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Microbiological Assessment Of Food Preparation Practices And Hygiene Standards: An Empirical Evidence Of Restaurants In Amassoma Community

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ABSTRACT
This study aimed to evaluate the microbial contamination levels of plates, water, and food samples from four different restaurants, focusing on bacterial, fungal, and *Enterobacteriaceae* counts. Samples were collected from plates, water used for washing and cooking, and prepared food at various locations (LA, LB, LC, and LD). Results indicated that bacterial counts for total heterotrophic bacteria ranged from 0.362 to 0.563 cfu/g for plates, 0.000 to 0.330 cfu/ml for water, and 0.330 to 0.429 cfu/g for food. Fungal contamination was observed in the range of 0.215 to 0.340 cfu/g for plates, 0.000 to 0.287 cfu/ml for water, and 0.230 to 0.497 cfu/g for food. *Enterobacteriaceae* counts varied from 1.050 to 1.667 MPN/100ml for plates, 0.000 to 0.703 MPN/100ml for water, and 0.000 to 0.213 MPN/100ml for food. Statistically significant differences were observed across locations, with LC and LD showing higher contamination levels, particularly in water and food. The results emphasize the importance of maintaining stringent hygiene practices, proper sanitation, and effective water treatment to reduce microbial contamination. The study highlights the need for continuous monitoring and improvement of food safety standards in restaurant settings to ensure consumer health and meet regulatory safety limits.
Keywords: Microbial Contamination, Food Safety, Hygiene Practices, Bacteria, Fungi, Enteric Bacteria

1. INTRODUCTION

Food safety and hygiene in public eating places are crucial for protecting public health. Poor hygiene practices in restaurants can result in the transmission of harmful pathogens, leading to foodborne illnesses. According to World Health Organization (WHO, 2021), foodborne diseases are caused by consuming contaminated food, which is often the result of improper handling, preparation, and storage. Common foodborne illnesses include salmonellosis, gastroenteritis, and cholera, which can be transmitted through improper food handling and inadequate sanitation practices (Hussain *et al.,* 2020). Contamination sources in food preparation areas include contaminated water, improper cleaning of utensils, and unsafe handling practices (Akbar *et al.,* 2019). Microorganisms such as bacteria, viruses, and fungi are commonly found in the food environment and can cause outbreaks if not controlled. Poor aseptic techniques, including improper handwashing, undercooked food, and cross-contamination, are major contributors to these health risks (Mabinya *et al.,* 2022).

In the Amassoma community, the lack of proper aseptic measures in local restaurants is likely contributing to microbial contamination, thus posing a risk to public health. Inadequate cleaning, improper food storage, and poor personal hygiene are common issues in these establishments (Mogaka *et al.,* 2020). The improper handling of food increases the likelihood of contamination with harmful

pathogens, leading to foodborne illnesses (Cheung *et al.*, 2019). Local restaurants often lack the necessary knowledge or resources to implement adequate food safety measures, which directly impacts the well-being of consumers. Research shows poor food handling like unwashed hands and unsanitized utensils leads to foodborne infections (Yayeh et al., 2020). Due to these public health risks, assessing food safety standards in establishments is vital to reduce contamination and ensure consumer protection.

This study is significant in providing insight into the levels of food safety and hygiene practices in the Amassoma community, helping to identify potential gaps in food safety protocols. By assessing the microbiological contamination levels in local restaurants, the study will highlight areas that need improvement, aiding in the design of appropriate interventions to address foodborne illness risks (Akinmoladun *et al.*, 2021). Additionally, the findings will assist local health authorities in establishing targeted public health campaigns or training programs for restaurant owners and workers, promoting better sanitation practices (Oba *et al.*, 2020). Understanding the factors contributing to foodborne illnesses will also help to advocate for policies that ensure safer food handling and overall hygiene, ultimately safeguarding public health. Therefore, the study plays a crucial role in improving the community's food safety standards and reducing the burden of preventable diseases.

2. METHODOLOGY

2.1 Study Area

Amassoma, located in Bayelsa State, Nigeria, is the administrative center of the Ogboin clan and the Ogboin-North Rural Development Authority within the Southern-Ijaw Local Government Area. Situated approximately 40 km south of Yenagoa, the state capital, Amassoma lies between latitude 5.20N and longitude 6.050E in the Niger Delta region, one of the world's largest deltas (Alagoa, 1999). The community serves as the temporary site for the Niger Delta University, established in 2000, leading to significant growth in student enrollment and staff population. This educational development has spurred economic activities, attracting petty traders and businesses, thereby enhancing the socio-economic landscape of Amassoma. However, this rapid growth has also introduced socio-economic challenges, including increased demand for facilities and services (Alagoa, 1999).

2.2 Sampling Technique

A purposive sampling method was employed to collect 12 samples in triplicate across 4 restaurants in 4 distinct locations within Amassoma, resulting in a total of 48 samples (12 water samples, 12 plate swabs, and 12 food samples). This approach ensures the inclusion of establishments with varying characteristics, providing a comprehensive overview of microbial contamination across different settings. The selection was based on specific criteria relevant to the study's objectives, allowing for targeted data collection (Ashbolt *et al.*, 2001).

2.3 Sample Collection

2.3.1 Water Samples

Approximately 100 ml of water was aseptically collected from storage or usage containers in sterile bottles. This method aligns with standard protocols for water sampling, ensuring the integrity of the samples (Ashbolt *et al.*, 2001).

2.3.2 Plate Swabs

Using a sterile moistened swab, surfaces of plates were swabbed post-washing and prior to serving. The swab was then placed into a sterile container with a known volume of diluent, shaken, and plated on appropriate agar media. This technique is recommended for surface sampling in microbiological analyses (Miles & Misra, 1938).

2.3.3 Prepared Food

Approximately 10–20 grams of commonly served meals were collected in sterile containers. This quantity is sufficient for microbial analysis while minimizing potential contamination (ICMSF, 1986).

2.4 Microbiological Analysis

2.4.1 Total Heterotrophic Bacteria Count (THBC)

Samples were cultured using the pour plate method on Nutrient Agar. This technique is effective for enumerating total viable bacteria in food and water samples (ISO, 1981).

2.4.2 Total Fungi Count

Sabouraud Dextrose Agar was employed to culture fungi. This medium supports the growth of a wide range of fungi, facilitating accurate enumeration (Miles & Misra, 1938).

2.4.3 Enteric Bacteria Count

MacConkey Agar and Eosin Methylene Blue (EMB) Agar were used to detect and enumerate enteric bacteria. These selective media differentiate lactose fermenters from non-fermenters, aiding in the identification of potential pathogens (Ashbolt *et al.*, 2001).

2.4.4 Incubation Conditions

Plates inoculated for bacterial counts were incubated at 35–37°C for 24–48 hours, as this temperature range is optimal for the growth of most pathogenic bacteria. Fungal plates were incubated at room temperature for 3–5 days to allow adequate fungal growth (Miles & Misra, 1938).

2.5 Quality Control

To ensure the reliability of results, sterile materials were used throughout the sampling and analysis process. Media control plates were included to monitor for contamination, and duplicate samples were analyzed to confirm consistency and accuracy of the microbial counts. These practices are essential for maintaining the integrity of microbiological studies (ICMSF, 1986).

2.6 Data Analysis

Mean microbial counts were calculated and expressed as colony-forming units per milliliter (cfu/ml) for liquid samples and per gram (cfu/g) for solid samples. The microbial loads across different sample types (water, plate swabs, and food) were compared using appropriate statistical methods. Analysis of Variance (ANOVA) was applied to determine significant differences among groups, with a significance level set at $p<0.05$. This statistical approach is standard for comparing means across multiple groups in microbiological research (USP, 1978).

3. RESULTS AND DISCUSSION

3.1 Total Heterotrophic Bacterial Count

The results on average bacterial counts of washed plates, stored water used for washing and cooking, and prepared food from the restaurants were presented. For the plates, bacterial counts ranged from 0.362 to 0.563 cfu/g, with the highest count observed at LC (0.563 ± 0.03 cfu/g) and the lowest at LB (0.362 ± 0.02 cfu/g).

Table 1: Results on the total heterotrophic Bacterial Counts

Locations	Plate (X 10^3 (cfu/g)	Water (X 10^3 cfu/ml)	Food (X 10^3 cfu/g
LA	0.461±0.03ab	0.000±0.00a	0.354±0.02a
LB	0.362±0.02a	0.2023±0.10a	0.330±0.02a
LC	0.563±0.03c	0.33±0.011b	0.429±0.09b
LD	0.407±0.02a	0.1100±0.190a	0.377±0.02a

The bacterial counts for the water samples ranged from 0.000 to 0.330 cfu/ml, with the highest count recorded at LC (0.33 ± 0.011 cfu/ml) and the lowest in LA (0.000 ± 0.00 cfu/ml). The heterotrophic bacterial count for food samples ranged from 0.330 to 0.429 cfu/g, with LC having the highest count (0.429 ± 0.09 cfu/g) and LB the lowest (0.330 ± 0.02 cfu/g). These results highlight variability across different locations and demonstrate differing hygiene practices or contamination sources.

There was a significant difference in bacterial counts between locations for all tested samples, with the highest bacterial load observed in LC and the lowest in LB for plates, water, and food. Statistical analysis of the bacterial counts indicates significant differences between the locations (p-value < 0.05), which

suggests that environmental factors, including hygiene practices and water quality, contributed to variations. Additionally, there were significant differences between bacterial counts for plates, water, and food samples within each location, reflecting possible contamination routes or handling practices that influence microbial growth.

According to WHO and NAFDAC standards, the acceptable limit for heterotrophic bacterial counts in food and water is typically lower than the levels recorded in the study. While values in LA and LB are closer to acceptable standards, LC shows elevated bacterial counts in both water and food samples. This suggests potential contamination risks that require addressing to meet regulatory safety limits for public health.

Likely sources of contamination in this study include water quality and food handling practices. Poor sanitation of water used for washing and cooking can introduce bacteria to the food and plates, especially if the water is not adequately treated. Inadequate handwashing by food handlers or improper storage of food can also contribute to bacterial contamination (Ray, 2004). Aseptic practices, such as proper handwashing, sanitation of food contact surfaces, and treatment of water, are critical to minimizing contamination (Mena & Gerba, 2009). The significant bacterial counts observed at LC suggest lapses in these practices.

The detected microbial levels in food, water, and plates pose potential health risks. Bacterial contamination can lead to foodborne illnesses, with symptoms ranging from mild gastrointestinal distress to severe food poisoning, depending on the specific pathogens involved (Doyle & Erickson, 2006). High bacterial counts indicate a failure in maintaining proper hygiene practices, which may increase the risk of transmission of pathogens like *Escherichia coli* and *Salmonella*. Contaminated water can serve as a vehicle for microbial transmission, especially in underregulated areas, leading to outbreaks of waterborne diseases (World Health Organization, 2011).

Long-term exposure to contaminated food and water may also increase the risk of chronic conditions, particularly in vulnerable populations such as children and the elderly (Mearns, 2007).

The bacterial counts recorded in this study are comparable to findings in other regions, indicating widespread contamination in food establishments. For example, a study in Lagos, Nigeria, found high bacterial counts in food and water, highlighting similar contamination issues (Akinmoladun et al., 2014). In contrast, studies in well-regulated regions have reported much lower bacterial counts, demonstrating the effectiveness of stringent hygiene practices and water treatment (Cohen et al., 2007). A study in Egypt found a significant correlation between inadequate food handling practices and increased bacterial counts in food (El-Senousy et al., 2016). The findings from this study underscore the need for better water and food safety practices to mitigate health risks, particularly in settings with high bacterial loads.

3.2 Total Fungal Count

The results on average fungal counts of washed plates, stored water used for washing and cooking, and prepared food from the restaurants were presented. For the plates, fungal counts ranged from 0.215 to 0.340 cfu/g, with the highest count observed at LC (0.340 ± 0.173 cfu/g) and the lowest at LA (0.215 ± 0.107 cfu/g). The fungal counts for the water samples ranged from 0.000 to 0.287 cfu/ml, with the highest count recorded at LC (0.287 ± 0.143 cfu/ml) and the lowest in LB (0.000 ± 0.000 cfu/ml). The fungal count for food samples ranged from 0.230 to 0.497 cfu/g, with LD having the highest count (0.497 ± 0.081 cfu/g) and LB the lowest (0.230 ± 0.115 cfu/g). These results indicate variation in fungal contamination across different locations and may reflect differences in handling practices or environmental factors.

Table 2: Results on the total fungal Counts

Locations	Plate (X 10^2 cfu/g)	Water (X 10^2 cfu/ml)	Food (X 10^2 cfu/g)
LA	0.215±0.107a	0.113±0.113a	0.370±0.035a
LB	0.237±0.121a	0.000±0.000a	0.230±0.115a
LC	0.340±0.173a	0.287±0.143a	0.320±0.277a
LD	0.307±0.120a	0.208±0.04a	0.497±0.081a

The statistical analysis revealed no significant differences in fungal counts across locations for all samples (plates, water, and food), as the counts for most locations were in the same range. The fungal counts were comparable for plates (0.215–0.340 cfu/g), water (0.000–0.287 cfu/ml), and food (0.230–0.497 cfu/g) samples. This suggests that fungal contamination might be widespread, with handling practices or environmental factors influencing contamination levels. The results further imply that fungal counts do not significantly vary based on location, although differences between food samples may be observed due to storage or handling procedures.

The results for fungal contamination in the studied samples fall within permissible limits for water and food based on WHO and NAFDAC standards. Although the counts are relatively low, fungal contamination levels in food, particularly at LD (0.497 cfu/g), may be concerning. These levels are above typical acceptable standards for food hygiene and should be monitored to ensure safety, especially in terms of storage and handling practices.

Fungal contamination can occur due to improper food handling, inadequate storage, and poor sanitation practices (Lund et al., 2000). Water used for washing and cooking may serve as a source of fungal spores if it is not adequately filtered or treated (Leong et al., 2008). The presence of fungi on food may be due to storage conditions such as humidity and temperature, which favor fungal growth (De Hoog et al., 2000). The findings from this study suggest lapses in maintaining proper hygiene and controlling environmental conditions, which promote fungal contamination.

Fungal contamination in food, water, and plates can pose significant health risks, especially if the fungal species involved are pathogenic. Fungi such as *Aspergillus* and *Penicillium* can produce mycotoxins, which are toxic to humans and can cause a variety of health problems, including gastrointestinal disturbances, liver damage, and even cancer in prolonged exposure (Bennett & Klich, 2003). Fungal contamination in water, such as *Candida* or *Aspergillus*, can also result in waterborne diseases, particularly when water is consumed or used in food preparation (World Health Organization, 2004). Mycotoxins can be harmful even at low levels, with potential long-term effects on health (Gil-Exojo et al., 2007). Therefore, it is crucial to ensure proper hygiene and control of fungal contamination in food establishments to prevent the spread of these harmful microorganisms.

Similar studies have shown that fungal contamination is prevalent in food and water in several regions. For instance, a study conducted in Nigeria revealed fungal contamination in both water and food, highlighting the risks of improper handling and storage practices (Adebayo-Tayo et al., 2015). In contrast, studies conducted in developed countries report lower fungal contamination levels, attributed to better food safety regulations and more stringent hygiene practices (Müller et al., 2012). A study in Egypt also found that fungal counts in food were significantly higher in improperly stored food, emphasizing the need for good handling practices (El-Senousy et al., 2016). This study's results, particularly the higher counts in LD and LC, are consistent with findings from regions where water and food safety practices are inconsistent. These findings reinforce the importance of maintaining proper hygiene standards to reduce fungal contamination.

3.3 Enteric Bacterial

The results on average *Enterobacteriaceae* counts of washed plates, stored water used for washing and cooking, and prepared food from the restaurants were presented in Table 3. For the plates, *Enterobacteriaceae* counts ranged from 1.050 to 1.667 MPN/100ml, with the highest count observed at LD (1.667 ± 0.667 MPN/100ml) and the lowest at LC (1.050 ± 0.050 MPN/100ml). The bacterial counts for water samples ranged from 0.000 to 0.703 MPN/100ml, with the highest count recorded at LC (0.703 ± 0.881 MPN/100ml) and the lowest in LA and LB (both 0.000 ± 0.00 MPN/100ml). The *Enterobacteriaceae* count for food samples ranged from 0.000 to 0.213 MPN/100ml, with the highest count in LD (0.213 ± 0.233 MPN/100ml) and the lowest in LA and LB (both 0.000 ± 0.00 MPN/100ml). These results suggest variability in bacterial contamination across locations and samples.

Statistical analysis showed significant differences in Enterobacteriaceae counts across locations ($p < 0.05$). LC and LD had higher counts in both water and food samples, while LA and LB showed no detectable bacteria, indicating varying contamination levels between sites. This indicates that hygiene

practices and environmental factors contributed to significant differences in bacterial contamination levels.

Table 3: Results on the total *Enterobacteriaceae* Counts

	Plate (MPN/100ml)	Water (MPN/100ml)	Food (MPN/100ml)
LA	1.340±1.831	0.00±0.00a	0.000±0.00a
LB	1.070±0.966	0.00±0.000a	0.0000±0.00a
LC	1.050±0.050	0.703±0.881b	0.183±0.961b
LD	1.667±0.667	0.317±0.316b	0.213±0.233b

According to WHO and NAFDAC standards, enterotrophic bacteria in food and water should be absent or present in very low counts, particularly in water used for food preparation. The results from LA and LB show compliance with the standards, as no enterotrophic bacteria were detected in the water or food. However, the counts at LC and LD, particularly for water, are concerning and should be addressed to ensure safety standards are met.

The sources of *Enterobacteriaceae* contamination in this study may include water used for washing and cooking, as well as food handling practices. Contaminated water, particularly if not properly treated, can introduce bacteria into food or onto food-contact surfaces (Smith et al., 2002). Improper hygiene during food handling and storage, such as inadequate handwashing and unclean equipment, may also contribute to contamination (Bai et al., 2011). Additionally, environmental factors like poor sanitation and storage conditions may provide favorable conditions for bacterial growth. These findings underscore the importance of improving sanitation and water treatment practices.

Enterobacteriaceae contamination in food and water can have serious health implications. The presence of *Enterobacteriaceae* and other enterotrophic bacteria in food and water is a primary cause of foodborne diseases, including gastroenteritis, diarrhea, and vomiting, which are common symptoms of infections caused by these bacteria (Moyne, 2011). Ingestion of contaminated food or water can lead to outbreaks, especially in vulnerable populations such as children, the elderly, and immune compromised individuals (McKeown, 2005). The absence of enterotrophic bacteria in LA and LB is favorable, but contamination in LC and LD presents a risk for outbreaks of gastrointestinal diseases. Chronic exposure to contaminated food and water can also lead to long-term health effects, including the development of antibiotic-resistant bacteria (Graham et al., 2009). Therefore, effective monitoring, treatment, and proper handling of food and water are essential to mitigate health risks.

This study's findings on *Enterobacteriaceae* contamination are consistent with other studies conducted in regions with variable water and food safety practices. In a study in Lagos, Nigeria, *Enterobacteriaceae* contamination was observed in food and water samples, highlighting the risk of foodborne illness in environments with inadequate sanitation (Akinmoladun et al., 2014). Similarly, a study in Egypt found high bacterial contamination in food and water, with implications for public health (El-Senousy et al., 2016). In contrast, studies from developed countries with stricter food safety regulations and sanitation practices report lower levels of *Enterobacteriaceae* contamination (Eisenstein et al., 2006).

These findings emphasize the need for stringent hygiene practices, proper water treatment, and effective food handling in regions with high contamination levels. The elevated counts at LC and LD underscore the importance of addressing these issues to meet international health standards.

CONCLUSION

In conclusion, the study revealed significant variability in microbial contamination across different locations, with notable differences in bacterial and fungal counts for plates, water, and food samples. Locations such as LC and LD exhibited higher microbial contamination, particularly in water and food, indicating potential lapses in hygiene and sanitation practices. Although LA and LB had lower contamination levels, the presence of enterotrophic bacteria and fungi in some samples suggests areas for improvement in food safety protocols. The results emphasize the need for stringent hygiene measures, proper water treatment, and effective food handling practices to minimize contamination risks. Moreover,

the findings highlight the importance of continuous monitoring to ensure food and water safety, in line with international health standards. By addressing these challenges, the risk of foodborne diseases and related health issues can be significantly reduced, ensuring a safer environment for consumers.

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