Stocking density of Rotifera, *Hexarthra mira* on water quality and production

I Ardi¹,³, S Cahyaningsih¹ and E Setiadi²

¹Research Institute for Ornamental Fish Aquaculture, Depok, West Java, Indonesia
²Research Institute for Freshwater Aquaculture and Fisheries Extension, Bogor, West Java, Indonesia
³Corresponding author: ardiidil@yahoo.com

Abstract. Live feed (rotifer) has an important role in fish seed production. The type of rotifers used in this study is *Hexarthra mira*. *Hexarthra mira* is included in monocyclic species which only has sexual periods in autumn and in winters. *Hexarthra mira* is found in a dormant egg. Water quality greatly affects the growth and survival rate. This study was aimed to determine the effect of *Hexarthra mira* on water quality in culture media on population and production. Different stocking density as treatments were as followed: A) 1 ind/ml; B) 2 ind/ml; C) 3 ind/ml; and D) 4 ind/ml. The results showed that *Hexarthra mira* density can influence (P<0.05) water quality on the pH parameter. High stocking density (C and D) were significantly higher than that of A and B in term of population and production (P<0.05).

1. Introduction

Rotifer is one of natural food which plays an important role, especially, for larval rearing due to many sizes of rotifer and becomes suitable for any species of fish larvae. Generally, rotifer has a body length ranges from 60-273 μm and 92-170 μm in body width [1]. On the other hand, rotifer moving is very slow, thus it easy to catch by fish larvae and easy to culture in order to adequate for seed production.

Rotifer consists of *Monogononta*, *Bdelloidea*, dan *Seisonidea*. *Monogononta* class comprise of 1500 species, *Bdelloidea* class comprise of 350 species, and *Seisonidea* class comprise of 2 primitive species. *Hexarthra mira* belongs to *Monogononta* class (Biology wise 2018). Classification of *Hexarthra mira* based on [2] consisted of Kingdom *Animalia*, Phylum *Rotifera*, Class *Eurotatoria*, subclass *monogononta*, ordo *Flosculariaceae*, family *Hexarthridae*, genus *Hexarthra*, species *Hexarthra mira*. *Hexarthra mira* has two symmetric cilia with C tape shape, *Cingulum* is tape cilia which appears around the mouth but cilia is smaller than that of *trochus*. Body of *Hexarthra mira* characterized by cone shape and possesses two medians (dorsal and ventral), two *anterior-lateral* and two *posterior-lateral* [3].

*Hexarthra mira* involving monocyclic and only has a sexual period in the autumn season wherein winter season *Hexarthra mira* find in egg dormant. Optimum water temperature for *Hexarthra mira* is ranged from 13 to 28°C, pH > 7. Water quality optimal could keep the survival of *Hexarthra mira* [4]. One way to achieve of availability of *Hexarthra mira* as a live food to supply for seed production should increase the stocking density of *Hexarthra mira* culture. Therefore, research focused on stocking density and water quality is needed. The pH value is an important parameter for aquatic organisms including Rotifera because the pH as a limiting factor parameter and also such as temperature and dissolved oxygen (DO). High stocking density has been led to degrading water
quality due to waste products by species culture. Thus, the objective of this experiment is to determine the optimal stocking density on population growth, production, and water quality.

2. Materials and methods

2.1. Culture container and rotifer test
Twelve of plastic containers with a size of 49.5 cm length × 34 cm width × 25.5 cm height were used. Each plastic container is fulfilled by water with a volume of 30l with pH value that was the same for all treatments. Rotifer, *Hexarthra mira* was used as a rotifer test

2.2. Inoculant adding and feeding *Hexarthra mira*
Inoculant was adding on day 1 after water quality checking. Adding *Hexarthra mira* inoculant into the container based on the treatment. Adding *Hexarthra mira* should free contaminant. Formulation feed was fed based on a procedure by Redjeki [1]. To maintain the pH stable, the 1 g of lime flour was adding into the media for all treatments.

2.3. Sample of *Hexarthra mira*
The container that content of *Hexarthra mira* was homogenized beforehand then 1litter was taken to homogenized again. Thereafter, 10 ml was taken for a sample then homogenized and 1 ml was taken for counting the density, after that the sample was stocked into the container culture again.

2.4. Water quality parameter
Water quality parameters such as temperature, Oxygen dissolved (DO), ammonia, nitrite, nitrate, total dissolved solids (TDS), and conductivity were measured using a water checker. Water quality parameters were measured at the beginning (D0) and by the end of the experiment (D14).

2.5. Experimental design and data analysis
The experiment using completed randomize design with four treatments and 3 replicates were performed. The different stocking density of *Hexarthra mira* was used as a treatment. The treatments of this experiment were as followed:

- a) *Hexarthra mira* 1 rotifer/ml
- b) *Hexarthra mira* 2 rotifer/ml
- c) *Hexarthra mira* 3 rotifer/ml
- d) *Hexarthra mira* 4 rotifer/ml

Data analysis using ANOVA and post hoc using Duncan’s test were applied. All data were analyzed using SPSS 17 statistic software.

3. Results and discussion

3.1. Results

3.1.1. Population and production of *Hexarthra mira*
Based on the result of different stocking density of *Hexarthra mira* on population and production data is shown in Figures 1 and 2.
Figure 1. Population of *Hexarthra mira* cultured at different stocking density. A) 1 ind/ml; B) 2 ind/ml; C) 3 ind/ml; and D) 4 ind/ml.

The population of *Hexarthra mira* for 14 days cultured showed increased with increasing culture period at all treatments. At 4 ind/ml (D) (550,000±17,320.5 ind) was the highest in a population of *Hexarthra mira* where the lowest 320,000±17,320.5 ind) found at 1 ind/ml (A) (Figure 1). The total production of *Hexarthra mira* cultured at 4 ind/ml (D) (430,000±17,320.5 ind) showed the highest while the lowest found at 1 ind/ml (A) (290,000±17,320.5 ind). Statistical analysis revealed that 3 (C) and 4 ind/ml (D) were better than that of 1 ind/ml (A) and 2 ind/ml (B) in terms of population growth and total production.

3.1.2. Water quality parameters

Water quality parameters such as temperature, DO, pH, ammonia, nitrite, TDS, and alkalinity during the cultured period of *Hexarthra mira* are shown in Table 1.

Table 1. Water quality parameters (temperature, DO, pH, ammonia, nitrite, TDS, and alkalinity) during a culture period of *hexarthra mira*.

| Treatment | Temperature (°C) | DO (ppm) | pH | Ammonia (ppm) | Nitrite (ppm) | TDS (ppm) | Conductivity (μS/cm) |
|-----------|-----------------|----------|----|--------------|--------------|-----------|---------------------|
| A         | 30.60±1.015a    | 6.74±0.011a | 8.09±0.011a | ND           | ND           | 127.1±1.2741a | 254.17±4.366a       |
Remarks: The values followed by the same superscript letter did not significantly different (P>0.05) *ND = Not Detected.

3.2. Discussion

The population of Hexarthra mira at all treatments showed increasing during the culture period (Figure 1). The increasing population occurred on days 3 where C and D treatments were better than that of A and B treatments. This indicated that different stocking density affects the production related to the number of initial stocking. Schroder [5] stated that the life cycle of Rotifera has heterogenic character namely parthenogenesis and periodically sexual reproduction. At the time of unsuitable, the rotifer becomes egg form dormant. Schrode [6] reported that the sexual reproduction test and dormant egg of Hexarthra mira, the hatching dormant egg become female and the next generation is parthenogenesis female. The life cycle of Hexarthra sp. It is different from Rotifera. Dormant egg of rotifer will produce males in the next generation. Male of Hexartra sp. can be reproduced by a mate. The development of Hexarthra sp. egg was ranged from 0.3-1.1 days with temperature ranging from 12-30ºC. Juvenile development was ranged from 0.5-2.1 days with temperature was the same as egg hatching and develop. Hexarthra sp. had shortened the life cycle than that of the other rotifer.

Unsuitable of environment can inhibit growth. However, the water quality parameters in this experiment were within the optimal ranged (Table 2). Galkovskaja [7] reported that the optimal temperature for Hexarthra sp. culture was ranged from 22-32ºC. Furthermore, the maximum respiration rate of Hexarthra mira at the temperature of 32°C while the decrease in respiration rate occurs at a temperature of 37°C. Temperature is correlated to dissolved oxygen, were increasing in temperature would be followed by decrease oxygen dissolved [8]. Londono [9] reported that the DO value was ranged from 5.57 – 8.72 ppm. pH value showed increased with increasing culture period. At the beginning (day 0), the pH value was 7.66 at all treatments where on day 14 the pH value increased with increasing stocking density. Different pH values at the present experiment might be caused by stocking density. Redjeki [10] stated that pH in the water influences by CO2 concentration and acid compound. This condition, CO2 will bond H+ become bicarbonate (HCO3-) as an initial reaction that affects the pH value in the water will decrease. However, in the present experiment showed that the pH value was increased with increasing stocking density. This condition may be the adding of lime flour into rearing media that influenced the increasing pH value. Herzig [10] suggested that Hexarthra sp. can grow with pH vale ranged from 7.8- 9.0. Geel [4] found that Hexarthra sp. can grow optimal at pH value is 7. Anyway, the present experiment showed that the Hexarthra sp was still growth even if the pH value was 8.91. This indicated that this rotifer was tolerated to pH value in base condition.

Ammonia is a metabolism product and degrades organic compounds by bacteria. Ammonia (NH₃) is toxic for aquatic organisms including rotifer. Ammonia is related to pH and temperature, the toxicity of ammonia becomes very toxic. Therefore, ammonia concentration should less than 0.1mg/L [8]. Redjeki [1] reported that free ammonia affects the number of rotifers. However, ammonia and nitrite concentrations in the present experiment are not detected. It is mean that those values were very low concentrations. Therefore, the present experiments suggested that ammonia and nitrite did not affect Hexarthra sp for growth and survival. Nitrite concentration should below 0.001 mg/liter [8].

Total dissolved solid is dissolved solids in the water that related to conductivity. Increasing TDS influenced by natural resources with a range from 200.85 μS/cm – 333.24 μS/cm [11]. In the present experiment, the TDS value at all treatments is still within the optimal range. Conductivity is water ability correlated to electricity. Conductivity influenced by kind and ion concentration [12]. Geel [13] stated that conductivity is correlated with aquatic productivity. The present experiment shows that conductivity at all treatments is not significantly different (P>0.05).

4. Conclusion

Based on the result can be concluded were as followed: Stocking density did not affect the population growth but the best production at a density of 3 individuals/ml. The high density of Hexarthra mira culture affects the pH value where increasing stocking density led to increasing pH value due to bicarbonate acid might be bound to adding the lime flour into the media during the culture period that containing calcium resulted in calcium bicarbonate form.
5. References

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