# DEVELOPMENT OF MICROBIAL POPULATION DURING THE FERMENTATION EVIRONMENTAL CONTROL OF MACKEREL (*Rastrelliger sp.*)

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#### **ABSTRACT**

Salt fermentation can control the growth of pathogenic microbes in fish. In this study, several types of salt (refined salt, brick salt, and krosok salt) were used to control the growth of pathogenic microbes in mackerel (*Rastrelliger sp.*). The effect of controlled fermentation environment and appropriate type of salt was determined by measuring bacterial growth, salt content, moisture content, and acidity (pH) during the fermentation period. At the end of the observation, it was noted that mackerel fish contained total of 7.2×10<sup>7</sup> - 7.9×10<sup>7</sup> CFU/g microbes, salt content of 14.2-14.3%, moisture content of 55.5-53.5%, and pH of 5.3-5.5. The results of environmental control are able to support the fermentation microbial population in the active phase (log phase), induce the microbe ready to enter the fermentation phase for aroma and flavor formation. However, the different types of salt do not affect the salt content, air content, total microbes, and acidity (pH) of mackerel during the fermentation process. The information obtained expected to be used as a guide to determine the length of time to control the fermentation environment.

Keywords: Time of Fermentation Control, Salt, Microbial Population, Mackerel Fish

# PERKEMBANGAN POPULASI MIKROBA SELAMA PENGENDALIAN LINGKUNGAN FERMENTASI IKAN KEMBUNG (Rastrelliger sp.)

## **ABSTRAK**

Fermentasi menggunakan garam dapat mengendalikan pertumbuhan mikroba pembusuk pada ikan. Dalam penelitian ini beberapa jenis garam (garam halus, garam bata, dan garam krosok) digunakan untuk dapat mengendalikan pertumbuhan mikroba pembusuk pada ikan kembung (*Rastrelliger sp.*). Jenis garam yang sesuai ditentukan dengan mengevaluasi pertumbuhan bakteri serta mengukur kadar garam, kadar air, dan kadar keasaman (pH) selama masa fermentasi ikan. Pada akhir pengamatan tercatat bahwa ikan kembung mengandung total mikroba sebanyak 7.2×10<sup>7</sup> - 7.9×10<sup>7</sup> CFU/g, kadar garam sebanyak 14.2-14.3%, kadar air sebanyak 55.5-53.5%, dan pH 5.3-5.5. Hasil penelitian menunjukkan bahwa pengendalian lingkungan mampu mendukung populasi mikroba fermentasi dalam fase aktif (*log phase*), sehingga siap memasuki phase fermentasi untuk pembentukan aroma dan citarasa. Namun, dari hasil juga dapat diamati bahwa perbedaan jenis garam tidak berpengaruh terhadap kadar garam, kandungan air, total mikroba, serta kadar keasaman (pH) dari ikan kembung selama proses fermentasi. Informasi yang diperoleh diharapkan dapat digunakan sebagai pedoman untuk menentukan lama pengendalian lingkungan fermentasi.

Keywords: Waktu pengendalian fermentasi, Garam, Populasi mikroba, Ikan kembung

#### **INTRODUCTION**

Mackerel (Rastrelliger sp.) is one of the most widely consumed of fishery commodities in Indonesia (Aminah 2016; Siswanti, Agnesia dan Baskara, 2016). Naturally, fresh fish contains microbes known as *micro-floras* which can be beneficial or harmful. Without controlling, spoilage microbes will growth better than other microbes. The process for controlling spoilage microbe are needed before the

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fish consumed by human. Processing is one of way to obstruct the spoilage microbe growth and prolong the fish shelf-life. One of the processes is fermentation, this process was used fermentation microbes to obstruct or stop the spoilage process growth. In Indonesia, Peda Kembung is one of traditional fermentation product which uses lactic acid bacteria in the processing (Fajri, Sukarso, & Rasmi 2014).

Controlling the fermentation environment is intended to create environmental conditions where spoilage microbes can not live, while fermentation microbes grow and develop well. Salt is one of the ingredients that widely used to control the fermentation process in various types of fishery products such as fish sauce (Gao, Li, Xia, Xu, dan Liu, 2020) and Jambal Roti salted fish (Karyantina, Anggrahini, Utami dan Rahayu 2020). In the manufacturing process, fermentation microbes play a major role in the order to process of the typical flavors produced (Das, Kumar, dan Nayak 2020). The development of the fermentation microbial population is one of the success indicators of the fermentation process. Therefore, environmental conditions during the fermentation process are important to be studied to evaluating the growth of fermentation microbes.

This study aim was to evaluate the growth of fermentation microbes during the environment controlling process. Several types of salt (refined salt, brick salt, and krosok salt) were used to control the growth of spoilage microbes in mackerel (*Rastrelliger sp.*) The effect of controlled fermentation environment and appropriate type of salt was determined by evaluating bacterial growth also measuring the salt content, moisture content, and acidity (pH) during the fermentation period.

#### **METHODOLOGY**

#### **Materials**

Mackerel (Rastrelliger sp.) weighing  $120 \pm 0.04$  g and three types of salt (refined salt, brick salt, and krosok salt) used for making Peda Kembung were obtained from Caringin Central Market, Bandung, Indonesia. The mackerel fish are transported using a cool box that has been given shaved ice, to maintain its freshness until its ready to processed.

## Environmental control process

The environmental control process was carried out using fine salt, mashed brick salt, and krosok salt. Fine salt is used as an environmental controller because bacteria are difficult to grow in a salt-rich environment. The process of salting was done by dry salting method with a concentration of 25% of the fish weight. The salting process was done by evenly smearing salt throughout the fish body. The fish then put into an airtight jar and then stored at room temperature for 7 days.

# Calculation of total lactic acid bacteria, salt content, moisture content and pH.

Total Lactic Acid bacteria was calculated based on the number of living microbial populations (Indonesian National Standard (SNI) 7545.1, 2009). The determination of the salt content was carried out by a filtration process using the Mohr method (AOAC, 1971) with standard solutions of AgNO3 and K2CHO4 as indicators. Moisture content analysis was performed using an oven method (AOAC, 2000). Acidity (pH) measurements were carried out using a pH meter (Hanna series H198107).

# Data analysis

The study was designed using a completely randomized design with three types of treatment and three replications. The calculated data were then analyzed using ANOVA with a significant level of 0.05 using the SPSS version 24.0 application (SPSS Inc., Chicago, IL, USA).

# RESULT AND DISCUSSION

#### **Total Bacteria**

The population of lactic acid bacteria tends to increase with the addition of salt and the fermentation environment. At the second day of environmental control, the growth of fermented microbes had not been seen. At this stage the fermented microbes are still adapting to their new environment. Entering the third day, the growth of fermented microbes tends to increase with the

fermentation days. Based on this curve, the fermentation microbial population still tends to increase because it is in the growth phase (log phase). The type of salt used in fermentation control has no effect on the fermentation microbial population. The iodine content in refined salt affects the growth of fermented microbes, while the natural flora contained in krosok salt is able to stimulate the growth of fermented microbes (Amalia, Dwiyanti & Haitami, 2016).

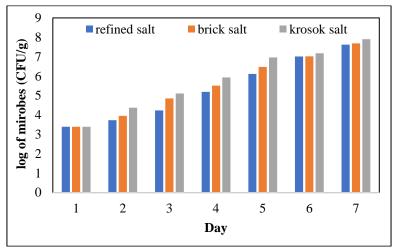


Figure 1. Log of fermented microbial population in mackerel with salt type treatment during the environmental control process.

#### Salt content

The salt content in fish meat increased significantly on the second day (Figure 2). The type of salt did not give a significant effect (p>0.05) on the salt content that entered the fish meat. The salt content ranged from 14.2 to 14.3% at the end of the observation. During the controlling process, the fluid that leaves the fish's body will liquefy the salt crystals. Salt crystals will form Na<sup>+</sup> and Cl<sup>-</sup> ions and enter the tissue of fish flesh. During the process of fluids from the fish's body, these ions will seep into the fish meat. Apart from being antimicrobial, these ions will cause a salty aroma and taste (Istania, Arpi, & Hasanuddin, 2019).

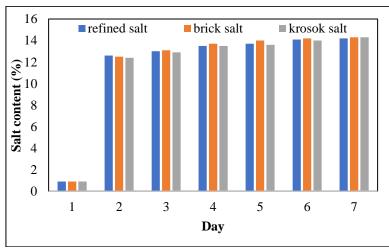


Figure 2. Salt content of mackerel with salt type treatment during the environmental control process.

#### Moisture content

The moisture content of the fermentation control fish meat has decreased from 73.5% to 53.5% (Figure 3). Changes in moisture content during the fermentation control process in all types of treatment were not significantly different (p>0.05). In general, the addition of salt can reduce the moisture content of fish meat (al-Rubai, Hassan, & Eskandder., 2020).

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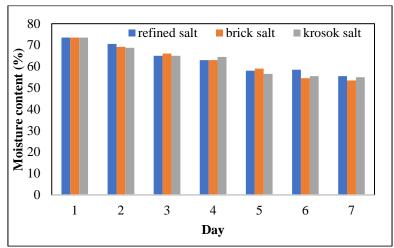


Figure 3. The change of moisture content of mackerel with salt type treatment during the environmental control process.

# Degree of acidity (pH)

The type of salt had no significant (p>0.05) effect on the acidity (pH) of fish meat (Figure 4). The degree of acidity is related to the overhaul of carbohydrate compounds into lactic acid compounds (Aristawati & Hasanuddin, 2016) which have an acidic pH value. At the end of the observation, the pH value of the fish meat was recorded between 5.3 and 5.5. The lowest pH value produced in mackerel with the addition of krosok salt. Krosok salt has not undergone further treatments it still contains natural micro-flora including fermented microbes. These microbes will transform carbohydrate compounds into lactic acid compounds (Desniar, Poernome, & Wijartur, 2009). The more lactic acid compounds formed, the more pH value will decrease.

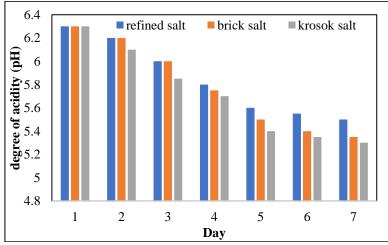


Figure 3. The change of degree of acidity (pH) of mackerel with salt type treatment during the environmental control process.

## Discussion

Control of the fermentation environment was aim to grow fermented microbes and simultaneously inhibit spoilage and pathogens. During the control of the fermentation environment, it can be seen that the growth of fermented microbes continued to show an increasing pattern (Figure 3). This is an indication that environmental conditions are in accordance with the needs of fermented microbes (Gassem, 2019). Another thing that can be observed is the average number of growth bacteria has decreased on days 5, 6, and 7. This happens because lactic acid bacteria may experience plasmolysis due to increased osmotic pressure when more salt permeates the fish's body (Hadiyanti & Wikandari , 2013). Even so, the number of fermented microbes has reached an adequate number and is in the active phase (log phase), induce the microbe ready to enter the fermentation phase for aroma and flavor formation. Study by Zang et al. (2020) show that lactic acid bacteria was shows a positive correlation with flavor and indicated to changes the organic acids during the fermentation process

The increased of salt content in fish (Figure 2) during the observation period also supported the fermentation process. The entry of Na<sup>+</sup> and Cl<sup>-</sup> ions into the fish meat will inhibit the activity and growth of spoilage microbes, but supports fermentation microbial life. This is because the Na<sup>+</sup> and Cl<sup>-</sup> ions in salt are toxic to spoilage microbes (Prasetyo et al., 2012).

Other indicators that can be used to show the growth of fermented microbes are moisture content and pH value. A decrease in moisture content (Figure 3) in fish meat can cause microbial activity to be inhibited due to lack of moisture (Aristyan et al., 2014). The moisture discharge from the fish meat causes free water to decrease and inhibit the microbe growth. Fermentation microbes will break down the carbohydrate compounds into simpler compounds (Arfianty, Farisi, & Ekowati, 2017). The compound is lactic acid which is acidic (Fajri, Sokarso, & Rasmi, 2014). The presence of lactic acid compounds will reduce the pH of fish meat. In the process of environmental control, more and more lactic acid compounds are formed causing the pH continues to decline (Figure 4).

From the overall observed data, the difference in salt types did not show a significant difference (p>0.05). However, the salt and fermentation time carried out still support the fermentation process that occurs. The same thing was found in the study of Anto et al. (2019), the results showed that the duration of fermentation had a significant effect (p<0.05) on protein, pH, water content, total lactic acid batteries and microbes after the 3rd day of fermentation.

#### **CONCLUSIONS**

This study aims to evaluate the controlled environment by using salt in the fermentation process of mackerel (Rastrelliger sp.). At the end of the observation, it was noted that mackerel fish contained a total of 7.2×10<sup>7</sup>-7.9×10<sup>7</sup> CFU/g of microbes, salt content of 14.2-14.3%, moisture content of 55.5-53.5%, and pH of 5.3-5.5. The results of environmental control are able to support the fermentation microbial population in the active phase (log phase), induce the microbe ready to enter the fermentation phase for aroma and flavor formation. However, from the results observed shows that the different types of salt did not affect the salt content, moisture content, total microbes, and acidity (pH) of mackerel during the fermentation process. Further research can be carried out to see the effect of fermentation using salt on the aroma and taste of Peda Kembung.

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