Utilization of garlic as traditional fish handling in Molluccas Islands: case study on layang fish (*Decapterus macrosoma*, BLECKER)

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**Abstract.** Fish form part of a highly nutritional diet and provide a source of essential proteins. However, they can rapidly degrade, particularly when in a fresh state and not handled properly. Degradation can occur either biochemically or microbiologically. Various methods can be implemented to maintain the quality of fresh fish; for example, the utilization of natural preservatives such as garlic. Garlic is a herb which has been proven to inhibit the growth of microorganisms. Tubers of garlic contain antimicrobial compounds such as allicin (diallyl thiosulfinate) and ajoene. This study aimed to determine the effect of immersion duration in a garlic solution on the freshness level of shortfin scad fish. The Total Volatile Base Nitrogen (TVBN) values ranged from 25.33 mg%N to 78.66 mg%N, the bacterial TPC (Total Plate Count) was 7.5x10¹ to 1.6x10⁵ CFU/g, and the pH 5.9 to 8.0. Visibility ranged from 4.64 to 8.33, odor 3.47 to 8.75, and texture 3.62 to 8.20. This study found that immersion treatment using garlic can maintain fish quality for eight hours, with a 20-minute immersion treatment obtaining optimal parameter values.

1. **Introduction**

Fish degradation occurs mainly due to the activity of microorganisms. In addition, enzymatic and chemical reactions also cause fish quality degradation. Fresh fish must be handled in a way which maintains their quality for as long as possible. As it is not feasible to completely avoid decomposition, handling techniques aim to minimize it. For example, higher environmental temperatures lead to increased microbial degradation rates [1], thus, fish must be stored at low temperatures.

According to [2], the naturally high decomposition process of fish means that they cannot be consumed in locations far away from where the fish are caught or produced without any processing or preservation. Preservation methods include cooling, salting, drying, and fumigation. Cooling is one of the most helpful ways to prolong fish quality. Ice is commonly used to maintain fish freshness during handling and distribution/transportation. At room temperature (± 25º C), fish generally only last between 6-12 hours, while ice treatment can maintain their quality for up to 1-2 weeks. At temperatures around 0º C, bacterial growth and enzyme activity are significantly inhibited [3]. However, the availability of ice is sometimes limited, resulting in fish handlers turning to illegal methods, such as formalin solution.

A better approach may be to use natural products. Some traditional ingredients that are often used by people in Maluku islands for handling fresh fish such as Ginger (*Zingiber officinale*), turmeric (*Curcuma sp.*), Picung (*Penguin edule*), atung fruit seeds (*Parinarium glaberimum*), durian seeds (*Durio zibethinus*), cabbage fermentation solution (*Brassica oleracea* L. var capitata) and sirih leaf (*Piper betle* L), have all been shown to have the potential to inhibit the decomposition process of fish [4, 5, 6, 7, 8, 9,10]. One of the ingredient used in this research is garlic. Garlic inhibits bacterial growth more effectively than ginger and turmeric. Garlic extract (*Allium sativum*) can inhibit fly...
infestation [5, 11]. This study aimed to determine the effect of garlic solution immersion duration on the freshness level of layang fish.

2. Materials and Methods

2.1. Raw material preparation

“Layang” fish were purchased from a fish market and washed to remove dirt. Fish were treated by immersion in 6% garlic solution for 10, 15, and 20 minutes. Non-immersed fish served as control. Following immersion, fish were drained and stored at room temperature for 16 hours, with 8-hour time intervals being used for storage, and the fish were then analyzed.

2.2. Experiment design

The experimental design used in this study was a Factorial Randomized Complete Random Design with 2 main factors: immersion period and storage duration (with 3 levels for each factor). Each immersion treatment in garlic solution was repeated three times. Parameters that were analyzed in this study included Total Volatile Base Nitrogen (TVBN), Total Plate Count (TPC), pH and subjective parameters (appearance, odor, and texture).

2.3. Data analysis

Data were processed using Variety Analysis and continued with a Least Significant Difference (LSD) test. Data were processed using the SAS 6.12 program. For analysis of subjective parameters (physical appearance, odor, and texture), Friedman and Multiple Comparison tests were used.

3. Results and Discussion

3.1. Total Volatile Base Nitrogen (TVBN)

Long treatment of immersion, duration of storage and interaction between the two treatments had a significant effect (P <0.05) on shortfin scad fish quality. Fifteen minutes immersion had the highest TVBN value, and was significantly different to immersion times of 10 and 20 minutes, while the 16 hour long storage treatment had the highest TVBN value and was significantly different from the other two storage treatments (Figure 1).

![Figure 1. TVBN value during storage of layang fish after immersed in garlic solution.](image-url)
During storage, TVBN tended to increase. This was due to decomposition of proteins derived from the product itself, as well as contamination resulting from storage conditions. Increased TVBN values were also caused by an increase in volatile bases or compounds (such as ammonia and trimethylamine) in fish meat during storage. According to [3], the degradation of protein compounds (peptides and free amino acids) by enzymes and bacteria in fish meat leads to the formation of such volatile compounds. For example, the degradation of alanine via a dehydrogenation reaction can produce ammonia. Choline is decomposed into trimethylamine oxide by a dehydrogenation enzyme, and is then reduced to trimethylamine. According to [5, 11], garlic extract solution can delay the synthesis of TVBN. This is probably because TVBN is not only produced by microbial decomposition, but also due to the effect of enzymatic degradation.

3.2. Number of bacterial colonies based on Total Plate Count (TPC)
The average bacterial burden from all immersion treatments, based on TPC result, ranged from $7.5 \times 10^1$ - $1.6 \times 10^5$ CFU/g (Figure 2). The duration of immersion, storage and interaction of the two treatments had a significant effect on bacterial TPC in shortfin scad fish ($P < 0.05$). An immersion time of 15 minutes had the highest TPC value, which was significantly different to immersion times of 10 and 20 minutes, while the 16-hour storage treatment had the highest storage TPC value and was also significantly different from the other two storage treatments.

![Figure 2. TPC value during storage of layang fish after immersed in garlic solution.](image)

TPC values generally tended to increase with longer storage times. Garlic immersion treatment led to acceptable TPC values in shortfin scad fish for all subsequent storage treatments, with values being below the cut-off TPC value for fishery products – no more than $5 \times 10^5$ CFU/g. Garlic extract can inhibit the growth of Gram-positive and Gram-negative bacteria. [5] showed that 5% garlic extract inhibits bacterial growth equivalent to 100 g / ml tetracycline. The garlic compound allicin can inhibit bacterial growth by preventing the synthesis of DNA and cell proteins. Bacteria known to have a role in fish quality deterioration include *Acinetobacter* spp., *Achromobacter* spp., *Pseudomonas* spp., *Moraxella* spp., *Aeromonas* spp., *Flavobacterium* spp., *Shewanella* spp., as well as several other Gram-negative bacteria. Gram-positive bacteria such as *Bacillus* spp., *Micrococcus* spp., *Clostridium*
spp., Corinebacterium spp., and Lactobacillus spp. also play a role in fish decomposition, being commonly found in rotten fish [3].

3.3. Potential of Hydrogen (pH)
The average value of shortfin scad fish pH in final storage hours ranged from 5.9 to 8.0. The lowest and highest pH values were each obtained from immersion treatments of 10 minutes with a storage time of 0 hours, and immersion treatment of 15 minutes with a storage time of 16 hours, respectively. The storage treatment and the interaction of both treatments gave a significant effect (P<0.05), but the immersion duration did not significantly affect pH values. The 16-hour storage time had the highest pH value and was significantly different from the storage treatments of 0 and eight hours (Fig. 3).

![Figure 3. pH value during storage of layang fish after immersed in garlic solution.](image)

Fish pH values during storage at room temperature tended to increase. This is due to the activity of microorganisms that break down amino acids into alkaline compounds such as ammonia, causing acid and base compounds to interact and produce inconsistent pH values. pH increases due to fish decomposition during long storage durations. Inconsistent pH values were also expected to be influenced by immersion duration.

3.4. Subjective parameters
The subjective parameters analyzed in this study were fish appearance, odor, and texture (Table 1). The highest (8.33) and lowest (4.64) appearance values for fish were obtained from treatments of 10 minutes immersion with 0 hours storage, and 15 minutes of immersion with 16 hours storage, respectively. Immersion treatment and storage duration had a significant effect on the appearance value of shortfin scad fish. The highest ranking of appearance value (25.0) was obtained from 10 minutes of immersion with 0 hours storage duration. The lowest appearance value (5.0) was obtained from 10 and 15 minutes of immersion with 16 hours storage duration. Highest (8.75) and lowest (3.47) shortfin scad fish odor values were obtained from the interaction of 10 minutes immersion with 0 hours storage, and 10 minutes immersion with 16 hours storage, respectively. The interaction of immersion duration in the garlic solution and the duration of storage significantly influenced the value.
of fish odor. The highest value (27) was obtained using 10 minutes immersion with 0 hours storage, and the lowest value (7.5) came from 20 minutes immersion with 16 hours storage.

The highest (8.20) and lowest (3.62) texture values of shortfin scad fish were each produced by the interaction of 10 minutes immersion with 0 hours storage, and 10 minutes immersion with 16 hours storage, respectively. Immersion duration and storage time had a significant effect on the texture value of shortfin scad fish. The highest value (27.0) was obtained from 10 minutes immersion with 0 hours storage, and the lowest value (3.0) resulted from a 10-minute immersion with a subsequent 16-hour storage time.

Table 1. Subjective parameter value on various treatments of immersion and storage duration.

| Experimental Treatments | Subjective Parameter |
|-------------------------|----------------------|
|                         | Appearance | Odor  | Texture |
| Immersion 10 min, Storage 0 h | 6.1      | 8.75  | 8.20    |
|                         | 8 h       | 7.33  | 6.33    | 6.01    |
|                         | 16 h      | 5.15  | 3.47    | 3.62    |
| Immersion 15 min, Storage 0 h | 8.21     | 8.47  | 7.77    |
|                         | 8 h       | 6.90  | 5.67    | 5.09    |
|                         | 16 h      | 4.64  | 3.87    | 3.77    |
| Immersion 20 min, Storage 0 h | 8.31     | 8.49  | 7.85    |
|                         | 8 h       | 6.49  | 6.60    | 5.14    |
|                         | 16 h      | 4.73  | 3.87    | 3.93    |

During storage at room temperature, the appearance values of fish for all treatments decreased. During storage, fish products are damaged due to fat oxidation, discoloration, protein changes as a result of bacterial activity and biochemical processes due to enzymes contained in fish, thus affecting appearance, odor, and texture.

During storage at room temperature, fish odor values tended to decrease. At the beginning of storage, the fish odor was still fresh mixed with the aroma of garlic. The distinctive smell of garlic is different for each immersion treatment. At the end of storage, the unpleasant odor of the fish could be due to the distinctive aroma of sulfur from garlic that can neutralize and reduce the smell of rotten fish. The decrease of odor values was generated by the degradation of proteins and amino acids by bacteria. In addition, during the storage of food, bacterial growth can change food’s composition, causing an unpleasant odor. Furthermore, [5] stated that the decrease of fish odor value is caused by the discharge of proteins, such as free amino acids, due to the activity of protease enzymes derived from spoilage bacteria which then produce metabolites that cause unpleasant odors in fish; for example, the indol odor generated from tryptophan. In addition, further degradation of free amino acids, oxidation and fat hydrolysis resulting in free fatty acids such as lauric acid and myristic acid can also cause unpleasant odors in fish.

Texture values decreased with increasing storage time. Low quality of shortfin scad fish is identified by changes in meat texture, which becomes flabby, and damage to the fish abdominal wall; this is as a result of bacterial and enzyme activity, protein degradation, and fat oxidation. After death, enzyme activity in the fish body increases with storage time, with fish meat becoming soft as it loses elasticity.

4. Conclusion
From the results of this research, it is concluded that immersion in garlic solution can maintain fish quality for eight hours storage, with a 20-minute immersion treatment obtaining the best objective parameter values.
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