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Growth weight and reproductive cycle in the mussel (*Mytilus galloprovincialis*) from Cala Iris sea of Al Hoceima (Northern Morocco)

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Abstract

This work aims to evaluate a link among weight variations of Mediterranean mussel *Mytilus galloprovincialis/M. galloprovincialis*, condition index and the environmental parameters. In order to determine the growth weight parameters (total weight, fresh and dry mass tissue weight as well as fresh and dry shell weight) of the bivalve, sampling *M. galloprovincialis* were collected from a mussel farm located along the coastline of Al Hoceima (Moroccan Mediterranean Sea), during the period 2016. The environmental parameters were recorded at the same sampling site. Results show that, the physicochemical parameters affect the growth weight parameters, condition index and the reproductive cycle of *M. galloprovincialis*. The condition index variation is a function of weight, which is itself a function of the gonadal development as well as the availability of phytoplankton. The period of maximum gain weight was observed in summer season, which correspond to the high condition index value, and was associated with the maturity of gonads. The lowest values of CI observed in February (8.71) and May (8.63) coincided with the two major spawning periods. Knowledge of the gametogenic cycle and physicochemical factors that may affect them may constitute a basic data, necessary to choose mussel farming sites, and for a good and sustainable commercial exploitation of bivalves in this part of the Mediterranean Sea.

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Introduction

Marine organisms, especially bivalve molluscs, are a good source of nutrients. The consumption of marine mussels provides an inexpensive source of protein and essential nutrient with high biological value for human consumption. Blue growth is a generally accepted long-term strategy to support sustainable growth in the marine sectors. Farming in open ocean/sea waters has been identified as one potential option to increase seafood production, and it has been a focus of international attention for more than a decade (Costa-Pierce, 2010; Azpeitia *et al.,* 2016; Ren *et al.,* 2019; Azizi *et al.,* 2020).

The Mediterranean Sea is a favorable food source because of the large natural river supply of phosphorus and nitrogen, essential nutrients for marine plants and microalgae (Azizi *et al.,* 2018a), which constitutes an important nutritional source for shellfish. Also, the sea is suitable due to the favorable salinity, temperature, large coastal areas, food availability, reproductive potential, and socio-economic conditions for mussel cultivation (Rouane-Hacene *et al.,* 2015; Benali *et al.,* 2017; Azizi *et al.,* 2018a; Taha *et al.,* 2018).

The common mussel *Mytilus galloprovincialis* has created the interest of many biologists for its economic and malacological importance. In Morocco, this interest is particularly increased as an aquaculture species who's the development of its exploitation in the Moroccan Mediterranean is provided by the Halieutis Plan (ANDA, 2017). The growth of mussels is an important biological factor to consider in any mussel farm. This growth is broadly defined as a change in the individual's body shape and biomass. It is particularly dependent, in mussels, on the environmental conditions in which they lead a sedentary life. Mussels respond to these environmental conditions through both morphological and physiological adaptations (Kefi *et al.,* 2014; Kerdoussi *et al.,* 2017). Authors (Okumuş and Stirling 1998; Azizi *et al.,* 2018b; Taha *et al.,* 2018) had indicated that mussel growth is a function of a number of environmental factors, mainly temperature, salinity, phytoplankton and local hydrodynamics. En relation to the physiological status of mussels, Chelyadina *et al.,* (2018) indicated that different parameters of growth show variations being closely related with different phases of the reproductive cycle for *Mytilus galloprovincialis*. The weight of mussels undergoes a seasonal cycle characterized by periods of loss and gain of animal weight reflecting the stage of gonad development (Orban *et al.,* 2002; Kefi *et al.,* 2014; Bhaby, 2015). Azizi *et al.* (2018a) shown that *Mytilus* lost up to 40% of the body weight during the spawning period. The purpose of this study was to examine the seasonal changes in the weight growth parameters and condition index of *M. galloprovincialis* in relation to the environmental factors in the Cala Iris bay of Al Hoceima (Northern Morocco). The relation between the weight growth and the gonadal development cycle of *M. galloprovincialis* was also studied.

Materials and methods

Study area, and sample collection

The Mediterranean mussel *M. galloprovincialis* and seawater were obtained from the mussel farming installed in the Cala Iris Bay of Al Hoceima. Waters and mussel samples were collected monthly in 2016, from Junuary to December, from five sampling sites (A, B, F, J and I) (Fig. 1), to ensure homogeneous distribution of samples. Water was sampled by hand into several bottles according to the analytical specifications and transported at +4°C in a cold box to the laboratory until analysis.

Environmental variables

Temperature, salinity, dissolved oxygen and turbidity in sea water were recorded *in situ* at all sites by means of a multiparameter device (HQd Series portable meters, HACH, Safety Mark), at the same moment that the mussels were sampled. In the laboratory, samples were immediately filtered using Millipore membrane filter paper (Whatman GF/C filters) and chlorphyll *a* were measured by a colorimetric methods according to standard methods recommended for marine waters (Aminot and Chaussepied, 1983).

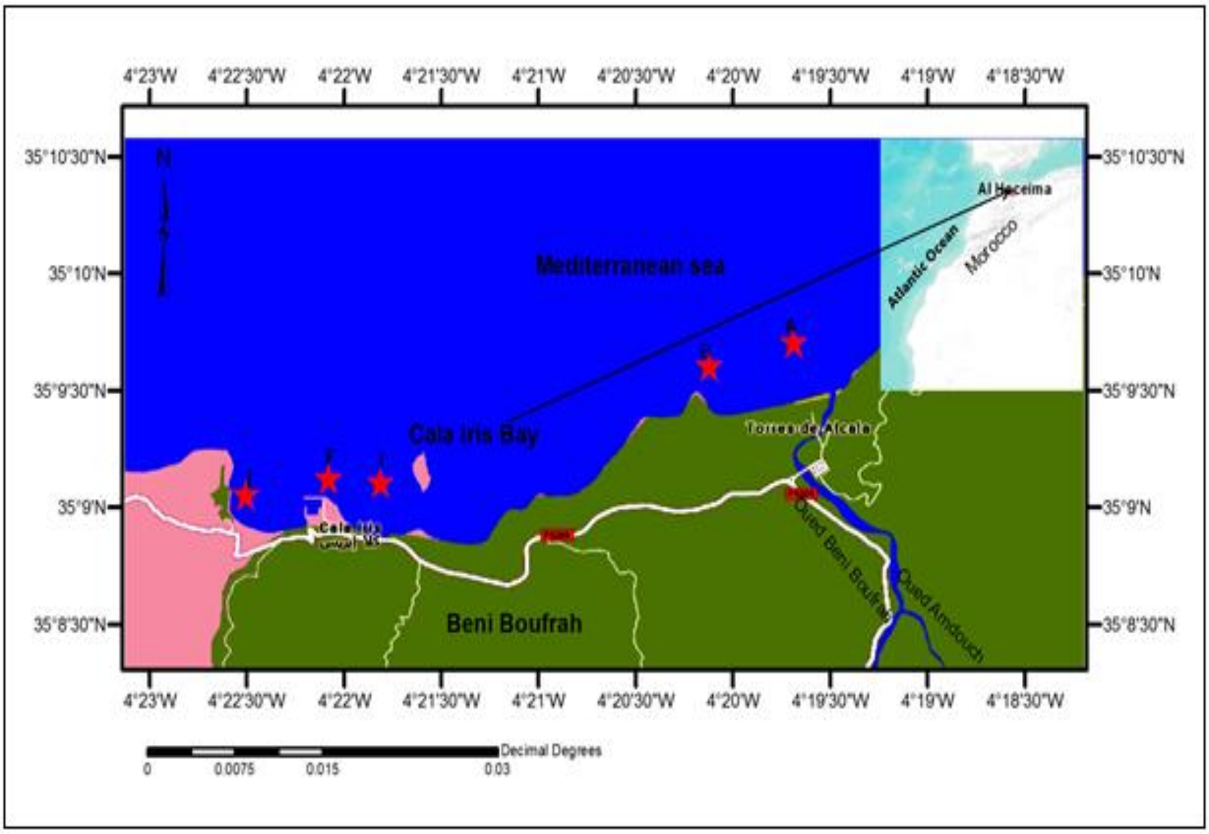

Fig. 1. Geographical location of mussel sampling sites along the mussel farming from coastal areas of Al Hoceima (Northern Morocco).

Growth and condition Index

Fifty-eight samples of bivalves (*M. galloprovincialis*) were collected monthly from five sampling sites (A, B, F, J and I) (Fig. 1), and immediately transported to the laboratory under refrigeration (3 ± 1^{0}C) and were brushed, washed and processed on the same day. For each harvested mussel, the weight measurements including, total weight (TW; g; whole body weight) and a weight of the different components (fresh shell weight (FSW; g) and dry shell weight (DSW; g), fresh flesh weight (FFW; g) and dry flesh weight (DFW; g)) were carried out to the nearest 0.001 g, using an analytical balance. Shell and flesh dry weight were obtained by constant heating at 103 °C until a stable weight was reached The condition index gives us a clear idea about physiological status of the individuals in a given population (Kefi *et al.,* 2014), and permit estimation of the gonadal tissues lost during the reproductive cycle (Chelyadina *et al.,* 2018).

The condition index (CI) of Mediterranean mussel was calculated as the ratio of dry flesh weight (DFW) to total weight (TW) (CI = (DFW/TW) ×100) (Galvao *et al.,* 2015; Chelyadina *et al.,* 2018). The condition index allows us to follow the different stages of the *Mytilus galloprovincialis* reproductive cycle (Azpeitia *et al.,* 2017). The improvements in accuracy and speed of measurements which are possible with the Cl method permit quite large samples of mussels to be evaluated individually, with a high degree of precision (Kerdoussi *et al.,* 2017).

Statistical analysis

All statistical data analysis was done using STATGRAPHICS plus 5. The data were expressed as means ± standard deviations (mean ±SD).

Results and discussion

The temporal variations of the seawater parameters (temperature, salinity, dissolved oxygen, Chlorophyll *a*, and turbidity) recorded throughout the sampling period are summarized in Table 1. Seawater salinity varied between 35.62 and 40.17 psu, observed in January and July during the experimental period of

2016, respectively. These differences between seasons founded in salinity, revealing the influence of freshwater coming from runoffs (Beiras *et al.,* 2003; Pinto *et al.,* 2015; Azizi *et al.,* 2018a). Temperature ranged between 18.16 and 20.67 °C observed in February and August, respectively.

Table 1. Mean monthly variations of the environment parameters of seawater recorded in the Cala Iris bay of Al Hoceima.

Month	T (°C)	Sal (psu)	O_2 (mg l^{-1})	Chl *a* (µg/l)	Turbidity
December	17.46	36.67	9.64	0.552	0.15
January	17.08	35.62	9.45	0.534	0.61
February	16.18	36.32	11.88	0.574	0.93
March	16.55	37.36	11.55	0.534	0.52
April	17.64	37.60	10.63	0.534	0.21
May	17.91	37.39	10.12	0.675	0.74
June	19.82	39.41	11.92	0.582	0.34
July	20.40	40.17	11.20	0.596	0.46
August	20.67	39.38	11.17	0.597	0.60
September	20.23	38.32	11.14	0.335	0.25
October	19.79	38.30	10.95	0.367	0.37
November	19.71	38.39	10.73	0.278	0.22

T: temperature; Sal: salinity; O_2: dissolved oxygen; Chl *a*: Chlorophyll *a*.

The lowest dissolved oxygen was recorded in winter season with a peak in January (9.45 ± 0.338 mg/l), and the highest value was observed in June (11.93 ± 0.217 mg/l). These high values reveal well oxygenated waters in the mussel farming from the Al Hoceima coast regions. Turbidity values were fluctuate between 0.21 and 0.93, recorded in April and February, respectively. Recorded chlorophyll *a* content shows fluctuations between 0.278 µg/l in November, and 0.675 µg/l in May. Previous investigators (Ojea *et al.;* 2004; Lemaire *et al.,* 2006; Ivanov *et al.,* 2007; Kefi *et al.,* 2014) have shown that physicochemical parameters and nutritional status, in the coastal marine environment, can affect growth performance and reproductive cycle of bivalves. The environmental parameters of waters comply with a fundamental role in the reproductive activity and different parameters of growth for mussels. García *et al.* (2016) has indicated that mussel growth is a function of a number of environmental parameters, mainly food and temperature. The environment influences the somatic and reproductive tissue growth of mussels (Pazos *et al.,* 1997; Ceballos-Vázquez *et al.,* 2000; Kerdoussi *et al.,* 2017). Total weight of the Mediterranean mussel *M. galloprovincialis* obtained from the mussel farming installed in the coastal areas of Al Hoceima oscillates between 22.11 and 48.55 g observed in May and July months, respectively. This parameter shows peaks in January (30.45 g), April (34.35 g), and July (48.55 g) (Fig. 2).

Fresh flesh weight of mussels from the mussel farming of Al Hoceima regions shows fluctuation from 4.04 g (May) to 9.10 g (July) (for an average weight of 6.16 g).

Regarding shell weight of *M. galloprovincialis* collected from Al Hoceima coastline, values ranged between 8.96 and 18.60 g observed in November and July, respectively (average weight = 11.98 g) with peaks in January (13.97 g), April (12.69 g) and July (18.60 g).

With regards to the dry flesh weight of mussels from the mussel farming of Al Hoceima coastline, it oscillates between 0.69 and 1.66 g observed in May and July, respectively. The dry flesh weight shows a decresed values in March (0.79 g), May (0.69 g) and November (0.81 g).

Dry weight values of shell recorded to the collected mussels from the Al Hoceima coasts are between 8.58

and 17.67 g observed in November and July months, respectively (for an average weight of 11.37 g).

Variations in the annual flesh and shell growth weight patterns of marine bivalve mollusks are controlled by both environmental (temperature, salinity and phytoplankton abundance) (Bayne and Newell, 1983; Jaramillo and Navarro 1995; Marsden and Pilkington 1995; Wu *et al.*, 2017; Chelyadina *et al.*, 2018) and physiological factors (gametogenesis) (Bayne and Worrall, 1980; Vishwajeet *et al.*, 2015).

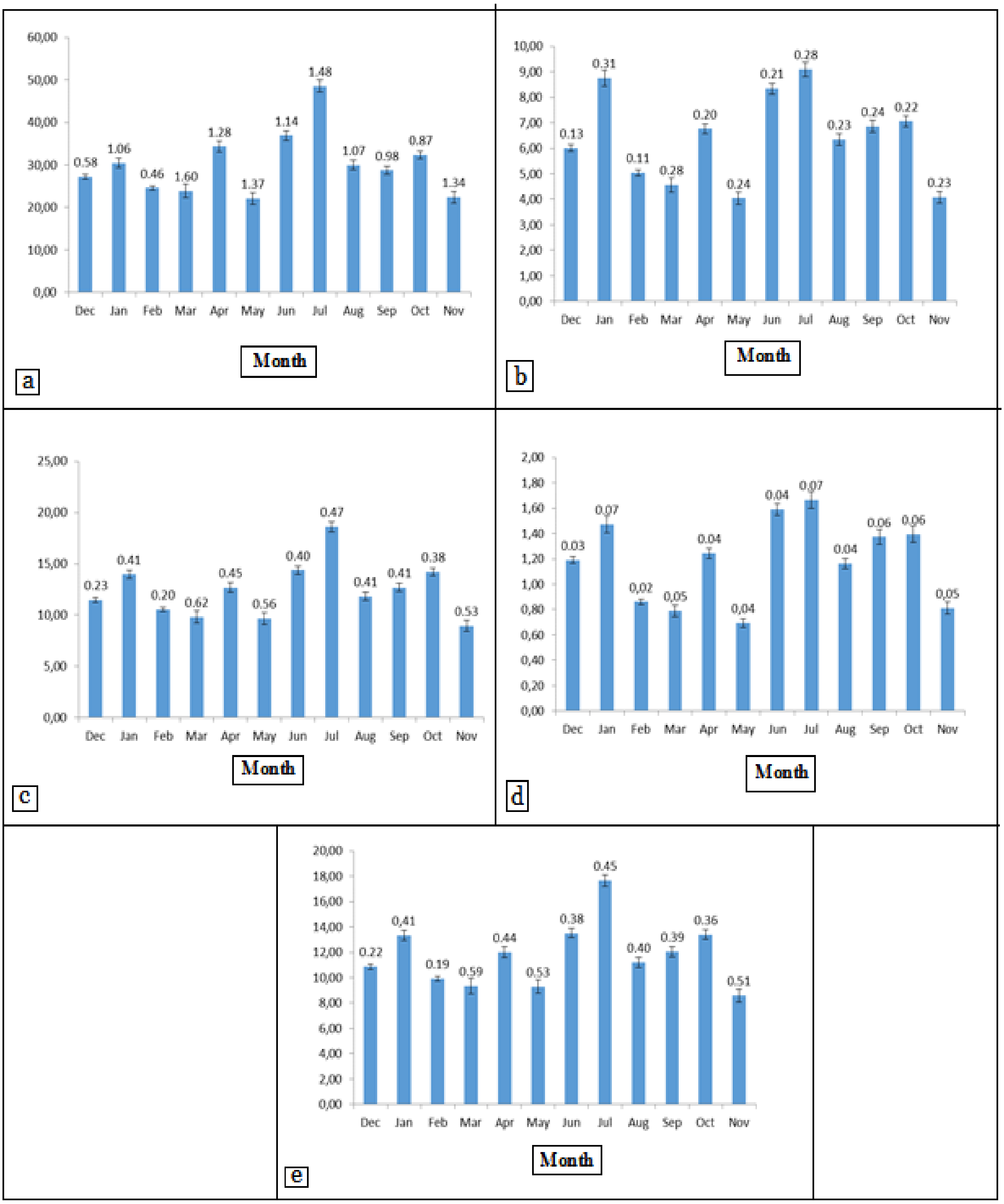

Fig. 2. Monthly distribution of different weights (mean ± SD) of the mussel *M. galloprovincialis* (a: total weight (TW); b: fresh flesh weight (FFW); c: fresh shell weight (FSW); d: dry flesh weight (DFW), and e: dry shell weight (DSW)) from Al Hoceima Coastal region during the sampling period of 2016.

Different parameters of flesh and shell weight patterns for individual mussels show seasonal variations in the Cala Iris sea of Al Hoceima, with low total, flesh and shell weights during late winter, spring and summer. Authors (Jantz and Neumann, 1998) found similar results in the growth of the Zebra Mussel in the River Rhine. In a study conducted on the Tokyo Bay of Japan, Okaniwa *et al.* (2010), reported that cultured mussels (*M. galloprovincialis*) show the lowest shell growth during winter and spring is possibly controlled by combined exogenous (lower seawater temperatures and phytoplankton) and endogenous (the reproductive effort required for gametogenesis and spawning) factors. Bayne and Worrall (1980) showed that the growth rate of *Mytilus edulis* in the sites near Polumoth (England) was related to the environmental factors such as temperature and food availability.

These two parameters also appear among the main physicochemical factors cited by Sokołowski *et al.* (2010), which affect the soft tissue weights of mussel *Perna perna* distributed along the coastline of the Gulf of Aden (Yemen). In the present work, the soft tissues gain weight coincided with the high abundance of feed.

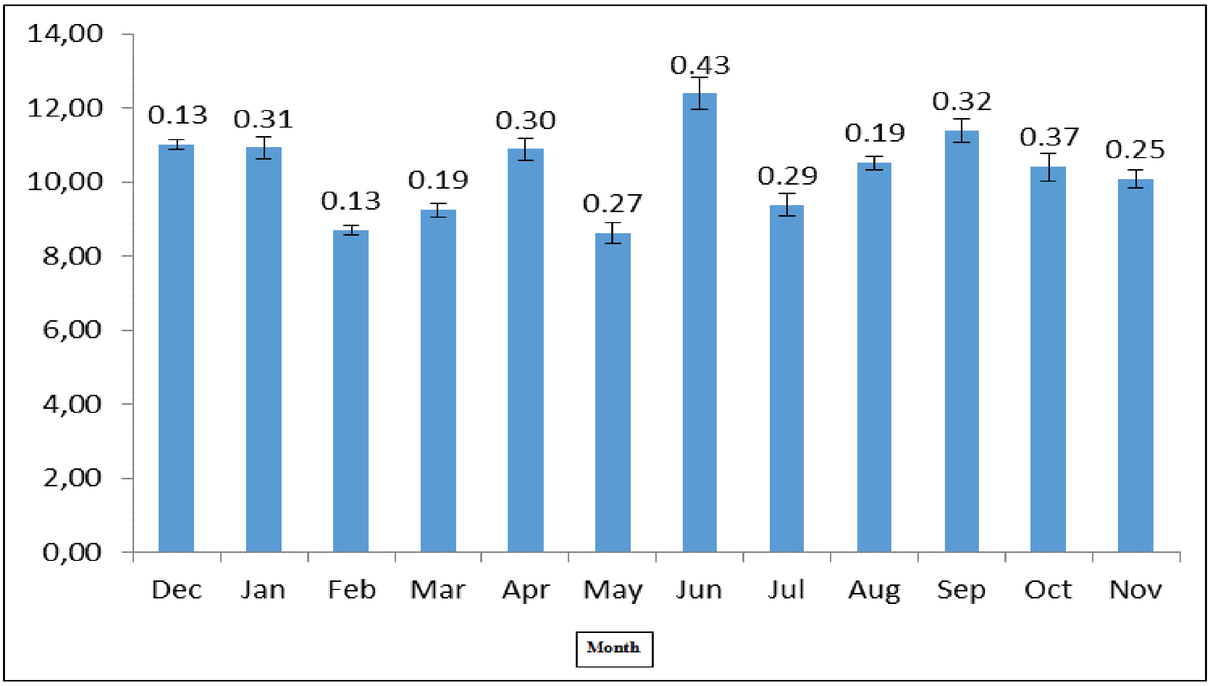

Fig. 3. Monthly evolution of the mussel M. galloprovincialis condition index (CI) (mean ± SD) collected from the Al Hoceima coastline during the sampling period of 2016.

Changes in body weight for standardized specimens are important to determine the onset of spawning periods. Jantz and Neumann (1998) found that the beginning of the spawning season was marked by a decrease in the relative weight of mussels. The visceral sac revealed the strongest decrease of dry weight, due to the release of gametes. The remaining tissues also lost weight, because of the remobilization of storage substances during reproduction (Borcherding, 1995), which indicated the clear evidence of the decreased in body weight values during this period. The endogenous factor as gametogenesis can affect the somatic tissue growth. Authors (Kefi *et al.*, 2014; Vishwajeet *et al.*, 2015) found that during the gametogenesis, the somatic growth decreases once the energy is allocated to the gonadal production.

As a consequence of the scaling of physiological processes in relation to body weight, the correction of dry tissue weights to an individual of standard weight is a common method for describing the reproductive cycle of mussel species (Borcherding, 1991; Dorgelo and Kraak, 1993; Khafage *et al.*, 2019).

Many authors (Aloui-Bejaoui *et al.* 2002; Kefi *et al.* 2014) have highlighted the value of studying dry flesh weight when determining the weight changes of mussels *M. galloprovincialis*, as well as when studying the gametogenic cycle indicated by the condition index. The fluctuations of dry flesh weight for mussels collected from the Al Hoceima coastline can depend largely on the degree of the sexual cells maturity, and thus have the advantage of explaining a loss or gain of weight tissue mass, without taking into account either changes of weight relating to the growth, or the volume of water retained by the bivalve during its reproductive cycle. A similar observation was noted by Ojea *et al.* (2004) who indicated that the lowest values of CI coincided with the minimum values of dry weight for different tissues. However, Galvao *et al.* (2015), determining the CI in mussels by using both soft tissue fresh and dry weights, they found that the water content does not play an important role in the parameter analyzed, suggesting that soft tissue fresh weight could also be used in the CI estimation. Previous works (Galvao *et al.* 2015; Kerdoussi *et al.* 2017) have been suggested requirement of standardizing the water content mussels' soft tissue, since mussels increase water uptake when the living condition is not favorable.

Condition index (CI) of the collected mussel *Mytilus galloprovincialis* from the sampling sites of Al Hoceima coastline ranged from 8.63 to 12.38 (with a mean of 10.23) observed in May and June months, respectively(Fig. 3). The CI indicate peaks in January (10.93), April (10.89) and Jun (12.38).

The CI values coincided with the fatting period, which include the resumption phase of gametogenesis activity and gonadal maturity. This index can serve both to determine the quality of bivalve products and to characterize the biological status (reproduction and growth) of mussels (Vishwajeet *et al.*, 2015). Chelyadina *et al.* (2018) used the index to describe the gametogenic stages of bivalve molluscs. Several authors agree to interpret as a beginning sexual cells maturation, an increase in the average values of this index and its sudden fall as an emission of gametes (Paulet *et al.,* 1988; Scudiero *et al.,* 2014).

In our study, the lowest CI observed in February (8.71), May (8.63) and July (9.39) coincided with the spawning season for *M. galloprovincialis*. These data are in agreement with Id Halla *et al.* (1997) observation about the spawning season of *Mytilus galloprovincialis* in the Bay of Agadir on the Atlantic coast of Morocco. Ojea *et al.* (2004) also observed a rapid decrease in the tissue weights and condition index at the beginning of the gamete releases. Reduction in this index is synchronous with development of gonad.

This phenomenon may be associated with the use of the somatic tissue reserves for gametes production (Gabbott, 1975; Zaba, 1981; Paulet *et al.,* 1988). Therefore, in estimating the mussels' weight-growth, we assumed February, May and, with less frequency, July to be the critical months for mussel-weight decrease due to gamete release.

The condition index reflects the physiological condition of the living organism. The seasonal fluctuation of this condition index can provide an idea about the gonad state and the progress of reproductive cycle of *Mytilus galloprovincialis*, and these seasonal variations depends on the environmental parameters such as temperature and abundance of food. In our study, we observed an increased in the food availability and suspended matter in February, May and July/August, which indicated the period of the gamete releases. Similar results were revealed by various authors (Kennedy, 1977; Villalba, 1995; Suárez *et al.,* 2005; Cuevas *et al.,* 2015), which noticed that the lowest CI coincides with highest levels of phytoplankton and suspended matter, whilst bivalves spawn during these periods.

Dix and Ferguson (1984) also observed a major spawning period in late winter, early spring and subsequent minor spawning during summer in *Mytilus edulis* from Tazmania. Authors indicated that food supply and seawater temperature constitute the principal factors controlling the timing and duration of gametogenesis. Urrutia *et al.* (1999) and Ojea *et al.* (2004) showed that in seawater when food is abundant, the surplus energy may be shared among the gonad development and somatic tissues growth of the mussel. According to authors (Villalba, 1995; Urian, 2009; Azpeitia *et al.* 2016) seasonal variations in the condition index of *Mytilus galloprovincialis* result from complex interactions among many factors as the nutrition, temperature and salinity, on the metabolic activities of the bivalve, and particularly on the gonad development and growth.

Conclusion

The mussel *M. galloprovincialis* has reproductive cycle that extends throughout the year, with two major spawning periods in winter and spring and subsequent minor spawning during summer.

The season of maximum weight gain was observed in summer period for the mussel collected from the Al Hoceima seawater, which coincide with the highest value of condition index, and was associated with the sexual cells maturity. Our study suggests that variations in environmental factors (phytoplankton and suspended matter) in the farming mussels of Al Hoceima coastline have marked effects on growth weight and gonadal cycle of *M. galloprovincialis*. Furthermore, studies are required to examine the effects of environmental factors on mussel bivalves to further evaluate the aquaculture potential in this region.

Conflict of Interest

The authors declare that they have no conflict of interest.

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