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EFFECTS OF MORPHOMETRIC VARIABLES AND NATURAL MICROBIAL LOAD ON THE SURVIVAL RATE OF LIVE *Mytilus galloprovincialis* DURING POSTHARVEST COLD STORAGE

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Abstract

This study investigated the relationships between morphometric characteristics, microbiological parameters, pH changes, and survival rates of Mediterranean mussels (*Mytilus galloprovincialis*) during post-harvest storage at +4°C. Mussels were categorized into three size groups based on shell length: large (63.92-80.19 mm), medium (48.51-62.63 mm), and small (30.08-47.75 mm). Total heterotrophic aerobic bacteria (THAB), *Enterobacteriaceae*, and *Vibrio* sp. counts were monitored over a 16-day storage period, along with pH changes and survival rates. Results demonstrated significant size-dependent variations in survival, with larger specimens showing superior viability throughout storage. THAB counts increased continuously across all groups, while *Enterobacteriaceae* levels remained relatively stable in medium and small groups. *Vibrio* sp. counts decreased in large specimens but remained stable in other groups. Strong negative correlations were observed between survival rates and both THAB (r: -0.854, $p<0.001$) and *Enterobacteriaceae* (r: -0.777, $p<0.001$) counts. Principal component analysis revealed that the first two components explained 83% of total variance,

with physical dimensions primarily influencing PC1 (56.5%) and yield-related traits, pH, and THAB characterizing PC2 (26.5%). By day 16, medium and small groups reached 0% survival, while large specimens maintained partial survival. These findings suggest that mussel size significantly influences post-harvest survival and should be considered when developing storage protocols. Future research should focus on optimizing storage conditions through combined approaches of temperature control, modified atmospheres, and periodic water exposure, particularly considering morphometric variables.

Keywords: Mediterranean mussel, post-harvest storage, size-dependent survival, microbial dynamics, shelf life, morphometric traits

Introduction

Bivalve mollusks, particularly the Mediterranean mussel (*Mytilus galloprovincialis*), represent a significant sector in global aquaculture, with annual production exceeding 1.9 million tones (FAO, 2022). These organisms are highly valued for their exceptional nutritional profile, including high-quality proteins (approximately 11.9g/100g), essential minerals (particularly zinc and iron), and omega-3 fatty acids (primarily EPA and DHA) (Panayotova et al., 2021; Peycheva et al., 2021; Lopez et al., 2022). However, their filter-feeding nature presents unique challenges for post-harvest quality and food safety, as their tissue composition directly reflects seawater conditions and environmental exposures (Colakoglu et al., 2010, 2011). These organisms can accumulate various risk factors from seawater, including persistent organic pollutants, trace elements, biotoxins, and microbiological agents, potentially affecting both the bivalves themselves and consumers through zoonotic pathways and bioaccumulation (Künili et al., 2021a; Künili et al., 2021b; Balkaya et al., 2023; Künili, 2023; Künili et al., 2023; Çolakoglu et al., 2014, 2020). The post-harvest handling of live mussels involves complex interactions between biological, chemical, and environmental factors that significantly influence product quality and safety. Seawater environmental parameters, which directly influence bacterial communities even within semi-closed coastal structures (Künili & Ateş, 2021), can significantly impact product quality. During storage and transportation, mussels face various stressors that can affect their survival rates and market value. While regulatory frameworks, such as European Union directives (EU, 2005), establish monitoring protocols for major risk factors in production areas, the relationship between mussel size, microbiological dynamics, and survival during storage remains inadequately understood (Künili, 2024). Depuration processes, whether through flow-through, closed-cycle, or semi-closed systems, serve as crucial intervention points for enhancing bivalve mollusk quality and safety (Colakoglu et al., 2014; Künili & Colakoglu, 2019; Künili & Dinc, 2024). However, post-depuration storage conditions, particularly temperature control and bacterial load management, significantly impact product shelf life and safety. Recent research has highlighted the importance of size-dependent metabolic scaling in stress tolerance, suggesting that larger specimens may exhibit enhanced survival capabilities during storage due to lower mass-specific metabolic rates (André et al., 2021; Ibarrola et al., 2022; Benjamin et al., 2023). The microbial ecology of stored mussels presents a critical dimension in post-harvest quality management (Azizan et al., 2022; Künili et al., 2023; Schoinas et al., 2023). Total heterotrophic aerobic bacteria, *Enterobacteriaceae*, and *Vibrio* species populations not only indicate product safety but also serve as predictors of shelf life (Jozić et al., 2017; Rosaria et al., 2023; Odeyemi et al., 2023). Additionally, tissue pH dynamics during storage have emerged as crucial indicators of mussel viability and microbial activity, with pH values above 6.8 often associated with increased bacterial proliferation and decreased product quality (Hirabayasi et al., 2022; Wang et al., 2023).

Climate change impacts on marine environments and increasing consumer awareness of food safety have amplified the importance of understanding these post-harvest dynamics (Dinç et al., 2022; Çolakoğlu et al., 2022). Despite extensive research on individual aspects of mussel storage, a comprehensive understanding of the relationships between morphometric characteristics, microbiological parameters, and survival rates remains elusive. This study addresses this knowledge gap by investigating the interplay between mussel size categories, bacterial population dynamics, pH changes, and survival rates during refrigerated storage at +4°C. Through detailed monitoring and principal component analysis (PCA), we aim to identify key predictors of post-harvest viability and establish improved protocols for maintaining product quality and safety during storage.

Materials and Methods

Materials

Mediterranean mussel (*Mytilus galloprovincialis*) samples were collected at 1–10 m depths by hand during SCUBA dives. A total of 10 kg sample was collected in December 2022 from the coasts of Çanakkale, Türkiye. Samples were firmly packed with wire meshes and transported to the laboratory via an ice–cooled insulated box within 1–2 hours.

Methods

Morphological measurements and meat yield

To determine the effect of morphological properties on the survival time and rate of the samples, as well as to assess their relationship with meat quality during storage, morphometric dimensions were measured. These dimensions included the longest shell length (X-axis), width (Y-axis), height (Z-axis), and total weight, comprising the shell, intravalvular liquid, and meat of the species. The measurements were conducted using a digital vernier caliper (accuracy: 0.01 mm) and an electronic balance (d: 0.01 g, Max: 3100 g, Acculab, ALC-3100). Meat yield (MY) was calculated with the following equation:

$$MY\ (\%) = [(MW / TW) \times 100]$$

All morphometric measurements were performed from the sub-sample specimens from the same batch representing the similar dimensions of the samples saved for main trials to avoid unnecessary prolonged retention without pack and storage conditions that possibly affect the overall results.

Figure 1. *Mytilus galloprovincialis*, samples at different size and morphometry used in this study.

Product Preparation

After measuring the morphometry, mussel samples were divided into three groups according to their lengths and labelled as large (L), medium (M), and small (S) sizes (Figure 1). The grouped

samples were then subjected to packaging that mimicked commercial methods for the evaluation trials, ensuring quality and safety during marketing.

Before the packaging steps, all samples were visually checked and cross-checked through direct contact reflection to ensure that all specimens were alive. Following this, all specimens were wiped with clean paper towels to remove excessive seawater from the shells and any juice from deceased specimens, if present.

For the packaging step, standard methods for marketing live bivalve molluscs were employed. The packaging method involved using stretch polyethylene films for 1-3 kg packing for each group, ensuring that there were no excessive gaps between the specimens.
All prepared products were immediately moved to a +4°C refrigerator with 80% relative humidity for storage. One pack from the stored samples was taken for each examination and subjected to analysis.

Survival Rate
To determine the survival rate, different packages in small portions were prepared as mentioned above. The survival rate was evaluated from each pack, without disturbing the other packs, to mimic the conditions during storage and transportation in a real market. Survival rates were calculated based on the total number of specimens in each package and the total weight measured before the storage began for each pack, following the equation:

$$SR\ (\%) = \frac{\mathrm{TS} - \mathrm{AS}}{\mathrm{TS}}\ x\ 100$$

Where, SR represents the survival rate, TS is the total number of specimens in the package, and AS is the total number of alive specimens in the package.

Each pack was cut open with a sterile knife, and the contents were distributed to a clean and sterilised bench using alcohol and a bunsen burner. After the specimens were distributed, they were touched with clean and sterile pens to assess whether they exhibited a reflex that indicated the shell was closing evenly or if it remained firmly closed. After waiting a few minutes, the specimens were checked again to see if the closing shell reflex, which sometimes indicates deceased specimens, occurred under the storage conditions.

Once all specimens were checked visually and by touch, a comparison between small (<2 cm), intermediate (3-5 cm), and large (>5 cm) live specimens was conducted, and they were directly subjected to microbiological analysis.

Microbiological Analysis
All packaging materials were sanitized with alcohol prior to sample processing. Live specimens were removed from their packages, and their shells were thoroughly cleaned of dirt and debris before being surface-sterilized with alcohol. Shell margins were cleaned using a sterile knife. A combined sample of meat and intervalvular liquid (10 g total) was diluted in 90 ml physiological saline solution (peptone water) and homogenized using a Stomacher 400 Circulator (Seward, UK) for 2 minutes at low speed. The homogenates were then subjected to serial dilutions using physiological saline solution.

For microbiological analysis, samples were first cleaned of sand, mud, and macroorganisms, with shell surfaces sterilized using alcohol-soaked cotton and a sterile knife. Intravalvular fluid

and mussel meat (10 g) were weighted in 90 ml of peptone water containing 0.8% NaCl and 1% bacterial peptone. Following homogenization at 4000 rpm for 4 minutes, samples were serially diluted in peptone water to 10^{-6}, and 1 ml of each dilution was spread on three replicate Petri dishes following the spread plate method (FDA, 1998).

For bacterial identification, Thiosulfate Citrate Bile Sucrose (TCBS) Agar was used to determine *Vibrio* species, with plates incubated at 30°C for 36 hours. Selected colonies were transferred to Marine Agar and incubated at 25°C for 36 hours for purity assessment and preliminary identification. *Enterobacteriaceae* were enumerated using Endo Agar with incubation at 35°C for 24-36 hours. Selected strains were subcultured for purity checks and identification. All PCA cultures were incubated at 25°C for 24 hours, and biochemical tests were performed on purified colonies for species confirmation.

Statistical Analysis

All data in this study were obtained in triplicate. Basic descriptive statistics for morphometric parameters and meat yield across groups (L, M, and S) were calculated. A one-way analysis of variance (ANOVA) was conducted to determine differences between groups, with results expressed as mean ± SD. The Tukey test was utilised for multiple comparisons between groups ($p < 0.05$). Correlations and distances between bacterial levels and factors were assessed using Pearson's correlation analysis and principal component analysis in Minitab 17 (Minitab, LLC, USA). Significance levels for differences between factors were set at 0.01 and 0.05.

Results

In this study, the morphometric characteristics, pH, and microbiological load of Mediterranean mussels were assessed during post-harvest storage at +4°C to determine their influence on mussel survival. Samples were categorized into three size groups based on shell length: large (L), medium (M), and small (S) and their morphometric characteristics are presented in Table 1.

Table 1. Morphometric parameters of *Mytilus galloprovincialis* specimens, including total numbers (N) of large (L), medium (M), and small (S) sizes, with standard deviations (±SD).

		Total Weight (g)	Length (mm)	Width (mm)	Height (mm)	Meat Weight (g)	Shell Weight (g)	Meat Yield (%)
L (N: 83)	Min.	21.17	63.92	21.44	22.78	4.01	15.47	13.40
	Max.	49.37	80.19	40.02	31.03	9.82	21.19	25.51
	Mean	33.83^{b}	69.50^{b}	35.40^{b}	26.45^{b}	6.16^{b}	17.25^{b}	18.58^{b}
	±SD	8.82	4.06	5.18	2.63	1.60	2.12	3.60
M (N: 91)	Min.	12.84	48.51	23.89	17.68	1.62	5.19	9.79
	Max.	25.65	62.63	34.02	25.26	5.66	14.17	27.64
	Mean	18.37^{a}	53.71^{a}	29.58^{a}	20.52^{a}	3.31^{a}	9.00^{a}	18.39^{a}
	±SD	4.02	4.75	2.40	2.38	0.96	2.46	5.23
S (N:122)	Min.	3.46	30.08	18.37	11.32	0.63	1.32	14.65
	Max.	11.07	47.75	26.85	20.97	4.78	6.77	48.19
	Mean	7.57^{c}	39.05^{c}	21.39^{c}	15.42^{c}	1.86^{c}	3.90^{c}	24.79^{c}
	±SD	2.61	5.04	2.35	2.57	1.02	1.30	9.42

The total weight ranges were 21.17-49.37 g for group L, 12.84-25.65 g for group M, and 3.46-11.07 g for group S. Shell lengths ranged from 63.92-80.19 mm, 48.51-62.63 mm, and 30.08-

47.75 mm for groups L, M, and S, respectively. While groups L and M showed similar meat yields of 18%, group S exhibited a significantly higher yield of 24%.

Figure 2 presents the microbiological analysis results and survival rates of the sample groups. Microbiological analyses revealed distinct patterns across size groups during the 16-day storage period. Total heterotrophic aerobic bacteria (THAB) counts increased continuously in all groups, with initial loads of 3.78, 4.08, and 2.77 Log cfu/g for groups L, M, and S, respectively. Group S, despite having the lowest initial THAB load, showed the most substantial increase over time.

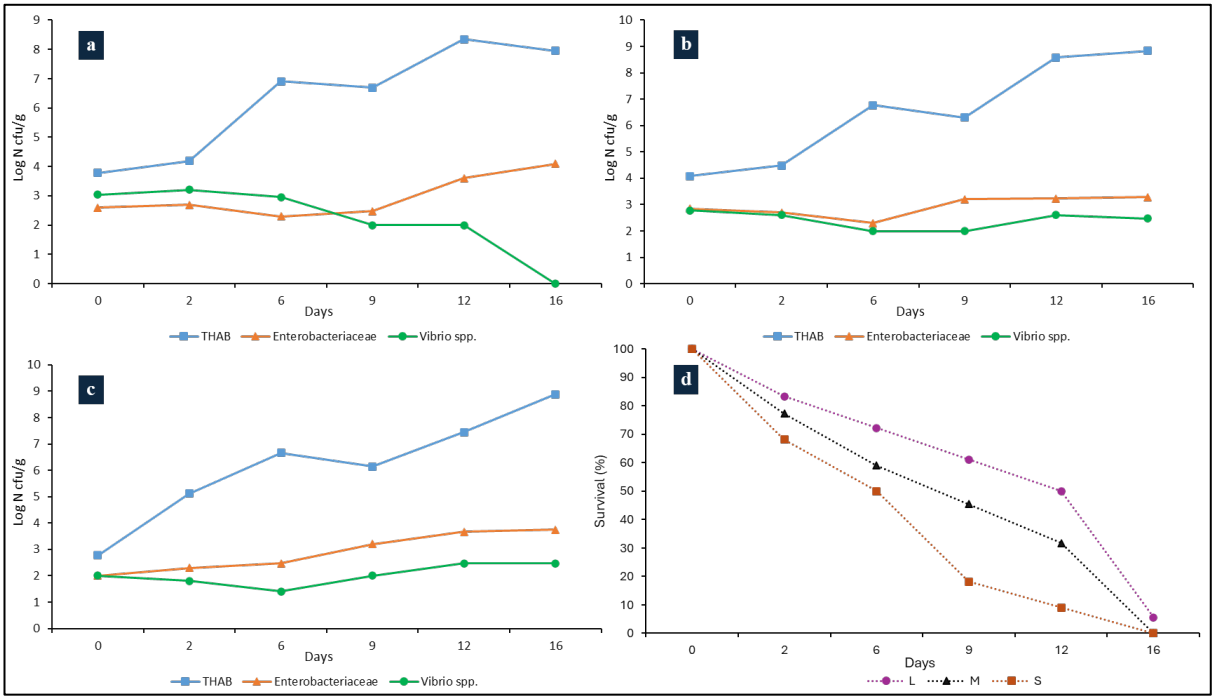

Figure 2. Microbiological analysis results of large (L) size (**a**), medium (M) size (**b**) and small (S) size (**c**) of *Mytilus galloprovincialis*, and their survival rates by sizes during the refrigerated (+4°C) storage (**d**).

Enterobacteriaceae levels exhibited minimal increase in group L and remained near baseline in groups M and S, with initial loads of 2.60, 2.84, and 2.00 Log cfu/g, respectively. *Vibrio* sp. counts (initial loads: 3.04, 2.77, and 2.00 Log cfu/g for L, M, and S groups) remained stable in groups M and S, while decreasing in group L to undetectable levels by day 16. During the first two days, only THAB levels showed significant increases across all groups, while *Enterobacteriaceae* and *Vibrio* levels remained stable.

Survival rates correlated with bacterial load changes, particularly affecting group S, which showed marked decline from early storage stages. By day 16, groups M and S reached 0% survival, while group L maintained some living specimens.

Among the parameters affecting the living conditions of organisms, pH is crucial for the survival of both the host organism, such as mussels, and the microorganisms present in the host when alive or as a food product post-mortem. The pH shifts in mussel samples by group are summarized in Figure 3.

The pH values ranged from 6.68-7.05, 6.68-7.46, and 6.68-7.46 for groups L, M, and S, respectively. A significant pH decrease was observed across all groups by day nine, with group L showing significantly lower pH than groups M and S after 16 days of storage.

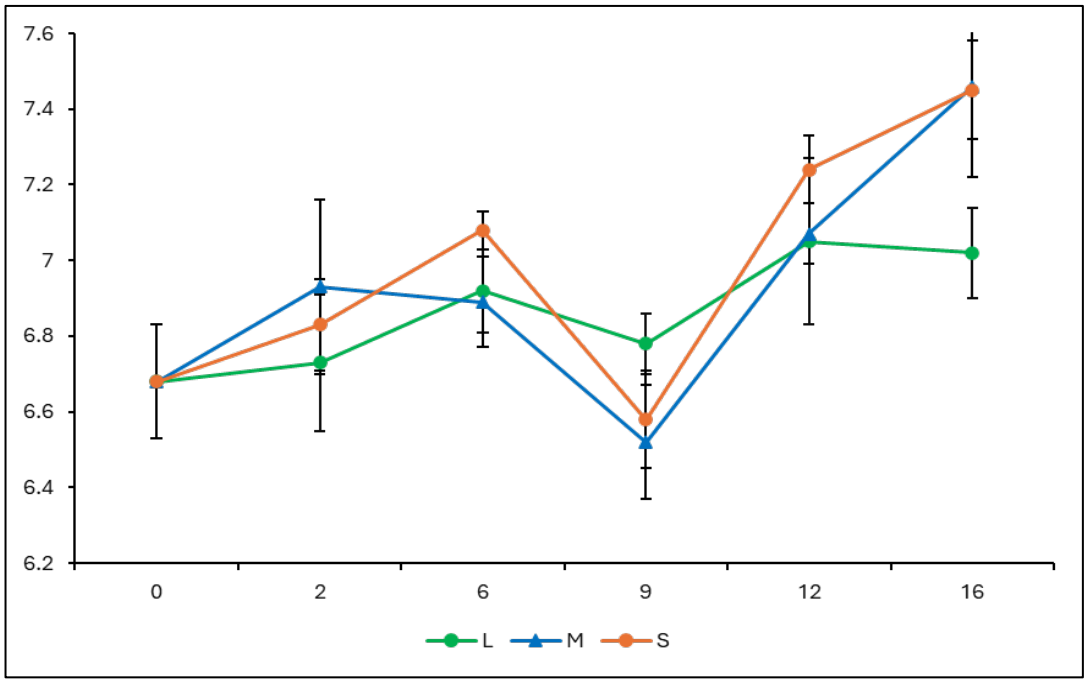

Figure 3. pH shifts of live *Mytilus galloprovincialis* specimens by large (L), medium(M) and small (S) sizes during refrigerated (+4ºC) storage.

To determine the relationships between groups, morphometric characteristics, pH, microbiological changes, and survival rates, Pearson correlation was applied to all variables. Correlation distance between the variables affecting survival rate was presented Figure 4.

	(L-M-S)	TW	Lenght (x)	Width (y)	Height (z)	MW	SW	MY	pH	THAB	Entr.	Vib
TW	-0.987**											
Lenght (x)	-0.993**	0.982**										
Width (y)	-0.994**	0.973**	0.986**									
Height (z)	-0.985**	0.977**	0.974**	0.987**								
MW	-0.977**	0.972**	0.963**	0.970**	0.989**							
SW	-0.991**	0.988**	0.984**	0.983**	0.994**	0.992**						
MY	-0.859**	-0.811**	-0.872**	-0.863**	-0.784**	-0.744**	-0.792**					
pH	0.175	-0.168	-0.18	-0.206	-0.243	-0.215	-0.204	0.012				
THAB	-0.031	0.054	0.04	0.004	-0.075	-0.055	-0.021	-0.197	0.733**			
Entr.	-0.044	0.057	0.041	-0.012	-0.073	-0.042	-0.006	-0.191	0.482*	0.664**		
Vib.	-0.101	-0.003	0.115	0.125	0.14	0.142	0.112	-0.111	-0.056	-0.276	-0.307	
Survival	-0.257	0.234	0.26	0.294	0.363	0.32	0.303	-0.063	-0.666**	-0.854**	-0.777**	0.394

L-M-S: Groups by sizes; large(L), medium(M), and small(S), TW: total weight, MW: meat weight, SW: shell weight, MY: Meat yield, Vib: *Vibrio* sp., Surv: Survival Rate, THAB: total heterotrophic aerobic bacteria, Entr: *Enterobacteriaceae*

Figure 4. Correlation heatmap constructed using Pearson's correlation coefficient, illustrating significant relationships ($p < 0.05$*; $p < 0.01$**) among size groups (L-M-S), morphometric parameters, pH, microbiological load, and survival rates of live *Mytilus galloprovincialis* during refrigerated storage at +4ºC.

Pearson correlation analysis revealed significant relationships between all measured parameters. Size groups showed significant correlations with morphological parameters. pH demonstrated strong positive correlations with THAB (r: 0.733, $p<0.01$) and *Enterobacteriaceae* (r: 0.482, $p<0.05$) counts, while exhibiting a strong negative correlation with survival rate (r: -0.666, $p<0.001$). Similarly, survival rate showed strong negative correlations with THAB (r: -0.854, $p<0.001$) and *Enterobacteriaceae* (r: -0.777, $p<0.001$) counts. Notably, *Vibrio* sp. counts, which generally declined during storage, showed no significant correlations with any variables, including survival rate.

Principal component analysis of variables that showing each variable effect on the survival rate with a power illustrated in the Figure 5.

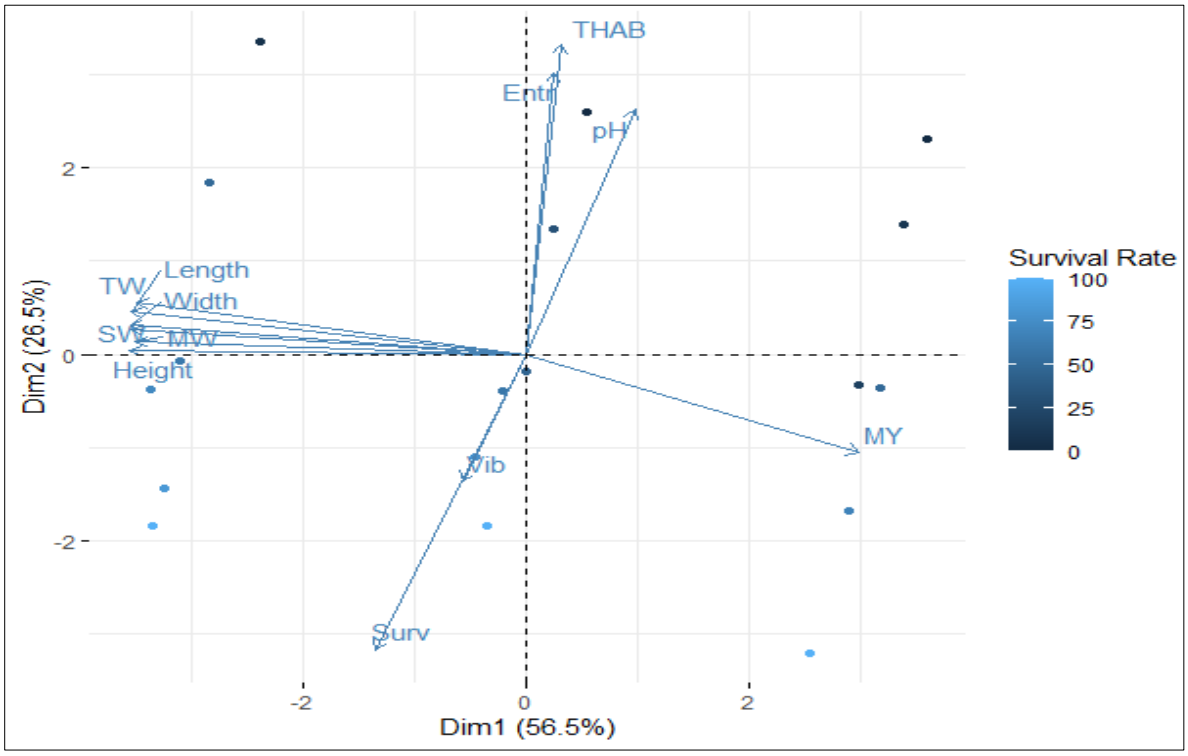

Figure 5. PCA biplot illustrates the relationship between variables and observations. Arrows indicate original variables, with length and direction representing loadings on PC1 (Dim1: x-axis) and PC2 (Dim2: y-axis). Individual observations are shown as points, positioned according to their scores on the first two components. MY: Meat yield, Vib: *Vibrio* sp., Surv: Survival Rate, TW: total weight, SW: shell weight, MW: meat weight, THAB: total heterotrophic aerobic bacteria Entr: E*nterobacteriaceae*.

Principal component analysis (PCA) revealed that the first two principal components (PC1 and PC2) explained 83% of the total variance (PC1: 56.5%, PC2: 26.5%). The survival rate exhibited opposite effects to THAB, *Enterobacteriaceae*, and pH, as indicated by their contrasting positions in the PCA biplot. Meat yield emerged as a distinct variable, opposing morphometric characteristics while aligning with survival and *Vibrio* sp. in PC2, and with *Enterobacteriaceae*, THAB, and pH in PC1. The PCA results demonstrated that PC1 was primarily influenced by physical dimensions (total weight, length, width, height) and shell characteristics (MW and SW), while PC2 was characterized by yield-related traits, pH, and THAB. The hierarchical clustered heatmap further confirmed these relationships, highlighting

size-related factors as primary drivers of variation, followed by pH, microbiological, and yield-related characteristics (Figure 6).

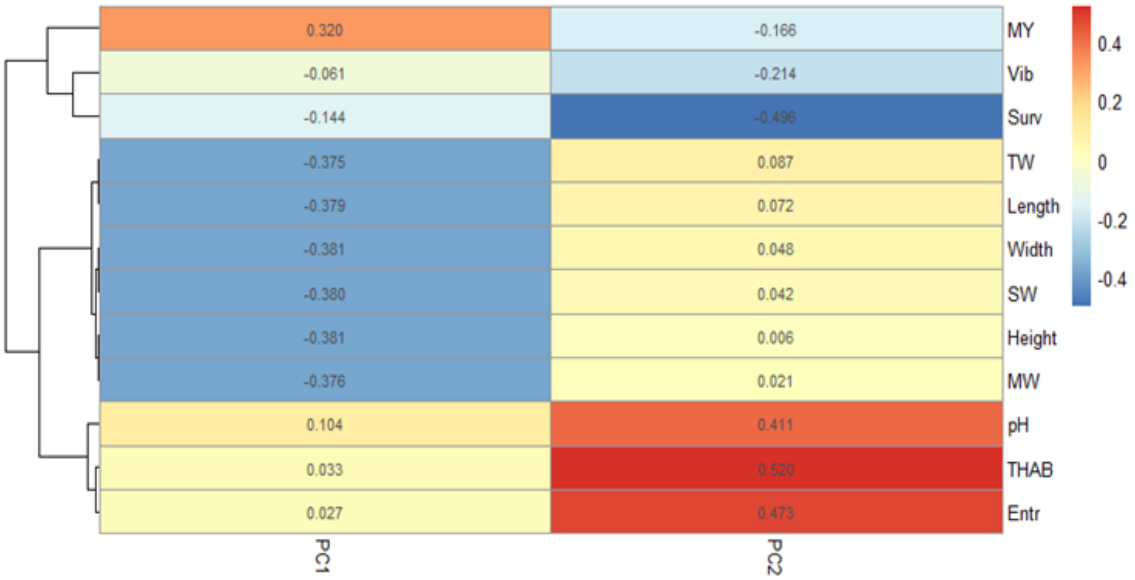

Figure 6. Hierarchical cluster heatmap showing variable loadings across principal components (PCs). The color intensity indicates loading strength and direction, with red representing positive loadings and blue indicating negative loadings. The dendrogram on the left illustrates the hierarchical clustering of variables based on their loading patterns, grouping those with similar contributions. Each row corresponds to a variable, and each column corresponds to a principal component, with the percentage of explained variance indicated for each PC, highlighting the underlying relationships in the data. MY: Meat yield, Vib: *Vibrio*, Surv: Survival Rate, TW: total weight, SW: shell weight, MW: meat weight, THAB: total heterotrophic aerobic bacteria Entr: *Enterobacteriaceae*.

The hierarchical clustered heatmap provided additional insights into the relationships between variables through their loading patterns across principal components. The heatmap visualization revealed distinct clustering patterns, with variables grouped based on their contribution similarities. The color gradient, ranging from red (positive loadings) to blue (negative loadings), illustrated the strength and direction of variable contributions to each principal component. This visualization demonstrated clear variable clusters: morphometric parameters (length, width, height, and total weight) formed one distinct cluster, showing strong positive loadings (red) in PC1, while microbiological parameters (THAB, *Enterobacteriaceae*) and pH formed another cluster with moderate to strong positive loadings in both PC1 and PC2.

The PCA results demonstrated that PC1 was primarily influenced by physical dimensions (total weight, length, width, height) and shell characteristics (MW and SW), while PC2 was characterized by yield-related traits, pH, and THAB. The dendrogram on the left side of the heatmap further supported these relationships by hierarchically organizing variables based on their loading patterns, effectively grouping those with similar contributions to the principal components. This hierarchical organization revealed that size-related parameters formed the most cohesive cluster, followed by microbiological parameters, while meat yield and survival rate showed more distinct patterns, appearing as separate branches in the dendrogram.

These multivariate analyses collectively highlighted size-related factors as primary drivers of variation, followed by pH, microbiological, and yield-related characteristics. The hierarchical clustering pattern provided additional evidence for the complex interactions between physical, chemical, and microbiological parameters in determining mussel survival during storage.

Discussion

The findings of this study regarding mussel mortality and microbial growth during storage align with several previous investigations while also revealing some notable differences. Our results for total heterotrophic aerobic bacteria (THAB) were slightly higher than those reported by Bernárdez & Pastoriza (2013), who found total viable bacteria counts ranging from 3.85 to 5.24 Log cfu/ml after 9 days of storage. However, the mortality rates observed in our study for groups M and L (54.55% and 48.99%, respectively) closely corresponded to their findings of approximately 49% mortality by day 9, suggesting similar patterns of deterioration despite differences in bacterial loads. Differences may be originated by the morphometric characteristic including meat yields, which is used effienctly to understand of bivalve mollusc biological responses both morphometric and environmental variables (Çolakoğlu et al., 2024).

The importance of storage temperature and oxygen conditions in maintaining mussel quality has been consistently demonstrated across multiple studies. Wang et al. (2023) identified 5°C as an optimal storage temperature for maintaining mussel quality for up to 7 days, based on comprehensive analysis of mortality rates, enzymatic activity, and bacterial parameters. This finding provides context for our storage conditions and suggests potential optimization strategies for future research. Barrento et al. (2013) were demonstrated the oxygen demand and ammonia excretion in live mussels (*Mytilus edulis*) with subjecting mussel out of the water and reimmersing at different temperatures and post-harvest conditions to establish optimal storage parameters for aquaculture. Similarly, Bernárdez & Pastoriza (2013) demonstrated that oxygen-rich packaging combined with storage at 2°C resulted in lower mortality rates compared to reduced oxygen atmospheres and higher storage temperatures.

The temporal pattern of mortality in our study showed significant alignment with recent findings by Suplicy et al. (2023), who observed a marked decrease in survival rates after two days of storage at 4°C. This consistent pattern across studies suggests a critical timepoint for mussel viability under refrigerated conditions, regardless of specific storage methods. However, more recent research by Tuckey et al. (2023) has demonstrated that alternative storage methods, such as periodic recirculated seawater immersion, can achieve notably lower mortality rates (2.4%) over extended storage periods, indicating potential areas for improvement in current storage protocols.

Regarding microbial dynamics, our findings can be contextualized within the broader literature on bacterial control methods. For example, passing from refrigerated temperature to frozen storage resulting significant decrases in seafood by storage time (Chakma et al., 2022). In addition to temperature and storage time, improvement packing can be another important factor affecting the quality. Rey et al. (2012) demonstrated that slurry ice packaging systems effectively reduced microbial loads in various shellfish, including mussels, compared to conventional methods. Additionally, modified atmosphere packaging (MAP) and vacuum packaging have shown promise in controlling microbial growth, particularly for specific bacterial groups such as Pseudomonas spp. and H_2S-producing bacteria (Goulas et al., 2005).

The pH changes observed in our study differed notably from those reported by Ratnawati et al. (2023), who observed a gradual decrease in pH from 6.3-6.6 to 5.7-6.0 over 16 days of storage. This divergence suggests that different packaging atmospheres and storage conditions may significantly influence biochemical changes during storage, warranting further investigation into the relationship between storage parameters and pH stability.

Recent research has also highlighted the importance of considering pathogenic bacteria in storage protocols. Nuñal et al. (2023) demonstrated that while *E. coli* levels in mussels typically decrease within 6 hours post-harvest, other pathogens like *Vibrio parahaemolyticus* may gradually increase over time. These findings emphasize the need for comprehensive microbiological monitoring during storage and suggest that different bacterial species may respond differently to storage conditions.

The seasonal variation in mussel quality and storage response, as noted by Tuckey et al. (2023) and demonstrated in practice by Theodorou et al. (2019), adds another layer of complexity to storage optimization. Besides, they also revealed that mussels can be successfully re-immersed in farm seawater for extended periods, even at temperatures approaching 28°C, suggests that traditional storage parameters may need to be adjusted based on seasonal conditions and local practices.

These findings collectively suggest that while current storage methods can maintain mussel quality for limited periods, there is significant potential for optimization through the integration of various approaches, such as temperature control, modified atmospheres, and periodic water immersion. Future research should focus on combining these methods to develop more effective storage protocols that can extend shelf life while maintaining product quality and safety.

Conclusion

This study revealed significant relationships between morphometric characteristics, microbiological parameters, and survival rates of Mediterranean mussels during refrigerated storage at +4°C. Key findings demonstrated that larger mussels exhibited superior survival rates throughout the storage period, while small-sized specimens showed rapid decline despite their higher meat yield of 24%. Microbiological analyses revealed continuous increases in total heterotrophic aerobic bacteria across all size groups, with strong negative correlations between survival rates and bacterial counts (THAB: r: -0.854; *Enterobacteriaceae*: r: -0.777). Furthermore, pH demonstrated significant correlations with both bacterial growth and survival rates, with large specimens showing notably lower pH values by the end of the storage period. While this research provides valuable insights into mussel storage dynamics, several limitations should be considered in future investigations. The study was restricted to a single storage temperature and conventional storage conditions, without exploring modified atmosphere packaging or periodic water immersion techniques. Additionally, the scope of microbial analysis was limited to selected bacterial groups, leaving room for more comprehensive pathogenic studies. Future research should focus on developing integrated storage solutions that combine temperature control, atmospheric modification, and periodic water exposure, while considering seasonal variations in mussel quality. Such investigations would benefit from including broader microbial analyses and enzymatic activity measurements to develop more effective storage protocols for maintaining mussel quality and safety during post-harvest storage.

Ethical approval
As the studied material is invertebrate, the ethical approval is not necessary for this study.

Informed consent
Not available.

Data availability statement
The authors declare that data can be provided by the corresponding author upon reasonable request.

Conflicts of interest
There is no conflict of interest for publishing this study.
The corresponding author is responsible on behalf of all authors' declaration.

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Contribution of authors
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