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Microbiological Status of Three Quality Categories Tiger Shrimp After Land Distribution Process (Case Study: Selili Fishing Port, Samarinda, Indonesia)

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ABSTRACT

Tiger shrimp (*Panaeus monodon*) is a crucial fishery product in East Kalimantan, Indonesia, distributed within 12 h by land routes, leading to quality deterioration due to microbial contaminants. This study aimed to analyze the presence of microbial contaminants in tiger shrimps and detect risk factors that cause the presence of bacterial contaminants in shrimp. The sampling used in this study was a purposive sampling of tiger shrimp based on three organoleptic quality categories (good (1), moderate (2), and poor (3)) and analyzed with several specific media. Risk factor observations were performed during the handling process. The highest total bacteria and coliforms were found in shrimp 3, at $30.25x10^6$ cfu/g and >1100 MPN/g, respectively. Based on the distribution of bacteria in each shrimp, shrimps 2 and 3 were contaminated by six types of bacteria: *Pseudomonas* sp., *Aeromonas* sp., *Salmonella/Shigella*, *Klebsiella* sp., *E. coli*, and *S. aureus*. *E. coli*, *Klebsiella* sp., and *Salmonella/Shigella* bacteria were not found in shrimp 1. The presence of these bacteria plays a role in the deterioration of tiger shrimp. Bacteria in shrimp is caused by the equipment and materials used, worker behavior, and environmental conditions that do not pay attention to hygiene during the tiger shrimp handling process. These results suggest that the long distribution process of tiger shrimp, coupled with poor handling, leads to bacterial contamination, resulting in quality degradation in the form of tiger shrimp spoilage, requiring a particular strategy to minimize bacterial contamination.

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1. Introduction

Fishery products are perishable and require careful handling. Fish and fishery products, especially those categorized as fresh raw materials, contribute to diseases caused by pathogenic bacteria, biotoxins, histamines, viruses, and parasites [1]. Fish and fishery products have been identified as media for the growth of pathogenic bacteria, which can subsequently cause human diseases. These bacteria include *Salmonella* spp., *Listeria monocytogenes*, *Vibrio* spp., *Yersinia* spp., *Clostridium botulinum*, *Staphylococcus aureus*, and *Aeromonas* spp. [2]–[6]. In addition to being pathogenic and causing diseases in humans, bacteria also act as spoilage agents in fishery products [7]. Spoilage bacteria can produce decarboxylase enzymes that significantly convert free histidine into histamine, leading to poisoning in individuals who consume it. This poses a problem for food safety [8].

Food safety, especially in fishery products, is of utmost importance for consumer health and is directly related to farming and food production stages. Consuming fishery products contaminated with pathogenic bacteria is a significant cause of hospitalization and death, particularly in developing countries. Like other types of food products, fishery products are susceptible to contamination by pathogenic bacteria [4]. Pathogenic microorganisms and spoilage bacteria can contaminate fishery products at any time during handling and throughout the supply chain [9]. Several studies conducted in other countries have detected the potential danger of spoilage bacteria caused by various factors, such as contamination from humans or the environment, distribution of fishery products (transportation), packaging, and personal hygiene of workers [10].

Distribution is part of the marketing of fishery products, and most fishery product distributions are not exempt from quality deterioration during the distribution process. Several physical, environmental, and biological factors cause a decline in the quality of fishery commodities. Physical factors include improper handling, unsuitable conditions during fish capture and storage, and transportation that does not meet standards, leading to quality deterioration owing to microbial contamination [11]. Environmental factors include cleanliness in handling areas, and the environment is considered the most crucial determinant of the microbiota in fish and fishery products [12]. It is difficult to detect the presence of pathogenic bacteria; therefore, cleanliness is crucial. The quality and safety of fishery products can be directly influenced by the lack of hygienic practices in fish handling and contact with workers who may carry pathogenic bacteria, including uncleaned working equipment [13].

Biological factors include contamination with pathogenic bacteria in fishery products, posing a risk to the safety of fishery products. The risk to fishery product safety lies in the contamination of pathogenic bacteria from the environment where the food is caught and contaminated by consumers before consuming fishery products [14]. Biological factors play a crucial role in quality deterioration, and bacterial contamination accelerates the decay of seafood products. East Kalimantan's flagship cultural commodity is tiger shrimp (*Panaeus monodon*). The tiger shrimp cultivated here is marketed globally as well as locally. Local marketing extends to several areas around Samarinda City, such as Balikpapan City, Bontang City, Kutai Kartanegara Regency, and West Kutai Regency, particularly the Barong Tongkok Market (Melak). The distribution of tiger shrimps is conducted via land routes using open truck units, with a

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travel time of approximately 12 h. After undergoing a series of handling activities during the shipping process, tiger shrimp are marketed locally in Melak. However, some shrimps that arrive at Melak experience a decline in quality following organoleptic testing [15], [16]. To validate the decline in quality resulting from biological factors, further testing in the form of microbiological analysis is needed to determine the presence of contaminant microbes that cause a decrease in shrimp quality. Therefore, this study aimed to analyze the presence of contaminant microbes in tiger shrimp categorized based on organoleptic tests and detect the risk factors causing these contaminant bacteria in shrimp.

2- Materials and methods

Samples and Media

The materials used in this study included three samples of good-quality shrimp, three samples of moderate-quality shrimp, and three samples of poor-quality shrimp [15]. Mediums used are Tryptic Soy Broth (TSB) (Merck, US), a coliform testing medium consisting of Lactose broth (LB) (Merck, US), Brilliant green bile broth (BGLBB) (Merck, US), and Eosin methylene blue (EMB) agar ((Merck, US),. The selective differential media used were Deoxycholate lactose sucrose (DCLS) agar (Merck, US), Cystine lactose electrolyte deficient (CLED) agar (Merck, US), and Glutamate starch phenol (GSP) agar ((Merck, US).

Research Design and Location

This study was conducted at the Barong Tongkok Market in Melak, West Kutai Regency. The type of research is descriptive, aiming to describe the condition of bacterial contamination in tiger shrimps that have undergone distribution processes within 12 h via land routes using open truck units covered with tarpaulins to transport cool boxes containing shrimps and other fisheries commodities packed with ice cubes. Samples of shrimp collected from Melak were subsequently tested at the Microbiology Laboratory of IPB Bogor. The selection of this location was based on the distance of the distribution location compared to the others, allowing for quality degradation testing of shrimp that have undergone distribution.

Sample Collection

Sampling was conducted using random purposive sampling, where shrimp were classified based on their quality as good, moderate, or poor after organoleptic testing [15], [16]. Sampling was performed carefully to avoid increasing bacterial contamination. Three shrimp were sampled in good condition, three in moderate condition, and three in poor condition. Subsequently, the shrimp samples were separated based on their quality, washed with clean water, placed in plastic bags, and subjected to freezing treatment in boxes before being transported to the testing laboratory. Freezing was conducted to prevent bacterial cell division, which could accelerate the decay process in shrimps.

Detection of Bacteria Presence Based on Most Probable Number (MPN)

The MPN method consists of three stages: presumptive, confirmed, and complete. The presumptive test used LBDS (lactose broth double strand) and LBSS (lactose broth single strand) media. A confirmation test was performed using BGLBB media. The MPN coliform value was calculated using the MPN index table. The completed test was conducted using EMB Agar media. Incubation was carried out for 24 h at 35 °C, and bacterial colonies were observed (metallic green colonies indicate *Escherichia coli*).

Detection of Total Bacteria Presence and Bacterial Types on Specific Media

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All samples (1 g), with two replicates, were serially diluted (10^{-1}–10^{-7}) using sterile distilled water. Subsequently, the diluted samples were tested using the spread plate method on TSB, GSP, CLED, and DCLS agar media poured into petri dishes. Petri dishes containing the samples were incubated at 35 °C for 18-24 hours. Red colonies grown on GSP agar medium were identified as *Pseudomonas* sp. and yellow colonies as *Aeromonas* sp. [17]. Bacterial colonies exhibited different morphological characteristics depending on the type of bacteria on DCLS agar (Table 1) and CLED agar (Table 2).

Observation of Risk Factors during the Distribution Process of Tiger Shrimps

Various physical, chemical, and biological factors cause a decline in the quality of tiger shrimp. Observations are needed to determine the emerging risks and identify the risk factors causing the decrease in the quality of tiger shrimp during the distribution process from PPI Selili Samarinda to the Barong Tongkok Market, Melak. The process of tiger shrimp delivery activities was obtained through observation and interviews with actors involved in logistics activities from PPI Selili, delivery services, and the Barong Tongkok Market. This was followed by hazard potential identification and hazard potential assessment to determine the potential hazards causing the risk of quality decline in tiger shrimp during delivery [15], [16].

Data Analysis

The data presented are the mean values and standard deviations from three trials analyzed using Microsoft Excel 2021.

3- Results and discussions

Microbial Presence Based on Total Plate Count and Most Probable Number

The shrimp analyzed by total plate count indicated that all three samples had high bacterial counts. In this study, shrimp classified as good, fair, or poor quality contained many bacteria in the TPC test. The bacterial counts in the shrimp ranged from $58.5x10^5$ to $30.25x10^6$ cfu/g. The lowest bacterial count was observed in sample 1, whereas the highest was in sample 3. The TPC test was conducted to determine the microbial count in a product by counting the bacterial colonies grown on agar media [18]. The total population of each sample was further analyzed for coliform microbial contamination, an indicator of product safety. Based on the results, two good- and fair-quality shrimp samples had a small amount of coliform microbial contamination. In contrast, shrimp 3, which was of poor quality, was contaminated with coliform bacteria in high numbers in shrimp 3 (>1100 MPN/g) (Table 3). This indicates that poor-quality shrimp contain higher levels of contamination. Coliform bacteria are microorganisms that cause environmental pollution or poor sanitation due to domestic waste [19]. Coliform bacteria are a group of bacteria originating from human and animal feces that are present in large numbers, making them commonly used as indicators of food and water quality [20].

Coliform bacteria belong to the family *Enterobacteriaceae* and are divided into fecal and non-fecal groups. Fecal coliforms serve as indicator bacteria, indicating the presence or absence of pathogenic bacterial contamination. This is because the presence of fecal coliform colonies correlates positively with the presence of pathogenic bacteria. Contamination with these bacteria can increase the incidence of diarrheal diseases [21]. Fewer coliforms indicate better water quality [19]. In shrimp 1, shrimp 2, and shrimp 3, no bacteria classified as fecal coliforms or *E. coli* were found. Further testing was conducted to ascertain the types

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of bacteria present in the three shrimp samples using specific media.

Microbial Presence Based on Specific Media

Further analysis of the presence of bacteria in these three shrimp categories was conducted using specific media, indicating the potential presence of bacteria that cause a decrease in shrimp quality. The first step involves using a DCLS medium, which is known to detect bacteria of the *Enterobacteriaceae* group [22]. *Enterobacteriaceae* are opportunistic pathogenic bacteria that constitute the normal flora capable of residing in the human body, including in the intestine. They can be found in the soil, water, and air, as well as in the digestive tract of humans and animals. The presence of these bacteria can serve as an indicator of contamination of animal or human feces [23]. Based on testing on a specific DCLS medium, several types of bacteria, such as *Proteus* sp., *Salmonella/Shigella* sp., and *Escherichia coli*, were found in the shrimp samples. *Proteus* sp. bacteria were only found in shrimp sample 1 at 3280±16.8 cfu/g. *Salmonella/Shigella* sp. was found in shrimp sample 2 (3100±7.4 cfu/g) and shrimp sample 3 (3860±8.1 cfu/g). *E. coli* was also found in shrimp 2 (40±5.0 cfu/g) and shrimp 3 (450±9.0 cfu/g) (Figure 1). The quantities of *E. coli* and *Salmonella/Shigella* were higher in poor-quality shrimp, indicating that these bacteria are correlated with shrimp spoilage.

E. coli can be transmitted from the feces of infected humans or animals to new hosts, including seafood products, through environmental reservoirs such as hands, water, and soil [24], [25]. *Salmonella* spp. play an important role in food contamination. Contamination with this bacterium can occur through the fish production chain due to inadequate handling, hygiene, or contact with contaminated water. This bacterium was not initially reported as a biological contaminant in fish [26]. *Salmonella* contributed to bacterial contamination in 7.5% of the outbreak-related cases in Brazil in 2016, whereas it caused 7,728 cases of foodborne illnesses in the United States in 2015, accounting for 15.89% of the incidents [27]. In Europe, 4,362 foodborne illness outbreaks have been reported, mainly caused by *Salmonella* spp., accounting for 21.8% of all outbreaks in 2015 [28]. In humans, *Salmonella* can cause gastroenteritis, bacteremia, and more serious diseases, such as typhoid fever and typhoid fever [29]. In addition to *Salmonella* spp., *Shigella* spp. also play a role in various incidents of death due to acute diarrhea, known as shigellosis [30]. This species is commonly found in water, such as *S. flexneri* [31].

These bacteria can survive in soil and water and contaminate fish and seafood products, making it crucial to provide information about the contamination process and the spread of these bacteria. This allows for tracking of microbial sources present at fish-cutting locations [32]. The presence of microbes in freshly caught seafood reflects the microbial content of the water in which they reside; the more contaminated the water, the greater the variability of these microbes [33]. Therefore, environmental factors, such as water quality, play a crucial role in *Salmonella* and *Shigella* contamination in fish/shrimp and pose a significant risk to those consuming fish caught in contaminated waters without sanitation controls [29].

Another specific medium used to detect the presence of contaminant microbes is the CLED medium. On CLED medium, we successfully identified several types of microbes, including *Proteus* sp., *S. aureus*, *E. coli*, and *Klebsiella* sp. Similar to the testing on the DCLS medium, *Proteus* sp. bacteria were found only in shrimp sample 1 at 3000±28.28 cfu/g. *S. aureus* bacteria were found in all three samples, with the highest count in shrimp 2 at 1550±35.4 cfu/g and the lowest in shrimp 3 at 395±19.5 cfu/g. *E. coli* bacteria were not found in shrimp sample 1, and the highest count was found in shrimp sample 2 at 855±18.1 cfu/g. *Klebsiella* sp. bacteria were not found in sample 1, and the highest count was found in shrimp sample 3 at 3000±20.0 cfu/g (Figure 2).

The CLED medium was also used to detect the presence of other bacteria, capable of detecting various types such as *Escherichia coli*, *Klebsiella pneumoniae*, *Salmonella* sp., *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Enterococcus* sp., and *Proteus* sp., thus increasing the diversity of the types and quantities of microbes found [22], [34]. The CLED medium is a selective and differential medium for contaminating bacteria associated with the urinary tract, often found as contaminants in water. *S. aureus* is a pathogenic bacterium also commonly found in fishery products. It is the third most common cause of foodborne illnesses worldwide [35]. This bacterium is one of the most common agents causing food poisoning outbreaks [36]. The presence of *S. aureus* is relatively high in seafood products, corresponding to reports of staphylococcal food poisoning caused by raw seafood products in many countries [37]. Microbial growth is influenced by environmental factors such as pH, temperature, and water activity. In Japan, foods most commonly associated with staphylococcal food poisoning are sushi (raw fish) and foods containing multiple ingredients [38]. S. aureus is often detected in meat products in Italy [39]. In Korea, it has been found that 19.8% of raw fish are contaminated with *S. aureus* [14]. In China, *S. aureus* threatens public health and is often associated with postharvest and processing procedures, leading to product recalls due to contamination [40].

Bacteria *Pseudomonas* sp. and *Aeromonas* sp. were also found in shrimp samples in significant amounts based on analysis of the GSP medium. Both types of bacteria were found in all three shrimp samples. Shrimp sample 3 was the highest contaminated sample with *Pseudomonas* sp. bacteria ($300 x 10^3 \pm 30.0$ cfu/g) and *Aeromonas* sp. bacteria ($244.5 x 10^3 \pm 33.23$ cfu/g) (Figure 3). The GSP medium is specific for *Pseudomonas* and *Aeromonas* bacterial groups. Bacterial colonies that grew yellow were classified into the *Aeromonas* group, whereas colonies that grew red were classified into the *Pseudomonas* group [17]. *Pseudomonas* sp. and *Aeromonas* sp. are often found in seafood products, leading to infections in live seafood commodities and decreased quality of dead seafood commodities. *Pseudomonas* and *Aeromonas* are commonly found in soil and aquatic environments. In addition to being spoilage bacteria in fish, *P. aeruginosa* is also pathogenic to humans, as it can cause infections when the host's defense function is abnormal. This bacterium can also cause food poisoning due to enterotoxins that disrupt the human digestive system. Typically, digestive disturbances such as nausea, vomiting, diarrhea, and abdominal cramps occur [41].

Aeromonas spp. is a common cause of disease in fish, with two strains, *A. hydrophila*, and *A. salmonicida*, the most frequent culprits. In addition to causing diseases in fish, contamination with seafood products containing these bacteria can also lead to human illnesses. Infections in humans are typically caused by *A. veronii*, *A. hydrophila*, and *A. caviae* (*A. punctata*), resulting in bacteremia, gastroenteritis, or even septicemia in individuals with both robust and weakened immune systems [5], [42]. Several outbreaks related to seafood products caused by *Aeromonas* spp. have been reported in India and Bangladesh. *Aeromonas* spp. contribute to epidemic ulcerative syndrome (EUS) in various types of fish, significantly damaging the quality of

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seafood products. Contamination of seafood products is caused by the colonization of the intestines by these bacteria in marine environment [43]. *Aeromonas* strains are widely distributed in various aquatic environments, including wastewater and natural water bodies, such as lakes, rivers, estuaries, and urban drinking water. Additionally, these bacteria can survive in soil, food, and animals [5]. Shrimp, a seafood product, is highly susceptible to contamination by *Aeromonas* spp. bacteria in the water in which they grow and are cultivated, even though seafood products have been processed to kill these types of bacteria. However, shrimp and other seafood products remain contaminated with these bacteria, indicating that processing facilities play a crucial role in the contamination of seafood products by *Aeromonas spp.* [43]. The bacterial distribution in each sample varied greatly based on the analysis of these three specific media types. Four types of bacteria dominated sample 1: *Pseudomonas* sp., *Aeromonas* sp., *Proteus* sp., and *Staphylococcus aureus*. *E. coli*, *Klebsiella* sp., and *Salmonella/Shigella* were not found in shrimp sample 1 (Figure 4). Six types of bacteria dominated sample 2: *Pseudomonas* sp., *Aeromonas* sp., *Salmonella/Shigella*, *Staphylococcus aureus*, *Klebsiella* sp., and *Escherichia coli*. Sample 3 also contained six types of bacteria from different orders: *Pseudomonas* sp., *Aeromonas* sp., *Salmonella/Shigella*, *Klebsiella* sp., *E. coli*, and *S. aureus* (Figure 4).

Factors Causing Contamination and Quality Decline Risks in Shrimp

Contaminant bacteria in shrimp samples can be identified based on risk factors during handling activities, such as loading and unloading processes, shipping, and handling at the unloading site. Direct field observations indicate several potential risks associated with contaminating shrimp bacteria. Identifying these potential risks is divided into three aspects: equipment and materials, worker behavior, and environmental conditions. The main potential risks associated with contaminating bacteria in tiger shrimp include the cleanliness of the equipment used, use of dirty water, use of ice (quantity and form), workers' behavior that does not prioritize cleanliness, and dirty environmental conditions (Table 4).

The presence of contaminating bacteria significantly affects the shelf life and quality of the harvested shrimp. Contaminant bacteria can grow rapidly, possess endogenous enzymatic activity, or produce unsaturated fatty acids owing to chemical degradation (auto-oxidation), resulting in shrimp meat spoilage [44]. Microbial contamination in tiger shrimp arises from careless handling practices, such as washing water and ice of unknown origin, which can contaminate shrimp. Factors contributing to bacterial contamination include the water used in the washing process to clean equipment during the postharvest activities of fishery products. Washing shrimp during handling processes using contaminated water can lead to microbial contamination, even after washing. Water has been widely reported as a medium capable of forming microbiomes in fish and fishery products [45]. Using rinse water and packaged ice from contaminated sources contributes to high levels of bacterial contamination in fishery products, particularly in Asian countries [46]. Our previous findings revealed that the washing water for fishery products at Selili Port, where these shrimp samples originated, was contaminated with bacteria such as coliform bacteria, fecal coliforms, *S. aureus, Proteus* sp., *E. coli*, *P. aeruginosa*, *Aeromonas* sp., and *Salmonella/Shigella* [47]. Several pathogens, including bacteria, viruses, and protozoa, can contaminate water. According to the World Health Organization (WHO), 80% of all

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diseases in developing countries are caused by contaminated water [48]. Monitoring water sources and identifying sanitation risks are top priorities in many developing countries, as is the sanitation supervision of fishery products in urban and rural areas. Therefore, it is crucial to investigate the quality of natural water sources and their effects on food safety [49].

In addition, using ice in shipping processes of unknown origin also leads to microbial contamination of transported shrimp. The provision of ice is a common choice among the methods for preserving shrimp products in the long term. Ice can inhibit microbial growth and slow enzymatic activity, thus preserving taste and nutritional value. However, it should be emphasized that while ice can maintain product quality, it cannot improve shrimp quality [50]. Bacteria that contaminate a product cannot be entirely eradicated by icing or freezing water [51]. When the ice layer is removed from the shrimp product, the shrimp experiences an increase in temperature. Subsequently, bacteria multiply rapidly, further exacerbating shrimp spoilage [52]. Based on field observations, the workers did not add or replace the melted ice, thereby increasing the temperature inside the storage containers.

Low sanitation and hygiene during processing and handling. Sanitation and hygiene factors during handling and processing of fishery products must be considered to avoid food poisoning due to bacterial contamination [53]. One of the causes of bacterial contamination in fishery products is the cleanliness of workers during the handling process, which can lead to significant shrimp contamination. This includes direct contact with shrimp (e.g., slicer, conveyor belt, and knife), indirect contact (e.g., floor, drain, and wall), personal contact (e.g., aprons, gloves, and boots), and other factors (e.g., air, ice, and water) [26]. This contact results in bacteria on conveyor belts, water samples from fish landing sites, and fish sorting machines spreading these bacteria throughout the fish processing area. This causes contamination during the initial stages before the distribution process is conducted [54]. Neglecting basic hygiene practices is a significant factor in the occurrence of cross-contamination of products [55].

In addition to water, ice, and team member behavior, several other factors influence the risk of microbial contamination in seafood products, including where the seafood commodities are caught, the species cultivated, farming practices, processing, and cultural habits. Microbiological hazards are associated with poor hygiene practices and domestic waste disposal. For example, waste contamination carries environmental pollutants into river water [56]. Contamination of seafood products in processing facilities can occur during all stages of processing, including transportation, washing with contaminated water, peeling, filleting, contact with ice used for preservation, cutting boards, knives, and trays [26]. Therefore, adequate sanitation conditions during the processing process, including handling hygiene, cleanliness of equipment such as tables and other tools, and the use of clean and chlorinated water throughout all processing stages, are crucial to avoid cross-contamination [57].

Cross-contamination associated with raw and processed food through the contact surfaces of these objects is considered a significant hazard, and several authors have reported the presence of *Salmonella* on stainless steel surfaces [9]. Salmonellosis incidents resulting from consuming seafood products have been a concern for public health agencies in several countries owing to a significant increase in seafood consumption, especially raw products, thus raising the risk of pathogen exposure, particularly among vulnerable groups such as the elderly,

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pregnant women, and infants [58], [59]. *Shigella* and *Salmonella* are also groups of fecal coliform bacteria that can be detected in tiger shrimp. The presence of these bacteria is closely related to poor water conditions, allowing bacterial contamination from polluted water used for shrimp washing during unloading activities at fishing ports. The washing water comes from the Mahakam River, which is densely populated by human activities, compounded by ice cubes, the water source of which is also unknown for shrimp distribution to the Barong Tongkok Market.

4- Conclusions

Shrimp samples contain various types of bacterial contaminants that can degrade the quality of tiger shrimp that have undergone the distribution process. The bacteria found included coliform bacteria, *Pseudomonas* sp., *Aeromonas* sp., *Staphylococcus aureus*, *Proteus* sp., *Escherichia coli*, *Salmonella/Shigella*, *and Klebsiella* sp. Three types of bacteria were not found in good-quality shrimp: *E. coli*, *Klebsiella* sp., and *Salmonella/Shigella*. Decayed shrimp, namely shrimps 2 and 3, were dominated by six types of bacteria, namely *Pseudomonas* sp., *Aeromonas* sp., *Salmonella/Shigella*, *Klebsiella* sp., *E. coli*, and *S. aureus*, and the highest number of each type was found in shrimp 3, which was of poor quality. The presence of bacterial contaminants in the shrimp was attributed to three main factors: the use of unclean equipment and materials, the behavior of workers who did not pay attention to hygiene, and poor environmental conditions during the tiger shrimp handling process. These results suggest that the long distribution process of tiger prawns coupled with poor handling leads to bacterial contamination, resulting in reduced quality in the form of tiger prawn spoilage, which requires a particular strategy to minimize bacterial contamination.

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Table 1- The characteristics of bacterial colonies on DCLS agar medium [22]

Microorganism	**Specification**	**Characteristic reaction**
Salmonella/Shigella	Good growth	Colourless/pale pink colonies
Proteus vulgaris	Moderate growth	Colourless/pink colonies and small formation of a precipitate
Escherichia coli	Inhibited	Pink-red colonies

Table 2- Morphology of bacterial colonies on CLED Agar medium [22]

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Type of bacteria	Growth morphology
Escherichia coli	Colonies are dark yellow, and the medium is yellow
Klebsiella	Colonies are yellow to whitish-blue, slimy, and the medium is yellowish
Proteus	Colonies are translucent blue, and the medium changes to greenish-blue to moderate blue
Pseudomonas aeruginosa	Green colonies with a characteristic and rough surface, and the edges are coarse
Enterococcus	The colonies are yellow with a diameter of 0.5 mm, and the medium is yellow
Salmonella	The colonies are smooth and blue in colour
Staphylococcus aureus	The colonies are dark yellow, uniform in colour, and the medium turns yellow

Table 3- Bacterial count based on total plate count and Most Probable Number (MPN)

Sample	**Total bacteria (CFU/g)**	**Coliform (MPN/ g)**	**Coliform Fecal (MPN/g)**	***E. coli***
Shrimp 1	$58.5 \times 10^5 \pm 7.78$	>21	0	Negatives
Shrimp 2	$86.0 \times 10^5 \pm 4.24$	>21	0	Negatives
Shrimp 3	$30.25 \times 10^6 \pm 21.08$	>1100	0	Negatives

Table 4- Identification of potential risks for the deterioration of post-harvest quality of tiger shrimp

Aspects	**Code**	**Risk Potential**
Equipment and Materials	1	Dirty baskets
	2	Ice cubes type
	3	Using river water in the tub containing shrimp
	4	Not using running water for rinsing
	5	Dirty carts
	6	Dirty containers
	7	Dirty buckets
	8	Dirty cool box
	9	A small amount of ice cubes is added
	10	Dirty display table
	11	Insufficient ice was provided during the display
	12	The table is not provided with partitions between products
	13	Dirty weighing scale
Worker Behavior	14	Not using gloves
	15	Not wearing boots
	16	Washing on wet floors
	17	Cleaning the floor while the cool box containing shrimp is open
	18	Disposing of shrimp washing water near the Cool box containing shrimp to be shipped
	19	Not adding ice to the cool box
	20	Disposing of melted ice from the cool box onto the cool box storage area floor.

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Environmental Conditions	21	Dirty and wet floor
	22	The washing area is located near the waste disposal
	23	The loading process is carried out near the drainage ditch
	24	Rats were found at the loading location
	25	There is a lot of garbage in the unloading area
	26	The display table is not dry, and there are still residues, such as fish scales that are not cleaned thoroughly and water puddles in the corners of the table

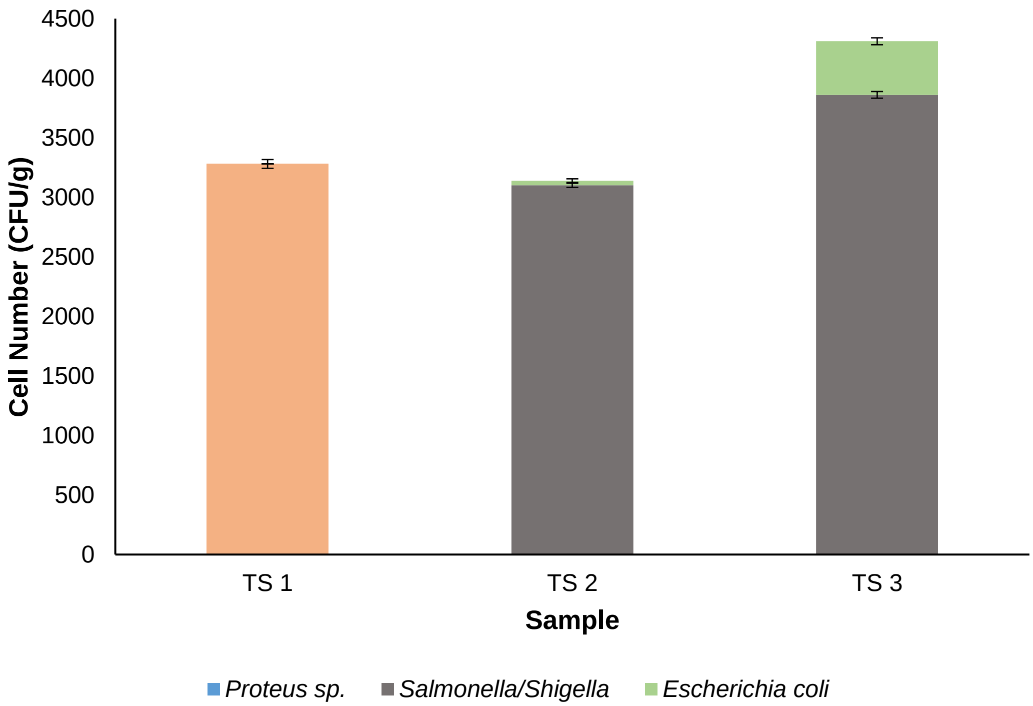

Figure 1- Types of bacteria found in shrimp samples after being grown in DCLS medium.

[DOI: 10.22034/FSCT.21.155.103]

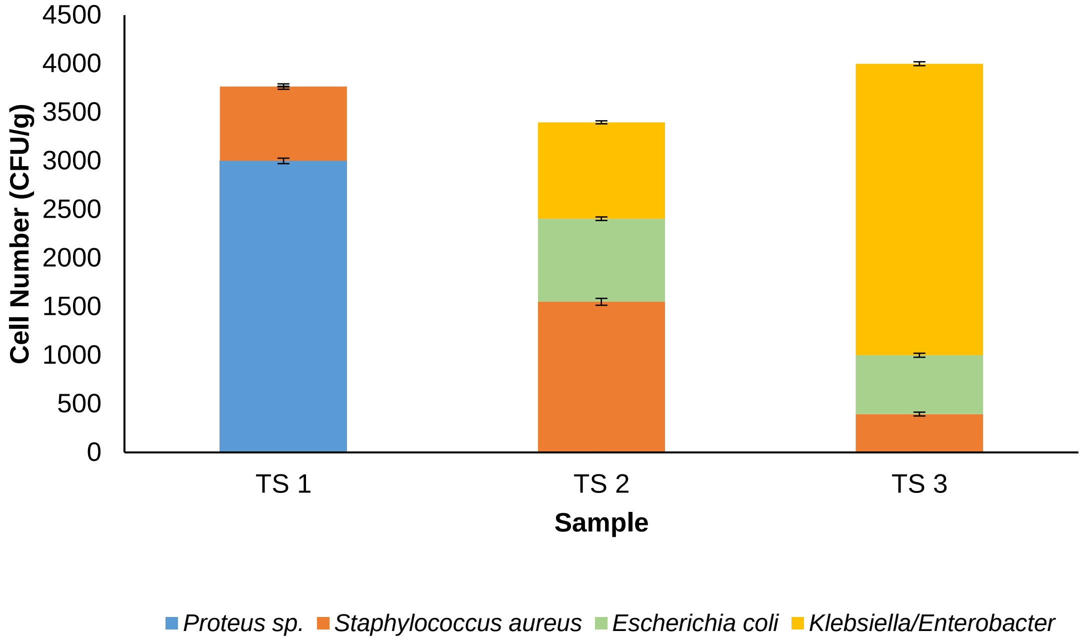

Figure 2- Types of bacteria found in shrimp samples after being grown in CLED medium.

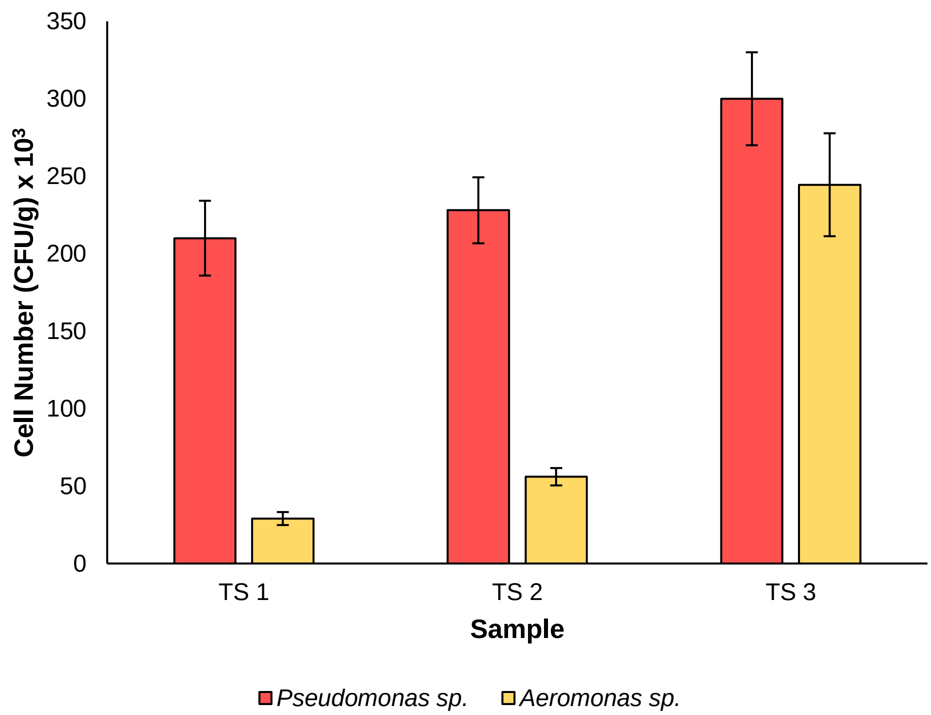

Figure 3- *Pseudomonas* sp. and *Aeromonas* sp. were found in shrimp samples after being grown on GSP medium.

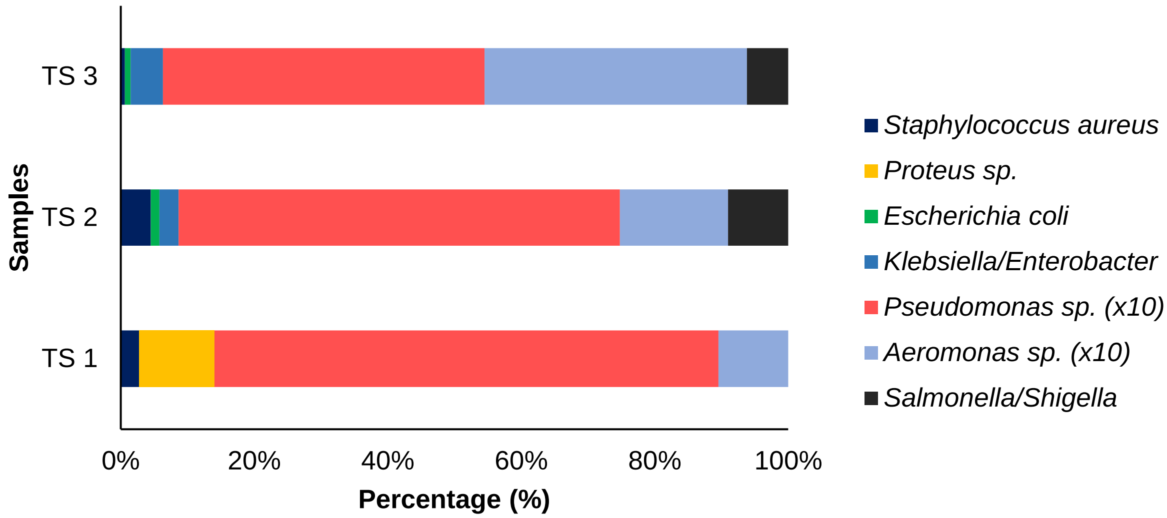

Figure 4- Distribution of bacteria types in each shrimp sample.

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