#### Bottled Milkfish (Chanos chanos) Enhanced with Different Seaweed Species

Remely A. Sanidad Violeta C. Funtanilla Katrina S. Sarazawa Ilocos Sur Polytechnic State College

#### ABSTRACT

The study was conducted at Ilocos Sur Polytechnic State College to determine the acceptability of bottled milkfish (Chanos chanos) enhanced with different seaweed species. This is an experimental research utilizing different treatments, namely To (control),  $T^1$  (1 tsp. pulverized Hydroclathrus clatrathus + basic ingredients),  $T^2$  (1 tsp. pulverized Euchema + basic ingredients,  $T^3(1 \text{ tsp. pulverized Gracilaria + basic})$ ingredients),  $T^3$  (1 tsp. pulverized Sargassum + basic ingredients). The sample products were assessed by 20 evaluators. Findings showed that T1 is the best among all the treatments in terms of appearance, aroma, texture, color and taste. Microbial load analysis of the products harbor below tolerable limit indicating that the products are safe for human consumption. Analysis of Variance showed that there is a significant difference among the treatments in terms of aroma, texture and taste. Proximate analysis also showed that the product contains 1.26 percent ash, 50.42 percent crude fat, 9.06 percent crude protein and 38.44 percent moisture content. Addition of powdered Hydroclathrus clathratus in the bottled milkfish in oil enhanced the sensory qualities of the formulated product. It is safe for human consumption. Bottled milkfish containing 50 percent beef and 50 percent sea cucumber added to the basic recipe have an ash content of (1.26 percent), crude fat (50.42 percent) crude protein (9.06 percent)and 38.44 percent moisture. The presence of seaweeds in the recipe is comparable in terms of return on investment. Bottled milkfish in oil enhanced with Hydroclathrus clathratus is recommended for commercialization. Researchers may further test the nutrient analysis particularly on the iodine content. Inviting more customers to buy the products, proper labelling and a training program are highly recommended.

**Keywords:** Hydroclatrhrus clatrathus, Euchema, Sargassum, Gracilaria, proximate analysis

#### INTRODUCTION

Canning as a process of preservation still remains as one of the best methods of preventing spoilage that results from microbial action. It is a process which produces a commercially sterilized product that is shelf-stable for several months. The initial bacterial load in the product is reduced to an acceptability low level depending upon the organism the process is designed to destroy (Espejo-Hermes, 2010). The processing steps using bottles are essentially similar to those using cans, but some modifications are necessitated because of the different thermal characteristics of glass and the sealing mechanism used. Glass containers are hermitically sealed by using a metal closure into which has been placed a plastisol-lining compound that acts as a sealant between the finished glass and the metal cap. Processing of food in glass containers is done by putting the containers under water. It is a counter-balanced retort. Glass containers are often chosen by home canning due to the number of advantages i.e. re-usable, easy to open and re-seal, no special sealing equipment needed, easy to inspect the quality of the product and minimize chemical reactions between the food material and the container due to its inert property. Meat, poultry and fish are low acid foods. They must be processed in a pressure canner to assure safety (Schafer, 2016).

Canning is a popular method of preserving seafood. When canned properly, seafood production is high in quality and safe to eat. It is important to pack and process seafood as directed to guarantee safety. When fish are landed they contain, in their gut and on their skin a million of bacteria which if allowed to grow and multiply will cause a rapid loss of the fresh quality and eventually results to spoilage. The purpose of canning is to use heat alone or a combination with other means of preservation, to kill or inactivate all microbial contaminants, irrespective of their source and to package the product in hermitically sealed containers so that it will be protected from recontamination. While prevention of spoilage underlies all canning operations, the thermal process also cooks the fish and in some cases leads to bone softening, changes without which canned fishery products would not develop their characteristics of sensory qualities.

In order to make the products absolutely safe, canned fish manufacturers or processors must be sure that the thermal processes given to the products are sufficient to eliminate all pathogenic spoilage micro-organisms. Of these Clostridium botulinum is undoubtedly the most notorious, for if able to reproduce inside the sealed container, it can lead to the development of a potentially lethal toxin (FAO, 2011).

The main objective of fish canning is to yield a product that may be stored for a considerable period of time, at the end in which it will be interesting and safe to eat. Fish deteriorate after death due to enzymes and bacteria. Both enzymes and bacteria can be permanently inactivated by development and provided that reinfection does not occur, heat processed food can be kept indefinitely (FAO, 2016).

Milkfish or bangus in the Philippines is the only living member of the family Chanidae. It has compressed and elongated body shape, with a symmetrical and streamlined appearance. The color of its body is olive green, with silver flanks and semi -darker fins. It has dorsal, falcate pectoral and sizable forked caudal fins.

The traditional uses of seaweeds as food and to a lesser extent, as animal feed and fertilizer supplement remain important, but in most parts of the world, these are raw materials for certain chemical products that marine algae are now chiefly valued. The special role of seaweeds is food in the far east and the prospect for a wider contribution of seaweeds to human nutrition. Seaweeds are rich source of phytochemicals having anti-oxidant and antimicrobial properties. Presence of fibers and minerals helps in improving the mineral content and reduce the salt content. The adding of seaweeds or their extracts to food products will help in reducing the utilization of chemical preservatives (Gupta and Abu-Ghannam, 2011).

Seaweed as enhancer draws out the existing flavor of any food that added to its savoury or sweetness. It acts as natural form of monosodium glutamate boosting the flavor of everyday food and adding umami or taste appeal. Even though it is not salty, it can be used to replace salt in recipes. Typically replacing added salt by at least half or completely.

Dried Gracilaria, locally known as tamsaw or kawkawayan, can be used as an ingredient for shanghai, lomi, siomai, empanada, pandesal, pastillas, pizza crust, hopia, chips and more. Apart from the nutritional value of seaweeds, it is also a source of agar, a gelatinous extract used as gelling and stabilizing agent in food. Hydroclathrus clathratus is a seaweed species complicatedly hemispherical with numerous round to oval-shaped holes of various diameters. The perforation margins roll up and inwards. The plants grow on rocks and gravel in lower intertidal zone along relatively calm shorelines. In addition to its distinctive flavor, milkfish has many benefits as a source of good nutrition for human health. According to the tests that have been carried out, as written by Deniarko, (as cited in Corpuz, 2017) the benefits of milkfish as a source of nutrition contains omega-3 than the other types of fish.

The sea- vegetables are of nutritional interest because of low calorie content but rich in vitamins, minerals and dietary fibers. Seaweeds, which have traditionally been used by the Western food industry for their polysaccharide extractive alginate, carrageenan and agar also contain compounds with potential nutritional benefits. These were approved in France for human consumption as vegetables and condiment thus opening new opportunities for the food industry (Fernandez-Martin, 2009).

This study was conceptualized to evaluate the sensory characteristics of the canned milkfish enhanced with different kinds of seaweeds (*Hydroclathrus clatrathus, Euchema, Gracilaria* and *Sargassum*). Specifically, it tried to: identify the microbial load content of the different treatment formulations of bottled milkfish using seaweeds as enhancer and determine the sensory characteristics of bottled milkfish enhanced with different kinds of seaweeds in terms of appearance, aroma, texture, color, and taste. It also determined the significant differences between and among the product formulated, identified the proximate analysis of the best product formulated and

lastly, determined the direct material cost and return of investment of the different product formulations.

# METHODOLOGY

This study used experimental research utilizing a Completely Randomized Design (CRD). Preparation of the products were done properly to avoid the intervention of the variables. Three trials were conducted to represent the replications. Five treatments were used in oil bottled milkfish as follows:

- T<sup>0</sup> basic ingredients (control)
- T<sup>1</sup>- basic ingredients + 1 tbsp *Hydroclathrus*
- T<sup>2</sup>- basic ingredients + 1 tbsp *Euchema*
- T<sup>3</sup>- basic ingredients + 1 tbsp *Gracilaria*
- T<sup>4</sup>- basic ingredients + 1 tbsp *Sargassum*

#### **Experimental Procedure**

A. Preparation of Tools and Equipment

The equipment, utensils and tools used were washed thoroughly and sanitized before the actual canning. These underwent air drying and were arranged properly according to use in the preparation table to make accessible for the researchers.

A.1. Selection of Raw Materials.

The quality and flavor of the bottled product depend largely on the selection of good quality raw material and the manner in which it is handled before canning and packing. Only fresh fish were used ready to process. Fresh milkfish were purchased to the nearby market to assure the product is safe for canning. Seaweeds used in the research were also purchased, cleaned into the running water to remove dirty particles.

A.2. Preparation of Raw Materials.

Seaweeds were dried under the heat of the sun for two days depending on weather temperature. When thoroughly dried, these seaweeds were pulverized using mortar and pestle. The fish were also eviscerated (internal organs, tails and scales were removed). Then were cut into bottle length sizes and washed.

# A.3. Soaking.

Salting of fish is carried out mainly to enhance the flavor of the final product. Salting the fish was done through brining utilizing one part salt to nine parts water for 15 minutes to remove the fishy odor.

# A.4. Draining.

Drain the fish, spread in a tray then partially dry under the heat of the sun.

A.5. Filling into Bottles.

Fish were placed into bottles manually. Uniform and accurate filling of food is required to maintain headspace. To ensure that the liquid consistency of the pack remains uniform and to maintain constant weight of the product. The standard headspace must be less than 6 percent of content volume.

# A.6. Cooking.

Heat sterilization is aimed to destroy the spores of Clostridium which are capable of producing a lethal toxin. The destruction of spores is generally accepted as the minimum standard for processing medium and low-acid canned foods. One hundred twenty one OC (1210C) as reference temperature range gives an additional safety margin to compensate for temperature-measurement inaccuracies. The product can be processed in a pressure cooker for home canning by using the heat-sterilizing equipment mentioned. Then put the fish in the glass bottle, along with the dried pulverized seaweed, iodized salt and pepper. Pour corn oil to cover all ingredients and place in the pressure cooker for 2 hours (Make sure when the pressure cooker starts a sizzling noise, turn down the heat to lower temperature and cook for 2 hours.

# A.7. Cooling.

The containers are cooled immediately after processing to avoid being overcooked. Bottled products were air-cooled by leaving them on racks to cool.

# A.8. Storage and Labelling.

Storage of canned products at high temperatures (>350C) must be avoided to prevent the growth of thermophilic spores which might survive the usual "botulinum" process. This was done by storing the products at room temperature.

Seaweeds used in the Experiment which serve as Treatments



Figure 1. Hydroclathrus



Figure 3. Sargassum sp.

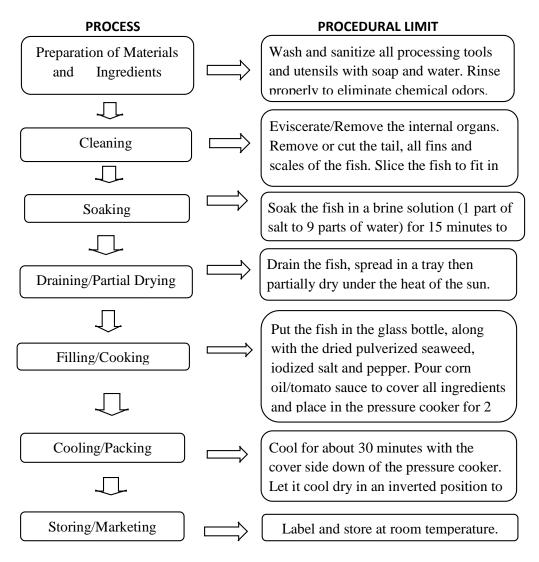


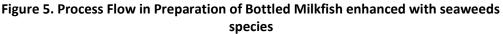
Figure 2. Euchema sp.



Figure 4. Gracilaria sp.

The seaweeds above were identified and validated by three experts of the Philippine Council for Aquatic and Marine Research Development (PCAMRD).





# **Microbial Analysis**

The formulated bottled milkfish in oil were first tested for Microbial Load analysis to determine the minimum microbial load content of the products. All treatments were sent to MMSU Microbial Laboratory, Batac, Ilocos Norte. When the product was found to have no microbes, the different products formulated proceeded in the conduct of sensory evaluation.

# **Evaluation of the Product**

Prior to the conduct of sensory evaluation, the researchers sought permission to the concerned officials in the conduct of the testing. Canning of milkfish was done thrice to represent the replication. Twenty respondents were purposively selected and were instructed to conduct sensory evaluation of the products based on the prepared score card. The score card contained the different criteria on how to rate the products using the 5-point Likert Scale and are characterized as follows:

Numerical Rating	Mean Range	Descriptive Rating
5	4.20 - 5.00	Very Much Acceptable
4	3.40- 4.19	Very Acceptable
3	2.60- 3.39	Moderately Acceptable
2	1.80- 2.59	Fairly Acceptable
1	1.0 - 1.79	Unacceptable

The best formulations were further tested for nutrient content of the product. Cost and return analysis were also computed to identify which of the formulations has the highest return of investment.

All the data gathered were treated using weighted mean, Analysis of Variance and further tested with Tukey-Kramer Test to determine the significant differences between and among treatments.

# **RESULTS AND DISCUSSION**

Microbial Load Content Analysis

To further examine the bottled milkfish, it underwent microbial load analysis and detection pathogens. Different tests of bacteria like *Escherichia coli*, *Salmonella sp., Listeria monocytogenes, Staphylococcus aureus* and molds were tested to determine the presence and absence of the microbes.

Products Formulated						
Food Product	Microbial Load (Total Plate) x10 <sup>7</sup>	Detection of Pathogens				
		Escherichia coli	Salmonella sp.	Listeria monocytogenes	Staphylococcus aureus	Molds
T0 – Control	8.0	Negative	Negative	Negative	Negative	Negative
T1- 1 tsp pulverized <i>Hydroclathrus</i> + basic ingredients	9.0	Negative	Negative	Negative	Negative	Negative
T2- 1 tsp pulverized <i>Euchema</i> + basic ingredients	10.0	Negative	Negative	Negative	Negative	Negative
T3- 1 tsp pulverized <i>Gracilaria</i> + basic ingredients	9.0	Negative	Negative	Negative	Negative	Negative
T4- 1 tsp pulverized <i>Sargassum</i> + basic ingredients	10.0	Negative	Negative	Negative	Negative	Negative

# Table 1 Microbial Load Analysis and Detection of Pathogens of the Different Products Formulated

The result of the analysis showed that the products have negative results which means that no pathogens were detected and these harbour microbial load below tolerable limit. This signifies then that the products are safe for human consumption and ready for evaluation purposes.

# Table 2 Sensory Evaluation of the Bottled Milkfish enhanced with different Seaweeds Species

Species					
Parameter	Experimental Lot	Mean	Descriptive Rating		
Appearance	T0- Control	4.30	VMA		
	T1- Hydroclathrus	4.35	VMA		
	T2-Euchema	4.29	VMA		
	T3-Gracilaria	4.35	VMA		
	T4-Sargassum	4.07	VA		
Aroma	T0- Control	4.07	VA		
	T1- Hydroclathrus	4.35	VMA		
	T2-Euchema	4.19	VA		
	T3- Gracilaria	4.22	VMA		
	T4-Sargassum	3.55	VA		
Texture	T0- Control	4.23	VMA		
	T1- Hydroclathrus	4.48	VMA		
	T2-Euchema	4.12	VA		
	T3-Gracilaria	4.23	VMA		
	T4-Sargassum	4.03	VA		

Parameter	Experimental Lot	Mean	Descriptive Rating	
Color	T0- Control	4.38	VMA	
	T1- Hydroclathrus	4.27	VMA	
	T2-Euchema	4.10	VA	
	T3-Gracilaria	4.20	VMA	
	T4-Sargassum	3.93	VA	
Taste	T0- Control	4.68	VMA	
	T1- Hydroclathrus	4.29	VMA	
	T2-Euchema	4.18	VA	
	T3-Gracilaria	4.18	VA	
	T4-Sargassum	3.78	VA	
egend:	Mean Range	Description		
	4.20 - 5.00	Very Much A	Very Much Acceptable (VMA)	
	3.40- 4.19	Very Acceptable (VA)		
	2.60- 3.39	Moderately Acceptable (VA)		
	1.80- 2.59	Fairly Accept	able (FA)	
	1.0 - 1.79	Unacceptable	e (U)	

# Appearance

The sensory characteristics of bottled milkfish along appearance showed that addition of *Hydroclathrus clathratus* and *Gracilaria* registered the same result with a mean rating of 4.35 respectively with a descriptive rating of **Very Much Acceptable** while the presence of *Sargassum* in the canned product appeared **Very Acceptable** Aroma.

In terms of aroma,  $T^1$  and  $T^3$  have the highest mean rating of 4. 35 and 4.22 respectively and described as **Very Much Acceptable** while  $T^0$  and  $T^2$  and  $T^4$  described as **Very Acceptable**. This indicates that the presence of *Hydrochathrus clathratus* and *Gracilaria* in the recipe enhanced the aroma of the bottled milkfish.

# Texture

The presence of Hydroclathrus clathratus in the recipe obtained the highest mean rating (4.48) described as **Very Much Acceptable** followed by presence of *Gracilaria* and the control. This further proves that the presence of powdered *Hydroclathrus clathratus* and *Gracilaria* further enhanced the smoothness of the finished products as compared to the presence of *Sargassum* and *Euchema* in the recipe.

# Taste

In terms of taste, the control  $(T^0)$  garnered the highest mean rating of 4.68 followed by *Hydroclathrus clathratus*  $(T^1)$  which was described as **Very Much Acceptable**. The rest of the treatments appeared **Very Acceptable**. This further elaborates that the presence of seaweeds affects the tasting quality of the products

to the panel of evaluators. This could be due to the presence of gelatinous substances in the seaweeds when further cooked in a pressurized temperature.

Analysis of Variance (ANOVA) on the Significant Differences of Bottled Milkfish Enhanced with Different Kinds of Seaweeds

Table 3
Analysis of Variance on the Significant Different Differences between and Among
Treatments

Appearance					
Source	df	SS	MS	f-value	Pr>F
Model	6	0.70382667	0.11730444	2.95 NS	0.0796
Error	8	0.31781333	0.03972667		
Total	14	1.02164000			
Aroma					
Model	6	2.30936000	0.38489333	8.86*	0.0035
Error	8	0.34741333	0.04342667		
Total	14	2.65677333			
Texture					
Model	6	0.72564000	0.12094000	10.44*	0.0020
Error	8	0.09265333	0.01158167		
Total	14	0.81829333			
Color					
Model	6	1.43054667	0.23842444	6.04 NS	0.0517
Error	8	0.31561333	0.03945167		
Total	14	1.74616000			
Taste					
Model	6	1.72881333	0.28813556	11.91*	0.0013
Error	8	0.19356000	0.02419500		
Total	14	1.92237333			

\*Significant at .01 level

Based on the result, it was found out that there are significant differences in the use of seaweeds as enhancer in terms of aroma, texture and taste while no significant differences appeared in terms of appearance and color. When tested further to Tukey Kramer Test,  $T^1$  (*Hydroclathrus clathratus*) compared to  $T^3$  (*Gracilaria*) or an addition of Sargassum, a significant difference was observed. The differences was manifested by the aroma, texture and taste of the products. This further implied that addition of *Hydroclathrus clathratus* in the bottled milkfish in oil enhanced the aroma, texture and taste of the product. This was manifested by the findings of Wang (2010) that Hydroclathrus clathratus are a good source of marine compounds with potential applications in food and medicine.

Proximate Analysis of the Most Acceptable Products					
Analysis Name Result (Milkfish in oil)					
Ash	1.26 %				
Crude Fat	50.42 %				
Crude Protein (N 6.25)	9.06 %				
Moisture	38.44 %				

Table 4 Proximate Analysis of the Most Acceptable Products

The table shows the result of the nutrient analysis of the bottled milkfish enhanced with *Hydroclathrus clathratus*.

Based on the conducted proximate analysis, it shows that  $T^1$  (*Hydroclathrus clathratus*) of bottled milkfish in oil enhanced with seaweeds has 1.26 percent of ash, 50.42 percent crude fat, 9.06 percent crude protein and 38.44 percent of moisture. The crude fat which is 50.42 percent has the highest nutrient content while ash which is 1.26 percent has the lowest content. This further indicates that the presence of large amount of oil serves as preservative and the process is done through milkfish in oil.

 Table 5

 Direct Material Cost and Return on Investment of Bottled Milkfish (Chanos chanos)

 Enhanced with Different Seaweed Species

Enhanced with Different Seaweed Species					
Particulars	Control (T <sup>o</sup> )	Hydroclathrus (T¹)	Euchema (T²)	Gracilaria (T³)	Sargassum (T <sup>4</sup> )
Sales	4,000.00	4,080.00	4,080.00	4.080.00	4,080.00
No. of bottle	80.00	80.00	80.00	80.00	80.00
Price	50.00	51.00	51.00	51.00	51.00
Expenses (Php)	1,200.00	1,200.00	1,200.00	1,200.00	1,200.00
Milkfish	-	1,200.00	1,200.00	1,200.00	1,200.00
Seaweeds		5.00	5.00	5.00	5.00
Corn oil	400.00	400.00	400.00	400.00	400.00
Iodized Salt	10.00	10.00	10.00	10.00	10.00
Black pepper	10.00	10.00	10.00	10.00	10.00
bottle	1,200.00	1,200.00	1,200.00	1,200.00	1,200.00
Labor	250.00	250.00	250.00	250.00	250.00
Gasul	100.00	100.00	100.00	100.00	100.00
Total Expenses	3,170.00	3,175.00	3,175.00	3,175.00	3,175.00
Net income	830.00	905.00	905.00	905.00	905.00
ROI	26.18%	28.50%	28.50%	28.50%	28.50%

Based on the ROI computation, it is found out that addition of different species of seaweeds obtained higher percentage (28.50 percent) as compared to the control recipe (26.18 percent). Though the price of the product increases by P1.00 because of the inclusion of seaweeds in the recipe, the advantages can be seen on the nutrient derived from seaweeds as sources of iodine and medicine.



Figure 6. The finished product that underwent different analysis

#### CONCLUSIONS

The formulated products are safe for human consumption, no pathogens were detected. Bottled milkfish containing 50 percent beef and 50 percent sea cucumber added to the basic recipe have an ash content of 1.26 percent, crude fat (50.42 percent) crude protein (9.06 percent) and 38.44 percent moisture. There are significant differences in the use of seaweeds as enhancer in terms of aroma, texture and taste while no significant differences appeared in terms of appearance and color. Addition of *Hydroclathrus clathratus* in the bottled milkfish in oil enhanced the aroma, texture and taste of the product. The presence of *Hydroclathrus clathratus*, *Sargassum* and *Gracilaria* in the recipe are comparable in terms of return on investment.

#### RECOMMENDATIONS

The researchers recommend that another study be conducted utilizing other species of seaweeds. There should be further testing on the nutrient analysis particularly on the iodine content. Bottled milkfish in oil enhanced with *Hydroclathrus clathratus* is recommended for commercialization. To invite more customers to buy the products, proper labelling is recommended. Lastly, a training program is highly recommended to further disseminate the result to consumers for them to appreciate the nutrient present in the canned products and an avenue for commercialization of the finished products.

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