	1 (12.5)	7 (87.5)	4 (100.0)	0 (0.0)
Levofloxacin (5)	7 (100.0)	0 (0.0)	11 (91.7)	1 (8.3)	3 (37.5)	5 (62.5)	0 (0.0)	4 (100.0)
Streptomycin (25)	4 (57.1)	3 (42.9)	8 (66.7)	4 (33.3)	5 (62.5)	3 (37.5)	2 (50.0)	2 (50.0)
Nalidixic acid (15)	6 (85.7)	1 (14.3)	9 (75.0)	3 (25.0)	5 (62.5)	3 (37.5)	3 (75.0)	1 (25.0)
Trimethoprim-sulfamethoxazole (25)	5 (71.4)	2 (28.6)	10 (83.3)	2 (16.7)	6 (75.0)	2 (25.0)	3 (75.0)	1 (25.0)
Tetracycline (5)	6 (85.7)	1 (14.3)	11 (91.7)	1 (8.3)	7 (87.5)	1 (12.5)	3 (75.0)	1 (25.0)
Aztreonam (30)	4 (57.1)	3 (42.9)	2 (16.7)	10 (83.3)	5 (62.5)	3 (37.5)	3 (75.0)	1 (25.0)
Piperacillin-tazobactam (110)	2 (28.6)	5 (71.4)	1 (8.3)	11 (91.7)	3 (37.5)	5 (62.5)	2 (50.0)	2 (50.0)
Amikacin (30)	1 (14.3)	6 (85.7)	3 (25.0)	9 (75.0)	1 (12.5)	7 (87.5)	3 (75.0)	1 (25.0)

Table 3: Mean Multiple Antibiotic Resistance Index of Bacterial Isolates

Bacterial Isolate	Mean MARI
<i>Shigella flexneri</i>	0.54
<i>E. coli</i>	0.52
<i>Klebsiella pneumoniae</i>	0.48
<i>Salmonella</i> Enteritidis	0.62

DISCUSSION

The present study isolated four Gram-negative bacterial species from the skin and nasal samples of goats in Abakaliki, Nigeria, comprising *Escherichia coli* 12(38.7%), *Klebsiella pneumoniae* 8 (25.8%), *Shigella flexneri* 7(22.6%), and *Salmonella* Enteritidis 4(12.9%). The predominance of *E. coli* among the isolates aligns with findings from Anueyiagu *et al.* (2025), who reported *E. coli* as the most prevalent isolate (71%) in ruminants with subclinical mastitis

in Plateau State, Nigeria. The presence of these enteric bacteria on the skin and nasal mucosa of goats is significant, as these sites are not traditional reservoirs for such organisms, suggesting fecal contamination from the animals' environment or from handling practices. The higher isolation rate from skin samples (23 isolates) compared to nasal samples (8 isolates) indicates that the skin may serve as a more substantial reservoir for Gram-negative bacteria in goats, likely due to direct contact with

contaminated bedding, soil, or handlers. Abreham *et al.* (2019) similarly documented the presence of *E. coli* in goats and sheep, with 20% of fecal samples from goats testing positive for *E. coli*, further supporting the role of small ruminants as reservoirs for this bacterium.

The detection of *Salmonella* Enteritidis (12.9%) in our study warrants comparison with findings from other regions. Manishimwe *et al.* (2021) highlighted that *Salmonella* spp., although an indicator of possible animal illness, can frequently be harbored by animals without outward signs of sickness, given its natural residence in their intestines and potential presence in their feces. This observation is particularly relevant to our findings, as the goats sampled in Abakaliki markets did not exhibit overt clinical signs of salmonellosis, yet *Salmonella* was isolated from their skin and nasal surfaces. Microorganisms present in feces have the potential to easily contaminate various parts of an animal's body, including its fur, as well as the immediate surroundings, such as pens, water, and soil (Manishimwe *et al.*, 2021). This mechanism likely explains the presence of *Salmonella* on goat skin in our study, where inadequate hygiene practices in market settings facilitate fecal-oral and fecal-skin contamination cycles. While *Salmonella* infections in animals may present with symptoms such as diarrhea, lethargy, vomiting, or fever (CDC, 2022), the absence of these signs in the sampled goats underscores the challenge of identifying asymptomatic carriers that can serve as reservoirs for zoonotic transmission. Notably, humans can contract salmonellosis by handling animals, particularly through direct touch, and handling contaminated food and unintentionally transferring the bacterium from hands to mouths can lead to *Salmonella* infections (Manishimwe *et al.*, 2021). This finding directly supports the public health concern raised in our study, that goats in Abakaliki markets may serve as reservoirs for transmission of pathogenic

bacteria to humans through direct contact, slaughtering, and meat consumption.

Hawwas *et al.* (2022) in the Suez Canal Area, Egypt, reported that *Salmonella* occurrence in goat feces was 7%, with *S. Enteritidis* being the most frequently identified serotype in both goats and human stool samples. Critically, they found that humans in contact with sheep and goats had a significantly higher prevalence of *Salmonella* infection (28%) compared to those with no animal contact (7.3%) ($\chi^2 = 6.728$, $p < 0.05$), demonstrating the zoonotic transmission potential of goat-associated *Salmonella*. In Maiduguri, Nigeria, Mustapha *et al.* (2024) investigated the prevalence of *Salmonellae* infections in sheep and goats and reported a prevalence of 14% in goats from rectal swab samples, with 41.66% of positive isolates from goats. This prevalence is comparable to our finding of 12.9% for *Salmonella* Enteritidis. Their study also highlighted an important epidemiological consideration the potential role of lizards in the transmission cycle of *Salmonellae* to small ruminants, as lizards were found in the vicinity of small ruminant houses and tested positive for *Salmonellae* (12% prevalence). This finding suggests that environmental reservoirs, including reptiles, may contribute to the persistence and transmission of *Salmonella* in goat production systems, a factor that warrants consideration in Abakaliki where goats are raised in open market environments.

The detection of *Shigella flexneri* (22.6%) from goat skin and nasal samples in this study represents a finding of considerable public health importance. This aligns with recent genomic evidence from Motlhaping *et al.* (2025) in South Africa, who conducted the first whole-genome study of *Shigella* species isolated from small ruminants and manure in the North West Province. Their investigation revealed that *S. flexneri* strains from livestock environments could not be assigned to known sequence types, suggesting novel or uncharacterized lineages circulating in small ruminant populations. Li *et al.* (2022) similarly

identified the presence of *Shigella* spp. in both goats and sheep, confirming that these small ruminants can harbor this pathogen. The detection of *Shigella* spp. in farms raises concerns about the risk of shigellosis and gastrointestinal infections in food animals (Li *et al.*, 2022). While *Shigella* infections are commonly associated with human illness, documented cases indicate occurrences in goats (Ishlak *et al.*, 2022). With the potential for zoonotic transmission, animals can serve as reservoirs and transmit pathogens to humans. *Shigella* infections in humans are often linked to the ingestion of contaminated food or water (Nomeh *et al.*, 2026; Orji *et al.*, 2025; Oke *et al.*, 2024). However, documented evidence has revealed instances where human illnesses originate from exposure to *Shigella*-infected livestock or their excrement (Buskirk *et al.*, 2022). This emphasizes an alternative pathway of transmission, highlighting the potential risk associated with direct or indirect contact with *Shigella*-contaminated animals or their environmental surroundings. *Shigella* transmission to humans can occur through direct or indirect contact with animals including goats and sheep, emphasizing the importance for individuals involved in livestock-related activities or exposed to livestock feces to take precautionary measures, such as wearing gloves and maintaining good hygiene (Buskirk *et al.*, 2022).

The prevalence of *Shigella* spp. in goats observed in our study (22.6%) is comparable to findings from smallholder farms in the United States, where Basnet and Kilonzo-Nthenge (2024) investigated antibiogram profiles of pathogenic bacteria in goat and sheep feces on smallholder farms in Tennessee and Georgia. Their study reported that 27% of goats tested positive for *Shigella* spp., with overall *Shigella* prevalence across both sheep and goats at 35.0%. The higher *Shigella* prevalence in their study compared to ours may reflect differences in sampling sites fecal samples versus skin and nasal swabs as *Shigella* is primarily an

enteric pathogen. Nonetheless, the detection of *Shigella* on goat skin and nasal surfaces in our study suggests that these sites may serve as alternative reservoirs or indicate fecal contamination during handling and housing. The presence of *Klebsiella pneumoniae* (25.8%) in our study is consistent with its recognition as an opportunistic pathogen in both veterinary and human medicine (Uzoeto *et al.*, 2025; Nomeh *et al.*, 2023). The variation in isolate distribution between sample types; skin yielding predominantly *E. coli* and *Shigella flexneri* while nasal samples showed higher proportions of *Klebsiella* may reflect differences in ecological niches. The nasal cavity may select for different bacterial populations than skin, with *Klebsiella* potentially better adapted to mucosal surfaces. This pattern warrants further investigation using molecular typing methods to determine whether isolates from different sites represent the same clonal populations or distinct strains.

Regarding the antibiotic susceptibility profile, all isolates (100%) demonstrated resistance to amoxicillin-clavulanic acid, with high resistance rates also observed against ceftazidime, ceftriaxone, azithromycin, and levofloxacin across the bacterial species. The high resistance to β -lactam antibiotics observed in our study aligns with findings from Basnet and Kilonzo-Nthenge (2024), who reported that antibiotic resistance among goat isolates was primarily against ampicillin (79.4%), vancomycin (65.1%), and gentamycin (63.6%), with significantly lower resistance to tetracycline (41.6%) and imipenem (21.8%). In contrast, our study revealed higher tetracycline resistance (75-91.7%), reflecting geographic variation in antibiotic usage patterns and selection pressures between Nigeria and the United States, where stricter antibiotic stewardship policies are enforced. The existence of tetracycline-resistant bacteria and resistant genes in animal fecal samples has also been confirmed by several studies (Turner *et al.*,

2022; Osorio *et al.*, 2023). Tetracycline, oxytetracycline, and doxycycline stand out as the most employed tetracyclines in veterinary medicine (Nwode *et al.*, 2026b). The affordability and broad-spectrum nature of tetracyclines have contributed to their extensive utilization in various applications, which likely explains the high tetracycline resistance rates observed in our study (Nwode *et al.*, 2026a; Nwode *et al.*, 2026b). The 100% resistance to amoxicillin-clavulanic acid across all isolates in our study is particularly concerning. Hawwas *et al.* (2022) reported that *Salmonella* isolates from goats in Egypt showed high susceptibility to ciprofloxacin and norfloxacin but exhibited resistance to ampicillin and amoxicillin, consistent with our findings of β -lactam resistance. Similarly, Mairiga *et al.* (2024) reported that *Salmonella* isolates from goats in Maiduguri were highly sensitive to ciprofloxacin (100%) and ofloxacin (100%), but resistant to ampicillin and amoxicillin. This pattern of β -lactam resistance appears consistent across Nigerian and Egyptian goat populations, suggesting widespread use of these antibiotics in livestock production without veterinary oversight.

The susceptibility patterns observed in our study varied among bacterial species. Notably, *Shigella flexneri* isolates demonstrated complete susceptibility (100%) to imipenem but low level resistance to imipenem was found in other Gram-negative bacteria in our study. However, instances of carbapenem resistance have been documented in livestock, including goats and sheep in Southeast Asia and Jordan (Khalifa *et al.*, 2021; Obaidat *et al.*, 2023). In recent time, resistance to imipenem raises significant concerns in healthcare settings, limiting the efficacy of this vital antibiotic in treating serious infections (Peteret *et al.*, 2025; Uzoeto *et al.*, 2025; Ogba *et al.*, 2025; Nomeh *et al.*, 2023; Ogba *et al.*, 2022). The presence of antimicrobial-resistant bacteria resulting from the administration of antibiotics such as imipenem to goats poses a potential food

safety hazard. Although carbapenems are not licensed in livestock or veterinary fields and are not commonly tested in animals (Ramírez-Castillo *et al.*, 2023), our study underscored the noteworthy aspect of bacterial isolates' resistance to carbapenems. The persistence and dissemination of carbapenem-resistant bacteria in the environment pose significant and interconnected risks to the health of goats, and the farm environment (Joseph *et al.*, 2023). Despite the low resistance observed in bacterial isolates in this study, the prevalence of Carbapenem-resistant Enterobacteriaceae (CRE) and other resistant Gram-negative bacteria is on the rise, posing a substantial threat to both human and animal health. Carbapenems are acknowledged as one of the most potent classes of antibiotics, typically reserved as a last-line treatment option for severe bacterial infections (Peteret *et al.*, 2025). They exhibit broad-spectrum activity, proving more effective than other β -lactams, especially against resistant bacteria (Aurilio *et al.*, 2022).

The multiple antibiotic resistance index (MARI) values in our study ranged from 0.48 for *Klebsiella pneumoniae* to 0.62 for *Salmonella* Enteritidis, all exceeding the 0.2 threshold established by Edemekong *et al.* (2022) and Agwu *et al.* (2026) as indicative of high-risk antibiotic exposure. These values reflect the cumulative effect of antibiotic selection pressure in the study area and are consistent with findings from Motlhaping *et al.* (2025), whose genomic analysis provided genetic insight into how such resistance profiles emerge. Their study demonstrated that *Shigella* species from small ruminants possess a core set of resistance determinants involving β -lactams (*ampC1*, *ampC*, *ampH*), macrolides (*mphB*), polymyxins (*eptA*, *pmrF*), and multidrug efflux pumps (AcrAB-TolC, Mdt, Emr, Kpn families), along with regulatory systems (*marA*, *hns*, *crp*, *baeRS*, *evgAS*, *cpxA*, *gadX*) that modulate resistance expression. The presence of these diverse resistance mechanisms in goat-associated Gram-

negative bacteria underscores the complexity of addressing antimicrobial resistance in livestock production systems.

All isolates in our study were resistant to three or more antibiotic classes and were therefore classified as multidrug-resistant (MDR) according to John-Onwe *et al.* (2023). This finding aligns with Basnet and Kilonzo-Nthenge (2024), who reported that 89.6% of bacterial isolates from goat feces exhibited multidrug resistance, with 52% demonstrating resistance to at least five antimicrobial classes. The high prevalence of MDR bacteria in goat populations across geographically distinct regions Nigeria, the United States, and South Africa suggests that goats serve as important reservoirs for multidrug-resistant bacteria globally, highlighting the need for a coordinated One Health approach to surveillance and intervention.

CONCLUSION

This study successfully investigated the prevalence, antibiotic susceptibility profile, and multiple antibiotic resistance index of Gram-negative bacteria isolated from skin and nasal samples of goats bred in Abakaliki, Ebonyi State, Nigeria. Four clinically significant Gram-negative bacterial species were recovered from the sampled goats, with *Escherichia coli* being the most prevalent, followed by *Klebsiella pneumoniae*, *Shigella flexneri*, and *Salmonella* Enteritidis. The higher recovery of isolates from skin samples compared to nasal samples indicates that the

skin serves as a more substantial reservoir for these organisms in the studied goat population. Regarding antibiotic susceptibility, all isolates exhibited resistance to amoxicillin-clavulanic acid. High resistance rates were also observed against ceftazidime, ceftriaxone, azithromycin, and levofloxacin, though susceptibility patterns varied by bacterial species.

Notably, *Shigella flexneri* demonstrated complete susceptibility to imipenem, while *Salmonella* Enteritidis was fully susceptible to levofloxacin, indicating that some critically important antibiotics retain efficacy against specific pathogens in this setting. The calculated mean multiple antibiotic resistance index values for all bacterial species exceeded the high-risk threshold, signifying that the isolates originated from sources with considerable antibiotic exposure pressure. Consistent with this high-risk profile, every isolate demonstrated resistance to three or more antibiotic classes and was classified as multidrug-resistant. This study establishes that goats in Abakaliki markets harbor multidrug-resistant Gram-negative bacteria with high antibiotic resistance indices on their skin and nasal surfaces. These findings provide baseline data on the prevalence and resistance profiles of these organisms in this previously uncharacterized goat population, highlighting the zoonotic transmission risks and the urgent need for antimicrobial stewardship and One Health surveillance in Nigeria's livestock sector.

REFERENCES

- Abreham, S. T., Cox, A., and Sisay, T. (2019). *Escherichia coli* O157:H7: distribution, molecular characterization, antimicrobial resistance patterns and source of contamination of sheep and goat carcasses at an export abattoir, Mojo, Ethiopia. *BMC Microbiology* 19: 215.
- Adesola, R. O., Bakre, A. A., Adekanmbi, A. O., Ogunro, B. N., Ogundijo, O. A., Hamzat, A., Hossain, D., Aribana, M. A., and Balogun, L. A. (2025). Molecular and epidemiological characterization of ESBL-producing *Escherichia coli* from captive wild birds in zoological gardens in Nigeria. *Microbiology Insights* 18: 1-12.
- Adibe-Nwafor, J. O., Uduku, N. D., Iroha, C. S., Ibiam, F. A., Onuora, A. L., Nwafor, K. A., Peter, I. U., and

- Iroha, I. R. (2023). Distribution and antibiotic resistance profile of extended spectrum beta-lactamase producing *Escherichia coli* from fish farms within Abakaliki Metropolis. *Advances in Research* 24(5): 175-184.
- Agwu, F. O., Iroha, C. S., Onoura, A. L., Peter, I. U., & Iroha, I. R. (2026). Gender-Based Differences in Antibiotic Resistance Profiles of Multidrug-Resistant *Pseudomonas aeruginosa* Isolates from a Nigerian Tertiary Hospital: A Comparative Statistical Analysis. *Journal of International Research in Medical and Pharmaceutical Sciences*, 21(2):171–182.
- Anueyiagu, K. N., Agusi, E. R., Kabantiyok, D., Ayanbimpe, G. M., and Ikeh, E. I. (2025). Zoonotic potential of ESBL-producing coliforms in pastorally managed ruminants with subclinical mastitis in Plateau State, Nigeria. *Frontiers in Antibiotics* 4: 1632264.
- Aurilio, C., Sansone, P., Barbarisi, M., Pota, V., Giaccari, L. G., Coppolino, F., et al. (2022). Mechanisms of action of carbapenem resistance. *Antibiotics* 11(3): 421.
- Basnet, A., and Kilonzo-Nthenge, A. (2024). Antibigram profiles of pathogenic and commensal bacteria in goat and sheep feces on smallholder farm. *Frontiers in Antibiotics* 3: 1351725.
- Basnet, A., and Kilonzo-Nthenge, A. (2024). Antibigram profiles of pathogenic and commensal bacteria in goat and sheep feces on smallholder farm. *Frontiers in Antibiotics* 3: 1351725.
- Buskirk, A. D., Ndungo, E., Shimanovich, A. A., Lam, D., Blackwelder, W. C., Ikumapayi, U. N., et al. (2022). Mucosal immune profiles associated with diarrheal disease severity in *Shigella*- and enteropathogenic *Escherichia coli*-infected children enrolled in the global enteric multicenter study. *mBio* 13: e00538-22.
- CDC (2022). The biggest antibiotic-resistant threats in the U.S. Centers for Disease Control and Prevention.
- CLSI (2020). Performance standards for anti-microbial susceptibility testing. 30th Edition (M100). Clinical & Laboratory Standard Institute, Berwyn, PA.
- Demirci, M., Yiğın, A., Altuner, D., and Çelik, S. (2019). *Salmonella* spp. and *Shigella* spp. detection via multiplex real-time PCR and discrimination via MALDI-TOF MS in different animal raw milk samples. *Nigerian Journal of Clinical Practice* 22(8): 1083-1090.
- Edemekong, C. I., Chukwujekwu, A. G., Okorie, M. E., Udenweze, E., Obodoechi, I. F., Ogbonna, I. P., and Iroha, I. R. (2025). Evaluation of antibacterial activity of selected medicinal plants on extended spectrum β -lactamase producing *Salmonella enterica* serovar Typhimurium. *UMYU Scientifica* 4(1): 28-36.
- Edemekong, C. I., Iroha, I. R., Thompson, M. D., Okolo, I. O., Uzoeto, H. O., Ngwu, J. N., Mohammed, I. D., Chukwu, E. B., Nwuzo, A. C., Okike, B. M., Okolie, S. O., and Peter, I. U. (2022). Phenotypic characterization and antibiogram of non-oral bacteria isolates from patients attending dental clinic at Federal College of Dental Technology and Therapy Medical Center Enugu. *International Journal of Pathogen Research* 11(2): 7-19.
- Ejikeugwu, C., Esimone, C., Iroha, I., and Adikwu, M. (2018). First detection of FOX-1 AmpC β -lactamase gene expression among *Escherichia coli* isolated from abattoir samples in Abakaliki, Nigeria. *Oman Medical Journal* 33(3): 243-249.

- Hawwas, H. A. E., Abd El-Halim, H., and El-Gohary, A. (2022). *Salmonella* serovars in sheep and goats and their probable zoonotic potential to humans in Suez Canal Area, Egypt. *Acta Veterinaria Scandinavica* 64: 17.
- Ishlak, A., Ehmid, M., Toweir, H., Mohammed, A., and Mansour, A. (2022). Bacterial diversity in goat milk produced in suburbs Benghazi - Libya. *International Journal of Scientific and Research Publications* 12.
- Ishlak, A., Mustafa, M., and Abubakar, S. (2022). *Shigella* infections in small ruminants: A review of documented cases and public health implications. *Veterinary World* 15(3): 621-630.
- John-Onwe, B. N., Aniokete, U. C., Ibiam, F. A., Peter, I. U., Iroha, C. S., and Iroha, I. R. (2023). Dissemination of multidrug-resistant, extensively drug resistant and pandrug-resistant *Pseudomonas aeruginosa* isolates among in-patients and out-patients in a multi-profile health care settings. *Journal of Advances in Microbiology* 23(10): 109-115.
- Joseph, A. J., Yakubu, A., and Mallam, I. (2025). Antibiotic resistance of bacteria isolated from smallholder Red Sokoto and West African Dwarf breeds of goats faeces in Nasarawa State, Nigeria. *Nigerian Journal of Animal Production*: 398-401.
- Joseph, I. S., Okolo, I. O., Udenweze, E. C., Nwankwo, C. E., Peter, I. U., Ogbonna, I. P., and Iroha, I. R. (2023). Comparison of antibiotic-resistant pattern of extended spectrum beta-lactamase and carbapenem resistant *Escherichia coli* isolates from clinical and non-clinical sources. *Journal of Drug Delivery and Therapeutics* 13(7): 107-118.
- Khalifa, H. O., Oreiby, A., Abd El-Hafeez, A. A., Abd El Latif, A., Okanda, T., Kato, Y., et al. (2021). High β -lactam and quinolone resistance of Enterobacteriaceae from the respiratory tract of sheep and goat with respiratory disease. *Animals* 11 (8): 2258.
- Kumavath, R., Gupta, P., Tatta, E. R., Mohan, M. S., Salim, S. A., and Busi, S. (2025). Unraveling the role of mobile genetic elements in antibiotic resistance transmission and defense strategies in bacteria. *Frontiers in Systems Biology* 5: 1557413.
- Li, J., Yan, H., Chen, J., Duan, C., Guo, Y., Liu, Y., et al. (2022). Correlation of ruminal fermentation parameters and rumen bacterial community by comparing those of the goat, sheep, and cow in vitro. *Fermentation* 8(9): 427.
- Mairiga, I. A., Muhammad, M., and Adamu, L. (2024). Prevalence of Salmonellae infections in sheep and goats and the possibilities of lizards in infection transmission cycle in Maiduguri and environs. *Journal of Sustainable Veterinary and Allied Sciences* 6(3): 156-161.
- Manishimwe, R., Moncada, P. M., Musanayire, V., Shyaka, A., Scott, H. M., and Loneragan, G. H. (2021). Antibiotic-resistant *Escherichia coli* and *Salmonella* from the feces of food animals in the East Province of Rwanda. *Animals* 11(4): 1013.
- Motlhaping, T., Ramatla, T., Thekisoe, O., Khasapane, N. G., and Lekota, K. E. (2025). Genomic insights into *Shigella* species isolated from small ruminants and manure in the North West Province, South Africa. *Applied Microbiology and Biotechnology* 109(1): 238.
- Nomeh, L. O., Federica, O. I., Joseph, O. V., Moneth, E. C., Ogba, R. C., Nkechi, O. A., Peter, I. U., Akpu, P. O., Edemekong, C. I., and Iroha, I. R. (2023). Detection of carbapenemase-producing *Escherichia*

- coli* and *Klebsiella pneumoniae* implicated in urinary tract infection. *Asian Journal of Research in Infectious Diseases* 2(1): 15-23.
- Nomeh, O. L., Peter, I. U., Ogba, R. C., Otu, Q., Mbafo, M., Ogobuchi, D., and Nworie, O. (2026). Antibacterial efficacy of *Ficus ottoniifolia* and *Zanthoxylum zanthoxyloides* leaf extracts against bacterial isolates from contaminated drinking water sources in Ikwo South, Nigeria. *GSC Advanced Research and Reviews* 26(1): 94-109.
- Nwode, V. F., Iroha, C. S., Edemekong, C. I., Peter, I. U., and Iroha, I. R. (2026b). High carriage of multidrug-resistant, biofilm-forming MRSA phenotype in poultry and swine in Southeast Nigeria. *World Journal of Biological and Pharmaceutical Research* 10(1): 006-014.
- Nwode, V. F., Peter, I. U., Iroha, C. S., Edemekong, C. I., and Iroha, I. R. (2026a). Culture-based identification and molecular confirmation of vancomycin-resistant *Staphylococcus aureus* (VRSA) from porcine rectal swabs in Ebonyi State, Nigeria. *Asian Journal of Microbiology and Biotechnology* 11(1): 97-106.
- Nwojiji, E. C., Okolo, I. O., Osuji, C. A., Aniokete, U. C., Akomolafe, S. O., Okekeaji, U., Edemekong, C. I., Peter, I. U., and Iroha, I. R. (2025). Molecular detection of ampicillinase blaEBCM, blaFOX and blaDHAM resistant genes in multi-drug resistant Gram-negative bacteria. *GSC Biological and Pharmaceutical Sciences* 31(2): 113-122.
- Nwosu, U. O., Ibiam, F. A., Amadi-Ibiam, C. O., Aniokete, U. C., Iroha, C. S., Peter, I. U., and Iroha, I. R. (2023b). Antimicrobial resistance profile of *Salmonella enterica* serovar Typhi isolated from human clinical samples in Ebonyi State, Nigeria. *Journal of Advances in Medical and Pharmaceutical Sciences* 25(9): 11-17.
- Nwosu, U. O., Ibiam, F. A., Amadi-Ibiam, C. O., Iroha, C. S., Edemekong, C. I., Peter, I. U., and Iroha, I. R. (2023a). Fecal carriage of extended spectrum beta-lactamase and fluoroquinolone resistant gene in non-typhoidal *Salmonella enterica* isolates from food producing animals and humans. *Journal of Drug Delivery and Therapeutics* 13(9): 128-134.
- Obaidat, M. M., Tarazi, Y. H., and AlSmadi, W. M. (2023). Sheep and goats are reservoirs of colistin resistant *Escherichia coli* that co-resist critically important antimicrobials: first study from Jordan. *Journal of Food Safety* 43: e13023.
- Ogba, R. C., Akpu, P. O., Nwuzo, A. C., Peter, I. U., Nomeh, L. O., and Iroha, I. R. (2022). Antibiotic susceptibility profile of clinical isolate of carbapenem resistant *Pseudomonas aeruginosa*. *South Asian Journal of Research in Microbiology* 14(2): 14-23.
- Ogba, R. C., Lovette, N., Nwite, O. P., and Edemekong, C. I. (2025). Antimicrobial resistance patterns of *Salmonella enterica* serovar Typhi in blood culture of typhoid fever patients: a recent trend in tertiary hospital in Enugu. *International Journal of Advanced Research in Medicine* 7(2): 12-18.
- Oke, B., Peter, I. U., Chinenye, C. O. L., Okpaga, A. O., Chukwuemeka, N. M., John-Onwe, B. N., Mbam, N. F., and Iroha, I. R. (2024). Distribution and antibiotic resistance profile of Enterobacteriaceae isolates from well water in the rural communities of Ezza South Local Government Area of Ebonyi State. *International Journal of Pathogen Research* 13(4): 22-34.

- Orji, J. O., Nwenyim, I. J., Emeruwa, A. P., Ogbonna, I. P., Aniokete, U. C., Uchechukwu, O., Awoke, O. O., and Ekuma, P. U. (2025). Bacteriological quality and antibiotic susceptibility profiles of bacteria in raw and processed meat sold for consumption in Abakaliki, Nigeria: a public health concern. *South Asian Journal of Research in Microbiology* 19(7): 12-25.
- Osorio, V., Sabater i Mezquita, A., and Balcázar, J. L. (2023). Comparative metagenomics reveals poultry and swine farming are hotspots for multidrug and tetracycline resistance. *Environmental Pollution* 322: 121239.
- Peter, I. U., Ngwu, J. N., Edemekong, C. I., Ugwueke, I. V., Uzoeto, H. O., Joseph, O. V., Mohammed, I. D., Mbong, E. O., Nomeh, L. O., Ikusika, B. A., Ubom, I. J., Inyogu, J. C., Ntekpe, M. E., Obodoechi, I. F., NseAbasi, P. L., Ogbonna, I. P., Didiugwu, C. M., Akpu, P. O., Alagba, E. E., Ogba, R. C., and Iroha, I. R. (2022a). First report prevalence of livestock acquired methicillin resistant *Staphylococcus aureus* (LA-MRSA) strain in South Eastern, Nigeria. *IOSR Journal of Nursing and Health Science* 11(1): 50-56.
- Peter, I. U., Obike, O. C., Ngwu, J. N., Emeruwa, A. P., Okolo, I. O., and Mohammed, I. D. (2025). Prevalence of biofilm-forming and carbapenemase-producing Gram-negative bacilli colonizing indwelling urinary catheters of patients. *UMYU Scientifica* 4(2): 270-284.
- Ramírez-Castillo, F. Y., Guerrero-Barrera, A. L., and Avelar-González, F. J. (2023). An overview of carbapenem-resistant organisms from food-producing animals, seafood, aquaculture, companion animals, and wildlife. *Frontiers in Veterinary Science* 10: 1158588.
- Turner, A., Schubert, H., Puddy, E. F., Sealey, J. E., Gould, V. C., Cogan, T. A., et al. (2022). Factors influencing the detection of antibacterial-resistant *Escherichia coli* in fecal samples from individual cattle. *Journal of Applied Microbiology* 132: 2633-2641.
- Uzoeto, H., Edemekong, C. I., Ogbonna, I. P., and Peter, I. U. (2025). Antimicrobial resistance profiles of biofilm-forming *E. coli*, *Pseudomonas aeruginosa*, and *Klebsiella pneumoniae* isolates from chronic wound infection. In *Proceedings of the 4th International Electronic Conference on Antibiotics*. Basel, Switzerland: MDPI.