

**IMPROVEMENT THE Ca-Mg MINERAL RATIO OF MEDIA
CULTIVATION ON STRESS LEVELS IN FRESHWATER PRAWNS
(*Macrobrachium rosenbergii*)**

**Peningkatan Rasio Mineral Ca-Mg Pada Media Pemeliharaan Terhadap Tingkat
Stress Pada Udang Galah (*Macrobrachium rosenbergii*)**

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ABSTRACT

Freshwater prawns (*Macrobrachium rosenbergii*) are a freshwater species that is intensively cultivated. Calcium (Ca) and magnesium (Mg) are important factors in supporting the biological and physiological activities of freshwater prawns. The presence of minerals in the form of calcium and magnesium in the environment will be utilized by giant prawns, especially after the molting process. Calcium and magnesium important for the osmoregulation process and carapace formation of freshwater prawns. This research aims to see the best ratio of minerals in the form of calcium and magnesium to environmental stress in terms of the parameters of calcium and magnesium content in the prawns body, osmotic gradient, oxygen consumption level, and water quality during rearing. The experimental design used in this research was a completely randomized design (CRD) with five treatments and three replications. The treatment given consisted of increasing Ca-Mg in the media: A: 30 mg l-1 Ca + 0 mg l-1 Mg (1:0); B: 30 mg l-1 Ca + 15 mg l-1 Mg (1:0.5); C: 30 mg l-1 Ca + 30 mg l-1 Mg (1:1); D: 30 mg l-1 Ca + 45 mg l-1 Mg (1:1.5) and E: 30 mg l-1 Ca + 60 mg l-1 Mg (1:2). Based on the result, treatment C: 30 mg l-1 Ca + 30 mg l-1 Mg (1:1) can reduce osmotic gradient values, oxygen consumption levels and glucose levels.

Key words: Ca-Mg ratio, environmental stress, *Macrobrachium rosenbergii*

ABSTRAK

Udang galah (*Macrobrachium rosenbergii*) merupakan spesies air tawar yang gencar untuk dibudidayakan. Kalsium (Ca) dan magnesium (Mg) merupakan faktor penting dalam menunjang aktifitas biologi dan fisiologi udang galah. Keberadaan mineral berupa kalsium dan magnesium di lingkungan akan dimanfaatkan oleh udang galah terutama setelah berlangsungnya proses molting. Kalsium dan magnesium berperan dalam proses osmoregulasi dan pembentukan karapas udang galah. Penelitian ini bertujuan untuk melihat rasio mineral berupa kalsium dan magnesium yang terbaik terhadap stress lingkungan ditinjau dari parameter

kandungan kalsium dan magnesium di tubuh udang, gradien osmotik, tingkat konsumsi oksigen, dan kualitas air selama pemeliharaan. Rancangan percobaan yang digunakan dalam penelitian ini adalah rancangan acak lengkap (RAL) dengan lima perlakuan dan tiga ulangan. Perlakuan yang diberikan berupa peningkatan Ca-Mg pada media: A: 30 mg l⁻¹ Ca + 0 mg l⁻¹ Mg (1:0); B: 30 mg l⁻¹ Ca + 15 mg l⁻¹ Mg (1:0,5); C: 30 mg l⁻¹ Ca + 30 mg l⁻¹ Mg (1:1); D: 30 mg l⁻¹ Ca + 45 mg l⁻¹ Mg (1:1,5) dan E: 30 mg l⁻¹ Ca + 60 mg l⁻¹ Mg (1:2). Berdasarkan data hasil penelitian, perlakuan C: 30 mg l⁻¹ Ca + 30 mg l⁻¹ Mg (1:1) dapat menurunkan nilai gradien osmotik, tingkat konsumsi oksigen dan kadar glukosa.

Kata Kunci: rasio Ca-Mg, stress lingkungan, *Macrobrachium rosenbergii*

INTRODUCTION

Freshwater prawn (*Macrobrachium rosenbergii*) is a high-value aquatic species with promising potential for aquaculture. As a species with broad environmental tolerance, its successful cultivation heavily relies on proper water quality and nutritional management. One critical factor in the maintenance medium is the balance of calcium (Ca) and magnesium (Mg) mineral ratios, which significantly influence growth, molting, and stress resistance in giant freshwater prawns (Miglio et al., 2021). The Ca-Mg ratio in water plays a crucial role in regulating physiological processes, such as exoskeleton formation and osmoregulation.

An imbalance in this ratio can lead to metabolic disorders, increased stress, and reduced productivity. Consistent with the findings of Rasid et al. (2021), an optimal Ca-Mg combination has been shown to improve reproductive performance and larval quality in giant freshwater prawns. Stress remains a primary challenge in intensive prawn farming, particularly under fluctuating environmental conditions. High stress levels are often associated with decreased immune resilience, elevated disease risks, and higher mortality rates (Liu et al., 2022). Therefore, a holistic approach is needed to mitigate stress through the management of maintenance media, including the regulation of the Ca-Mg ratio.

Prawn farming make the Ca and Mg serve specific roles. Calcium is essential for exoskeleton mineralization, while magnesium is involved in enzymatic regulation and energy metabolism. The influence of mineral ratios on stress is also linked to the prawns' ability to maintain osmotic homeostasis. Mineral imbalances can increase osmotic pressure, disrupting metabolic stability and reducing resistance to environmental changes, such as fluctuations in temperature, salinity, and oxygen levels (Kamaruding & Abdullah, 2021). This study aims to evaluate the effects of increased Ca-Mg mineral ratios in maintenance media on stress levels in giant freshwater prawns. This approach is expected to provide practical guidelines for water quality management, supporting sustainable aquaculture practices.

METHODS

Place and Time

The research was carried out from July to August 2024 at the Environmental Laboratory of the Aquaculture Study Program, Department of Fisheries and Marine Sciences, Faculty of Agriculture, University of Mataram.

Design

This study utilized a laboratory experimental method. The experimental design applied was a Completely Randomized Design (CRD) with five treatments and three replications. All treatments involved the addition of calcium at a concentration of 30 mg L⁻¹, based on Zaidy (2007). Meanwhile, magnesium was added at concentrations of 0, 15, 30, 45, and 60 mg L⁻¹ for each treatment. The Ca-Mg ratio levels tested in this experiment were as follows: A: Ca 30 mg L⁻¹ + Mg 0 mg L⁻¹ (1:0); B: Ca 30 mg L⁻¹ + Mg 15 mg L⁻¹ (1:0.5); C: Ca 30 mg L⁻¹ + Mg

30 mg L⁻¹ (1:1); D: Ca 30 mg L⁻¹ + Mg 45 mg L⁻¹ (1:1.5); and E: Ca 30 mg L⁻¹ + Mg 60 mg L⁻¹ (1:2).

The experimental setup included 15 glass aquariums measuring 50x30x30 cm, each equipped with an aeration system and a water heater. The aquariums were covered with black plastic to create a shaded environment, as prawns are nocturnal and highly active during the night. Additionally, each aquarium was fitted with shelters made of PVC pipe sections, matching the number of prawns stocked in each aquarium.

The maintenance medium for the giant freshwater prawns consisted of calcium-enriched water with magnesium added according to the specified treatments. The calcium (CaOH₂) and magnesium (MgSO₄) concentrations in the medium were achieved through dilution. Prior to dilution, calcium and magnesium levels were analyzed to determine the exact quantities required to meet the treatment concentrations. Strong aeration was applied during the dilution process to aid oxygen dissolution. Calcium and magnesium solutions were prepared weekly and stored in tanks, as water changes of 50% were performed weekly. The water volume for each aquarium was maintained at 30 liters.

The test animals used were giant freshwater prawns (*Macrobrachium rosenbergii*) with an initial body weight of 1±0.04 g and a length of 4.15±0.03 cm. The prawns were stocked at a density of 10 individuals per aquarium. Commercial feed containing 35% protein was provided ad libitum three times daily (morning, noon, and evening). The rearing period for this experiment lasted 35 days.

Sampling for water quality data was conducted every seven days, on days 0, 7, 14, 21, 28, and at the end of the experiment (day 35). A 300 ml water sample was collected from each aquarium using sampling bottles. These samples were analyzed to measure parameters such as temperature, pH, dissolved oxygen (DO), total ammonia nitrogen (TAN), hardness, and alkalinity. At the end of the study, observations were made on blood glucose levels, oxygen consumption rates, osmotic gradients, and calcium and magnesium content in the prawn's body.

Data Analysis

Experimental data were analyzed using Mini Tab Version 16. Analysis of variance (ANOVA) with a 95% confidence interval, if there was a significant effect, a further Tukey test was carried out (Steel & Torrie, 1993). Data is presented in the form of tables and graphs. liking for sensory testing (hedonic scale) with specifications, appearance, color, aroma, texture and taste.

RESULT

Glucose Levels

The results of analysis of variance (ANOVA) showed that the treatment had a significant effect on the glucose levels of giant prawns (P<0.05).

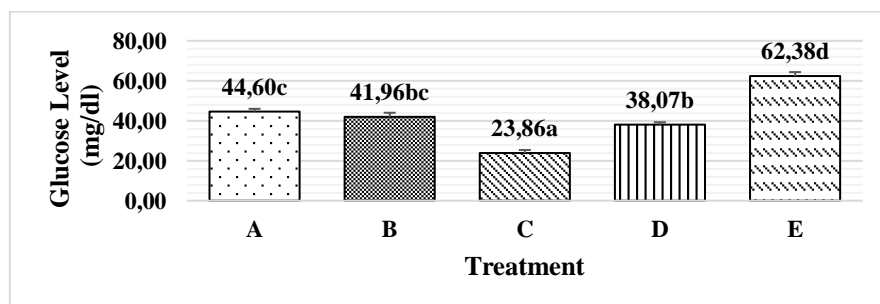


Figure 1. Glucose Level for 35 days of cultivation

Oxygen Consumption Rate

The results of analysis of variance (ANOVA) showed that the treatment had a significant effect on the oxygen consumption rate of giant prawns ($P < 0.05$).

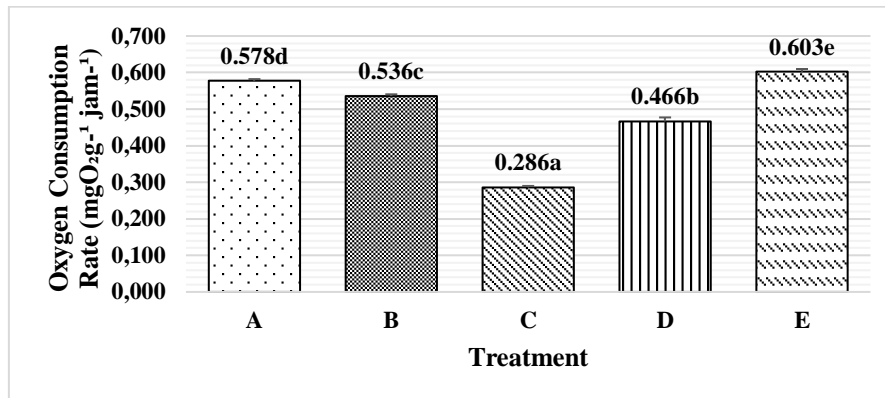


Figure 2. Oxygen consumption rate for 35 days of cultivation

Osmotic Gradient

The results of analysis of variance (ANOVA) showed that the treatment had a significant effect on the osmotic gradient of giant prawns ($P < 0.05$).

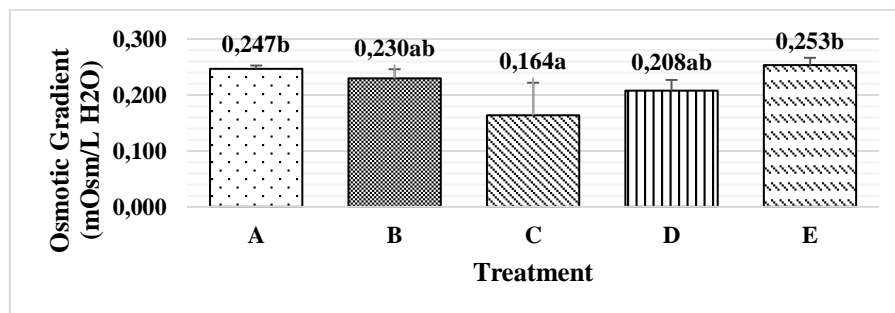


Figure 3. Osmotic gradient for 35 days of cultivation

Calcium Body Content

The results of analysis of variance (ANOVA) showed that the treatment had a significant effect on the calcium body content of giant prawns ($P < 0.05$).

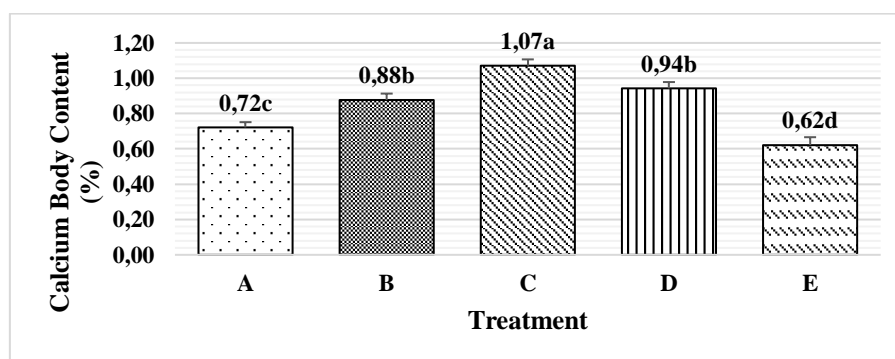


Figure 4. Calcium body content for 35 days of cultivation

Magnesium Body Content

The results of analysis of variance (ANOVA) showed that the treatment had a significant effect on the magnesium body content of giant prawns ($P < 0.05$).

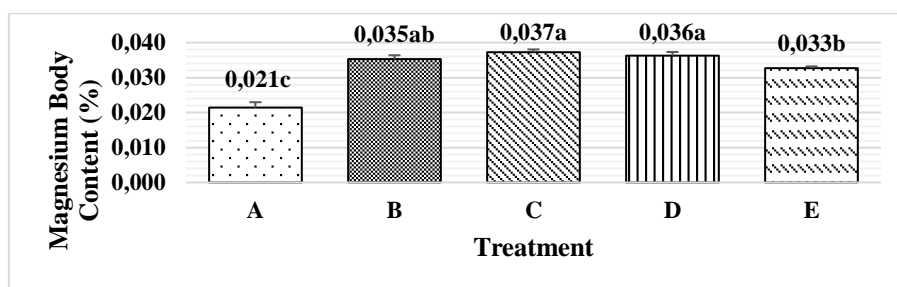


Figure 4. Magnesium body content for 35 days of cultivation

Water Quality During Cultivation

The range of physical and chemical water quality parameters during the experimental period was still reasonable range for the survival of giant prawns (Boyd & Zimmermann, 2000). Data on the range of water quality values in the experiment are presented in Table 1.

Table 1. Water quality during 35 days of cultivation

Parameter	Range
Temperature (0C)	27.32-28.41
pH	7.14-7.83
NH3 (mg/L)	0.0093-0.0126
Hardness (mg/L)	315.04-397.20
Alkalinity (mg/L)	44.52-48.25
DO	5.20-6.23

DISCUSSION

The physiological response to stress in prawns involves a significant increase in energy demand to cope with allostatic loads. This energy is derived from the mobilization of metabolic resources such as glucose and proteins, which are utilized to meet the requirements of anaerobic metabolism and osmotic homeostasis during stress periods (McEwen & Wingfield, 2003; Lorenzon et al., 2008). In this study, differences in blood glucose levels among treatments reflect variations in stress levels due to the different Ca-Mg ratios in the maintenance medium. Treatment C, with the lowest glucose level (23.86 mg/dL), indicates that this mineral combination reduces glucose mobilization as a stress response, suggesting that a 30:30 mg/L Ca-Mg ratio supports better metabolic stability. Conversely, treatment E (Ca 30 mg/L + Mg 60 mg/L) showed the highest glucose levels (62.38 mg/dL), likely due to osmotic imbalance caused by excessive magnesium. This aligns with previous studies indicating that high magnesium concentrations can disrupt osmoregulatory functions in crustaceans (Galkanda-Arachchige et al., 2021).

The increase in glucose levels during stress is also associated with the activation of stress hormones, such as cortisol or catecholamines, which stimulate glycogenolysis to provide rapid energy. However, excessive glucose levels can lead to lactate accumulation, potentially damaging tissues and reducing the long-term adaptability of prawns to environmental changes if not addressed (Jiang et al., 2023). These metabolic responses highlight the importance of managing the Ca-Mg ratio in the maintenance medium. An optimal mineral ratio not only supports growth but also enhances stress tolerance by balancing energy mobilization and metabolic stability. Further studies are needed to evaluate the interactions between mineral

ratios and other environmental factors, such as temperature and salinity, to optimize giant freshwater prawn aquaculture outcomes.

Low oxygen consumption rates in treatment C (Ca 30 mg/L + Mg 30 mg/L), at $0.286 \text{ mgO}_2\text{g}^{-1} \text{ h}^{-1}$, indicate less energy used for metabolism and more energy allocated for growth. According to Rubalcaba et al. (2020), oxygen consumption rates can serve as a parameter to assess the metabolic rate of aquatic organisms. Environmental factors, such as temperature and activity levels, influence oxygen consumption rates. Lower oxygen consumption suggests less metabolic energy expenditure and potentially greater energy availability for growth. In contrast, treatment E (Ca 30 mg/L + Mg 60 mg/L) recorded the highest oxygen consumption rate at $0.603 \text{ mgO}_2\text{g}^{-1} \text{ h}^{-1}$ (Figure 2).

In adapting to their environment, fish exhibit tolerance and resistance within a specific range of environmental variations. Osmotic fluctuations in environmental conditions trigger regulatory mechanisms to maintain internal environmental stability (Bal et al., 2021). Osmotic gradient, the difference between the osmolarity of prawn bodies and their medium, indicates comfort levels in prawns. Lower osmotic gradient values reflect a more isoosmotic state, where the body fluid concentration matches the medium. Treatment C (Ca 30 mg/L + Mg 30 mg/L) achieved the lowest osmotic gradient value of 0.164 mOsm/L H₂O, whereas treatment E (Ca 30 mg/L + Mg 60 mg/L) had the highest value of 0.253 mOsm/L H₂O (Figure 3).

The presence of magnesium affects calcium absorption in prawns. Observations show that the highest calcium content in prawn bodies was recorded in treatment C (Ca 30 mg/L + Mg 30 mg/L) at 1.07%. Conversely, the lowest calcium content was found in treatment A (Ca 30 mg/L + Mg 0 mg/L) (Figure 7). Insufficient or excessive magnesium concentrations hinder calcium absorption. This aligns with Rastegari et al. (2023), who reported that excessively high mineral levels impede mineral transfer from the environment to the organism. Increased mineral levels correspond to enhanced growth rates in prawn weight. As prawn weight increases, mineral content in the exoskeleton also rises (Eddy et al., 2020). The highest magnesium content in prawn bodies in this experiment was observed in treatment C (Ca 30 mg/L + Mg 30 mg/L) at 0.037%, which was not significantly different from treatment D (Ca 30 mg/L + Mg 45 mg/L) at 0.036%. However, a significant difference was found with treatment E (Ca 30 mg/L + Mg 60 mg/L) at 0.33%, suggesting that excessive magnesium inhibits its absorption for exoskeleton formation (Figure 4).

CONCLUSION

Based on the results a calcium and magnesium ratio of 30 mg l⁻¹ Ca + 30 mg l⁻¹ Mg (1:1) can reduce the osmotic gradient value, oxygen consumption rate and glucose levels.

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REFERENCES

- Bal, A., Panda, F., Pati, S. G., Das, K., Agrawal, P. K., & Paital, B. (2021). Modulation of Physiological Oxidative Stress and Antioxidant Status by Abiotic Factors Especially Salinity In Aquatic Organisms. *Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology*, 241, 108971.
- Boyd, C., & Zimmermann, S. (2000). Grow-out Systems-Water Quality and Soil Management. *Freshwater Prawn Culture: The Farming of Macrobrachium Rosenbergii*, 221–238.
- Eddy, M., Tbib, B., & Khalil, E.-H. (2020). A Comparison of Chitosan Properties After Extraction from Shrimp Shells by Diluted and Concentrated Acids. *Heliyon*, 6(2).
- Galkanda-Arachchige, H. S. C., Roy, L. A., & Davis, D. A. (2021). The Effects of Magnesium

- Concentration in Low-Salinity Water on Growth of Pacific White Shrimp (*Litopenaeus vannamei*). *Aquaculture Research*, 52(2), 589–597.
- Jiang, Q., Ji, P., Ao, S., Gao, X., & Zhang, X. (2023). Effects of Starvation and Refeeding on Glucose Metabolism and Immune Responses in *Macrobrachium rosenbergii*. *Marine Biotechnology*, 25(3), 447–462.
- Kamaruding, N. A. A., & Abdullah, M. I. (2021). Effects of Handling Stress and Moulting Cycle on Reproductive Performance and Digestive Capacity of Female Giant Freshwater Prawn, *Macrobrachium rosenbergii*. *International Aquatic Research*, 13(3), 195.
- Liu, B., Gao, Q., Liu, B., Song, C., Sun, C., Liu, M., Liu, X., Liu, Y., Li, Z., & Zhou, Q. (2022). Application of Transcriptome Analysis to Understand the Adverse Effects of Hypotonic Stress on Different Development Stages in the Giant Freshwater Prawn *Macrobrachium rosenbergii* Post-larvae. *Antioxidants*, 11(3), 440.
- Lorenzon, S., Giulianini, P. G., Libralato, S., Martinis, M., & Ferrero, E. A. (2008). Stress Effect of Two Different Transport Systems on the Physiological Profiles of The Crab Cancer *pagurus*. *Aquaculture*, 278(1–4), 156–163.
- McEwen, B. S., & Wingfield, J. C. (2003). The Concept of Allostasis in Biology and Biomedicine. *Hormones and Behavior*, 43(1), 2–15.
- Miglio, M. C., Zaga, B., Gastelu, J. C., Severi, W., & Peixoto, S. (2021). Survival and Metamorphosis of Giant River Prawn *Macrobrachium rosenbergii* Larvae in A Commercial Recirculation System With Artificial Seawater. *Aquaculture Research*, 52(12), 6063–6073.
- Rasid, R., Brown, J. H., Pratoomyot, J., Monroig, O., & Shinn, A. P. (2021). Interaction and Effects of Graded Levels of Dietary Phytic Acid on The Moulting Performance, Whole Body Mineral Composition Aand Availability on the Freshwater Prawn *Macrobrachium rosenbergii*. *Aquaculture*, 537, 736466.
- Rastegari, H., Nadi, F., Lam, S. S., Ikhwanuddin, M., Kasan, N. A., Rahmat, R. F., & Mahari, W. A. W. (2023). Internet of Things in Aquaculture: A Review of the Challenges and Potential Solutions Based on Current and Future Trends. *Smart Agricultural Technology*, 4, 100187. <https://doi.org/10.1016/j.atech.2023.100187>
- Rubalcaba, J. G., Verberk, W. C. E. P., Hendriks, A. J., Saris, B., & Woods, H. A. (2020). Oxygen Limitation May Affect the Temperature and Size Dependence of Metabolism in Aquatic Ectotherms. *Proceedings of the National Academy of Sciences*, 117(50), 31963–31968.
- Steel, R. G. D., & Torrie, J. H. (1993). *Prinsip dan Prosedur Statistika*. Jakarta: Gramedia Pustaka Utama.