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Stock mapping, size structure and biological parameters of the clam *Polititapes aureus* in the shellfish production area of the southern Tunisian waters (Central Mediterranean)

by

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Abstract

The clam *Polititapes aureus* is one of the most abundant shellfish species in the southern Tunisian waters. Its current exploitation status and management are becoming a major concern for fishing industry in Tunisia. The significant ecological role of the species and possible future commercial benefits require a better knowledge of its stock. This research is the first attempt to investigate its current status in an area with the largest shellfish production. The obtained results showed a scattered distribution. The stock density ranged from 0 to 124 ind. m^{-2}, and biomass values varied from 0 to 300 g m^{-2}. This results in a remarkable biomass of 201.2 ($\pm$ 64.6) t and high abundance reaching 91.3 $\pm$ 32.9 million individuals, estimated an area of 4182 ha. The species distribution was also investigated, with the size ranging from 4.9 to 35.34 mm. The overall sex ratio (F:M) was 1.26:1, which significantly deviated different from parity (1:1). The main environmental factors were identified and several abiotic parameters were found to strongly affect the spread of the clam species. The clam reproduce well and is expected to almost meet the domestic market demand through artisanal fishery.

Key words: edible shellfish species, *Polititapes aureus*, stock mapping, population structure, sex ratio, morphometric relationships, south Tunisia

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1. Introduction

Shellfish production is a major contributor to the Tunisian fishery economy. Bivalves can play an important role in addressing the scarcity and rising prices of protein sources in Tunisia. The Tunisian coast stretches over long distances, so it becomes necessary to look at the exploitation of bivalves occurring in these areas. Intertidal bivalves are conspicuous members of productive areas. Unfortunately, shellfish exploitation has so far focused only on the clam *Ruditapes decussatus* (Linnaeus, 1758), which is extensively harvested from natural populations in Tunisia. This species is important to the economy, especially in terms of employment and the trade balance of the country through its exports.

The venerid *Polititapes aureus* (= *Venerupis aurea*) (Gmelin, 1971) is endemic to the Mediterranean Sea (Fouda & Abou-Zied 1990). It has migrated through the Suez Canal and successfully colonized the Tunisian coasts. The clam *P. aureus* belongs to the most common marine mollusk species found in the intertidal zone on the southern Tunisian coast including vast mudflats and sandy beaches favorable for infaunal bivalves (Ben Salem et al. 2002). Natural populations of *P. aureus* occur in sandy and silty-sandy sediments in bays, estuaries, coastal lagoons and other sheltered environments, living on tidal flats and below the low-tide level to a depth of 1 m. The species also plays an important role in ecosystems. It is a filter feeder pumping water through its siphons at the sediment level (Vilela 1950).

Despite its considerable economic importance, no studies have been carried out in Tunisia on the spread of the mollusk *P. aureus*. Consequently, the status of this species is still poorly understood and there is a knowledge gap in stock assessment. The literature review clearly indicates a lack of studies focusing on the stock size and biology of *P. aureus* in southern Tunisia. Stock mapping is necessary to indicate the location of major bivalve beds along the southern coastal areas, which may have potential commercial value. This study can provide basic knowledge in different fields of environmental research and constitute a first step for any future work and can be useful as a reference for research on in marine invertebrates. It presents a method for quantitative assessment of the bivalve species conducted in the area with the highest shellfish production. The species may be important due to its high exploitation potential. Therefore, this paper is the first attempt to obtain basic information on *P. aureus* by investigating its population structure, sexuality, geographical distribution and stock size in relation to the effects of certain abiotic factors.

2. Materials and methods

2.1. Study area

The southern coast of Sfax, located in the Gulf of Gabes (south Tunisia), extends for 135 km (Fig. 1). The climate is arid and semiarid Mediterranean, largely influenced by the gentle topography and maritime exposure (Chamtouri et al. 2008). Both the wide and shallow continental shelves are topographically regular. The bottom slopes slightly toward the sea, with a depth of 60 m occurring 110 km off the coast (Ben Othman 1973). The substrate in the inshore area is mainly silty sand (Derbali et al. 2016) covered in some places with the seagrass *Cymodocea nodosa* (Ucria) Ascherson and *Zostera noltei* Hornemann. The littoral zone of Sfax is characterized by a benthic community of an exceptional bionomy, consisting of extensive meadows of magnoliophytes *Posidonia oceanica* and *C. nodosa* (Ben Mustapha & Hattour 2013). Their leaves provide a suitable substrate for the establishment and growth of shellfish.

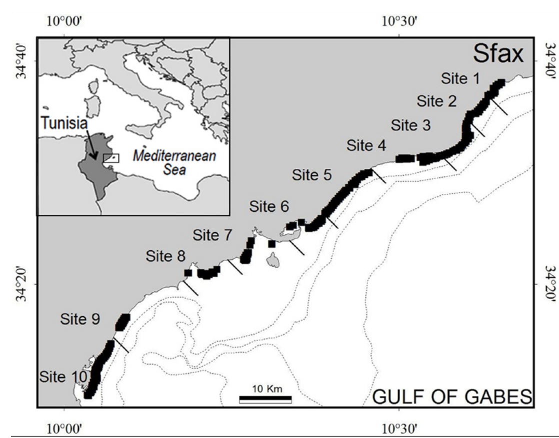

Figure 1

Map of the study area. The location of sampling transects is indicated

2.2. Sampling protocol and operations

Sampling operations were conducted from May 2012 to August 2014 within the 135 km shellfish production area (Sfax coasts, Gulf of Gabes) in south Tunisia. The sampling area was roughly divided into ten sites based on the occurrence of clams (Fig. 1). Transects were systematically run in the sampling area during low tides. Samples were collected every 50 m along transect lines from the extreme high tide to the extreme low tide. Along transects, 4–10 sites were sampled in quadrats (0.25 m^2) using a shovel. Large specimens were collected by hand and small ones using a 2 mm mesh sieve. During the sampling period, seawater temperature and were measured near the bottom immediately after sampling using a multi-parameter kit (Multi 340 i/SET). To complete the study on the clam distribution, an ecological survey was carried out at all study sites. Specific interactions between abiotic and biotic factors affecting the spatial distribution of the *P. aureus* population were investigated by analyzing granulometric characteristics, organic matter content and the structure of shellfish communities recorded during the sampling period. Samples of sediment were collected from each site to a depth of 5 cm. Samples were pooled for each site and used for granulometric analysis (Buchanan 1984). Results were expressed as % gravel (> 2 mm), % coarse sand (2–710 μm), % medium sand (710–250 μm) and % fine sand (< 250 μm).

2.3. Data analysis

Samples were placed in labelled plastic bags, and then preserved in a 7% formaldehyde solution. In the laboratory, the material was sorted and washed to remove all attached organisms and other debris. Individuals were identified, counted and shell length (SL) was measured with a digital caliper to the nearest 0.01 mm and weighed on a toploading digital balance (precision of 0.001 g) to determine total fresh weight (TW). The obtained dataset was registered and maps were drawn. After identification to the species level, data were pooled at sampling sites to obtain mean densities (ind. m^{-2}) and mean biomass (g m^{-2}) per site, and then pooled across the sampling sites to assess stocks based on the kriging method using ArcView v. 3.2 software, according to the following equation (Gulland 1969):

$$B_i = N_i \times \frac{A_i}{a_i} \times \frac{1}{X_e}$$

where B_i – represents the total biomass of clams; N_i –mean abundance in a sample; A_i – the whole study area; a_i – the swept area and X_e – the proportion retained.

Statistical analysis was performed to investigate the effect of site on SL and abundance using one-way ANOVA. Similarities between the sites in terms of abundance and biomass were investigated using cluster analysis (group average). In addition, the harmonic Spearman correlation coefficient was also applied to identify any significant correlation between density and biomass of clams at each site. The results are presented as means ± standard error (SE) and the significance level used in the tests was $p < 0.05$.

In biological studies, clam species were examined for sexuality, with all specimens subjected to (1) macroscopic examination of gonads and (2) microscopic examination of gametes. The visceral mass was then teased apart and smears of the visceral wall with attached gonads were examined at 100× magnification and the sex of individuals was determined. The sex ratio (expressed as a number of females per males; F:M) was determined. Statistically significant deviations from a balanced sex ratio of 1:1 were assessed by the χ^2 test, with statistical significance considered at $p < 0.05$ (Zar 1996). The statistical package STATISTICA v. 6.0 was used.

3. Results

3.1. Occurrence and distribution

P. aureus was found in various substrates throughout the study area, including a silty-sand substrate covered in some areas with the marine seagrass *C. nodosa* or *Z. noltii*, or mixed vegetation consisting of these two seagrasses. A total of 143 transects were run from the extreme high water tide to the extreme low water tide, corresponding to a total of 966 samples. Data on sediment parameters for all sites related mainly to silty-sandy substrates, except site 3 (Table 1). Most of the sampled sites were covered with the seagrass *C. nodosa* and *Z. noltii* (> 65%). During the sampling period, the highest temperature values were recorded in July (27°C), whereas the lowest values were recorded in February (12°C). Salinity measurements showed an annual fluctuation between 36 PSU in winter and 48 PSU in summer.

A total of 966 replicates were taken during sampling. Mean stock levels for each site are presented in Table 2. In general, there were significant fluctuations in the distribution of clams across the sites, with a density ranging from 0 to 124 ind. m^{-2} and biomass from 0 to 300 g m^{-2} (Figs 2 & 3). The density of clams does not show normal distribution (Kolmorogov–Smirnov test, $p < 0.05$) and was not homogeneous (Leven's test, $p < 0.05$). In addition, pairwise comparisons between density and biomass levels indicated that data obtained for all production sites were significantly different (Kruskal–Wallis median test, $p < 0.05$).

3.2. Stocks assessment

The total population of the shellfish under study was estimated at 201.2 ± 64.6 t (total fresh weight), with mean biomass of about 5.3 ± 0.8 g m^{-2} and total abundance of over 91.3 ± 32.9 million individuals.

Table 1

Sediment type and organic matter content recorded in the shellfish production area of Sfax (south Tunisia)

Sites	% Gravel	% Coarse	% Medium sand	% Fine sand	% Silt/clay	Organic matter content (%)
Site 1	0	5.68	22.31	62.08	9.93	14.02
Site 2	0.03	3.62	25.82	63.12	7.41	11.03
Site 3	0.42	4.72	26.06	68.02	0.78	19.27
Site 4	0.11	7.52	27.63	61.17	3.57	18.11
Site 5	1.11	11.53	26.02	50.05	11.29	16.01
Site 6	4.71	12.97	24.19	38.72	19.41	8.54
Site 7	2.42	9.82	28.02	46.48	13.26	19.01
Site 8	4.82	10.93	28.30	41.81	14.14	6.72
Site 9	0.10	1.52	19.12	79.18	0.08	22.13
Site 10	0.09	2.54	38.15	57.19	2.03	17.14

Table 2

Surface area, the number of transects and replicates and stock levels (means ± SE) of *Polititapes aureus* in the shellfish production area of Sfax (south Tunisia)

Sites	Surface (ha)	% of total surface	Number of transects	Number of replicates	Mean density (ind. m^{-2}) ± SE	Mean biomass (g m^{-2}) ± SE
Site 1	505	12.08	18	130	0.92 ± 0.3	2.74 ± 1.0
Site 2	265	6.34	11	62	1.68 ± 0.7	6.12 ± 2.9
Site 3	146	3.49	12	50	0	0
Site 4	379	9.06	16	82	1.32 ± 0.4	5.38 ± 1.5
Site 5	535	12.79	24	168	0.90 ± 0.2	1.34 ± 0.3
Site 6	310	7.41	9	62	1.61 ± 0.4	3.08 ± 0.9
Site 7	960	22.95	11	72	3.83 ± 1.8	6.28 ± 1.9
Site 8	600	14.35	6	42	1.24 ± 0.5	4.07 ± 1.7
Site 9	87	2.08	8	72	1.0 ± 0.4	2.80 ± 1.0
Site 10	395	9.45	28	226	5.65 ± 1.3	11.88 ± 3.1
Total	4182	100	143	966	2.34 ± 0.4	5.28 ± 0.8

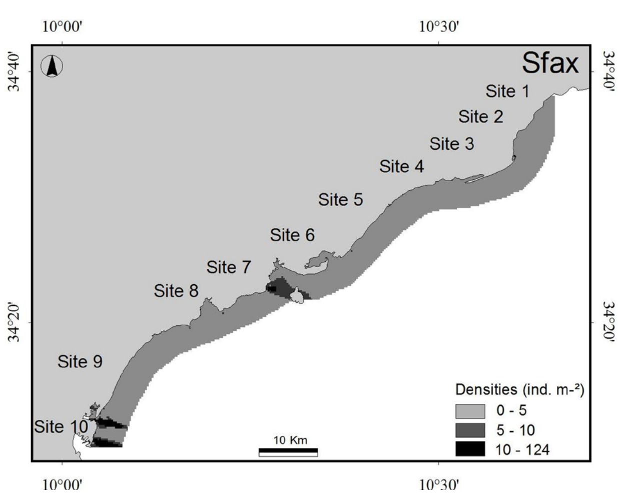

Figure 2

Polititapes aureus: spatial distribution of densities in the littoral zone of Sfax (south Tunisia)

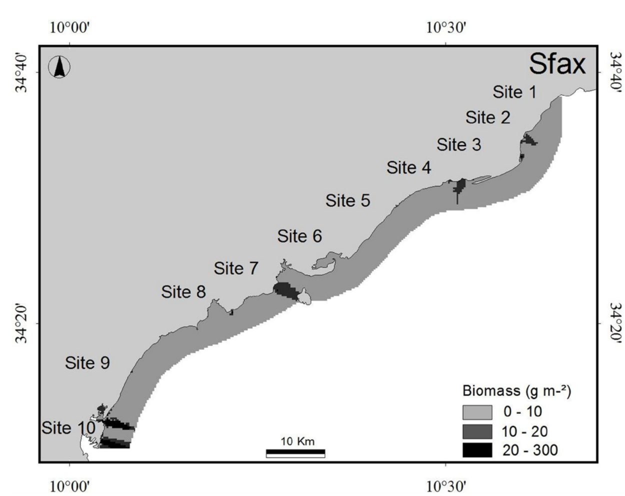

Figure 3

Polititapes aureus: spatial distribution of biomass in the littoral zone of Sfax (south Tunisia)

The mean density per replicate was 2.34 ± 0.4 ind. m^{-2}. In total, 565 specimens were harvested at the most interesting sites (sites 1, 2 and 4 to10) covering 1777 ha. No clams were found at site 3. The distribution of all mean density and biomass values are presented in Table 2.

There were clear differences in stock levels in the sampling area (Fig. 4). In terms of density, there was a significant difference between all sampling sites ($p < 0.05$) except for zone 1 for sites 2, 4, 5 and 6 ($p > 0.05$). As for abundance, the total biomass levels significantly varied between the sampling sites ($p < 0.05$). Spearman's correlation coefficient was applied to compare stock values for each site. The results showed strong a positive correlation between abundance and biomass (R > 0.84). In addition, cluster analysis (group average) of the sites, applied to compare similarities between the sites, identified a core group that spanned all sites (Fig. 5). The analysis of similarity tests showed that the aforementioned group and sites 7 and 10 differed significantly (global R greater than 0.9; $p < 0.05$). This was mainly due to *P. aureus* stocks being most abundant at sites 7 and 10 compared to the other sites.

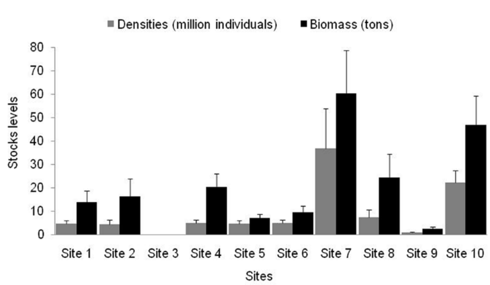

Figure 4

Polititapes aureus stock levels in the colonized zones and their standard errors (± SE) in the littoral zone of Sfax (south Tunisia)

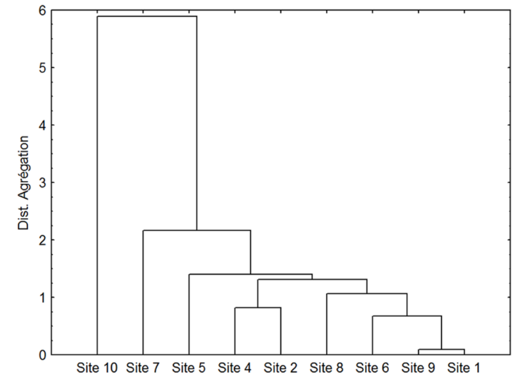

Figure 5

Polititapes aureus: similarity dendrograms for the colonized zones (average group) in the littoral zone of Sfax (south Tunisia)

3.3. Population structure

The size frequency distribution of *P. aureus* was determined at all sites (Table 3). The distribution of clams was analyzed with respect to their size, which varied markedly between the sites. The clam population was not uniformly distributed: the distribution pattern of smaller specimens was relatively heterogeneous throughout the study area, while larger individuals were more geographically restricted to sites 4 and 10. Mean sizes ranged from 18.41 ± 1.14 mm at site 5 to 26.10 ± 0.43 mm at site 2. In general, the sizes of specimens varied between 4.9 and 35.34 mm SL. Almost the entire population was assigned to size classes (15–30 mm), which represented 88% of all samples. Large sizes (> 30 mm) accounted for only 1.4% (Fig. 6). The shell length-weight relationship for all data (565 individuals) showed a positive correlation (R = 0.808; $p < 0.001$). The relationship was: $TW = 0.0004SL^{2.692}$ (Fig. 7).

Table 3

Polititapes aureus. Variability of minimum (Min.), maximum (Max), average (Mean), standard error (SE) and mode values of shell length (SL) in the shellfish production area of Sfax (south Tunisia), N (number of harvested specimens)

Sites	N	Min. SL (mm)	Max SL (mm)	Mean SL (mm)	SE	Mode
Site 1	30	8.30	29.15	23.82	0.86	27.00
Site 2	26	21.30	29.80	26.10	0.43	27.00
Site 3	0	0	0	0	0	0
Site 4	27	6.70	32.65	25.06	1.11	25.00
Site 5	38	6.70	27.10	18.41	1.14	10.00
Site 6	25	9.50	30.70	20.43	1.21	19.00
Site 7	69	8.15	30.25	19.16	0.79	24.00
Site 8	13	14.00	30.50	25.95	1.15	26.00
Site 9	18	17.50	29.45	23.85	0.93	27.00
Site 10	319	4.90	35.34	22.34	0.26	23.00

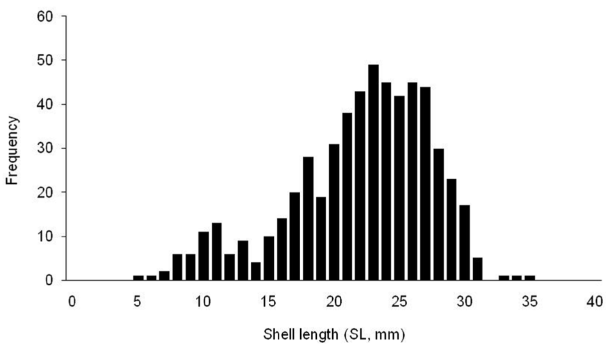

Figure 6

Polititapes aureus: length–frequency distribution from the Sfax coastline (south Tunisia)

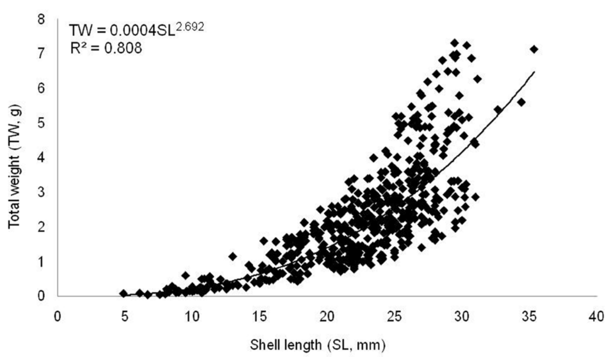

Figure 7

Polititapes aureus: relationship between shell length (SL) and total weight (TW) in the littoral zone of Sfax (south Tunisia)

3.4. Sex ratio

We identified 292 females (52%), 231 males (41%) and 42 individuals of indeterminate sex (7%) in the samples of clams. For both sexes, individuals presented wide range of sizes, both in terms of shell length (4.9–35.34 mm) and total weight (0.04–8.28 g). The overall sex ratio (F:M = 1.26:1) significantly deviated from parity (F:M = 1:1; χ^2-test, $p < 0.001$).

4. Discussion

The current study reports for the first time the spread of the clam *P. aureus* in the largest shellfish production area in the south of Tunisia by investigating the stock levels, population distribution, size structure and sex ratio. The findings of this study indicate an exceptional stock of this species, resulting in a remarkable biomass of 201 t of the total fresh weight and a high level of abundance, reaching over 91 million individuals. In general, this study was aimed at ascertaining whether the stock of *P. aureus* is large enough to start management of its stocks. Stock assessments of soft-bottom clams are scarce in the literature. The available information on the population stock assessment and distribution is insufficient. What is known about this species is limited to preliminary studies conducted in the Bou Ghrara Lagoon (64 t and a high level of abundance reaching over 156 million individuals; Derbali 2006). A similar gradient can be observed for another clam species (*R. decussatus*) with a total fresh weight of about 891 t and a high level of abundance reaching over 261.7 million individuals estimated in the same study area. Thus, our results showed that the variance within and between the sampling sites was large and confidence limits of biomass and density were correspondingly wide.

Stock mapping showed a fluctuation of *P. aureus* habitats in the colonized area. The stock appears to have varied considerably between the strata. The population seems to have been influenced by strong interactions at different levels (e.g. physicochemical, edaphic and hydrological factors operating in the study area). Stergiou et al. (1997) clarified that temperature and food resources are the most important factors affecting phenotypic differences in growth patterns and maximum size in different marine organisms. Elevated water temperatures and phytoplankton levels occurring in the sampling area may promote rapid growth rates in bivalve species. Due to the high water temperatures (12–27°C) and shallowness, salinity remains fairly stable throughout the year. The highest salinity was during summer (48 PSU) and often in winter (36 PSU). This result indicates that salinity remain high throughout the year. The maximum value recorded in this study is much higher than that recorded in Lake Vouliagmeni where salinity varied between 17 and 18 PSU (Chintiroglou et al. 2000). The increased salinity appears to have a major effect on the composition of the fauna and has reduced their standing crop (Fishar 2000; El-Shabrawy 2001).

The clam population appears to have been strongly affected by abiotic factors. During the present study, hydrodynamic conditions were found to be similar within the sampling area and it can be assumed that the relative population growth is affected by other environmental parameters such as sediment type, organic matter content, burrowing behavior of bivalve species and their subsequent strategies to counter dislocation and avoid predation. In fact, some interesting connections were detected between environmental conditions and bivalve behavior. Elevated water temperatures and phytoplankton levels may contribute to rapid growth rates in many local bivalve species (Dridiet al. 2007; Enríquez-Díaz et al. 2009; Sobral & Widdows 2014).

Additional mechanisms structuring clam populations include soft bottoms and organic matter content. The high diversity of shellfish species is particularly interesting when considering the relative organic matter content (11–22.13%; Table 1), depth (0–1 m) and silty-sand bottom. These factors can provide ecological conditions that are able to maintain highly diverse reef communities in the colonized area. Indeed, the absence of the clam *P. aureus* in the area characterized by sandy substrate (site 3) prevented the species from reproducing. Except for site 3, the clam *P. aureus* is omnipresent throughout the study area. The colonized sites maintain a high diversity compared to other areas of southern Tunisian waters, where only a few species (*Gibbula ardens*, *Cerithium vulgatum*, *Cerithium scabridum* and *Bittium reticulatum*) were found on rock and hard substrates. This low diversity seems to be attributable primarily to seabed characteristics. This hypothesis may be supported by the relationship between the sedentary nature of some shellfish species such as *P. aureus*and the substrate.

As regards faunal diversity and substrate types, this species was more frequent and abundant in areas sheltered by seagrass *C. nodosa* and *Z. noltei*, covering more than 65% of the silty- sandy seabed. Heterogeneity of the geographical distribution of *P. aureus* was found to be significantly correlated with the distribution of seagrass. This positive correlation was probably related to the main organic source provided by *C. nodosa*. The *C. nodosa* detritus was the richest in organic carbon and was the dominant source of primary organic matter. This is a result indicating that the colonized sites currently provide good conditions for the proliferation of clams. It can thus be concluded that this species improves the food resources and dissolved oxygen levels. The same conclusion was reached by Vilela (1950) for natural populations of *R. decussatus* in Portugal. Several studies have confirmed the correlation between patterns of community structure and primary production. In particular, local abundance and biomass of filter-feeders was correlated with both intertidal productivity and nearshore primary productivity (Menge & Olson 1990; Bustamante et al. 1995). Other parameters such as light and tides may also contribute (Drummond et al. 2006; da Costa et al. 2013). Seasonal metabolic activities in bivalves are usually the result of interactions between food availability, environmental conditions, growth and gametogenic cycles (Gabbott 1983; Derbali 2011).

The results on the distribution of clams may explain the dominance of this species in these particular water bodies. They also show the importance of environmental factors in controlling the density and subsequently the biomass of shellfish assemblages. This study showed that the clam population was not uniformly distributed: smaller specimens are dispersed in a relatively heterogeneous pattern throughout the study area, while larger individuals were more geographically restricted to sites 4 and 10. The mean sizes ranged from 18.41 ± 1.14 mm at site 5 to 26.10 ± 0.43 mm at site 2. The size range in populations varied between the sites. Size ranges of some bivalve species vary greatly across studies, complicating comparisons between species from different geographical areas. Size fluctuations may be related to abiotic factors that vary from one habitat to another. The growth, development and survival of bivalves are generally controlled by physical and chemical parameters, temperature in particular. This conclusion is the same as that given by Laing et al. (1987) and Jara-Jara et al. (1997) for *R. decussatus* from Conwy (UK) and Galicia (NW Spain), respectively.

In this study, the clam population exhibited characteristics similar to those described for other bivalve species. The condition factor estimated from the length–weight relationship can provide an indication of the "well-being" of a given species and can be an indicator of food abundance for a species in a given area or time (Mzighani 2005). For many bivalve species, several environmental factors are known to affect shell morphology and relative proportions, such as latitude (Beukema & Meehan 1985), depth (Claxton et al. 1998), shore and tidal levels (Franz 1993), water currents and turbulence (Hinch & Bailey 1988; Fuiman et al. 1999), wave exposure (Akester & Martel 2000), type of bottom (Claxton et al. 1998) and sediment type (Newell & Hidu 1982). Thereafter, Tlig-Zouari et al. (2009, 2010) and Derbali et al. (2011, 2012) from Tunisia argued that these parameters are known to be potentially related to environmental conditions and vary within bivalve species and between localities.

Sexual maturity of bivalves is classified according to microscopic features, such as the presence of gametes in gonads and the degree of their development, though sometimes it can be determined from macroscopic features, i.e. the appearance and colour of gonads (Lucas 1965). Similar trends were observed in other bivalve families, such as Pectenidae, Mytilidae, Limidae and Cardiidae, both sex and the degree of gonad development can be determined based on the color of gonads (Lubet 1959; Derbali et al. 2009a,b; 2012, 2014). Accordingly, the overall sex ratio (F:M = 1.26:1) significantly deviate from parity (F:M = 1:1;

$p < 0.001$). The same sequence of events was reported for the surf clam *Mactra stultorum* from the Gulf of Gabes (south Tunisia). On the other hand, different trends were found for the clam *R. decussatus* from the present study area. Elsewhere, similar trends were observed for *P. aureus* from Lake Qarun in Egypt (Kandeel 2017).

Finally, this study provides crucial information on the spread of potentially commercially valuable *P. aureus* in the major shellfish production area in Tunisia by investigating the stock size, distribution, size structure and sexuality. In addition, this paper is the first attempt to provide new information on its stock levels and parameters responsible for its variability. The distribution of *P. areus* was found to be strongly affected by many abiotic factors. For any production purposes, further work is required to investigate other biological parameters to accurately monitor the exploitation of clams. The adoption and implementation of rules, such as limiting the size of clams, will be required to protect this new exploitable fishery resource similar to *R. decussates* natural populations.

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