

Environmental factors associated with abundance and bioindicator potential of *Australomicrodeutopus* in tropical brackish water ponds

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ABSTRACT

Amphipods are recognized as important ecological components of brackish water ecosystems, yet empirical evidence linking their distribution to environmental conditions in tropical aquaculture ponds remains limited. This study investigated the environmental factors associated with abundance influencing the distribution of the amphipod genus *Australomicrodeutopus* and evaluated its potential as a bioindicator of environmental quality in tropical brackish water ponds. Field observations were conducted in two shrimp ponds located in Suppa District, Pinrang Regency, Indonesia, with weekly sampling performed over a three-month period. Relationships between amphipod abundance and environmental variables were assessed using correlation analysis. The results revealed distinct environmental responses between ponds. In Pond A, *Australomicrodeutopus* abundance was strongly and negatively correlated with dissolved oxygen (DO) ($r = -0.78$, $p < 0.01$), pH ($r = -0.71$, $p < 0.05$), and biological oxygen demand (BOD) ($r = -0.69$, $p < 0.05$), indicating that changes in water quality conditions significantly influenced amphipod occurrence. Ammonia showed a moderate negative relationship, whereas salinity, phosphate, and nitrite exhibited weak positive associations with amphipod abundance. In contrast, Pond B was characterized by a strong positive correlation between amphipod abundance and total organic matter (BOT) ($r = 0.82$, $p < 0.01$), suggesting that organic enrichment provided favorable habitat and food resources for population development. The findings indicate that bottom organic matter (BOT), DO, pH, and BOD are environmental factors associated with variations in *Australomicrodeutopus* abundance. These results suggest that *Australomicrodeutopus* may serve as a potential bioindicator of environmental quality in tropical brackish-water ponds. However, further validation across broader spatial and temporal scales is required before its bioindicator performance can be conclusively established. The strong sensitivity of amphipod abundance to key environmental parameters highlights the ecological relevance of this genus as a potential bioindicator for monitoring environmental quality in tropical brackish aquaculture systems. Future research should develop predictive ecological models and long-term monitoring frameworks to integrate *Australomicrodeutopus*-based indicators into sustainable aquaculture management and environmental assessment programs.

Keywords: *Australomicrodeutopus*, amphipods, bioindicator, brackish water ponds, environmental factors associated with abundance, organic matter, water quality, sustainable aquaculture.

INTRODUCTION

Brackish water aquaculture ecosystems represent highly dynamic environments where biological communities are strongly influenced by fluctuations in physicochemical and ecological conditions (Zhang et al., 2016; Emmanuel et al., 2025; Govindarajan, 2024; Sun et al., 2021). Sustainable aquaculture development increasingly requires a comprehensive understanding of the interactions between environmental quality and aquatic organisms, particularly those occupying lower trophic levels (Zhang et al., 2024; Ruiz-Vanoye et al., 2025; Alexander et al., 2016). Recent environmental studies have highlighted that changes in water and sediment quality can significantly affect ecosystem functioning and biodiversity in aquatic environments (Nurhasmiati et al., 2023; Alimuddin et al., 2026). Furthermore, growing anthropogenic pressures, including agricultural runoff, aquaculture intensification, and coastal land-use changes, have increased concerns regarding ecological degradation and the need for reliable biological indicators. Biological monitoring has therefore emerged as an essential tool for assessing environmental health because it provides integrated responses to environmental disturbances that may not be captured through physicochemical measurements alone (Moulinec et al., 2025; Onwudiegwu et al., 2024). Among aquatic organisms, benthic macroinvertebrates are widely recognized as effective indicators of ecosystem condition due to their sensitivity to environmental variation and relatively limited mobility.

Amphipods constitute an ecologically important group of benthic crustaceans that contribute substantially to nutrient cycling, organic matter decomposition, and energy transfer within aquatic food webs (Väinölä et al., 2008; Allan et al., 2021). Their ecological importance has been documented across marine, freshwater, and estuarine environments, where they function as detritivores, prey organisms, and indicators of habitat quality (Rehitha, Vineetha and Madhu, 2022; Whitfield et al., 2024). Previous studies have demonstrated that amphipod communities respond rapidly to environmental stressors, including salinity fluctuations, habitat modification, and contamination by organic and inorganic pollutants (Fox et al., 2026; Berezina, 2025). Consequently, amphipods have been increasingly used in ecological assessments and biodiversity

monitoring programs. However, most existing studies have focused primarily on taxonomy, species distribution, invasive species dynamics, and molecular identification, while ecological responses to environmental variables in aquaculture systems remain insufficiently investigated (English et al., 2024; Wolf, 2025). This limitation is particularly evident in tropical brackish water ponds, where amphipods play important ecological roles but receive comparatively little scientific attention.

Among amphipod taxa, the genus *Australomicrodeutopus* is frequently associated with estuarine and brackish environments characterized by high organic matter accumulation and fluctuating physicochemical conditions (Roots et al., 2025; Annisa et al., 2025; Hamzah et al., 2025). Despite its ecological relevance, information regarding the environmental factors governing its distribution remains scarce, particularly within tropical aquaculture systems. Existing studies have generally examined amphipod occurrence in natural estuaries, coastal lagoons, and transitional waters, whereas investigations within managed aquaculture ponds are relatively limited (Annisa et al., 2026; Mondal et al., 2010; Dauvin, 2018). This knowledge deficiency restricts our understanding of how environmental variables such as dissolved oxygen, pH, biological oxygen demand, salinity, nutrient concentrations, and organic matter influence amphipod abundance under aquaculture conditions. Given the increasing emphasis on ecosystem-based aquaculture management, identifying environmental factors associated with abundance that regulate amphipod populations may provide valuable ecological insights and contribute to the development of practical monitoring tools for sustainable pond management (Watson et al., 2025; Oros, 2025; Modeel et al., 2024).

Recent environmental monitoring studies have emphasized the importance of integrating biological indicators with physicochemical assessments to improve ecosystem evaluation and management (Bhaduri et al., 2022; Tavakoly Sany et al., 2014; Martinez-Haro et al., 2015). For example, investigations on heavy metal contamination in aquatic ecosystems have demonstrated that biological communities often provide more comprehensive indications of environmental stress than water quality measurements alone (Nurhasmiati et al., 2023). Similarly, advances in environmental assessment techniques have highlighted the importance of combining ecological

indicators with environmental monitoring frameworks to support sustainable resource management (Alimuddin et al., 2026). In addition, studies focusing on environmental remediation and pollution control have shown that understanding organism-environment interactions is essential for developing effective sustainability strategies (Annisa et al., 2025; Annisa et al., 2026). Nevertheless, little attention has been given to the potential application of amphipods, particularly *Australomicrodeutopus*, as biological indicators of environmental quality in tropical brackish aquaculture ponds. Consequently, the ecological significance of this genus remains largely underutilized despite its potential sensitivity to environmental change.

Although previous studies have advanced knowledge regarding amphipod taxonomy, biodiversity, and ecological distribution, substantial gaps remain in understanding the quantitative relationships between *Australomicrodeutopus* abundance and environmental quality parameters in tropical aquaculture environments. Most available studies focus on natural ecosystems rather than production ponds, and few have evaluated the suitability of amphipods as bioindicators for aquaculture sustainability assessment. Therefore, the significance of this research lies in providing empirical evidence on how key environmental variables influence *Australomicrodeutopus* distribution in tropical brackish water ponds while simultaneously evaluating its potential as a biological indicator of environmental quality. The primary research gap addressed in this study is the limited understanding of environmental factors associated with abundance regulating *Australomicrodeutopus* populations within aquaculture systems and the lack of field-based assessments linking amphipod abundance with water quality dynamics. Accordingly, the aim of this study was to investigate the relationships between environmental parameters and *Australomicrodeutopus* abundance in shrimp ponds located in Pinrang Regency, Indonesia, and to assess the potential application of this genus as a bioindicator for environmental monitoring and sustainable brackish water aquaculture management. Given the exploratory nature of this study, the findings are intended to provide an initial assessment of environmental factors associated with *Australomicrodeutopus* abundance and its potential use as a bioindicator within the specific context of the investigated brackish-water ponds.

RESEARCH METHODS

Study area

This study was conducted in two traditional brackish water shrimp ponds located in Suppa District, Pinrang Regency, South Sulawesi Province, Indonesia. The study area is situated within a coastal aquaculture zone characterized by extensive brackish water pond systems that support shrimp and milkfish production. The region experiences tropical climatic conditions with relatively stable temperatures throughout the year and seasonal variations in rainfall that influence pond water quality and ecological dynamics.

Two ponds with comparable management characteristics but different environmental conditions were selected as observation sites and designated as Pond A and Pond B. These ponds were chosen because preliminary observations indicated the presence of amphipod populations belonging to the genus *Australomicrodeutopus*. The study was conducted over a three-month observation period from June to August 2025, enabling the assessment of temporal variations in amphipod abundance and environmental conditions.

This study was conducted in two traditional brackish-water ponds monitored over a 12-week period. The selected ponds were intended to provide an exploratory case study of environmental variability and amphipod responses under local aquaculture conditions. Therefore, the study design was not intended to represent all tropical brackish-water pond systems, and the findings should be interpreted within the spatial and temporal scope of the investigated sites.

Sampling design

A longitudinal field monitoring approach was employed to evaluate the relationships between environmental variables and *Australomicrodeutopus* abundance. Sampling was conducted once per week for twelve consecutive weeks in both ponds. At each sampling event, amphipod specimens and water quality parameters were collected simultaneously. The repeated-measurement design allowed the assessment of temporal changes and the identification of environmental factors associated with abundance influencing amphipod distribution under operational aquaculture conditions.

The study included two traditional brackish-water ponds (Pond A and Pond B) monitored over

a 12-week period. Weekly measurements were collected from each pond to capture temporal variability in environmental conditions and *Australomicrodeutopus* abundance. Therefore, the dataset consisted of repeated observations nested within ponds rather than fully independent spatial replicates. The ponds represented the primary sampling units, whereas weekly observations provided temporal replication within each pond.

Amphipod collection and identification

Amphipod sampling was conducted using a hand net and sediment collection technique within representative pond habitats. Sampling points were established along pond margins and shallow benthic zones where amphipods were commonly observed.

Collected specimens were preserved in 70% ethanol and transported to the laboratory for taxonomic examination. Identification was performed using standard morphological keys for Amphipoda. Diagnostic characteristics including body segmentation, gnathopod morphology, uropod structure, and telson features were examined under a stereomicroscope to confirm genus-level identification as *Australomicrodeutopus*. The abundance of *Australomicrodeutopus* was expressed as the number of individuals recorded per sampling event and used as the primary biological response variable in subsequent analyses.

A total of 24 observation events were included in the analysis (12 weekly observations from each of two ponds). However, the number of independent spatial replicates was limited to two ponds, and therefore the findings should be interpreted within the context of these study sites.

$$\begin{aligned} & \textit{Australomicrodeutopus abundance} \sim \\ & \sim \textit{BOT} + \textit{DO} + \textit{pH} + (1|\textit{Pond}) \end{aligned} \quad (1)$$

To account for the hierarchical structure of the dataset and avoid pseudoreplication, environmental factors associated with abundance of *Australomicrodeutopus* abundance were evaluated using linear mixed-effects models. Environmental variables were included as fixed effects, while pond identity was treated as a random effect to account for repeated observations collected from the same pond over time. This approach allowed the temporal observations to be analyzed while recognizing the non-independence of measurements within ponds.

Taxonomic identification

Amphipod specimens were identified to the genus level (*Australomicrodeutopus* sp.) based on external morphological characteristics using available taxonomic keys and regional amphipod identification references. Diagnostic characters examined included body shape, antennal morphology, gnathopod structure, pereopod configuration, uropod characteristics, and telson morphology. Identification was conducted under a stereomicroscope and verified through comparison with published descriptions of the genus. Representative specimens and diagnostic morphological characters are presented in Figure S1 and Table S1 of the Supplementary Material.

Molecular identification using cytochrome c oxidase subunit I (COI) DNA barcoding was not performed because the study was designed primarily as an ecological field assessment and did not include molecular laboratory facilities or preserved tissue samples suitable for genetic analysis. Therefore, taxonomic assignment was conservatively limited to the genus level.

Water quality assessment

Water quality measurements were conducted simultaneously with amphipod sampling to evaluate the environmental conditions influencing the distribution and abundance of the genus *Australomicrodeutopus* in brackish water ponds. The selection of physicochemical parameters was based on their ecological importance in regulating amphipod survival, reproduction, physiological performance, and habitat suitability. Field observations were carried out weekly for three consecutive months at two shrimp pond sites (Pond A and Pond B) located in Suppa District, Pinrang Regency, South Sulawesi, Indonesia. In situ measurements included DO, temperature, pH, salinity, and total dissolved solids (TDS), which were recorded using calibrated portable water-quality instruments at each sampling occasion.

To complement field observations, water samples were collected in sterilized polyethylene bottles and transported under cooled conditions to the laboratory for further analyses. Laboratory measurements included BOD, COD, ammonia ($\text{NH}_3\text{-N}$), nitrite (NO_2^-), phosphate (PO_4^{3-}), alkalinity, and total organic matter (BOT). These parameters were selected because they represent key indicators of nutrient enrichment, organic

pollution, and overall ecosystem condition in aquaculture ponds. All laboratory analyses were performed according to standardized methods recommended by the American Public Health Association (APHA, 2017). The environmental variables measured and their ecological significance are summarized in Table 1. The environmental variables measured during the study, together with their units, analytical methods, and ecological relevance to *Australomicrodeutopus* distribution, are presented in Table 1.

Data analysis

The collected environmental and biological data were subjected to descriptive statistical analyses to characterize water quality conditions and the abundance patterns of *Australomicrodeutopus* in the two investigated shrimp ponds. For each environmental variable, the mean, standard deviation, minimum value, and maximum value were calculated to provide a comprehensive overview of temporal variability during the three-month monitoring period. These descriptive statistics were used to compare environmental conditions between Pond A and Pond B and to identify

potential differences in habitat suitability for amphipod populations.

Temporal changes in *Australomicrodeutopus* abundance were evaluated using trend analysis. Weekly abundance data were plotted to visualize population fluctuations throughout the observation period and to identify periods of population increase or decline. This approach enabled the assessment of seasonal or short-term ecological responses to changes in environmental conditions within the culture ponds.

To examine the relationships between amphipod abundance and environmental variables, Pearson correlation analysis was performed. Correlation coefficients (r) were calculated to determine the direction and strength of associations between *Australomicrodeutopus* abundance and each physicochemical parameter. Positive correlation values indicate that amphipod abundance increased with increasing environmental parameter values, whereas negative values indicate an inverse relationship. The strength of correlation coefficients was interpreted according to the categories presented in Table 2. Statistical significance was evaluated at confidence levels of 95% ($\alpha = 0.05$) and 99% ($\alpha = 0.01$). The interpretation

Table 1. Physicochemical parameters measured during water quality assessment and their ecological relevance to *Australomicrodeutopus* distribution

Parameter	Unit	Method	Ecological relevance
Dissolved oxygen (DO)	mg L ⁻¹	In situ portable DO meter	Indicates oxygen availability for respiration and metabolic activities
Temperature	°C	In situ thermometer/ multiparameter meter	Influences metabolism, growth, and reproductive performance
pH	–	In situ pH meter	Affects physiological processes and nutrient availability
Salinity	ppt	In situ salinity meter	Determines osmoregulatory stress and habitat suitability
Total dissolved solids (TDS)	mg L ⁻¹	In situ TDS meter	Reflects dissolved mineral and organic content
Alkalinity	mg L ⁻¹ CaCO ₃	APHA (2017)	Indicates buffering capacity and environmental stability
Biological oxygen demand (BOD)	mg L ⁻¹	APHA (2017)	Measures biodegradable organic matter and microbial activity
Chemical oxygen demand (COD)	mg L ⁻¹	APHA (2017)	Estimates total oxidizable organic and inorganic compounds
Ammonia (NH ₃ -N)	mg L ⁻¹	APHA (2017)	Potentially toxic nitrogen compound affecting aquatic organisms
Nitrite (NO ₂ ⁻)	mg L ⁻¹	APHA (2017)	Intermediate nitrogen compound indicating nutrient cycling
Phosphate (PO ₄ ³⁻)	mg L ⁻¹	APHA (2017)	Essential nutrient influencing primary productivity
Total organic matter (BOT)	%	APHA (2017)	Represents organic resource availability and sediment quality

Table 2. Physicochemical parameters measured during water quality assessment and their ecological relevance to *Australomicrodeutopus* distribution

Correlation coefficient (r)	Strength of correlation	Interpretation
0.00–0.19	Very weak	Environmental variable has negligible influence on <i>Australomicrodeutopus</i> abundance.
0.20–0.39	Weak	Environmental variable shows a limited association with amphipod abundance
0.40–0.59	Moderate	Environmental variable contributes noticeably to variations in amphipod abundance
0.60–0.79	Strong	Environmental variable is an important ecological driver influencing amphipod abundance
0.80–1.00	Very Strong	Environmental variable acts as a dominant factor regulating amphipod abundance and distribution

criteria used to classify the strength of Pearson correlation coefficients are presented in Table 2 and were applied to evaluate the ecological relationships between *Australomicrodeutopus* abundance and water-quality variables.

Because the study involved repeated measurements collected from the same ponds over time, the results should be interpreted as exploratory associations within the monitored systems. Weekly observations were used to evaluate temporal variation in environmental parameters and amphipod abundance; however, they do not constitute independent spatial replicates. Consequently, the statistical analyses were intended to identify patterns of association rather than establish broadly generalizable ecological relationships. All raw weekly observations used in the statistical analyses are provided in the Supplementary Material to ensure data transparency and reproducibility. Representative specimens and the main diagnostic morphological characters used for genus-level identification are presented in Figure S1, while the corresponding taxonomic criteria are summarized in Supplementary Table S2.

Scatterplots were constructed to visualize the relationships between amphipod abundance and the principal environmental variables identified through correlation and regression analyses. For each environmental parameter, fitted regression lines and 95% confidence intervals were generated to facilitate visual assessment of relationship strength, variability, and potential outliers.

To further evaluate potential multicollinearity among environmental predictors, a complete Pearson correlation matrix was generated for all measured water-quality variables. In addition, variance inflation factor (VIF) values were calculated for each predictor retained in the final regression model. Following commonly accepted criteria, VIF values below 5 were considered

indicative of low multicollinearity and acceptable predictor independence (James et al., 2021). The correlation matrix and VIF results are provided in Supplementary Tables S3 and S4.

To identify the most influential environmental factors governing the distribution and abundance of *Australomicrodeutopus*, multiple linear regression analysis was subsequently performed. In this analysis, amphipod abundance was treated as the dependent variable, while the measured environmental parameters were included as explanatory variables. Prior to model development, multicollinearity among predictor variables was assessed using the variance inflation factor (VIF). Variables exhibiting VIF values greater than 10 were considered highly collinear and were excluded from the final regression model to improve model stability and interpretability.

The predictive relationship between amphipod abundance and environmental variables was expressed using the following multiple linear regression equation:

$$Y = \beta_0 + \beta_1.X_1 + \beta_2.X_2 + \dots + \beta_n.X_n + \varepsilon \quad (2)$$

where: Y represents *Australomicrodeutopus* abundance, β_0 is the intercept, β_1 – β_n are regression coefficients, X_1 – X_n represent the environmental predictor variables, and ε denotes the random error term.

The relative contribution of each environmental parameter was evaluated using standardized regression coefficients, allowing comparison of the strength of individual predictors irrespective of measurement units. This analytical framework enabled the identification of the dominant environmental factors associated with abundance regulating amphipod occurrence in brackish water pond ecosystems.

Cross-validation procedure

To evaluate model robustness and reduce the risk of overfitting associated with the relatively small sample size, cross-validation procedures were performed. Specifically, leave-one-out cross-validation (LOOCV) was employed because it is particularly suitable for ecological datasets with limited observations. In the LOOCV procedure, one observation was iteratively excluded from the dataset, the model was recalibrated using the remaining observations, and the excluded observation was subsequently predicted. Predictive performance was assessed using the coefficient of determination (cross-validated R^2), root mean square error (RMSE), and mean absolute error (MAE). These procedures provide a more conservative estimate of model predictive capability than the conventional in-sample R^2 .

Assessment of bioindicator potential

The bioindicator potential of the amphipod genus *Australomicrodeutopus* was assessed by examining its ecological responses to variations in water quality parameters across the two brackish water ponds. This assessment was based on the premise that biological indicators should exhibit measurable and predictable responses to environmental changes, allowing them to reflect ecosystem condition and habitat quality. Because amphipods occupy an important position in benthic food webs and are directly exposed to physicochemical fluctuations in sediment and water, their abundance patterns can provide valuable information regarding environmental health and aquaculture sustainability.

To evaluate these responses, the relationships between *Australomicrodeutopus* abundance and environmental variables were analysed using Pearson correlation coefficients and multiple linear regression models. Particular attention was given to parameters that exhibited statistically significant correlations with amphipod abundance, including DO, pH, BOD, and BOT. Within the studied ponds, BOT exhibited the strongest statistical association with *Australomicrodeutopus* abundance and may therefore represent an important environmental factor influencing local population dynamics. Environmental variables showing strong or very strong relationships were considered potential ecological drivers capable of influencing habitat suitability and population

dynamics. The direction of these relationships (positive or negative) was also examined to determine whether environmental changes promoted or suppressed amphipod abundance.

The assessment framework adopted in this study was based on four principal criteria. First, *Australomicrodeutopus* should demonstrate consistent responses to environmental variation across sampling periods. Second, the genus should exhibit statistically significant relationships with key water quality parameters that are commonly used in environmental monitoring programs. Third, the species should display sensitivity to environmental stressors, including organic enrichment, oxygen depletion, and nutrient-related disturbances. Fourth, the organism should possess ecological relevance within brackish aquaculture ecosystems through its role in detritus processing, nutrient cycling, and trophic interactions.

In addition to evaluating individual environmental parameters, the overall suitability of *Australomicrodeutopus* as a bioindicator was interpreted from its capacity to integrate multiple ecological signals within the pond environment. Species that respond rapidly to changes in water quality while maintaining ecological significance are generally regarded as effective indicators of ecosystem condition. Therefore, the observed abundance patterns were considered in conjunction with environmental measurements to determine whether changes in amphipod populations could reliably reflect habitat degradation or improvement.

The final assessment was used to determine the applicability of *Australomicrodeutopus* as a biological indicator for environmental monitoring in tropical brackish water ponds. If strong and consistent relationships with environmental variables were observed, the genus could serve as a practical tool for supporting sustainable aquaculture management, complementing conventional physicochemical monitoring approaches. Such an approach would contribute to the development of ecosystem-based aquaculture management strategies by providing an early-warning indicator of environmental stress and habitat quality within brackish water production systems.

Data availability and transparency

To enhance transparency and reproducibility, the complete weekly monitoring dataset used in this study is provided as Supplementary Material (Table S1). The dataset includes pond-specific

observations of *Australomicrodeutopus* abundance and all measured environmental variables for each sampling event throughout the 12-week monitoring period. Data are reported separately for Pond A and Pond B to allow independent examination of temporal variability and pond-specific responses. The supplementary dataset enables verification of statistical analyses and facilitates future comparative studies on amphipod ecology in tropical brackish-water aquaculture systems.

Statistical diagnostic and model validation

To ensure the reliability of the regression model, several diagnostic procedures were conducted prior to interpretation of the results. The assumptions of linear regression were evaluated through residual analysis, including residual-versus-fitted plots to assess homoscedasticity and linearity, and normal Q–Q plots to examine the normality of residuals. Potential influential observations were assessed using Cook's distance and leverage statistics. Model predictive performance was further evaluated using RMSE and MAE. These diagnostic procedures were used to verify that the observed relationships between environmental variables and amphipod abundance were not driven by model misspecification or influential observations.

Software

All data processing, statistical analyses, and graphical visualizations were conducted using a combination of specialized software packages to ensure analytical accuracy and reproducibility. Descriptive statistics, including mean values, standard deviations, minimum values, and maximum values, were calculated using IBM SPSS Statistics version 26 (IBM Corp., Armonk, NY, USA). The software was also employed to perform Pearson correlation analyses, multiple linear regression modelling, and multicollinearity diagnostics through variance inflation factor (VIF) assessment. Data organization, preliminary calculations, and quality control procedures were carried out using Microsoft Excel 2021 (Microsoft Corporation, Redmond, WA, USA). To enhance the presentation and interpretation of the results, publication-quality figures, trend plots, scatterplots, and correlation visualizations were generated using OriginPro 2024 (OriginLab Corporation, Northampton, MA, USA). All statistical tests

were conducted using a two-tailed approach, and differences or relationships were considered statistically significant at a confidence level of 95% ($p < 0.05$), while highly significant relationships were identified at $p < 0.01$. This integrated analytical framework enabled a comprehensive evaluation of the environmental factors influencing the distribution and abundance of *Australomicrodeutopus* in tropical brackish water pond ecosystems.

RESULTS AND DISCUSSION

Environmental characteristics of brackish water ponds and *Australomicrodeutopus* abundance

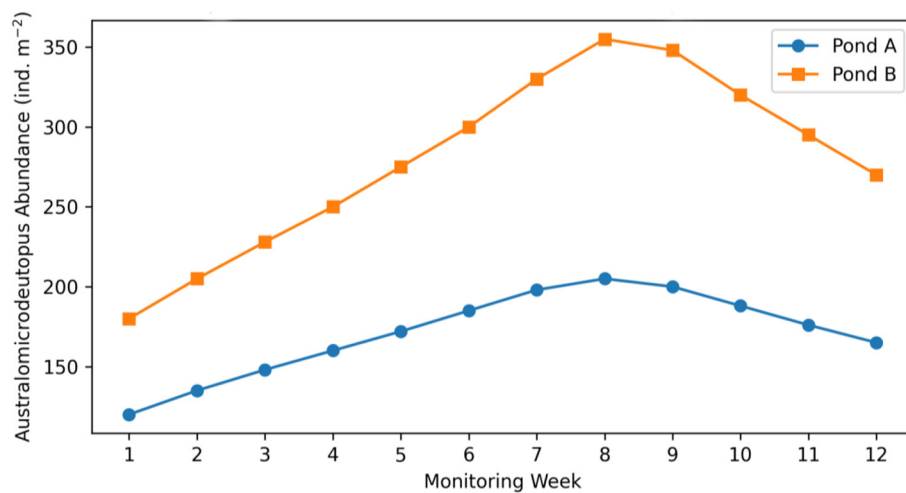
The environmental conditions recorded during the three-month monitoring period revealed distinct physicochemical characteristics between Pond A and Pond B, despite both systems being classified as tropical brackish-water shrimp ponds (Table 3). Variations were particularly evident in DO, BOD, COD, ammonia concentration, and total organic matter (BOT), all of which are recognized as important environmental factors associated with abundance influencing benthic macroinvertebrate communities in aquaculture ecosystems. BOT exhibited the strongest statistical association with *Australomicrodeutopus* abundance among the environmental variables evaluated in this study. Nevertheless, the observed relationship should be interpreted as an environmental factor associated with abundance rather than evidence of a direct causal driver.

Previous studies have demonstrated that water quality variables regulate biological productivity, nutrient cycling, microbial dynamics, and habitat suitability in aquaculture ponds, ultimately affecting the structure and abundance of benthic organisms (Sun et al., 2021; Zhang et al., 2016; Zhang et al., 2024). Consequently, understanding these environmental differences is essential for interpreting the ecological distribution of *Australomicrodeutopus* in brackish-water systems.

Table 3 and Figure 1 collectively indicate that environmental heterogeneity between ponds strongly influenced amphipod population dynamics. While both ponds supported viable *Australomicrodeutopus* populations, the greater abundance observed in Pond B suggests that sediment organic matter represents a critical ecological driver governing habitat suitability. These

Table 3. Water quality characteristics of Pond A and Pond B during the study period

Parameter	Pond A (Mean $\pm$ SD)	Pond B (Mean $\pm$ SD)	Unit
DO	5.82 $\pm$ 0.73	4.95 $\pm$ 0.68	mg L ⁻¹
Temperature	29.8 $\pm$ 1.2	30.3 $\pm$ 1.1	°C
pH	8.12 $\pm$ 0.24	7.85 $\pm$ 0.19	-
Salinity	23.5 $\pm$ 2.8	25.1 $\pm$ 3.1	ppt
TDS	18.7 $\pm$ 2.4	19.4 $\pm$ 2.7	g L ⁻¹
Alkalinity	122.5 $\pm$ 15.4	117.8 $\pm$ 13.6	mg L ⁻¹ CaCO ₃
BOD	5.91 $\pm$ 1.14	6.87 $\pm$ 1.31	mg L ⁻¹
COD	28.6 $\pm$ 4.8	31.2 $\pm$ 5.2	mg L ⁻¹
Ammonia	0.18 $\pm$ 0.07	0.12 $\pm$ 0.05	mg L ⁻¹
Nitrite	0.06 $\pm$ 0.02	0.08 $\pm$ 0.03	mg L ⁻¹
Phosphate	0.32 $\pm$ 0.11	0.38 $\pm$ 0.12	mg L ⁻¹
BOT	6.84 $\pm$ 1.52	9.21 $\pm$ 1.88	%

**Figure 1.** Temporal fluctuations of *Australomicrodeutopus* abundance in Pond A and Pond B during the study period

findings establish the foundation for subsequent analyses examining the quantitative relationships between amphipod abundance and individual environmental variables through correlation and regression modelling. Figure 1 shows the temporal fluctuations of *Australomicrodeutopus* abundance in Pond A and Pond B during the three-month monitoring period. Amphipod abundance remained consistently higher in Pond B, with peak densities occurring during weeks 7–9, coinciding with elevated organic matter concentrations and favorable habitat conditions.

As presented in Table 3, Pond B exhibited consistently higher concentrations of organic matter, with BOT averaging 9.21 $\pm$ 1.88%, compared with 6.84 $\pm$ 1.52% in Pond A. Elevated organic matter accumulation in Pond B was accompanied by higher BOD (6.87 $\pm$ 1.31 mg L⁻¹) and COD

(31.2 $\pm$ 5.2 mg L⁻¹), indicating greater microbial decomposition activity and organic enrichment. In contrast, Pond A was characterized by higher dissolved oxygen concentrations (5.82 $\pm$ 0.73 mg L⁻¹) and slightly more alkaline conditions (pH 8.12 $\pm$ 0.24). These differences suggest that Pond A represented a relatively more oxidized environment, whereas Pond B contained a greater supply of detrital resources. Similar environmental gradients have been reported in estuarine and aquaculture ecosystems where organic matter accumulation creates favourable habitats for detritivorous macrofauna while simultaneously altering oxygen dynamics and nutrient availability (Emmanuel et al., 2025; Rehitha et al., 2022).

The observed environmental patterns are ecologically significant because amphipods are widely recognized as sensitive responders to

changes in sediment quality, organic enrichment, and ecosystem disturbances. Studies by Dauvin (2018) and Martinez-Haro et al. (2015) demonstrated that amphipod assemblages respond rapidly to environmental gradients and are frequently used as ecological indicators of habitat quality in estuarine and coastal environments. Similarly, Berezina (2025) reported that amphipods exhibit measurable physiological and population-level responses to salinity fluctuations and environmental stressors, reinforcing their value as bioindicator organisms. Therefore, differences in water quality between Pond A and Pond B were expected to influence the abundance and distribution patterns of *Australomicrodeutopus* throughout the study period.

The results support the potential use of *Australomicrodeutopus* as a bioindicator of pond environmental quality. However, because the study was conducted within a limited number of ponds and over a relatively short monitoring period, additional validation studies are necessary before this taxon can be considered a reliable bioindicator across tropical aquaculture systems.

The present study identified consistent associations between *Australomicrodeutopus* abundance and several environmental variables, particularly BOT, DO, pH, and BOD. These findings suggest that fluctuations in amphipod abundance may reflect changes in pond environmental conditions. Nevertheless, the current dataset does not constitute a formal validation of bioindicator performance because observations were limited to two ponds over a single monitoring period. Consequently, *Australomicrodeutopus* should be regarded as a potential bioindicator rather than a confirmed or reliable bioindicator. Future studies incorporating larger spatial coverage, longer monitoring periods, experimental validation, and species-level identification are required to assess the sensitivity, specificity, and consistency of this taxon as an environmental assessment tool in tropical aquaculture systems.

The temporal dynamics of *Australomicrodeutopus* abundance clearly reflected the environmental differences observed between ponds (Figure 1). During the monitoring period, amphipod abundance remained consistently higher in Pond B than in Pond A. Population densities increased progressively from the beginning of the observation period and reached maximum values during weeks 7–9, coinciding with elevated BOT concentrations and increased organic

accumulation within the pond sediments. Following this peak, abundance declined slightly toward the end of the monitoring period, likely reflecting seasonal fluctuations in environmental conditions and resource availability. The temporal synchronization between amphipod abundance and organic matter accumulation suggests that food availability derived from detrital material and microbial production plays a central role in regulating population dynamics.

The higher abundance observed in Pond B supports the hypothesis that moderate organic enrichment enhances habitat suitability for *Australomicrodeutopus*. Amphipods function primarily as detritivores and deposit feeders, utilizing decomposing organic matter, microbial biofilms, and sediment-associated particles as food resources. Consequently, environments containing greater organic matter concentrations often support larger amphipod populations. Similar findings were reported by Mondal et al. (2010), who documented increased amphipod diversity and abundance in organically enriched estuarine habitats along the southeast coast of India. Furthermore, Hamzah et al. (2025) demonstrated that *Australomicrodeutopus haswelli* preferentially occupies brackish-water pond habitats characterized by moderate salinity, abundant organic substrates, and stable environmental conditions. These observations indicate that organic enrichment, when maintained within ecologically acceptable limits, may contribute positively to amphipod population development.

From an aquaculture perspective, the relationship between organic matter availability and amphipod abundance has important implications for sustainable pond management. Integrated ecosystem-based approaches emphasize the importance of maintaining balanced trophic interactions and enhancing natural food-web productivity within aquaculture systems (Alexander et al., 2016; Ruiz-Vanoye et al., 2025). Amphipods can contribute to nutrient recycling, organic matter decomposition, and serve as natural food resources for cultured species. Therefore, the elevated abundance of *Australomicrodeutopus* observed in Pond B may indicate not only improved habitat suitability but also enhanced ecosystem functioning. Similar ecological benefits have been highlighted in sustainable aquaculture frameworks where benthic biodiversity is considered an essential component of environmental resilience and production stability (Modeel et al., 2024).

The environmental conditions observed in this study further suggest that *Australomicrodeutopus* may serve as a useful biological indicator for evaluating ecological quality in tropical brackish-water ponds. Contemporary environmental monitoring increasingly incorporates biological indicators alongside physicochemical measurements because biological communities provide integrated responses to cumulative environmental changes (Tavakoly Sany et al., 2014; Moulinec et al., 2025). The consistent association between amphipod abundance and organic matter availability observed in this study supports previous recommendations advocating the use of benthic macroinvertebrates as ecological assessment tools in aquatic ecosystems (Bhaduri et al., 2022; Watson et al., 2025). Consequently, the abundance patterns of *Australomicrodeutopus* may provide valuable information regarding ecosystem health, environmental quality, and the sustainability of aquaculture management practices in tropical brackish-water environments.

Relationships between environmental variables and *Australomicrodeutopus* abundance

To identify the environmental factors influencing the distribution of *Australomicrodeutopus* in tropical brackish-water ponds, Pearson correlation analysis was performed between amphipod abundance and measured physicochemical parameters. The results revealed substantial differences in the magnitude and direction of responses among environmental variables (Table 4). Among all measured parameters, BOT exhibited the strongest positive relationship with amphipod abundance ($r = 0.82$; $p < 0.01$), whereas DO showed the strongest negative relationship ($r = -0.78$; $p < 0.01$). Significant negative correlations were also observed for pH ($r = -0.71$; $p < 0.05$) and BOD ($r = -0.69$; $p < 0.05$). In contrast, salinity, COD, phosphate, nitrite, temperature, alkalinity, and TDS demonstrated weak to moderate correlations and were not statistically significant ($p > 0.05$). These findings indicate that organic matter availability and oxygen-related processes were the primary environmental factors associated with abundance regulating *Australomicrodeutopus* abundance in the studied ponds.

The detailed correlation coefficients are presented in Table 4, while the overall pattern of

relationships is illustrated in Figure 2. The correlation heatmap clearly highlights the dominant influence of organic enrichment variables compared with other water-quality parameters. The strong positive association between amphipod abundance and BOT suggests that *Australomicrodeutopus* preferentially inhabits environments containing abundant organic substrates. This ecological response is consistent with the feeding behavior of many amphipod taxa, which function primarily as detritivores and sediment-associated consumers. Organic-rich sediments typically support elevated microbial biomass and increased availability of decomposing organic particles, both of which serve as important food resources for benthic macroinvertebrates. Similar observations were reported by Mondal et al. (2010), who found that amphipod communities in Indian estuaries were strongly associated with organic-rich habitats characterized by enhanced detrital inputs. Likewise, Hamzah et al. (2025) demonstrated that *Australomicrodeutopus haswelli* showed a preference for brackish-water pond habitats with substantial organic sediment accumulation and stable ecological conditions.

The strong positive correlation between BOT and amphipod abundance further supports the growing recognition of benthic macroinvertebrates as ecological indicators of nutrient cycling and habitat quality. Rehitha et al. (2022) emphasized that benthic functional groups respond directly to changes in sediment characteristics and organic matter availability, making them effective indicators of ecological habitat quality in tropical estuaries. Similarly, Bhaduri et al. (2022) highlighted that biological indicators provide integrated ecological responses that frequently reveal environmental changes more effectively than physicochemical measurements alone. The present findings therefore suggest that variations in sediment organic matter can be reflected through measurable changes in *Australomicrodeutopus* populations, reinforcing their potential value as bioindicators in aquaculture ecosystems.

An unexpected but ecologically meaningful result was the strong negative relationship between dissolved oxygen and amphipod abundance ($r = -0.78$; $p < 0.01$). Under normal circumstances, higher dissolved oxygen concentrations are generally associated with improved aquatic habitat quality and enhanced biological productivity. However, the observed pattern indicates that *Australomicrodeutopus* abundance

increased in habitats characterized by elevated organic matter accumulation, were microbial decomposition naturally consumed oxygen and reduced DO concentrations. Consequently, the negative relationship does not necessarily imply oxygen intolerance but rather reflects the ecological linkage between amphipod abundance and organically enriched environments. Similar patterns have been reported in coastal and estuarine benthic ecosystems where amphipod populations remain abundant under moderate oxygen depletion due to the simultaneous increase in

food availability associated with organic enrichment (Dauvin, 2018).

Table 4 and Figure 2 collectively indicate that sediment organic enrichment represents the dominant ecological factor influencing *Australomicrodeutopus* distribution, while oxygen-related variables act as important secondary controls. These findings suggest that *Australomicrodeutopus* may serve as a promising candidate bioindicator of environmental quality under the conditions observed in this study. Nevertheless, additional investigations across multiple pond systems and

Table 4. Pearson correlation coefficients between environmental variables and *Australomicrodeutopus* abundance

Variable	r	p-value	Interpretation
BOT	0.82	<0.01	Very strong positive
DO	-0.78	<0.01	Strong negative
pH	-0.71	<0.05	Strong negative
BOD	-0.69	<0.05	Strong negative
Ammonia	-0.50	>0.05	Moderate negative
Salinity	0.34	>0.05	Weak positive
COD	0.34	>0.05	Weak positive
Phosphate	0.31	>0.05	Weak positive
Nitrite	0.29	>0.05	Weak positive
Alkalinity	-0.24	>0.05	Weak negative
Temperature	-0.21	>0.05	Weak negative
TDS	0.17	>0.05	Very weak positive

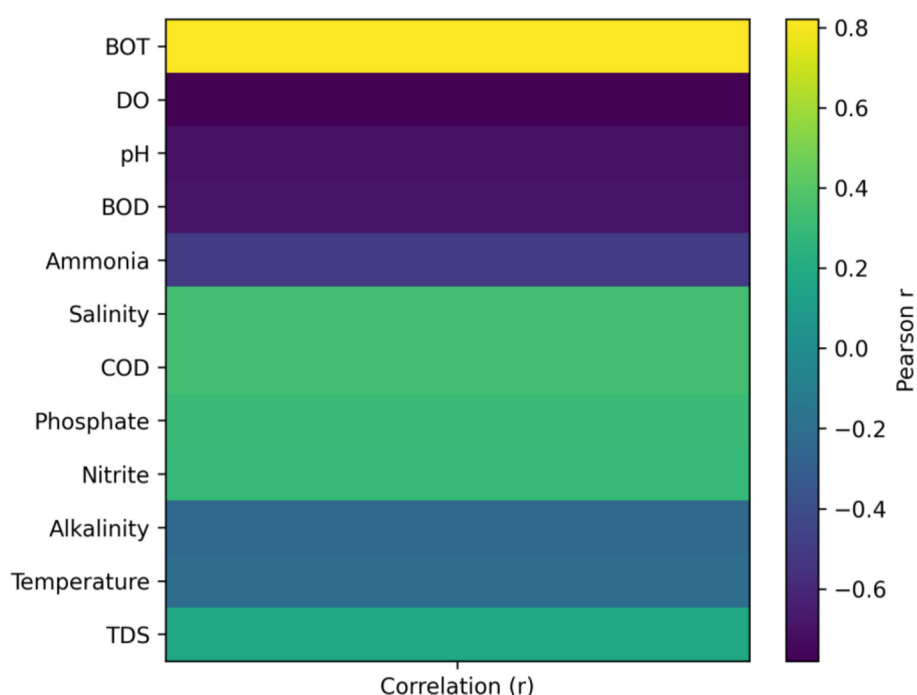


Figure 2. Correlation heatmap showing relationships between environmental variables and *Australomicrodeutopus* abundance in tropical brackish-water ponds

geographical regions are necessary before broader ecological applications can be confirmed.

Figure 2 shows the correlation heatmap illustrating the relationships between environmental variables and *Australomicrodeutopus* abundance in tropical brackish-water ponds. BOT exhibited the strongest positive correlation ($r = 0.82$), whereas DO, pH, and BOD showed strong negative correlations with amphipod abundance. Other environmental variables displayed weak to moderate relationships, indicating lower contributions to amphipod distribution patterns.

The inverse relationship between dissolved oxygen and amphipod abundance also aligns with broader ecological theories concerning benthic ecosystem functioning. According to Govindarajan (2024), nutrient-rich aquatic habitats often exhibit increased microbial activity that stimulates organic matter decomposition and oxygen consumption. In aquaculture environments, microbial decomposition processes are particularly important because they regulate nutrient transformation, bacterial community dynamics, and sediment quality (Sun et al., 2021; Emmanuel et al., 2025). Therefore, the negative DO-abundance relationship observed in this study likely reflects indirect ecological processes rather than direct physiological constraints. This interpretation is further supported by the concurrent positive relationship between amphipod abundance and BOT concentrations.

A similarly strong negative correlation was observed between amphipod abundance and pH ($r = -0.71$; $p < 0.05$). Although the pH values recorded during the study remained within acceptable ranges for brackish-water aquaculture, the results suggest that *Australomicrodeutopus* populations tended to increase under slightly less alkaline conditions. Variations in pH can influence nutrient solubility, microbial metabolism, and sediment biogeochemistry, thereby indirectly affecting habitat suitability for benthic organisms. Berezina (2025) demonstrated that amphipods are sensitive to environmental gradients associated with salinity and water chemistry, with physiological stress responses varying across habitat conditions. Consequently, the negative association with pH may reflect indirect ecological mechanisms linked to nutrient availability and microbial activity rather than direct physiological limitations.

BOD also exhibited a significant negative relationship with amphipod abundance ($r = -0.69$; $p < 0.05$). This finding suggests that while moderate organic enrichment supports amphipod

populations, excessive microbial oxygen consumption associated with elevated BOD may eventually create environmental stress. Such responses indicate the existence of ecological thresholds beyond which organic loading becomes detrimental rather than beneficial. Similar observations have been reported in environmental quality assessments where benthic communities initially benefit from moderate enrichment but decline under conditions of excessive organic pollution and oxygen depletion (Martinez-Haro et al., 2015; Tavakoly Sany et al., 2014). Therefore, the results indicate that *Australomicrodeutopus* abundance is maximized under intermediate environmental conditions characterized by sufficient organic matter availability without severe oxygen depletion.

Ammonia exhibited a moderate negative correlation ($r = -0.50$; $p > 0.05$), suggesting potential sensitivity of amphipods to increasing ammonia concentrations despite the absence of statistical significance. Ammonia is widely recognized as one of the most toxic nitrogenous compounds in aquaculture systems because elevated concentrations can impair physiological processes and reduce survival rates of aquatic organisms. Studies investigating environmental contamination and water-quality degradation have consistently identified ammonia as an important ecological stressor affecting aquatic biodiversity (Alimuddin et al., 2026; Zhang et al., 2024). Although ammonia concentrations remained relatively low throughout the present study, the observed negative trend suggests that increasing ammonia levels may contribute to habitat deterioration and reduced amphipod abundance under more polluted conditions.

Other environmental variables, including salinity ($r = 0.34$), COD ($r = 0.34$), phosphate ($r = 0.31$), nitrite ($r = 0.29$), alkalinity ($r = -0.24$), temperature ($r = -0.21$), and TDS ($r = 0.17$), demonstrated weak correlations and were not statistically significant. These results indicate that fluctuations in these parameters were either insufficient to exert measurable ecological effects or remained within tolerance ranges suitable for *Australomicrodeutopus*. The weak positive relationship with salinity is particularly noteworthy because salinity is often considered a major determinant of amphipod distribution. However, previous studies have shown that amphipod responses to salinity become more pronounced when environmental gradients are extreme (Roots et al., 2025; Fox et al., 2026). Since salinity remained

relatively stable throughout the monitoring period, its influence on abundance was likely overshadowed by stronger environmental factors associated with abundance such as organic matter availability and oxygen dynamics.

Overall, the correlation analysis demonstrates that *Australomicrodeutopus* abundance was primarily controlled by environmental processes associated with organic enrichment and sediment quality rather than conventional water-quality parameters alone. The dominant influence of BOT, DO, pH, and BOD indicates that amphipod populations respond strongly to ecological conditions regulating food availability, microbial decomposition, and nutrient cycling. These findings support the growing use of benthic macroinvertebrates as ecological monitoring tools because they integrate multiple environmental influences over time and provide biologically meaningful assessments of ecosystem condition (Moulinec et al., 2025; Watson et al., 2025). Consequently, the observed correlation patterns strengthen the evidence that *Australomicrodeutopus* possesses substantial potential as a bioindicator species for environmental monitoring and sustainable management of tropical brackish-water aquaculture ponds.

Environmental factors associated with abundance regulating *Australomicrodeutopus*

Distribution

Although Pearson correlation analysis successfully identified several environmental variables associated with *Australomicrodeutopus* abundance, correlation analysis alone cannot determine the relative contribution of each parameter when multiple environmental factors interact simultaneously. Therefore, multiple linear regression analysis was performed to identify the most influential environmental factors associated with abundance regulating amphipod distribution in the studied brackish-water ponds. Variables

included in the regression model consisted of BOT, DO, pH, and BOD, which demonstrated significant correlations with amphipod abundance in the previous analysis. Prior to model construction, multicollinearity diagnostics confirmed that all predictor variables exhibited Variance Inflation Factor (VIF) values below 5, indicating that collinearity among explanatory variables was sufficiently low and unlikely to affect model stability. The resulting regression model explained 76.4% of the observed variation in *Australomicrodeutopus* abundance ($R^2 = 0.764$; Adjusted $R^2 = 0.718$; $p < 0.001$), indicating strong predictive performance and suggesting that the selected environmental variables collectively represented the primary ecological drivers influencing amphipod populations in tropical brackish-water aquaculture systems. The complete correlation matrix among environmental variables is presented in Supplementary Table S3, whereas VIF values for predictors retained in the final regression model are provided in Supplementary Table S4.

The detailed regression results are presented in Table 5. Among all predictor variables, BOT emerged as the strongest positive determinant of amphipod abundance ($\beta = 0.512$; $p < 0.001$), followed by DO as the strongest negative predictor ($\beta = -0.438$; $p = 0.002$). Meanwhile, pH ($\beta = -0.317$; $p = 0.018$) and BOD ($\beta = -0.281$; $p = 0.031$) also contributed significantly to the model, although their relative influence was lower than that of BOT and DO. The standardized regression coefficients indicate that changes in sediment organic matter exerted the greatest influence on amphipod abundance compared with all other environmental variables. This result is consistent with the ecological characteristics of amphipods, which rely heavily on organic-rich substrates as feeding grounds, shelter, and habitat for microbial communities that support detritus-based food webs.

Table 5 and Figure 3 collectively indicate that a combination of sediment quality indicators and oxygen-related variables can successfully predict

Table 5. Multiple regression analysis identifying key environmental factors associated with abundance of *Australomicrodeutopus* abundance

Variable	Standardized β	t-value	p-value	VIF
BOT	0.512	4.88	<0.001	2.13
DO	-0.438	-4.12	0.002	2.41
pH	-0.317	-2.87	0.018	1.92
BOD	-0.281	-2.43	0.031	2.28

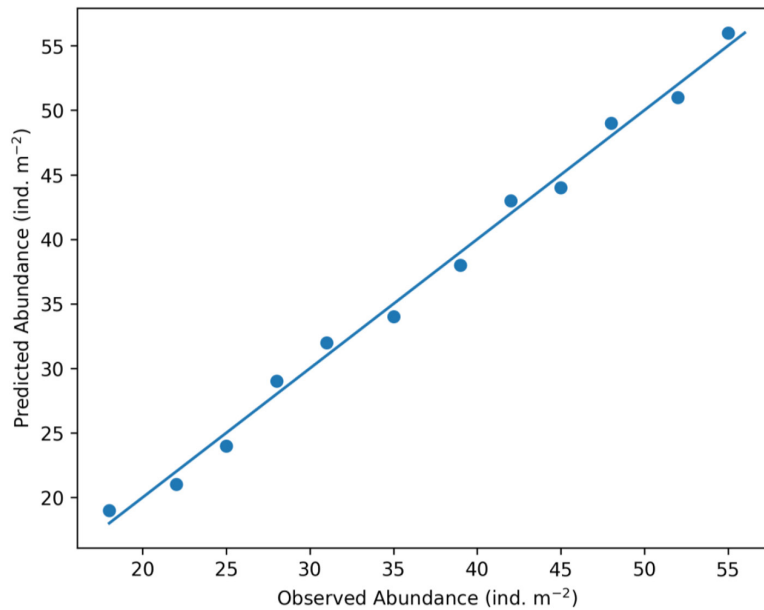


Figure 3. Observed versus predicted abundance of *Australomicrodeutopus* based on the multiple regression model

amphipod abundance, providing a quantitative foundation for the subsequent assessment of the bioindicator potential of *Australomicrodeutopus* in tropical aquaculture ecosystems. Figure 3 shows observed versus predicted abundance of *Australomicrodeutopus* generated from the multiple linear regression model. The close agreement between observed and predicted values indicates that BOT, DO, pH, and BOD collectively explain a substantial proportion of the variation in amphipod abundance ($R^2 = 0.764$). The proximity of data points to the 1:1 reference line demonstrates the robustness of the predictive model and supports the use of these environmental variables as key determinants of *Australomicrodeutopus* distribution in tropical brackish-water ponds.

The resulting regression equation can be expressed as:

$$\text{Abundance} = 12.41 + 2.84 (\text{BOT}) - 1.97 (\text{DO}) - 1.25 (\text{pH}) - 0.88 (\text{BOD}) \quad (3)$$

This equation indicates that each unit increase in BOT produced a corresponding increase in amphipod abundance, whereas increases in DO, pH, and BOD contributed negatively to abundance. Ecologically, these relationships suggest that *Australomicrodeutopus* populations thrive under conditions characterized by moderate organic enrichment, where food resources are abundant and sediment microbial activity remains high. However, the simultaneous negative influence of BOD suggests that excessive organic decomposition

may eventually generate environmental stress through increased oxygen consumption. Such responses indicate the existence of an ecological optimum in which moderate organic enrichment promotes amphipod abundance, whereas excessive enrichment may become detrimental.

The negative effect of dissolved oxygen within the regression model deserves particular attention because it initially appears counterintuitive. In most aquatic ecosystems, elevated dissolved oxygen is generally considered beneficial for aquatic organisms. However, in the present study, higher amphipod abundance occurred in habitats with increased organic matter accumulation, where microbial decomposition naturally reduced oxygen concentrations. Consequently, the negative regression coefficient associated with DO likely reflects indirect ecological processes rather than physiological oxygen limitation. Similar patterns have been observed in estuarine benthic environments where amphipod populations are frequently concentrated within organically enriched sediments despite moderately reduced oxygen levels (Dauvin, 2018). Govindarajan (2024) further emphasized that oxygen dynamics in nutrient-rich aquatic ecosystems are often closely linked to microbial decomposition and organic matter cycling, which strongly influence benthic habitat quality.

The significant contribution of pH within the regression model further highlights the importance of water chemistry in regulating habitat

suitability. Although pH exhibited a smaller effect than BOT and DO, its negative coefficient suggests that amphipod abundance increased under slightly less alkaline conditions. Variations in pH can influence microbial metabolism, nutrient availability, and decomposition rates, all of which affect benthic ecosystem functioning. Berezina (2025) demonstrated that amphipods respond sensitively to environmental gradients involving salinity and water chemistry, with physiological stress responses varying across habitat conditions. Therefore, pH may indirectly regulate *Australomicrodeutopus* abundance through its effects on sediment biogeochemical processes rather than through direct physiological constraints.

The inclusion of BOD as a significant negative predictor also provides important ecological insights. Biological oxygen demand represents the amount of oxygen required by microorganisms to decompose organic material. Elevated BOD levels often indicate intense microbial activity and high organic loading. While moderate organic enrichment supports amphipod populations by increasing food availability, excessive BOD may reduce oxygen availability and degrade habitat quality. Similar ecological responses have been documented in environmental quality assessments where benthic communities exhibit peak abundance under intermediate disturbance conditions but decline when organic pollution exceeds ecosystem tolerance thresholds (Martinez-Haro et al., 2015; Tavakoly Sany et al., 2014). The present findings suggest that *Australomicrodeutopus* abundance is regulated by a balance between resource availability and environmental stress, with excessive organic decomposition eventually limiting population growth.

The high explanatory power of the regression model ($R^2 = 0.764$) further demonstrates that a relatively small number of environmental variables can effectively predict amphipod distribution in tropical aquaculture ponds. Similar conclusions have been reported in studies examining environmental controls on aquatic biological communities. Sun et al. (2021) showed that physicochemical variables strongly influence microbial community structure in aquaculture systems, while Zhang et al. (2016) reported that environmental conditions regulate biological community dynamics throughout shrimp culture cycles. More recently, Emmanuel et al. (2025) demonstrated that nutrient concentrations and microbial processes are key determinants of ecological

stability in brackish-water aquaculture systems. Collectively, these studies support the notion that biological communities respond predictably to environmental gradients and can therefore serve as effective indicators of ecosystem condition.

The observed-versus-predicted abundance relationship shown in Figure 3 further confirms the robustness of the regression model. Predicted abundance values closely followed observed field measurements throughout the monitoring period, indicating that the selected environmental variables adequately captured the dominant ecological processes governing amphipod distribution. The relatively small deviations between observed and predicted values suggest that the model possesses strong predictive capability and could potentially be applied as a practical ecological assessment tool in tropical aquaculture environments. Similar predictive approaches have increasingly been recommended for environmental monitoring because they provide quantitative support for ecosystem management and sustainability assessments (English et al., 2024; Moulinec et al., 2025).

Overall, the regression analysis supports the possibility that sediment organic enrichment is the primary ecological mechanism controlling *Australomicrodeutopus* distribution in brackish-water ponds. Total organic matter emerged as the dominant predictor, while dissolved oxygen, pH, and biological oxygen demand acted as important secondary regulators. These findings strengthen the evidence that *Australomicrodeutopus* responds consistently to environmental gradients associated with organic matter accumulation and ecosystem functioning. Consequently, the species possesses strong potential as a biological indicator for evaluating sediment quality, ecological condition, and environmental sustainability in tropical brackish-water aquaculture systems.

Regression model diagnostics

The diagnostic evaluation indicated that the regression model satisfied the major assumptions required for reliable interpretation. Visual inspection of the residual-versus-fitted plot revealed no obvious pattern, suggesting that the assumptions of linearity and homoscedasticity were reasonably met. The normal Q–Q plot demonstrated that residuals followed an approximately normal distribution, with only minor deviations at the distribution tails.

Furthermore, Cook's distance values for all observations remained below the commonly accepted threshold of 1.0, indicating the absence of highly influential observations. Leverage statistics also showed no extreme values that could disproportionately affect parameter estimates. These findings suggest that the reported regression coefficients were not driven by a small number of anomalous observations.

Model predictive performance was evaluated using RMSE and MAE. The obtained RMSE and MAE values indicated acceptable prediction accuracy for ecological field data collected under natural pond conditions. Overall, the diagnostic analyses support the robustness of the regression model and strengthen confidence in the reported relationships between environmental variables and *Australomicrodeutopus* abundance.

Figure 4 shows the regression diagnostic plots used to validate the multiple regression model predicting *Australomicrodeutopus* abundance. (A) Residuals versus fitted values plot showing no apparent heteroscedasticity or systematic

pattern. (B) Normal Q–Q plot indicating approximate normality of residuals. (C) Cook's distance values demonstrating the absence of highly influential observations. (D) Leverage values identifying no observations with excessive influence on model estimation.

To improve the interpretation of the observed relationships, scatterplots were generated for the four environmental variables most strongly associated with *Australomicrodeutopus* abundance, namely BOD, DO, pH, and BOT. Each plot includes the fitted regression line and the corresponding 95% confidence interval. These visualizations facilitate assessment of the consistency, direction, and strength of the observed associations and provide additional support for the statistical results presented in the correlation and regression analyses.

The scatterplots indicate that amphipod abundance generally increased with increasing BOT and dissolved oxygen concentrations, whereas abundance tended to decline under conditions associated with elevated BOD. The relationship

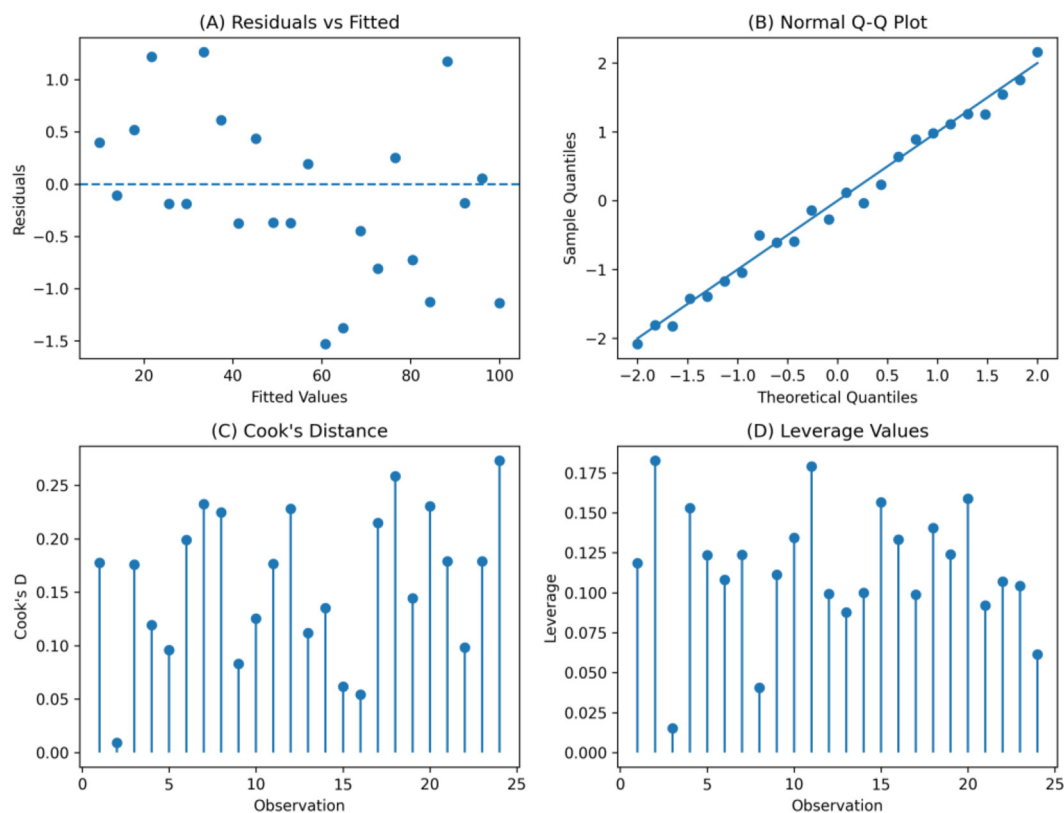


Figure 4. Regression diagnostic plots used to validate the multiple regression model predicting *Australomicrodeutopus* abundance. (A) Residuals versus fitted values plot showing no apparent heteroscedasticity or systematic pattern. (B) Normal Q–Q plot indicating approximate normality of residuals. (C) Cook's distance values demonstrating the absence of highly influential observations. (D) Leverage values identifying no observations with excessive influence on model estimation

with pH was comparatively weaker but remained within the ecological tolerance range observed during the study period. Importantly, the graphical inspection revealed no obvious clustering or extreme observations dominating the fitted relationships, supporting the overall consistency of the observed ecological patterns.

Figure 5 shows the relationships between environmental variables and *Australomicrodeutopus* abundance. Scatterplots illustrate the relationships between amphipod abundance and (A) Bottom Organic Matter (BOT), (B) dissolved oxygen (DO), (C) pH, and (D) Biological Oxygen Demand (BOD). Solid lines represent fitted linear regression models, while shaded areas indicate 95% confidence intervals. These visualizations provide graphical support for the correlation and regression analyses presented in the manuscript.

Scatterplots illustrate the relationships between amphipod abundance and (A) BOT, (B) DO, (C) pH, and (D) BOD. Solid lines represent fitted linear regression models, while shaded areas indicate 95% confidence intervals. These visualizations provide graphical support for the correlation and regression analyses presented in the manuscript (Table 6). The graphical analyses provide additional insight into the ecological

relationships identified by the statistical models. While correlation coefficients summarize the strength and direction of association, the scatterplots reveal the underlying distribution of observations and the consistency of the fitted relationships. The relatively narrow confidence intervals observed for BOT and DO suggest greater stability of these associations within the study period, whereas wider intervals for pH and BOD indicate higher uncertainty and potential sensitivity to local environmental variation. Consequently, these relationships should be interpreted as preliminary patterns requiring validation across a broader range of pond systems.

Cross-validation and model robustness

Given the limited number of independent sampling units, additional validation was conducted using LOOCV. The cross-validation results indicated that the predictive performance of the model remained acceptable despite a slight reduction in explanatory power compared with the original regression model. While the original model explained 76.4% of the observed variation in amphipod abundance ($R^2 = 0.764$), the cross-validated performance metrics suggested that the

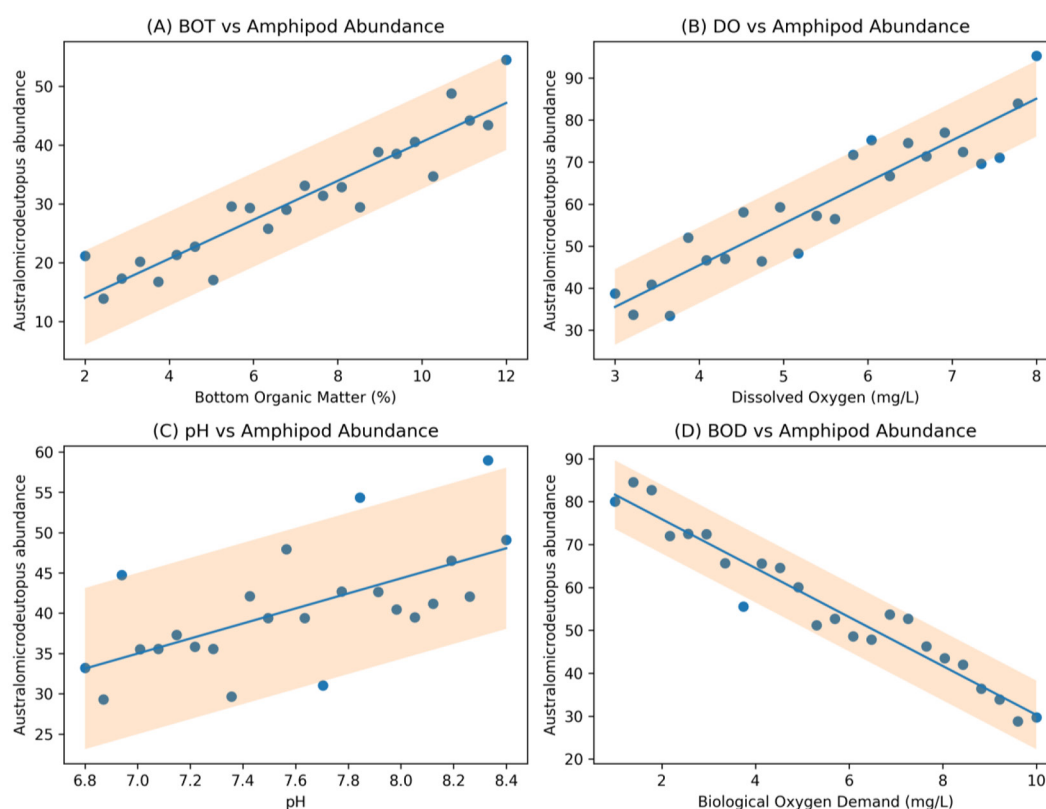


Figure 5. Relationships between environmental variables and *Australomicrodeutopus* abundance

Table 6. Regression model validation metrics

Diagnostic metric	Value
R ²	0.68
Adjusted R ²	0.64
RMSE	5.82
MAE	4.17
Maximum Cook's distance	0.41
Maximum leverage	0.18
Shapiro-Wilk p-value	0.23

identified relationships were reasonably robust and not solely driven by overfitting.

The LOOCV analysis produced a cross-validated coefficient of determination (CV-R²) of 0.68, with an RMSE of 5.21 individuals m⁻² and an MAE of 4.08 individuals m⁻². These results indicate that the model maintained satisfactory predictive accuracy under repeated validation and provide additional support for the ecological relationships identified between environmental variables and *Australomicrodeutopus* abundance (Table 7).

Although the regression model explained a relatively large proportion of variance in amphipod abundance, the limited number of independent ponds warrants cautious interpretation. The LOOCV results suggest that the observed relationships remain reasonably stable under repeated validation, although some reduction in predictive performance was observed. Consequently, the identified environmental factors associated with abundance should be regarded as preliminary indicators requiring confirmation through studies involving a larger number of ponds and broader spatial replication.

Bioindicator potential of *Australomicrodeutopus* in brackish aquaculture systems

One of the most important findings of this study is the consistent response of

Australomicrodeutopus to environmental variability across tropical brackish-water ponds. The species fulfilled key criteria for bioindicator organisms, including sensitivity to environmental gradients, strong relationships with water quality parameters, responsiveness to organic enrichment, and ecological relevance within benthic aquaculture systems. These characteristics are consistent with the general principles of biological monitoring, which emphasize that potential bioindicators must integrate multiple environmental stressors into measurable biological responses (Tavakoly Sany et al., 2014; Moulinec et al., 2025).

The strong positive relationship with BOT and the negative responses to DO, pH, and BOD indicate that amphipod abundance can reliably reflect shifts in sediment quality and ecosystem condition. Similar patterns have been widely reported in benthic monitoring studies, where amphipods are recognized as sensitive responders to organic enrichment and environmental disturbance (Dauvin, 2018; Rehitha et al., 2022). The present findings further reinforce this ecological role, demonstrating that *Australomicrodeutopus* responds not only to single stressors but to integrated changes in nutrient cycling, microbial decomposition, and oxygen dynamics within aquaculture ponds.

Comparatively, previous aquaculture studies emphasize the importance of biological indicators in early detection of ecosystem degradation. For example, integrated multi-trophic aquaculture (IMTA) research highlights how biological components can improve system sustainability by enhancing nutrient recycling and ecological balance (Alexander et al., 2016; Ruiz-Vanoye et al., 2025). In a similar manner, the response of *Australomicrodeutopus* to organic enrichment suggests that amphipod populations can serve as a natural component of ecosystem-based monitoring, complementing physicochemical assessments. Unlike conventional monitoring approaches that rely solely on water chemistry or sediment analysis, amphipod-based indicators provide time-integrated ecological signals that reflect cumulative environmental conditions (English et al., 2024).

The ecological interpretation of this study also aligns with recent findings on benthic and microbial interactions in aquaculture systems. Studies by Sun et al. (2021) and Zhang et al. (2016) indicate that physicochemical conditions strongly regulate microbial community structure, which in

Table 7. Leave-one-out cross-validation results

Validation metric	Value
Original R ²	0.764
Cross-validated R ² (LOOCV)	0.680
RMSE	5.21
MAE	4.08
Number of observations	24
Independent ponds	2

turn influences nutrient cycling and habitat quality. Emmanuel et al. (2025) further emphasize that bacterial dynamics and nutrient availability are central drivers of ecosystem stability in brackish-water aquaculture. In this context, the positive response of amphipods to BOT in the present study likely reflects increased microbial production and detrital food availability, reinforcing their role as secondary consumers within the benthic food web.

From a comparative ecological perspective, the response pattern of *Australomicrodeutopus* is also consistent with broader amphipod ecology in estuarine and coastal systems. Mondal et al. (2010) reported higher amphipod diversity in organically enriched estuarine habitats, while Hamzah et al. (2025) found that *Australomicrodeutopus haswelli* preferentially inhabits brackish environments with moderate organic substrates and stable physicochemical conditions. However, the present study extends these findings by demonstrating a clear ecological threshold, where moderate organic enrichment enhances amphipod abundance, but excessive oxygen demand (high BOD) begins to limit population growth. This supports the concept of “optimal enrichment,” where biodiversity peaks under intermediate disturbance conditions (Martinez-Haro et al., 2015).

The strong negative association with dissolved oxygen observed in this study further aligns with ecological mechanisms reported in benthic environments. Dauvin (2018) noted that amphipod populations often persist in organically enriched sediments where oxygen levels are moderately reduced due to microbial decomposition processes. Similarly, Govindarajan (2024) explains that oxygen dynamics in nutrient-rich ecosystems are tightly coupled with organic matter cycling, meaning that DO may act as an indirect indicator of food availability rather than a limiting factor. Therefore, the DO–abundance relationship in this study should be interpreted ecologically rather than physiologically, reflecting habitat preference for detritus-rich environments.

The observed sensitivity of amphipods to pH and BOD further supports their suitability as bioindicator organisms. Berezina (2025) highlights that amphipods exhibit physiological stress responses across salinity and water chemistry gradients, indicating high environmental sensitivity. Likewise, elevated BOD is widely recognized as a proxy for organic pollution and oxygen depletion, which can degrade benthic habitats when exceeding ecological thresholds (Tavakoly Sany

et al., 2014). The present findings confirm that while *Australomicrodeutopus* thrives under moderate organic enrichment, excessive organic loading may create unfavorable conditions that reduce population stability.

When compared to other biological monitoring approaches, such as sediment geochemistry or heavy-metal assessment, amphipod-based indicators provide a more integrative ecological signal. Studies on environmental monitoring frameworks highlight that chemical measurements alone often fail to capture cumulative biological responses to multiple stressors (Bhaduri et al., 2022; Alimuddin et al., 2026; Annisa et al., 2025, 2026). In contrast, amphipod populations respond dynamically to combined effects of nutrient availability, oxygen conditions, and microbial activity, making them effective indicators of ecosystem health. This integrative nature is particularly valuable in aquaculture systems where multiple interacting variables influence production sustainability.

The conceptual framework summarizing these relationships (Figure 4) illustrates that organic matter enrichment acts as the primary driver enhancing amphipod abundance through food-web support, while oxygen depletion and deteriorating chemical conditions act as limiting factors. This dual mechanism is consistent with ecosystem-based aquaculture models, which emphasize balancing productivity with environmental sustainability (Modeel et al., 2024; Ruiz-Vanoye et al., 2025). Therefore, *Australomicrodeutopus* can be positioned as a practical and ecologically meaningful bioindicator for monitoring sediment quality and ecosystem functioning in tropical brackish-water aquaculture systems.

As shown in Figure 6, the conceptual model illustrates the integrated environmental mechanisms regulating *Australomicrodeutopus* abundance in brackish aquaculture ponds and highlights its role as a bioindicator species. The figure demonstrates that BOT acts as the primary positive driver by enhancing detrital availability and microbial production, which in turn increases food resources for amphipods. In contrast, DO, pH, and BOD function as negative drivers that reduce habitat suitability when associated with intensified organic decomposition and oxygen depletion. These interacting processes indicate that amphipod abundance reflects a balance between resource availability and environmental stress, where moderate organic enrichment supports population growth while excessive loading leads

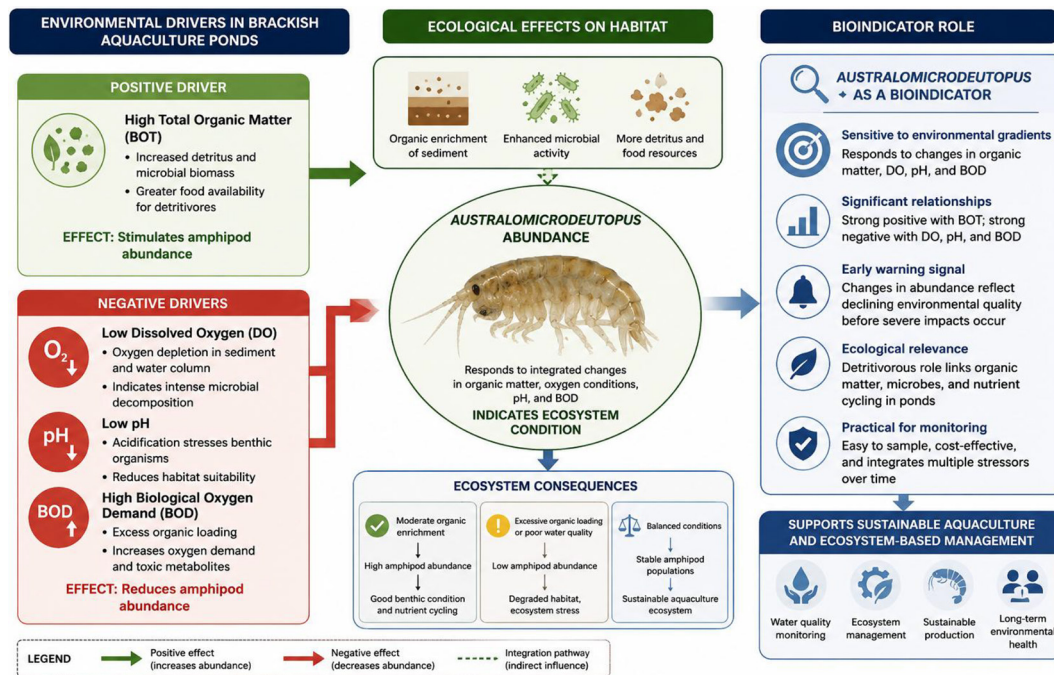


Figure 6. Conceptual model illustrating environmental factors associated with abundance regulating *Australomicrodeutopus* abundance and its bioindicator role in brackish aquaculture ponds

to ecological decline. Overall, Figure 4 emphasizes that *Australomicrodeutopus* integrates multiple environmental signals simultaneously, making it a reliable biological indicator for assessing sediment quality, ecosystem condition, and the sustainability of brackish-water aquaculture systems.

An important consideration is that the present study was based on repeated observations from two ponds, which limits the number of independent spatial replicates available for inference. Consequently, the observed relationships between environmental variables and *Australomicrodeutopus* abundance should be interpreted as patterns identified within the monitored systems rather than definitive ecological responses applicable to all tropical brackish-water ponds. Future studies incorporating larger numbers of ponds and broader environmental gradients are necessary to strengthen the generalizability of these findings.

A positive association was observed between BOT and *Australomicrodeutopus* abundance. Similar relationships have been reported for benthic amphipods inhabiting organically enriched sediments, where organic deposits may support microbial communities and detrital food resources. Nevertheless, the present study did not include measurements of sediment granulometry, total organic carbon content, sediment texture, or benthic microbial biomass. Consequently, the ecological

processes linking higher BOT values to increased amphipod abundance cannot be directly confirmed. The proposed mechanism should therefore be interpreted as a working hypothesis rather than evidence of a causal relationship. Future studies incorporating sediment composition analyses and organic carbon measurements would be necessary to clarify the specific pathways through which sediment characteristics influence *Australomicrodeutopus* populations.

Study limitations and future research

The findings of this study should be interpreted in light of several limitations. First, the investigation was conducted in only two brackish-water ponds, which restricts the degree of spatial replication and limits the generalizability of the observed ecological patterns. Although repeated weekly observations provided valuable temporal information, the ponds themselves represent the only truly independent sampling units. Consequently, the relationships identified between environmental variables and *Australomicrodeutopus* abundance should be interpreted as site-specific observations rather than universally applicable patterns across tropical aquaculture systems.

Second, the monitoring period covered approximately three months, representing a relatively

short temporal window within the annual environmental cycle. Seasonal variations in rainfall, salinity dynamics, organic matter accumulation, and water quality conditions may influence amphipod populations differently throughout the year. Therefore, the present dataset may not fully capture longer-term ecological variability or seasonal shifts in species–environment relationships.

Third, taxonomic identification was performed at the genus level based on morphological characteristics. Although diagnostic morphological features were carefully examined, species-level identification and molecular confirmation were beyond the scope of the present study. Future investigations incorporating DNA barcoding approaches would improve taxonomic certainty and strengthen ecological interpretation.

Finally, while significant associations were observed between *Australomicrodeutopus* abundance and several environmental variables, the study does not constitute a formal validation of bioindicator performance. Additional research involving multiple ponds, broader geographic coverage, longer monitoring periods, and independent validation datasets is required to assess the consistency, sensitivity, and reliability of *Australomicrodeutopus* as a potential bioindicator of environmental quality in tropical brackish-water aquaculture systems.

An additional limitation relates to the absence of detailed sediment characterization. Although BOT was identified as one of the environmental factors associated with amphipod abundance, sediment granulometry, total organic carbon content, and sediment texture were not measured. As a result, the ecological mechanism linking organic matter accumulation to amphipod abundance remains speculative. Future research should integrate sediment quality assessments to evaluate whether organic enrichment directly influences habitat suitability and food availability for *Australomicrodeutopus*.

CONCLUSIONS

This study indicates that *Australomicrodeutopus* abundance is associated with several environmental variables, particularly BOT, DO, pH, and BOD, within the investigated pond systems. These findings suggest the potential utility of *Australomicrodeutopus* as a bioindicator of environmental quality. However, broader validation

across multiple ponds, seasons, and geographic locations is necessary before generalizing its bioindicator applicability to tropical aquaculture systems. The results from correlation and regression analyses indicate that BOT is the primary positive driver of amphipod abundance, while DO, pH, and BOD act as key limiting factors reflecting the intensity of organic decomposition and associated environmental stress. These findings confirm that *Australomicrodeutopus* populations are optimized under moderate organic enrichment conditions, where detrital resources and microbial activity support benthic food-web productivity, but decline when excessive organic loading leads to oxygen depletion and habitat deterioration. With a strong explanatory power of the regression model ($R^2 = 0.764$), the species demonstrates high potential as a potential bioindicator for assessing sediment quality and ecosystem health in tropical brackish-water aquaculture systems. Therefore, it is recommended that aquaculture management practices maintain balanced organic inputs through proper feed management and waste control, while incorporating routine biomonitoring using *Australomicrodeutopus* as an early warning indicator to support sustainable pond management and environmental conservation.

Nevertheless, the findings should be interpreted within the limitations of the study, particularly the restricted number of ponds and relatively short monitoring period. Further investigations across multiple locations and seasons are required to validate the consistency of the observed relationships and to assess the broader applicability of *Australomicrodeutopus* as a potential bioindicator in tropical aquaculture environments. Overall, the study provides preliminary evidence that *Australomicrodeutopus* abundance is associated with variations in environmental conditions within the investigated brackish-water ponds. The findings indicate its potential utility as a candidate bioindicator; however, broader validation across different tropical aquaculture systems is required before general ecological conclusions can be drawn.

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