IJMAAS International Journal of Microbiology and Applied Sciences
Volume 2 issue 2 Sept, 2023
Research Article

Antibiotic Susceptibility Pattern of Bacteria Isolated from Laboratory Produced Fermented Beverage ("Burukutu") Produced from Guinea Corn (*Sorghum bicolor*)

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ABSTRACT

Burukutu, a traditional Nigerian fermented beverage produced from guinea corn (*Sorghum bicolor*), has gained popularity for its nutritional and cultural significance. However, the microbial quality and antibiotic susceptibility pattern of bacteria isolated from laboratory produced burukutu remain underexplored. This study aimed to isolate and identify bacteria from laboratory produced burukutu using standard analytical methods. Antibiotic susceptibility testing of bacterial isolates was carried out using the Kirby-Bauer disk diffusion method. Bacterial isolates were identified as *Bacillus subtilis, Bacillus cereus, Lactobacillus fermentum, Staphylococcus aureus, Streptococcus lactis* and *Escherichia coli.* Antibiotic susceptibility testing revealed varying degrees of resistance among the isolated strains. Notably, *Bacillus subtilis* and *Staphylococcus aureus* exhibited resistance to septrin and zinacef, respectively. *Bacillus cereus* and *Lactobacillus fermentum* were sensitive to all the antibiotics used. *Streptococcus lactis* was resistant to almost all the antibiotics except rocephin and amoxicillin while *Bacillus cereus* and *Lactobacillus fermentum* were sensitive to all the antibiotics used. These findings highlight the presence of antibiotic-resistant bacteria in laboratory-produced burukutu, which could pose potential health risks to consumers. The presence of these bacteria could be attributed to contamination from handlers during harvest and processing. It is therefore necessary to develop strategies to mitigate their prevalence in burukutu production. This study emphasizes the importance of monitoring and ensuring the safety of traditional fermented beverages to protect public health.

Keywords: *Sorghum bicolor*, "Burukutu", traditional fermented beverage, *S. aureus*, *E. coli*, antibiotic susceptibility.

Introduction

Burukutu is a traditional fermented beverage produced from guinea corn (*Sorghum bicolor*) with deep cultural roots in Nigeria and other West African countries. This millet-based drink has been consumed for generations and is known for its rich nutritional content and unique taste (Kolawole *et al*., 2007). Microorganisms play a pivotal role in the fermentation process of burukutu, contributing to its characteristic flavors and nutritional profile. However, the presence of pathogenic or antibiotic-resistant bacteria in laboratory-produced burukutu can pose serious public health concerns. This is particularly worrisome in a world grappling with antibiotic resistance, a global health crisis that threatens our ability to combat infectious diseases effectively (Achi, 2005).

The antibiotic susceptibility pattern of bacteria isolated from laboratory-produced burukutu is a topic of emerging interest, as it sheds light on the potential presence of antibiotic-resistant strains within this beverage. Understanding the susceptibility of these bacteria to commonly used antibiotics is crucial for assessing the safety of burukutu and for implementing strategies to mitigate any health risks associated with its consumption. Usually, *Lactobacillus* colonizes the human digestive tract, urinary tract and genital systems and very rarely causes any infection. Besides lacking pathogenicity, they confer several health benefits (Achi, 2005) which serves the purposes of probiotics; - Probiotics are globally consumed in food, dietary supplements, or as active components of a known medication, and are commercially available in different forms.

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However, there is a need to reassure their safety, especially in terms of spreading antibiotic resistance. There is a high possibility of horizontal gene transfer among bacteria in nature and further spread of these resistant strains between populations (Rina and Sonali, 2016). The last decade has witnessed an increase in the number of reports documenting antibiotic resistance in *Lactobacillus* strains. Although, they are safe, but there is concern towards possible mobility of resistance determinants to human and animal pathogenic and opportunistic bacteria. Few researchers acknowledge the presence of antibiotic resistance in *Lactobacillus* and appreciate the possibility of their co-administration with antibiotic therapy, ensuring replenishment of the healthy gut flora, which is otherwise at high risk (Abuajah *et al.,* 2015). However, this statement is divisive and a matter of conflict. The presence of resistance coding genes and transfer of the same through plasmids and conjugative transposons have also been reported in *Lactobacillus* species (Ahmed *et al*., 2012). Fermented milk products such as curd and yoghurt and human milk are among the common source of *Lactobacillus* that assures establishment and replenishment of healthy gut flora after antibiotic treatment to adults and infants, respectively (Alemu, 2009). It is important to understand the resistance profile of bacterial inhabitants of our foods. This study aims to investigate the microbial composition of laboratory produced burukutu and assess the antibiotic susceptibility patterns of the isolated bacteria. By doing so, we can gain valuable insights into the safety of this traditional fermented beverage and contribute to the broader conversation on food safety, antibiotic resistance, and public health. The findings of this research will inform both consumers and producers about potential risks associated with burukutu consumption and guide future efforts to ensure the safety and quality of this beloved West African beverage.

Materials and Methods

Sample collection and visual observation

Grains of guinea corn (*Sorghum bicolor*) were purchased from Mile III market, Diobu, in Port Harcourt Local Government Area of Rivers State. Visual observation of the sample was done to note the appearance of sample before and after preparation of the product.

Guinea corn grain is reddish-brown with black spot. The guinea corn sample was placed in a sterilized zip lock bag to avoid contamination and transported to the Department of Microbiology Laboratory, Rivers State University, Port Harcourt for preparation and analysis within two hours of collection.

Preparation of burukutu using (guinea corn)

Sifting of guinea corn sample

Sifting of the guinea corn sample was carried out by sieving with a sieve to sift off dirt, dust and chaffs probably acquired from harvest. Debris and stones were carefully removed from the grain at this stage to ensure a clean brew.

Steeping

About 1.5 kg of the guinea corn grain was weighed and was submerged in water and rinsed twice, and then eight (8) litres of sterilized water were added to the grain and allowed to soak for two days. To enable an even sprouting, the guinea corn was completely submerged for two days as to enable the grains absorb moisture that eventually enabled sprouting. This process enhances and gives the brew its unique taste and texture. After the soaking period, the grains were spread on a mat to allow aeration for a comfortable sprouting at a room temperature of 26ºC; the grains were stirred or turned frequently to enable even sprouting. This process of sprouting lasted for 3 days after observation of a desired sprout.

Mashing

The grains were milled/ground and mixed with water to form a mash. During the milling process, 200 ml of water was added continuously to yield a wort of 2 kg liquor after which it was heated at 100ºC intensively for 24 hours. This process was achieved by regulating the periods of water added and continuous adjustment of the fire source to achieve accurate timing. It was then allowed to cool down a bit to a temperature of 60ºC and then re-boiled again at 100ºC for 12 hours. This was to enable the grain enzymes decompose the starch and prepare it for the next phase.

The boiled wort was allowed to cool to about 45ºC before it was sieved off the chaff using a sieve with tiny pores to obtain fine smooth wort for the fermentation process.

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Fermentation of guinea corn wort

The wort obtained from the mashing and sieving process was mixed with 200ml of potable water to dilute its viscosity or its thickness (as to facilitate its fermentation) before inoculating with the foam from previously mature burukutu or a sack bag previously soaked in the foam gotten from already mature brew, obtained from local producers. The wort was allowed to ferment for two to five (2 – 5) days. Note that the longer the fermentation period the more the alcohol content of the resultant brew.

Mature Burukutu

After the fermentation of the brew, the product was transferred to a cleaner vessel where it was stored for consumption.

Determination of pH of the samples

The pH of the samples was determined using the pH meter. The pH meter was switched on for 30 minutes to allow for stabilization before taking measurements. The pH meter was calibrated against standard buffers before measuring the pH of mature burukutu sample.

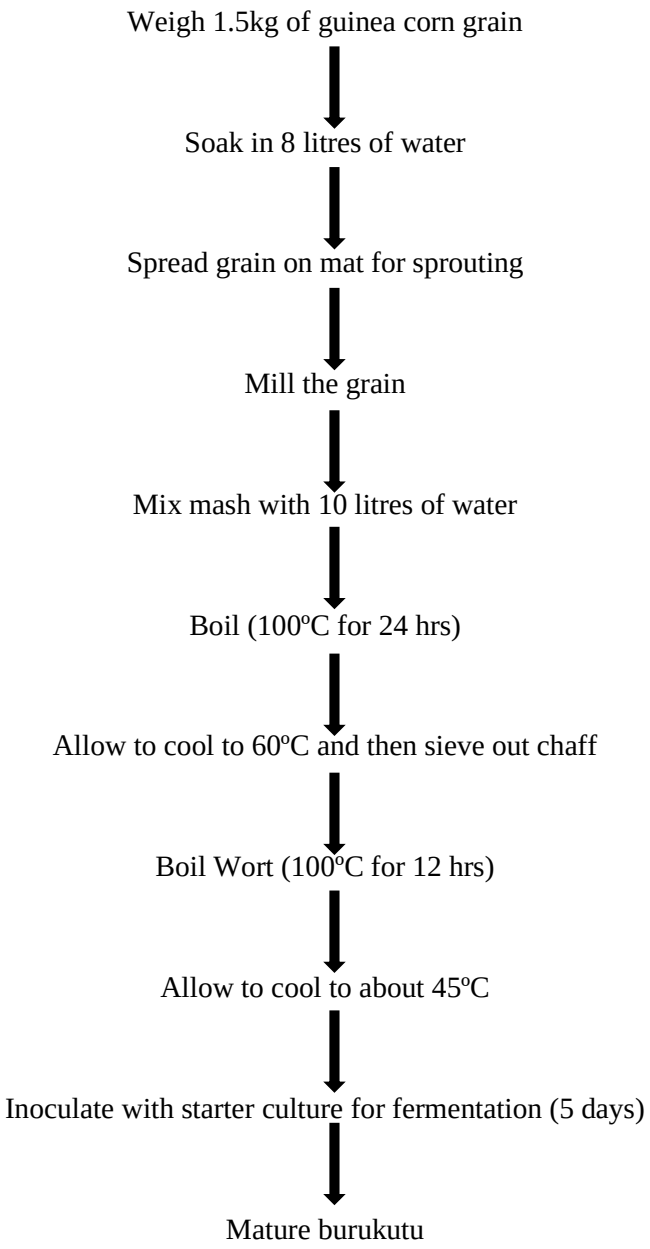

Fig 1: Flow diagram of the stages of production of Burukutu (By the Authors)

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Isolation, enumeration and identification of microorganisms

A ten-fold serial dilution of the sample was caried out. The 10^2, 10^3 and 10^4 dilution factors were plated using pour plate isolation technique. The plates were incubated at 37ºC for 24 hours. The colonies that appear different in shape, size, texture and colour on the incubated plates were sub-cultured into sterile nutrient agar plate and stored at 36°C for 24 hours. Pure culture obtained was then transferred into separate nutrient agar slant in the McCartney bottles and stored in the refrigerator as stock culture. The bacterial isolates were characterized based on colonial morphology, cellular morphology, staining reactions and biochemical tests.

Suspected *Lactobacillus* sp isolates were cultured on the de Man, Rogosa and Sharpe *agar* (MRS) to observe for the growth of *Lactobacillus* sp with a milky round raised gram positive *on* the medium which is the characteristics of the organism. Gram Staining, Motility test and Biochemical tests such as Indole, Methyl red, Voges Proskaur, Glucose, Lactose, Mannose, Sucrose and Citrate Utilization test were carried out to confirm isolates.

Antibiotic susceptibility testing

Antibiotic susceptibility of the bacteria isolated from the burukutu samples to commonly used antibiotics was determined on Mueller Hinton agar using the disc diffusion method according to the description of the Clinical and Laboratory Standards Institute (CLSI, 2010). Inoculum was prepared by direct colony suspension and growth methods depending on whether the bacterium is Gram-positive or Gram-negative. The standardized inoculum of each isolate with a uniform optical density was spread over Mueller Hinton agar plate using sterile cotton swabs.

The standard antibiotics discs were placed at the equidistance on the surface of inoculated agar plates. The susceptibility patterns of the isolates were determined by measuring the zone of inhibition in millimeter (mm) and interpreted according to CLSI as resistance, intermediate or susceptible.

The commercial antibiotics used for the study were; Erythromycin 5µg, Nalidixic acid 5µg, Streptomycin 10µg, Septrin 10µg, Perfloxacin 5µg, Ceporex 15µg, Augmentin 10µg, Ampicillin 5µg, Ciprofloxcin 10µg, Gentamycin 15µg, Norfloxacin 13µg, Trivid 20µg, rocephin 25µg, Zinacef 10µg, and Rocephin 25µg.

Arithmetical determination of multiple antibiotic resistance (MAR) index

Arithmetical method was carried out to determine the determination of MAR index by analyzing the antibiotic susceptibility/resistance pattern of the bacteria against antibiotics. The expression of MAR index is estimated thus:
MAR = a/b, where a is the number of antibiotics to which the test isolate demonstrated resistance and b is the total number of antibiotics to which the test isolate has been assessed for susceptibility. Multiple Antibiotic Resistance (MAR) index is a valid and useful method of tracking especially bacterial infections and drug resistance. Bacteria having MAR index ≥ 0.2 originate from a high-risk source of contamination where several antibiotics are used (Sandhu *et al.*, 2016).

Results

Table 1 presents the bacterial counts in colony forming units (CFU) of various samples (whole grains, sprouted grains, uncooked mash, and boiled wort during the production process of laboratory produced burukutu.

Total Heterotrophic Bacteria Count (THB): Ungrounded grain has the highest THB count at 3.1×10^6 CFU/ml, followed by sprouted grains at 2.8×10^6 CFU/ml. Uncooked mash and boiled wort have lower THB counts, indicating a lower population of these types of bacteria.

Lactobacillus Count (LC): *Lactobacilli* are a group of bacteria often associated with fermentation processes. Uncooked mash has the highest LC at 2.4×10^4 CFU/ml, followed by sprouted grains at 1.9×10^4 CFU/ml Ungrounded grain and boiled wort have significantly lower LC counts.

Vibrio Count (VC): *Vibrio* bacteria are a type of Gram-negative bacteria. Sprouted grains have the highest VC at 1.1×10^4 CFU/ml, while uncooked mash has a lower but still significant VC count. Ungrounded grain and boiled wort show no detectable *Vibrio* bacteria.

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Table 1: Bacterial count (CFU/g or CFU/ml) of processed samples during production of Burukutu

Sample	Bacterial count (CFU/g or CFU/ml) of samples during production of Burukutu					
	THB ($\times10^6$)	**Lactobacilli ($\times10^4$)**	***Vibrio*** **($\times10^4$)**	**Total *Staph*** **($\times10^4$)**	**Total Coliform ($\times10^4$)**	**Faecal Coliform ($\times10^3$)**
Unground grain	3.1±0.03	0.0±.0.00	0.0±0.00	2.3±0.22	2.3±0.09	1.4±0.4
Sprouted grains	2.8±0.7[b]	1.9±0.07[b]	1.1±0.02[b]	2.7±0.22 [b]	2.3±0.09 [c]	1.1±0.02 [b]
Uncooked mash	2.5±0.8[b]	2.4±0.9[c]	0.0±0.0[a]	0.29±0.09[a]	1.9±0.14 [b]	0.65±0.35[ab]
Boiled wort	0.65±0.28[a]	0.80±0.14[a]	0.0±0.0	0.0±0.0	0.0±0.0	0.0±0.0[a]

*Mean with the same superscript along the column is not significantly different ($p\leq0.05$)
Key: THBC = Total Heterotrophic Bacteria.

Total Staphylococcal Count (TSC): *Staphylococcus* bacteria are common inhabitants of various environments. Ungrounded grain has the highest TSC at 2.3×10^4 CFU/ml, while uncooked mash and sprouted grains also have notable counts. Boiled wort has the lowest TSC count.

Total Coliform Count: Coliform bacteria are often used as indicators of fecal contamination. Sprouted grains and uncooked mash have similar TCC counts at 2.3×10^4 CFU/ml and 1.9×10^4 CFU/ml respectively, indicating potential fecal contamination. Ungrounded grain has a slightly lower count, while boiled wort has no detectable coliforms.

Faecal Coliform Count (FCC): Faecal coliforms are a subgroup of coliform bacteria associated with fecal contamination. Sprouted grains and uncooked mash have FCC counts, with sprouted grains having a higher count. Ungrounded grain and boiled wort show lower or no FCC counts.

Table 2 provides insights into the microbial counts at different times during fermentation of the guinea corn wort to produce Burukutu, a traditional Nigerian fermented beverage. Total Heterotrophic Bacteria Count (THB) on Day 1, the THB count is the highest at 2.6×10^6 CFU/ml, but it decreases significantly over the fermentation period, reaching 0.15×10^6 CFU/ml on Day 5. *Lactobacillus* Count (LC): *Lactobacilli* are lactic acid bacteria often associated with the fermentation process. LC remains relatively stable throughout the fermentation, with counts ranging from 0.085×10^5 CFU/ml on Day 4 to 2.4×10^5 CFU/ml on Day 3. *Vibrio* and total Staphylococcal counts were not recorded during the period of fermentation.

Table 2: Microbial counts during fermentation of the guinea corn wort to produce Burukutu

Fermentation Time	Bacterial Count (CFU/ml) of fermented wort samples during production of Burukutu					
	THB ($\times10^6$)	***Lactobacilli*** **($\times10^5$)**	***Vibrio*** **($\times10^2$)**	**Total *Staph*** **($\times10^4$)**	**Total Coliform ($\times10^4$)**	**Faecal Coliform ($\times10^2$)**
Day 1	2.6±0.07[d]	1.8±0.07[c]	0.0±0.0	0.0±0.0	1.24±0.2	13.0±0.1[d]
Day 2	1.4±0.08[c]	1.9±0.0[c]	0.0±0.0	0.0±0.0	2.17±0.2	8.5±0.7[c]
Day 3	0.97±0.28[b]	2.4±0.07[d]	0.0±0.0	0.0±0.0	1.8±0.30	6.0±1.4[bc]
Day 4	0.33±0.07[a]	0.085±0.007[b]	0.0±0.0	0.0±0.0	0.25±0.9	2.5±0.07[ab]
Day 5	0.15±0.08[a]	0.70±0.04[a]	0.0±0.0	0.0±0.0	0.11±0.4	0.0±0.0[a]

*Mean with the same superscript along the column is not significantly different ($p\leq0.05$)
Key: THBC = Total Heterotrophic Bacteria.

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Total coliform count initially started low ($\times 10^4$) on Day 1 but increased on subsequent days, reaching 2.17×10^4 CFU/ml on Day 2. However, it decreased again to 0.11×10^4 CFU/ml on Day 5. Faecal coliform count were notably high ($13.0\pm0.1\times10^2$ CFU/ml) on Day 1, indicating possible initial contamination. However, these counts dropped significantly as fermentation progresses and became undetectable (0.0×10^2 CFU/ml) on Day 5.

The antibiotic susceptibility testing presented in Table 3 reveals varying degrees of resistance among the isolated strains. Notably, *Bacillus subtilis* and *Staphylococcus aureus* exhibited resistance to septrin and zinacef, respectively. *Bacillus cereus* and *Lactobacillus fermentum* were sensitive to the entire antibiotics used. *Streptococcus lactis* was resistant to almost all the antibiotics except rocephin and amoxicillin. While *Escherichia coli* showed resistance to erythromycin.

The Multiple Antibiotic Resistance Index of bacteria isolated from burukutu sample, is shown on Table 4. The study revealed that the isolates had multidrug resistance index of between 0.5 and 0.7 respectively.

Table 3: Antibiotic Sensitivity Pattern of Bacterial Isolates to Commercial Antibiotics

Isolates/antibiotics	CN	APX	Z	AM	A	CPX	S	SXT	E	PEF
Bacillus Subtilis	13.0	15.5	7.5	5.0	13.5	20.0	15.0	R	13.0	16.0
Bacillus cereus	20.5	21.0	19.5	20.0	20.0	22.0	21.5	19.5	23.0	29.5
Lactobacillus fermentum	23.5	23.0	22.0	21.0	21.0	29.0	22.0	22.0	21.0	24.5
Staphylococcus aureus	16.5	16.0	R	19.5	19.5	25.0	23.0	16.5	20.0	28.0
Streptococcus lactis	R	R	R	19.5	19.5	R	R	R	R	R
Escherichia coli	25.5	20.0	15.0	23.0	23.0	21.0	16.0	26.0	R	23.0

Key: A = Androcephin, PEF = Pefloxacin, GN = Gentamicin, APX = Ampiclox, Z = Zinacef, AM = Amoxicillin, R = Rocephin, CPX = Ciprofloxacin, S = Streptomycin, SXT = Septrin, E = Erythromycin.

Table 4: Multiple Antibiotics Resistant (MAR) Indices of Bacteria

MAR Index	Number (%)
0.1	0(0.00)
0.2	0(0.00)
0.3	0(0.00)
0.4	0(0.00)
0.5	3(50.0)
0.6	0(0.00)
0.7	3(50.0)

Key: MAR = Multiple Antibiotic Resistance

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Discussion

The present study has revealed the bacterial counts of samples during the production process of burukutu and during the fermentation of the guinea corn wort to produce burukutu. The total heterotrophic bacteria (THB) counts are indicative of the overall microbial load in the samples. The higher THB counts in whole grains and sprouted grains suggest that these samples have a higher population of microorganisms capable of utilizing organic carbon sources. This is expected, as these grains are less processed and thus more suitable substrate for (conducive to) microbial growth. Conversely, uncooked mash and boiled wort exhibit lower THB counts, indicating reduced microbial populations, likely due to the heat treatment during the malting and boiling processes (Atter *et al*., 2014). *Lactobacilli* are beneficial lactic acid bacteria frequently associated with fermentation. The highest LC count in uncooked mash indicates that this sample is rich in *Lactobacilli*. Sprouted grains also show significant counts of *Lactobacilli*, suggesting the presence of these bacteria, possibly due to natural fermentation processes during sprouting. In contrast, the lower LC counts in ungrounded grain and boiled wort may be attributed to the absence of specific fermentation conditions or treatments that favor *Lactobacillus* growth (Ogbonna *et al*., 2016). *Vibrio*, typically found in water and marine environments, are detected in sprouted grains and uncooked mash, albeit at relatively low levels. The absence of *Vibrio* in ungrounded grain and boiled wort suggests that these environments may not support the growth of these bacteria. The presence of *Vibrio* might be attributed to water sources used during grain processing or environmental contamination (Economopoulou *et al*., 2017). *Staphylococci* are widespread in various environments. The presence of notable Total Staphylococcal counts in ungrounded grain, uncooked mash, and sprouted grains may indicate environmental contamination or the natural occurrence of these bacteria on grains. The lower Total Staphylococcal count in boiled wort may result from the heat treatment process, which can reduce microbial loads. Coliform bacteria, often used as indicators of fecal contamination, are present in sprouted grains and uncooked mash, with similar Total coliform counts. This suggests potential fecal contamination during processing or storage. Ungrounded grain has a slightly lower count, while boiled wort shows no detectable coliforms, which is a positive indication of safety (Critzer and Doyle, 2010).

Faecal coliform, a subgroup of coliform bacteria associated with fecal contamination, are detected in sprouted grains and uncooked mash. The higher faecal coliform count in sprouted grains suggests a greater level of contamination compared to uncooked mash. The absence of FCC in ungrounded grain and boiled wort is a favorable sign of lower fecal contamination. Microbial counts in the grain samples vary depending on the sample type and processing methods. These findings are essential for evaluating the microbial safety and quality of the grains, particularly when they are used as ingredients in fermentation processes like those involved in burukutu production (Achi, 2005).
Proper sanitation, storage, and processing techniques can help mitigate potential microbial risks and ensure the safety of the final product. The fermentation process begins with a relatively high THB count, indicating the presence of a diverse population of microorganisms. As fermentation progresses, there is a significant decrease in THB count. This decline suggests that the microbial community is changing, and specific microorganisms are dominating the fermentation process. The reduction in THB count can be attributed to competition among microorganisms, the production of inhibitory compounds, and changes in environmental conditions like pH and nutrient availability. *Lactobacillus* counts are relatively stable throughout fermentation, indicating that these lactic acid bacteria are actively involved in the fermentation process. Lactobacilli play a crucial role in the production of lactic acid, which contributes to the characteristic flavor and preservation of burukutu (Togo *et al*., 2002).

The absence of *Vibrio* and Staphylococcal counts suggests that these specific groups of bacteria may not be significant contributors to burukutu fermentation or may not have survived the fermentation conditions. The fermentation starts with a relatively low TCC count, suggesting minimal initial fecal contamination. There is a notable increase in Total coliform count on the second day, indicating a potential increase in coliform bacteria. This might be due to the introduction of contaminants during the initial stages of fermentation. The TCC decreases significantly by the end of fermentation, suggesting that the fermentation process reduces the population of coliform bacteria, possibly due to the acidic conditions and competition with other microorganisms (Generose *et al*., 2016). The initial faecal coliform count is notably high, indicating possible fecal contamination during the early stages of fermentation.

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By the fifth day, the faecal coliform count becomes undetectable, indicating that the fermentation process effectively eliminates faecal coliforms. The microbial counts during burukutu fermentation reflect the dynamic nature of the process. Lactic acid bacteria, particularly lactobacilli, play a pivotal role in shaping the microbial community and contributing to fermentation's safety and quality (Lyumuegabe *et al.,* 2012). The decrease in total heterotrophic bacteria and coliform counts over time suggests that fermentation helps reduce the microbial load and enhance the preservation of the beverage. The absence of counts of *Vibrio* and *Staphylococcus* indicates a specific microbial profile associated with burukutu fermentation (Ogunbanwo *et al.,* 2013).

Monitoring these microbial changes is essential for ensuring the safety and quality of the final product. Bacillus subtilis exhibited resistance to Septrin. This resistance may be due to specific genetic factors in the strain that confer resistance to the antibiotics present in Septrin. *Staphylococcus aureus* showed resistance to Zinacef. This resistance could be attributed to the strain's genetic makeup and its ability to produce enzymes or proteins that inactivate or modify the antibiotic. Both *Bacillus cereus* and *Lactobacillus fermentum* were found to be sensitive to all the antibiotics used. This sensitivity suggests that these strains are susceptible to the antibiotics tested and do not possess mechanisms of resistance against them. *Streptococcus lactis* exhibited resistance to almost all the antibiotics, except rocephin and Amoxicillin.

The susceptibility of *Streptococcus lactis* to rocephin and Amoxicillin suggests that these antibiotics might be effective treatment options for infections caused by this strain. The resistance to other antibiotics indicates that *Streptococcus lactis* has developed mechanisms to evade those specific drugs. *Escherichia coli* showed resistance to Erythromycin. Resistance to erythromycin may result from the presence of specific genes, such as those encoding efflux pumps or enzymes that modify the antibiotic, rendering it ineffective against this strain. The results of antibiotic susceptibility testing highlight the varying degrees of resistance among the isolated bacterial strains. It is important to note that antibiotic resistance can arise due to genetic mutations or the acquisition of resistance genes from other bacteria (Yuceer *et al.,* 2016).

Monitoring and understanding the resistance profiles of bacterial strains are crucial for effective treatment and the prevention of antibiotic resistance dissemination in clinical and environmental settings. Additionally, these findings can inform healthcare practitioners about suitable antibiotic choices when dealing with infections caused by these specific strains. The MAR Index values of 0.5 to 0.7 among bacteria isolated from Burukutu samples highlight the presence of multidrug-resistant strains. This finding emphasizes the need for vigilance in food safety practices, responsible antibiotic use, and ongoing research to address antibiotic resistance concerns in food production and public health.

In conclusion, various bacterial strains were isolated from the burukutu samples, including *Bacillus subtilis*, *Bacillus cereus*, *Lactobacillus fermentum*, *Staphylococcus aureus*, *Streptococcus lactis*, and *Escherichia coli*. The isolated strains exhibited different resistance profiles to the tested antibiotics. Notably, *Bacillus subtilis* and *Staphylococcus aureus* showed resistance to specific antibiotics, while *Bacillus cereus* and *Lactobacillus fermentum* were sensitive to all antibiotics. *Streptococcus lactis* displayed resistance to most antibiotics except rocephin and Amoxicillin while *Escherichia coli* was resistant to Erythromycin. The presence of antibiotic-resistant bacteria in burukutu raises concerns about the potential transmission of antibiotic resistance to consumers and the environment. Antibiotic resistance is a global public health challenge. The presence of antibiotic-resistant bacteria in traditional fermented beverages like burukutu highlights the need for increased awareness and responsible antibiotic use in both healthcare and agriculture to combat the rise of antibiotic resistance. Further studies and surveillance of antibiotic resistance in fermented foods and beverages are warranted to better understand the extent of the issue and to develop strategies for mitigating antibiotic resistance risks in food production. In summary, the antibiotic susceptibility pattern of bacteria isolated from laboratory produced burukutu provides valuable insights into the microbial safety and quality of this traditional beverage. These findings emphasize the importance of responsible antibiotic use, stringent quality control, and ongoing research to address antibiotic resistance concerns in food production.

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Citation: Idoko *et al*. (2023). Antibiotic susceptibility pattern of bacteria isolated from laboratory produced fermented beverage ("burukutu") produced from guinea corn (*Sorghum bicolor*). *International Journal of Microbiology and Applied Sciences*. *2(2)*: 85 - 93.