COMPARATIVE CHARACTERIZATION OF THE SPAWNING ENVIRONMENTS OF EUROPEAN ANCHOVY, *ENGRAULIS ENCRASICOLUS*, AND ROUND SARDINELLA, *SARDINELLA AURITA* (ACTINOPTERYGII: CLUPEIFORMES) IN THE EASTERN COAST OF TUNISIA

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Background. The distribution of early ontogenic stages of small pelagic fishes is important for understanding the dynamics of their fluctuating populations. To fill a gap in the knowledge we conducted a comparative analysis on the distribution of eggs and larvae of the exploited populations of European anchovy, *Engraulis encrasicolus* (L.) and round sardinella, *Sardinella aurita* (Valanciennes, 1847), in the eastern coast of Tunisia.
Materials and methods. A multidisciplinary survey was carried out in the summer of 2008, from June 23 through July 9. Samples were taken over a grid of 71 stations. Temperature and salinity profiles were recorded at each station by CTD casts and water samples were taken by means of a Rosette equipped with Niskin bottles. Ichthyoplankton was sampled by oblique tows with a Bongo net of 60 cm mouth diameter and 335-µm mesh nets. To specify the preferred spawning environment the quotient values were estimated for each variable.
Results. The eggs and larvae of both species represented an important proportion of the ichthyoplankton, around 50% for each ontogenic stage. Anchovy mainly spawned in the shelf edge, north-east of Kuriate Island, with a minor spawning area near Cape Bon and Kelibia. The main spawning area of the round sardinella was in the warmer waters. It was located near the major spawning ground of anchovy. Anchovy eggs were scarce in the coastal zones and in the south of the study area, whereas round sardinella eggs were scarce in the north. The main spawning grounds of anchovy coincided with the area of higher zooplankton abundance while round sardinella spawning grounds correspond with those areas of highest chlorophyll-a and zooplankton. This distribution suggests a propensity for food availability, for both adults and larvae, as the location of spawning grounds of these species.
Conclusion. Both anchovy and sardinella spawned mainly in deeper waters within the limits of the continental shelf. Each species appeared to have a specific strategy for spawning that was influenced by physical and biological variables.

Keywords: anchovy, round sardinella, Ionian Sea, spawning environments

INTRODUCTION

Among the small pelagic species that inhabit the eastern coast of Tunisia, European anchovy, *Engraulis encrasicolus* (Linnaeus, 1758) and round sardinella, *Sardinella aurita* (Valanciennes, 1847), stand out for their abundance and commercial importance in fisheries. Along the Tunisian coast, spawning of anchovy and round sardinella occurs mainly in summer (Gaamour unpublished[**], Khemiri unpublished[***], Zarrad unpublished[****]). More specifical-

ly, the anchovy spawning extends from April through September and peaks in June and July. Round sardinella spawns from May to September with a peak in July–August (Tsikliras et al. 2010), which after that of *E. encrasicolus*. The competition with anchovy and sardine (European pilchard), *Sardina pilchardus* (Walbaum, 1792), two dominant species competing with each other for space and food (Larrañeta 1960, Demir 1965), could be responsible for the short reproductive phase of *S. aurita*, since it ends

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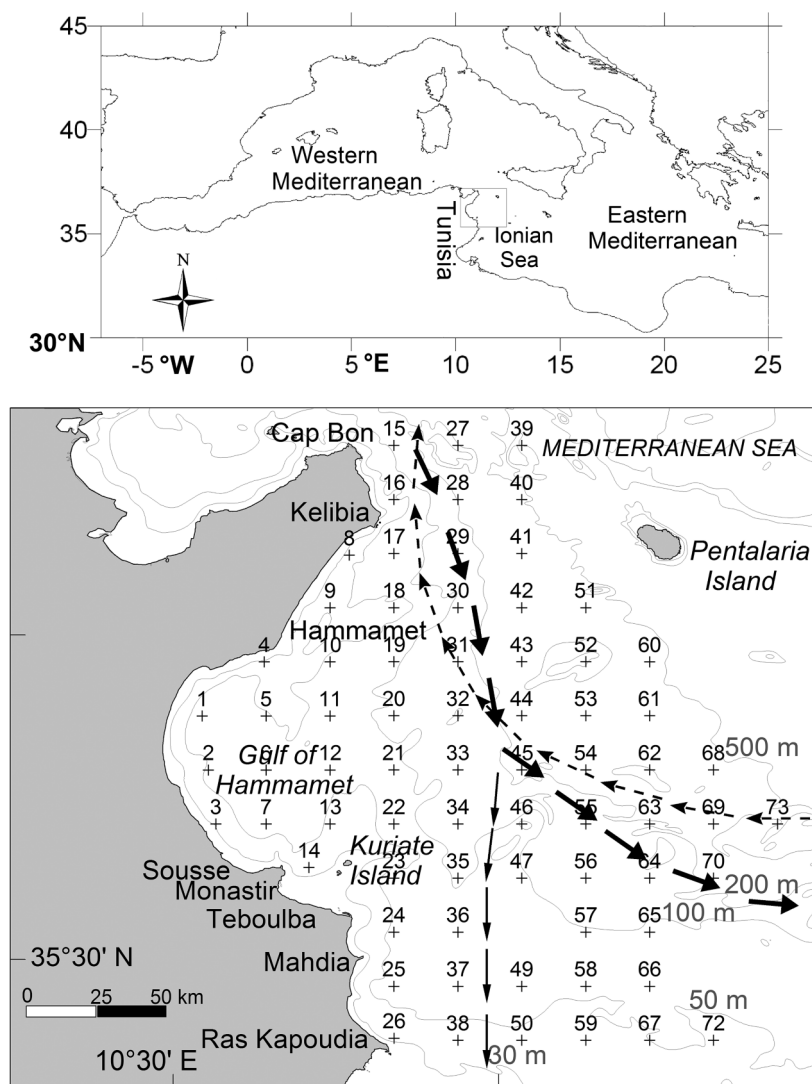
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by the time the sardine spawning season begins, when water temperatures start to decline (Aldebert and Tournier 1971, Palomera et al. 2007).

The eastern coast of Tunisia is located south of the Sicilian Channel constituting the boundary between the western- and eastern Mediterranean (Fig. 1). The Strait of Sicily plays a key role in the circulation between these two Mediterranean basins (Astraldi et al. 1999). The surface Atlantic waters coming from the Strait of Gibraltar flow close to the northern Tunisian coast and the Levantine intermediate waters fills the whole bottom section of the strait (Grazoli and Maillard 1979, Lermusiaux and Robinson 2001, Astraldi et al. 2002, Hamad et al. 2005). The low salinity of PSS 37.03 to 37.15 (Sammari et al. 1999) of surface Atlantic waters detected off Tunisian coasts derives from the eastward current of Atlantic waters flowing through the Algerian Basin (Béthoux 1980, Millot 1999). Once it reaches the Strait of Sicily it splits into two dis-tinct currents: one flowing near Cape Bon and the other, very narrow, flowing over the Sicilian shelf close to the south-western Sicilian coast (Astraldi et al. 1996). The Levantine intermediate waters constitute the main compo-nent of the intermediate current returning to the Atlantic Ocean and is characterized by saltier (PPS 38.7) and warmer water masses (> 14°C) (Guibout 1987). It is com-posed of two quite distinct water types (Astraldi et al. 1996). One of them is the classical Levantine intermediate waters, which fills the deeper parts of both sections and has its core at about 300 m. The other water mass is cold-er and recognizably closer to the bottom in the Tunisian side of the Strait.

There is no river runoff in the eastern coast of Tunisia. Downwelling occurs along this coast on the opposite side of the Strait of Sicily (Agostini and Bakun 2002). Accordingly, the Ekman field is directed so as to trans-port water from the upwelling (enrichment) zone towards

Fig. 1. Map of the Tunisian coastline showing location of the study area, pre-vailing circulations (→: Atlantic waters and ---›: Levantine intermediate waters, according to Lermusiaux and Robinson 2001, Astraldi et al. 2002, Hamad et al. 2005), and the sampling stations (+)

** Gaamour A. 1999. La sardinelle ronde (Sardinella aurita Valenciennes, 1847) dans les eaux tunisiennes: Reproduction, croissance et pêche dans la région du Cape Bon. PhD thesis, Bretagne Occidentale University, France.
*** Khemiri S. 2006. Reproduction, âge et croissance de trois espèces de téléostéens pélagiques des côtes tunisiennes: Engraulis encrasicolus, Sardina pilchardus et Boops boops. PhD thesis, Pierre and Marie Curie University, France.
**** Zarrad R. 2007. Distributions spatio-temporelles des œufs et des larves de l'anchois *Engraulis encrasicolus*, de la sardinelle *Sardinella aurita* et de la sardine *Sardina pilchardus* dans le golfe de Tunis et relations avec les paramètres environnementaux. PhD thesis, Carthage University, Tunisia.

the downwelling (concentration) zone along the east coast of Tunisia.

The knowledge on the variable distribution of eggs and larvae of small pelagic fishes is considered important for understanding the dynamics of their fluctuating populations. However, historical data on the spawning habitats of anchovy and round sardinella off southern coasts of Mediterranean including the Tunisian eastern coast are scarce. No recent data are available on the anchovy eggs distribution and the only single paper was published over 30 years ago by Ktari-Chakroun (1979) who sampled scattered stations along the tuniso-sard and siculo-tunisien channels. There have been some recent contributions dealing with anchovy egg and larval distribution in adjacent areas (Cuttitta et al. 2003, García Lafuente et al. 2005, Zarrad et al. 2006, Zarrad unpublished*) and on the early life stages of round sardinella (Regner 1977, Tsikliras et al. 2005, Zarrad unpublished*).

In view of the limited information on the spawning ecology of anchovy and round sardinella along the eastern coast of Tunisia and the importance of the fisheries, the objects of this study were to localize and to characterize the spawning areas of each species. We also discuss the hypotheses for similarities and differences in the preferred spawning environments.

MATERIALS AND METHODS

Sampling methods. The study area was located off the eastern coast of Tunisia (Ionian Sea, Eastern Mediterranean). Its western part covered the Gulf of Hammamet, extending from Kelibia in the north to Ras Kapoudia in the south, including also the Island of Kuriate (Fig. 1).

A multidisciplinary survey, named ESPOIRS 9, was carried out from 23 June to 9 July, on board of *R/V HANNIBAL*. Samples were taken over a grid of 71 stations (10 × 10 miles equidistant) distributed over the shelf and slope (Fig. 1). Temperature and salinity profiles were recorded at each station by CTD casts using a Sea Bird 911+. Water samples were taken by means of a Rosette equipped with Niskin bottles, to analyse chlorophyll-a. The samples for chlorophyll-a were filtered through Whatman GF/C glass fibre filters, which were frozen at –20°C. Chlorophyll-a was determined using the conventional technique proposed by Lorenzen (1967). After 90% acetone extraction, chlorophyll-a (Chl-a) was measured colorimetrically using a Jenway model 6405UV/Visible spectrophotometer.

Ichthyoplankton was sampled by oblique tows with a Bongo net of 60 cm mouth diameter, fitted with 335-μm mesh nets. The vessel speed was 1.54 m · s^{-1} (3 knots). Maximum sampled depth was 100 m, wherever possible, or about 5 m above the bottom where the water was shallower. The net mouth was fitted with a Hydro-Bios flowmeter to estimate the volume of the water filtered. Plankton samples were preserved in 4% buffered formalin. Zooplankton was sampled by plankton net associated with the Bongo net. The mesh size was 100 μm and the mouth diameter was 25 cm. A flowmeter was attached in

the mouth to measure the volume of filtered water. The samples were stored in a frozen state at –20°C. The dry weight was determined by drying the aliquot of the zooplankton sample in an electric oven at a temperature of 60ºC during 72 h. The values of zooplankton dry weight (ZDW) were expressed in mg per 10 m^2. In the laboratory, anchovy eggs and larvae were sorted and counted with the aid of a binocular microscope.

Data analysis. The stability of water column (SWC = Dd · Dz^{-3}) was calculated as the density (kg · m^{-3}) difference (Dd) between the surface and the maximum sampling depth divided by the depth (m) difference (Dz) between surface and maximum sampling depth (Peterson et al. 1988). The mixed layer depth (MLD) is defined as the depth (m) of the layer immediately below the sea surface at which the water temperature is 0.5°C lower than that of surface waters (Nishikawa and Yasuda et al. 2008).
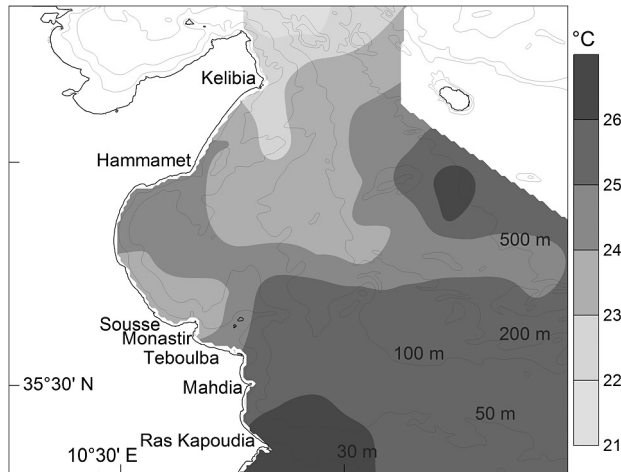
The abundance values of eggs and larvae at each station were standardised to numbers beneath 10 m^2 of sea surface area as described by Smith and Richardson (1977). Environmental variables, egg and larval abundance distributions were mapped using the Surfer software package (Golden Software Inc.), applying the kriging interpolation method and the isotropic linear variogram model.

The abundance of eggs and larvae by species found for each environmental variable class (e.g., 0.5°C for temperature, 0.1 for salinity) was expressed as a percentage of the total abundance values of all stations sampled (Eggs or larvae). This value was then divided by the frequency of occurrence of that particular environmental variable within the class (van der Lingen et al. 2001, Twatwa et al. 2005). The number of classes in every environmental variable ensured that maximum occurrence per class did not exceed 25% of all measurements. Quotient values (Q_C) were plotted against environmental variables, reflecting 'selection' ($Q_C > 1$) or 'avoidance' ($Q_C < 1$) for a specific environmental variable range (category). Quotients were smoothed using three-point running means centred on the second datum (van der Lingen et al. 2001). Moreover, a non-parametric Kolmogorov–Smirnov test for goodness of fit (Zar 2010) was used to compare the cumulative frequency distribution of anchovy eggs or larvae per category of environmental variable against the distribution curves of that environmental variable. The null hypothesis was that no difference existed between them (i.e., observed anchovy distribution was random with respect to that environmental variable).
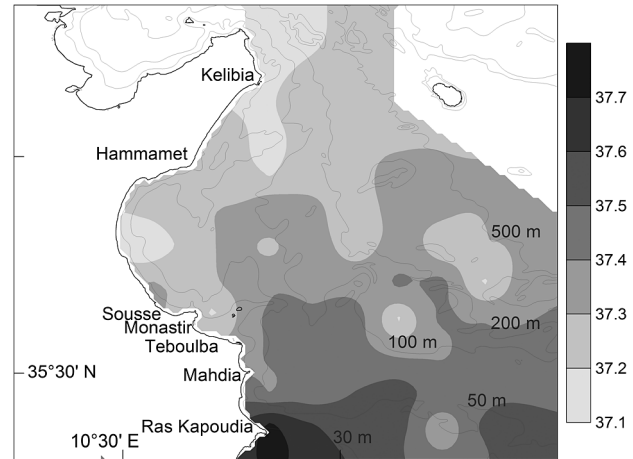
RESULTS

Environmental conditions. Positive gradients of sea surface temperature (SST) and sea surface salinity (SSS) were observed from north (off Cape Bon) to south (off Mahdia), where highest SST and SSS were recorded (Figs. 2 and 3). Half of the southern part of the study area was dominated by warm waters. Higher temperatures (> 26°C) were observed offshore, south of Pentalaria Island. The mean SST was 24.55 ± 1.11°C, with a difference between

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Fig. 2. Horizontal distribution of sea surface temperature along the eastern coast of Tunisia

Fig. 3. Horizontal distribution of sea surface salinity (PSS) along the eastern coast of Tunisia
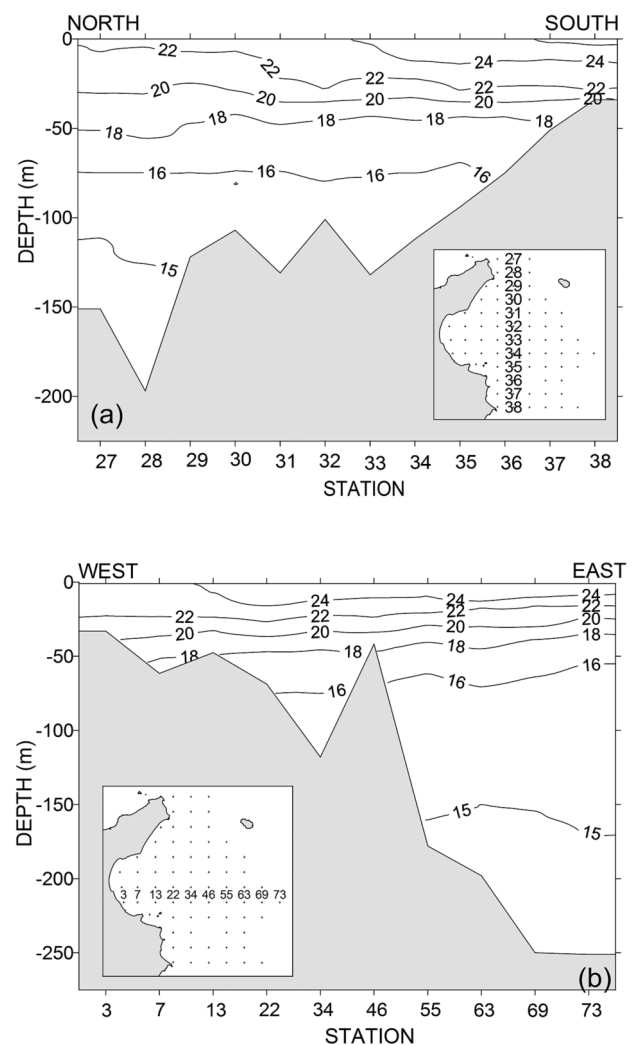
minimum and maximum values of 4.64°C. The SSS ranged between PSS 37.12 and 37.73. Vertical distribution of temperature (transects: north-south and west-east) showed highly stratified waters (Fig. 4). The thermocline was found between 40 and 50 m, with temperatures above 18°C in the upper layers and between 14 and 15°C in deeper waters, near the bottom (200 to 250 m). The stability of water column was higher in the south and near the coast (Fig. 5). However, the mixed layer depth showed a horizontal heterogeneity (Fig. 6).

The horizontal distribution of the chlorophyll-a at surface (Fig. 7) showed high heterogeneity. The highest concentrations (> 1.2 mg · m^{-3}) were recorded in the north east of Monastir and Mahdia. The average value was 0.49 mg · m^{-3} (±0.31). The highest zooplankton biomass patches (> 30 g per 10 m^2) were found near the coast Mahdia and offshore Monastir (Fig. 8), with concentration values over 40 g per 10 m^2. A moderate zooplankton biomass (> 20 g per 10 m^2) occurred in the area east of Kelibia. Nevertheless, the majority of the study area was dominated by lower values (< 10 g per 10 m^2).

Egg and larval abundances and distributions. Anchovy and round sardinella eggs represented 30% (mean 163 eggs per 10 m^2; SD = 631) and 23% (mean 129 eggs per 10 m^2; SD = 381) of the total fish eggs of the ichthyoplankton catch, respectively. Larvae were at the proportion of 27% (mean 86 larvae per 10 m^2; SD = 195) for anchovy and 23% (mean 73 larvae per 10 m^2; SD = 151) for round sardinella.

The highest concentration of anchovy eggs (4935 eggs per 10 m^2) was found over the shelf edge northeast of Kuriate Island at 70 miles from the coast (Fig. 9a). The next highest egg concentration (> 1000 eggs per 10 m^2) were found off the coasts between Cape Bon and Kelibia and east of Hammamet. Anchovy egg abundance were much lower or practically null in the coastal areas and in the southern part of the study area where the see was deeper than 100 m. Round sardinella eggs were practically nonexistent in the northern part of the study area (Fig. 9b).

Fig. 4. Vertical distribution of temperature (°C), (a) north–south and (b) west–east along the eastern coast of Tunisia

Higher concentrations of round sardinella eggs (> 1000 eggs per 10 m^2) were located east of Kuriate Island (depths 157 to 182 m), near the stations where highest anchovy eggs values were found. The second most abundant area (> 500 eggs per 10 m^2) was found in the east of Mahdia, 25 miles offshore.

Anchovy larvae were mostly concentrated northeast of Kuriate Island (> 1000 per 10 m^2) (Fig. 10a). They were scarce to absent near the coastline. Round sardinella larvae were concentrated (> 500 larvae per 10 m^2) north of Sousse (in the Gulf of Hammamet) and south-east of Teboulba (Fig. 10b). Larvae of both species were scarce or absent in the northern part of the study area, as well as in offshore waters beyond 220 m depth.

Egg and larval distributions and environmental conditions. The anchovy and sardinella selected (Egg quotient values > 1) for spawning areas over depths greater than 120 m (Fig. 11, Table 1), and with the same ranges of SSS and SWC. The sardinella spawned only in warmer waters in contrast with anchovy that could spawn in both colder and warmer waters. For the larvae, the quotient values (Fig. 12) showed similarities between both species for the SST, SSS, SWC, and Chl-a. The

Kolmogorov–Smirnov analysis indicated that anchovy eggs were not random with respect to SWC, MLD, and ZDW (Table 1). The sardinella eggs were not random with depth, SST, and ZDW. The larvae of both species were not random for depth. Significant differences were shown for anchovy larvae with MLD and Chl-a and for sardinella larvae for depth and SST.

DISCUSSION

The typical summer season hydrographic conditions observed during the studied period, with stratified waters and relatively high SST (mean = 24.55°C) correspond to the anchovy and round sardinella spawning scenarios described by several authors in other Mediterranean areas (Palomera and Sabatés 1990, Gaamour unpublished[*], Khemiri unpublished[*], Zarrad unpublished[*]). The positive gradient of SST and SSS from north to south seems to be related to the entrance of Atlantic waters at Cape Bon, as showed by Brandhost (1977) and Sammari et al. (1999). This current can form a frontal zone between two water masses with different densities; the surface waters of recent Atlantic origin, less saline, and the already modified and saltier surface Mediterranean waters that can
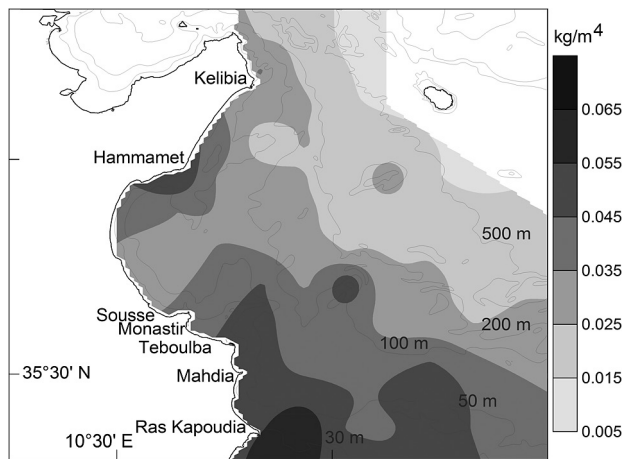

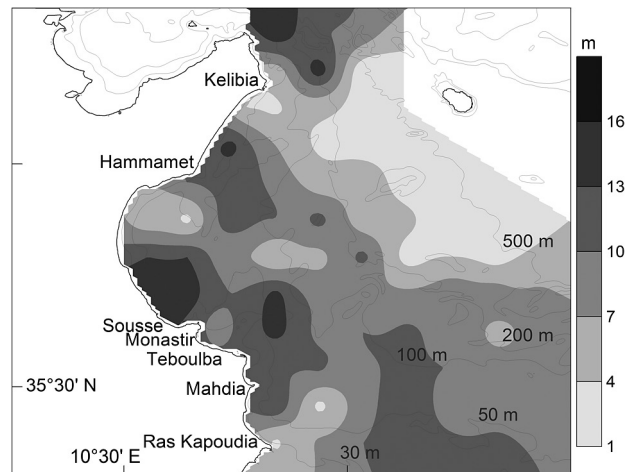**Fig. 5.** Stability of water column along the eastern coast of Tunisia

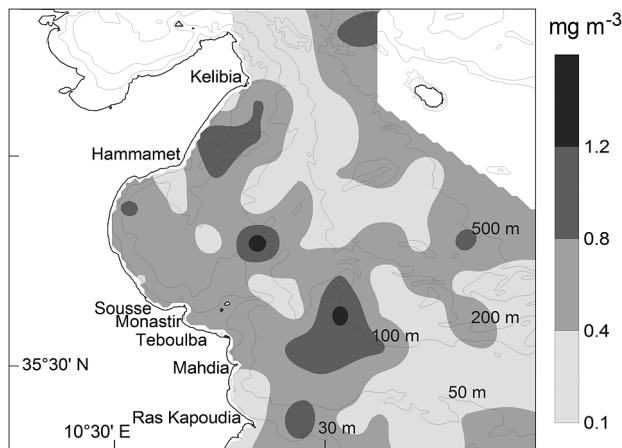**Fig. 6.** Mixed layer depth along the eastern coast of Tunisia

Fig. 7. Horizontal distribution of chlorophyll-a at surface along the eastern coast of Tunisia
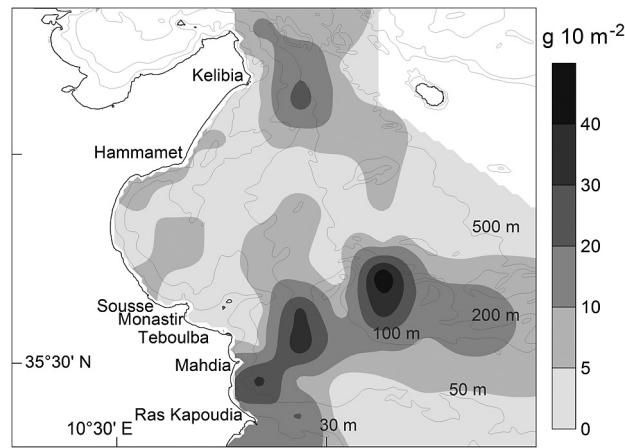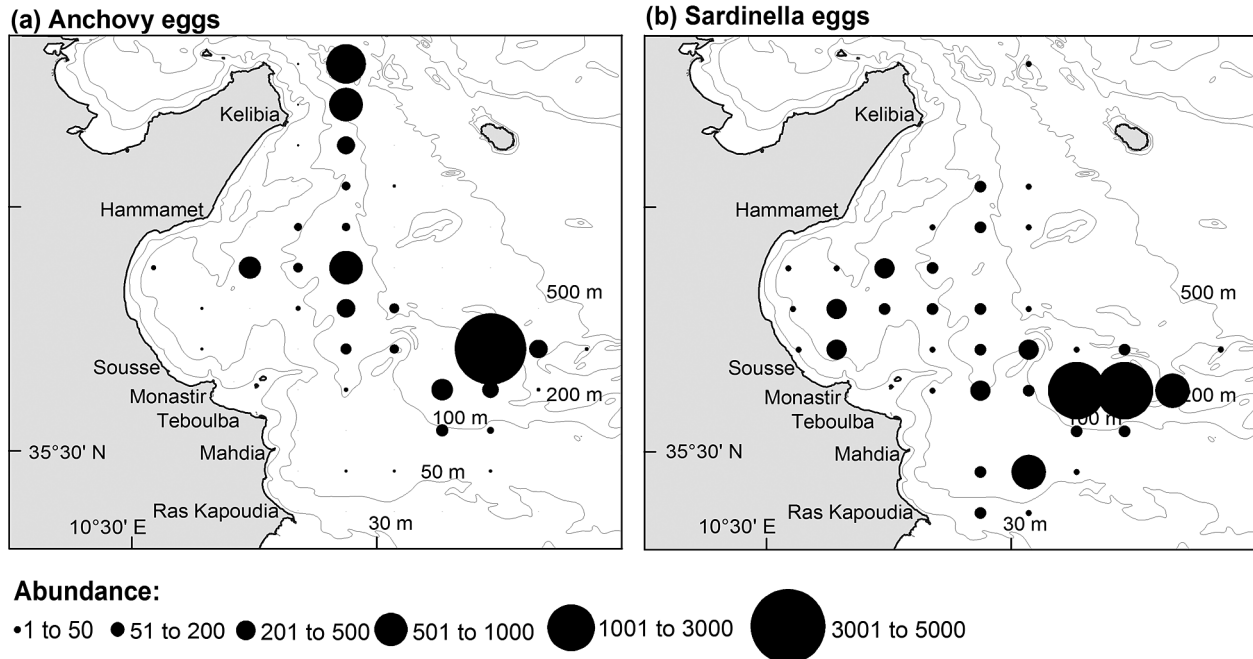

Fig. 8. Horizontal distribution of zooplankton dry weight along the eastern coast of Tunisia
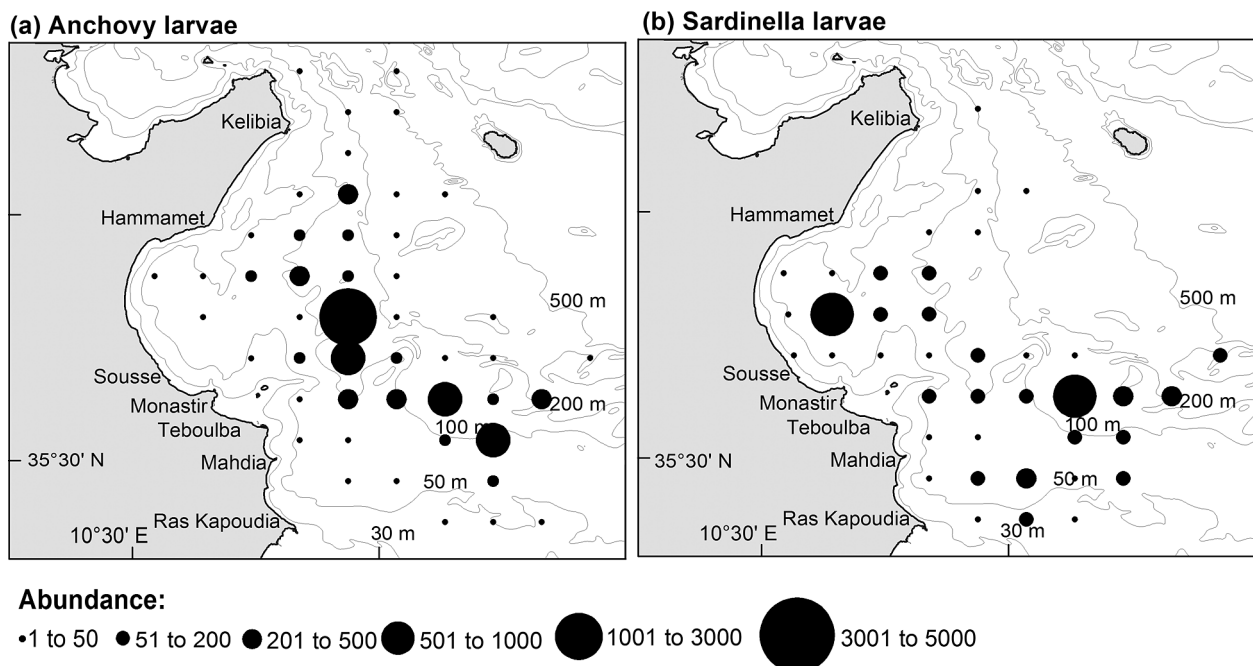
[*] See footnote on page 10.

determine the spatial distribution of fish larvae communities, as observed in different zones of western and central Mediterranean (Cuttitta et al 2003, Alemany et al. 2006).

The high proportions represented by anchovy and round sardinella eggs and larvae in the analyzed samples (around 50%) are in agreement with previous studies carried out in Tunisian waters (Zarrad et al. 2006, Zarrad unpublished[*]), and reflect the abundance of these species in the Tunisian waters.

The main spawning area of sardinella localized in the southern part of the study area witch seems to be a selection of an area with warmer waters. However, the anchovy can spawn in different water temperatures and had two main spawning areas in the north and in the south. The spawning of the round sardinella in the warmest period of the year (Gaamour unpublished[*], Khemiri unpublished[*]) and the preference for the warmest areas for spawning can be explained by the tropical origin of this species. This is

(a) Anchovy eggs

(b) Sardinella eggs

Abundance:
• 1 to 50   ● 51 to 200   ● 201 to 500   ● 501 to 1000   ● 1001 to 3000   ● 3001 to 5000

Fig. 9. Horizontal distribution of (a) anchovy, *Engraulis encrasicolus*, and (b) round sardinella, *Sardinella aurita*, eggs along the eastern coast of Tunisia (expressed in egg number per 10 m^2)

(a) Anchovy larvae

(b) Sardinella larvae

Abundance:
• 1 to 50   ● 51 to 200   ● 201 to 500   ● 501 to 1000   ● 1001 to 3000   ● 3001 to 5000
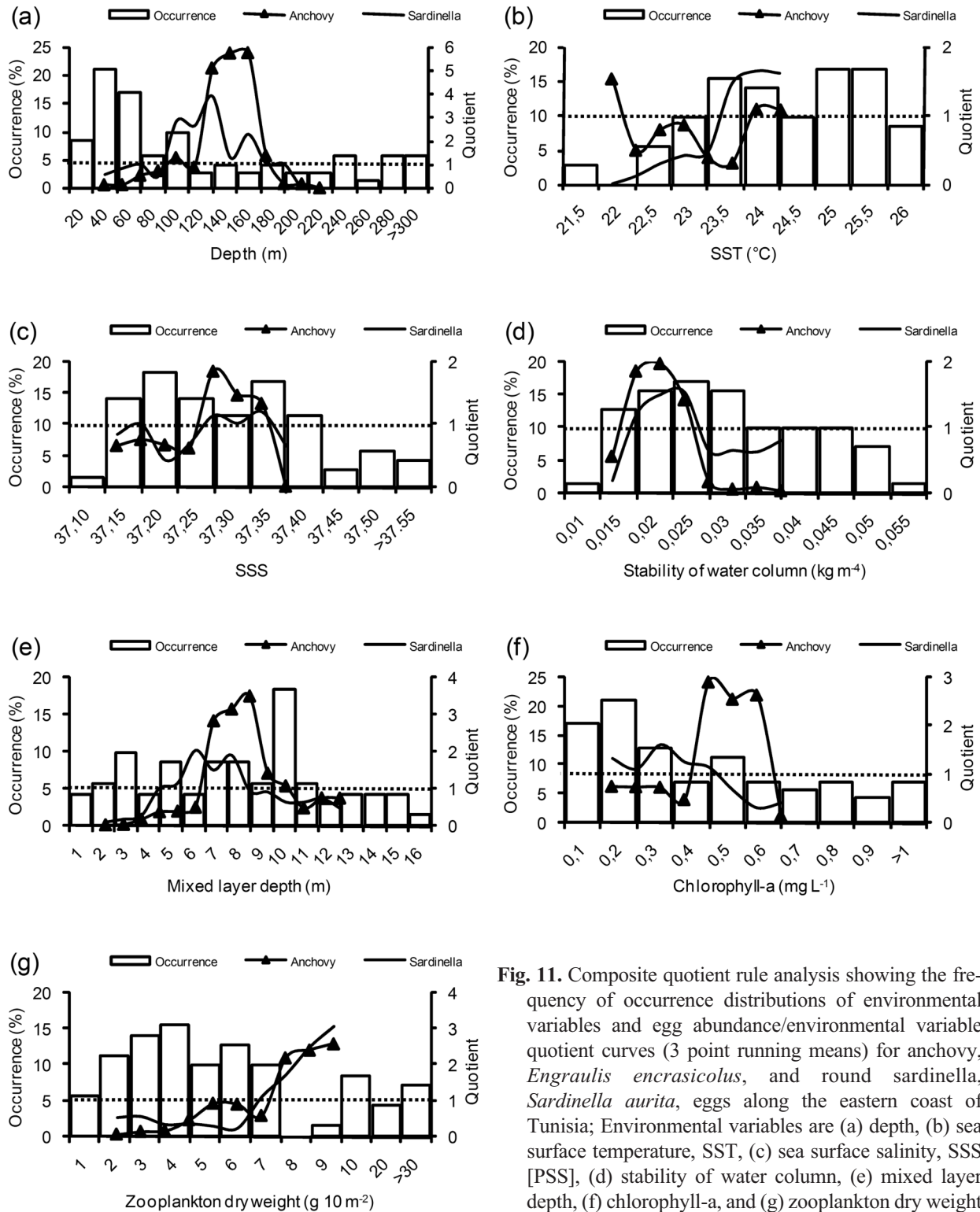
Fig. 10. Horizontal distribution of (a) anchovy, *Engraulis encrasicolus*, and (b) round sardinella, *Sardinella aurita*, larvae along the eastern coast of Tunisia (expressed in number of larvae per 10 m^2)

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* See footnote on page 10.

in agreement with the other observations reported in the Mediterranean by Palomera and Sabatés (1990) and Palomera et al. (2007) and in the NE Atlantic by Ettahiri et al. (2003) and Berraho et al. (2005). The latter authors showed that, in the southern coast of Morocco, the intense spawning of anchovy was in the north and in the south but for sardinella it was only on the south by the effect of water temperature.
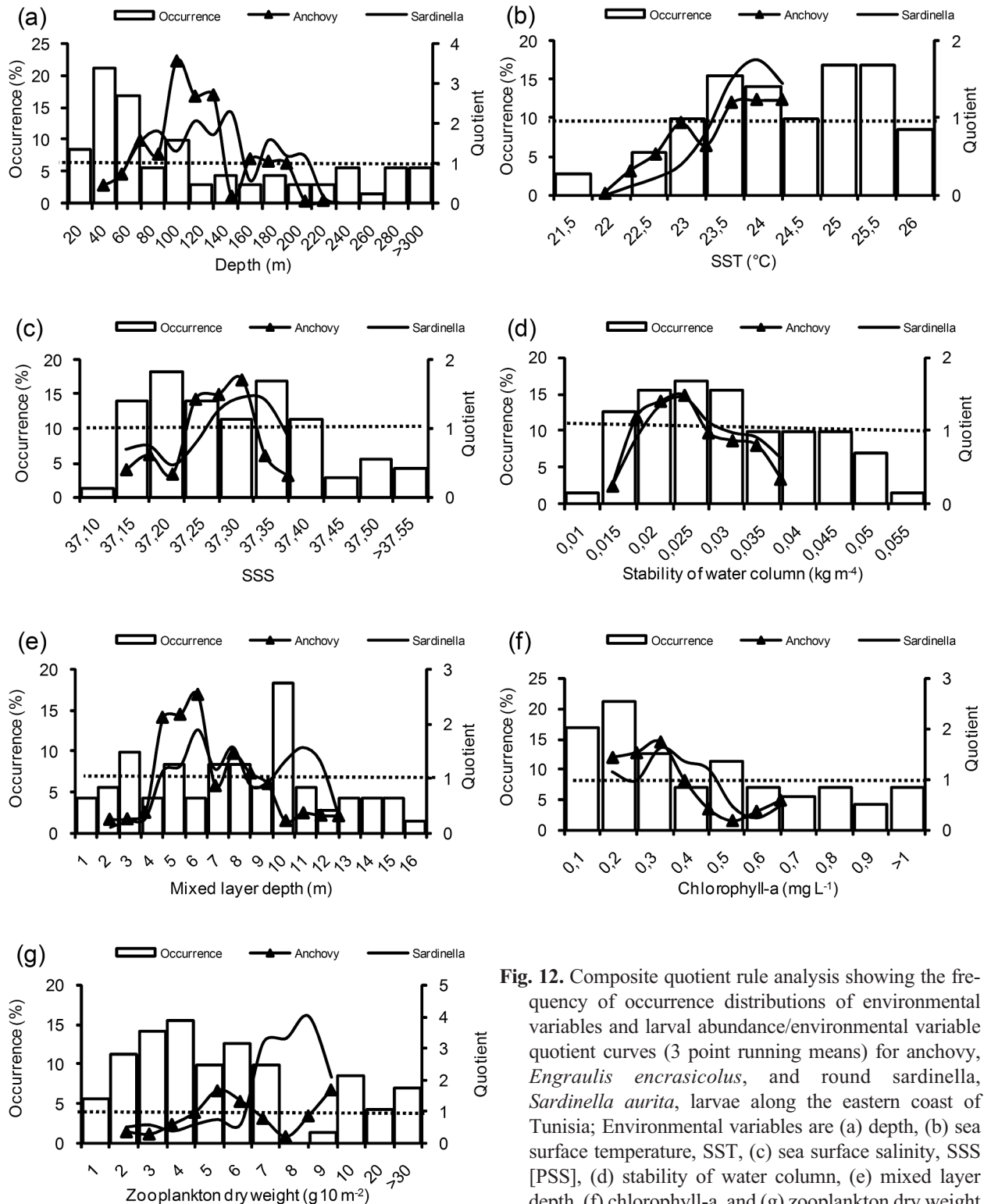
However, in spite of this difference in the spatial distribution of the early life stages of both species there are some similarities. Both the anchovy and the sardinella spawned mainly over the shelf, as expected in these small pelagics, but showed higher concentrations near the shelf edge. This could be related to the high prey concentration. Indeed, the main spawning grounds of anchovy overlapped with the areas of high zooplankton biomass and those of the round sardinella

Fig. 11. Composite quotient rule analysis showing the frequency of occurrence distributions of environmental variables and egg abundance/environmental variable quotient curves (3 point running means) for anchovy, *Engraulis encrasicolus*, and round sardinella, *Sardinella aurita*, eggs along the eastern coast of Tunisia; Environmental variables are (a) depth, (b) sea surface temperature, SST, (c) sea surface salinity, SSS [PSS], (d) stability of water column, (e) mixed layer depth, (f) chlorophyll-a, and (g) zooplankton dry weight

overlap with areas of high values of both chlorophyll-a and zooplankton. These distributions could be merely a result of adult distributions since they are plankton feeders (Tudela and Palomera 1995, 1997, Coombs et al. 1997, Plounevez and Champalbert 2000, Tsikliras et al. 2005). However, they could also identify a spawning strategy directed to maximize larval survival through increasing food availability for larvae (Tudela et al. 2002, Morote et al. 2008).

In the Mediterranean Sea, the spawning habitats of anchovy seem to be confined to shelf edges, where various kinds of enrichment process may occur (Palomera and Sabatés 1990, Palomera 1992, Olivar et al. 2001, Cuttitta et al. 2006). In the Bay of Biscay, anchovy select stable habitats related to river plumes, shelf edge fronts, and oceanic eddies, where increased biological production potentially occurs (Motos et al. 1996). We note that no

Fig. 12. Composite quotient rule analysis showing the frequency of occurrence distributions of environmental variables and larval abundance/environmental variable quotient curves (3 point running means) for anchovy, *Engraulis encrasicolus*, and round sardinella, *Sardinella aurita*, larvae along the eastern coast of Tunisia; Environmental variables are (a) depth, (b) sea surface temperature, SST, (c) sea surface salinity, SSS [PSS], (d) stability of water column, (e) mixed layer depth, (f) chlorophyll-a, and (g) zooplankton dry weight

Table 1

Results of the quotient values analysis and the Kolmogorov–Smirnov (KS)
to test the null hypothesis that the distrubution of eggs and larvae of *Engraulis encrasicolus*
and *Sardinella aurita* in the eastern coast of Tunisia is random with respect to the environmental variables

DS	EV [unit]	df	*Engraulis encrasicolus*			*Sardinella aurita*		
			Range	Preference	dmax	Range	Preference	dmax
Eggs	Depth [m]	15	42–280	120; 160–220	0.303[NS]	33–418	120–200	0.309[s]
	SST [°C]	10	21.85–26.18	21.5–22; 25–25.5	0.250[NS]	22.70–26.28	24.5–25.5	0.483[ss]
	SSS [PSS]	10	37.17–37.50	37.35–37.45	0.299[NS]	33.12–37.62	37.35–37.45	0.193[NS]
	SWC [kg·m^{-4}]	10	0.019–0.054	0.020–0.030	0.518[ss]	0.013–0.054	0.020–0.030	0.249[NS]
	MLD [m]	16	3–15	8–11	0.359[ss]	3–16	7–9	0.251[NS]
	Chl-a [mg·L^{-1}]	10	0.16–1.57	0.6–0.8	0.277[NS]	0.16–1.57	0.2–0.6	0.316[NS]
	ZDW [g · 10 m^{-2}]	12	1.27–35.69	9–51	0.371[s]	1.27–51.09	9–30	0.361[s]
Larvae	Depth [m]	15	42–418	80–160; 200–240	0.422[ss]	33–280	80–180; 220–260	0.348[s]
	SST [°C]	10	21.85–26.18	24.5–25.5	0.324[NS]	23.26–26.28	24.5–25.5	0.441[s]
	SSS [PSS]	10	37.17–37.54	37.30–37.40	0.273[NS]	37.12–37.62	37.35–37.45	0.180[NS]
	SWC [kg·m^{-4}]	10	0.126–0.054	0.020–0.030	0.216[NS]	0.019–0.062	0.025–0.035	0.198[NS]
	MLD [m]	16	1–15	5–7 ; 9–10	0.303[s]	3–16	5–9 ; 12–14	0.251[NS]
	Chl-a [mg·L^{-1}]	10	0.16–1.57	0.2–0.4	0.380[s]	0.16–1.57	0.2–0.6	0.326[NS]
	ZDW [g · 10 m^{-2}]	12	1.27–51.09	6–7 ; 20–51	0.186[NS]	1.27–51.09	8–20	0.188[NS]

DS = developmental stage; EV = environmental variable; df = degree of freedom; Range = range of occurrence of the fish stage; Preference = range with quotient values >1; dmax = maximum difference between the observed and theoretical cumulative proportions (range: 0–1); SST = sea surface temperature; SSS = sea surface salinity; PSS = practical salinity scale; SWC = stability of water column; MLD = mixed layer depth; Chl-a = chlorophyll-a; ZDW = zooplankton dry weight; [s] = significant $P < 0.05$; [ss] = significant $P < 0.01$; [NS] = Non-significant.

permanent and important river exists in the east coast of Tunisia. Therefore, it could be expected that anchovy spawning in this area should mainly occur in localities where enrichment processes occur, as those associated with the shelf edge (depths around 200 m), as reported in this study and by the findings of Ktari-Chakroun (1979). Other spawning areas for anchovy, such as those localized near Cape Bon and Kelibia could be associated with the intrusion of Atlantic waters, whose interaction with resident waters may produce frontal structures that enhance productivity.

Summing up, both anchovy and sardinella spawned mainly in deeper waters near the limits of the continental shelf. The anchovy spawned mainly in the northern and in the southern parts of in the eastern coast of Tunisia, whereas the round sardinella spawned mainly in the southern part with warmer waters and high values of chlorophyll-a and zooplankton. The spawning areas of both species in the south were adjacent.

It should be emphasized that in this work we presented a snapshot result from a single sampling survey. Therefore, we recommend to carry out a series of surveys in the future in order to analyze intra- and inter annual fluctuations in the spatial location and extension of anchovy and sardinella spawning habitats and in the abundance of eggs and larvae, as well as in deepening the analysis of environmental changes on such variability.

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