Effects of Salting Pretreatment on Quality and Safety of Shrimp (*Parapenaeopsis Spp.*). Powder.

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Abstract

Shrimps are well known as one of the healthiest seafood options, providing a huge amount of protein, vitamins, and minerals. However, shrimp is a highly perishable food, thus attempts to make innovative and healthy products may improve per capita consumption while lengthening the nutrient content shelf life. This research focused on the effects of different pretreatment salt concentrations at 0, 3, 6, and 9% on nutritional composition, microbial activity, and sensory evaluation of the shrimp powder samples. The analyses were performed in triplicate using the Association of Official Analytical Chemicals (AOAC) method for proximate analysis and physical analysis was used to determine pH, color, and water activity. The sensory attributes like saltiness, color, odor, texture, and aftertaste were measured using the hedonic scale method. Initial findings demonstrated significant differences in proximate composition, particularly protein content, ranging from 13 to 46% (w/w). There were also notable variations in color attributes among the shrimp powder samples. The lightness value varied, reflecting differences in the brightness levels of the powder, ranging from light to dark shades. The findings also revealed there were no significant differences in the total plate count among the shrimp powder samples which ranged from 3.50 to 2.69 Log CFU/g except shrimp powder treated with 0% salt concentration sample exhibited slightly higher counts compared to others which is 4.93 Log CFU/g. Sensory evaluation revealed variations in saltiness, color, odor, texture, and aftertaste profiles, with certain samples exhibiting stronger saltiness, color, odor, texture, and aftertaste, while others had milder attributes. These findings show that the most preferable salt concentration for brining shrimp is 3% because it can preserve protein content and have the best acceptability during sensory evaluation.

Introduction

Locally known as “udang minyak” in Terengganu, Malaysia or sharp rostrum shrimp is one of 21 species of genus *Parapenaeopsis* (Sakai & Shinomiya, 2011). Shrimp is highly valued for its commercial importance in the seafood industry. It is harvested for both domestic consumption and international trade, primarily for its meat, which is known for its delicate flavor and firm texture.

After harvesting, fresh shrimp should be kept frozen or processed quickly to reduce product deterioration. Even though the freezing method was the most efficient method for preserving marine products, it is costly and cannot maintain the quality for a long time. Shrimps are susceptible to rapid deterioration due to their high protein and moisture content. The moisture content of fresh aquatic products ranges from 75 to 90%. They are very sensitive to heat and highly perishable. Even if there is no external contamination, if they are not preserved after harvest, they spoil in less than 24 hours. The shrimp industry alone is dumping about 50 to 60% waste from acquired amounts around the world that include a lot of bioactive compounds such as protein/peptides, chitin/chitosan, pigments, enzymes, lipids, minerals, and vitamins (Nirmal et al., 2020).

After being caught, the shrimp are immediately kept in cold storage to increase its shelf life. However, cold storage might not be easily available in locations with poor access to electricity (Akanor et al., 2016). Many methods have been applied to dry aquatic products to reduce the moisture content to a level low enough to prevent microbial growth that may contribute to food contamination.
spoilage, but those methods cause deterioration of product quality. Various studies have shown that certain drying methods cause undesirable colour due to excessive reactions during drying (Lin et al., 2022). Pretreatment was used before the drying process to quicken the process, reduce energy consumption, and increase product quality. An example of pretreatment used was salting. Even though salting has long been a common pretreatment for marine products, there is no standard salt concentration recommended for pretreatment of the shrimp to achieve the most palatable salt content. Despite that, there is a shortage of data regarding the effect of different salt concentration treatments on the nutritional quality and safety of the shrimp.

Due to susceptibility to microbial deterioration, aquatic products like fish and seafood must be dried with proper precautions. An alternative preservation method usually used in extending the shelf life of marine products like shrimp is drying because drying can prevent microbial growth and enzymatic reactions. Thus, numerous drying methods have been invented to dry marine products, for example cold air drying (CAD), hot air drying (HAD), vacuum freeze drying (VFD), microwave drying (MD), microwave vacuum drying (MVD), and sun drying (SD) (Lin et al., 2022). Prior to drying, seafood products are frequently salted, blanched, or soaked in antioxidant or preservative solutions. These pretreatment techniques can minimize drying time, boost product yield, give the product a distinct flavor and taste, and extend the product's shelf life (Qiu et al., 2019). High-quality dried products are obtained when the qualities of fresh foods such as color, flavor, nutrients, rehydration, appearance, and uniformity during the drying process are protected (Zhang et al., 2015).

As Jayasinghe et. al, (2010) stated in their study regarding the influence of different processing methods on the quality and shelf life of dried shrimp that shrimp treated at 5% salt concentration for 30 minutes had the best preference during sensory evaluation. In addition, shrimp powder produced from foam-mat drying methods results showed as the temperature increased, drying time, water activity, oil adsorption and color decreased (Hamzeh et al., 2019). Furthermore, shrimps powder produced by grilled methods showed higher nutritional content and antioxidant activity (AlFaris et al., 2022). Another study on stability of dried shrimp produced by freeze-dried showed a lower level of antioxidant parameters such as peroxide value and thiobarbituric acid reactive substance value (TBARS) (Li et al., 2019). Shrimp powder can be used as an ingredient of seafood products that have been developed, including soups, burgers, nuggets, sausages, sauces, snacks, and desserts (Hamzeh et al., 2019).

This research was carried out because there is a growing concern about fresh aquatic products spoiling quickly within 24 hours of harvest. Thus, aquatic products can be brined and dried to reduce moisture, which prevents microbial growth and undesirable chemical reactions involving enzymes that cause spoilage. Furthermore, the salting technique with drying is a highly effective and efficient preservation technique for aquatic products that will improve the quality of shrimp powder. This might increase food security by reducing post-harvest losses due to spoilage and waste. Additionally, it makes the product storable at room temperature, which makes it easier to store, transport, and use the products. Apart from that, a product that has a unique flavor, color, odor, and texture combination that is highly accepted by consumers is also important to make it stand out in the market and be used for routine quality control of the products. Thus, this research aimed to analyze the proximate and physical composition, determine the microbial activity, and evaluate the sensory attributes of shrimp powder treated under different salt concentrations.

Materials and Methods
Preparation of Shrimp Powder

The sharp rostrum shrimp (Parapenaeopsis spp.) were bought at fisherman market Pantai Beting Lintang, Besut, Terenganu. The samples were labeled and kept at -10 °C until analysis. The shrimps were divided into four equal portions and undergo wet salting treatment in four different brine concentrations in 0, 3, 6, and 9% (w/v) at a 1:6 ratio for 30 minutes at room temperature as mentioned by Lin et al., (2022) with a slight modification. Sodium chloride (NaCl), and distilled water was used to make the brine solution. Each sampling point's samples were brined in a separate, closed container. After 30 minutes, the salted shrimp were blanched at a temperature of 100 °C for 7 minutes. The drying process was done at a temperature of 60 °C for 16 hours using an oven as indicated by Rabie et al., (2016). The dried samples were ground into powder and stored appropriately for future analysis.

Methods
Proximate Analysis

The shrimp powder sample was used for the determination of proximate composition including moisture, crude protein, ash content, fat, and carbohydrate based on the Association of Official Analytical Chemicals method (AOAC, 2000). The analyses were performed in three replications. Moisture content was measured using a thermogravimetric technique. Ash content was determined using the dry ashing method. Fat content was determined using the Soxhlet method and crude protein was determined using the Kjeldahl method. Carbohydrate content was calculated as the sum of all protein, fat, ash and moisture minus 100.

Physical Analysis
pH Value

A pH meter (Fisherbrand™ Accumet™ AE150 Benchtop pH Meter, UK) was used to measure the pH value of shrimp powder. Each measurement was made triplicate for each salt concentration. Prior to analysis, 2
g of shrimp powder were mixed with 10 mL of distilled water at a sample-to-water ratio of 1:5 (w/v) for two minutes. Before the measurement, the pH meter was calibrated using standard buffers with a pH value of 7.0 (Noor & Jirarat, 2022).

Determination of Water Activity

Water activity (aw) of shrimp powders was determined using a water activity meter (METER Aqualab PAWKIT, Portable Water Activity Meter, Pullman, USA). About 3 g of the sample was placed in the instrument cup, and aw was measured automatically after starting the program. The data was collected in triplicate (Hamzeh et al., 2022).

Color Measurement

The color of the shrimp powder sample was measured using a Chromameter (Model CR-400, Minolta, Osaka, Japan). The color was expressed in CIExLab system L*, a*, and b* values, where L* denotes lightness on a 0-100 scale from black to white. The positive values of a* and b* indicate redness and yellowness while negative values of a* and b* indicate greenness and blueness. Before the analysis, this instrument was calibrated with white reference tiles. The sample was placed on the white tray with the color reader on it and L*, a*, b* values were then recorded. Each sample’s reading was carried out in triplicate (Noor & Jirarat, 2022).

Microbial analysis

Total plate count method was used to determine the microbiological quality of the samples. Ten grams (10 g) of shrimp powder sample was used for microbiological analysis. Samples were homogenized for 1 minute using a laboratory paddle blender (Stomacher® 400 Circulator, UK) with 90 ml of sterile peptone water to give a 10^1 dilution. The filtrate was used for 6-fold serial dilution by transferring 1 ml of the diluent to test tubes containing 9 ml sterile peptone. Aliquots (0.05 ml) of appropriate dilutions were transferred separately to plates of dried sterile Nutrient Agar (Merck, Germany) for total heterotrophic bacteria counts. Spread plate inoculation was carried out using a clean bent glass spreader. Bacteria plates were incubated at 37 °C for 48 hours. Plates were observed for colony growth after incubation. Colonies were counted, and the average was calculated, recorded, and reported as colony forming units per gram (CFU/ g) (Nwosu et al., 2020).

Sensory evaluation

Samples were evaluated by 40 semi trained panels using 7 points hedonic scale grading in the hedonic test ranging from 1 = dislike very much to 7 = like very much. This hedonic test was used to directly measure the degree of product acceptance by the panels. The attributes that were measured during the test were saltiness, color, odor, texture, and aftertaste. The shrimp powder samples with four different salt concentrations (0%, 3%, 6%, and 9%) were presented in a cup labeled with a random three-digit code to the 40 semi trained panelists. The panelists measured the attributes of the samples by filling out the score sheet consisting of a 1 to 7 hedonic scale based on their preference. The data obtained were analyzed using One Way Analysis of Variance (ANOVA) to determine the significant difference in the acceptability of the sample. All data was provided as mean values with standard deviations (Gatti et al., 2011).

Statistical Analysis

The data obtained from the analysis was used to conduct the basis of statistical analysis. The data were determined using the SPSS (Statistical Package for Social Science) software by One Way Analysis of Variance (ANOVA). All data were presented as mean values ± standard deviations (SD) of three replicates for each analytical determination.

Results and Discussion

Proximate Composition

Shrimp are classified as Crustacea and had impressive nutritional value and antioxidant activity at fresh condition and were affected by pre-treatment and processing methods that applied to prolong the shelf life and quality of the products (Akonor et al., 2016). They also stated the proximate composition of the freshness of shrimps is 73.77% moisture, 3.75% fat, 2.81% ash, and 18.39% protein. Table 1 shows the results of proximate composition of shrimp powder prepared using four different salt concentrations. After the salting and drying process of shrimp, the moisture content was 38.06% in non-salted shrimp powder (0% salt concentration) and were 10.35%, 21.19%, and 31.85% in salted shrimp powder at 3, 6 and 9% salt concentration. The lowest value of moisture content was found in shrimp powder brined with 3% salt solution while the highest value was found in shrimp powder brined with 9% salt solution. The changes in moisture contents of shrimp powder are due to the greater concentration difference of salt between the inside and outside of the shrimp muscle which initiates the osmosis dehydration process. Osmotic dehydration generates countercurrent mass transfer fluxes of solutes and water, specifically solute infusion into the shrimp muscle and water outflow from the shrimp muscle to the surrounding solution. The key technique of osmotic dehydration allows for the removal of water from the product as well as the alteration of its functional qualities through the impregnation of chosen solutes (Ahmed et al., 2016). It is also due to the hygroscopic property of salt that gives the ability for salt to absorb moisture from surrounding which increases the moisture content in shrimp powder with increasing concentration of salt solution. This property is due to its ionic nature, where sodium and chloride ions attract water molecules. In humid environments, salt can either clump together or dissolve, demonstrating this characteristic (Zhang et al., 2020).
The ash content of the shrimp powder increased as moisture content decreased due to salt infusing into the shrimp flesh during the osmosis process. Salt can raise the amount of minerals in food because sodium chloride, which is a source of sodium and chloride ions, makes up most of the salt. When salt is added to food, it may increase the overall mineral content, especially sodium, and chloride, in the ash that is produced. As a result, foods with greater salt content may have higher ash content (Henney et al., 2010).

Dried shrimp powder with 3% salt concentration shows the highest protein content which is 45.78%. During the salting process, the changes in protein structure such as protein denaturation occurred when brine concentration was raised due to the protein salting-out. The salting process entails the loss of water and uptake of salt. Protein salting out, also known as protein precipitation or protein fractionation, is a phenomenon that occurs when proteins undergo precipitation and separate from a solution in the presence of high salt concentrations. When a salt, such as ammonium sulphate or sodium chloride, is added to a protein solution, it disrupts the electrostatic interactions between the proteins and the surrounding water molecules. This disruption causes the proteins to lose solubility and aggregate together, forming a precipitate (Mohamed, 2019). Shrimp with high salt content also have lower protein resistance to heat denaturation, which causes protein denaturation to occur more quickly (Niamnuy et al., 2007). While shrimp powder treated with 0% salt concentration showed the lowest protein content at 13.81%. Drying without salting can lead to the denaturation of proteins, which involves the disruption of their native structure. The removal of water during the drying process can cause proteins to unfold and lose their functional and structural properties. This can result in changes to the texture and quality of the protein, including potential loss of solubility and reduced functionality. Moreover, without salting pretreatment, the rate of protein denaturation increases causing a loss of protein content in the food sample. The presence of salt is necessary for the proper folding, stability, and enzymatic activity of these proteins (Sinha & Khare, 2014).

It has been shown in Table 1 that the fat content in shrimp powder is 1.86% for 0% salt concentration, 2.40% for 3% salt concentration, 2.48% for 6% salt concentration, and 2.30% for 9% salt concentration, respectively. Increasing salt concentration did not significantly affect the fat content in shrimp because salt does not directly affect the fat content in food, its impact on flavor, texture, and overall palatability (Fellendorf et al., 2015).

Carbohydrate content shows a significant difference when increasing salt concentration during brining the shrimp where it decreases with increasing salt concentration. Salting primarily increases taste and preserves food rather than the carbohydrate content. Salt itself does not directly decrease the carbohydrate content of food. However, carbohydrates can be water-soluble, and during salting the carbohydrate in shrimp flesh which mainly consists of a small amount of glycogen, can leach out into the brine solution during the brining process. Increased water elimination through osmosis leads to greater carbohydrate losses in food products due to their solubility in water. As water leaves the food during osmosis, it carries dissolved carbohydrates with it, resulting in a loss of carbohydrates from the food matrix (Henney et al., 2010).

### Physical Properties

Astaxanthin, a red carotenoid found in shrimp, interacts with lots of different proteins. Shrimp’s red color is caused by the release of individual astaxanthin from the carotenoproteins when proteins are denatured (Niamnuy et al., 2007). Table 2 shows the color variations of dried shrimp powder in terms of L*, a*, and b*. The a* value of shrimp powder, indicating the redness, slightly decreased with the concentration of salt solution in the range of 4.98 to 3.26. However, the concentration of salt solution did not significantly affect b* (yellowness) value of shrimp powder. Dried shrimp powder with 3% and 6% salt gave equal L* value which is 75.00 and 74.80. This is because shrimp with higher salt concentrations had faster Maillard browning reactions, which reduced their lightness (L*) (Niamnuy et al., 2007). Apart from that, the water activity of shrimp powder decreases from 1.08 to 0.66 with increasing salt concentration. By combining the dehydration carried on by simple drying with the addition of salt, water activity (aw) is reduced, which in turn decreased or inhibited microbiological and enzymatic activity. This results in a considerable preservation impact on the product.
(Barcenilla et al., 2022). The average pH value of shrimp powder after brining and drying lowered equally regardless of the salt concentration and does not show a significant difference when increasing salt concentration. According to Binici & Kaya., (2017), the pH decreases due to lactic acid arising from the death of the aquatic animals. It also may be due to the increase of acid concentration as water migrants outside the muscle and acidic compounds in salt diffuse into the shrimp muscle.

Microbial growth (Total Plate Count)

Aquatic items can be preserved (increased shelf-life and microbiological stability) by reducing the water activity with the addition of a solute like salt and dehydrating with simple evaporation. Water molecules become trapped among Na+ and Cl ions, making them inaccessible for other purposes such as chemical or enzymatic reactions, or for microorganisms’ growth. This inhibits spoilage and pathogenic bacteria, which extends the shelf-life and safety of foods (Barcenilla et al., 2022). In an analysis performed on shrimp powder treated under different salt concentrations, the results showed the microbial count of shrimp powder are not significantly different as the salt concentration used increased. Table 3 showed that there were no significant differences in the total plate count among the shrimp powder samples treated with salt where the results of the Total Plate Count (TPC) analysis revealed a TPC value of 3.50 Log CFU/g for 3% salt concentration and 2.69 Log CFU/g for 6% and 9% salt concentration respectively, while for shrimp powder treated with 0% salt concentration sample exhibited higher counts which is 4.93 Log CFU/g compared to others. For most ready-to-eat foods, the recommended TPC is less than 6 Log CFU/g (Food Act 1983). This range ensures that the product is relatively free from harmful bacteria and indicates good hygienic practices during processing and storage. This indicates the presence of a moderate microbial load in the sample (Balzaretti et al., 2013). The TPC value for shrimp powder treated with 0, 3, 6 and 9% concentration of salt showed no significant difference and falls within the acceptable range which is less than 6 Log CFU/g, while it is slightly higher for shrimp powder that is not treated with salt (0%). The relatively higher TPC value suggests the need for closer attention to hygienic practices during the processing, handling, and storage of the shrimp powder. It is important to note that the TPC value alone does not provide information about the specific types of bacteria present or their potential for causing foodborne illnesses. Therefore, additional testing, such as specific pathogen identification, may be necessary to assess the safety of the product in terms of pathogenic bacterial contamination.

Sensory Evaluation

The sensory evaluation of the shrimp powder involved assessing its saltiness, color, aroma, texture, and aftertaste which is mainly based on consumer preference. The appearance of the shrimp powder was evaluated in terms of its color, texture, and overall visual appeal. The powder should exhibit a fine powdery texture, with a pale pink or orange color, which was visually appealing to the consumer. Its aroma should have a characteristic seafood aroma, without any abnormal or unpleasant odors, such as rancidity or ammonia-like smells. In terms of taste, the powder should have a mild saltiness and umami flavor without any undesirable aftertaste. These sensory attributes indicate that the shrimp powder is high quality and likely to be well-accepted by consumers.

Table 2. Physicochemical properties of shrimp powder prepared using four different salt concentrations.

<table>
<thead>
<tr>
<th>Salt Concentration (%)</th>
<th>Color</th>
<th>Water Activity</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>L*</td>
<td>a*</td>
<td>b*</td>
</tr>
<tr>
<td>0%</td>
<td>64.55±1.99^b</td>
<td>6.97±1.10^a</td>
<td>21.29±3.13^a</td>
</tr>
<tr>
<td>3%</td>
<td>75.00±3.57^a</td>
<td>4.98±0.35^b</td>
<td>21.51±1.13^a</td>
</tr>
<tr>
<td>6%</td>
<td>74.80±0.39^a</td>
<td>3.26±0.25^c</td>
<td>20.43±0.71^a</td>
</tr>
<tr>
<td>9%</td>
<td>66.63±0.88^b</td>
<td>4.25±0.15^b,c</td>
<td>19.16±0.61^a</td>
</tr>
</tbody>
</table>

Results given as mean values ± standard deviations (SD) of three replicates for each analytical determination. Different letters in the same column indicate a significance difference by Duncan’s multiple range test at p < 0.05.

Table 3. Total plate count of shrimp powder prepared using four different salt concentrations.

<table>
<thead>
<tr>
<th>Salt Concentration</th>
<th>Log CFU/g</th>
</tr>
</thead>
<tbody>
<tr>
<td>0%</td>
<td>4.93±0.32^a</td>
</tr>
<tr>
<td>3%</td>
<td>3.50±0.23^b</td>
</tr>
<tr>
<td>6%</td>
<td>2.69±0.55^b</td>
</tr>
<tr>
<td>9%</td>
<td>2.69±0.12^b</td>
</tr>
</tbody>
</table>

Results given as mean values ± standard deviations (SD) of three replicates for each analytical determination. Different letters in the same column indicate a significance difference by Duncan’s multiple range test at p < 0.05.
received the highest score in each attribute during hedonic test which supported the findings in proximate composition that showed lowest moisture content at 3% salt concentration, thus contributing to its quality and consumer preference. While shrimp powder treated with 0% salt concentration imparts the lowest organoleptic score in terms of saltiness, texture, and aftertaste. Attribute ratings for shrimp powder increased significantly after the shrimp were treated with salt. Salt is commonly used to enhance the flavor and appeal of food due to its ability to improve positive sensory characteristics and mitigate the perception of bitterness and sweetness. Additionally, salting aids in the volatilization of specific food compounds, resulting in an intensified aroma, thus contributing to a more aromatic and flavorful culinary experience (Barcenilla et al., 2022). Despite that, increasing salt concentration in shrimp powder can also decrease the organoleptic score as shown in Figure 1, the organoleptic score decreases from 4.90 to 4.40 when salt concentration increases from 3% to 9%. Excessive salt in food can overpower other flavors and make the food taste excessively salty. This can mask or diminish the nuances of other taste components, leading to an unbalanced and unpleasant flavor profile (Henney et al., 2010).

Figure 1. Sensory attributes of the shrimp powder prepared using four different salt concentrations.

Conclusion
In conclusion, dried shrimp powder with 3% salt concentration shows the highest protein content, while shrimp powder treated with a 0% salt concentration shows the lowest protein content due to loss during drying. The combination of salting and drying reduces the water activity of the shrimp powder and results in low microbiological growth. Dried shrimp powder treated with salt does not show a significant difference in colony count compared to shrimp that does not treat with salt. Thus, the most preferable salt concentration for brining shrimp is 3% because it can preserve protein content, the TPC value falls within the acceptable range, and has the best acceptability during sensory evaluation. Overall, 3% salt solution plays a crucial role in the preservation, flavor enhancement, and sensory characteristics of shrimp powder, but its concentration should be carefully controlled to maintain optimal quality and consumer acceptance. In the future, more addition of other beneficial ingredients should be added to the shrimp powder to improve its taste and quality.

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References