PROJECT REPORT

ON

EFFECT OF BITTER LEAF ON MICROBIAL LOAD AND SENSORY PROPERTIES OF OGI.

BY:

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CERTIFICATION

This is Project work has been read and approved as meeting part of the requirements of Science Laboratory Technology, Institution of Applied science, Kwara State Polytechnic, Ilorin. In partial fulfillments of therequirement for the award of National Diploma (ND) in Science Laboratory Technology.

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DEDICATION

This report is dedicated to Almighty God and magnify God for his infinite mercy that my path to date. He is the director and builder of my life.

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My praise is due to Almighty GOD we praise him and thank him for giving us the

strength and knowledge to complete this project work and also for our continued existence on the earth.

I appreciate the effort of my supervisor MRS. T.O. ADEBOYE whose patience, support and encouragement have been the driving force behind the success of this research work. She gave useful corrections, recommendations, advice and always ensures that excellent research is done thank you very much for your support ma, i really appreciate your efforts.I sincere gratitude goes to the (HOD) DR USMAN ABDULKAREEM and other members of staff of the department of Science Laboratory Technology, kwara state polytechnic, Ilorin, for their constant cooperation, and encouragement throughout the program.

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ABSTRACT

The study investigates the effect of bitter leaf (Vernoniaamygdalina) on microbial load and sensory properties of ogi, a traditional Nigerian fermented cereal gruel. Bitter leaf is incorporated into ogi at varying concentrations to assess its antimicrobial and sensory attributes. Results show that bitter leaf significantly reduces microbial load in ogi, particularly against pathogenic bacteria and fungi. Sensory evaluation reveals that ogi samples with bitter leaf exhibit acceptable taste, aroma, and overall acceptability, although higher concentrations may impact flavor profile. The findings suggest bitter leaf's potential as a natural preservative in ogi production, enhancing food safety while maintaining sensory quality. This research contributes to developing innovative, herbal-based preservation methods for traditional African foods.

CHAPTER ONE

1.0 Introduction

Ogi is a traditional Nigerian fermented cereal-based food widely consumed across the country. It plays a vital role in both nutrition and culture, especially among infants and adults. Traditionally made from cereals such as maize, sorghum, or millet, ogi undergoes a natural fermentation process that enhances its taste, texture, and nutritional profile. It serves as a weaning food for infants, a staple breakfast meal, and sometimes as a dietary option for individuals recovering from illness due to its easy digestibility(Adebayo et al., 2020).

In recent years, attention has turned towards improving the safety, shelf life, and nutritional value of ogi through the incorporation of natural preservatives. One such potential additive is bitter leaf (Vernoniaamygdalina), a plant known for its antimicrobial and antioxidant properties. The inclusion of bitter leaf in ogi production has been investigated as a promising approach to reducing microbial load, enhancing food safety, and enriching its nutritional content. This chapter provides a background to the study, highlights the relevance of sorghum and bitter leaf in ogi production, and sets the stage for the research on the effects of bitter leaf incorporation on the microbial load and sensory properties of ogi.

1.1 Background

Ogi, commonly consumed in Nigeria and other parts of West Africa, is a fermented food product traditionally prepared from cereals such as maize, sorghum, or millet. The fermentation process, driven by a variety of microorganisms, enhances the food's palatability, safety, and nutritional properties. Among these cereals, sorghum stands out for its high fiber, antioxidant content, and adaptation to Nigeria's climatic conditions.

 Fig 1: Sorghum plant

Source: (Okoye et al., 2020).

On the other hand, bitter leaf (Vernoniaamygdalina) is a medicinal plant frequently used in African traditional medicine and local cuisine. Known for its characteristic bitter taste,

it contains numerous bioactive compounds including flavonoids, saponins, and terpenoids. These compounds have demonstrated antimicrobial, antioxidant, and anti-inflammatory effects, making bitter leaf a viable natural additive for enhancing the health benefits and shelf life of food products.

Fig 2: Bitter leaf

Source: (Ezugwu et al., 2021)

Several studies (Adebayo et al., 2020; Ezugwu et al., 2021) have shown that bitter leaf extracts possess significant antimicrobial properties, which can be harnessed to reduce

the microbial load inogi. This could help improve food safety, extend shelf life, and potentially enhance the organoleptic (sensory) properties of the product. Research has also indicated that the addition of bitter leaf may influence taste, aroma, and appearance-factors critical to consumer acceptance.

Moreover, bitter leaf is locally abundant and affordable, which supports its integration into traditional food processing practices. Investigating the effects of bitter leaf on ogi not only contributes to food safety but also opens up opportunities for developing value-added products that combine traditional knowledge with scientific innovation.

Sorghum

Sorghum (Sorghum bicolor) is a drought-tolerant cereal grain widely grown in Nigeria and across sub-Saharan Africa. It serves as one of the main ingredients in ogi production, especially in regions where maize is less dominant. Sorghum is rich in carbohydrates, dietary fiber, proteins, and polyphenols, which contribute to its antioxidant activity (Ezeonu et al., 2022).

Its fermentation during ogi production improves nutrient bioavailability and digestibility. Sorghum ogitends to have a distinctive flavor and darker color compared to maize ogi. The grain's composition makes it suitable for improving the health benefits of ogi, particularly in regions facing food insecurity and malnutrition.

Bitter Leaf (Vernoniaamygdalina)

Bitter leaf is a leafy vegetable and medicinal herb widely used in Nigeria for culinary and therapeutic purposes (Ezugwu et al., 2021). It is characterized by its intensely bitter

taste, which is attributed to its phytochemical content, including alkaloids, flavonoids, terpenoids, and saponins. These bioactive compounds offer a range of health benefits, such as antimicrobial, anti-inflammatory, and antioxidant activities. In the context of food preservation, bitter leaf has gained attention for its ability to inhibit microbial growth. Its incorporation into fermented foods like ogi has been shown to reduce spoilage and enhance safety without the need for artificial preservatives. Additionally, bitter leaf contributes to the nutritional value of food products by supplying micronutrients such as iron, calcium, and vitamin C.

1.1.1 Preparation of ogi

Ogi is traditionally prepared through a fermentation process involving various grains like maize, sorghum, or millet. According to Adebayo et al. (2020), the preparation of ogi involves soaking grains in water to facilitate fermentation, followed by milling into a paste or flour. The paste or flour is then fermented for several days, which enhances the nutritional value and flavor of ogi (Oluwafemi & Oluwafemi, 2022). Onwuka et al. (2021) noted that the fermentation process involves various microorganisms, including lactic acid bacteria, which contribute to ogi's characteristic flavor and texture.

1.1.2 Health Benefits of Ogi

The fermentation process involved in ogi production enhances its nutritional value and provides several health benefits. Ogi is a good source of probiotics, which are beneficial microorganisms that promote gut health and boost the immune system. The probiotics in ogi help to maintain a healthy gut microbiome, which is essential for proper digestion, absorption of nutrients, and immune function. Additionally, ogi is rich in antioxidants,

which help to protect the body against oxidative stress and related diseases. Consuming ogi has been associated with several health benefits, including improved digestion, enhanced immune function, and reduced risk of chronic diseases such as heart disease, diabetes, and certain cancers. The fiber content in ogi also helps to promote satiety, reduce cholesterol levels, and regulate blood sugar levels.

Furthermore, ogi is a good source of essential minerals such as iron, zinc, and potassium, which are important for maintaining healthy red blood cells, immune function, and blood pressure (Okoro et al., 2022). Iron deficiency is a common nutritional disorder in Nigeria, and consuming ogi can help to address this issue. Zinc plays a crucial role in immune function, wound healing, and protein synthesis, while potassium helps to regulate blood pressure and maintain healthy heart function.

Ogi also contains other essential nutrients such as B vitamins, vitamin E, and other minerals like magnesium and phosphorus. These nutrients are important for maintaining healthy skin, hair, and nails, as well as supporting energy metabolism and bone health.

The fermentation process involved in ogi production also enhances the bioavailability of nutrients, making them easier for the body to absorb. This is particularly important for individuals with compromised digestive systems or malabsorption issues. Incorporating ogi into one's diet can have numerous health benefits, particularly in developing countries where access to nutrient-dense foods may be limited (Onwuka et al., 2021). Ogi is a nutritious, affordable, and accessible food option that can help to address nutritional deficiencies and promote overall health and well-being (Ezugwu et al., 2021).

Moreover, ogi can be used as a base for other food products, such as snacks, beverages, and even pharmaceutical products, due to its versatility and nutritional value (Nwachukwu et al., 2023). Overall, ogi is a nutritious food that offers numerous health benefits, making it an excellent addition to a balanced diet, as supported by various studies (Oluwafemi & Oluwafemi, 2022).

1.1.3 Side Effects and Allergies

While ogi is a nutritious food that provides several health benefits, some individuals may experience side effects or allergic reactions to its consumption. One potential side effect of ogi consumption is gastrointestinal discomfort, including bloating, gas, and stomach cramps. This is often due to the fermentation process involved in ogi production, which can be difficult for some individuals to digest (Oluwafemi and Oluwafemi, 2022). Some people may also be allergic to the ingredients used in ogi production, such as corn or millet.

Symptoms of an ogi allergy can range from mild to severe and may include hives, itching, swelling, stomach cramps, diarrhea, and difficulty breathing. In severe cases, ogi allergy can cause anaphylaxis, a life-threatening allergic reaction that requires immediate medical attention (Nwachukwu et al., 2023). If you experience any symptoms of an ogi allergy, it's essential to consult a healthcare professional for proper diagnosis and treatment.

Additionally, ogi may also be contaminated with mycotoxins, which are toxic compounds produced by fungi that can grow on grains during storage. Mycotoxins can cause a range of health problems, including liver damage, kidney damage, and immune

system suppression (Okoye et al., 2020). Individuals with weakened immune systems, such as those with HIV/AIDS or undergoing chemotherapy, may be more susceptible to the adverse effects of mycotoxins.Furthermore, individuals with celiac disease or gluten intolerance may need to be cautious when consuming ogi made from gluten-containing grains such as wheat or barley. Ogi made from gluten-free grains such as corn or millet may be a safer option for these individuals (Adeyeye et al., 2021). It's crucial for individuals with gluten intolerance or celiac disease to choose ogi products that are certified gluten-free to minimize the risk of adverse reactions.

Ogi can also be high in phytates, which can inhibit the absorption of minerals such as iron and zinc (Onwuka et al., 2022). Individuals with mineral deficiencies may need to take steps to minimize phytate intake or enhance mineral absorption. This can be achieved by soaking, sprouting, or fermenting ogi to reduce phytate levels or consuming foods high in vitamin C, which can enhance iron absorption.

To minimize the risk of adverse effects associated with ogi consumption, it's essential to:

1. Choose ogi products from reputable manufacturers that adhere to good manufacturing practices.
2. Store ogi properly to prevent contamination and spoilage.
3. Cook ogi thoroughly to reduce the risk of foodborne illnesses.
4. Consult a healthcare professional if you experience any adverse symptoms after consuming ogi.

1.1.4 Micro Organisms Causing Spoilage of Ogi

Ogi, a traditional Nigerian fermented cereal-based food, is susceptible to spoilage by various microorganisms. The spoilage of ogi can result in changes in its texture, taste, and aroma, making it unacceptable for consumption. According to Omemu et al. (2007), the microorganisms responsible for ogi spoilage are primarily starch-degrading bacteria. Two main bacteria that cause ogi spoilage are Bacillus megaterium and Bacillus subtilis.

Bacillus megaterium has higher amylase activity, which breaks down the starch in ogi, leading to spoilage. This bacterium thrives best at a temperature of 40°C and pH 4 (Adebayo et al., 2018). Bacillus subtilis also degrades starch in ogi and has optimal growth at 40°C and pH 2. These bacteria can cause significant changes in ogi's texture and flavor, making it unpalatable.Other microorganisms that can cause ogi spoilage include fungi such as Aspergillus and Penicillium. According to Okoye et al. (2020), these fungi can produce mycotoxins, which can be toxic to humans and animals. Mycotoxins can cause a range of health problems, including liver damage, kidney damage, and immune system suppression. The growth of these fungi can be influenced by factors such as temperature, humidity, and storage conditions.

The control of ogi spoilage requires proper handling and storage practices. The use of clean equipment, proper sanitation, and storage in airtight containers can help to prevent contamination and spoilage of ogi (Nwachukwu et al., 2023). Additionally, the use of natural preservatives such as spices and herbs can also help to extend the shelf life of ogi. Some studies have shown that certain spices and herbs have antimicrobial

properties that can inhibit the growth of microorganisms responsible for ogi spoilage (Onwuka et al., 2021).Proper storage conditions, such as low temperature and low humidity, can also help to prevent the growth of fungi and bacteria that cause ogi spoilage. Furthermore, the use of modified atmosphere packaging or vacuum packaging can also help to extend the shelf life of ogi by reducing the oxygen levels and preventing the growth of microorganisms (Ezugwu et al., 2021).

Overall, the control of ogi spoilage requires a combination of proper handling, storage, and preservation practices. By understanding the microorganisms responsible for ogi spoilage and the factors that influence their growth, manufacturers and consumers can take steps to prevent spoilage and extend the shelf life of ogi.

1.1.5 Factors Affecting the Spoilage of Ogi

Several factors can contribute to the spoilage of ogi, including temperature, moisture, and handling practices. According to Omemu et al. (2007), temperature and moisture are critical factors that affect the growth of microorganisms in ogi. When ogi is exposed to high temperatures and moisture, it creates an ideal environment for microorganisms to grow, leading to spoilage.

The type of cereal used to produce ogi can also impact its spoilage rate. For example, ogi made from maize may be more susceptible to spoilage than ogi made from sorghum or millet (Adebayo et al., 2018). The type of cereal used can affect the microbial load and shelf life of ogi due to differences in nutrient content, moisture absorption, and enzymatic activity.

Handling and storage practices are also critical factors that can contribute to ogi spoilage. Poor handling and storage practices can lead to contamination and spoilage of ogi (Nwachukwu et al., 2022). Proper handling and storage practices, such as storing ogi in airtight containers and keeping it in a cool, dry place, can help extend its shelf life.

Other factors that can contribute to ogi spoilage include pH, water activity, and enzymatic activity. The pH and water activity of ogi can affect the growth of microorganisms and enzymatic activity, leading to spoilage (Okoye et al., 2020). For instance, ogi with high water activity can support the growth of microorganisms, while ogi with low pH can inhibit the growth of certain microorganisms.Additionally, enzymatic activity can also contribute to ogi spoilage. Enzymes such as amylases and proteases can break down the starches and proteins in ogi, leading to changes in texture and flavor (Onwuka et al., 2021). The activity of these enzymes can be influenced by factors such as temperature, pH, and moisture.

To prevent ogi spoilage, it's essential to control these factors. This can be achieved by:

1. Storing ogi in airtight containers to prevent moisture and contamination.
2. Keeping ogi in a cool, dry place to reduce microbial growth.
3. Using proper handling practices to prevent contamination.
4. Monitoring the pH and water activity of ogi to prevent microbial growth.
5. Using natural preservatives or additives to extend shelf life.

1.1.6 Preservation of Ogi

Ogi, a traditional Nigerian fermented cereal-based food, requires proper preservation techniques to extend its shelf life and maintain its quality. Various methods can be employed to preserve it which include, heat treatment, refrigeration, and the use of natural preservatives. Heat treatment can effectively reduce the microbial load and extend its shelf life (Adebayo et al., 2020).

Heat Treatment and Refrigeration

Heat treatment and refrigeration are common methods used to preserve ogi:

- Heat Treatment: Heat treatment involves heating ogi to a high temperature to kill microorganisms and extend its shelf life. Heat treatment can effectively reduce the microbial load and improve its safety (Omemuet al., 2007).
- Refrigeration: Refrigeration involves storing at low temperatures to slow down the growth of microorganisms.Refrigeration can effectively extend the shelf life and maintain its quality (Nwachukwuet al., 2022).

Natural Preservatives

Natural preservatives, such as spices and herbs, can also be used to preserve ogi:

- Spices and Herbs: Certain spices and herbs, such as garlic and ginger, have antimicrobial properties that can help to preserve it. The use of natural preservatives can effectively extend the shelf life and improve its safety (Okoyeet al., 2020).

Other Preservation Methods

Other preservation methods, such as packaging and storage, can also impact the shelf life of ogi:

- Packaging: Proper packaging can help prevent contamination and spoilage of ogi. Packaging ogi in airtight containers can help extend its shelf life (Adetuyiet al., 2019).
- Storage: Proper storage conditions, such as low temperatures and low humidity, can also help extend the shelf life of ogi.

1.1.7 Natural Preservation of Ogi and Their Advantages

Ogi, a traditional Nigerian fermented cereal-based food, can be preserved naturally using various methods. Natural preservation methods can improve the safety and quality of ogi (Adisa and Enujiugha, 2020). One such method is fermentation, which involves encouraging the growth of beneficial microorganisms.

Advantages of Natural Preservation Methods

Natural preservation methods offer several advantages, including improved safety, enhanced nutrition, and extended shelf life. Natural preservation methods can reduce the risk of contamination and spoilage, (Omemuet al., 2007). Additionally, fermentation can increase the nutritional value of ogi and create new compounds with health benefits.

Types of Natural Preservatives in Ogi

Some natural preservatives found in ogi include lactic acid bacteria (LAB) and antimicrobial compounds. According to Okoye et al. (2020), LAB produce lactic acid,

which creates an acidic environment that inhibits the growth of pathogens and spoilage organisms. LAB also produce antimicrobial compounds, such as bacteriocins, that help eliminate pathogens and spoilage organisms.

The antimicrobial properties of LAB in ogi can be attributed to several factors, including:

1. Acidification: LAB produce lactic acid, which lowers the pH of ogi and creates an environment that is unfavorable for the growth of pathogens and spoilage organisms.
2. Production of antimicrobial compounds: LAB produce compounds such as bacteriocins, which have been shown to inhibit the growth of a wide range of microorganisms.
3. Competition for nutrients: LAB compete with pathogens and spoilage organisms for nutrients, making it difficult for them to survive and grow.

The use of LAB as natural preservatives in ogi offers several benefits, including:

1. Improved safety: LAB can help prevent the growth of pathogens and reduce the risk of foodborne illnesses.
2. Extended shelf life: LAB can help extend the shelf life of ogi by inhibiting the growth of spoilage organisms.
3. Preservation of nutritional quality: LAB can help preserve the nutritional quality of ogi by preventing the degradation of nutrients.

1.1.8 Benefits of Natural Preservation Methods

The benefits of natural preservation methods include consistent quality, improved safety, and increased efficiency. Using starter cultures in ogi production can ensure consistent quality and flavor, reduce the risk of contamination and spoilage, and accelerate the fermentation process (Nwachukwu et al., 2022). Starter cultures can also enhance the nutritional value of ogi, improve its texture and flavor, and reduce the risk of mycotoxins (Okoye et al., 2020). Additionally, starter cultures can help extend the shelf life of ogi by controlling the growth of spoilage organisms. This can lead to increased efficiency and innovation in ogi production, as well as opportunities for developing functional foods and improving food safety (Onwuka et al., 2021). Overall, natural preservation methods using starter cultures can play a crucial role in maintaining the quality and safety of ogi.

1.2 Preparation of Bitter Leaf

Bitter leaf (Vernoniaamygdalina) is typically prepared by washing, chopping, or crushing the leaves to release their bioactive compounds. The leaves can be used fresh or dried for later use. Bitter leaf extracts can be prepared using various solvents, such as water or ethanol, to extract its antimicrobial and antioxidant properties (Ezugwu et al., 2021).

1.2.1 Health Benefit of Bitter Leaf

Bitter leaf (Vernoniaamygdalina) offers numerous health benefits due to its richness in essential nutrients, antioxidants, and phytonutrients. Studies have shown that bitter leaf possesses antioxidant properties, which can help combat oxidative stress and reduce the risk of chronic diseases (Okoro et al., 2022). Its anti-inflammatory effects may also alleviate conditions like arthritis and heart disease (Adebayo et al., 2020).

Bitter leaf supports immune function due to its vitamin C content, helping the body fight off infections and illnesses (Onwuka et al., 2021). The dietary fiber in bitter leaf promotes digestive health by preventing constipation and supporting regular bowel movements (Nwachukwu et al., 2023).

1.2.2 Side Effect and Allergies of Bitter Leaf

Bitter leaf may cause allergic reactions in some individuals, particularly those with sensitivities to plants in the Asteraceae family (Adebayo et al., 2020). Symptoms of allergic reactions can include skin rashes, itching, and respiratory issues. Additionally, the bitter compounds in bitter leaf may cause stomach upset, nausea, or diarrhea in some individuals, especially when consumed in large quantities (Onwuka et al., 2021).

Excessive consumption of bitter leaf may also lead to gastrointestinal discomfort, and individuals with pre-existing medical conditions or taking medications should consult the healthcare professional before using bitter leaf (Okoro et al., 2022).

1.2.3 Micro Organisms Causing Spoilage to Bitter Leaf

Bitter leaf can be susceptible to spoilage caused by various microorganisms, including bacteria, fungi, and yeast. According to studies, some common microorganisms that can cause spoilage of bitter leaf include Aspergillus spp. (Adebayo et al., 2020), Fusarium spp. (Onwuka et al., 2021), Bacillus spp. (Okoro et al., 2022), and Rhizopus spp. (Nwachukwuet al., 2023). These microorganisms can cause decay, discoloration, and off-flavors in bitter leaf, leading to reduced quality and shelf life.

Proper handling, storage, and preservation techniques can help minimize the growth of

these microorganisms and extend the shelf life of bitter leaf (Ezugwu et al., 2021).

1.2.4 Factors Affecting the Spoilage of Bitter Leaf

The spoilage of bitter leaf can be affected by various factors, including temperature, humidity, and light exposure (Adebayo et al., 2020).Temperature affects bitter leaf spoilage as high temperatures facilitate the growth of microorganisms, leading to decay and spoilage (Adebayo et al., 2020). Humidity plays a role as excessive moisture creates an ideal environment for microbial growth, causing bitter leaf to spoil faster (Onwuka et al., 2021). Light exposure contributes to spoilage as it can cause degradation of nutrients and promote microbial growth, reducing the quality and shelf life of bitter leaf (Okoro et al., 2022).

Poor handling practices, such as bruising or crushing, damage bitter leaf and create entry points for microorganisms, increasing the risk of spoilage (Ezugwu et al., 2021). Improper storage conditions, including high temperatures, high humidity, or exposure to light, contribute to spoilage by creating an environment conducive to microbial growth (Nwachukwu et al., 2023). Contamination with microorganisms, soil, or other substances leads to spoilage as it introduces pathogens that can cause decay and spoilage (Adebayo et al., 2020).

1.2.5 Preservation of Bitter Leaf

Bitter leaf can be preserved through various methods, including drying (Adebayo et al., 2020), freezing (Onwuka et al., 2021), and refrigeration (Nwachukwu et al., 2023). Drying bitter leaf can help reduce moisture content, inhibiting microbial growth and

spoilage (Adebayo et al., 2020). Freezing bitter leaf can also help preserve its nutritional value and flavor by slowing down microbial growth (Onwuka et al., 2021).

Refrigeration can help maintain the quality and freshness of bitter leaf by keeping it at a low temperature, thereby slowing down microbial growth (Nwachukwu et al., 2023). Additionally, proper packaging and storage can also help preserve bitter leaf by protecting it from contamination and environmental factors (Ezugwu et al., 2021).

1.3 Aim and Objectives of the Study

Aim:

The aim of this study is to investigate the effect of bitter leaf (Vernoniaamygdalina) on the microbial load and sensory properties of ogi, a traditional Nigerian fermented cereal-based food.

Objectives:

The specific objectives of this study are:

1. To evaluate the effect of bitter leaf addition on the microbial load of ogi during fermentation and storage.
2. To assess the impact of bitter leaf on the sensory properties of ogi, including taste, aroma, texture, and overall acceptability.
3. To investigate the optimal level of bitter leaf addition that achieves a balance between microbial safety and sensory acceptability of ogi.

1.4 Statement of the problem

The traditional method of ogi production involves a wild fermentation process that can lead to contamination and spoilage, resulting in variable product quality and food safety concerns (Adebayo et al., 2020). The lack of control over microorganisms during fermentation can compromise the safety and quality of ogi, making it prone to spoilage and reducing its shelf life (Onwuka et al., 2021).

The presence of pathogenic microorganisms in ogi can pose health risks to consumers, particularly infants and the elderly (Nwachukwu et al., 2023). Therefore, there is a need to explore natural and effective methods to improve the safety and quality of ogi.

1.5 Justification

The study on the effect of bitter leaf on microbial load and improving sensory properties of ogi is justified because bitter leaf's antimicrobial properties can improve food safety and quality (Adebayo et al., 2020). Additionally, bitter leaf's rich nutrient profile can enhance ogi's nutritional value (Onwuka et al., 2021).

Furthermore, extending ogi's shelf life through the use of bitter leaf can reduce food waste and improve food security (Nwachukwu et al., 2023). Improving ogi's quality can also increase consumer confidence and demand, benefiting producers and marketers (Ezugwu et al., 2021).

1.6 Literature Review

Ogi is a traditional Nigerian fermented cereal-based food made from various grains, including maize, sorghum, and millet. The type of grain used can impact the nutritional composition, bioactive compounds, and sensory properties of ogi.

1.6.1 Maize

Maize is a widely cultivated crop in Nigeria and is commonly used for ogi production. It is a good source of carbohydrates, fiber, and some essential minerals like phosphorus and potassium (Adebayo et al., 2020). Maize contains bioactive compounds like ferulic acid and other phenolic acids, which have antioxidant properties (Okoro et al., 2022). The high carbohydrate content in maize makes it an excellent source of energy. Additionally, maize contains vitamins like thiamin, folate, and vitamin C, which are essential for various bodily functions (Adebayo et al., 2020).

1.6.2 Sorghum

Sorghum is another popular grain used for ogi production. It is rich in antioxidants, fiber, and minerals like iron and zinc (Onwuka et al., 2021). Sorghum contains various bioactive compounds, including tannins and phenolic acids, which have been linked to several health benefits (Adeyemi et al., 2022). Sorghum is also a good source of protein and has a relatively low glycemic index, making it suitable for people with diabetes (Onwuka et al., 2021). The bran of sorghum is particularly rich in antioxidants and other bioactive compounds.

1.6.3 Millet

Millet is a small-grained cereal that is widely cultivated in Nigeria. It is a good source of protein, fiber, and minerals like calcium and iron (Nwachukwu et al., 2023). Millet contains bioactive compounds like phenolic acids and flavonoids, which have antioxidant and anti-inflammatory properties (Oluwafemi & Oluwafemi, 2022). Millet is also rich in B vitamins and other essential nutrients. Its small grain size and high nutrient content make it an excellent choice for ogi production.

1.6.4 Cultivation and Production

The cultivation and production of these grains vary across Nigeria, depending on factors like climate, soil type, and agricultural practices (Adebayo et al., 2020). Maize is widely cultivated in the southern regions, while sorghum and millet are more commonly grown in the northern regions due to their drought tolerance (Onwuka et al., 2021). The production of these grains is also influenced by factors like pest management, irrigation, and fertilizer application. Proper agricultural practices, such as crop rotation, timely planting, and integrated pest management, can significantly impact the yield and quality of these grains.

Climate change and variability can also affect grain production, making it essential for farmers to adopt climate-resilient agricultural practices (Nwachukwu et al., 2023). Additionally, access to improved seeds, fertilizers, and irrigation systems can enhance grain production and quality.

In Nigeria, smallholder farmers play a significant role in grain production, and their adoption of sustainable agricultural practices can impact food security and livelihoods (Oluwafemi & Oluwafemi, 2022). Supporting smallholder farmers through training, extension services, and access to markets can help improve grain production and quality.

Overall, the cultivation and production of grains for ogi production require careful consideration of various factors to ensure high-quality grains that meet the nutritional and sensory needs of consumers.

Fig 3: Varieties of corn

Source: (Nwachukwu et al., 2023).

1.6.5 Nutritional Composition

The nutritional composition of ogi can vary depending on the type of grain used. Generally, ogi is a good source of carbohydrates, fiber, and some essential minerals like iron, zinc, and potassium (Nwachukwu et al., 2023). The fermentation process involved in ogi production can also impact its nutritional composition, increasing the bioavailability of some nutrients like vitamins and minerals (Oluwafemi & Oluwafemi, 2022). Ogi made from different grains may have varying levels of protein, fiber, and other nutrients. For example, ogi made from sorghum may have higher levels of antioxidants and fiber compared to ogi made from maize (Onwuka et al., 2021).

Understanding the nutritional composition of ogi is crucial for developing products that meet the nutritional needs of consumers. Ogi can be a valuable source of nutrients for people of all ages, particularly in regions where it is a staple food. The nutrient content of ogi can also be influenced by factors like grain type, processing methods, and storage conditions (Adebayo et al., 2020).

Additionally, ogi's nutritional composition can be enhanced through fortification or supplementation with other nutrient-rich ingredients. This can help address micronutrient deficiencies and improve overall health outcomes (Nwachukwu et al., 2023). Further research is needed to fully understand the nutritional benefits and limitations of ogi and to develop products that are tailored to specific nutritional needs.

Summary of the literature review

Ogi is a traditional Nigerian fermented cereal-based food made from grains like maize, sorghum, and millet, each impacting its nutritional composition and sensory properties. Maize is rich in carbohydrates, fiber, and essential minerals like phosphorus and potassium. Sorghum is high in antioxidants, fiber, and minerals like iron and zinc, with a low glycemic index. Millet is a good source of protein, fiber, and minerals like calcium and iron.

The nutritional composition of ogi varies depending on the grain used, but it's generally a good source of carbohydrates, fiber, and essential minerals. Fermentation increases nutrient bioavailability. Factors like climate, soil type, and agricultural practices influence grain production and quality.

Ogi's nutritional value can be enhanced through fortification or supplementation with nutrient-rich ingredients, addressing micronutrient deficiencies and improving health outcomes.

CHAPTER TWO

MATERIALS AND METHODOLOGY

2.0 Introduction

This chapter outlines the materials, equipment, and experimental procedures used to

Investigate the effect of bitter leaf (Vernoniaamygdalina) on the nutritional composition and shelf life of wet-milled sorghum it includes details on the research desıgn, samples preparation, data collection and analytical techniques employed.

2.1 Study Area

This study was conducted using raw white sorghum purchased from local market, at Oke Oyi Kwara State, Nigeria, and fresh bitter leaf obtained from Odo Ita in Ilorin.

Microbiological and chemical Analysıs were conducted at Microbiology and Chemistry Laboratory of Kwara State Polytechnic and Central Research Laboratory of University of

Ilorin Nigeria.

2.2 Materials Used

Bitter leaf (Vernoniaamygdalina), Ogi (traditional Nigerian fermented cereal-based food), Cereal grains (e.g., maize, sorghum, or millet), Water, Incubator, Autoclave, Microscope, and Sensory evaluation panel.

2.2.1 Sample Collection

Sorghum grains (Sorghum bicolor) was purchased from a local market, Oke Oyi KwaraState. Fresh bitter leaves (Vernoniaamygdalina) was sourced from Odo Ota in Ilorin

2.2.2 Chemicals and Reagents

- Ethanol for sterilization
- Analytical grade media used are: Nutrient Agar. MacConkey Agar, Yeast Extract, Sabouraud Dextrose Agar (SDA), de Man, Rogosa and Sharpe Agar (MRS), (For microbial cultivation and fungal growth analysis).

2.2.3 Equipment

Petri-dishes, inoculating loops. Refrigerator, Incubators, Hot air oven, Test tube, Beakers Conical flask, Grinder Gas cooker, Pot, Cups,¡Spoons and different container for

Samplıng.

2.2.4 Sample Collection

The Sorghum sample was purchased from the market placedin a clean, sterile polythene

Bag to prevent contaminationand transported to the Microbiology Laboratory for Analysis.

2.3 Sample Preparation

The sorghum sample was manually sorted to remove dirt and unwanted particles, while

The bitter leaf was thoroughly washed with clean water to eliminate surface contaminants.The sorghum was then divided into three different containers with the followingCompositions.

- SBC1: 308.5g of sorghum + 0.7g of bitter leaf
- SBC2: 308.0g of sorghum + 1.0g of bitter leaf
- SBC3: 307.5g of sorghum + 1.25g of bitter leaf

Each sample was soaked in 400ml of sterıle deionized water and allowed to ferment tor

48 hours under ambient conditions.

2.3.1 Control Setup

Two additional control samples were prepared:

- Control 1: 310g of sorghum soaked in potable/clean water

- Control 2: 310g of sorghum soaked in distilled water

2.3.2 Milling of the Sample

After 48 hours of fermentation, the steeping water was decanted from each sample.

Additional bıtter leaf was added in the same proportion as the inıtial setup;

- Sample 1: +0.7g of bitter leaf
- Sample 2: +1.0g of bitter leaf
- Sample 3: +1.25g of bitter leaf

The sample were milled using 400ml of water, the control samples also had their steep water decanted before milling.

2.4 Sterilization of Glass wares

All glassware were washed with soap and rinsed with distilled water. All the glass ware were drained out and packed with aluminum foil to prevent the entry of air and to avoid contamination and they were packed into the hot air oven. Then the oven was maintained at 45°C for 15 minutes of sterilization. Others were cleaned and properly kept in a carton. The autoclave and centrifuge were properly covered. Inoculating loop was heat to redness with spirit lamp and workbench was wiped with 70% ethanol.

2.5 Preparation of Media

Five different media were prepared for microbial analysis: MRS agar (33.6g/L), MacConkey agar (23.5g/L), SDA (32.5gL) Yeast Extract (11.5g/L), and Nutrient agar (14g/L). Each agar type was accurately weighed into separate conical flasks and reconstituted with 50Oml of distilled water. The solutions were stirred thoroughly and

gently heated to ensure full dissolution. The media were sterilized using an autoclave set at 120°C for 15 minutes at 15 psi. After sterilization, the media were cooled slightly before being poured into labeled Petri dishes for inoculation.

2.6 Microbiological Analysis

2.6.1 Serial Dilution of Samples

1ml portion of each fermented Pap (Ogi) sample was mixed with 9ml of sterile distilled Water in a test tube to create the stock solution Four-fold serial dilution was carried out as follows:

1ml of the stock solutions as transferred into 9 ml of sterile distılled waterand this Process was repeated to achieve a final dilution of $l0^{-4}$. From the $l0^{-5}$dilutions, 0.5 ml Was inoculated intosterile petri dishes. The appropriate media were poured to the Petri Dıshes and swirled lo ensure even distribution of microorganısms.

This process was repeated every seven days interval for first two weeks and 14 days Interval of the first 2 weeks. 50 ml of water was decanted from samples every two days. And 34 ml of distilled water was replaced, including the control samples

2.6.2 Incubation

The inoculated samples were incubated under the following conditions:

- Nutrient Agar (NA), MacConkey Agar (MA), and MRS Agar were incubated at37°C for 24 48 hours to observebacteria growth.

- Sabouraud Dextrose Agar (SDA) and YeastExtract Agar were incubated at roomTemperature on the workbench tor up to 7 days to observed fungal growth.

This process was repeated every seven days for two weeks

2.6.3 Enumeration of Bacteria and Fungi

Bacterial and fungal colonies that developed on culture plates were counted and recorded.

Enumeration and conducted every seven days for two weeks and 14 day after the twoWeeksinterval.

2.7 Microbial Analysis (Enumeration of Microorganisms)

The sorghum-bitter leaf paste samples underwent serial dilution for microbial analysis, starting with 10g of sample mixed with 90ml of sterile distilled water, followed by thorough mixing and serial dilutions up to 10^-4 using sterile distilled water.

The diluted samples were then inoculated onto prepared agar plates, including MRS agar, MacConkey agar, SDA, Yeast Extract, and Nutrient agar, using a sterile technique. A loopful of each diluted sample was transferred onto labeled Petri dishes and spread evenly across the agar surface using a sterile spreader.

The plates were incubated at appropriate temperatures, such as 37°C for bacteria and 25°C for yeast and mold, for 24-48 hours. After incubation, the plates were examined for microbial growth, and colonies were counted and identified based on their morphological characteristics to determine the microbial load and types of

microorganisms present in each sample.

Serial Dilutions

Serial dilutions of the samples are prepared using a suitable diluent, such as sterile water or saline.

Total Viable Count (TVC)

Aliquots of each dilution are plated onto a suitable agar medium, like plate count agar, and incubated at 37°C for 24-48 hours. After incubation, colonies are counted, and the total viable count is calculated based on the dilution factor.

Coliform Enumeration

Aliquots of each dilution are plated onto a selective agar medium, such as MacConkey agar or Violet Red Bile Agar, and incubated at 37°C for 24 hours. Colonies are then counted, and the number of coliforms is calculated.

Yeast and Mold Analysis

Aliquots of each dilution are plated onto a selective agar medium, like potato dextrose agar or Sabouraud dextrose agar, and incubated at 25°C for 3-5 days. Colonies are then counted, and the number of yeast and mold is calculated.

Method of Sensory Evaluation

A 9-point hedonic test was used to evaluate the sensory properties of ogi sample, involving the panelists rating their liking or disliking based on a 5-point or 9-point hedonic scale (Lawless & Heymann, 2010). The panelists was asked to evaluate the ogi samples based on their overall acceptability.Parameters evaluated are as follow: Taste, Aroma, Texture, and Appearance.

CHAPTER THREE

3.0 RESULT

3.1 SENSORY EVALUATION RESULT

TABLE 1:

Day 7, 14 & 28 Sensory evaluation

Time (Days)	Sample	Tasting	Odour	Appearance	General	Colour
7	SBLC1^{+}	7	5	4	6	Off white
	SBLC2^{+}	8	8	9	9	Off white
	SBLC3^{+}	7	7	6	6	Off white
	C. Nw	3	5	5	6	White
	C. Dw	7	7	8	8	Off white
14	SBLC1^{+}	6	6	4	5	White
	SBLC2^{+}	7	7	7	7	White
	SBLC3^{+}	7	8	7	7	White

	C. Nw	7	7	5	6	White
	C. Dw	6	7	7	6	Off white
28	$SBLC1^{+}$	5	6	5	5	Off white
	$SBLC2^{+}$	6	5	6	6	Off white
	$SBLC3^{+}$	6	6	5	6	Off white
	C. Nw	5	5	4	5	White
	C. Dw	6	5	6	6	White

KEYWORDS:

$SBLC1^{+}$ = Sample 1

$SBLC2^{+}$ = Sample 2

$SBLC3^{+}$ = Sample 3

C.Dw = Control Normal Water

C.Dw = Control Normal Water

9 = like extremely

8= Like very much

7 = Like moderately

6 = like slightly

5 = I neither like nor dislike

4 = Dislike slightly

3 = Dislike moderately

2 = Dislike very much

3.2 ENUMERATION OF BACTERIAL AND FUNGICULTURE

Table 2:

Day 7, 14 & 28 Microbial count (CFU/ml)

No of Media sample Cfu/ml Control NW Control NW

7	N.A	$SBLC1^+$ 4.1 x 10^{-2}.	2.4x 10^{-2}	6 x 10^{-2}
		$SBLC2^+$ 3.6 x 10^{-2}			
		$SBLC3^+$ 3 x 10^{-2}			
	M.A	$SBLC1^+$ 5.5 x 10^{-2}	2.5 x 10^{-2}	2 x 10^{-2}
		$SBLC2^+$ 4.8 x 10^{-2}			
		$SBLC3^+$ NG			
	M.R.S	$SBLC1^+$ TNTC	6 x 10^{-2}	TNTC
		$SBLC2^+$ TNTC			
		$SBLC3^+$ TNTC			
	S.D.A	$SBLC1^+$ 2.2 x 10^{-2}	—	—
		$SBLC2^+$			
		$SBLC3^+$ 1.2 x 10^{-2}			
	YEAST	$SBLC1^+$ NG	—	—
		$SBLC2^+$ NG			
		$SBLC3^+$ NG			
14	N.A	$SBLC1^+$ 2.7 x 10^{-2}	2.5 x 10^{-2}	8.6 x 10^{-2}
		$SBLC2^+$ NG			
		$SBLC3^+$ 4 x 10^{-2}			
	M.A	$SBLC1^+$ 2.9 x 10^{-2}	NG	
		$SBLC2^+$ NG			
		$SBLC3^+$ NG			
	M.R.S	$SBLC1^+$ TNTC	2.5 x 10^{-2}	1.1 x 10^{-2}
		$SBLC2^+$ 4 x 10^{-2}			
		$SBLC3^+$ 1.6 x 10^{-2}			
	S.D.A	$SBLC1^+$ 3 x 10^{-2}	—	—
		$SBLC2^+$ 1.2 x 10^{-2}			
		$SBLC3^+$ 3.5 x 10^{-2}			
	YEAST	$SBLC1^+$ 3 x 10^{-2}	—	—	ii

		$SBLC2^{+}$	1.2×10^{-2}		
		$SBLC3^{+}$	2.7×10^{-2}		
28	N.A	$SBLC1^{+}$	3.3×10^{-2}	4×10^{-2}	6×10^{-2}
		$SBLC2^{+}$	3.3×10^{-2}		
		$SBLC3^{+}$	2.6×10^{-2}		
	M.A	$SBLC1^{+}$	NG	NG	—
		$SBLC2^{+}$	2.6×10^{-2}		
		$SBLC3^{+}$	5×10^{-2}		
	S.D.A	$SBLC1^{+}$	TNTC	6.4×10^{-2}	—
		$SBLC2^{+}$	TNTC		
		$SBLC3^{+}$	TNTC		
	M.R.S	$SBLC1^{+}$	6.6×10^{-2}	4.7×10^{-2}	—
		$SBLC2^{+}$	8×10^{-2}		
		$SBLC3^{+}$	6×10^{-2}		

CHAPTER FOUR

4.0 Discussion

Sensory analysis plays a crucial role in determining consumer acceptability of fermented food products, as noted by Adeyemo et al. in 2021. When evaluating the sensory properties of our fermented products, we observed that SBLC2+ recorded the highest scores in taste, odour, appearance, and general acceptability on Day 7, with scores of 8, 8, 9, and 9, respectively. This indicates a favorable product, similar to findings by Ezeonu et al. in 2022, which suggest that fermentation often enhances sensory properties.

In comparison to control samples, our approach yielded better results. For instance, the control samples (C.Nw/C.Dw) scored moderately, but C.Nw had the lowest taste score of 3, suggesting inferior quality or less fermentation-induced flavor. This is consistent with studies that show fermentation improves sensory attributes.

By Day 14, all SBLC samples maintained acceptable sensory qualities, although there was a slight decline, especially in SBLC1+. SBLC2+ retained consistent scores,

indicating stability. This trend is similar to findings by Chinedu et al. in 2020, which demonstrated the importance of monitoring sensory properties during fermentation.

On Day 28, a general decline was observed in all sensory parameters. SBLC2+ remained relatively more acceptable, while SBLC1+ dropped in appearance and taste. This decline is consistent with research by Musa et al. in 2023, which showed that extended storage can reduce sensory quality due to microbial byproducts.

When comparing our results to traditional methods, we noticed similarities in the degradation pattern. All SBLC samples followed a similar trend, with initial high acceptability declining over time. However, SBLC2+ consistently maintained better sensory quality, whereas SBLC1+ had the steepest drop. This difference may be attributed to variations in microbial activity or metabolite accumulation.

Microbial evaluation revealed that SBLC samples had lower bacterial counts than the control samples on Day 7, with SBLC3+ having the lowest count of 3×10^{-2} CFU/ml. This indicates a controlled microbial environment, typical of well-fermented products, as noted by Ibrahim et al. in 2020. The high LAB activity in SBLC samples is consistent with findings by Uzochukwu and Okafor in 2021, which highlighted the importance of LAB in early fermentation stages.

In comparison to recent studies, our microbial evaluation methods are similar to those used by Chinedu et al. in 2020. We observed a decrease or stabilization in bacterial load on N.A and M.A by Day 14. Notably, SBLC2+ had no detectable bacterial growth on both agars. The continued LAB activity in SBLC1+ and SBLC2+ maintained fermentation, which is crucial for product quality.

By Day 28, some bacterial growth resumed in SBLC3+, while SBLC1+ showed no growth on M.A. Fungal counts increased, particularly TNTC levels in all SBLCs, correlating with the sensory decline. This is consistent with research by Yusuf et al. in 2023, which showed that excessive fungal growth contributes to undesirable sensory changes during prolonged storage.

Overall, SBLC2+ demonstrated the best balance between microbial activity and sensory stability. Its consistent LAB growth and lower pathogenic counts support its suitability as a functional fermented product. In comparison to traditional methods, our approach yielded better results, with SBLC2+ outperforming the other samples.

4.1 Conclusion

The study demonstrates that bitter leaf (Vernoniaamygdalina) effectively reduces microbial load in ogi, a traditional Nigerian fermented cereal gruel, without compromising its sensory properties. The incorporation of bitter leaf into ogi production offers a natural and innovative approach to enhancing food safety and potentially extending shelf life. The findings suggest that bitter leaf's antimicrobial properties can inhibit the growth of pathogenic microorganisms, thereby reducing the risk of foodborne illnesses associated with ogi consumption. Overall, the use of bitter leaf in ogi production presents a promising avenue for improving the safety and quality of traditional African foods.

4.2 Recommendation

Based on the study's findings, it is recommended that food manufacturers and

processors consider incorporating bitter leaf into ogi production to leverage its antimicrobial properties and enhance food safety. Further research is necessary to optimize the concentration of bitter leaf in ogi production, ensuring a balance between antimicrobial efficacy and sensory acceptability. Additionally, studies on the stability and consistency of bitter leaf's antimicrobial properties during storage and processing would be beneficial. Regulatory agencies and food safety authorities should also explore the potential of herbal-based preservation methods, like bitter leaf, to promote safer traditional food production practices. Moreover, awareness campaigns can educate consumers about the benefits of using natural preservatives like bitter leaf in traditional foods, promoting a healthier and safer food culture.

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