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Research article

First maturity size of the Black Sea anchovy and its implications on the age-based stock assessment models

Gizem AKKUŞ*, , Ali Cemal GÜCÜ

Middle East Technical University, Institute of Marine Sciences, P.O.Box 28, 33731, Erdemli-Mersin, Türkiye.

* Corresponding Author: e-mail: gizemakkuss@gmail.com

Abstract: This study aims to estimate the Black Sea anchovy's first maturity length and its relationship with the ambient temperature to investigate possible consequences of global warming on the stock. The study also evaluated how changes in maturity size/age affect the stock assessment model and scientific advice given by the Scientific, Technical, and Economic Committee for Fisheries (STECF), an advisory board of the European Commission, for the sustainable exploitation of the stock. A total of 7218 anchovies sampled during the scientific surveys conducted in 2013, 2014, 2015, 2018, and 2020 covering the southern Black Sea suggested that the L_{50} of the species was 7.6 ±0.07 cm in TL. On average, 30% of the anchovy were found to be matured at age zero, representing 8% of the population. It was also noted that the zero-age-maturity rate was the highest during the warmest year. When the XSA model, which was chosen by the STECF in 2017 to assess the status of the stock was re-run with the newly estimated parameters, the Spawning Stock Biomass was estimated to be 1.5 times higher than the value used to produce scientific management advice. While maturity information significantly affects the stock representation of the model in the short run; in the long run, the management policies produced with these results may cause the stock to be mismanaged. Thus, the results must be considered to enhance the quality of the scientific advice given to manage the Black Sea anchovy stock.

Keywords: L50, XSA, fisheries management, spawning stock biomass, global warming.

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Introduction

Black Sea anchovy (*Engraulis encrasicolus*, Linnaeus, 1758) is a fast-growing, short-lived, and batch-spawner fish. The majority of the population reaches maturity at age one and its spawning period lasts from the middle of May to the second half of August which peaks from the middle of June to the end of July (Chashchin et al., 2015; Galatchi et al., 2015; Ünsal, N., 1989; Lisovenko and Andrianov, 1996). Nevertheless, it is well known that the Black Sea anchovy has enormous reproduction potential. Depending on water temperature, food availability, and body size, each female can spawn more than 50 times a year with an average egg number of 200,000 per spawning season (Lisovenko and Andrianov, 1996). Seawater temperature, on the other hand, is an important environmental factor that causes fish to become sexually mature at a younger age (Lowerre-Barbieri et al.,2011). Moreover, according to Lisovenko and Andrianov (1996) and Chashchin et al. (2015), approximately 3% of the anchovy population in the Black Sea, attains sexual maturity and starts spawning after two or three months from the completion of their larval stage.

This high reproductive potential stock is one of the most abundant and priority-managed fish species in the Black Sea region, with its unique life history characteristics and high commercial value in the basin

(FAO, 2022). Indeed, for more than a decade, the Scientific, Technical, and Economic Committee for Fisheries (STECF, 2011:2017) and the General Fisheries Commission for the Mediterranean (GFCM; since 2014) have been catalyzing the comprehensive scientific stock assessment studies across the Black Sea. Lately, age-structured statistical stock evaluation models have been the primary tools used to anticipate anchovy stock conditions. The eXtended Survival Analysis (XSA) model (Shepherd, 1999) is one of these models derived from Virtual Population Analysis (VPA). It differs from VPA in its ability to use the complete information year-class strength from the catch-at-age data, its sensitivity to observation errors in the final year, its tuning procedure that approximates the value of the observed index data and calculated model results, and its estimate of the fishing mortality rate (F) from recent years (Darby and Flatman 1994; Shepherd 1999). The last accepted assessment of the Black Sea anchovy was performed using XSA (GFCM, 2018).

Assessment models provide important biological reference points that help assess the status of fish stocks and guide managers toward sustainable management practices. One of the crucial indicators for determining the state of a stock is the Spawning Stock Biomass (SSB) which is the total weight of all sexually mature fish in the stock (FAO, 1996; Heino et al., 2013). SSB is estimated within the assessment model based on the final stock in the matrix of the number, and the inputs of mean weights per individual by age and maturity (Lassen and Medley, 2001; Osio et al., 2018). Among them, the maturity of the Black Sea anchovy will be the main subject of this study.

On the other hand, L_{50}, which means 50% of the individuals within a given fish population attain sexual maturity, is a length-based life-history characteristic used as a proxy to evaluate the targeted population's reproductive status and reproductive potential. From a fisheries perspective, the average length of fish in a population should be over L_{50} to ensure self-sufficient productivity and sustainability (Morgan, 2018). It is the indicator used in the management measures such as minimum landing size to avoid over-exploitation of younger individuals.

Maturity information is one of the life history parameters of fish populations estimated externally and used as input within the XSA model. In the current stock assessment models applied to the Black Sea anchovy, the fixed maturity percentage over the age classes for each year is used (STECF, 2017; GFCM, 2018). The use of fixed maturity may be a precautionary measure to avoid extra bias, given that the determination of maturity stage requires expertise and no standard macroscopic and histological staging protocol for anchovy is accepted and agreed upon by all Black Sea countries. However, to ensure an accurate estimation of the SSB and effective management of the overall stock, it is crucial also to prioritize the consideration of varying proportions of early-maturing individuals over time.

This study aimed to draw attention to the importance of first maturity information that has been ignored so far as one of the basics of fish stock assessment and management. For this, L_{50} for the Black Sea anchovy was estimated. The annual maturity schedule for the sampling years was calculated and its relation to summer seawater temperature was tested. In addition, a sensitivity test was conducted to analyze how the estimated annual maturity affects the predicted outcome of SSB predicted by XSA. The test compared maturity schedules from the current study with the one used in the last accepted STECF assessments. Given the proportion of mature zero-year-old anchovies in the population, the results of this study could help enhance the quality of the Black Sea anchovy stock assessment.

Materials and Methods

During the fishery-independent scientific surveys held in 2013, 2014, 2015, 2018, and 2020, a total of 7218 anchovy samples were collected by using a mid-water trawl on board RV Bilim-2 belonging to the Middle East Technical University (METU). Sampling stations represent coastal and offshore regions of the Southern Black Sea (40.9º – 43.4º N and 28.0º - 41.4º E) covering the Turkish Exclusive Economic Zone (EEZ). A wide temporal and spatial sampling area helped to avoid possible variability due to the differential distribution of mature and immature fish. The summer cruises were performed considering the peak of spawning of the Black Sea anchovy, which is in July (Lisovenko and Andrianov, 1996). Correspondingly, while estimating the maturity proportion over the ages, it was assumed that the peak of anchovy spawning occurs at precisely the same period each year.

All collected samples were analyzed at the METU, Institute of Marine Sciences, Marine Biology and Fisheries Laboratory, Turkey. The total length (±0.1 cm) was measured. Sex and maturity stages were determined macroscopically for the stock assessment purpose (Ferreri et al., 2009; Min et al., 2022). For staging, the ICES WKSPMAT REPORT (2008) protocol was used as a guide with its six-stage of maturity: stage I (immature or rest), stage II (developing), stage III (imminent spawning), stage IV (spawning), stage V (partial post-spawning), and stage VI (spent).

Size at first maturity (L_{50}) Estimation

The maturity stages were converted to binary maturity categories. The fish were considered mature if they were in maturity stages 2 to 6 and immature if they were in maturity stage 1. For maturity ogive estimation, the logistic regression was fitted to the individual total fish length (cm) and maturity stage data using a Generalized Linear Model (GLM) with a binomial error structure. The logit was employed as the link function (ICES, 2019; Min et al., 2022).

The proportion of mature individuals-at-length was fitted by

$$P = \frac{1}{1 + e^{-(a+b*TL)}}$$

where **P** is the proportion of the mature individuals for each size class; **a** and **b** are constant defining the shape and location of the fitted curve; **TL** is the total length in cm. The infection point (L_{50}) in the logistic curve was estimated by (Tiraşin et al., 2020):

$$L50 = \frac{-a}{b}$$

For maturity size estimation and its confidence limits, non-parametric bootstrapping was performed (3000 resamplings) using the logit transformations by logistic regression coefficients.

Maturity proportion-at-age estimation

The ages of 2468 sampled fish were determined. Age estimations were conducted using the sagittal otoliths immersed in 70% ethyl alcohol. They were analyzed under the reflected light against a black background using a binocular microscope with a magnification of 25X by following the Common Age Reading Protocol for the Black Sea anchovy prepared by Black Sea experts (GFCM, 2019). The maturity schedule of individuals belonging to each age group in the population was calculated. This maturity-at-age information was used as the input for the stock assessment models.

Annual average seawater temperature

In-situ temperature measurements (with an accuracy of 0.002°C) were obtained during the summer cruises in 2013, 2015, and 2018 using a CTD water column (Seabird SBE 911 Scientific Inc.). Since the anchovy in the Black Sea is a thermophilic species and stays over the thermocline, the average temperature at a depth of 4-9 m, which represents the mixed layer over the thermocline, was used in the analysis.

SSB Estimation

In the last STECF (2017) assessment meeting, management advice given by the expert group is based on the results of the XSA model. For running the model and estimating the SSB, the required input data: landings, catch-at-ages, weight-at-ages, natural mortality (M), tunings, and the maturity schedule were taken from the last accepted assessment attained by STECF (2017). In another scenario with different maturity schedules, data was set up in this study by averaging the maturity percentage over the ages. The model was run separately with sensitivity to the maturity schedule.

The required input data to run the XSA were prepared in Lowestoft format. XSA was run under R version 3.6.2 (R Core Team, 2019) with the *FLXSA*, *FLCore*, *FLEDA*, *FLAssess*, *FLash*, and *FLBRP* packages from FLR Project (https://flr-project.org/#packages) and *lattice*, *iterators*, and *ggplot2* packages from CRAN Repository (https://CRAN.R-project.org/package). All statistical analyses were done under the R version 4.0.5 (R Core Team, 2021), using the packages of FSA, *magrittr*, *dplyr*, *lubridate*, *car* and *ggplot2*.

Results

L_{50} and Maturity proportion-at-age-zero

The estimated L_{50} with 95% CI for the Black Sea anchovy is 7.6 ±0.07 cm in TL and L_{95} is 9.1 ± 0.12 cm in TL (df=7216, p<0.001; Table1, Figure 1). Sexual differences in estimated L_{50} were observed. While the L_{50Male} is 7.52 ±0.09 cm in TL (df= 3307, p<0.001), $L_{50Female}$ is 7.66 ±0.05 cm in TL (df= 3907, p<0.001). Yet, the smallest observed mature females and males were both at 6.0 cm in total length.

The maturity schedule over the ages was calculated for all sampling years (Table 2). The results indicate that the Black Sea anchovy is sexually fully-mature at age 1 (97-100%). However, the proportion of mature individuals at age 0 is highly variable, and its rate changes annually. Considering all years, on average, 30% of the zero-age-old anchovy start breeding in the year they were born. This rate is about 8% of the entire population.

Table1. Summary table for the results of L50 estimation for the years 2013: 2015, 2018, and 2020

	Estimated L_{50}	L_{50} Lower Limit of CI	L_{50} Upper Limit of CI	Estimated L_{05}	L_{05} Lower Limit of CI	L_{05} Upper Limit of CI	Estimated L_{95}	L_{95} Lower Limit of CI	L_{95} Upper Limit of CI
Overall	7.60	7.53	7.67	6.09	5.95	6.23	9.10	8.98	9.21
Female	7.66	7.55	7.76	6.18	6.08	6.29	9.15	8.99	9.27
Male	7.52	7.43	7.63	5.99	5.87	6.1	9.06	8.9	9.2

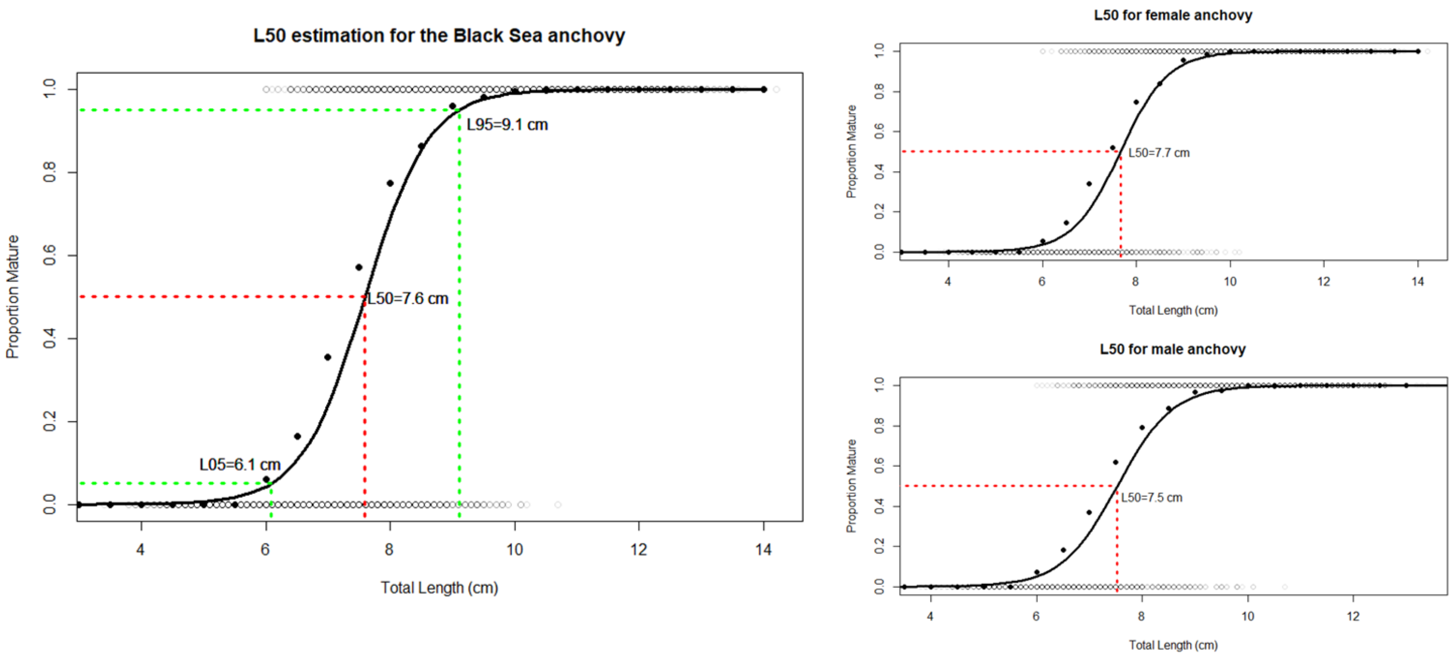

Figure 1. The length estimation at which 50% of Black Sea anchovy individuals are sexually mature (female: right-upper panel and male: right-lower panel).

Table 2. Percentage of the mature individuals over age classes

Years	% Of Mature at 0-Age-Classes	% Of Mature at 1-Age-Classes	% Of Mature at ≥2-Age- Classes
2013	38	97	100
2014	13	88	100
2015	38	98	100
2018	65	100	100
2020	42	93	100
All	30	97	100
STECF* (2017)	0	100	100

* *Scientific Technical Economic Committee for Fisheries*

Environmental parameters

In 2013, the average seawater temperature measured at a depth of 4–9 meters was 25.0 °C, with a 95% confidence interval of [24.5, 25.6]. In 2015, the mean temperature was 25.6 °C, with a 95% confidence interval of [25.0, 26.1]. Finally, in 2018, the mean temperature was 26.7 °C, with a 95% confidence interval of [25.0, 26.1]. Average temperatures over the years changed in parallel with the percentage of mature individuals at age zero (Figure 2). What is noteworthy is that 2018 the year with the highest percentage of young matures compared to other years (Table 2), was also the year warmer than 2013 and 2015 (Pairwise comparisons using t-tests; p-value < 0.05) (Figure 2, (ANOVA; $F(2, 37) = [16.9]$, $p<0.001$)).

SSB Estimation Results

The two prepared maturity schedules were used to compare the XSA model's SSB estimations. One of them was used in the last accepted assessment of STECF (2017), and the other was estimated from the result of this study to be 0.30, 0.97, 1.00, 1.00, and 1.00 for the ages of 0, 1, 2, 3, and 4 (plus-group), respectively (Table 2). As a result, while the common trends are principally congruent (Figure 3), the SSB estimates from the different model runs are significantly different (Welch Two Sample t-test, $p<0.05$). According to this finding, the stock assessment carried out by STECF in 2017 did not account for the ratio of mature fish at age zero, leading to an underestimation of the SSB status.

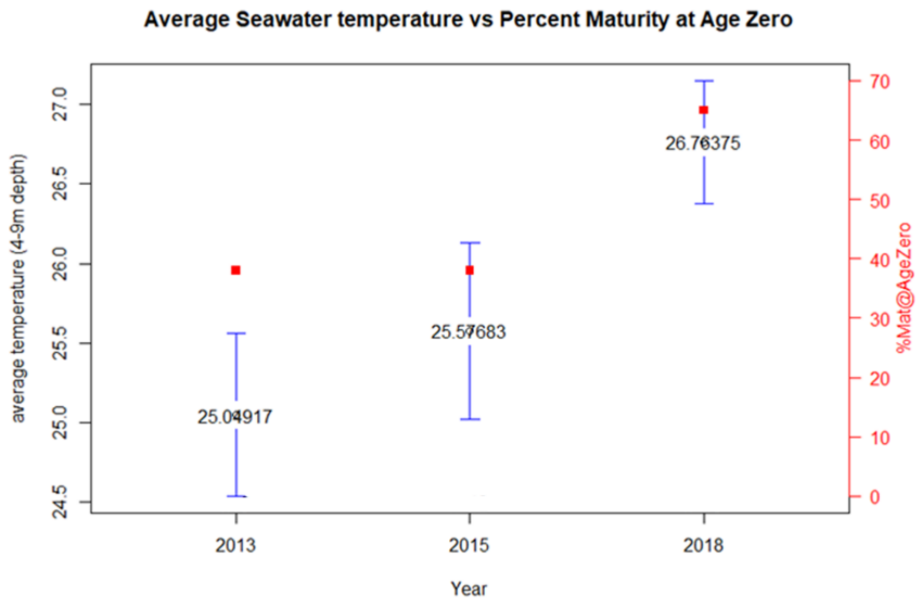

Figure 2. The average seawater temperature for 4-9 m depth (primary axis) and the percentage of fishes achieving sexual maturity at age zero (secondary axis, red squares) for 2013, 2015, and 2018.

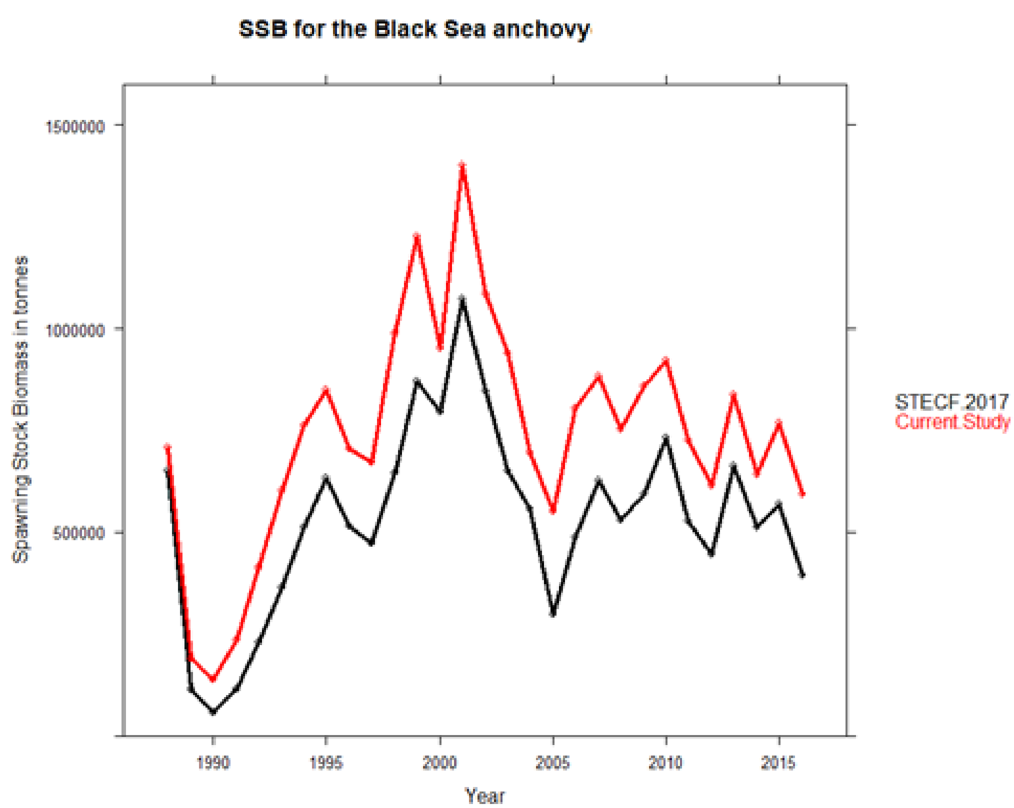

Figure 3. SSB results, estimated by XSA for 1988-2016 with two different maturity datasets.

Discussion

This research offers supplementary information for enhancing anchovy stock management by estimating the first maturity length and rate of mature fish at age zero. This study also attempted to draw attention to the impact of maturity information on spawning stock biomass estimation and hence management applications.

The length at first maturity (7.6 cm in TL) estimated in this study suggested a significant decrease compared to previous studies based on samples collected from the southern Black Sea. Such that in the study carried out by Ünsal in 1989, the first maturity length was given as 10 cm TL. In the studies conducted in the following years, it was estimated as 9.3 cm TL (Kayalı, 1998) and 8 cm in (Artüz, 2003). It is very likely that the difference originated from the sources of the samples. Most of the studies addressing the maturity size were based on the commercial catch in which the size groups smaller than the minimum landing size of 9 cm TL (Üstündağ, 2010) were not fully represented. However, recent studies displayed some signs of a decrease in the first maturity size. According to the L_{50} estimates for the years 2002–2018 shared by Çiloğlu and Şahin (2022), the first maturity sizes varied between 8.57 and 10.53 cm in TL.

In the northern Black Sea, however, the first maturity size estimates were closer even smaller to the one estimated in this study. Lisovenko and Andrianov (1996) estimated the length at first maturity within 5.5-6.5 cm in fork length. In addition, Chashchin et al. (2015) reported the first maturity length for the Black Sea anchovy as a standard length of 6 cm. Converting the given fork and standard lengths to the total fish length (Chesalin et al., 2020), 6-7 cm range and 7 cm were obtained, respectively.

The regulation of a minimum landing size for anchovies has pros because it allows them to spawn in the sea before being exposed to the fishery. This helps to prevent recruitment overfishing, which can have a negative impact on the abundance of anchovies (Lleonart, 2005; Pastor et al., 2002). Yet, it also has some cons. It is widely recognized that fish stocks that are overexploited have a tendency to reach maturity at a smaller size (De Ross et al., 2006; Dieckmann and Heino, 2007; Rochet, 1998; Trippel, 1995;). Fish that are larger in size and mature at a slower rate are at a higher risk of being caught in the size-selective fishery. The reduction in older fish in the stock can lead to an accumulation of smaller and younger fish in the population that are less affected by selective fishing (Arula et al., 2017; Hixon et al., 2014; Trippel,1995; Wright and Trippel, 2009). Early-mature-smaller fish tend to lay smaller and fewer eggs compared to older and larger fish during spawning (Arula et al., 2017; Hixon et al., 2014). The succession of the eggs and larva from the early-matured fish evolves the population's genotype towards the smaller in size and early-matured individuals (Arula et al., 2017; Law, 2007; Trippel 1995). This situation might be one of the reasons for the decreasing trends in the first maturity size of the Black Sea anchovy. These young adult trends can accumulate and be permanent in the long run. In the worst-case scenario, it may cause the adult body size to decrease in the population of the Black Sea anchovy. Thus, overfishing causing early maturation can have an impact on recruitment abundance and stock productivity, ultimately threatening the sustainability of the stock. The best example of this phenomenon is the Atlantic cod, *Gadus morhua*. Overexploitation caused the stock to suffer from fewer in number (collapse), decreasing fish size and age at maturity, lower fecundity, and decreased egg number. (Trippel, 1995 and the references therein).

The decreasing trend in fish size and in size at first maturity may eventually increase the probability of fish being mature in their first year of life. Indeed, the results of this study also show that, on average, 30% of zero-year-old individuals (Table 2) are actively spawned at age zero. In fact, the estimated zero-year-old anchovy population in this study is relatively higher than the estimated 25% of the total anchovy population sampled by Mikhailov (1992) in August 1987 before the collapse of the anchovy fishery. Furthermore, considering the rate of zero-year-old and mature anchovy in the total sampled population, it was found to be 8%, higher than the 3% previously predicted by Lisovenko and Andrianov in 1996.

Temperature, on the other hand, plays a crucial role in creating the ideal environmental conditions for growth and reproduction. It can influence the rate of growth and gametogenesis by stimulating the production and release of hormones (Lowerre-Barbieri et al., 2011; Pankhurst, 2016). Thus, increasing temperature will drive somatic growth and shorten the time being reach sexual maturity (Jonsson et al., 2013). Temperatures within the years at the sampling sites show significant inter-annual variability. In 2018, when average seawater was relatively warmer, a relatively high percentage of maturity at age zero was observed. In these circumstances, it can be assumed that temperature can be a triggering factor in the phase transition of fish from the immature to the mature state

(Lowerre-Barbieri et al., 2011; Pauly, 2019). However, this triggering effect may also be evaluated as a stress factor in fish biology by increasing oxygen demands (Pauly, 2019; Trippel, 1995). Bergmann's rule (Bergmann, 1848) may be the reason that lay behind the increase in young mature anchovy at age zero, especially in 2018 (Hattab et al., 2021).

Moreover, Gill-Oxygen Limitation Theory (GOLT) by Pauly (1984; 2021), can also explain the high percentage of mature Black Sea anchovy individuals at age zero in high-temperature years of the Black Sea. Some studies show that temperature increase decreases the body size of the anchovy, such as the European anchovy (*Engraulis encrasicolus*) in the Mediterranean region (Hattab et al., 2021). In addition, Yoneda et al. (2015) showed that Japanese anchovy (*Engraulis japonicas*) living in high-temperature regimes mature at a much smaller size and younger age when compared to ones living in colder regions. Therefore, in the Black Sea anchovy case, in order to truly comprehend the underlying cause, this situation should be investigated in detail.

So, all these possible reasons and the expected global warming effects on the Black Sea (Barange et al., 2018) may eventually affect the first maturity length, size of fish in the population, and stock productivity. Therefore, accurately monitored first maturity data is also essential for estimating the Spawning Stock Biomass (Heinisch et al., 2014). This study used a maturity schedule sensitivity test, which revealed that when maturity information is underestimated, the SSB is also underestimated. In a scenario where the decreasing first maturity length, the increasing rate of zero-age mature fish, and therefore the decreasing average size of anchovy in the population will continue, even if abundance increases, biomass will be underestimated.

Finally, according to Arula et al. (2017), "commercial fisheries can only be managed sustainably if population characteristics such as age and size at first maturity stabilize over time." By this and by the results of this current study, it can be inferred that the Black Sea anchovy, being the largest stock in the Mediterranean and the Black Sea deserves close monitoring to better manage fishery targeting this precious stock

Conclusion

It can be concluded that the anchovies in the Black Sea reach sexual maturity at smaller sizes. Therefore, decreasing first maturity size and the increasing rate of mature young in the population should be considered as forewarning signals for the future of the Black Sea anchovy stock. Maturity information containing many details about the stock should be monitored annually as an indication of changes in the fishery pattern and environmental conditions. In addition, considering climate change and its impact on fish populations; it also reveals the fact that maturity information can be a tool to understand the future of the stock. Therefore, a standard maturity staging method applied by all Black Sea countries can be a good starting point for further investigations and their compatibilities. Because, if maturity information used in the stock assessment studies does not reflect the stock's current condition, the risk of under or overestimating the SSB over time will have arisen. In the long run, it will inevitably lead to inadequate management plans and affect the sustainable management of the stock.

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Ethical Approval

During the study, no treatment/experiment was implemented on the live animal. All sampling and laboratory work on fish complied with the Republic of Turkey Ministry of Agriculture and Forestry animal welfare laws, guidelines, and policies as approved by the Republic of Turkey Ministry of Agriculture and Forestry Central Fisheries Research Institute, and the Middle East Technical University Animal Experiments Local Ethics Committee (KAMAG-110G124).

Conflicts of Interest

The authors declare no competing interests.

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