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Cultivation Techniques of *Thalassiosira* sp. at PT. Bumi Persada Gemilang Shrimp Hatchery, Central Pertiwi Bahari, North Galesong, South Sulawesi

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Abstract
The maintenance of larvae in hatcheries must consider seed quality, one aspect of which involves implementing standard operating procedures for appropriate feeding. Thalassiosira sp. has a complex nutritional content that supports growth, making it selected as a natural feed for vaname shrimp larvae. Objective. This final project aims to describe the technique of culturing Thalassiosira sp. and to identify obstacles as well actions that can be taken during the Thalassiosira sp. culturing process. Method. The culturing stages are conducted in a stepwise manner, starting from the smallest volume, namely from laboratory scale, intermediate scale, to mass scale. Results. The peak population in the bottle container occurred on day 3, reaching 940,000 cells/ml. This was followed by culturing in gallon 1 media at 590,000 cells/ml and gallon 2 media at 400,000 cells/ml. Subsequently, at the intermediate scale, the peak population occurred on day 2 with 81,000 cells/ml, and finally at the mass scale, it reached 162,000 cells/ml on day 2. Conclusion. The harvesting process is conducted during the logarithmic phase to obtain maximum biomass.
Keywords: *Thalassiosira sp., Natural feed, Peak population, Culture.*

1. INTRODUCTION

Hatchery production is a crucial initial stage in the shrimp production cycle, with the aim of producing high-quality shrimp postlarvae (Lestari et al., 2022). The rearing of shrimp postlarvae in hatcheries must pay close attention to seed quality, one of which is by implementing appropriate feeding standards (Kamil et al., 2023). Feed is one of the most important factors because it directly affects the growth and development of the cultured shrimp. One of the determining factors for successful rearing is the quality of natural feed, including its quantity, type, and concentration (Lante & Herlinah, 2015). Based on the developmental stage of the shrimp, the feed provided should be adjusted to the size of the mouth opening and the nutritional requirements of the feed.

One type of natural feed for vannamei shrimp larvae that has a complex nutritional composition is Thalassiosira sp. Its nutritional content includes protein at 21.85–37%, fat at 2.41–10%, and carbohydrates at 17–21% (Erlangga et al., 2021). According to Devianti et al. (2022), Thalassiosira sp. is used as a natural feed for shrimp larvae because it has a size range of 4–32 µm, which is suitable for the shrimp mouth opening and is easily digested from the nauplius to zoea stages. The provision of Thalassiosira sp. as natural feed can also improve survival rate, metamorphosis percentage, and digestive enzyme activity in vannamei shrimp larvae (Mustofa et al., 2024).

To ensure the availability of Thalassiosira sp. as natural feed, a cultivation process is required. The cultivation of Thalassiosira sp. requires appropriate nutrients to support its

growth. Therefore, the cultivation process requires several inorganic elements in the form of macronutrients and micronutrients. The required macronutrients include nitrogen (N) and phosphorus (P) (Dewi, 2017), while the required micronutrients include iron (Fe), copper (Cu), manganese (Mn), zinc (Zn), cobalt (Co), and boron (B) (Erlangga et al., 2021). The success of Thalassiosira sp. cultivation is also influenced by water quality parameters, such as salinity, temperature, and pH, as well as light intensity and photoperiod to support the photosynthesis process (Prasetyo et al., 2022).

Therefore, appropriate cultivation techniques are required to support the growth of Thalassiosira sp. One of the hatcheries that conducts Thalassiosira sp. cultivation is PT. Bumi Persada Gemilang Shrimp Hatchery Centralpertiwi Bahari. The cultivation techniques for Thalassiosira sp. include laboratory-scale, intermediate-scale, and mass-scale cultivation. The purpose of this final report is to describe proper Thalassiosira sp. cultivation techniques to ensure an adequate supply of natural feed for vannamei shrimp larvae.

2. METHODOLOGY

2.1 Sample Collection

This study was conducted at PT. Bumi Persada Gemilang Shrimp Hatchery Centralpertiwi Bahari, North Galesong, Takalar, South Sulawesi, for a period of three months. The equipment and materials used in the Thalassiosira sp. cultivation process at PT. Bumi Persada Gemilang Shrimp Hatchery Centralpertiwi Bahari were as follows:

Table 1. Equipment Used for Thalassiosira sp. Cultivation

No.	Equipment	Function
1	1-liter glass bottle	First-stage culture vessel
2	15-liter gallon container	Second- and third-stage culture vessel
3	1,000 mL graduated cylinder	Measuring the volume of culture medium and starter culture
4	Micropipette	Collecting *Thalassiosira* sp. samples
5	Microscope	Observing and identifying the cells being cultured
6	*Haemocytometer*	Determining the cell density of *Thalassiosira* sp.
7	*Cover glass*	Maintaining sample flatness
8	*Hand counter*	Counting the density of *Thalassiosira* sp.
9	*Autoclave*	Sterilizing equipment and culture media
10	Digital balance	Weighing materials
11	Aeration tubing	Supplying oxygen
12	T-joint	Connecting the aeration pipe and tubing
13	Pipette	Directing air from the aeration tubing
14	Filter bag	Filtering seawater
15	pH meter	Measuring pH and temperature
16	Refractometer	Measuring salinity
17	Blower	Supplying oxygen to the culture medium
18	Aeration pipe	Delivering oxygen from the blower
19	Tubing	Delivering seawater into the culture vessel

Table 2. Culture Materials for Thalassiosira sp.

No.	Materials	Purpose
1	Thalassiosira sp.	Inoculum
2	Freshwater	Washing culture equipment
3	Seawater	Culture medium
4	Chlorine (bleach)	Water sterilization

No.	Materials	Purpose
5	Thiosulfate	Chlorine neutralization
6	Distilled water (Aquades)	Replacement for sterile freshwater
7	Chlorine test	Testing residual chlorine concentration
8	70% alcohol	Antiseptic and disinfectant
9	Silicate	Nutrient source for Thalassiosira sp.
10	EDTA	Nutrient source for Thalassiosira sp.
11	DSP	Nutrient source for Thalassiosira sp.
12	$NaNO_3$	Nutrient source for Thalassiosira sp.
13	KNO_3	Nutrient source for Thalassiosira sp.
14	Urea	Nutrient source for Thalassiosira sp.
15	AGP	Nutrient source for Thalassiosira sp.

2.2 Sterilization of Culture Equipment and Materials

There are two principles of sterilization, namely physical and chemical sterilization. Physical sterilization was performed using moist-heat sterilization with an autoclave, while chemical sterilization was carried out using chlorine (bleach) for the sterilization of equipment and culture media.

2.2.1 Equipment Strerillization

All equipment used during the *Thalassiosira* sp. culture process, whether directly or indirectly in contact with the *Thalassiosira* sp. inoculum, was sterilized beforehand. Equipment used during the initial culture stage, such as 1-liter glass bottles, sample tubes, pipettes, and aeration hoses, was thoroughly washed using a mixture of detergent and iodine and then allowed to dry. Detergent functions to help remove dirt from equipment surfaces by reducing the surface tension of water, allowing water to spread more easily and penetrate stains and dirt (Afrianto & Muqsith, 2014). After drying, physical sterilization was carried out using an autoclave at 121°C for approximately 15 minutes. Other equipment, such as gallons, measuring cylinders, aeration hoses, and air stones, was sterilized using a mixture of detergent and iodine. The cleaned equipment was placed on a rack to dry. The culture rack was sterilized using 70% alcohol and then wiped until clean and dry. The intermediate- and mass-culture tanks were sterilized by rinsing them with freshwater and scrubbing all tank surfaces using a scouring pad with a detergent and iodine solution until algae and dirt were removed. The tanks were then rinsed again with freshwater and allowed to dry for approximately two days before use.

2.2.2 Material Sterillization

The materials used as the culture medium for the 1-liter bottle culture included seawater filtered through a filter bag, purified freshwater or distilled water, EDTA fertilizer, silicate, and AGP. These materials were mixed in a 20-liter bucket consisting of 16 liters of seawater and 4 liters of freshwater to obtain a salinity of 28 ppt. Fertilizers were added to the culture medium according to the specified dosage (Table 4) and mixed using aeration. The medium was then transferred into the bottles at 700 mL per bottle and sterilized using an autoclave at 121°C for approximately 15 minutes. For seawater sterilization during the Gallon 1 and Gallon 2 culture stages, seawater was first filtered using a filter bag. The seawater was then collected in a 500-liter storage tank and mixed with freshwater to obtain a salinity of 28 ppt, followed by the addition of chlorine at a concentration of 25 ppm. The medium was aerated and mixed for 24 hours. When added to water, chlorine reacts by killing or inhibiting the growth of microorganisms and is therefore used as a sterilizing agent for the culture medium prior to use.

The medium was subsequently neutralized using thiosulfate at a dosage of 15 ppm. For sterilization of the culture water at the intermediate scale, seawater was filtered using a filter bag. At the mass scale, two sterilization methods were used, namely filtration using a filter bag and chlorination at 10 ppm, followed by neutralization with thiosulfate at a dosage of 1.5–2 ppm. Before the culture medium was used, a chlorine test was conducted to ensure that no residual chlorine remained in the culture medium.

2.3 Fertilizer Preparation

The fertilizers used during the culture process were applied at different dosages according to the culture medium and volume used. At the laboratory scale, liquid fertilizers were used; therefore, the fertilizers were dissolved in 1,000 mL of distilled water. The composition of the liquid fertilizer used at the laboratory scale is presented in Table 3.

Table 3. Composition of liquid fertilizer at the laboratory scale (SOP of PT. Bumi Persada Gemilang Shrimp Hatchery Centralpertiwi Bahari)

No.	Fertilizer	Dosage
1	EDTA	20 g
2	Silicate	135 g
3	Thiosulfate	30 g

The liquid fertilizer presented in Table 3 was dissolved in 1,000 mL of distilled water and then transferred into a glass bottle and covered with aluminum foil. The dissolved fertilizer was sterilized using an autoclave at 121°C for approximately 15 minutes. The fertilizer dosages applied according to the culture medium are presented in Table 4.

Table 4. Fertilizer dosages used during laboratory-scale culture

Type of Fertilizer	Bottle	Gallon 1	Gallon 2
EDTA	0.36 mL	5 mL	5 mL
Silicate	0.54 mL	10 mL	10 mL
AGP	0.71 mL	10 mL	10 mL

At the intermediate and mass culture scales, fertilizers were measured according to the predetermined dosages, as presented in Table 5. Urea, $NaNO_3$, KNO_3, and DSP were dissolved in a bucket containing 3 liters of freshwater and aerated for 12 hours. Silicate was dissolved separately in a bucket containing 3 liters of freshwater and aerated for 12 hours. After dissolution, pure AGP was added and the solution was mixed again until homogeneous.

Table 5. Fertilizer dosages at the intermediate and mass culture scales

Fertilizer	**Intermediate (7 tons)**	**Mass (28 tons)**
Urea	120 g	400 g
$NaNO_3$	800 g	1,600 g
KNO_3	300 g	480 g
DSP	40 g	170 g
Silicate	270 mL	810 mL
AGP	150 mL	200 mL

2.4 Thalassiosira sp. Culture

2.4.1 1 Liter Bottle Culture

The initial stage of Thalassiosira sp. culture was carried out using 1-liter bottles. This stage began by mixing water with a salinity of 28 ppt and fertilizer in a 20-liter bucket. The fertilizer dosage used is presented in Table 4. The medium was mixed until homogeneous using aeration and then transferred into sterile glass bottles at a volume of 700 mL per bottle. The bottle openings were tightly covered with aluminum foil, and the culture media were sterilized using an autoclave at 121°C for approximately 15 minutes. The culture media were then allowed to cool. The aluminum foil was removed, and Thalassiosira sp. inoculum was added to 12 culture bottles, with 200 mL of inoculum added to each bottle. The bottles were then covered again with aluminum foil, followed by the installation of pipettes and aeration hoses. The bottles were placed on a culture rack equipped with lamps as a light source, providing a light intensity of 4,200 lux. The bottle culture was maintained for 2–3 days or until the highest cell density was reached, as indicated by a darker color of the culture medium and confirmed through cell counting.

2.4.2 Gallon 1 Culture

The Gallon 1 culture was initiated by adding sterile seawater into four gallons, with 10 liters of seawater in each gallon. Thiosulfate was initially added at a dosage of 10 mL, followed by fertilizer addition according to the dosage presented in Table 4. Thalassiosira sp. inoculum obtained from the bottle culture was then transferred into the gallons at a rate of three bottles per gallon. The culture process was maintained for two days until the highest cell density was reached, as indicated by a darker cell color and confirmed through cell counting.

2.4.3 Gallon 2 Culture

The Gallon 2 culture was initiated using treated seawater that had been filtered through a filter bag. The seawater was distributed into 12 gallons, with each gallon containing 10 liters, and aeration was provided. Thiosulfate was added to the Gallon 2 culture medium at 5 ppm or 10 mL. The addition of thiosulfate was intended to neutralize residual chlorine in the culture medium. Fertilizer was subsequently added according to the dosage presented in Table 4. Then, 5 liters of Thalassiosira sp. inoculum from Gallon 1 were added. The gallons were placed on a culture rack maintained at 21°C and equipped with aeration and lamps as a light source. The culture process was maintained for two days.

2.4.4 Intermediate-Scale Culture

The Thalassiosira sp. inoculum used for the intermediate-scale culture was obtained from the Gallon 2 culture, with a volume of 15 liters per gallon and six gallons added to each tank. Each tank contained 6 tons of seawater, which was then mixed with fertilizer according to the dosage presented in Table 5. The culture tanks were equipped with eight aeration points, with a distance of 40 cm between each aeration point. Six gallons of culture were transferred into each intermediate culture tank to increase the biomass of microalgal cells, with the expectation of obtaining a greater cell biomass than that obtained at the laboratory scale. Harvesting at this stage was carried out after two days of culture.The Gallon 2 culture was initiated using treated seawater that had been filtered through a filter bag. The seawater was distributed into 12 gallons, with each gallon containing 10 liters,

and aeration was provided. Thiosulfate was added to the Gallon 2 culture medium at 5 ppm or 10 mL. The addition of thiosulfate was intended to neutralize residual chlorine in the culture medium. Fertilizer was subsequently added according to the dosage presented in Table 4. Then, 5 liters of *Thalassiosira* sp. inoculum from Gallon 1 were added. The gallons were placed on a culture rack maintained at 21°C and equipped with aeration and lamps as a light source. The culture process was maintained for two days.

2.4.5 Mass Scale Culture

The mass-scale culture used Thalassiosira sp. inoculum obtained from the intermediate-scale culture. The mass culture consisted of 21 tons of seawater and 7 tons of Thalassiosira sp. inoculum. The inoculum was transferred from the intermediate tank to the mass culture tank using a pump assisted by hoses and equipped with aeration. The mass-scale culture was conducted to produce a larger biomass of Thalassiosira sp. sufficient to meet the natural feed requirements of shrimp postlarvae.

2.5 Growth Monitoring

Growth monitoring was conducted throughout the culture process to determine cell proliferation and the quality of the cultured *Thalassiosira* sp. cells. Growth monitoring was performed by measuring cell density every morning. The initial step in cell counting involved collecting samples from each culture medium. At the laboratory scale, samples were collected into sample tubes using a dropper, whereas at the intermediate and mass scales, 50-mL sample tubes were used and samples were collected using a dipper. Growth monitoring was performed by counting cells using a haemocytometer under a microscope at 10× magnification. A hand counter was also used to facilitate the cell-counting process. Each culture scale used a different counting method and formula. The formulas used to calculate *Thalassiosira* sp. cell density are presented in Table 6.

Table 6. Formula for calculating Thalassiosira sp. density according to the SOP of PT. Bumi Persada Gemilang Shrimp Hatchery Centralpertiwi Bahari

Sample Level	Counting Area	Algal Density (cells/mL)
Bottle	1 large square (center)	Cell count × 10^4
Gallon	1 large square (center)	Cell count × 10^4
Intermediate	9 large squares	(Cell count ÷ 9) × 10^4
Mass	9 large squares	(Cell count ÷ 9) × 10^4

Phytoplankton density was determined by counting the number of cells contained within one large square of the haemocytometer at the laboratory scale and nine large squares at the intermediate and mass scales. If **N** represents the total number of cells counted, the value of $\mathbf{N \times 10^4}$ is then divided by the number of large squares counted. The calculation procedure is illustrated in Figure 1.

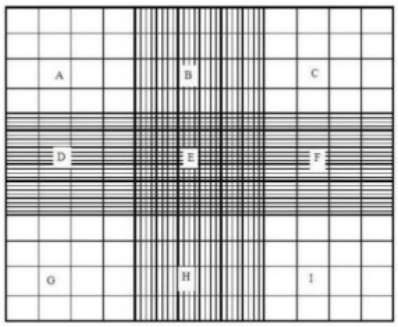


Figure 1. Haemocytometer (Winasi, 2012)

Cell growth rate is calculated to determine cell proliferation over a specific period. The cell growth rate is calculated using the formula employed in Fogg's (1965) study, as cited in Fadila *et al.* (2021), namely:

$$K = \frac{Wt - Wo}{\Delta t}$$

where:

K = specific growth constant

Wt = number of cells on day t

Wo = number of cells on day 0

Δt = time from 0 to t (days)

2.6 Water Quality Monitoring

Water quality measurements were also conducted during the culture process to ensure the culture medium remained within standard parameters, thereby supporting optimal growth of *Thalassiosira* sp. The water quality parameters measured included pH, temperature, and salinity. Water quality measurements on a laboratory scale are performed at the early stages of the culture, while those on an intermediate and large-scale are conducted in the morning throughout the culture period. A refractometer is used to measure salinity. A pH meter is used to measure temperature and pH. Water quality measurements on an intermediate and large-scale use a dipper to avoid contamination from the water quality measuring instruments. During the culture process, it is also necessary to monitor for any issues that arise, such as contamination caused by other microorganisms or protozoa. This is done by examining the culture medium and *Thalassiosira* sp. cells under a microscope to check for any abnormalities or contamination.

2.7 Harvesting Technique

Harvesting is performed when the density of the cultured *Thalassiosira* sp. has reached its population peak. Before harvesting, an observation using a microscope is conducted to assess cell quality and density, thereby determining whether growth has reached the population peak. This check is performed at every stage of the culture prior to harvesting. If peak population density has not yet been reached, there are still residual nutrients in the culture medium, so the culture period can be extended. However, if peak population density has been reached, harvesting must be carried out immediately; otherwise, delayed harvesting

will result in cell death, leading to a decline in the quality *of Thalassiosira* sp. Declining quality is indicated by clumping, damage, and cell colors that differ from normal.

Harvesting during the culture process is carried out by transferring the culture from the bottle medium to gallon 1, from gallon 1 to gallon 2, and from gallon 2 to the intermediate scale, then transferring it to the mass culture tank using a pump and pipes. Culture activities on a laboratory scale using bottle media last 3–4 days, while those using gallon 1, gallon 2, intermediate-scale, and mass-scale media last 3 days. This is done to achieve the highest density at each culture scale and to prevent a death phase. During each *Thalassiosira* sp. harvest, hygiene must be maintained by performing appropriate sterilization procedures to prevent cross-contamination and to preserve the quality of the resulting cells.

2.8 Data Analysis

The data collected for this final project were analyzed descriptively. The analysis focused on actual problems and phenomena by describing the facts studied as they were observed in the field. Both primary and secondary data were used. Primary data was obtained through active participation and observation to gather facts via direct interviews with technicians possessing both knowledge and field experience (Herdayati & Syahrial, 2019). Meanwhile, secondary data is data obtained indirectly from sources such as journals, books, and literature reviews related to Thalassiosira sp. culture techniques (Linarwati et al., 2016)

3. RESULT AND DISCUSSION

3.1 Result

Based on the results of the *Thalassiosira* sp. natural feed culture conducted progressively, starting from the smallest volume at the laboratory scale, followed by the intermediate scale and finally the mass culture scale, cell density data were obtained from the laboratory-scale cultures, namely the bottle culture medium (Figure 2), Gallon 1 and Gallon 2 (Figure 3), as well as from the intermediate and mass culture scales (Figure 4).

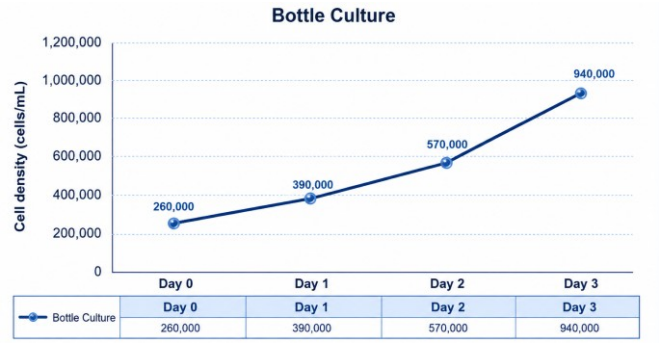
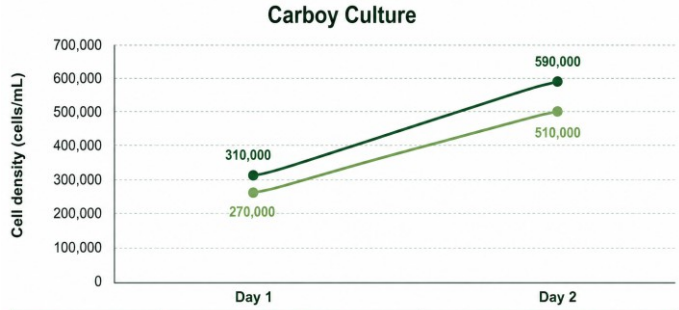


Figure 2. Growth curve of *Thalassiosira* sp. in the bottle culture medium

Based on the growth curve presented in Figure 4, the cell density of *Thalassiosira* sp. in the bottle culture medium increased from the initial inoculation (Day 0). The initial cell density of 260,000 cells/mL increased by 130,000 cells/mL on Day 1, reaching 390,000 cells/mL. This stage represents the adaptation phase, during which *Thalassiosira* sp. adjusts to its new environment (Angraeni et al., 2019). The growth rate obtained was 680,000 cells/mL over a three-day culture period. From Day 1 to Day 3, cell density increased rapidly, resulting

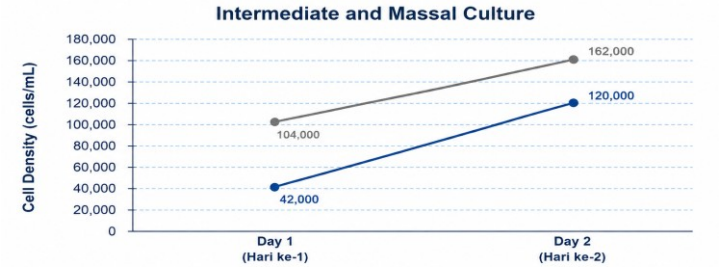
in a cell density of 940,000 cells/mL. This stage is referred to as the logarithmic phase, characterized by a rapid increase in cell density, which causes the nutrients in the culture medium to decrease rapidly (Meria et al., 2021). Harvesting or subsequent culture is conducted during this phase to prevent cell mortality caused by reduced light intensity penetrating the culture medium, as well as competition for oxygen and nutrients.



Treatment	Day 1 (cells/mL)	Day 2 (cells/mL)
Carboy 1	310,000	590,000
Carboy 2	270,000	510,000

Figure 3. . Growth curve of Thalassiosira sp. In carboy 1 and carboy 2.

Based on the data in the graph in Figure 5, growth in gallon 1 media occurred from day 1 to day 2, with an increase of 280,000 cells/mL. The daily growth rate in Gallon 1 was 90,000 cells/mL. The cell count in Gallon 1 was 590,000 cells/mL on day 2. Growth in Gallon 2 also occurred from Day 1 to Day 2, with a lower cell count compared to Gallon 1; however, the total volume of medium in Gallon 2 was greater, at 12 gallons. The cell growth rate in gallon 2 increased by 240,000 cells/mL, with the initial cell count of 270,000 cells/mL rising to 510,000 cells/mL on the second day of the culture. Thus, the logarithmic phase occurred on the second day of the culture in the gallon-based medium.



Culture Type	Day 1 (cells/mL) (Hari ke-1)	Day 2 (cells/mL) (Hari ke-2)
Intermediate	42,000	120,000
Massal	104,000	162,000

Figure 4. . Growth curves of Thalassiosira sp. in intermediate-scale and mass-scale cultures.

Based on the results of cell density calculations for *Thalassiosira* sp. cultured in intermediate and mass scales (Figure 6), data showed that the density continued to increase from day 1 to day 2 of the culture. The initial seeding density on the intermediate scale, which was 42,000 cells/mL, increased on day 1 by 78,000 cells/mL, reaching 120,000 cells/mL on day 2. The initial seeding density at the mass scale, which was 104,000 cells/mL, increased by 58,000 cells/mL on day 1, reaching 162,000 cells/mL on day 2. Harvesting was conducted on day 2 of the culture because continuing the culture beyond this point would lead to a phase of declining growth rate and eventual cell death. Cell division in intermediate- and large-scale cultures occurs more slowly due to limited light

penetration into the tank surface, resulting in a slower division process (Wardani *et al*., 2022). During the culture period, water quality is a critical factor to monitor, as it affects the growth rate of *Thalassiosira* sp. cells. Based on water quality measurements—specifically pH, temperature, and salinity taken during the culture period, the data presented in Table 7 are as follows:

Table 7. Water quality measurement data for laboratory, intermediate, and large-scale cultures

Culture Medium	Temperature (°C)	pH	Salinity (ppt)
Bottle	21	8.1	28
Gallon 1	21	8.1	28
Gallon 2	21	8.1	28
Intermediate	30–30.2	9	33–34
Mass Culture	29.7–29.9	9	33–34

3.2 Discussion

The growth of Thalassiosira sp. is characterized by a gradual change in culture color, which becomes increasingly darker as cell density increases. Growth monitoring was conducted to determine cell density and assess the quality of Thalassiosira sp. using microscopic observations. The population peak (logarithmic phase) in the laboratory-scale culture using bottle media occurred on the third day, whereas in Gallon 1, Gallon 2, intermediate-scale, and mass-scale cultures, the population peak occurred on the second day. The cell density obtained at the end of the culture period or prior to harvesting was 940,000 cells/mL in the bottle medium, followed by 590,000 cells/mL in Gallon 1 and 510,000 cells/mL in Gallon 2. At the intermediate scale, the population peak occurred on the second day, with a cell density of 120,000 cells/mL, while at the mass scale, the cell density reached 162,000 cells/mL on the second day. The population peak (logarithmic phase) represents the period when the culture reaches its highest cell density and is considered the appropriate phase for harvesting and subsequent inoculation into the next culture medium to increase cell production. According to Mufidah et al. (2017), harvesting should be conducted when sufficient nutrients remain available to support microorganism growth. Therefore, harvesting during the exponential phase can help prevent excessive competition for nutrients among Thalassiosira sp. cells.

The differences in cell density observed at each culture stage may be influenced by water quality parameters, including temperature, pH, light intensity, and salinity. Water quality is one of the key factors determining the success of Thalassiosira sp. cultivation. Based on the water quality measurements presented in Table 7, the temperature, pH, and salinity during the culture period were generally within suitable ranges for microalgal growth. Temperature measurements at the laboratory scale were 21°C, while the intermediate-scale culture ranged from 30 to 30.2°C and the mass-scale culture ranged from 29.7 to 29.9°C. These values were generally within the reported suitable range for Thalassiosira sp. growth. Baek et al. (2011), as cited in Wahyudi et al. (2022), reported that Thalassiosira sp. can grow within a temperature

range of 10–30°C. The laboratory-scale pH was 8.1, which is consistent with the findings of Wahyudi et al. (2022), who reported that a pH range of 8.0–8.5 is suitable for Thalassiosira sp. growth. Meanwhile, the pH at the intermediate and mass culture scales ranged from 9.0 to 9.1, which was within the optimal range of approximately 8.9–9.7 reported for these culture scales (Yuliana, 2017). The increase in pH may be associated with increasing microalgal cell growth.

Salinity measurements using a refractometer showed a salinity of 28 ppt at the laboratory scale, while salinity at the intermediate and mass culture scales ranged from 33 to 34 ppt. According to Sanjaya and Danakusuma (2018), the suitable salinity range for Thalassiosira sp. cultivation is 25–35 ppt. Therefore, the salinity values recorded during the culture process were within the suitable range for Thalassiosira sp. growth. Overall, the temperature, pH, and salinity observed during the culture process were generally suitable for supporting the growth of Thalassiosira sp.

Light intensity is another important factor influencing microalgal growth. During the laboratory-scale culture, the recorded light intensity was approximately 4,200 lux. Erlangga et al. (2021) reported that the suitable light intensity range for natural feed culture is 900–3,000 lux, while the optimal light intensity for Thalassiosira sp. growth is approximately 5,000–12,000 lux. Light intensity exceeding 12,000 lux may negatively affect the population density and quality of Thalassiosira sp. cells. Thus, the light intensity recorded in this study was relatively close to the reported optimal range, although it remained slightly below the recommended intensity for maximum growth.

One of the common challenges in Thalassiosira sp. cultivation is contamination by other microorganisms, particularly protozoa. Protozoan contamination is frequently encountered in microalgal cultures and can damage the cell walls of microalgae, thereby reducing culture quality and productivity (Neordjito et al., 2019). Therefore, appropriate preventive measures are required to maintain culture quality, including the sterilization of all equipment and culture media used during laboratory-, intermediate-, and mass-scale cultivation. Proper sterilization and culture management are essential to minimize contamination and maintain the growth and quality of Thalassiosira sp. throughout the culture process.

4. CONCLUSION & SUGGESTION

The culture technique of Thalassiosira sp. at PT. Bumi Persada Gemilang Shrimp Hatchery, Centralpertiwi Bahari, was carried out at three culture scales. The laboratory scale consisted of three stages: culture in 1-liter bottles, 13-liter gallon containers (Gallon 1), and 15-liter gallon containers (Gallon 2). The culture was then continued at the intermediate scale with a volume of 7 tons and, finally, at the mass culture scale with a volume of 28 tons. Harvesting was conducted during the logarithmic phase to obtain maximum biomass. The peak population density of Thalassiosira sp. in the 1-liter bottle culture was reached on the third day, with a density of 940,000 cells/mL. In Gallon 1, the peak density was reached on the second day at 590,000 cells/mL, while in Gallon 2, it reached 510,000 cells/mL on the second day. At the intermediate scale, the peak density was 120,000 cells/mL on the second day, whereas at the mass culture scale, the density reached 162,000 cells/mL on the second day of culture. The culture of Thalassiosira sp. at PT. Bumi Persada Gemilang Shrimp Hatchery, Centralpertiwi

Bahari, requires improved handling through the implementation of better biosecurity measures and sterilization procedures during the preparation of culture equipment and media. These measures aim to prevent Thalassiosira sp. cultures from becoming contaminated by other microorganisms, such as protozoa, during the culture process.

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