Antibiotic Susceptibility Profiling and Multiple Antibiotic Resistance (MAR) Index of Bacteria Isolated from Soils of Selected Waste Dump Sites in Rivers State, Nigeria

Wilcox, O.A*, Wemedo, S. A., Douglas, S. I. and Ugboma, C.J.
Department of Microbiology, Rivers State University, Port Harcourt, Nigeria.
*Corresponding Author: *oluyemisi.wilcox@ust.edu.ng*

ABSTRACT

Antibiotics are a class of antimicrobials specifically used to combat bacterial infections. This study was aimed at antibiotic susceptibility profiling and determination of multiple antibiotic resistance (MAR) index of bacteria isolated from soils of selected waste dump sites in Rivers State. Soil samples were collected from three dumpsites and three control locations for duration of twelve calendar months (May 2023 to April 2024). Bacteria were isolated, characterized and identified using standard microbiological techniques and antibiotic susceptibility tests were conducted on the isolates using the disk diffusion technique. The bacteria isolated were: *Rhizobacter* spp, *Kluyvera* spp, *Escherichia coli*, *Chryseomonas* spp, *Klebsiella* spp, *Shigella* spp, *Salmonella* spp, *Psuedomonas* spp, *Vibrio* spp, *Cronobacter* spp, *Bacillus* spp, *Paenibacillus* spp, *Nesterenkonia* spp, *Rathayibacter* spp, *Sinomonas* spp, *Actinomyces* spp, and *Staphylococcus* spp. The isolates exhibited high resistance to the antibiotics, showing multidrug resistance with multiple antibiotic resistance (MAR) index of 0.2 to 1.0. Highest resistance was exhibited by *Rhizobacter* spp and *Chryseomonas* spp. Highest MAR Index of 1.0 was observed in *Rhizobacter* spp, *Klebsiella* spp, *Chryseomonas* spp, *Bacillus* spp, *Nesterenkonia* spp, *Sinomonas* spp and *Actinomyces* spp. Lowest MAR Index of 0.2 was observed in *Kluyvera* spp. The most frequent MAR Index with 30.5 % prevalence was 0.8. This is an indication of overuse or inappropriate antibiotic use. The results of this study suggest that waste dump sites contain many bacteria that are resistant to antibiotics. This is of public health concern. There is a global health challenge now as there is a wide emergence of multidrug resistance within populations of microorganisms. It is recommended that the Government/ Health ministry educate the populace about the possible risk of indiscriminate antibiotic use. Antibiogram should also be conducted regularly with wider variation of antibiotics tested till antibiotics that the bacteria would be highly susceptible to are discovered.

Keywords: Waste Dump Sites, Bacteria, *Klebsiella* spp, Multiple Antibiotic Resistance (MAR) Index, Public Health.

Introduction

Waste management in Nigeria is not regulated. Most solid waste dumpsites are located close to residential areas. The wastes dumped have contents that may have detrimental potential and they cause pollution of the air, soil and water. There are scavengers and other waste handlers at the dumpsites. These and other people within the vicinity are at potential risk health wise as there are organisms which may be present which could cause infections/ diseases (Nasir, *et al*., 2024).

Antibiotics were initially described to be organic substances produced by various species of microorganisms (bacteria, fungi, actinomycetes) that hinder the development of other microorganisms and may destroy them in due time (Muteeb *et al*., 2023**)**.

The definition has been slightly modified because synthetic antibiotics that can accomplish functions identical to those performed by natural antibiotics have been produced (Kalidas and Agrawal, 2020). Antibiotics are much more frequently used than any other class of antimicrobials (Folarin and Adedoyin, 2024). Some antibiotics completely kill or destroy other bacteria (bactericidal) while others only hinder their growth (bacteriostatic). Although antibiotic generally refers to antibacterial, antibiotic compounds are differentiated as antibacterials, antifungals and antivirals to reflect the group of microorganisms they antagonize. The probable points of difference amongst the antibiotics may be physical, chemical, pharmacological properties, antibacterial spectra, and mechanism of action.

They have made it possible to cure diseases caused by bacteria, such as pneumonia, tuberculosis, and meningitis, and they save the lives of millions of people around the world (Etebu and Arikekpar, 2016).

It is crucial to perform antibiotic susceptibility tests because it guides the physician in the administration of antimicrobial medications. Antibiogram of isolates from soils of waste dumps is important to be able to observe sensitivity/ resistance trends among the isolates as resistant bacteria can be transferred to humans and ultimately to animals and vice versa (Salam *et al*., 2023). The aim of this study therefore, is antibiotic susceptibility profiling and the determination of the multiple antibiotic resistance (MAR) Index of some bacteria isolated from soils of selected waste dump sites in Rivers State.

Materials and Methods

Study Area

The study was conducted in Rivers State within Port Harcourt and Aluu. Samples were collected from three solid waste dumpsites and three control locations : Allu Dumpsite (4.90702ºN 6.96342ºE), Allu Control (4.90478ºN 6.96352ºE), Elijiji Dumpsite (4.83965ºN 7.0597ºE), Elijiji Control (4.83994ºN 7.05767ºE), Boskel Dumpsite (4.85888ºN 7.0821ºE) and Boskel Control (4.85861ºN 7.08197ºE), for twelve months from May 2023 to April 2024. Soil samples were collected with the aid of soil auger at depth of 0 – 15 cm. Soil samples were placed in sterile polythene bags and put into cool boxes containing ice and transported to the Microbiology Laboratory of the Rivers State University for analysis.

Characterization and Identification of Bacterial Isolates

Tenfold serial dilution of the soil samples were performed as described by Tantray *et* al. (2023). The 10^{-1} dilution was obtained by transferring one gram (1g) of soil sample into 9ml sterile normal saline. One milliliter (1.0 ml) was transferred to 9ml sterilized normal saline after shaking the 10^{-1} dilution to get the 10^{-2} dilution. Serial dilutions were made up to 10^{-4} dilution. An aliquot of zero point one milliliter (0.1ml) of the 10^{-4} dilution was inoculated onto sterilized media plates. Plates were incubated at 37ºC for 24 – 48 hours.

The colonies that developed were counted and calculated as colony forming units per gram (CFU/g). Streak plate technique was employed in subculturing the colonies that developed onto appropriate medium until pure isolates were obtained. Pure isolates were maintained on Nutrient agar slants and stored in the fridge until further analysis.

Morphological, microscopic and biochemical tests were conducted on the pure isolates. The tests carried out include Gram stain, motility, catalase, oxidase, indole, citrate, methyl red, Voges-Proskauer, urease, salt tolerance, nitrate reduction, sulphate reduction, starch hydrolysis and sugar fermentation for characterization (Chauhan and Jindal, 2020**)**. Reference was made to Bergey's Manual of Determinative Bacteriology (Holt, 1993) and confirmation was by Automated Biometric Identification System (ABIS) online.

Antibiotic Susceptibility Tests

Antibiotic susceptibility tests were performed to determine which antibiotics were effective against particular bacteria isolate. The disk diffusion method was used. The isolates were tested for susceptibility to the following antibiotics: NF- Nitrofurantoin (300µg), CXM- Cefuroxime (30µg), CRO- Ceftriaxone sulbactam (45µg), ACX- Ampiclox (10µg), ZEM- Cefexime (5µg), LBC-Levofloxacin (5µg), AUG- Amoxicilin Clavulanate (30µg), CTX- Cefotaxime (25µg), IMP- Imipenem/Cilastatin (10/10µg), OFX- Ofloxacin (5µg), GN- Gentamicin (10µg), NA- Nalidixic acid (30µg), ERY- Erythromycin(15µg), AZN- Azithromycin (15µg), CIP- Ciprofloxacin (5µg); with concentrations as recommended by the National Committee for Clinical Laboratory Standards (CLSI, 2023). Freshly grown bacteria (24hours on nutrient agar) were inoculated into sterile 10ml normal saline till turbidity matching 0.5 McFarland standard was obtained (Tagesu, 2018). Sterile swabs were used in inoculating Mueller Hinton agar plates, swabbing the plates in different directions. Antibiotic discs were aseptically placed with sterilized forceps onto the surface of the agar plates after a few minutes. Plates were incubated at 37ºC for 24 hours. Where zones of inhibition were observed, they were measured in millimeters on the underside of the plate. Diameters of zones were compared with the CLSI (2023) limits and the isolates were classified as Sensitive (S), Intermediate (I) or Resistant (R) (Gajic *et al*., 2022).

The results of the antibiotic susceptibility testing were used to determine the isolates' antibiotic profile and used to calculate the MAR (Multiple Antibiotic Resistance) Index (Chauhan and Jindal, 2020**)**.

Calculation of Multiple Antibiotic Resistance (MAR) Index

The a/b formula was used to determine the multiple antibiotic resistance (MAR) index for each isolate, where "a" is the total number of antibiotics to which the isolated bacteria is resistant and "b" is the total number of test antibiotics that the isolated bacteria were tested against (Cookey and Otokunefor, 2016).

Results

The tests for identification and characterization conducted on the isolates revealed the following bacteria were present in the soil samples: *Staphylococcus* spp, *Bacillus* spp, *Paenibacillus* spp, *Shigella* spp, *Salmonella* spp, *Vibrio* spp, *Escherichia coli*, *Pseudomonas* spp, *Klebsiella* spp, *Kluyvera* spp, *Rhizobacter* spp, *Cronobacter* spp, *Chryseomonas* spp, *Actinomyces* spp, *Nesterenkonia* spp, *Rathayibacter* spp and *Sinomonas* spp.

There were varying responses to the antibiotics with different percentages of susceptibility and resistance exhibited by the different bacteria. The antibiotic susceptibility profiles of some of the bacterial isolates to some antibiotics are presented in Tables 1 to 17.

Susceptibility and resistance exhibited by the identified bacteria were observed as follows: *Rhizobacter* spp was 47.1% and 41.2 % susceptible to Ofloxacin and Levofloxacin. There was 100% resistance to Nitrofurantoin, Cefuroxime, Ampiclox, Cefexime, Amoxicillin Clavulanate, Cefotaxime, Imipenem, Gentamicin and Nalidixic Acid respectively (Table 1).

Kluyvera spp was 46.4% and 35.7% susceptible to Gentamicin and Ofloxacin, showed resistance as follows: 92.9% to Ampliclox, 85.7% to Cefuroxime and Imipenem, 75% to Nitrofurantoin, Amoxicillin Clavunalate and Nalidixic Acid, 71.4% to Cefexime, 67.9% to Cefotaxime, 64.3% to Ceftriaxone Sulbactam and Levofloxacin respectively (Table 2).

Table 1: Antibiotic Susceptibility Profile of *Rhizobacter* spp isolated from soil (n = 17)

Antibiotics	Reaction of *Rhizobacter* spp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
NF	0	0	17 (100)
CXM	0	0	17 (100)
CRO	0	6 (35.3)	11 (64.7)
ACX	0	0	17 (100)
ZEM	0	0	17 (100)
LBC	7 (41.2)	5 (29.4)	5 (29.4)
AUG	0	0	17 (100)
CTX	0	0	17 (100)
IMP	0	0	17 (100)
OFX	8 (47.1)	3 (17.6)	6 (35.3)
GN	0	0	17 (100)
NA	0	0	17 (100)

Note: Numbers before the bracket represent the frequency while the numbers inside the bracket represent the percentage; n = Number of species;

Key: NF- Nitrofurantoin, **CXM-** Cefuroxime, **CRO-** Ceftriaxone sulbactam, **ACX-** Ampiclox, **ZEM-** Cefexime, **LBC-** Levofloxacin**, AUG-**Amoxicilin Clavulanate**, CTX-** Cefotaxime, **IMP-** Imipenem/Cilastatin, **OFX-** Ofloxacin, **GN-** Gentamicin, **NA-** Nalidixic acid, **ERY-** Erythromycin, **AZN-** Azithromycin, **CIP-** Ciprofloxacin.

Table 2: Antibiotic Susceptibility Profile of *Kluyvera* spp isolated from soil (n = 28)

Antibiotics	Reaction of *Kluyvera* spp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
NF	6 (21.4)	1 (3.6)	21 (75.0)
CXM	4 (14.3)	0	24 (85.7)
CRO	9 (32.1)	1 (3.6)	18 (64.3)
ACX	2 (7.1)	0	26 (92.9)
ZEM	4 (14.3)	4 (14.3)	20 (71.4)
LBC	7 (25.0)	3 (10.7)	18 (64.3)
AUG	7 (25)	0	21 (75)
CTX	8 (28.6)	1 (3.6)	19 (67.9)
IMP	4 (14.3)	0	24 (85.7)
OFX	10 (35.7)	3 (10.7)	15 (53.6)
GN	13 (46.4)	13 (46.4)	2 (7.1)
NA	5 (17.9)	2 (7.1)	21 (75.0)

Escherichia coli was 43.9% and 42.1% susceptible to Imipenem and Nitrofurantoin, exhibited resistance as follows: 98.2% to Cefotaxime, 68.4% to Nalidixic Acid, 63.2% to Cefuroxime, 61.4% to Ceftriaxone Sulbactam and Ampiclox respectively (Table 3).

Table 3: Antibiotic Susceptibility Profile of *Escherichia coli* isolated from soil (n = 57)

Antibiotics	Reaction of *Escherichia coli*		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
NF	24 (42.1)	8 (14.0)	25 (43.9)
CXM	17 (29.8)	4 (7.0)	36 (63.2)
CRO	10 (17.5)	12 (21.1)	35 (61.4)
ACX	5 (8.8)	7 (12.3)	45 (61.4)
ZEM	11 (19.3)	13 (22.8)	33 (57.9)
LBC	20 (35.1)	14 (24.6)	23 (40.4)
AUG	13 (22.8)	11 (19.3)	33 (57.9)
CTX	0	1 (1.8)	56 (98.2)
IMP	25 (43.9)	11 (19.3)	21 (36.8)
OFX	16 (28.1)	13 (22.8)	28 (49.1)
GN	14 (24.6)	12 (21.1)	31 (54.4)
NA	8 (14.0)	10 (17.5)	39 (68.4)

Chryseomonas spp was 50% and 40% susceptible to Levofloxacin and Ofloxacin and showed 100% resistance to Nitrofurantoin, Cefuroxime, Ceftriaxone Sulbactam, Ampiclox,, Cefexime, Amoxicillin Clavulanate, Cefotaxime, Imipenem, Gentamicin, Nalidixic Acid, Erythromycin, and Azithromycin respectively (Table 4).

Table 4: Antibiotic Susceptibility Profile of *Chryseomonas* spp isolated from soil (n = 10)

Antibiotics	Reaction of *Chryseomonas* spp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
NF	0	0	10 (100)
CXM	0	0	10 (100)
CRO	0	0	10 (100)
ACX	0	0	10 (100)
ZEM	0	0	10 (100)
LBC	5 (50.0)	2 (20.0)	3 (30.0)
AUG	0	0	10 (100)
CTX	0	0	10 (100)
IMP	0	0	10 (100)
OFX	4 (40.0)	4 (40.0)	2 (20.0
GN	0	0	10 (100)
NA	0	0	10 (100)

Klebsiella spp was 63.6% and 54.5% susceptible to Imipenem and Nitrofurantoin. Resistance was observed to Cefuroxime (100%), Nalidixic Acid (100%), Ampiclox (90.9%), Levofloxacin (72.7%) and Ceftriaxone Sulbactam (63.6%), (Table 5).

Table 5: Antibiotic Susceptibility Profile of *Klebsiella* spp isolated from soil (n = 22)

Antibiotics	Reaction of *Klebsiella* spp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
NF	12 (54.5)	1 (4.5)	9 (40.9)
CXM	0	0	22 (100)
CRO	5 (22.7)	3 (13.6)	14 (63.6)
ACX	0	2 (9.1)	20 (90.9)
ZEM	0	0	22
LBC	0	6 (27.3)	16 (72.7)
AUG	8 (36.4)	2 (9.1)	12 (54.5)
CTX	0	0	22 (100)
IMP	14 (63.6)	3 (13.6)	5 (22.7)
OFX	9 (40.9)	7 (31.8)	6 (27.3)
GN	7 (31.8)	4 (18.2)	11 (50.0)
NA	0	0	22 (100)

Shigella spp was 56.5% susceptible to Cefotaxime and Gentamicin. Resistance was observed as follows: 100% to Cefuroxime, Cefexime and Ofloxacin, 95.7% to Nitrofurantoin and 69.6% to Levofloxacin (Table 6).

Table 6: Antibiotic Susceptibility Profile of *Shigella* spp isolated from soil (n = 23)

Antibiotics	Reaction of *Shigella* spp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
NF	0	1 (4.3)	22 (95.7)
CXM	0	0	23 (100)
CRO	11 (47.8)	4 (17.4)	8 (34.8)
ACX	0	8 (34.8)	15 (65.2)
ZEM	0	0	23(100)
LBC	0	7 (30.4)	16 (69.6)
AUG	7 (30.4)	3 (13.0)	13 (56.5)
CTX	13 (56.5)	1 (4.3(	9 (39.1)
IMP	7 (30.4)	5 (21.7)	11 (47.8)
OFX	0	0	23 (100)
GN	13 (56.5)	4 (17.4)	6 (26.1)
NA	11 (47.8)	1 (4.3)	11 (47.8)

Salmonella spp was 57.1% and 47.6% susceptible to Imipenem and Ceftriaxone Sulbactam. Resistance was also observed: 100% to Nitrofurantoin, Amoxicillin Clavulanate and Cefotaxime, 95.2% to Cefuroxime, 90.5% to Ampiclox, 76.2% to Ofloxacin and 61.9% to Nalidixic Acid (Table 7).

Table 7: Antibiotic Susceptibility Profile of *Salmonella* spp isolated from soil (n = 21)

Antibiotics	Reaction of *Salmonella* spp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
NF	0	0	21 (100)
CXM	0	1	20 (95.2)
CRO	10 (47.6)	5 (23.8)	6 (28.6)
ACX	1 (4.8)	1 (4.8)	19 (90.5)
ZEM	9 (42.9)	7 (33.3)	5 (23.8)
LBC	2 (9.5)	7 (33.3)	12 (57.1)
AUG	0	0	21 (100)
CTX	0	0	21 (100)
IMP	12 (57.1)	6 (28.6)	3 (14.3)
OFX	2 (9.5)	3 (14.3)	16 (76.2)
GN	6 (28.6)	6 (28.6)	9 (42.9)
NA	4 (19.0)	4 (19.0)	13 (61.9)

Pseudomonas spp was 50% susceptible to Gentamicin and Ceftriaxone Sulbactam. Resistance was observed: 97.1% to Ampiclox, 85.3% to Amoxicillin Clavulanate, 76.5 to Nalidixic Acid, 73.5% to Nitrofurantoin and Levofloxacin, 70.6% to Ofloxacin and 67.6% Cefuroxime (Table 8).

Table 8: Antibiotics Susceptibility Profile of *Pseudomonas* spp isolated from soil (n = 34)

Antibiotics	Reaction of *Pseudomonas* spp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
NF	2 (5.9)	7 (20.6)	25 (73.5)
CXM	8 (23.5)	3 (8.8)	23 (67.6)
CRO	17 (50.0)	9 (26.5)	8 (23.5)
ACX	0	1 (2.9)	33 (97.1)
ZEM	13 (38.2)	5 (14.7)	16 (47.1)
LBC	8 (23.5)	1 (2.9)	25 (73.5)
AUG	3 (8.8)	2 (5.9)	29 (85.3)
CTX	15 (44.1)	5 (14.7)	14 (41.2)
IMP	11 (32.4)	5 (14.7)	18 (52.9)
OFX	3 (8.8)	7 (20.6)	24 (70.6)
GN	17 (50.0)	8 (23.5)	9 (26.5)
NA	6 (17.6)	2 (5.9)	26 (76.5)

Vibrio spp was 69% and 55.2% susceptible to Nalidixic acid and Ceftriaxone Sulbactam respectively Resistance was observed: 100% to Amoxicillin Clavulanate, 96.6% to Ampiclox, 89.7% to Nitrofurantoin, 75.9% to Cefuroxime, 65.5% to Levofloxacin and 62.1% to Cefexime (Table 9).

Table 9: Antibiotics Susceptibility Profile of *Vibrio* spp isolated from soil (n = 29)

Antibiotics	Reaction of *Vibrio* spp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
NF	0	3 (10.3)	26 (89.7)
CXM	4 (13.8)	3 (10.3)	22 (75.9)
CRO	16 (55.2)	7 (24.1)	6 (20.7)
ACX	0	1 (3.4)	28 (96.6)
ZEM	7 (24.1)	4 (13.8)	18 (62.1)
LBC	7 (24.1)	3 (10.3)	19 (65.5)
AUG	0	0	29 (100)
CTX	9 (31.0)	7 (24.1)	13 (44.8)
IMP	14 (48.3)	5 (17.2)	10 (34.5)
OFX	10 (34.5)	6 (20.7)	13 (44.8)
GN	11 (37.9)	6 (20.7)	12 (41.4)
NA	20 (69.0)	4 (13.8)	5 (17.2)

Cronobacter spp was 45.5% susceptible to Nitrofurantoin, Ceftriaxone Sulbactam, Levofloxacin and Gentamicin. Resistance was observed: 100% to Ampiclox, Amoxicilin Clavulanate and Nalidixic Acid, 90.9% to Cefo taxime and 63.6% to Cefuroxime (Table 10).

Table 10: Antibiotic Susceptibility Profile of *Cronobacter* spp isolated from soil (n = 11)

Antibiotics	Reaction of *Cronobacter* spp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
NF	5 (45.5)	3 (27.3)	3 (27.3)
CXM	0	4 (36.4)	7 ((63.6)
CRO	5 (45.5)	5 (45.5)	1 (9.1)
ACX	0	0	11 (100)
ZEM	0	6 (54.5)	5 (45.5)
LBC	5 (45.5)	1 (9.1)	5 (45.5)
AUG	0	0	11 (100)
CTX	0	1 (9.1)	10 (90.9)
IMP	0	6 (54.5)	5 (45.5)
OFX	3 (27.3)	3 (27.3)	5 (45.5)
GN	5 (45.5)	3 (27.3)	3 (27.3)
NA	0	0	11 (100)

Bacillus spp were susceptible to Imipenem (44%) and Amoxicilin Clavulanate (42%). Resistance was observed to Cefexime (98%), Ceftriaxone Sulbactam (88%), Cefuroxime (86%), Cefotaxime (82%), Levofloxacin(64%) & Azithromycin(62%) (Table 11).

Table 11: Antibiotic Susceptibility Profile of *Bacillus* sp isolated from soil (n = 50)

Antibiotics	Reaction of *Bacillus* sp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
GN	16 (32.0)	12 (24.0)	22 (44.0)
CTX	0	9 (18.0)	41 (82.0)
CRO	0	6 (12.0)	44 (88.0)
ZEM	0	1 (2.0)	49 (98.0)
LBC	9 (18.0)	9 (18.0)	32 (64.0)
CIP	16 (32.0)	14 (28.0)	20 (40.0)
IMP	22 (44.0)	6 (12.0)	22 (44.0)
CXM	4 (8.0)	3 (6.0)	43 (86.0)
AZN	8 (16.0)	11 (22.0)	31 (62.0)
OFX	17 (34.0)	12 (24.0)	21 (42.0)
ERY	16 (32.0)	6 (12.0)	28 (56.0)
AUG	21 (42.0)	3 (6.0)	26 (52.0)

Susceptibility was observed in *Paenibacillus* spp to Ciprofloxacin (46.7%) and Azithromycin (40%). There was 100% resistance to Gentamicin, Ceftriaxone Sulbactam, Cefexime and Cefuroxime, 86.7% to Erythromycin, 73.3% to Amoxicilin Clavulanate, 60% to Levofloxacin and Ofloxacin (Table 12).

Table 12: Antibiotic Susceptibility Profile of *Paenibacillus* sp isolated from soil (n = 15)

Antibiotics	Reaction of *Paenibacillus* sp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
GN	0	0	15 (100)
CTX	4 (26.7)	3 (20.0)	8 (53.3)
CRO	0	0	15 (100)
ZEM	0	0	15 (100)
LBC	0	6 (40.0)	9 (60.0)
CIP	7 (46.7)	3 (20.0)	5 (33.3)
IMP	2 (13.3)	5 (33.3)	8 (53.4)
CXM	0	0	15 (100)
AZN	6 (40.0)	5 (33.3)	4 (26.7)
OFX	5 (33.3)	1 (6.7)	9 (60.0)
ERY	0	2 (13.3)	13 (86.7)
AUG	4 (26.7)	0	11 (73.3)

Nesterenkonia spp were 22% susceptibile to Imipenem and Gentamicin. They were 100% resistant to Cefexime, Levofloxacin, Cefuroxime, Amoxicilin Clavulanate. There was also resistance to Gentamicin (77.8%), Azithromycin (72.2%), Ceftriaxone Sulbactam (66.7%), Ciprofloxacin (66.7%) (Table 13).

Table 13: Antibiotic Susceptibility Profile of *Nesterenkonia* spp isolated from soil (n =18)

Antibiotics	Reaction of *Nesterenkonia* spp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
GN	4 (22.2)	0	14 (77.8)
CTX	1 (5.6)	7 (38.9)	10 (55.6)
CRO	0	6 (33.3)	12 (66.7)
ZEM	0	0	18 (100)
LBC	0	0	18 (100)
CIP	1 (5.6)	5 (27.8)	12 (66.7)
IMP	4 (22.2)	10 (55.6)	4 (22.2)
CXM	0	0	18 (100)
AZN	0	5 (27.8)	13 (72.2)
OFX	1 (5.6)	9 (50.0)	8 (44.4)
ERY	2 (11.1)	8 (44.4)	8 (44.4)
AUG	0	0	18 (100)

Rathayibacter spp were susceptible to Ciprofloxacin (50%). They were 100% resistant to Gentamicin, Cefotaxime, Ceftriaxone Sulbactam, Cefexime, Imipenem and Cefuroxime. (Table 14).

Table 14: Antibiotic Susceptibility Profile of *Rathayibacter* spp isolated from soil (n =12)

Antibiotics	Reaction of *Rathayibacter* spp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
GN	0	0	12 (100)
CTX	0	0	12 (100)
CRO	0	0	12 (100)
ZEM	0	0	12 (100)
LBC	0	7 (58.3)	5 (41.7)
CIP	6 (50.0)	4 (33.3)	2 (16.7)
IMP	0	0	12 (100)
CXM	0	0	12 (100)
AZN	4 (33.3)	4 (33.3)	4 (33.3)
OFX	0	7 (58.3)	5 (41.7)
ERY	3 (25.0)	3 (25.0)	6 (50.0)
AUG	4 (33.3)	1 (8.3)	7 (58.3)

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Sinomonas spp were susceptible to Ciprofloxacin (50%), Levofloxacin (40.9%). Resistance was observed for the following antibiotics: 100% to Cefexime, Imipenem, Cefuroxime and Amoxicilin Clavulanate, 86.4% to Cefotaxime, 81.8% to Ceftriaxone Sulbactam, 77.3% to Azithromycin and 68.2% to Gentamicin (Table 15).

Table 15: Antibiotic Susceptibility Profile of
***Sinomonas* spp isolated from soil (n = 22)**

Antibiotics	Reaction of *Sinomonas* spp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
GN	2 (9.1)	5 (22.7)	15 (68.2)
CTX	0	3 (13.6)	19 (86.4)
CRO	3 (13.6)	1 (4.5)	18 (81.8)
ZEM	0	0	22 (100)
LBC	9 (40.9)	1 (4.5)	12 (54.5)
CIP	11 (50.0)	0	11 (50.0)
IMP	0	0	22 (100)
CXM	0	0	22 (100)
AZN	0	5 (22.7)	17 (77.3)
OFX	8 (36.4)	7 (31.8)	7 (31.8)
ERY	7 (31.8)	2 (9.1)	13 (59.1)
AUG	0	0	22 (100)

Actinomyces spp were 45.5% susceptible to Cefuroxime and Amoxicilin Clavulanate. They were 100% resistant to Cefotaxime and Cefexime, 72.3% resistant to Ceftriaxone Sulbactam, 63.6% to Gentamicin, Levofloxacin and Ciprofloxacin (Table 16).

Staphylococcus spp exhibited susceptibility of 80.3%, 46.1%, 43.4% and 40.8% to Amoxicillin Clavulanate, Ofloxacin, Levofloxacin, Ciprofloxacin respectively was observed. Resistance was observed to Cefexime (89.5%), Azithromycin (81.6%), Cefuroxime (77.6%), Erythromycin (77.6%), Imipenem (64.5%) (Table 17).

The Multiple antibiotic resistance (MAR) index of the bacterial isolates ranged from 0.2 – 1.0. High MAR Index of 0.8 was the most frequent, with 30.5 % prevalence, followed by isolates with 0.7(20.4%), 0.6(15.7%), 0.5(13.1), 0.4(8.3%). 0.9(6.0%), 1.0(3.0%) and 0.3(2.8%). Isolate with 0.2 had the least occurrence (0.2%) The MAR Index of the isolated bacteria are presented in Tables 18 to 20.

Table 16: Antibiotic Susceptibility Profile of
***Actinomyces* spp isolated from soil (n = 11)**

Antibiotics	Reaction of *Actinomyces* spp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
GN	3 (27.3)	1 (9.1)	7 (63.6)
CTX	0	0	11 (100)
CRO	0	3 (27.3)	8 (72.7)
ZEM	0	0	11 (100)
LBC	2 (18.2)	2 (18.2)	7 (63.6)
CIP	4 (36.4)	0	7 (63.6)
IMP	2 (18.2)	3 (27.3)	6 (54.5)
CXM	5 (45.5)	1 (9.1)	5 (45.5)
AZN	0	5 (45.5)	6 (54.5)
OFX	0	5 (45.5)	6 (54.5)
ERY	4 (36.4)	4 (36.4)	3 (27.3)
AUG	5 (45.5)	0	6 (54.5)

Table 17: Antibiotic Susceptibility Profile of
***Staphylococcus* spp isolated from soil (n =76)**

Antibiotics	Reaction of *Staphylococcus* spp		
	Susceptible n(%)	Intermediate n(%)	Resistant n(%)
GN	29 (38.2)	12 (15.8)	35 (46.1)
CTX	24 (31.6)	15 (19.7)	37 (48.7)
CRO	30 (39.5)	13 (17.1)	33 (41.4)
ZEM	1 (1.3)	7 (9.2)	68 (89.5)
LBC	33 (43.4)	9 (11.8)	34 (44.7)
CIP	31 (40.8)	8 (10.5)	37 (48.7)
IMP	18 (23.7)	9 (11.8)	49 (64.5)
CXM	2 (2.6)	15 (19.7)	59 (77.6)
AZN	8 (10.5)	6 (7.9)	62 (81.6)
OFX	35 (46.1)	17 (22.4)	24 (31.6)
ERY	10 (13.2)	7 (9.2)	59 (77.6)
AUG	61 (80.3)	3 (3.9)	12 (15.8)

Note: Numbers before the bracket represent the frequency while the numbers inside the bracket represent the percentage; n = Number of species;

Key: NF- Nitrofurantoin, **CXM-** Cefuroxime, **CRO-** Ceftriaxone sulbactam, **ACX-** Ampiclox, **ZEM-** Cefexime, **LBC-** Levofloxacin**, AUG-**Amoxicilin Clavulanate**, CTX-** Cefotaxime, **IMP-** Imipenem/Cilastatin, **OFX-** Ofloxacin, **GN-** Gentamicin, **NA-** Nalidixic acid, **ERY-** Erythromycin, **AZN-** Azithromycin, **CIP-** Ciprofloxacin.

Table 18: Multiple Antibiotic Resistance Index and Resistance (%) of the Bacterial Isolates from Soil Sample

MAR Index	Bacterial Isolates from Soil Sample 1				
	Rhizobacter spp	*Kluyvera* spp	*Escherichia coli*	*Klebsiella* spp	*Chryseomonas* spp
0.1					
0.2		1(3.6)			
0.3		1(3.6)	1(1.8)		
0.4			8(14.0)		
0.5		4(14.3)	15(26.3)		
0.6		1(3.6)	12 (21.0)	6(27.3)	
0.7		4 (14.3)	9(15.8)	8(36.4)	
0.8	12(70.6)	11(39.2)	11(19.3)	7(31.8)	6 (60.0)
0.9	2 (11.8)	5(17.8)	1(1.8)		3(30.0)
1.0	3(17.6)	1(3.6)		1(4.5)	1(10.0)

Note: Numbers before the bracket represent the frequency while numbers inside the bracket represent the percentage.

Table 19: Multiple Antibiotic Resistance Index and Resistance (%) of the Bacterial Isolates from Soil Sample 2

MAR Index	Bacterial Isolates from Soil Sample 2				
	Shigella spp	*Salmonella* spp	*Cronobacter* spp	*Vibrio* spp	*Pseudomonas* spp
0.1					
0.2					
0.3		1(4.7)	1(9.1)		3(8.6)
0.4	2 (8.7)		1(9.1)	7(24.1)	3(8.6)
0.5	2 (8.7)	2 (9.5)	5(45.4)	6 (20.7)	7(20)
0.6	6 (26.1)	5 (23.8)	1 (9.1)	5 (17.2)	4(11.4)
0.7	4 (17.4)	7(33.3)	1(9.1)	7(24.1)	10(28.5)
0.8	9 (39.1)	4(19.2)		4(13.8)	7(20)
0.9		2 (9.5)	2(18.2)		1(2.9)
1.0					

Note: Numbers before the bracket represent the frequency while numbers inside the bracket represent the percentage.

Table 20: Multiple Antibiotic Resistance Index and Resistance (%) of the Bacterial Isolates from Soil Sample 3

MAR Index	Bacterial Isolates from Soil Sample 3						
	Bacillus spp	*Paenibacillus* spp	*Nesterenkonia* spp	*Rathayibacter* spp	*Sinomonas* spp	*Actinomyces* spp	*Staphylococcus* spp
0.1							
0.2							
0.3	1(2.0)					1(9.1)	5(6.6)
0.4	7(14.0)	2(13.3)				2(18.2)	9 (11.8)
0.5	7(14.0)	1(6.7)	2(11.1)		1(4.5)	1(9.1)	10 (13.1)
0.6	11(22.0)		1(5.6)	2(16.6)	1(4.5)	3(27.2)	16 (21.0)
0.7	9 (18.0)	3(20)	6(33.3)	5(41.7)	5(22.7)	1(9.1)	15(19.7)
0.8	12 (24.0)	8(53.3)	8(44.4)	5(41.7)	12(54.5)	1(9.1)	19(25)
0.9		1(6.7)			2(9.0)		2(2.6)
1.0	3(6.0)		1(5.6)		1(4.5)	2(18.2)	

Note: Numbers before the bracket represent the frequency while numbers inside the bracket represent the percentage.

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Discussion

This present study has revealed that, most of the bacteria isolated in this study exhibited resistance to the antibiotics they were tested against. There was susceptibility at varying levels to the antibiotics tested but there was more resistance than susceptibility to the antibiotics during this study. The highest susceptibility observed was 80.3% by *Staphylococcus* spp to Amoxicillin Clavulanate. Lower susceptibilities were exhibited by other bacteria to the antibiotics tested: *Vibrio* spp was 69% susceptible to Nalidixic acid, *Klebsiella* spp was 63.6% to Imipenem/cilastin. Susceptibilities exhibited to the antibiotics by some of the other bacteria were: *Salmonella* 57.1% to Imipenem/cilastin, *Shigella* 56.5% to Cefotaxime, *Pseudomonas* 50% to Gentamicin and Ceftriaxone Sulbactam, *Rhizobacter* 47.1% to Ofloxacin, *Kluyvera* 46.4% to Gentamicin, *Actinomyces* 45.5% to Cefuroxime, *Bacillus* 44% to Imipenem/cilastin and *Escherichia coli* 43.9% to Imipenem/cilastin.

The high resistance pattern observed within the bacterial isolates could be because antibiotics are casually used by humans and on animals, resulting over time in development of resistance. Resistance can be transferred through food chain, human to animal contact and environment (Al-Ghamdi *et al*., 2023). This eventually gets into waste dump sites as part of the components of the dumpsites are of faecal origin, which are produced by humans and animals that have bacteria that have developed resistance to antibiotics.

The observed results could also be possibly because some of them possess innate ability to resist antibiotics or have acquired resistance to antibiotics mainly as a result of misuse. Antibiotics are also used on animals to prevent disease and to enhance growth. This also leads to increase in antibiotic resistance. Resistance to antimicrobials has become a great threat to public health and is gradually becoming a global epidemic (Adegoke *et al*., 2017). The varied levels of susceptibility and resistance to antibiotics observed could be because of the different sources of isolation and differences in strains within the bacterial species in all the sampling locations (Osman and Elhaig, 2020).

In this study, it was observed that the identified bacteria had susceptibility range to the tested antibiotics of 1.3% to 100% and resistance range of 5.9% to 100%.

This varies from the study of Bashir *et al*., (2021) who observed susceptibility range of 20% to 100% and resistance range of 13.3% to 100% to tested antibiotics. They also observed that none of the bacteria that they identified were completely resistant or sensitive to all the antibiotics tested. Some of the isolates identified in this study could be potential pathogens. The antibiotics that are supposed to be used in treating illnesses caused by them have been observed from this study to be inadequate to treat them because of high level of resistance exhibited by most of them. Many have developed/acquired resistance to the antibiotics. Resistance observed in the bacteria to the antibiotics may be traced to ability to produce enzymes which can modify or inactivate the antibiotics (Egorov *et al*., 2018).

Therefore, as wastes are continually discharged into waste dumpsites, there is possibility of continuous spread of bacteria that are resistant to antibiotics and it becomes more and more difficult to treat infections further aggravating the global health problems as a result of antibiotic resistance (Breijyeh *et al*., 2020).

It was observed that the isolates in this study showed high resistance to ß-lactam antibiotics. This agrees with the studies of Nyandjou *et al.,* (2019) and Olusolape *et al.* (2023) who also observed resistance to ß-lactam antibiotics. Olusolape *et al.* (2023) also reported that all bacterial isolates were susceptible to Gentamicin and Ofloxacin. Most of the bacterial isolates in this study were resistant to Gentamicin and Ofloxacin. It is noteworthy that most of the isolates in this study were resistant to most of the antibiotics tested. This is of great concern as it is potentially dangerous for public health. When antibiotics are resistant, it implies that it would be difficult to treat diseases and unusual diseases could emerge (Nwobodo *et al*., 2022). The antibiotic resistance exhibited may be linked to acquisition of antibiotic resistance genes (Brown-Jaque *et al*., 2015).

Antibiotics that had been effective in treatment are now ineffective in treating the infections they were able to adequately treat in the past. The dumpsites have become prospective 'springs' of bacteria that are antibiotic resistant. In developing countries including Nigeria, there is a high level of non completion of medication cycles, there are sub-standard drugs and self medication (Akinnawo, *et al*., 2021). These may also contribute to development of antibiotic resistance.

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Monitoring of antibiotic resistance of isolates from dump site soils should be done on a continuous basis in order to detect pathogens that are resistant and are just emerging and the resistance trends.

Multiple Antibiotic Resistance (MAR) is the resistance of isolate to at least four different antibiotics or more than two different classes of antibiotics. Multiple Antibiotic Resistance (MAR) index: the a/b formula was used to determine the multiple antibiotic resistance index (MAR index) for each isolate, where "a" is the total number of antibiotics to which the isolated organism is resistant and "b" is the total number of test antibiotics that the isolated organisms were tested against (Cookey and Otokunefor, 2016). The antibiotic susceptibility test results were used to calculate the MAR index. A high-risk source of contamination where antibiotics are primarily used is indicated by a MAR index value greater than 0.2, whereas a source associated with low antibiotic use is indicated by a MAR index value less than 0.2 (Ayandele *et al*., 2020). The results of the antibiotic susceptibility testing were used to determine the isolates' antibiotic profile and multidrug resistance. It was observed that all the isolates in this study were multidrug resistant with multiple antibiotic resistance index (MAR Index) 0.2 to 1.0. High MAR Index of 0.8 was the most frequent, with 30.44 % prevalence. This is in agreement with Odum *et al.* (2020) who reported MAR Index of 0.8 for 31.15% of the tested isolates. Nyandjou *et al*., (2019) reported MAR Index of 0.2 to 0.7. The results of this study suggest widespread antibiotic resistance. The bacteria exhibited high resistance against all classes of antibiotics tested: β lactams, quinolones, aminoglycosides, macrolides and nitrofuran. MAR index was greater than 0.2, which is an indication of overuse or inappropriate antibiotic use. This suggests possible spread of persistent bacterial infections among the resident population. Exhibition of bacterial resistance to multiple drugs imply that the antibiotics are no longer potent and can no longer treat sickness/ illness (Nwobodo *et al*., 2022).

Multi-drug resistant pathogenic bacteria have emerged in recent time presumably due to the extensive use of chemotherapeutic agents in both humans and animals. The Multiple Antibiotic Resistance (MAR) Index is used in assessing the risk of bacteria indicating areas with high antibiotic abuse rates. It helps understand the broader impacts of antibiotic resistance spread.

Bacteria with a high MAR index are likely from environments like soil, hospitals or water (Chilaka *et al*., 2022, Osundiya *et al*., 2023). These environments contain more of these resistance genes (Perry *et al*., 2014)**.** Multidrug resistance within populations of microorganisms has become a great global challenge. The microorganisms have devised strategies to avoid the effects of antibiotics (Mwaikono *et al.,* 2015). Studies have reported that dumpsites 'shelter' large populations of multidrug resistant microorganisms (Mwaikono *et al.,* 2015; Idahosa *et al.*, 2017)

Conclusion

The bacteria isolated from the soils of the waste dumpsites in this present study exhibited high resistance to the antibiotics tested. Highest susceptibility observed was by *Staphylococcus* spp: 80.3% to Amoxicillin Clavulanate. Highest resistance of 100% was exhibited by *Rhizobacter* spp and *Chryseomonas* spp to Nitrofurantoin, Cefuroxime, Ampiclox, Cefexime, Amoxicilin Clavulanate, Cefotaxime, Imipenem, Gentamicin and Nalidixic acid.

The bacterial isolates were multidrug resistant. MAR Index was from 0.2 to 1.0, with the most frequent being 0.8 which suggests that antibiotics are misused and this could result in escalation of antibiotic resistance. It is recommended that the Government/ Ministry of Health should educate the populace about the possible risk of indiscriminate antibiotic use. Antibiogram should also be conducted regularly with wider variation of antibiotics tested till antibiotics that the bacteria would be highly susceptible to are discovered.

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