

**African Journal of Life Sciences Research  
(AJLSR)**

E-ISSN 2978-3496 (Online); ISSN 2978-3488 (Print)

Indexed by SABINET

Volume 1, Number 1, June 2025

Pp 9-36

**Molecular Characterization and Antibiotic Susceptibility  
of Bacteria Associated with Cassava Farmlands from  
Igbariam Rural Communities in Anambra State, Nigeria**

DOI: <https://doi.org/10.31920/2978-3496/2025/v1n1a1>

**Ruth Asikiya Afunwa**

*Department of Pharmaceutical Microbiology and Biotechnology  
Faculty of Pharmaceutical Sciences  
Chukwuemeka Odumegwu Ojukwu University  
Igbariam Anambra State, Nigeria  
[ra.afunwa@coou.edu.ng](mailto:ra.afunwa@coou.edu.ng)  
[dr Ruthafunwa@yahoo.com](mailto:dr Ruthafunwa@yahoo.com)*

**Frances Ngozi Olisaka**

*Department of Biological Sciences  
Faculty Of Natural Sciences and Environmental Studies  
Godfrey Okoye University  
Enugu State, Nigeria*

**Ekenedilichukwu Godson Igboamazu**

*Department of Pharmaceutical Microbiology and Biotechnology  
Faculty of Pharmaceutical Sciences  
Chukwuemeka Odumegwu Ojukwu University  
Igbariam Anambra State, Nigeria*

**Mmesoma Vivian Ikezuagu**

*Department of Pharmaceutical Microbiology and Biotechnology  
Faculty of Pharmaceutical Sciences  
Chukwuemeka Odumegwu Ojukwu University  
Igbariam Anambra State, Nigeria*

**Martin Chukwunonso Nwofia**

*Department of Pharmaceutical Microbiology and Biotechnology  
Faculty of Pharmaceutical Sciences  
Chukwuemeka Odumegwu Ojukwu University  
Igbariam Anambra State, Nigeria*

**Philip Chibuikem Onwukwe**

*Department of Pharmaceutical Microbiology and Biotechnology  
Faculty of Pharmaceutical Sciences  
Chukwuemeka Odumegwu Ojukwu University  
Igbariam Anambra State, Nigeria*



**Francis Ayodele Gbadamosi**

*Department of Biological Sciences  
Faculty of Natural Sciences and Environmental Studies  
Godfrey Okoye University  
Enugu State, Nigeria*

---

**Abstract**

Cassava (*Manihot esculenta* Crantz) is a key staple crop for millions of people worldwide, particularly in tropical and subtropical regions. It is well-known for its ability to thrive in poor soil and drought conditions, making it a vital resource for food security especially in third world nations. However, the economic viability of cassava farming is frequently threatened by microbial pathogens that lead to diseases like cassava bacteria blight and root rot. The aim of this study is to carry out molecular characterization and antibiotic characterization of bacteria associated with cassava tubers in the rural communities at Igbariam, Anambra state, Nigeria. Cassava tubers were collected from five different farmlands and were processed by fermentation. The fermented cassava tubers were cultured in MacConkey, cetrimide, and mannitol salt agars. The isolates were identified by their morphological features, biochemical tests, DNA analysis and sequencing. Antibiotic susceptibility testing was performed on Mueller-Hinton agar after standardizing to 0.5 McFarland standard. The diameter of the zones of inhibition was measured (in mm) after incubation and the results interpreted by EUCAST charts. A total of 42 isolates comprising *Alcaligenes faecalis* (14) 33.3%, *Pseudomonas*

*aeruginosa* (13) 30.95% and *Pseudomonas putida* (15) 35.71% were identified. The isolates were resistant to cefixime, nitrofurantoin, ampicillin, amoxicillin-clavulanate, ceftriaxone, imipenem, and cefuroxime. The isolates showed sensitivity to gentamicin, azithromycin, ofloxacin and levofloxacin. The findings contribute to the understanding of some microorganisms that make up the microbiome associated with cassava tubers. By studying these bacteria, beneficial microorganisms that promote plant health, enhance nutrient uptake, or provide natural resistance against pathogens can be identified.

**Keywords:** *Cassava, Bacteria, Disease, PCR, DNA analysis*

## Introduction

Food security remains a pressing issue in both developed and developing nations, intensifying global focus on agriculture. Beyond food production, agriculture promotes environmental sustainability, biodiversity, and economic growth, contributing to hunger eradication, resource management, and job creation (Alabi et al., 2011; Obi et al., 2022; Yadav et al., 2022; Younas et al., 2022; Chinyere et al., 2022).

Cassava (*Manihot esculenta* Crantz or *Manihot utilissima* Phol), a key crop in Africa, Asia, and Latin America, plays a vital role in food and economic security (Bayata, 2019; Halake & Chinthapalli, 2020; Simonyan, 2015; Zhou et al., 2023). It is widely processed into food products—flour, garri, fufu, sweeteners—and industrial items like textiles and adhesives (Adebayo-Oyetoro et al., 2013; Ono & Tanimaki, 2021). Despite its utility, cassava contains toxic compounds such as linamarin and cyanogenic glycosides, which require proper processing to avoid health risks (Adebayo-Oyetoro et al., 2013).

Cassava is commercially cultivated due to its nutritional value and adaptability for food processing (Balogun et al., 2021; Simonyan, 2015). Its high demand underscores the need to understand the microbial communities associated with cassava tubers, as these can influence both crop health and food safety. Advanced molecular tools like high-throughput sequencing and metagenomics allow in-depth analysis of microbial populations, revealing both culturable and non-culturable species (Bokulich et al., 2023; Koren et al., 2023., De Souza et al., 2023; Singh et al., 2021).

Molecular characterization identifies both pathogens and beneficial microbes involved in spoilage, storage, and disease resistance. This information supports improved breeding and disease management

strategies (Oduor et al., 2022; Wu et al., 2023). Integrating techniques such as PCR, NGS, and metagenomics is key to sustainable cassava production.

This study aims to characterize bacterial isolates and their antibiotic resistance profiles in cassava tubers from Igbariam, Awkuzu, Ukwulu, Umudioka, and Otoko. It will expand the limited data on cassava's microbial ecology and guide future agricultural practices (Marín et al., 2023).

## **Materials and Methods**

### **Study Area**

The survey was conducted across five different zones within the Awkuzu metropolis and its environs in Anambra State, Nigeria. These zones include the cassava farmlands located in Chukwuemeka Odumegwu Ojukwu University (COOU), Igbariam; as well as in the communities of Awkuzu, Ukwulu, Umudioka, and Otoko. The study area lies within the geographical coordinates of latitudes 5°59.99'–6°00.00'N and longitudes 6°13'–6°56'E (Ezenwaji et al., 2017). This region is situated in Southeastern Nigeria and is known for its significant cassava production, owing to its fertile soils and favourable tropical climate. The study was carried out between June and August 2024 at the Pharmaceutical Microbiology and Biotechnology Laboratory of the Faculty of Pharmaceutical Sciences in the University.

### **Sample Collection and Processing**

Ten cassava tubers were collected from five zones—COOU Igbariam, Awkuzu, Ukwulu, Umudioka, and Otoko—in Anambra East LGA, Anambra State, Nigeria. Only healthy, insect-free tubers were selected. Samples were placed in sterile bags, labeled, and transported to the Pharmaceutical Microbiology and Biotechnology Laboratory, COOU (Renner et al., 2024). Tubers were aseptically peeled, washed, and cut into approximately 3 cm pieces, then fermented in sterile vessels with distilled water for four days at 25 °C (Balogun et al., 2022). The samples from the soaked cassava water were serially diluted to give tenfold ( $10^{-3}$ ) dilutions. Each 1 ml of the cassava water sample was agitated with 9 ml of distilled water to ensure a homogenous mixture. Subsequently, a tenfold serial dilution of the homogenates was made in nutrient broth such that each

broth diluent test tube contained 9 ml. Afterwards, the broth sample tubes were incubated at 35 °C for 24–48 hours (Chetan *et al.*, 2017).

### **Preparation of Culture Media**

All media used in this study were prepared and sterilized according to the manufacturer's instructions. The media used are Nutrient broth, Nutrient Agar, MacConkey Agar, Mannitol Salt Agar, Cetrimide Agar and Mueller-Hinton Agar.

### **Microbiological analysis**

With the aid of sterile syringes, 0.2 ml of the respective diluents of broth-cultured specimens were aseptically collected and inoculated onto the surfaces of the various solidified Nutrient agar plates. Thereafter, it was incubated for 24 hours at 37°C. The number of colonies on the plates with distinct characteristics after incubation were noted and counted (Nwakoby *et al.*, 2021).

### **Isolation of the Pure Cultures of Bacteria**

With the aid of a sterile wire loop, a colony from each respective Nutrient agar plates was picked and streaked accordingly in a series of parallel and non-overlapping lines on the surfaces of the well-groomed and labeled sterile petri dishes containing MacConkey Agar, Mannitol Salt Agar, and Cetrimide Agar, and incubated for 24–48 hours at 35°C (Chetan *et al.*, 2017).

### **Identification of Bacteria Isolates**

The pure bacterial isolates were identified based on their morphological and biochemical analysis. Morphological tests like color, shape, height, consistency, and margin assessments as well as biochemical tests were conducted to confirm the results obtained from the examination of the pure culture isolates (Parija, 2012).

### **Indole Test**

The indole test involved incubating isolates in peptone water at 37 °C for 24 hours, adding Kovac's reagent, and observing colour change. A red ring indicated a positive result (Chinyere *et al.*, 2022).

## Citrate Utilization Test

The isolates were inoculated by stab technique onto a slope of Simmons's citrate solid media and incubated at 37°C for 24 hours. Growth with a blue colour on the slant indicates a positive test and no growth or growth without any colour change indicates a negative test (Nwakobyet *al.*, 2021; Parija, 2012).

## Oxidase Test

A 1% solution of oxidase reagent, freshly prepared was soaked onto a piece of filter paper and then moistened with sterile distilled water. A sterile wire loop was used to pick isolates and spread them over the filter papers. Formation of deep purple colour change within 10 seconds (an indophenol blue), indicates a positive test for oxidation complement (Chinyere *et al.*, 2022; Parija, 2012).

## Catalase Test

About 5 drops of 3% hydrogen peroxide ( $H_2O_2$ ) were emulsified with a 24-hour-old sample colony on a sterile test slide. The slides were placed against a dark background and observed for immediate effervescence or bubbles representing a positive test because of the breakdown of  $H_2O_2$  by the catalase enzyme to produce oxygen bubbles (Chinyere *et al.*, 2022; Parija, 2012).

## Antibiotics Susceptibility Testing (AST)

Antibiotic susceptibility testing was performed using the Kirby-Bauer disk diffusion method. Standardized bacterial inoculum was swabbed on Mueller-Hinton agar plates. Antibiotic-impregnated discs were placed on the surface and incubated at 37°C for 18–24 hours. The susceptibility of each isolate to each antibiotic was shown by a clear zone of growth inhibition measured millimeters and they were interpreted using a standard chart, EUCAST 2023 (European Committee on Antimicrobial Susceptibility Testing).

## **Molecular Characterization of Bacteria Isolates**

### **Bacterial DNA Extraction**

Bacterial cells were pelleted by centrifuging 5 mL of culture at 10,000 x g for 12 minutes and re-suspended in 500 µL of guanidine hydrochloride. Proteinase K was added for protein digestion and equal volume ethanol was added to the lysate, and the solution was transferred to a spin column. Centrifugation at 10,000 x g allowed DNA to bind to the column. Purified DNA was eluted by adding 70 µL of TE buffer and centrifuging.

### **PCR Amplification**

Universal bacterial primers 27F and 1492R, which target the 16S rRNA gene, were prepared in a cocktail 27F (5'- **GAGTTTGATCMTGGCTCAG-3'**) and 1492R (5'**TACGGYTACCTTGTTACGACTT-3'**). The total reaction volume was 25 µL, including template DNA, primers, and PCR Master Mix. The thermal cycling conditions were set as follows: Initial denaturation at 94°C for 5 minutes, 35 cycles of denaturation at 94°C for 30 seconds, annealing at 56°C for 30 seconds, and extension at 72°C for 45 seconds and Final extension at 72°C for 7 minutes, followed by an indefinite hold at 4°C (Green & Sambrook, 2012).

### **Agarose Gel Electrophoresis**

A 2% agarose gel was prepared by dissolving agarose in TAE buffer and adding ethidium bromide for DNA visualization. PCR products were loaded into the gel and electrophoresed at 100V for 35 minutes. A 100-bp DNA ladder was used as a marker. DNA bands were visualized under UV light using an Accuris UV Transilluminator (Green & Sambrook, 2012).

### **Sequencing**

PCR products were cleaned enzymatically using EXOSAP and sequenced using the Brilliant Dye™ Terminator Cycle Sequencing Kit V3.1. The sequences were analyzed using a BLAST search to identify the bacterial species (Green & Sambrook, 2012).

Results

Identified Bacteria Isolates from Specimens Cultures

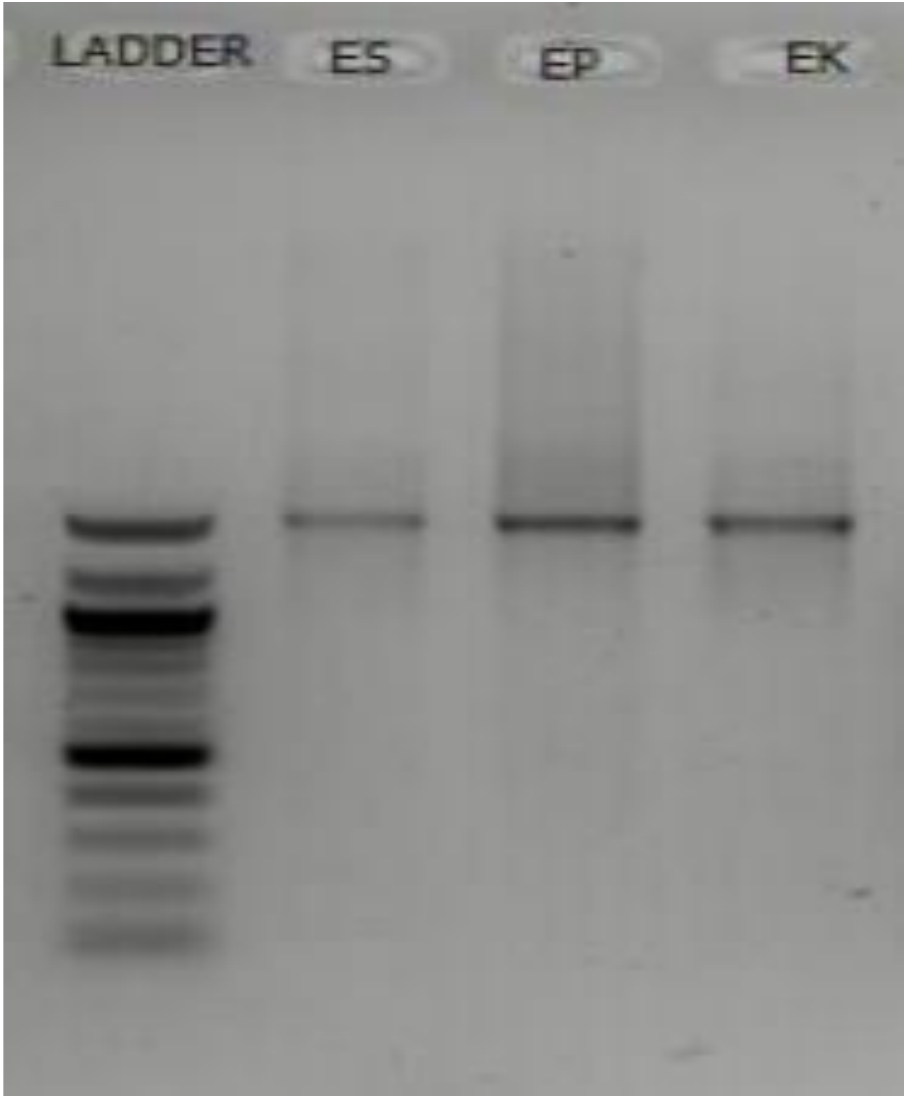
A total of 42 strains of bacteria isolates were obtained from the fermented cassava tubers and identified as *Alcaligenes faecalis* (14) 33.3%, *Pseudomonas aeruginosa* (13) 30.95% and *Pseudomonas putida* (15) 35.71% as shown on tables 1.

TABLE 1: Morphological and Biochemical Features of Bacteria Isolates from sample

MEDIUM	COLONY FEATURES	IND	CIT	CAT	NUMBER OF ISOLATES
Macconkey agar	Red, Solitary, Sticky, Convex, Non-hemolytic, Slimy	+	+	-	14
Mannitol salt agar	Yellow, entire, smooth, shiny, solitary, convex	+	-	+	13
Cetrimide agar	Green, Shiny, Mucoid, Convex	-	+	+	15

Key:  
IND = Indole test  
CIT = Citrate test  
CAT = Catalase test  
CT= Cassava Tubers  
+ = Positive  
- = Negative





**Figure 1:** Agarose gel electrophoresis showing PCR amplification of the 16S rRNA gene (~1500 bp) from bacterial isolates obtained from cassava tuber samples. Lane **L**: DNA ladder (molecular weight marker); Lane **ES**: amplified product from sample ES; Lane **EP**: amplified product from sample EP; Lane **EX**: amplified product from sample EX. All samples showed distinct bands at approximately 1500 bp, indicating successful amplification of the 16S rRNA gene region.

**Table 2: Antibiotics susceptibility test results of *Pseudomonas aeruginosa* with the Inhibition Zone Diameter measured in mm**

Isolates	AZN	AUG	CXM	CIP	CRO	GN	LBC	IMP	ZEM	OFX
<i>Pseudomonas aeruginosa</i>	0	0	0	19	0	12	0	0	0	0
<i>Pseudomonas aeruginosa</i>	09	0	0	0	0	0	15	0	0	0
<i>Pseudomonas aeruginosa</i>	0	0	18	0	0	14	25	0	0	0
<i>Pseudomonas aeruginosa</i>	0	0	0	0	0	0	0	0	0	19
<i>Pseudomonas aeruginosa</i>	0	0	0	0	0	0	15	15	0	0
<i>Pseudomonas aeruginosa</i>	12	0	0	19	0	17	16	0	0	0
<i>Pseudomonas aeruginosa</i>	19	0	15	21	0	13	27	20	0	20
<i>Pseudomonas aeruginosa</i>	19	0	0	12	0	12	10	0	0	0
<i>Pseudomonas aeruginosa</i>	0	0	0	0	0	0	17	0	0	09
<i>Pseudomonas aeruginosa</i>	0	0	0	17	0	0	15	0	0	0
<i>Pseudomonas aeruginosa</i>	21	0	15	0	0	14	15	0	0	0
<i>Pseudomonas aeruginosa</i>	15	0	0	0	0	0	20	0	0	0
<i>Pseudomonas aeruginosa</i>	0	0	0	0	0	16	16	0	0	0

**Keys** →AZN– Azithromycin (15 µg), AUG– Amoxicillin-clavulanic acid (30 µg), CXM– Cefuroxime (30 µg), CIP– Ciprofloxacin (5 µg), CRO– Ceftriaxone (30 µg), GN– Gentamycin (10 µg), LBC– Levofloxacin (5 µg), IMP– Imipenem (10 µg), ZEM– Cefixime (5 µ), OFX–Ofloxacin (5 µg)

Isolated bacteria	AUG	CRO	ACX	IMP	GN	CXM	NF	LBC	ZEM	OFX
<i>Alcaligenes faecalis</i>	0	0	0	0	0	0	0	23	0	15
<i>Alcaligenes faecalis</i>	0	0	0	0	17	0	0	16	0	0
<i>Alcaligenes faecalis</i>	0	0	0	0	0	0	0	0	0	22
<i>Alcaligenes faecalis</i>	0	0	0	0	10	0	0	21	0	10
<i>Alcaligenes faecalis</i>	0	0	0	0	21	0	0	29	0	0
<i>Alcaligenes faecalis</i>	0	0	0	0	15	0	0	18	10	0
<i>Alcaligenes faecalis</i>	0	0	0	0	0	0	0	25	10	15
<i>Alcaligenes faecalis</i>	0	0	0	0	0	0	0	22	15	0
<i>Alcaligenes faecalis</i>	0	0	0	0	12	0	0	15	0	0
<i>Alcaligenes faecalis</i>	0	0	0	0	14	0	0	17	0	0
<i>Alcaligenes faecalis</i>	0	0	0	0	0	0	0	26	0	15
<i>Alcaligenes faecalis</i>	0	0	0	0	0	0	0	22	0	0
<i>Alcaligenes faecalis</i>	0	0	0	0	0	0	0	21	0	10
<i>Alcaligenes faecalis</i>	0	0	0	0	15	0	0	18	0	12
<i>Pseudomonas putida</i>	0	0	0	0	0	0	0	10	0	18
<i>Pseudomonas putida</i>	0	0	0	0	0	0	0	20	0	12
<i>Pseudomonas putida</i>	0	0	0	0	0	0	0	10	0	17
<i>Pseudomonas putida</i>	0	0	0	0	14	0	0	22	0	16
<i>Pseudomonas putida</i>	0	0	0	0	0	0	0	20	0	0
<i>Pseudomonas putida</i>	0	0	0	0	0	0	0	10	0	0
<i>Pseudomonas putida</i>	0	0	0	0	0	0	0	0	0	0

<i>Pseudomonas putida</i>	0	0	0	0	0	0	0	0	0	15
<i>Pseudomonas putida</i>	0	0	0	0	0	0	0	16	0	0
<i>Pseudomonas putida</i>	0	0	0	0	10	0	0	0	0	17
<i>Pseudomonas putida</i>	0	0	0	0	0	0	0	19	0	10
<i>Pseudomonas putida</i>	0	0	0	0	15	0	0	0	0	0
<i>Pseudomonas putida</i>	0	0	0	0	11	0	0	0	0	19
<i>Pseudomonas putida</i>	0	0	0	0	17	0	0	0	0	0
<i>Pseudomonas putida</i>	0	0	0	0	17	0	0	0	0	0

**Table 3: Antibiotics susceptibility test results of *Alcaligenes faecalis* with the Inhibition Zone Diameter measured in mm**

**Keys** → AZN– Azithromycin (15 µg), AUG–Amoxicillin-clavulanic acid (30 µg), CXM–Cefuroxime (30 µg), CIP–Ciprofloxacin (5 µg), CRO– Ceftriaxone (30 µg), GN–Gentamycin (10 µg), LBC– Levofloxacin (5 µg), IMP– Imipenem (10 µg), ZEM– Cefixime (5 µ), OFX– Ofloxacin (5 µg)

**Discussion**

This study investigated the microbial composition of cassava farmlands in Anambra State, Nigeria, identifying three major bacteria: *Alcaligenes faecalis*, *Pseudomonas aeruginosa*, and *Pseudomonas putida*. The microbial isolation was conducted in two phases: biochemical tests (indole, catalase, oxidase, coagulase) and molecular studies using 16S rRNA sequencing for definitive identification. The isolates comprised *Pseudomonas putida* (36%), *Alcaligenes faecalis* (33%), and *Pseudomonas aeruginosa* (31%), consistent with previous findings in Benin and Delta State (Dike et al., 2022; Igbinosa & Igiehon, 2015; Adomi et al., 2020).

A similar study by Kandasamy et al. (2015) identified *Pseudomonas putida* in cassava wastewater. *Pseudomonas* spp. are frequently associated with cassava fermentation due to their enzymatic role in cyanide reduction (Balogun et al., 2021; Bankole et al., 2022). Aremu et al. (2010) demonstrated that *Pseudomonas aeruginosa* utilizes cassava-derived reducing sugars for polyhydroxy butyrate production during fermentation. The presence of

*Pseudomonas* spp. in cassava tubers may also stem from farmland contamination with cassava effluents, influencing soil microbial diversity and pH (Igbiosa & Igiehon, 2015). *Alcaligenes faecalis* was also isolated, in agreement with Obire *et al.* (2021), and its occurrence in cassava and plantain flours was also reported by Oyeyinka & Oyeyinka (2018).

Molecular techniques have increasingly been used to study microbial communities in agricultural environments. For instance, Orji *et al.* (2019) and Okonko *et al.* (2020) used 16S rRNA sequencing to detect *Pseudomonas aeruginosa* in food and water samples, affirming its prevalence in environmental matrices. Babalola *et al.* (2018) also identified *Pseudomonas putida* in cassava effluents and soils, reinforcing its relevance in bioremediation. Likewise, Singh *et al.* (2021) and Biswas *et al.* (2017) identified *Alcaligenes faecalis* in agricultural soils using molecular methods, emphasizing its nitrogen-cycling role and bioremediation potential.

Antibiotic susceptibility testing followed EUCAST (2025) guidelines to evaluate resistance profiles of *Pseudomonas aeruginosa*, *Pseudomonas putida*, and *Alcaligenes faecalis*. *Pseudomonas aeruginosa* exhibited 100% resistance to cefixime, amoxicillin-clavulanic acid, ceftriaxone, cefuroxime, ciprofloxacin, ofloxacin, and azithromycin; 92.4% resistance to imipenem and gentamicin; and 76.9% to levofloxacin. Minor susceptibility was noted to levofloxacin (7.6%) and gentamicin (7.6%), with intermediate response to imipenem. These findings align with Urgancı *et al.* (2022) and Pang *et al.* (2019).

*Alcaligenes faecalis* showed 100% resistance to amoxicillin-clavulanic acid, ceftriaxone, azithromycin, imipenem, cefuroxime, cefixime, ofloxacin, and ciprofloxacin; 92.8% resistance to gentamicin; and 42.8% to levofloxacin. Huang (2020) and Moscoso *et al.* (2023) similarly reported multidrug resistance in *Alcaligenes faecalis*.

*Pseudomonas putida* demonstrated 100% resistance to amoxicillin-clavulanic acid, ceftriaxone, imipenem, cefuroxime, levofloxacin, cefixime, ofloxacin, ciprofloxacin, and azithromycin, and 93% resistance to gentamicin.

*Pseudomonas aeruginosa* showed notable resistance to multiple antibiotics including azithromycin, beta-lactams, and fluoroquinolones. However, some sensitivity was noted to ciprofloxacin (12–21 mm), gentamicin (12–17 mm), and levofloxacin (10–27 mm). These findings reflect earlier studies which report resistance of *Pseudomonas aeruginosa* to beta-lactams and partial susceptibility to aminoglycosides and fluoroquinolones.

*Alcaligenes faecalis* demonstrated relatively lower resistance, with notable sensitivity to levofloxacin (15–29 mm), gentamicin (10–21 mm), and

ofloxacin (10–22 mm). Resistance to beta-lactam antibiotics was consistent with its known intrinsic resistance profile.

*Pseudomonas putida* was resistant to most antibiotics, yet showed moderate sensitivity to levofloxacin (10–22 mm), ofloxacin (10–19 mm), and gentamicin (10–17 mm), suggesting fluoroquinolones, particularly ofloxacin, may be viable for treatment.

Antibiotic resistance was further evaluated by counting the number of antibiotics each organism resisted. *Pseudomonas aeruginosa* and *Pseudomonas putida* resisted five antibiotics and were sensitive to three, while *Alcaligenes faecalis* also resisted five but was sensitive to four, indicating slightly lower resistance overall. The ability to survive antibiotic-exposed environments suggests an adaptive advantage. Studies by Odu & Adeniji (2013) and Nwancho et al. (2014) similarly reported widespread antibiotic resistance in cassava ecosystems, emphasizing the broader implications for food safety and public health.

## Conclusion

The presence of *Pseudomonas aeruginosa*, *Pseudomonas putida*, and *Alcaligenes faecalis* in cassava farmlands poses serious health and economic risks. These opportunistic pathogens can cause infections, particularly in vulnerable individuals, and exhibit high antibiotic resistance. Contaminated cassava may threaten food safety and reduce product shelf life, resulting in financial losses. *Alcaligenes faecalis*, though useful in waste treatment, is emerging as a public health concern. These findings highlight the need for improved agricultural practices, regular microbial monitoring, and further research on resistance mechanisms to ensure the safety and quality of cassava products in Nigeria.

## References

- Abdullahi Taiwo, J. H., Oluwabukola Kudirat, A., Wakili, T., & Jimoh, F. A. (2024). Evaluation of the Potential of Immobilized Cyanide-Degrading Bacteria for the Bioremediation of Cassava Mill Effluent. *Jordan Journal of Biological Sciences*, 17(3).
- Abubakar, M., Mohammed, I. U., Hamisu, A., and Mohammed, M. T. (2019). Severity of cassava mosaic disease in North East, Nigeria. *Journal of Innovative Research in Life Sciences*, 1(1): 1–8. Accessed from <https://www.researchgate.net/publication/371305762> on May 16, 2024.
- Adebayo-Oyetoro, A. O., Oyewole, O. B., Obadina, A. O., and Omemu, M. A. (2013). Microbiological safety assessment of fermented cassava flour “lafun” available in Ogun and Oyo States of Nigeria. *International Journal of Food Science*, 2013: 1–5.
- Adegbehingbe, K. T., Fakoya, S., Marcus, B., Oluyemi, Adeleke, B. S., Fagbohun, O. S., & Adejoro, D. O. (2019). Antibacterial Properties of the Predominant Microorganisms Isolated from Fermenting Cassava Tubers during fufu Production against Selected Enteropathogenic Bacteria. *European Journal of Nutrition & Food Safety*, 287–296. <https://doi.org/10.9734/ejnf/s/2019/v9i330068>
- Adesemoye, A. O., Adeyemo, M. O., & Afolabi, A. S. (2023). Interactions between pathogenic fungi and microbial communities in cassava: Implications for disease management. *Plant Pathology*, 72(1), 1-12.
- Adetunji, C. O., Akande, S. A., Oladipo, A. K., Salawu, R. A., and Onyegbula, A. F. (2017). Determination of the microbiological quality and proximate composition of fermented cassava food products sold in Ilorin-west local government area, Nigeria. *Ruhuna Journal of Science*, 8(2): 76.
- Adomi, Patience & Morka, Emmanuel. (2020). Microbial and Physicochemical Characteristics of Cassava Mill Effluents Receiving Soil in Abraka and Environs, Delta State. *FUPRE Journal of Scientific and Industrial Research* Vol.4, (1), January 2020 ISSN: 2579-1184 (Print) ISSN: 2578-1129
- Alabi, O. J., Kumar, P. L., and Naidu, R. A. (2011). Cassava mosaic disease: a curse to food security in Sub-Saharan Africa. In *The American Phytopathological Society* (pp. 1–16). APSnet Features. Accessed from <https://doi.org/10.1094/APSnetFeature-2011-0701> on May 16, 2024.

- Alene, D. (2013). Economic impacts of cassava research and extension in Malawi and Zambia. *Journal of Development and Agricultural Economics*, 5(11): 457–469.
- Aremu, Layokun, & Solomon. (2010). Production of Poly (3-hydroxybutyrate) from cassava starch hydrolysate by *Pseudomonas aeruginosa* NCIB 950. *American Journal of Scientific and Industrial Research*, 1(3), 421–426. <https://doi.org/10.5251/ajsir.2010.1.3.421.426>
- Balogun, B. O., Adeleke, B. S., and Owoseni, I. I. (2021). Characterization of bacteria isolates from fermented cassava steeping water. *International Journal of Applied Biology*, 5(2): 190–199.
- Bakker, P. A. H. M., Pieterse, C. M. J., & van Loon, L. C. (2019). Induced systemic resistance by fluorescent *Pseudomonas* spp. *Phytopathology*, 109(2), 227–235.
- Balog, un, O. B., Adeleke, B. S., & Owoseni, I. (2021). Characterization of bacteria isolates from fermented cassava steeping water. *International Journal of Applied Biology*, 5(2), 190–199.
- Banjaw, D. T., and Regessa, D. (2017). Cassava integrated pest management: review on cassava mosaic disease and mealybug. *Journal of Biology, Agriculture and Healthcare*, 7(13): 10–16.
- Bayata, A. (2019). Review on nutritional value of cassava for use as a staple food. *Science Journal of Analytical Chemistry*, 7(4): 83.
- Boher, B., Alabi, O. J., Ogunsanya, P., & Legg, J. P. (2022). Molecular tools for the rapid detection of cassava pathogens. *Journal of Plant Pathology*, 104(2), 235–248.
- Bokulich, N. A., Louis, R. R., & Boucher, S. M. (2023). Microbial diversity and function in cassava: Insights from metagenomic studies. *Frontiers in Microbiology*, 14, 1018123.
- Binsker, U., K̄asbohrer, A., and Hammerl, J. A. (2022). Global colistin use: a review of the emergence of resistant Enterobacterales and the impact on their genetic basis. *FEMS Microbiology Reviews*, 46(1): 1–37.
- Biswas, S., et al. (2017). Isolation and molecular characterization of *Alcaligenes faecalis* from wastewater and agricultural soils. *Bioremediation Journal*, 21(3), 197–209.
- Bochner, B. R. (2009). Global phenotypic characterization of bacteria. *FEMS Microbiology Reviews*, 33(1): 191–205.
- CDC. (2019). Antibiotic resistance threats in the United States, 2019. In *Atlanta, GA: U.S. Department of Health and Human Services, CDC* (pp. 1–150).



- Ceballos, H., Ramirez, J., Bellotti, A. C., Jarvis, A., & Alvarez, E. (2020). Adaptation of cassava to changing climates. *Tropical Plant Biology*, 13(2), 87-104.
- Ceballos, H., Pizarro, M., & Gaitán-Sánchez, M. (2021). Metagenomic analysis of microbial diversity in cassava tubers: Implications for food security. *Applied and Environmental Microbiology*, 87(8), e00271-21.
- Chetan, D. M., Raghavendra, H. L., and Prithviraj, H. K. (2017). Isolation and characterization of bacteria from solid waste. *International Journal of Research and Scientific Innovations*, 4(5): 63–68.
- Chinyere Constance Ezemba, G. A Agu, E. J Archibong, M. Ezeokoli, V.N Anakwenze, A.S Ezemba, & Oluchi Judith Osuala. (2022). Isolation and identification of microorganisms associated with fermented (Manihot esculenta Crantz) for the production of Akpu. *International Journal of Frontline Research in Multidisciplinary Studies*, 1(1), 008–015. <https://doi.org/10.56355/ijfrms.2022.1.1.0026>
- Chibucos, M. C., Zweifel, A. E., Herrera, J. C., Meza, W., Eslamfam, S., Uetz, P., Siegele, D. A., Hu, J. C., and Giglio, M. G. (2014). An ontology for microbial phenotypes. *BMC Microbiology*, 14(1): 1–8.
- Chikoti, P. C., Mulenga, R. M., Tembo, M., and Sseruwagi, P. (2019). Cassava mosaic disease: a review of a threat to cassava production in Zambia. *Journal of Plant Pathology*, 101(3): 467–477. Accessed from <https://doi.org/10.1007/s42161-019-00255-0> on May 16, 2024.
- Chikoti, P. C., and Tembo, M. (2022). Expansion and impact of cassava brown streak and cassava mosaic diseases in Africa: a review. *Frontiers in Sustainable Food Systems*, 6: 1–12.
- Chinyere, M. Q., Sunday, S. J., Antip, T. M., Yilyok, C. W., Idris, S., and Nanbyen, D. (2022). Bacteriological quality and public health implications of garri sold in Saturday Market in Langtang North Town, Plateau State. *Journal of Research in Environmental and Earth Sciences*, 8(6), 41–49.
- De Souza, R. M., Ferreira, C. M., & Lima, D. (2023). Characterization of the microbiome in cassava tubers using metagenomic sequencing. *Microbial Ecology*, 86(3), 421-432.
- Dike, K.S., et al. (2022). Analysis of bacterial communities of three cassava-based traditionally fermented Nigerian foods (abacha, fufu, and garri). *Letters in Applied Microbiology*, 74(3), 452-462.
- Dutilh, B. E., Backus, L., Edwards, R. A., Wels, M., Bayjanov, J. R., and Van Hijum, S. A. F. T. (2013). Explaining microbial phenotypes on a genomic scale: Gwas for microbes. *Briefings in Functional Genomics*, 12(4): 366–380.

- Edet, I. V., Akpomie, T. M., Augustine, A. U., Samoh, T. F., Ekam, E. J., and Nzeqbuna, D. D. (2023). Comparative analysis on the proximate composition of processed cassava products obtained from January to March, 2023 in Lafia Town, Nigeria. *Chemical Science International Journal*, 32(4): 50–63.
- Elenwo, M., Maduka, N., and Odu, N. N. (2019). Antibigram testing of bacterial isolates from cassava, yam and plantain flours and shelf life studies of the products sold in some markets in Port Harcourt, Nigeria. *International Journal of Pathogen Research*, 2(4): 1–11.
- Ethica, Stalis & Oedjijono, Oedjijono & Semiarti, Endang & Widada, Jaka & Raharjo, Tri. (2017). Characterization of moaC and a nontarget gene fragments of food-borne pathogen *Alcaligenes* sp. JG3 using degenerate colony and arbitrary PCRs. *Journal of Food Safety*. 37. e12345. 10.1111/jfs.12345
- EUCAST. (2024). Breakpoint Tables for Interpretation of MICs and Zone Diameters\_Version 14.0. In *The European Committee on Antimicrobial Susceptibility Testing* (pp. 1–115). Accessed from <https://www.eucast.org> on May 16, 2024.
- Ezeji, L. A., Adediji, A. O., Nkere, C. K., Ogbe, O. C., Onyeka, J. T., and Atiri, G. I. (2023). Viruses associated with cassava mosaic disease and their alternative hosts along Nigeria-Cameroon border. *African Crop Science Journal*, 31(3): 263–277.
- Ezenwaji, E. E., Nzoiwu, C. P., and Umeogu, C. C. (2017). Contributions of rainfall and other meteorological factors to building collapse in urban areas of Anambra State. *Environmental Review*, 6(1): 45–55.
- Farva, K., Sattar, H., Ullah, H., Raziq, A., Mehmood, M. D., Tareen, A. K., Sultan, I. N., Zohra, Q., and Khan, M. W. (2023). Phenotypic Analysis, Molecular Characterization, and Antibigram of Caries-Causing Bacteria Isolated from Dental Patients. *Microorganisms*, 11(8): 1952.
- Food and Agriculture Organization (FAO). (2023). FAO statistical yearbook 2023. Rome: Food and Agriculture Organization of the United Nations.
- Fregene, M., Cuellar, W. J., & Huerta, M. (2022). Advancements in cassava breeding and the role of microbial interactions. *Plant Biotechnology Journal*, 20(4), 843-857.
- Frías-De León, M. G., Zavala-Ramírez, M., Córdoba, S., Zúñiga, G., Duarte-Escalante, E., Pérez-Torres, A., Zepeda-Rodríguez, A., López-Martínez, I., Buitrago, M. J., and Del Rocío Reyes-Montes, M. (2011). Phenotypic characteristics of isolates of *Aspergillus* section *Fumigati*

- from different geographic origins and their relationships with genotypic characteristics. *BMC Infectious Diseases*, 11(3000): 116.
- Gomes, A. A., Santos, A. L., Oliveira, R. R., & Silva, T. S. (2022). Role of endophytic bacteria in promoting cassava growth under biotic stress. *Microbial Ecology*, 84(3), 517-529.
- Green, M. R., & Sambrook, J. (2012). *Molecular Cloning: A Laboratory Manual* (4th ed.). Cold Spring Harbor Laboratory Press.
- Guanghua Yang, Yucui He, Zhiqiang Cai, Xiyue Zhao, Liqun Wang and Li Wang (2011) Isolation and characterization of *Pseudomonas putida* WLY for reactive brilliant red x-3b decolorization. *African Journal of Biotechnology* Vol. 10(51), pp. 10456-10464, 7 September, 2011 Available online at <http://www.academicjournals.org/AJB> DOI: 10.5897/AJB11.1440 ISSN 1684-5315 © 2011 Academic Journals
- Halake, N. H., & Chinthapalli, B. (2020). Fermentation of traditional African cassava-based foods: Microorganisms role in nutritional and safety value. *Journal of Experimental Agriculture International*, 42(9), 56-65. <https://doi.org/10.9734/jeai/2020/v42i930587>
- Hamad, A. A., Sharaf, M., Hamza, M. A., Selim, S., Hetta, H. F., and El-Kazzaz, W. (2022). Investigation of the bacterial contamination and antibiotic susceptibility profile of bacteria isolated from bottled drinking water. *Microbiology Spectrum*, 10(1): 1-7.
- Hasan, M. J., Nizhu, L. N., & Rabbani, R. (2019). Bloodstream infection with pandrug-resistant *Alcaligenes faecalis* treated with double-dose of tigecycline. *IDCases*, 18, e00600. <https://doi.org/10.1016/j.idcr.2019.e00600>
- Huang, C. (2020). Extensively drug-resistant *Alcaligenes faecalis* infection. *BMC Infectious Diseases*, 20(1), 833. <https://doi.org/10.1186/s12879-020-05557-8>
- Igbinosa, E. O., & Igiehon, O. N. (2015). The Impact of Cassava Effluent on the Microbial and Physicochemical Characteristics on Soil Dynamics and Structure. *Jordan Journal of Biological Sciences*, 8(2), 107-112. <https://doi.org/10.12816/0027556>
- Inetianbor, J. E., Ikenebomeh, M. J., and Udochukwu, U. (2017). Microbiological quality of commercially ready to eat masa sold in Kano, Nigeria. *American Journal of Food, Nutrition and Health*, 2(5): 26-30.
- Kandasamy, S., Dananjeyan, B., Krishnamurthy, K., & Benckiser, G. (2015). Aerobic cyanide degradation by bacterial isolates from cassava factory wastewater. *Brazilian Journal of Microbiology*, 46(3), 659-666. <https://doi.org/10.1590/S1517-838246320130516>

- Kibemo, B. (2017). Isolation, identification, and characterization of some fungal infectious agents of cassava in Southwest, Ethiopia. *Advances in Life Science Technology*, 54: 16–28.
- Koren, S., O'Rourke, S. M., & Edwards, R. (2023). High-throughput sequencing reveals diverse microbial communities in cassava tubers. *Applied and Environmental Microbiology*, 89(5), e00512-23.
- Koza, N. A., Adedayo, A. A., Babalola, O. O., and Kappo, A. P. (2022). Microorganisms in plant growth and development: roles in abiotic stress tolerance and secondary metabolites secretion. *Microorganisms*, 10(8): 1–20.
- Lateef, A., and Ojo, M. O. (2016). Public health issues in the processing of cassava (*Manihot esculenta*) for the production of lafun and the application of hazard analysis control measures. *Quality Assurance and Safety of Crops and Foods*, 8(1): 165–177.
- Liu, S., Wang, H., & Feng, J. (2021). Random amplified polymorphic DNA analysis in microorganisms: Applications and limitations. *Microbiology Insights*, 14, 1-10.
- Loiko, N., Kanunnikov, O., & Litti, Y. (2023). Use of *Alcaligenes faecalis* to Reduce Coliforms and Enhance the Stabilization of Faecal Sludge. *Sustainability*, 15(16), 12580. <https://doi.org/10.3390/su151612580>
- Magiorakos, A. P., Srinivasan, A., Carey, R. B., Carmeli, Y., Falagas, M. E., Giske, C. G., Harbarth, S., Hindler, J. F., Kahlmeter, G., Olsson-Liljequist, B., Paterson, D. L., Rice, L. B., Stelling, J., Struelens, M. J., Vatopoulos, A., Weber, J. T., and Monnet, D. L. (2012). Multidrug-resistant, extensively drug-resistant and pandrug-resistant bacteria: an international expert proposal for interim standard definitions for acquired resistance. *Clinical Microbiology and Infection*, 18(3): 268–281.
- Marín, C., Yáñez-Mansilla, E., & Carvajal, M. (2023). Spatial distribution of pathogenic fungi in postharvest cassava tubers studied by FISH. *Plant Disease*, 107(1), 12-19.
- Mary Nkongho Tanyitiku (2024), Microbiological Contamination of Fermented Cassava Products Sold in Local Markets, Yaounde (Cameroon) *African Journal of Agriculture and Food Science* 7(2), 1-14. DOI:10.52589/AJAFSDPAMM68W
- Meena, M., Swapnil, P., Barupal, T., and Sharma, K. (2020). Encyclopedia of Animal Cognition and Behavior. In *Encyclopedia of Animal Cognition and Behavior*. Downloaded from <https://doi.org/10.1007/978-3-319-47829-6on> May 16, 2024

- Minato, N., Kuhara, S., & Nakamura, S. (2021). The potential of next-generation sequencing in cassava disease diagnostics. *Frontiers in Plant Science*, 12, 637058.
- Moscoso, J., Silva, I., Afonso, M., & Nunes, P. (2023). *Alcaligenes Faecalis*, An unexpected agent of urinary tract infection in a 14-Years-Old Boy. *International Journal of Medical Reviews and Case Reports*, 0, 1. <https://doi.org/10.5455/IJMRCR.172-1671379368>
- Mukumbira, R., Rey, C., & Mashingaidze, A. B. (2021). Cassava root rot diseases: A review of research priorities. *Plant Disease*, 105(11), 3073-3084.
- Nadia, H. S., Paul, K., Cornelius, M. W., and Elijah, M. A. (2021). Phenotypic evaluation of cassava genotypes (*Manihot esculenta*) under moisture stress. *African Journal of Agricultural Research*, 17(6): 836–843.
- Nazarov, P. A., Baleev, D. N., Ivanova, M. I., Sokolova, L. M., and Karakozova, M. V. (2020). Infectious plant diseases: etiology, current status, problems and prospects in plant protection. *Acta Naturae*, 12(3): 46–59.
- Night, G., Asimwe, P., Gashaka, G., Nkezabahizi, D., Legg, J. P., Okao-Okuja, G., Obonyo, R., Nyirahorana, C., Mukakanyana, C., Mukase, F., Munyabarenzi, I., and Mutumwinka, M. (2011). Occurrence and distribution of cassava pests and diseases in Rwanda. *Agriculture, Ecosystems and Environment*, 140(4): 492–497.
- Nuwamanya, E., Mbabazi, R., Tumuhimbise, R., & Kawuki, R. (2021). Cassava genetic diversity and its implication on breeding for starch quality. *Plant Breeding*, 140(1), 91-100.
- Nwakoby, N. E., Ezeogo, J. I., Orji, M. U., and Ejimofor, C. F. (2021). Isolation and identification of bacteria and fungi from cassava mill effluent in Afikpo, Ebonyi State, Nigeria. *South Asian Journal of Research in Microbiology*, 10(4): 18–28.
- Nwanaforo, E., Udensi, J. U., Anyanwu, E. C., Opara, M. C., Egwuogu, G. C., Duru, C. C., and Anyanwu, C. O. (2024). Proximate and microbial analysis of yellow, white and spoilt garri sold in Owerri, Imo State, Nigeria. *Archives of Current Research International*, 24(3): 138–145.
- Nwancho, R.M., et al. (2014). Public health issues in the microbiological quality of cassava products. *Journal of Food Quality*, 37(4), 245-252.
- Nyaka, N. A. I. C., Kammegne, D. P., Ntsomboh, N. G., Mbenoun, M., Zok, S., and Fontem, D. (2015). Isolation and identification of some pathogenic fungi associated with cassava (*Manihot esculenta* Crantz) root rot disease in Cameroon. *African Journal of Agricultural Research*, 10(50): 4538–4542.

- Obi, P. U., Mohammed, Y. M., Okeke, K. S., Ibrahim, N. J., Ajayi, M. A., Benedict, A. U., and Umar, M. (2022). Microbial analysis and sensory attributes of garri produced and marketed in Bida, Niger State, Nigeria. *Journal of Applied Sciences and Environmental Management*, 26(6): 1007–1013.
- Obire, Omokaro and Deeyah, Faith Epsibari (2021). Effect of Cassava Processing Effluent on Microbial Diversity and Physicochemical Constituents of Soils. *J Agri Horti Res.* 4(3): 92-97.
- Odom, T. C., Udensi, E. A., and Nwanekezi, E. C. (2012). Microbiological qualities of hawked retted cassava fufu in Aba Metropolis of Abia State. *Nigerian Food Journal*, 30(1): 53–58.
- Odu, N.N., &Adeniji, A.O. (2013). Antibigram Testing of Bacterial Isolates from Cassava, Yam, and Plantain Flours and Shelf Life Studies of the Products Sold in Some Markets in Port Harcourt, Nigeria. *American Journal of Microbiological Research*, 1(4), 101-106. Retrieved from <https://www.sciepub.com/AJMR/content/1/4/>
- Oduor, G. I., Monda, G. H., & Fregene, M. (2022). Genomic characterization of cassava-associated pathogens: Implications for disease management. *Plant Pathology*, 71(4), 817-830.
- Olopade, B. K., Oranusi, S., Ajala, R., and Olorunsola, S. J. (2014). Microbiological quality of fermented cassava (garri) sold in Ota, Ogun State, Nigeria. *International Journal of Current Microbiology and Applied Sciences*, 3(3): 888–895.
- Ogbonna, O. E., Ayodele, M. A., & Samuel, O. M. (2023). Integrative omics approaches in cassava improvement: From genotyping to phenotyping. *\*Current Plant Biology\**, 28, 100254.
- Ogunjobi, A. A., Ayandiran, T. A., & Olabode, T. B. (2020). Metagenomic analysis of cassava-associated bacterial communities. *Microbial Genomics*, 6(10), e000380.
- Okogbenin, E., Ntawuruhunga, P., Fregene, M., & Mbanaso, E. N. (2021). Cassava pathogen resistance: Advances and prospects. *Advances in Agronomy*, 172, 143-192.
- Ono, L. T., and Taniwaki, M. H. (2021). Fungi and mycotoxins in cassava (*Manihot esculenta* Crantz) and its products. *Brazilian Journal of Food Technology*, 24, e2020240. Accessed from <https://doi.org/10.1590/1981-6723.24020> on May 16, 2024.
- Orji, M.U., et al. (2019). Bacterial contaminants and their antibiotic susceptibility patterns in selected ready-to-eat foods in Nigeria. *Letters in Applied Microbiology*, 70(1), 150-155.

- Otoo, E., Afful, T., & Eshun, J. (2020). Characterization of bacterial communities in cassava tubers using 16S rRNA gene sequencing. *Journal of Microbiology*, 58(6), 476-485.
- Oyeyinka, S.A., & Oyeyinka, A.T. (2018). Microbiological Quality Assessment of Pupuru and Plantain Flours in Akure Metropolis, Nigeria. *Food and Nutrition Sciences*, 9(8), 1010-1020.
- Pang, Z., Raudonis, R., Glick, B. R., Lin, T.-J., & Cheng, Z. (2019). Antibiotic resistance in *Pseudomonas aeruginosa*: Mechanisms and alternative therapeutic strategies. *Biotechnology Advances*, 37(1), 177–192. <https://doi.org/10.1016/j.biotechadv.2018.11.013>
- Parija, S. C. (2012). Textbook of Microbiology and Immunology (S. M. Bhattacharya, S. Nasim, S. Kumar, & S. Dutta, Eds.; 2nd Edit). Elsevier: Reed Elsevier India Private Ltd.
- Parker, N., Schneegurt, M., Tu, A.-H. T., Forster, B. M., & Lister, P. (2017). Microbiology (1st Edit). Openstax, Rice University.
- Ploetz, R. C., Ventura, J. A., & Gomes, A. M. (2022). Novel cassava pathogens and disease management strategies. *Annual Review of Phytopathology*, 60, 285-306.
- Pradhan, P., and Tamang, J. P. (2019). Phenotypic and genotypic identification of bacteria isolated from traditionally prepared dry starters of the Eastern Himalayas. *Frontiers in Microbiology*, 10, 1–15. Accessed from <https://doi.org/10.3389/fmicb.2019.02526> on May 16, 2024.
- Renner, N., Ginika, A., and Ifeanyi, O. (2024). Isolation, multiplication and preservation of cassava fermenting microorganisms. *Journal of Life and Bio Sciences Research*, 5(01), 01–07. Accessed from <https://doi.org/10.38094/jlbsr501110> on May 16, 2024.
- Różewicz, M., Wyzińska, M., and Grabiński, J. (2021). The most important fungal diseases of cereals—problems and possible solutions. *Agronomy*, 11(4): 714.
- Segun, A., Ayandiji, A., Emuoyi Bofarhe, O., Emuoyi Bofarhe, J. O., Adebayo, S., Demeji, O., and James, O. (2019). Detection and classification of cassava diseases using machine learning. *International Journal of Computer Science and Software Engineering (IJCSSE)*, 8(7): 2409–4285.
- Silva, A., Zhang, C., & Li, X. (2022). Functional metagenomics of cassava tuber microbiota: Insights into enzymatic activities. *Food Microbiology*, 106, 104092.

- Simonyan, K. J. (2015). Cassava post-harvest processing and storage in Nigeria: A review. *African Journal of Agricultural Research*, 9(53), 3853–3863. <https://doi.org/10.5897/AJAR2013.8261>
- Singh, A., Verma, J. P., & Mishra, A. (2021). Metagenomics and its application in soil microbial diversity analysis: A review. *Journal of Advanced Research in Microbiology*, 12(4), 57–68.
- Singh, B. K., Trueman, H., & Ritz, K. (2023). The microbial metagenome of cassava tubers: New insights into plant-microbe interactions. *Soil Biology and Biochemistry*, 165, 108514.
- Singh, R., et al. (2021). Molecular tools for identification of *Alcaligenes faecalis* isolated from agricultural soil samples. *Environmental Science and Pollution Research*, 28(12), 15423–15431.
- Soro, M., Tiendrébéogo, F., Pita, J. S., Traoré, E. T., Somé, K., Tibiri, E. B., Nèya, J. B., Mutuku, J. M., Simporé, J., and Koné, D. (2021). Epidemiological assessment of cassava mosaic disease in Burkina Faso. *Plant Pathology*, 70(9): 2207–2216.
- Sujakhu, C., Olee, A., Aryal, C., & Gautam, S. (2016). Isolation and identification of pathogenic bacteria from college premises. St. Xavier's College Maitighar, Kathmandu, Nepal.
- Sundh, I., Del Giudice, T., and Cembalo, L. (2021). Reaping the benefits of microorganisms in cropping systems: is the regulatory policy adequate? *Microorganisms*, 9(7): 1–18.
- Tena D, Fernández C, Lago MR. *Alcaligenes faecalis*: an unusual cause of skin and soft tissue infection. *Jpn J Infect Dis*. 2015;68(2):128-30. doi: 10.7883/yoken.JJID.2014.164. Epub 2014 Nov 25. PMID: 25420652.
- Tian, Y., Zhao, Y., & Li, X. (2019). Limitations of traditional microbiological methods in cassava pathogen detection. *Plant Disease*, 103(6), 1340–1346.
- The European Committee on Antimicrobial Susceptibility Testing. Breakpoint tables for interpretation of MICs and zone diameters. Version 15.0, 2025. <https://www.eucast.org>.
- Thomas, B. T., Effedua, H. I., Davies, A., and Oluwadun, A. (2019). Prevalence of antibiotic-resistant bacteria in dried cassava powder (garri) circulating in Ogun State, Nigeria. *Academia Arena*, 2012(1): 4.
- Torkpo, S. K., Gafni, Y., Danquah, E. Y., and Offei, S. K. (2018). Incidence and severity of cassava mosaic disease in farmer's fields in Ghana. *Ghana Journal of Agricultural Science*, 53: 61.
- Tutun, S., & Yurdakul, Özen. (2023). Importance of *Pseudomonas aeruginosa* in Food Safety and Public Health. *Turkish Journal of*



- Agriculture - Food Science and Technology*, 11(10), 2016–2026.  
<https://doi.org/10.24925/turjaf.v11i10.2016-2026.6155>
- Urgancı, N. N., Yılmaz, N., Koçer Alaşalvar, G., & Yıldırım, Z. (2022). *Pseudomonas aeruginosa* and Its Pathogenicity. *Turkish Journal of Agriculture - Food Science and Technology*, 10(4), 726–738.  
<https://doi.org/10.24925/turjaf.v10i4.726-738.4986>
- Usta Atmaca H, Akbas F. A Extensively drug-resistant *Pseudomonas putida* bacteremia that was resolved spontaneously. *J Infect Dev Ctries*. 2019 Jun 30;13(6):577-580. doi: 10.3855/jidc.11213. PMID: 32058993.
- Wang, Q., Chai, Q., Dou, X., Zhao, C., Yin, W., Li, H., and Wei, J. (2024). Soil microorganisms in agricultural fields and agronomic regulation pathways. *Agronomy*, 14(4): 669. Accessed from <https://doi.org/10.3390/agronomy14040669> on August 22, 2024.
- Wang, L., Xu, F., & Ma, Y. (2020). Denaturing gradient gel electrophoresis (DGGE) in microbial ecology studies. *Critical Reviews in Microbiology*, 46(3), 254-264.
- Wasi, S.M., et al. (2013). Molecular identification and characterization of *Pseudomonas putida* from contaminated soils. *Journal of Environmental Science and Technology*, 6(4), 136-142.
- Wood, S. J., Kuzel, T. M., & Shafikhani, S. H. (2023). *Pseudomonas aeruginosa*: Infections, Animal Modeling, and Therapeutics. *Cells*, 12(1), 199. <https://doi.org/10.3390/cells12010199>
- Wu, Q., Zhang, Y., & Wang, J. (2023). Exploring the fungal microbiome of cassava tubers through metagenomic sequencing. *Fungal Biology*, 127(2), 122-134.
- Yadav, A. N., Kour, D., Abdel-Azeem, A. M., Dikilitas, M., Hesham, A. E. L., and Ahluwalia, A. S. (2022). Microbes for agricultural and environmental sustainability. *Journal of Applied Biology and Biotechnology*, 10: 1–5.
- Younas, T., Muhammad, U., Gondal, A. H., Aziz, H., Khan, M. S., Jabbar, A., Shahzad, H., Panduro-Tenazoa, N. M., Jamil, M., and Areche, F. O. (2022). A comprehensive review on impact of microorganisms on soil and plant. *Journal of Bioresource Management*, 9(2): 109–118.
- Zárate-Chaves, C. A., Gómez de la Cruz, D., Verdier, V., López, C. E., Bernal, A., and Szurek, B. (2021). Cassava diseases caused by *Xanthomonas phaseoli* pv. *manihotis* and *Xanthomonas cassavae*. *Molecular Plant Pathology*, 22(12): 1520–1537.
- Zhao, Y., Zhang, H., & Wang, J. (2022). Microbial diversity in cassava: From tuber to fermented products. *Journal of Applied Microbiology*, 132(2), 543-555.

Zhou, W., Ananga, A., Ukuku, D. O., and Aryee, A. N. A. (2023). High salt concentration affects the microbial diversity of cassava during fermentation, as revealed by 16S rRNA gene sequencing. *Fermentation*, 9(8). Accessed from <https://doi.org/10.3390/fermentation9080727> on August 22, 2024.

## Appendix

### 16S rRNA GENE SEQUENCING RESULTS

#### Sample EK

**Organism:** *Alcaligenes faecalis* strain LCU-MCB-22-001

**GenBank Accession Number:** OP114642.1

**Sequence Identity:** 100%

#### Partial 16S rRNA Sequence:

AAGTCGAACGGCAGCGCGAGAGAGCTTGCTCTCTTGGCGGC  
GAGTGGCGCACGGGTGAGTAATATATCGGAACGTGCCCCGAT  
AGCGGGGGATAACTACTCGAAACAGTGGCTAATACCGCATA  
CGCCCTACGGGGGAAAGGGGGGGATCGCAAGACCTCTCACT  
ATTGGAGCGGCCGATATCGGATTAGCTAGTTGGTGGGGTAA  
AGGCTCACCAAGGCTACGATCCGTAGCTGGTTTGGAGAGGAC  
GACCAGCCACACTGGGACTGAGACACGGCCCAAACCTCCTAC  
GGGAGGCAGCAGTGGGGAATTTTGGACAATGGGGGAAACC  
CTGATCCAGCCATCCCGCGTGTATGATGAAGGCCTTTCGGGTT  
GTAAAGTACTTTTGGCAAAGAATAAAAAGGTATCCCCTAATAC  
GGGATACTGCTGACGGTATCTGCAGAATAAGCACCGGCTAA  
CTACGTGCCAGCAGCCGCGGTAATACGTAGGGTGCAAGCGT  
TAATCGGAATTACTGGGCGTAAAGCGTGTGTAGGCGGTTTCG  
GAAAGACAGATGTGAAATCCCAGGGCTCAACCTTGGAACCTG  
CATTTTAACTGCCGAGCTAGAGTATGTCAGAGGGGGGGTAG  
AATTCACAGTGTAGCAGTGAAATGCGTAGATATGTGGAGGA  
ATACCGATGGCGAAGGCAGCCCCCTGGGATAATACTGACGC  
TCAGACACGAAAGCGTGGGGAGCAAACAGGATTAGATACCC  
TGGTAGTCCACGCCCTAAACGATGTCAACTAGCTGTTGGGG  
CCGTTAGGCCTTAGTAGCGCAGCTAACGCGTGAAGTTGACC  
GCCTGGGCAGTACGGTCGCAAGATTAAAACTCAAAGGAATT  
GACGGGGACCCGCACAAGCGGTGGATGATGTGGATTAAATTC  
GATGCAACGCGAAAAACCTTACCTACCCTTGACATGTCTGGA  
AAGCCGAAGAGATTTTGGCCGTGCTCGCAAGAGAACCGGAAC  
ACAG

**Sample ES**

**Organism:** *Pseudomonas aeruginosa* strain Ps.ADMC09

**GenBank Accession Number:** MK598336.1

**Sequence Identity:** 87.93%

**Partial 16S rRNA Sequence:**

TTATAGATTTTGTCTCTGATATGAGCGGCGGGTATGCCTA  
CTAAATGCAAATCGAGGGGCGGAGAGAGAGTACTCTCCTGT  
TGTCAGCGGCGGCGGGGTGAGTTATTTATTGGGATCTGCCT  
GATAGGGGGGAAAAACGTCCGGAAACGGGCGCTAATACCG  
CATAAGTCCTGTGGGGGGGAAGGGGGGGGGTTTTCGGACCTT  
TCGCTATCAGATGAGCCCATGTGCGATTAGTTAGTTGGTGG  
GGTAAAGGCTTACCTAGGCGACGATCCGTAACCTGGTTTGAG  
AGGATGATCAGCCACCCTGGAACCTGAGACACGGTCCCGACT  
CCTACGGGAGGCA  
GCAGTGGGGAATATTGGACAATGGGCGAAAGCCTGATCCA  
GCCATCCCGCGTGTGTGAAGAAGGCCITTGGGTGTAAAGT  
ACTTTAAGTTGGGAGGAAGGGCAGTAAGTTAATACCGTGGT  
CTTTTGACGTTTCCAACAAAAAAGCACCGGCTAATTTCTTGC  
CACCAGCCCCGGTAATACTAAGGGGGCAAGGGTTTATTGGA  
ATTTTTGGGGGTAAAAAGGGGGTAGGGGGTTTATCAATTTG  
GATGTGAAAACCTCGGGCCTAACCTGGGAAATGCATCCAAA  
ACTGGTGAGCTAGAGTCAGGTAGAGGGAGGTAGAATTTTCAT  
GTGTAGCGGTGAAATGGGTAAAATTTGGGAGGAAAAACCGG  
TGGGGAAGGCGGCCTCCTGGACATATCTTGCCCTTAGGTCA  
GCAAGCGTGGGGGGCGAACC GGATTAGATACCTCCGTGTT  
CCAACCCCCAACGGATGTTGAATATGGCGTTGGGGGTCCTT  
GAGGTTTTGGTTGCGCGAGTTAACGCGTAATTTCTCCCGCCTG  
GGGAGTACGGCCGCAAGGTAAAACTCAATGAATTGACGGG  
GGCCCGCACAAAGCGGTGGAGCATGTGGTTTAATTCAAAGCA  
ACGCGAAGAACCTTACCTGGTCTTGACATGCTGAGAACTTTTC  
CAAAGATGGATTGGTGCTTTCGGGAACTCTGACACATGGCT  
GATGGCTGTCTGATGCTCGGGTTGTGAATGGTGGGTAAAGTC  
CCGAACGAAGCCAAACCTTATCCTTTGTTGCAGCATTAAGGG  
CCGAATTTCTAAGGAGACTGCCCCGAGACAAACGGAAGGAAGG  
GGGATGACGTAAAGTCATCATGGTCCTTACGGCCAGGGCTA  
CCACCTGCTACCATGGACGAAACAAAAGGGTGCCACCCCGC  
GAGGGGGGAGCTAATCCCATAAAACCGATCGTAGTTTCGGATC  
GGGTCTGCAACTCGACTTCCTGAAACCGGATTCGCTAGTAAT  
CGTGAATCAAAATGGTACGGTGAATACCTTCCCGGGCCTTGT  
ACAAACCGCCCGTCACCCCATGGGAGTGGGTTGCTACAAAA

GCACTAATTTAACCGTCACGAGGACGGCTCCCACGATGTGAT  
TCTTGACTGCGGTGACCCAAACAAGGCC

**Sample EP**

**Organism:** *Pseudomonas putida strain SB19*

**GenBank Accession Number:** MZ430405.1

**Sequence Identity:** 84.82%

**Partial 16S rRNA Sequence:**

GCGTTAGCTGCGGACTGAGGGAGACCCACCGGCTATCGACA  
TCGTTTACCGGGGACTACCAGGGAACTAATCCTGGTTGGTCC  
CCACGCTTTCCACCTCAGTGCAGTATGTCCAGGGGGGCCCTT  
CCCACGGGTTCCTTCCTATTTTACCATTTCCCGTACCCAGAAA  
TTCCACCCCCCTTCCATACTTAGCTTCGGTTTGTGGATGTTCCCG  
GTGAGCCCGGGGTTTACATCAACTTAACAACCCCTACCGCG  
CTTTACCCCAAATTCATAACGCTTGCCCCGTATTACCGGGT  
GTGGCACAGATTAGCCGGTGCTTATTCCTGTGGAAACGCAAA  
AAAGGATTAACCTTACTGCCCTTCCTCCCAACTAAAGTGCTTTA  
CAACCAAGACCTTCTTCAACACGCGGGATGGTGATCAGGCTT  
CGCCCATTTGTCAAATTCCCCACTGTGCCTCCCGTAGGAGCTG  
GACCGTGTCTCAGTCCAGGTGACTGATCATCCTCTCAACAGT  
ACGGATCGTCGCTAGGTAGCATTACCTCACCTACTACTAATC  
GACCTGGCTCATCTGATAGCGCAAGGCCGAAGGTCCCCTGC  
TTTCTCCCGAGGACATGCGGTATTAGCGCCTTTCAGAGTTCC  
CCCACTACAGGCAGATCCTATGATTACTCACCCGTCCGCCGC  
TACAAGGAAATCCCGTCTCCGTCTGCAGTGTAGCCTGACCA  
CCACGTCAATTCTGAACAGATCAACTCTACAACGT