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Research Article

Temporal Dynamics in Density of the Freshwater Mussel, *Lamellidens marginalis*, in a Lentic Habitat of Bangladesh

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Abstract

Freshwater mussel, *Lamellidens marginalis* (Lamarc, 1819), is a commercially important species in Bangladesh. It is widely used in pearl, lime, and poultry food production. Information about temporal variation of *L. marginalis* population in aquatic ecosystems is inadequate in Bangladesh. Hence, it is difficult to plan farming and harvesting of *L. marginalis*. A year-long study was conducted at a culture pond in Mymensingh, Bangladesh to understand the temporal variation of *L. marginalis* population during April 2023–March 2024. Quadrat method was used to estimate its natural population in the pond of 502 m^2 area. Mussels were collected monthly by hand-picking from eight distant quadrats across the pond, each having an area of 4 m^2. Their mean population in the pond varied from 988 to 2071 individuals where minimum being in September and maximum being in April. The pond's water quality parameters–depth, temperature, and pH–were simultaneously monitored to correlate with mussel population. The annual mean depth, temperature, and pH were 1.51 m, 23.59 °C, and 7.37, respectively. Depth was found to be significantly negatively correlated (Spearman's ρ = -0.97, P < 0.001) with population size. No substantial correlation was found between population and temperature or population and pH. Biometric characteristics of the mussels were also recorded during the study. The mean shell length, width, and height were 92.10, 44.05, and 20.27 mm, respectively. Overall, this study provides a basis for farming and harvesting quality mussels in a lentic habitat. The collected data have been released publicly via GitHub to facilitate future mussel population dynamics research.

Introduction

Freshwater mussels are important ecologically, commercially, and nutritionally (Souza et al. 2024; Siddique et al. 2020; Yaghubi et al. 2021). Due to their macro size and abundance, they can significantly influence the surrounding benthic ecosystem. Bangladesh has four major species of freshwater pearl mussels: *Lamellidens marginalis, Lamellidens corrianus*, *Lamellidens phenchooganjensis,* and *Lamellidens jenkinsianus* (Siddique et al. 2024). Among them, *L. marginalis* is known to produce the best quality pearls. It is also used in lime, poultry, and aquaculture food production (Siddique et al. 2020). On the other hand, farmed mussels are a viable source of high-quality protein, omega-3 fatty acids, B-12, zinc, and iron (Yaghubi et al. 2021). Since mussels feed by filtering water, they absorb heavy metals and harmful particles suspended in water. This filtration makes the water clear and lets the sunlight penetrate deeper towards the bottom. The increased photons facilitate benthic plants to perform photosynthesis and enhance primary productivity (Souza et al. 2024). As a result, energy and oxygen are produced for other aquatic organisms. The sunlight boosts growth of phytoplankton and zooplankton that enhances fish food production. Overall, mussels are considered as a valuable indicator of water quality (Crotty et al. 2023). Their filter feeding process precipitates a large volume of material on marsh surfaces and thereby helps it grow (Crotty et al. 2023). This accretion has the potential to prevent bank erosion. Moreover, their shells provide habitat for insects, small fish, and plants (Crotty et al. 2023). All the benefits come at no feed cost. Because of all the environmental, economic, and food values, knowing mussel's population at different times of the year would be helpful to plan farming and harvesting. But information about temporal variation of *L. marginalis*

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population in aquatic ecosystems is inadequate in Bangladesh.

Quadrat method of mussel population estimation has been popular for its simplicity and objectivity. A quadrat is a frame laid on the bottom bed of a water body to mark an area where organisms would be sampled. The frame can be square, rectangular, or circular. The quadrats should be spatially segregated (Radinger et al. 2019) so that the samples are independent from one another and representative enough of the entire community. A recent article established the reliability of quadrat method in estimating mussel population by comparing with true numbers along Norwegian coast (Reamon et al. 2024). Scientists sampled mussels in Ireland (Harrison et al. 2024) and Bangladesh (Ollard et al. 2024) using square quadrats. A past work also assessed the efficacy of a quadrat-based mussel sampling protocol in Canadian rivers (Reid and Morris 2017). Because of its established effectiveness, the quadrat method can be used to estimate mussel population.

A lentic habitat, in this case the culture pond, experiences a great seasonal variation of physicochemical factors that affects the water quality. The water quality parameters we recorded are: depth, temperature, and pH. Some literature reported effects of our chosen water quality parameters on other freshwater mussels. Quagga mussel growth was greatest at 15 m depth but lower below 45 m in Lake Ontario (Elgin et al. 2022). Besides, depth was suggested as a good predictor for *Echyridella menziesii* density in warm monomictic lakes in New Zealand (Cyr et al. 2017). On the other hand, adverse effects of elevated water temperature (20– 35 °C) on juvenile mussels were studied in a laboratory setting (Ganser et al. 2013). Heart rate was found to decline at higher temperatures for multiple mussel species (Ganser et al. 2013). Moreover, multiple thermal thresholds have been reported that would cause long-term mortality in glochidia and juvenile mussels across Tar River Basin, North Carolina (Pandolfo et al. 2024). pH is another critical physicochemical factor that affects mussel physiology. Its effect on the growth, survival, and reproduction of quagga mussels was studied using water from lakes in the Western United States (Seitz et al. 2023). However, effects of the above water quality parameters on *L. marginalis* density in a natural environment are still unknown.

There are some publicly available mussel datasets. For example, NOAA has a mussel watch program that samples oysters and mussels as surrogates of water pollution and bioaccumulation from over 400 sites across the United States (National Centers for Coastal Ocean Science 2025). *Mytilus* mussel data from the northern Gulf of Alaska reports mussel counts, size measurements, and sampling sites' layout (U.S. Geological Survey 2024). Another effort was made to compile a traits dataset of several freshwater mussels of Margaritiferidae and Unionidae families but that was based on published sources and no field work (Hopper et al. 2023). To the best of our knowledge, no dataset is present yet about the natural population dynamics of *L. marginalis*.

Effects of environmental factors on the population dynamics of a different freshwater mussel, *Parreysia corrugata*, was investigated in a lotic habitat (Haque et al. 2024). Another work showed monthly changes in glochidia larvae of *L. marginalis* in a wetland (Salam et al. 2024). A recent work presented the decline of freshwater mussels in water bodies in the capital city of Bangladesh (Ollard et al. 2024). That work sampled mussels from the water bodies in the year of 2023 and compared them against their prior samples from 2010. However, seasonal changes of mussel population in a lentic habitat remain an unexplored topic. To the best of our knowledge, this is the first attempt to record such changes in a culture pond i.e., lentic habitat.

Given the above scope, our objectives were to: 1) Estimate the natural population of mussels in a lentic habitat. 2) Infer the mathematical associations among water quality parameters and mussel density. 3) Release the first publicly available *L. marginalis* population dynamics dataset.

Methods

A quadrat sampling method was used to estimate the natural population of the freshwater mussel *Lamellidens marginalis*. The mussels were sampled at a culture pond in Mymensingh, Bangladesh during April 2023–March 2024. They were collected monthly by hand-picking from eight random quadrats placed on the pond bed. First, Kruskal-Wallis test was used to show that the eight samples are uniform. Then, their numbers were averaged to compute density and estimate total population in the pond. Meanwhile, three water quality parameters–depth, temperature, and pH of the pond were recorded on the day of sampling. Spearman's rank correlation test was used to deduce mathematical associations among the above water quality parameters and mussel density. To work with live mussels, the guidelines of animal ethics provided by the ethical and animal welfare committee of Bangladesh Agricultural University were followed (BAURES ethical clearance certificate number PR/102/ESRC/FISH/2025).

Figure 1. The dot (24.69930°N, 90.41437°E) on Mymensingh shows the location of study site (https://www.openstreetmap.org/copyright). The rectangle on Bangladesh shows Mymensingh district. The inset shows part of Asia.

Study Site

The study was conducted at Borobilarpar (24.69930°N, 90.41437°E), Mymensingh, Bangladesh (Fig. 1). The study pond (Fig. 2) was man-made. It was rectangular in shape having length and width 25.1 m and 20 m, respectively (Fig. 3). Its surface area was 502 m^2. It had nearly uniform depth with an annual average of 1.51 m. This pond was selected for its regular shape, freshwater mussel (*L. marginalis*) abundance, and a favorable ecosystem. The pond was rain-fed and free from surface aquatic vegetation. It was well exposed to sunlight. Its water was used for household chores such as bathing and laundry. Its water color appeared greenish throughout the study period. Various freshwater fishes were cultured in this pond including silver carp, mrigal carp, tilapia, rohu, etc.

Figure 2. The study pond at Borobilarpar, Mymensingh, Bangladesh.

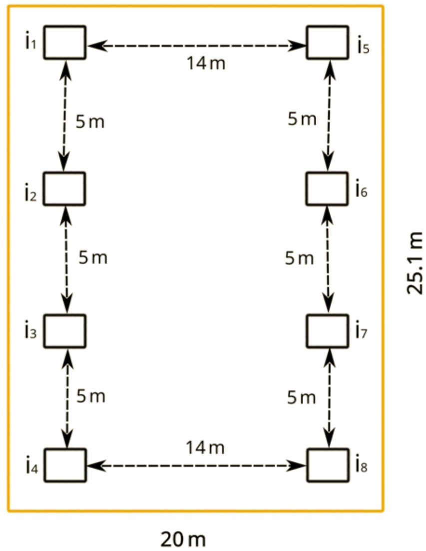


Figure 3. Illustration of the study pond's dimensions, eight quadrats, and their spatial segregation.

Sampling Framework

For estimating the natural population of *L. marginalis* in the pond, mussel samples were collected during April 2023–March 2024 on the 15th day of each month. The sampling time was 9–4 pm. The quadrat method of sampling was used. Four two-meter bamboo sticks were tied together to form a square quadrat of 4 m^2 area. The quadrats were placed on the bottom mud at eight different locations e.g., i_1, i_2, …, i_8 (Fig. 3) in different months. The choice of eight samples is well established in literature (Miller et al. 2017). It was mathematically shown that 2.1– 8.0 samples can detect 90–99% mussel species in a site regardless of abundance (Miller et al. 2017). Like our approach, three random 1 m^2 quadrats over a rectangular block were used to estimate mussel population in Canadian rivers (Reid and Morris 2017). While placing our quadrats, special attention was given to maintain adequate spatial segregation to prevent autocorrelation (Radinger et al. 2019, Ollard et al. 2024). An example placement along the perimeter of the pond (Fig. 3) shows their vertical distances 5 m and horizontal distances 14 m. The quadrats were placed 1 m from the pond shore. Afterward, each quadrat was thoroughly searched by hand to collect mussels up to a sediment depth of 6 inches. The search duration was 30 minutes. Such hand-picking approach was followed by a previous work (Chowdhury et al. 2016). During collection, the samples were stored in separate containers labeled after the eight quadrats. Then the samples were promptly transported to the Aquatic Ecology Laboratory at Bangladesh Agricultural University, Mymensingh for further analysis.

Measurement of Morphological Dimensions

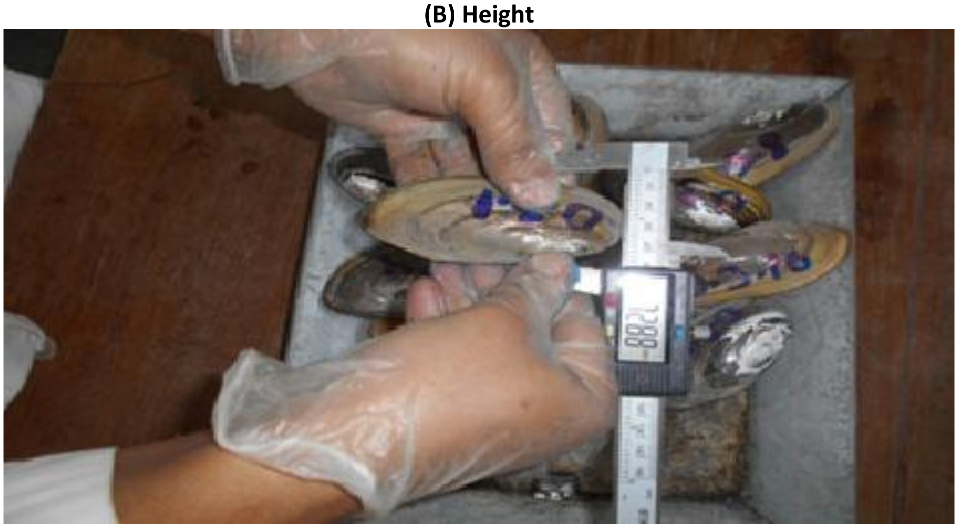
In the laboratory, the mussels were thoroughly washed with running tap water to remove dirt. Then, they were wiped dry using absorbent paper. Finally, their morphological dimensions were measured by digital Vernier calipers (model: Heng Liang, China) to an accuracy of 0.01 cm. The measured dimensions (Fig. 4) were shell length, height, and width as defined in prior literature (Haque et al. 2024).

Monitoring of Water Quality Parameters

Eight concomitant water quality parameters were recorded during the study period with a view to drawing analogy with temporal variation of mussel population. The parameters were depth, temperature, and pH. These were recorded monthly on the sampling day. Eight readings for each parameter–one at the center of every quadrat (Fig. 3) was taken. The average of the eight readings was reported as the monthly value of each parameter. The depth of the pond was read using a bamboo pole by settling it on the bottom vertically. The surface of the water was marked with chalk. After bringing it to the shore, accurate reading was taken using a measuring tape. A Celsius thermometer was used for measuring water temperature. The thermometer was put directly in pond water for 3 minutes to obtain its temperature. pH is an important quality metric of a culture pond. It indicates acidity-alkalinity state of the lentic habitat. It was measured by a pH meter (model: pHep® by Hanna Instruments, USA).

(A) Length

(B) Height



(C) Width

Figure 4. Measurement of morphological dimensions of a *Lamellidens marginalis*.

Estimation of Mussel Population

Our primary problem was to estimate the total mussel population in the pond from collected samples every month. Freshwater pearl mussels (*L. marginalis*) were sampled every month during April 2023–March 2024 using eight quadrats. Let i_n be the number of samples from n^{th} quadrat in each month. Then, for quadrat area a and pond area A, total mussel population N for that month is given by Eq. 1. $N = \frac{\sum_{n=1}^{8} i_n}{8 \times a} \times A$ (Eq.1)

It can also be written as Eq. 2 where D is the average mussel population density. The latter is defined as the number of mussels per unit area for that month.

$$N = D \times A \quad \text{(Eq.2)}$$

$$D = \frac{\sum_{n=1}^{8} i_n}{8 \times a} \quad \text{(Eq.3)}$$

The density to population estimation formulae should hold true when the underlying mussel distribution is uniform across the area. This is a conventional assumption and was utilized in prior literature for mussel sampling (Chowdhury et al. 2016). We went one step further. It was shown that our quadrat samples are independent from one another. In other words, there are no substantial differences among our quadrat samples. This was achieved using Kruskal-Wallis test over eight quadrats' samples: i_1, i_2, …, i_8. Under the condition of uniformity and indifference, Eq. 2 holds true.

Relation Between Mussel Population and Water Quality Parameters

Our secondary problem was to assess relationships between mussel population and water quality parameters. For this task, Spearman's rank test was used. It is a non-parametric test that makes no assumption about normality of the involved variables. Hence, it is appropriate for our use case. This method ranks the involved dependent and independent variables and computes correlation from the paired ranks. In month t, the dependent variable was mussel population density D_t and independent variables were water depth, temperature, and pH. One independent variable x was considered at a time to calculate Spearman's rank correlation ρ (Best and Roberts 1975).

$$\rho = 1 - \frac{6\sum_{t=1}^{n}\left(d_t^x\right)^2}{n(n^2-1)} \quad \text{(Eq.4)}$$

$$d_t^x = |\, rank(D_t) - rank(x_t)\, | \quad \text{(Eq.5)}$$

Here, d_t^x is the difference in rank between D_t and one independent variable x_t (depth, temperature, or pH) in month t; n is the number of observations: 12 for 12 months.

Data Analysis

All data analyses were performed using R version 4.4.2 (R Core Team 2024). Kruskal-Wallis test was performed using *kruskal.test* function. It was used to show that the eight quadrats' samples came from the same distribution. By virtue of that, we could estimate total mussel population N from density D in Eq. 2. On the other hand, Spearman's rank test was performed using *cor.test* function. It was used to show correlation between mussel population density and water quality parameters according to Eq. 4. Both functions came from *stats* package version 4.4.2. Scatter plots (Fig. 6) of dependent and independent variables along with Spearman's rank correlation ρ were obtained using *ggscatter* function of *ggpubr* package version 0.6.0 (Kassambara 2023). All R codes used in analyses were publicly released for reproducibility of results and facilitation of future research. Those are available at https://doi.org/10.5281/zenodo.14996734.

Table 1. Monthly variation of mussel population in study pond.

Month	Number of mussels per quadrat								Population density (no./m^2)	Total population size
	i_1	i_2	i_3	i_4	i_5	i_6	i_7	i_8	$D = \frac{\sum_{n=1}^{8} i_n}{8 \times a}$	$N = D \times A$
Apr 2023	21	13	16	20	14	16	16	16	4.13	2071
May 2023	15	12	13	16	13	14	13	14	3.44	1726
Jun 2023	13	8	13	12	10	12	11	11	2.81	1412
Jul 2023	8	11	12	9	10	12	10	9	2.53	1271
Aug 2023	8	9	8	7	8	10	9	9	2.13	1067
Sep 2023	9	7	8	9	8	7	8	7	1.97	988
Oct 2023	10	10	11	11	12	10	11	12	2.72	1365
Nov 2023	15	12	13	14	12	13	14	14	3.34	1679
Dec 2023	17	13	12	18	14	13	16	15	3.69	1851
Jan 2024	14	12	15	14	14	13	14	14	3.44	1726
Feb 2024	16	15	14	15	14	15	16	15	3.75	1883
Mar 2024	17	13	18	16	14	17	17	16	4.00	2008
Mean±SD	-	-	-	-	-	-	-	-	3.16 ± 0.72	1587 ± 360

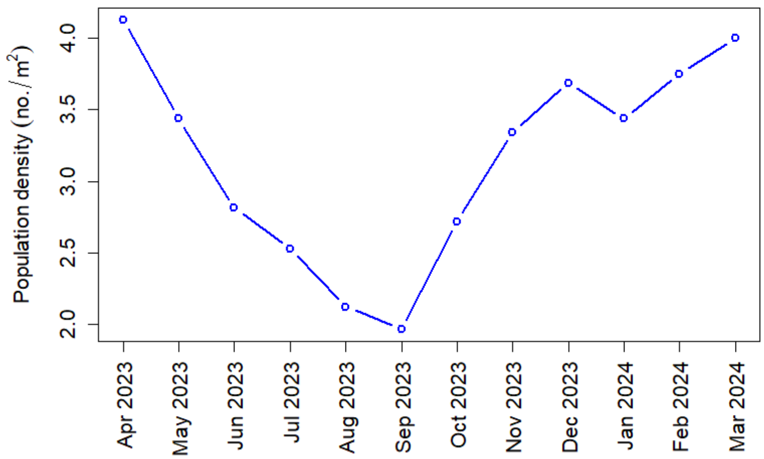


Figure 5. Monthly variation of mussel population density (D_t) in study pond.

Results

Mussel Population Estimation

From the culture pond in Fig. 1, the freshwater pearl mussels (*Lamellidens marginalis*) were collected on the 15th day of each month during April 2023–March 2024. The mussels were sampled from eight distant quadrats (Fig. 3). The positions of the quadrats were varied every month. During the study, a total of 1214 mussels were sampled and brought to our laboratory for further analysis. The quadrat-wise collections (i_1, i_2, …, i_8), population density per unit area (D), and estimated total population (N) of the whole pond are noted in Table 1. Area of each square quadrat (a) was 4 m^2; area of the rectangular pond (A) was 502 m^2.

As discussed in methods, the sample to population estimation formulae (Eqs 2–3) will hold true if the eight quadrats' samples came from the same distribution. To show that there are no significant differences among the quadrat samples, Kruskal-Wallis test was used. The result (χ^2 = 5.52, df = 7, P = 0.60) indicates that there were no significant differences among quadrats. In other words, all samples came from the same population distribution. Therefore, Eqs 2–3 could be applied to estimate the total population (Table 1).

Overall, *L. marginalis* population was highest in April (D = 4.13 no./m^2, N = 2071 no.) and lowest in September (D = 1.97 no./m^2, N = 988 no.). Monthly variation of mussel population density (D_t) is shown in Fig. 5.

Morphological Dimensions

A total of 1214 mussels were collected during the study. The morphological dimensions of all samples were measured. Their mean shell length, width, and height were 92.10 ± 6.26, 44.05 ± 3.48, and 20.27 ± 1.48 mm, respectively.

Table 2. Monthly variation of water quality parameters (*x*) in study pond.

Month (t)	Water quality parameters (x)			Population density (D_t)
	Depth (meter)	Temperature (°C)	pH	no./m^2
Apr 2023	1.15	25.75	7.63	4.13
May 2023	1.35	26.13	7.65	3.44
Jun 2023	1.56	26.34	7.67	2.81
Jul 2023	1.77	26.17	7.72	2.53
Aug 2023	1.93	27.12	7.64	2.13
Sep 2023	2.10	27.35	7.70	1.97
Oct 2023	1.55	24.50	7.44	2.72
Nov 2023	1.46	21.26	6.81	3.34
Dec 2023	1.42	15.67	6.76	3.69
Jan 2024	1.35	18.50	6.41	3.44
Feb 2024	1.25	21.61	7.68	3.75
Mar 2024	1.19	22.67	7.30	4.00
Mean ± SD	1.51 ± 0.30	23.59 ± 3.71	7.37 ± 0.45	3.16 ± 0.72

Water Quality Parameters

Three water quality parameters: depth, temperature, and pH were recorded on the sampling day of every month during the study period April 2023–March 2024. Their monthly variations are shown in Table 2. The mean of each parameter is defined as the arithmetic average of its monthly values. Therefore, the mean pond depth was 1.51 m with range being [1.15 (April 2023), 2.10 (September 2023)] m. The mean water temperature was 23.59 °C with range being [15.67 (December 2023), 27.35 (September 2023)] °C. Finally, the mean pH was 7.37 with range being [6.41 (January 2024), 7.72 (July 2023)].

Correlation Between Variation of Population Density and Water Quality Parameters

The correlations between monthly variation of *L. marginalis* population density and the water quality parameters were calculated using Spearman rank method (Eqs 4–5). The correlation coefficients ρ, corresponding *P*-values and *S* statistics are shown in Table 3. Meanwhile, Fig. 6 takes on a broader approach of illustrating all attributes in one place: 1) data points representing dependent and independent variables, 2) a regression line fitted over the data points, 3) 95% confidence region of the regression line, 4) ρ and *P*-values.

As seen in Table 3 and Fig. 6, monthly variation of mussel population density was found to be significantly negatively correlated with pond depth (Spearman's rank test: S = 563.97, ρ = -0.97, P < 0.001). But no significant correlation was found between population density and temperature (Spearman's rank test: S = 468.32, ρ = -0.64, P = 0.026), or between population density and pH (Spearman's rank test: S = 409.22, ρ = -0.43, P = 0.162).

Table 3. Spearman's rank test between variation of population density and water quality parameters in study pond.

Water quality parameter (*x*)	*S* statistic	Spearman's rank correlation (ρ)	*P*-value
Depth	563.97	-0.97	<0.001
Temperature	468.32	-0.64	0.026
pH	409.22	-0.43	0.162

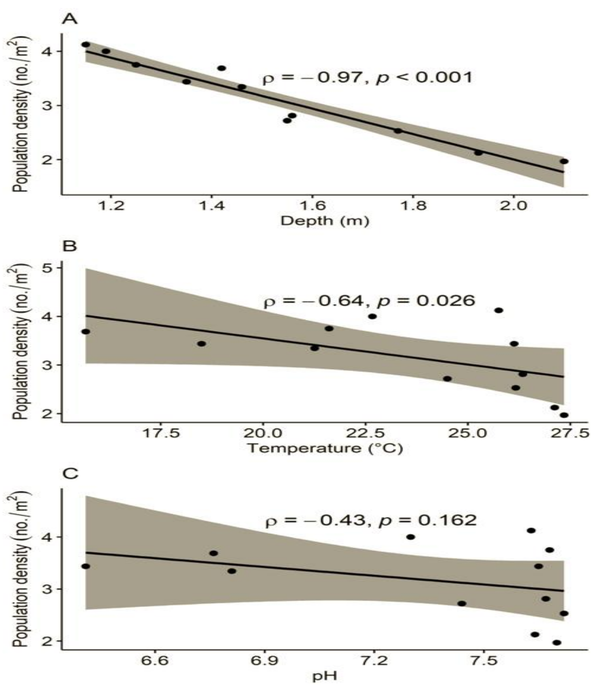


Figure 6. Spearman rank correlation between variation of population density and (A) depth (ρ = -0.97, P < 0.001), (B) temperature (ρ = -0.64, P = 0.026), (C) pH (ρ = -0.43, P = 0.162).

Discussion

The quadrat method of sampling mussels is a popular method (Harrison et al. 2024, Ollard et al. 2024, Reamon et al. 2024). A quadrat sampling method was applied to assess temporal variation of mussel population in a lentic habitat i.e., culture pond. To the best of our knowledge, this is the first attempt at yearly monitoring of mussel population in a constrained yet natural environment. During the study, April 2023–March 2024, a total of 1214 freshwater pearl mussels (*Lamellidens marginalis*) were sampled from the culture pond using eight quadrats. As apparent from Fig. 5 and Table 1, mussel population was higher in March–April (D = 4–4.13 no./m^2, N = 2008–2071 no.) and lower in August–September (D = 1.97–2.13 no./m^2, N = 988–1067 no.). The annual average density was 3.16 ± 0.72 no./m^2. This variation should be related to the life cycle of pearl mussels–how long they take to become adults from glochidia. Though we did not track sampled mussels' ages, from the little standard deviations of their shell dimensions, we can assume that their growth period was the same. Let the period be T months. Given that, higher population in March–April should mean their glochidia was released T months ago when release rate was high. Contrarily, lower population in August–September should mean their glochidia was released T months ago when release rate was low. For insight about glochidia release timings, we refer the reader to relevant literature (Siddique et al. 2020, Salam et al. 2024)–which are not the scope of this study. But for mussel harvesting purposes, the calendar that we compiled (Fig. 5 and Table 1) should be adequate for a fisheries manager's knowledge.

Identity of Population Distributions of Quadrats

A novel contribution of our work is to show the identity of underlying population distributions of the eight quadrats using Kruskal-Wallis test. Instead of directly averaging the eight quadrats' samples to obtain pond-wide mussel population density, first, we statistically showed that they came from the same distribution. Kruskal-Wallis test is agnostic to the actual distributions–may that be normal or uniform. It shows that the samples came from identical distributions. Such identity is necessary for arithmetically averaging samples.

Effect of Water Quality Parameters

Fig. 6 (A) shows significant negative correlation (Spearman's rank test: S = 563.97, ρ = -0.97, P < 0.001) between variation of mussel population density and depth of pond water. The alignment of data points along the regression line and tightness of its confidence region confirms the strength of correlation. Besides, *L. marginalis* population showed a harmonious decrease with increasing depth. It was highest in April 2023 (D = 4.13 no./m^2, N = 2071 no.) at 1.15 m depth, and lowest in September 2023 (D = 1.97 no./m^2, N = 988 no.) at 2.10 m depth. The depth of the pond varied between 1.15–2.10 m. When the depth was lower, probably more nutrients were present in pond bed creating a favorable atmosphere for more mussels to mature. It could also be related to less rain around April in Bangladesh that allowed more pollutants in the water for mussels to feed on. Such a negative association between lake depth and mussel growth was also reported before (Elgin et al. 2022).

Fig. 6 (B) shows no significant correlation (Spearman's rank test: S = 468.32, ρ = -0.64, P = 0.026) between variation of mussel population density and temperature of pond water. The lack of significance can be realized from the scatter of data points and the expanse of regression line confidence region. *L. marginalis* population was highest in April 2023 (25.75 °C, D = 4.13 no./m^2, N = 2071 no.), and lowest in September 2023 (27.35 °C, D = 1.97 no./m^2, N = 988 no.). The little difference (1.60 °C) between the two extremes may be the reason why this experiment did not observe any significant association between population density and temperature. Moreover, at highest temperature (27.35 °C, September 2023) and lowest temperature (15.67 °C, December 2023) during the year-long study, the population densities were 1.97 and 3.69 no./m^2, respectively. Though these values indicate negative association, population at other temperatures did not support its significance. Given that, monthly temperature did not appear to have any significant effect on mussel population variation. Previous literature studied effects of elevated temperature on juvenile and adult mussels as an interaction of temperature × time (Ganser et al. 2013, Alter et al. 2025) in laboratory settings. But such interaction is impractical for a lentic habitat and particularly for Bangladesh where extreme heat does not continue for more than a couple weeks. This country is blessed with a moderate climate where there is a frequent turnover of hot-warm-rainy days. As a result, mussels do not experience prolonged heat that would reach the level of mass mortality and significantly impact pond population. If anything, glochidia release would be impacted by monthly temperature (Siddique et al. 2020, Salam et al. 2024) that may impact the number of adult mussels T months later. The value of T remains a matter of exploration as glochidia become adults over several months. Meanwhile, one sample each month was probably not enough to characterize the monthly average of temperature in a changing climate. Likely for the above reasons, water temperature did not show any significant association with mussel population.

pH is an important parameter that affects the survival, growth, and reproduction of mussels (Seitz et al. 2023). Acidic media (pH < 7) threatens the assemblage of $CaCO_3$ crystals in mollusk shells (Rajan and Vengatesen 2020). But deposition of $CaCO_3$ is essential for strengthening their calcareous structures. An alkaline media (pH > 7) is necessary for $CaCO_3$ deposition. However, Fig. 6 (C) shows no significant correlation (Spearman's rank test: S = 409.22, ρ = -0.43, P = 0.162) between variation of mussel population density and pH of pond water. The lack of significance can be realized from the scatter of data points and the expanse of regression line confidence region. *L. marginalis* population was highest in April 2023 (D = 4.13 no./m^2, N = 2071 no.) at pH 7.63, and lowest in September 2023 (D = 1.97 no./m^2, N = 988 no.) at pH 7.70. The little pH difference (0.07) may be the reason why this study did not observe any significant association between population density and pH. Moreover, during the year-long study, the pH of the pond varied only between 6.41–7.72. The pragmatic explanation behind such a little variation lies in the culture pond–its pH was well maintained near equilibrium (7.0) to facilitate farming of various fishes. Lime ($CaCO_3$) was regularly applied to this pond to maintain equilibrium pH. Hence, the environment of the culture pond could not show any significant association between variation of pH and mussel population. Nevertheless, for *L. marginalis* farming, an optimum pH of 8.0 should be maintained that was found to maximize its filtration rate (Islam et al. 2020). A similar range (7.72–8.75) was reported as suitable for growth and reproduction of *L. marginalis* (Niogee et al. 2019).

Optimal Harvesting Strategy for Mussels

So far, we have indicated that mussels should be harvested in March–April (D = 4–4.13 no./m^2, N = 2008–2071 no.) and not in August–September (D = 1.97–2.13 no./m^2, N = 988–1067 no.). Our recommendation was corroborated by the shell dimensions measured throughout the study. Samples collected in April were larger (length = 103.46, width = 49.77, height = 23.08 mm) and samples collected in September were smaller (length = 83.79, width = 38.73, height = 17.79 mm).

Conclusion

This work showed the application of quadrat method in estimating freshwater pearl mussel (*Lammellidens marginalis*) population in a culture pond. To the best of our knowledge, this is the first such attempt in Bangladesh. Mussel samples were collected monthly for a whole year (April 2023– March 2024). The estimated population showed peaks in March–April and valleys in August–September. Simultaneously, depth, temperature, and pH of the pond water were recorded. Spearman's rank test showed a strong negative correlation between mussel population and pond depth. The density of mussels was higher in shallow water than deep. Temperature seemed to have no effect on mussel population. Since pH was regularly maintained near equilibrium in the culture pond, it did not show any correlation with mussel population either. All data and codes are released publicly via GitHub to facilitate future research on mussel population dynamics. Overall, this study serves as a fisherman's guide to harvesting *L. marginalis* and maintaining a congenial pond environment for its growth. Future research should investigate the nutrient content in bottom substrate to determine what exactly causes mussel population variation at various depths. Another year of mussel samples could be collected to confirm 2023–2024's yield pattern. Other variables involved in mussel's life cycle: glochidia release timing, availability of host fishes in lentic habitat, etc. should be considered to more precisely explain temporal dynamics in density.

Data Availability

The collected data of mussel samples, water quality parameters, and biometric characteristics of mussel shells are available via GitHub at https://doi.org/10.5281/zenodo.14996734.

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Author Contributions

MMR and BC participated in field sampling and lab work. FF performed all statistical analysis on R, created the GitHub repository, and led the writing of this manuscript. TB provided technical support for data visualization and dissemination. FF, TB, and MJU edited the manuscript.

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