Product Development of Bangus (*Chanos chanos*) enriched with Guso (*Eucheuma sp.*) Chorizo (BAGURIZO)

ABSTRACT

The research aimed to assess how varying percentages of seaweed gel affect the sensory attributes of Milkfish Sausage. Treatments included a control (Treatment 1) and four experimental groups with different seaweed gel percentages (Treatment 2: 2%, Treatment 3: 4%, Treatment 4: 6%, Treatment 5: 8%). Completely Randomized Design (CRD) was employed, with ANOVA utilized for data analysis. Thirty untrained participants from SPAMAST, comprising faculty, staff, and students, were randomly chosen for sensory evaluation, irrespective of gender or age. Evaluation criteria encompassed color, aroma, taste, texture, and overall acceptability using descriptive and hedonic scales. While no significant variations were observed in aroma, taste, or texture, color and overall acceptability showed notable differences.

Regarding microbial analysis post-storage for three days, Treatment 2 and Treatment 5 exhibited significant differences. Additionally, three treatments underwent proximate analysis, focusing on potassium, crude protein, moisture, ash, crude fat, magnesium, zinc, and iron. Magnesium levels increased with higher seaweed gel proportions, while moisture content decreased. Other components displayed varying trends.

The study suggests further investigations into cooking loss, physico-chemical parameters, moisture content, and shelf-life of seaweed-enriched Milkfish Sausage to comprehensively understand its properties and potential applications.

KEYWORDS: Milkfish (Bangus), Seaweed (Guso), Sausage

INTRODUCTION

The Philippine fish processing industry began with traditional methods like drying and smoking, later advancing to modern techniques such as canning [8,14-18]. Growing consumer demand for premium meat prompts the development of meat products enriched with health-enhancing additives [37,19-25]. The latest advancement in the country involves the production of value-added items crafted from minced fish or surimi, such as fish sausage, meeting consumer preferences for healthier meat options with reduced fat, sodiu, cholesterol, nitrates, calories, and additional health-promoting components, commonly available in supermarkets [39, 26-30]. Sausage is the outcome of combining appropriate ingredients in precise proportions, alongside a well-defined structure and regulated production methods, with its quality consistently mirroring the condition of the raw materials and the manufacturing process. Sausage manufacturing is a well acknowledged method of combining ground meat with spices, because they are delectable, healthful, and portable qualities [2,5-10]. Milkfish, a key ingredient in fish sausage, is a significant food in Southeast Asia, notably in Indonesia, Taiwan, and the Philippines, recognized for its high-protein content. Therefore, possessing nutritional information about milkfish is crucial for both consumers and the fisheries processing industry to address [19-31-40]. On the contrary, seaweeds have broad applications ranging from human consumption, animal feed, dietary alternatives, to serving as gelling agents, stabilizers, thickening agents (hydrocolloids like agar, alginate, and carrageenan), and functional food additives [9-15]. Adding seaweed dietary fiber to fish sausage can provide dietary fiber to the product while also improving its quality and texture [10-14]. The research aimed to determine the development of seaweed enriched milkfish sausage. Specifically, it aimed to: (1) To determine the sensory qualities (color, aroma, flavor, texture, and general acceptability) of seaweed-enriched milkfish sausage; and (2) To determine the microbial content of seaweed-enriched milkfish sausage.

MATERIALS AND METHODS

Research Locale

The product seaweed-enriched milkfish sausage was conducted at SPAMAST Shared Service Facility (SSF) Center, Malita, Davao Occidental, Philippines, and sensory evaluation was conducted at Post-harvest Processing (Sensory Evaluation Room).

Research Design and Instrument

The experiment employed five treatments, each replicated three times, within a Completely Randomized Design (CRD) to assess the impact of individual treatments. The 60% fish was steady to all treatments while 40% is variable dependent on the combination of fats, seaweeds, and other ingredients.

Table 1. The different treatments of seaweed-enriched milkfish sausage

Treatments	Description
1 (Control)	60% Fish + 40% Fats (317g fats + 83g other ingredients)
2	60% Fish + 40% Fats (309g fats) + 2% (8g seaweed gel) + 83g other ingredients
3	60% Fish + 40% Fats (301g fats) + 4% (16g seaweed gel) + 83g other ingredients

4	60% Fish + 40% Fats (293g fats) + 6% (24g seaweed gel) + 83g other ingredients
5	60% Fish + 40% Fats (285g fats) + 8% (32g seaweed gel) + 83g other ingredients

Procurement of Raw Materials

Seaweed (*Eucheuma spp.*), milkfish and all other products were procured from the public market of Malita, Davao Occidental, Philippines. The procured materials were placed in a clean container with cover to avoid exposure to dust and ashes. The actual preparation and production of the product was conducted at SPAMAST Shared Service Facility (SSF) Center and sensory evaluation was conducted at Post-harvest Processing (Sensory Evaluation Room).

Preparation of Milkfish

Milkfish were washed thoroughly in running water to remove adhering dirt and other particles from the fish. The bones and visceral organs of the fish were separated. After segregation, the fish meat was wash thoroughly and sliced manually, and grounded using a blender.

Preparation of Seaweeds

Drying of Seaweeds

The fresh seaweed is washed and soaked first with rice washing water for twelve hours or overnight, the following day seaweeds were dried for 7 hours or more using a solar dryer depending on the presence of sunlight.

Seaweeds Powder

Dried seaweed is placed into a coffee or spice grinder and are grind finely using a blender. Ultimately, the seaweed powder was moved to an airtight container and kept in a cool, dry location.

Blending and Cooking/gelling

The 80 grams powdered seaweeds were blended with 400 ml water. Thereafter, seaweeds were boiled with water. While heating, the mixture was stirred occasionally in one direction until it would become sticky. Seaweed gel was poured in a plastic basin and set aside for further use [16-17].

Preparation of Pork Fats

Pork fats were weighted for the desired ratio of each treatment and washed thoroughly in running water to remove dirt and other particles. The pork fats were trimmed/cut into small pieces for easy grinding. After cutting/trimming, the pork fat was grinded using the meat grinder. The pork fat used is fresh and is subjected to refrigeration after every process to ensure that the bacteria would not spread rapidly.

Production of Seaweeds-Enriched Sausage

The following ingredients were utilized on production of seaweed enriched sausage for every treatment, namely 600 grams' fish meat; pork fat (according to treatment); seaweed gel (according to treatment); 20 grams' salt; 30 grams' sugar; 10 grams' garlic; 20 grams' vinegar; 6 grams' black pepper and 4 grams MSG.

The skinless and boneless milkfish were grounded. Pork fat was grounded separately. The ground fish and pork fat were mixed well with all other ingredients and were packed into casings. The finished product was labelled according to its treatment, then stored in the refrigerator.

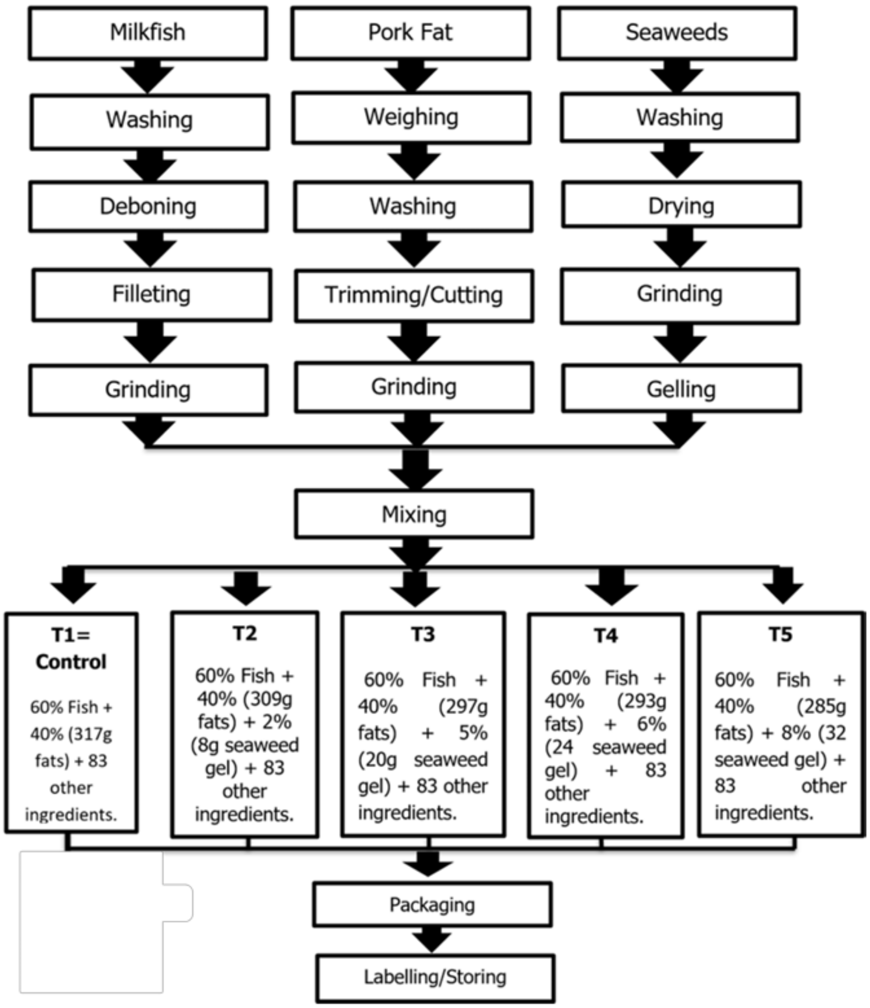

Fig. 1. Process flow chart of seaweed-enriched milkfish sausage.

Sensory Evaluation

The different samples of seaweed-enriched milkfish sausage were evaluated by 30 untrained selected respondents. The respondents were randomly from among the faculty, staff, and students of SPAMAST regardless of gender and age. The respondents were given coded samples of the products and a score sheet for the sensory evaluation (color, aroma, taste, and texture) and general acceptability. It used Hedonic scale in determining the Sensory qualities in color, texture, odor, taste and general acceptability. 60% of fish are steady to all treatments while 40% are variable dependent on the combination of fats and seaweeds [32].

Microbial Analysis

A single gram sample was thoroughly mixed with 9 milliliters of sterile saline solution containing 0.85% NaCl. Serial dilutions were carried out with the same diluents. The dilute suspension was poured on the standard plate count agar. All the inoculated plates were incubated for 24 to 48 hours at 28 0C- 37 0C. The colony forming units (CFU) were counted using a colony counter. Frequency of microbial count was assessed [41,14-20]

Statistical Tools and Analysis

All data from the experiment were subjected to Analysis of Variance (ANOVA) to evaluate the treatment effects. To determine significant difference among treatments, Tukey's test was utilized.

RESULTS AND DISCUSSION

Sensory Evaluation

A total of five attributes were evaluated for sensory evaluation namely: color, aroma, taste, and texture. Sensory evaluation was done using the descriptive and the hedonic scale ratings. General acceptability of the product was also determined using a nine-point hedonic scale.

Table 2. Descriptive mean sensory score of seaweed-enriched milkfish sausage for color, aroma, taste, texture, and general acceptability.

Treatments	Sensory Attributes				
	Color^{s}	Aroma^{ns}	Taste^{ns}	Texture^{ns}	General Acceptability^{s}
1 (Control)	3.84^{a}	3.69	3.69	3.38	7.47^{ab}

2	3.59[ab]	3.56	3.63	3.52	7.53[a]
3	3.56[a]	3.42	3.37	3.20	7.23[ab]
4	3.36[ab]	3.66	3.39	3.34	7.03[ab]
5	3.20[b]	3.79	3.47	3.53	6.89b

ns = not significant

s= significant

Descriptive Criteria

	Color:		Aroma:		Taste:		Texture:
5	Light brown	5	Absence of seaweed aroma	5	Absence of seaweed taste	5	More hard than soft texture
4	Slightly pronounced brown	4	Slightly pronounced seaweed aroma	4	Slightly pronounced seaweed taste	4	Slightly soft texture
3	Moderately pronounced brown	3	Moderately pronounced seaweed aroma	3	Moderately pronounced seaweed taste	3	Moderately soft texture
2	Pronounce brown	2	Pronounced seaweed aroma	2	Pronounced seaweed taste	2	Pronounce soft texture
1	Very pronounced brown	1	Very pronounce seaweed aroma	1	Very pronounce seaweed taste	1	Very soft texture

Hedonic Scale

General Acceptability:

9	Like extremely	4	Dislike
8	Like very much	3	Dislike moderately
7	Like moderately	2	Dislike very much
6	Like slightly	1	Dislike extremely
5	Neither like nor dislike		

Color

Treatment 1 with no addition of seaweed gel has a comparable effect to treatment 3 with 4% seaweed gel and significantly higher than the rest of the treatments. Treatments with 2%, 6% and 8% were comparable. Consequently treatment 1 to 3 were rated slightly pronounced brown while treatment 4 and 5 were rated moderately pronounced brown by the evaluators. The study's findings aligned with Munsu *et al.'s* research (2021), indicating that incorporating seaweed into chicken sausage affected its color characteristics, resulting in darker hues with increased seaweed content [21]. Incorporating dietary fiber notably influenced both water holding capacity and color, with increased fiber enhancing water retention and imparting color, particularly the lightness (L*) values, attributed to reduced fat and fiber incorporation [19-21]. Razali *et al.,* (2019), reported comparable result, in which utilizing seaweed (*Kappaphycus alvarezii*) in Malaysian food products which resulted to L* value significantly increased (became lighter) with increasing concentration of seaweed in fish sausages [31]. Another study by Tanase *et al.,* (2019), due to the presence of beta-carotene, chlorophyll a, chlorophyll b, and xanthophylls, green seaweeds range in color from yellow to green [35-40]. The presence of yellow pigments such as chlorophyll, phycopine, and xanthophyll in SP contributes to its dark hue, leading to a reduction in brightness observed in chicken sausages containing SP. These pigments overlap in spectrum with others like chlorophyll and beta-carotene.

Aroma

Constantly, all treatments showed no significant difference. As shown in Table 2, all treatments with an addition of seaweed gel were comparable to treatments with no addition of seaweed gel. Treatment with addition of 2% seaweed gel and the control were rated as slightly pronounced seaweed aroma while treatment with 4%, 6% and 8% were rated as moderately pronounced seaweed aroma. The fragrance characteristics of various varieties of seaweed vary and might be anything from fresh and grassy to saltwater and sulfurous. For instance, some seaweeds are rich in chemicals that include sulfur, which can give the dish a strong, sulfurous fragrance. Other seaweeds have a high concentration of aromatic chemicals including terpenes and aldehydes, which may give meals a fresh, flowery, or fruity flavor [7,9,10]

Taste

In this study, the impact of seaweed gel on product taste was deemed statistically insignificant ($p<0.05$), as indicated in Table 2. Although respondents rated the control group (Treatment 1) highest in sensory evaluation, followed by treatments 2,5,4, and then 3, no significant differences were observed. Maghfiroh (2000) suggested that food taste is influenced by its components such as protein, fat, and carbohydrates. Seaweeds inherently possess a salty taste due to their high mineral content, primarily potassium, serving as a healthy alternative to sodium in food [18]. Furthermore, certain seaweeds contain various flavor components that can be extracted through gentle processing, enhancing food flavor naturally. The unique chemical compositions of seaweeds can impact sensory perceptions, influenced by factors like species, season, and environment [20]

Texture

As to texture, the effects of seaweed gel were found to be not significant. Treatment 2 with 2 % seaweed gel exhibited a higher than the mean rating than the other treatments, but no significant difference was detected. Treatment 2 was rated as slightly soft texture which may be because hardness and chewiness increased with more addition of seaweed in the sausage formulation [22] still, all treatments were comparable. Consequently, among the five treatments, treatment 2 were rated slightly soft texture while the rest were rated as moderately soft texture by the evaluators. The soft texture observed is likely attributed to the presence of dietary fiber in seaweed, which undergoes disintegration upon heating during sausage cooking. Typically, dietary fiber is incorporated into foods to enhance texture, particularly insoluble fibers, known for their effective water holding and swelling properties [36-41].

General Acceptability

Treatment 2 with 2% of seaweed gel has a significant effect with treatment 5 with 8% of seaweed gel. Treatments with 4%, 6% and with no seaweed gel (control) were comparable. Consequently treatment 2 was rated as like very much while treatment 1, 3, 4 and 5 were rated like moderately by the evaluators. Results of the study showed that as the amount of seaweed gel increased there is a decrease in the overall acceptability of the product. Consumers showed a preference for sausages with lower seaweed content, consistent with their findings on the inclusion levels of various brown and red seaweeds in pork sausages, where evaluators favored nori>SS>IW>PP [8,14-18]. The result revealed that no significant difference detected for Aroma, taste, and texture. However, significant effects of adding seaweed gel at 4% were observed in color and 2% seaweed gel for general acceptability. Color shows significant difference since additional fiber (present in seaweed), water holding capacity increased, and color was impacted, notably lightness (L*) values since less fat and a more fiber was added. The same with the study of Munsu *et al.,* (2021), seaweed has changed the way chicken sausage looked in terms of color [22]. When additional seaweed was added, the chicken sausages became darker because of more seaweed it contains. The seaweed components phycoerythrin and phycocyanin are what changed the color of the chicken sausages (KA)

Microbial Analysis

Initial microbial counting was done immediately after the product was prepared to assess microbial quality of the product and to determine the trend of microbial count across the different treatments. As seaweed gel increased microbial count decreased. Statistically, there was a significant difference among treatments. Treatment 1 and 5 shows a significant difference of 0.033 was observed in 3 days of storage using Dunn's pairwise test.

Table 3. Data on treatment mean score of microbial count on the 10^3 days of storage.

Treatments	**Days of Storage**		
	Day1	**Day 2**	**Day 3**
1 (Control)	8×10^4	1.08×10^5	1.68×10^5
2	6×10^4	6.8×10^4	9.2×10^4
3	5.6×10^4	6×10^4	8.4×10^4

4	5.4 X 10^4	5.6 X 10^4	6.8 X 10^4
5	3.6 X 10^4	4.4 X 10^4	5.6 X 10^4
Grand Total			1.088 X 10^6

S=Signifant

Microbial analysis of the product also assessed its quality and safety for consumption. Results revealed that microbial count of the product decreases as incorporation of seaweed gel increases. This is mainly due to the antimicrobial property present in seaweed. Phenols, fatty acids, carbohydrates, proteins, and other minor chemical compounds have been identified as the active chemical components that give algae its antibacterial capabilities [12-15]. Even though most of the substances in seaweed that have antibacterial properties are mostly phenolic and polysaccharide components, and their mode of action may be stasis (inhibition of microbial growth) or cidal (direct microbial killing) [9-15]. The cutoff point between rotten and unspoiled may be seen as being below 106 CFU/g (log 6 CFU/g) [31]. The microbiological count in the current work was therefore acceptable. The ability of polyphenols to change microbial cell permeability, allowing the loss of macromolecules from the interior, or their ability to interfere with membrane function, leading to a loss of cellular integrity and eventual cell death, are two potential explanations for their antimicrobial effects [15].

Nutritional Analysis

The proximate compositions of milkfish sausage containing seaweed gel are shown in Table 4. There are 3 treatments subjected to nutritional analysis (Treatment 1, 2 and 3) which obtained the first 3 highest mean score of the general acceptability. Dry-Ashing-AAS method was performed for potassium, magnesium, zinc, and iron, Kjeldahl method for crude protein, Gravimetric for moisture and ash while Soxhlet method for crude fat.

Table 4. Proximate Composition per 500 grams of Seaweed-enriched Milkfish Sausage.

Parameters	**Seaweed-Enriched Milkfish Sausage**		
	Treatment 1	**Treatment 2**	**Treatment 3**
Potassium, %	0.13	0.08	0.17
Crude Protein, %	13.2	12.6	13.2
Moisture, %	58.6	57.5	52.1
Ash, %	3.0	3.0	2.8
Crude Fat, %	33.9	35.1	33.9
Magnesium, ppm	239	357	443
Zinc, ppm	8.1	7.9	11.2
Iron, ppm	26.0	26.4	26.4

Important variations were observed in 3 treatments, where magnesium exhibited higher results as addition of seaweed gel increases while moisture decreases as seaweed gel increases since seaweed contains less fat than pork. Related study by Wang et al., (2009), compared to a variety of seaweed inclusion, the seaweed-infused chicken sausages had less moisture than the control[38]. Additionally, the inclusion of dietary fiber—even in its dry form— reduced the samples' moisture content. Seaweed has a lower fat content than other plants, hence the treated samples' fat content considerably ($p>0.05$) decreased. Crude Protein and Crude fat for treatments 1 and 3 exhibited a higher result compared to treatment 2 since red seaweed (Rhodophyta) contains high protein [11-12] research investigated by 13.6% in U. lactuca, this outcome was better than the protein content [19-20] reported for the United States. Lactuca was 10.89% and Abirami and Kowsalya (2011) were 12.9%, although these figures were lower than those reported by Xiao-Lim et al. (2003), Ortiz et al. (2006), 37.2%, Abirami and Kowsalya (2011), 16.78–17.88%, and Abdel-Khaliq et al. (2014), 17.6%. According to Burtin (2003), compared to brown seaweeds (which range from 5% to 15%), the protein content of green and red seaweeds is often greater (ranging from 10% to 30%) [1-5]. The addition of 4% seaweed gel exhibited a higher result in Potassium and Zinc compared to the other treatments, treatment 1 and 2 for Ash and treatment 2 and 3 for Iron that exhibited a consistent result while Iron has a varying result across all 3 treatments.

CONCLUSIONS

Based on the result of the study, the following conclusions are drawn:

1. The addition of 2% seaweed gel has comparable effect with the control as to color. The addition of 8% seaweed gel has a comparable effect to treatment 2 (2%) seaweed gel as to general acceptability which make the result significant. But aroma, taste, and texture had no significant difference among treatments.
2. Addition of 8% seaweed gel is the most acceptable and had a comparable effect to treatment with addition of 2% seaweed gel.
3. Treatment 5 (32 grams) seaweed gel and control have a comparable effect on the microbial analysis of seaweed-enriched milkfish sausage in 3 days storage.

4. There are 8 components composed the proximate analysis, magnesium shows a highest result as incorporation of seaweed gel increases and moisture decreases as seaweed gel increase while the remaining components are at a varying level.

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