

## BACTERIAL COUNT COMPARISON IN THE HEPATOPANCREAS AND CULTURE MEDIA OF *Litopenaeus vannamei* AS HEALTH CONTROL

Perbandingan Jumlah Bakteri pada Hepatopankreas dan Media Budidaya Post Larva Udang Vanname (*Litopenaeus Vannamei*) sebagai Kontrol Kesehatan

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(Received January 13<sup>th</sup> 2026; Accepted February 19<sup>th</sup> 2026)

### ABSTRACT

*Vaname shrimp (Litopenaeus vannamei) is a leading Indonesian aquaculture commodity with high productivity and rapid growth. However, various obstacles arise in vaname shrimp cultivation, including environmental damage, declining water quality, competition for land use, and disease. Diseases that easily arise are generally caused by bacteria, viruses, and fungi. Disease prevention in shrimp can be done by controlling shrimp health using the Total Plate Count (TPC) method. Total Plate Count (TPC) is a fast, simple, inexpensive, and accurate method for diagnosing total bacteria. This method is carried out by counting the number of live bacteria in water samples and shrimp hepatopancreas. This research was carried out for 30 days from June 10, 2025, to July 10, 2025, at the National Research and Innovation Agency (BRIN), North Lombok. This study aims to determine the technique of controlling the health of vaname shrimp (L. vannamei) at the post-larval stage using the Total Plate Count (TPC) method. This study uses descriptive and statistical methods. The results obtained in this study showed that the values of the shrimp hepatopancreas and water samples showed a high bacterial range, but still within the optimal range. The high bacterial density in the water and shrimp hepatopancreas samples could be caused by poor water quality and the quality of the bacterial media, which began to deteriorate with increasing incubation time. This, in turn, led to an increase in organic matter in the media.*

*Keywords: Bacteria, Total Plate Count (TPC), Vaname Shrimp*

### ABSTRAK

Udang vaname (*Litopenaeus vannamei*) merupakan komoditas unggulan budidaya perikanan Indonesia yang memiliki produktivitas cukup tinggi dan pertumbuhan pesat. Namun, munculnya berbagai kendala pada budidaya udang vaname antara lain, kerusakan lingkungan, penurunan kualitas air, persaingan pemanfaatan lahan dan penyakit. Penyakit yang mudah timbul umumnya disebabkan oleh bakteri, virus dan jamur. Pencegahan penyakit pada udang dapat dilakukan melalui pengontrolan kesehatan udang dengan menggunakan metode *Total Plate Count (TPC)*. *Total Plate Count (TPC)* adalah metode diagnosis total bakteri yang cepat,

sederhana, murah dan memberikan hasil yang akurat. Metode ini dilakukan dengan melakukan perhitungan jumlah bakteri hidup pada sampel air dan hepatopankreas udang. Praktik Kerja Lapangan ini dilaksanakan selama 30 hari pada 10 Juni 2025 sampai 10 Juli 2025 di Badan Riset dan Inovasi Nasional (BRIN), Lombok Utara. Penelitian ini bertujuan untuk mengetahui teknik pengontrolan kesehatan udang vaname (*L. vannamei*) pada stadia post larva dengan metode *Total Plate Count* (TPC). Penelitian ini menggunakan metode deskriptif dan statistik. Hasil didapatkan pada penelitian ini adalah nilai pengecekan dengan hepatopankreas udang dan air sampel menunjukkan kisaran bakteri yang tinggi namun masih dalam kisaran optimal. Tingginya kepadatan bakteri pada sampel air dan hepatopankreas udang dapat disebabkan oleh kualitas air yang tidak baik dan kualitas media tebar bakteri yang mulai memburuk yakni dengan semakin lama waktu inkubasi yang digunakan. sehingga, menyebabkan peningkatan bahan organik pada media.

Kata Kunci: Bakteri, *Total Plate Count* (TPC), Udang Vaname

## INTRODUCTION

Vaname shrimp (*Litopenaeus vannamei*) is a leading Indonesian aquaculture commodity with relatively high productivity and rapid growth. This is because this type of shrimp has several advantages over other shrimp, including rapid growth, a relatively short maintenance period, and high economic value (Rivaldo *et al.*, 2021). According to the Directorate General of Aquaculture, Ministry of Maritime Affairs and Fisheries (KKP) in 2021, national vaname shrimp production in 2019-2020 reached 856,753 tons and is targeted to reach 2 million tons per year by 2024. However, in recent years, shrimp farming has encountered various obstacles, including environmental damage, declining air quality, competition for land use, and disease. In 2022, clinical supervision and observations at the Fish Quarantine Center for Quality Control and Safety of Fishery Products (BKIPM) found a high mortality rate caused by disease, reaching 5.5% (Hamjah *et al.*, 2024).

Diseases in vaname shrimp can be caused by bacteria, viruses and fungi. Disease in shrimp can occur if there is an imbalance between shrimp and pathogens. In shrimp farming, diseases that easily arise are generally caused by the bacteria *Vibrio* sp. (Amelia *et al.*, 2020). According to Panjaitan *et al.* (2015) and Ningrum & Diniariwisan (2024), the shrimp stage most susceptible to disease is the post-larva stage. In this stage, the shrimp's immune system is not yet fully developed, making them more susceptible to various diseases, including those caused by bacteria, viruses, and parasites. Post-Larva (PL) is the final stage before the shrimp enter the juvenile phase, where they resemble adult shrimp without significant changes in shape (Rasuliyanasari & Diniariwisan, 2024). At this stage, the shrimp have complete body organs and are ready for culture.

Shrimp disease prevention can be achieved by monitoring shrimp health using the Total Plate Count (TPC) method. TPC is a rapid, simple, inexpensive, and accurate method for diagnosing total bacteria. This method involves counting the number of live bacteria in water samples and the shrimp's hepatopancreas. A high TPC value indicates a high level of contamination in the shrimp (Syafrina *et al.*, 2022). So that later by carrying out health control using the Total Plate Count (TPC) method, it is hoped that will be possible to identify and detect bacteria in vaname shrimp farming. Therefore, the aim of this research activity is to determine the bacterial count comparison in the hepatopancreas and media culture using the TPC method as a technique for controlling health of vaname post-larva stage at the National Research and Innovation Agency (BRIN), North Lombok.

## METHODS

### Time and Place

This activity was carried out for 30 days on June 10<sup>th</sup> - July 10<sup>th</sup>, 2025. This activity was carried out at the Marine and Land Bioindustry Research Center, National Research and Innovation Agency (BRIN), North Lombok.

### Tools and Materials

The tools used are aerator, autoclave, bunsen, petri dish, scissor, hot plate, incubator, microtube, micro scraper, spreader, analytic scale, vortex and yellow tip. The materials used in this activity are culture sample water, alcohol, aluminum foil, distilled water, shrimp hepatopancreas, Phosphate Buffered Saline (PBS) solution, latex, Thiosulfate Citrate Bile Salt Sucrose agar (TCBS) media, Marine Agar (MA) spiritus media and tissue.

### Research Method

The method used is a descriptive and statistical method with treatments tested using different samples, namely shrimp hepatopancreas and vaname shrimp cultivation water samples. The samples used three replications each with the type of media: Thiosulfate Citrate Bile Salt Sucrose Agar (TCBS) and Marine Agar (MA) with different incubation times of 24 hours and 48 hours. Furthermore, the data obtained was processed using Microsoft Excel accompanied by a literature review.

### Research Procedure

#### TPC Media Preparation

Media preparation is the process of designing, compiling, and producing bacterial growth media for use in microbiology. This process involves several steps, including sterilizing equipment, determining the media to be used, selecting materials, and dissolving the media. The purpose of media preparation is to provide a suitable environment for bacterial growth and reproduction. According to Wati (2018), media is a material consisting of a mixture of nutrients that are useful for cultivating microbes. Media frequently used in shrimp health monitoring using the Total Plate Count (TPC) method are Marine Agar (MA) and Thiosulfate Citrate Bile Salts Sucrose (TCBS) media. Marine Agar (MA) is a growth medium used to support the growth of marine bacteria. This medium contains nutrients such as peptone and yeast extract, which can support the growth of bacteria used in observation. Furthermore, Thiosulfate Citrate Bile Salts Sucrose (TCBS) media is a selective medium used to isolate and identify bacteria of the *Vibrio* genus, which are commonly found in marine waters and shrimp farming, making it suitable for microbiological research (Lilatussyifa *et al.*, 2020).

#### Water & Hepatopancreas Sample Preparation

- Water Sample

The water was taken from the culture tanks using small bottles. The conditions in the tanks were monitored, ensuring that the sampling point was representative of the overall condition of the tanks. Water and shrimp samples were collected once a week for the three weeks of observation.

- Hepatopancreas Sample

Shrimp hepatopancreas samples were taken directly during testing to ensure freshness. The vaname shrimp were collected while still alive and transported to the laboratory. The collected shrimp were then dissected, and the hepatopancreas was removed using sterile scissors and placed in an eppendorf flask. The eppendorf flask was weighed before and after inserting the hepatopancreas to determine the shrimp's hepatopancreas weight. Weighing was performed using an analytic scale. The hepatopancreas is an organ composed of liver and

pancreatic cells. In the digestive system, the hepatopancreas functions to produce digestive enzymes and regulate metabolism in the shrimp.

### **Sample Dilution**

Sample dilution is the process of reducing the concentration of a solute in a solution by adding a solvent. Sample dilution is performed to reduce the density of microorganisms in the sample, allowing for accurate colony counts and avoiding colony clumping, which can complicate counting.

- **Water Sample Dilution**

0.1 ml water sample was taken using a micropipette and then inserted into an eppendorf tube filled with 1 ml of Phosphate Buffered Saline (PBS) solution as a solvent. The sample was vortexed so that the sample water and Phosphate Buffered Saline (PBS) solution were mixed evenly or homogeneously. After that, 0.1 ml was taken from the first Eppendorf tube with a micropipette and transferred to the second Eppendorf tube (101) filled with 1 ml of PBS solution and then vortexed again.

- **Hepatopancreas Sample Dilution**

The shrimp hepatopancreas that had been taken was placed in an eppendorf tube and ground using a micro pestle. After that, 1 ml of Phosphate Buffered Saline (PBS) solution was added and vortexed to mix the shrimp hepatopancreas sample and PBS solution evenly or homogeneously. Next, 0.1 ml of the sample was taken with a micropipette and transferred to a second Eppendorf tube (101) filled with 1 ml of PBS solution and then vortexed again.

### **Bacterial Inoculation**

Bacterial inoculation is performed using a bacterial culture method using universal or selective media. The method used is the surface plate method using the spread plate method. Bacterial isolation is performed in a Laminary Air Flow to avoid contamination. The media used are Marine Agar (MA) to identify common bacteria and Thiosulfate Citrate Bile Salts Sucrose (TCBS) to identify vibrio bacteria.

### **Bacterial Count**

The number of bacterial growths was counted using a colony counter, recorded, and calculated using a formula. Bacterial density data was recorded in CFU/ml. Bacterial concentrations were measured according to the National Standards Agency (SNI 7388:2009), with the maximum TPC limit for shrimp being  $3 \times 10^5$  CFU/g, with a colony between 30-300. Bacterial calculation formula according to Fatayati *et al.* (2023) are as follows:

$$\text{TPC} = \frac{\text{Number of Bacterial Colony} \times \text{Dilution Factor}}{(\text{Sample Volume} / \text{Stocking Volume})}$$

### **Water Quality Measurement**

Water quality measurement is a crucial process to ensure good quality water for vaname shrimp farming. The water quality parameters for vaname shrimp culture media must be optimal. The parameters influencing cultivation are pH, temperature, and salinity. Water quality measurements using a water checker were conducted once a week for a three-week observation period. The one-week sampling intervals were intended to determine changes in water quality in the cultivation tanks and their impact on the morphology of the vaname shrimp.

## **RESULTS**

The data of hepatopancreas weight and eppendorf using analytical scales are presented in Table 1 as follows:

Table 1. Hepatopancreas Weight

Week	Hepatopancreas		
	Initial Weight (g)	Final Weight (g)	Organ Weight (g)
1	0.7 g	1.7 g	1.0 g
2	0.8 g	1.9 g	1.1 g
3	0.7 g	1.8 g	1.1 g

Based on the results of inoculation of shrimp hepatopancreas samples and cultured water samples carried out with a 101 dilution, vibrio bacteria and common bacteria were found on Thiosulfate Citrate Bile Salts Sucrose (TCBS) and Marine Agar (MA) media. Colonies grew like spots around the media. From the treatment on TCBS media, green and yellow bacteria were found, while bacteria on MA media were white. The results of the observations obtained bacteria in Figure 1 are as follows:

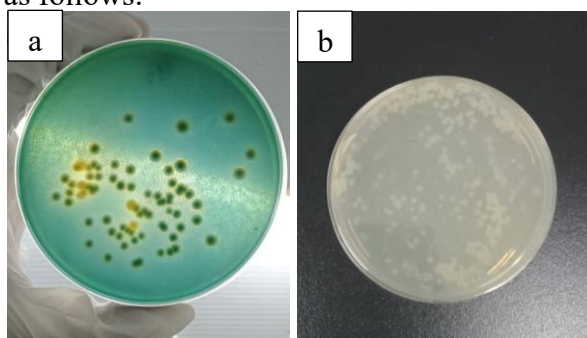


Figure 1. TCBS Bacterial Media (a); MA Bacterial Media (b)

Bacteria growing on TCBS and MA media, respectively, after 24 and 48 hours of incubation. The differences in the number of vibrio and common bacteria are shown in the following table:

Table 2. Total Bacterial Count of Hepatopancreas

Total Bacterial Count						
Week	Repetition	Colony Number (1 x 10 <sup>1</sup> )	Total Bacterial/mL	Total Bacterial/g	Average	STDEV
24 hours	1	300	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>	2.57 x 10 <sup>5</sup>	7.51 x 10 <sup>4</sup>
	2	170	2 x 10 <sup>5</sup>	2 x 10 <sup>5</sup>		
	3	300	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>		
2	1	180	2 x 10 <sup>5</sup>	2 x 10 <sup>5</sup>	1.66 x 10 <sup>5</sup>	5.34 x 10 <sup>4</sup>
	2	211	2 x 10 <sup>5</sup>	2 x 10 <sup>5</sup>		
	3	107	1 x 10 <sup>5</sup>	1 x 10 <sup>5</sup>		
3	1	215	2 x 10 <sup>5</sup>	2 x 10 <sup>5</sup>	2.11 x 10 <sup>5</sup>	7.96 x 10 <sup>4</sup>
	2	289	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>		
	3	130	1 x 10 <sup>5</sup>	1 x 10 <sup>5</sup>		
48 hours	1	300	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>	2.94 x 10 <sup>5</sup>	1.04 x 10 <sup>4</sup>
	2	282	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>		
	3	300	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>		
2	1	216	2 x 10 <sup>5</sup>	2 x 10 <sup>5</sup>	2.46 x 10 <sup>5</sup>	2.74 x 10 <sup>4</sup>
	2	270	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>		
	3	251	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>		
3	1	297	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>	3.14 x 10 <sup>5</sup>	3.21 x 10 <sup>4</sup>

Total Bacterial Count						
Week	Repetition	Colony Number (1 x 10 <sup>1</sup> )	Total Bacterial/mL	Total Bacterial/g	Average	STDEV
	2	351	4 x 10 <sup>5</sup>	4 x 10 <sup>5</sup>		
	3	294	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>		

Table 3. Total Vibrio Count of Hepatopancreas

Total Vibrio Count						
Week	Repetition	Colony Number 1 x 10 <sup>1</sup>	Total Bacterial/mL	Total Bacterial/g	Average	STDEV
24 hours	1	300	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>	2.98 x 10 <sup>5</sup>	2.41 x 10 <sup>4</sup>
	2	321	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>		
	3	273	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>		
2	1	49	5 x 10 <sup>4</sup>	5 x 10 <sup>4</sup>	2.30 x 10 <sup>4</sup>	2.27 x 10 <sup>4</sup>
	2	7	7 x 10 <sup>3</sup>	7 x 10 <sup>3</sup>		
	3	13	1 x 10 <sup>4</sup>	1 x 10 <sup>4</sup>		
3	1	48	5 x 10 <sup>4</sup>	5 x 10 <sup>4</sup>	4.70 x 10 <sup>4</sup>	2.85 x 10 <sup>4</sup>
	2	18	2 x 10 <sup>4</sup>	2 x 10 <sup>4</sup>		
	3	75	8 x 10 <sup>4</sup>	8 x 10 <sup>4</sup>		
48 hours	1	269	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>	2.54 x 10 <sup>5</sup>	6.15 x 10 <sup>4</sup>
	2	306	3 x 10 <sup>5</sup>	3 x 10 <sup>5</sup>		
	3	186	2 x 10 <sup>5</sup>	2 x 10 <sup>5</sup>		
2	1	52	5 x 10 <sup>4</sup>	5 x 10 <sup>4</sup>	2.50 x 10 <sup>4</sup>	2.38 x 10 <sup>4</sup>
	2	7	7 x 10 <sup>3</sup>	7 x 10 <sup>3</sup>		
	3	16	2 x 10 <sup>4</sup>	2 x 10 <sup>4</sup>		
3	1	36	4 x 10 <sup>4</sup>	4 x 10 <sup>4</sup>	3.73 x 10 <sup>4</sup>	2.00 x 10 <sup>4</sup>
	2	18	2 x 10 <sup>4</sup>	2 x 10 <sup>4</sup>		
	3	58	6 x 10 <sup>4</sup>	6 x 10 <sup>4</sup>		

The bacterial density obtained on each medium then needs to be averaged each week for 24-hour and 48-hour incubation periods. The total bacterial count is shown in the following graph:

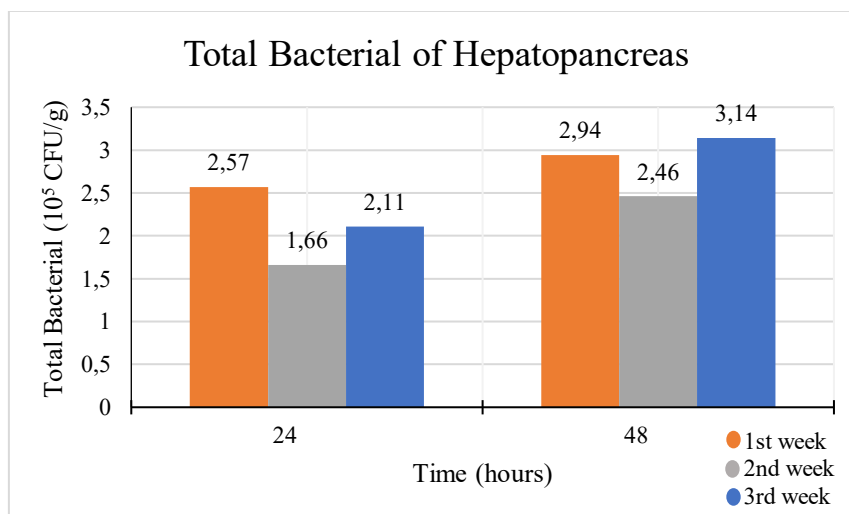


Figure 2. Total Bacterial Count Hepatopancreas Graph

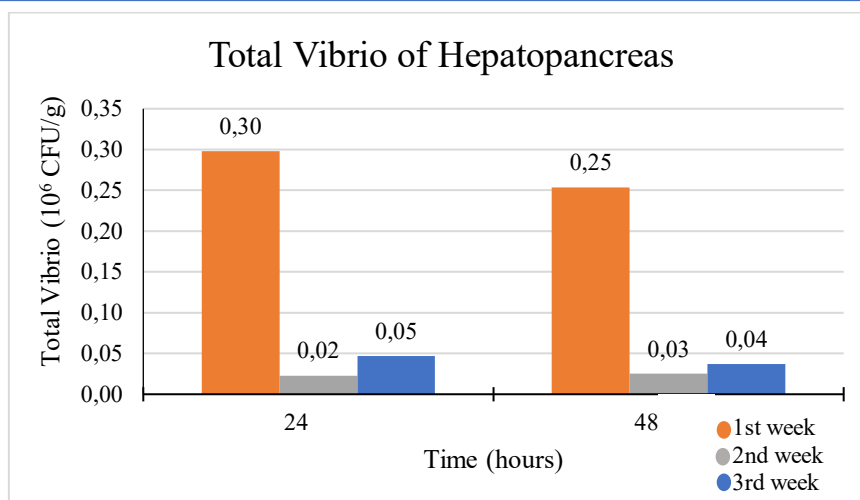


Figure 3. Total Vibrio Count Hepatopancreas Graph

Table 4. Total Bacterial Count of Water

Total Bacterial Count					
Week	Repetition	Colony Number $1 \times 10^1$	Total Bacterial/mL	Average	STDEV
24 hours 1	1	121	$1.21 \times 10^5$	$1.21 \times 10^5$	$1.45 \times 10^4$
	2	135	$1.35 \times 10^5$		
	3	106	$1.06 \times 10^5$		
2	1	300	$3.00 \times 10^5$	$2.47 \times 10^5$	$6.61 \times 10^4$
	2	268	$2.68 \times 10^5$		
	3	173	$1.73 \times 10^5$		
3	1	TMTC			
	2	TMTC			
	3	TMTC			
48 hours 1	1	135	$1.35 \times 10^5$	$1.49 \times 10^5$	$1.40 \times 10^4$
	2	149	$1.49 \times 10^5$		
	3	163	$1.63 \times 10^5$		
2	1	300	$3.00 \times 10^5$	$2.94 \times 10^5$	$9.81 \times 10^3$
	2	283	$2.83 \times 10^5$		
	3	300	$3.00 \times 10^5$		
3	1	TMTC			
	2	TMTC			
	3	TMTC			

Table 5. Total Vibrio Count of Water

Total Vibrio Count					
Week	Repetition	Colony Number $1 \times 10^1$	Total Bacterial/mL	Average	STDEV
24 hours 1	1	19	$1.90 \times 10^4$	$3.67 \times 10^4$	$2.89 \times 10^4$
	2	21	$2.10 \times 10^4$		
	3	70	$7.00 \times 10^4$		
2	1	15	$1.50 \times 10^4$	$1.27 \times 10^4$	$6.81 \times 10^3$
	2	18	$1.80 \times 10^4$		

Total Vibrio Count					
Week	Repetition	Colony Number $1 \times 10^1$	Total Bacterial/mL	Average	STDEV
	3	5	$5.00 \times 10^3$		
3	1	TFTC			
	2	TFTC			
	3	TFTC			
48 hours 1	1	15	$1.50 \times 10^4$	$2.70 \times 10^4$	$2.08 \times 10^4$
	2	15	$1.50 \times 10^4$		
	3	51	$5.10 \times 10^4$		
2	1	16	$1.60 \times 10^4$	$1.37 \times 10^4$	$2.52 \times 10^3$
	2	14	$1.40 \times 10^4$		
	3	11	$1.10 \times 10^4$		
3	1	TFTC			
	2	TFTC			
	3	TFTC			

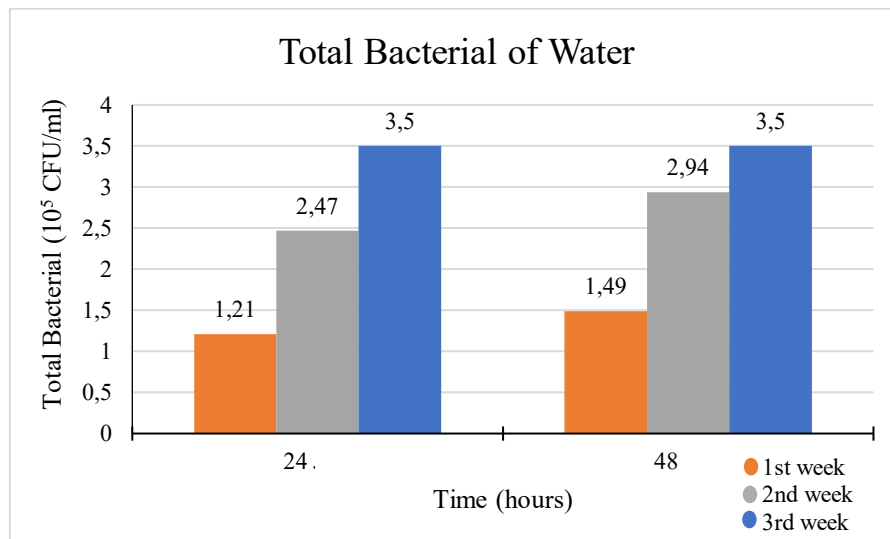


Figure 4. Total Bacterial Count of Water Graph

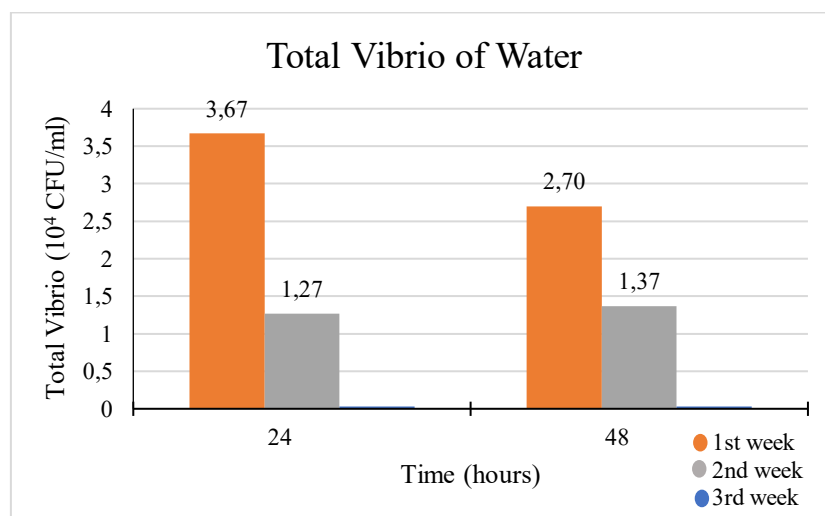


Figure 5. Total Vibrio Count of Water Graph

Based on the results of observations of the presence of bacteria using agar media, it is linked to several environmental qualities including measuring temperature, salinity and pH factors. The results of checking the water quality of vaname shrimp cultivation during 3 weeks of observation based on the 2016 National Quality Standards SNI are presented in Table 6, as follows:

Table 6. Water Quality Measurement

No	Parameter	Unit	Week 1	Week 2	Week 3	Average	Quality Standard
1.	Salinity	ppt	32.28	33.54	32.51	32.78	30-33
2.	Temperature	°C	25.8	25.6	25.6	25.7	25-31
3.	pH	-	8.45	8.23	8.61	8.43	7.5-8.5

## DISCUSSION

### Total Bacterial Count of Hepatopancreas

Bacterial growth increased with the longer incubation time used. In the first week of 24-hour incubation, the number was  $2.57 \times 10^5$  CFU/gr and in the 48-hour incubation, the number was  $2.94 \times 10^5$  CFU/gr. In the second week of 24-hour incubation, the number was  $1.66 \times 10^5$  CFU/gr and increased to  $2.46 \times 10^5$  CFU/gr after 48 hours of incubation. Meanwhile, in the third week of 24-hour incubation, the number was  $2.11 \times 10^5$  CFU/gr and in the 48-hour incubation, the number was  $3.14 \times 10^5$  CFU/gr. The increase in bacterial density can be attributed to the longer incubation period, which was 48 hours. Incubating bacteria for 48 hours allows for a longer exponential growth rate compared to 24 hours, resulting in a higher bacterial population. During this phase, bacteria can divide continuously, resulting in a significant increase in the population. This is in line with the opinion of Andayani *et al.* (2024), who stated that the increase in bacterial count was caused by the quality of the culture medium, which began to deteriorate after 48 hours of incubation, resulting in an increase in organic matter. Therefore, the total number of common bacteria in Marine Agar (MA) media with shrimp hepatopancreas samples increased each week, specifically after 48 hours of incubation.

### Total Vibrio Count of Hepatopancreas

The results obtained from the calculation of total Vibrio hepatopancreas bacteria in whiteleg shrimp showed that the bacterial density decreased with the longer incubation time used. In the first week of 24-hour incubation, the number was  $0.30 \times 10^6$  CFU/gr and decreased in the 48-hour incubation to  $0.25 \times 10^6$  CFU/gr. The bacterial density in the second week of 24-hour incubation was  $0.02 \times 10^6$  CFU/gr and increased in the 48-hour incubation to  $0.03 \times 10^6$  CFU/gr. Meanwhile, in the third week of 24-hour incubation, the number was  $0.05 \times 10^6$  CFU/gr and decreased to  $0.04 \times 10^6$  CFU/gr after 48 hours of incubation. The increase and decrease in the density of vibrio bacteria in the hepatopancreas of shrimp using TCBS media may be due to the media's high nutrient content, which can support the growth of vibrio bacteria. However, in the first and second weeks, with an incubation period of 48 hours, bacterial density decreased. This could be due to the bacteria's diameter increasing with increasing incubation time, which causes them to divide and grow, thus decreasing bacterial density. This is in line with the opinion of Martsiningsih *et al.* (2023), that variations in incubation time for 6-48 hours can affect the results of bacterial diameter formation so that it can become wider or larger and reduce the level of bacterial density.

### Total Bacterial Count of Water

Bacterial growth in the 1st week of 24-hour incubation was  $1.21 \times 10^5$  CFU/ml and increased in the 48-hour incubation to  $1.49 \times 10^5$  CFU/ml. In the 2nd week of 24-hour

incubation, it was  $2.47 \times 10^5$  CFU/ml, increasing in the 48-hour incubation to  $2.94 \times 10^5$  CFU/ml. Meanwhile, in the 3rd week, it increased beyond the normal limit in the 24-hour and 48-hour incubation, reaching more than  $3 \times 10^5$ , which caused TMTC (Too Many to Count). However, bacterial growth in weeks 1 and 2 showed that bacterial density remained within the normal range for intensive indoor cultivation, thus remaining within the optimal range for further cultivation. Cultivation activities such as feeding and water quality can contribute to the bacterial population decline. Another impact can be caused by contamination during the spread of bacterial growth media, which can cause some pathogenic microorganisms to exceed the limit. This aligns with the opinion of Siregar & Huda (2019), who stated that sterilization is necessary when spreading media because bacterial density can be caused by contamination from other bacteria during the spread or inoculation process. The increase in other bacteria can also be caused by the choice of Marine Agar (MA) media which allows unwanted bacteria to be carried along and grow together with the bacteria to be tested so that the number of colonies cannot be counted.

### **Total Vibrio Count of Water**

TCBS media is a selective and differential media for the growth of vibrio bacteria. This media contains bile salts that function to inhibit the growth of microorganisms other than the vibrio sp. bacterial group. The results of the data analysis obtained where the total culture water test data for vibrio sp. bacteria in the test sample meets the requirements for shrimp cultivation, which is below  $3 \times 10^5$  CFU/ml with a value of  $3.67 \times 10^5$  CFU/ml in the first week with a 24-hour incubation period and a decrease of  $2.70 \times 10^5$  CFU/ml in 48 hours of incubation. In the second week of 24-hour incubation, the count was  $1.27 \times 10^5$  CFU/ml, and in the 48-hour incubation, the count was  $1.37 \times 10^5$  CFU/ml. Meanwhile, in the third week, the count was TFTC (Too Few to Count), meaning the bacteria grew too little to be counted on the media. In the 24-hour and 48-hour incubations, the count was only  $0.03 \times 10^5$  CFU/ml. This condition can be caused by the good water quality of the vaname shrimp cultivation water, which prevents the growth of many vibrio bacteria, and it can also be caused by the agar media used during stocking. According to Amrullah et al. (2023), the decrease in total Vibrio bacteria in water samples can be caused by the condition of the bacterial growth media, the temperature of the agar media being hot or cold during the spreading process, the incubation time being too long, and the natural characteristics of the microorganisms.

### **Water Quality**

Water quality control data indicates a relatively modest change in water temperature. However, the cultivation temperature is relatively low, at  $25.7^\circ\text{C}$ , compared to the Indonesian National Standard (SNI) of  $28-31^\circ\text{C}$ . The optimal temperature for shrimp farming is  $26-30^\circ\text{C}$  (Wahyuni *et al.*, 2024). A temperature drop that is still tolerable for organisms will be followed by a decrease in metabolic rate and photosynthetic activity of natural food (phytoplankton). Likewise, water temperature will affect survival, morphological growth, reproduction, behavior, the molting process, and metabolism of shrimp. According to Arsad *et al.* (2018), the optimal temperature range for vaname shrimp growth is  $28-31^\circ\text{C}$ , and they grow well at  $24-34^\circ\text{C}$ . Therefore, the cultivation water temperature is still within the normal range for further cultivation.

The next water quality factor to consider is the water's acidity (pH), which is related to the dissolution of certain compounds. The degree of acidity is often interpreted as the water condition that can be acidic, alkaline and neutral (Murtadha *et al.*, 2025). The pH value obtained for cultivation was 8.43, which meets the 2016 SNI quality standard of 7.5-8.5. According to Supriatna *et al.* (2020), the increase in pH value that occurs can be caused by the lower  $\text{CO}_2$  content in the cultivation tank because it is used by plankton for photosynthesis

which produces O<sub>2</sub> interacting with H<sub>2</sub>O to release OH<sup>-</sup> ions which cause alkaline waters and a tendency for pH to rise, so that the pH level of vaname shrimp cultivation water fluctuates according to the ongoing photosynthesis and respiration activities.

The salinity value obtained was 32.78, which is in accordance with the 2016 SNI quality standard, which is 30-33 ppt. Salinity values can fluctuate significantly. This can be due to unpredictable weather changes. A decrease in salinity values can occur due to rain, which causes salinity to decrease, resulting in changes in salt concentration due to the presence of freshwater. Meanwhile, an increase in salinity values can be caused by environmental factors and inappropriate cultivation media management, including high evaporation, low rainfall, and ocean currents that can spread salt.

### CONCLUSION

The conclusion of this activity is that health control in shrimp farming using the Total Plate Count (TPC) method and water quality monitoring is optimal for assessing shrimp health. Testing values using shrimp hepatopancreas and water samples showed a high bacterial count, but still within the optimal range. The high bacterial density in water samples and shrimp hepatopancreas may be caused by poor water quality and the deteriorating quality of the bacterial media, which increases with increasing incubation time.

### ACKNOWLEDGEMENT

The author would like to express his gratitude for the successful implementation of this project thanks to the support of various parties. This includes the Aquaculture Study Program University of Mataram and the National Research and Innovation Agency (BRIN) in North Lombok, where the project took place. The author also thanked the field supervisors and supervising lecturers who assisted throughout the project.

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