

**COMPARISON OF BACTERIAL LOAD ASSOCIATED WITH DRY AND  
WET FUFU SOLD IN BENIN CITY**

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**UNIVERSITY OF BENIN  
BENIN CITY**

**SEPTEMBER, 2023.**

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**A RESEARCH PROJECT SUBMITTED TO THE DEPARTMENT OF  
MICROBIOLOGY, FACULTY OF LIFE SCIENCES, UNIVERSITY OF  
BENIN, BENIN CITY, IN PARTIAL FULFILLMENT OF THE  
REQUIREMENT FOR THE AWARD OF DEGREE B.Sc. (HONS) IN  
MICROBIOLOGY, UNIVERSITY OF BENIN, BENIN CITY.**

**SEPTEMBER, 2023**

## **CERTIFICATION**

This is to certify that this project work was carried out by Oruaroghene Daniel OMOSOMOFA in the Department of Microbiology, Faculty of Life Sciences, University of Benin, Benin City under my supervision.

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**PROF B. A. OMOGBAI**  
(Project supervisor)

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**Date**

## **APPROVAL**

This project work was carried out by Oruaroghene Daniel OMOSOMOFA in partial fulfillment of the award of a Bachelor of Science, B.Sc (Hons) degree in the Department of Microbiology, University of Benin, Benin City.

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**PROF. (MRS) F. I. AKINNIBOSUN**  
(Head of Department)

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**Date**

## **DEDICATION**

This project work is dedicated to God Almighty for his unconditional love, faithfulness and mercy, and to my parents Mr and Mrs Omosomofa for their unending love and support.

## ACKNOWLEDGEMENTS

I want to first appreciate God almighty for His mercies and grace that has brought me so far; for helping me realize many of my dreams within this great citadel of learning. To him be all glory and adoration.

I am deeply indebted to my supervisor, Prof. B. A. Omogbai for his invaluable guidance, mentorship, and expertise throughout this project. The unwavering support, insightful feedback, and patience have been instrumental in shaping the direction of my work. I am grateful for the opportunities provided, the constructive discussions, and the trust placed in me. Your belief in my abilities has been a driving force behind my accomplishments, and I am grateful for the knowledge and skills I have gained under your supervision.

I would like to express my heartfelt gratitude to my parents, Mr and Mrs Nathaniel Omosomofa whose unwavering love, encouragement, and sacrifices have been the foundation of my journey. Your belief in me and constant support have given me the strength to pursue this project and reach new heights. Thank you for always being there for me, for cheering me on during the challenging times, and for instilling in me the values of hard work and determination.

Furthermore, I would like to acknowledge the contributions of my friends and course-mates who have supported me throughout this project. Your encouragement, discussions, and shared experiences have been a source of inspiration and motivation.

Lastly, I would like to express my gratitude to all the researchers, authors, and individuals whose work and insights have laid the foundation for my project. Your contributions have been instrumental in shaping my understanding and providing a solid framework for my research

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## ABSTRACT

Fufu is a starchy dish made from ingredients such as cassava, yam, or plantain. Fufu plays a significant role in African cuisine and cultural traditions, often served alongside various soups, stews, and sauces. The aim of this study was to carry out the comparative analysis of the bacterial load associated with dry and wet fufu sold in Benin City. Six (6) different samples were collected from Fagcoop supermarket in University of Benin, Benin City. Three dry Fufu samples and three wet Fufu samples were obtained. Serial dilution was carried out using the dilution of  $10^3$ ,  $10^4$  and  $10^5$  for Nutrient agar, *Salmonella* and *Shigella* agar (SSA) and De Man, Rogosa and Sharpe agar (MRS) agar media were used. The bacterial isolation of fufu samples was carried out on the three agar mediums using the pour plate method. Cultural, morphological and biochemical test were employed for the identification of the isolates. Biochemical tests carried out include urease test, citrate utilization test, hydrogen sulphide test, indole test, oxidase test, catalase test and sugar fermentation test. Using disc diffusion methods, antibiotic susceptibility test was carried out for the bacterial isolates. In this study, species of *Corynebacterium*, *Micrococcus*, *Bacillus*, *Salmonella* and *Pseudomonas* were among the various bacterial genera isolated of which *Salmonella* is pathogenic. *Lactobacillus* sp tested *Pseudomonas* sp tested negative for all sugar fermentation tests while *Bacillus subtilis* and *Lactobacillus* sp tested positive for all sugar fermentation tests. The antibacterial activity test with Gram negative and Gram positive antibiotic discs as well as chitosan was evaluated. It was observed that chitosan had inhibitory effect on all the microorganisms isolated from the dry and wet fufu samples with zones of inhibition ranging from 6mm (at 25 mg/ml) to 48mm (at 100mg/ml). The zone of inhibition for *Corynebacterium* sp ranged from 16mm-36mm. The evaluated antibiotic resistance in the isolated bacteria shows the need for more usage of antibiotic in the industrial production of fufu to help minimize the chances of fufu being contaminated by pathogenic microorganisms. The consumption of wet fufu and dry fufu is not advised since *Salmonella* spp (causative organism of Salmonellosis) was isolated from sample 2 of wet fufu and sample 1 of dried fufu. Proper hygienic practices should be carried out to avoid the contamination of fufu with pathogenic organisms such as *Salmonella* spp.

## CHAPTER ONE

### 1.0

### INTRODUCTION

#### 1.1 Background of Study

Fufu is a staple food widely consumed in West and Central Africa, particularly in countries like Nigeria, Ghana, Cameroon, and Ivory Coast. It holds a central place in the region's culinary traditions. Fufu is not a single dish but rather a type of food made from starchy ingredients such as cassava, yam, plantains, or cocoyam. A method of the preparation of fufu involves boiling or steaming the starchy ingredient until it becomes soft and then pounding or mashing it to achieve a smooth, dough-like consistency (Atlas *et al.*, 1986). This labor-intensive process is often done manually using a mortar and pestle, although mechanical methods are also employed. Fufu is known for its versatility, as it can be paired with a variety of soups, stews, or sauces. It is typically served alongside dishes like egusi soup, groundnut soup, or palm nut soup, depending on the region and local preferences. Fufu has cultural minary significance, as it plays a vital role in social gatherings, celebrations, and daily meals across many African communities. The preparation methods and preferred starchy ingredients can vary from one region to another, resulting in diverse flavors and textures that make fufu a fascinating and beloved part of African cuisine. (Achi and Akomas, 2006).

Fufu is a starchy dish made from ingredients such as cassava, yam, or plantain. Fufu plays a significant role in African cuisine and cultural traditions, often served alongside various soups, stews, and sauces (Ezeronye *et al.*, 2003). When it comes to Fufu, there are two main variations: dry Fufu and wet Fufu. The distinction between these two types lies in their preparation methods and consistency. Dry Fufu is typically made by drying and milling starchy ingredients into a fine powder, while wet Fufu involves boiling and mashing the ingredients into a smooth paste. There

has been growing interest in studying the microbial composition of various foods, including Fufu. Bacteria play a crucial role in food safety and can have both positive and negative effects on human health. Understanding the types and presence of bacteria in different types of Fufu can provide insights into food safety considerations, fermentation processes, and potential health benefits (Iwuoha *et al.*, 1996).

Fufu is a staple food in African cuisine, particularly in West and Central Africa. It carries cultural significance, being deeply rooted in African culinary traditions and often shared during social gatherings (Singh *et al.*, 2011). Fufu provides carbohydrates, dietary fiber, and essential micronutrients, contributing to the nutritional value of African diets. It is versatile, with regional variations and various accompaniments. Fufu also has economic importance, supporting agriculture, trade, and export opportunities. Overall, fufu plays a vital role in African culinary heritage, promoting cultural identity and showcasing the diversity of African cuisine. There are different variations between dry and wet fufu. The variations include the preparation method, texture, cooking time, regional preference and serving method (Cardoso *et al.*, 2008).

## **1.2 Aim and Objectives**

My aim of this study was to carry out the comparative analysis of the bacterial load associated with dry and wet fufu sold in Benin City.

The specific objectives were to;

1. Determine the microbial diversity
2. Identify specific bacterial differences
3. Assess food safety implications
4. Investigate factors influencing bacterial composition
5. Provide insights for quality improvement

## CHAPTER TWO

### 2.0

### LITERATURE REVIEW

#### 2.1 Fufu

Fufu is a fermented wet-paste made from cassava. Fufu is made by steeping whole or cut peeled cassava roots in water to ferment for a maximum of three days, depending on ambient temperature. During steeping, fermentation decreases the pH, softens the roots, and helps to reduce potentially toxic Cyanogenic compounds. When sufficiently soft, the roots are taken out, broken by hand, and sieved to remove the fibers. At present, processors sieve manually by adding water to the retted mass on nylon or cloth screens. The fiber produced as a by-product is sold for animal feed, either in its wet form or after sun-drying. The sieved mass is allowed to sediment in a large container for about 24 hours. After sedimentation, the water is poured off while the fine, clean sediment (mainly starch) is dewatered using a high powered Press. The cake is then sifted before drying (Carlsson *et al.*, 1999). Apart from being easy to prepare the consumable form, dried fufu has the advantages of having a longer shelf life, being more convenient to store, and less bulky. When cooked, fufu is a creamy/white smooth textured product. When properly packaged and stored, dried fufu flour has a shelf-life of six months or more (Tamand *et al.*, 2016).

#### 2.2 Regional Variation of Fufu

Fufu is a traditional staple food in several countries in West and Central Africa, including Ghana, Nigeria, Cameroon, Sierra Leone, and Côte d'Ivoire. While the basic concept of fufu remains the same across these regions, there are notable variations in terms of ingredients, preparation methods, and cultural significance. These regional differences contribute to the diverse flavors, textures, and names associated with fufu. For example the Ghanaian fufu, Nigerian fufu, Cameroonian fufu, Sierra Leonean fufu and Ivorian fufu (Coker and Achi, 2018).

## 2.3 Types of Fufu

There are two types of fufu. They are the wet fufu and the dry fufu.

### 2.3.1 Wet Fufu

Wet Fufu is a traditional African staple food that holds significant cultural and culinary importance. It is consumed in various countries across the continent, with slight variations in preparation and ingredients based on regional preferences. Wet Fufu is known for its distinct texture, flavor, and its role as a versatile accompaniment to a wide range of soups, stews, and sauces (Coker and Achi, 2018).

Fufu, in general, refers to a dough-like food made from starchy ingredients such as cassava, yam, or plantain. Wet Fufu specifically refers to the variant that undergoes a boiling and mashing process, followed by the addition of water to achieve a smoother and slightly liquid consistency compared to its dry counterpart (Davies *et al.*, 2008).

The preparation process of wet Fufu involves selecting high-quality starchy ingredients and boiling them until they become soft and easily mashable. The softened ingredients are then mashed and kneaded to create a smooth paste. Water is gradually added to adjust the consistency, resulting in a wet, cohesive mixture. One distinguishing aspect of wet Fufu is the fermentation process it undergoes. Microorganisms, particularly lactic acid bacteria (LAB), play a crucial role in the fermentation of wet Fufu. These bacteria convert sugars present in the starchy ingredients into lactic acid, which lowers the pH of the mixture and contributes to its unique flavor profile and texture (Hernande *et al.*, 1995). Wet Fufu's microbial characteristics are shaped by the fermentation process. LAB, known for their beneficial properties, are often present in wet Fufu and contribute to its preservation and sensory attributes. However, it's important to consider potential risks associated with bacterial contamination and the proper handling and storage of wet Fufu to maintain food safety (Carlsson *et al.*, 1999).

In this discussion, we will explore the preparation process of wet Fufu, the role of fermentation and microbial characteristics, as well as the food safety considerations associated with this traditional African dish. By understanding the microbial aspects and cultural significance of wet Fufu, we can appreciate its rich heritage while ensuring safe and enjoyable culinary experiences (Lateef *et al.*, 2006).

### **2.3.1.1 Production Process of Wet Fufu**

The production process of wet fufu with fermentation typically involves the following steps:

#### **(a). Selection and Preparation of Raw Materials**

The main ingredient for wet fufu production is usually cassava. The cassava roots are harvested and thoroughly cleaned to remove dirt and other impurities (Abruha *et al.*, 2016).

#### **(b). Peeling and Washing**

The cassava roots are peeled to remove the outer skin. The peeled roots are then washed to further remove any remaining dirt or debris (Abruha *et al.*, 2016).

#### **(c). Grating**

The peeled and washed cassava roots are grated using a mechanical grater or a traditional grater made of metal or wood. This process breaks down the cassava into smaller pieces, creating a mash-like consistency (Abruha *et al.*, 2016).

#### **(d). Fermentation**

The grated cassava mash is transferred to a fermentation vessel, which can be a large container or a specially designed fermentation pit. The mash is allowed to ferment for a period ranging from 1 to several days, depending on the desired level of fermentation. During fermentation, natural microorganisms present in the cassava, such as lactic acid bacteria, yeast, and other beneficial bacteria, convert the starches in the cassava into simpler sugars and organic acids. This

fermentation process contributes to the characteristic flavor, texture, and aroma of the fufu (Adegoke and Babalola, 1988)

**(e). Pressing**

After fermentation, the fermented cassava mash, now known as “dough,” is pressed to remove excess liquid and obtain a more solid consistency. This can be done using a traditional press, where the dough is placed in a cloth bag and squeezed to extract the liquid, or using a mechanical press (Adegoke and Babalola, 1988).

**(f). Shaping and Cooking**

The pressed dough is divided into smaller portions and shaped into round or cylindrical balls. These balls are then cooked by boiling or steaming until they become soft and fully cooked. The cooking time may vary depending on the size of the fufu balls (Carlsson *et al.*, 1999).

**(g). Pounding or Mashing**

Once cooked, the fufu balls are pounded or mashed using a mortar and pestle or a specialized pounding tool. This process involves repeatedly pounding or mashing the balls to break them down and create a smooth, cohesive fufu dough (Achi and Akomas, 2006).

**(h). Final Preparation**

The pounded or mashed fufu dough is shaped into serving portions or larger portions for storage. It is traditionally served with various soups or stews. (Cardoso *et al.*, 2008).

### **2.3.2 Dry Fufu**

Dry fufu, also known as instant fufu or powdered fufu, is a convenient and ready-to-use alternative to traditional fufu made from fresh cassava or other starchy ingredients. Dry fufu is produced by processing cooked fufu dough into a dehydrated form that can be reconstituted with water when needed. The process of making dry fufu involves cooking the fufu dough traditionally, which is usually made by boiling or steaming cassava, plantains, or yams until they become soft. The

cooked dough is then pounded or mashed to achieve the desired texture. The resulting fufu dough is then dehydrated using methods such as drum drying or freeze drying to remove the moisture content. Once the moisture is removed, the dehydrated fufu dough is ground into a fine powder. The powder is then packaged and sealed, ready for sale and consumption. To prepare dry fufu for consumption, it is typically mixed with hot water and stirred until it forms a smooth, thick consistency similar to traditional fufu. Dry fufu offers the advantage of convenience and longer shelf life compared to traditional fufu, as it can be stored for extended periods without refrigeration. It provides an easy and quick way to prepare fufu dishes, especially in areas where fresh ingredients are not readily available. It's important to note that the specific process and ingredients used to make dry fufu may vary depending on the brand or manufacturer (Lateef *et al.*, 2006).

### **2.3.2.1 Production Process of Dry Fufu**

#### **(a). Sorting**

Sorting is the initial step in the production process of fufu flour, where the cassava roots are carefully examined and selected to ensure that only wholesome roots are used for further processing. This step is crucial as it helps to eliminate any damaged, diseased, or substandard roots, ensuring that the final product meets the required quality standards. Here is a more detailed explanation of the sorting process (Achi and Akomas, 2006).

#### **(b). Weighing**

Weighing is an important step in the production process of fufu flour, where the sorted cassava roots are accurately measured to determine the quantity of cassava being processed. This step ensures that the right amount of cassava is used, allowing for better control over the production process. Here is a more detailed explanation of the weighing process (Bamidele *et al.*, 2015).

**(c). Peeling**

Peeling is a significant step in the production process of fufu flour, where the outer peels or skins of the cassava roots are removed. This process is essential to eliminate any dirt, impurities, or toxins that may be present on the surface of the roots. Here is a brief explanation of the peeling process (Achi and Akomas, 2006).

**(d). Washing**

Peeling in the production process of fufu flour involves removing the outer peels or skins of cassava roots. This step is important to eliminate dirt, impurities, and potential toxins. Peeling can be done manually or with the help of mechanical equipment, improving efficiency and ensuring food safety (Bamidele *et al.*, 2015).

**(e). Soaking**

Soaking is a step in the production process of fufu flour where the peeled cassava roots are immersed in clean water for a period of time. This process helps to soften the roots, making them easier to process and reducing the required cooking time. It also aids in the removal of bitter compounds and improves the texture of the final product. (Bamidele *et al.*, 2015).

**(f). Pulping and sifting**

Pulping is a step in the production process of fufu flour where the soaked cassava roots are ground or mashed into a pulp. This is typically done using mechanical equipment such as grinders or blenders. The pulping process breaks down the cassava roots into a smooth consistency, facilitating further processing. Sifting, on the other hand, involves passing the pulped cassava through a fine mesh or sieve to separate the fiber or coarse particles from the smooth pulp. This helps to achieve a finer texture and remove any undesirable components. The sifted pulp is

collected, while the coarse particles are discarded or used for other purposes (Achi and Akomas, 2006).

**(g). Sedimentation**

Sedimentation is a natural process that occurs during the production of fufu flour. After pulping the cassava roots, the resulting mixture is allowed to sit undisturbed for a period of time. During this time, solid particles in the mixture settle to the bottom due to their weight, while lighter particles and liquid form a layer on top. This separation process is called sedimentation. The heavier solids, consisting of starch particles and other impurities, settle down as sediment at the bottom of the container. The liquid layer, which contains water and some dissolved components, remains on top. The sedimentation process is crucial as it helps to remove impurities and separate the desired starch from the liquid. After sedimentation, the liquid layer is carefully drained or decanted, while the settled sediment is further processed to extract the starch. This starch is then dried and ground to produce the final fufu flour product (Achi and Akomas, 2006).

**(h). Dewatering**

Dewatering is a process that involves removing excess water from a substance or material. In the context of fufu flour production, dewatering refers to the step where the starch sediment obtained from sedimentation is further processed to reduce its moisture content. After the sedimentation process, the starch sediment contains a significant amount of water. Dewatering is performed to remove this water and obtain a drier starch product. This is typically achieved through methods such as pressing, centrifugation, or filtration. During dewatering, the starch sediment is subjected to mechanical pressure or centrifugal force to squeeze out the water. Alternatively, it may be filtered through a porous medium that allows water to pass through while retaining the starch particles. The goal of dewatering is to reduce the moisture content of the starch to a suitable level

for further processing and storage. The dewatered starch is then typically dried to remove any remaining moisture, resulting in a stable and shelf-stable fufu flour product (Bamidele *et al.*, 2015).

**(i). Granulating**

Granulating is the process of converting small particles or powders into larger granules or pellets. In fufu flour production, granulating involves agglomerating dried and powdered fufu flour into larger particles. This improves flowability, reduces dust formation, and enhances the dispersibility of the flour when mixed with water. Granulation can be achieved through wet granulation using liquid binders or dry granulation through compacting the powder under pressure. The resulting granules are easier to handle, package, and prepare for fufu consumption (Achi and Akomas, 2006).

**(j). Drying**

Drying is a crucial step in the production process of fufu flour, where the wet or moist material is subjected to controlled conditions to remove moisture and achieve a desired level of dryness. In the context of fufu flour production, drying refers to the process of removing the remaining moisture from the granulated fufu flour. Drying is important for several reasons. It enhances the shelf life of the fufu flour by preventing the growth of microorganisms and reducing the risk of spoilage. It also improves the texture, stability, and quality of the final product (Akyildiz *et al.*, 2006).

There are various methods of drying that can be employed, including air drying, sun drying, drum drying, and vacuum drying. These methods involve exposing the fufu flour to heat or airflow to facilitate moisture evaporation. The specific drying method used may depend on factors such as the scale of production, available equipment, and desired product characteristics. During the drying process, the fufu flour is spread out in a thin layer or passed through drying equipment,

allowing the moisture to evaporate. The temperature, airflow, and duration of drying are carefully controlled to ensure efficient moisture removal without negatively affecting the quality of the flour. Once the drying process is complete, the fufu flour reaches a moisture content suitable for storage and consumption. It is then packaged and stored in a dry environment to maintain its quality until it is used to prepare fufu. Overall, drying plays a vital role in fufu flour production by extending shelf life, enhancing quality, and ensuring a stable product that can be stored and used over an extended period of time (Coker and Achi, 2018).

#### **(k). Sieving**

The sieving step in fufu flour production involves passing the dried and granulated fufu flour through a sieve or mesh to remove any remaining lumps or coarse particles. This process helps to achieve a finer and more uniform texture in the final product. During sieving, the fufu flour is poured onto the sieve, and gentle agitation or shaking is applied to allow the smaller particles to pass through the sieve's openings. The larger particles or lumps that cannot pass through the sieve are retained and discarded or reprocessed. Sieving ensures that the fufu flour is free from any undesirable coarse or uneven particles, resulting in a smoother and more consistent texture. It also improves the dispersibility of the flour when mixed with water, making it easier to prepare fufu without clumps. The sieved fufu flour is collected and packaged for storage or distribution, ready to be used in the preparation of fufu dishes (Achi and Akomas, 2006).

#### **(l). Milling**

The milling process in fufu flour production involves grinding or pulverizing the dried and sieved fufu flour into a fine powder. This step helps to further refine the texture and particle size of the flour, making it easier to dissolve and cook when preparing fufu. (Singh *et al.*, 2011).

During milling, the fufu flour is fed into a milling machine or grinder, which applies mechanical force to break down the particles into smaller sizes. The milling equipment may utilize rotating blades, burrs, or other mechanisms to achieve the desired level of fineness. The duration and intensity of the milling process can vary, depending on the desired particle size and the specific characteristics of the fufu flour. The goal is to obtain a smooth and uniform powder that dissolves easily in water without forming lumps. Once the milling process is complete, the fufu flour is typically collected and packaged for storage or distribution. The finely milled flour is ready to be used in the traditional method of cooking fufu, where it is mixed with water and cooked into a thick, starchy paste. Milling plays a crucial role in fufu flour production by ensuring the desired texture, ease of preparation, and overall quality of the final product (Adegoke and Babalola, 2006).

#### **(m). Blending**

Blending is a step in fufu flour production that involves combining different ingredients to create a homogeneous mixture. In the context of fufu flour, blending refers to the process of mixing the finely milled fufu flour with other ingredients, such as water or other additives, to create a uniform blend. During blending, the fufu flour is typically measured and added to a mixing vessel or container. Water or other liquids are gradually added to the flour while agitating or stirring the mixture. This ensures that the water is evenly distributed and thoroughly incorporated into the flour particles (Davies *et al*, 2008).

Blending may also involve the addition of other ingredients, such as flavorings, preservatives, or fortificants, depending on the desired characteristics of the final fufu flour product. These additional ingredients are carefully measured and added during the blending process to achieve the desired taste, shelf life, or nutritional profile. The duration and intensity of blending may vary depending on the specific recipe or production requirements. The goal is to achieve a

homogeneous blend where all the ingredients are evenly distributed throughout the mixture. Once the blending process is complete, the fufu flour mixture is typically packaged or further processed based on the intended use or market requirements. The blended fufu flour is then ready for cooking or distribution. Blending is an essential step in fufu flour production as it ensures the uniform distribution of ingredients, enhances the flavor and consistency of the final product, and allows for customization based on desired characteristics (Achi and Akomas, 2008).

**(n). Packaging**

Packaging is the process of enclosing a product, such as fufu flour, in suitable materials for protection, preservation, and convenient handling. In fufu flour production, packaging refers to the step where the final product is placed into containers or bags to be stored, transported, and sold. The packaging of fufu flour serves several purposes. It protects the flour from moisture, air, light, and other external factors that could degrade its quality or contribute to spoilage. It also helps to maintain the freshness, flavor, and nutritional value of the flour over an extended period. The packaging materials used for fufu flour are typically selected to be durable, food-safe, and resistant to moisture and pests. Common packaging options include plastic bags, paper bags, or rigid containers. The packaging materials may also be designed to provide information about the product, such as nutritional information, cooking instructions, and branding. During the packaging process, the fufu flour is carefully weighed and filled into the packaging containers. The containers are then sealed or closed to ensure the integrity and safety of the product. Quality control measures may be implemented to monitor the packaging process and ensure that the proper quantity of fufu flour is accurately packaged (Achi and Akomas, 2008).

Proper packaging is crucial in fufu flour production as it helps to extend the shelf life, maintain product quality, and provide consumers with a convenient and safe product. The packaged fufu

flour is ready for storage, distribution, and ultimately for use in preparing fufu dishes (Carlsson *et al.*, 1999).

## **2.4 Microorganisms Involved in the Production of Fufu**

### **2.4.1 Dry Fufu**

Microorganisms involved in the production of dry fufu include; *Lactobacillus acidophilus*, *Lactobacillus plantarum*, *Lactobacillus fermentum*, *Lactobacillus casei*, *Lactobacillus delbrueckii*, *Streptococcus thermophilus*, *Clostridium pefringenes*, *Staphylococcus aureus*

#### **2.4.1.1 *Lactobacillus acidophilus*:**

*Lactobacillus acidophilus* is a beneficial lactic acid bacteria commonly found in fermented foods. It contributes to the fermentation process by converting sugars in the starchy ingredients into lactic acid, which helps lower the pH and creates an acidic environment that inhibits the growth of harmful bacteria (Donald *et al.*, 1989).

#### **2.4.1.2 *Lactobacillus plantarum*:**

*Lactobacillus plantarum* is another lactic acid bacteria commonly involved in food fermentation. It helps convert sugars into lactic acid, contributing to the sour taste and preservation of the fufu. *L. plantarum* also produces antimicrobial compounds that inhibit the growth of undesirable microorganisms (Donald *et al.*, 1989).

#### **2.4.1.3 *Lactobacillus fermentum*:**

*Lactobacillus fermentum* is a lactic acid bacteria that participates in the fermentation of fufu. It contributes to the production of lactic acid, which helps acidify the fermenting mixture and creates an environment unfavorable for the growth of spoilage organisms (Achi and Akomas, 2006).

#### **2.4.1.4 *Lactobacillus casei*:**

*Lactobacillus casei* is a lactic acid bacteria known for its probiotic properties. It aids in the fermentation process by producing lactic acid and various enzymes that contribute to the breakdown of complex carbohydrates in the starchy ingredients (Adegoke and Babalola, 1988).

#### **2.4.1.5 *Lactobacillus delbrueckii*:**

*Lactobacillus delbrueckii* is a lactic acid bacteria commonly involved in the fermentation of various foods. It helps convert sugars into lactic acid, contributing to the acidity and characteristic flavor of the fermented fufu (Lateef *et al.*, 2006).

#### **2.4.1.6 *Streptococcus thermophilus*:**

*Streptococcus thermophilus* is a thermophilic lactic acid bacteria. It aids in the fermentation process by converting lactose into lactic acid, contributing to the acidity and flavor development of the fufu (Adegoke and Babalola, 1988).

#### **2.4.1.7 *Clostridium perfringens*:**

*Clostridium perfringens* is a pathogenic bacterium that can cause foodborne illness. It is not desirable in the fermentation process of fufu production. Strict hygiene practices, such as proper sanitation and control of fermentation conditions, are essential to prevent the growth of *C. perfringens* and other harmful bacteria (Adegoke and Babalola, 1988).

#### **2.4.1.8 *Staphylococcus aureus*:**

*Staphylococcus aureus* is another pathogenic bacterium associated with food poisoning. It is not a desired organism in fufu fermentation. Proper hygiene and sanitation practices are crucial to prevent contamination with *S. aureus* and ensure the safety of the final product (Achi and Akomas, 2006).

### **2.4.2 Wet Fufu**

Microorganisms involved in the production wet fufu include; *Enterococcus faecium*, *Enterococcus faecalis*, *Bacillus subtilis*, *Bacillus licheniformis*, Lactic acid bacteria (LAB), Coliform bacteria

#### **2.4.2.1 *Enterococcus faecium***

*Enterococcus faecium* is a type of lactic acid bacteria. In fufu fermentation, it can contribute to the production of lactic acid, which helps acidify the mixture and create an environment that inhibits the growth of harmful bacteria. *Enterococcus faecium* is generally considered beneficial and can contribute to the flavor development of the fufu (Donald *et al.*, 1989).

#### **2.4.2.2 *Enterococcus faecalis***

*Enterococcus faecalis* is another lactic acid bacteria commonly found in the intestinal tract of humans and animals. While it can contribute to lactic acid production, *Enterococcus faecalis* is also associated with opportunistic infections and can pose a risk if present in high numbers. Strict hygiene practices and control of fermentation conditions are necessary to prevent the overgrowth of *Enterococcus faecalis* (Donald *et al.*, 1989).

#### **2.4.2.3 *Bacillus subtilis***

*Bacillus subtilis* is a spore-forming bacterium commonly found in the environment. It is not a typical organism involved in traditional fufu fermentation. However, if present, it can contribute to the breakdown of complex carbohydrates and the production of enzymes that aid in the fermentation process. Strict control of fermentation conditions and good hygiene practices are necessary to prevent the growth of undesirable bacteria like *Bacillus subtilis* (Donald *et al.*, 1989).

#### **2.4.2.4 *Bacillus licheniformis***

*Bacillus licheniformis* is another spore-forming bacterium that is not commonly associated with fufu fermentation. If present, it can contribute to the breakdown of complex carbohydrates and the production of enzymes that affect the fermentation process. However, the presence of *Bacillus*

*licheniformis* is generally not desired in fufu fermentation due to its potential to cause spoilage or other undesirable effects (Donald *et al.*, 1989).

#### **2.4.2.5 Lactic acid bacteria (LAB)**

Lactic acid bacteria, including various species of *Lactobacillus* and other LAB, play a significant role in fufu fermentation. They convert sugars in the starchy ingredients into lactic acid through fermentation, which helps lower the pH, create an acidic environment, and contribute to flavor development. LAB also produce antimicrobial compounds that inhibit the growth of undesirable microorganisms (Donald *et al.*, 1989).

#### **2.4.2.6 Coliform bacteria**

Coliform bacteria, including *Escherichia coli* and other related species, are a group of bacteria commonly associated with the intestinal tracts of humans and animals. While some coliform bacteria are harmless, the presence of coliforms in fufu fermentation can indicate unsanitary conditions and potential fecal contamination. Strict hygiene practices and control measures are essential to prevent the presence of coliform bacteria in the fermentation process (Donald *et al.*, 1989).

### **2.5 Hygienic Practices carried out to avoid the Contamination of Fufu Production Process**

Maintaining proper hygiene practices is crucial to avoid contamination during fufu production. Here is a list of hygiene practices to follow:

#### **2.5.1 Personal Hygiene**

Hands should be thoroughly washed with soap and clean water before and after handling any ingredients or utensils to maintain proper hygiene. Clean and properly sanitized utensils, bowls, and equipment should be used during the entire fufu production process. Clean and appropriate

protective clothing, such as gloves and aprons should be worn to prevent any potential contamination (Obadina *et al.*, 2010).

### **2.5.2 Ingredient Handling**

The starchy ingredients, such as cassava, yam, or plantains, should be ensured that they are fresh and free from any signs of spoilage or mold. Ingredients should be washed thoroughly before peeling or processing them. The ingredients should be stored in clean, dry, and well-ventilated areas to prevent contamination and spoilage (Obadina *et al.*, 2010).

### **2.5.3 Water Quality**

Clean and potable water should be used for washing ingredients, cooking, and mixing the fufu. If using well water or other non-treated water sources, ensure that it is tested regularly for safety and treated if necessary (Obadina *et al.*, 2010).

### **2.5.4 Cleaning and Sanitization**

All work surfaces, utensils, and equipment should be cleaned and sanitized before and after use. Use of food-grade sanitizers or a mixture of water and bleach to disinfect surfaces effectively is also a good sanitary practice. Special attention should be paid to equipment such as mortars, pestles, or blenders used for pounding or grinding the ingredients (Adegoke and Babalola, 1988).

### **2.5.5 Fermentation Process**

If fermenting the fufu, create a controlled environment that promotes proper fermentation and inhibits the growth of harmful bacteria. This includes using clean fermentation vessels and ensuring appropriate temperature and humidity levels. Covering the fermentation vessels with clean lids or coverings to prevent contamination from insects, dust, or other external sources (Adegoke and Babalola, 1988).

### **2.5.6 Storage and Preservation**

Storage of fufu in clean and airtight containers to protect it from contamination by pests or airborne microbes. Keeping the storage area clean, dry, and well-ventilated to prevent the growth of mold or bacteria (Obadina *et al.*, 2010).

### **2.5.7 Good Manufacturing Practices (GMP)**

Following general principles of GMP, including proper sanitation, training of staff, separation of raw and cooked ingredients, and regular inspection of the production area (Hernandez *et al.*, 1995).

## **2.6 Chitosan**

Chitosan is a polysaccharide of marine origin that is produced from chitin of the exoskeletons of crustaceans. Chitosan is a linear polymer formed by d-glucosamine and N-acetyl-d-glucosamine. This biopolymer and its derivatives are considered promoters of diverse biological activities, including antioxidant, antihypertensive, anti-inflammatory, anticoagulant, antitumoral, antimicrobial, hypocholesterolemic, and antidiabetic effects ((Kumar *et al.*, 2004)

### **2.6.1 Source**

Chitosan is a polysaccharide derived from chitin, which is found in the exoskeletons of crustaceans such as shrimp, crab, and lobster. It is also found in the cell walls of certain fungi and the cuticles of insects. Chitosan is typically obtained by deacetylating chitin through a chemical process, which removes the acetyl groups from the chitin molecule. The resulting chitosan has different properties and applications compared to chitin (Kwaeon *et al.*, 2003).

### **2.6.2 Physicochemical characteristics and applications of chitosan**

The composition and size of the polymer chains of chitosan vary depending on their origin and the chitin deacetylation method. According to Rinaudo, (2006), the solid chitosan is a semi-crystalline

polymer of white or slightly yellow appearance. Chitosan is insoluble in water, in alkaline solutions, and in organic solvents. Chitosan is considered one of the most valuable polymer for biomedical and pharmaceutical applications due to its biodegradability, biocompatibility, antimicrobial, non-toxicity, and anti-tumor properties. Nanoparticles, microspheres, hydrogels, films, and fibers are typical chitosan based forms for biomedical and pharmaceutical applications. Examples of such applications include nasal, ocular, oral, parenteral and transdermal drug delivery (Kumar *et al.*, 2004).

## **CHAPTER THREE**

### **3.0 MATERIALS AND METHODS**

#### **3.1 Collection of Samples**

Dry fufu samples were purchased at Fagcoo supermarket in the University of Benin and at Uselu market. 3 wet fufu samples and 3 dry fufu samples totaling 6 samples. The samples were taken to the laboratory for analysis.

#### **3.2 Preparation of Culture Media**

The media used were prepared according to the manufacturer's instructions. The media used were SSA Agar, MRS agar and Nutrient Agar.

##### **3.2.1 Preparation of *Salmonella* and *Shigella* Agar**

63 grams of Salmonella Shigella Agar (SSA) powder was dissolved in 1 liter of distilled water in a conical flask after which it was covered with cotton wool and aluminum foil paper. It was mixed thoroughly and sterilized for 10 minutes. The medium was cooled to 45-50°C and then dispensed aseptically into sterile petri dishes in the laminar flow.

##### **3.2.2 Preparation of Nutrient Agar**

28 grams of nutrient agar (NA) powder was dissolved in 1 liter of distilled water in a conical flask covered with cotton wool and aluminum foil paper. It was mixed thoroughly and sterilized by autoclaving at 121°C for 15 minutes. The medium was cooled to 45-50°C and then dispensed aseptically into sterile petri dishes in the laminar flow.

### **3.3 Preparation of MRS Agar (De Man, Rogosa and Sharpe Agar)**

Sixty two (62g) grams of MRS agar powder was dissolved in 1 liter of distilled water in a conical flask covered with cotton wool and aluminum foil paper. It was mixed thoroughly and sterilized by autoclaving at 121°C for 15 minutes. The medium was cooled to 45-50°C and then dispensed aseptically into sterile petri dishes in the laminar flow.

### **3.4 Isolation of bacteria**

1g of the sample was weighed and placed in 9ml sterile water. The aliquot was then transferred aseptically to sterile petri plates. The prepared agar (for bacteria growth) was poured in aseptically and incubated at 37°C for 24 hours. After successful growth of microorganisms, the colonies were counted with a colony counter and the results per dilution count were recorded. The number of colony forming unit per milliliter was calculated with the formula:

$$\text{Cfu/g} = \frac{\text{number of colonies}}{\text{volume plated} \times \text{dilution factor}}$$

### **3.5 Sub-culturing of Bacterial Isolates**

One single colony was identified and re-streaked as a primary inoculant on the surface of a nutrient agar plate medium. Pure cultures were checked from nutrient agar plates. After achieving a pure culture, the same colony was streaked onto a nutrient agar slant. These cultures were incubated at 37°C for 24 hours.

### **3.6 Cultural characteristics**

Each colony morphology e.g., size, shape, margin, elevation, consistency, color, transparency was determined.

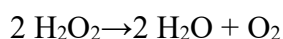
### **3.7 Morphological Test**

#### **3.7.1 Gram staining**

Smears of the bacterial isolates were prepared and heat fixed on clean grease free slides. The smears were stained for one minute with crystal violet. This was washed out with distilled water. The slides were flooded with dilute Grams' iodine solution for one minute. This was washed off with distilled water and the smears were decolorized with 95% alcohol for 30 seconds and rinsed off with distilled water. The smears were then counter stained with safranin solution for one minute. Finally, the slides were washed off with distilled water, air dried and observed under oil immersion objective.

#### **3.7.2 Catalase Test**

This is a test to detect the presence or absence of catalase enzyme. The catalase enzyme catalyses the breakdown of hydrogen peroxide to release free oxygen gas and the formation of water. A few drops of freshly prepared 3% hydrogen peroxide were added onto the bacterial isolates smeared on a slide. The production of gas bubble indicated catalase enzyme positive.

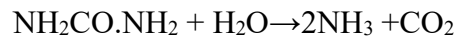


#### **3.7.3 Oxidase Test**

A piece of filter paper was wet with a few drops of the dilute (1%) solution of oxidase reagent (tetramethyl-pphenylenediamine-dihydrochloride) which was prepared by standard procedure. A bit of growth from the nutrient agar slant was obtained using sterilized platinum wire loop and smeared on the wet piece of paper. Development of an intense purple color by the cells within 30 seconds indicates a positive oxidase test.

#### **3.7.4 Urease Test**

The urease test is used to determine the ability of an organism to split urea in the presence of the enzyme urease. The bacterial isolates were inoculated into slants of urea medium and incubated at 37°C for 24-48 hours. Urease positive cultures produced a red-pink colour due to changes in the colour of the indicator



### **3.7.5 Citrate Utilization Test**

This test is based on the ability of some organisms to utilize citrate as a sole source of carbon. This was carried out by inoculating the test organism in test tube containing Simon's citrate medium and this was incubated at 37°C for 24 - 48 hours. The development of deep blue colour after incubation indicates a positive result.

### **3.7.6 Hydrogen Sulphide (H<sub>2</sub>S) Test**

Hydrogen sulphide production can be detected by incorporating a heavy metal salt containing (Fe<sup>2+</sup>) or lead (Pb<sup>2+</sup>) ion as H<sub>2</sub>S indicator to a nutrient culture medium containing cysteine and sodium thiosulfate as the sulphur substrates. Hydrogen sulphide, a colourless gas, when produced reacts with sulphur metal salt (ferrous sulphate) forming a visible insoluble black sulphide precipitate.

### **3.7.8 Indole Test**

Indole test is performed to determine the ability of the organism to split tryptophan molecule into indole. This test is performed to help differentiate species of the family enterobacteriaceae.

Kovac's reagent which contains hydrochloric acid, dimethylaminobenzaldehyde and amyl alcohol is used. Inoculate broth with the test organism and incubate for 18 – 24 hours at 37°C. Add 5ml of Kovac's reagent down the inner wall of the tube. Development of bright red colour at the interface

of the reagent and the broth within seconds after adding the reagent is indicative of the presence of indole and is a positive test while absence is negative.

### **3.7.9 Sugar Fermentation Test**

Each of the isolates was tested for its ability to ferment a given sugar with the production of acid and gas or acid only. The growth medium comprised of peptone water, sugar (1%) and the indicator (bromocresol purple). The mixture was dispensed into test tubes and sterilized by autoclaving at 121°C for 15 minutes. After sterilizing, tubes were allowed to cool and then inoculated with the isolates and incubated at 37°C for 24hrs. Acid and gas production or acid only were observed after about 24 hours of incubation. Acid production was indicated by the change of the medium from purple to yellow colour indicated a positive test. Sugars used are: lactose, sucrose, glucose, fructose, maltose, starch and sorbitol.

### **3.8 Antibiotic susceptibility test**

Test organisms will be subjected to antibiotics sensitivity test using the Kirby Bauer disc diffusion on Mueller Hinton Agar. Ten (10) different commercial antibiotic discs will be used. The antibiotic discs will be carefully and firmly placed on the inoculated plates using a sterile pair of forceps. The plates will be inverted and incubated for 37°C for 24 h. The diameter of the zone of inhibition will be measured in millimeters (mm) using a meter rule. The experiments will be carried out in triplicates to minimize probability of error

### **3.9 Preparation of Mueller Hinton Agar**

Suspend 38g of Mueller Hinton agar powder in 1L of distilled water. Mix and dissolve them completely. Sterilize by autoclaving at 121°C for 15 minutes. Pour the liquid into the petri dish

and wait for the medium to solidify. Be sure to prepare the agar in the clean environment to prevent any contamination.

### **3.10 Preparation of Chitosan**

**Stock solution:** 10g of Chitosan was weighed and dissolved in 100ml of sterile distilled water.

This resulted in a concentration of 100mg/ml..... (A)

50ml of the stock was taken and diluted with 50ml of sterile distilled water to yield a 50mg/ml..... (B)

50ml of the 50mg/ml was taken and further diluted with 50ml of sterile distilled water to yield a 25mg/ml..... (C)

## **CHAPTER FOUR**

### **RESULTS**

Table 4.1 shows the result for total heterotrophic bacteria count from 6 fufu samples in three different agars. The agars are: Nutrient agar, Salmonella and Shigella agar (SSA) and De Ma, Rogosa and sharpe agar (MRS). The wet fufu sample 2 had the highest heterotrophic bacteria count of  $9.16 \times 10^{-6}$  (cfu/g) in the nutrient agar while the lowest was the dry fufu sample 1 with heterotrophic count of  $2.36 \times 10^{-6}$  (cfu/g). For *Salmonella* and *Shigella* agar, the heterotrophic bacteria count ranged from  $1.0 \times 10^{-3}$  (cfu/g) to  $3.0 \times 10^{-3}$  (cfu/g). While for De Man, Rogosa and sharpe agar the heterotrophic bacteria count ranged from  $1.0 \times 10^{-4}$  (cfu/g) to  $4.6 \times 10^{-4}$  (cfu/g).

Table 4.2 shows the result of the cultural, morphological and biochemistry tests of bacterial isolates. Isolates in the study include: *Corynebacterium* spp, *Corynebacterium violaceu*, *Micrococcus* sp, *Pseudomonas* spp, *Bacillus subtil*, *Lactobacillus* sp, *Salmonella* sp.

Table 4.3 shows the records of the distribution pattern of the bacterial isolates form dry and wet fufu The most occurring bacterial isolate is *Corynebacteriun* sp with an occurrence percentage of 33.33%

Table 4.4 and table 4.5 shows antibiotic susceptibility test of the bacterial isolates. The result showed that for Gram positive bacteria, *Bacillus subtilis* had MAR index of 9 as the highest and *Corynebacteria* had the lowest MAR index of 0.1. While for Gram negative isolates, *Salmonella* had the highest MAR index of 5 while *Pseudomonas* spp had the least with MAR index of 0.1.

Table 4.6 shows the various concentration of susceptibility of isolates to chitosan. The zone of inhibition exhibited by the bacterial isolates when tested with 100mg/ml of chitosan ranged from 16mm – 48mm. The zone of inhibition exhibition by bacterial isolates when tested with 50mg/ml of chitosan ranged from 12mm – 14mm. While the zone of inhibition exhibited by bacterial isolates when tested with 25mg/ml of chitosan ranged from 6mm-20mm.

**Table 4.1: Colony Count for Fufu samples (Wet and Dry) in cfu/g**

<b>Sample Name</b>	<b>Heterotrophic bacteria count (cfu/g)(NA)</b>	<b>Heterotrophic bacteria count (cfu/g) (SSA)</b>	<b>Heterotrophic bacteria count (cfu/g) (MRS)</b>
<b>WET FUFU 1</b>	4.13×10 <sup>6</sup>	0.00	4.60×10 <sup>4</sup>
<b>WET FUFU 2</b>	9.16×10 <sup>6</sup>	3.00×10 <sup>3</sup>	1.00×10 <sup>4</sup>
<b>WET FUFU 3</b>	3.86×10 <sup>6</sup>	1.00×10 <sup>3</sup>	0.00
<b>DRIED FUFU FLOUR 1</b>	2.36×10 <sup>6</sup>	0.00	0.00

<b>DRIED FUFU FLOUR 2</b>	$3.86 \times 10^6$	$2.00 \times 10^3$	$4.00 \times 10^4$
<b>DRIED FUFU FLOUR 3</b>	$5.60 \times 10^6$	$1.00 \times 10^3$	$3.00 \times 10^4$

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NA : Nutrient agar

SSA: *Salmonella* and *Shigella* agar

MRS: De Man Rogosa and Sharpe agar

**Table 4.2: Cultural, Morphological and Biochemical Characteristics of Bacterial Isolates**

<b>Cultural Characteristics</b>	<b>1</b>	<b>2</b>	<b>3</b>	<b>4</b>	<b>5</b>	<b>6</b>	<b>7</b>	<b>8</b>	<b>9</b>
Shape	Irregular	Circular	Circular	Rhizoid	Irregular	Circular	Circular	Circular	Circular
Size	Small	Small	Small	Large	Medium	Small	Small	Punctiform	Punctiform
Elevation	Flat	Low convex	Flat	Low convex	Flat	Flat	Flat	Flat	Flat
Transparency	Translucent	Opaque	Opaque	Transparent	Translucent	Opaque	Opaque	Translucent	Translucent
Margin	Undulate	Entire	Entire	Filiform	Undulate	Entire	Entire	Entire	Entire
Colour									
Agar 1 (NA)	Cream	Purple	Cream	Cream	Cream	-	-	-	-
Agar 2 (SSA)	-	-	-	-	-	-	-	Cream	Cream
Agar 3 (MRS)	-	-	-	-	-	Off white	White	-	-
<b>Morphological</b>									
Gram stain	+ve	-ve	+ve	+ve	-ve	+ve	+ve	-ve	-ve
Cell type	Rod	Cocco-bacillus	Cocci	Rod	Rod	Rod	Rod	Rod	Rod
Cell arrangement	Cluster	Pairs	Single	Chains	Single	Chains	Single	Single	Pairs
<b>Biochemical</b>									
Urease	-	-	-	-	-	-	-	+	+
Indole	-	-	-	-	-	-	-	-	-
Citrate	-	+	-	-	+	+	-	+	+
Catalase	+	+	+	+	+	+	-	+	+
H <sub>2</sub> S	+	-	-	+	+	-	-	-	-
Oxidase	-	-	+	-	+	-	-	-	-
<b>Sugar fermentation</b>									
Lactose	-	-	-	-	-	+	+	-	-
Sucrose	-	-	-	-	-	+	+	-	-
Glucose	+	+	-	+	-	+	+	+	+
Fructose	+	-	-	+	-	+	+	-	-
Maltose	+	-	-	+	-	+	+	-	-
Starch	-	-	-	-	-	+	+	-	-
Sorbitol	-	-	-	-	-	+	+	+	+
<b>Isolates</b>	<i>Corynebacterium</i> sp1	<i>Chromobacterium</i> <i>violaceum</i>	<i>Micrococcus</i> sp.	<i>Corynebacterium</i> sp2	<i>Pseudomonas</i> sp.	<i>Bacillus</i> <i>subtilis</i>	<i>Lactobacillus</i> sp.	<i>Salmonella</i> sp1	<i>Salmonella</i> sp2

**Table 4.3: Distribution pattern of bacteria isolates from dry and wet fufu samples**

Probable Organism	Source						Percentage frequency of occurrence
	Wet fufu 1	Wet fufu 2	Wet fufu 3	Dried fufu flour 1	Dried fufu flour 2	Dried fufu flour 3	
<i>Corynebacterium</i> sp.	+	+	-	-	-	-	33.33%
<i>Chromobacterium violaceum</i>	-	-	-	-	-	+	16.66%
<i>Micrococcus</i> sp.	-	-	-	-	+	-	16.66%
<i>Corynebacterium</i> sp.	-	-	+	-	-	-	16.66%
<i>Pseudomonas</i> sp.	-	-	-	+	-	-	16.66%
<i>Bacillus subtilis</i>	+	-	-	-	-	-	16.66%
<i>Lactobacillus</i> sp.	-	-	-	-	+	-	16.66%
<i>Salmonella</i> sp <sup>1</sup>	-	+	-	-	-	-	16.66%
<i>Salmonella</i> sp <sup>2</sup>	-	-	-	+	-	-	16.66%

**Table 4.4: Antibiotics Sensitivity Test of Gram positive bacteria from dry and wet fufu samples**

Isolates	PEF	CN	APX	Z	AM	R	CPX	S	SXT	E	MAR Index
<i>Corynebacterium</i> sp1	14(I)	14(I)	10(R)	14(I)	14(I)	16(I)	16(I)	14(I)	16(I)	12(I)	0.1
<i>Micrococcus</i> sp.	18(S)	20(S)	14(I)	16(I)	14(I)	10(R)	10(R)	10(R)	12(I)	14(I)	0.3
<i>Corynebacterium</i> sp2	10(R)	12(I)	4(R)	4(R)	6(R)	10(R)	20(S)	10(R)	12(I)	8(R)	0.7
<i>Bacillus Subtilis</i>	4(R)	12(I)	0(R)	0(R)	0(R)	10(R)	0(R)	0(R)	0(R)	10(R)	0.9
<i>Lactobacillus</i> sp.	10(R)	2(R)	0(R)	0(R)	0(R)	18(S)	20(S)	22(S)	4(R)	16(I)	0.6

**Keys**

MAR = Multiple Antibiotic Resistance index

PEF = Pefloxacin

CN = Gentamycin

APX = Ampliclox

Z = Zinnacef

AM = Amoxicillin

R = Rocephin

CPX = Ciproflxacin

S = Streptomycin

SXT = Septrin

E = Erythromycin

MAR = Multiple Antibiotic Resistance index

**Table 4.5: Antibiotics Sensitivity Test of Gram negative bacteria from dry and wet fufu samples**

<b>ISOLATES</b>	<b>SXT</b>	<b>CH</b>	<b>SP</b>	<b>CPX</b>	<b>AM</b>	<b>AU</b>	<b>CN</b>	<b>PEF</b>	<b>OFX</b>	<b>S</b>	<b>MAR Index</b>
<i>Chromobacterium violaceum</i>	10(R)	10(R)	12(I)	14(I)	16(I)	20(S)	22(S)	20(S)	18(S)	14(I)	0.2
<i>Pseudomonas sp.</i>	14(I)	14(I)	16(I)	16(I)	16(I)	20(S)	18(S)	18(S)	18(S)	8(R)	0.1
<i>Salmonella sp</i> <sup>1</sup>	12(I)	6(R)	18(S)	20(S)	8(R)	0(R)	16(S)	14(I)	12(I)	12(I)	0.3
<i>Salmonella sp</i> <sup>2</sup>	4(R)	20(S)	16(I)	16(I)	8(R)	0(R)	10(R)	14(I)	18(S)	10(R)	0.5

**NB:** Resistance (R) = 0-10mm, Intermediate (I) = 11-16mm, Sensitive (S) = 17mm and above

**Keys**

SXT = Septrin

CH = Chloramphenicol

SP = Sparifloxacin

CPX = Ciproflxacin

AM = Amoxacillin

AU = Augmentin

CN = Gentamycin

PEF = Pefloxacin

OFX = Tarivid

S =Streptomycin

MAR Index = Multiple Antibiotics Resistance index

**Table 4.6: Antibacterial activity of Chitosan on bacteria isolates from dry and wet fufu samples**

<b>Concentration</b>  <b>Organisms</b>	<b>Zone of Inhibition (mm)</b>		
	<b>(100mg/ml)</b>	<b>(50mg/ml)</b>	<b>(25mg/ml)</b>
<i>Corynebacterium</i> sp.	20	18	10
<i>Chromobacterium violaceum</i>	36	16	6
<i>Micrococcus</i> sp.	16	14	10
<i>Corynebacterium</i> sp.	34	18	6
<i>Pseudomonas</i> sp.	28	18	10
<i>Bacillus subtilis</i>	20	12	6
<i>Lactobacillus</i> sp.	48	44	20
<i>Salmonella</i> sp <sup>1</sup>	44	22	20
<i>Salmonella</i> sp <sup>2</sup>	36	22	6

## CHAPTER FIVE

### 5.0

### DISCUSSION

Comparing the bacterial composition between dry and wet fufu samples is an important area of study as it provides insights into the microbial characteristics of these traditional food products. By analyzing the microbial diversity in each sample type, researchers can gain a better understanding of the range of bacterial species present and their relative abundance. The processing methods used for dry and wet fufu production have a significant impact on the bacterial composition. During the course of the project work, the results shown after the isolation of bacterial samples showed that 2 dry fufu samples and 3 wet fufu samples had the presence of *Corynebacterium* sp and 1 dry and wet fufu sample each had the presence of *Salmonella* spp present. Some articles state that the presence of microorganism such as *Salmonella* spp is as a result of improper hygienic practices (Obadina *et al.*, 2010). If proper hygienic practices are not carried out, it can lead to contamination by pathogenic microorganisms present in the fufu and also lead to diseases such as cholera after consumption of contaminated fufu food. One pathogenic microorganism found in a dry fufu sample during the course of my research is *Salmonella* spp. Also, the fermentation process involved in wet fufu production creates an environment conducive to the growth of specific bacteria, leading to different microbial profiles compared to the drying process used for dry fufu. Understanding these differences can help optimize processing techniques to enhance desired microbial populations or control the growth of undesirable bacteria (Adegoke and Babalola, 1988). From test carried out during the course of my project, the dry fufu. An important aspect of comparing bacteria between dry and wet fufu is assessing the food safety implications. Certain bacterial species may indicate the presence of pathogens or spoilage organisms. By identifying and quantifying these organisms,

researchers can evaluate potential risks associated with each sample type and develop appropriate food safety measures to mitigate them.

The microbial composition of fufu also has a direct impact on its quality and shelf life. Undesirable bacterial species can contribute to spoilage, leading to off-flavors, texture changes, and reduced shelf life. By comparing the bacterial populations in dry and wet fufu, it becomes possible to identify specific bacteria associated with these issues and develop strategies to extend the product's freshness and quality.

Moreover, the microbial composition of fufu can influence its sensory attributes, such as taste and aroma. Different bacterial species can produce various metabolites during fermentation, which may contribute to the unique flavor profiles of fufu. Understanding these relationships can help producers create fufu varieties with specific flavor profiles that align with consumer preferences.

The findings from comparing bacterial populations in dry and wet fufu can also provide insights for process optimization. For example, if certain bacteria contribute to desirable characteristics in wet fufu, such as improved flavor or texture, techniques can be developed to promote the growth of these bacteria in dry fufu or vice versa. This can lead to the development of new production methods or starter cultures to enhance the quality and consistency of fufu products (Ezeronye, 2003).

In summary, comparing bacteria between dry and wet fufu samples is a valuable area of research that can provide insights into microbial diversity, food safety implications, quality attributes, and process optimization. By understanding the microbial characteristics of these food products.

## **5.1 Conclusion**

In conclusion, proper hygienic practices should be carried out to avoid the contamination of fufu with pathogenic organisms such as *Salmonella* spp. and *Bacillus subtilis*.

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