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Original Article

Quality Evaluation of Clarias Catfish Fermented Sausage Manufactured by Pediococcus acidilactici 0110<TAT-1 Starter Culture at Different Level of NaCl

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ABSTRACT

This study was aimed to obtain the best level of NaCl concentration with emphasis on the physico-chemical of Clarias fermented sausage. Three different Clarias catfish fermented sausage manufactures (1, 2, and 3% NaCl added) were used to obtain the quality along storage for 28 days. The physiochemical characters of sausage were affected by NaCl addition, NaCl content, water activity, pH, weight loss, water content, water holding capacity; and the higher of protein and fat content. While microbiology was affected by aerobic plate count, total pathogen bacteria, and lactic acid bacteria. Total pathogen bacteria was effectively inhibited on sausage with addition of 1 % NaCl after 7 days storage, however sausage with addition of 2 and 3 % NaCl was effectively inhibited before 7 days. Sensory evaluation indicated that of 2 % NaCl was acceptable to all parameters.
Keywords: Clarias catfish, Fermented Sausage, lactic Acid Bacteria, Sensory Evaluation, Quality.

INTRODUCTION

Fish product was high in protein and low of fat content makes its commodity to be exploited by diversification as an alternative of energy intake against the risk of degenerative diseases [1]. Sausage as a product, where fresh fish meat mixed with some additives, and then put in casings and processed by heating [2], many factors such as aw, temperature, redox potential, low pH, NaCl or nitrite could inhibit the growth of pathogenic organisms and bacterial spoilage [3]. Lactic acid bacteria are highly beneficial in inhibiting the growth of pathogenic bacterial flora. Addition of lactic acid produced has antibacterial activity against several floras [4].

The NaCl used, as well as preservation of the processing also maintain the stability of micro-organisms during fermentation, it is also able to reduce the discharge of AW participated in the fish so that the product becomes more dry. The use of curing include: salt, sugar solution, were affected on the concentration of the soluble protein substances and water absorption [5]. The salt added to the smoked product was repair the flavor and soften fish tissues. NaCl is more effective, safer, and cheaper application in fishery products [5]. NaCl added was effect on the activity of bacteria [6]. Salt can be denaturation of the protein surface, and the dry product, denatured proteins that form a layer on the skin that resembles the surface of the product, thus protecting the inside of the fish and keep the smell of smoke in the fish [5].

Fermented sausage processing was done to the meat of livestock and poultry. As for the fishery products some have done, such as the fish Jangiles [6]; Mullet [7]; Sardine [8]; Carp [1]; catfish hybrid [2]. Of the research report turned out mostly using formulas beef cattle, regardless NaCl levels and the most appropriate fat, especially the quality of sensory and physico-chemical.

This study was aimed to obtain the best level of NaCl with emphasis on Clarias fermented sausage supported by the physico-chemical and microbiological.

MATERIALS AND METHODS

Bacterial culture

Pediococcus acidilactici 0110<TAT-1 was obtained from IUC (Inter University Centre), Gajahmada University, Indonesia. This bacteria was cultured in MRS broth (Oxoid) at 30-32 °C. Bacterial cells was harvested

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after 24 hours of incubation, and dissolved in 0.1% sterile peptone to obtain the desired density using a spectrophotometer.

Sausage preparation

Clarias catfish (Clarias sp) fresh was obtained from Batu breeders, East Java, beheaded, skinned and fileted, then chopped using a blender (Phillip), and preblending for 24 hours. Formula sausage to 1000 grams of catfish were: NaCl (appropriate treatment), 0.2 g sodium nitrate, sodium nitrite 0.1 g, 4 g sucrose, glucose 3 g, 3 g fructose, 1 g of white pepper, black pepper 1 g, 0.7 g galangal, 0.7 g ginger, cinnamon 0.6 g, 0.5 g garlic, and cloves 0.5 g. Recipes are drawn based [7], with the modification process by [9]. All these ingredients are mixed with catfish chopped, and then add the starter culture of P. acidilactici 108 cfu / ml, 2 ml for 500 g of meat, the next batter put collagen casing 2 cm of diameter and 10 cm of long, pre-incubation, fumigation, and incubated at commercial temperature (15-22 º C) for 28 days. Experimental variables were: 1, 2, and 3% of NaCl level.

Physico Chemical Analysis

Water activity (aw) was measured using Thermoconstanter TH-200. The pH was measured by a digital PHM 210. WHC based on the ratio of the water that comes out with the initial moisture content of sausages [10]. Losses based on the percentage ratio of the final weight to initial weight [11]. A level of protein, fat, and water refers to the AOAC [12].

Microbiological Analysis

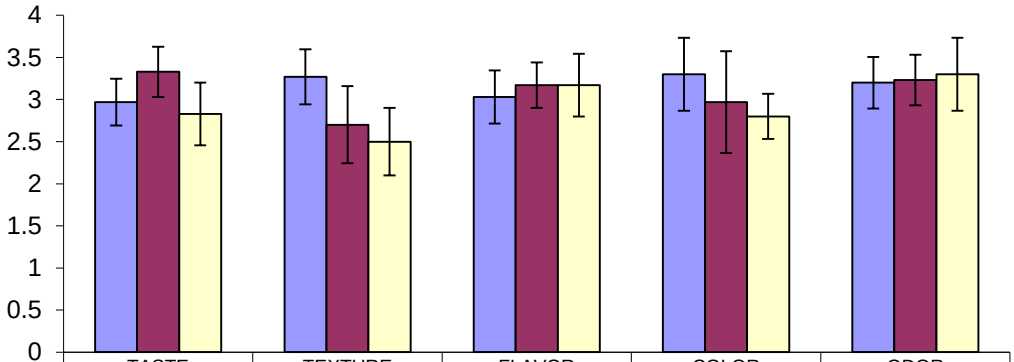
Sausage samples (10 g) were homoginated for 2 minutes in 90 ml of 0.1% peptone sterile by Stomacher. Homogenate serially diluted in 0.1% sterile peptone and used to calculate the APC on Plate Count Agar (PCA, Oxoid) with dilution method. Lactic acid bacteria (catalase-negative and Gram-positive colonies) were analyzed on MRS agar (Oxoid). Enterococcus bacteria on the VRBD media (Oxoid) [13]. All of that plate was incubated aerobically. MRSA and PCA were incubated at 30 ° C for 24-48 hours. The other plate was incubated at 37 ° C for 48 hours.

Data Analysis

The data were analyzed descriptively based on the standard deviation (SD) among the three independent variables in the experiment, using Microsoft Excel v.3.0. To determine the best treatment are using effective index by DeGarmo [14].

RESULTS and DISCUSSION

In this experiment were emphasized on the sensory quality of sausages at the end of the maturation period (28 days), were aims to obtain the most preferred level of salt by the panelists. The histogram and the mean results of sensory analysis (ISO: 8589, 1988) is given in Fig. 1; WHC values, shrinkage weight, protein content, fat, water in Table 1. Data Aw. PH, APC, total BAL, and bacterial pathogens (not shown).



	TASTE	TEXTURE	FLAVOR	COLOR	ODOR
1 %	2.97	3.27	3.03	3.3	3.2
2 %	3.33	2.7	3.17	2.97	3.23
3 %	2.83	2.5	3.17	2.8	3.3

Figure 1. Histogram of sensory evaluation of sausage at level of NaCl, analyzed after 28 days incubation.

The data and histogram in Fig. 1 were showed that the sausage at the 2 % of NaCl level had the highest sense of the value (most preferred), and the lowest at 3% level (least preferred) by the panelists, while the best treatment for all parameters at the end of found that 2% salt level had the highest value of affectivity index, i.e. 0.613. This condition is due to the salt level is too high causing the sausage to be salty, while too low was increasingly the felt bland, or depending on where the product is marketed. Good and bad taste of a product depends on the taste buds of a dominant society in which the product is distributed, the weather or the climate of an area, eating habits, as well as cultural and aesthetic factors. In the United States, to reduce the overly salty taste is usually improved with the addition of glucose or other additives that are allowed by the USDA-FSIS.

The analysis of flavor in Fig. 1 are indicating that the lower of the salt levels get low product value; higher levels of NaCl got meat matrix more compact, and the air cavity formed are smaller and decreases. This situation is due to the flavor which is a complex sensory reaction to describe the taste, smell (odor), and the texture of a product is not detected by the panelists, who allegedly because was covered by the smell of smoke. Odor were more sensitive senses of smell to estimate revenues generated component, usually divided into oxidation of fatty acids which include alkynes, aldehydes, cetones and carboxylic acid groups [15]. Sausage flavor detected by the panelists was dominated by the smell of spices and smoke, odor or flavor result is the result of bacterial metabolism of the most important determinants of flavor sausages are also not detected

The lower level of salt causes the color value or the sausage lightest (Fig. 1). Products with 1% NaCl had the highest average color value, amounting to 3.30 (the brightest), and 3% NaCl are lowest (darkest), which is 2.80 of value. This is due to the color which is the sensory properties affected when NaCl is reduced. Reduction of salt in sausage products can reduce the appearance of a brighter color to 50% on product. Decreased color on the processed fish was reported by Birkeland and Bjerkeng [16], are caused by associated carytenoids with higher tissue protein extracted into astaxanthin with higher salt solution used. In the case of salmon with a 1-6 M NaCl visible loss of color along with increased astaxanthin extracted at pH 7.

Table 1. Experiments of NaCl levels on the physico-chemical characters of fish fermented sausage using Pediococcus acidilactici starter culture during incubation

Parameter	Level of NaCl (% weight)	Incubation (days)			
		0	7	14	28
WHC (%)	1	28,50±0,90	58,23±0,71	27,85±0,71	27,84±0,79
	2	27,85±0,73	27,54±0,77	27,21±0,69	27,53±0,76
	3	27,83±0,76	27,42±0,82	26,63±0,88	26,73±0,69
Weight loss (%)	1	17,49±0,45	17,61±0,52	17,75±0,41	17,85±0,81
	2	16,80±0,43	17,39±0,37	17,59±0,42	17,65±0,48
	3	16,54±0,60	17,19±0,54	17,38±0,44	17,89±0,63
Protein content (% wb)	1	16,03±0,50	16,34±0,47	16,76±0,39	16,79±0,43
	2	16,37±0,58	16,89±0,58	17,10±0,56	17,18±0,60
	3	16,77±0,65	16,93±0,54	17,51±0,72	17,38±0,42
Fat content (% wb)	1	5,56±0,13	5,30±0,11	5,20±0,16	5,24±0,16
	2	5,67±0,21	5,38±0,18	5,29±0,13	5,31±0,15
	3	6,81±0,21	6,58±0,27	6,52±0,22	6,51±0,16
Water content (%)	1	70,01±1,89	69,43±1,49	68,62±1,49	68,61±1,65
	2	68,62±1,53	67,98±1,63	67,29±1,47	67,95±1,60
	3	68,59±1,61	67,72±1,72	66,05±1,86	66,28±1,45

Note: Data had shown from the average of three independent variables experiments, with SD.

The value of weight loss, protein content, water content (Table 1), aw, and pH were not different at each observation during incubation, but differently to the APC, LAB, and total count of bacterial pathogens (data not shown). The content of pathogenic bacteria do not grow on sausages 14 and 28 days of incubation due the value of AW from three products is quite low (<0.88). Some strains of Listeria monocytogenes is able to grow at aw below 0.9 and pH below 4.4, and there is a relationship between aw, NaCl, lactic acid and L. Monocytogenes growth [17].

Increased protein levels is due to decreased water and fat content of sausages, and the highest get in product 3% salt, because salt soluble proteins that play a role in the strength of the emulsion extracted were more than 2 and 3%. This condition indicates that the emulsion product becomes more unstable with the decline of salt sausage; due to the decline in the NaCl content of the product affects the binding properties of the processed meat tissue. Increasing concentrations above 0.6 M NaCl, the bound water would enhance electrostatic rejection until myofibrillar structures were destroyed. This effect is very useful in the improvement in the manufacture of sausage emulsions, because the salt will issue myofibrillar protein bound in water, so it can act as an emulsifier for the fat particles [18].

Reduction of salt causes the extracted proteins becomes less and inhibits protein denaturation by heat, mainly to the protein network which affects the binding characteristics of processed meat products [19], states that the higher salt levels cause an increase in the ionic strength of the product and increase the extracted proteins, resulting in increased interaction between the polypeptide chains during the process of fermentation, so the gel matrix formed into a more stable, which in emulsion stability becomes increasingly.

Protein networks are often classified as sarcoplasmic proteins and myofibrillar proteins (myosin, actin and actomyosin), and the connective tissue or stroma proteins (collagen). Sarkoplasmic protein is soluble in water or saline solution, which myofibrillar proteins are soluble salt concentrations >0.3 M. Myosin and actomyosin are

requiring high salt concentrations for extraction [20]. In addition there are reports indicating that myofibrillar proteins are protein water-soluble [18].

Reduction in salt dough is consequences for the stability of the product, because the extraction of salt-soluble proteins was reduced [21]. Reduction of up to 0.58% salt had wider capillaries compared to 2.82%, with successive ripening shrinkage 56-35%. Higher ionic strength was reported increase the electrostatic repulsion and causes myofibrillar structural changes shown by the emergence of a more susceptible to discharge more water [17].

Table 2. Experiments of NaCl levels on the microbiology characters of fish fermented sausage using *Pediococcus acidilactici* starter culture during incubation

Parameter	Level of NaCl (% weight)	Incubation (days)			
		0	7	14	28
APC (log)	1	6,24±0,11	6,96±0,10	7,12±0,15	7,68±0,25
	2	6,23±0,18	6,78±0,30	6,95±0,24	7,60±0,21
	3	6,23±0,23	7,27±0,26	7,59±0,23	6,77±0,24
LAB (%)	1	4,74±0,12	5,43±0,16	5,21±0,14	5,24±0,14
	2	4,71±0,18	5,32±0,14	5,39±0,13	5,38±0,18
	3	5,45±0,18	5,40±0,13	5,46±0,14	5,45±0,11
Enterobac teriaceae (log)	1	4,25±0,04	2,20±0,17	*	*
	2	3,80±0,03	*	*	*
	3	3,78±0,05	*	*	*

Note: Data had shown from the average of three independent variables experiments, with SD. *). No detected bacteria.

Based on the Table 2, the content of LAB did not differ in incubation 7, 14, and 28 days. APC at 14 and 28 days of incubation is different, while the total content of a pathogen bacterial was not found from incubation 7 to 28 days at level 2 and 3% salt. For 14 and 28 days all salt levels found pathogenic bacteria colonies. This gives an indication that the fermentation process at that time was able to eliminate pathogenic bacteria until there are no colonies at all.

From the above observations give a clear picture that the solubility and degradation of proteins affects the sensory character of the final product, so the levels of saltiness products are not only influenced by the presence of NaCl alone.

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