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Research Article

BURONG TILAPIA: FACTORS ON THE FERMENTATION PROCESS AND ITS MICROBIOTA

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Abstract: Fermented fish products hold significant prominence in the Philippines. However, its potential risks for human consumption are faced as a result of inadequate awareness of microbiological standards in its preparation. The objectives of this study are: (1) to determine the microbiota and fermentation process of *burong tilapia*, (2) to analyze how microbial species is influenced by fermentation factors such as salt content, temperature, and proper sanitation, and (3) to evaluate how it can impact food safety for human consumption. The data gathering procedure in this case study included the sample preparation of *burong tilapia* and its microbiological analysis such as Aerobic Plate Count, *E. coli* Count, and Yeast and Molds Count. This study revealed that the population of aerobic microorganisms in *burong tilapia* was relatively high with the Aerobic Plate Count result of 2.1 x 107 CFU/g, indicating significant bacterial presence. On the other hand, a relatively low level of *E. coli* and Yeast and Molds count was detected having <25 CFU/g and <10 CFU/g, respectively. Moreover, it was found that low salt content, non-ideal temperature conditions and failure to strictly observe sanitation practices, all influenced the growth and proliferation of unwanted microorganisms. These results emphasize how crucial it is to optimize fermentation factors in order to guarantee both the food quality and safety of fermented fish products for human consumption. Furthermore, this case study highlights that consumer education is needed in promoting awareness and acceptance of fermented fish products in the market.

Keywords: *Fermentation of fish, Aerobic plate count, Food quality and safety*

INTRODUCTION

In Asia, fermented foods are essential components of diets because it provides nutritional balance and food security. However, in a developing country like the Philippines, the popularity of these food products is attributed to the socio-economic status or living conditions of people since other processing methods, such as freezing and canning, are generally expensive for Filipinos (Olympia, 1992). Other than that, the high level of humidity in the country

Figure 1. Burong Tilapia Food Product

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makes fermentation on its tropical islands relatively basic giving them regular access to fermented foods.

Figure 2. Buro in a sealed container

"*Buro*" as in "*pagbuburo*," a Filipino word for fermentation, is the old Filipino style of fermenting food with varied concentration of salt (Amoyan, 2019). The Bicol region in the Philippines is known for practicing this preservation of food called "buro". Tilapia or commonly known in English as St. Peter's fish is the main medium of the understudied. This kind of freshwater fish is abundant in the Philippines and it is usually grown in cages in a fish farms.

Animal-based fermented food made of fish can be divided into two groups. The first group includes those containing high concentrations of salt—about 15 to 20 percent in the final product. This group consists of bagoong (fish paste) and patis (fish sauce). These products are generally used as condiments. The second group includes *burong isda* (fermented rice fish mixture) and *burong hipon*, also known as *balao balao* (fermented shrimp rice mixture). In the Bicol region, similar fermented fish products are known as hingmay wherein small Bolinao fish is used, and *kuyog* which uses small rabbitfish (*danggit*) and is uniquely characterized by adding ginger (Nuncio et. al., 2020).

Freshwater fish is used in the preparation of *burong isda* and then it is mixed with salt, about 5 to 6 tablespoons per 500 grams (Katz, 2012). Sanchez (1999) detailed that the ratio of rice and fish is at 65:35 and the fermentation is carried out for at least 5 months at 26°C to 30°C.

This article argued to provide specific data on the microbiota of *burong tilapia* strengthened by microbiological analysis and further determine its quality and safety for human consumption. Specifically, the purpose of this study is to answer the following questions: What is the level of microbial species present in *burong tilapia*? How the microbial content of *burong tilapia* does is influenced by these fermentation factors: a. salt content; b. temperature; c. proper sanitation? and how does the microbial species in *burong tilapia* impact its safety for human consumption?

Recent events in food security in the country has been the most crucial issues the government are currently addressing, hence, processes like this help secure food from spoilage and address the sustainable development goal number two on zero hunger, 3 on good health and well-being and number 14 on life below water. With these as answering the call of SDGs, the timely and relevant conduct of this study is put forth.

METHOD

This article focused on qualitative and quantitative data using multiple case analyses of the variables, such as the primary and secondary data recording and analysis from the laboratory results to lived experiences of the traditional fish fermentation processes in Bicol region, Philippines. Data recordings were utilized during the sample preparation and fermentation process, which were the basis in analyzing the factors affecting fermentation, such as: (1) salt content, (2) temperature, and (3) proper sanitation. The records were subject to analysis according to the laboratory results of the sample, which were further utilized in

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identifying the microbiota of *burong tilapia* and its safety for human consumption. Two (2) food technologists and (2) chemistry laboratory experts certified the phases and the results of the data after careful and standard mechanisms were observed and obtained. The study runs for six (6) months in the laboratory, which worked with strict supervision.

The data-gathering procedure in this study included the sample preparation of *burong tilapia* and its microbiological analysis, such as aerobic Plate Count, *E. coli* count, and yeast and molds count. Sample preparation was conducted at the laboratory facility at Cadlan, Pili, Camarines Sur, on April 10, 2024. The recipe of the sample is based on an integrated research-based recipe and traditional way of its preparation. Raw ingredients such as rice, tilapia, and salt were purchased in the local market of Pili, Camarines Sur. The proportion of the raw ingredients was: 65% of cooled cooked rice and 35% tilapia mixed with 10% of salt. With 750 grams of total sample, this is equivalent to 455 grams of cooled cooked rice mixed with 25 grams of salt and 245 grams of tilapia slices mixed with 50 grams of salt: 1. Rice was cooked a day before the preparation, and was mixed with the corresponding amount of salt during preparation; 2. The tilapia was washed, eviscerated, deboned and its meat was chopped into medium sizes. It was mixed with the corresponding amount of salt, and was set aside for at least 30 minutes; 3. The rice and tilapia samples were mixed in a bowl thoroughly and was later packed into a wide-mouth sealable jar sterilized in boiling water for 15 minutes. Space at the top was left for expansion. Afterwards, the jar was sealed; 4. After 11 days, on January 21, 2024, 300 grams of the sample were brought to the food testing laboratory for microbiological analysis.

The three tests utilized for the microbiological analysis are enumerated below. These tests used the Petrifilm method, which is a microbial testing technique which involves the use of Petrifilm plates, which are thin, flexible, and self-contained culture media films. Moreover, procedures followed in the conduct of the analysis are based on Official Methods of Analysis of AOAC (Association of Official Agricultural Chemists). *International*. 1.Aerobic Plate Count (APC). The aerobic plate count is an indicator commonly used in the food industry to determine the level of microorganisms in a certain product (Maturin, 1998). 2. E. coli Count - The E-coli count is an indicator of fecal contamination in food, water, and environmental samples, as high levels of *E. coli* may indicate the presence of fecal matter and potential pathogens. 3. Yeast and Molds Count Quantifying fungi and mold and yeast contents can be a form of assessment of microbiological quality (Oliveira, et. al., 2012).

RESULTS AND DISCUSSION

The level of microbial species presents in *burong tilapia* is shown in table 1 below. The results of this presence of microbial content were certified observed and corrected by the experts in the field of microbiology.

Table 1. Microbial Species Level in *Burong tilapia*

Sample	Parameters	Result	Level	FDA Standard
750 grams	Aerobic Plate Count (APC)	2.1×10^7 CFU/g(Failed)	High	5×10^5 CFU/g
	E. Coli Count	<25 CFU/g(Negative)	Low	10 CFU/g
	Yeast and Molds Count	<10 CFU/g(Passed)	Low	10^3 CFU/g

Table 1 reveals the data results of tests conducted to determine the level of microbial species in the 750-gram *burong tilapia* sample. The Aerobic Plate

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Count (APC) in the sample of *burong tilapia* of 2.1 × 10^7 CFU/g (colony-forming unit per gram) indicates a relatively high level of aerobic bacteria. This exceeded the acceptable limit, suggesting potential spoilage that compromises the safety and quality of the sample. The excessive microbial load could lead to spoilage, a foul odor, and degradation of the texture of the product. On the other hand, the *E. Coli* counts with less than 25 CFU/g being negative, an indicator of fecal contamination and unsanitary processing (Feng et al., 2020), were not detected. *E. Coli* presence is a critical indicator of fecal contamination and unsanitary handling, and its absence is a positive outcome. Further, despite being considered an aerobic bacterium too, *E. coli* result is different from the APC since there has been a separate microbiological analysis test conducted to detect such. During the tests, specific pathogens were not observed due to the testing capacity of the food laboratory.

Similarly, the yeast and mold count with less than 10 CFU/g being passed indicates a relatively low level of fungal contamination in the *burong tilapia*, which suggests a minimal risk. Fungal growth can negatively affect the sensory characteristics and safety of the product.

Despite the positive aspects of low *E. Coli* and yeast and mold counts, the high APC poses a significant concern. An excessive number of aerobic bacteria can potentially generate toxins harmful to consumers. This highlights the need for stricter quality control measures throughout the *burong tilapia* production process. These acceptable limits or levels are based on the Food and Drug Administration Guidelines for the Assessment of Microbiological Quality of Processed Foods in the Philippines**.**

Microbial content of *burong tilapia* be affected by the fermentation factors such as salt content

Table 2. NaCl (Sodium chloride) presence in the *burong tilapia*

Sample	Salt Content
750 grams of *burong tilapia*	75 grams (10%)

Table 2 shows the salt content of *burong tilapia* sample. The level of salt content added in the rice and tilapia is 75 grams, encompassing 10% of the total sample.

Appropriate salt concentrations can alter the microbial community composition of fermented foods (Lee, et. al., 2021). The APC result suggests that there are approximately 21,000,000 CFUs of aerobic bacteria per gram in the *burong tilapia* sample. This indicates a relatively high level of aerobic bacterial contamination. In this study, *burong tilapia* is considered as a fermented fish product with relatively low concentration, therefore, at low levels, salt did not sufficiently suppress the growth of aerobic bacteria.

Salt has the property of removing the water presence in any of the food hence the lesser the microorganisms lived since water is the niche of microorganism.

Table 3 shows the recorded room temperature during the storage of *burong tilapia* samples from January 11 to May 11, 2024. Room temperature measurements were taken regularly at exact times to determine temperature variations. The temperature ranged from 26°C to 29°C, with an average temperature of 27.2°C. Indicating that the medium and the facility adheres to standard climatic suitable place for the experimentation processes ensuring no harmful microorganism may affect the result of the *buro*.

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Table 3. Temperature of the processes of fermentation (As observed and recorded in the facility)

Fermentation Duration (Summation by month)	Time Checked	Temperature (°C)
January 2024	6:35 AM – 4:55 PM	26 °C
February 2024	6:35 AM – 4:58 PM	28 °C
March 2024	6:33 AM – 4: 51 PM	27 °C
April 2024	6:30 AM – 4: 53 PM	28 °C
May 2024	6:35 AM – 4: 48 PM	27 °C
	Average Temperature	**27.2 °C**

The temperature during fermentation significantly impacts the microbial content of *burong tilapia*. Studies have shown that warmer temperatures favor specific bacterial communities compared to cooler environments (Lu, et. al., 2019). Ideal fermentation temperatures for fermented fish products lie around 28-30°C (Sanchez, 1999). At this range, there's a dominance of lactic acid bacteria (L.A.B.) like Lactobacillus spp. Lower temperatures can hinder the growth of L.A.B., potentially leading to the proliferation of undesirable microorganisms. This can compromise the safety qualities of *burong tilapia*. Conversely, excessively high temperatures can also be detrimental. As observed in the reference study by Cabello-Olmo et al. (2020), higher temperatures of 37°C above significantly reduced the overall bacterial load for fermented fish products. This suggests that temperatures exceeding the optimal range might negatively impact the total microbial count in *burong tilapia*, if not controlled.

On Sanitation

Proper sanitation practices that were practiced during the sample preparation of *burong tilapia*. There were five areas investigated such as personal hygiene, dishwashing, utensils and equipment, food storage and dry storage, and food preparation, which consisted of a total of 19 proper sanitation practices distributed in each of the areas. Such practices followed by the food preparers were: (1) proper and frequent hand washing during the preparation; (2) cleaning utensils every time after use; (3) bowls and plates were used to transport ingredients; (4) the table for preparation is placed 6 to 8 inches off the floor; (5) the sample was protected from contamination through removing unnecessary objects in the area; (6) food contact surfaces are properly washed before every use; (7) clean reusable towels were used for cleaning utensils and surfaces; and (8) others. Therefore, the majority of the proper sanitation practices were strictly observed.

Table 4. Proper Sanitation Practices during the Preparation of *Burong tilapia*

Areas of Proper Sanitation Practices	No. of Proper Sanitation Practices FOLLOWED	No. of Proper Sanitation Practices NOT FOLLOWED
Personal Hygiene (6)	6	0
Dishwashing (3)	3	0
Utensils and Equipment (3)	3	0
Food Storage and Dry Storage (4)	4	0
Food Preparation (3)	3	0

Inadequate hygiene conditions and the lack of standards for safety controls in the preparation of fermented foods can result in the presence of pathogenic microorganisms (Skowron, et. al., 2022). The value of *E. coli* count detected upon the microbiological analysis indicates the presence of less than 25

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colony-forming units per gram (<25 CFU/g). Meanwhile, the value of yeast and molds detected upon the microbiological analysis indicates the presence of less than 10 colony-forming units per gram (<10 CFU/g). However, there are still loopholes when conducting fermentations, even if you followed strictly the processes and measures, microorganisms has its ability to proliferate in such conditions that are beyond the limits when it is not stationed by the researchers. For both results, this indicates a low level of *E. coli*, and yeast and molds detected in the sample. In this case study, only minimal proper sanitation practices were followed, therefore, this indicates that such measures do not significantly affect the growth of the aforementioned pathogens in *burong tilapia*.

Impact its safety for human consumption

Table 5 shows the data result from the microbiological analysis conducted to analyze and determine the microbial species in *burong tilapia* and its safety for human consumption.

Table 5. APC Result of *Burong tilapia* in Comparison to FDA Standards

Parameters	Result (CFU/g)	FDA Accepted Level (CFU/g)	FDA Level of Rejection (CFU/g)
Aerobic Plate Count (APC)	2.1×10^7	5×10^5	10^7

APC with 2.1 x 107 CFU/g or 21,000,000 CFU/g result shows a high level of aerobic bacteria present in the sample. This indicates that the sample may have been exposed to conditions that promote bacterial growth even if the observed standards and high hygiene were followed. This includes several conditions why it happened, and these maybe due to not properly exercising hygiene during food handling, poor storage conditions, specifically the improperly sealed container, or unconditioned processing environments while the medium is under observation. The study of Silva et. al. (2023) discussed that factors contributing to high aerobic plate counts (APC) in food samples include hygiene practices, storage conditions, and processing environments.

With the presented scenario above, still a high aerobic plate count indicates potential spoilage or the presence of harmful pathogens, making the *burong tilapia* unsafe for human consumption. The standard aerobic plate count in a food is typically less than 10,000 CFU/g. Lower aerobic plate counts are considered safer for food consumption. However, due to the high aerobic plate count, the sample of *burong tilapia* is contaminated with unwanted bacteria, which may cause foodborne illnesses and may pose health risks (Gizaw, 2019). This result shows the consumption of *burong tilapia* is not safe due to its health-risk-related factors.

Meanwhile, the result of the *E. coli* count in the sample shows a lower level, indicating that it is within acceptable safety limits. The low count suggests that the sample has not been significantly exposed to conditions promoting E. coli contamination, such as poor hygiene during food handling or unsanitary processing environments. The yeast and mold count with <25 CFU/g result shows a low level in *burong tilapia*, which indicates a minimal risk to the products present in the sample. This low count indicates that there is very little mold and yeast in this sample, making it within acceptable safety limits for human consumption. A low count of mold and yeast in food is generally a good thing as it indicates that

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there won't be any harmful organisms or severe spoilage that might threaten the safety and quality of the meal. These low counts reduce spoilage and toxins, ensuring quality and a longer shelf life for human consumption (Gilbert et al., 2000).

The result indicates that minimal proper sanitation practices performed during the process and fermentation does not totally affect the growth and production of microbial species such as *E. coli* and yeast and mold. However, the overgrowth of aerobes or aerobic microorganisms indicates improper hygiene practices. Therefore, cleanliness and proper food handling must be observed to control the growth of aerobes and to ensure that *burong tilapia* is safe for human consumption and prevent the risk of foodborne illnesses.

Manufacturers of fermented food should consider the following strategies to optimize the quality and safety of *burong tilapia* and the like: (1) increasing salt content for at least more than 10% of the total rice and fish mixture is crucial to inhibiting unwanted bacteria growth; (2) Maintaining an ideal temperature condition; within the ideal range of 28-30°C is essential. Regulate fermentation temperatures through the use of thermometers or temperature-controlled storage; and (3) regularly clean and sanitize equipment and surfaces, along with good hand hygiene.

CONCLUSIONS

As a result, this study demonstrated that the *burong tilapia* sample had more aerobic bacteria than was considered safe. This suggests that food may rot as a result of careless handling or insufficient fermentation procedures. This poses risks to the safety and quality of the product. However, the low concentrations of yeast, molds and *E. coli* all point to a low level of fecal matter and fungal infection. These results emphasize how crucial it is to follow appropriate fermentation procedures and enforce stronger cleanliness regulations in order to guarantee both the quality and safety of *burong tilapia* for human consumption.

While *burong tilapia* holds a cultural significance as a traditional fermented food that is widely enjoyed in Bicol, Central Luzon and other parts of the Philippines, its safety for consumption depends on proper handling and fermentation processes. This study emphasizes the significance of maintaining appropriate fermentation conditions, such as salt content, temperature and sanitation, as well as maintaining food safety requirements to reduce pathogenic microbial growth that may harm consumers.

DECLARATIONS

This section should include as follow:

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