

Sodium content in fermented shrimp paste with varying packaging materials

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Abstract

Shrimp and fish paste are fermented products with high amounts of added salt. Sodium (Na^+) ions in salt are growth factors for halophilic lactic acid bacteria (LAB), essential in shrimp paste. However, an overabundance of Na^+ negatively affects the human body and may increase blood pressure and the risk of heart attacks, strokes, kidney damage, and other health issues. Commercial shrimp paste can be found in various packaging materials. The permeability of the packaging material affects air availability during fermentation, which affects LAB growth and sodium levels. This study aimed to determine the effect different packaging materials have on rebon (*Acetes* sp.) shrimp paste sodium levels, precisely that of teak leaf, paper, and plastic. A completely randomized design and an experimental laboratory were used for the experiments. The data were analyzed using ANOVA and Honest Significant Differences tests. The different packaging materials significantly affected ($P < 5\%$) the acidity, sodium levels, and total plate count (TPC) of LAB but did not affect the water activity or salt content. Shrimp paste consisted of $1.32 \pm 0.12\%$ sodium when packaged in teak leaves, $1.51 \pm 0.08\%$ in paper packaging, and $1.69 \pm 0.07\%$ in plastic packaging. Teak leaves packaging resulted in shrimp paste with the lowest sodium content because of its many pores and high gas permeability, thus supporting the growth of microbes that use Na^+ .

1. Introduction

Preserved shrimp paste, known as *terasi* in Indonesia, is made by fermenting, drying, and grinding shrimp. It usually comes in a block shape with a grainy texture, a solid and distinctive aroma, and a rich flavor (Li *et al.*, 2022). In Indonesia, rebon shrimp paste processing usually involves adding salt and sugar, but it is sometimes done without adding ingredients (Rianingsih *et al.*, 2021). Sodium (Na^+) ions in salt are growth factors for halophilic lactic acid bacteria (LAB), essential in shrimp paste. However, an overabundance of Na^+ negatively affects the human body and may increase blood pressure and the risk of heart attacks, strokes, kidney damage, and other health issues. As people become increasingly concerned about what they eat, which may affect their health, including their salt intake, low-salt or low-sodium diets are becoming popular.

Commercial shrimp paste can be found in various packaging materials. The permeability of the packaging material affects air availability during fermentation, which affects LAB growth and sodium levels. The fermentation process is primarily influenced by two

factors: the addition of salt and the packaging material. Adding salt during production can enhance the flavor, act as a bacteria selector, and promote halophilic bacteria growth (Rahmayati *et al.*, 2014). However, because salt is composed of Na^+ and chloride (Cl^-) ions, it also extracts water from cells by binding with hydrogen ions. Packaging materials vary in gas permeability and, therefore, air availability, which positively affects the growth of microorganisms (Cayre *et al.*, 2005).

Terasi comes in various types of packaging, with common ones being plastic, paper, and leaves. The rebon shrimp paste in banana leaf, teak leaf, paper, and plastic packaging was exposed to varying levels of aeration during fermentation, which affected the acidity and sensory quality of the paste. The largest LAB colonies were found in the pastes with teak leaf and banana leaf packaging because the leaves' relatively high permeability allowed relatively much oxygen. Furthermore, the packaging type determines the gas permeability and the shrimp paste's sodium content.

According to Umami *et al.* (2018), teak leaves have

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larger pores and higher permeability than plastic. The permeability determines the level of aeration during fermentation, so teak leaf packaging creates a favorable environment for LAB — which has anaerobic and facultative growth — to flourish. Plastic packaging, on the other hand, is airtight and has a low permeability. The low air exchange rate restricts LAB growth (Cayre *et al.*, 2005). Wrapping a product in a leaf and letting it ferment is comparable to storing it in a dark room (a condition for fermentation). The leaves block light but allow for aeration through their pores.

Salt attracts water, which increases its osmotic pressure. It also facilitates the growth of microorganisms in foodstuffs. Adding salt to foodstuffs reduces acidity because H^+ ions in water and Cl^- ions in salt can form HCl (hydrogen chloride) compounds (Karim *et al.*, 2014). According to Majid *et al.* (2014), salting decreases water activity (a_w), affecting the product's shelf life and quality. Aristyan *et al.* (2014) confirmed that adding salt to rebon (*Acetes* sp.) shrimp paste decreases water activity. Bacteria in shrimp paste need water activity to grow. Low water activity causes bacterial cells to be less productive at the beginning of the static phase and to die before their death phase.

Bacterial cells metabolize to survive and grow. LAB continues metabolizing during the fermentation of shrimp paste. Halophilic LAB requires mineral salt compounds (Liu *et al.*, 2022) to be used in balancing the osmotic pressure of cells and surrounding mediums, so halophilic LAB in shrimp paste can survive high salt concentrations. Moderately halophilic and halotolerant bacteria can balance osmotic pressure to avoid the influence of denaturation by environmental salts by accumulating salts and osmolytes (organic molecules) in the cytoplasm.

Salt can also act as a medium selector. The environmental conditions in shrimp paste with added salt may only support the growth of halotolerant or halophilic bacteria. The fermentation process of rebon shrimp paste involves the presence of salt and halotolerant LAB that act as preservatives. The formation of specific flavors in rebon shrimp paste is caused by halophilic bacteria in aerobic and anaerobic processes decomposing proteins, carbohydrates, and fats. Therefore, salt, during the fermentation of shrimp paste, has a dual function, such as being a preservative and establishing flavors. According to Koesoemawardani (2013), salt also suppresses the growth of decaying bacteria and pathogens in *rusip* through osmolarization, allowing the salt to thin the walls of microbial cells. They state that the longer the fermentation, the fewer the contaminant microbes are because LAB is dominant during the early phases of fermentation. This study, therefore, aimed to

determine the effect of different types of packaging on sodium levels in rebon shrimp paste.

2. Materials and methods

2.1 Shrimp paste and packaging samples

Rebon shrimp paste was obtained from a traditional Demak Regency, Central Java, Indonesia processing unit. The packaging materials were teak leaf, paper (A4, 70 g), and plastic polyethylene (0.04 mm thick).

2.2 Shrimp paste production and analysis

Samples of *terasi* were produced in and obtained from small-scale household industries in Demak. Fresh rebon shrimp was washed with tap water and dried under the sun until dry. Salt 6% w/w was added, and the solution was ground and mashed until mixed. The rebon shrimp paste was again dried under the sun for 7 hrs and fermented overnight. The following day, the mixture was dried under the sun until fully dry. The paste was then wrapped tightly in the different packaging materials and fermented further for 30 days. The rebon shrimp paste was then analyzed for water activity, LAB total plate count (TPC), acidity, salt, and sodium levels.

2.3 Water activity

Water activity measurements were done using an a_w -meter (Rotronic Hygropalm HP23-AW-A, Rotronic Instruments, Crawley, UK).

2.4 Lactic acid bacteria Count

The LAB TPC analysis involved MRS agar media, 1% $CaCO_3$, and 0.01% sodium azide. The pour plate method was used for plating, and incubation was done at 37°C for 48 hrs. Growing colonies were selected based on their ability to produce clear zones (Dewi *et al.*, 2020).

2.5 Acidity testing

Acidity testing was done per the Indonesian National Standard of 2004. Samples were chopped, weighed to 5 g, and placed in a beaker glass to which 10 mL of distilled water was added and stirred until homogenous. The acidity of the samples was then measured, observed, and recorded by dipping the tip of the pH meter into the diluted sample. Each measurement was calibrated with distilled water.

2.6 Salt content

The salt content levels were determined per the Indonesian National Standard of 01-2359:1991. Samples weighing 1–3 g were placed with a pipette in a 250 mL Erlenmeyer with 20 mL of HNO_3 and set to simmer on a

hotplate. Next, 50 mL of halogen-free water was added, and the mixture was cooled to room temperature. Finally, a 3 mL K_2CrO_4 indicator was added and titrated with 0.1 N $AgNO_3$ until it showed an even, light-brown color. The 0.1 N $AgNO_3$ volume was used for the titration and was included in the salt content formula.

2.7 Sodium content

Sodium levels were analyzed with an atomic absorption spectrophotometer (AAS) (Shimadzu, Japan) at 589 nm (Association of Official Analytical Chemists (AOAC), 2005).

2.8 Statistical analysis

A completely randomized design and an experimental laboratory were used for the experiments. The data were analyzed with ANOVA and Honest Significant Differences ($p < 0.05$) tests.

3. Results and discussion

3.1 Water activity

Water activity in rebon shrimp paste before fermentation was 0.65 a_w . Water activity after 30 days of fermentation and with the different packaging materials are shown in Table 1. The paper, teak leaf, and plastic packaging materials did not substantially affect water activity. While the teak leaf packages had wider surface pores than plastic, they could still protect the paste from the outside environment, and the water activity value remained low. Pathogenic bacteria and mold growth can be prevented at 0.59–0.60 a_w , and according to Frazier and Westhoff (1988), to prevent the growth of food-damaging molds, water activity should be below 0.70 a_w -optimum conditions for the growth of *Staphylococcus aureus* and *Salmonella* spp. Appear at 0.990–0.995 a_w and at 0.995 a_w for *Escherichia coli*. Microorganisms that can grow at high solution concentrations, such as sugar and salt, have a low minimum limit of a_w .

3.2 Lactic acid bacteria count

The TPC helps quantify LAB growth in fermented products. LAB TPC after 30 days of fermentation and with the varying types of packaging material are shown in Table 1. In the early stages of fermentation and before packaging, the LAB counted 2.51 log CFU/g. After packaging and leaving it for 30 days, the highest bacteria count was found in the sample with teak leaf packaging, such as 7.59 log CFU/g, followed by paper packaging with 6.66 log CFU/g and plastic packaging with 5.34 log CFU/g. Aristyan *et al.* (2014) found TPC-values in rebon shrimp paste of 2.9×10^6 CFU/g for comparison. Cayre *et al.* (2005) reported that packaging materials with high gas permeability allow LAB to dominate over

other microorganisms.

Fermented products such as shrimp paste have several microflorae essential in manufacturing, like the LAB group and other bacteria. In this study, water activity in shrimp paste tended to be low at approximately 0.6 a_w . Such conditions limit the survival and growth rates of salt-tolerant bacteria. Li *et al.* (2022) reported that Carnobacteriaceae and Staphylococci are abundant in shrimp paste, and Yao *et al.* (2020) found that *Tetragenococcus* play an essential role in forming the aroma of shrimp paste.

3.3 pH

The initial acidity in rebon shrimp paste before fermentation was pH 7.63. Shrimp paste with plastic packaging had the lowest pH; the highest pH was found in the sample with teak leaf packaging. The sample with paper packaging was similar to the other two samples. It may be explained by the relatively limited oxygen permeability of plastic wrapping, which limits air penetration and inhibits bacterial growth. The polyethylene plastic used in this study is commonly used for packaging because of its low oxygen permeability of approximately 4.5 times. The limited oxygen availability can inhibit the growth of aerobic microbes (Gholizadeh *et al.*, 2006). LAB and other bacteria that produce volatile base products are also inhibited, which results in a lower pH in the sample. Teak leaves had the opposite effect. Among the packaging materials in this research, it has the most significant gas permeability, and aerobic bacterial growth was relatively high. Table 1 confirms that teak leaf packaging resulted in a higher pH. The count of bacteria that produce volatile base products may have been higher than that of acid-producing bacteria.

Another mechanism that reduced the pH of samples post-fermentation was salt. Water that binds with Cl ions forms HCl, and at the end phase of the fermentation period, the formation of lactic acid and HCl causes the pH to decrease. Aside from the formation of HCl compounds, a low pH value can also be caused by organic acids from bacterial metabolites. According to Fadilah *et al.* (2018), microbes produce organic acids during fermentation, which decreases the pH. As a result of these reactions, the longer the fermentation period, the lower the pH. This statement is reinforced by Juliarsi *et al.* (2018), who found that as the fermentation period continued, the acidity of sambal masin increased. Another cause for the increase of acidity in sambal masin is that the NaCl compound of salt decomposes into its constituent molecules, such as Na^+ and Cl^- ions, because Na^+ ions are necessary for LAB to grow, and Cl^- ions bind to free water, reducing its availability.

3.4 Salt content

The salt contents after the 30-day fermentation period and with the different types of packaging materials are shown in Table 1. The analysis of rebon shrimp paste showed a salt content of 11.64% before fermentation and 8.28 to 8.34% after fermentation. The salt content in the shrimp paste was relatively low. According to the Indonesian National Standard (2016), the minimum required salt content of shrimp paste is 12–20%. Salt affects the safety of a food product because it has an antibacterial effect. Aristyan (2014) reported that various salt levels during the manufacturing of rebon shrimp paste resulted in varying product characteristics. The less salt was added, the higher the presence of *S. aureus* became until it failed to meet the Indonesian National Standard of a maximum of 1×10^3 CFU/g. Salt concentrations in this study's rebon shrimp paste with different packaging materials showed no significant differences. Despite their varying permeability, Teak leaf, paper, and plastic packaging did not affect the salt content.

3.5 Sodium levels

Sodium levels were determined by calculating the absorbance of sodium ions in the prepared samples. Sodium levels after the 30-day fermentation period and with different packaging materials ranged from 1.32 to 1.69%, as shown in Table 1. According to Hwang *et al.* (2017), sodium levels in seafood during fermentation are 1.58–2.41 mg/100 g, equivalent to 0.0158–0.0241%.

Sodium levels of rebon shrimp paste with teak leaf packaging were lower than with plastic packaging. It is linked to the LAB TPC results, as the higher the total LAB TPC, the lower the sodium levels. LAB uses sodium to maintain the difference in cells' pressure and environment. Sodium levels during fermentation, therefore, decrease. According to Setati (2010), most halophilic and halotolerant bacteria survive in high-salinity environments by accumulating water-soluble organic compounds with a low molecular weight (compatible dissolved substances) to counteract the damaging effects of high salinity on cell physiology and loss of cell water. Sodium in paste is not only sourced from added salt but also comes from the raw material itself.

4. Conclusion

Different packaging materials for shrimp paste resulted in different sodium levels, LAB TPC, and pH values. The teak leaf packaging was found to have the lowest sodium content, followed by paper and plastic packaging.

Conflict of interest

The authors declare no conflict of interest.

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Table 1. Chemical and microbiological characteristics of shrimp paste with different packaging.

Packaging material	a_w	Log (CFU/g)	pH	Salt content (%)	Sodium content (%)
Paper	0.60±0.01 ^a	6.66±1.01 ^{ab}	6.71±0.02 ^{ab}	8.28±0.43 ^a	1.51±0.08 ^{ab}
Teak leaf	0.59±0.01 ^a	7.59±0.57 ^b	6.72±0.03 ^b	8.33±0.25 ^a	1.32±0.12 ^a
Plastic	0.60±0.02 ^a	5.34±0.81 ^a	6.52±0.13 ^a	8.34±0.67 ^a	1.69±0.07 ^b

Values are presented as mean±SD. Values with different superscripts within the same column are statistically significantly different (P<0.05).

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