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The reproduction and growth of the sardine *Sardina pilchardus* in West Mediterranean, Morocco

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ABSTRACT

The sardine, *Sardina pilchardus* (Walbaum, 1792), is the most abundant fish species in the eastern part of the Moroccan Mediterranean Sea. The biological indicators of reproduction (sex-ratio, gonado-somatic index "GSI", length at first sexual maturity "L_{50}") and growth (parameters of growth of Von Bertalanffy) were calculated from the treatment of the 529 individuals under-sampled during the period of study from January to December 2019. The size classes' frequencies distribution of the commercial samples is understood between 7.5 and 20 cm of the total length (TL). Sex-ratio (males: females) total is lightly in favor of the females. According to the present results of seasonal variations in maturity stages and gonado-somatic index (GSI), the reproductive season of *S. pilchardus* collected, is extended from January to April for both sexes and the fish length at first sexual maturity (L_{50}) is 13.29 cm (TL). Growth results indicated a major allometry growth for which the weight increases proportionately faster than the length.

INTRODUCTION

Morocco ranks among the countries with the most fish in the world thanks to its two sea fronts, Mediterranean and Atlantic, and a coastline that extends over 3,500 km. Indeed, the fishing sector is the main engine of Morocco's economy with catches reaching more than one million tons, a GDP of 2 to 3%, and nearly 700,000 direct and indirect jobs generated **(MPM. 2018)**. For rational exploitation of these resources, assessments of their stocks and determination of their state of exploitation are essential. To this end, basic biological studies are necessary.

Sardines remain the flagship product of the fisheries sector, with national production exceeding 850,000 tons annually. It also represents more than half of the total Moroccan fisheries production (52%) **(MPM, 2018)**.

In this context, it was aimed to study the biology of sardine (*Sardine pilchardus*) landed by fishing in the eastern region of the Moroccan Mediterranean Sea. This particular specie was chosen because of its economic importance and the fact that it is the main of small pelagic fish landed by fisheries.

MATERIALS AND METHODS

1. Location of sampling sites

Biological sampling of sardines was carried out at the ports of Al-Hoceima, Nador and Ras Kebdana **(Figure, 1)**.

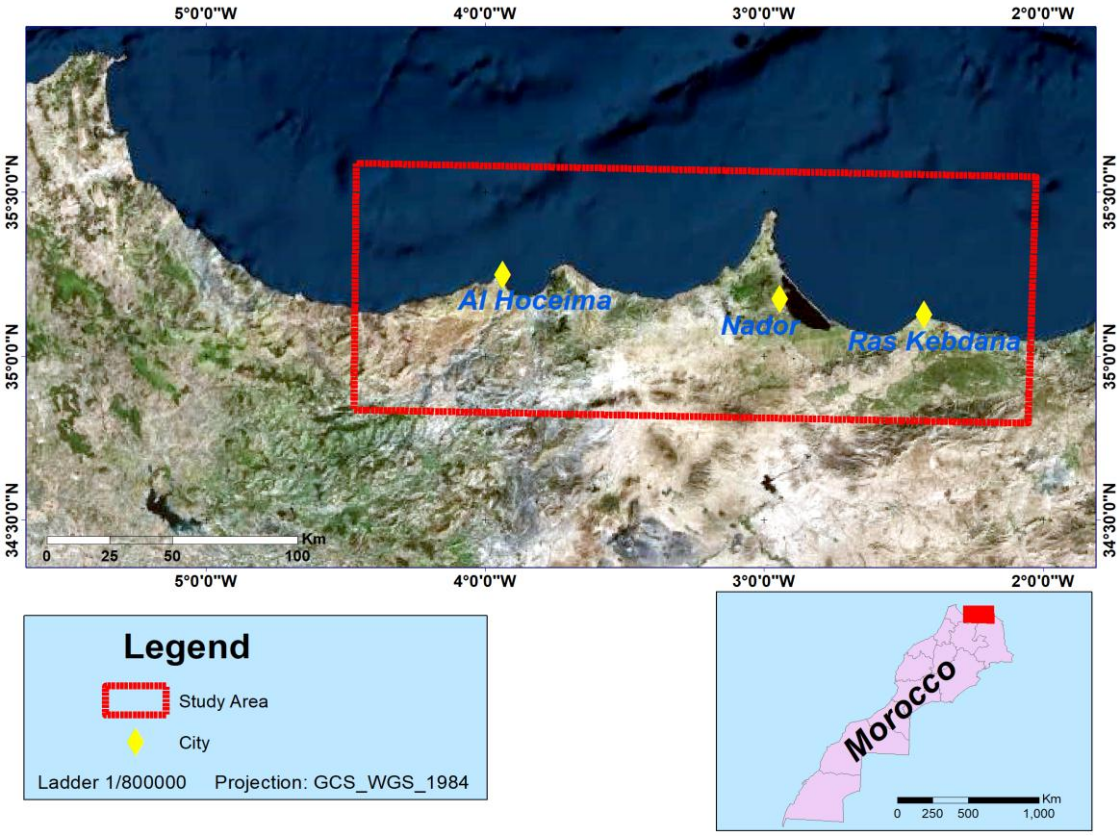

Figure 1: Sampling sites at sea Mediterranean

2. Sampling method

For sardine and small pelagic fishes in general, the sampling strategy followed is systematic with simple random sampling. The methodology for biological sampling of small pelagic species consists in principle of randomly selecting the first vessel from the first ten and then sampling the landing units at intervals of ten in the order of entry of the vessels. It should be noted that this sampling shall cover $1/10^{th}$ of the number of vessels having landed on the same day.

Then a random sample of approximately 3 kg of fish is taken from each sampled vessel. Surveys of fishing efforts are carried out in parallel (name of vessel, number, time of departure and arrival, number of positive and negative operations, distance/coast, and distance/port). This sample will be used for size measurement and a sub-sample is taken for the collection of biological parameters.

The main biological parameters collected per individual are: total length, total weight, gonad weight, eviscerated weight, sex, stage of sexual maturity, gonad weight and finally, otoliths are collected for age reading.

3. Reproductive biology

3.1. Sex-ratio

The sex ratio is a parameter that makes it possible to assess the stock's demographic structure and spawning biomass **(Kartas & Quignard, 1984).** It is considered to be the proportion of males and females in the sample. It is calculated as follows:

$$\% \text{ males} = (n_1/N) * 100 \qquad \% \text{ females} = (n_2/N) * 100$$

Where:
N: Sample size
$\mathbf{n_1}$: number of male individuals
$\mathbf{n_2}$: number of female individuals

The variation in the sex ratio was carried out by month and by size class.

3.2. Stages of maturity

Determination sexual maturity stage had been carried out macroscopically using a five-stage sexual maturity scale **(Holden & Raitt, 1974).** These stages of sexual maturity had different characteristics**:** stage I (Immature or inactive), stage II (spent), stage III (Active or developing), stage IV (Active-ripe or developed) and stage V (Spawning or ripe-running).

3.3. Gonado Somatic Index (GSI)

The monthly evolution of the gonadosomatic index allows us to determine the laying period. This ratio is calculated by the following relationship:

$$\% \text{ GSI} = (Wg/Ws) * 100$$

Where:
Wg: gonads weight (in grams)
Ws: somatic weight (eviscerated weight) (in grams)

3.4. Size at first sexual maturity (L_{50})

The determination of L_{50} (length at which 50% of the fish are mature) was made by grouping the individuals sampled during the main spawning season by sex and size class. Then, the proportion of mature individuals in each size class was calculated. The threshold of sexual maturity is set at stage III, which corresponds to the beginning of the gonadal development phase **(FAO, 1978)**.

Size-proportion pairs of mature individuals are fitted to the symmetrical sigmoid logistic curve **(Pope *et al*., 1983** and **Delgado & Fernandez, 1985)** whose mathematical expression is as follows:

$$P = 1 / (1+e^{-(a + b * L)})$$

With,

P: Proportion of masts by size class, **L**: Total length, **a**: Original order and **b**: Slope

Parameters **a** and **b** are obtained, after the logarithmic transformation of expression, by the method of least squares **(Sokal & Rholf, 1979)**.

$$Ln (P / (1-P)) = a + b * L$$

The representation of the maturity ogive is carried out by considering all pairs of values except those with a proportion: P = 0 and P = 1.

$$L_{50} = -a / b$$

The age of first sexual maturity is then deduced using the growth model established on sardines.

4. Growth biology

4.1. Relative growth or length-weight relationship

Most fish have an allometric relationship between fish size and weight **(Beverton & Holt, 1957)**. This relationship is influenced by food availability, gonad development and reproduction **(Fréon *et al,* 1979)**. It is very useful in fisheries biology to estimate the changes that growth can cause in the morphology of the species. It is translated into an equation of the type:

$$W = aL^b$$

Where:
W: Weight of fish in grams.
L: Total length of the fish in centimeters.
a: Constant. **b**: Coefficient of allometry.

4.2. Linear growth

In population dynamics, the model of **Von Bertalanffy (1938)** is chosen to express linear growth mathematically. This model adjusts both from the age of recruitment and from the age at first capture. In addition, it is the easiest to integrate into production equations (**Daget & Le Guen, 1975**). Its expression is as follows:

$$L_t = L\infty\ (1 - e^{-k\ (t-t_0)})$$

Where:

L_t: Length of fish at time (in year).

$L\infty$: Asymptotic length that a fish would expect at the theoretical infinite age. **L** should not be systematically confused with the maximum size actually reached by the fish.

K: Coefficient representing the metabolism of fish. It is the slope of adjustment between size and instantaneous increase in size. In the derivative, K sets the mode of decreasing growth rate as size increases.

T_0: The theoretical age at which the fish would be zero in size (the curve cuts the abscissa axis), but at birth the larvae or juveniles already have a length that is not zero.

RESULTS

1. Sampling intensity

During the study period a total of 111 samples were taken for size measurement, of which more than 11,707 individuals were measured. In addition, 529 fish were collected for the biological study **(Table 1)**.

Table 1: Intensity of biological sampling of sardines

Months	Number of samples	Number of individuals measured	Number of individuals Under-sampled
January	12	1412	40
February	18	1900	80
March	11	1235	60
April	9	912	60
May	5	440	80
June	6	528	30
July	5	460	30
August	6	540	20
September	10	1180	30
October	12	1430	29
November	9	890	30
December	8	780	40
Global	**111**	**11707**	**529**

2. Size structure

The size frequencies of sardines in the year 2019 allowed us to determine the size composition of the sardines landed. The sizes range from 7.5 to 20 cm.

The distribution of sardine size frequencies is bimodal; the main mode being located at 13.5 cm and the secondary mode at 11 cm **(Figure 2)**.

3. Reproductive biology

3.1. Sex-ratio

The monthly distribution of the sex ratio of sardines landed in the Eastern Mediterranean region of Morocco is illustrated in **Figure (3)**; the overall sex ratio is slightly in favor of females. Indeed, the percentage of females in the sampled population was 53%, while that of males is 47%. The monthly evolution of the sex ratio showed that the frequency of females and males varied respectively between 26% and 80%, and between 20% and 74%.

3.2. Maturity stages

These Individuals in maturity stages 1 and 2 are considered immature while individuals in the upper stages are considered mature. The mature stages are dominant from November to April while the immature stages dominate between May and October **(Figure 4)**.

3.3. Gonado- Somatic Index (G.S.I.)

The gonado-somatic index also known as the "maturity coefficient" reflects the change in gonad weight relative to the weight of eviscerated fish. The monitoring of the monthly variation in the GSI during the year 2019, allows us to have information on the development and maturation of the gonads, as well as on the laying period.

The monthly change in the GSI is almost similar for both sexes. The GSI curve peaks in January-February and then declines until May. The GSI then begins to rise from October. The egg-laying period would mainly take place between January and April **(Figure, 5)**.

3.4. Size at first sexual maturity (L_{50})

The establishment of “proportions mature individuals per size class” curve made it possible to conclude on the size at first maturity of Mediterranean sardines from the Al Hoceima region to Ras Kebdana. This size is taken as the length for which 50% of the individuals are mature. The calculation of L_{50} was based on data for both genders combined. The value of L_{50} obtained is 13.29 cm **(Figure 6)**.

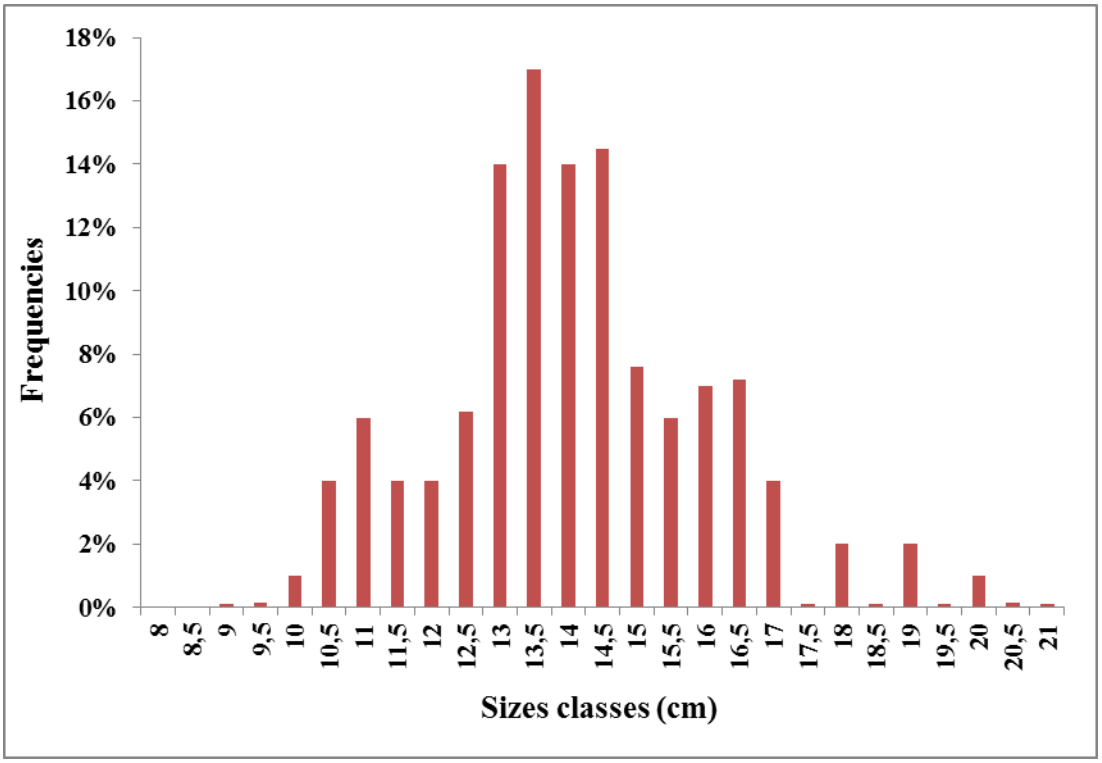

Figure 2: Frequency distribution of sardine sizes

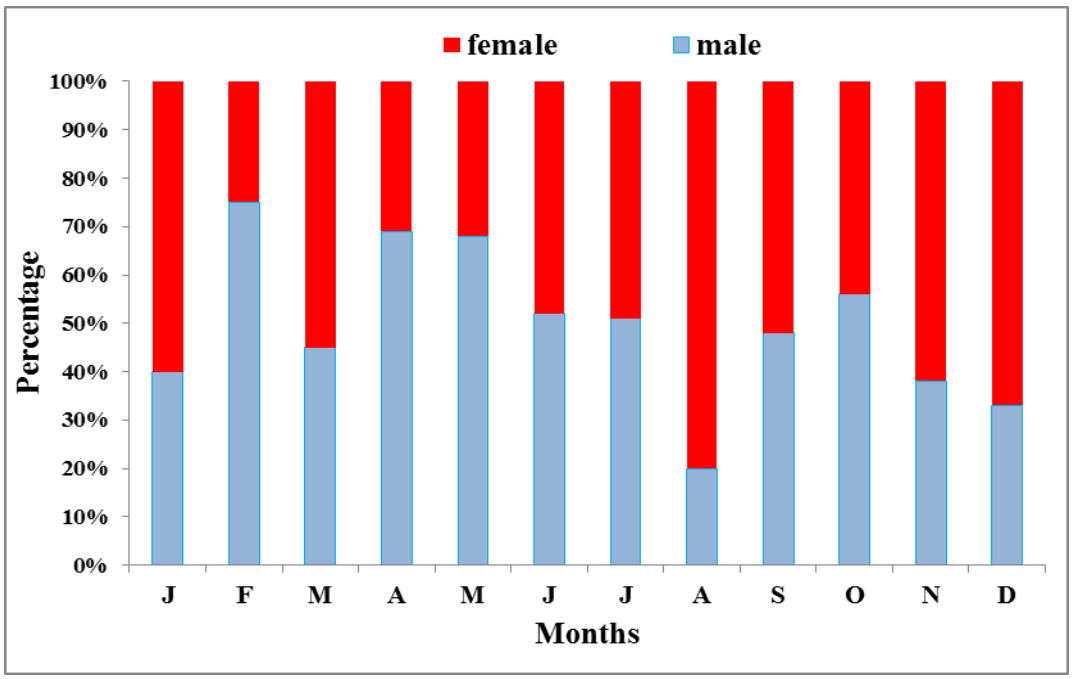

Figure 3: Distribution of relative frequencies of male and female sardines in the Eastern Mediterranean region of Morocco.

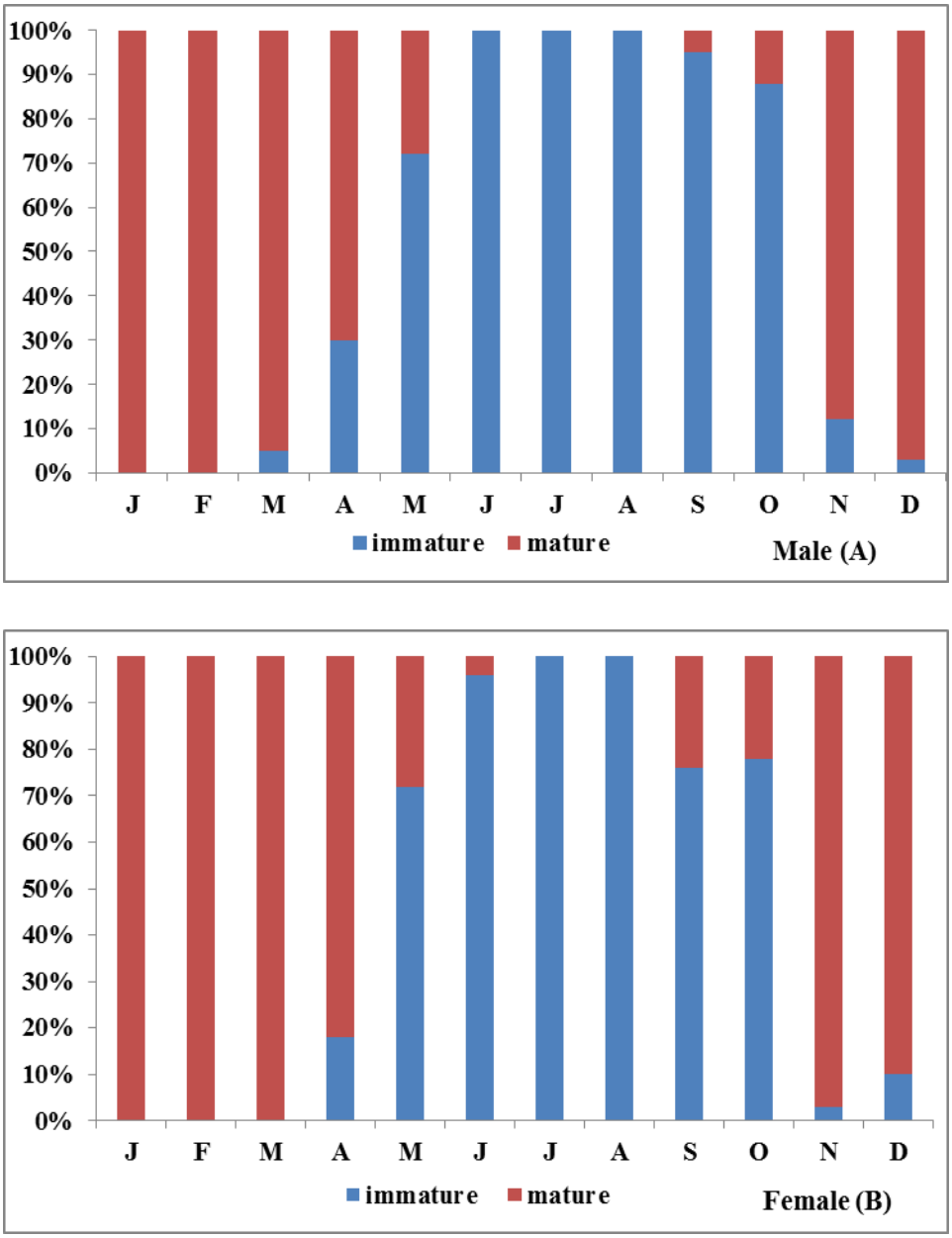

Figure 4: Monthly variation in sexual maturity stages in male (A) and female (B) sardines during 2019.

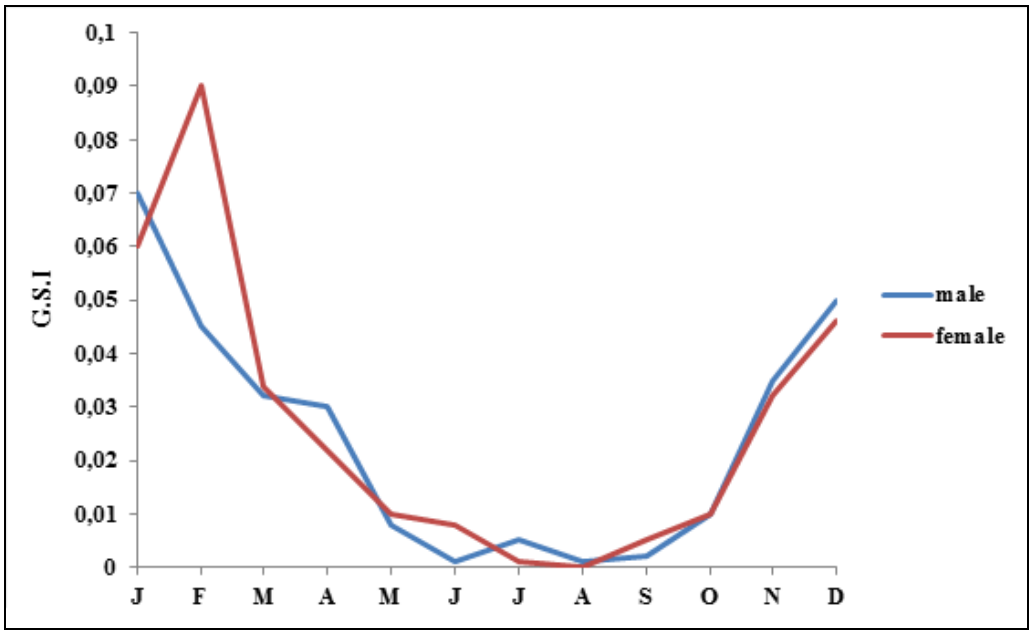

Figure 5: Monthly variation in the GSI for sardines in the studied region

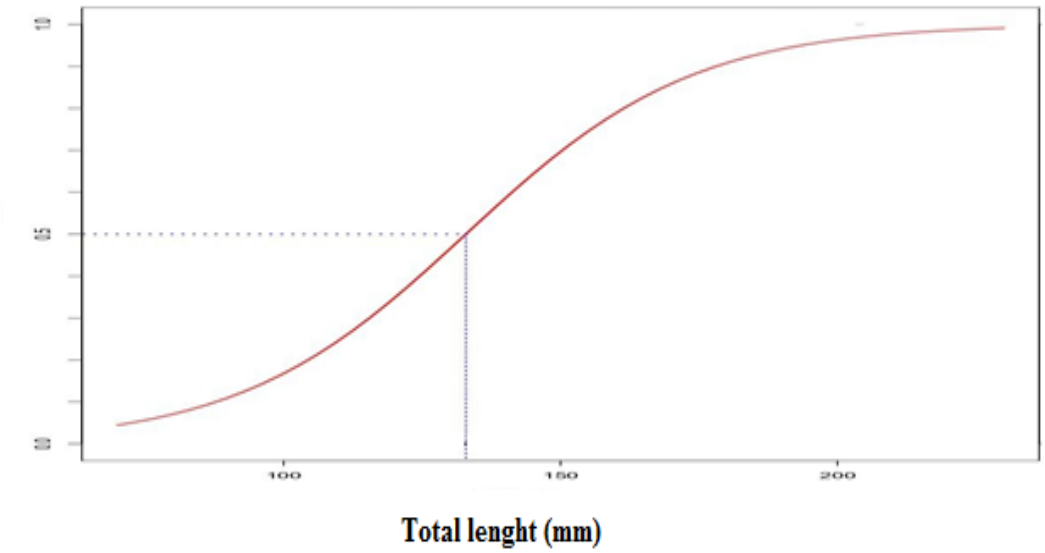

Figure 6: Length at first of sexual maturity of the Eastern Mediterranean sardine

4. Growth biology

4.1. Length-weight relationships

- **Length-weight relationship by sex**

The length-weight relationships were established separately by sex for all samples during the study period. The correlation coefficient, being high for both sexes,

shows a strong correlation between weight and height. The allometry coefficient (b) is slightly greater than 3, indicating a major allometry in both sexes **(Figures, 7 & 8)**.

As a result, the sardine shows a weight growth proportionally higher than its growth in size. The length-weight relationships for the different sexes are therefore:

$$W = 0.0073 * L^{3.0056} \quad \textbf{(for females)}$$

$$W = 0.0068 * L^{3.0363} \quad \textbf{(for males)}$$

Where **W**: total weight, **L**: total length

4.2. Linear growth

- **Direct method (age reading)**

The individual age estimation allowed us to determine the average individual sizes for all sardines sampled in the study area. We then fitted the Von Bertalanffy model equation to the pairs of mean age-length values calculated over all individuals **(Figure, 9)**.

- **Growth curve**

The parameters of Von Bertalanffy's equation, expressing linear growth as a function of time for all samples, obtained by direct reading of age are as follows:

L∞: 20.21 cm, **K**: 0.47 and $\mathbf{t_0}$: -1.93

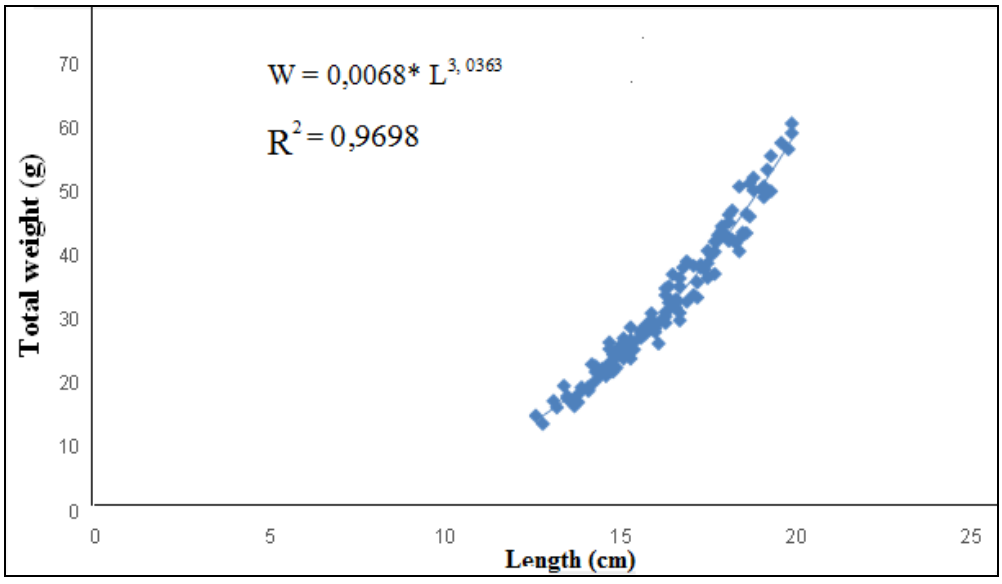

Figure 7: Length-weight relationship of male sardines landed between January and December 2019

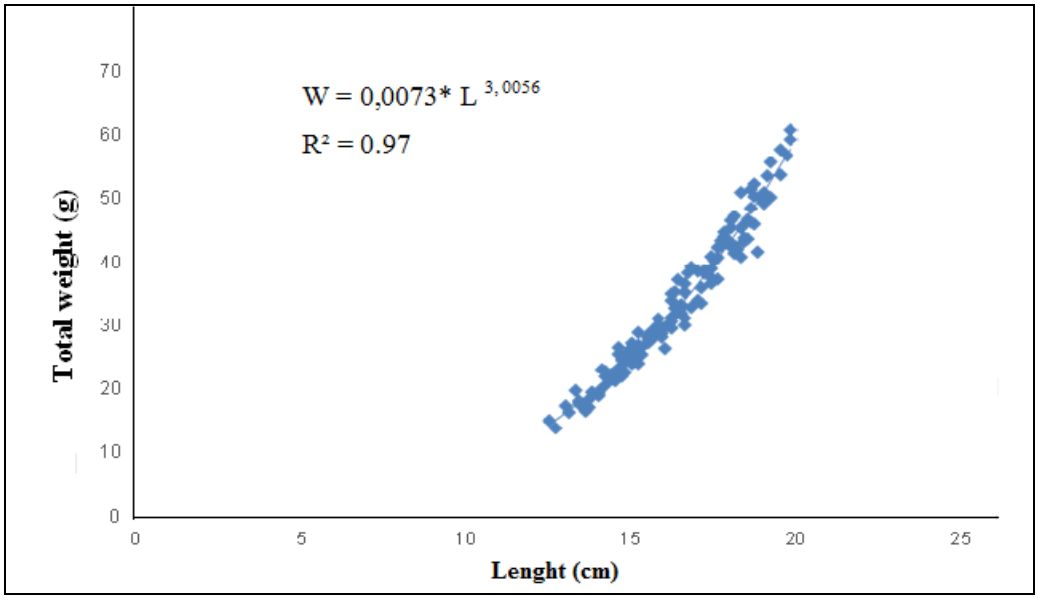

Figure 8: Length-weight relationship of female sardines landed between January and December 2019

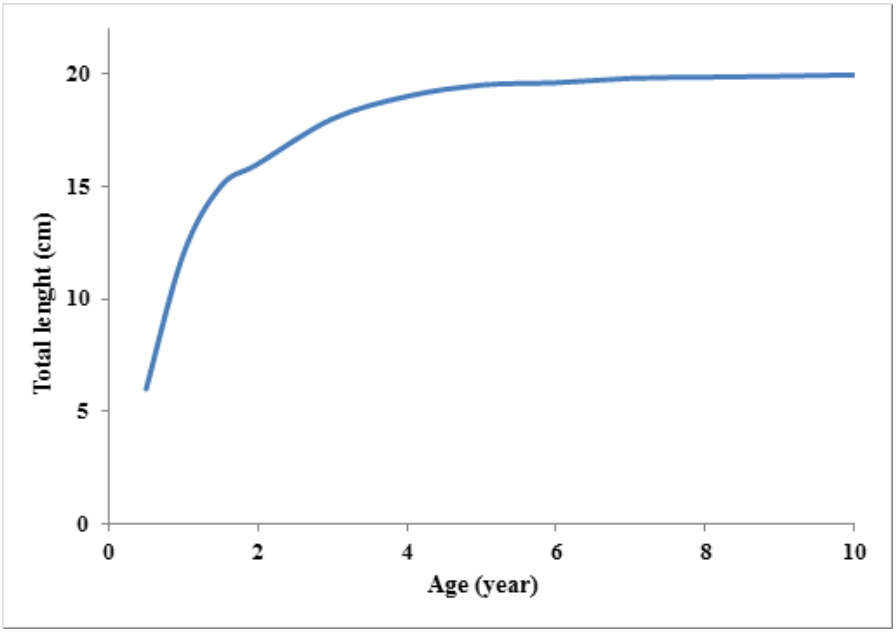

Figure 9: Linear growth curve from age reading (growth ring reading on otolith) versus size.

DISCUSSION

I. Biological indicators

In the Mediterranean area, the size structure of the sardine landed by the purse seine fleet is bimodal. It is marked by a clear dominance of average sizes. The main mode is 13.5 cm, while the second mode corresponds to 11 cm. The size range of sampled individuals is wide (9 - 20 cm). The average length is 13.58 cm and the average weight is 20.79 cm.

II. Growth biology

The Parameters **a** and **b** of the size-weight relationship are highly sensitive to the numbers of fish sampled monthly and the size composition of each sample as reported by **Fréon (1988)**. These two parameters have an antagonistic action in the determination of weight for a given size, i.e., an increase in one is partially offset by a decrease in the other. This was also reported by **Belvèze** (**1984)**.

According to **Amenzoui (2010),** Moroccan sardines show a growth isometry in males and majoring in females, the same result has been observed by **(Kartas, 1981)** in Tunisia and **(Mendes *et al*., 2004)** in Portugal. These differences are generally related to the maturation process, release of sexual products and feeding as observed by **(Furnestin, 1957)** and **(Somoue, 2004)** or hydrological differences in the environment **(Makkaoui, 2008)**.

Comparison of Von Bertalanffy's parameters (L∞, k) in *S. pilchardus* from the Eastern Mediterranean region of Morocco shows that these are higher or lower than those obtained in other regions **(Table 2)**. This can be explained by:

-Differences in sampling areas, years and periods and maximum sample sizes in the different regions.

-The differences observed, due to the heterogeneity of the methods used such as direct reading of otoliths or scales, back calculation, size frequency analysis, the individual cohorts or synthetic cohorts or age reading that is not uniform.

III. Reproductive biology

The overall sex ratio of sardines landed in the area showed a dominance of females over males. This dominance can be explained by the fact that the spatial distribution of the two sexes is not identical. On the other hand, this difference in the sex

ratio distribution as a function of size indicating a high femininity rate for large sizes has been reported **(Lee, 1961**
; Bouchereau, 1981; Kartas, 1981; Belvèze, 1984; Pérez *et al.*, 1992; Abad & Giraldez, 1993; Amenzoui *et al.*, 2006 and **Khemiri, 2006).** This biological characteristic seems to be a general rule in Clupeiformes as it has been observed in other species such as the flat sardinella, *Sardinella maderensis* **(Boëly, 1979),** Venezuelan sardinella (**Fréon *et al.*, 1997)** and the round sardinella, *Sardinella aurita* **(Gaamour, 1999)**.

The monthly variation in sexual maturity stages during the year 2019 showed that there are no great differences between males and females. For both sexes, immatures are encountered practically all year round, particularly between May and October. On the other hand, the proportion of mature females is high practically during the November-April period, which corresponds to the egg-laying season.

Table 2: Summary representation and comparison of sardine growth parameters obtained in different regions of the world.

Region	L ∞ (mm)	t0 (year)	K	Age (year)	Authors
Algeria	18.9	-	0.46	5	**Brahimi et al. (1998)**
Adriatic	20.5	-0.5	0.46	1-8	**Sinovcic et al. (2008)**
North West Mediterranean	19.9	-2.73	0.35	0-8	**Morales-Nin & Pertierra (1990)**
W. Mediterranean (Alicante)	22	-	0.29	-	**Larrañeta (1975)**
Moroccan Atlantic	21.6	–0.129	0.88	-	**Delgado et al. (1981)**
Study area	20.21	-1.93	0.47	1- 6	**Present work**

During the study period, the variation in the GSI showed that the laying period could take place between January and April. Indeed, this period is characterized by a high maturity of the female gonads. Previous histological studies have confirmed the reproductive period determined by the GSI in *S. pilchardus*. These results are comparable to those reported by **(Pinto & Andreu, 1957)** and modified by **(Mouhoub, 1986)** along the Algerian coast, which indicated that *S. pilchardus* generally reproduces during the same period:

- Between December and March, in the Algerian **(Djabali *et al.*, 1989)** and Oranese coasts **(Tomasini *et al.*, 1989)**;
- Between January and March, in the Bay of Annaba and on the Atlantic coast **(Bedairia & Dejbar, 2009)**.

CONCLUSION

The aim of this study is to examine the biology of *Sardina pilchardus* (Walbaum, 1792) fished in the eastern region of the Moroccan Mediterranean Sea. On the one hand, studying sardine's biology was addressed by the analyze of growth and reproduction of these species and it should be noted that knowledge of sardine's biology, as well as the fishery state, were taken into account as key elements in order to study the state of the stock and to ensure rational and sustainable management. On the other hand growth studies showed that sardines have rapid growth and a short life span. Studying the linear growth of sardines (sexes combined) through calculating Von Bertalanffy's parameters revealed that growth coefficient K is 0.47 and asymptotic length $L\infty$ is 20.21 cm by the direct age reading method.

Sardines are multiple breeders that lay several times during the same breeding season. It is noted that the sex ratio is in favor of females and the size at first sexual maturity in sardines has been estimated at L_{50}=13.29 cm total length.

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