Chapter 14

Closed Aquaculture System: Zero Water Discharge for Shrimp and Prawn Farming in Indonesia

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Abstract

This chapter focuses on the development and application of zero water discharge (ZWD) system, which has become an alternative solution to conventional methods of aquaculture production. With this system, it is expected to answer many issues in aquaculture cultivation, such as environmental damage, disease outbreak, and land-use change, and to create a sustainable aquaculture cultivation system. ZWD system is an improved batch system with an emphasis on microbial manipulation in rearing tank. The principle of microbial selection is based on the role of each microbial component in nutrient cycle in the rearing tank. This chapter contains in detail how methods and stages are performed in order to conduct this system, including design of construction system, cultivation of microbial components, initial conditioning of this system, and microbial manipulation. The performance of the system was tested in crustacean culture such as white shrimp and giant freshwater prawns, and it showed that the system can increase the average survival rate of 10–20%. In addition, the technical and economic feasibility of this system was evaluated to illustrate the production efficiency upon the application of this system in the industry.

Keywords: closed aquaculture system, zero water discharge, white shrimp, prawn, microbial loop, microbial manipulation

1. Introduction

Driven with the increment of human population in the world, trend of total world fishery production has increased from 2009 to 2014 with an average growth rate of about 2.77% per year [1]. This growth mostly came from aquaculture sector instead of capture practice with annual growth rate of 8.8% [2]. Global aquaculture production has reached 73.8 million tons in 2014 with an estimated value of USD160.2 billion. It shared about 44.14% of total fishery production. In the next decade (2025), FAO predicted that aquaculture sector would share 52% of the total fishery production [1]. Along with the prediction, Indonesia has a great potential to develop the aquaculture sector. Indonesia is one of main producers of both capture and aquaculture fishery commodities because it is supported by its geographical condition. Indonesia is an archipelagic country that has great potential in fisheries sector. It consisted of 17,500 islands and located between two big oceans, Pacific and Indian Ocean. Moreover, Indonesia is a country crossed by equator line and ranked as world's 4th longest coastline, which indicates a high diversity of aquatic organisms, including marine biota [3]. So, there are many fishery commodities grown in Indonesia. Currently, Indonesia ranks as the second top both capture and aquaculture producers after People's Republic of China, contributing 6.48 and 14.36 million tons, respectively, to worldwide production [1]. One of main commodities is crustaceans that produced both capture and aquaculture practices. In fact, most of productions were obtained from aquaculture. In 2014, shrimp capture production only contributed about 30% of the total shrimp production or approximately 273,133 tons [4]. Shrimp commodities rank as the top by annual total aquaculture production from aquatic animal.

Most of shrimp production is dominated by white shrimp (*Litopenaeus vannamei*) which is also exported to several countries in the world, such as United States of America, Japan, People's Republic of China, United Kingdom, Malaysia, etc. [5]. Trend of white shrimp production has increased significantly with an average growth of 22.46%. This increment production was due to ease of cultivation practice, in case of availability of seed, cultivation period, and more resistance to environmental changes. Another species, giant freshwater prawn has the opportunity to become a main commodity due to high economic value. In 2013, prawn production reached approximately 3.171 tons, which has been cultivated in several site, such as West Kalimantan, Bali, West Java, and East Java [6]. Although it is still small in number compared to white shrimp, production volume continued to rise in recent years. Ministry of Maritime Affairs and Fisheries Republic of Indonesia seriously began promoting the cultivation of prawn, started in 2015, they have allocated a national capital budget for prawn production up to Rp 275.2 billions [7].

However, a high production scale does not ensure sustainability of shrimp aquaculture industry, because currently most shrimp farms use conventional culture practices, such as batch or flow-through system. It is true that conventional shrimp rearing strategies are still widely applied and profitable due to its simplicity and acceptable production cost, but since the cultivation relies on natural environment with less control to water quality and disease or predation, this condition leads to unpredictable culture performances [8]. Furthermore, the accumulation of harmful substances in culture water from uneaten feed and excretion (e.g., ammonium and nitrite) is very likely exceeds the tolerance limits, causing a decrement of culture survival rate and thus affecting overall shrimp productivity in conventional culture system [9]. Besides, the system is considered as not environmentally friendly, because untreated effluent water can pollute the surrounding aquatic environment [8]. In term of space requirement, the system occupies a large production area and requires close distance to coastal area to ensure seawater access. These circumstances contribute to impractical shrimp farming

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industry and its sustainability in the near future. These problems urge an improvement of better aquaculture technology, which can support the culture's sustainability, with regard to water quality and culture performance, good hygiene, as well as high culture efficiency in terms of space utilization, water sources, and feed.

One alternative technology called zero water discharge (ZWD) system has been developed to resolve the above-mentioned problems [10–13]. ZWD is a sustainable intensive culture technology, which is environmentally friendly as it maintains water quality, therefore prevents pathogen spreading as well as wastewater discharge, which is rich in nutrients, to the environment [14]. The ZWD system allows limiting or reducing water usage, by implementing microbial consortium with various important roles, such as recycling nitrogen compound in the culture water and cleaning harmful nitrogen substances prior partial or total reuse of the water.

2. ZWD principle

Water body is habitat for all aquatic animals, including shrimp and prawn. Consequently, the key for success cultivation is to keep the habitat favorable for shrimp to grow. So that, it is crucial to maintain water quality in tolerance range for shrimp growth. Water quality includes physical, chemical, and biological parameters particularly temperature, dissolved oxygen, and toxic nitrogen substance concentrations [15]. Temperature and dissolved oxygen parameters can be manipulated by physical treatment such as using aerator and water heater, while toxic nitrogen substances have dealt with biological treatment system usually utilizing microbial-based treatment.

Toxic nitrogen substances produced from excretion activity of shrimp and their feed residue, such as ammonium and nitrite, disturb metabolic balance of the shrimps, making them more prone to disease that causes several disadvantages, including reduced body weight, increased mortality, and eventually decrease production yield [16–18]. As this has become one major problem in aquaculture, ammonium and nitrite removal management is a major concern in ZWD system. In natural aquatic ecosystems, microorganism present in water body maintains a balance concentration of each nitrogen compounds. As ammonium and nitrite concentration in intensive aquaculture systems build up much faster than in natural ecosystems, we cannot rely on naturally occurring microorganisms in the ponds. Their low population size cannot cope with the rate of ammonium accumulation, and therefore, addition of microorganism is needed.

This system uses the principle of microbial loops adapted from natural ecosystems. Toxic nitrogen substances present in ammonium and nitrite form can be converted into nitrate which is less toxic substance through consecutive nitrification microbial process. ZWD system aims to improve water quality through recycling chemical waste [19]. While conventional system (e.g. flow-through) requires a continuous new water supply to avoid waste accumulation in the culture, ZWD recycles ammonium, nitrite, and nitrate using microorganism consortia, and therefore, it reduces water usage significantly. Ammonium, nitrite, and nitrate level can be maintained using addition of heterotrophic bacteria, nitrifying bacteria, and microalgae, regularly [13].

2.1. State of the art

Based on the principle explained earlier, the most crucial thing is the selection of microbial components that have functions in maintaining water quality and are harmless to the animals being cultivated. In addition, selected microbes may act as probiotics such as to counteract pathogenic attacks from *Vibrio* sp. in shrimp farming [19]. Since this system refers to nutrient cycles in aquatic habitat, the selected microbes should have a role in the alteration of toxin substances into harmless substance produced in the cultivation system. The system emphasizes nitrogen nutrient cycle because nitrogen toxin is very dangerous if it accumulates excessively.

Figure 1 shown below is an example for the estimation of nutrient cycle and microbial loop that occur in the ZWD system [13]. The greatest accumulation of toxic compounds in cultivation is from animal feed and feces. These compounds are mostly organic matters, which can be degraded by heterotrophic bacteria into inorganic compounds. Inorganic compounds, such as ammonium and nitrite, which became the focus, have to be removed. The ammonium and nitrite should be converted into less harmful compounds such as nitrate by oxidation. Microbes that can do the oxidation process from inorganic compounds are litoautotrophic bacteria [20]. There are two stages of the oxidation processes: (1) the conversion from ammonium to nitrite and (2) nitrite to nitrate. Ammonium-oxidizing bacteria (AOB) convert ammonium to nitrite, for example, Nitrosomonas, and nitrite-oxidizing bacteria (NOB) convert nitrite to nitrate, such as Nitrobacter [21]. Even though nitrate is a harmless substance, the tolerance range in aquaculture system is no more than 200 ppm [22]. Therefore, it is necessary to search microbes that can utilize nitrate. Some microalgae can use nitrate as a source of nitrogen, so that addition of microalgae is important in this system. In addition, at the trophic level, microalgae act as

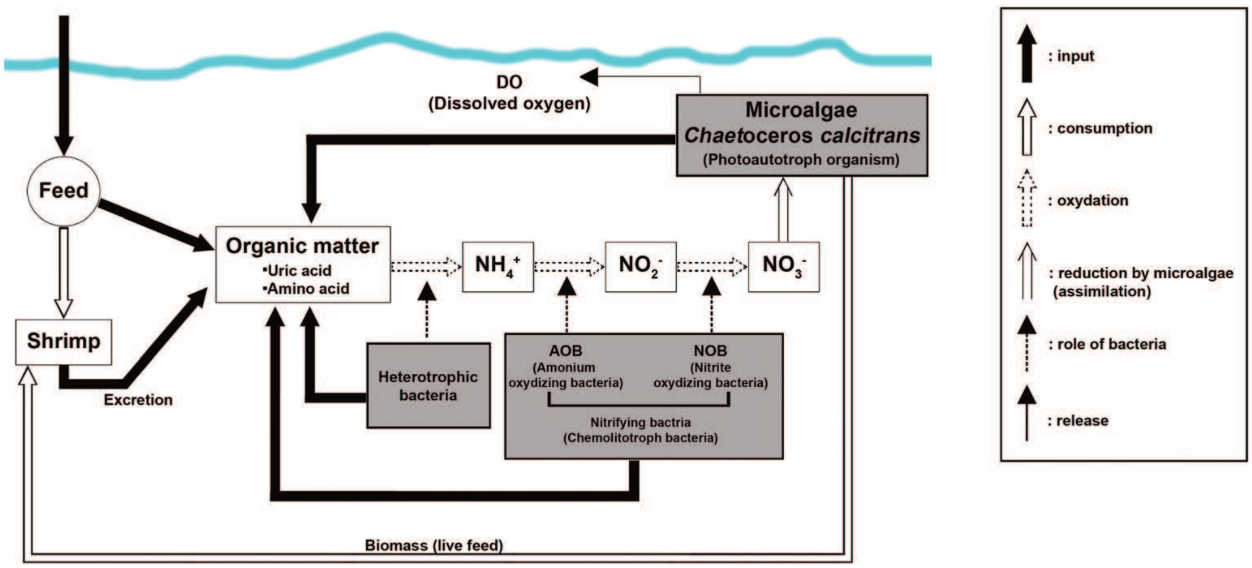

Figure 1. Schematic of nutrient cycle in ZWD system [13].

producer that also provides oxygen that can raise the DO level, and their biomass may be possible as food source for cultivated animals in the system.

Because the ZWD system relies on microbial components added to the system, different animal cultures will have different microbial components added, and the system must be favorable for microbial components to live. For a simple example, we have to consider about native microbial habitat; marine microbes will be suitable for marine animal farming and freshwater microbes for freshwater animal farming. **Figure 1** is an example of ZWD system in the cultivation of white shrimp, so the microbial components used are marine heterotrophic bacteria, marine AOB, marine NOB, and marine microalgae (*Chaetoceros calcitrans*).

2.2. Distinctive characteristics in ZWD system compared to other microbial-based systems

As the aquaculture industry grows rapidly in the world, it encourages research to create technology that leads to the sustainability in aquaculture industry. One of main research areas is the utilization of microbes that are now widely used in aquaculture industry. It can play a role as a food source such as microalgae for the larval phase [23, 24], maintain water quality such as using ammonium as a source of nitrogen for microbial metabolism [21, 25, 26], fight against disease such as immunostimulant that trigger antibodies or directly interact antagonistically with pathogen [19, 27, 28].

The application of microbes in aquaculture is conducted into microbial-based closed systems, such as the ZWD system. The term of zero water discharge has many versions; it can be zero water exchange [14, 29–31], limited water discharge [32, 33], minimal discharge system [34], minimal effluent discharge [35], minimal exchange system [36], etc. All such systems have the same principle that is minimizing water use and re-recycling water used by involving the role of microbes. ZWD system is an improvement from batch system with an emphasis on microbial manipulation in rearing tanks. ZWD system can be interpreted as no water discharge during culture period, additional water that put into the system is to balance water level due to water losses caused siphoning and evaporation. It is approximately 2% of culture volume in every 6 weeks [13].

So far, the existing microbial form used may be in consortium, biofilm, periphyton, biofloc forms or has separated compartments such as biofilter in recirculation aquaculture system (RAS). In ZWD system, the form of microbial used is consortia that have been added regularly to the system during cultivation period. The purpose of additional microbial consortia regularly is to control microbial loop works in appropriate way. In addition, the presence of microbial control is to keep dominancy of selected microbes that play a role in predicted microbial loop. However, to maintain the availability of microbial cultures, the system must be equipped with separated microbial cultivation facilities. Consequently, there is control to maintain microbial culture from contamination and to keep the microbes in their optimum growth. **Table 1** below is a summary of the characteristics of each microbial-based closed system.

Microbial-based. closed system	Characteristics	References
Zero water discharge	– Low/no water discharge – Improved system from batch system – Emphasize in microbial manipulation – Nitrogen toxic compound removal by microbial loop system – Microbial consortia added regularly to the system – Microbial component is kept dominant in the system – Need additional compartment for separated microbial cultivation	[13]
Biofloc	– Low/no water discharge – Improved system from batch system – Add carbon source to enhance heterotrophic bacteria consortium – Emphasize in C/N ratio in the system – 'waste' Nitrogen is converted to high concentration of total suspended solid (microbial biomass) that can act as highly protein feed for cultured animal – Consider well mixing and aeration to compensate BOD in the system	[26, 37, 38]
Periphyton	– Low/no water discharge – Improved system from batch system – Need organic substrate i.e. bamboo to periphyton attachment – Input organic matter i.e. manure and chemical fertilizers to trigger periphyton growth – Sometimes, needs additional carbon source to maintain C/N ration in the system – Periphyton acts as nitrogen toxic removal system and food source for cultured animal	[39–41]
Biofilm	– Low water discharge – Improved system from batch system – Nitrogen toxic compound removal was done by formed biofilm during culture period – No control of microbial consortia in biofilm – Biofilm can also be a food source for cultured animal	[18]
Defined biofilm	– Biofilm production needs additional reactor and attachment substrate – Defined microbial consortia in biofilm (predominantly nitrifying bacteria) – Main purpose is to remove nitrogen toxic substance in system – Can be applied in the system or in external unit i.e. biofilter	[42, 43]
RAS	– No water discharge – Many treatment process involved including physical and chemical treatment – Microbial compartment is in biofilter – Biofilter has defined microbial consortia – Isolated and clear-water system – Main purpose is biologically secured and hygiene aquaculture product – Investment cost and operational cost is higher than other systems	[44–46]
Green water technique	– Low water discharge – Use batch system – Mostly autotrophic microalgae used as microbial component in the system – Utilized chemical fertilizer and organic waste to trigger phytoplankton grow – No control in microbe community in the system – Main purpose is to provide natural food for cultured animal	[47, 48]

Table 1. Characteristics of microbial-based system.

3. Preparation of ZWD system

Proper designed systems and good microbial management are important parts to optimize production efficiency in intensive cultivation using ZWD system. This section will describe the main and supporting facilities of ZWD systems for crustacean cultivation, particularly white shrimp and giant freshwater prawn. Besides, the selection of microbial components that are suitable for shrimp and prawn culture and how to prepare the cultivation will be explained in this section.

3.1. Design construction and facility

As ZWD system is an improvement system of batch culture, the main facilities provided are similar to batch system. ZWD system installation can be constructed in a rectangular or circular culture tank that is equipped with several basic utilities commonly used in aquaculture. Here is the list of facilities for ZWD system.

1. Culture tanks for nitrifying, heterotrophic bacteria, and microalgae culture. Separation is necessary for easier maintenance purpose. Proper maintenance is critical to keep optimum performance of microbial components. Tank sizes of nitrifying bacteria and microalgae are suggested to have minimal capacity about 20% from culture tanks, while tank size of heterotrophic bacteria is suggested to have minimal capacity about 2.5% from culture tanks.
2. Aeration equipment including aerator, silicon hose, and air stone. The aerator provides continuous oxygen supply with airflow rate of 28 L/min [49]. Proper aeration is critical, not only to provide oxygen to shrimp for effective feed utilization and growth, but as importantly to oxidize liquid, solid, and gaseous waste in the system. Oxygen level in water must be maintained between 4 and 6 ppm [44]. However, 6 ppm is recommended to support optimum growth. With high inputs of feed, there is higher demand for oxygen by shrimps and by the microbial community in the water.
3. A net covering is used to avoid pollutant entry to culture pond, and it is more important to reduce water evaporation, which can affect salinity level significantly. In addition, covering reduces light penetration through the water column to suit intensity level for the microalgae population in water.
4. A thermometer to monitor daily culture temperature.
5. Feeding trays to administer and control sufficient daily feed amount.
6. $CaCO_3$ and gravel, as a substrate for nitrifying bacteria attachment as well as a buffering agent.

In addition, shelter is required for some crustacean such as prawn (*Macrobrachium rosenbergii* De Man). Prawn is much more aggressive than shrimp. There is a risk of cannibalistic behavior that emerges when prawns are cultured at high density, especially during their grow-out phase and molting period [11]. Without shelter, they do not have enough niche for each individual. Several shelters that have been proved in previous studies were textile vertical substrate [11] and cubical bamboo shelters [51]. These shelters have been proven to improve prawn culture productivity. **Figure 2** below represents the components of ZWD system.

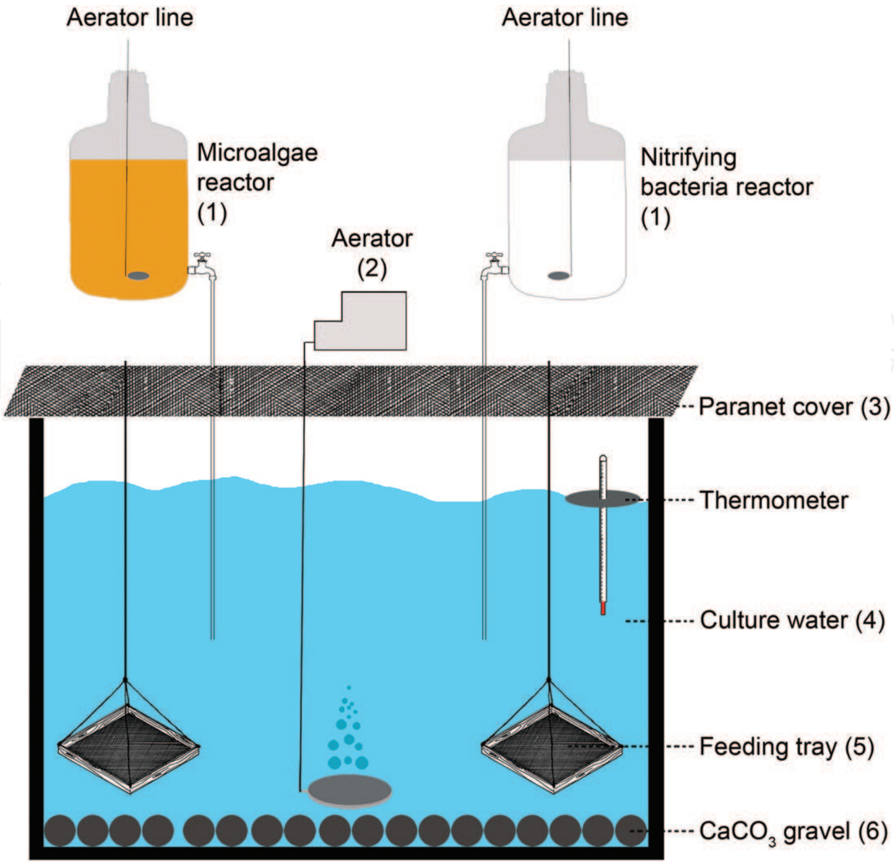

Figure 2. Basic facilities of ZWD system [13, 50].

3.2. Microbial components

The microbial component of ZWD system can be understood through three key functional groups: nitrifying bacteria, autotropic microalgae, and heterotrophic bacteria. When managed correctly, a diverse healthy microbial community contributes directly and indirectly to shrimp nutrition and growth while processing excess nitrogen waste in the system. Once established, the community becomes stable, competitively excluding harmful opportunistic pathogen and therefore improving health and immune competence of shrimps. The key to maximize these benefits is in understanding and managing the microbial community in the system.

3.2.1. Nitrifying bacteria

Nitrifying bacteria live in a wide variety of habitats, including soil, freshwater, seawater, rocks, and sediment. Nitrifying bacteria are widely used in aquaculture practice and usually in the form of ammonium-oxidizing bacteria (AOB) and nitrite-oxidizing bacteria (NOB). AOB derive energy through the process of catabolism of ammonium into nitrite; the bacteria included are genera *Nitrosomonas, Nitrosococcus, Nitrosospira, Nitrosolobus*, and *Nitrosovibrio*. While NOB oxidize nitrite to nitrate, the bacteria included are genera *Nitrobacter, Nitrococcus, Nitrospira,* and *Nitrospina*. The bacteria are classified into litoautotrophic bacteria because they use inorganic compounds as a source of energy and CO_2 as a carbon source [52].

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According to previous studies, the most common nitrifying bacteria that were stable during cultivation period were *Nitrosomonas* sp. and *Nitrobacter* sp. [13]. *Nitrosomonas* and *Nitrobacter* are Gram-negative and aerobe obligate that is used as final electron acceptor. The bacteria can grow and multiply as individual units or in the form of biofilms. *Nitrosomonas* reproduces with binary fission, while *Nitrobacter* reproduces with budding. *Nitrosomonas* and *Nitrobacter* have different generation time, *Nitrosomonas* is every 8 h, and *Nitrobacter* is every 12 h. After 72 h, the population size of *Nitrosomonas* will be eight times greater than the population size of the *Nitrobacter* [53].

Several inhibition factors affect the nitrification process. Inhibitors may be short-term or long-term impact to their enzymatic activity. Some factors that can inhibit the rate of nitrification, there are alkaline pH, temperature, oxygen, salinity, organic and inorganic compounds, substrate for attachment and sunlight [53]. From mentioned factors, sunlight is an important factor to be taken into attention because sunlight can decrease the activity of bacteria *Nitrosomonas* and *Nitrobacter* in oxidizing ammonium and nitrite compounds [54].

The cultivation of nitrifying bacteria was performed in Winogradsky medium and in strong aeration with no light conditions (covered with black plastic). At the beginning of cultivation, bacterial culture of nitrification is activated by adding 10 ppm of ammonium. Ammonium levels are measured daily until it reaches 0 ppm. Furthermore, the activity of nitrifying bacteria is enhanced by continuously increasing the ammonium level up to 50 ppm. Later, the culture was scaled up to 10 L and then up to 500 L. The culture substrate used was $CaCO_3$ and gravel. **Figure 3** below shows schematic reactor for nitrifying bacteria cultivation.

3.2.2. Autotrophic microalgae

Besides nitrifying bacteria, microalgae are also an important component in ZWD system. Through their metabolism, microalgae take up nitrate obtained from final nitrification process by nitrifying bacteria as a nitrogen source [13]. Microalgae have the ability to conduct photosynthesis, which captures energy from light to synthesize organic carbon from inorganic carbon (CO_2). It accumulates organic carbon in forms of starch or other carbohydrates. Along

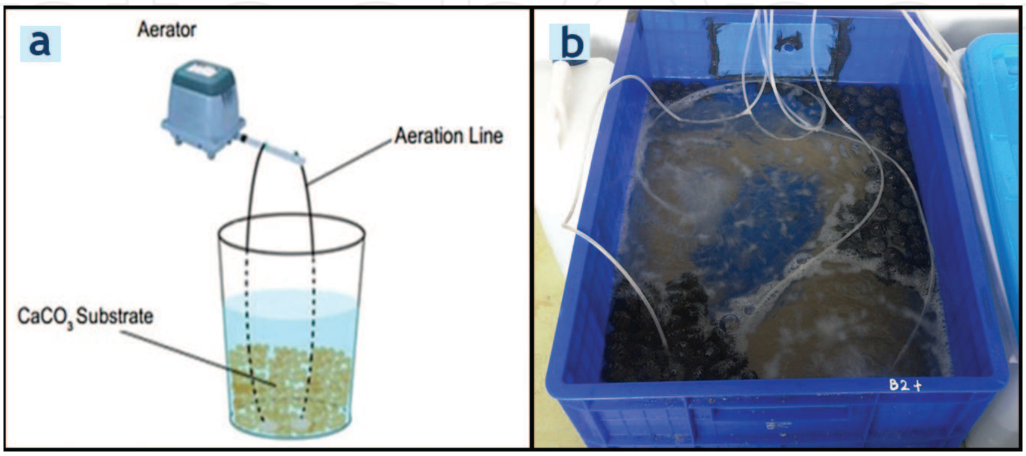

Figure 3. (a) Schematic diagram of nitrifying bacteria reactor [11, 13]; and (b) real nitrifying bacteria culture.

with other physiological processes, microalgae produce high-quality vitamin and minerals [58]. Moreover, microalga also has a good nutritional composition for aquaculture animals. Microalgae usually serve as a live feed at larval or early juvenile stage. Selected microalgae must have rapid growth rates, can cultivate to mass culture, and are stable growth to any environmental fluctuations.

Several species of microalgae that have been documented to success cultivation since 1997 are *Isochrysis* sp., *Pavlova lutheri, C. calcitrans, Chaetoceros muelleri, Chaetoceros gracilis, Thalassiosira pseudonana, Skeletonema* spp., *Tetraselmis suecica, Navicula* spp., *Nitzschia* spp. [55–59]. In practice, diatoms such as *C. calcitrans, C. muelleri*, or *C. gracilis* proved to increase shrimp productivity. Moreover, cell wall of these microalgae contains silicate, which is an important mineral for building exoskeleton of shrimps [60]. In addition, as microalgae cell density increases throughout their growth, it reduces light penetration to water body, so shrimp is not directly exposed to light (i.e., shading effects). Shading effect improves the production of shrimp, even though their exact mechanism of action remains unclear [61].

Microalgae begin to be cultivated from small scale (1 L) to large scale approximately 500 L. The cultured microalgae are diatoms (*C. calcitrans, C. muelleri,* and *C. gracilis*) for white shrimp and *Chlorella* sp. for prawn. For diatoms, medium used is f/2 medium [62] for stock culture up to 1 L, while medium used for *Chlorella* stock culture is Bold's Basal Medium [63]. Commercial media consisted of chemical fertilizer are used for large scale. Fertilizer must comprise a source of nitrogen, phosphate, silicate, and a small portion of the mineral. Examples of fertilizers used are NPK, Urea, ZA, mineral concentrates, etc. The cultivation uses batch system with condition parameter as follows: temperature is at interval 25–30°C, light intensity 3000–5000 lux, pH between 7.0–8.5, and aeration rate 3 L/min. Initial density is 10^5 CFU/mL and is incubated for 7–10 days until density reaches approximately 10^6 CFU/mL. **Figure 4** shows 1 L stock culture and 500 L scale-up culture for diatom *C. calcitrans*.

3.2.3. Heterotrophic bacteria

Heterotrophic bacteria can also uptake ammonium and nitrate as their nitrogen source [64]. The main advantage of adding particular heterotrophic bacteria is related to growth rate of heterotrophic bacteria that exhibit much faster than nitrifying bacteria [65]. Just like nitrifying

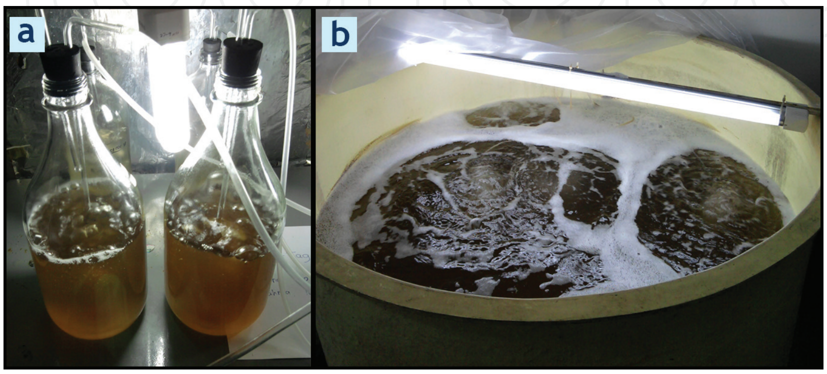

Figure 4. (a) Small scale and (b) large scale of *Chaetoceros calcitrans* in batch system.

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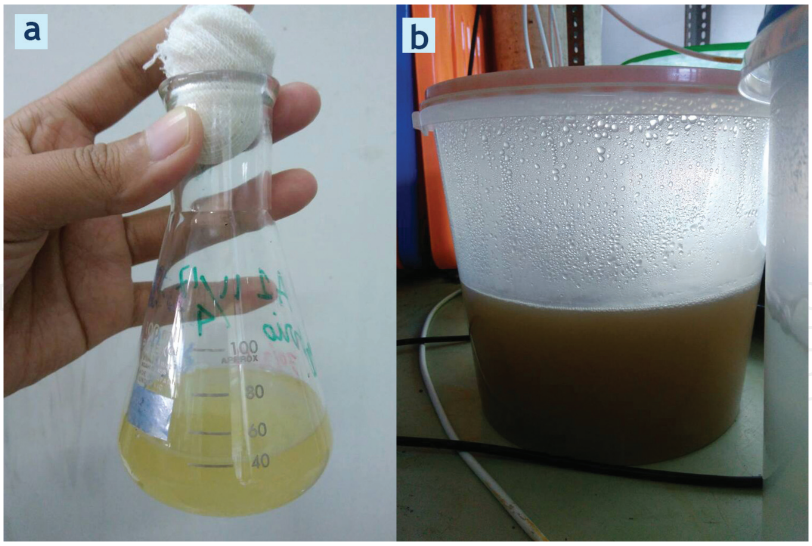

Figure 5. (a) Culture stock and (b) large scale cultivation of *Bacillus megaterium*.

bacteria, species of heterotrophic bacteria varies among different psychochemical conditions. Most common predominant species are *Bacillus megaterium* and *Bacillus flexus* [66]. *B. megaterium* is an example of well-studied bacteria for aquaculture application. The bacteria secretes high amount of extracellular enzymes, such as protease, carbohydrolase, and lipase, that can increase feed intake and digestibility in shrimp [67]. Other species that are also beneficial are *Shewanella algae* and *Halomonas aquamarina*. These natural-occurred bacteria have been studied and found to increase shrimp weight significantly [19].

Recent research progress proved that several heterotrophic bacteria genus (e.g. *Bacillus* sp. and *Pseudomonas* sp.) associated with microalgae through a mutual symbiosis. Although these heterotrophs have no direct role in controlling nitrogen cycle, their presence was proven to suppress growth of *Vibrio* spp. Mechanism of pathogenicity was done by quorum sensing [68]. This pathogen causes shrimp's mortality only when their population reaches a certain number of cells at least 10^6 CFU/mL [69]. Therefore, dominance of friendly heterotrophic bacteria can avoid pathogenic bacteria growth in the water, minimizing the risk of pathogen infection to the shrimp's gut.

Medium for culturing heterotrophic bacteria is nutrient broth medium for stock culture, while commercial medium is made from beef broth and ammonium chloride for large-scale cultivation. Heterotrophic bacteria were cultivated up to 15 L and then incubated for 24 h or until they reached the cell density of 10^7 CFU/mL. **Figure 5** below shows heterotrophic bacteria *B. megaterium* that are cultivated for culture stock and large-scale reactor.

4. Conditioning of ZWD system

There are several steps in conditioning of ZWD system; they are (1) microbial maturation in culture animal tanks; (2) acclimatization and stocking of shrimp or prawn; (3) feeding management; and (4) microbial manipulation that will be described in detail below.

4.1. Microbial maturation in rearing tank

Rearing tank conditioning was started by adding limestone ($CaCO_3$) and gravel at the bottom of the tank as substrate for nitrifying bacteria and then fill with disinfected seawater or freshwater (depends on cultured animal habitat). Afterward, 2% v/v of nitrifying bacteria consisted of AOB, and nitrite NOB at 10^6 CFU/mL is inoculated in the tank. 0.3 gr NH_4Cl (approx. 1 ppm of NH_4^+) is added per tank as ammonium source. After NH_4^+ and NO_2^- concentration was about 0 ppm, NO_3^- concentration was rising. It shows that AOB and NOB activities to convert ammonium to nitrate works. About 2% v/v of microalgae, i.e., *C. calcitrans* at 10^6 cell/mL and 0.05% v/v of heterotrophic bacteria, i.e., *B. megaterium* at 10^7 CFU/mL are inoculated into each tank. After the water turns to brownish color, the tank was ready to be used.

4.2. Acclimatization and stocking shrimp and prawn

After microbial maturation, we entered to the next steps or we can acclimatize shrimp or prawn in the same time with tank maturation. White shrimp PL-10 was acclimatized at room temperature (25 ± 1°C), while prawn was acclimatized from PL-40. Afterward, white shrimp was usually stocked at intensive farming using stocking density of 400 ind/m^3 and prawn at stock 60 ind/m^2. White shrimp and prawn were usually conducted for 90 and 60 days culture period, respectively.

4.3. Feeding management

Feed management was done by creating an estimation of daily feed (blind feeding; **Table 2**). The amount of feed was determined according to the mean body weight, estimated survival rate, and feeding rate, where:

$$Daily\ Feed\ (gr) = SD \times ABW \times FR \times SR \quad (1)$$

With: SD is the stocking density (Ind/tank), ABW is the average weight of shrimp (gr), SR is survival (%), and FR is feeding rate.

The feed was placed on the feed tray (ancho) and then checked to gain daily information of feeding accuracy (**Table 3**).

Average body weight (gr)	Feeding rate (%)	Survival rate (%)	Feeding tray monitoring intervals (h)
<1	10.0	100	3.5
1–3	8.0	98	3.5
3–5	6.0	96	2.5
5–7	5.0	94	2
7–9	4.0	92	2

Table 2. Blind feeding in super intensive white shrimp cultivation at 25 ± 1°C.

Ancho	Uneaten feed				Results	Decision
	1	2	3	4		
A	0	0	0	0	4/4	Add more 5–10%
B	0	0	0	+	3/4	Sufficient
C	0	0	+	+	2/4	Subtracted 5%
D	0	+	+	+	1/4	Subtracted 10–15%
E	+	+	+	+	0/4	Subtracted 20–30%

Table 3. Strategy to measure feeding efficiency following common feeding procedures created by Shrimp Club Indonesia in Lampung [personal communication].

Total consumption of feed and shrimp condition can be monitored via ancho based on the remaining amount of feed in the ancho. Feeding frequency was four times a day, given at 09:00, 12:00, 16:00, and 21:00 [13].

4.4. Microbial manipulation

This stage explores the characteristic of ZWD system that is similar to green water technique. Microbial components consisted of nitrifying bacteria, microalgae, and heterotrophic bacteria are added into the system every 2 weeks. The additional of microbial components is to maintain water quality and to balance microbial cycles in the system. In addition, microbial components are also given into the tank, if the ammonium concentration significantly rises. Similar to maturation step, nitrifying bacteria, microalgae, and heterotrophic bacteria were also added in the same volume to maintain the system performance.

5. Monitoring ZWD system

Monitoring is to evaluate the performance of ZWD system. Some parameters in the ZWD system that must be monitored are water quality and the growth performance of shrimp or prawn.

5.1. Water quality parameter

Maintaining water quality is important in aquaculture system because water is habitat of aquatic animal so it should be monitored periodically. Factors that affect water quality are temperature, dissolved oxygen, pH, and inorganic nitrogen concentrations. Water quality parameters are divided into two groups: psychochemical and microbiological parameters.

5.1.1. Psychochemical parameters

5.1.1.1. Temperature

The optimum range of water temperature allows aquatic organisms to perform metabolism and growth. Temperature is an important water quality parameter, because it can affect the amount of dissolved oxygen budget in water and increase the rate of chemical reaction. If temperature value exceeds the tolerance limit of the cultures animal, then it leads the animal die. A good temperature range for aquaculture is 25–32°C for the tropics [70]. The critical temperature for living water organisms ranges from 35 to 40°C. Various regions in Indonesia have average air temperature during the day between 12.8–38°C, and the difference depends on the elevation above sea level.

5.1.1.2. Dissolved oxygen (DO)

Dissolved oxygen is one of limiting factor for aquatic animals. Changes in oxygen concentration can have a direct effect to their respiration, if the oxygen is insufficient, the animal will be death. The number of organic compounds present in water body influences the amount of dissolved oxygen content. Organic compound is produced by microorganisms, consequently increasing biological oxygen demand (BOD), so that oxygen concentration is reduced in water body [71]. The minimum DO value that can be tolerated by crustacean is 4 mg/L, if less than that number, the shrimp will die. The recommended DO range for cultivation is 4–6 mg/L [44].

5.1.1.3. pH

pH is a value for expressing the concentration of hydrogen ions (H^+) in water. Water with a pH less than 4 and higher than 9.5 can cause death to living creatures and reduce aquatic productivity [71]. Water pH fluctuates with dissolved CO_2 and has an inverse relationship pattern; the higher the CO_2 content of the water, the pH will decrease and vice versa. This fluctuation will decrease when water contains $CaCO_3$ salt. The tolerance for aquatic life to pH depends on many factors including temperature, dissolved oxygen concentration, variations of differentiated anions and cations, species, and biota life cycle. The nonoptimal pH of water affects the growth and reproduction [72].

5.1.1.4. Inorganic nitrogen compounds

Inorganic nitrogen compounds are often found in water body in the form of ammonia, ammonium, nitrite, and nitrate. These compounds are strongly influenced by the oxygen content in water, when oxygen decreases, the ammonia formation increases. Naturally, the ammonia present in water is the result of animal metabolism and the decomposition of organic matter by bacteria. Ammonium tolerance for shrimp does not exceed from 3.95 ppm [9].

Another nitrogen form is nitrites. Naturally, such compounds are usually found in very little amount in water, because nitrite is unstable when there is oxygen. Other compounds are nitrate, which is the main nitrogen form in natural waters. Nitrate is one of the important compounds in the process of protein synthesis in animals and plants. The concentrations of nitrite and nitrate suggested in aquaculture were less than 25.7 ppm [73] and 200 ppm [22], respectively.

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5.1.2. Microbiological parameter

Aquatic microbes have an important role in aquatic ecosystems. These microbes can affect the health of aquatic animals and occupy key positions in the food chain by providing edible nutrient for the next higher trophic level of aquatic life. In addition, the microbes assist the biochemical reactions that recycle most of the elements in the aquatic environment as well as in the soil. The amount of microbes in the water depends on the amount of organic compounds present in water body that usually interprets in biological oxygen demand (BOD) and chemical oxygen demand (COD) index. Higher organic matter causes dissolved oxygen content to be smaller because microbes use oxygen to oxidize organic matter.

5.2. Growth performance of shrimp or prawn: biological parameters

Production performance is also evaluated by a number of biological parameters. However, the list describes only for most priority parameters; there are survival rate (SR), average daily growth (ADG), total biomass (Wt), and food conversion ratio (FCR). Here are the formulas to calculate important biological parameters.

a. Survival rate is a survival index of cultured animals in a cultivation process from the beginning of the animal stocked until the animal harvested. Survival was calculated using equation:

$$SR = \frac{Nt}{No} \times 100\% \tag{2}$$

Where, SR = survival rate (%), No = initial shrimp number (ind), Nt = final shrimp number (ind), t = culture period (day).

b. Average body weight represents the average of individual weight for the entire shrimp population, it can be done by measuring individual shrimp weight that the numbers follow statistical rule (n). ABW was calculated as follow:

$$ABW = \frac{\sum_{i=1}^{n} Wi}{n} \tag{3}$$

Where, ABW = average body weight (gr), Wi = body weight of the i-th shrimp (gr), n = number of shrimp or prawn measured (ind)

c. Average daily growth is average weight gained each day. ADG was calculated as follow:

$$ADG = \frac{Wt - Wo}{t} \tag{4}$$

Where, ADG = average daily growth (gr/day), Wt = total biomass at harvest (gr), Wo = total initial biomass at stocking (gr), t = culture period (day).

d. Total biomass is total weight of cultured animals at harvest. The total biomass was calculated as follows:

$$Wt = \sum_{i=1}^{n} Wi \quad (5)$$

Where, Wt = total biomass (gr), Wi = body weight of the i-th shrimp (gr)

e. Food conversion ratio (FCR) indicates a ratio of efficiency of feed, which is converted into animal body mass. FCR was calculated as follow:

$$FCR = \frac{Total\ feed\ given\ during\ culture\ period\ (kg)}{Total\ biomass\ (kg)} \quad (6)$$

6. ZWD performance in shrimp and prawn cultivation

Commodities used in this ZWD system were white shrimp (*L. vannamei*) and giant freshwater prawn (*M. rosenbergii* De Mann). The ZWD system was examined in pilot scale for both commodities, but the system was only applied in white shrimp at industrial scale. Here are the detailed explanations of each performance.

6.1. White shrimp cultivation

6.1.1. Pilot scale

Research on the performance of ZWD system on white shrimp nursery at pilot scale has been done [13]. The ZWD system was compared to batch system as control based on performances of water quality and biological parameters in the same stocking density (approx. 400 ind/m^3). Microbial components used were nitrifying bacteria consortia (*Nitrosomonas* sp. and *Nitrobacter* sp.) and microalgae *C. calcitrans*.

After 90 days culture period, all growth performance including average body weight, total biomass, survival rate, and specific growth rate was significantly higher in ZWD than those of batch systems (**Table 4**). In contrast, food conversion ratio in ZWD system was significantly lower than that of conventional culture system (1.27 and 4.10, respectively). Based on data, the FCR value of ZWD system was still tolerance range of shrimp (1.5–2.6).

Parameter	Batch	ZWD
Final ABW (g)	5.45 ± 0.28	8.24 ± 0.84*
SR (%)	27.22 ± 2.09	90.82 ± 2.5*
Total biomass (g)	160.48 ± 6.62	923.38 ± 42.15*
SGR (%)	7.24 ± 0.05	7.7 ± 0.11*
FCR	4.10 ± 0.66	1.27 ± 0.29*

*Significant difference $p < 0.05$.

Table 4. Biological performance during 90-day cultivation period [13].

Water quality during 90-day cultivation period was still in tolerance levels for white shrimp in both systems, as seen in **Table 5**. As a concern, threshold value of ammonium in ZWD systems was higher than batch system, it was 0.69 ppm compared to 0.59 ppm, but it was not significantly different ($p > 0.05$). High ammonium level was caused by higher feed input in the ZWD system, it was about 44% compared to the batch system. A large feed input would affect the increment of ammonium level in culture, but ZWD system has rapid ammonium breakdown capacity that was accomplished through microbial manipulation. Moreover, nitrite level also did not differ significantly in both systems. These levels suggested that the ammonium and nitrite breakdown capacity of ZWD systems are higher than batch system. In addition, better ammonium and nitrite breakdown capacity were shown through nitrate level that was higher in ZWD system after 90-day culture period. The nitrate level in ZWD system reached 42.9 mg/L, while in batch system, it was 14.17 mg/L.

Besides the benefits of nitrifying bacteria and microalgae in maintaining water quality, they also indirectly inhibit pathogenic bacteria *Vibrio* spp. growth. Microbiological assessment shown that excessive organic matter did not increase the number of *Vibrio* spp. Previous research reported that marine diatom, such as *C. calcitrans,* has the ability to secrete fatty acids and esters, antibacterial compounds which inhibit several heterotrophic bacteria growth, such as *Vibrio* spp. [74]. Another analysis of predominant bacteria found in shrimp cultivation using ZWD system reported that the water contained following species, based on the most dominant bacteria found *B. flexus*, *Geobacillus stearothermophilus*, five species of *Bacillus* sp., *Pseudomonas oleovorans, Pseudomonas peli,* and *Xenorhabdus nematophilus* [66]. *Bacillus* sp. known as probiotic bacteria suggested that ZWD system not only achieved acceptable psychochemical parameters, but also microbiological parameters support shrimp growth as well.

6.1.2. Industrial scale

Research conducted for grow out white shrimp cultivation using ZWD system at industrial scale has been applied in UD. Populer, Gresik, East Java [50]. The research used PL-17 white shrimp as cultured animal at low salinity water (5 ppt) in ZWD system. Culture period was 70 days for three different stocking densities; there were 200, 300, and 400 ind/m^3 further referred as SD200, SD300, and SD400.

Parameter	Batch	ZWD	Tolerance range
Temperature (°C)	25.96–30.63		25–32°C for tropic area [70]
pH	7.63–8.80		4–9.5 [71]
DO (mg/L)	7.42 ± 0.52	6.81 ± 0.5	> 4 mg/L [44]
NH_4^+ (mg/L)	0.20–0.59	0.07–0.69	< 3.95 mg/L [9]
NO_2^- (mg/L)	0–3.20	0–3.15	< 25.7 mg/L [73]
NO_3^- (mg/L)	1.38–14.17	1.04–42.9	< 200 mg/L [22]

Table 5. Water quality measurement during 70-day culture period [13].

Based on the research, biological parameters have been documented as shown in **Table 6**. Interestingly, survival rate has a value inversely to total biomass. Treatment SD400 has the lowest survival rate of 70.59 ± 6.15% but has the highest total biomass, which is 44.13 ± 4.44 kg. It was because in higher stocking density has a higher stress level than lower density culture, such as space competition, higher cannibalism level, etc. If the total number of living individuals was calculated, SD400 has the largest number of about 280 individuals, and this number was larger than SD200 and SD300, which were 186 and 237 individuals, respectively. In addition, given feed was the highest proportion compared to other two treatments, so that nutrient sources were greater too, it can be seen from its size distribution, SD400 has a size of 100–150 ind/kg of 95.85%. Specific growth rate and FCR did not differ significantly. Based on the value of productivity, three treatments have reasonable value for cultivation conducted in Indonesia that was 1.72–2.0 kg /m^3 for 100–120 days of culture on stocking density of 60–300 ind/m^3.

For water quality parameters, all parameters had acceptable tolerance limits. Among all treatments, there was no significantly difference ($p > 0.05$). **Table 7** showed the measurements for water quality for 70 days of culture period.

6.2. Prawn cultivation

The performance of ZWD system on the nursery phase and grow-out phase of giant freshwater prawn (*M. rosenbergii* De Mann) was evaluated using addition of two types of shelters: three-dimensional cubical bamboo shelters (on nursery phase experiment) [51] and vertical textile shelters (on grow-out phase experiment) [11].

6.2.1. ZWD system and additional bamboo shelters

Research was conducted at laboratory scale using PL40 giant freshwater prawn taken from commercial hatchery in Sukamandi, West Java [51]. Microbial components used were nitrifying bacteria and *Chlorella* sp. that inhabit in freshwater. The research used ZWD system with addition of cubical bamboo shelter that has dimension of 0.6 x 0.6 x 0.2 m. Experimented

Parameters		**Stocking Densities (ind/m^3)**		
		200	**300**	**400**
SR (%)		93.52 ± 3.32^a	79.11 ± 5.81^b	70.59 ± 6.15^b
SGR (%)		4.64 ± 0.14^a	4.22 ± 0.24^a	4.40 ± 0.25^a
FCR		1.05 ± 0.07^a	1.06 ± 0.08^a	1.14 ± 0.14^a
Total biomass (kg)		27.7 ± 1.55^a	36.25 ± 3.01^b	44.13 ± 4.44^c
Size distribution (%)	100–150 ind/kg	86.77^a	91.93^{ab}	95.85^b
	150–250 ind/kg	13.23^a	8.07^{ab}	4.15^b
Productivity (kg/m^3)		1.39	1.81	2.21

Note: Means of values with same superscript along rows are significantly different ($p < 0.05$).

Table 6. Biological parameter measurement during 70-day culture period [50].

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Parameter	SD200	SD300	SD400	Tolerance range
Temperature (°C)	29.3–30.1	29.8–30.1	30.4–30.9	25–32°C
pH	7.61–8.27	7.71–8.36	7.27–8.38	4–9.5
DO (mg/L)	4.90–8.50	5.00–8.00	5.60–7.80	> 4 mg/L
NH_4^+ (mg/L)	0–0.5	0–3.0	0–0.30	< 3.95 mg/L
NO_2^- (mg/L)	0–5.0	0.2–3.0	0.2–5.0	< 25.7 mg/L
NO_3^- (mg/L)	5.0–35.0	5.0–30.0	5.0–25.0	< 200 mg/L

Table 7. Water quality measurement during 70-day culture period [50].

treatments in the system were differences in the number of bamboo shelters installed in culture tank (2 x 1 x 0.4 m) and variation of stocking density. There were no shelter as control (C), two shelters (20% of water volume) (CB1), and four shelters (40% of water volume) (CB2) (**Figure 6**). Stocking density in sheltered tank was two times higher than control, and it was 60 ind/m^2. The cultivation was conducted for 28 days.

At the end of cultivation period, growth performance of prawns with shelters has better performance than control (no shelter). Treatment of CB2 produced final total biomass of 196 ± 0.09 gr, specific growth rate of 8.24% gr/day, final mean body weight of 2.17 ± 0.89 gr, and final mean body length of 6.50 + 0.91 cm. Shelter installment also resulted in a significant higher survival rate that was significantly different to control, as seen in **Table 8**. It was due to the availability of a larger territory area, vertically and horizontally, which will reduce contact possibility of prawns that was reared at high stocking densities, and at minimum contact with each other, cannibalistic behavior was suppressed. In addition, culture productivity increased with system using shelter, both CB1 and CB2 (30 and 39%, respectively). This suggests that shelters installation with the addition of microbial consortium improves prawn cultivation productivity.

Based on water quality parameters, ZWD system balances dissolved nitrogen concentration in water during cultivation period. pH value, dissolved oxygen (DO), ammonium nitrogen (NH_4^+),

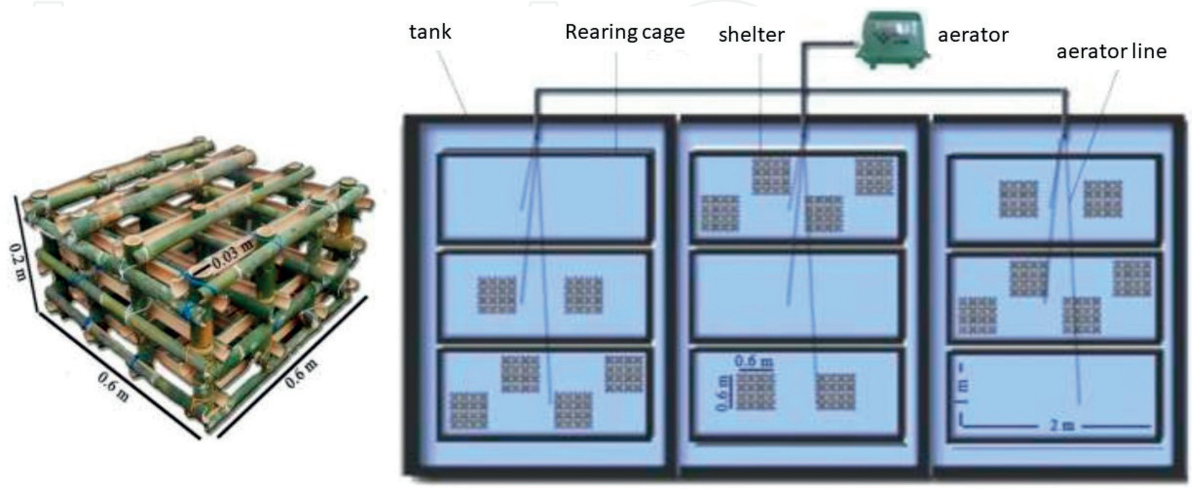

Figure 6. Cubical-bamboo used as shelters (left); cultivation scheme for prawn cultivation using bamboo shelters (right) [51].

Parameters	Control	CB1	CB2
Total biomass (gr)	141 ± 0.03^{a}	183 ± 0.05^{b}	196 ± 0.09^{b}
SGR (gr/day)	7.74	7.88	8.24
Survival rates (%)	77	90	92
Productivity increase compared to control (%)	—	30	39

Note: Different letters in the same column denote a significant difference ($p < 0.05$).

Table 8. Growth performance during 28-day cultivation period [51].

nitrite nitrogen (NO_2^-), and nitrate nitrogen (NO_3^-) remain stable during the culture period, keeping it in tolerance range of prawns (**Table 9**).

6.2.2. ZWD system and additional textile vertical substrate

Prawn (*M. rosenbergii* De Man) cultivation on the grow-out phase takes a period of 60 days, longer than nursery phase, which is only 14 days. For longer period of water quality maintenance, several inoculation of nitrifying bacteria suspension (10^5 CFU/mL) was needed. Prawn cultivation was conducted using five different stocking densities: 30, 40, 50, 60, 70 ind/m^2 into each pond (2 x 1 m^2) [11]. Shelter used in this research was textile vertical substrate that was placed in culture tank as seen in **Figure 7**.

Bodyweight, body length, specific growth rate, and survival rate were measured during 60-day cultivation (**Table 10**). As stocking density increases, prawn growth rate and survival rate decrease, due to competition of resources and risk of cannibalism. In final measurement, the highest mean bodyweight and body length were achieved by 70 ind/m^2 stocking density (11.46 ± 4.52 g and 10.70 ± 1.50 cm, respectively). Specific growth rate and survival rate from all treatments range between 1.393–2.569%/day and 67.1–76.3%, respectively. Overall, biological performance did not differ significantly among other stocking densities ($p > 0.05$).

From these results, the presence of a microbial component in rearing tank, mostly attached on solids, such as $CaCO_3$, and the textile vertical substrate surface directly improves water quality. In addition, textile vertical shelters reduce aggressive behavior of prawns due to secured spaces for each individual.

Parameters	Control	CB1	CB2
pH	7.47–8.45	7.40–8.07	7.13–7.96
DO (mg/L)	6.43–8.10	6.37–747	5.87–7.63
NH_4^+ (mg/L)	0.041–0.121	0.033–0.022	0.044–0.116
NO_2^- (mg/L)	0.011–0.156	0.012–0.237	0.011–0.210
NO_3^- (mg/L)	13.33–53.22	5.00–44.97	8.33–37.491

Table 9. Water quality parameter during 28-day cultivation period [51].

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Ammonium concentration during cultivation was maintained acceptable for prawn culture. On early stage of cultivation period, ammonium reached to almost undetectable level but gradually increased to 0.09–0.12 mg/L, observed in all treatments. The highest ammonium concentration (0.12 mg/L) was obtained in the highest stocking density (60 ind/m^2), followed respectively by the lower stocking densities (**Table 11**). Nitrite concentration during cultivation remained below the tolerance limit of freshwater prawn (1 mg/L). Nitrite concentration was gradually decreased during the cultivation from 0.064–0.066 mg/L to 0.015–0.032 mg/L in all treatments, showing nitrification process of conversion of nitrite to nitrate. Nitrate concentration remained stable between 53.9–71.0 mg/L in all treatments. Overall, DO levels during

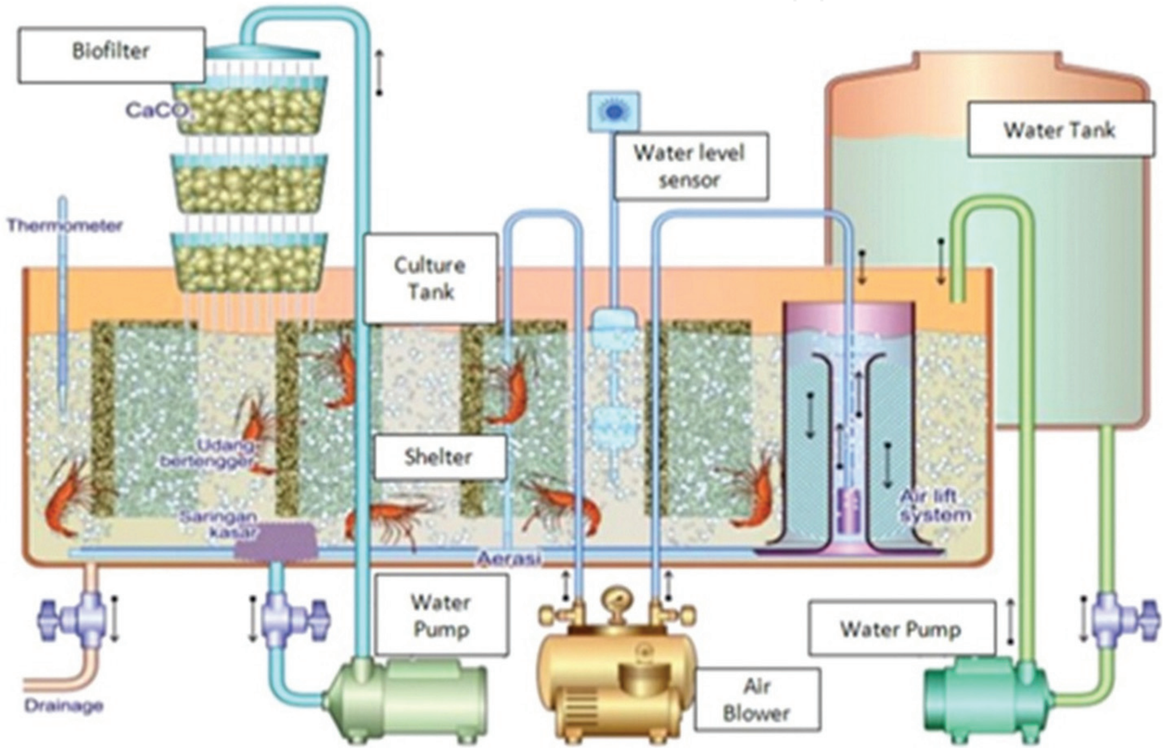

Figure 7. Cultivation scheme for prawn cultivation on grow-out phase using vertical textile shelters [11].

Parameters	Treatment (ind/m^2)				
	30	40	50	60	70
Mean body weight (g)	11.37 ± 4.96	9.34 ± 3.82	10.80 ± 5.62	10.98 ± 5.86	11.46 ± 4.52
SGR (% per day)	2.569	1.393	2.105	1.916	1.893
Survival rate (%)	78.3	76.3	70.0	70.0	67.1
Total biomass (g)	534.48	532.57	734.06	922.3	974.37
Total feed (g)	526.75	625.87	864.49	1080.5	1334.14
Feed conversion ratio (FCR)	0.99	1.18	1.18	1.17	1.37

Table 10. Prawn growth performance during 60-day cultivation period [11].

Parameters	Stocking densities (ind/m^2)				
	30	40	50	60	70
pH	6.74–8.42	6.74–8.15	6.31–7.91	6.42–7.95	6.52–7.96
DO (mg/L)	5.6–8.3	5.15–8.3	5.2–8.4	4.3–8.5	3.7–8.4
NH_4^+ (mg/L)	0.044–0.089	0.045–0.101	0.044–0.116	0.042–0.123	0.044–0.092
NO_2^- (mg/L)	0.015–0.066	0.017–0.064	0.021–0.066	0.027–0.066	0.032–0.065
NO_3^- (mg/L)	23.5–56.3	22.9–54.0	30.6–63.0	36.6–71.0	25.8–56.6

Table 11. Water quality parameters during 60-day cultivation period [11].

cultivation remained within tolerance range of prawn (min. DO is 4 mg/L) [44]. DO level decreased with response to increasing prawn growth, and the lowest level was 3.7 mg/L in 70 ind/m^2. pH level was relatively the same among all treatments, ranged between 7.71–7.96. From these parameters, it can be concluded that water quality during cultivation is suitable for the grow-out phase of prawns.

Based on microbiological parameters, there was a significant difference in microbial diversity between batch and ZWD system. In batch system, the water contained various species during cultivation period such as *Xenorhabdus japonica, Bacillus megaterium, Micrococcus luteus,* and *Bacillus amyloliquefaciens*, whereas in ZWD system, *B. megaterium* dominated from second week to the end of cultivation period. Batch system showed a fluctuation on bacterial dominance, which probably due to various abiotic and biotic factors, and therefore, tanks were prone to pathogen infection. In contrast with ZWD system, *B. megaterium*, which has been proven to benefits prawn growth, constantly dominated water during cultivation. Its dominance limited other bacteria domination, including pathogenic bacteria as well. Total pathogen bacteria particularly *Vibrio* spp. was counted. *Vibrio* spp. in rearing tank reached 10^0–10^3 CFU/mL during cultivation in both systems. Based on the results, *Vibrio* spp. abundancy below 10^6 CFU/mL was safe for prawn [69].

It was proven that the use of nitrifying bacteria can maintain good water quality, and textile vertical substrate can support a higher stocking density, better growth, larval survival rate, and profit of prawn *M. rosenbergii* de Man during the grow-out phase.

7. Technical and economic feasibility analysis

7.1. Technical feasibility analysis

Technical feasibility is an assessment of supporting factors in cultivation. It is concern in natural, social and cultural factors for successful cultivation [50]. The technical feasibility of parameters using ZWD system consists of site selection in terms of topography and structure, quantity and quality of water sources, accessibility, available of production facilities such as electricity source, seed producers, and distance to government research facility.

Research in technical feasibility has been conducted in white shrimp culture using ZWD system at grow-out stage. Based on criteria, north coastal areas of East Java Province are suitable for white shrimp urban aquaculture using ZWD system, such as Tuban, Lamongan, Gresik, Sidoarjo, Pasuruan, Probolinggo, and Situbondo, etc. The sites were included in the following criteria:

- Geographically, the sites are adjacent to the sea, therefore easy to get seawater
- Environmental condition is suitable for shrimp growth such as temperature and humidity ranged between 22 and 34°C and 50–86%, respectively
- Land topography is included into lowland making it possible to construct shrimp farming
- The sites are close to domestic market in Surabaya as capital cities of East Java province and Lamongan that had many cold storage companies (19.8%) and fish processing units (23.7%) compared to total available units in Indonesia
- There is no social conflict of interest

7.2. Economic feasibility analysis

The economic feasibility was analyzed to calculate the overall cost and profitability from real implementation of ZWD system at industrial scale in Gresik (East Java, Indonesia) [50]. White shrimp juveniles were stocked on different stocking densities, providing that varying stocking densities can affect financial calculation, considering operational as well as investment expenses. Assumptions used were to produce 1000 kg shrimp/cycle during 10 years production period.

The results showed that the best biological feasibility was in stocking density at 400 ind/m^3. The lowest operational and investment cost was stocking density at 400 ind/m^3, because it needed the least area to produce 1000 kg/cycle. So, this economic feasibility takes the best performance in 400 ind/m^3. Based on calculation, operational cost consisted of shrimp seeds, feed, labor, electricity, seawater, algae and probiotics, chemical and disinfectants, harvesting, packaging and delivery, and depreciation costs that has the proportion as seen in **Figure 8**. The operation cost at 400 ind/m^3 reached Rp 44,227,125, while the highest component contributions to the investment costs were production ponds cost (36–42%) and land purchasing (21–24%) and total cost reached Rp 318,230,000.

Financial projections were calculated to predict the break event point. Profit could be calculated by subtracting the total revenue with production cost. Assumed that there were four production cycles per year, in which 1000 kg shrimps were produced per cycle with duration of 3 months, in 1 year, the farm would produce 4000 kg of fresh shrimp with total revenue Rp 240,000,000. The production cost in 400 ind/m^3 stocking density was achieved Rp 40,227,125 per cycle or Rp 160,908,500 per year and has the highest profit Rp 79,091,500 among all stocking density treatment. A total of 400 ind/m^3 stocking densities treatment will achieve a payback period after 4 years of operation.

To assess the economic feasibility of ZWD system, financial ratios were calculated. Financial ratio analyzed consisted of NPV, IRR, B/C ratio, and Pay Back Period (PBP). Based on financial

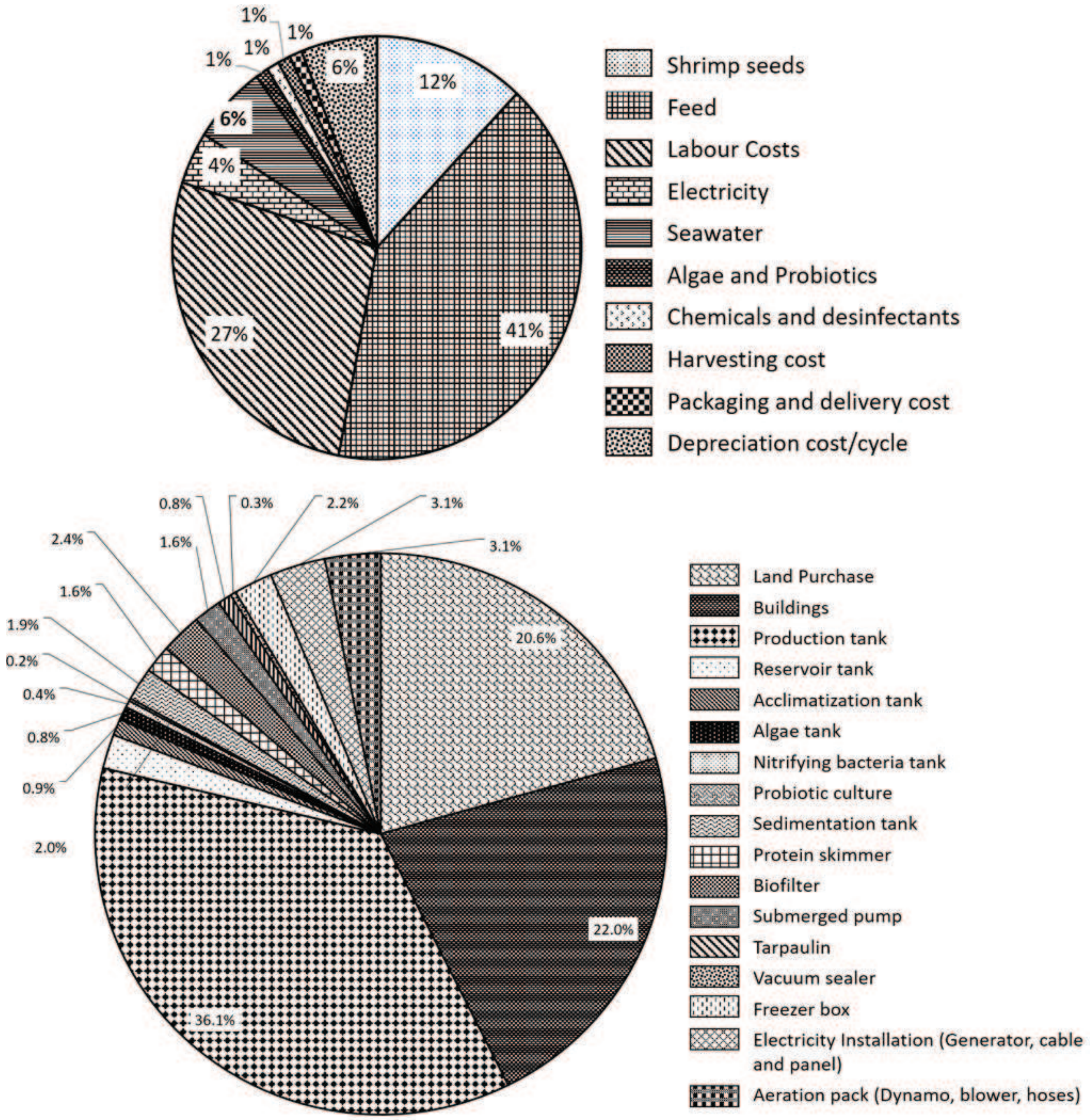

Figure 8. (above) Diagram of operational cost and (below) diagram of investment cost components (in percentages) at 400 ind/m^3 [50].

analysis, the project was financially feasible if the NPV is positive, IRR value is higher than discount factor, and B/C ratio value is higher than one [75]. The financial ratio calculations were presented in **Table 12**. It can be clearly seen that 400 ind/m^3 stocking density was financially feasible, because it has positive NPV (Rp 69,439,955), IRR value that was higher than discount factor (15.49%), and B/C ratio that was higher than 1 (1.22).

Parameter	SD (400 ind/m^3)
Investment cost	Rp 318,230,000
Revenue	Rp 60,000,000
Production cost/cycle	Rp 44,227,125
Profit/cycle	Rp 15,722,875
Profit/kg shrimp	Rp 15,773
BEP (kg)	**2804**
Net present value (NPV)	**69,439,955**
B/C ratio	**1.22**
Pay back period (year)	**4.02**
Internal rate of return (IRR) (%)	**15.49**

Table 12. Financial ratio calculation of ZWD system at 400 ind/m^3 stocking density to produce 1000 kg shrimp/cycle using low salinity [50].

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