Experimental aquatic food chain system: Enhancing *Daphnia magna* as natural feed using eutrophic waters

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**Abstract.** An experimental aquatic food chain system was tested to utilize eutrophic water for producing useful natural feed. Water flea (*Daphnia magna*) was the main focus in this food chain system, in which it was grown on algal biomass bloom in eutrophic water, and harvested following its function as fish feed. The eutrophic waters were originated from a small lake Situ Rawa Kalong and catfish ponds located in Bogor. *Daphnia* was grown in an aquarium filled with the eutrophic waters from the two localities and reared without artificial feed, with an initial density of 5 individuals/L. The experiment was conducted with three replications for 19 days. Sampling was carried out five times to observe the *Daphnia* development. The original chlorophyll content in the aquarium of catfish pond and Rawa Kalong water were 257.64 and 211.91 mg/m³, respectively. It was observed that *Daphnia* consumed algal biomass to support their growth, indicated by the increase of the chlorophyll content in the water was followed by the rise of the *Daphnia* density to reach a maximum of 245 individuals/L at day 15 for the catfish aquarium, and then overgrazing occurred which resulted in a decrease in the chlorophyll content which was observed from day 12. The density of *Daphnia* in Rawa Kalong water at day 19 was 152 individuals/L and kept increasing. In the experimentation of *Daphnia* as natural feed, *Daphnia* was grown in four concrete ponds and fed with fish pellets. The *Daphnia* biomass was harvested when it reached a productivity of 1.67 g/m²/day. The feed conversion ratio (FCR) of fish pellets into *Daphnia* biomass increased with the age of the culture, with the highest value of 0.46 at day 36. Five Bada fish (*Rasbora maninjau*) per aquarium were then grown for 60 days, fed daily with progressive amount of *Daphnia* from 1.5 to 4.0 g, and the growth was compared with the control which was fed with an increasing amount of fish pellet. Fish length and weight were observed every 20 days to calculate the growth rate, weight gain, and feed conversion ratio. The results showed that the growth rate of the fish ranged from 0.24–0.98% per day. Fish pellets stimulated a higher growth in the initial phase of growth, but decreased rapidly with the fish age, while *Daphnia* provided a more stable growth rate. Based on its lower FCR value, *Daphnia* is more efficient as fish feed. Thus, *Daphnia* proved to be a suitable natural food source for *Rasbora* cultivation. Overall, our results indicated that the experimental aquatic food chain system composing of *Daphnia* as the first level consumer grazing on algal biomass, followed by *Rasbora* as second-level consumer devouring of *Daphnia* not only is an efficient and flexible system for reducing the nutrient level in eutrophic waters but also can support a stable growth of cultured fish.
1. Introduction

Eutrophic waters contain excessive amounts of nitrogen and phosphorus that pose a significant water quality problem. Anthropogenic activities have been pointed out as the primary sources of nutrient enrichment and cause of eutrophication. However, eutrophication is also a natural aging process for lakes and reservoirs and involves an increase in nutrient concentration and sediment thickness [1]. Nutrient removal and several mechanical treatment methods, such as the activated sludge process [2] have been developed to prevent eutrophication. However, these treatments take up a lot of space and are economically expensive. Also, they cannot be applied to non-point source pollution [3].

The current trend of work in ecological treatments to remove nutrients from water is the development of environmentally friendly approach which principally involves natural processes to lower the cost and reducing any adverse impacts, such as artificial wetlands [4] and floating islands [5]. Nevertheless, the nutrient removal efficiencies of these ecotechnologies are lower than that of mechanical treatment [3].

*Daphnia magna* is a planktonic crustacean that belongs to the Phyllopoda (sometimes called Branchiopoda), which are characterized by flattened leaf-like legs used to produce water current for the filtering apparatus. *Daphnia* is filter feeders [6] that filter suspended solids in the water and taking in microorganisms such as phytoplankton and other organic matters for their food.

In eutrophic waters, high levels of organic matter and nutrients stimulate the growth of microorganisms, such as phytoplankton. In this ecosystem, *Daphnia magna* plays an essential role in the food chain as a first-level consumer to transfer the energy to a higher trophic level [7]. The filter feeder entity of the animal was used to eliminate microorganisms present in the water and consequently remove the nutrients from the water column. Moreover, in the food chain, it transforms the biomass of various microorganisms producers into a larger form available to the animal consumers of higher level [8].

This study involves an experimental food chain system composed of phytoplankton and zooplankton which was utilized to remove nutrients from nutrient-rich eutrophic waters. The potential of *Daphnia* to improve lake water quality was assessed through its capability to remove phytoplankton and suspended materials from water while utilizing the biomass for natural fish feed to provide an added value to the efforts of lake water rehabilitation.

2. Materials and Methods

2.1. *Daphnia growth in eutrophic waters*

The water used as a growth medium for *Daphnia magna* was taken from catfish ponds located within the Research Center for Limnology LIPI and from a small lake Situ Rawa Kalong in Depok, West Java. *Daphnia* was grown in a 40 x 30 x 30 cm³ aquarium equipped with an aeration system and filled with 15 L of eutrophic water (figure 1). The initial density of *Daphnia* inoculated into the water was five adults per L. The experiment was carried out for 19 days without commercial feeding. The research was performed with three replications for both types of eutrophic water. During the investigation, the *Daphnia* population was sampled five times to determine its density.

Water quality parameters measured every day included temperature, pH, dissolved oxygen (DO), conductivity, total dissolved solids (TDS), and turbidity, while the concentration of chlorophyll and TSS was measured twice a week.
2.2. Daphnia as a potential natural fish feed

2.2.1. Daphnia productivity

The Daphnia population was grown for 36 days in four concrete tanks measuring 4 x 1 x 1 m$^3$ filled with eutrophic water of Lake Maninjau (figure 2), which was previously filtered using a fine sieve. During the experiment, Daphnia was fed with fish pellets (Prima Feed PF 500, PT Matahari Sakti). The population of Daphnia was allowed to grow for several days until it reached a level of density sufficient to be harvested regularly, which is one tank a day in the four-day rotation. Harvesting was done by scooping around 1/3 of the volume of the container. The biomass harvested was then weighed and the data obtained were processed to calculate the growth rate and productivity.

2.2.2. Daphnia as fish feed

Five freshwater fish Rasbora maninjau (figure 3) per aquarium were grown in six aquariums measuring 60 x 40 x 40 cm$^3$ and filled with 60 L of tap water. The aquariums were equipped with an aeration system. The experimental fish were fed with live Daphnia, whereas to the fish control, the commercial pellets were given (Prima Feed PF 500, PT Matahari Sakti). The fish were fed once a day. The calculation and adjustment of the amount of feed given were done every week. The experiment was carried out with three replications for 60 days with observations every 20 days on the length and weight of the fish. The data obtained were then processed to calculate the specific growth rate (SGR), weight gained (WG), and feed conversion ratio (FCR) according to the formula as follows:

- SGR = $\left[\frac{\ln W_t - \ln W_i}{T}\right] \times 100$  

- WG = $\left[\frac{W_t - W_i}{W_i}\right] \times 100$  

- FCR = $\frac{\text{Total feed intake (g)}}{\text{Total net weight gained (g)}}$
where \( W_t \) = biomass final weight, \( W_i \) = biomass initial weight, \( T \) = harvesting period

The difference in pellet administration between 10 and 20 g on \( Daphnia \) growth was analyzed by Anova single factor using excel with 95% confidence, while the difference in feeding between pellets and \( Daphnia \) against fish WG and FCR were also analyzed by Anova single factor using excel with 95% confidence. Supporting data of water quality were analyzed descriptively.

3. Results and Discussion

3.1. \( Daphnia \) growth in eutrophic waters

Both types of water used to culture the \( Daphnia \) population were categorized as eutrophic based on the chlorophyll content measured before the study, which was 59.3 mg/m3 for the catfish pond water and 75.9 mg/m3 for Situ Rawa Kalong water. The total nutrient status index (TNI) was used by Yang et al. [9] to assess the eutrophication status of the lake. TNI was the sum of indexes of all nutrient parameters, including chlorophyll concentration. A water body was classified eutrophic when its TNI value ranged 61–100, obtained from the equation \( \text{TNI} = 10 (2.5 + 1.086 \ln \text{Chl}) \). For catfish pond and Situ Rawa Kalong waters, the values of TNI were 85.3 and 83.2, respectively, that justified the eutrophic status of both types of water. The chlorophyll concentration reflects the nutrient content in the water that essential for algal growth. Therefore, the measurement of the chlorophyll concentration was taken to represent nutrient content in the water. At the same time, observation on TSS parameter showed a dynamic pattern most similar to the dynamic chlorophyll pattern, which indicated the suspended solid mainly consisted of phytoplanktons.

There was a relationship between the \( Daphnia \) population and chlorophyll concentration observed. The growth of the \( Daphnia \) population depends on the availability of phytoplankton as its food. Based on the dynamic’s population principle, at the beginning of the study, an increase in the chlorophyll concentration in the water was followed by the rise in \( Daphnia \) density until it reached the maximum that occurred on day 15 in the catfish pond aquarium (figure 4 and 6). However, the decrease in the chlorophyll concentration has started on day 12 which indicated the overgrazing of the phytoplankton by \( Daphnia \). Hence, after day 12, the growth rate of algae could not keep up with the grazing pace by \( Daphnia \) that resulted in the decline of algal population reflected in the chlorophyll concentration. Therefore, the decrease in the chlorophyll concentration was followed by a reduction in \( Daphnia \) density.

Catfish ponds showed to be a better growth media for \( Daphnia \), with a maximum density of 245 individuals/L at day 15, while the frequency of \( Daphnia \) in Rawa Kalong aquarium was still increasing at day 19 at 152 individuals/L. Both types of water tested were able to support the growth and development of \( Daphnia \) during the 19-day trial. \( Daphnia \) density obtained from a previous experiment [8] was 192 individuals/L from the initial mass of 20 individuals/L. The maximum density obtained at day six by Chrismadha & Widoretno [8] was 9.6 times its initial density. In this experiment, despite the initial mass of four times lower than the research of Chrismadha & Widoretno [8], the maximum frequency was 1.28 times higher, even though it was achieved 2.5 times longer, and

![Freshwater fish Rasbora maninjau employed in this study.](image.jpg)
it was 49 times its initial density of 5 individuals/L (figure 4). In figure 5, it is shown that the frequency of *Daphnia* in the fish pond reached its maximum at day 15 with the juvenile density was almost triple the density of adults, while the *Daphnia* density in Situ Rawa Kalong was still increasing and at day 19 the proportion of the juvenile density was more than double the density of adults. The higher proportion of juveniles can be attributed to food limiting conditions so the youth had less chance to grow up.

Water quality parameters consisted of temperature, pH, DO, conductivity, TDS, and turbidity are shown in figure 7. The temperature fluctuated in the range of 24.6–27.3°C. This temperature range was following the optimal temperature (24–28°C) for *Daphnia* [10]. The dissolved oxygen (DO) content was relatively stable in the range of 5.5–7.7 mg/L, while the pH ranged from 7.0 to 9.6. It is mainly following Lavens and Sorgeloos [11] which stated that the optimum pH for mass culture of *Daphnia* was between 7 and 8, temperature about 25°C and dissolved oxygen levels above 3.5 mg/L. The conductivity and TDS, however, increased during the experiment, indicating the release of ionic minerals that occurred along with organic matter degradation in the water. Turbidity in the fish pond aquarium increased until day 5, from 10.40 to 23.56 NTU, then decreased with the formation of microorganism flocks at the bottom of the aquarium. Water turbidity in the Situ Rawa Kalong aquarium was very high (54.49 NTU) at the beginning of the experiment, but then decreased at day 5 to 8.25 NTU, and then rose again to 40.22 NTU on the day 19. The formation of microorganism flocks at the bottom of the aquarium occurred in all aquariums (figure 7).

**Figure 4.** *Daphnia* density (individuals/L) during the experiment.
Figure 5. The density of juvenile and adult *Daphnia* in the fish pond and Situ Rawa Kalong during the experiment; FP = Fish pond, RK = Rawa Kalong.

Figure 6. Chlorophyll (mg/m³) and TSS content (mg/L) in the experimental water media.
Figure 7. Water quality parameters measured consisted of temperature, pH, DO, conductivity, TDS, and turbidity.

3.2. Daphnia as a potential natural fish feed
3.2.1. Daphnia productivity
This experiment showed that Daphnia grew well in Lake Maninjau water when fed with fish pellets. The average productivity level of Daphnia was 1.67 g/m²/day (figure 8a). Fish pellets used as feed strongly supported the growth of Daphnia, as could be seen from the feed conversion ratio (FCR),
which improves with age of the culture (figure 8b). The best obtained FCR value was 0.46 which means 0.46 kg of feed was needed to produce 1 kg of Daphnia biomass at day 36. This very low value of FCR is possible because fish pellets utilized have a maximum water content of 10%, while the water content inside the body of Daphnia magna reaches 86.4–97.3% [12][13]. Therefore, the Daphnia biomass harvested in this experiment came mostly from its water content. The lower FCR value may also be attributed to organic material accumulation followed the regular supply of fish pellets. Single-factor ANOVA analysis (95 % confidence) revealed that the growth of Daphnia resulted from the daily administration of fish pellets at 20 g was significantly greater than the fish pellet given at 10 g. Thus, to exploit the potential of Daphnia as natural fish feed, it is necessary to adapt the culture with feeding technology.

![Figure 8.](image-url)

(a) Comparison of the amount of feed given and Daphnia biomass produced. (b) Feed conversion ratio (FCR) of Daphnia cultivation for 36 days.

Measurement of the water quality parameters in the Daphnia rearing tanks included water temperature, pH, DO, and TDS, is shown in figure 9. The temperature fluctuated with an interval of 23.9 –28.1°C. DO was relatively stable in the range of 4.64–5.71 mg/L during the first two weeks, then tend to decrease until the end of the experiment from 5.30 to 2.11 mg/L on day 56. There was also pH fluctuation observed in the range of 7.2–9.3, whereas TDS experienced an increase during the study from 85 to 180 mg/L. DO declination and TDS inclination can be possibly attributed to organic accumulation in the water resulting from the pellet feed accumulation. During the experimental time, however, all water quality parameter measurements are still within the normal range for Daphnia growth.
3.2.2. Performance of Daphnia as fish feed

The results showed that the growth rate of *Rasbora* during the two months of study ranged from 0.24–0.98% per day (figure 10a). A higher fish growth rate was observed during the initial phase up to day 20, but after that a slightly higher growth rate was obtained by the Daphnia fed fish (figure 10b). This lower growth rate at the initial phase of Daphnia fed fish can be attributed to adaptation process, as the fish was previously used to have pellets for the meal. After passing through adaptation phase, the fish began to take advantage of better nutritional value of Daphnia, which was manifested by that of slightly better growth. Better nutritional value of Daphnia has been previously reported, particularly the protein contents, which was as much as 1.18–4.10% of the fresh biomass, while in the dried biomass it was 31–61% [12][13][14]. Also, a study by Maulidiyanti et al. [15] showed that fresh Daphnia contains 86.6% water, 6.3% protein, 0.6% fat, 4.1% carbohydrates, and 1.2% ash. This range of protein content is equivalent to Artemia so that it can replace the expensive Artemia as natural fish feed.

**Figure 9.** Water quality parameters in the Daphnia rearing tanks.
As fish feed, *Daphnia* has advantages in addition to its nutritional content due to its range of sizes that suitable to the mouth opening of fish since the early juvenile stage until the late young stage. Furthermore, *Daphnia* contains a broad spectrum of digestive enzymes such as proteinases, peptidases, amylases, lipases, and cellulase, which can serve as exoenzymes in the gut of the fish and fish larvae [14].

The water quality parameters measured in the aquariums of *Rasbora* rearing that included temperature, DO, pH, and TDS are shown in figure 11. In general, temperatures fluctuated but increased from 23.1 to 27.8°C, but are still within the normal range for fish growth. DO rise from 5.6 to 7.5 mg/L from day 35 until the end of the study. The value of pH fluctuates in the range of 7.8-9.8, while TDS shows the highest value at day 49 (62.3 mg/L) and then drops to 56.7 mg/L at the end of the experiment.
4. Conclusion
This study showed the potential of utilizing eutrophic water to produce water flea (Daphnia magna) biomass as an integral part of eutrophication abatements. The produced biomass was comparably useful to that of available commercial feed to be used as a natural feed in fish farming. Although need to take some adaptation phase, a slightly better growth was shown by the Daphnia fed fish comparing to that had conventional pellet meal. Additional fish pellets can be used to increase the growth of Daphnia if the lake water is not sufficiently eutrophic.

5. References
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