Chapter 5
Animal-Based Fermented Foods in Tropical Countries: Functional Aspects and Benefits

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ABSTRACT

Tropical countries are rich in fermented animal foods, such as meat paste, shrimp paste, ronto, dadih, Nem chua, and chin som mok. The salt addition (2.4-3.0%) and carbon sources resulted in fermentation process at room temperature in tropical countries. The abundance of Salinococcus spp. during dough preparation and Lentibacillus spp. during fermentation contributes to the distinctive taste and umami of the shrimp paste. Lactic acid bacteria isolated from fermented animal foods have the potential as probiotics. Probiotics can play a role in increasing antioxidant activity and antimicrobial properties. Corynebacterium sp, Bacillus subtilis, and Lactobacillus plantarum were designated as functional starter cultures that could inhibit the growth of pathogenic bacteria (Staphylococcus aureus, Salmonella sp. and Escherichia coli). Animal based fermented foods in tropical countries are very diverse and have functional properties for health, related to antioxidant, probiotic, and antimicrobial properties.

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INTRODUCTION

Currently, a healthy lifestyle is becoming a trend in the world community. Therefore, the existence of fermented foods is needed. The fermented foods and their modifications continue to grow. In tropical countries, traditional fermented foods are very varied, both plant-based and animal-based fermented foods. Most fermented animal-based foods are sourced from livestock meat and seafood. Some examples of fermented animal foods that are well known in tropical countries are meat paste (*Petis*), shrimp paste, *Ronto*, *Cincalok*, *Dadih* (Indonesia), *Nem Chua* (Vietnam), and *Chin Som Mok* (Thailand). Indonesia is a maritime country with two-thirds of its territory consisting of water. As a maritime country, Indonesia has enormous fishery potential. Indonesia's marine fisheries potential is 6.4 million tons annually, with an average production of 4.88 million tons annually (76.3%) (Rinto, 2018). This paper aims to describe various examples of animal-based fermented foods that are developed in tropical countries, their fermentation technique, and their functional properties and benefits for health and product quality.

BACKGROUND

This chapter describes various types of fermented animal food from tropical countries, which are sourced from livestock, fish, and other marine products. The themes presented in this chapter include the types and names of fermented foods, fermentation techniques, and their functional properties. Functional properties include the benefits for health when the food is consumed or benefits for the product development, specifically on flavor and sensory characteristics of the product.

The diversity of processed products contributes to the culinary richness of tropical countries, the majority of which are developing countries. This processed product is also an original ethnic food in tropical countries. Knowledge of the product contributes to food sustainability and serves as a consideration for the authorities in food policy. Therefore, the theme of this chapter is closely related to the theme of the proposed book, namely Food Sustainability, Environmental Awareness, and Adaptation and Mitigation Strategies for Developing Countries.

FERMENTED MEAT (PETIS)

Meat fermentation is quite popular so far, this product is a product of microbial activity in meat-based media that produces distinctive aroma or flavor characteristics. In America, it is known as pepperoni, which is a fermented sausage for pizza, which is the manufacturing process at the time of mixing raw materials at a temperature of 2-3°C, while the fermentation process is at a temperature of 20-28°C. In Europe, it is known fermentation of meat sausages derived from beef, pork, or poultry, the manufacture of which there is one with dry or semi-dry fermentation.

Types of Fermented Meat

In Indonesia there is a fermentation called *Petis* as fermented meat, this product is in the form of a sweet, salty, or sweet-salty taste pasta (according to taste) with the addition of salt, sugar, and spices or spices. Petis products are usually used as seasonings or flavorings, as a complement to food or raw materials for chili sauce, and also as side dishes. The way it is made is that the meat is finely ground and then salted as much as 15 – 20% (w/w), then cooling is carried out at a temperature of 40 °C for 6 hours. After that fermentation takes place spontaneously at room temperature for 48 hours. After fermentation is separated between solids and liquids, it is this liquid that is then added with spices and flavors (salty, sweet, or salty-sweet) and thickened with starch flour by heating at a temperature of 70–80 °C for some time until cooked (Anonym, personal communication, 2004).

Based on the fermentation process and the types of meat fermentation products that exist today, they are divided into 3 parts namely dry fermentation, semi-dry fermentation, and fermentation without drying.

Dry fermentation is characterized by Aw ranging from 0.82 – 0.86 and a final pH of less than 5.3. In this fermentation, dehydration occurs so that the water content becomes about 20 – 30% (w/w), usually, if the fermentation takes place both the bioamines formed are less than fermentation without any addition. In this dry fermentation, there must be the addition of salt, the content of which varies depending on the taste of the maker. Other ingredients added are sugar (some are without the addition of sugar), spices, and some are given nitrite or nitrate to maintain the color to make it more attractive. In the drying process, some use fumigation or without fumigation, and there is a ripening process with molds or maturation without molds. The duration of the fermentation process has three variations in time, namely short fermentation between 1-4 weeks, medium fermentation between 12 – 14 weeks, and long fermentation, which is more than 4 months. This long process depends on the magnitude of the fermented material (meat), the larger the material it takes the longest. For fermentation temperature depends on the process and duration of fermentation carried out, in a short fermentation process without fumigation the temperature is less than 15 °C, while those without fumigation range from 15-26 °C. In the medium-time fermentation process without fumigation, the fermentation temperature ranges from 20 – 25 °C, while in fermentation with fumigation between 32 – 38 °C, the fermentation process takes a long time the temperature is maintained at 25 °C. In dry fermentation this usually has physical properties that can be cut before consumption (sliceable), examples of products that often exist are *Salamis* (Germany, Denmark, and Hungary), *Genoa* (Italy), *Chorizo* (Spain), Summer Sausage (USA), *Thuringer* (Germany), *Sausion Sec* (France), *Thai Nam* (Thailand), and large-shaped ones such as whole meat cured ham (Hammes et al., 2003).

Semi-dry fermentation is characterized by Aw ranging from 0.92 – 0.94 and the pH at the end of fermentation is less than or equal to 0.53. In this semi-dry fermentation, dehydration also occurs but the water content is more than that of the dry system, which is around 40% (w/w). Just like dry fermentation in semi-dry fermentation, the bioamine content will also decrease if the fermentation takes place properly when compared to the fermentation process without any additives. As in dry fermentation the temperature and duration of fermentation depend on the fermentation system carried out, but in this semi-dry fermentation, there are three divisions of time, namely short, medium, and long fermentation. At short fermentations between 1 – 4 weeks, the fermentation temperature is less than 15 °C in the process by fumigation, while the one with fumigation ranges from 15 – 26 °C. The fermentation process is medium in time between 12 – 14 weeks, with a temperature of 20 – 25 °C in fermentation without fumigation, while those with fumigation range from 32 – 38 °C. For a long fermentation time usually months (more than 4 months) at a temperature of 25 °C, the fermentation time depends on the magnitude of the fermented meat. In this semi-dry fermentation, the same as other fermentations of meat, there is always the addition of salt, due to its properties that can serve as an agent for microbiota selection during fermentation. In this process, there is also the addition of spices, sugar (without sugar), and nitrites or nitrates which function to maintain the color to remain attractive, in addition to being a preservative. Examples of products are the same as those with dry fermentation at short fermentation times, namely summer sausage (US) and *Thuringer* (Germany), while for medium fermentation *Salamis, Genoa, Saucision,* and *Chorizo*. An example of a longtime process is cured ham whose basic ingredients can be beef or pork (Hammes et al., 2003).

Fermentation without drying is characterized by high water content and very minimal dehydration occurs, the water content ranges from 34 – 42% (w/w), with Aw between 0.95-0.96, and the pH at the end of fermentation is less than 0.5. The fermentation time is very short only 2 – 4 days with a fermentation temperature of less than 25 ^{o}C, in the process without maturation with molds as well as physical properties can be smeared or flattened (spreadable). Examples of products that are already known in Europe are *Streech Mectwurst* (Germany), *Sobrada* (Spain), and *Petis* (Indonesia) (Pramono, et al, 2009)

Lactic Acid Bacteria as a Functional Starter Culture in Meat Fermentation

The use of functional starter culture in the fermented food industry is currently being developed (De Vuyst, 2000). The term functional starter culture is a starter that has several functional properties when used. The functional properties in question can play a role in increasing product safety and or improving organoleptic properties, technology, nutritional aspects, or health-added value. The application of this functional culture starter must show these functional properties but can maintain the natural properties of the product, as well as its health aspects (De Vuyst and Leroy, 2004).

Fermentation of meat has a purpose (1) to increase product safety through the inactivation of pathogenic bacteria, (2) to increase product stability, namely being able to extend shelf life and inhibit putrefactive bacteria, (3) product diversification; and (4) to improve functional properties (Lucke, 2000). One of the functional starter cultures in meat fermentation is lactic acid bacteria that can improve nutritional aspects, namely increasing bioactive peptides and suppressing bioamine production. Meanwhile, from the aspect of food safety, it can produce components that can play a role in extending storage. For the functional properties of organoleptic, that is, it plays a role in suppressing the formation of "off-flavor" (De Vuyst and Leroy, 2004).

In spontaneous fermentation of meat, there is a very large variation of the microbiota, resulting in a very high variety of quality, resulting in unstable quality. On the other hand, the modern fermented meat industry requires high and stable product quality, minimal risk of quality variation, and the sensory characteristics of the product are getting better, this cannot be met by spontaneous fermentation. For this reason, it is necessary to develop the potential for controlled fermentation of meat with starter cultures whose properties are known.

The research that Santos et al. (2001) carried out showed fermentation of pork sarcoplasmic muscle protein with starter yeast *Debaryomyces hansenii* CECT 12487 isolated from sausages could increase the amount of carnosine. Fermentation was carried out with a salt content of 0.5% (w/w) at pH 6.5 with a temperature of 27 °C for 4 days there was an increase in the amount of carnosine 28.54 g/500 L of extract. This shows the potential for the use of fermentation to increase the amount of carnosine in meat-based media.

Jae-Young et al. (2005) conducted a study on the spontaneous fermentation of oyster sauce at a temperature of 25 °C with a salt content of 25% (w/w), and there was an increase of 18.7% in the amount of carnosine for 4 months. In this study, there was no mention of the type of microbe that played a role in the amount of quantitative increase, but it was suspected to be a halophilic lactic acid bacterium that played a role in increasing the amount of carnosine because it was able to survive at high salt conditions (25%).

Fermentation of the koji system in mackerel fish was performed by Yin (2005) using *Aspergillus oryzae* BRCC 30118 and *Aspergillus sojae* BRCC 30103 with 0.5% NaCl at a temperature of 25 °C for 4 days. Fermentation conditions are carried out with a combination of 50% chopped mackerel (mince), 3% rice flour, as well as 47% water. The fermentation results showed that there was a 10-fold increase in the amount of carnosine which from the beginning was not detected to 10.08 mg/100g with the starter *Aspergillus oryzae* BRCC 30118, while in fermentation with starter *Aspergillus sojae* BRCC 30103 there was an increase in the amount of carnosine by 9.74 times from the beginning also undetected to 9.74 mg/100g.

The increase in the number of carnosine also occurred in a study conducted by Yin et al. (2005) on mackerel-based fermentation. Fermentation is carried out by combining with proteases from *Aspergillus oryzae* which are then fermented with *Pediococcus pentasaceus* type S and L. In the fermentation of *Pediococcus pentasaceus* type S, carnosine was increased by 17 times from the initial amount of only 8.33 mg/100g to 140.91 mg/100g, while in the combined fermentation of type S and L, carnosine was increased by 25x from the beginning of 8.33 mg/100g to 206.19 mg/100g. Utilization of lactic acid bacterial fermentation can increase the amount of carnosine as a bioactive peptide beneficial to health.

The results of research by Pramono, et al., (2009) showed that isolates of *Pediocoocus acidilacticii* YDA3 and *Pediococcus pentosus* YDA4 can be used as starter cultures for fermentation of petis liquid because they can improve quality and maintain stability, maintain product safety, and have the potential to improve the functional properties of *Petis*.

The ability of the two isolates to improve quality and maintain stability because it can suppress the growth of putrefactive bacteria and suppress the formation of "off-flavor" component indicators, namely TVN (total volatile based-nitrogen) and TMA (trimethyls amin), maintaining guaranteed product safety because they can suppress the growth of pathogenic bacteria and histamine-producing bacteria, as well as playing a role in increasing the amount of carnosine so that it has the potential to improve the functional properties of the *Petis* produced.

NEM CHUA

Nem Chua is a processed pork product originating from Vietnam which is fermented for 2 to 4 days at room temperature. *Nem Chua* is a meat paste shaped into cubes, topped with sliced onions, and wrapped in banana leaves and *Psidium guajava*. Packaging aims to create anaerobic fermentation conditions and prevent contamination from pathogenic microbes. *Nem Chua* is usually homemade, in rural Vietnam. The shelf life at room temperature is 5 days, while at refrigerator temperature it is up to a month. *Nem Chua* fermented products produce lactic acid bacteria (LAB) isolates, including the genus *Lactobacillus*, *Coccus*, *Kocuria spp*, and *Staphylococcus*. These lactic acid bacteria are useful in producing antimicrobial compounds and bioactive components. These antimicrobial compounds will inhibit the growth of spoilage microorganisms and pathogens. In addition, lactic acid bacteria also contribute to the development of the unique and distinctive sensory characteristics of *Nem Chua* (Nguyen et al., 2013).

Staphylococcus and *Kocuria spp* play a role in the development of the color and taste of *Nem Chua*. This is due to the degradation of free amino acids during the fermentation process and inhibition of the oxidation of unsaturated free fatty acids.

FERMENTED PRODUCTS BASED ON FISH

Fish contain high water content ranging from 70-80%. Fish also contains food components in carbohydrates, proteins, fats, and minerals. The high water content and composition of the fish cause the fish to experience a deterioration in quality (perishable foods) easily. Protein is the largest component after water found in fish. In addition, the high water content in fish and the nutritional components of fish cause fish to be easily damaged by microbial activity. As a result, numerous attempts were undertaken to handle and treat fish to increase their shelf life. Efforts to extend the shelf life of fish have been carried out both traditional and modern. Several attempts to extend the shelf life of fish include drying, salting, and fermentation.

Fermentation can be done spontaneously or by adding a certain microbial starter. Microbes that play a role in fish fermentation are lactic acid bacteria. Fermentation breaks down complex organic compounds such as carbohydrates, proteins, and fats into simpler forms without oxygen (anaerobic). Fishery fermented products are spread all over the territory of Indonesia, with different characteristics in each region. The fermentation process aims to extend the shelf life while at the same time producing a product with a different taste from the raw material. As a result, fermented products tend to have a more sour taste.

Various fermentation techniques are applied to various types of fish to produce the characteristics of fermented products. Types of fish and different fermentation processes in each region such as Wood Fish (Aceh), *Bisasam* (South Sumatra), *Budu* (West Sumatra), *Naniura* (North Sumatra), *Cincalok* (Riau), *Rusip* (Bangka Belitung), Picungan (Banten), Terasi, Peda (Java), Wadi (Kalimantan), and Bekasang and *Tembalo* (Sulawesi) produce different fermented products. Therefore, the fresher or better quality of the fish used to make fermented products, the better the quality.

Budu

Budu is made from mackerel, but other fish types can also be used. Huge sea fish with white flesh, such as Talang-Talang Fish (*Chorinemus tala*), Mackerel (*Scomberomorus guttatus*), and other types of fish are used. *Budu* fish products are named after the type of fish used such as *Talang budu* fish, *Tenggiri budu* fish, or other *Budu* fish. The purpose of hanging this fish is to allow the blood from fresh fish to drop, lowering the fish's liquid weight. Mackerel used as a preserved product of *Budu* fish must meet several criteria. Mackerel, for instance, cannot be used if it has been mixed with ice if the *Tenggiri* fish has been cooled down with ice.

The process of making *Budu* fish in the community has traditionally been unregulated since the amount of salt and spices utilized varies. Equally, the place and conditions used during the manufacturing process vary based on the habits of each fish processor. According to Marrysa Derec, (2022) the process of making *Budu* fish is as follows:

1. Cleaning The mackerel is cleaned from head to tail. Because putrefactive bacteria in fish are mostly found in the digestive tract, specifically the stomach and gills, cleaning the fish's stomach is intended.
2. Hanging. The hanging process is carried out for 24 hours until the fish meat is swelling. The purpose of hanging is so that the fish's blood goes down until it disappears. If the blood is still present in the fish, it will cause itching for consumers who have eaten fish when it has become a culinary product. The time in hanging this fish must be considered because the fish should not go through the swelling; eventually, the meat will not stick together or break. On the other hand, dividing the fish flesh will be difficult if the fish is less than optimal inflating. The resulting product will not be as expected because the organoleptic value is not good.
3. Fish meat cleavage. The purpose of cleaving the fish meat is to remove all of the bones from the fish. By removing all types of bones found in fish, boneless fish is produced, resulting in only pure fish meat in *Budu* fish products.
4. Giving salt to the fish that has been evenly split. There are several specialized techniques for applying salt to salt. The first aspect is the composition of the salt given to the fish. If the *Budu* fish makers are familiar with or competent in determining the level of salt to be given, this salt will usually produce an optimal product. Aside from that, the size of the fish used for processed products influences the salt composition used during salting. The second consideration is the type of salt used. Sand salt is the type of salt used. Sand salt has been mashed and dried in such a way that it is in the form of sand.

5. Seasoning. The provision of cooking spices on processed products of *Budu* fish depends on the expertise of the local community. Each community has its secret in giving seasoning to *Budu* fish. Some people provide seasonings such as Ajinomoto to add to the taste. After giving the Ajinomoto, it is combined with the addition of a little sugar so that the salty taste of the salt is not too prominent in the *Budu* fish products.
6. Incubation (fermentation). Incubation of fish products takes time, depending on the fermented fish's size. In general, if the size of the fish used is large and thick, incubation is carried out for 3 hours. On the contrary, if the size is small and thin, the time required only reaches 1 hour. The purpose of incubating fish is so that the salt that has been given can dissolve and spread to all parts of the fish meat, and the salty taste of fish products can be achieved. In addition, salting also aims to make the resulting product more durable and durable due to a microbiological process in *Budu* fish products, where salt will inhibit the growth of spoilage microbes and pathogens in fish.
7. Cleaning II. Cleaning the fish from the remaining salt left on the surface of the fish meat during fermentation. Increase the organoleptic value of the resulting product, where the fish will look clean like fish products without salt.
8. Drying fish. Drying fish aims to make fish products dry, free from the water content contained in the product during fermentation. Apart from that, drying also aims to make the fish meat and fish together. Drying is also related to the shape of the product to be produced. The time required for drying depends on the quality of the existing sun. Optical drying generally takes 48 hours according to the sun's state at the drying time.

The use of sufficient salt (certain concentration) aims to absorb the glucose liquid contained in the fish and inhibit the growth of unwanted bacteria. Appropriate temperature settings must also be considered during fermentation to maintain the viability of lactic acid bacteria.

The aroma of *Budu* is produced by methyl ketone, butyraldehyde, ammonia, amino, and compounds other anonymous compounds due to fat oxidation. Even though fat oxidation can cause rancidity, if the process is not too continuous, it will produce a distinctive aroma that consumers prefer.

Lactic acid fermentation is the process used in *Budu* fish production. According to Susalam et al. (2022), lactic acid bacteria isolated from *Budu* fish have the potential as probiotics. Lactic acid bacteria are the dominant bacteria in the fermentation process, which occurs spontaneously. Microorganisms release a variety of metabolites during fermentation, including protease, which hydrolyzes the proteinaceous raw materials. Some manufacturers cook the resulting sauce at the end of the fermentation period, while others do not (Ilyanie et al., 2022).

Maslami et al. (2018) found that lactic acid bacteria isolated from *Budu* can produce glutamic acid, improving broiler carcasses' quality by increasing broiler meat's color and aroma. Aisman et al. (2019) added that lactic acid bacteria from *Budu* can produce gamma amino butyric acid (gaba), which can reduce the effect of stress on broilers with high cage densities. Liasi et al. (2009) asked lactic acid bacteria isolated from *Budu* fish in the form of *Lactobacillus* (*Lactobacillus casei* LA17, *Lactobacillus plantarum* LA22, and *L. paracasei* LA02), and the highest population was *Lactobacillus paracasei* LA02. The lactic acid bacteria showed antimicrobial activity. *Lactobacillus plantarum* and one strain of *Streptococcus faecalis* are also lactic acid bacteria isolated from *Budu* fish. These lactic acid bacteria have the potential to produce lactate which is a group of antibiotics that act as antimicrobials (Ohhira, 1990). *L. plantarum* and *L. paracasei* are potential probiotic lactic acid bacteria isolated from *Budu* (Ilyanie et al., 2022).

The well method can evaluate the *Budu* lactic acid bacteria's ability to fight against microbes. The isolates were grown in MRS broth at 37 °C and centrifuged at 12,000 x g for 10 minutes at 4 °C to create cell-free culture supernatants for antibacterial tests. The antimicrobial activity of the cell-free culture supernatant was determined using agar.'s well diffusion assay. Aliquots of supernatants (100 L) were placed in wells (6 mm diameter) cut in cooled soft nutrient agar plates (25 mL) that had been seeded with the appropriate indicator strains (1 percent v/v). The diameters of the growth inhibition zones were measured after 24 hours. The agar-well diffusion assay was used to determine the inhibitory spectrum of *Lactobacilli* isolates. The plates were incubated in an optimum condition for the growth of the target microorganism. After 24 hours, the growth inhibition zones' widths were assessed. To ascertain the inhibitory spectrum of *Lactobacill*i isolates, the agar-well diffusion experiment was utilized.

Lactic acid bacteria isolated from *Budu* fish showed antibiotic resistance, according to Liasi et al. (2009). According to Liasi et al. (2009) lactic acid bacteria isolated from *budu* fish were resistant to aminoglycoside antibiotics (amikacin, kanneomycin, neomycin, and streptomycin). In addition, lactic acid bacteria are also resistant to gram-negative spectrum antibiotics (nalidixic acid). Resistance to these antibiotics is usually intrinsic, so the genes are not transferred to pathogenic or bacterial flora.

Marlida et al. (2021) stated that microorganisms such as yeast *Saccharomyces cerevisiae* were also found in *Budu* fermentation. Another use for yeast is as a probiotic is known. Using universal yeast agar, which contains 3.0 g.L.-1 of malt extract, 3.0 g.L.-1 of yeast extract, 10.0 g.L.-1 of glucose, 5.0 g.L.-1 of peptone, and 15.0 g.L.-1 of agar, it was possible to isolate the yeast from *Budu*. Following procedures were used to isolate the *Budu* yeast.; the *Budu* (1 g) was added to 9 mL of 0.9% NaCl (saline) solution and mixed thoroughly for 60 s. Serial dilution was then carried out in saline solution and spread plated onto universal yeast agar. The yeast universal agar was composed of 3.0 g.L-1 malt extract, 3.0 g.L-1 yeast extract, 10.0 g.L-1 glucose, 5.0 g.L-1 peptone, and 15.0 g.L-1 agar. The spread-plated yeast universal agar was incubated for 72 h at 28 °C. Presumptive yeast showed white-to-yellow colonies under the microscope. Such isolates were randomly selected and further purified on yeast universal agar. Yeasts showing the typical appearance of *Saccharomyces* (white-to-yellow colonies) were selected. The selected yeast strains were purified by successive streaking on the universal media. Three isolates were maintained at -80 °C in 20% (v/v) glycerol (Hi-Media) (Marlida et al., 2021).

Naniura

Naniura is a fermented fish product from North Sumatra and one of the typical foods of the Toba Batak tribe. *Naniura* is a traditional food that is different from other traditional foods. The difference between *Naniura* and other fermented fish products is that if other foods have a cooking process, either boiled, steamed, fried, or baked, *Naniura* does not go through a cooking process.

Naniura is made using carp (*Cyprinus carpico*) based on the fermentation process, namely the use of microorganisms in food processing. This food is served at major ceremonies such as weddings and funerals. But some make it at gatherings, birthday celebrations, and other family events. *Naniura* is a fermented food product that uses microorganisms in food processing.

Naniura is served without going through cooking using fire, but only with the addition of the prepared spices and *asam jungga* until it becomes soft. Soaked asam jungga (*Citrus jambhiri*) can make raw fish less fishy and tough like raw fish (Silalahi, 2006). The type of acid used to make *Naniura* is *jeruk jungga* (*Citrus jambhiri*).

The Batak people generally make goldfish *Naniura*, using three pieces of asam jungga in 1 kg of carp. Several other types of acids that can be used as alternative acids are *jeruk nipis, jeruk kasturi*, and *jeruk purut*. Because the price of *asam jungga* is also relatively high and difficult to find on the market. The acidic conditions in *Naniura* allow the growth and development of acid-fast bacteria that can prevent the development of spoilage microbes.

Jeruk jungga or some call it *unte jungga* has a shape similar to *jeruk purut*, and tastes almost the same as lime (*jeruk nipis*). The aroma of oranges is also more fragrant than lime. The Batak tribe uses *jeruk jungga* in *Naniura* cuisine because of the high acidity by applying the juice of jungga jungga to fresh carp or *mujair* and allowing them to ripen (Febrian et al., 2016). The dominant acid in orange juice is citric acid, with a pH of 2.2 - 2.5, which can inhibit growth or kill bacteria by damaging bacterial cells to reduce microbial growth, especially pathogenic microbes.

The addition of orange juice in the manufacture of *Naniura* aims to reduce fish's pH value and microbial growth so that *Naniura* is safe for consumption. A low pH in food (<4.5) can inhibit the growth of pathogens so that food can be stored longer than food with a neutral pH (Aloysius et al., 2019).

The length of the soaking process is one of the crucial factors in making *Naniura*. The immersion time is related to the penetration time of the acid into the fresh carp. Soaking fish in acid can affect the nutritional components of carp as the main raw material in the manufacture of *Naniura*. During immersion, there will also be physical changes in goldfish color, aroma, taste, and texture. In addition, ceding carp into *Naniura* fish can inhibit and kill pathogenic bacteria that cannot survive (Haro, 2019).

According to Haro (2019), the manufacture of *Naniura* is done by first preparing the essential ingredients and spices. Then, the carp is washed, scaled, cut in half wide, and removed from the entrails and gills. The carp is washed with cold boiled water and then drained until the water no longer drips. Oranges are also cleaned, cut in half crosswise, squeezed with an orange squeezer, and filtered, and the juice is taken. The spices used to make *Naniura* are shallots, garlic, ginger, *kencur* (*Kaempferia galanga*), and turmeric. Shallots, garlic, and candlenuts are roasted separately until the aroma is fragrant, then mashed. Ginger, *kencur*, and turmeric are grated and squeezed to get the juice. Next, cayenne pepper and *andaliman* are blended. After being steamed, *rias* is mashed. Except for cosmetics and candlenuts, all spices are evenly combined and stirred.

The drained fish is placed in a container and then doused with tamarind juice in a ratio of 1g of jungga acid juice to 1.8 g of carp meat, and 3% salt is added. The fish was soaked for 7 hours. Then, the fish is added with all the spices and allowed to stand for 1 hour after adding the spices. The immersion of fish is done in a container that is not tightly closed. Fish meat soaked in orange juice or acid has a pH of up to 4, most likely not to be overgrown by bacteria, so the fish is safe for consumption (Febrian et al., 2016). *Naniura* is usually consumed without going through the cooking process after fermenting raw carp-based ingredients for 3 hours.

The processing of carp *Naniura* occurs by acids which can cause changes in protein structure with specifications for the color of the fish flesh to be pale white, and myotomes are visible. Myotomes that are visible indicate the presence of protein decomposition due to enzymatic processes that cause changes in texture and appearance of fish and affect pH. The hydrolysis of fish meat protein occurs because the acid causes fish to be consumed without cooking (Indah Turnip, 2017). Febrian et al. (2016) stated that the total microbes of the goldfish *Naniura* will decrease during the immersion time due to the more organic acids from the orange juice penetrating the fish meat tissue as the soaking time increases.

Aloysius et al. (2019) showed that *Naniura* contains lactic acid bacteria with probiotic potential and inhibits pathogens *S. aureus*, *E. coli*, and *S. typhi*. However, the concentration of acid is also different and the length of stay on the total bacterial colonies in the manufacture of carp *Naniura*, resulting in the total bacterial colonies decreasing and maintaining the life of *Lactobacillus sp* while the *Streptococcus sp* bacteria will die (Pasaribu et al. 2015). Total microbes of fresh carp used in the manufacture of carp *Naniura* were 5.5563 log CFU/g. The results of the research by Febrian et al. (2016) stated that the total microbes of goldfish *Naniura* up to 6 hours of immersion still met the requirements of SNI. Based on SNI 01-2779.1-2006, it is indicated that the total plate number in fresh fish is a maximum of 5 x 10^5 colonies/g or 5.6990 log CFU/g (BSN, 2006).

The longer the immersion time, the lower the protein content of the *Naniura* carp. Due to the ability of the acid to penetrate the carp meat, the free water contained in the fish meat is pushed out, and the amount of acid that enters the fish meat increases as the length of immersion of fish meat in acid and spices *Naniura* (Febrian et al., 2016). The acid in fish meat will denature protein which can cause coagulation and release water so that water in fish meat will decrease. The decrease in protein content of carp *Naniura* is influenced by the pH of carp *Naniura*, which decreases along with the longer immersion time. Acidic conditions can accelerate the breakdown of protein into short-chain peptide groups or amino acids easily soluble in water, causing the protein content of the material to decrease (Febrian et al., 2016).

All the lactic acid bacteria isolates had inhibition activity against the α-glucosidase enzyme. The enzyme has the role of degrading polysaccharides into monosaccharides. Thus inhibition makes the isolates can be an alternative way of treating type 2 diabetes. Furthermore, the exopolysaccharide produced by the lactic acid bacteria protects the lactic acid bacteria from hard conditions such as dehydration and acidity, even bile acid.

Bekasam

Bekasam is a traditional fermented fish that tastes sour and is widely known in various parts of Indonesia, especially in South Sumatra (Rusmana, Suwanto, and Mubarik, 2012; Rinto et al., 2015). In addition, *Bekasam* is a source of animal protein, so it has the potential to be used as food that is suitable for consumption by the people of Indonesia. *Bekasam* has other names that are different in some areas, including Pedas, Bekasang, Peda, and Wadi.

Bekasam is generally made from fresh or brackish water fish such as catfish, snakehead fish, tilapia, carp, wader fish, and mujair. The process of making *Bekasam* is still done traditionally by applying spontaneous fermentation. *Bekasam* is made by fermenting fish using high salt content with the addition of rice as a carbohydrate source with a specific ratio and fermenting for 5-7 days. The addition of salt generally ranges from 15-20% of the weight of fresh fish. There is no standard process for making excavate, so each region has its process stages.

In principle, the process of making *Bekasam* is carried out in 3 stages, namely; (1) salting, (2) addition of carbohydrates, and (3) fermentation. Making *Bekasam* begins with preparation, including cleaning the gills and washing the stomach contents. Furthermore, salting is done; the fish is mixed with mixed ingredients, then the fermentation is continued for 5 to 7 days. *Bekasam* is thought to have antihypertensives due to the formation of bioactive peptides resulting from protein degradation during the fermentation process of the *Bekasam* (Wikandari et al., 2012).

The container for making *Bekasam* is generally by placing fish that has been added with salt and a source of carbohydrates in a jar. The jar is closed, so there is only a small cavity between the lid and the fish to be fermented. Because lactic acid bacteria are expected to ferment, fish can grow in low oxygen conditions. In addition to rice as a source of carbohydrates, the maker of *Bekasam* also uses other carbohydrate sources, including roasted rice, cassava, sticky rice, flour, and so on (Omega, 2016). The addition of carbohydrates in the manufacture of *Bekasam* aims to stimulate microbial growth.

Microbes that play a role in the fermentation of *Bekasam* include lactic acid bacteria. Lactic acid bacteria involved in the fermentation of acid reflux include the genera *Streptococcus, Lactobacillus*, and *Staphylococcus* (Lestari et al., 2018). *Lactobacillus plantarum*, *Lactobacillus pentosus* and *Pediococcus pentosaseus* (Wikandari et al. 2012).

Carbohydrates are broken down by lactic acid bacteria producing lactic acid, acetic acid, propionic acid, and ethyl alcohol. These compounds are beneficial as preservatives and give the tamarind product a sour taste and distinctive aroma. Rice is widely used in making *Bekasam.* Lactic acid bacteria have proteolytic enzymes that can degrade fish protein into peptides and amino acids. Lactic acid bacteria are also included in the amylolytic microorganism group, so starch, the primary substrate, will serve as the initial substrate for lactic acid bacteria. Adding a significant carbohydrate source allows lactic acid bacteria to grow well on these foodstuffs.

Salt is a microorganism selector that can minimize the presence of spoilage microbes to extend the shelf life of used products. However, salt can bind water materials and indirectly increase the material's osmotic pressure, so only certain microorganisms can grow. This phenomenon is a selector for several spoilage microbes that cannot tolerate salt (Priyanto and Djajati. 2018).

During the fermentation process, Lactic acid bacteria also produce bioactive components that function for health. Lactic acid bacteria, as in the manufacture of *Bekasam* can be used to improve the nutritional quality (digestibility) of the *Bekasam* and increase its functional value of the *Bekasam*. The functional properties of *Bekasam* as a cholesterol-lowering agent can be seen from the lovastatin content in *Bekasam.*

Lactic acid bacteria isolated from seluang fish shells produce lovastatin as an inhibitor of cholesterol synthesis (Rinto et al. 2015; Wikandari & Yuanita 2014).

Lovastatin belongs to the statin compound known as monacolin K or mevinolin. Rinto and Thenawidjaja (2016) stated that lovastatin acts as a competitive inhibitor for the enzyme HMG-CoA (3-hydroxy-3 methylglutaryl Coenzyme A) reductase. This enzyme determines cholesterol biosynthesis to help reduce cholesterol levels in the blood. Seluang fish extract contained lovastatin on average, ranging from 165.08 to 248.27 ppm (Lestari et al. 2018). Lactic acid bacteria metabolites from fermented tamarind act as antimicrobials, so *Bekasam* products have a longer shelf life when compared to fresh fish (Rusmana, Suwanto, & Mubarik, 2012).

Bekasam is also known to have antihypertensive activity caused by the activity of Angiotensin Converting Enzyme (ACE) inhibitor peptides resulting from proteolytic degradation during fermentation. Lactic acid bacteria and their fermentation products can lower blood pressure and produce bioactive peptides that can inhibit the activity of Angiotensin I Converting Enzyme (ACE). This enzyme plays a role in regulating blood pressure in the Renin-Angiotensin system. The activity of ACE inhibitors produced by lactic acid bacteria is in line with the increase in the number of peptides resulting from proteolytic degradation of lactic acid bacteria (Wikandari et al., 2012). ACE inhibitory activity is thought to be related to the formation of oligopeptides. Proteolytic lactic acid bacteria produce dipeptides, and tripeptides can influence the ACE inhibitory activity. Lactic acid bacteria isolated from *Bekasam* can produce ACE inhibitor activity of 51.77% - 65.75% (Wikandari et al., 2012). ACE inhibitory activity's magnitude correlates with the increase in peptides formed during fermentation.

Lactic acid bacteria isolated from tamarind are also known to have antimicrobial abilities, thereby increasing the product's shelf life. This durability is due to lactic acid bacteria inhibiting spoilage and pathogenic bacteria. Lactic acid bacteria can produce metabolites such as organic acids (lactic and acetic acid), hydrogen peroxide, diacetyl, and bacteriocins (Rusmana, Suwanto, & Mubarik, 2012).

Bacteriocin is an antimicrobial protein produced by lactic acid bacteria. The bacteria closely related to the bacteria that produce the bacteriocin can kill by bacteriocin. Therefore, bacteriocins are safe if consumed and can be applied as food preservatives. Bacteriocins have antagonistic properties against several pathogenic bacteria such as *Listeria*, *Clostridium*, *Staphylococcus*, *Bacillus spp*, *Brochotrix*, *Aeromonas*, and *Vibrio spp*. Therefore, bacteriocins can potentially be used to control bacterial contaminants in food products (Darbandi et al. 2021).

Organic acids are preservatives commonly used in food, and GRAS has a broad spectrum as an antibacterial agent. Organic acids are effective in preserving food because, in addition to antibacterial activity, organic acids also act as a sour taste enhancer (Rusmana, Suwanto, & Mubarik, 2012). Crude extract of bacteriocin isolates of lactic acid bacteria was able to inhibit *Staphylococcus aureus*, *Escherichia coli*, and *Salmonella sp*.

Ronto

Ronto is a traditional fermented shrimp product popular on the coast of South Kalimantan, made from a mixture of *Rebon* (*Acetes sp.*), salt, and rice, fermented for two weeks at room temperature. Comparing *Rebon*, salt, and rice 7:1:2. Fermentation has been performed anaerobically for two weeks (Khairina et al. 2016). *Ronto* processors use different types of doses in determining the ratio between salt shrimp: and rice so that the quality of the product between processors also varies (Khairina et al., 2013). *Ronto* is a chili sauce mixture's side dish, flavoring, and ingredient.

Ronto has a characteristic pungent sour, salty and strong smell of salt with the aroma of fermented shrimp. The taste of *Ronto* is a mixture of sour, salty, and savory flavors with a robust fermented shrimp flavor. *Rebon* has a light pink, acidic taste with a strong savory flavor of fermented shrimp. The texture of the *Ronto* is like porridge with a suspension of crushed rice and shrimp meat.

Processors of *Ronto* use different doses to determine the ratio between shrimp and salt and rice so that the product quality between processors is also various. Small-scale *Ronto* processing (<5 kg) uses a small bowl as a measure. While processing with a capacity of more than 10 kg, the dose used is buckets or cans with a volume of 1 – 3 kg, the difference in the amount used will affect the ratio of salt and rice added so that it will impact the quality of the resulting product (Soetikno, et al. 2018)

Small-scale *Ronto* processing (<5 kg) uses a small bowl as a measure. While processing with a capacity of more than 10 kg, the dose used is buckets or cans with a volume of 1 – 3 kg, the difference in the dose used will affect the ratio of salt, and rice added so that it will affect the quality of the products produced. The addition of carbohydrates in fish processing helps the growth of lactic acid bacteria (Rhee et al. 2010; Adams 2011). The percentage of salt and rice used in *Ronto* processing with a ratio of rebon: salt: rice = 7:1:2 is about 11.47% salt and 19.67% rice (Khairina et al. 2016b). The difference in salt and rice given affects the Aw loss value. The water content and water activity in the material affect the growth and metabolism of microbes that play a role in *Ronto* fermentation.

Various processed fishery products always use salt as a preservative. The amount of salt given other than as a preservative affects the sensory quality of the *Ronto*, especially the taste. High salt is a barrier for consumers because of the very salty taste, but high salt products tend to have a better shelf life. Therefore, determining the optimum amount of salt for the *Ronto* fermentation process needs to be known to obtain durable *Ronto* with sensory properties acceptable to consumers.

The decrease in pH is an indicator of the success of the fermentation process. The pH of *Ronto* generally decreases rapidly if the raw material is easily fermentable carbohydrates. Rice is the most commonly used carbohydrate source. Rice or roasted rice allows the saccharification process by amylolytic bacteria and other bacteria.

The color change of *Ronto* during Fermentation is related to the content of carotene and astaxanthin in the skin and flesh of rebon shrimp. Protein binding and Shrimp shell binding compounds decompose during fermentation causing carotene and astaxanthin bonds to be released, resulting in a color change. During Fermentation, shrimp protein undergoes proteolysis by a group of indigenous protease bacteria that decompose protein into short chain peptides and free amino acids (Chaijan & Panpipat, 2012). These compounds form specific flavors and flavors in fermented shrimp products (Faithong et al., 2010). Various biochemical reactions occur during shrimp fermentation, resulting in the development of color, aroma, taste, and texture of the shrimp fermented product. Proteolysis and hydrolysis cause the release of astaxanthin from protein bonds. Releasing astaxanthin produces a color change from pink (reddish-pink) to orange. When the rebon prawns are still fresh, the color is gray to pale white, and then the color will slowly change to pale pink with increasing intensity until it becomes light pink. Astaxanthin, also known as red xanthophyll with the chemical formula (3,3'-dihydroxy-i,i-carotene-4,4'-dione), plays a role in forming specific colors of crustaceans (Rodriquez et al. 2010).

The difference in salt concentration affects the resulting fallout's total acid and pH value. The pH value generally will decrease rapidly if the raw material is easily fermentable carbohydrates. Rice is the best carbohydrate source. Rice or roasted rice allows the saccharification process by amylolytic bacteria and other bacteria (Fernandez, 2009). High total acid affects the taste of *Ronto*, so it will be less sensory favorable.

Protein decomposition during fermentation is related to forming a total volatile basis that produces *Ronto* aroma. Khairina et al. (2017) reported an increase in the total volatile base value during 12 days of fermentation, namely 150 mg N/100 g sample. The total volatile base value was in line with the Sik-Khae study (Rhee et al., 2011)

The difference in salt and rice affected the Aw *Ronto*. The results of the analysis of diversity showed that there was a significant difference between treatments. The lowest Aw value was indicated by the control treatment. The *Ronto* fermentation process causes a decrease in the water activity and occurs significantly starting on the 4th day of fermentation and the 12th day, reaching 0.83 (Khairina et al., 2016). A w value drops influenced by the salt and water content of the product during fermentation (Abbas et al. 2009). Professionals use water activity in foodstuffs in product development, quality control, and food safety, making it an essential criterion for evaluating and controlling food safety and quality. Water activity decreases during the *Ronto* fermentation process. The water content and water activity in the material influence the growth and metabolism of microbes that play a role in fermentation.

The optimum Aw value for bacteria is 0.8 – 0.9, yeast 0.7-0.8, and mold < 0.7 if the Aw value is lower than this value, the metabolic system and growth of spoilage and pathogenic microbes will be disturbed because it is unable to grow at low Aw. This condition is very beneficial in food preservation, including fermented foods such as *Ronto*.

Thus, the fermentation process in animal-based fermented food must pay attention to the water activity conditions of the ingredients. Water activity can be adjusted by adding sugar, salt, flour or reducing the water content. This is important for the success of the fermentation process and prevention of contamination.

FUTURE RESEARCH DIRECTIONS

Exploration of the potential of animal fermented food is still very broad to be done. Especially for traditional and local fermented products in tropical countries. Examples of such foods are Nem Chua and Cin Som Mok.

CONCLUSION

Fermented animal-based foods in tropical countries are very varied and have the potential to be observed further. The fermentation results produce bioactive components that act as antioxidants, anti-hypertensives, and antibacterial.

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