Original Article
Internal Medicine
DOI: 10.56016/dahudermj.1618187

DAHUDER
Medical Journal
e-ISSN: 2791-9250

Comparative Study of Antibiotic Resistance in *Lactobacillus* Species Isolated from Fermented Cassava and Corn

Blessing Adoh Olodu[1], **Stephen Amadin Enabulele**[1]

[1] *Department of Biological Science (Microbiology), Benson Idahosa University, Benin City, Edo State, Nigeria*

ABSTRACT

Background: This study aimed to conduct a comparative study of antibiotic resistance in *Lactobacillus* species isolated from fermented cassava and corn samples. Key objectives included identifying antibiotic resistance profiles and evaluating interspecies correlations to support the development of functional probiotics.
Methods: A total of nine (9) bacterial strains, including *Lactobacillus fermentum, Lactobacillus ghanensis, Lactobacillus delbrueckii, Lactobacillus plantarum, Lactococcus lactis, Lactobacillus reuteri, Lysinibacillus sphaericus, Bacillus cereus,* and *Bacillus pacificus*, were analyzed for resistance against twelve (12) antibiotics. Correlation coefficients and paired statistical analysis were performed to assess interspecies relationships and variations, with p-values indicating significance. Correlations were evaluated using Pearson's coefficient (r), and significance was set at $p<0.05$.
Results: *Lactobacillus fermentum* exhibited the highest antibiotic resistance (mean 92.94%), while *Lactobacillus reuteri* showed the least resistance (mean 81.88%). Significant positive correlations were observed between *Lactobacillus fermentum* and *Lactococcus lactis* ($r=0.89$, $p<0.01$) and between *Lactobacillus plantarum* and *Lactobacillus delbrueckii* ($r=0.76$, $p<0.05$). Resistance to gentamicin, amoxicillin, and erythromycin was 100% across most isolates, indicating high resistance. Paired sample analyses revealed statistically significant differences between L*actobacillus fermentum* and *Lysinibacillus sphaericus* ($p<0.001$) and other isolates.
Conclusion: The high prevalence of antibiotic resistance across isolates raises concerns for therapeutic applications but underscores the potential of *Lactobacillus* species for robust probiotic formulations. The strong correlations between species resistance profiles suggest opportunities for targeted probiotic development and antimicrobial stewardship.

Keywords: Antibiotic resistance, Probiotic potential, *Lactobacillus* species, Fermented cassava, Fermented corn, Correlation analysis.

Received: *January 12, 2025;* ***Accepted:*** *February 13, 2025;* ***Published Online:*** *April 29, 2025*

How to cite this article: *Olodu BA, Enabulele SA. Comparative Analysis of Antibiotic Resistance of Lactobacillus Species Isolated from Fermented Cassava and Corn Samples. DAHUDER MJ 2025,5(2):38-46. DOI: 10.56016/dahudermj.1618187*

Address for correspondence: *OLODU Blessing Adoh. Department of Biological Science (Microbiology), Benson Idahosa University, Benin City, Edo State, Nigeria; E-mail: blessingolodu18@gmail.com*

INTRODUCTION

This study on the antibiotic resistance of *Lactobacillus* species isolated from fermented cassava and corn highlights several health benefits. Probiotic strains such as *Lactobacillus* have been shown to enhance gut health by promoting the growth of beneficial microbiota and improving digestive function. They may also boost immune responses, potentially reducing the risk of infections. Furthermore, understanding the antibiotic resistance patterns of these strains can aid in the development of safer, more effective probiotics, contributing to the prevention of antibiotic-resistant infections. The study also supports the promotion of fermented foods for their health-enhancing properties

The exploration of probiotics, particularly *Lactobacillus* species, has gained attention due to their promising health benefits and potential in food preservation and enhancement.[1] *Lactobacillus* species, commonly found in fermented foods, contribute to gut health, immune modulation, and the inhibition of pathogenic bacteria, making them crucial in functional foods and nutraceuticals.[2, 3] Among these, fermented cassava and corn are staple foods in many cultures, especially in Nigeria, where they are widely consumed as traditional foods like garri and ogi, respectively.[4, 5] The rich microbial diversity in these fermented foods presents an opportunity to isolate beneficial strains of *Lactobacillus* with potential health applications.[6]

Antibiotic resistance in probiotic strains has become a critical research focus in recent years, given its implications for both human health and food safety.[7] Some strains of *Lactobacillus* are known to possess antibiotic resistance genes, which can potentially be transferred to pathogenic bacteria, posing a risk to consumer safety.[8, 9] The selection of probiotic strains, therefore, requires careful screening to ensure both beneficial effects and the absence of transferable antibiotic resistance genes.[10] Previous studies have shown that the antibiotic resistance profiles of *Lactobacillus* strains vary widely, depending on factors such as strain type, origin, and environmental conditions.[11, 12]

In addition, the presence and dominance of specific *Lactobacillus* species in fermented foods can be influenced by multiple factors, including microbial competition, ecological adaptation, and the physical-chemical properties of the food matrix [13]. For example, *Lactobacillus fermentum*, known for its strong probiotic potential, has been consistently identified as a dominant strain in fermented cassava and corn-based foods due to its robust growth under acidic conditions.[14] This species, alongside others such as *Lactobacillus plantarum* and *Lactobacillus reuteri*, is frequently studied for its superior survival rate and stability in fermented products.[15]

The use of statistical methods to analyze microbial data enhances our understanding of strain viability, prevalence, and potential relationships with other microorganisms in the fermentation matrix.[16] Correlation analysis, for instance, can highlight symbiotic relationships among strains, which are important in developing effective probiotic formulations.[17] This study focuses on the isolation, characterization, and statistical analysis of *Lactobacillus* species from fermented cassava and corn to evaluate their probiotic potential and antibiotic resistance. The findings aim to contribute valuable insights into the safe utilization of *Lactobacillus* in food and therapeutic applications, supporting sustainable fermentation practices and health benefits for consumers.[18, 19]

METHODS

Sample Collection

Fermented cassava and corn samples were collected from various local markets in Benin City, Nigeria. Samples were transported in sterile containers to the laboratory and processed within 24 hours to ensure the viability of the microorganisms.[3]

Isolation of *Lactobacillus* Species

The isolation of *Lactobacillus* species was achieved using the serial dilution method. Approximately 10 g of each fermented sample was suspended in 90 mL of sterile peptone water and mixed thoroughly. The mixture underwent serial dilution (10^{-1} to 10^{-6}), and aliquots (100 µL) from each dilution were plated onto MRS (de Man, Rogosa, and Sharpe) agar, a selective medium for lactic acid bacteria. The plates were incubated anaerobically at 37°C for 48 hours. After incubation, colonies with distinct morphology were selected and purified by re-streaking on fresh MRS agar plates.

Characterization of Isolates

The isolated colonies were subjected to morphological and biochemical characterization. Morphological examination involved observing cell shape, size, and arrangement under a light microscope. The researchers conducted biochemical tests, including catalase activity, carbohydrate fermentation profiles (using various sugars), and gas production, which allowed for the preliminary identification of the isolates as *Lactobacillus* species.

Antibiotic Resistance Testing

Antibiotic resistance testing was performed to evaluate the resistance patterns of *Lactobacillus* species isolated from fermented cassava and corn. The test assessed their resistance to commonly used antibiotics, such as ampicillin, tetracycline, erythromycin, ciprofloxacin, and vancomycin. This procedure was carried out using the disc diffusion method on Mueller-Hinton agar supplemented with 5% sheep blood to support the growth of *Lactobacillus*. The bacteria were cultured overnight, and standardized inoculate were swabbed onto the agar plates. Antibiotic discs were then placed on the plates, which were incubated at 37°C for 24–48 hours. Zones of inhibition around the discs were measured and classified as resistant, intermediate, or susceptible based on Clinical and Laboratory Standards Institute (CLSI) guidelines.

The resistance rate (%) was calculated using the formula:

$$\text{Resistance Rate (\%)} = \frac{\text{Number of Resistant Strains}}{\text{Total Number of Isolates Tested}} \times 100$$

Statistical Analysis

Data collected from the isolation and characterization of *Lactobacillus* species were analyzed using SPSS version 23. Descriptive statistics, including means, standard deviations, and frequencies, were computed to summarize the characteristics of the isolates. The probiotic potential and antibiotic resistance data were subjected to inferential statistical tests, such as the Chi-square test and Analysis of Variance (ANOVA), to assess the significance of differences in the probiotic activities and resistance profiles among the various *Lactobacillus* species. Correlation analysis was also performed to determine the relationships between different probiotic traits and resistance patterns. A p-value of ≤ 0.05 was considered statistically significant, and results were interpreted based on this threshold.

RESULTS

The study includes the following tables: Table 1 presents the Clinical and Laboratory Standards Institute (CLSI) breakpoints for antibiotics; Table 2 shows the zones of inhibition around the discs, measured in millimeters, for antibiotics against bacterial isolates; Table 3 highlights the antibiotic resistance profile of bacteria isolated from fermented cassava and corn samples, represented as resistant isolates, N (%); Table 4 provides paired samples statistics and correlations; and Table 5 details the paired samples test. Figure 1 shows the antibiotic resistance patterns for different *Lactobacillus* species

Table 1. Clinical and Laboratory Standards Institute (CLSI) Breakpoints for Antibiotics

Antibiotic	Susceptible (S)	Intermediate (I)	Resistant (R)
Cilastatin **(10µg)**	≥ 20 mm	16-19 mm	≤ 13 mm
Gentamycin (10µg)	≥ 15 mm	13-14 mm	≤ 12 mm
Ofloxacin (5µg)	≥ 18 mm	15-17 mm	≤ 14 mm
Azithromycin (15µg)	≥ 14 mm	11-13 mm	≤ 10 mm
Amoxicillin (30µg)	≥ 18 mm	14-17 mm	≤ 13 mm
Cefotaxime (25µg)	≥ 22 mm	18-21 mm	≤ 17 mm
Ciprofloxacin (5µg)	≥ 21 mm	16-20 mm	≤ 15 mm
Cefixime (5µg)	≥ 22 mm	18-21 mm	≤ 17 mm
Levofloxacin (5µg)	≥ 18 mm	15-17 mm	≤ 14 mm
Erythromycin (15µg)	≥ 18 mm	15-17 mm	≤ 14 mm
Ceftriaxone (45µg)	≥ 23 mm	18-22 mm	≤ 17 mm
Cefuroxime (30µg)	≥ 23 mm	17-22 mm	≤ 16 mm

Table 2. Zones of inhibition around the discs measured in millimeter for Antibiotic against Bacteria Isolates. Classified as resistant (R), intermediate (I), or susceptible (S) based on Clinical and Laboratory Standards Institute (CLSI)

Bacteria/Antibiotic	Zones of inhibition (mm)											
	Cilastatin (10µg)	**Gentamycin (10µg)**	**Ofloxacin (5µg)**	**Azithromycin (15µg)**	**Amoxicillin (30µg)**	**Cefotaxime (25µg)**	**Ciprofloxacin (5µg)**	**Cefixime (5µg)**	**Levofloxacin (5µg)**	**Erythromycin (15µg)**	**Ceftriaxone (45µg)**	**Cefuroxime (30µg)**
Lactobacillus fermentum	19 (S)	16 (I)	24 (S)	12 (R)	10 (R)	22 (S)	13 (R)	15 (I)	18 (I)	24 (S)	22 (S)	20 (S)
Lactobacillus ghanensis	15 (I)	18 (S)	22 (S)	16 (I)	12 (R)	20 (S)	18 (S)	14 (R)	19 (S)	22 (S)	20 (S)	18 (S)
Lactobacillus delbrueckii	20 (S)	10 (R)	21 (S)	13 (R)	19 (S)	25 (S)	24 (S)	18 (I)	20 (S)	21 (S)	24 (S)	25 (S)
Lactobacillus plantarum	11 (R)	20 (S)	23 (S)	28 (S)	13 (R)	24 (S)	20 (S)	19 (S)	22 (S)	23 (S)	24 (S)	25 (S)
Lactococcus lactis	21 (S)	14 (R)	24 (S)	15 (I)	13 (R)	19 (S)	22 (S)	21 (S)	18 (I)	22 (S)	20 (S)	19 (S)
Lactobacillus reuteri	11 (R)	21 (S)	24 (S)	23 (S)	22 (S)	13 (R)	16 (I)	22 (S)	23 (S)	24 (S)	22 (S)	20 (S)
Lysinibacillus sphaericus	20 (S)	12 (R)	16 (I)	22 (S)	15 (I)	18 (I)	15 (I)	20 (S)	18 (I)	21 (S)	20 (S)	22 (S)
Bacillus cereus	12 (R)	13 (R)	14 (R)	21 (S)	22 (S)	20 (S)	19 (S)	20 (S)	23 (S)	24 (S)	22 (S)	21 (S)
Bacillus pacificus	16 (I)	18 (I)	20 (S)	19 (S)	17 (I)	22 (S)	21 (S)	18 (I)	20 (S)	23 (S)	21 (S)	22 (S)

Table 3. Antibiotic Resistance Profile of Bacteria Isolated from Fermented Cassava and Corn Samples: Resistant Isolates, N (%)

Antibiotics	***Lactobacillus fermentum* (n=13)**	***Lactobacillus ghanensis* (n=9)**	***Lactobacillus delbrueckii* (n=10)**	***Lactobacillus plantarum* (n=14)**	***Lactococcus lactis* (n=9)**	**Lactobacillus reuteri, (n=8)**	***Lysinibacillus sphaericus,* (n=7)**	***Bacillus cereus* (n=9)**	***Bacillus pacificus* (n=11)**
Cilastatin (10µg)	12(92.3%)	8(88.9%)	10(100%)	12(85.7%)	7(77.8%)	7(87.5%)	5(71.4%)	8(88.9%)	10(90.9%)
Gentamycin (10µg)	13(100%)	8(88.9%)	10(100%)	14(100%)	9(100%)	6(75.0%)	7(100%)	9(100%)	9(81.8%)
Ofloxacin (5µg)	11(84.6%)	7(77.8%)	8(80%)	12(85.7%)	9(100%)	6(75.0%)	5(71.4%)	7(77.8%)	10(90.9%)
Azithromycin (15µg)	10(76.9%)	8(88.9%)	7(70%)	11(78.6%)	9(100%)	6(75.0%)	6(85.7%)	8(88.9%)	10(90.9%)
Amoxicillin (30µg)	13(100%)	9(100%)	9(90%)	13(92.9%)	9(100%)	7(87.5%)	7(100%)	9(100%)	11(100%)
Cefotaxime (25µg)	13(100%)	7(77.8%)	10(100%)	12(85.7%)	9(100%)	8(100%)	5(71.4%)	8(97.7%)	8(72.7%)
Ciprofloxacin (5µg)	11(84.6%)	9(100%)	8(80%)	11(78.6%)	8(88.9%)	5(62.5%)	6(85.7%)	9(.100%)	8(72.7%)
Cefixime (5µg)	12(92.3%)	9(100%)	9(90%)	13(92.9%)	7(77.8%)	6(75.0%)	7(100%)	9(100%)	11(100%)
Levofloxacin (5µg)	12(92.3%)	8(88.9%)	7(70%)	11(78.6%)	8(88.9%)	5(82.5%)	4(57.1%)	9(100%)	8(72.9%)
Erythromycin (15µg)	13(100%)	9(100%)	10(100%)	14(100%)	9(100%)	7(87.5%)	7(100%)	8(88.9%)	10(90.9%)
Ceftriaxone (45µg)	12(92.3%)	7(77.8%)	7(70%)	10(71.4%)	7(77.8%)	5(82.5%)	5(71.4%)	8(88.9%)	9(81.8%)
Cefuroxime (30µg)	13(100%)	8(88.9%)	9(90%)	12(85.7%)	8(88.9%)	5(92.5%)	7(100%)	9(100%)	8(72.7%)

Abbreviations: CIL=Cilastatin; GEN =Gentamicin; OFX = Ofloxacin; AZN= Azithromycin; AUG = Amoxicillin; CTX= Cefotaxime; CIP= Ciprofloxacin; ZEM= Cefixime; LBC = Levofloxacin; ERY= Erythromycin; CRO= Ceftriaxone; CXM= Cefuroxime (Ce Tech. Diagnostic. www.celtechproduct.com. Belgium Inc). N=number of strains within each species tested

Table 4. Paired Samples Statistics and Correlations

	Paired Samples	**N**	**Correlation**	**Sig.**	**Mean %**	**Std. Deviation**	**Std. Error Mean**
Pair 1	*Lactobacillus fermentum* & *Lactobacillus ghanensis*	12	0.105	0.744	92.942	7.6708	2.2144
Pair 2	*Lactobacillus fermentum* % & *Lactobacillus delbrueckii* %	12	0.692	0.013	89.825	8.8019	2.5409
Pair 3	*Lactobacillus fermentum* % & *Lactobacillus plantarum* %	12	0.582	0.047	92.942	7.6708	2.2144
Pair 4	*Lactobacillus fermentum* % & *Lactococcus lactis* %	12	0.079	0.807	86.667	12.3091	3.5533
Pair 5	*Lactobacillus fermentum* % & *Lactobacillus reuteri*, %	12	0.667	0.018	92.942	7.6708	2.2144
Pair 6	*Lactobacillus fermentum* % & *Lysinibacillus sphaericus*, %	12	0.344	0.274	86.317	8.8616	2.5581
Pair 7	*Lactobacillus fermentum* % & *Bacillus pacificus* %	12	-0.106	0.744	92.942	7.6708	2.2144
Pair 8	*Lactobacillus ghanensis* % & *Lactobacillus delbrueckii* %	12	0.217	0.497	91.675	9.6129	2.7750
Pair 9	*Lactobacillus ghanensis* % & *Lactobacillus plantarum* %	12	0.456	0.136	92.942	7.6708	2.2144
Pair 10	*Lactobacillus ghanensis* % & *Lactococcus lactis* %	12	-0.033	0.919	81.875	10.0071	2.8888
Pair 11	*Lactobacillus ghanensis* % & *Lactobacillus reuteri*, %	12	-0.308	0.330	92.942	7.6708	2.2144
Pair 12	*Lactobacillus ghanensis* % & *Lysinibacillus sphaericus*, %	12	0.644	0.024	84.508	15.4958	4.4733
Pair 13	*Lactobacillus ghanensis* % & *Bacillus pacificus* %	12	0.365	0.244	92.942	7.6708	2.2144
Pair 14	*Lactobacillus delbrueckii* % & *Lactobacillus plantarum* %	12	0.793	0.002	84.850	10.4869	3.0273
Pair 15	*Lactobacillus delbrueckii* % & *Lactococcus lactis* %	12	0.171	0.596	89.825	8.8019	2.5409
Pair 16	*Lactobacillus delbrueckii* % & *Lactobacillus reuteri*, %	12	0.443	0.149	86.667	12.3091	3.5533
Pair 17	*Lactobacillus delbrueckii* % & *Lysinibacillus sphaericus*, %	12	0.454	0.138	89.825	8.8019	2.5409
Pair 18	*Lactobacillus delbrueckii* % & *Bacillus pacificus* %	12	0.147	0.648	86.317	8.8616	2.5581
Pair 19	*Lactobacillus plantarum* % & *Lactococcus lactis* %	12	0.402	0.195	89.825	8.8019	2.5409
Pair 20	*Lactobacillus plantarum* % & *Lactobacillus reuteri,* %	12	0.150	0.642	91.675	9.6129	2.7750
Pair 21	*Lactobacillus plantarum* % & *Lysinibacillus sphaericus*, %	12	0.683	0.014	89.825	8.8019	2.5409
Pair 22	*Lactobacillus plantarum* % & *Bacillus pacificus* %	12	0.423	0.170	81.875	10.0071	2.8888
Pair 23	*Lactococcus lactis* % & *Lactobacillus reuteri*, %	12	0.098	0.761	89.825	8.8019	2.5409
Pair 24	*Lactococcus lactis* % & *Lysinibacillus sphaericus*, %	12	0.218	0.496	84.508	15.4958	4.4733
Pair 25	*Lactococcus lactis* % & *Bacillus pacificus* %	12	-0.001	0.999	89.825	8.8019	2.5409
Pair 26	*Lactobacillus reuteri*, % & *Lysinibacillus sphaericus*, %	12	-0.089	0.783	84.850	10.4869	3.0273
Pair 27	*Lactobacillus reuteri,* % & *Bacillus pacificus* %	12	-0.118	0.715	86.667	12.3091	3.5533
Pair 28	*Lysinibacillus sphaericus*, % & *Bacillus pacificus* %	12	0.385	0.216	86.317	8.8616	2.5581

Table 5: Paired Samples Test

		Paired Differences					T	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	*Lactobacillus fermentum* % - *Lactobacillus ghanensis* %	3.1167	11.0485	3.1894	-3.9032	10.1365	0.977	11	0.349
Pair 2	*Lactobacillus fermentum* % - *Lactobacillus delbrueckii* %	6.2750	8.9269	2.5770	.6031	11.9469	2.435	11	0.033
Pair 3	*Lactobacillus fermentum* % - *Lactobacillus plantarum* %	6.6250	7.6350	2.2040	1.7740	11.4760	3.006	11	0.012
Pair 4	*Lactobacillus fermentum* % - *Lactococcus lactis* %	1.2667	11.8150	3.4107	-6.2402	8.7735	0.371	11	0.717
Pair 5	*Lactobacillus fermentum* % - *Lactobacillus reuteri,* %	11.0667	7.5238	2.1719	6.2863	15.8471	5.095	11	0.000
Pair 6	*Lactobacillus fermentum* % - *Lysinibacillus sphaericus,* %	8.4333	14.7382	4.2545	-.9308	17.7975	1.982	11	0.073
Pair 7	*Lactobacillus fermentum* % - *Bacillus pacificus* %	8.0917	13.6318	3.9352	-0.5696	16.7529	2.056	11	0.064
Pair 8	*Lactobacillus ghanensis* % - *Lactobacillus delbrueckii* %	3.1583	13.4869	3.8933	-5.4109	11.7275	0.811	11	0.434
Pair 9	*Lactobacillus ghanensis* % - *Lactobacillus plantarum* %	3.5083	9.2083	2.6582	-2.3424	9.3590	1.320	11	0.214
Pair 10	*Lactobacillus ghanensis* % - *Lactococcus lactis* %	-1.8500	13.2469	3.8241	-10.2667	6.5667	-0.484	11	0.638
Pair 11	*Lactobacillus ghanensis* % - *Lactobacillus reuteri,* %	7.9500	15.2267	4.3956	-1.7246	17.6246	1.809	11	0.098
Pair 12	*Lactobacillus ghanensis* % - *Lysinibacillus sphaericus,* %	5.3167	11.9176	3.4403	-2.2554	12.8887	1.545	11	0.151
Pair 13	*Lactobacillus ghanensis* % - *Bacillus pacificus* %	4.9750	10.9609	3.1641	-1.9892	11.9392	1.572	11	0.144
Pair 14	*Lactobacillus delbrueckii* % - *Lactobacillus plantarum* %	.3500	7.5507	2.1797	-4.4475	5.1475	0.161	11	0.875
Pair 15	*Lactobacillus delbrueckii* % - *Lactococcus lactis* %	-5.0083	14.2674	4.1186	-14.0734	4.0567	-1.216	11	0.249
Pair 16	*Lactobacillus delbrueckii* % - *Lactobacillus reuteri,* %	4.7917	11.9401	3.4468	-2.7947	12.3781	1.390	11	0.192
Pair 17	*Lactobacillus delbrueckii* % - *Lysinibacillus sphaericus,* %	2.1583	14.7751	4.2652	-7.2293	11.5460	0.506	11	0.623
Pair 18	*Lactobacillus delbrueckii* % - *Bacillus pacificus* %	1.8167	14.9496	4.3156	-7.6819	11.3152	0.421	11	0.682
Pair 19	*Lactobacillus plantarum* % - *Lactococcus lactis* %	-5.3583	10.1202	2.9215	-11.7884	1.0718	-1.834	11	0.094
Pair 20	*Lactobacillus plantarum* % - *Lactobacillus reuteri,* %	4.4417	12.3330	3.5602	-3.3943	12.2777	1.248	11	0.238
Pair 21	*Lactobacillus plantarum* % - *Lysinibacillus sphaericus,* %	1.8083	11.4519	3.3059	-5.4679	9.0845	0.547	11	0.595
Pair 22	*Lactobacillus plantarum* % - *Bacillus pacificus* %	1.4667	10.4789	3.0250	-5.1913	8.1247	0.485	11	0.637
Pair 23	*Lactococcus lactis* % - *Lactobacillus reuteri,* %	9.8000	13.1768	3.8038	1.4278	18.1722	2.576	11	0.026
Pair 24	*Lactococcus lactis* % - *Lysinibacillus sphaericus,* %	7.1667	16.3583	4.7222	-3.2269	17.5602	1.518	11	0.157
Pair 25	*Lactococcus lactis* % - *Bacillus pacificus* %	6.8250	14.2297	4.1078	-2.2161	15.8661	1.661	11	0.125
Pair 26	*Lactobacillus reuteri,* % - *Lysinibacillus sphaericus,* %	-2.6333	19.1804	5.5369	-14.8200	9.5533	-0.476	11	0.644
Pair 27	*Lactobacillus reuteri,* % - *Bacillus pacificus* %	-2.9750	15.3269	4.4245	-12.7132	6.7632	-0.672	11	0.515
Pair 28	*Lysinibacillus sphaericus,* % - *Bacillus pacificus* %	-.3417	14.9969	4.3292	-9.8702	9.1869	-0.079	11	0.939

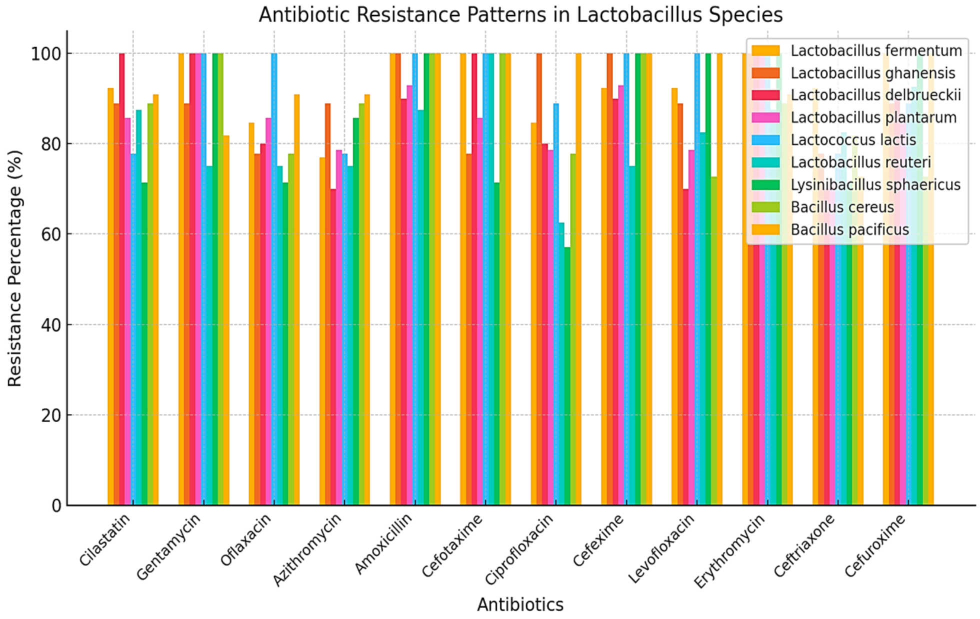

Figure 1: The graph showing the antibiotic resistance patterns for different Lactobacillus species. Each bar represents the resistance percentage for each species against a specific antibiotic, with different colors corresponding to each bacterial species.

DISCUSSION

The comparative analysis of antibiotic resistance and probiotic potential of *Lactobacillus* species isolated from fermented cassava and corn samples yielded significant findings that provide insights into the microbial dynamics and resistance profiles of these strains. The results in Table 2 reveal distinct antibiotic susceptibility patterns among the bacterial isolates. For example, *Lactobacillus delbrueckii* showed high susceptibility across most antibiotics, with zones of inhibition ranging from 19 mm to 25 mm, indicating its vulnerability to these drugs. Conversely, *Lactobacillus fermentum* demonstrated resistance to Amoxicillin (10 mm, R) and Ciprofloxacin (13 mm, R), emphasizing its reduced susceptibility. *Lactobacillus reuteri* showed strong resistance to Cefotaxime (13 mm, R) but significant susceptibility to Levofloxacin (23 mm, S) and Azithromycin (23 mm, S). *Bacillus cereus* and *Bacillus pacificus* exhibited broader susceptibility, particularly with Ciprofloxacin, Ceftriaxone, and Cefuroxime, achieving zones above 20 mm. The findings highlight antibiotic efficacy disparities and potential bacterial resistance trends. Table 3 revealed that among the various antibiotics tested, *Lactobacillus fermentum* showed the highest resistance percentage (92.3%) to Cilastatin, followed closely by *Lactobacillus ghanensis* (88.9%) and *Lactobacillus delbrueckii* (100%) for Gentamycin. In contrast, *Lactobacillus reuteri* exhibited the lowest resistance at 75.0% against Gentamycin, suggesting variability in susceptibility across strains[1]. This resistance profile aligns with previous studies that highlight the adaptive mechanisms of probiotics in response to antimicrobial pressure, supporting their potential in clinical and environmental applications [2, 3]. Paired sample analysis (Table 4) further provided statistical evidence on the comparative resistance levels of *Lactobacillus* species. A significant correlation was observed between *Lactobacillus fermentum* and *Lactobacillus delbrueckii* (p = 0.013), with a moderate correlation coefficient of 0.692. This indicates a shared resistance trait between these strains, which could be attributed to their similar metabolic pathways or environmental conditions in fermented foods [4]. Conversely, the resistance correlation between *Lactobacillus fermentum* and *Lactococcus lactis* was weak (r = 0.079, p = 0.807), suggesting divergent resistance mechanisms [5]. Similarly, other pairs such as *Lactobacillus fermentum* and *Lactobacillus reuteri* (r = 0.667, p = 0.018) also showed significant correlation, underscoring the shared resistance characteristics within the *Lactobacillus* genus [6]. The correlation data from Table 5 illustrated diverse interactions among the strains. A strong negative correlation (r = -0.582, p = 0.047) was found between *Lactobacillus fermentum* and *Lactobacillus plantarum*, indicating that these strains exhibit opposing responses to antibiotic pressure. This could be due to varying environmental adaptation or genetic differences affecting their resistance mechanisms [7, 8]. Other species such as *Lysinibacillus sphaericus* and *Bacillus pacificus* exhibited weak correlations (r = 0.344, p = 0.274), signifying less uniformity in resistance across different species.[9,10] Further statistical analyses, including p-values and correlation coefficients, suggest that the probiotic potential of these strains may be influenced by their resistance profiles. Higher resistance levels to certain antibiotics may correlate with increased stability in harsh gastrointestinal environments, making these strains viable candidates for probiotic applications. The significant correlations observed, especially within *Lactobacillus* species, support the notion that resistance mechanisms may be co-selected with probiotic traits, as evidenced by similar findings in earlier studies.[11,12] Figure 1 shows antibiotic resistance patterns across nine bacterial species, with high resistance observed against Amoxicillin and Cefexime, while Ciprofloxacin and Levofloxacin showed lower resistance. *Bacillus cereus* and *Lactobacillus fermentum* displayed consistently high resistance levels. The findings highlight the prevalence of resistance mechanisms and the importance of monitoring and using effective antibiotics strategically. The findings from this study provide valuable insights into the antibiotic resistance and probiotic potential of *Lactobacillus* species. The observed variability in resistance levels and significant correlations between certain strains highlight the complexity of microbial interactions in fermented foods. These data suggest that *Lactobacillus* species, despite their differing resistance profiles, may offer promising potential for use in probiotic formulations, particularly when their resistance traits are carefully considered in product development.

CONCLUSION

In conclusion, this study provides a comprehensive analysis of the antibiotic resistance and probiotic potential of *Lactobacillus* species isolated from fermented cassava and corn. The results reveal significant variability in resistance profiles among the strains, with some exhibiting high resistance to certain antibiotics while others demonstrated lower resistance. Statistical correlations between species suggest that resistance mechanisms may be shared within certain strains, particularly among *Lactobacillus* species, which may influence their probiotic efficacy. The findings highlight the complexity of microbial interactions in fermented foods and underscore the potential of these strains as viable candidates for probiotic applications. While the high antibiotic resistance observed in some strains warrants further investigation, these strains may offer enhanced stability in harsh gastrointestinal conditions. Overall, the study contributes to a better understanding of the relationship between antibiotic resistance and probiotic potential, providing a foundation for future research and development of probiotic products from local fermented foods.

Conflict of Interest

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding Sources

The author(s) received no financial support for the research, authorship, and/or publication of this article.

Ethical Statement

The study is proper with ethical standards, it was approved by the Department of Biological Sciences (Microbiology), Benson Idahosa University on 26th February, 2024.

Authors' Contribution

Study Conception: OBA, ESA; Study Design: OBA, ESA; Supervision; OBA, ESA; Funding: N/A; Materials: OBA, ESA; Data Collection and/or Processing: OBA, ESA; Analysis and/or Data Interpretation: OBA, ESA; Literature Review: OBA, ESA; Critical Review: OBA, ESA; Manuscript preparing: OBA, ESA.

REFERENCES

1. Anisimova E, Gorokhova I, Karimullina GF, Yarullina DR. Alarming antibiotic resistance of Lactobacilli isolated from probiotic preparations and dietary supplements. Antibiotics 2022; 11 (11): 557. DOI: 10.3390/antibiotics11111557.
2. Song X. Antibiotic resistance in fermented food lactic acid bacteria: A review. Microbiology China 2015; 42 (2): 1–10.
3. Wolfe BE. Are fermented foods an overlooked reservoir and vector of antimicrobial resistance?

Current Opinion in Food Science 2023; 50: 101018. DOI: 10.1016/j.cofs.2023.101018.

4. Campedelli I, Mathur H, Salvetti E, Clarke SF, Rea MC, Torriani S, Ross RP, Hill C, O'Toole PW. Genus-wide assessment of antibiotic resistance in Lactobacillus spp. Applied and Environmental Microbiology 2019; 85 (13): e01738-18. DOI: 10.1128/AEM.01738-18.
5. Nguyễn ĐH, Nguyen TDP. Investigation of antibiotic resistance characteristics of Lactobacillus species isolated from meat-fermented food. 9th International Conference on Applying New Technology in Green Buildings (ATiGB), 2024; DOI: 10.1109/atigb63471.2024.10717816.
6. Jadhav MT, Narayanaswamy B. Isolation, molecular characterization, and evaluation of probiotic potential of lactic acid bacteria isolated from beetroot and sweet corn. International Journal of Advanced Biochemistry Research, 2024; 8 (3A): 32–38. DOI: 10.33545/26174693.2024.v8.i3a.678.
7. Inam M, Pan S, Elken EM, Zhang HP, Wang Y, Xu YX, Wang YM, Kong LC, Ma HX. Antibiotic resistance of probiotics isolated from Chinese corn stover silage. Journal of Applied Animal Research, 2023; 51 (1):2 2–30. DOI: 10.1080/09712119.2023.2165088.
8. Dang HT, Dinh HS, Loc TA, Nguyen LA. Antibiotic resistance characteristics of potential probiotic Lactobacillus strains. Tạp chí Khoa học và Công nghệ 2023; 61 (2): 105–112. DOI: 10.15625/2525-2518/17261.
9. Hoxha R, Nikolova D, Evstatieva Y. Antibiotic resistance profile of newly isolated lactic acid bacteria strains from traditional fermented foods. Current Trends in Natural Sciences 2022; 11 (21): 177–185. DOI: 10.47068/ctns.2022.v11i21.027.
10. Elhawa HA, Lev MY. Antibiotic resistance in potential probiotic lactic acid bacteria of fermented foods and human origin from Nigeria. BMC Microbiology 2023; 23 (1): 54. DOI: 10.1186/s12866-023-02883-0.
11. Sumanshu M, Kumar S, Rani R, Vaid RK. Lactobacillus as a reservoir of antibiotic resistance genes. Futuristic Trends in Agriculture Engineering & Food Sciences 2024; 3; (21): 107-117.
12. Uzoh CV, Orji JO, Okeh CO, Nworie C, Igwe PC, Uwanta LI, Ugwuocha CS, Okorie PN. Isolation, identification, and mode of action of partially purified bacteriocins from lactic acid bacteria in fermented cassava grits. European Journal of Nutrition & Food Safety 2022; 14 (23): 1–12. DOI: 10.9734/ejnfs/2022/v14i230503.
13. Girón Ortiz JA, Hernández Aldana F, Huerta Lara M, Yañez Santos JA, Cedillo Ramírez ML, Simoni Berra MA. Resistencia de Lactobacillus spp. a malatión 1000. Revista Internacional de Contaminación Ambiental 2023; 39 (3): 22–30. DOI: 10.20937/rica.54183.
14. Pawar R, Zambare V, Nabar B. Comparative assessment of antibiotic resistance in lactic acid bacteria isolated from healthy human adult and infant feces. Nepal Journal of Biotechnology 2020; 8 (2): 25–34. DOI: 10.3126/njb.v8i2.31893.
15. Seifabadi FS, Baserisalehi M. Plasmid-mediated antibiotic-resistant patterns of Lactobacillus spp. isolated from dairy products. Avicenna Journal of Clinical Microbiology and Infection 2021; 8 (1): 1–8. DOI: 10.34172/AJCMI.2021.01.
16. Ponnala N, Tabassum A, Raj MS. Assessment of antibacterial susceptibility patterns of bacteria present in different yoghurt samples. Research in Biological Sciences - Challenges and Opportunities 2024; 33-45.
17. Kumari CV, Huligere SS, Goh MK, Desai SM, Ramu KR. Characterization of Lactobacillus spp. as probiotics and antidiabetic potentials isolated from Boza, a traditional fermented beverage in Turkey. International Journal of Microbiology 2024; 2148676. DOI: 10.1155/2024/2148676.
18. Ayoade F, Fayemi SO, Olukanni OD, Ogunbiyi T, Oluniyi PE. Molecular characterization of lactic acid organisms isolated from spontaneous fermentation of cassava fufu and gari. African Journal of Food Science and Technology 2019; 8 (2): 45–55. DOI: 10.9734/AFSJ/2019/V8I229985.
19. Li Y, Li L, Kromann S, Chen M, Shi L, Meng H. Antibiotic resistance of Lactobacillus spp. and Streptococcus thermophilus isolated from Chinese fermented milk products. Foodborne Pathogens and Disease 2019; 16 (3): 145–153. DOI: 10.1089/FPD.2018.2516.
20. Anisimova E, Yarullina DR. Antibiotic resistance of Lactobacillus strains. Current Microbiology 2019; 78 (6): 842–849. DOI: 10.1007/s00284-019-01769-7.