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Age Assessment and Growth Analysis of *Mugilcephalus* in the South Moroccan Atlantic

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ABSTRACT

This study is the first exploration aimed at estimating the age of *Mugilcephalus* (grey mullet) in the southern Atlantic region of Morocco using otolithometry, a sclerochronological technique for analyzing growth rings in otoliths that were collected from 122 specimens from this region. The primary objective was to understand the correlations between age, weight, and length of individuals to unveil growth parameters necessary to implement species stock assessment models. This study exhibited five distinct age groups ranging from 4 to 8. The growth parameters were estimated for both sexes combined ($L\infty$ = 949.21mm, K = 0.113, and t_0 = -1.772), and the von Bertalanffy equation was Lt = 94,921* [1 - e-0.113 (t+1.7727)]. The growth performance index (φ′) was 5.93, and T max was 26.54 years. The statistical analysis showed a significant correlation between age and length growth equations for both sexes, indicating that growth rates accelerated to a peak before slowing. There is a nonlinear relationship between age and length increment. The independent samples t-test revealed no significant difference in mean weight between males and females. Age analysis and growth in weight and length provide the necessary elements to model growth and carry out sustainable management of the stock of this species.

INTRODUCTION

Morocco, benefiting from a 200 nautical mile exclusive economic zone (EEZ) in the Atlantic and one of the world's most significant upwelling zones, stands out as one of the richest fishery zones, with a production potential exceeding 1.5 million tons annually **(Doukkali& Kamili, 2018)**. However, the sustainable management of these fisheries resources requires a deep understanding of the biology of the targeted species, emphasizing the crucial importance of sclerochronology **(Panfili *et al*., 2002)**. The study of the sclerochronology of *Mugilcephalus* provides essential scientific and economic data, allowing the identification of key management elements. This study is particularly groundbreaking since it represents the first comprehensive exploration of the age estimation of *Mugilcephalus* (Mullet) in the southern Atlantic region of Morocco. It aimed to estimate the age of *Mugilcephalus* in the study site using otolithometry, a sclerochronological technique for analyzing growth rings in the otoliths. Another objective was to compare the results obtained by the morphometric method for estimating age and growth parameters with the method based on otoliths. The present research contributes to understand the correlations between age, weight, and length of individuals to unveil specific growth patterns. These studies are crucial for the assessment and sustainable management of the *Mugilcephalus* stock, a species of a significant economic importance along the southern Atlantic coast of Morocco **(Doukkali& Kamili., 2018)**.

MATERIALS AND METHODS

In this study, only individuals whose otoliths were recovered are included, unlike the previous study conducted in 2022 **(Lamraouhi *et al,* 2022)**, where this procedure had not been applied.

Study area

The study was conducted in the southern Moroccan region, precisely between Cape Boujdour and Cape Blanc (Fig.1). This area is known for its significant mullet landings; eminently, it is an area characterized by a permanent upwelling phenomenon throughout the year **(Moujane *et al.*, 2011)**.

Fig. 1. Study area, south of Morocco **(Mapsland, 2024)**

Data collection

A total of 122 grey mullet specimens were sampled from various locations in the southern Atlantic region of Morocco between February 2017 and January 2018.The specimens were gathered from different sampling stations, from longliners, purse seiners, and small board. The sample individuals were identified, measured, and weighed in the laboratory.

1. Extraction of otoliths

Otoliths were collected in laboratory; the extraction of otoliths was performed through a transverse section of the head at the boundary of the upper edge of the operculum. They were obtained by a sagittal cut of the head. A total of 122 samples were examined, and 176 sagittal otoliths were collected. The otoliths underwent several treatments including extraction from fresh fish, focusing on the sagittae pair due to their large size and regular shape, followed by cleaning and drying, storing in plastic tubes such as "Eppendorf" tubes, preparation for analysis involving the use of thin sectioning technique to aid in estimating the age of the *Mugilcephalus* species, marking the nucleus of the otolith before embedding it in resin, fixing between layers of resin in an aluminum mold, drying resin molds, and then cutting thin sections using an "ISOMET 4000" saw, and finally reading and analyzing growth marks through observation of thin sections using an Olympus SZX7 stereoscopic microscope in reflected light, use of clearing liquids to enhance visibility of growth rings, and analysis of seasonal marks to determine the age of the fish, with a subsequent length increment analysis performed in *Mugilcephalus* to examine the influence of age on its length growth." To analyze the variation of the length increment according to age, the advanced regression techniques were used to determine the nature of the relationship between these two variables. Using polynomial, exponential and logarithmic regressions, the models were evaluated using the corrected Akaike information criterion (AICc).

Age estimation process

Some informative data were required to assign a precise age to the individual **(Panfili *et al.*, 2002)**, including the following points:

- Date of capture
- Individual birth date (if available)
- Key periods of formation for the considered growth mark
- Nature of the otolith edge

Generally, if:

- Opaque Edge: Age = Number of hyaline zones (N)
- Hyaline Edge:

- the age = Number of hyaline zones (N) if the capture date is between January and June
- the age = Number of hyaline zones - 1 (N-1) if the capture date is between July and December

2. Length-weight relationship

The weight growth model expresses the individual weight of individuals (Pt) as a function of their age. The equation was deduced from the height growth model, using the height/weight relationship.

The weight growth relationship was established from the two equations of the previous models. The weight growth relationship was then written as follows:

W_t= W∞* (1 - e -k (t-t_0))b Where: W_∞ = a*L_∞b

Where,Wt and P∞ are the weights (g) corresponding respectively to Lt and L_∞;K is the growth coefficient from the equation of **Von Bertalanffy (1938)**;b is the allometry coefficient extracted from the lenght-weight relationship.t: the age (years),t_0: the hypothetical age at zero length **(Bagenal, 1978)**.

The t- test was used to assess whether significant differences in weight existed between males and females.

Moreover, the Pearson correlation test was used to examine the relationship between age and weight. This test produces a correlation coefficient (r). The *P*-value was calculated, and it was associated with each coefficient to the statistical significance of the observed correlation. These analyses were carried out using the statistical software R.

A weight increment analysis was conducted to evaluate the impact of age on the weight gain in *Mugilcephalus*, analyzing how the weight growth rate evolved with the age of the species through this study.

The analysis of variance (ANOVA) was used as a statistical method to evaluate differences in the pea increment means between males and females. This analysis aimed to determine whether categorical variables, such as gender, have a significant effect on weight gain. ANOVA tests the null hypothesis that all group means are equal.

To examine the relationship between age and weight increase in the *Mugilcephalus*, first, a simple linear model was constructed, assuming that weight increases steadily with age. Then, a quadratic model was developed to incorporate the acceleration phase and the slowdown phase in weight gain. The analysis of variance (ANOVA) was employed to assess the relevance of these models.

Growth performance index and longevity

The growth performance index (φ') was estimated using the phi prime test formula: φ' = log(k) + 2 log(L∞), where k represents the growth coefficient and L∞ denotes the asymptotic length of the fish.On the other hand, the estimate of lifespan or longevity (Tmax) was determined by the approximate maximum age that a fish would reach, calculated as Tmax = 3/k, as proposed by **Taylor (1958)**.

RESULTS

1. Age estimation

The analysis of 122 sagittal otoliths from *Mugilcephalus* revealed the presence of five distinct age groups (IV, V, VI, VII, VIII) ranging from 4 to 8. The observed maximum age was approximately 8 years, while the minimum age was 4 years. The most frequent age groups were those of 4 years, with 57 individuals, and 5 years, with 52 individuals. Other age groups included 9 individuals aged 6 years, 2 individuals aged 7 years, and 2 individuals aged 8 years (Table 1 & Figs. 1, 2).

The distribution of age groups based on the size (total length in mm) of individuals, sexes combined for *Mugilcephalus* reveals a correlation between age and total length in the *Mugilcephalus* individuals.

Table 1. Age-length in *Mugilcephalus*

	Age group					
Lt (cm)	IV	V	VI	VII	VIII	**Total**
37	1					**1**
39	1					**1**
41	3					**3**
43	10					**10**
45	13					**13**
47	15	6				**21**
49	12	12				**24**
51	2	16				**18**
53		9	1			**10**
55		8	5			**13**
57		1	2			**3**
59			1	1		**2**
61				1		**1**
63					1	**1**
65					1	**1**
Total	**57**	**52**	**9**	**2**	**2**	**122**
Lt moy. (cm)	**45.77**	**51.15**	**55.66**	**60**	**64**	

Fig. 2. Sagittal otolith of *Mugilcephalus* from the 4-year age group: male, TL (total length) = 551mm, captured in November (The red dots represent hyaline areas)

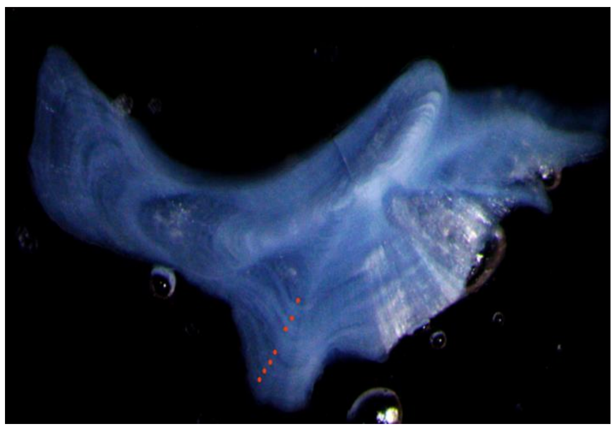

Fig. 3. Sagittal otolith of *Mugilcephalus* from the 7-year age group: female, TL (total length) = 559mm, captured in October

1.1. Length-age

The length growth curve of *Mugilcephalus* was adjusted using the von Bertalanffy model, combining age and length data from both sexes. The parameters of the linear growth relationship were as follows:

$$\mathbf{K = 0{,}113; \quad L_{\infty} = 949{,}21\ mm; \ t_0 = -1{,}7727}$$

and the equation of the von Bertalanffy growth model was:

$$\mathbf{L_t = 94{,}921^{*}\ [1 - e^{-0{,}113\ (t+1{,}7727)}]}$$

The von Bertalanffy equation allows for estimating the length (Lt) as a function of the age (t) of the fish, regardless of the gender (Fig. 4).

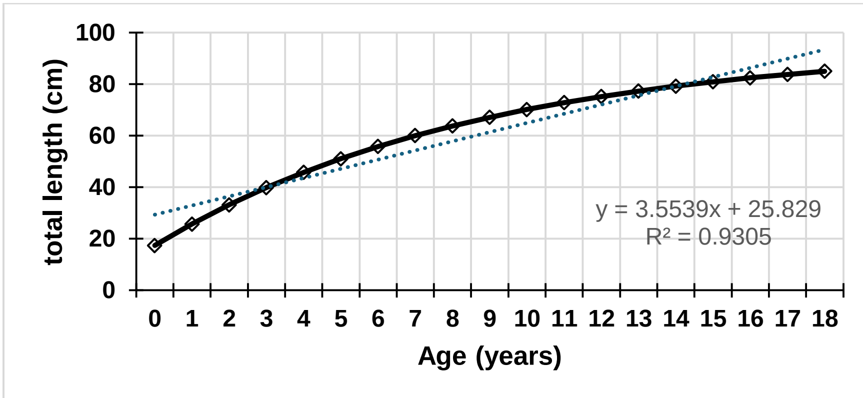

Fig. 4. von Bertalanffy curve for the linear growth of *Mugilcephalus* in the South Atlantic of Morocco

In the first few years of his life, the juvenile grey mullet has a rapid maturity. However, when it reaches maturity, the length growth slows down progressively. It was observed that, the increment length of *M. cephalus* (Fig. 5) is ventilated from 17,36 to 0.54cm for ages from 1 to 25 years. The graph in Fig. (4) shows the evolution of the increment with age, initially there is a significant increase, which quickly reaches a peak, then there is a gradual and sustained decrease. This trend reflects an initially rapid growth that slows over time.

The results of the statistical analysis revealed a nonlinear relationship between age and length increment. The exponential model stood out as the best performing, with the lowest corrected the Akaike information criterion (AIC) among the models tested. This indicates that growth declines exponentially with age. The polynomial model also showed a significant fit, but less optimal than the exponential. The linear and logarithmic models exhibited a poor performance, suggesting that these simple relationships do not adequately capture the observed growth dynamics.

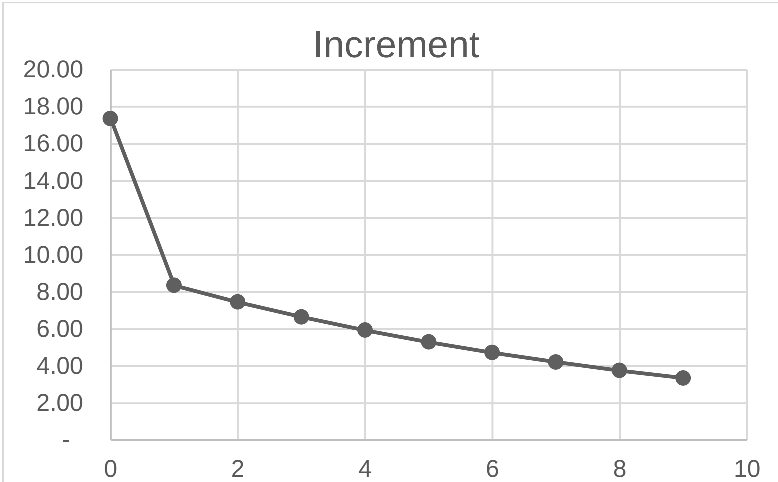

Fig. 5. Back calculation total length (cm) for *M. cephalus* in the study site

1.2. Weight growth- age

The equations below show the relationship between the size (length) and weight of *Mugilcephalus*.

Males:W =0,00002 L ^{2,8511} R=0,7444

Females: W = 0,00008 L^{2,6718} R= 0,8836

Both sexes: W= 0,00003 L^{2,8223} R= 0,8521

- **Absolute weight growth**

The von Bertalanffy equations describing weight growth related to the age (years) for *Mugilcephalus* are outlined in Fig. (6). The weight- length equations for the grey mullet were described as follows:

$$\mathbf{Wt = 6162{,}936\ [1 - e^{-0{,}113\ (t+1{,}7727)}]^{2{,}8511}}\ \text{males ;}$$

$$\mathbf{Wt = 7211{,}184\ [1 - e^{-0{,}113\ (t+1{,}7727)}]^{2{,}6718}}\text{females ;}$$

$$\mathbf{Wt = 7588{,}062\ [1 - e^{-0{,}113\ (t+1{,}7727)}]^{2{,}8223}}\text{both sexes combined.}$$

Females exhibit a faster growth rate than males. The females reach a higher asymptotic weight, potentially reaching up to 7211.184 grams, while males show a relatively moderate weight growth.

However, independent samples t-test revealed that there is no significant difference in the mean weight between males (2977.72) and females (3598.86), with a t-value = -1.1455, degrees of freedom of 50, and *P*-value is 0.2574.

The 95% confidence interval for the difference in means ranges from -1710.225, 467.947, which indicates that the observed difference is random.

The increment of weights at the end of life steadily increases until the age of 8, then it begins to decrease (Fig. 7).

The results of the Pearson correlation analysis show a strong relationship between age and weight for both sexes and sex combined (r = 0.990), males (r = 0.990), and females (r = 0.988). These results suggest that age and weight are linked in the population studied. The 95% confidence intervals for these correlations are very close to 1 for females, males, and both sexes combined. Then, the analysis of variance (ANOVA) was used to explore the effect of sex on weight increment, the results do not show a statistically significant difference between males and females. The F-value of 2.283 and the *P*-value of 0.137 do not allow us to reject the null hypothesis: there is no significant difference in the means between the sexes.

The degrees of freedom associated with the effect of gender are 1, with a total degree of freedom of 50. These results suggest that variation due to gender does not contribute significantly to the total variation observed in our variable of interest, supporting the idea that other factors may play a more important role.

To analyze the increase in weight with age in mugil, ANOVA was used to compare a simple linear model to a quadratic model.

The results obtained indicated that the quadratic model offers a better fit, as evidenced by:

- A significant decrease in the sum of squares of the residuals (RSS) from 181,463 to 51,840.
- A very high F-statistic of 57,511,
- A low *P*-value of 1.06e-07.

These results, marked by very strict significance codes (***), indicated that the relationship between age and weight increase is not simply linear but follows a curve where the weight increase accelerates until reaching a certain age before slowing down, for both sexes. This significant trend suggests the influence of other physiological or environmental factors that affect weight growth with age, which are common to both males and females.

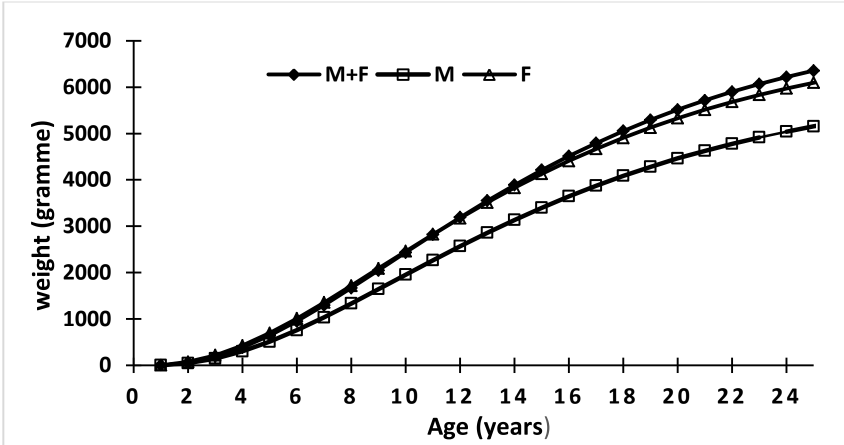

Fig. 6. Absolute weight growth of *Mugilcephalus* along the South Atlantic Moroccan coast

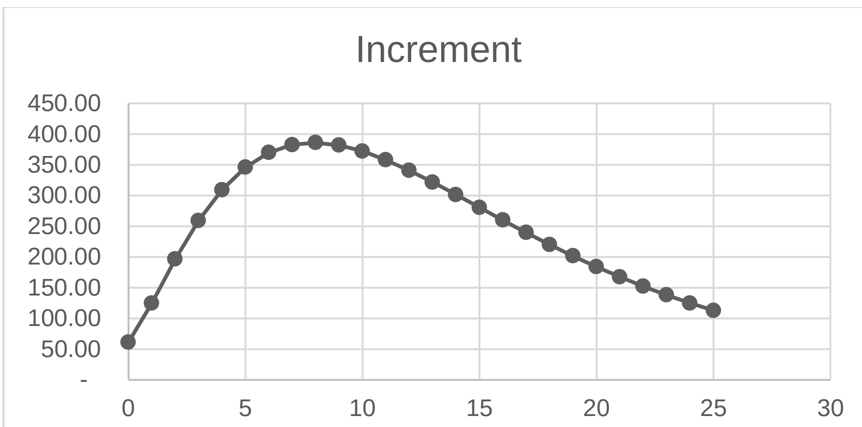

Fig.7. Back calculation total weight of *M. Cephalus* (g) along the South Atlantic Moroccan coast

2. Growth performance index (φ') and longevity (T max)

The growth performance index (φ') for *M. cephalus* in the southern Atlantic region was 5,93, and the estimate of life span (t max) of *M. cephalus* was 26,54 years.

DISCUSSION

The analysis of 122 sagittal otoliths from *Mugilcephalus* revealed the presence of five distinct age groups ranging from 4 to 8. This result is consistent with the data obtained from the method based on morphometric measurements **(Lamraouhi *et al.*, 2022)**. Moreover, the juvenile grey mullet has a rapid growth; however, as they reach maturity, their length growth progressively slows down. The growth of *M. cephalus* has variable rates, it may depend on animal's physiology, local condition and food resources availability **(Whitfield, 2012)**. Overall, growth is rapid in the first year, with fish typically reaching a fork length (Ls) of 140 to 180mm in the tropical and subtropical waters and a Ls of 130 to 160mm in more temperate regions **(Whitfield, 2012)**. (Table 2)

Table 2. *Mugilcephalus* age groups

Study area	**Method**	**N**	**Age group**								**Source**
			I	II	III	IV	V	VI	VII	VIII	
Tamiahua,Mexico	Otolith	232		24.6	28.2	31.7	34.9	37.5			Aguirre *et al.*, 1999
GüllükLagoon	Otolith	132	19.3	24.6	30.7	39.0	43.0				Hoşsucu, 2001
GulfofGökova	Scale	120	22.95	27.6	33.2	35.9	49.5				Kasımoğluan dYılmaz, 2011
KöyceğizLagoon	Otolith	291	13.5	20.6	27.2	31.9	36.0	39.8	43.2		Reis and Ateş, 2020
Southern Atlantic Morocco	Morphometric	281				45,25	50,2	57.9		63.1	Lamraouhi *et al.*, 2022

For L∞, the present study results are generally higher than those of other studies, except for the study of **Thakur (1967)** who reported a close value of 89.62cm. These differences could be due to variations in the maximum height reached by the populations studied or to methodological differences such as sample size, study period, and measurement techniques (Table 3). Additionally, environmental factors, availability of food resources, and fishing pressures may influence fish growth and explain these variations.

Table 3. Comparison of estimated parameters for the von Bertalanffy equation in *Mugilcephalus*

Study area	L∞(mm)	K(an^{-1})	T_0 (year)	Headcount	Source
Estuary System of Mahanadi (India)	896,2	0,15	0,2	2628	THAKUR, 1967
Bonny Estuary (Nigeria)	332	0,55	0,15	246	ALELEYE-WOKOMA *et al*., 2001
Mexico	630,8	0,22	-0,64	66	VINCENT, 2010
Fleuve Estuary Sénégal	687,8	0,42	-0,012	1660	SAAR *et al*., 2012
Chilika lagoon (India)	606	0,28	-0,74	267	SAHOO *et al*., 2012
Dams: BirMchergua and Joumine (Tunisia)	713,6	0,16	0,04	121	Mili, 2015
Köyceğiz Lagoon–Estuary (Mediterranean coast)	599,9	0.169	–0.0132	1195	Reis and Ateş, 2020

The range values of the growth performance index Φ′reported by **Ibanez (2016)** were between 2.421 to 3.468. However, the phi prime calculated in the South Atlantic region of Morocco is higher than in other studies, including the results obtained by **Benjedid *et al.*(2022)** in El Mellah lagoon, where Φ′=3.02. Yet, this high growth performance index of *M. cephalus* may be related to the positive conditions in the South Moroccan Atlantic region (Table 4).

Table 4. Estimated parameters of phi-prime and the von Bertalanffy equation in *Mugilcephalus*

Study area	L_∞ mm	K	phi-prime	Source	Method	Age group
El Mellah lagoon	588	0.3	3.02	Bendjedid *et al.*, 2022	Morphometric	
Bardawill lagoon	521,9	0.261	2.85	El Aiatt and salem, 2017	scales	0 to 6 years
South Atlantic Morocco	804	0,14	5,71	Lamraouhi *et al.*, 2022	Morphometric	4 to 9 years
South Atlantic Morocco	949	0,113	5,93	Current study	Otolith	4 to 8 years

CONCLUSION

The results of this study are of a particular importance in the management of the *Mugilcephalus* stocks, they provide a solid basis for establishing appropriate fishing quotas, defining legal minimum catch sizes and implementing effective conservation measures. The otolithometry's results of the present study made it possible to determine the age of the species since it provides an adequate information on the growth of *Mugilcephalus* in southern Morocco and contributes to better management of this fishery. The identification of five age groups within the studied population enabled a dynamic assessment of the mullet stock. However, there are still important perspectives to explore. From an ecological point of view, it is necessary to develop more in-depth research on the impact of environmental factors on the growth and age of *Mugilcephalus*. Scientifically, extending this study to different geographic regions would make it possible to compare the growth and age patterns of different populations, thereby improving our understanding of the ecological and evolutionary factors that influence them. Finally, on an economic level, all the results could guide sustainable management practices for fisheries regarding this species.The relationship between age and growth, established by the von Bertalanffy model, as well as the height-weight relationship, was described for both sexes.

Competing interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Declarations

Ethical Approval

Not applicable

Funding

Not applicable

Availability of data and materials

Not applicable

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