Version of Record: https://www.sciencedirect.com/science/article/pii/S0165783618303394
Manuscript_8e3922cdf3f21e6cc7726955a228d7ff

Variation in size at maturity by horse mackerel (*Trachurus trachurus*) within the central Mediterranean Sea: implications for investigating drivers of local productivity and applications for resource assessments

Rosalia Ferreri*[(a)], Richard S. McBride[(b)], Marco Barra[(a)], Antonella Gargano[(a)], Salvatore Mangano[(a)], Maurizio Pulizzi[(a)], Salvatore Aronica[(a)], Angelo Bonanno[(a)], Gualtiero Basilone[(a)].

[(a)] Consiglio Nazionale delle Ricerche – Istituto per lo studio degli impatti Antropici e Sostenibilità in ambiente marino (CNR- IAS), SS di Capo Granitola, Via del Mare, 3 – 91021 Campobello di Mazara (TP, Italy)

[(b)] NOAA Fisheries, Northeast Fisheries Science Center, Woods Hole (MA, USA)

Abstract

Understanding drivers of fish maturity are essential to predict the productivity, stability, and resiliency of exploited populations. In terms of horse mackerel (*Trachurus trachurus*) , in the eastern Atlantic Ocean and the Mediterranean Sea, size at maturity estimates date back to the 1940s and throughout its range. However, many older estimates may not reflect current maturity rates. This is because past methods were imprecise, or environmental or fishing effects may change maturity rates, and some stock areas have been overlooked. To address this data gap for the Central Mediterranean Sea, female horse mackerel size at maturity was estimated for two areas: the Strait of Sicily and the Tyrrhenian Sea. All ovaries were assigned a reproductive phase based on macroscopic methods, and the reliability of this method (94%) was validated with a microscopic method (i.e., gonad histology). Although the collected females exhibited similar condition, in terms of total body weight at a given length, as well as similar gonad-somatic indices, the size at median maturity in the Strait of Sicily was smaller (161 mm total length [TL]) than in the Tyrrhenian Sea (176 mm TL). Future sampling in a series of years with contrasting levels of productivity may help identify whether this is a broad latitudinal trend, or due to specific oceanographic drivers, such as estuarine outflow into the Tyrrhenian Sea or upwelling in the Strait of Sicily. Meanwhile, resource assessments should consider that these two areas represent two phenotypic stocks.

Keywords: Strait of Sicily; Tyrrhenian Sea; length at maturity; fish condition; gonad histology

***corresponding author:** rosalia.ferreri@cnr.it

1. Introduction

Small pelagic fishes comprise about one quarter of the global catch of fishes, and as such are economically and ecologically important at an international scale (Lleonart and Maynou, 2003; Checkley et al., 2009; Vázquez-Rowe et al., 2010). Despite their importance, the life history of most of these species is poorly known, and their highly variable recruitment complicates standard stock assessment models (Abaunza et al., 2003a; Barange et al., 2009). Therefore, new or improved estimates of model parameters should assist with sustainable management of living marine resources in both data-poor and data-rich situations. In the North Sea and north-eastern Atlantic, stocks of horse mackerel (*Trachurus trachurus*) have been defined for management and assessment purposes by the International Council for the Exploration of the Sea (ICES) since late 1970s (e.g., ICES, 1999, 2017). In contrast, this species has received little attention regarding sustainable exploitation and monitoring plans in the Mediterranean Sea, despite their ecological importance (Abaunza et al., 2003b; Rumolo et al., 2017). According to recommendations by the European Union (STECF, 2010) and the General Fisheries Commission for the Mediterranean (FAO-GFCM), management plans should follow an eco-systemic and multispecies approach, i.e. at pelagic ecosystem level (FAO, 2008; Fogarty, 2013). Therefore, many species, usually not includes in the management plans, are receiving renewed interest by the scientific community. Life history parameters, and particularly reproductive traits, are the basis for assessing population productivity and resilience, making them basic inputs into a stock assessment, e.g. maturity data to define the size of the mature biomass in a population.

Horse mackerel (HOM) is a pelagic fish with a broad latitudinal distribution in the northeast Atlantic Ocean (from the West African Cape Verde Islands to Norwegian Sea and North Sea), as well as in the Mediterranean and Black Seas (Abaunza et al., 2003b). It is a zooplanktivorous

(Rumolo et al., 2017), schooling species usually inhabiting the continental shelf (Iglesias et al., 2003; D'Elia et al., 2014). HOM is often caught close to the sea floor using pelagic trawls and purse seines (Abaunza et al., 2003a). Several overviews of HOM biology and ecology and a series of fishery assessments using age-structured models suggest this is a fairly data-rich species (e.g., Abaunza et al., 2003a, b, 2008; Gordo et al., 2008; ICES, 2017).

The spawning period of HOM displays high variability in both range and peak relative to the latitude, but in general, it is protracted (up to 8 months), usually with a peak in spring, in both the Atlantic Ocean and Mediterranean Sea (Abuanza et al., 2003a). Other reproductive traits demonstrate a possible latitudinal cline in the eastern Atlantic (Abaunza et al., 2003b, 2008) including the size at maturity (Abaunza et al., 1995; and reference therein). The estimate and variability of the length at which fishes reach sexual maturity is of particular interest, given its importance in stock assessment. There are over twenty maturity ogive estimates, particularly for females (Abaunza et al., 2003b [and references therein], 2008; Carbonara et al., 2012; see Supplementary Material [Table S3] for more details). However, most of these are quite old (> 20 years), and older parameters may no longer be appropriate given the likelihood that life history traits may be adaptive and change in response to changing environmental conditions (Gordo et al., 2008; McBride et al., 2013; Ganias et al., 2015). Moreover, HOM is characterized by great plasticity in the growth (Abaunza et al., 2003b), and size reached may vary among populations or over time for a given population (Brander, 1995). Variation in maturity-at-size and –at-age has direct implication of productivity fluctuation in many populations, with implications for fisheries management (Morgan, 2018). As a likely income breeder, size-a-maturity of HOM may vary on short time scales, perhaps by year-class (McBride et al., 2015). Older parameters may have also been measured using simpler, and less precise, methods. Many maturity schemes and methods have been used (Abaunza et al., 2003b), and few early efforts validated the maturity evaluations using gonad histology, now accepted as the best practice (Ferreri et al., 2009; Costa, 2009; Khoufi et al., 2014). In multiple-spawning fishes, specifically those with asynchronous ovary development such as the HOM, macroscopic determination of spawning phase is difficult without the support of microscopic examination, because of its subjectivity and variability. Nonetheless, macroscopic evaluation of gonads may still play a key role in the assessment of fishery resources if calibrated to the more precise measures from histological preparations of gonads, and training of sea-going technicians, because of the low cost of macroscopic methods can lead to high sample sizes (Ferreri et al., 2009). Obtaining a full size range of immature and mature sizes or ages can also be an obstacle, particularly for small fishes that may recruit to a fishery at about the same size or larger than maturity, an increasingly common situation as smaller, pre-spawning fishes are protected from harvest. A recent effort to collect synoptic data for HOM at 21 locations across a seascape level led to few immature individuals, particularly females (Abaunza et al., 2008).

Thus, there are several stocks of HOM in the eastern Atlantic, including the Mediterranean Sea, but associating the variability in maturity schedules among these stocks and sub-regional habitat productivity has been obscured by the imprecision of older methods, the difficulty in obtaining adequate samples for maturity classification, and the potential that size-at-maturity may change over time. The present study addresses the first two challenges: the frst with a validation study of maturity assignments data based on naked eye evaluation, referenced to microscopic examination of ovarian histological slides; and the second, by sampling in multiple years and including a stratified-random sampling to target the small, immature fish necessary to fit a model to the data. These data are then used to estimate and evaluate differences in the size-at-first maturity for HOM females between two areas of the central Mediterranean – the Strait of Sicily (SS) and the Tyrrhenian Sea (TY) – that heretofore have not been investigated.

2. Materials and Methods

2.1 Field methods

HOMs were collected during three consecutive years (2012 to 2014) and later in 2016 during May-August in two study areas (Table 1, Fig. 1). The months of sampling overlapped with the latter part of the spawning season in the eastern Mediterranean (Karlou-Riga and Economidis, 1996), which enhanced our ability to distinguish immature from mature but resting fish. The two sampling areas, both in the central Mediterranean Sea, were the Strait of Sicily (SS; Geographical Sub-Areas [GSA] 16 and GSA 15, as defined by General Fisheries Commission for the Mediterranean; GFCM, 2009) and the west Tyrrhenian Sea (TY), starting from the North of Sicily to Ligurian coast (GSA 9 and GSA 10; GFCM, 2009).

Sampling took place within the framework of combined Daily Egg Production Method (Parker, 1980) and echo-acoustic surveys aimed to evaluate abundance and distribution of small pelagic fish species, particularly *Engraulis encrasicolus* and *Sardina pilchardus* (Bonanno et al., 2014a, 2016; Barra et al., 2015; Basilone et al., 2017). Catches were taken from the research vessel “G. Dallaporta” by means of an experimental mid-water pelagic trawl (vertical opening of 8 m, cod-end mesh size of 18 mm), operating at 4.0 knots. The trawl is equipped with a monitoring system (Simrad ITI), for observing net position along the water column, vertical and horizontal mouth opening, and for testing the catch efficiency, during the whole sampling.

HOMs were collected with a simple random approach in the years 2012-2014, but when it was apparent the too few small individuals were being collected, in 2016, they were collected with a stratified-random approach (3 random fish per 1 cm stratified-length bins, for each sampling area). Individuals were measured on board for total length (TL; ± 1 mm), and total (TW) and somatic weight (SW) (± 0.01g), using a stable marine scale (0.01g). Individuals were dissected and the gonads were extracted, sexed and staged according to macroscopic features; the ovaries were preserved in buffered formalin (4%). Collected males were classified maturity by macroscopic characters (ICES, 2008a). However, the data were not validation by gonad histology, nor were gonad weights collected in a manner to corroborate the male maturity data (Supplementary Material). For this reason, all analyses were carried out considering only HOM females. In addition, the maternal condition appears more significant to investigate the reproductive potential (Marshall et al., 2000).

2.2 Reproductive phase evaluation

Macroscopic evaluation of the gonad was carried out at sea on freshly caught individuals. Although several classification schemes have been used in the past (Abaunza et al., 2003b; and references therein), the macroscopic assignment of reproductive phase was carried out directly on board according to the ovary features according to a six-class scale developed for *E. encrasicolus* (Ferreri et al., 2009). This scheme appeared suitable because both species showed gonochorism, asynchronous gonad development and indeterminate fecundity (Ferreri et al., 2016; Ganias et al., 2017). Briefly, the six macroscopic phases have been named: 1) immature; 2) early developing; 3) imminent spawning; 4) spawning; 5) partial post-spawning; 6) spent. Phases 1-2 were considered immature (spawning not imminent this season) and phases 3-6 were considered mature (they have contributed or are contributing to the reproduction this season).

Microscopic evaluation was carried out using gonad histology at the CNR-IAS laboratory. Ovaries were dried of surface moisture and weighed (OW; ± 0.001 g). A small part of ovarian tissue was dehydrated in ethyl alcohol, cleared in xylol, and embedded in paraffin. Sections (4 µm) were stained with Haematoxylin and counter-stained with Eosin (Hunter and Macewicz, 1985). Microscopic examination was used to identify the maturity stage of the most advanced group of oocytes and the presence of postovulatory follicles (POFs; Fig. 2). The earliest oocyte stage recorded was perinucleolar (PE) (Fig. 2a). A subsequent secondary growth oocyte stage was more developed, with cortical alveoli (AC), particularly located along the cellular membrane (Fig. 2b). The presence of cortical alveoli as the most-advanced oocyte stage was not deemed sufficient for imminent spawning in these samples, considered early developing by others (Brown-Peterson et al., 2011), but evaluated here as not likely to spawn in the current year (i.e., hormonally active but functionally immature). The spawning capable phase was characterized by vitellogenic stages, an early stage where the yolk partially filled the cytoplasm (V1) and a later stage where the yolk completely filled the cytoplasm (V2) (Fig. 2c). The final maturity stages were represented by oocytes in nucleus migration (NM) and, subsequently, hydrated (H) (Figs 2d and 2e). Oocyte development stages were used to classify six reproductive phases, according to Ferreri et al. (2009). Briefly, the six microscopic phases are: 1) immature, with only PE oocytes; 2) early developing, with some presence of oocyte with CA; 3) imminent spawning, with mature oocytes (V1 and V2); 4) spawning, with oocytes in NM or hydrated; 5) partial post-spawning, with evidence of occurred spawning (i.e., POFs at any reabsorption stage); 6) spent, with immature oocytes, old POFs and different stage of atresia. Phases defined by microscopic characters were designed to match phases defined by macroscopic characters.

2.3 Data analysis

Gonad development and condition were evaluated to evaluate possible differences among different sampling areas. The gonad-somatic index (GSI) was estimated as:

$$GSI = OW/SW*100,$$

where, OW = ovarian weight, and SW = somatic weight, both in g.

Differences in GSI between adjacent GSAs (i.e., between GSAs 9 and 10, and between GSAs 15 and 16) were assessed using the Mann-Whitney U test and were not statistically significant ($P > 0.1$). Consequently, samples from adjacent GSAs were pooled for the subsequent analyses, to increase the size of the dataset for each area and to compare only two study areas (TY and SS).

The length–weight relationship of HOM was obtained for SS and TY separately, as a measure of condition, using the equation:

$$TW = a^*TL^b,$$

where, TW = total weight (g), TL = total length (mm), a = intercept, and b = slope.

Since the TY region had several very large fish, much larger than SS, the analysis of the length-weight distribution in the two study areas were made on a subset of the data truncated at fish less than 280 mm TL for TY. This ensured that the large HOMs from Tyrrhenian Sea were not on some non-linear trend from the smaller ones, which would bias the comparisons between regions. A general linear model (GLM) was applied to test for differences on fish length, geographic area, and interaction between each, using the log-transformed data (logTW and logTL, respectively). The test was performed by R (vers. 3.3.0 [R Core Team, 2016]).

Validation of reproductive phase, as determined by macroscopic assignment, was evaluated by comparing to paired, independently-determined microscopic assignments for a subset of female HOMs ($n = 201$). These phases were compared directly in a two-way table and the potential bias in disagreements was evaluated by a Bowker's Test of Symmetry based on an α-value of 0.05 (Bowker, 1948; McBride, 2015). Phase-specific GSIs were also plotted to evaluate reproductive performance by sampling date and in each study area.

Size at maturity was predicted using the logistic model:

$$P_i = (1 + e^{-(\alpha + \beta x_i)})^{-1},$$

where, Pi is the probability of being mature at size x_i, and α and β are model parameters representing the intercept and slope of a linear form of this model. In R, this model was fitted using a binary logistic link function of generalized linear models (R, vers. 3.3.0 [R Core Team, 2016]). Macroscopic maturity phases 1-2 were considered immature, and phases 3-6 were considered mature. Area-specific maturity ogives are presented with 95% confidence intervals determined by bootstrapping 1,000 times. An information-theoretic approach was used to select among full (female size, sampling area [factor], and an interaction of both) and reduced models; the second-order Akaike's information criterion (AICc) was used to account for sample sizes in all comparisons.

3. Results

Female HOM collected in the SS were smaller than in the TY in both length (mean TL_{SS} = 190 mm; mean TL_{TY} = 195.32 mm; Table 1 and Supplementary Material [Tables S2]) and weight (mean TW_{SS} = 58.89 g; mean TW_{TY} = 68.84 g; Table 1). The length-weight relationship was plotted for each area separately (Fig. 3); GLM showed the differences were not significant ($p > 0.05$) for neither length, weight, nor weight at a given length between SS and TY.

Reproductive phases agreed well between macro- and microscopic methods. Among immature phases, macroscopic examination tended to be biased towards phase 2, while among mature phases, macroscopic method had difficulty identifying phases 5 and 6 (Table 2). However, the analysis by naked eye correctly recognized females as either immature or mature most of the time (189/201 = 94%; Table 2). Bowker's test of symmetry found no bias in the disagreements between the two methods when assigning ovary to either immature or mature phases ($\chi^2 = 0.33$, df = 1, p = 0.56). Given these results, the remaining analyses were performed using data by macroscopic method, to extend the observations to a wide amount of fishes (294 individuals), for which only macroscopic data were available.

All reproductive phases were commonly observed in both areas, except for the spent phase, which indicated the May-August sampling occurred throughout an active spawning period (Fig. 4). Phase-specific values of GSI were overlapping between areas, indicating similar trends in GSI evolution. As expected, the GSI increased from immature (phases 1 and 2) to mature individuals (from phase 3 onwards), reaching the highest values in hydrated females (phase 4). Similar values were recorded for phases 3 and 5, explained by the prevalence of vitellogenic oocytes (V1 and V2) as the most abundant cellular stage in ovaries of these two phases.

Logistic modelling relating total length and proportion of mature fishes suggested two differences between the two considered areas (Fig. 5). First, the median size at maturity, L_{50}, was smaller for females collected in the SS (161 mm; 95% CI: 153 - 169) than in the TY (176 mm; 95% CI: 171 - 181). Second, the shape of the ogive was more knife edged for the SS. For example, in terms of an odds ratio, SS females were half as likely to be mature at 100 mm TL, but twice as likely by 175 mm TL, and 9 times as likely by 250 mm TL, compared to the TY (Table 3). The full model – accounting for sampling area, fish size, and their interaction – was indistinguishable from the reduced model without the interaction terms (ΔAICc = < 1), but both were best models compared to the fully reduced model with only TL as a predictor (ΔAICc $\approx$ 6). Thus, area was an important proxy, indicating that HOM mature at a smaller median size and a narrower range of lengths in the Strait of Sicily compared to the Tyrrhenian Sea.

4. Discussion

This study reports not only a new maturity schedule for female HOM in the central Mediterranean Sea, where sampling had been lacking, but local variation between two maturity traits estimated from the Strait of Sicily and the Tyrrhenian Sea. This difference in maturity schedules was evident without differences in weight-at-length between regions, or in terms of the evolution of GSIs in relation to macroscopic phase or sampling date. Maturation occurred over a narrower size range and at a smaller L_{50} in the SS, which likely reflects the differences in habitat conditions between the two areas such as in primary production or temperature (Bonanno et al., 2016; Basilone et al., 2017), and represents reproductive (phenotypic) stock structure in this region of the Mediterranean Sea (McBride, 2014).

In both areas, the higher GSI values in mature individuals (phases 3, 4 and 5) confirm the spawning activity indicated by an increasing energy investment in the egg production. Similar conditions of females, highlighted by overlapped GSI values from SS and TY, agree with evidence found for other partial spawner species in Catalan Sea and the northern Tyrrhenian Sea in which, under normal environmental condition, the gonads have a similar development trend, although individuals inhabit regions with different features (i.e. *Merluccius merluccius*, Recasens et al., 2008).

Fish maturity, among other life history traits, is likely a reflection of the environmental productivity in each region (McBride et al., 2015). Future monitoring may uncouple possible hydrodynamic effects from a more general latitude effect, by sampling female HOM maturity in contrasting years of river flows, which would affect TY productivity (Bonanno et al., 2016), versus contrasting years of upwelling, which would affect SS productivity (Bonanno et al., 2014b). Generally, fishes of a particular species in low latitudes grow at a faster rate and mature earlier than those of the same species in high latitudes (Laevastu and Favorite, 1988) and these differences are attributable to the ability to adapt to large-scale patterns in environmental conditions (Beverton, 1992 Winton et al., 2014). However, the effects on reproductive behaviour and traits arising from broad latitude differences versus those due to meso- or micro-habitat variability require more attention than planned into the sampling design here (Recasens et al., 1998; Domínguez-Petit and Saborido-Rey, 2010). For instance, variations in reproductive traits in other small pelagic fishes vary between years by environmental factors, specifically species with indeterminate fecundity, e.g. *Engraulis mordax* (Hunter and Leong, 1981) and *E. encrasicolus* (Somarakis et al., 2004; Basilone et al., 2006).

Although the presence of a possible latitudinal effect in the Mediterranean Sea on reproductive traits seems not to have been investigated until now, differences in productivity were observed between the Eastern and Western parts of the basin (Karlou-Riga and Sinis 1997; Abaunza et al., 2008). Elsewhere (e.g., in the case of Agulhas Bank, South Africa), differences in L_{50} have been attributed to differences in the interpretation of maturity scale or a change in the biological characteristics of the population over time (Hecht, 1990). The lengths at maturity recorded for HOM during the present study (both areas) are lower than the L_{50} estimated in other central Mediterranean areas, Adriatic and Ionian Sea (Carbonara et al., 2012; see Supplementary Material [Table S3] for more details). Otherwise, the HOM in the northeast Atlantic showed increasing L_{50} with latitude, mainly explainable by differences in water temperature (Abaunza et al., 1995). Present L_{50} values are comparable with other estimates for the Mediterranean Sea, using both macro- and microscopic evaluation of gonads (Abaunza et al., 2003b [and reference therein], 2008; Carbonara et al., 2012; see Supplementary Material [Table S3] for more details). The only published reference for the TY reported a higher value, L_{50} = 190 mm, relative to our estimate of 176 mm TL. However, this previous value was estimated from macroscopic analysis without histological validation (Carbonara et al., 2012; see Supplementary Material [Table S3] for more details). No previous estimate had been reported for the SS.

Future monitoring of HOM appears warranted to investigate a working hypothesis about phenotypic stock structure in this part of the Mediterranean Sea, and the results presented here will guide best practices for allocating sampling effort. In particular, total lengths of HOMs sampled in both study areas ranged from 69 to 445 mm, but the functional lower limit was 121 mm when restricted to individuals for which gonad development is already in progress (sex is identifiable). This range is comparable with length range of HOMs caught by other fishing gears and in other coastal Mediterranean waters, such as the Turkish Sea (100 to 189 mm fork length; Erdoğan et al., 2016) and the Adriatic Sea (60 to 390 mm fork length; Jukić and Piccinetti, 1981; Alegría-Hernández , 1984). Future sampling will require extra effort to sample from the lower size range to conform to best practices that the samples used for the ogive evaluation are representative and cover the whole length range of the population (ICES, 2008b). Moreover, the presence of actively spawning females in our samples, including HOM with hydrated ovaries, met the general recommendation to use data collected during the spawning period for estimating the length at maturity (ICES, 2008b).

The macroscopic recognition of reproductive phase by visual examination of gonads is a rapid and inexpensive method for determining the reproductive status in fishes, even though the subjective judgment intrinsic of such method may reduce the data accuracy (Ferreri et al., 2009; Khoufi et al., 2014; Fogg et al., 2017). Nevertheless, a higher level of accuracy may be achieved, if a representative dataset is validated using histological examinations. Generally, macroscopic analysis is not satisfactory for distinguishing between immature, maturing and resting females

(Recasens et al., 2008; Costa, 2009; Ferreri et al., 2009). The literature is replete with examples of the difficultly in distinguishing between immature (phase I) and spent (phase VI, non-active but mature), because in both cases the ovaries are very small, without visible oocytes (Costa, 2009; Ferreri et al., 2009). Such misclassification erroneously affects estimation of the mature proportion of the stock, because resting females macroscopically considered as immature have already contributed to the spawning biomass of that year. During the present study, a good correspondence of immature females between macroscopic and histological reproductive phase identifications was observed. Nevertheless very few HOMs in phase VI have been sampled. Such good agreement appeared in contrast to the results obtained for HOM along the Portuguese coast (Costa, 2009) or other partial spawner fish species, like *Pterois volitans* (Fogg et al., 2017). Otherwise, present observations are in agreement with results obtained for *Merluccius merluccius*, which showed a good correspondence of immature females between macroscopic and histological reproductive phase identification (Khoufi et al., 2014).

Variations in maturation, growth, condition, and spawning stock biomass directly influence population productivity, highlighting the need to consider varying productivity into the stock assessment (Morgan, 2018). As recommended by the European Commission for Fisheries, the biological parameters involved in stock assessment models (i.e., mortality, growth and maturity ogive) should have reference points for each stock in each area monitored or needing to be monitored (STECF, 2010, 2016). The results herein are a substantial step forward, by defining the suitability of the macroscopic maturity evaluation and estimating L_{50} in two subregions of the central Mediterranean Sea. The resulting information improves the accuracy and precision of data available for management of HOM in two areas where until now no reference points were available. Future work should build on these best practices to investigate whether reproductive traits of HOM populations change among adjacent GSAs of central Mediterranean Sea, in response to identifiable environmental signals known to differ between the two subregions.

Acknowledgements

Surveys were co-funded by the Consiglio Nazionale delle Ricerche and the European Union through the Data Collection Framework (DCF – Reg. Ce. No.199/2008, No. 665/2008 and Commission Decision No. 949/2008). The Masters of the R/V ‘‘G. Dallaporta’’ and their crew are thanked for their work. RSM was funded by a National Marine Fisheries Service International Fellowship.

References

Abaunza, P., Fariña, A.C., Carrera, P., 1995. Geographic variations in sexual maturity of the hors mackerel, *Trachurus trachurus*, in the Galician and Cantabrian shelf. Sci. Mar. 59 (3–4), 211–222.

Abaunza, P., Fariña, A.C., Murta, A., 2003a. Applying biomass dynamic models to the Southern horse mackerel stock (Atlantic waters of Iberian Peninsula). A comparison with VPA-based methods. Sci. Mar. 67, 291-300.

Abaunza, P., Gordo, L., Karlou-Riga, C., Murta, A., Eltink, A.T.G.W., García Santamaría, M.T., Zimmermann, C., Hammer, C., Lucio, P., Iversen, S.A., Molloy, J., Gallo, E. 2003b., Growth and reproduction of horse mackerel, *Trachurus trachurus* (Carangidae). Rev. Fish Biol. Fish. 13, 27-61.

Abaunza, P., Gordo, L.S., García Santamaría, M.T., Iversen, S.A., Murta A.G., Gallo E., 2008. Life history parameters as basis for the initial recognition of stock management units in horse mackerel (*Trachurus trachurus*). Fish. Res. 89(2), 167-180.

Alegría-Hernández, V., 1984. Some aspect of horse mackerel (*Trachurus trachurus* L.) biology in the middle Adriatic. FAO Fish Rep. 290, 123-125.

Barange, M., Bernal, M., Cergole, M.C., Cubillos, L.A., Daskalov, G.M., Carryn L. deMoor (formerly Cunningham), C.L., De Oliveira, J. A. A., Dickey-Collas, M., Gaughan, D. J., Hill, K., Jacobson, L.D. Köster, F.W. , Massé, J., Ñiquen, M., Nishida, H., Oozeki, Y., Palomera, I., Saccardo, S.A., Santojanni, A., Serra, R., Somarakis, S., Stratoudakis, Y., Uriarte, A., van der Lingen, C. D., Yatsu, A., 2009. Current trends in the assessment and management of small pelagic fish stocks, pp. 191-255. In: Climate change and small pelagic fish. Ed. By Checkely, D., Alheit, J., Oozeki Y., et al. Cambridge University Press, Cambridge.

Barra, M., Petitgas, P., Bonanno, A., Somarakis, S., Woillez, M., Machias, A., Mazzola, S., Basilone, G., Giannoulaki, M., 2015. Interannual Changes in Biomass Affect the Spatial Aggregations of Anchovy and Sardine as Evidenced by Geostatistical and Spatial Indicators. *PLoS ONE*, 10(8), e0135808.

Basilone, G., Guisande, C., Patti, B., Mazzola, S., Cuttitta, A., Bonanno, A., Vergara, A.R., Maneiro I., 2006. Effect of habitat conditions on reproduction of the European anchovy (*Engraulis encrasicolus*) in the Strait of Sicily. Fish. Oceanogr. 15 (4), 271–280.

Basilone, G., Mangano, S., Pulizzi, M., Fontana, I., Giacalone, G., Ferreri, R., Gargano, A., Aronica, S., Barra, M., Genovese, S., Rumolo, P., Mazzola, S., and Bonanno, A. 2017. European anchovy (*Engraulis encrasicolus*) age structure and growth rate in two contrasted areas of the Mediterranean Sea: the paradox of faster growth in oligotrophic seas. Mediterr. Mar. Sci. 18(3), 504-516.

Bonanno, A., Giannoulaki, M., Barra, M., Basilone, G., Machias, A., Genovese, S., Goncharov, S., Popov, S., Rumolo, P., Di Bitetto, M., Aronica, S., Patti, B., Fontana, I., Giacalone, G., Ferreri, R., Buscaino, G., Somarakis, S., Pirounaki, M., Tsoukali S., Mazzola, S., 2014a. Habitat selection response of small pelagic fish in different environments. Two examples from the oligotrophic Mediterranean Sea. *PLoS ONE*, 9(7): e101498.

Bonanno, A., Placenti, F., Basilone, G., Mifsud, R., Genovese, S., Patti, B., Di Bitetto, M., Aronica, S., Barra, M., Giacalone, G., Ferreri, R., Fontana, I., Buscaino, G., Tranchida, G., Quinci E., Mazzola, S., 2014b. Variability of water mass properties in the Strait of Sicily in summer period of 1998-2013. Oceanogr. Sci. 10, 759-770.

Bonanno, A., Barra, M., Basilone, G., Genovese, S., Rumolo, P., Goncharov, S., Popov, S., Buongiorno Nardelli, B., Iudicone, D., Procaccini, G., Aronica, S., Patti, B., Giacalone, G., Ferreri, R., Fontana, I., Tranchida, G., Mangano, S., Pulizzi, M., Gargano, A., Di Maria A., and Mazzola, S., 2016. Environmental processes driving anchovy and sardine distribution in a highly variable environment: the role of the coastal structure and riverine input. Fish. Oceanogr. 25 (5), 471–490.

Beverton, R.J.H., 1992. Fish resources; threats and protection. Netherlands Journal of Zoology, 42: 139 –175.

Bowker, A.H., 1948. Test for symmetry in contingency tables. Journal of the American Statistical Association, 43, 572-574.

Brander, K.M., 1995. The effect of temperature on growth of Atlantic cod (*Gadus morhua* L.). ICES Journal of Marine Science, 52, 1–10.

Brown-Peterson, N.J., Wyanski, D.M., Saborido-Rey, F., Macewicz, B.J., Lowerre-Barbieri, S.K., 2011. A standardized terminology for describing reproductive development in fishes. Mar. Coast. Fish. 3, 52-70.

Carbonara, P., Casciaro, L., Bitetto, I., Spedicato, M.T., 2012. Reproductive cycle and length at first maturity of *Trachurus trachurus* in the Central-Western Mediterranean Sea. Biol. Mar. Mediterr. 19 (1), 204-205.

Checkley, D.M., Alheit, J., Oozeki, Y., Roy, C., 2009. Climate Change and Small Pelagic Fish. Cambridge University Press.

Costa, A.M., 2009. Macroscopic vs. microscopic identification of the maturity stages of female horse mackerel. ICES J Mar. Sci. 6, 509–516.

D'Elia, M., Patti, B., Bonanno, A., Fontana, I., Giacalone, G., Basilone, G., Fernandes, P.G., 2014. Analysis of backscatter properties and application of classification procedures for the identification of small pelagic fish species in the Central Mediterranean. Fish. Res. 149, 33–42.

Domínguez-Petit, R., Saborido-Rey, F., 2010. New bioenergetic perspective of European hake (*Merluccius merluccius* L.) reproductive ecology. Fish. Res. 104, 83–88.

Erdoğan, Z., Torcu Koç, H., Ulunehir, G., and Joksimović, A. 2016. Some biological properties of different populations of the Atlantic horse mackerel *Trachurus trachurus* (L.) in Turkish Seas. Acta Adriat. 57(1), 51 – 62.

FAO, 2008. Fisheries management. 2 The ecosystem approach to fisheries. 2.1 Best practices in ecosystem modelling for informing and ecosystem approach to fisheries. FAO Technical Guidelines for Responsible Fisheries, 4(2) Add. 1. Rome, 78 pp.

Ferreri, R., Basilone, G., D'Elia, M., Traina, A., Saborido-Rey, F., Mazzola, S., 2009. Validation of macroscopic maturity stages according to microscopic histological examination for European anchovy. Mar. Ecol. 30, 181–187.

Ferreri, R., Ganias, K., Genovese, S., Fontana, I., Giacalone, G., Bonanno, A., Mazzola, S., Aronica, S., Mangano, S., Basilone, G., 2016. Oocyte batch development and enumeration in the European anchovy (*Engraulis encrasicolus*). Mediterr. Mar. Sci. 17/3, 670-677.

Fogarty, M., 2013. The art of ecosystem-based fishery management. Can. J. Fish. Aquat. Sci. 71(3), 479–490.

Fogg, A.Q., Brown-Peterson, N.J., Peterson, M.S., 2017. Reproductive life history characteristics of invasive red lionfish (*Pterois volitans*) in the northern Gulf of Mexico. B. Mar. Sci. 93(3), 791-813.

Ganias, K., Lowerre-Barbieri, S.K., Cooper, W., 2015. Understanding the determinate – indeterminate fecundity dichotomy in fish populations using a temperature dependent oocyte growth model. J. Sea Res. 96, 1–10.

Ganias, K., Mouchlianitis, F.A., Nunes, C., Costa, A.M., Angélico, M.M., 2017. A reassessment of the fecundity type of Atlantic horse mackerel (*Trachurus trachurus*) in Atlantic Iberian waters (ICES division IXa) shows that indeterminate spawners can cease recruiting oocytes during their spawning season. ICES J. Mar. Sci. 74(1), 31–40.

GFCM, 2009. Establishment of Geographical Sub-Areas in the GFCM area amending the resolution GFCM/31/2007/2, RES-GFCM/33/2009/2.

Gordo, L.S., Costa, A., Abaunza, P., Lucio, P., Eltink, A.T.G.W., Figueiredo, I., 2008. Determinate versus indeterminate fecundity in horse mackerel. Fish. Res. 89, 181–185.

Hecht, T., 1990. On the life history of Cape horse mackerel *Trachurus trachurus capensis* off the south-east coast of South Africa. S. Afr. J. Marine Sci. 9, 317-326

Hunter, J.R., Leong, R., 1981. The spawning energetics of female northern anchovy, *Engraulis mordax*. Fish. B-NOAA 79, 215–230.

Hunter, J.R., Macewicz, B.J., 1985. Measurement of spawning frequency in multiple spawning fishes. pp. 79-94. In: An egg production method for estimating spawning biomass of pelagic fish: Application to the northern anchovy, *Engraulis mordax*. Ed. By R. Lasker, NOAA Tech. Rep. NMFS 36.

ICES, 1999. Report of the Working Group on Mackerel and Horse Mackerel Egg Surveys. ICES CM 1999/G:5, Ref.: D.

ICES, 2008a. Report of the Workshop on Small Pelagics (*Sardina pilchardus*, *Engraulis encrasicolus*) maturity stages (WKSPMAT). ICES CM 2008/ACOM:40. 82 pp.

ICES, 2008b. Report of the Workshop on Maturity Ogive Estimation for Stock Assessment (WKMOG), 3-6 June 2008, Lisbon, Portugal. ICES CM2008/ACOM: 33. 72 pp.

ICES, 2017. Final Report of the Working Group on Mackerel and Horse Mackerel Egg Surveys. WGMEGS Report 2017, 24-28 April 2017. Vigo, Spain. ICES CM 2017/SSGIEOM: 18. 134 pp.

Iglesias, M., Carrera P., Muiño, R., 2003. Spatio-temporal patterns and morphological characterization of multispecies pelagic fish schools in the North-Western Mediterranean Sea. Aquat. Living Resour. 16, 541–548.

Jukić, S., Piccinetti, C., 1981. Quantitative and qualitative characteristics of demersal resources, in the Adriatic Sea, with some population Dynamics estimates. FAO Fish. Rep. 253, 73-91.

Karlou-Riga, C., Economidis, P.S., 1996. Ovarian atretic rates and sexual maturity of horse mackerel, *Trachurus trachurus* (L.) in the Saronikos Gulf (Greece). Fish. Bull. U.S., 94 (I): 66-76.

Karlou-Riga, C., Sinis, A., 1997. Age and growth of horse mackerel, *Trachurus trachurus* (L.), in the Gulf of Saronikos (Greece). Fish. Res. 32, 157–171.

Khoufi, W., Ferreri, R., Jaziri, H., El Fehri, S., Gargano, A., Mangano, S., Ben Meriem, S., Romdhane, M.S., Bonanno, A., Aronica, S., Genovese, S., Mazzola, S., Basilone, G., 2014. Reproductive traits and seasonal variability of *Merluccius merluccius* from the Tunisian coast. J. Mar. Biol. Ass. UK. 94, 1545–1556.

Laevastu, T., Favorite, F., 1988. Fishing and stock fluctuations. Fishing News Books Ltd. Farnham, Surrey, England 239 pp.

Lleonart, J., Maynou, F., 2003. Fish stock assessments in the Mediterranean: state of the art. Sci. Mar. 67, 37–49.

Marshall, C.T., Yaragina, N.A., Adlandsvik, B., Dolgov, A.V., 2000. Reconstructing the stock-recruit relationship for Northeast Arctic cod using a bioenergetic index of reproductive potential. Can. J. Fish. Aquat. Sci. 57, 2433–2442.

McBride, R.S., Wuenschel, M.J., Nitschke, P., Thornton, G.,King, J.R., 2013. Latitudinal and stock-specific variation in size- and age-at-maturity of female winter flounder, *Pseudopleuronectes americanus*, as determined with gonad histology. J. Sea Res. 75, 41–51.

McBride, R.S., 2014. The continuing role of life history parameters to identify stock structure. pp. 77-107. In Stock Identification Methods (Second Edition). Ed. By Cadrin, S. X., Kerr, L. A., and Mariani, S. Academic Press, San Diego.

McBride, R.S., 2015. Diagnosis of paired age agreement: a simulation of accuracy and precision effects. ICES J. Mar. Sci. 72 (7), 2149-2167.

McBride, R.S., Somarakis, S., Fitzhugh, G.R., Albert, A., Yaragina, N.A., Wuenschel, M.J., Alonso-Fernández, A., Basilone, G., 2015. Energy acquisition and allocation to egg production in relation to fish reproductive strategies. Fish Fish. 16, 23-57.

Morgan, M.J., 2018. Understanding biology to improve advice for fisheries management. ICES J. Mar. Sci. 75(3), 923–931.

Parker, K., 1980. A direct method for estimating northern anchovy, *Engraulis mordax*, spawning biomass. Fish. Bull. U.S. 78, 541-544.

R Core Team, 2016. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

Recasens, L., Lombarte, A., Morales-Nin, B., Torres, G.J., 1998. Spatiotemporal variation in the population structure of the European hake in the NW Mediterranean. J. Fish Biol. 53, 387–401.

Recasens, L., Chiericoni, V., Belcari, P., 2008. Spawning pattern and batch fecundity of the European hake (*Merluccius merluccius* (Linnaeus, 1758)) in the western Mediterranean. Sci. Mar. 72 (4), 721-732.

Rumolo, P., Basilone, G., Fanelli, E., Barra, M., Calabrò, M., Genovese, S., Gherardi, S., Ferreri, R., Mazzola, S., Bonanno, A., 2017. Linking spatial distribution and feeding behavior of Atlantic horse mackerel (*Trachurus trachurus*) in the Strait of Sicily (Central Mediterranean Sea). J. Sea Res. 121, 47–58.

Somarakis, S., Palomera, I., Garcia, A., Quintanilla, L., Koutsikopoulos, C., Uriarte, A. Motos, L., 2004. Daily egg production of anchovy in European waters. ICES Journal of Marine Science, 61: 944-958.

STECF, 2010. Scientific, Technical and Economic Committee for Fisheries (STECF) - Report of the SGMOS-10-03 Working Group Development of the Ecosystem Approach to Fisheries Management (EAFM) in European seas. JRC61951. 146 pp.

STECF, 2016. Reports of the Scientific, Technical and Economic Committee for Fisheries (STECF) - Methodology for the stock assessments in the Mediterranean Sea (STECF-16-14). JCR102680. 168 pp.

Vázquez-Rowe, I., Iribarren, D., Moreira, M.T., Feijoo, G., 2010. Combined application of life cycle assessment and data envelopment analysis as a methodological approach for the assessment of fisheries. Int. J. Life Cycle Ass. 15, 272-283.

Winton, M.V., Wuenschel, M. J., McBride, R.S., 2014. Investigating spatial variation and temperature effects on maturity of female winter flounder (*Pseudopleuronectes americanus*) using generalized additive models. Can. J. Fish. Aquat. Sci. 71, 1279-1290.

Table

Table 1: Year and sampling month; Number of HOM (No. of samples) and number of females (No. of females) collected per each study year; minimum and maximum total length (TL); minimum and maximum total weight (TW); minimum and maximum of gonad weight (GW). All the data are presented for the two study areas, the Strait of Sicily (SS) and the Tyrrhenian Sea (TY).

	SS						*TY*					
Year	*Sampling month*	*No. of samples*	*No. of females*	*TL (mm)*	*TW (g)*	*GW (g)*	*Sampling month*	*No. of samples*	*No. of females*	*TL (mm)*	*TW (g)*	*GW (g)*
2012	June-July	112	34	69-262	2.92 – 131.75	0.19 – 9.44	July	105	42	92 – 435	5.72 – 562	0.10-5.21
2013	June	158	30	124 – 250	15.85 – 138.45	0.63 – 4.62	May – June	229	79	122-346	16.27-333	0.02-23.28
2014	July	36	11	142 – 287	20.36 - 177	1.09 – 14.19	June	182	49	117-394	12.71-500	0.05-8.66
2016	July	42	14	104 – 268	9.62 – 126.9	0.02 – 4.68	July-August	150	61	144-445	24.17-573	0.05-14.95

Table 2. Comparison of reproductive phase recognition by two methods, microscopic (Micro; by histology) versus macroscopic (Macro), for 201 female horse mackerel. Data from both study areas, Strait of Sicily and Tyrrhenian Sea, were combined. Reproductive phases 1-2 are immature, 3-6 are mature; agreements for immature or mature classifications for individual fish are in bold.

		Micro					
		1	*2*	*3*	*4*	*5*	*6*
Macro	*1*	**19**	**2**	2	-	-	-
	2	**47**	**3**	2	-	-	3
	3	3	2	**74**	**3**	**10**	**4**
	4	-	-	**6**	**11**	**4**	-
	5	-	-	**2**	**3**	**1**	-
	6	-	-	-	-	-	-

Table 3. Tabulation of proportion mature (P_{mat}), odds of being mature ($P_{mat}/P_{immature}$) at a given fish size for each sampling area, and the odds ratio of being mature in the Strait of Sicily (SS) versus the Tyrrhenian Sea (TY). Estimated parameters used here to calculate P_{mat} were α = -8.08 $\pm$ 2.80 (estimate $\pm$ standard error) and β = 0.0503 $\pm$ 0.0156 for the SS, and α = - 5.33 $\pm$ 0.982 and β = 0.0303 $\pm$ 0.00533 for the TY (see methods for full logistic equation).

	SS		*TY*		*SS/TY*
TL	P_{mat}	Odd_{mat}	P_{mat}	Odd_{mat}	*Odds Ratio*
100	0.045	0.047	0.091	0.100	0.472
125	0.143	0.167	0.176	0.214	0.779
150	0.369	0.586	0.313	0.456	1.284
175	0.673	2.060	0.493	0.973	2.117
200	0.879	7.243	0.675	2.075	3.490
225	0.962	25.470	0.816	4.426	5.755
250	0.989	89.568	0.904	9.440	9.488

Figures captions

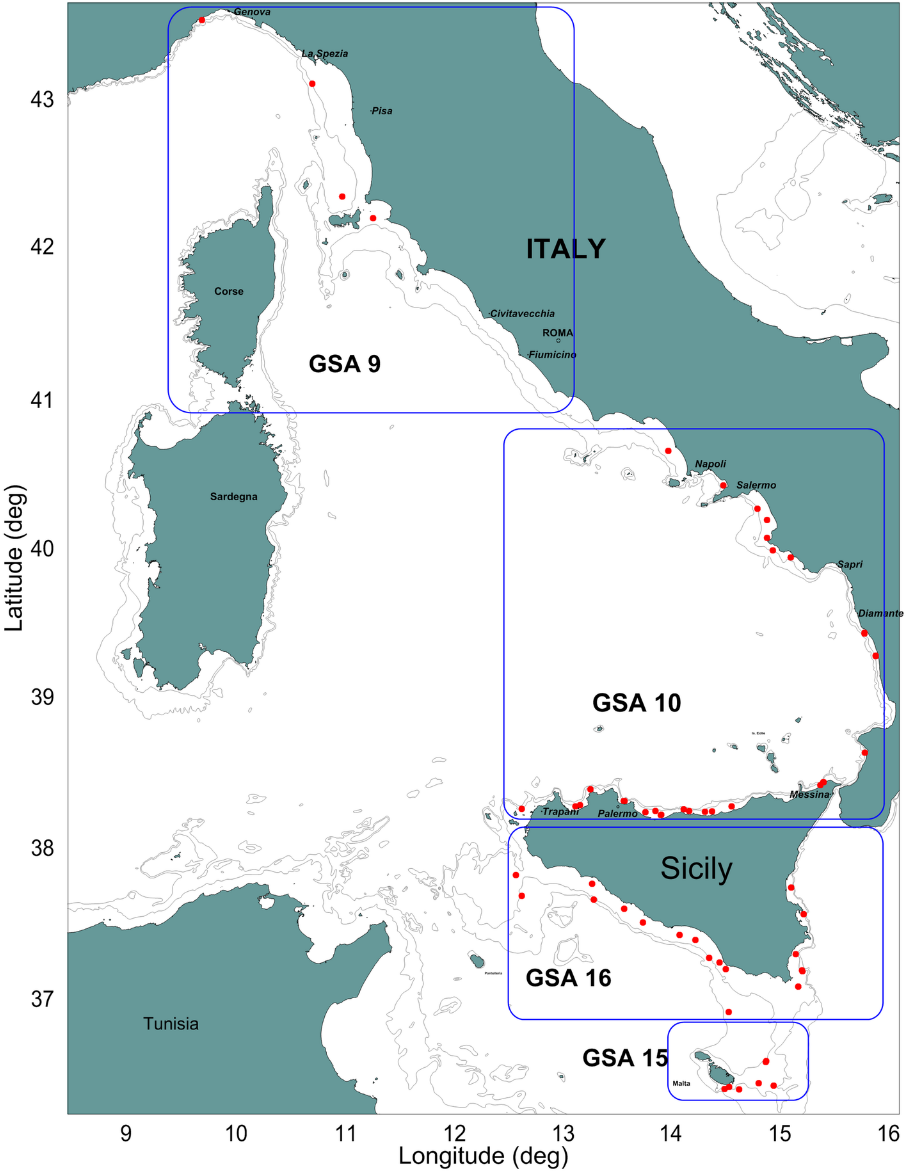
Fig. 1 Locations of fish trawls collecting female horse mackerels in the two study areas, Strait of Sicily (triangles) and Tyrrhenian Sea (circles) during four summer surveys (2012 to 2014 and 2016).

Fig. 2 Histological sections of oocytes stages in horse mackerel females: a) PE = perinucleolar oocyte stage; b) CA = cortical alveoli; c) V1 = partially vitellogenic oocytes and V2 = fully vitellogenic oocytes; d) NM = nucleus migration; e) H = hydrated oocytes; f) POF = post-ovulatory follicles.

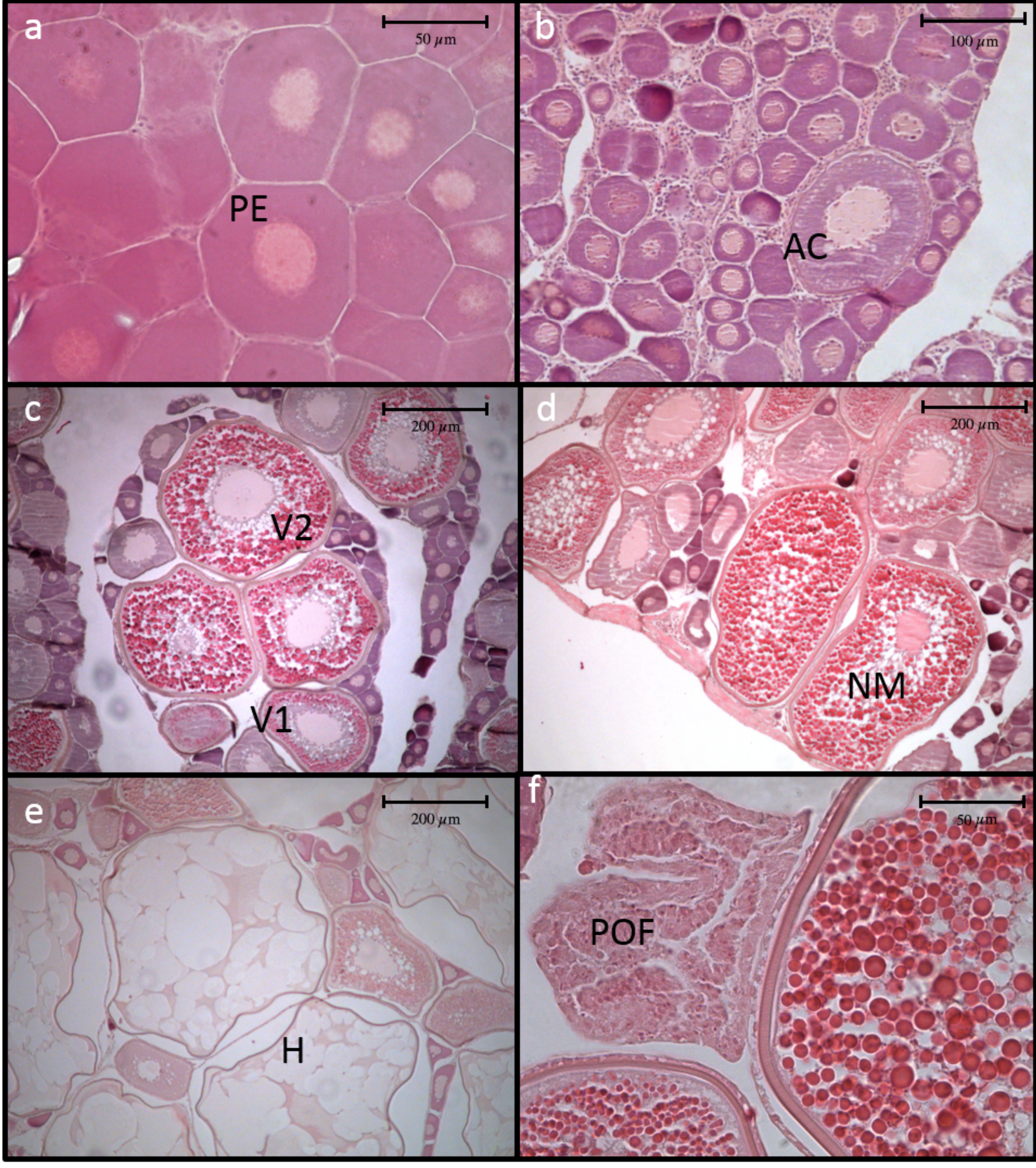
Fig. 3 Length (log Total Length) – weight (log [Total] Weight) relationship for horse mackerel females (< 280 mm TL) for two study areas, Strait of Sicily (SS) and Tyrrhenian Sea (TY).

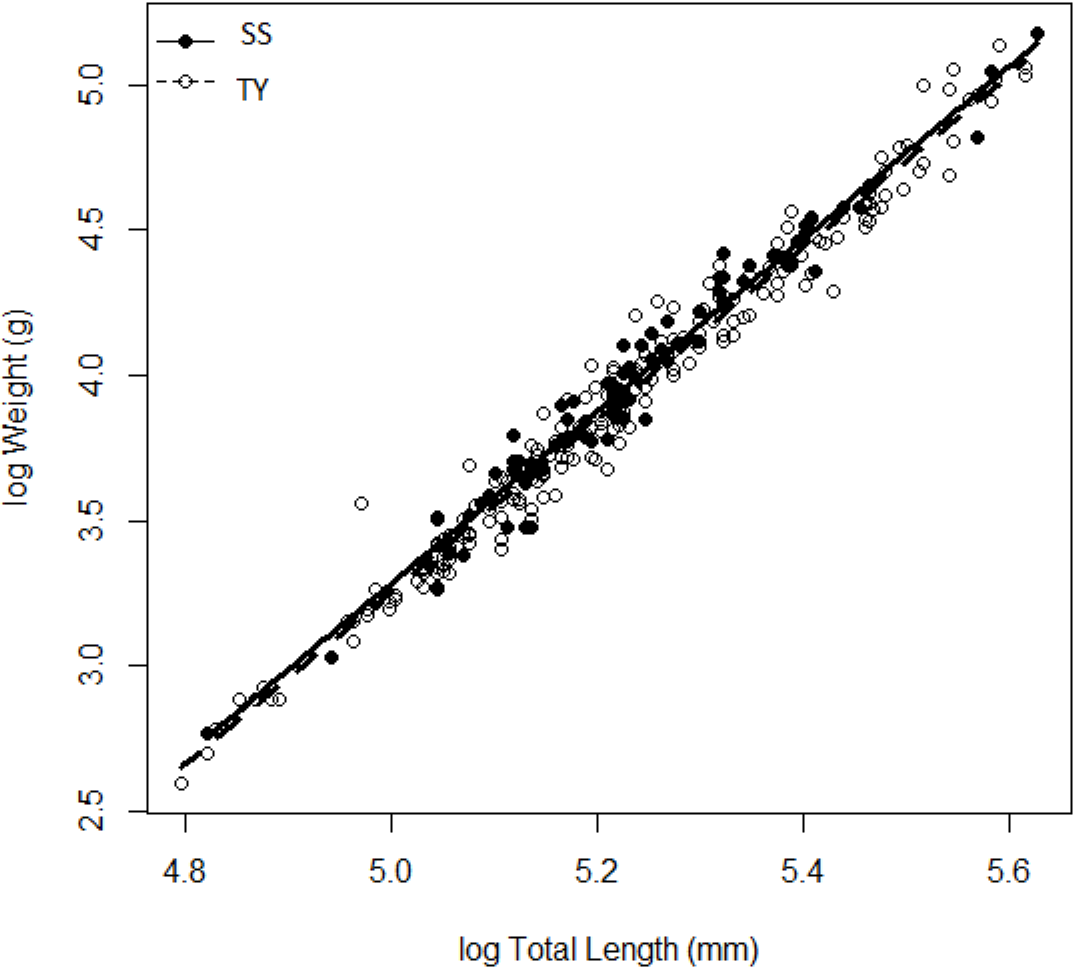
Fig. 4 Gonad-somatic index (GSI) of female horse mackerel by reproductive phase (1-6; macroscopic evaluation; top panel) and by sampling date (day of year; i.e., 152 = June 1; bottom panel). Data for individual fish are presented separately for two study areas, Strait of Sicily (SS) and Tyrrhenian Sea (TY; symbols by regions are staggered to be non-overlapping).

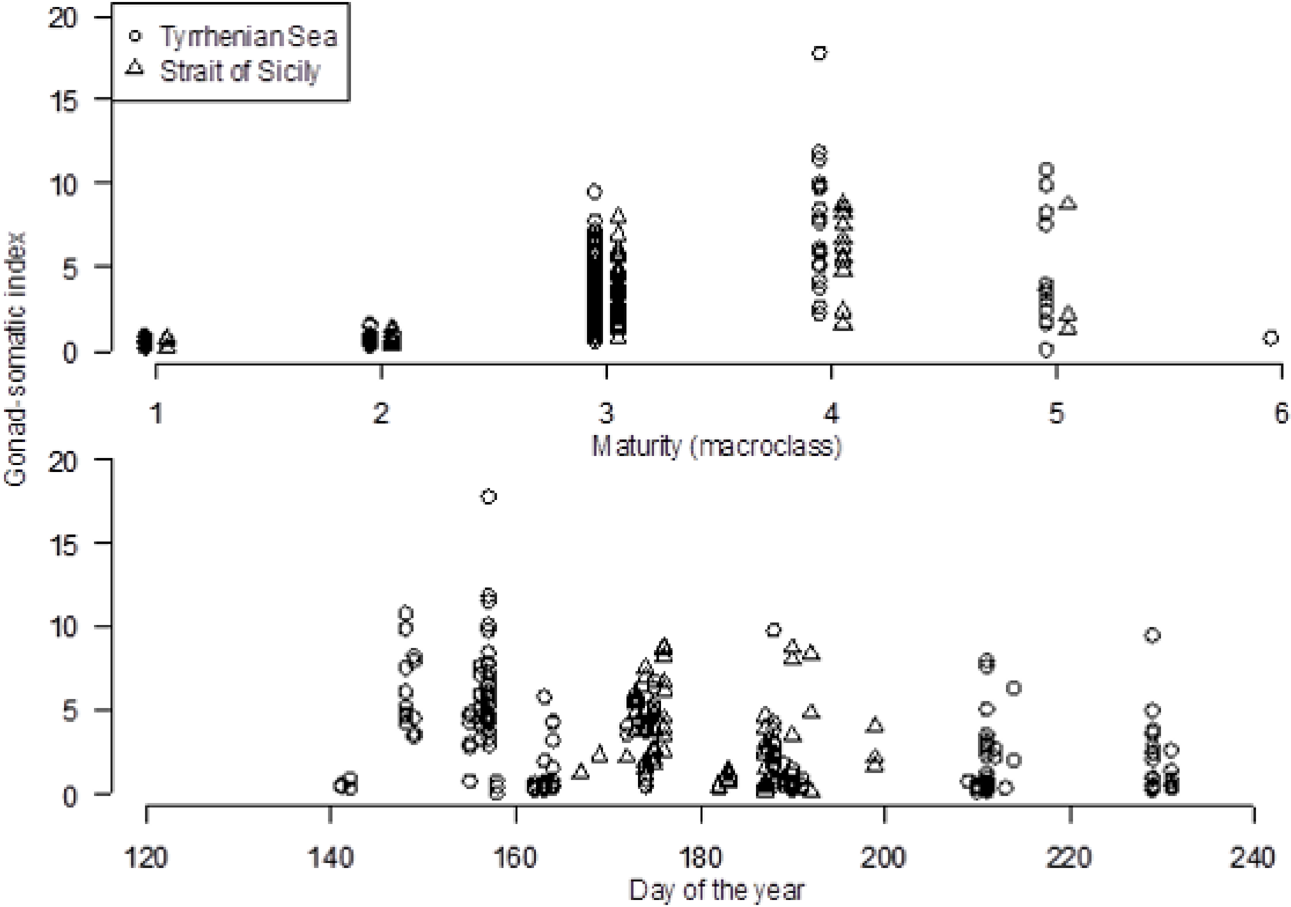
Fig. 5 Maturity ogives for female horse mackerel in two study areas, Strait of Sicily and Tyrrhenian Sea. The solid black line is the predicted proportions of mature females at length, the dashed blue lines are the 95% confidence limits, and the tick marks on the bottom and top axes indicate fish size data for immature and mature fish, respectively. Point estimates for mean size (± 95% c.l.) are estimated for 5% (L_5), 50% (L_{50}), and 95% (L_{95}) mature females. See Table 3 for more details about the logistic equation parameters for each area and odds ratios of being mature at a given size.

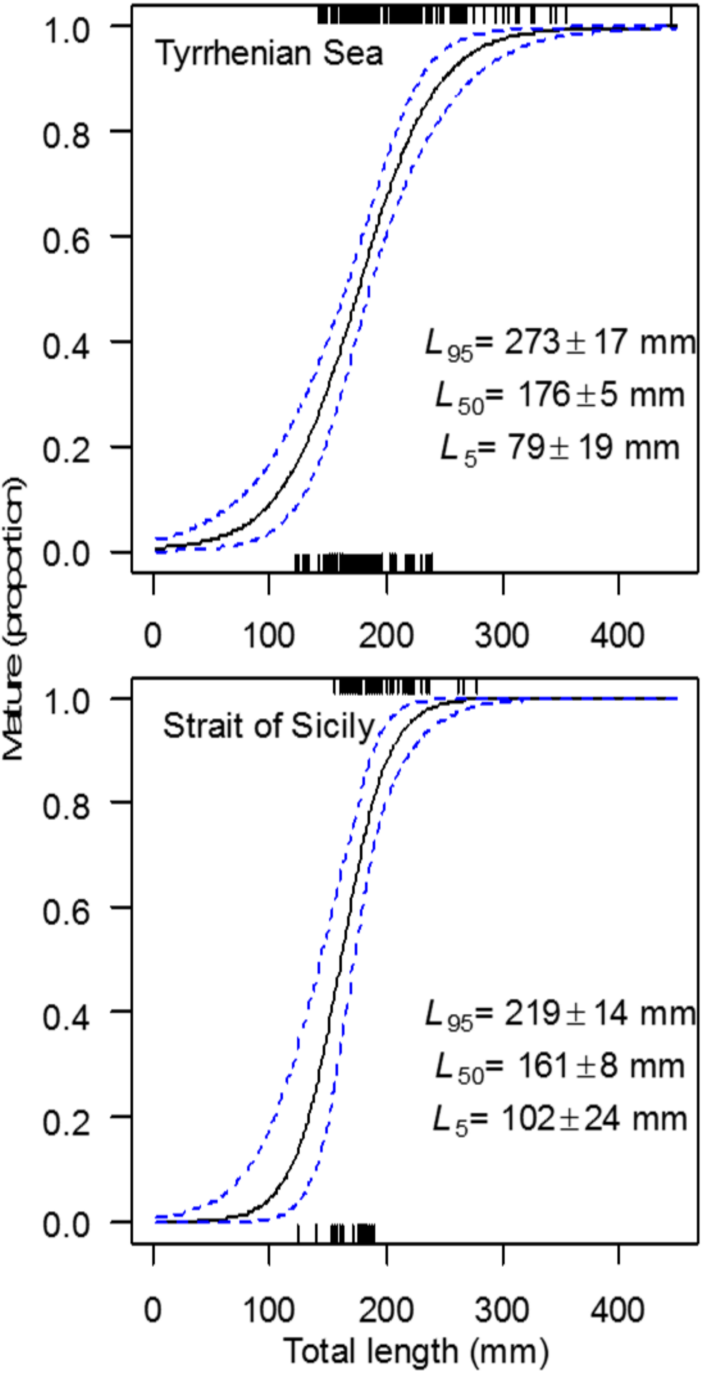


Genova
La Spezia
Pisa
ITALY
Corse
Civitavecchia
ROMA
Fiumicino
GSA 9
Sardegna
Napoli
Salermo
Sapri
Diamante
GSA 10
Is. Eolie
Messina
Trapani
Palermo
Sicily
Pantelleria
GSA 16
Tunisia
GSA 15
Malta
43
42
41
40
39
38
37
Latitude (deg)
9
10
11
12
13
14
15
16
Longitude (deg)


a
50 μm
PE
b
100 μm
AC
c
200 μm
V2
V1
d
200 μm
NM
e
200 μm
H
f
50 μm
POF


SS
TY
log Weight (g)
2.5
3.0
3.5
4.0
4.5
5.0
4.8
5.0
5.2
5.4
5.6
log Total Length (mm)


Tyrrhenian Sea
Strait of Sicily
Gonad-somatic index
20
15
10
5
0
1
2
3
4
5
6
Maturity (macroclass)
20
15
10
5
0
120
140
160
180
200
220
240
Day of the year


1.0
0.8
0.6
0.4
0.2
0.0
Tyrrhenian Sea
L_{95}= 273 ± 17 mm
L_{50}= 176 ± 5 mm
L_{5}= 79 ± 19 mm
0
100
200
300
400
Mature (proportion)
1.0
0.8
0.6
0.4
0.2
0.0
Strait of Sicily
L_{95}= 219 ± 14 mm
L_{50}= 161 ± 8 mm
L_{5}= 102 ± 24 mm
0
100
200
300
400
Total length (mm)