# AACL BIOFLUX

**Aquaculture, Aquarium, Conservation & Legislation**
**International Journal of the Bioflux Society**


## The influence of environmental parameters on the growth and meat quality of the Mediterranean mussel *Mytilus galloprovincialis* (Mollusca: Bivalvia)


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**Abstract**. In this study, changes of average monthly values of the condition index of Mediterranean mussel *M. galloprovincialis* were investigated at seven different locations in the area of Bay of Mali Ston, in the period from November 2008 to December 2009, as well as their relationship with the variation of monthly values of ecological parameters (salinity, temperature, total, organic and inorganic particulate matter). Concentrations of organic and inorganic particulate matter had the highest influence on the condition index at all locations, what indicates the importance of food availability, but also its usability that is influenced by the relation between the concentration of organic and inorganic particulate matter. Mean monthly values of meat indices of mussels at all locations showed statistically significant spatial and temporal variations. This fact indicates that it is necessary to investigate variations of the meat quality at number different locations within the same geografical area. That should be the way for the selection of the production locations within the larger farming areas, and for the strategical organization of the production in order to prolong the market supply with the high quality product. A significant statistical difference was recorded by comparison of the mean monthly values of meat indices with the regard to the dept (1 and 4 m) at different locations. The statistically significant differences in the meat quality of the Mediterranean mussel at the same location at different depths determined during our research, represent a significant economical problem. Producers will have harvest size mussels of different market quality at the same location and the same production unit. This shows that there is a need for applying appropriate equipment and production methods.


**Key words**: mediterranean mussel, condition index, enviromental parameters, meat quality, temperature, salinity.



**Sezetak**. U ovom radu su istrazivane promjene srednjih mjesecnih vrijednosti indeksa kondicije dagnje *Mytilus galloprovincialis* (Lamarck, 1819) na sedam razlicitih postaja u podrucju Malostonskog zaljeva, te njihov odnos s promjenom srednjih mjesecnih vrijednosti ekoloskih cimbenika (salinitet, temperatura, ukupna (TPM), organska (POM) i anorganska (PIM) partikularna tvar) u razdoblju studeni 2008 – prosinac 2009. Najveći utjecaj na vrijednost oba njerena indeksa kondicije na svim istrazivanim postajama imale su koncentracije organske i anorganske partikularne tvari, sto ukazuje na cinjenicu da pored dostupnosti hrane, na njezinu iskoristivost znacajno utjece i odnos izmedju koncentracije POM i PIM. Srednje mjesecne vrijednosti oba mjerena indeksa kondicije dagnji na svim istrazivanim postajama pokazale su statisticki znacajne prostorne i vremenske varijacije. Navedeno ukazuje da je na svakom geografskom području potrebno istraziti varijacije kvalitete mesa na vise uzgojnih mjesta. Samo na ovaj nacin moguce je unutar veceg proizvodnog podrucja pravilno izabrati najkvalitetnije uzgojne postaje, strateski organizirati proizvodnju i produljiti razdoblje snabdjevanja trzista skoljkasima najbolje kvalitete mesa. Rezultati usporedbe srednjih mjesecnih vrijednosti oba mjerena indeksa kondicije s obzirom na dubinu uzorkovanja po pojedinacnim postajama (1 and 4 m) su pokazali postojanje statisticki znacajnih razlika. U vrijeme kada se kakvoci proizvoda na trzistu posvecuje izuzetna pozornost, cini se potrebnim istraziti tehnoloske mjere koje ce na istoj proizvodnoj jedinici omoguciti ujednaceniju kvalitetu.


**Kljucne rijeci**: dagnja, indeks kondicije, cimbenici okolisa, kvaliteta mesa, temperatura, salinitet.


**Introduction**. Mali Ston Bay, located in the Eastern Adriatic, has been a famous area of shellfish cultivation for centuries. While the European flat oyster *Ostrea edulis* (Linnaeus, 1758) has been cultivated since the Roman era, production of the Mediterranean mussel

*Mytilus galloprovincialis* (Lamarck, 1819) was initiated in this area at the begining of 20th century (Benovic 1997).

Mussel farming in this region is based on the collection of larvae from their natural habitat. Collectors with an attached spat of this species remain in the water for an average of five to six months. They are then placed on growout installations until the shellfish reach market size. Ropes with mussels are usually 2.5 to 4 m long, and suspended vertically at a depth between 1 and 5 m. The growout phase is carried out between the sea bottom and surface (considered the mediterranian method), on fixed or floating installations. Lately, floating long lines are the most commonly used installations (Gavrilovic & Petrinec 2003; Gavrilovic et al 2011). Thanks to favorable conditions for shellfish growth, where the mussels (from spat settlement on collectors) reaches market size (> 60 mm) in 15 – 18 months, this area still presents the highest shellfish production basin in Croatia. For example, in the Black Sea, Turkey, it takes two years for the mussel *M. galloprovincialis* to reach its market size (Karayucel et al 2003, 2010), and in the Irish sea (Dare & Davies 1975) and Scotland (Karayucel & Karayucel 1997) it takes between 2.5 and 3 years.

According to Gosling (1992, 2003) shell length or, a more appropriate variable, flesh weight, can be used as indicators of shellfish growth. It is well known that the growth and quality of mussel meat shows a seasonal variation which is mainly influenced by spawning season, as well as by relevant local environmental factors such as hydrographic parameters and the availability of food (Medcof & Needler 1941; Baird 1966; Timet et al 1969; Mitin et al 1971; Hrs-Brenko 1978; Seed 1980; Orban et al 2002; Yildiz et al 2006; Karayucel et al 2010). Furthermore, shellfish handling during cultivation (shell cleaning of mud and fouling organisms, thinning), depth of cultivation and stocking density are important technological factors that could affect growth and commercial quality of mussels (Bolotin 1988; Fuentes et al 2000; Gosling 2003; Santacroce et al 2008).

One of the common methods for the evaluation of meat yield, i.e. the commercial quality of mussels, is the determination of condition indices in which the amount of flesh is related to the quantity of shell (Gosling 1992; Yildiz et al 2006). As shellfish are sesile organisms, their condition at one farming location depends on the combined influence of many different factors. Information about variations of mussel meat quality are necessary for production planning and harvest strategy. This paper describes the combined effects of various factors on the meat quality of the mussel *M. galloprovincialis* cultivated at seven sampling stations in the Mali Ston Bay (Fig. 1).


Fig. 1. Sampling locations in the Bay of Mali Ston.

**Materials and Methods**. This study was carried out from November 2008 to December 2009 at seven different longline farms (Sutvid, Brijesta, Duba, Banja, Kuta, Soca and Bistrina) in the Mali Ston Bay (see Fig. 1). Sampling locations were chosen using the criteria of oyster farm density in the Bay.

The Bay of Mali Ston is situated between the mainland of the Adriatic coast and the Peljesac peninsula in a northeast-southwest direction. The bay is elongated, with a number of coves (Roglic 1981). The whole bay is shallow (the depth in shellfish farming areas usually does not exceed 11 m, with a depth of 29 m at the deepest location). Annual values of temperature and salinity vary during the year, depending mostly on freshwater sources at different locations (underground water sources and river Neretva), with more extreme values recorded in the surface layers (IOR 2003).

Market sized mussel samples (60-80 mm) were collected at two depths (1 m and 4 m) seven times during the investigation period (November 2008, February, April, June, August, October, and December 2009). At each sampling time, and at each sampling station, 60 mussels (30 from a depth of 1 m, 30 from a depth of 4 m) were collected for the determination of two condition indices. At the same time, water samples for the determination of the seston quantity parameters (total – TPM, inorganic – PIM and organic particulate matter POM) were collected with a Niskin bottle from the depths of 1 and 4 m. Salinity and temperature at each sampling depth were measured with a WTW Cond 315i/SET probe.

Every individual specimen was cleaned of mud and biofouling organisms, and then weighed on a digital scale (to 0.0001 g) and sized. The shell was then opened, and the meat and shell were drained onto the filter paper and weighed. Dry meat and dry shell weight were measured after drying to a constant weight at 105 °C.

From these measurements two different condition indices were calculated. The first condition index (CI) was calculated according to the formula: dry meat weight / dry shell weight x 1000 (Mann 1978). The second condition index (MI) was calculated according to Fleury et al (2003) with the formula: wet meat weight / total shellfish weight x 100. Seston quantity parameters (TPM, PIM, POM, ratio of the organic fraction of seston f = POM/TPM, were analyzed according to the method described by Paterson et al (2003). The relationship between CI and MI and the environmental parameters at each station, and at each sampling depth, was determined by a multiple regression analysis (Zar 1999). A chi-square test was used to compare CI and MI values at different depths at each location. A PCA (Principal Components Analysis) was used to determine similarities between individual locations in relation to CI and MI values (Jolliffe 1986; Jackson 1991).

**Results and Discussion**. Results of this research show significant spatial and temporal variations of monthly means of all investigated parameters (condition indices and ecological parameters: temperature, salinity, total, organic and inorganic particulate matter) at the different sampling locations in the Bay. The same parameter values varied at different depths at the same location (see Figs 2-5).

The highest temperatures were recorded in August at all locations and the lowest, depending on different locations, were recorded in February or December (see Figure 2). The lowest temperature (11.7º C) was recorded at a depth of 1 m in December at the Bistrina location, and the highest (26.6º C) at the Kuta location in August. At the depth of 4 m, the lowest and the highest temperatures (12 and 26º C) were recorded in February and August at the Kuta location. Generally, the lowest values of salinity were measured in June at all locations, which could be due to periods of elevated rainfall. The highest measured salinity values, at both depths, showed spatial variations. The lowest value at 1 m was measured in June (21.6 psu) at the Sutvid location, and the highest in November 2008 in Duba (37.9 psu). At 4 m, the lowest salinity was recorded in May in Duba (29.2 psu), and the highest in December at the Brijesta location (38.9 psu) (see Figure 2).

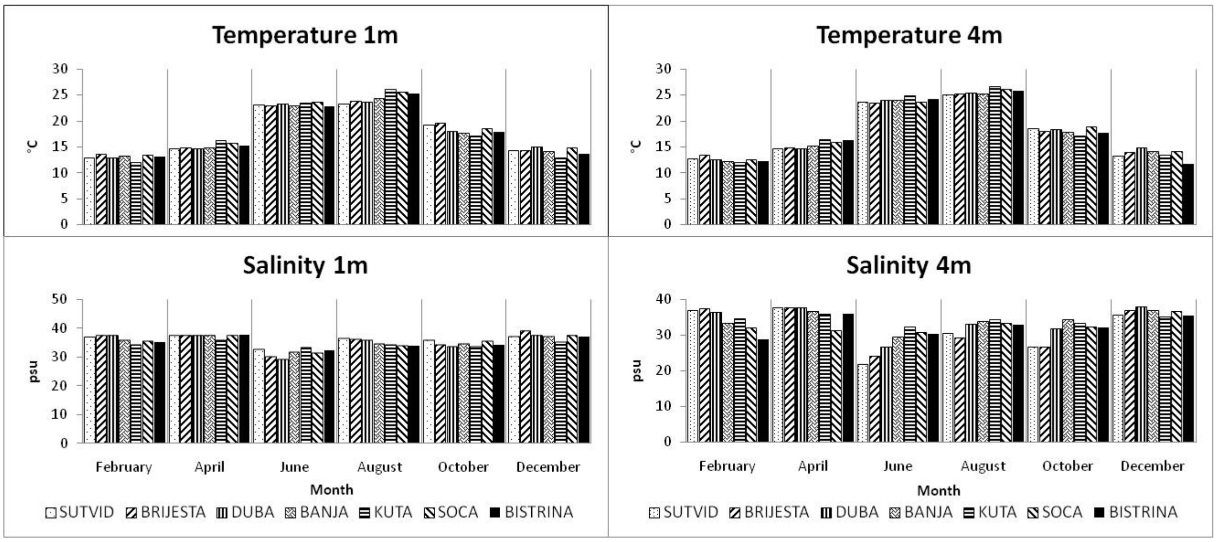


Figure 2. Monthly variation of temperature and salinity at different sampling times, different locations and two different depths in the Bay of Mali Ston.

Concentrations of all components of the particulate matter varied in relation to the location and the depth (see Figure 3). The highest concentration of TPM at 1 m was recorded in August at the Banja location (0.0832 $gL^{-1}$), and the lowest in October in Sutvid (0.0031 $gL^{-1}$). A high concentration of TPM measured in August at the location Banja was followed by the highest measured concentration of PIM (0.04863 $gL^{-1}$) and of POM (0.0346 $gL^{-1}$). At the same depth, the lowest concentration of PIM was measured in October in Soca (0.0014 $gL^{-1}$), and of POM in February in Hodilje (0.0009 $gL^{-1}$). The highest ratio of the organic fraction of seston (f) at the depth of 1 m, was recorded in February in Brijesta (0.5962), and the lowest in November 2008 in Hodilje (0.1107).

At the 4 m depth, the highest concentration of TPM was measured in August at the Kuta location (0.0233 $gL^{-1}$), and the lowest in October in Sutvid (0.0024 $gL^{-1}$). The highest concentration of PIM at the 4 m depth was recorded in August at the Brijesta location (0.0143 $gL^{-1}$), and the lowest, as well as the concentration of TPM, in October in Sutvid (0.0012 $gL^{-1}$). The highest concentration of POM at the same depth was recorded in December in Kuta (0.0126 $gL^{-1}$), and the lowest in November 2008 in Hodilje (0.0008 $gL^{-1}$). At this depth, the highest ratio of the organic fraction of seston (f) was in October at the location Sutvid (0.6169), and the lowest in November at the location Bistrina (0.1212) (see Figure 3).

Both calculated condition indices of mussels (CI and MI) also varied in relation to the sampling time, location and the depth. From the Figures 4 and 5, it is evident that at the different locations the maximal values of CI and MI were recorded at different sampling times. At the 1 m sampling depth, the highest CI was recorded in October at the location Duba (147.89), and the lowest in February in Sutvid (61.91). At the 4 m depth, the highest CI was recorded in April at the Banja location (139.84), and the lowest in February in Kuta (61.23).

The highest MI value of mussels at 1 m was recorded in June in Brijesta (25.61), and in October in Kuta (24.99) and Bistrina (24.15). At this depth, the lowest value was measured in February in Sutvid (10.65). At the 4 m depth the highest value was again measured in Brijesta (25.04), but at the 1 m depth, the highest value is measured not in June but in December. The high value was also recorded in Bistrina (24.37) in October. The lowest value was measured in February in Banja.

In spite of the relatively small area, the results of the PCA statistical analysis of similarity, in relation to the distribution of the mean monthly values of CI and MI, show significant differences. In relation to the values of CI at the 1 m depth, three groups of similar locations were established: Banja and Brijesta, Bistrina and Duba, Kuta and Sutvid. Two similar groups at 4 m were established: Banja and Kuta and Duba, Brijesta and Soca. Results of the comparison of MI at 1 m sampling depth showed two groups of similar locations: Sutvid and Banja, Bistrina, Duba and Kuta. For the 4 m depth, significant similarities were shown for only two locations: Sutvid and Duba.

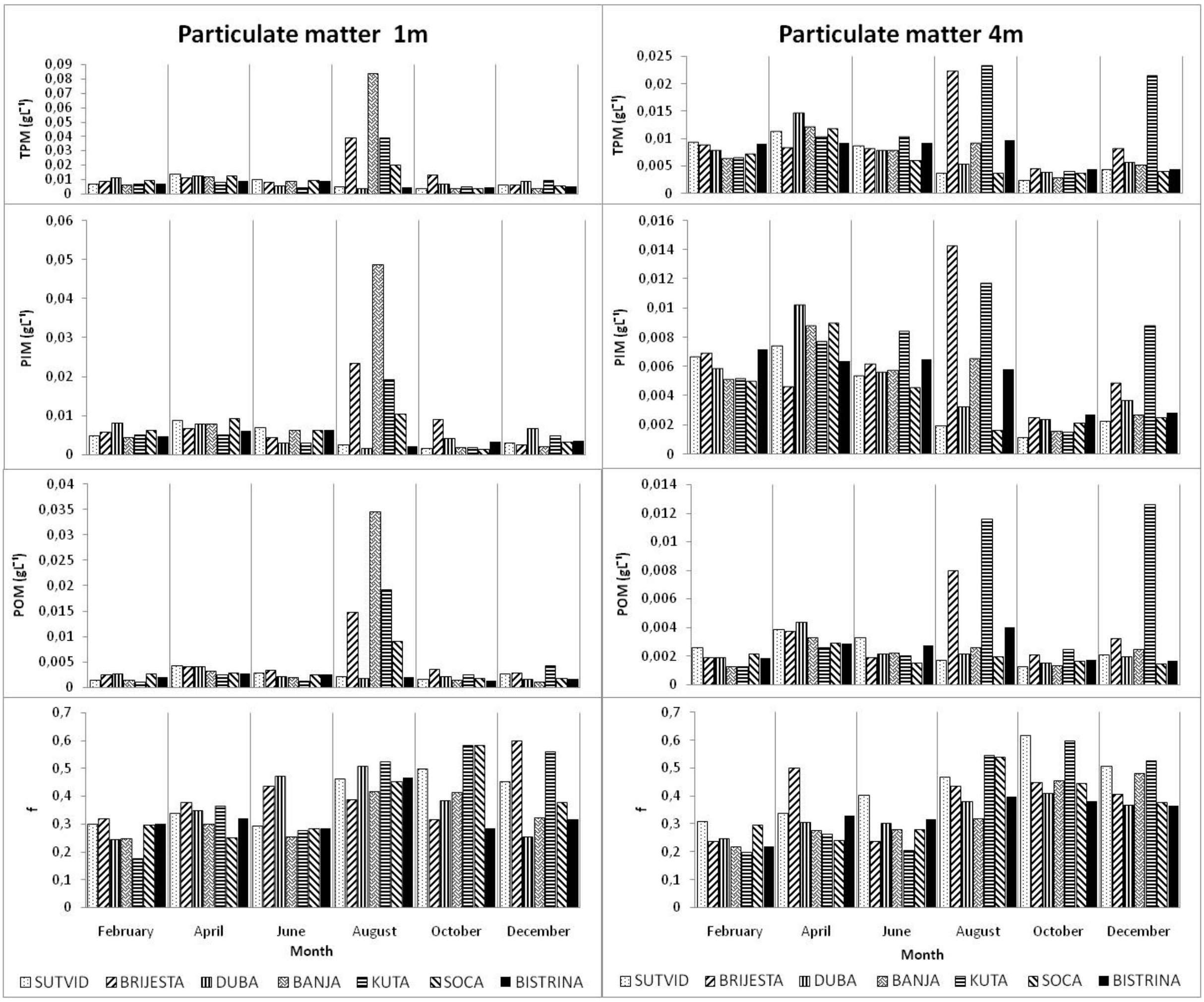


Figure 3. Monthly variation of particulate matter components at different sampling times, different locations and two different depths in the Bay of Mali Ston.

Maximum and minimum condition indices values of mussels reported at different geographical locations also show spatial and temporal variations. In Kilya Bay, Dardanelles, Yildiz et al (2006) found the highest values of CI and MI for mussels in May, while the lowest values were in July and in the winter period. Investigations at two different sites in Italy (at the northern Adriatic coast and in a sea lake along the Central Tyrrhenian coast) showed the highest condition indices of the mussels in March and April. A rapid decrease was observed in December in mussels from the Adriatic site, and in September, in mussels from the Tyrrhenian sea lake (Orban et al 2002). Results by Karayucel et al (2010) show that mussels wet meat weight in the Black sea, Turkey, increased from November to March while rapid decreases were noticed in April – May and October. As described by many other authors, a rapid decrease of mussel meat weight was prescribed to the occurence of the spawning season. Seasonal changes are due to a complex interaction of local environmental factors such as temperature, salinity, and food supply which are thought to influence somatic growth and the reproductive cycle (Gosling 1992). Yildiz et al (2006) reported a significant correlation between the condition index and levels of chlorophyll-a ($p<0.05$), and concluded that chlorophyll-a represents a determining factor for the condition indices values in the investigated area. Karayucel et al (2010) found that mussel meat yields in the Black Sea positively correlate with POM ($P<0.05$). In the Scottish Loch Etive the main environmental factors that effected condition indices of mussel *M. edulis* were temperature and salinity (Karayucel & Karayucel 1997).

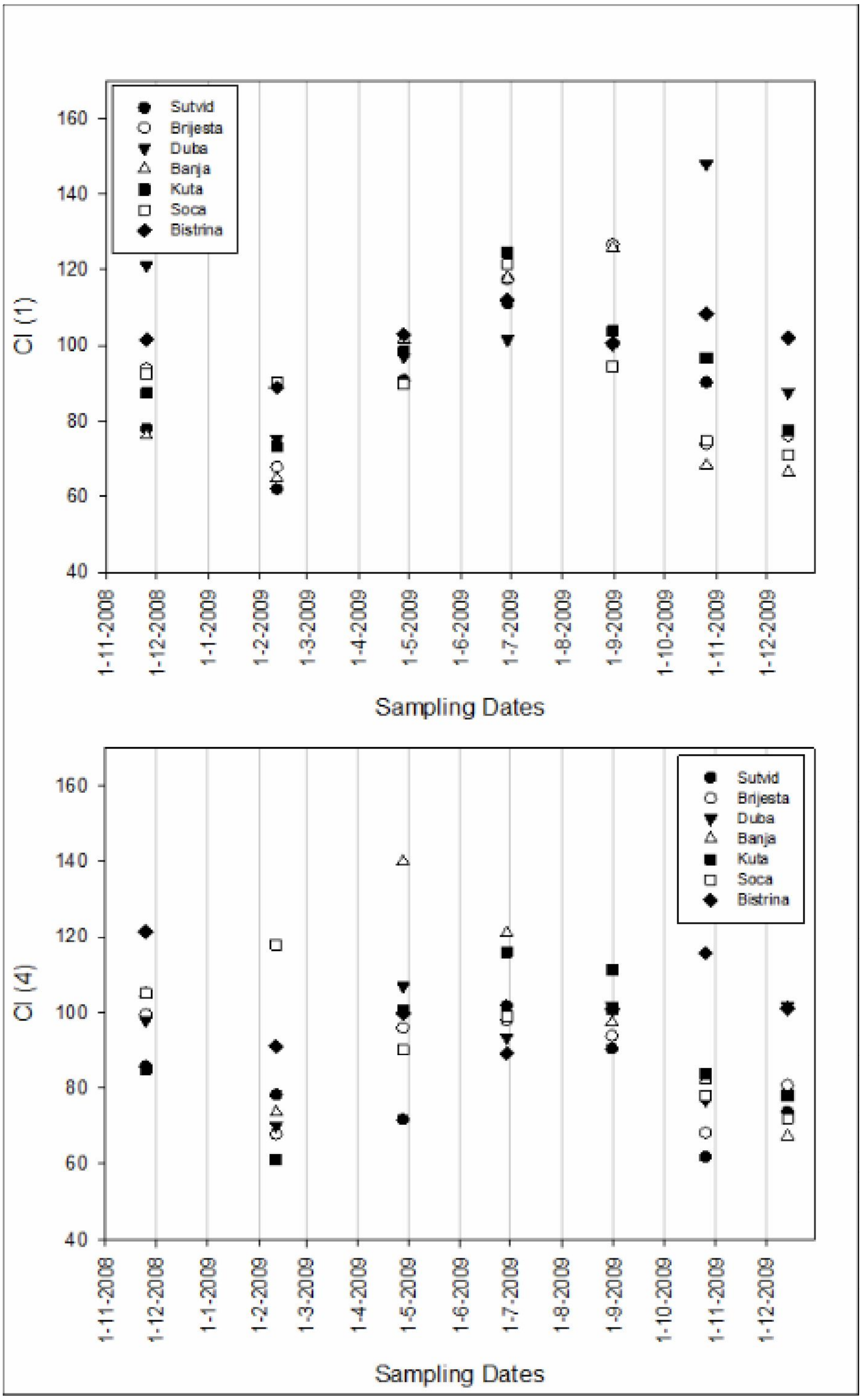


Figure 4. Monthly variation of CI of mussels at different sampling times, different locations and two different depths in the Bay of Mali Ston.

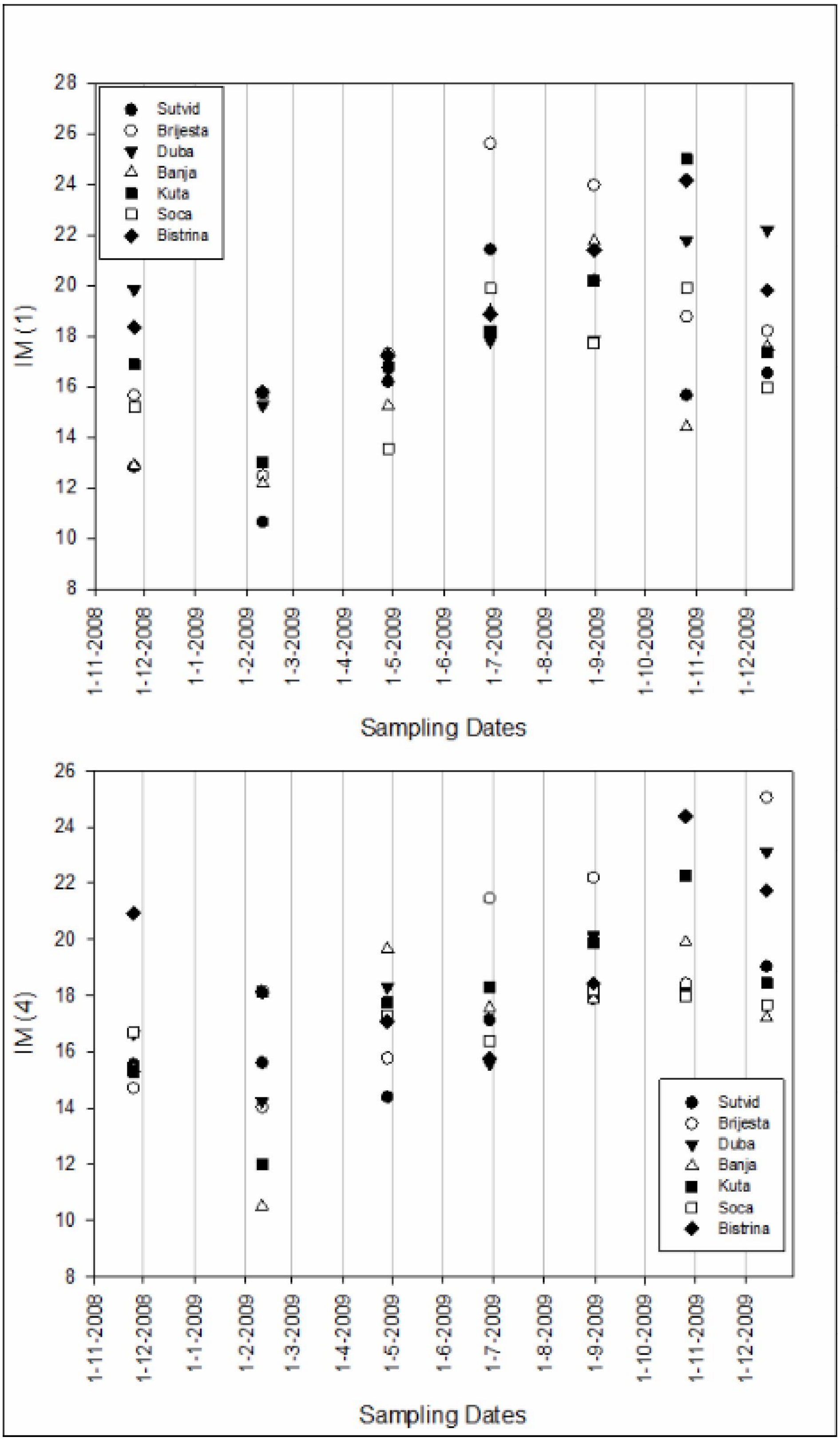


Figure 5. Monthly variation of MI of mussels at different sampling times, different locations and two different depths in the Bay of Mali Ston.

The results of a multiple linear regression (MRA) of this research data (see Table 1 and 2) also demonstrate the presence of seasonal differences. Among all investigated ecological parameters (temperature, salinity, TPM, PIM, POM), both components of the particulate matter (PIM and POM), as well as their relation, have the highest influence on CI and MI values at all investigated locations. Mussels feed on phytoplankton but also on zooplankton, bacteria, detritus and dissolved organic matter (Langdon & Newell 1996; McHenry et al 1979; McHenry & Birkbeck 1985; Prieur et al 1990; Jorgensen 1983; Manahan et al 1983; Devenport et al 2000). Therefore, for a better understanding of the combined influence of local parameters on meat quality, it is important to determine the concentration and relationship of the different individual components of the particulate matter. Our results confirm this, as well as the results of the laboratory research conducted by Jorgensen (1990) and Kiorboe et al (1981). Jorgensen (1990) concluded that optimal laboratory conditions for the growth of the mediterranean mussel must include the presence of mud particles (PIM) in concentrations that are higher that the concentration of suspended food particles. Experiments by Kiorboe et al (1981) showed that adding mud in concentrations 250 times higher than the concentration of algae stimulates the filtration rate of *M. edulis*. The same authors measured the highest filtration rate when the concentration of mud was 25-50 times higher than the concentration of algae. The assumption is that organic particles assist with the mechanical mastication of the feed in the area of the gastric shield and cristalyne style (Murken 1976).

Table 1

The relation between CI of Mediterranean mussel and investigated environmental factors at the individual locations according to the results of MRA

| *Sampling location* | *Equation (IC=)* | $r^2$ |
|---|---|---|
| SUTVID | 248.423-1.709*Datum-200.382*f-12494.429*PIM+ 22394.573*POM-2.738*Sal+ 1.136*Temp | 0.919 |
| BRIJESTA | -105.812-4.044*Datum-109.535*f-12757.058*PIM+17755.114*POM+4.073*Sal+ 7.630*Temp | 0.999 |
| DUBA | -26.034+ 2.681*Datum+676.968*f+31251.856*PIM-44936.144*POM-5.176*Sal-0.038*Temp | 0.999 |
| BANJA | 10.595+3.237*Datum+212.180*f+17079.448*PIM-33051.113*POM-.0479*Sal+ 0.172*Temp | 0.943 |
| KUTA | -329.088+4.530*Datum-40.665*f-2187.55*PIM-1058.733*POM+10.141*Sal+ 4.939*Temp | 0.999 |
| SOCA | 269.839-2.734*Datum-99.547*f-2291.676*PIM+5968.819*POM-4.004*Sal+ 0.194*Temp | 0.999 |
| BISTRINA | -58.292+9.459*Datum+147.556*f+ 12020.823*PIM+ 5536.226*POM+0.260*Sal-0.972*Temp | 0.999 |

Table 2

The relation between MI of Mediterranean mussel and investigated environmental factors at the individual locations according to the results of MRA

| *Sampling location* | *Equation (MI=)* | $r^2$ |
|---|---|---|
| SUTVID | 47.161+0.528*Date-55.908*f-3185.973*PIM+4962.080*POM-0.317*Sal+ 0.165*Temp | 0.999 |
| BRIJESTA | 423.175+11.354*Date-1091.004*f-48636.107*PIM+82926.739*POM+1.394*Sal+ 0.232*Temp | 0.999 |
| DUBA | -3.237+1.610*Date+20.852*f+1879.647*PIM-2482.467*POM-0.029*Sal+ 0.244*Temp | 0.999 |
| BANJA | -17.238+ 1.038*Date+ 13.814*f+ 1027.615*PIM-2064.689*POM+0.463*Sal+ 0.407*Temp | 0.999 |
| KUTA | 36.957+ 0.303*Date+ 37.908*f+ 1888.848*PIM-1708.513*POM-1.300*Sal+ 0.400*Temp | 0.999 |
| SOCA | 21.977-0.173*Date+6.082*f-173.560*PIM-294.507*POM-0.181*Sal+0.101*Temp | 0.999 |
| BISTRINA | -8.316+ 0.578*Date+ 36.434*f+ 1156.945*PIM-3146.449*POM+0.355*Sal+ 0.088*Temp | 0.999 |

A statistical comparison, using a chi-square test, of mean monthly values of CI of the mediterranean mussel cultivated at depths of 1 and 4 m, confirmed significant differences at all investigated locations. The comparison of MI, using the same method, gave similar results, except that significant differences in MI values were not found at the Kuta location. Based on these calculations, and on the variations of CI and MI depicted in Figures 3 and 4 it may be concluded that the Mediterranean mussel in the Bay of Mali Ston, cultivated at the same location, reaches the best meat quality at different periods of the year. Similar to the results recorded by Bolotin (1988) during his investigations in the Bay of Bistrina from 1984-1985, different values of CI for mediterranean mussels were measured at different depths (samples were taken from 1, 3, and 6 m, at the same location). Gosling (2003) also pointed out the importance of cultivation depth for growth and meat yield of shellfish. Fuentes et al (2000) have shown that depth of cultivation is a more important factor for the black mussel than the stocking density. Results of the mentioned investgation showed that mussels in the upper part of the water column, above the thermocline (2.5 m), were significantly larger and heavier than those cultivated in deeper water (7.5 m). Contrary to these findings, Marusic et al (2009) did not find a significant variation of CI in relation to the depth. They conducted eight months of sampling of the Mediterranean mussel from February to October, in the cove of Budava and in the Bay of Rasa, Croatia. Samples were taken at depths of 0.5 m and 2.5 m. It is possible that the depth range was not significant enough to show significant statistical differences.

**Conclusion**. All investigated environmental parameters influenced mean monthly values of the condition index of the Mediterranean mussel at all investigated locations. The most statistically significant influence involves both components of particulate matter (organic and inorganic). Besides the effect of food availability (organic component), the meat quality depends on the optimal relation between these two components.

The results of this research demonstrate that even in the same geographical area, a significant difference in meat quality of the Mediterranean mussel can develop as a result of spatial and seasonal environemental differences. This suggests that at each geographical area, it is necessary to investigate the variation of the meat quality at differing sites and at different depths. This is the only way to select the most suitable locations in a large production area, and strategically organize the production. Only a detailed knowledge of the differing environmental parameters at a number of potential production locations could secure the optimal market supply with the best meat quality for the longest period of the year.

The statistically significant differences in the meat quality of the Mediterranean mussel at the same location at different depths determined during our research, represent a significant economical problem. Producers will have harvest size mussels of different market quality at the same location and the same production unit. This shows that there is a need for aplying apropriate equipment and production methods.

**Acknowledgements**. We are grateful to Dubrovnik-Neretva County for the financing the MARIBIC's project "The characteristics of the farming locations for oysters and mussels on the basis of the monitoring of the growth, condition index, health status and sexual maturation of the farming population in the area of the Bay of Mali Ston".

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Received: 03 August 2011. Accepted: 14 September 2011. Published online: 16 September 2011.
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How to cite this article:
Gavrilovic A., Jug-Dujakovic J., Marinovic Bonacic A., Conides A., Bonacic K., Ljubicic A., Van Gorder S., 2011 The influence of environmental parameters on the growth and meat quality of the Mediterranean mussel *Mytilus galloprovincialis* (Mollusca: Bivalvia). AACL Bioflux **4**(5):573-583.