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Research Article

The Effects of Phytoplankton Concentration, Size of Mussel and Water Temperature on Feed Consumption and Filtration Rate of the Mediterranean Mussel (*Mytilus galloprovincialis* Lmk)

İbrahim OKUMUŞ, Nadir BAŞÇINAR, Mükerrem ÖZKAN
KTU Faculty of Marine Sciences, Department of Fisheries
61530 Çamburnu, Trabzon - TURKEY

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Abstract: The effects of phytoplankton concentration, size and water temperature on the feeding rate of the Mediterranean mussel (*Mytilus galloprovincialis*) from the Black Sea were investigated in the laboratory within a static system. Overall consumption (total cell/h) was significantly higher at the highest phytoplankton concentrations than at the lowest concentration ($p<0.05$), while the consumption and filtration rates declined from 0.62 to 0.23 l/h per mussel with increasing concentrations ($p<0.001$). Mussel size did not have a clear effect on total cell consumption, but filtration rates seemed to increase with mussel size ($p<0.05$). Although the consumption and filtration rates were slightly higher (81.1% and 0.83 l/h) at 18°C than those (70.6% and 0.62 l/h) at 22°C, the differences between the temperatures were not significant. The significance of the findings for coastal aquaculture and eutrophication was evaluated.

Key Words: *Mytilus galloprovincialis*, food consumption, filtration rate, phytoplankton concentration, mussel size, temperature

Akdeniz Midyesi (*Mytilus galloprovincialis* Lmk)'nin Besin Tüketimi ve Filtrasyonu Oranı Üzerine Fitoplankton Konsantrasyonu, Büyüklük ve Su Sıcaklığının Etkileri

Özet: Akdeniz midyesinin (*Mytilus galloprovincialis*) filtrasyon oranı üzerine fitoplankton konsantrasyonu, büyüklük ve su sıcaklığının etkileri incelenmiştir. Çalışma Karadeniz'den toplanan ergin midyelerle laboratuvarda statik ortamda yürütülmüştür. Toplam tüketim (toplam hücre/h), yüksek fitoplankton konsantrasyonunda önemli derecede yüksek ($P<0.05$) bulunmasına karşın, tüketim ve filtrasyon oranları, konsantrasyonun artmasıyla 0.62 l/h'den 0.23 l/h'e düşmüştür ($P<0.001$). Midye büyüklüğü, toplam hücre tüketimi üzerinde açık bir etki göstermemiş, fakat filtrasyon oranı midye büyüklüğü ile artmıştır ($P<0.05$). Tüketim ve filtrasyon oranları 22 °C'ye (%70.6 ve 0.62 l/h) oranla 18 °C'de (%81.1 ve 0.83 l/h) biraz yüksek olmasına rağmen, farklılık önemsiz bulunmuştur. Bulguların bivalve yetiştiriciliği ve ötrifikasyon kontrolü açısından önemi irdelenmiştir.

Anahtar Sözcükler: *Mytilus galloprovincialis*, besin tüketimi, filtrasyon oranı, fitoplankton konsantrasyonu, midye büyüklüğü, sıcaklık

Introduction

Mussels, as sessile and filter feeding coastal marine animals, play a significant role not only as a cheap and valuable seafood but also as a popular "bio-indicator" species in studies investigating environmental degradations, such as pollution, hypernutrification and eutrophication including toxic algae blooms (1-4). The production of mussels within aquaculture systems reached an annual value of 1.2 million tons corresponding to a economic value of over US$ 500 million (5). Unfortunately, aquaculture production seems to be leveling off due to the diminishing number of coastal sites suitable for the farming of these filter feeding organisms. Thus, there is a need for increasing the production efficiency in existing culture operations. Recently, the use of bivalve filter feeders in controlling coastal eutrophication and increasing primary production by processing sediment and cycling the nutrients has drawn considerable attention (3,6) and they have acquired an importance which surpasses their economic significance as a source of food.

Mussels, particularly *Mytilus*, are one of the most cosmopolitan marine organisms occurring in estuarine and ocean habitats. Thus, they show large differences in growth and production within and between species and populations of the same species, as well as over seasonal time scales in temperate latitudes (7,8). Among the major factors influencing feeding and growth, water temperature and food availability have been widely acknowledged (9-13). However, these factors may show

within and among site (combination of all biotic and abiotic factors), species, stock (genotype) specific or temporal (seasonal) variations (4,11,14,15). These studies showed that both food quality and quantity are the major factors influencing growth, and mussels exhibit a considerable ability for physiological compensation to variable food quality and quantity under variable environmental conditions. Thus, there seem to be justifiable needs for evaluating the wide interaction ranges of these factors.

The present paper submits the results of a preliminary study conducted on the effects of food concentration, mussel size, and water temperature on the feeding rate of the Mediterrannean mussel (*Mytilus galloprovincialis*) and evaluates the significance of the findings for bivalve aquaculture and eutrophication in coastal waters.

Materials and Methods

The experiments were carried out in the laboratory of KTU Faculty of Marine Sciences, Trabzon, in April 2000. Adult mussels (>50 mm shell length) of *Mytilus galloprovincialis* (Lmk.) were collected from walkways of marine fish cages in Yomra fishing port. They were carefully detached from the settlement surfaces, cleaned of fouling organisms and transferred to the faculty in a cooling box. On arrival they were placed in a fibreglass tank with a volume of around 300 l. The seawater in the tank was aerated continuously and changed once every two days. Water temperature in the tank was similar to that found in the sampling area. The mussels were fed twice daily with laboratory cultured phytoplankton (*Nannochloropsis oculata*; class: Eustigmatophyceae). This micro-alga was chosen because it was readily available from the turbot hatchery development project at the Central Fisheries Research Institute, its cell size (5 µm) is suitable for mussels (16), and it is one of the micro-algae utilized in bivalve hatcheries. Stock culture of the *Nannochloropsis* was maintained in 20-ml test tubes and semi-continuous mass-culture was conducted at around 16ºC in 5 l jars filled with filtered and UV sterilized brackish water. The culture unit was enriched with F-media and aerated continuously.

A simple experimental static (closed) system consisting of experimental chambers or flasks (500 ml) was set up to determine the filtration and consumption rates. Individual mussels were placed in experimental chambers after adjusting the desired water temperature and phytoplankton cell concentrations. After all mussels started feeding, phytoplankton concentrations were readjusted and water samples of around one drop or 0.03 ml from each chamber were collected every 15 min for 60 min. Before sampling the suspension in the chamber was mixed thoroughly without disturbing the mussel. Cell concentrations of *Nannochloropsis* were counted directly under a microscope (objective 40x), using a haematocytometer after cleaning the slide and cover-glass.

The effects of following variables on filtration rate and consumption were studied:

i) phytoplankton concentrations: low ($1280x10^3$ cell/ml), medium ($2256x10^3$ cell/ml) and high ($3016x10^3$ cell/ml); ii) mussel size: small (51.1 mm, 14.0 g), medium (63.2±1.1 mm, 26.2±5.03 g) and large (70.5 mm, 35.3 g); iii) water temperature: 18°C and 22°C.

The effects of phytoplankton concentrations and water temperatures were tested on medium size (63.2±1.1 mm, 26.2±5.03 g) mussels, while for size and temperature tests $1016x10^3$ cell/ml density was employed.

Consumption was estimated from reductions in cell numbers during the course of the tests, while filtration rate (FR), the volume of water filtered of particles (cells) per hour, was estimated by following the exponential decline in phytoplankton cell numbers (16):

$$FR\ (l/h) = V \times \log_e (Co/Ct)/t$$

where V: volume suspension in experimental chamber, Co: initial cell concentration, Ct: cell concentration at time, t.

All data analysis and statistical testing were carried out using the Minitab (8.1) statistical program. The mean and standard deviation (±s.d.) were calculated for all variables and one-way analysis of variance (ANOVA) was used to test for differences among variables. When significant differences ($P<0.05$) were found, a multiple comparison test (Tukey) was used to determine the different group(s).

Results

Mussels started feeding between 2 and 8 min, while the first faeces production appeared between 3 and 10 min after placing the animals into experimental chambers.

Thus, the time the last experimental mussel started feeding was taken as the beginning of the test.

Changes in cell concentrations and absolute consumption are shown in Figures 1 and 2, while filtration rates, and overall consumption values and rates are presented in the Table. Cell concentrations declined from $1280x10^3$, $2256x10^3$ and $3016x10^3$ cell/ml to $376x10^3$, $800x10^3$ and $1892x10^3$ cell/ml, respectively (Figure 1). Both consumption and filtration rates in all concentrations were highest during the first quarter of the trial, but stayed almost steady during the later three quarters (Figure 2). Overall consumption was significantly higher in the highest two concentrations, namely $2256x10^3$ and $3016x10^3$ cell/ml, than the lowest concentration over the 60 min ($p<0.05$), while the consumption rates exhibited a negative correlation ($r=0.91$) with increasing cell concentrations (Table). Filtration rates varied from 0.23 to 0.62 l/h per mussel and, similar to consumption rates, they also declined with increasing concentrations and exhibited significant differences between the groups ($p<0.001$).

This trial ended up with a single replicate due to gamete productions in one of the replicates. Cell concentrations were reduced from $1016x10^3$ to $264x10^3$, $136x10^3$ and $48x10^3$ cell/ml (Figures 3 and 4), which corresponds to an overall consumption of $440x10^6$, $376x10^6$ and $484x10^6$ cells by small (51.1 mm), medium size (64.3 mm) and large (70.5 mm) mussels, respectively. Concerning the total consumption, no significant differences were observed between the mussels. Filtration rates varied between 0.67 and 1.53 l/h, and seemed to increase with mussel size ($p<0.05$) (Table).

Medium size mussels (63.2±1.1 mm) reduced cell densities from $1016x10^3$ to $192x10^3$ cell/ml at 18°C and $1280x10^3$ to $376x10^3$ cell/ml at 20°C (Figures 5 and 6), and consumed 412±42.43 and 452±16.97 cell/h. The consumption and filtration rates were slightly higher (81.1% and 0.83 l/h) at 18°C than those (70.6% and 0.62 l/h) at 22°C (Table). None of the examined variables showed significant variations with water temperature.

Discussion

Laboratory measurements showed that phytoplankton concentration has a significant effect on the feed consumption and filtration rates of Mediterranean mussels. Overall consumption increased significantly with increasing plankton concentration, while the filtration rates declined. Filtration rates of phytoplankton cells or food particles by bivalve filter-feeders are dependent on particle or cell size, food quantity and quality, the size of the organism and environmental factors (15,17). Both field and laboratory studies revealed that below the pseudofaeces threshold ($0.5\text{-}2.0x10^7$ cells/l) mussels can retain all particles above 4 µm at close to 100% efficiency and at about 50% for cells at 2 µm (16). The filtration rates estimated in the

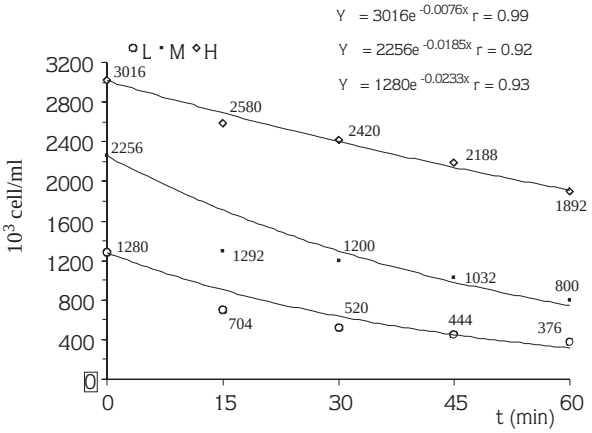

Figure 1. Declines in different initial cell concentrations (L: $1280x10^3$, M: $2256x10^3$ and H: $3016x10^3$ cell/ml) as a result of grazing by mussels (63.2±1.1 mm).

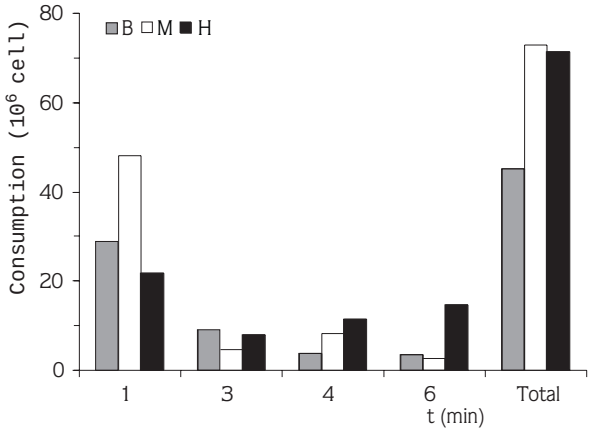

Figure 2. Absoulte consumption of mussels (63.2±1.1 mm) during sampling intervals and entire trial period in different initial concentrations (B: $1280x10^3$, M: $2256x10^3$ and H: $3016x10^3$ cell/ml).

Table. The effects of mussel size, phytoplankton concentrations and water temperatures on filtration rates (l/h) and total *Nannochloropsis oculata* cell consumption under laboratory conditions. Note: different superscript letters (a, b, c) indicate significant differences between the treatments.

t (min)	Concentration (x10^3 cell/ml)[1]			Mussel size[2]			Temperature (°C)[3]	
	1280	2256	3016	S	M	B	18	22
0-15	1.20±0.059^a	1.12.±0.013^a	0.31±0.028^b	1.25	1.19	1.90	1.34±0.057	1.20±0.057
15-30	0.61±0.000^a	0.35±0.000^b	0.13±0.047^c	0.61	0.84	0.31	1.10±0.092	0.61±0.000
30-45	0.32±0.004^a	0.30±0.021^a	0.20±0.043^b	0.89	0.13	3.32	0.17±0.099	0.32±0.007
45-60	0.33±0.000^a	0.51±0.037^b	0.29±0.032^c	1.27	0.53	0.58	0.72±0.099	0.33±0.007
0-60	0.62±0.41^a	0.55±0.40^a	0.23±0.083^b	1.01^{ab}	0.67^a	1.53^b	0.83±0.44	0.62±0.36
Total consumption (10^6 cell):								
0-60	452±16.97^a (70.6%)	728±59.40^b (64.5%)	714±161.22^b (47.5%)	440 (86.6%)	376 (74.0%)	484 (92.3%)	412±42.43 (81.1%)	452±16.97 (70.6%)

[1]: 63.2±1.1 mm mussels at 18°C

[2]: 1016x10^3 cell/ml concentration at 18°C

[3]: 63.2±1.1 mm mussels in 1016x10^3 cell/ml (18°C) and 1280x10^3 cell/ml (22°C)

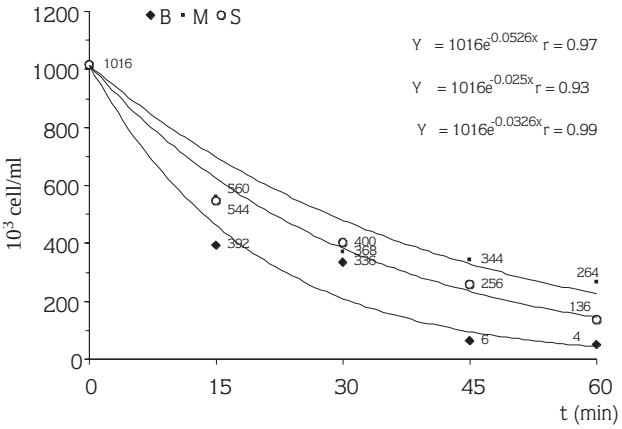

Figure 3. Reductions in initial cell concentration of 1016x10^3 cell/ml as a result of grazing by mussels in different sizes: Big (B): 70.5 mm; Medium (M): 64.3 mm and Small (S): 51.1 mm).

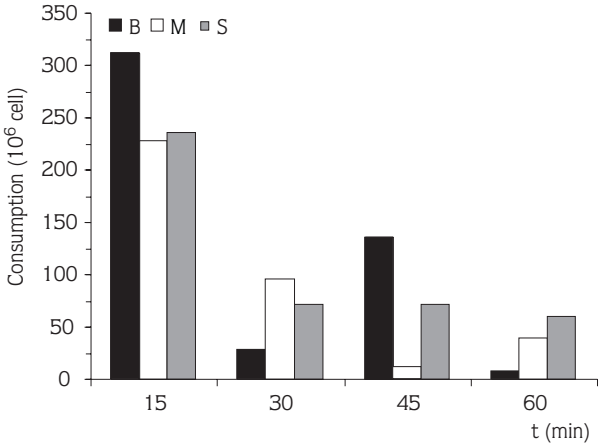

Figure 4. Absolute cell consumption values of different sized (B: 70.5, M: 64.3 and S: 51.1 mm) mussels at initial concentration of 1016x10^3 cell/ml.

current work were in the lower part of the range usually reported for mussels (12,13,15,16,18). This was mainly the result differences in food type and concentrations. Particularly those filtration values measured in the field where suspended material also consists of substantial amount of detritus and inorganic material can be quite high. Various factors controlling the filtration rate and different sets of experimental conditions make it difficult to compare the absolute filtration rates. Instead, comparison of patterns can be more realistic. In general, filtration rates of mussels are highest at low cell concentrations and decline at high concentrations (7,13,18). For example, Newell and Shumway (16) report that the filtration rate is low at <5x10^6 cells/l, maximum between 10-20x10^6 cells/l and declining above 20x10^6 cells/l. In particular, in laboratory experiments with pure algal culture, mussels have been observed to regulate their filtration rate in response to food concentrations. When the ingestion rate reaches its maximum, filtration rates are reduced. This decline in the filtration rate is not due to a reduction in the rate of food capture or consumption, but it is a consequence of the

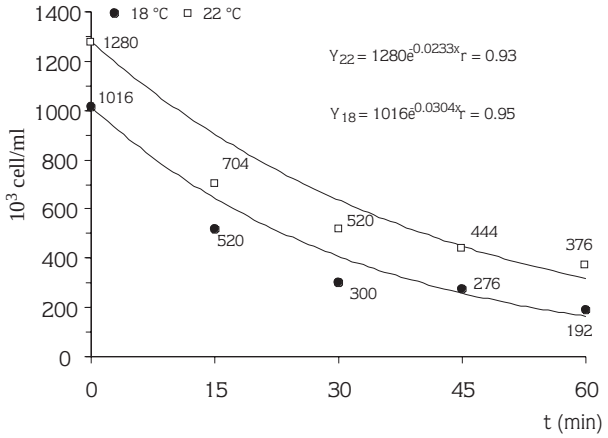

Figure 5. Removal of phytoplankton cells by mussels (63.2±1.1 mm) at 18 and 22°C.

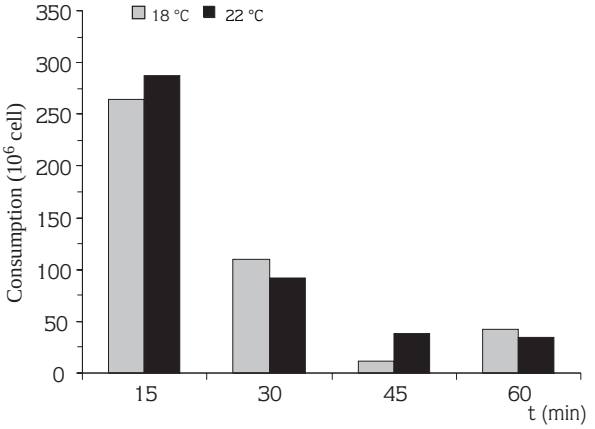

Figure 6. Absolute cell consumption values of medium size (63.2±1.1 mm) mussels at two temperatures.

increase in the rejection rate from labial palps. This results in a relatively constant consumption rate over a wide range of food concentrations (17,18). The results of the present work agree with this pattern. It means that mussels can easily meet their feed requirements by filtering less water and spending less energy in high food concentrations and vice versa in low concentrations.

Mussel size did not have a clear effect on total cell consumption, but filtration rates seemed to increase with mussel size. This was a expected result since the larger mussels have a large gill surface and thus a large food capturing potential.

The water temperatures tested did not have a significant effect on consumption and filtration rates, but values at 18ºC were slightly higher than those estimated at 22ºC. Because mussels are poikilothermic, their metabolic rates increase with temperature. Thus, water temperature certainly plays an important role in the growth of marine mussels through feeding activity and physiological energetics (7,10,11,13,17). In general, rates of consumption and growth increase with temperature up to an optimum and then decline at higher temperatures. The optimum temperature range for mussels is 16-25ºC, but this range varies throughout the geographical range, e.g. it is higher for mussels originating from lower latitude populations, for instance 20-25ºC for *M. galloprovicialis* from Arcachon, France (19). Walne (20) reported a gradual increase in the filtration rate with temperature, but the highest temperature tested was 20º, while Denis et al. (13) did not observe any differences in filtration rates between 20 and 26ºC. The present study was performed in a spring month when the seawater temperature was around 10-13ºC. This previously adapted water temperature might be the reason for higher filtration rates of mussels at 18ºC.

These results may emphasize the importance of the food for the growth and production of mussels, and also in exploiting the mussel culture not only for seafood production, but also ecological manipulations. Mussels are herbivorous filter feeders and play the same role as sheep and cows cultivated in terrestrial meadows. Their culture is an extensive system or ecological manipulation which requires no artificial feeding but natural suspended organic material. This means, in contrast to finfish aquaculture, which add waste and nutrients to environment, that bivalve culture is the net remover of suspended organic particulates (8). They certainly play an important role in coastal waters by the filtration of large quantities of organic suspended material, and the reduction and potential local depletion of the phytoplankton concentration. Therefore, there is a serious risk of over-exploitation of the coastal ecosystems with modern suspended culture technology as already experienced in areas such as Galicia (Spain), Wadden Sea (Netherlands) and Killary Harbour (Ireland) (8,15, 21). For example, according to Cabanas et al. (22), 30% of the carbon, 42% of the nitrogen, and 60% of the chlorophyll contained in the suspended organic matter present in the water flowing through mussel culture rafts is ingested by the mussels. However, mussels also cause the formation and remineralization of biodeposits and the

release of nutrients to the water column (23). These impacts normally extend to areas outside the natural beds or culture sites and the filtration pressure may act as a stabilizer of natural eutrophication control by limiting the phytoplankton biomass (1,3,6). These facts show that the sustainability of mussel culture certainly depends on the carrying capacity of the local ecosystem, and site selection is clearly very important, especially in relation to environmental pollution and primary production. Thus, laboratory and field studies related to feeding ecology bivalves under various environmental conditions can make a major contribution to the site-selection for culture, management of natural stocks, the understanding of limiting factors for bivalve growth and evaluation of their role in coastal ecosystem management. The filtration rate is estimated on the basis of laboratory measurements and is extrapolated to field conditions for comparisons of the different sites.

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