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Monitoring Water Quality in Indoor Mud Crab (*Scylla serrata*) Farming Systems

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Abstract: *This study investigated the effects of two different water sources, artificial seawater prepared with Red Sea Salt and natural brackish water from Kampot, on the water quality dynamics, growth performance, and survival of mud crab (Scylla serrata) cultured in a recirculating aquaculture system (RAS) under indoor conditions. The experiment was conducted over 60 days using a controlled setup where individual mud crabs were stocked separately to reduce stress and cannibalism. Key water quality parameters, including temperature, salinity, pH, dissolved oxygen (DO), ammonia, and nitrite, were monitored daily using calibrated instruments. Growth and survival performance were assessed at the end of the trial, and statistical differences between treatments were analyzed using one-way ANOVA. Results showed that both water sources maintained stable water quality, with salinity around 30 ppt and DO levels above 5 mg/L. Ammonia and nitrite remained at non-toxic levels due to proper system management. However, water temperatures frequently exceeded 35 °C, a threshold known to induce thermal stress in Scylla serrata. Crabs cultured in artificial seawater exhibited significantly higher final weight and weight gain ($p < 0.05$) compared to those reared in brackish water, indicating a more favorable environment for growth. Despite this, survival rates were relatively low in both treatments (45% and 40%), likely due to elevated temperatures affecting molting and immune function. These findings demonstrate that artificial seawater is a viable substitute for natural water in indoor mud crab farming and may enhance growth outcomes. However, the study also highlights the need for temperature control strategies to improve survival rates. This research contributes practical insights for optimizing water quality management in tropical aquaculture systems and supports the development of sustainable, high-efficiency indoor farming practices for mud crab.*

Keywords: Aquaculture, Artificial Seawater, Growth Performance, Mud Crab, Water Quality

1. INTRODUCTION

Aquaculture is one of the fastest-growing food production sectors globally, playing a crucial role in enhancing food security, generating economic opportunities, and supporting rural development [1]. In Southeast Asia, including Cambodia, aquaculture is recognized as a strategic component for poverty alleviation and sustainable livelihoods, especially in coastal and riverine communities [2], [3]. Among various high-value aquaculture species, mud crabs (*Scylla spp.*) are particularly prized due to their rich nutritional profile, rapid growth rates, and strong consumer demand in both domestic and international markets [4].

Traditionally, mud crab farming in Cambodia has been practiced in coastal provinces such as Kampot, Kep, and Koh Kong, utilizing extensive and semi-intensive systems like earthen ponds and mangrove pens [5]. However, these traditional methods are increasingly challenged by environmental degradation, climate variability, disease outbreaks, and inconsistent yields [6]. To overcome these challenges, indoor aquaculture systems have emerged as a promising alternative, offering several advantages such as year-round production, enhanced biosecurity, better environmental control, and higher stocking density potential [7]. Despite these benefits, the success of indoor mud crab farming is highly dependent on maintaining optimal water quality parameters [8].

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Mud crabs are sensitive to changes in temperature, salinity, dissolved oxygen (DO), pH, ammonia, and nitrite levels [9]. Deviations from optimal ranges can impair physiological functions, suppress immune responses, reduce growth, and increase mortality, particularly during molting phases [10]. In addition to system design, the selection of an appropriate water source is a critical factor in indoor RAS operation. Natural brackish water collected from coastal areas is commonly used in mud crab farming; however, its use involves transportation costs, potential contamination risks, and variability in chemical composition. Artificial seawater prepared using commercial sea salt mixtures provides a practical alternative by allowing standardized and consistent ionic composition under controlled conditions [11], [12]. Despite its potential advantages, limited studies have directly compared artificial seawater and natural brackish water in supporting water quality stability, growth performance, and survival of mud crab under indoor tropical conditions [13].

Indoor mud crab farming depends heavily on maintaining stable water quality, as fluctuations in temperature, salinity, pH, dissolved oxygen, ammonia, and nitrite have been shown to significantly influence growth performance, physiological stability, and survival of *Scylla* serrata [14], [15]. However, limited data exist on how artificial seawater compares with natural brackish water in supporting these parameters under indoor RAS conditions in tropical climates. This knowledge gap makes it difficult for farmers especially those far from coastal areas to select suitable water sources and optimize system management.

Artificial seawater prepared with Red Sea Salt provides a practical alternative for indoor mud crab farming, especially for inland facilities located far from coastal water sources. Collecting natural brackish water requires transportation, labor, and time, which increases operational costs and introduces risks of contamination during transit. By contrast, Red Sea Salt allows farmers to create consistent, high-quality seawater on-site by dissolving the salt mixture in freshwater. This approach ensures stable ionic composition, reduces dependence on natural water collection, and supports year-round indoor aquaculture even in urban or non-coastal regions.

Salinity is a key environmental factor influencing the survival and physiological performance of mud crabs [16]. Because tolerance to salinity can vary depending on stock condition and culture environment, a preliminary laboratory salinity trial was conducted to determine a suitable salinity level for the mud crab stock used in this study prior to the main recirculating aquaculture system (RAS) experiment.

Mud crab (*Scylla serrata*) was selected for this study due to its high economic importance in Cambodia and its sensitivity to environmental changes, particularly temperature and salinity fluctuations [17], [18]. Therefore, this study aimed to compare artificial seawater prepared using Red Sea Salt and natural brackish water from Kampot in terms of water quality dynamics, growth performance, and survival under indoor recirculating aquaculture system (RAS) conditions. The findings are expected to provide scientific evidence to support the development of sustainable and efficient indoor mud crab farming practices in tropical regions. The research also explores the use of artificial sea salt mixtures and comparisons with natural brackish water from Kampot. By providing empirical data and practical insights, this study contributes to the development of more sustainable and efficient indoor farming practices for *Scylla spp.* in Cambodia.

2. METHODOLOGY

2.1 Mud crab farming system

The study was conducted in a controlled indoor facility using a Recirculating Aquaculture System (RAS) tailored for mud crab (*Scylla serrata*) culture. The RAS consisted of a series of fiberglass tanks connected through a centralized filtration system, which included mechanical filtration (for removing solids), biological filtration (to convert ammonia to less toxic compounds), and UV sterilization (to minimize microbial contamination). Each tank was equipped with an aeration system to ensure sufficient dissolved oxygen levels throughout the culture period [19]. To optimize animal welfare and reduce aggressive behavior, each crab was housed individually in separate rearing units, commonly referred to as "crab condos". This method prevents physical contact, especially during vulnerable molting periods, which are associated with high cannibalism risk in communal systems [20]. Individual housing allowed precise monitoring of growth and health for each crab, but it was used here for research purposes, in practical farming, crabs can be kept in shared tanks with proper management.

Before stocking, all tanks and system components were thoroughly cleaned and disinfected using a diluted chlorine solution, followed by multiple rinses with freshwater to eliminate any residual chemicals. The entire system was operated under ambient tropical indoor temperatures ranging between 30–35 °C. Continuous aeration and water circulation were maintained to mimic natural tidal conditions and prevent stratification or stagnation [13].

The recirculating aquaculture system (RAS) used in this study had an overall footprint of approximately 3.5 m × 2.5 m, with a total operational water volume of approximately 2,000 L. The system was designed to support up to 200 individually housed mud crabs, although 100 crabs were stocked per treatment during this experiment.

Water circulation followed a closed-loop sequence. Water from the crab housing tanks first passed through a mechanical filtration unit for solid waste removal. It then flowed into a biological filtration chamber (biofilter) to facilitate nitrification, converting ammonia to nitrite and nitrate. After biofiltration, water passed through a UV sterilization unit for microbial control before returning to the culture tanks.

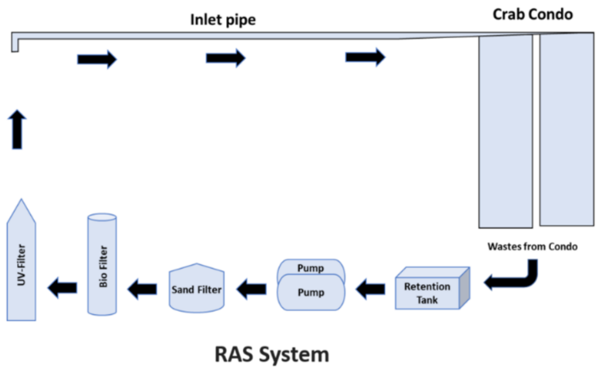

Fig. 1. Schematic diagram of the indoor recirculating aquaculture system (RAS) for mud crab culture showing water flow direction from culture tanks to mechanical filter, biofilter, UV sterilizer, and return line. The system includes a main circulation pump and a separate aeration pump.

Two pumps were used in the system:
(1) a main circulation pump (flow rate: approximately 2,000 L h^{-1}) responsible for continuous water movement through the filtration system, and
(2) a separate aeration pump supplying air stones in each crab housing unit to maintain dissolved oxygen above 5 mg L^{-1}.

Water entered the crab housing units through an inlet pipe with a controlled flow rate of approximately 15–20 L per minute, ensuring sufficient exchange without causing stress to the crabs. Each individual crab condo measured approximately 30 cm × 25 cm × 25 cm, allowing sufficient space for molting and minimizing aggressive interactions.

2.2 Preparation of water treatments

Two types of water were used as treatments: artificial seawater and natural brackish water.

- Natural brackish water was collected from Kampot, an estuarine coastal region where seawater mixes with freshwater. The water was pre-filtered and sterilized to remove suspended solids and pathogens before being introduced into the RAS.
- Artificial seawater was prepared by dissolving Red Sea Salt into dechlorinated freshwater to reach a salinity of 30 ppt, a level considered optimal for mud crab farming [11]. The amount of salt required was calculated using the formula:

$$\text{Salt(kg)} = \text{Volume (l)} \times \frac{\text{Target Salinity (g/l)}}{1000} \quad \text{(Eq. 1)}$$

Artificial seawater was prepared by dissolving a commercial marine salt mixture (Red Sea Salt) into dechlorinated freshwater to achieve a target salinity of 30 ppt. This salinity level was selected based on published optimal ranges for Scylla serrata culture and confirmed by the preliminary laboratory salinity trial conducted in this study [17].

The required salt mass was calculated using (Eq. 1), and exactly 3.0 kg of Red Sea Salt was dissolved in 100 L of freshwater. The solution was mixed under continuous aeration for 30 minutes to ensure complete dissolution and homogeneous ionic distribution. Final salinity was measured using a calibrated handheld salinity meter and recorded as 30.0 ± 0.2 ppt prior to use [14].

Red Sea Salt was selected because it is widely used in aquaculture research and practice due to its consistent and standardized ionic composition, which allows for reproducible preparation of seawater and minimizes variability associated with natural water sources. After preparation, the artificial seawater was allowed to equilibrate to room temperature (25–30 °C) before being introduced into the recirculating aquaculture system (RAS) [21].

Fig. 2. Commercial artificial sea salt (Red Sea Salt) used for the preparation of experimental seawater, selected for its standardized ionic composition and suitability for controlled aquaculture experiments.

Artificial seawater was prepared using a commercial marine salt mixture (Red Sea Salt®, Red Sea Ltd., Israel), which is widely available from aquarium supply stores and aquaculture equipment suppliers. This product is specifically formulated to replicate the ionic composition of natural seawater and contains balanced concentrations of major ions such as sodium (Na^+), chloride (Cl^-), magnesium (Mg^{2+}), calcium (Ca^{2+}), potassium (K^+), sulfate (SO_4^{2-}), and trace elements essential for marine organisms. In contrast, common cooking salt (sodium chloride, NaCl) contains primarily NaCl with minimal or no essential trace minerals. Therefore, cooking salt cannot be used to prepare artificial seawater for aquaculture purposes, as it does not provide the complete mineral composition required for osmoregulation, molting, and physiological stability in mud crabs. The use of formulated marine salt ensures consistent water chemistry and better suitability for marine aquaculture systems.

2.3 Laboratory salinity trial (preliminary base experiment)

Prior to the main RAS experiment, a laboratory-based salinity trial was conducted to provide a preliminary indication of a suitable salinity level for the mud crab stock used in this study. Five salinity treatments (15, 20, 25, 30, and 40 ppt) were prepared using Red Sea Salt. Each treatment consisted of a single plastic container (15 L capacity) stocked with one mud crab (initial weight 110–115 g). Because only one crab was used per treatment (n = 1), the trial had extremely limited replication and was intended only as a preliminary observation rather than a statistically robust experiment. The purpose of this trial was to provide an initial reference for selecting a practical salinity level for the subsequent recirculating aquaculture system (RAS) experiment.

The experiment was conducted under indoor laboratory conditions. Containers were filled with shallow water sufficient to submerge the crabs, reflecting their benthic behavior, and were continuously aerated. Salinity was adjusted gradually at a rate not exceeding 2 ppt per day and verified using a handheld salinity meter. Crabs were fed twice daily, uneaten feed and wastes were removed daily, and partial water exchange (10–20%) was performed when necessary to maintain water quality. The trial lasted 14 days, after which survival and weight change were recorded. A schematic view of the laboratory salinity experimental setup is shown in **Figure 3**.

Fig. 3. Laboratory salinity trial setup showing individual housing of mud crabs in plastic containers (15 L capacity) with shallow water depth and continuous aeration. One crab was stocked per container to avoid cannibalism and allow individual monitoring of survival and growth.

Based on the survival response observed during the laboratory trial, a salinity of 30 ppt was selected for use in the subsequent RAS experiment.

2.4 Experimental design

The experiment was conducted at the Institute of Technology of Cambodia’s aquaculture facility. A completely randomized design (CRD) was employed with two treatments:

- T1: Artificial seawater using Red Sea Salt
- T2: Natural brackish water from Kampot

Each treatment included 100 mud crabs, individually stocked in separate tanks under the same feeding and maintenance regime [16].

The experimental system consisted of two independent RAS units corresponding to the two treatments (T1: artificial seawater and T2: natural brackish water). Each RAS unit accommodated 100 individual rearing compartments (“crab condos”), resulting in a total of 200 individual housing units. A total of 200 mud crabs were stocked, with 100 crabs assigned to each treatment and one crab per compartment to prevent cannibalism and allow accurate monitoring of individual growth and survival.

Fig. 4. RAS System

The RAS used in this study was a commercially available aquaculture system adapted for mud crab culture under indoor conditions. Minor modifications were made to accommodate individual crab housing units ("crab condos") for experimental control.

Fig. 5. Mud Crab in Condo

2.5 Feeding regime

Mud crabs were fed twice daily at 08:00 and 17:00 throughout the 60-day culture period. The primary diet consisted of freshly chopped marine trash fish, occasionally supplemented with small shrimp to enhance dietary diversity and stimulate feeding response[5].

The daily feeding ration was initially set at 6% of individual body weight per day, based on recommendations for juvenile and sub-adult Scylla serrata under controlled culture conditions. Given the mean initial body weight of approximately 107 g, the initial ration corresponded to approximately 6.0–6.5 g feed per crab per day, divided equally between the two feeding times. Feeding rates were adjusted weekly (5–8% body weight range) following weight measurements and behavioral observations, particularly during pre-molt and post-molt stages when feeding activity typically fluctuates.

The proximate composition of marine trash fish (wet weight basis) typically contains 45–55% crude protein, 5–10% lipid, and high moisture content (70–75%), providing sufficient dietary protein to support tissue growth, molting, and exoskeleton synthesis. Mud crabs require relatively high dietary protein (≥40%) due to their carnivorous feeding habits and rapid growth potential. The use of natural marine feed ensured high palatability and adequate essential amino acid availability.

Uneaten feed was siphoned and removed 1–2 hours after feeding to minimize organic loading and prevent excessive nitrogen accumulation within the recirculating aquaculture system (RAS). Controlled feeding management was essential to limit ammonia (NH_3-N) production derived from protein catabolism and uneaten feed decomposition, thereby maintaining stable water quality conditions throughout the experiment.

2.6 Water quality monitoring

Water quality was monitored and maintained to meet the optimal conditions in **table 1** reported for *Scylla serrata* culture, ensuring proper growth, molting, and survival. The parameters and their target ranges were selected based on previous studies [17], [18], [15] and [22] are summarized in table 1.

Table 1. Optimal water quality parameters for Scylla serrata culture

Parameter	Optimal Range	Purpose	Reference
Temperature	28-35 ℃	Prevent thermal stress, support metabolism	[17], [19], [20]
Salinity	28-25 ppt	Suitable for juveniles and adults	[17], [23], [24]
pH	7.5-8.5	Maintain physiological stability	[21]

Dissolved oxygen	>5 mg/l	Prevent hypoxia	[6], [21]
Ammonia	<0.1 mg/l	Avoid gill damage and immunosuppression	[9]
Nitrite	<0.5 mg/l	Avoid disruption of hemolymph oxygen transport	[25]

Maintaining optimal water quality is essential because deviations from these ranges can negatively affect molting, metabolism, immune function, and survival of mud crabs. By controlling these parameters, environmental stress is minimized, ensuring that the experimental results accurately reflect the effects of the tested water sources rather than uncontrolled water conditions. This approach directly supports the research objective of evaluating the impact of different water sources on mud crab growth and survival.

Water quality was closely monitored and maintained to ensure optimal conditions for the growth and survival of Scylla serrata in the indoor recirculating aquaculture system (RAS). Salinity levels were measured using a HANNA Salinity Tester, while pH was recorded with a Mettler Toledo pH meter. Dissolved oxygen (DO) and temperature were monitored using a HANNA Multiparameter HI 9829. Concentrations of nitrogenous compounds, specifically nitrite (NO_2^-) and ammonia (NH_3-N), were analyzed using a Spectrophotometer DR-3900. Measurements were taken daily, and any deviations from ideal ranges were addressed immediately through water exchange or system adjustment to maintain stable water quality parameters throughout the experimental period [14].

2.7 Data collection and analysis

All experimental data, including individual mud crab weights, survival rates, and daily water quality parameters, were systematically recorded and organized using Microsoft Excel [23]. Two key performance indicators were calculated:

Weight Gain (g) = Final Weight − Initial Weight (Eq. 2)

$$\text{Survival Rate \%} = \frac{\text{Number of Crabs Survived}}{\text{Initial Number of Crabs}} \times 100 \quad \text{(Eq. 3)}$$

Data were recorded and processed using Microsoft Excel. Statistical analysis was performed using one-way ANOVA to determine significant differences between treatments. A significance level of $p < 0.05$ was applied [26].

Statistical comparisons between the two treatments were performed using one-way analysis of variance (ANOVA) to evaluate differences in final weight, weight gain, survival rate, and mean water quality parameters. The purpose of the ANOVA was to determine whether the type of water source (artificial seawater vs. natural brackish water) had a statistically significant effect on growth performance and survival of Scylla serrata under indoor RAS conditions.

Prior to ANOVA, assumptions of normality and homogeneity of variance were tested using the Shapiro–Wilk test and Levene's test, respectively. Independence of observations was ensured through individual housing of crabs in separate compartments. When assumptions were satisfied, parametric ANOVA was applied. A significance level of $p < 0.05$ was used to determine statistical significance. All statistical analyses were conducted using Microsoft Excel.

For growth and survival analysis, each individual crab was considered an experimental unit ($n = 100$ per treatment), whereas water quality parameters were analyzed based on three replicated sampling measurements obtained during the monitoring period ($n = 3$).

2.8 Cost Comparison of Water Source Preparation

A cost comparison was conducted to evaluate the economic feasibility of artificial seawater and natural brackish water for indoor mud crab farming.

Artificial seawater was prepared using Red Sea Salt. The market price of Red Sea Salt was USD 35 per 7 kg (USD 5 per kg). Approximately 3 kg of salt were required to prepare 100 L of seawater at 30 ppt salinity, resulting in a preparation cost of approximately USD 15 per 100 L (USD 150 per 1,000 L), excluding freshwater and electricity costs.

Natural brackish water collected from Kampot cost USD 3 per 30 L, with an additional transportation cost of USD 3 per 30 L. Therefore, the total cost was USD 6 per 30 L, equivalent to approximately USD 20 per 100 L (USD 200 per 1,000 L).

The comparison indicates that artificial seawater was more cost-effective than transported natural brackish water under the conditions of this study. Additionally, artificial seawater eliminates transportation dependency, reduces contamination risks during transport, and provides consistent salinity and ionic composition. These advantages make artificial seawater a practical and economically viable option for inland indoor mud crab farming systems.

A simplified cost comparison is presented in **Table 2.**

Cost Component	Artificial Seawater	Natural Brackish Water
Cost per 100 L	USD 15	USD 20
Cost per 1,000 L	USD 150	USD 200
Transportation required	No	Yes
Contamination risk	Low	Moderate
Suitability for inland farming	High	Limited

3. RESULTS AND DISCUSSION

3.1 Laboratory Salinity Trial

The laboratory salinity trial revealed a strong survival response of mud crabs to salinity under low-temperature conditions. Only crabs maintained at 30 ppt survived the full 14-day experimental period. Survival outcomes from the laboratory salinity trial were quantified over the 14-day experimental period. Survival was 100% at 30 ppt (n = 1), while 0% survival was recorded at 15, 20, 25, and 40 ppt within the first 7–10 days of exposure. Due to complete mortality in all but one treatment, statistical comparison among salinity levels was not feasible.

These results must be interpreted with caution because the trial was conducted at a low ambient laboratory temperature of approximately 15 °C, which is substantially below the optimal thermal range for tropical mud crabs. Low temperature is known to suppress metabolic activity and impair osmoregulatory function, thereby increasing sensitivity to salinity stress. Under such suboptimal thermal conditions, salinity levels closest to the species natural range are more likely to support short-term survival.

Therefore, the observed survival at 30 ppt reflects a combined temperature salinity tolerance response rather than an isolated salinity effect. This preliminary trial was used solely to guide salinity selection for the subsequent RAS experiment, which was conducted under higher and more representative culture temperatures. Exposure to low temperature can suppress metabolic activity and impair osmoregulatory capacity, thereby increasing sensitivity to salinity stress.

Under such conditions, salinity levels closest to the species' optimal range are more likely to support short-term survival. The consistent survival observed at 30 ppt suggests that this salinity provided the greatest physiological stability for the experimental stock under combined temperature and salinity stress [24]. Similar interactions between temperature and salinity tolerance have been reported for mud crabs and other portunid species. Based on these findings, 30 ppt was selected as the artificial seawater salinity for the subsequent RAS experiment conducted under higher and more typical culture temperatures [25].

3.2 Initial Water Quality Characteristics of the Two Water Sources

The initial physicochemical characteristics of the two water sources before their introduction into the RAS are presented in Table 3 Natural brackish water showed slightly greater variability in dissolved nutrients, reflecting its origin from an open estuarine environment. In contrast, artificial seawater prepared using Red Sea Salt exhibited more controlled and uniform ionic composition because it was formulated from freshwater using a standardized salt mixture.

After entering the RAS, both water sources converged toward similar water-quality conditions due to continuous filtration, aeration, and biological nitrification processes. This demonstrates the buffering and stabilizing function of the RAS, which minimized natural variability and maintained parameters within suitable ranges for mud crab culture.

Table 3. Initial water quality of the two water sources before RAS operation.

Parameter	Artificial Seawater (Prepared)	Natural Brackish Water (Kampot)	Interpretation
Temperature (°C)	29.5 ± 0.5	30.2 ± 0.7	Similar ambient origin
Salinity (ppt)	30.0 (adjusted)	28–30	Natural fluctuation
pH	8.1 ± 0.1	7.7 ± 0.2	Slightly lower in natural water
Dissolved oxygen (mg/L)	6.2 ± 0.3	5.8 ± 0.4	Comparable
Ammonia (mg/L)	<0.05	0.08 ± 0.02	Higher background in natural water
Nitrite (mg/L)	<0.01	0.03 ± 0.01	Environmental origin

3.3 Water quality parameters

Water quality plays a critical role in the success of indoor Scylla serrata culture. Table 4 presents the mean values (± SD) of the key parameters recorded over the 60-day experimental period in both artificial seawater (Red Sea Salt) and natural brackish water treatments.

Statistical analysis using one-way ANOVA showed that temperature ($F = 0.12$, $p = 0.74$), dissolved oxygen ($F = 2.31$, $p = 0.19$), salinity ($F = 3.02$, $p = 0.14$), pH ($F = 1.67$, $p = 0.24$), and ammonia ($F = 0.96$, $p = 0.37$) did not differ significantly between artificial seawater and natural brackish water treatments ($p > 0.05$). Nitrite concentration, however, showed a statistically significant difference ($F = 5.21$, $p = 0.048$), although values remained within non-toxic levels for S. serrata culture.

No significant differences were observed in temperature, dissolved oxygen, salinity, pH, or ammonia concentrations between the two treatments, indicating that both artificial seawater and natural brackish water provided comparable environmental conditions suitable for mud crab growth. The measured salinity values, 30.83 ± 0.42 ppt in the Red Sea Salt treatment and 29.60 ± 0.55 ppt in the brackish water treatment, fall within the optimal salinity range reported for S. serrata, demonstrating effective salinity management in the recirculating aquaculture system (RAS). Nitrite concentrations differed significantly ($p < 0.05$) between treatments, with higher levels in the Red Sea Salt group. Although the nitrite levels remained below harmful thresholds, this variation could be attributed to differences in microbial populations or biofiltration efficiency between artificial and natural water sources [27]. Continuous monitoring and appropriate system management are essential to minimize nitrite accumulation. Overall, the stable water quality parameters observed in both treatments suggest that the use of artificial seawater (Red Sea Salt) is a viable alternative to natural brackish water for indoor mud crab farming under RAS conditions [12].

Table 4. Result of Water quality parameters (mean ± SD) recorded during the 60-day culture period of Scylla serrata under two water sources. Water quality measurements were obtained from three sampling events during the RAS monitoring schedule ($n = 3$). Statistical comparisons between treatments were performed using one-way ANOVA ($\alpha = 0.05$). F- and p-values indicate differences between artificial seawater and natural brackish water treatments.

Water Quality Parameter	Red Sea Salt (Mean ± SD)	Brackish Water (Mean ± SD)	F-value	p-value
Water Temperature (°C)	35.17±0.83	35.05 ± 0.78	0.12	0.74
Dissolved oxygen (ppm)	5.42±0.13	5.30 ± 0.17	2.31	0.19
Salinity (ppt)	30.83±0.42	29.60 ± 0.55	3.02	0.14
pH	8.01±0.22	7.89 ± 0.18	1.67	0.24
Ammonia (mg/L)	0.12 ± 0.05	0.15 ± 0.07	0.96	0.37
Nitrite (mg/l)	0.05±0.10	0.02 ± 0.07	5.21	0.048

Note: Significant difference at $p < 0.05$.

3.4 Growth performance and survival under RAS conditions

Growth data over the culture period are summarized in Table 5.

Table 5. Result of Growth performance and survival of Scylla serrata during the 60-day culture period under two treatments. Each crab was treated as an independent experimental unit ($n = 100$ per treatment). Values are expressed as mean ± SD and compared using one-way ANOVA ($\alpha = 0.05$).

Water Quality Parameter	Red Sea Salt	Brackish Water	F-value	p-value
Initial Weight (g)	107.5±4.5	107.2 ± 4.8	0.02	0.89
Final Weight (g)	201.0±18.19	181.5 ± 11.50	6.74	0.032
Weight Gain (g)	93.5±13.7	74.3 ± 11.2	7.11	0.028
Survival Rate %	45	40	1.56	0.25

Significant difference at $p < 0.05$.

Growth performance of *Scylla serrata* (Forsskål, 1775) during the 60-day indoor RAS experiment is presented in Table 3. Crabs reared in artificial seawater (Red Sea Salt) exhibited higher final body weight (201.0 ± 18.19 g) and greater weight gain (93.5 ± 13.7 g) compared with those cultured in natural brackish water (181.5 ± 11.50 g; 74.3 ± 11.2 g). These results indicate that artificial seawater provided a more favorable ionic environment for growth under controlled RAS conditions.

Despite improved growth performance, survival rates were relatively low in both treatments (45% in artificial seawater and 40% in brackish water). This mortality pattern is not attributable to salinity or water source, as all measured water quality parameters—salinity, dissolved oxygen, pH, ammonia, and nitrite—remained within acceptable ranges for S. serrata throughout the experiment (Table 2). Instead, the reduced survival is best explained by consistently elevated water temperatures, which frequently ranged between 35–36 °C during the RAS culture period.

Temperatures above the optimal thermal range for S. serrata (approximately 28–32 °C) are known to induce physiological stress, disrupt molting processes, and suppress immune function, leading to increased mortality. The similar survival rates observed in both treatments further confirm that temperature, rather than water source, was the dominant limiting factor affecting survival. Therefore, while artificial seawater enhanced growth performance, effective temperature regulation is essential to fully realize its benefits in indoor mud crab farming systems [28].

It is important to distinguish between the two experimental phases conducted in this study, which were characterized by markedly different thermal conditions. The preliminary laboratory salinity trial was performed at a low ambient temperature of approximately 15 °C, where reduced survival was associated with cold stress interacting with salinity tolerance. In contrast, the main RAS experiment was conducted under tropical indoor conditions, with water temperatures consistently maintained at 35–36 °C. Mortality observed during the RAS experiment was therefore associated with chronic high-temperature stress, not low temperature. These results demonstrate that S. serrata survival is sensitive to temperature extremes at both ends of the thermal spectrum, and that the causes of mortality differ fundamentally between the laboratory salinity trial and the RAS growth experiment.

3.5 Integrated Relationship Between Water Source, Water Quality, and Crab Performance

To clarify the linkage between water source, water-quality behavior, and biological responses, the results were evaluated as an interconnected system rather than as isolated variables. Although artificial seawater and natural brackish water differed in their original characteristics, these differences became less pronounced once the water entered the recirculating aquaculture system (RAS). Continuous mechanical filtration, biological nitrification, and aeration regulated key parameters and maintained them within similar ranges throughout the experiment.

This indicates that the RAS functioned as a stabilizing unit that minimized environmental variability, allowing both water sources to provide comparable physicochemical conditions. Despite this convergence, mud crabs reared in artificial seawater showed greater weight gain, suggesting that the consistency of ionic composition in formulated seawater reduced osmotic stress and supported more efficient metabolic performance.

In contrast, survival patterns were similar in both treatments because mortality was primarily associated with elevated temperature rather than water chemistry. Temperature remained outside the optimal range for Scylla serrata and acted as an overriding environmental constraint independent of water source.

These findings demonstrate that in indoor mud crab culture, water source influences growth indirectly through stability of water quality, whereas survival is more strongly governed by thermal conditions and system-level management.

3.6 Temperature and survival trends

Figure 6 shows the temporal relationship between water temperature and survival rate during the RAS culture period. Mean water temperature remained consistently high, ranging from 35.0 to 36.0 °C (mean ± SD), exceeding the optimal thermal range for S. serrata. Survival declined progressively over time in both treatments, with final survival rates of 45% in artificial seawater and 40% in brackish water.

Statistical analysis revealed a significant negative association between temperature and survival ($p < 0.05$), indicating that elevated temperature was the primary factor driving mortality during the RAS experiment. Other water quality parameters remained within acceptable ranges and did not show a significant relationship with survival, confirming that temperature stress, rather than water source or salinity, was the dominant limiting factor.

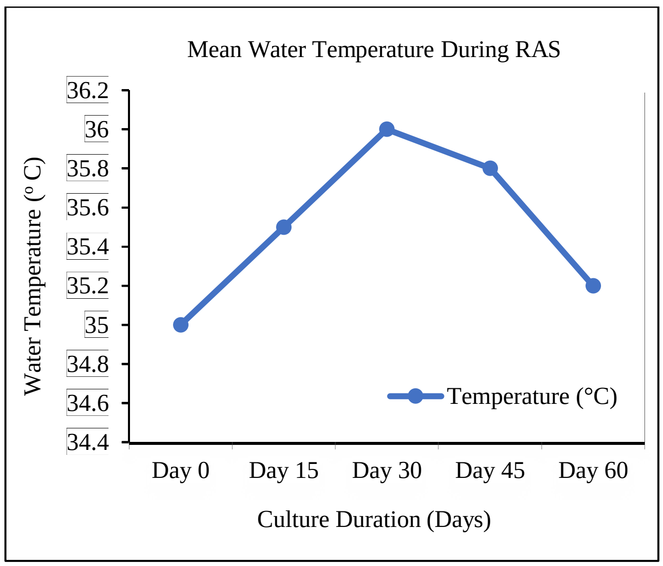

Fig. 6a. Mean water temperature (°C) recorded during the 60-day indoor RAS culture period.

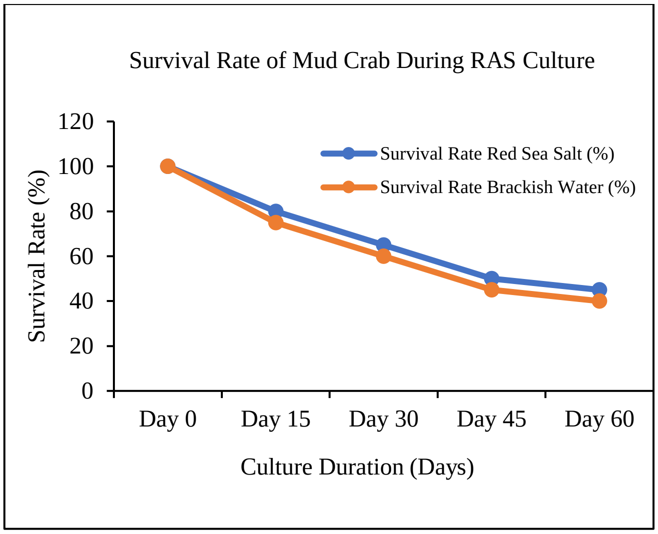

Fig. 6b. Survival rate (%) of *Scylla serrata* (Forsskål, 1775) cultured in artificial seawater and natural brackish water during the 60-day RAS experiment (n = 100 per treatment).

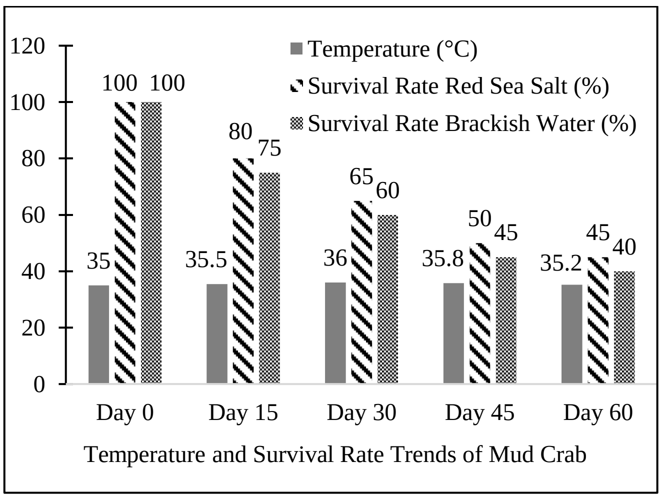

Fig. 6. Mean water temperature (°C) and survival rate (%) of Scylla serrata (Forsskål, 1775) during the 60-day indoor RAS experiment. Temperature values are presented as mean ± SD (n = 100 crabs per treatment). Survival rates represent cumulative survival percentages. A significant negative relationship was observed between water temperature and survival rate ($p < 0.05$).

Correlation analysis showed a negative relationship between temperature and survival, confirming that elevated temperature was the major limiting factor in this study. Other water quality parameters remained relatively stable and were comparable to natural estuarine conditions, indicating that temperature not salinity, pH, DO, ammonia, or nitrite was primarily responsible for reduced survival. This finding highlights the need to control thermal conditions in indoor RAS systems in tropical regions. These findings underline the need for temperature regulation mechanisms in indoor mud crab aquaculture systems in tropical regions. Strategies such as water chillers, improved ventilation, thermal insulation, or passive cooling via shading could help maintain water temperatures within the optimal range and improve survival outcomes [28].

Artificial seawater supported stable water quality and enhanced growth performance compared to natural brackish water. However, survival was below 50% in both treatments due to temperature stress. Therefore, the practical viability of artificial seawater depends on effective temperature management within the indoor RAS.

Future improvements in RAS design, such as automated temperature control, molting stage synchronization, and biofilter optimization, could significantly enhance survival and productivity. Additionally, long-term studies assessing reproductive performance and cost-efficiency under different water conditions will provide further insights into system scalability.

3.7 Relationship Between Water Quality Parameters, Survival, and Growth of Mud Crab

The analysis showed that temperature was the only parameter exceeding natural habitat ranges, consistently reaching 35–36 °C, which is higher than the optimal thermal tolerance of Scylla serrata (28–32 °C) [23]. This resulted in a strong negative association between temperature and survival, confirming that thermal stress was the primary factor causing mortality in both treatments.

In contrast, salinity (29.60–30.83 ppt), pH (7.89–8.01), dissolved oxygen (5.30–5.42 mg/L), ammonia (0.12–0.15 mg/L), and nitrite (0.02–0.05 mg/L) all remained within acceptable or near-optimal ranges commonly found in natural estuarine habitats where S. serrata thrives. These parameters showed weak or negligible relationships with survival, indicating that they were not limiting factors in this study.

Comparison with natural habitat values (Table 4) further supports this conclusion: temperature was the only parameter outside the natural range, while all other water quality indicators matched typical environmental conditions of mud crab ecosystems.

Table 6. Comparison of water quality parameters recorded in this study (mean ± SD or observed range) with reported natural habitat ranges of *Scylla serrata*. [11]

Parameter	This Study (Mean ± SD / Range)	Natural Habitat Range	Interpretation
Temperature (°C)	35.0–36.0	28–32	Above optimal → thermal stress
Salinity (ppt)	29.60–30.83	25–35	Within natural range
pH	7.89–8.01	7.5–8.5	Within natural range
Dissolved oxygen (mg/L)	5.30–5.42	5–7	Within natural range
Ammonia (mg/L)	0.12–0.15	<0.1–0.3	Acceptable
Nitrite (mg/L)	0.02–0.05	<0.5	Acceptable

Note: Values are presented as mean ± SD when appropriate; temperature is expressed as the observed range during the 60-day RAS culture period.

Comparison with reported natural habitat conditions (Table 4) indicates that water temperature was the only parameter that consistently exceeded the optimal range for Scylla serrata. Throughout the RAS experiment, temperatures remained between 35.0 and 36.0 °C, which is above the recommended thermal range of 28–32 °C for normal physiological functioning, molting success, and immune competence in mud crabs.

Elevated temperature is known to increase metabolic demand while simultaneously reducing oxygen solubility, potentially leading to energetic imbalance, impaired molting, and increased mortality. This is consistent with the observed decline in survival over time and the negative relationship between temperature and survival rate (Fig. 6). Statistically, temperature showed the strongest association with survival compared to other water quality parameters, which remained within acceptable or near-natural ranges and showed no clear relationship with mortality.

In contrast, salinity, pH, dissolved oxygen, ammonia, and nitrite levels closely matched values commonly reported for estuarine habitats of S. serrata and were maintained within biologically acceptable limits. These findings suggest that mortality in both treatments was primarily driven by chronic thermal stress rather than water source or chemical water quality. Therefore, effective temperature regulation is critical for improving survival in indoor RAS-based mud crab culture, even when other water quality parameters are well controlled.

4. CONCLUSIONS

This study evaluated the effects of two water sources, T1 (artificial seawater prepared using Red Sea Salt) and T2 (natural brackish water from Kampot), on water quality dynamics, growth performance, and survival of Scylla serrata cultured in an indoor recirculating aquaculture system (RAS). The results showed that both water sources maintained acceptable ranges for key water quality parameters, including salinity, pH, dissolved oxygen, ammonia, and nitrite, indicating that both systems are technically suitable for indoor mud crab culture. However, crabs reared in artificial seawater exhibited higher final body weight and greater weight gain compared with those cultured in natural brackish water. This suggests that formulated seawater may provide more stable and favorable ionic conditions that support improved growth performance under controlled indoor culture conditions.

Despite these positive growth outcomes, survival rates were relatively low, reaching 45% in the artificial seawater treatment and 40% in the brackish water treatment. The consistently high water temperature recorded during the RAS experiment (35–36 °C) exceeded the optimal thermal range for S. serrata (28–32 °C) and was likely a major factor contributing to increased mortality. In addition to thermal stress, other factors may also have influenced survival, including molting-related mortality, handling stress during stocking and monitoring, feeding management practices, and possible system design limitations associated with individual housing units. Therefore, although artificial seawater demonstrates strong potential as a practical alternative to natural brackish water for inland or indoor mud crab farming, improving temperature regulation and overall culture management will be essential to enhance survival and system productivity in tropical RAS environments.

Future research should focus on developing cost-effective temperature control strategies, such as improved ventilation, shading, or water-cooling systems, as well as optimizing feeding management and system design to reduce stress during molting periods. Additional long-term studies evaluating the economic feasibility and scalability of artificial seawater use will also be important for supporting sustainable indoor mud crab aquaculture development in Cambodia and other tropical regions.

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REFERENCES

[1] T. H. E. S. Of, *The State of World Fisheries and Aquaculture 2022*. 2022. doi: 10.4060/cc0461en.

[2] O. M. Joffre, S. Freed, J. Bernhardt, S. J. Teoh, S. Sambath, and B. Belton, “Assessing the Potential for Sustainable Aquaculture Development in Cambodia,” *Front. Sustain. Food Syst.*, vol. 5, no. October, 2021, doi: 10.3389/fsufs.2021.704320.

[3] T. H. E. S. Of, *World fisheries and aquaculture in review*, vol. 35, no. 3. 1999.

[4] Fao, *The future of mariculture: a regional approach for responsible development in the Asia-Pacific region*, no. March. 2006.

[5] P. T. Le, C. T. Ton, N. T. T. Thuy, S. H. K. Nguyen, and M. N. Binh, “Effects of different feeds on the growth performance and survival rates of juvenile mud crab Scylla paramamosain Estampador, 1950 (Crustacea: Portunidae),” *AACL Bioflux*, vol. 17, no. 6, pp. 3052–3062, 2024.

[6] J. C. BAYLON, “The combined effects of salinity and temperature on the survival and development of zoea, megalopa and crab instar larvae of mud crab, Scylla tranquebarica (Fabricius 1798),” *Asian Fish. Sci.*, vol. 26, no. 1, pp. 14–25, 2013, doi: 10.33997/j.afs.2013.26.1.002.

[7] S. G. Pati, B. Paital, F. Panda, S. Jena, and D. K. Sahoo, “Impacts of Habitat Quality on the Physiology, Ecology, and Economical Value of Mud Crab Scylla sp.: A Comprehensive Review,” *Water (Switzerland)*, vol. 15, no. 11, 2023, doi: 10.3390/w15112029.

[8] N. A. Kasan *et al.*, “A NOVEL STUDY ON THE EFFECT OF RAPID BIOFLOC AS PELLET FEED ON THE SURVIVAL RATE AND WATER QUALITY OF MUD CRAB, Scylla olivacea CULTURE,” *J. Sustain. Sci. Manag.*, vol. 17, no. 2, pp. 46–54, 2022, doi: 10.46754/jssm.2022.02.005.

[9] W. Cheng, I. S. Hsiao, and J. C. Chen, “Effect of ammonia on the immune response of Taiwan abalone Haliotis diversicolor supertexta and its susceptibility to Vibrio parahaemolyticus,” *Fish Shellfish Immunol.*, vol. 17, no. 3, pp. 193–202, 2004, doi: 10.1016/j.fsi.2004.03.004.

[10] E. T. Quinitio and F. D. Parado-Estepa, *BIOLOGY AND HATCHERY OF MUD CRABS Scylla spp.* 2008.

[11] C. E. Boyd, *General relationship between water quality and aquaculture performance in ponds*. Elsevier, 2017. doi: 10.1016/B978-0-12-804564-0.00006-5.

[12] Y. Tal, H. J. Schreier, K. R. Sowers, J. D. Stubblefield, A. R. Place, and Y. Zohar, “Environmentally sustainable land-based marine aquaculture,” *Aquaculture*, vol. 286, no. 1–2, pp. 28–35, 2009, doi: 10.1016/j.aquaculture.2008.08.043.

[13] J. Pitakphongmetha, W. Suntiamorntut, and S. Charoenpanyasak, “Internet of things for aquaculture in smart crab farming,” *J. Phys. Conf. Ser.*, vol. 1834, no. 1, 2021, doi: 10.1088/1742-6596/1834/1/012005.

[14] S. Pedapoli and K. R. Ramudu, “Effect of water quality parameters on growth and survivability of mud crab (Scylla tranquebarica) in grow out culture at Kakinada coast, Andhra Pradesh,” *~ 163 ~ Int. J. Fish. Aquat. Stud.*, vol. 2, no. 2, pp. 163–166, 2014, [Online]. Available: www.fisheriesjournal.com

[15] M. N. Syafaat, M. N. Azra, F. Mohamad, and C. Z. Che-ismail, “Thermal Tolerance and Physiological Changes in Mud Crab , Scylla paramamosain Crablet at Different Water Temperatures,” pp. 1–14, 2021.

[16] B. D. Paterson and D. L. Mann, *Mud Crab Aquaculture*. 2011. doi: 10.1002/9781444341775.ch4.

[17] I. M. Ruscoe, C. C. Shelley, and G. R. Williams, “The combined effects of temperature and salinity on growth and survival of juvenile mud crabs ˚ l) (Scylla serrata Forska,” vol. 238, pp. 239–247, 2004, doi: 10.1016/j.aquaculture.2004.05.030.

[18] M. N. Syafaat *et al.*, “A review of the nursery culture of mud crabs, genus scylla: Current progress and future directions,” *Animals*, vol. 11, no. 7, 2021, doi: 10.3390/ani11072034.

[19] D. Chavande, S. Bagde, S. Sinha, T. Seth, and H. Singh, “Sustainable Mud Crab Farming : Vertical Crab Culture Technology with Re-circulatory Aquaculture System”.

[20] K. Karal Marx, A. Rathipriya, J. K. Sundaray, and M. Muthu Abishag, “Water Quality Management in Aquaculture,” *Broodstock Manag. Fish Seed Prod.*, no. July, pp. 119–134, 2020, doi: 10.1201/9781003111689-13.

[21] W. Quality and E. Ronmental, “Chapter 2,” 1995.

[22] A. M. Win, H. T. K. Nguyen, E. T. Nguyen, H. T. T. Do, and T. Phuong, “Effects of combined temperature and salinity on growth and digestive enzymes of mud crab (Scylla paramamosain) from

larvae to juvenile," vol. 14, no. 5, pp. 2672–2687, 2021.

[23] A. Ferreira, L. Marquez, E. Almansa, J. P. Andrade, A. Lorenzo, and P. M. Domingues, "The use of alternative diets to culture juvenile cuttlefish, Sepia officinalis: effects on growth and lipid composition," *Aquac. Nutr.*, vol. 16, no. 3, pp. 262–275, 2010, doi: 10.1111/j.1365-2095.2009.00661.x.

[24] N. Romano and C. Zeng, "Osmoregulation in decapod crustaceans : implications to aquaculture productivity , methods for potential improvement and interactions with elevated ammonia exposure," *Aquaculture*, vol. 334–337, pp. 12–23, 2012, doi: 10.1016/j.aquaculture.2011.12.035.

[25] H. Manimaran, P. Chidambaram, C. Antony, and K. Ravaneswaran, "Effect of Salinity Regimes on Growth Performance , Digestive Enzyme Activity and Histological Structure of Scylla serrata," pp. 1–8, 2025, doi: 10.18805/IJAR.B-5670.Submitted.

[26] Eddiwan, T. Dahril, Adriman, Budijono, Efawani, and Y. Harjoyudanto, "Study of Growth and Survival of Mud Crab (Scylla serrata, Forskal) with Different Salinity Levels in culture media," *IOP Conf. Ser. Earth Environ. Sci.*, vol. 934, no. 1, 2021, doi: 10.1088/1755-1315/934/1/012015.

[27] R. Crab, Y. Avnimelech, T. Defoirdt, P. Bossier, and W. Verstraete, "Nitrogen removal techniques in aquaculture for a sustainable production," *Aquaculture*, vol. 270, no. 1–4, pp. 1–14, 2007, doi: 10.1016/j.aquaculture.2007.05.006.

[28] R. Nurdiani and C. Zeng, "Effects of temperature and salinity on the survival and development of mud crab, Scylla serrata (Forsskål), larvae," *Aquac. Res.*, vol. 38, no. 14, pp. 1529–1538, 2007, doi: 10.1111/j.1365-2109.2007.01810.x.