

**Original Article****Open Access****Prevalence and pattern of carriage of Enterobacterales isolates among food handlers in Nnewi metropolis, southeast Nigeria***¹Okwuanaso, C. B [0000-0003-4125-7155](https://orcid.org/0000-0003-4125-7155), ²Onyemelukwe, N. F., and ¹Ofodile, C. A [0000-0003-0664-9698](https://orcid.org/0000-0003-0664-9698).¹Department of Medical Microbiology and Public Health, Faculty of Medical Laboratory Science, Nnamdi Azikiwe University, Nnewi Campus, Nnewi, Nigeria²Department of Medical Laboratory Science, College of Health Sciences and Technology, University of Nigeria, Enugu Campus, Nigeria*Correspondence to: cb.okwuanaso@unizik.edu.ng; +2348030820722**Abstract:****Background:** There is a public health concern about carriage of Enterobacterales among food workers and their potential role for transmission of food-borne illnesses. Food contamination by Enterobacterales could happen at any point of the production, processing, distribution, and preparation processes. This challenge is especially severe in developing countries like Nigeria. The objective of this study is to determine the prevalence and pattern of carriage of Enterobacterales by food handlers in Nnewi metropolis, southeast Nigeria.**Methodology:** This was a cross-sectional study that employed a non-probability sampling technique to recruit 178 food handlers who gave their consent and were available at the sampling area during the time of the study. Stool, urine, nasal and hand swab samples were collected from 115, 116, 120 and 173 food handlers respectively for microbiological analysis using conventional culture isolation and biochemical identification by Analytical Profile Index (API) 20E (bioMérieux). The antimicrobial susceptibility testing of the isolates was carried out on the identified isolates by the disk diffusion method. Descriptive statistics were carried out on the data.**Results:** Forty-four (25.4%) of the hand swab samples from 173 food handlers, 39 (32.5%) of the nasal swabs from 120 food handlers, 67 (58.3%) of the stool samples from 115 food handlers, and 29 (25.0%) of the urine samples from 116 food handlers, were positive for Enterobacterales isolates. The frequency of Enterobacterales isolation was significantly higher from stool samples compared to other samples ($\chi^2=40.032$; $p<0.0001$), indicating a higher carriage rate in the gastrointestinal tracts of the food handlers. Across all the samples, a total of 179 Enterobacterales were isolated from the 101 (56.7%) positive food handlers. The frequency of isolation in descending order is *Escherichia coli* 23.2% (n=41), *Klebsiella* spp 18.1% (n=32), *Enterobacter* spp 15.3% (n=27), *Citrobacter* spp 10.7% (n=19), *Raoultella* spp 7.3% (n=13), *Serratia* spp 5.6% (n=10), *Salmonella* spp 3.9% (n=7), *Kluyvera* spp 3.9% (n=7), *Shigella* spp 2.8% (n=5), *Proteus* spp 2.8% (n=5), *Cronobacter* spp 1.7% (n=3), *Erwinia* spp 1.1% (n=2), *Pantoea* spp 1.1% (n=2), *Hafnia* spp 1.1% (n=2), and *Yersinia* spp 1.1% (n=2). The Enterobacterales isolates were resistant to cefotaxime (83.0%), amoxicillin-clavulanic acid (79.1%), cefuroxime (76.8%), cefixime (75.1%), imipenem-cilastatin (74.6%), ceftazidime-avibactam (73.3%), ceftriaxone-sulbactam (71.2%) and levofloxacin (70.5%).**Conclusion:** Food handlers in this study had a high carriage rate of resistant Enterobacterales pathogens, which can be transmitted to unsuspecting consumers, through the food processing and handling chains.**Keywords:** Carriage; Enterobacterales; Food handlers; Nnewi; Nigeria

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Copyright 2025 AJCEM Open Access. This article is licensed and distributed under the terms of the Creative Commons Attribution 4.0 International License <http://creativecommons.org/licenses/by/4.0/>, which permits unrestricted use, distribution and reproduction in any medium, provided credit is given to the original author(s) and the source. Editor-in-Chief: Prof. S. S. Taiwo**Prévalence et profil de portage des isolats d'Enterobacterales parmi les manipulateurs d'aliments dans la métropole de Nnewi, au sud-est du Nigéria***¹Okwuanaso, C. B [0000-0003-4125-7155](https://orcid.org/0000-0003-4125-7155), ²Onyemelukwe, N. F., et ¹Ofodile, C. A [0000-0003-0664-9698](https://orcid.org/0000-0003-0664-9698).¹Département de Microbiologie Médicale et de Santé Publique, Faculté des Sciences de Laboratoire Médical, Université Nnamdi Azikiwe, Campus de Nnewi, Nnewi, Nigéria²Département des Sciences de Laboratoire Médical, Collège des Sciences de la Santé et de la Technologie, Université du Nigéria, Campus d'Enugu, Nigéria*Correspondance à: cb.okwuanaso@unizik.edu.ng; +2348030820722

Résumé:

Contexte: Le transport d'Enterobacterales parmi les travailleurs du secteur alimentaire et leur rôle potentiel dans la transmission de maladies d'origine alimentaire constituent un problème de santé publique. La contamination des aliments par les Enterobacterales peut survenir à n'importe quel stade des processus de production, de transformation, de distribution et de préparation. Ce défi est particulièrement grave dans les pays en développement comme le Nigéria. L'objectif de cette étude est de déterminer la prévalence et le schéma de transport d'Enterobacterales par les manipulateurs d'aliments dans la métropole de Nnewi, dans le sud-est du Nigéria.

Méthodologie: Il s'agissait d'une étude transversale qui a utilisé une technique d'échantillonnage non probabiliste pour recruter 178 manipulateurs d'aliments qui ont donné leur consentement et étaient disponibles dans la zone d'échantillonnage au moment de l'étude. Des échantillons de selles, d'urine, de prélèvements nasaux et de mains ont été prélevés respectivement auprès de 115, 116, 120 et 173 manipulateurs d'aliments pour une analyse microbiologique utilisant l'isolement par culture conventionnelle et l'identification biochimique par l'indice de profil analytique (API) 20E (bioMérieux). Le test de sensibilité aux antimicrobiens des isolats a été réalisé sur les isolats identifiés par la méthode de diffusion sur disque. Des statistiques descriptives ont été réalisées sur les données.

Résultats: Quarante-quatre (25,4%) des échantillons de prélèvements manuels provenant de 173 manipulateurs d'aliments, 39 (32,5%) des prélèvements nasaux provenant de 120 manipulateurs d'aliments, 67 (58,3%) des échantillons de selles provenant de 115 manipulateurs d'aliments et 29 (25,0%) des échantillons d'urine provenant de 116 manipulateurs d'aliments étaient positifs pour les isolats d'Enterobacterales. La fréquence d'isolement des Enterobacterales était significativement plus élevée dans les échantillons de selles que dans les autres échantillons ($\chi^2 = 40,032$; $p < 0,0001$), ce qui indique un taux de portage plus élevé dans le tractus gastro-intestinal des personnes manipulant des aliments. Sur l'ensemble des échantillons, un total de 179 Enterobacterales ont été isolés chez les 101 (56,7%) personnes manipulant des aliments positifs. La fréquence d'isolement par ordre décroissant est la suivante: *Escherichia coli* 23,2% (n=41), *Klebsiella* spp 18,1% (n=32), *Enterobacter* spp 15,3% (n=27), *Citrobacter* spp 10,7% (n=19), *Raoultella* spp 7,3% (n=13), *Serratia* spp 5,6% (n=10), *Salmonella* spp 3,9% (n=7), *Kluyvera* spp 3,9% (n=7), *Shigella* spp 2,8% (n=5), *Proteus* spp 2,8% (n=5), *Cronobacter* spp 1,7% (n=3), *Erwinia* spp 1,1% (n=2), *Pantoea* spp 1,1% (n=2), *Hafnia* spp 1,1% (n=2) et *Yersinia* spp 1,1% (n=2). Les isolats d'Enterobacterales étaient résistants à la céfotaxime (83,0%), à l'amoxicilline-acide clavulanique (79,1%), à la céfuroxime (76,8%), à la céfixime (75,1%), à l'imipénème-cilastatine (74,6%), à la céftazidime-avibactam (73,3%), à la céftriaxone-sulbactam (71,2%) et à la lévofloxacine (70,5%).

Conclusion: Les personnes manipulant des aliments dans cette étude avaient un taux élevé de portage d'agents pathogènes Enterobacterales résistants, qui peuvent être transmis à des consommateurs sans méfiance, par le biais des chaînes de transformation et de manipulation des aliments.

Mots clés: Portage; Entérobactéries; Manipulateurs d'aliments; Nnewi; Nigéria

Introduction:

The family Enterobacteriaceae are facultatively anaerobic non-spore forming Gram-negative bacteria that primarily inhabit the gastrointestinal system of human and animal and are major contributors to nosocomial and community-acquired infections. They typically cause infections of the respiratory system, urinary tract, and wounds (1). There has been growing interest in public food consumption in recent years. However, food is a necessary component for human health and survival. Its importance, therefore, cannot be overemphasized. Microorganisms such as Enterobacteriaceae are regarded as indicator bacteria for the microbiological quality of food and the hygiene state of a manufacturing and handling process but can readily contaminate food. Customers are also in danger from Enterobacteriaceae-contaminated food.

Food handlers are frequently reported to harbor these bacterial species, and a range of environmental conditions can easily contaminate food during manufacturing and handling. Food contamination can happen at any stage of the production, processing, distribution, and preparation processes. Food hand-

lers' personal cleanliness, food hygiene knowledge, and food hygiene practices all have a significant impact on the likelihood that food may get contaminated (2).

Over 250 distinct food borne illnesses exist globally. A wide range of bacteria, viruses, and parasites are responsible for majority of these infectious disorders. Other foodborne illnesses may be poisonings caused by toxic substances such as enterotoxins produced by certain bacteria. *Shigella*, *Enterobacter*, *Citrobacter*, *Yersinia*, *Salmonella*, *Campylobacter*, *Listeria*, and pathogenic *Escherichia coli* are all potential candidates for this (3). These bacteria can be found on the skin of food handlers and can spread to cooked, wet, protein-rich foods.

Majority of the population in most African nations, particularly Nigeria, eat food from public restaurants and vending centers (4). The occupation of food handlers put them in danger of contracting and spreading infectious diseases. Actions to improve the occupational health and safety of food handlers can be influenced by knowledge of the incidence of Enterobacteriaceae in this population. To protect the health of the public, stop the spread of antibiotic resistance, and enhance the occu-

pational health and safety of this workforce, it is crucial to conduct research on the prevalence, antimicrobial susceptibility and pattern of Enterobacteriaceae among food handlers in Nnewi metropolis, southeast Nigeria.

Materials and method:

Study setting, design and participants:

This was a cross-sectional study conducted on asymptomatic food handlers in the 4 quarters of Nnewi metropolis (Uruagu, Umudim, Nnewichi and Otolo), southeast Nigeria.

Ethical consideration:

Ethical approval was obtained from the Ministry of Health, Anambra State, Nigeria, before the commencement of the study. All enrolled food handlers provided written informed consent.

Sample size determination:

The Leslie Fischer's formula, $n = Z^2 pq / d^2$ (5) for calculating sample size in population larger than 10,000 was used, where n is the estimated minimum sample size, Z is the 95% confidence interval level of significance (1.96), p is the proportion of food handlers (54.0%, 0.54) who adhere to proper food safety and hygiene, obtained from a previous study (6), q is the complementary probability (1- p), and d is the precision set at 0.05. This gave a calculated sample size of 382.

Inclusion and exclusion criteria:

All food handlers who had static food vending establishments and gave informed consent were included in the study. Food handlers under the age of 18 years, those with fever, diarrhoea, currently on antibiotic treatment, and those who did not complete the questionnaires administered were excluded.

Method of sampling:

A non-probability sampling technique was used to recruit all eligible food handlers within the study area. A pilot survey was first conducted in all the 4 quarters of Nnewi to identify volunteer participants.

Data and sample collection:

A total of 178 food handlers were recruited into the study over the period of the study. Socio-demographic data and food handlers' knowledge of carriage and antibiotic use were collected with a structured questionnaire.

Urine and fecal samples were collected into sterile containers while hand/nasal swab samples were collected with sterile swabs emulsified in normal saline. Nasal swabs were collected from a total of 120 food handlers, urine from 116, hand swabs from 173, and stool samples from 115 food handlers, giving a total of 524 samples. The samples were

transported to the laboratory within 1 hour of collection.

Microbiological cultures:

Urine sample:

A loopful of the urine sample was aseptically inoculated on cystine lactose electrolyte deficient (CLED) agar and incubated at 37°C for 24 hours. Subculture was done on MacConkey agar plates and incubated at 37°C for 24 hours as previously described (7).

Stool sample:

One gram of the stool specimen was enriched into 9 ml of buffered peptone water. A pea-sized stool sample was transferred into 9ml of Selenite F broth (Liofilchem) and incubated overnight at 37°C. A loopful of each enrichment medium was then sub-cultured onto MacConkey agar (Liofilchem) for detection of lactose fermenters (LF) and non-lactose fermenters (NLF). Subculture was also done on *Shigella-Salmonella* agar (SSA) (Liofilchem) for isolation of *Shigella* and *Salmonella* species. Plates were incubated for 24 hours at 37°C.

Swab samples:

Nasal and hand swab samples were cultured on MacConkey agar and SSA, which were incubated at 37°C for 24 hours.

Biochemical identification:

Biochemical identification of the Enterobacterales isolates from the culture plates was done using analytical profile index 20E (API 20E) (bioMérieux) and the isolates were identified to species level using the APIWEB™ V5.0 software.

Antibiotic susceptibility testing (AST) of bacterial isolates:

The antibiotic susceptibility test (AST) on the isolates was carried out using the modified Kirby-Bauer disc diffusion method in accordance with the CLSI guideline (8). About 3-5 colonies from overnight cultures on nutrient agar plates were emulsified in 5ml physiological saline, and the turbidity was adjusted to 0.5 McFarland standards, which corresponds to $\sim 1.5 \times 10^8$ CFU/ml.

The standardized inoculum suspension was inoculated on Mueller-Hinton (MH) agar using a sterile swab. Using a dispenser, the antibiotic discs were aseptically placed on the inoculated MH agar. A 30-minute pre-diffusion period was permitted, and the plates were then incubated for 24 hours at 37°C.

The diameter of zone of inhibition for each isolate was measured in millimeters, and interpreted as resistant, intermediate or sensitive based on the CLSI guideline (8). The antibiotics used include amoxicillin-clavulanic acid (AMC, 30µg), cefuroxime (CXM, 5µg), cefota-

xime (CTX, 5µg), ceftazidime-avibactam (CAZ, 30µg), ceftriaxone-sulbactam (CRO, 45µg), cefixime (5µg), gentamicin (GN, 10µg), nitrofurantoin (300µg), nalidixic acid (NA, 30µg), ofloxacin (OFX, 5µg), levofloxacin (5µg), imipenem-cilastatin (IMP, 10/10µg), and meropenem (MRP, 10 g).

Statistical analysis:

The data on the sociodemographic and knowledge of the food handlers as well as the microbiological data were analyzed in a computer Minitab version 21.2 with the statistical

package for the social sciences (SPSS) version 27.0.

Results:

The socio-demographic characteristics of the 178 food handlers is shown in Table 1. The age group 24-30 years represented the largest proportion of the food handlers (24.2%, n=43), with a mean age of 38.8 years, and predominantly females (82%, n=146). A total of 131 (74.0%) have post-primary education level and had worked in the food industry for 5 years or longer.

Table 1: Socio-demographic characteristics of selected food handlers in Nnewi metropolis, Nigeria

Characteristics	Frequency	Percentage (%)
Age group (years)		
16 - 20	14	7.9
21 - 25	29	16.3
26 - 30	43	24.2
31 - 35	27	15.1
36 - 40	19	10.7
41 - 45	15	8.4
46 - 50	10	5.6
51 - 55	9	5.1
56 - 60	7	3.9
61 - 65	5	2.8
Total	178	100
Mean age (± SD) (years)	38.8 ± 12.5	
Gender		
Female	146	82
Male	32	18
Total	178	100
Marital status		
Single	87	48.9
Married	91	51.1
Total	178	100
Level of education		
No Formal Education	26	14.6
Post-primary	21	11.8
Post-secondary	106	60
Tertiary	25	14
Total	178	100
Duration of food vending (years)		
≤ 5	86	48.3
> 5	92	51.7
Total	178	100

Table 2: Frequency of the Enterobacterales isolates from food handlers with respect to sample types in Nnewi metropolis

Sample type	Frequency	Percentage	χ^2	p value
Hand swab (n=173)	44	25.4	40.032	<0.0001*
Nasal swab (n=120)	39	32.5		
Stool (n=115)	67	58.3		
Urine (n=116)	29	25.0		
Total (n=524)	179	34.2		

* = statistically significant

Table 2 shows the frequency of the Enterobacterales isolated from the food handlers according to sample type. Of the hand swab samples collected from 173 food handlers, 44 (25.4%) were culture positive; of the nasal swabs collected from 120 food handlers, 39 (32.5%) were culture positive; of the stool samples collected from 115 food handlers, 67 (58.3%) were culture positive; and of the urine samples collected from 116 food handlers, 29 (25.0%) were culture positive for Enterobacterales isolates. The frequency of Enterobacterales isolation was significantly different with respect to the sample types ($\chi^2=40.032$; $p<0.0001$)

Table 3 shows that across all the samples (n=524) from the 178 food handlers, a total of 179 Enterobacterales were isolated from the 101 positive food handlers, out of which 46 (45.5%) were colonized by 2 or more Enterobacterales isolates from different type of specimen. The frequency of isolation in descending order is *E. coli* 23.2% (n=41), *Klebsiella* spp 18.1% (n=32), *Enterobacter*

spp 15.3% (n=27), *Citrobacter* spp 10.7% (n=19), *Raoultella* spp. 7.3% (n=13), *Serratia* spp 5.6% (n=10), *Salmonella* spp 3.9% (n=7), *Kluyvera* spp 3.9% (n=7), *Shigella* spp 2.8% (n=5), *Proteus* spp 2.8% (n=5), *Cronobacter* spp 1.7% (n=3), *Erwinia* spp 1.1% (n=2), *Pantoea* spp 1.1% (n=2), *Hafnia* spp (1.1% (n=2), and *Yersinia* spp 1.1% (n=2) (Table 3).

Table 4 shows the frequency and type of Enterobacterales isolated from hand swab samples of the food handlers. The Enterobacterales isolates include *E. coli* 6 (3.4%), *Klebsiella* spp 11 (6.1%), *Enterobacter* spp 6 (3.4%), *Citrobacter* spp 2 (1.1%), *Raoultella* spp 5 (2.8%), *Serratia* spp 5 (2.8%), *Kluyvera* spp 1 (0.6%), *Salmonella* spp 1 (0.6%), *Shigella* spp 2 (1.1%), *Cronobacter* spp 1 (0.6%), *Pantoea* spp 1 (0.6%), *Erwinia* spp 2 (1.1%), and *Hafnia* spp 1 (0.6%), while no Enterobacterales was isolated from the hand swab samples of 135 (75.4%) of the food handlers including those who were not sampled.

Table 3: Frequency of the Enterobacterales isolation among the food handlers in Nnewi metropolis, Nigeria

Enterobacterales	Frequency	Percentage
<i>Escherichia coli</i>	41	23.2
<i>Klebsiella</i> spp	32	18.1
<i>Enterobacter</i> spp	27	15.3
<i>Citrobacter</i> spp	19	10.7
<i>Raoultella</i> spp	13	7.3
<i>Serratia</i> spp	10	5.6
<i>Salmonella</i> spp	7	3.9
<i>Kluyvera</i> spp	7	3.9
<i>Shigella</i> spp	5	2.8
<i>Proteus</i> spp	5	2.8
<i>Cronobacter</i> spp	3	1.7
<i>Erwinia</i> spp	2	1.1
<i>Pantoea</i> spp	2	1.1
<i>Hafnia</i> spp	2	1.1
<i>Yersinia</i> spp	2	1.1
Total	179	100.0

Table 4: Type of Enterobacterales isolated from hand swab samples of food handlers in Nnewi metropolis, Nigeria

Enterobacterales	Frequency	Percentage
<i>Escherichia coli</i>	6	3.4
<i>Klebsiella</i> spp	11	6.3
<i>Enterobacter</i> spp	6	3.4
<i>Citrobacter</i> spp	2	1.1
<i>Raoultella</i> spp	5	2.8
<i>Serratia</i> spp	5	2.8
<i>Salmonella</i> spp	1	0.6
<i>Kluyvera</i> spp	1	0.6
<i>Shigella</i> spp	2	1.1
<i>Cronobacter</i> spp	1	0.6
<i>Erwinia</i> spp	2	1.1
<i>Pantoea</i> spp	1	0.6
<i>Hafnia</i> spp	1	0.6
No isolate/sample	135	75.4
Total	179	100.0

Table 5: Type of Enterobacterales isolated from nasal swab samples of food handlers in Nnewi metropolis, Nigeria

Enterobacterales	Frequency	Percentage
<i>Escherichia coli</i>	6	3.4
<i>Klebsiella</i> spp	7	3.9
<i>Enterobacter</i> spp	7	3.9
<i>Citrobacter</i> spp	9	5.0
<i>Raoultella</i> spp	1	0.6
<i>Serratia</i> spp	2	1.1
<i>Proteus</i> spp	4	2.2
<i>Shigella</i> spp	1	0.6
<i>Pantoea</i> spp	2	1.1
No isolate/sample	140	78.2
Total	179	100.0

Table 6: Type of Enterobacterales isolated from stool samples of food handlers in Nnewi metropolis, Nigeria

Enterobacterales	Frequency	Percentage
<i>Escherichia coli</i>	18	10.1
<i>Klebsiella</i> spp	10	5.5
<i>Enterobacter</i> spp	12	6.7
<i>Citrobacter</i> spp	7	3.9
<i>Raoultella</i> spp	5	2.8
<i>Serratia</i> spp	2	1.1
<i>Kluyvera</i> spp	3	1.8
<i>Salmonella</i> spp	5	2.8
<i>Proteus</i> spp	1	0.6
<i>Cronobacter</i> spp	1	0.6
<i>Hafnia</i> spp	1	0.6
<i>Yersinia</i> spp	2	1.1
No isolate/sample	112	62.6
Total	179	100.0

Table 7: Type of Enterobacterales isolated from urine samples of food handlers in Nnewi metropolis, Nigeria

Enterobacterales	Frequency	Percentage
<i>Escherichia coli</i>	11	6.1
<i>Klebsiella</i> spp	4	2.2
<i>Enterobacter</i> spp	2	1.1
<i>Citrobacter</i> spp	1	0.6
<i>Raoultella</i> spp	3	1.7
<i>Serratia</i> spp	1	0.6
<i>Kluyvera</i> spp	3	1.7
<i>Salmonella</i> spp	1	0.6
<i>Shigella</i> spp	2	1.1
<i>Cronobacter</i> spp	1	0.6
No isolate/sample	150	83.8
Total	179	100.0

Table 5 shows the frequency and type of Enterobacterales isolated from nasal swab samples of the food handlers. Enterobacterales isolated includes *E. coli* 6 (3.4%), *Klebsiella* spp 7 (3.9%), *Enterobacter* spp 7 (3.9%), *Citrobacter* spp 9 (5.0%), *Raoultella* spp 1 (0.6%), *Serratia* spp 2 (1.1%), *Proteus* spp 4 (2.2%), *Shigella* spp 1 (0.6%), and *Pantoea* spp 2 (1.1%), while no Enterobacterales was isolated from the nasal swab samples of 140 (78.2%) of the food handlers including those who were not sampled.

Table 6 shows the frequency and type of Enterobacterales isolated from stool samples of the food handlers. Enterobacterales isolated includes *E. coli* 18 (10.0%), *Klebsiella* spp 10 (5.5%), *Enterobacter* spp 12 (6.7%), *Citrobacter* spp 7 (3.9%), *Raoultella* spp 5 (2.8%), *Serratia* spp 2 (1.1%), *Kluyvera* spp 3 (1.8%), *Salmonella* spp 5 (2.8%), *Proteus*

spp 1 (0.6%), *Cronobacter* spp 1 (0.6%), *Hafnia* spp 1 (0.6%), and *Yersinia* spp 2 (1.1%), while no Enterobacterales was isolated from the stool samples of 112 (62.6%) of the food handlers including those who were not sampled.

Table 7 shows the frequency and type of Enterobacterales isolated from urine samples of the food handlers. Enterobacterales isolated includes *E. coli* 11 (6.1%), *Klebsiella* spp 4 (2.2%), *Enterobacter* spp 2 (1.1%), *Citrobacter* spp 1 (0.6%), *Raoultella* spp 3 (1.7%), *Serratia* spp 1 (0.6%), *Kluyvera* spp 3 (1.7%), *Salmonella* spp 1 (0.6%), *Shigella* spp 2 (1.1%), and *Cronobacter* spp 1 (0.6%), while no Enterobacterales was isolated from the urine samples of 150 (83.8%) of the food handlers including those who were not sampled.

Table 8: Knowledge of food handlers' carriage of bacteria and antibiotic use in Nnewi metropolis, Nigeria

Factors	Yes (%)	No (%)
Have you ever been tested for carriage of Enterobacteriaceae?	76 (42.7)	102 (57.3)
Have you ever taken antibiotics in the past 6 months?	108 (60.7)	70 (39.3)
Did you complete the antibiotics dosage?	26 (14.6)	152 (85.4)
Do you know what measures can be taken to prevent antibiotic resistance?	59 (33.1)	119 (66.9)

Table 9: Antimicrobial susceptibility profiles of Enterobacterales isolated from food-handlers in Nnewi metropolis, Nigeria

Antibiotics	Resistant (%)	Intermediate (%)	Susceptible (%)
Cefotaxime (5µg)	147 (82.0)	10 (5.6)	22 (12.3)
Nalidixic acid (30µg)	140 (78.0)	16 (8.9)	23 (12.8)
Amoxicillin clavulanate (30µg)	140 (78.0)	17 (9.5)	22 (12.3)
Cefuroxime (5µg)	136 (76.0)	18 (10.0)	25 (14.0)
Cefixime (5µg)	133 (74.0)	11 (6.1)	35 (19.6)
Imipenem/cilastatin (10/10µg)	132 (73.0)	21 (11.7)	26 (14.5)
Nitrofurantoin (300µg)	132 (73.0)	14 (7.8)	33 (18.4)
Ceftazidime-avibactam (30µg)	129 (72.0)	16 (8.9)	34 (19.0)
Ceftriaxone-sulbactam (45µg)	126 (70.4)	25 (14.0)	28 (15.6)
Levofloxacin (5µg)	124 (70.5)	28 (14.2)	27 (15.3)
Meropenem (10µg)	113 (63.1)	18 (10.0)	48 (26.8)
Ofloxacin (5µg)	107 (60.0)	27 (15.0)	45 (25.0)
Gentamicin (10µg)	105 (58.7)	33 (18.4)	41 (22.9)

Table 8 shows the knowledge of food handlers in Nnewi metropolis with respect to antibiotic use and previous testing for carriage, with 76 (42.7%) of the 178 food handlers reported to have been previously tested for carriage of Enterobacterales. However, 108 (60.7%) of the handlers had taken antibiotics in the past six months, while only 26 (14.6%) completed their prescribed courses. Furthermore, the table reveals a substantial knowledge gap regarding preventive measures against antibiotic resistance, with only 59 (33.1%) of the handlers aware of such measures, while 119 (66.9%) lack this crucial knowledge.

Table 9 provides an overview of the antimicrobial susceptibility profiles of Enterobacterales isolates. The data reveals high resistance rates among the Enterobacterales isolates to several commonly used antibiotics. Notably, the highest resistance rate was observed for cefotaxime (82.1%), followed by nalidixic acid (78.2%), and amoxicillin-clavulanate (78.2%). Resistance rates were equally high for other antibiotics including cefuroxime (76.0%), cefixime (74.0%), ceftazidime-avibactam (72.0%), ceftriaxone-sulbactam (70%), nitrofurantoin (73.0%), imipenem-cilastatin (73.0%), meropenem (63.1%), ofloxacin (60%),

levofloxacin (70.5%) and gentamicin (58.7%).

Discussion:

Until recently, it was thought that the ESBL-producing organisms were healthcare-associated or hospital-acquired pathogens, primarily affecting patients who had previously visited hospitals or other healthcare institutions for care. However, in recent years, Enterobacterales isolates that are potential ESBL producers have either moved from hospitals to the community and are being identified in seemingly healthy people who had never before interacted with the healthcare system.

The members of the Enterobacterales isolated in our study had previously been implicated in food borne diseases and the frequency described for *E. coli* (23.2%), *Klebsiella* spp (18.1%), *Enterobacter* spp (15.3%), and for other members of the family Enterobacterales have been reported in Ondo State and Awka south, Nigeria (9,10), where *E. coli*, *K. pneumoniae*, and *E. cloacae* were the top 3 most isolated pathogens, with frequencies of 17.9%, 8.9%, and 6.7%, respectively. Other studies have reported high prevalence of *E. coli* among food handlers (11), which may be

explained by the fact that *E. coli* is a typical commensal in the gastrointestinal tract and majority of the study participants appeared to be in good health and did not exhibit any symptoms of illness.

In our study, we isolated *Salmonella* but not *Shigella* from stool samples of the food handlers (although *Shigella* was isolated from other samples), which contrasts the finding of the study conducted in Kenya by Juma (11) but agrees with Omemu and Oloyede (12) who reported that 5.5% of food handlers employed by small companies in urban region of Abeokuta, Nigeria had *Salmonella* spp isolated from their stool samples. The differences in the findings of these studies could be due to variations in the personal cleanliness, food hygiene knowledge and practice or health of the food handlers. Our findings of not isolating *Shigella* from the stool samples also agrees with the reports of studies from Jordan (13) and Addis Ababa (14). However, *Shigella* prevalence of 14.7% and 5.04% were reported from Kano, Nigeria (16) and Haramaya, Eastern Ethiopia (17) respectively.

Our study showed high resistance rate among the Enterobacterales to several commonly used antibiotics. Notably, the highest resistance rate was reported for cefotaxime (82.1%) followed nalidixic acid (78.2%) and amoxicillin-clavulanate (78.2%). Resistance was equally high for other antibiotics including cefuroxime (76.0%), cefixime (74.0%), nitrofurantoin (73.0%), imipenem-cilastatin (73.0%), ceftazidime-avibactam (72.0%), levofloxacin (70.5%), ceftriaxone-sulbactam (70.0%), meropenem (63.1%), ofloxacin (60.0%), and gentamicin (58.7%). These high resistance rates may have occurred from widespread use or misuse of these antimicrobial agents as shown in our results where 14.6% of the 108 food handlers who had taken antibiotics in the previous 6 months did not complete their antimicrobial doses, a practice that has been associated with emergence of bacterial resistance.

The high antimicrobial resistance rate in our study will pose significant challenge to the treatment of infections caused by members of the family Enterobacterales in our environment. Some of the major risk factors for emergence and spread of antimicrobial-resistant strains worldwide are self-medication and failure of compliance with the recommended doses of the prescribed antibiotics (18). However, the high resistance rates observed in our study may not be explained by these factors alone, as other confounding factors along with inappropriate antimicrobial use may be in operation (19).

Our study is limited by the fact that we could only recruit 178 food handlers, which make the generalization of our study findings to be difficult. However, our findings remain valid for the study setting.

Conclusion:

The current study found that a significant number of asymptomatic food handlers were carriers of Enterobacterales strains, that are highly resistant to commonly used antibiotics. Antibiotic resistance was widely distributed across various sample sources. Accordingly, close monitoring of food handling processes is necessary to avoid foodborne illnesses and identify newly emerging antibiotic resistant bacteria, particularly in a developing country like Nigeria.

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Contributions of authors:

OCB designed the study, OCB wrote the protocol; OCA and OCB contributed to the literature search; OCA performed the statistical analysis of data; OCB, ONF and OCA contributed in discussion; OCB produced the initial manuscript draft; ONF supervised the work; OCB wrote the final manuscript; OCA proofread the manuscript and all authors approved the final manuscript submitted for publication.

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Conflict of interest

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