



Stock identification: Advancing in the knowledge of stock structure as a requirement for stock assessment

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STOCK IDENTIFICATION: ADVANCING IN THE KNOWLEDGE OF STOCK STRUCTURE AS A REQUIREMENT FOR STOCK ASSESSMENT

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ABSTRACT

The uncertainty of stocks identity remains an important challenge for the assessment and management of most West African fishery resources. The stock identification of certain demersal coastal species was considered as a key activity within the Working Package 1 of DEMERSTEM: “Stock identity and assessment”. Three cases of study were selected, by neighbouring countries: 1) *Epinephelus aeneus* and *Penaeus notialis* in Mauritania and Senegal (the Gambia and Guinea-Bissau also included for *P. notialis*); 2) *Pagrus caeruleostictus* and *Pseudotolithus elongatus* in Guinea-Bissau and Guinea; 3): *Pseudotolithus senegalensis* and *Pagellus bellottii* in Côte d’Ivoire and Ghana. A holistic approach was used for stock identification, using a number of complementary techniques: life history traits for all species, morphometry (body shape using truss network for all species and otolith shape for fish species) and genetic analysis only for *E. aeneus* and *P. notialis*. The selected species were sampled for biology in their distribution areas of the countries of each case of study during at least one annual cycle between 2019 and 2021, following standard protocols established for each technique.

The two morphometric techniques showed reliable information for stock identification. Life history traits results were not conclusive for determining the stocks structure, but their study has contributed to improve or provide new knowledge of some biological parameters and features of the species in the studied areas. Genetics results were obtained for *E. aeneus*, being the first time that molecular markers were used to decipher the genetic structure of this species in West African waters. In addition to the information on stock identification, this study has contributed with new genetic information of four of the studied species in West Africa.

Morphometry and genetic results are consistent for *E. aeneus*, both showing one single stock in Mauritania and Senegal. Oppositely, low estimated variability in the body shape within samples of *P. notialis* suggests that each sampled country-area forms a phenotypically homogeneous group, with clear differences to the others. For the rest of species, the following phenotypically homogeneous groups can be distinguished by morphometry, at least at country level: *P. caeruleostictus* and *P. elongatus* (one stock in Guinea-Bissau and another in Guinea, for each species); *P. senegalensis* and *P. bellottii* (one stock in Côte d’Ivoire and another in Ghana, for each species).

Based on the results of our studies, stock assessment and management of all species but *E. aeneus*, should be based on a more detailed resolution of their populations than the one currently used in CECAF. The consideration of the independent stocks proposed might contribute to a more accurate knowledge of the stock status in order to achieve its sustainable exploitation. In addition, the extension of this study to longer periods and to neighbouring areas is highly recommended to determining appropriate geographic boundaries, needed to define the structure and distribution of these West African stocks.

1 INTRODUCTION

The Component 3 of the PESCAO program aims the improvement of the management of marine resources at the regional level, building the resilience of marine and coastal ecosystems to perturbations. Among the three intervention priorities identified for that, the project DEMERSTEM (DEMERSal EcosySTEMs) aims to give answer to Priority 1, that is, improving the knowledge on the state of shared fish stocks and/or fisheries of common interest. DEMERSTEM specifically targets the study of demersal stocks and fisheries in West Africa, with the ultimate goal of providing the basis for better scientific advice on fisheries management that leads to sustainable exploitation in the jurisdictional waters (EEZs) of the coastal states selected as case studies (Mauritania-Senegal, Guinea Bissau-Guinea and Côte d'Ivoire-Ghana).

The uncertainty of stocks identity remains an important challenge for the assessment of West African fishery resources. In general, in the Fishery Committee for the Eastern Central Atlantic (CECAF), resources are assessed as national or shared stocks, but without any biological basis for this structuring. Of the 37 demersal stocks evaluated in the CECAF Working Groups, 51% are considered as shared stocks between two or more EEZs for assessment purposes (FAO, 2023).

Taking this into consideration, the study of the identity of certain demersal coastal stocks was considered as a key activity within the Working Package 1 of DEMERSTEM: "Stock identity and assessment", which aims to improve knowledge about the selected demersal stocks and, in particular, to solve the problems of stock identification, together with the improvement of quality of the data used for assessments, both following the recommendations recurrently made by CECAF.

The studied species were selected by the involved coastal states, matching one or more of the following criteria: i) they are assessed as shared stocks; ii) they are commercially important species; iii) they are emblematic species for artisanal fisheries; iv) some of them are target or by-catch species of European fleets operating under Sustainable Fisheries Partnership Agreements (SFPA) with some countries involved (i.e: *Penaeus notialis* in Mauritania and Guinea-Bissau); v) they are species that have coastal nurseries.

The species selected for each case study are (see Figure 1.1):

1- For Mauritania and Senegal:

- the white grouper (locally called "Thiof") *Epinephelus aeneus*
- the southern pink shrimp *Penaeus notialis* (the Gambia and Guinea-Bissau were also included for this species)

2- For Guinea-Bissau and Guinea:

- the bluespotted seabream *Pagrus caeruleostictus*
- the Bobo croaker *Pseudotolithus elongatus*

3- For Côte d'Ivoire-Ghana:

- the cassava croaker *Pseudotolithus senegalensis*
- the red pandora *Pagellus bellottii*

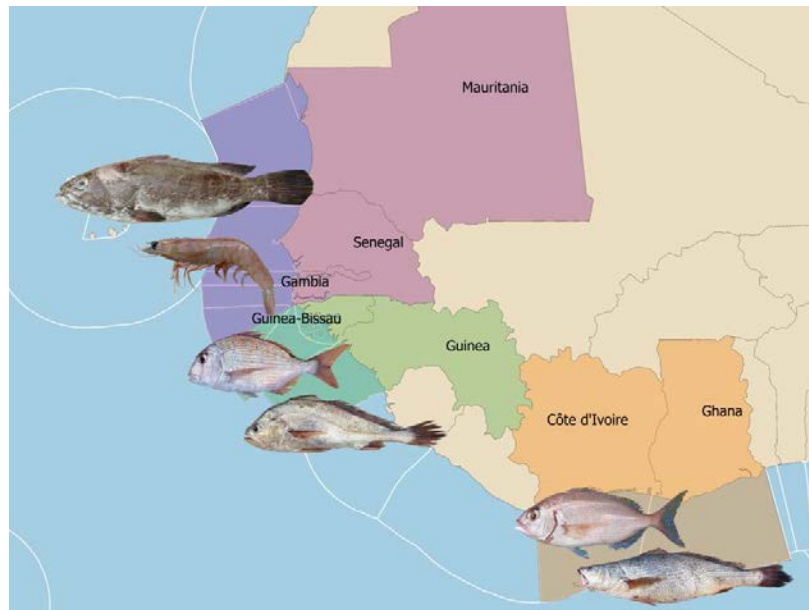


Figure 1.1.- Species studied under the DEMERSTEM project, by case study.

Describing a stock is complex and it is necessary to combine analyses from several biological disciplines, such as genetics, morphometry and life history traits, among others (Abaunza et al., 2008 b). The use of multiple techniques for fish stock identification seems to be adequate for those species with complex stock identities (Begg and Waldman, 1999). In principle, it is expected that applying all the techniques on the same specimen would facilitate the interpretation of the results and comparison of the performance of the different techniques (Waldman et al., 1997), and would partly reduce the uncertainty associated with observational studies (Abaunza et al., 2008).

Given the clear links among life history traits, individual fitness and increasing population rates (Swain et al., 2005), the understanding of the life history traits is a fundamental first step to identify stocks, before applying more specific stock identification analyses (Pawson and Jennings, 1996; Begg et al., 1999; Begg, 2005). In general, the stock should be identified in terms of the spawning area and the spawning behaviour (Gulland, 1971). When analysing life history traits it is important to keep in mind that larger body size benefits fish in migratory processes in terms of energy and swimming capacity (Dodson, 1997). Size also has a close relationship with reproductive parameters, such as length at first maturity and fecundity. Therefore, to compare the production parameters of different areas we have to take into account the possible migratory behaviours, as well as the spatio-temporal stability in the parameters studied (Begg et al., 1999).

The body morphometrics was based on the truss network measurements, i.e. a series of measurements between landmarks that form a regular pattern of contiguous quadrilaterals or cells across the body form (Strauss and Bookstein, 1982; Winans, 1984). The truss network imposes no restrictions on the direction of variation or localization of shape changes (Rawat et al., 2017). The truss network system is highly effective in capturing information about the shape of an organism (Cavalcanti et al., 1999), increasing the likelihood of extracting morphometric differences between species or stocks being used for their differentiation (Cadrin and Friedland, 1999; Cadrin, 2000). This contributes to a better knowledge of the population structure of the species, enabling better management of its fisheries (Erguden et al., 2009).

Together with body shape, otolith shape analysis has become an effective tool for stock identification. Otolith shapes were shown to be species-specific (e.g. L'Abée-Lund, 1988; Stransky and MacLellan, 2005), and in many cases, geographic variation in otolith shapes could be related to stock differences (e.g. Campana and Casselman, 1993; Begg and Brown, 2000; Stransky, 2005). In fish, hard body parts

like scales, otoliths, and bones tend to grow at the same rate to somatic growth. By analysing the shape of otoliths from different fish populations, researchers can differentiate between groups of fish that have had different growth experiences (Brophy, 2013).

The application of stock identification methods can reveal inconsistencies between the spatial structure of biological populations and the definition of stock units used in assessment and management. The aim of the genetic analyses is to identify the geographical boundaries of the stocks using microsatellite markers. Genetic methods have been long recognized for their usefulness in defining stock structure of fish (Ward, 2000). Microsatellites are currently widely used to assess genetic structure primarily due to the very high levels of genetic variation that are often detected at individual microsatellite loci, the large number of loci that can be screened, and their relative ease of analysis, particularly with automated sequencers (Wirgin and Waldman, 2005).

Knowing the identity of stocks is a key issue for assessment and management. The stock definitions of the species concerned in our studies are usually based on geographical boundaries of individual or neighbouring countries. However, stock definitions should evolve as management requirements do, and should be revisited when the need arises or when breakthrough technologies become available (Begg and Waldman, 1999). The objective of this study is therefore, to provide a biological basis to define the stock structure of the six species selected in four cases of studies in order to be used for improving their assessments to provide the scientific basis for better management from the six coastal states involved.

2 MATERIAL AND METHODS

Identifying the stocks of the selected species consists of defining the populations and validating or not its stock unit (species + distribution area) from a biological point of view.

The identity of the stocks of the species selected in the different case studies was analysed following a holistic approach, using a series of complementary techniques:

1. Life history traits (mainly weight related parameters; reproduction parameters and features).
2. Morphometry. Two main techniques were used:
 - Morphometric measurements of the individuals (body shape)
 - Otolith shape, for fish species.
3. Genetic analysis, for *E. aeneus* and *P. notialis*.

The same samples were used for all the above techniques, in order to facilitate the interpretation of the results.

2.1 SAMPLING SCHEME

The selected species were sampled for biology in their distribution areas of the countries of each case of study. Samplings were carried out:

- a) During research surveys (Table 2.1.1).
- b) Samples from the commercial fleet, obtained from landings or markets, by buying the samples in several landings sites, previously defined.

Samplings during research surveys have the advantage that can be exactly geo-referenced and the disadvantage that they are limited in time to the period/s when the survey were carried out and only in certain countries (see Table 2.1.1). Oppositely, samplings from the commercial fleet have the advantage that they could be regularly carry out, with some exceptions, especially relate due to the COVID pandemic, but the disadvantage that the exact fishing area is not known and only approximate fishing areas could be inferred depending on the landing site.

Table 2.1.1- Number of individuals of target species sampled during surveys carried out in 2019 -2020, by country and area.

Country	Area	Survey	Month	Year	GPW	SOP	BSC	PSE	PSS	PAR
Mauritania	North	NANSEN	September	2019	31					
			October	2019	19					
		AWAM	September	2019		59				
			October	2019		42				
	South	NANSEN	September	2019	8					
			November	2019	1					
Guinea- Bissau	North	Bissau 1219	November	2019		362	1		42	111
			December	2019						9
	South	Bissau 1219	November	2019		42	36			56
			December	2019						20
		NANSEN	September	2019			14			
			October	2019						
Guinea	North	GLC	September	2020			40	45		23
			October	2020			137			26
		NANSEN	September	2019			150			
	South	GLC	September	2020			151			26
			October	2020			120			76
		NANSEN	September	2019			163			

For biological and length monthly samplings of the selected species specimens, main sampling areas were defined by country, on the basis of previous knowledge on their distribution area and biology provided by the experts of each country. Figure 2.1.1 shows the sampling stations from surveys and sampling ports for each target species for the selected cases of study.

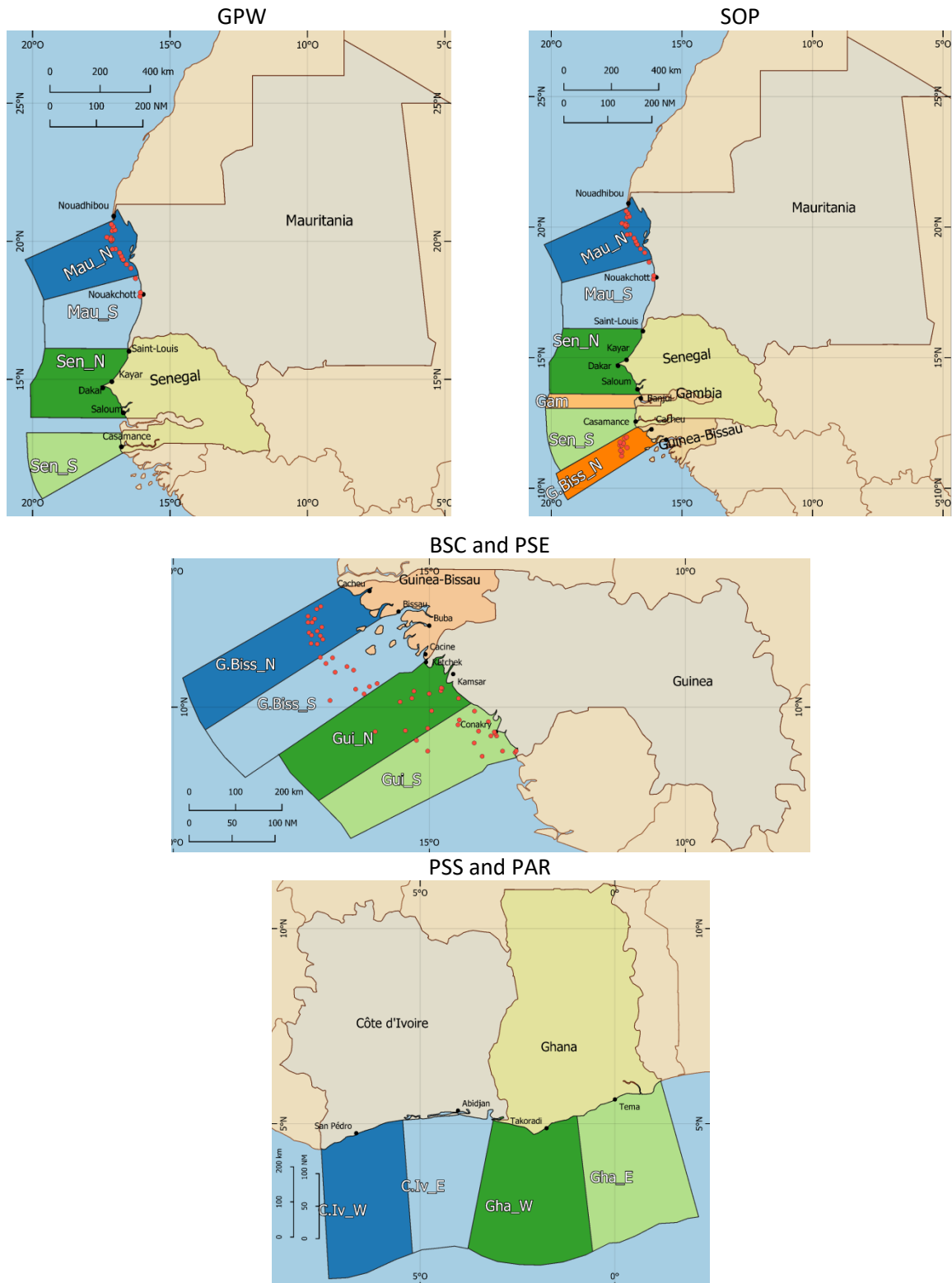


Figure 2.1.1. - Sampling ports and surveys sampling stations by country and area (North- South or West-East) for *E. aeneus* GPW in Mauritania and Senegal (top right), *P. notialis* in Mauritania, Senegal, Gambia and Guinea-Bissau (top left), *P. caeruleostictus* and *P. elongatus* in Guinea-Bissau and Guinea (middle) and *P. senegalensis* and *P. bellottii* (bottom).

A minimum number of individuals per area were established to be monthly sampled for life history traits and bi-annually for genetics and morphometry (pictures for body shape studies) (see sampling Protocol in García Isarch et al. (2020)). In addition, otoliths were extracted from all sampled fish individuals for studies of otolith shape.

Tables in Annex 1 shows the different typed of sampling carried out (biological sampling, pictures for morphometry and genetics) by month and sampling area for each target species. It should be noted that many gaps were detected for certain species and countries. Some of them could be attributed to COVID related issues, as many lacking samples in March-April 2020. Other gaps were due to institutional administrative issues, as it was the case of Côte d'Ivoire, which started sampling six months after the planned starting month, or Guinea-Bissau, with no sampling of *P. elongatus* during the period March-June 2020. There were planned samplings in certain areas, where the species in question are not usually landed, as it is the case of *P. caeruleostictus* in Guinea-Bissau North. The lack of sampling in Mauritania- South, especially for *P. notialis* and for other species in certain months/areas and are for reasons unknown. In addition, the original plan was that samplings from the same species were synchronized between different areas, which were somehow difficult for the same reasons above. Because of that, for morphometry and genetics samplings, extended periods were considered instead the initially planned sampling months.

2.2 LIFE HISTORY TRAITS

The number of individuals sampled for life history traits, by month, sex and country/area are summarized in tables of Annex 2, for each target species. For each sampled individual, parameters registered were:

- Total length (fish) TL (cm), length class of 1 cm
- Cephalothorax length (shrimps) CL (mm), length class of 1 mm.
- Total weight: Wt (g);
- Gutted or eviscerated weight Wg (g)
- Gonad weight (Wgo)
- Sex: male (1), female (2) or indeterminate (3)
- Maturity:
 - scale 1-5, for fish.
 - scale 1-4 (female) and 1-2 (male), for shrimps.

See the protocols established in García-Isarch et al. (2020).

The number of sampled individuals by area and their length range was sometimes variable from one country/area to another. Thus, similar length ranges were tried to be used for parameter comparisons between country/areas. The final number of specimens and length ranges studied by country/area and by sex is shown in different tables of weight- related parameters and reproduction parameters-features, by species.

Weight- related parameters

The estimated weigh-related parameters were:

- 1) **Weight-length relationships**, by sex and for combined sexes. They were calculated for the gutted weight (for the five fish species) and for the total weight (for *P. notialis*). The regression function used was:

$$W = a(L)^b$$

where:

- W = gutted weight [W_g (g)] for fish or total weight [W_t (g)] for shrimps
- L = TL (cm) for fish or CL (mm) for shrimps
- a , b = parameters of the regression

- 2) **Conversion factor** (for fish), by sex and for combined sexes. It is the relationship between the total weight (W_t) and the gutted weight (W_g), related by a linear function with values “0” to intercept with the x-axis:

$$W_t = aW_g$$

where:

- a = parameter of the regression

- 3) **Condition factor**: – Le Cren’s (1951) relative condition factor (K):

$$K = W/aL^b$$

where:

- K = Le Cren’s condition factor
- W = is W_g (gutted weight, g) for fish or W_t (total weight, g) for shrimps
- L = TL (cm) for fish or CL (mm) for shrimps
- a , b = parameters of the regression

K was estimated for mature females, considering matures those females with length (L) higher than the L_{50} . Median values were estimated by quarter as no enough data were available for obtaining representative monthly females estimates of K in most species.

Reproduction parameters and features

- 1) **Sex-ratio**. The sex ratio was estimated and analyzed for all the sampling periods and by length.
- 2) The **spawning period** was determined by analyzing the seasonal changes of the monthly information of the following parameters over one-year period:
 - **Macroscopic maturity stage** of females. The percentages by month of potentially mature specimens were calculated by grouping the maturity stages:
 - o III, IV and V, relative to the total, in fish
 - o III, IV relative to the total, in females shrimps.

- **Gonadosomatic index** (GSI) of mature individuals:

$$GSI = (W_{go}/W) \times 100$$

where:

- W = is W_g (gutted weight, g) for fish or W_t (total weight, g) for shrimps.

Only individuals above the length at first maturity (L_{50}) determined in previous studies were used.

There were cases with no consistency between the spawning periods determined by higher proportion of mature females by month and those identified by using greater GSI values by month. In those cases, the period indicated by the GSI was predominantly considered to determine the spawning season, as the macroscopic staging of females can be considered more subjective (since

it depends on the interpretation of the maturity scale by each researcher), while GSI, based on weights, is more objective.

- 3) **Maturity ogives** by length were constructed using only data collected during the spawning periods obtained here. Maturity ogives were estimated with the sizeMat R package (Torrejón-Magallanes, 2020). The morph_mature function in sizeMat uses the logit approach.
- 4) **Length at first maturity.** The size at 50% maturity (L50) was estimated as the length at which a randomly chosen specimen has a 50% chance of being mature. In the regression analysis, X (length) is considered the explanatory variable and the classification of maturity CS (inmatures: 0, matures: 1) is considered the response variable (binomial). The variables are fitted to a logit function with the form:

$$P_{CS} = \frac{1}{1 + e^{-(\hat{\beta}_0 + \hat{\beta}_1 * X)}}$$

where:

- PCS is the probability of an individual of being mature at a determinate X (length),
- β_0 (intercept) and β_1 (slope) are parameters estimated.
- The L50 is calculated as:

$$L_{50} = -\frac{\hat{\beta}_0}{\hat{\beta}_1}$$

Statistical analysis

Regression parameters of Weight-Length allometric growth and condition factor for each zone/sex were estimated applying an ANCOVA model to the log transformed total weight (TW) and total length (TL) for fish or carapace length (CL) for *P. notialis*. Then, a pairwise test was applied to compare the allometric growth coefficient of each country-area and sex using Tukey's range test method with 95% family-wise.

Potential differences between country-areas in other weight related parameters were analysed by pairwise Wilcoxon tests applying the Bonferroni method to adjust p values to multiple comparisons.

L50 estimated for each species were compared by country-area, by analysing potential overlapping of their confidence intervals.

2.3 MORPHOMETRY

2.3.1 Body shape

Biological sampling for morphometry studies based on body shape was conducted biannually, from a pre-selected number of the same individuals sampled for life history traits during the established periods. Pictures of each individual were taken following the sampling protocols for morphometry (García-Isarch et al., 2020). Table 2.3.1 shows the sampling periods and number of individuals analyzed for body shape, by species. Tables in Annex 1 show the months sampled for morphometry for each species and sampled site.

Table 2.3.1.- Sampling periods and number of individuals analyzed for body shape, by species.

Species	Sampling periods			N. individuals
	1st period	2nd period	Year	
<i>E. aeneus</i> (GPW)	February-March	October-December	2019-2020	329
<i>P. notialis</i> (SOP)	February-March	August-October	2020	228
<i>P. caeruleostictus</i> (BSC)	January	July-October	2020	249
<i>P. elongatus</i> (PSE)	January-March	November	2020	242
<i>. senegalensis</i> (PSS)	May	October-November	2020-2021	190
<i>P. bellottii</i> (PAR)	February-May	September-November	2020-2021	212

Image acquisition and measurement of truss distances

Before its biological sampling, each specimen was placed with its left side of the body up on a white flat background and photographed, by using a digital camera.

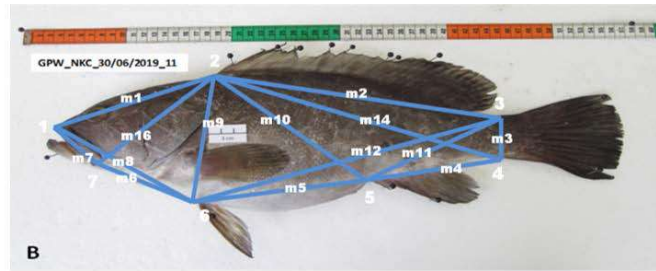
Three groups of species are identified, depending of the number of anatomical landmarks to be used. These landmarks are located along that left side contour and are clearly distinguishable points of the species, corresponding mainly to the fin insertion points (in fish) or to the inter-segment points (in shrimps) and similar to those used in other truss network analyses (Marini et al., 2017; Rawat et al., 2017). After the location of the landmarks in each digital image, the truss network (Strauss and Bookstein, 1982) on the fish body was constructed by interconnecting them to form a total number of truss measurements. The number of anatomical landmarks and measurements by group of species are in Table 2.3.2. Figures 2.3.1, 2.3.2 and 2.3.3 show the locations of the landmarks for constructing the truss network system for each group of species considered.

Table 2.3.2.- Number of landmarks and measurements by group of species.

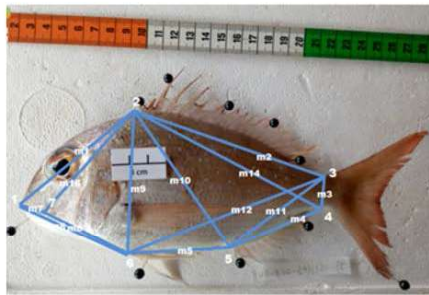
GROUP	SPECIES	N. landmarks	N. measurements
Fish with 1 dorsal fin	<i>E. aeneus</i> <i>P. caeruleostictus</i> <i>P. bellottii</i>	7	14
Fish with 2 dorsal fins	<i>P. elongatus</i> <i>P. senegalensis</i>	8	16
Shrimps	<i>P. notialis</i>	21	47

The fork length (in the sparid species *P. caeruleostictus* and *P. bellottii*) or the total length (in the Pseudotolitus species and in *E. aeneus*) of each specimen was also measured in the images for standardization purposes (Schneider et al., 2012). For *P. notialis*, the longest measurement, the transversal carapace length (m55) of each specimen was also measured in the images for standardization.

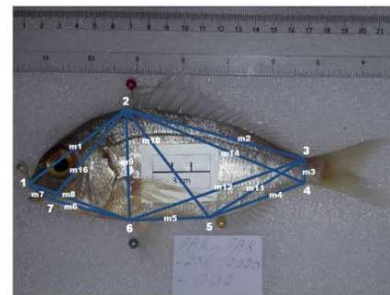
The extraction of the measurements from the digital images of specimens was conducted from Image J (Schneider et al., 2012). Correction of errors, such as those from problems from the use of ruler for performing the measurements, image resolution and quality, was performed.



E. aeneus

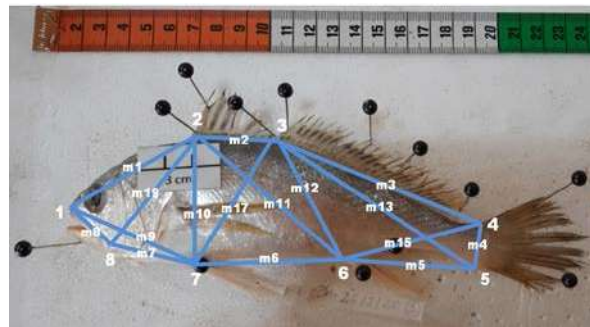


P. caeruleostictus

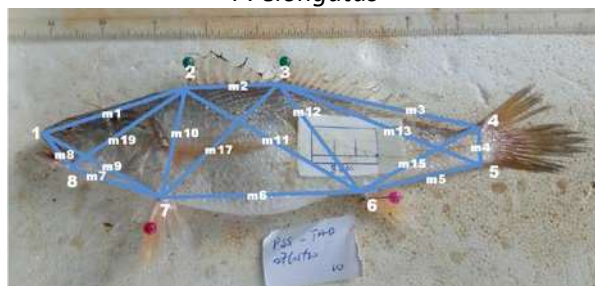


P. bellottii

Figure 2.3.1.- Location of the 7 landmarks for constructing the truss network system of fish species with one dorsal fin. Landmarks refer to (1) anterior tip of snout at upper jaw; (2) origin of dorsal fin; (3) anterior-dorsal point of the caudal fin; (4) anterior-ventral point of the caudal fin; (5) origin of anal fin; (6) origin of ventral fin; (7) most posterior point of maxillary.

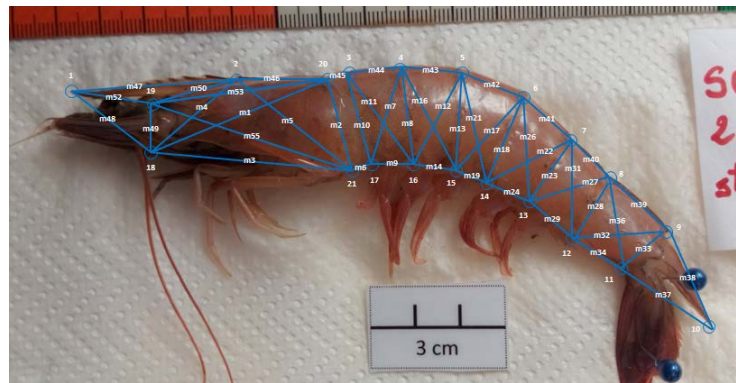


P. elongatus



P. senegalensis

Figure 2.3.2.- Location of the 8 landmarks for constructing the truss network system of fish species with two dorsal fins. Landmarks refer to (1) anterior tip of snout at upper jaw; (2) origin of first dorsal fin; (3) origin of second dorsal fin; (4) anterior-dorsal point of the caudal fin; (5) anterior-ventral point of the caudal fin; (6) origin of anal fin; (7) origin of ventral fin; (8) most posterior point of maxillary.



P. notialis

Figure 2.3.3.- Location of the 21 landmarks for constructing the truss network system of *P. notialis*. Landmarks refer to (1) base of the rostrum; (2) upper teeth the first rostrum; (3) upper of the first segment base; (4) upper of the second segment base; (5) upper of the third segment base; (6) upper of the fourth segment base; (7) upper of the fifth segment base; (8) upper of the sixth segment base; (9) upper of the final segment base; (10) caudal base; (11) end of the sixth segment bottom; (12) end of the fifth segment bottom; (13) end of the fourth segment bottom; (14) end of the third segment bottom, (15) end of the second segment bottom; (16) end of the first segment bottom; (17) base of the first walk leg; (18) base of the antenna; (19) base of carapace; (20) upper end of carapace; (21) end of carapace bottom.

Statistical analysis

All truss measurements were log transformed and the outliers, if any, were removed before further analysis (Jolicoeur, 1963).

Transformation of the absolute measurements into size-independent shape variables was performed following the formula (Reist, 1985):

$$M_{\text{trans}} = \log M - b(\log L - \log L_{\text{mean}})$$

where:

- M_{trans} = transformed truss measurement.
- M = original truss measurement.
- L = length, being:
 - Total Length (TL) for *E. aeneus*, *P. elongatus* and *P. senegalensis*
 - Fork Length (FL) for the sparids *P. caeruleostictus* and *P. bellottii*
 - Transversal cephalothorax length (TCL) for *P. notialis*
- L_{mean} = overall mean length.
- b = within-group slope of the geometric mean regression calculated

Classification and multivariate analysis of shapes (PCA, LDA)

A Principal Components Analysis (PCA) using SPSS Statistics 17.0 was performed using the transformed truss measurements to obtain the principal (factor) components. Further, a few factor components were retained for rotation procedure (varimax rotation) and this was based on the cumulative

proportions of variances and scree plot. The rotation procedure in PCA helps to identify variables that form biologically meaningful groupings with significant contribution to the retained factor components. When the factor loading of a truss variable is 0.4 or greater, that variable is considered to load heavily on that factor (Hatcher, 1994). The applicability of the PCA to the truss measurements studied was analyzed using the Bartlett's sphericity test. PCA model was checked if significant (KMO test) was.

Linear discriminant analysis (LDA) was used to predict and classify each specimen to their respective sampling areas based on their shape differences, i.e. truss variables with high loadings on factor components (Pazhayamadam et al., 2015). It was performed using SPSS Statistics 17.0 through a stepwise selection process where the variables are added to the LDA model when its F value exceeds the entry criteria (by default, 3.84). The classification of individuals by cross-validation of the LDA was carried out and the number and percentage of correctly re-classified specimens (confusion matrices) were obtained including information on actual (in rows) and predicted (in columns) studied areas. Analysis of variance (ANOVA) was applied to the factor scores of samples to determine whether the effects of area are significant.

The scatterplot of factors and box-plots were performed using the Paleontological Statistics (PAST) (Hammer et al., 2001).

2.3.2 Otolith shape

Photographing and extraction of the contour of the otolith

Sagittal otoliths of fish target species were extracted during biological samplings on a monthly basis. A number of otoliths were photographed biannually for shape analysis, when possible choosing them from individuals sampled during the same months than for body shape. Table 2.3.3 shows the number of otoliths photographed and used for the analysis of shape in the selected months, by species, country and area.

Otoliths from selected individuals were photographed with high contrast, by placing them on a black background to favour the reflection of light when the pictures were taken. Only unbroken were chosen for photography.

The outlines of the otoliths were extracted automatically from the images using an *ad hoc* RScript (Denechaud et al., 2020; Smoliński et al., 2020) that incorporates the Momocs package (Bonhomme et al., 2014) within the R environment. Previously, digital images of otoliths in TIFF format had to be converted to JPG format with an image converter.

The otolith contours were converted into a series of (x, y) coordinates that represent the shape. A binary version of the images, the otolith outlines and the extracted coordinates in an R object and an image with the contour were obtained (Figure 2.3.4). The latter were used to detect possible errors from the extraction.

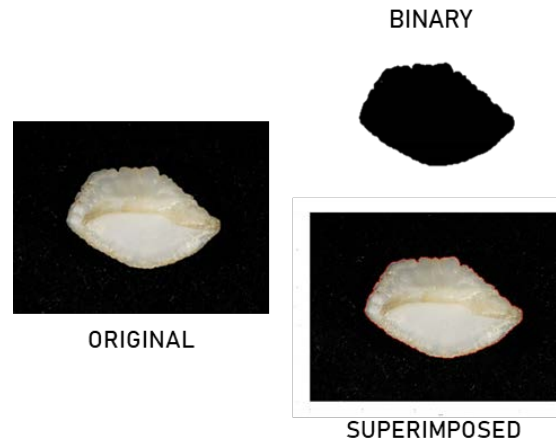


Figure 2.3.4.- Graphical results of otolith contour extraction. Example: *P. bellottii* (PAR).

Table 2.3.3.- Number of otoliths used by species, country and zone photographed and used in the stock identification analysis by otolith shape.

Specie	Country	Zone	n° otoliths photographed	n° otoliths used in shape analysis (left)	Sample months
GPW	Mauritania	Nouadhibou (North)	81	66	March/October
		Nouakchott (South)	74	59	March/October
	Senegal	Kayar (North)	96	39	March/October
		Saloum (South)	78	66	March/October
BSC	Guinea Bissau	Buba (South)	113	123	January/July
	Guinea	Kamsar (North)	110	92	January/October
		Conakry (South)	128	120	January/October
PSE	Guinea Bissau	Cacheu (North)	134	134	January/November
		Cacine (South)	117	117	January/November
	Guinea	Kamsar (North)	140	140	March/November
		Conakry (South)	130	130	March/November
PSS	Côte d'Ivoire	San Pedro (West)	99	67	March/October
		Abidjan (East)	57	39	May/October
	Ghana	Tadkoradi (West)	115	44	May/November
		Tema (East)	67	37	May/November
PAR	Côte d'Ivoire	San Pedro (West)	120	88	March/November
		Abidjan (East)	124	96	May/November
	Ghana	Tadkoradi (West)	64	-	May/November
		Tema (East)	39	32	May/November

Fourier analysis

Once all the otolith contours were obtained, the elliptical Fourier analysis (EFA) was used to measure the otolith outlines. EFA is a technique based on the decomposition of the two-dimensional shape into a sum of mathematical sinusoidal functions called Fourier harmonics. Each harmonic is described by four coefficients (A, B, C, D). It is recommended for multivariate analysis to reduce the number of harmonics to avoid collinearity between shape descriptors. For this, the RScript calibrates the Fourier coefficients by calculating the cumulative Fourier power.

Usually, the first 20 harmonics contain the important information so the number of harmonics that reaches 99% of cumulated Fourier power of a maximum of 20 harmonics were chosen to summarize

the otolith shape. To check the correct choice of the number of harmonics, the script allows to visualise the reconstruction of a randomly selected otolith from the dataset.

After calibration, the otolith outlines are converted to Fourier coefficients. The process considers the scaling of the otoliths to the same size, so to normalize otoliths for size, orientation and starting point, the first three coefficients, A1, B1, C1 were taken as fixed values. This scaling ensures all contours are the same size, preventing analysis based on size. For this purpose, this step allows crossing with biological data and scale the outlines to the same size.

This approach enables the shapes of otoliths to be represented in a mathematical form that can be analysed statistically, in the next step, which can provide insights into patterns of shape variation among different individuals or stocks.

Classification and multivariate analysis of shapes (PCA, LDA)

As for body shape, otolith shape was studied through multivariate analysis included in the RScript used. First, a PCA was carried out to assess the overall variance of otolith shape. Then, a LDA was performed to predict and classify each otolith into their respective sampling areas based on shape differences, specifically using Fourier Descriptors. Complementary to the LDA, the script calculates the importance of the variables (Fourier descriptors) considered in the LDA and the confusion matrix to shows the relationship between the true sampling areas (in rows) and the predicted areas by the model. Finally, by evaluating the accuracy of the model, the percentage of correctly re-classified is obtained.

Additionally, a Covariate Assisted Principal (CAP) was carried out. CAP is a multivariate analysis technique that combines elements of PCA and LDA, and is used to explore and visualize patterns in multivariate data with independent or covariate variables. In this way, it is possible to evaluate whether or not there is a possible allometric effect in the results. In this step, in order to remove the allometric effect, an ANOVA test was used to detect and remove from the dataset the Fourier descriptors having significant interaction with size and its classification in a zone. Therefore, it is necessary to correct this effect and repeat the PCA and LDA analyses.

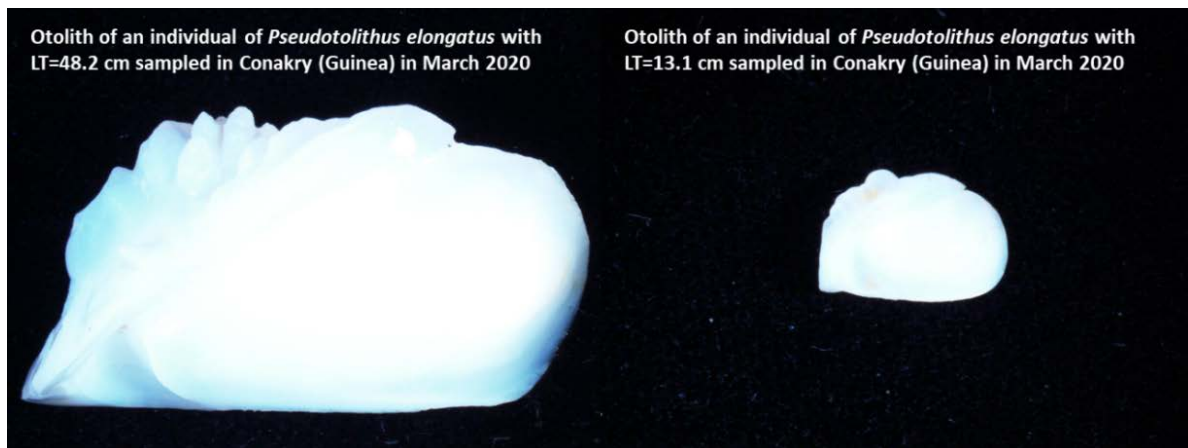


Figure 1.3.5.- Differences in shape of otolith due to size. Example: *P. elongatus* (PSE).

2.4 GENETICS

The genetic structure of the populations was analysed by using microsatellite markers. Identification was done by mitochondrial and nuclear DNA sequencing to obtain a precise identification. The selection of the two species was based on the availability of their barcodes either in the BOLD database or in the Barcode database.

As the genetic information about all target species of the study is scarce, samples obtained from other species were used to obtain new genetic information about them. The molecular identification of the species with samples available was done by sequencing several fragments of nuclear and mitochondrial DNA to get an accurate identification. All the sequences obtained will be uploaded into public databases.

2.4.1 Application of microsatellite markers to decipher geographic boundaries of stocks

The characterization of genetic diversity at microsatellite loci was done to identify the geographical limits of the stocks. Two species were selected for these analyses, both of them with previously developed genetic markers: the “thiof” *E. aeneus* and the southern pink shrimp *P. notialis*.

Selection of microsatellite loci for two species: Epinephelus aeneus and Penaeus notialis

To detect geographical boundaries of *E. aeneus* and *P. notialis* stocks, microsatellite markers were selected from the literature based on described variability. Heterologous microsatellite markers chosen to characterize genetic diversity in *E. aeneus* were obtained from sequences containing a minimum of (CA)₁₀ repeats from different *Epinephelus* species detected in the NCBI database by Dor et al., (2014) and are shown in Table 2.4.1. Microsatellite loci selected to characterize diversity in *P. notialis* are shown in Table 2.4.2.

Table 2.4.1. Genetic tool designed with selected heterologous microsatellite markers for *E. aeneus* (GPW).

PCR	Locus	Forward primer	Reverse primer	Bibliography
M1	ARO1105	TGATAGCTTTACATGCACTCA	CTGAACCTCACCTGAAA	Dor et al., 2014
M1	ARO1045	CACGAAGTATTTGGCTGAT	GAGAAAAGTGGCAATATTTGAC	Dor et al., 2014
M1	ARO1083	CCGGTTCTTCTCTCTCCC	TTACTGTTGATTGAGTTGTTGT	Dor et al., 2014
M1	ARO1084	GGGTTTATTTCAAAGGTCAG	CCCAATGAGGTGTTCAATAT	Dor et al., 2014
M2	ARO1003	GTGCAAGGCAAGCTGTGTTA	AGCAGGCATCTTGTTATCTGG	Dor et al., 2014
M2	ARO1120	CTCTGATGCTGTTTACACAAC	TCTCCATCGAAGGTAAAGG	Dor et al., 2014
M2	ARO1137	ATGGGTATAATTAGGACACACT	AGGAAAAGGAGGGAGGAAA	Dor et al., 2014
M3	ARO1131	TGTGTGTCAGAGGTGGGTT	TGAATTTCACTGCATGTTTC	Dor et al., 2014

Table 2.4.2. Genetic tool designed with selected microsatellite markers for *P. notialis* (SOP)

PCR	Locus	Forward primer	Reverse primer	Reference
M1	PnS03	TGCTAAATAAAAGTTTCTCGGTGAG	AAGCTTGATTTGCGTGTCG	Robainas-Barcia et al. (2002)
M2	PnS04	CGATTTGCAGAACCCGTTTA	GGGGGAGGGGTTAGAAAGAG	Robainas-Barcia et al. (2002)
M3	PnS18	GTCTTATCAAACCCAAAGG	GAACCAAGTCCCGGCCCTCTGC	Robainas-Barcia et al. (2008)
	PnS20	CTTCCATATTCGCATGATGG	ACCCGGGATCAAGCCCTTGC	Robainas-Barcia et al. (2002)
	PnS01	TGCTGTTTGTGAGTCTT	TGGCATGTTGCAGACAGTCC	Robainas-Barcia et al. (2008)

Characterization of genetic diversity

Microsatellite genetic data were examined with GenAEx software v6.5 (Peakall and Smouse, 2006, 2012) to obtain allele frequency and estimate genetic diversity through observed (HO) and expected (HE) heterozygosity. The inbreeding coefficient F_{IS} (Weir and Cockerham, 1984) and compliance to equilibrium of Hardy–Weinberg (HW) were studied with GENEPOP 4.7 (Raymond and Rousset, 1995; Rousset, 2008). Probability values were corrected (Weir, 1996) due to low counts for certain genotypes with the Markov Chain Monte Carlo (MCMC) approximation (involving 10,000 dememorization steps, 1,000 batches and 10,000 iterations per batch).

Deviations from expected proportions in HW equilibrium imply non-random mating, selection for certain genotypes, mutations, or small population sizes. Under HW equilibrium assumptions, the difference between expected and observed heterozygosities can be measured with the inbreeding coefficient F_{IS} (Weir and Cockerham, 1984).

Clustering of genetic diversity

Clustering of genetic diversity was investigated with Discriminant Analysis of Principal Components (DAPC) and Bayesian algorithms to proportionally assign individuals to inferred population clusters. In the DAPC approach we used the R package adegenet (Jombart, 2008) in RStudio with R free software to study differences among clusters identified from genotyped sardines. STRUCTURE v2.3.4 software (Pritchard et al., 2000) was used for the Bayesian approach assuming an ancestry admixture correlated allele frequency model (Falush et al., 2003), and considering prior sampling location information. Running parameters were set to a burn-in of 2.5×10^4 followed by a MCMC simulation of 5×10^4 runs simulating K 1 to N populations (N was 4 for GPW and 6 for SOP) in 25 iterations. STRUCTURE results were explored with Structure Harvester (Earl and von Holdt, 2012) and Clumpak (Kopelman et al., 2015) was used to detect the consensus solutions for the K clusters that best fit the Bayesian algorithm used by STRUCTURE.

2.4.2 Molecular Identification of species

The molecular identification of these species was done by sequencing a fragment of mitochondrial DNA to get an accurate identification.

For the species for which samples were available (*E. aeneus* and *P. notialis*, *P. bellottii* and *P. senegalensis*), a fragment of the Cytochrome Oxidase I (COI) mitochondrial gene was sequenced to both, obtain genetic information and for its the BOLD database of the Barcode of Life. All the sequences obtained will be uploaded into public databases.

COI amplification:

- ***E. aeneus***: Amplification of the COI gene fragment of *E. aeneus* mtDNA was carried out in a 20 μ l reaction mixture with 1X buffer, 1.5mM $MgCl_2$, 0.5 μ l of dNTPs, 15 pmol of Forward and Reverse primer respectively, 1U of Taq DNA polymerase (BioTaq), 1 μ l of sample DNA and the remainder of Milli-Q water. The primers used were FishF2 and FishR2 (Ward et al., 2005).
- ***P. notialis***: In the case of *P. notialis*, the mixture differed in that 2.5mM $MgCl_2$ was used and the primers used were LCO1490 and HC02198 (Folmer et al., 1994).
- ***P. bellottii***: Amplification of the COI gene fragment was carried out in a 20 μ l reaction mixture with 1X buffer, 1.5mM $MgCl_2$, 0.5 μ l of dNTPs, 10pmol of Forward and Reverse primer respectively, 1U of Taq DNA polymerase, 1 μ l of sample DNA and the remainder of Milli-Q water. The primers used were FishF2 and FishR2 (Ward et al., 2005).

PCR amplifications and visualization

PCR amplifications for the three species above were carried out on a SureCycler 8800 thermal cycler (Agilent technologies) under the following conditions: an initial denaturation at 95°C for 5 min, a cycle of 40 denaturation repeats at 95°C 30 s, hybridization at 55 °C 1 min and 72 °C elongation 1 min, followed by a final extension at 72°C for 10 min. The amplified product in both cases was checked on a 2% agarose gel by ethidium bromide staining. Once visualized, it was purified with exonuclease (Exo I) and alkaline phosphatase (FastAP Thermosensitive) and cycled for 15 min at 37°C and 15 min at 85°C. Finally, Sanger sequencing was performed on an ABI 3730 xl sequencer at CACTI (University of Vigo).

The sequences were visualized using the Chromas program (Technelysium), discarding those that presented background noise or were very unreadable. The sequences were examined and assembled with the program BioEdit7 (Hall, 1999) and aligned using the multiplex algorithm CLUSTAL W. The sequences were quality trimmed after alignment. Once the sequences were revised, they were compared with the reference sequences archived in GenBank using the BLAST algorithm (Altschul et al., 1990). The MEGA X program (Tamura et al., 2007) was used to perform the phylogenetic analysis. In the case of *E. aeneus* and *P. bellottii*, distance, mean diversity and diversity between populations were calculated using the p-distance model with a bootstrap value of 1000 replicates. For *P. notialis*, the MEGA X program (Tamura et al., 2007) was used to perform the genetic analysis.

For *E. aeneus* and *P. notialis*, trees were obtained by Maximum Likelihood (ML), Neighbor-Joining (NJ) and Maximum Parsimony (MP) methods with a bootstrap value of 1000 replicates applying the Hasegawa-Kishino-Yano (HKY) model in the case of ML and p-distance in the case of NJ and MP. In both cases the T92 model was applied. For *P. bellottii*, phylogenetic inference was carried out by Maximum Likelihood (ML) method with a bootstrap value of 1000 replicates.

For *P. senegalensis*, none of the amplification conditions tested nor the primer pairs produced an amplicon amenable to sequencing. Visualization on agarose gel gave the image of an asymmetric PCR, which led us to suspect genome rearrangement. To identify the problem, it was decided to sequence the entire mitochondrial genome. The procedure is detailed below.

- 1) Samples and DNA isolation. Two samples of *P. senegalensis*, both from Tema (Ghana), (PSS_TAD19/10/19_23 and PSS_TAD19/10/19_42), both from Tema (Ghana), were selected for mitochondrial genome sequencing. Genomic DNA isolation from tissue samples was carried out using the Quick-DNA 96 Plus kit (Zymo Research) according manufacturer's instruction. A negative extraction control was included. DNA was quantified using the Qubit High Sensitivity dsDNA Assay (Thermo Fisher Scientific).
- 2) Library preparation and sequencing. The gDNA libraries were prepared following Carøe & Bohman (2020) to avoid false assignment of sequences, with minor modifications. The libraries were purified using the Mag-Bind RXNPure magnetic beads (Omega Biotek), following manufacturer's instructions. Libraries sequencing was performed in a fraction of NovaSeqPE150 run (Illumina), with a total output of 5 Gb per library. Genomic sequencing was carried out by AllGenetics & Biology SL (www.allgenetics.eu).
- 3) Raw data pre-processing, de novo assembly and annotation. Sequencing raw data were first quality-checked using the software FastQC v0.11.5 (Babraham Bioinformatics, 2010). Trimmomatic 0.39 (Bolger et al., 2014) was used to remove the adapters and low-quality regions, and regions with shorter lengths were also discarded. After this quality trimming, another quality-check was carried out (FastQC), to check that only high-quality reads were selected (Supplementary material 1).
- 4) Mitochondrial genomes *de novo* assembling and annotation was performed with the Geneious Prime 2022 software (www.geneious.com), using *Pseudotolithus typus* (NC_056258) and *Pseudolithus elongatus* (NC_044717) as reference mitochondrial genomes. The MITOchondrial

genome annotation Server (MITOS2) (Bernt et al., 2013) was used to automatically annotate the mitochondrial genomes selecting RefSeq 63 Metazoa as reference database and the vertebrate mitochondrial genetic code. The protein prediction method from Al Arab et al. (2017) was enabled.

- 5) Phylogenetic analysis. Phylogenetic analysis using a total of 28 mitogenomes of *Scienidae* family were performed based on genetic distance model Tamura-Nei and Neighbor-Joining methods, with *Conger erebennus* as outgroup. This mitogenomes were downloaded from the Mitochondrial Genome Database of Fish (MitoFish 3.75; <http://mitofish.aori.u-tokyo.ac.jp>).

3. RESULTS

Results from the different studies and species from this project are detailed in different manuscripts being produced within the framework of this project (García-Isarch et al., a, b, in prep.; Landa et al., a, b, in prep.; Pérez et al., a, b, in prep.).

3.1. LIFE HISTORY TRAITS

3.1.1. *Epinephelus aeneus* (GPW)

A total of 2080 individual of *E. aeneus* were sampled for obtaining life history traits during the period September 2019 - January 2021. Table 2.1 in Annex 2 shows the total number of individuals analysed in the biological sampling, by sex, length range (cm of TL) and country/area for *E. aeneus*. Not enough information of males was available to estimate representative life history traits for this sex, so it was not possible to make comparisons of males between areas. Figure 3.1.1 shows the number of individuals sampled by month and country/area, for females and combined sexes.

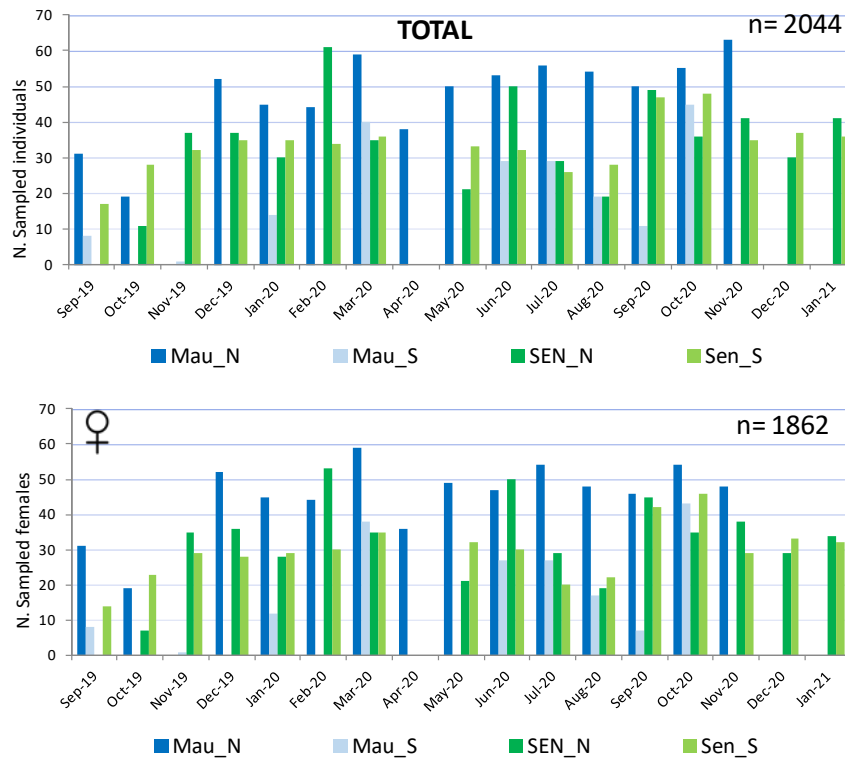


Figure 3.1.1.- Number of *E. aeneus* (GPW) sampled by month and country/area, for combined sexes (top) and for females (bottom).

The number of individuals sampled by quarter and country-area are shown in Table 3.1.1. *E. aeneus* in Mauritania-South was clearly undersampled in relation to the other areas.

Table 3.1.1.- Number of individuals of *E. aeneus* (GPW), sampled by quarter in Mauritania and Senegal, by area (North and South).

QUARTER	Mau_N	Mau_S	Sen_N	Sen_S
Quarter 1	156	54	178	167
Quarter 2	145	30	71	80
Quarter 3	199	69	97	133
Quarter 4	197	49	208	247
Total	697	202	554	627

Minimum, maximum, mean and median values of length are summarized and plotted by zone in Annex 3.

Weight-related parameters

Higher lengths of *E. aeneus* were sampled in Mauritania North (median value of 61.5 cm TL) compared to the other three areas (media values ranging from 45.8-49.7 cm TL) (Annex 3). To make possible any comparison of weight related parameters, a common length range between the four country-areas of 28-80 cm TL was selected.

Weight-length relationships

The results of the gutted weight (Wg)–total length (Lt) relationship, their coefficients of determination, sample size and length and weight ranges for each country-area are shown by sex and for combined sexes in Table 3.1.2 and plotted for combined sexes in Figure 3.1.2.

Table 3.1.2.- Parameters of the gutted weight-length relationships for *E. aeneus* (GPW) estimated in Mauritania and Senegal, by area (North and South).

MALES				
<i>E. aeneus</i> (GPW)	Mau-N	Mau-S	Sen-N	Sen-S
a	–	–	0.006	0.005
b	–	–	3.17	3.23
n	5	6	32	41
R ²	–	–	0.99	0.99
Length range	57.0-79.5	41.3-67	28.2-58.5	28.4-57.3
Gutted weighth range	1960-6350	885-4400	246-2585	217-2233
Weigth range	2140-6950	1035-5100	262-2846	223-2374
FEMALES				
<i>E. aeneus</i> (GPW)	Mau-N	Mau-S	Sen-N	Sen-S
a	0.005	0.004	0.007	0.008
b	3.21	3.27	3.13	3.09
n	632	177	492	472
R ²	0.99	0.99	0.99	0.99
Length range	28.4-80.0	28.2-77.0	28.2-78.8	28.0-80.0
Gutted weighth range	260-8150	190-6600	243-6638	166-7050
Weigth range	269-8950	200-7300	256-7958	194-7900
SEX COMBINED				
<i>E. aeneus</i> (GPW)	Mau-N	Mau-S	Sen-N	Sen-S
a	0.005	0.005	0.007	0.008
b	3.21	3.26	3.13	3.10
n	669	193	525	537
R ²	0.99	0.99	0.99	0.99
Length range	28.4-80.0	28.2-77.0	28.2-78.8	28.0-80.0
Gutted weighth range	260-8150	190-6600	243-6638	166-7050
Weigth range	269-8950	200-7300	256-7958	194-7900

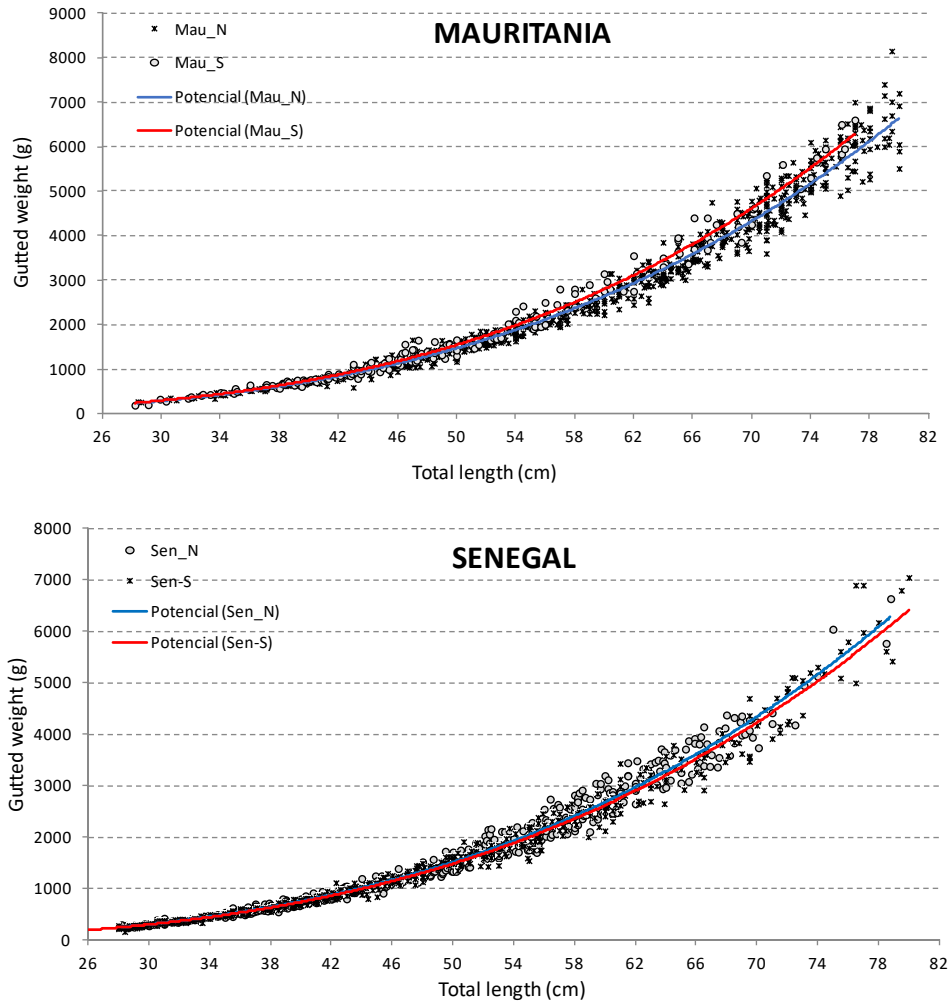


Figure 3.1.2.- Gutted weight–length relationships estimated for *E. aeneus* (GPW) in Mauritania (top) and Senegal (bottom), by country-area and combined sexes.

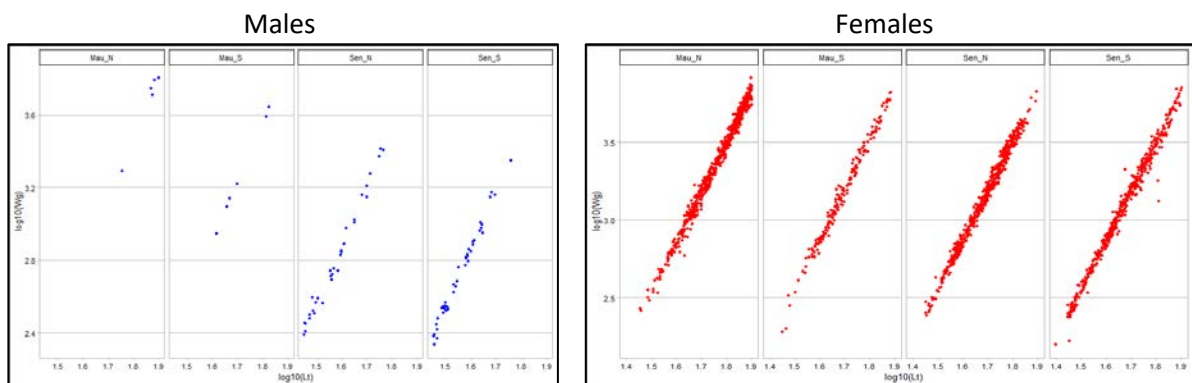


Figure 3.1.3.- Gutted weight–length relationships (log values) estimated for *E. aeneus* (GPW) in Mauritania and Senegal, by area (North and South) and by sex.

The regression lines indicate a close relationship and similar parameters for different country-areas, with slight differences between slopes between Mauritania and Senegal (3.2 and 3.1, respectively for sex combined). However, significant differences ($p < 0.05$) in the slopes between both countries in the weight–length regressions were found by pairwise comparisons using Tukey’s range test, while no

significant differences were found within the north and south area within each country (Section 4.1.1. in Annex 4). The same results were obtained for females in both countries. It should be reminded that the number of males was considered insufficient for the estimation of weight related parameters and therefore, to make any potential comparison between country-areas.

Weight conversion factor

The conversion factors, sample sizes for total weight (Wt)–gutted weight (Wg) linear relationships in each country-area (sex combined) are shown in Table 3.1.3.

Table 3.1.3.- Parameters of the total weight (Wt)–gutted weight (Wg) linear relationships for *E. aeneus* (GPW) estimated in Senegal and Mauritania, by area (North and South).

<i>E. aeneus</i> (GPW)	Mau-N	Mau-S	Sen-N	Sen-S
b	0.98	0.98	0.98	0.99
N	669	196	526	539
R ²	0.997	0.998	0.996	0.997

Similar Wt–Wg conversion factors were obtained in the analyzed country-areas, as the slope ranged between 0.98–0.99, and no significant differences between the four country-areas were found when applying the Tukey’s range test (Section 4.1.2. in Annex 4).

Condition factor

The Le Cren’s condition factor (K) was estimated for mature females, by quarter and country-area (Figure 3.1.4). The seasonal trend of the median values of K was similar among the country-areas, showing no significant differences in most areas and quarters, with the greatest values during the second quarter (with the exception of Senegal North) and the lowest in the third quarter. Significant differences were only found between both areas of Senegal (in the first quarter), and between Mauritania South and the other three areas (in the third quarter) (section 4.1.3 in Annex 4).

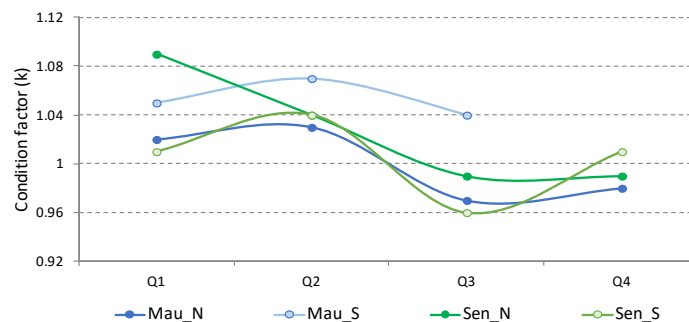


Figure 3.1.4.- Quarterly variation of the median values of the Le Cren’s condition factor (K) in mature females of *E. aeneus* (GPW) in Mauritania and Senegal, by area (North and South).

Reproduction parameters and features

Sex-ratio

Sex-ratio (male:female) of *E. aeneus* ranged between 1:11 (92% of females) in Senegal-South to 1:110 (99% of females) in Mauritania- North (Table 3.1.4). The proportion of females was higher than 89% in all studied country-areas, decreasing gradually from North to South.

Table 3.1.4.- Sex ratio values estimated for *E. aeneus* (GPW) in Mauritania and Senegal, by area (North and South).

Sex-ratio	Mau_N	Mau_S	Sen_N	Sen_S
% Females	99%	97%	92%	89%
M:F	1:110	1:31	1:11	1:8

The sex ratio was analyzed by length. The percentage of females was higher than 80% for most length classes, and lower than this proportion for individuals < 30 cm, where they were present (in Senegal) (Figure 3.1.5). While in Mauritania the proportion of females was close to 100% for fish length from 35 cm onwards, both areas of Senegal show a progressive increase in the percentage of females from the 25 cm range up to 45 cm (where they are close to 100%).

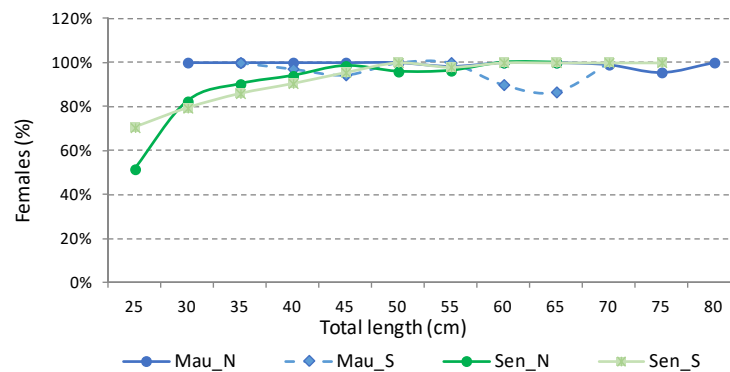


Figure 3.1.5.- Female percentage of for *E. aeneus* (GPW) by length class (5 cm) in Mauritania and Senegal, by area (North and South).

Spawning period

The monthly percentage of mature and immature females of *E. aeneus* and the monthly mean of their GSI is shown in Figure 3.1.6. This GSI was estimated only for females higher than 49 cm, which is the size at first maturity estimated for the thiof in Senegalese waters (Ndiaye et al., 2013). No mature males were found during the study period in any of the country-areas.

Some inconsistencies were found in Senegal between the spawning period showed by the highest percentages of mature females and that indicated by the greater values of the GSI. In these cases, the spawning period indicated by the GSI was considered.

Although there were some months with no data, results suggested that the thiof can reproduce in Senegal all year around, while in Mauritania the reproduction seems to be more concentrated in a time period (Figure 3.1.6). In both countries and areas one main spawning peak could be determined, occurring in July-August in Mauritania (both North and South) and being more extended in Senegal (May-August in the North and May-September in the South).

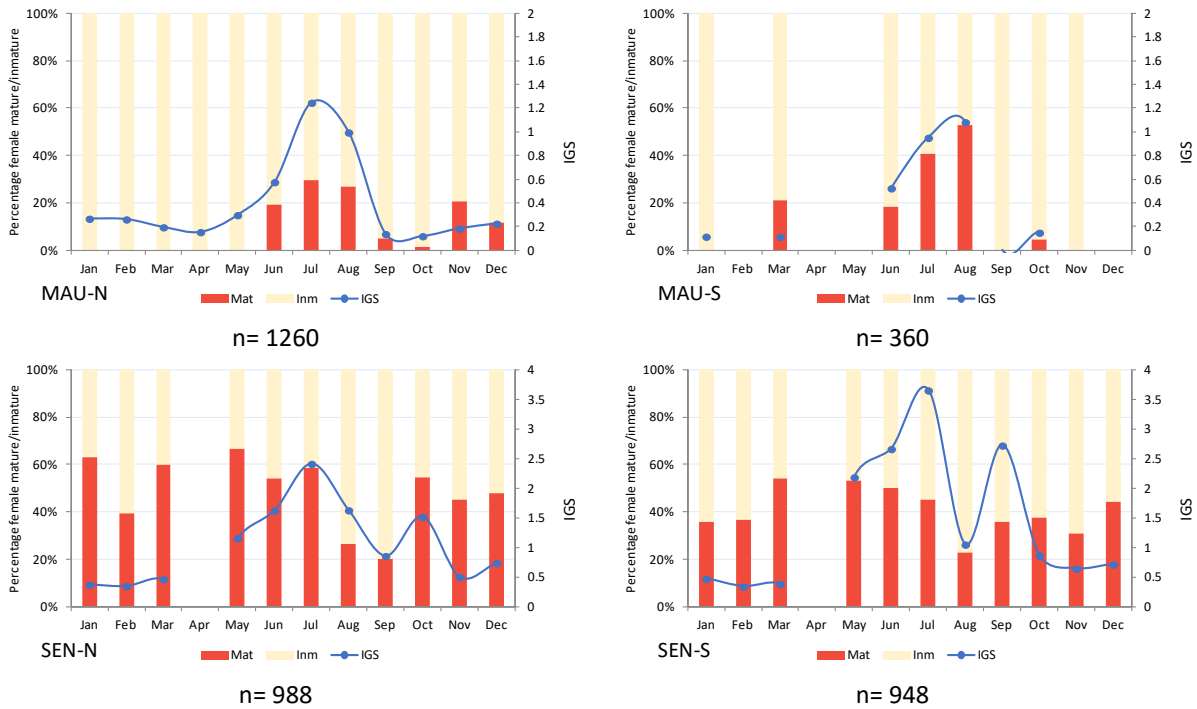


Figure 3.1.6.- Monthly percentage of mature/immature females and mean GSI of *E. aeneus* (GPW) in Mauritania (top) and Senegal (bottom) by area (North and South).

Maturity ogives

The maturity ogives of females in Senegal (North and South) are shown in Figure 3.1.7. Not enough information was available to estimate a representative maturity ogive of *E. aeneus* for any of the Mauritanian areas. Apart from the low number of females considered for the estimation, especially in Mauritania South, bigger thiof sizes were predominately sampled in that country, with median values of females sampled around 62 cm TL. This median value is much higher than the L₅₀ obtained in Senegal and those estimated from previous studies of *E. aeneus* (Table 3.1.5).

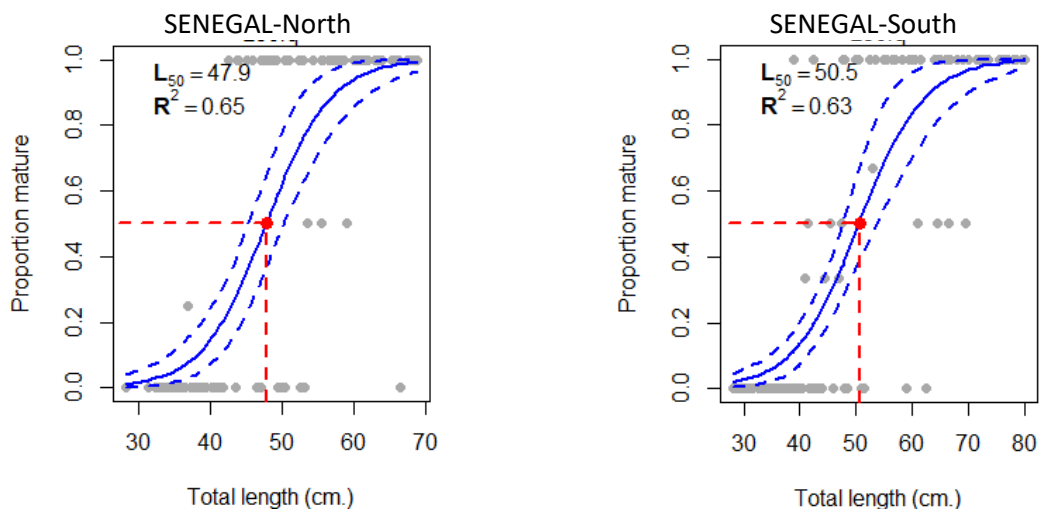


Figure 3.1.7.- Maturity ogives of females of *E. aeneus* (GPW) in Senegal (North and South).

Length at first maturity

The lengths at first maturity (L50), their confidence intervals and the number of females used for the estimations in their correspondent spawning peaks are shown in Table 3.1.5. The L50 for females in Senegal South was slightly higher than in Senegal North, but the confidence intervals of the L50 in both areas overlapped and therefore, they were not considered significantly different.

Table 3.1.5.- Spawning periods and peaks, L50s and their confidence intervals, number of individuals and their length range (LT in cm) of females of *E. aeneus* (GPW) in Mauritania and Senegal by area (North and South).

FEMALES GPW	Mau_N	Mau_S	Sen_N	Sen_S
Spawning period	Jun-Dec	March-Oct	All year	All year
Spawning peak	Jul-Aug	Jul-Aug	May-Aug	May-Sep
L50 (Confidence intervals), cm	—	—	47.9 (45.4 - 50.3)	50.5 (47.7 - 53.7)
N	102	44	119	148
Length range (median), cm	43.0 – 80.0 (61.6)	40.1 – 76.3 (61.7)	28.2 – 68.9 (49.5)	28 – 80 (46.0)

3. 1. 2. *Penaeus notialis* (SOP)

A total of 6759 individuals of *P. notialis* were sampled for estimating life history traits during the period September 2019 - March 2021. Table 2.2 in Annex 2 shows the total number of individuals analysed in the biological sampling, by sex, length range (mm of CL) and country/area for *P. notialis*. Unfortunately, not all country-areas were sampled all months, as established in the sampling protocol and the sampled period was not synchronized in all countries, as planned. This makes more difficult the comparisons among the studied areas (Figure 3.1.8). Table 3.1.6 shows the number of individuals sampled by quarter and country-area. Mauritania-South is clearly undersampled with no sampling during the first semester of the year. Indeed, according to IMROP samplers, landings of this species in Nouakchott (in southern Mauritania) can come from several fishing areas, mainly including Senegal and Guinea-Bissau, unlike landings in Nouadhibou (in northern Mauritania) where shrimps are known to come from the northern zone. This fact has hampered sampling in the Mauritania-South. This, samples correctly allocated to Mauritania South were only obtained with from a scientific observation mission onboard a shrimp trawler in August 2020 (Meissa, *pers. comm.*). Gambia lacks sampling during the second quarter.

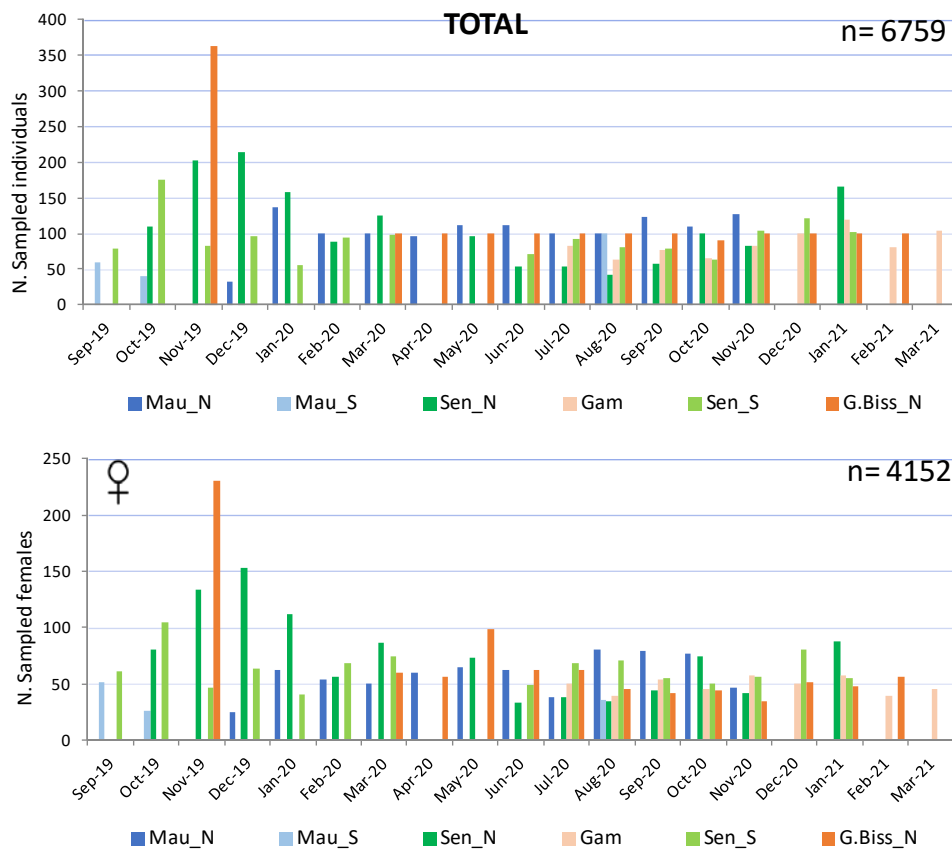


Figure 3.1.8.- Number of *P. notialis* (SOP) sampled by month and country/area, for combined sexes (top) and for females (bottom).

Table 3.1.6.- Number of individuals of *P. notialis* (SOP), sampled by quarter in Mauritania and Senegal, by area (North and South).

QUARTER	Mau_N	Mau_S	Sen_N	Gam	Sen_S	G.Biss_N
Quarter 1	337	–	538	303	348	300
Quarter 2	320	–	151	–	71	300
Quarter 3	323	159	153	222	332	300
Quarter 4	269	41	708	248	641	653
Total	1249	200	1550	773	1392	1553

Minimum, maximum, mean and median values of length are summarized and plotted by zone in Table 3.2.1. and Figure 3.2. 1. of Annex 3.

Weight-related parameters

Differences between the lengths of *P. notialis* sampled in different areas were found, being especially significant between Guinea-Bissau (median value of 19.5 mm CL for sex combined) and the other areas (median value ranging from 28.5 to 31.7 mm CL, without considering Mauritania South due to sampling limitations) (Annex 3). Thus, to make possible the comparison of weight related parameters, a common length range between the different country-areas was selected: 16-50 mm CL both for females and the total sampled population, and 16-34 mm CL for males (Table 3.1.7).

Weight–length relationships

Table 3.1.7.- Parameters of total weight-cephalothorax length relationships estimated for *P. notialis* (SOP) in Mauritania, Senegal-Gambia and Guinea Bissau, by country-area.

MALES						
<i>P. notialis</i> (SOP)	Mau-N	Mau-S	Sen-N	Gam	Sen-S	G. Biss
a	0.001	0.007	0.002	0.001	0.001	0.003
b	2.86	2.31	2.75	2.93	2.78	2.62
n	506	64	468	326	423	455
R ²	0.94	0.82	0.93	0.94	0.93	0.90
Length range (mm LC)	17.6-34	20.9-31.3	16.0-33.9	16.0-34.0	16.1-33.9	16-33.2
Weigth range (g)	5.3-30.4	7.6-17.7	3.1-28.7	3.2-29.9	3.2-28.06	2.9-21.7
FEMALES						
<i>P. notialis</i> (SOP)	Mau-N	Mau-S	Sen-N	Gam	Sen-S	G. Biss
a	0.002	0.003	0.002	0.001	0.002	0.002
b	2.66	2.62	2.73	2.79	2.73	2.72
n	738	36	1032	436	903	534
R ²	0.97	0.95	0.98	0.97	0.98	0.93
Length range (mm LC)	21-50.0	21.2-37.2	16.2-50.0	16.0-45.0	16.4-50.0	16.0-38.2
Weigth range (g)	7.3-87.0	7.8-35.0	2.2-74.8	2.9-61.0	2.6-93.6	2.2-36.0
SEX COMBINED						
<i>P. notialis</i> (SOP)	Mau-N	Mau-S	Sen-N	Gam	Sen-S	G. Biss
a	0.003	0.004	0.002	0.001	0.002	0.002
b	2.64	2.47	2.71	2.81	2.73	2.68
n	1269	100	1521	765	1339	989
R ²	0.97	0.93	0.98	0.96	0.98	0.92
Length range (mm LC)	17.6-50.0	20.9-37.2	16.0-50.0	16.0-45.0	16.1-50.0	16.0-38.2
Weigth range (g)	5.3-87.0	7.6-35.0	2.2-74.8	2.7-61.0	2.6-93.6	2.2-36.0

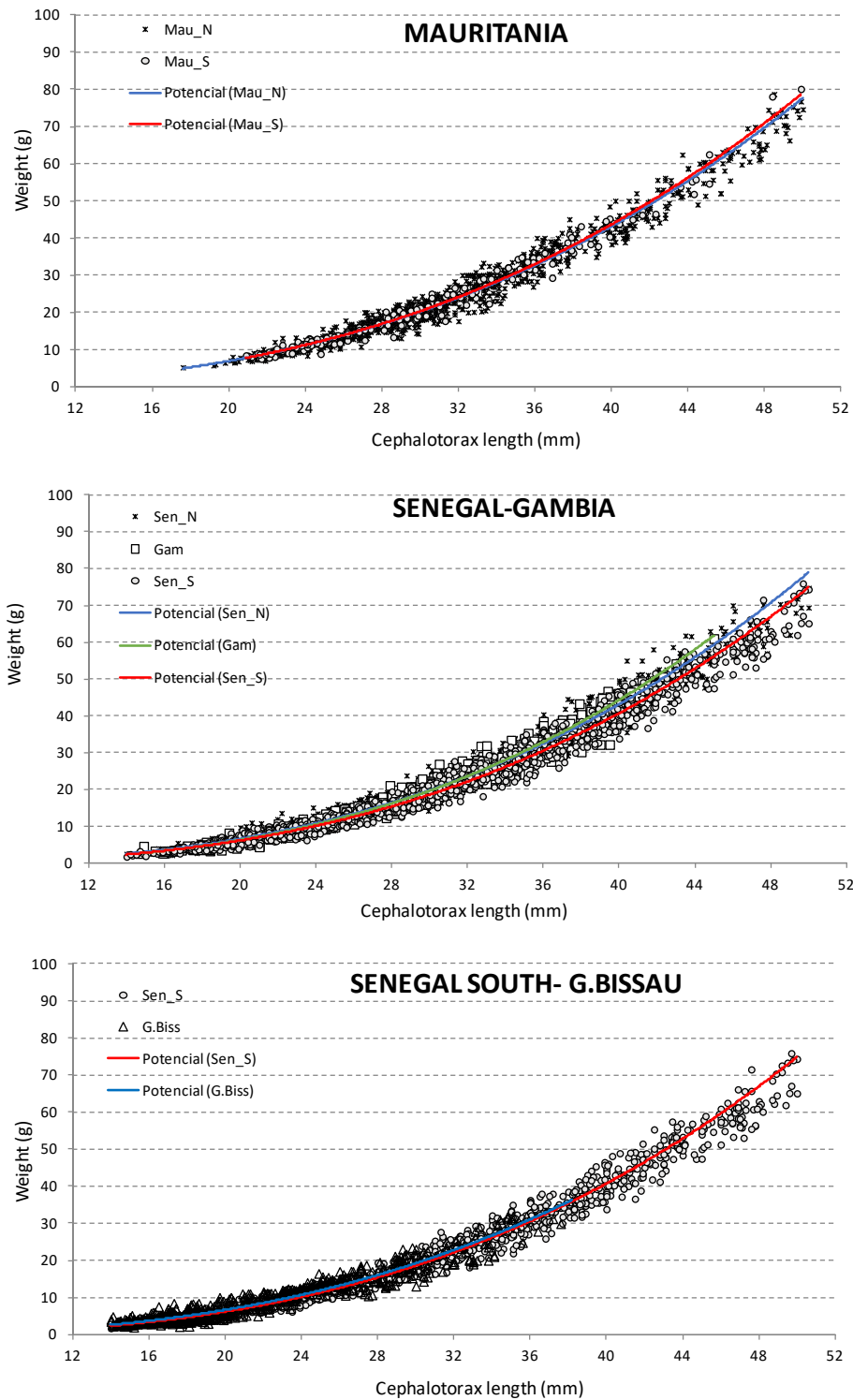


Figure 3.1.9.- Total weight–cephalothorax length relationships estimated for *P. notialis* (SOP) in Mauritania (top), Senegal-Gambia (middle) and Senegal South- Guinea Bissau (bottom), by country-area and combined sexes.

The results of the total weight (W) – total length (CL_t) relationship, their coefficients of determination, sample size and length and weight ranges for each country-area are shown by sex and for sex combined in Table 3.1.7 and plotted for sex combined in Figure 3.1.9. It should be noted the small number of individuals of Mauritania South and the smaller individuals sampled in Guinea-Bissau.

The regression lines indicate a close relationship for most country-areas (Figure 3.1.10). The values of the slopes vary between 2.5 and 2.8 for sex combined. The smallest values were always estimated in Mauritania-South, probably related to the low number of sampled individuals, while highest values were observed in Gambia. The slope values for males ranged from 2.6 (in Guinea-Bissau) and 2.9 (Mauritania-North and Gambia), and for females from 2.7 (in North Mauritania, Senegal, and Guinea-Bissau) to 2.8 (in Gambia), not considering Mauritania South, due to the low number of sampled individuals. The pairwise comparison analysis carried out by the Tukey's range test showed significant differences between Mauritania North compared with Senegal (North and South) and with Gambia. When separated analysis are conducted by sex, significant differences were found between Guinea-Bissau and Mauritania North, Gambia and Senegal South and between Senegal North and Gambia (for males) and between Mauritania North and Gambia (for females) (Section 4.2.1. in Annex 4).

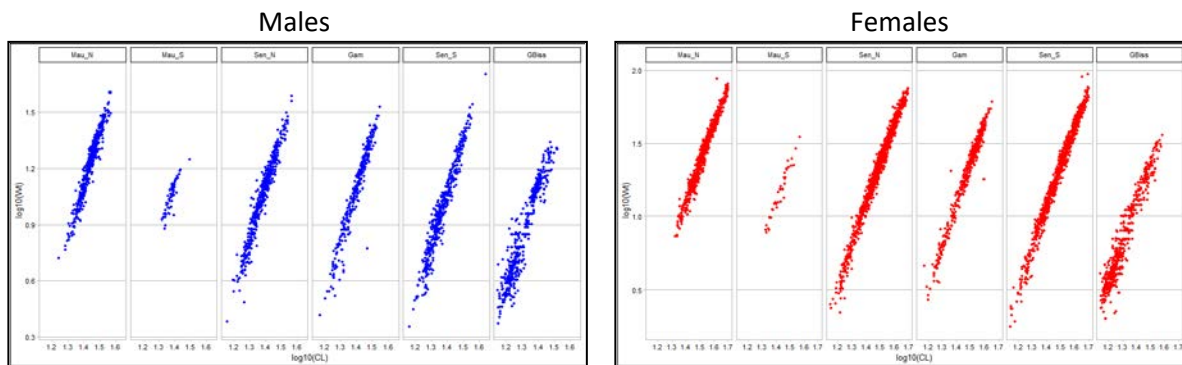


Figure 3.1.10.- Total weight– cephalothorax length relationships (log values) of *P. notialis* (SOP) in Mauritania, Senegal, Gambia and Guinea-Bissau, by sex and by area.

Condition factor

The Le Cren's condition factor (K) was estimated for mature females of *P. notialis*, by quarter and country-area (Figure 3.1.11). The trend of the median values of K was quite similar in most areas with available information, with the exception of Guinea-Bissau. In Mauritania North, the highest and lowest mean K values were found in the first and third quarter, respectively, while in both areas of Senegal they occurred in the second and fourth quarters. When comparing the K values of the different areas by quarter, significant differences are found: a) between Senegal-South and Mauritania-North, Senegal-North and Gambia in the first quarter), b) between Guinea-Bissau and the other sampled areas in the second quarter, c) between Senegal South and Gambia, Guinea-Bissau in the third quarter, and d) between Senegal-South and the northern areas of both Mauritania and Senegal in the fourth quarter (section 4.2.2. in Annex 4).

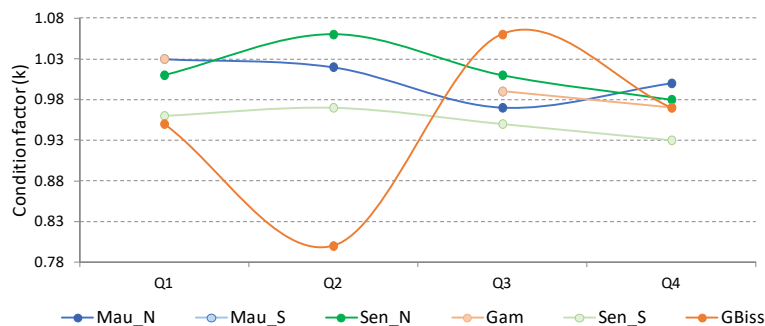


Figure 3.1.11.- Quarterly variation of the median values of the Le Cren's condition factor (K) in mature females of *P. notialis* (SOP) estimated in Mauritania, Senegal, Gambia and Guinea-Bissau, by area.

Reproduction parameters and features

Sex-ratio

Without considering Mauritania-South, as it comes from one single sampling month, sex-ratio (male:female) ranged between 1:1.3-1.1.4 (57-58% of females) in Mauritania- North, Gambia and Guinea- Bissau and 1-2.2 (68% of females) in Senegal, both North and South (Table 3.1.8).

The sex ratio was analyzed by length (Figure 3.1.12). In general, males dominate at the smaller length classes (< 22 mm CL), with the exception of the smallest sizes (< 14 mm CL), only registered in Guinea-Bissau, that were sexed as females. Oppositely, females dominate at bigger sizes (> 30 mm in all country-areas). The sex-ratio between 22-30 mm varied between different areas. Most individuals larger than 38 mm were females in all country-areas. This is in accordance with the known sexual size dimorphism between sexes for the species, with bigger females than males.

Table 3.1.8.- Sex ratio values estimated for *P. notialis* (SOP) in Mauritania, Senegal, Gambia and Guinea-Bissau, by area.

Sex-ratio	Mau_N	Mau_S	Sen_N	Gam	Sen_S	G. Biss
% Females	58%	36%	68%	57%	68%	58%
M:F	1:1.4	1:0.6	1:2.2	1:1.3	1:2.2	1:1.4

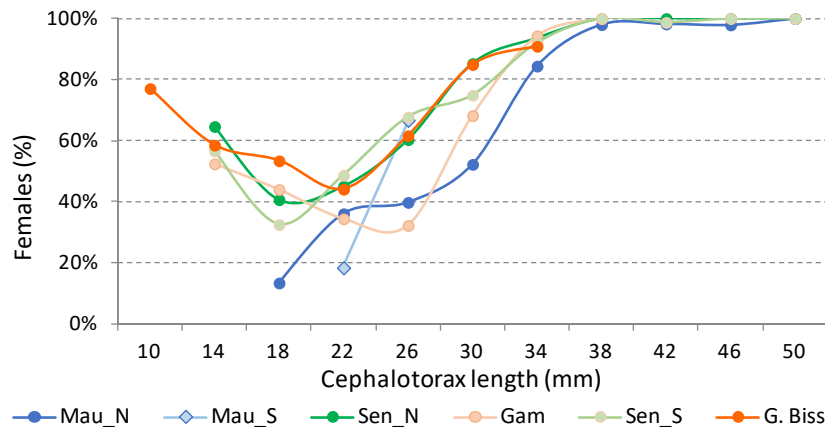


Figure 3.1.12.- Female percentage of *P. notialis* (SOP) by cephalothorax length class (4 mm) in Mauritania, Senegal, Gambia and Guinea-Bissau, by area.

Spawning period

The monthly percentage of mature and immature females of *P. notialis* and the monthly mean of their GSI, when available, are shown in Figure 3.1.13. This GSI was estimated only for females bigger than 25 mm CL, which is the size at first maturity estimated for this species in Senegalese waters (L’homme L. & Garcia, S., 1984).

Some inconsistencies were found in Senegal between the spawning period showed by the highest percentages of mature females and the one indicated by the greater values of the GSI. In these cases, the spawning period indicated by the GSI was considered.

Not enough information was available to identify the spawning period of the species in Mauritania South and in Guinea-Bissau. In the first case, only one month was sampled. In Guinea-Bissau, no mature females were registered during landing samplings and mature individuals were only recorded during a survey in November 2019.

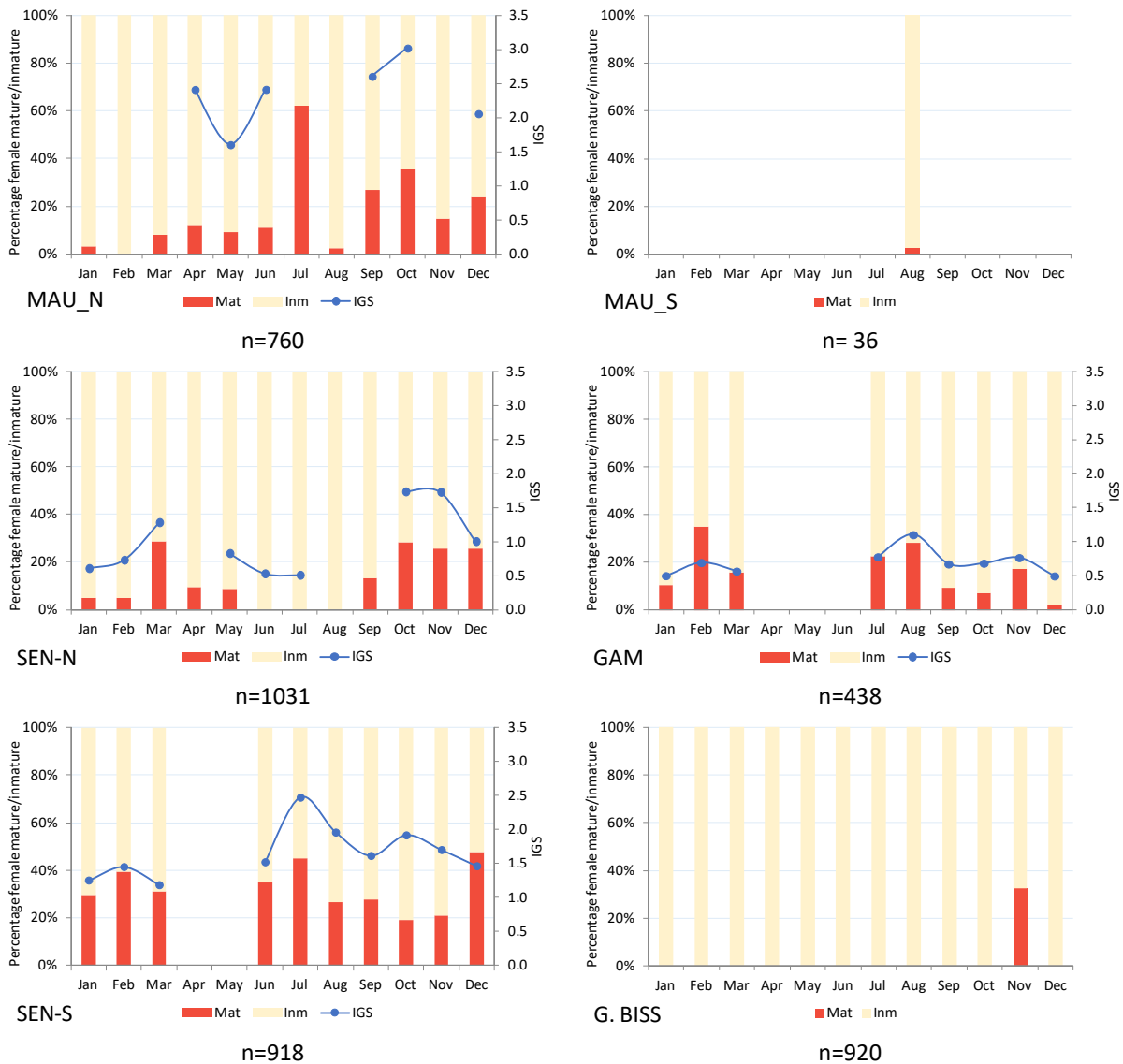


Figure 3.1.13.- Monthly percentage of mature/immature females and mean GSI of *P. notialis* (SOP) in Mauritania, North and South (top), Senegal North and Gambia (middle) and Senegal South-Guinea Bissau North (bottom).

The information available suggests that *P. notialis* can reproduce all year around, at least in Mauritania and Senegal-South. Mature females usually occur throughout the year in Senegal-North although they were not found in our study during June-August because usually during that period, fishermen catching *P. notialis* concentrate their activity into the estuaries, where only juveniles are found (Thiaw, *pers.comm.*), fact that was confirmed by the length frequency distribution during those months. Two main spawning peaks can be detected in the studied areas where most completed information is available. In general, a main and more extended spawning peak occurs during the warm season, being variable in duration (2-3 months, between June and November, depending on the area). A secondary peak occurs during the cold season, in February or March (in Gambia and Senegal North) and from December to August (in Senegal South). Only in Mauritania North, the two spawning peaks occur at the onset and at the end of the warm season (July and October) (see Table 3.1.9).

Maturity ogives

The maturity ogives of females by country-areas are shown in Figure 3.1.14. Not enough information was available to estimate a representative maturity ogive for the species, neither in Mauritania South nor in Guinea-Bissau.

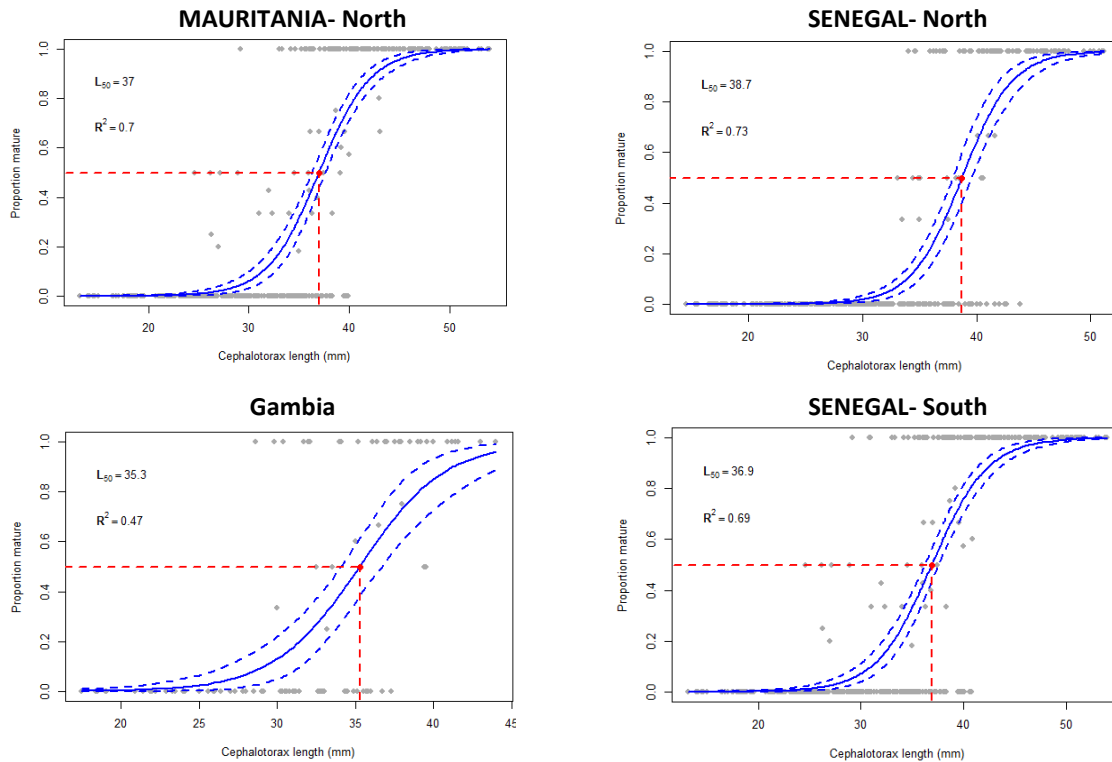


Figure 3.1.14.- Maturity ogives of females of *P. notialis* (SOP) in Mauritania-North and Senegal-North (top), and Gambia and Senegal-South (bottom).

Length at first maturity

The lengths at first maturity (L_{50}), their confidence intervals and the number of females used for the estimations in their correspondent spawning peaks are shown in Table 3.1.9.

Reliable estimations of L_{50} s for females of *P. notialis* were obtained for Mauritania-North, Senegal (North and South) and Gambia, while they are not available for Mauritania-South and Guinea-Bissau, due to the sampling limitations. The L_{50} values ranged between 35.3 mm CL in Gambia and 38.7 mm CL in Senegal-North. There is overlapping of the L_{50} confidence intervals between Mauritania-North, Gambia and Senegal-South but not with any of them and Senegal-North. The values obtained are much higher than those obtained in previous studies in the area (L'homme, 1981; García-Isarch et al., 2021).

Table 3.1.9.- Spawning period and peaks, L50s and their confidence intervals, number of individuals used for their estimation and their length range (CL in mm). Females of *P. notialis* (SOP) in Mauritania, Senegal, Gambia and Guinea-Bissau, by area.

FEMALES SOP	Mau_N	Mau_S	Sen_N	Gam	Sen_S	G.Biss
Spawning period	All year	—	Sept-May	All sampled months	All sampled months	Nov ^{*1}
Spawning peak	– Jul – Sept-Oct	Aug ^{*2}	– Mar – Oct-Nov	– Feb – Jul-Aug	– Dec-Mar – Jun-Aug	Nov ^{*1}
L50 (Confidence intervals), mm CL	37.0 (36.3 - 37.6)	—	38.7 (37.9 - 39.6)	35.3 (34.1 - 36.7)	36.9 (36.2 - 37.5)	—
N	569	36	416	128	569	265
Length range (median) (mm CL)	19.9 - 53.2 (34.6)	21-2-37.2 (27.1)	11.6 - 51 (30.7)	17.5 - 44 (33)	12.5 – 58.9 (34.6)	12.4-37.0 (24.9)

^{*1} Only one month with mature females (survey sampling)

^{*2} The only month with sampling.

3. 1. 3. *Pagrus caeruleostictus* (BSC)

A total of 3727 individuals of blue spotted seabream *P. caeruleostictus* were sampled for obtaining life history traits during the period September 2019 - February 2021. Table 2.3 in Annex 2 shows the total number of individuals analysed in the biological sampling, by sex, length range (cm of TL) and country/area. The number of individuals sampled by month and country/area, for females and combined sexes are shown in Figure 3.1.15. Biological samplings of *P. caeruleostictus* could not be performed in Guinea-Bissau North, as initially planned (García-Isarch et al., 2020). The number of individuals sampled by quarter and country-area are shown in Table 3.1.10. Second and third quarters were undersampled in Guinea-Bissau, in relation to the other studied areas.

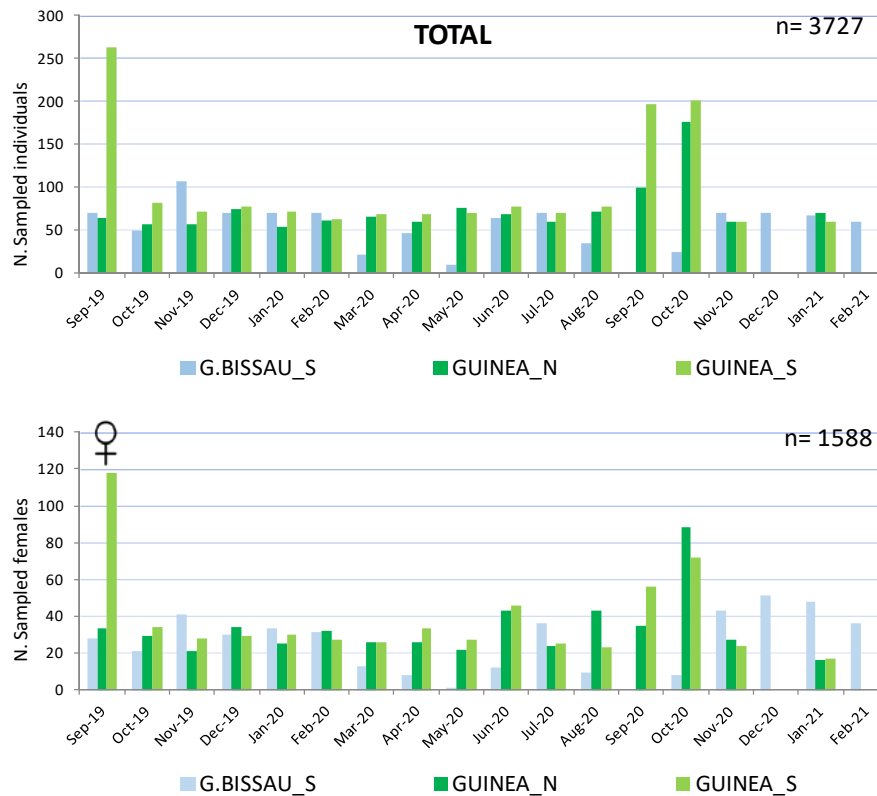


Figure 3.1.15.- Number of *P. caeruleostictus* (BSC) sampled by month and country/area, for combined sexes (top) and for females (bottom).

Table 3.1.10.- Number of individuals *P. caeruleostictus* (BSC), sampled by quarter in Guinea-Bissau and Guinea, by area (North and South).

QUARTER	G.Biss_S	Gui_N	Gui_S
Quarter 1	289	250	262
Quarter 2	120	205	215
Quarter 3	175	295	607
Quarter 4	391	425	493
Total	975	1175	1577

Minimum, maximum, mean and median values of length are summarized and plotted by zone in Table 3.3.1. and Figure 3.3.1. of Annex 3.

Weight-related parameters

Individuals from Guinea-Bissau were clearly smaller than those from Guinea, with median values of 20.5 cm TL and 23.5-24.5 cm TL, respectively (Annex 3). Thus, while a common length range of 10-34 cm was selected to estimate the weight-related parameters in the four country-areas (Table 3.1.11, Figure 3.1.16), a more slightly restricted length range (between 13 and 31 cm TL) was used for comparisons among areas.

Weight–length relationships

The results of the gutted weight (Wg)–total length (Lt) relationship, their coefficients of determination, sample size and length and weight ranges for each country-area are shown by sex and for sex combined in Table 3.1.11 and plotted for sex combined in Figure 3.1.16.

Table 3.1.11.- Parameters of the gutted weight–length relationships for *P. caeruleostictus* (BSC) estimated in Guinea-Bissau and Guinea, by area.

MALES			
<i>P. caeruleostictus</i>	G.Biss_S	GUI_N	GUI_S
a	0.019	0.029	0.028
b	2.91	2.80	2.81
n	443	459	637
R ²	0.98	0.99	0.98
Length range (TL, cm)	11.0-33.7	16.0-34.0	16.0-34.0
Gutted weight range (g)	16-580	75-672	68-637
Weight range (g)	19-588	78-712	72-662
FEMALES			
<i>P. caeruleostictus</i>	G.Biss_S	GUI_N	GUI_S
a	0.020	0.033	0.030
b	2.88	2.76	2.78
n	452	545	481
R ²	0.96	0.98	0.98
Length range (TL, cm)	13.0-32.9	16.6-34.0	13.0-34.0
Gutted weight range (g)	26-452	75-585	53-588
Weight range (g)	33-491	79-623	56-641
SEX COMBINED			
<i>P. caeruleostictus</i>	G.Biss_S	GUI_N	GUI_S
a	0.019	0.029	0.025
b	2.90	2.80	2.84
n	973	1178	1310
R ²	0.97	0.97	0.97
Length range (TL, cm)	11.0-33.7	10.0-34.0	10.2-34.0
Gutted weight range (g)	16-580	18-672	18-637
Weight range (g)	19-588	19-712	20-662

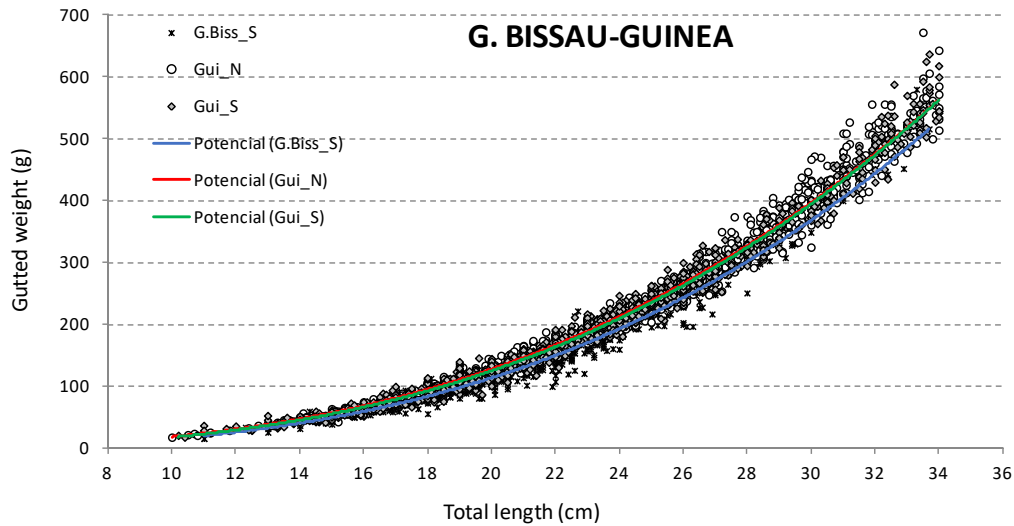


Figure 3.1.16.- Gutted weight–length relationships estimated for *P. caeruleostictus* (BSC) estimated in Guinea-Bissau and Guinea, by area and combined sexes.

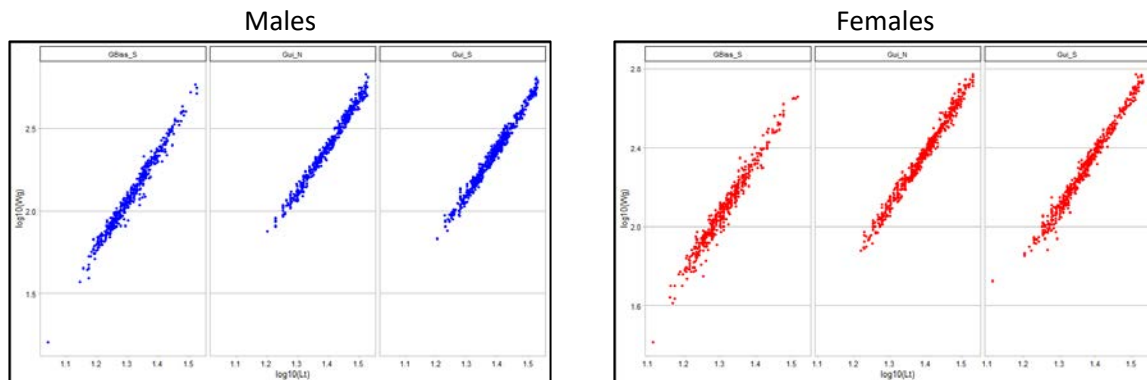


Figure 3.1.17.- Gutted weight–length relationships (log values) estimated for *P. caeruleostictus* (BSC) in Guinea-Bissau and Guinea, by area and by sex.

The regression lines indicate similar weight–length relationship for combined sexes between the two areas in Guinea (North and South), and close to that of Guinea-Bissau South (Figure 3.1.16). This pattern was also found both for males and females, separately, being the slope always higher in Guinea-Bissau (Table 3.1.11). Significant differences ($p < 0.05$) were found between the slopes of the three areas (Section 4.3 in Annex 4). However, when analysing by sex, no significant differences are found between North and South Guinea (for both males and females) (Section 4.3.1 in Annex 4).

Weight conversion factor

The conversion factors, sample sizes for total weight (Wt)–gutted weight (Wg) linear relationships in each country-area (sex combined) are shown in Table 3.1.12.

Similar annual values, ranging between 0.99–1.00, were obtained for the Wt–Wg conversion factors between the three sampled areas. However, significant differences were found by the pairwise comparison between Guinea-Bissau and the two areas in Guinea (Section 4.3.2 in Annex 4).

Table 3.1.12.- Parameters of the total weight (Wt)–gutted weight (Wg) linear relationships for *P. caeruleostictus* (BSC) estimated in Guinea-Bissau and Guinea, by area.

SEX COMBINED			
<i>P. caeruleostictus</i> (BSC)	G.Biss_S	Gui_N	Gui_S
b	1.00	0.99	0.99
N	973	1178	1310
R ²	0.991	0.991	0.991

Condition factor

The Le Cren’s condition factor (K) was estimated for mature females of *P. caeruleostictus*, by quarter and country-area (Figure 3.1.18). The trend of the median values of K was similar in Guinea-Bissau South and Guinea North, with the greatest and lowest values during the third and first quarter, respectively. Guinea South shows the highest K in a previous (second) quarter. Similarities between the two Guinean areas, in the first, second and fourth quarters were found by the pairwise comparison and no similarities were found between these two areas and Guinea-Bissau in any quarter.

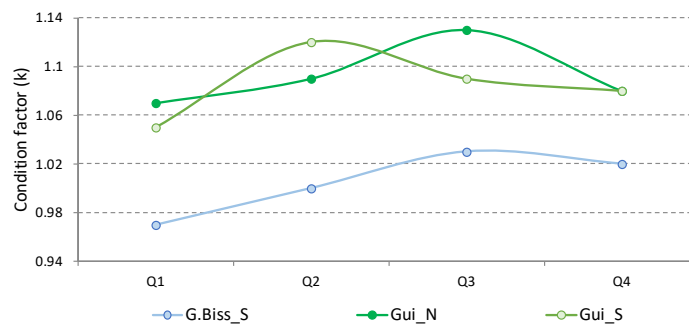


Figure 3.1.18.- Quarterly variation of the median values of the Le Cren’s condition factor (K) in mature females of *P. caeruleostictus* (BSC) estimated in Guinea-Bissau and Guinea, by area.

Reproduction parameters and features

Sex-ratio

Sex-ratio (male:female) of *P. caeruleostictus* ranged between 1:08 (45% of females) in Guinea-South to 1:1.1 (53% of females) in Guinea-North (Table 3.1.13).

The analysis of sex ratio by length shows a similar pattern in large individuals (> 28 cm) in both Guinean areas with a higher proportion of males. In smaller lengths of blue spotted seabreams, a more different pattern between the studied areas was found (Figure 3.1.19), with more abundance of females at length ranges below 28 cm TL in northern Guinea and 24 cm TL in southern Guinea. In Guinea-Bissau, where the length range sampled was much smaller, the female proportion was only higher between 18 and 22 cm TL.

Table 3.1.13.- Sex ratio values estimated for *P. caeruleostictus* (BSC) estimated in Guinea-Bissau and Guinea, by area.

Sex-ratio	G.Biss_S	Gui_N	Gui_S
% Females	50%	53%	45%
M:F	1:1	1:1.1	1:0.8

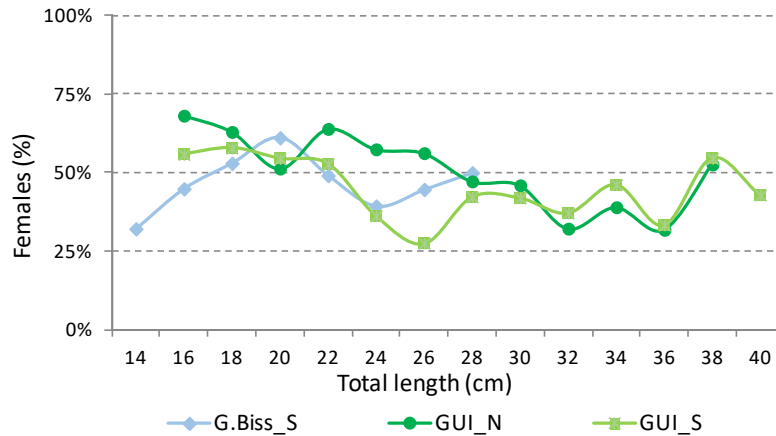


Figure 3.1.19.- Female percentage of *P. caeruleostictus* (BSC) by length class (2 cm) in Guinea-Bissau and Guinea, by area.

Spawning period

The monthly percentage of mature and immature females and the monthly mean of their GSI of *P. caeruleostictus* are shown in Figure 3.1.20. This GSI was estimated only for females higher than 18.7 cm, which is the size at first maturity estimated for females in Guinea-Bissau by Kromer et al. (1994).

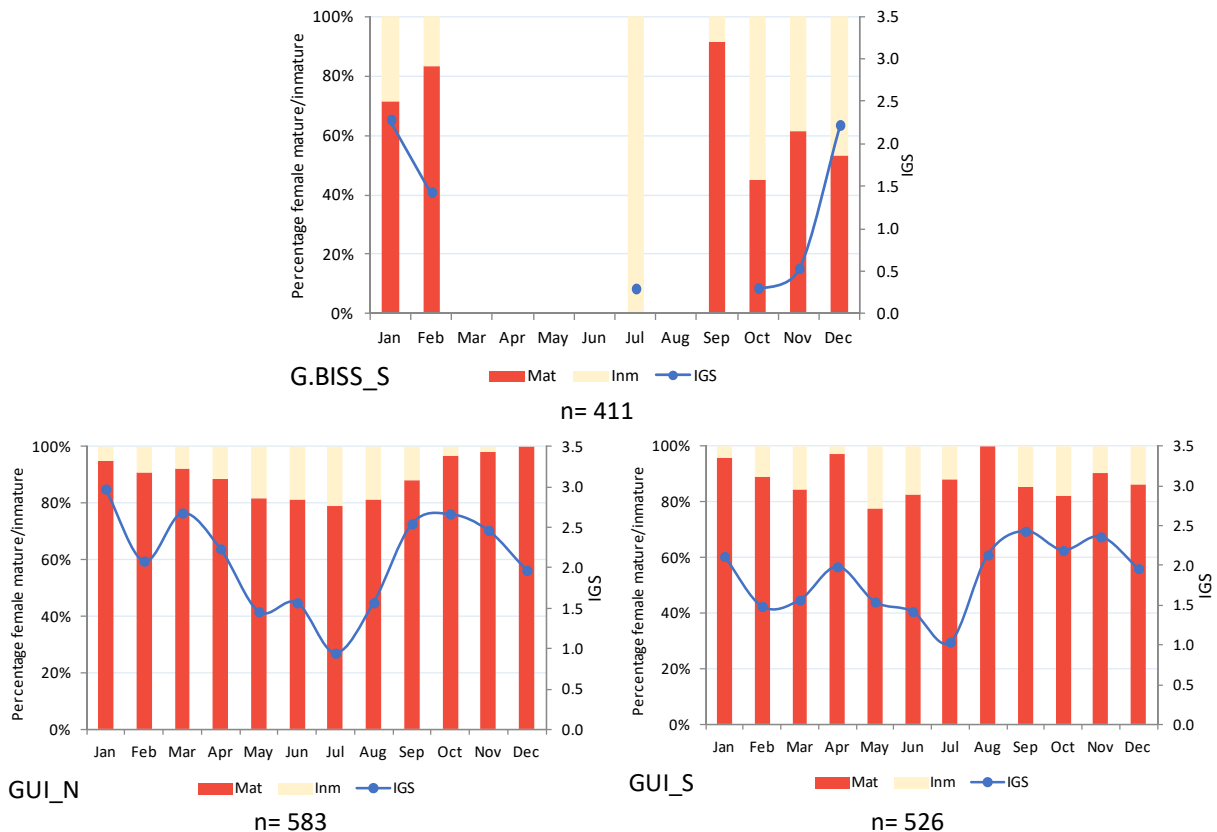


Figure 3.1.20.- Monthly percentage of mature/immature females and mean GSI of for *P. caeruleostictus* (BSC) in Guinea-Bissau (top) and Guinea, by area (bottom).

The information of *P. caeruleostictus* in Guinea-Bissau South is quite limited as only 7 months were sampled. In this country/area, high proportions of mature females were found in most sampled

months, with the exception of July, when all females were immature. Large proportions of mature females (> 70%) were found during all months in both sampled areas of Guinea.

However, the seasonal trends in this species were more marked using female GSI than maturity proportions. The mean GSI and maturity showed an overall similar trend in northern Guinea, with lower values between May and August compared to higher values found between September and April. In southern Guinea, higher GSI was also found in the second half of the year, between August and January. GSI in southern Guinea Bissau showed the highest values between December and at least until February, but did not show consistency with the estimated maturity proportions.

The species spawns throughout the year, with at least one main spawning peak. The main spawning peak occurred from December to February in Guinea-Bissau South and the potential existence of a second peak cannot be exactly determined, due to the lack of sampling during March-June and August. In Guinea, the main spawning peak occurred from August (in the South) or September (in the North) to January. A secondary peak seems to be observed in March (in the North) and one month later in the South (Figure 3.1.20).

Therefore, there seems to be a greater similarity between the spawning periods observed between both areas of Guinea than that in Guinea Bissau South.

Maturity ogives

The maturity ogives of females in Guinea-Bissau South and Guinea South are shown in Figure 3.1.21. The fitting of the maturity at length observations to the model predictions in both areas was quite poor, especially for Guinea-Bissau. Not quality information was available to estimate a representative maturity ogive for the species in Guinea North, as most sampled size classes are mature during the spawning peaks. Without any trend in the proportion-mature estimates it is not impossible to fit a logistic curve, and L50 cannot be estimated (e.g. Taylor et al., 2018).

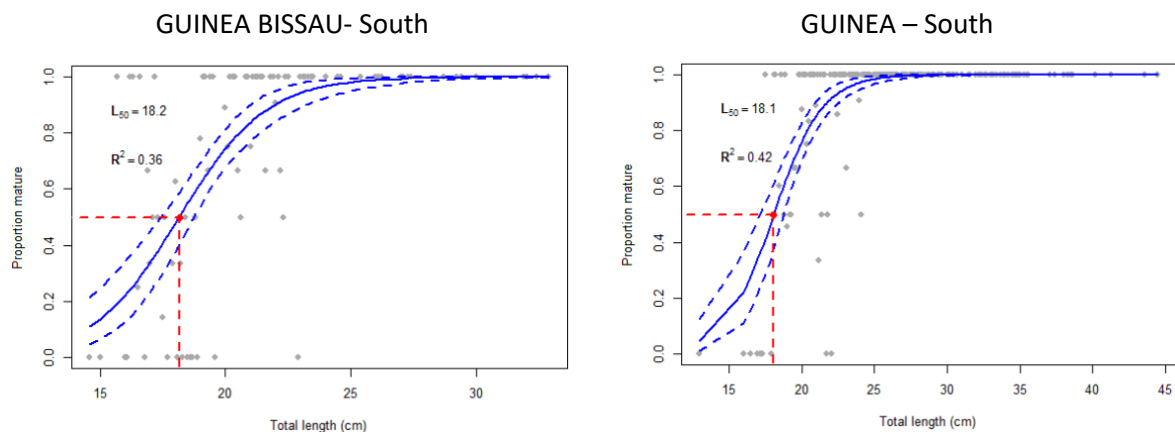


Figure 3.1.21.- Maturity ogives of females of *P. caeruleostictus* (BSC) in Guinea-Bissau South (left) and Guinea South (right).

Length at first maturity

With the data available, lengths at first maturity (L_{50}) of females of *P. caeruleostictus* could be only estimated in the southern areas of Guinea-Bissau and Guinea. L_{50} could not be estimated for females in Guinea North, for aforementioned reasons. The estimates of L_{50} , their confidence intervals and the number of females used for the estimations in their correspondent spawning peaks are shown in Table 3.1.14.

Similar values of 18.1-18.2 were estimated in Guinea South and Guinea-Bissau South.

Table 3.1.14.- Spawning periods and peaks, L50s and their confidence intervals, number of individuals and their length range (LT in cm) of females of *P. caeruleostictus* (BSC) in Guinea-Bissau (South) and Guinea (North and South).

FEMALES BSC	G.Biss_S	Gui_N	Gui_S
Spawning period	6 of 7 sampled months	All year	All year
Spawning peak	Dec-Feb....	- Sept-Jan - March	- Ago-Jan - April
L50 (Confidence intervals), cm	18.2 (17.4 - 18.8)	–	18.1 (17.2 - 18.8)
N	228	353	375
Length range (median), cm	14.6 - 32.9 (20)	16.9 - 50.4 (23.6)	13- 44.5 (23.9)

3. 1. 4. *Pseudotolithus elongatus* (PSE)

A total of 4384 individuals of bobo croaker *P. elongatus* were sampled for life history traits during the period September 2019- February 2021. The last three months were additionally sampled by Guinea-Bissau. However, no samplings were carried out in Guinea-Bissau North between March-August 2020. The total number of individuals analysed in the biological sampling by sex, length range (cm of TL) and country/area are shown in Table 2.4 in Annex 2. The number of individuals sampled by month and country/area, for females and combined sexes is shown in Figure 3.1.22.

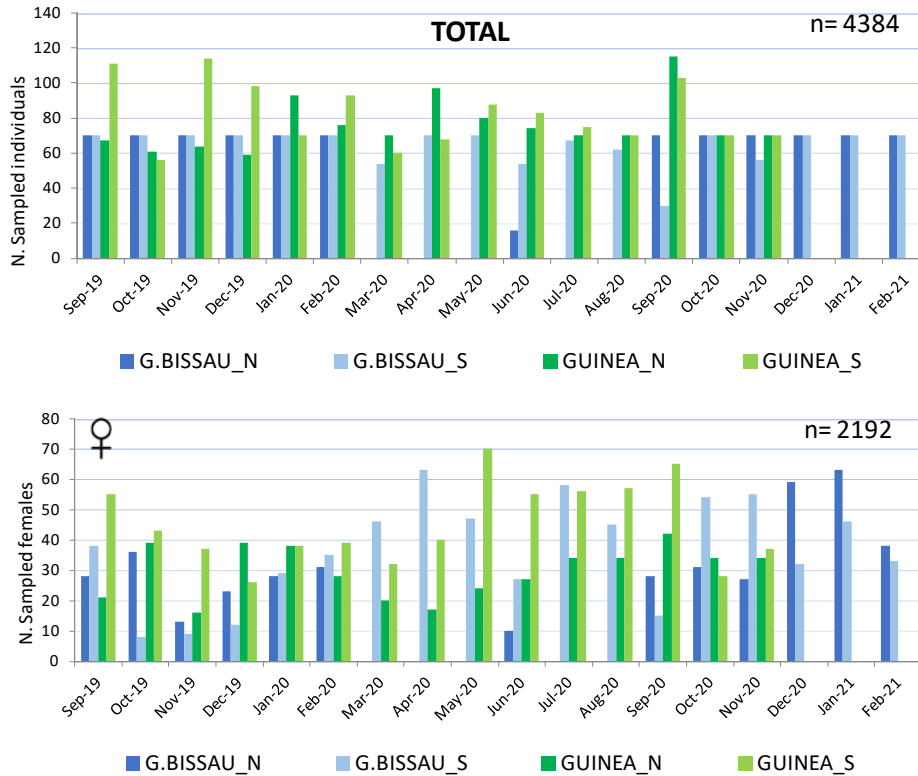


Figure 3.1.22.- Number of sampled individuals of *P. elongatus* (PSE) by month and country-area, for all combined sexes (top) and for females (bottom).

Table 3.1.15.- Number of individuals of *P. elongatus* (PSE), sampled by quarter in Guinea-Bissau and Guinea, by area (North and South).

QUARTER	G.Biss_N	G.Biss_S	Gui_N	Gui_S
Quarter 1	280	334	181	223
Quarter 2	16	194	184	239
Quarter 3	140	229	299	359
Quarter 4	420	406	248	406
Total	856	1163	912	1227

Minimum, maximum, mean and median values of length are summarized and plotted by zone in Table 3.4.1. and Figure 3.4.1. of Annex 3.

Weight-related parameters

The individuals of *P. elongatus* sampled in North Guinea-Bissau were smaller than in the other three areas (median values of 21.6 cm and 23.0-24.2 cm TL, respectively) (Annex 3). Thus, a common length range of 11-35 cm TL was selected to provide the parameters of the length-weight relationships while for comparison among areas, a more restrictive length range of 17.8-30.5 cm TL was used.

Weight-length relationships

The results of the gutted weight (Wg)–total length (Lt) relationship of *P. elongatus*, their coefficients of determination, sample size and length and weight ranges for each country-area are shown by sex and for sex combined in Table 3.1.16 and plotted in Figure 3.1.23. Many inconsistencies were found in data from Guinea-Bissau, and thus, only those individuals for which the two variables potentially correlated were considered for the analysis.

The regression lines and slopes indicate similar relationships between the two areas (North and South) in Guinea and in Guinea-Bissau North, with values around 2-9-3.0. The pairwise comparisons made showed no significant differences between the four country-areas for combined sexes (Section 4.4.1. in Annex 4). However, when analysing by sex, significant differences were found between northern Guinea-Bissau and the other country-areas (for males) and between northern Guinea-Bissau and the two southern areas of both countries (for females).

Table 3.1.16.- Parameters of the gutted weight-length relationships for *P. elongatus* (PSE) estimated in Guinea-Bissau and Guinea, by area (North and South).

MALES				
<i>P. elongatus</i> (PSE)	G.Biss_N	G.Biss_S	GUI_N	GUI_S
a	0.005	0.004	0.006	0.007
b	3.12	3.20	3.08	3.07
n	313	412	346	306
R ²	0.88	0.96	0.97	0.97
Length range	14.3-32.7	11.0-34.0	15.5-35.0	17.0-35.0
Gutted weight range	20-275	6-338	35-408	42-401
Weight range	21-288	6-354	37-441	44-421
FEMALES				
<i>P. elongatus</i> (PSE)	G.Biss_N	G.Biss_S	GUI_N	GUI_S
a	0.012	0.003	0.010	0.005
b	2.83	3.25	2.95	3.14
n	318	566	382	1142
R ²	0.87	0.97	0.97	0.97
Length range	16.5-32.0	11.0-35.0	14.5-35.0	16.0-35.0
Gutted weight range	22-251	6.8-356	38-429	32-648
Weight range	24-280	7.5-392	41-473	35-675
SEX COMBINED				
<i>P. elongatus</i> (PSE)	G.Biss_N	G.Biss_S	GUI_N	GUI_S
a	0.008	0.004	0.007	0.007
b	2.99	3.23	3.07	3.03
n	653	1013	1045	2096
R ²	0.88	0.97	0.99	0.98
Length range	14.3-32.7	11.0-35.0	11.0-35.0	11.0-35.0
Gutted weight range	20-275	6-356	9-429	9-648
Weight range	21-288	6-392	10-473	10-675

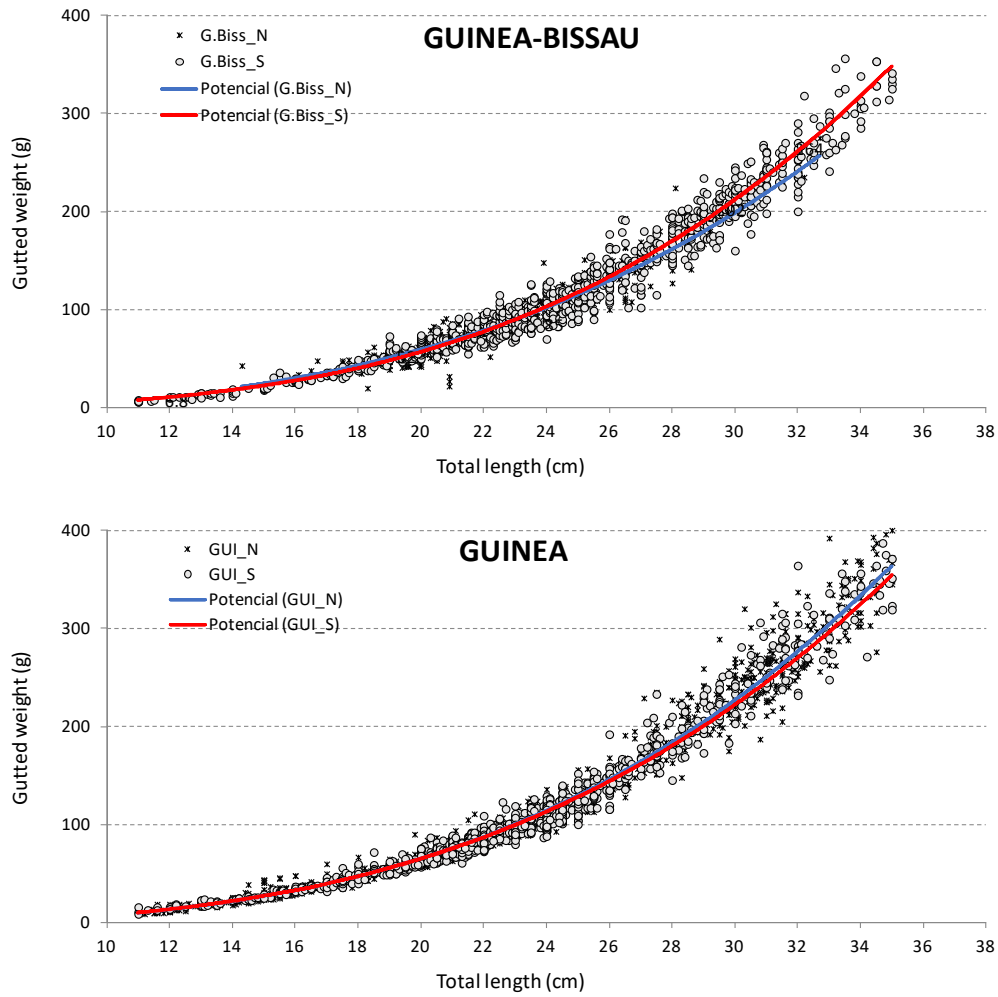


Figure 3.1.23.- Gutted weight–length relationships estimated for *P. elongatus* (PSE) estimated in Guinea-Bissau and Guinea, by area (North and South) and combined sexes.

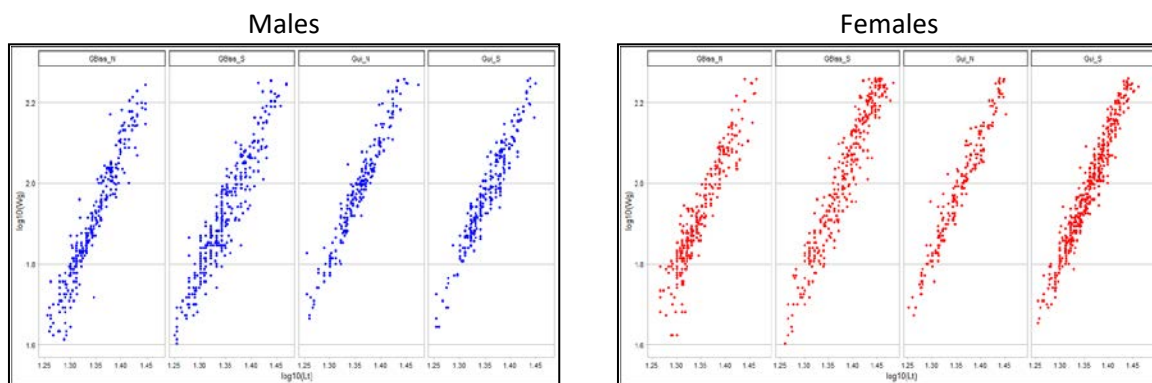


Figure 3.1.24.- Gutted weight–length relationships (log values) estimated for *P. elongatus* (PSE) in Mauritania and Senegal, by area (North and South) and by sex.

Weight conversion factor

The conversion factors, sample sizes for total weight (Wt)–gutted weight (Wg) linear relationships of *P. elongatus* in each country-area (sex combined) are shown in Table 3.1.17.

Similar Wt–Wg conversion factors were obtained in the analyzed country-areas, between 0.97 and 0.99, although significant difference ($p < 0.05$) between Guinea North and South was obtained by the pairwise comparisons.

Table 3.1.17.- Parameters of the total weight (Wt)–gutted weight (Wg) linear relationships for *P. elongatus* (PSE) estimated in Guinea-Bissau and Guinea, by area (North and South).

SEX COMBINED				
<i>P. elongatus</i> (PSE)	G.Biss_N	G.Biss_S	Gui_N	Gui_S
B	0.98	0.99	0.99	0.97
N	636	915	813	917
R ²	0.958	0.990	0.999	0.977

Condition factor

The Le Cren’s condition factor (K) was estimated for mature females of *P. elongatus*, by quarter and country-area (Figure 3.1.25). The trend of the median values of K was similar for both areas in Guinea, but different to those in Guinea-Bissau. In Guinea, the higher mean K was during the last and first quarter, while in Guinea-Bissau-South, the highest values was observed later, in the second quarter. No information was available for the second quarter in Guinea-Bissau North. When comparing the median values by quarter, significant differences were found in all areas with the exception of North and South Guinea in the first and fourth quarter. Oppositely, for the second quarter, no significant differences were found among all areas except among the two areas in Guinea. Finally, for the third quarter, significant differences were found between Guinea-Bissau North and the two areas in Guinea (section 4.4.3 in Annex 4).

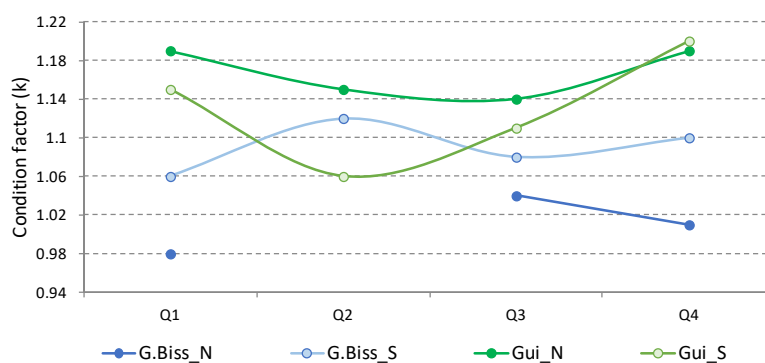


Figure 3.1.25.- Quarterly variation of the median values of the Le Cren’s condition factor (K) in mature females of *P. elongatus* (PSE) in Guinea-Bissau and Guinea, by area (North and South).

Reproduction parameters and features

Sex-ratio

Sex-ratio (male:female) of *P. elongatus* ranged between 1:1 (50% of females) in Guinea-Bissau North to 1:2.1 (67% of females) in Guinea South (see Table 3.1.18). It should be noted that sampled length range is smaller in Guinea-Bissau than in Guinea.

Table 3.1.18.- Sex ratio values estimated for *P. elongatus* (PSE) estimated in Guinea-Bissau and Guinea, by area (North and South).

Sex-ratio	G.Biss_N	G.Biss_S	Gui_N	Gui_S
% Females	50%	59%	55%	67%
M:F	1:1	1:1.4	1:1.2	1:2.1

In general, females predominated at sizes higher than 28 cm in all areas, and from this size an overall increase in the proportion of females is observed as the fish grows (Figure 3.1.26). For smaller lengths, sex-ratios show different patterns in the four country-areas.

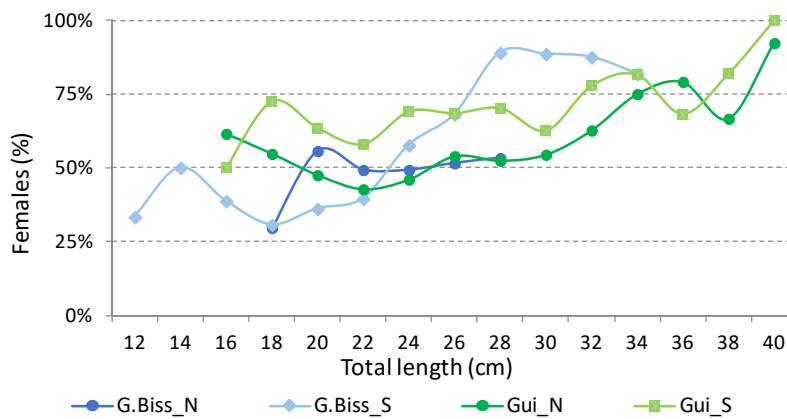


Figure 3.1.26.- Female percentage of *P. elongatus* (PSE) by length class (2 cm) in Guinea-Bissau and Guinea, by area (North and South).

Spawning period

The monthly percentage of mature and immature females of *P. elongatus* and the monthly mean of their GSI are shown in Figure 3.1.27. This GSI was estimated only for females bigger than 21 cm, which is the size at first maturity estimated for females in Guinea-Bissau previously by Kromer et al. (1994).

Biological information of *P. elongatus* from Guinea-Bissau North was quite uncompleted, lacking data from five months of the year. In the rest of the country-areas, high proportions of mature females were found during most of the analysed months. A combination of the knowledge provided by the proportion of mature females and the highest mean values of the GSI were considered to determine the spawning peaks of the species, as sometimes the GSI was lacking for certain months (as in Guinea-Bissau South). In those cases that both figures were inconsistent, the GSI was mainly used to define the spawning season, taking into account that it is based on objective measures (weights) while maturity scaling by macroscopic observation is more subjective and sometimes difficult for samplers.

The species spawns throughout the year, with two spawning peaks in Guinea-Bissau South and Guinea (North and South). The first peak seems to take place at the end of the dry season, mainly between March to April (in Guinea South) or to May (in Guinea-Bissau South and Guinea North). The scarce information from Guinea-Bissau North only seems to indicate an earlier start (February) of this first

spawning peak in this area. The second spawning peak seems to occur during the rainy season, starting in June or July and lasting to August (in Guinea) or September (in Guinea-Bissau South) (Table 3.1.19).

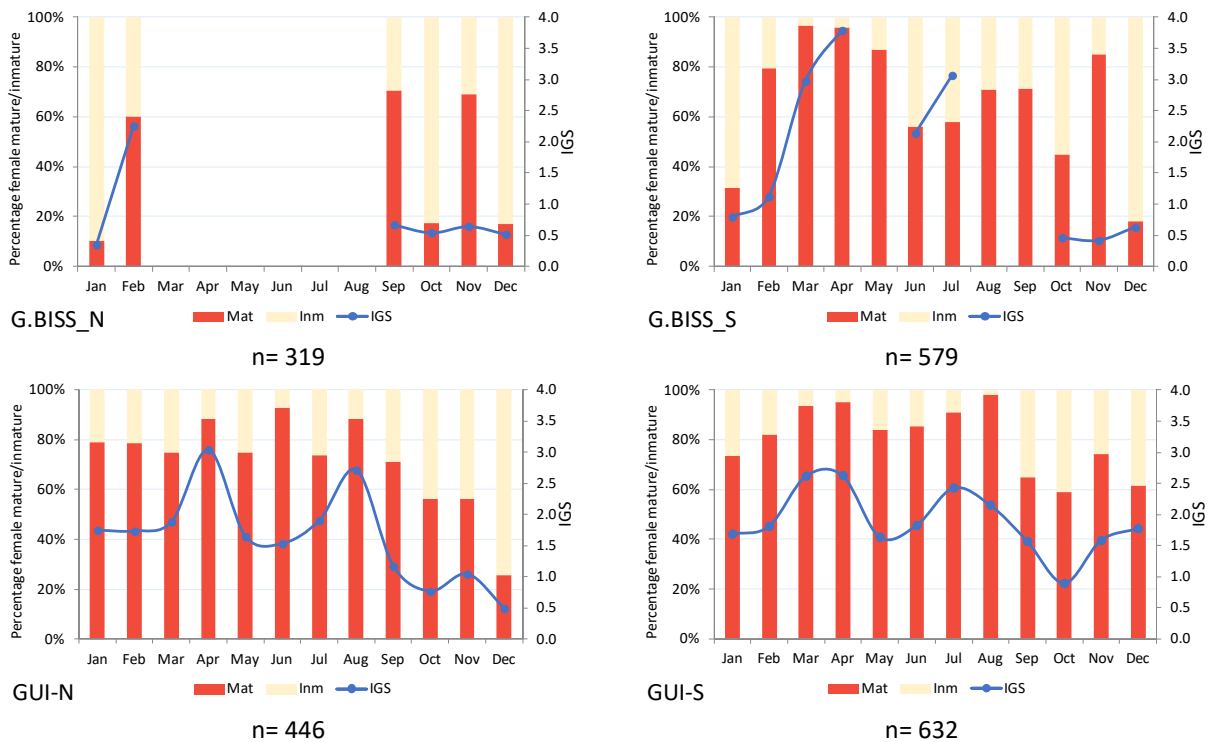


Figure 3.1.27.- Monthly percentage of mature/immature females and mean GSI of for *P. elongatus* (PSE) in Guinea-Bissau (top) and Guinea (bottom), by area, North (left) and South (right).

Maturity ogives

The maturity ogives of females of bobo croakers in the northern areas of both Guinea-Bissau and Guinea are shown in Figure 3.1.28. While the ogive from Guinea North showed a relatively good fit, a poor fitting of the observations of maturity at length to the model predictions was observed in Guinea-Bissau North. Not quality or enough information was available to estimate a representative maturity ogive for the species in any of the southern areas analysed.

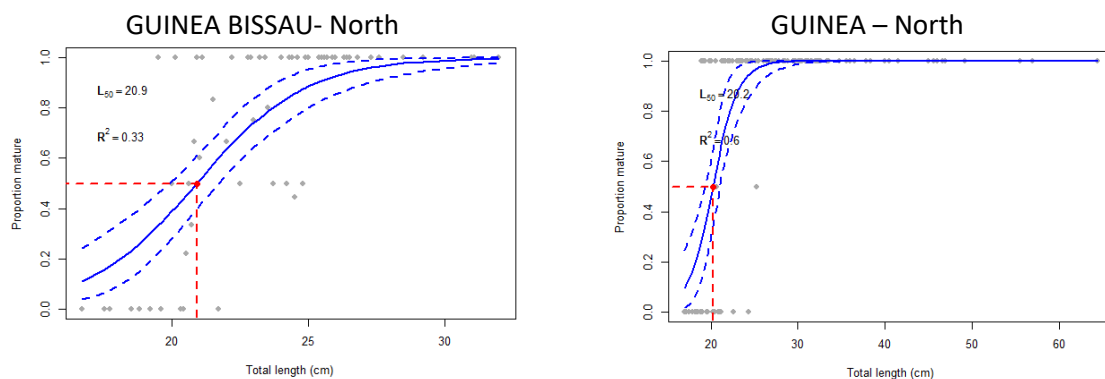


Figure 3.1.28.- Maturity ogives of females of *P. elongatus* (PSE) in Guinea-Bissau North (left) and Guinea North (right).

Length at first maturity

The lengths at first maturity (L50), their confidence intervals and the number of females used for the estimations in their correspondent spawning peaks are shown in Table 3.1.19.

Reliable estimations of L50s for females of *P. elongatus* were obtained for the two northern areas, while no robust estimates are available for the southern areas. The L50 values are quite similar in both northern areas (20.2 and 20.9 cm TL in Guinea and Guinea-Bissau, respectively) showing overlapping of their confidence intervals.

Table 3.1.19.- Spawning period and peaks of females of *P. elongatus* (PSE) in Guinea-Bissau and Guinea, by area (North and South).

FEMALES	G.BISSAU_N	G.BISSAU_S	GUINEA_N	GUINEA_S
Spawning period	6 of 7 months sampled	All year	All year	All year
Spawning peaks	Feb + Sep + Nov (uncompleted)	March-May/ Jul-Sep	March-May/ Jul-Ago	March-April/ Jun-Ago
L50 (Confidence intervals), mm CL	20.9 (19.9 – 21.7)	–	20.2 (19.3 – 21)	–
N	130	259	129	240
Length range (median) (mm CL)	16.7 – 32.0 (22.9)	24.0 – 42.3 (29)	17 – 64.5 (24.7)	16.6 – 60.2 (25.5)

3. 1. 5. *Pseudotolithus senegalensis* (PSS)

A total of 2744 individuals of Cassava croaker *P. senegalensis* were sampled for estimating life history traits during the period October 2019-March 2021. Figure 3.1.29 shows the number of individuals sampled by month and country/area, for females and for sex-combined. As the first biological samplings were performed later in Côte d'Ivoire, the synchronized sampling period among the two countries was from May 2020 to February 2021 (Figure 3.1.29). Table 2.5 in Annex 2 shows the total number of individuals sampled for biology, by sex, length range (cm of TL) and country/area.

The number of individuals sampled by quarter and country-area is shown in Table 3.1.20. Côte d'Ivoire West lacks sampling during the second quarter, due to COVID related lockdown that affected for a longer period to the industrial fleet, which is the one targeting this species in the area (Tapé, *pers. comm.*). Ghana West is undersampled during the first semester, partly related to COVID and partly explained by the absence of the species in the fishery (Ansong, *pers. comm.*) that was previously described in the same area by Okyere & Blay (2020) and attributed to the migration of fish to deeper waters from April to June.

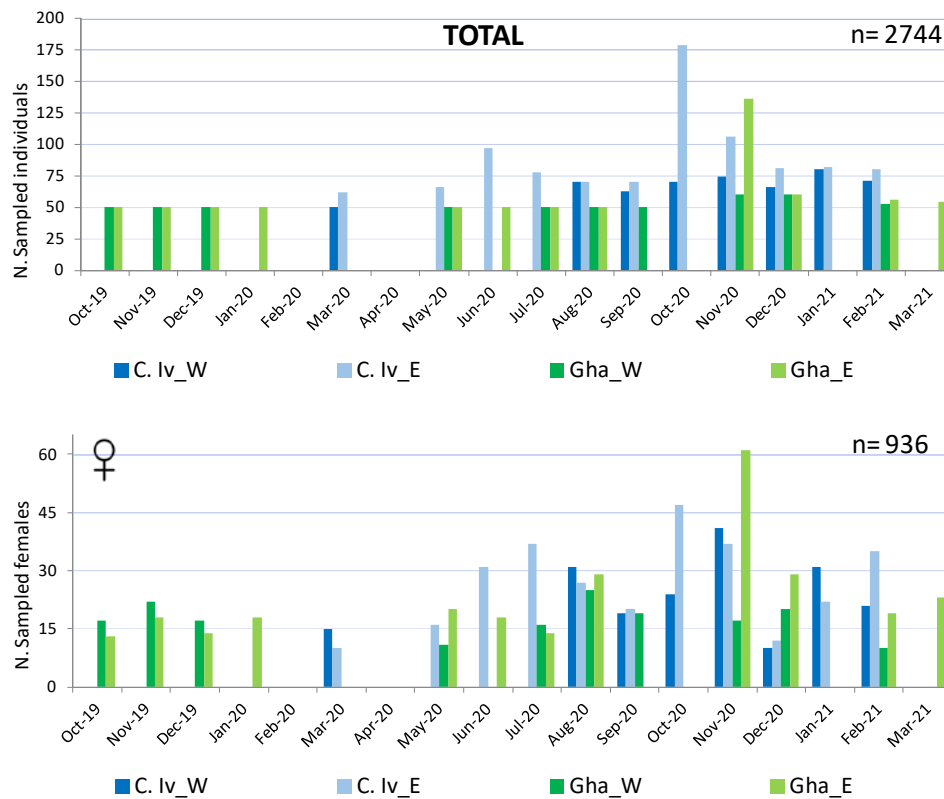


Figure 3.1.29.- Number of *P. senegalensis* (PSS) sampled by month and country-area, for combined sexes (top) and for females (bottom).

Table 3.1.20.- Number of individuals of *P. senegalensis* (PSS), sampled by quarter in Côte d'Ivoire and Ghana, by area (West and East).

QUARTER	C.Iv_W	C.Iv_E	Gha_W	Gha_E
Quarter 1	201	224	53	160
Quarter 2	–	163	50	100
Quarter 3	133	218	150	100
Quarter 4	210	366	270	346
Total	544	971	523	706

Minimum, maximum, mean and median values of length are summarized and plotted by zone in Table 3.5.1. and Figure 3.5.1. of Annex 3.

Certain differences were found in the length of the sampled *P. senegalensis* individuals among zones. This was especially patent between West and East Côte d'Ivoire, with median values of 26.5 cm and 24.4 cm TL, respectively. This fact is explained by the different type of fisheries developed in each area: while *P. senegalensis* is mainly caught by coastal industrial trawlers in the western area, it is fished by artisanal fisheries (mainly gillnets) in the East (Tapé, *pers. comm.*). This means that they are fished at different depth ranges, inhabited by different fractions of the population: smaller individuals in or near the estuaries, where they are fished by artisanal fleets and bigger individuals at deeper waters, where they are fished by the industrial fleet.

Weight-related parameters

Taking into account the difference sizes in different areas of Côte d'Ivoire explained above, a common length range of 17-40 cm was selected for comparison among areas.

Weight-length relationships

The results of the gutted weight (Wg)–total length (Lt) relationship of *P. senegalensis*, their coefficients of determination, sample size and length and weight ranges for each country-area are shown by sex and for combined sexes in Table 3.1.21 and plotted for combined sexes in Figure 3.1.30.

Table 3.1.21.- Parameters of the gutted weight-length relationships for *P. senegalensis* (PSS) estimated in Côte d'Ivoire and Ghana, by area (West and East).

MALES				
<i>P. senegalensis</i> (PSS)	C.Ivoire_W	C.Ivoire_E	Ghana_W	Ghana_E
a	0.002	0.003	0.013	0.003
b	3.36	3.30	2.84	3.28
n	264	602	286	365
R ²	0.98	0.98	0.90	0.95
Length range (TL, cm)	17.0-40.0	17.0-40.0	18.5-38.0	18.0-40.0
Gutted weight range (g)	24-633	28-651	43-440	41-810
Weight range (g)	25-701	29-712	51-450	43-840
FEMALES				
<i>P. senegalensis</i> (PSS)	C.Ivoire_W	C.Ivoire_E	Ghana_W	Ghana_E
a	0.003	0.003	0.010	0.003
b	3.33	3.26	2.92	3.31
n	143	276	173	270
R ²	0.98	0.98	0.85	0.93
Length range (TL, cm)	17.0-40.0	17.0-40.0	18.0-36.0	18.0-40.0
Gutted weight range (g)	25-569	30-530	44-330	43-640
Weight range (g)	26-638	31-601	45-340	46-710
SEX COMBINED				
<i>P. senegalensis</i> (PSS)	C.Ivoire_W	C.Ivoire_E	Ghana_W	Ghana_E
a	0.002	0.003	0.009	0.003
b	3.36	3.30	2.96	3.28
n	409	885	521	684
R ²	0.96	0.97	0.88	0.91
Length range (TL, cm)	17.0-40.0	17.0-40.0	17.4-38.0	17.0-40.0
Gutted weight range (g)	16-633	28-651	38-440	40-810
Weight range (g)	18-701	29-712	40-450	41-840

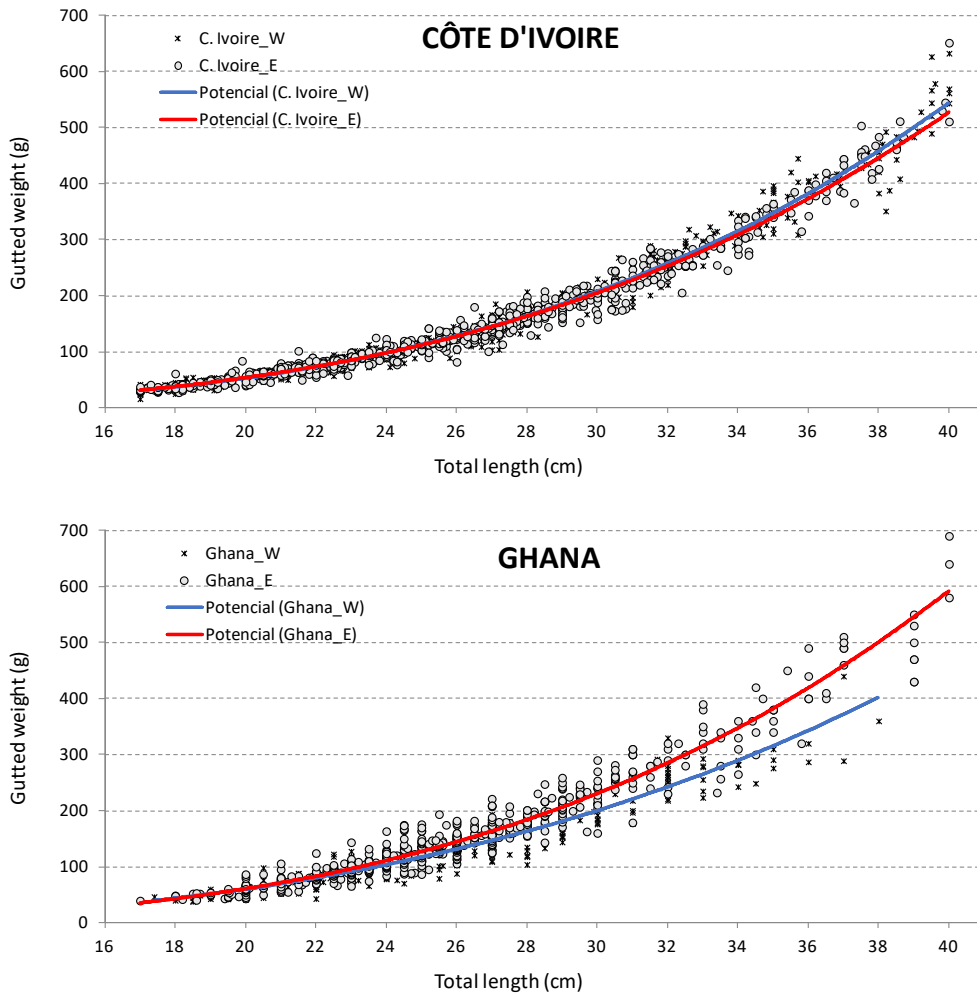


Figure 3.1.30.- Gutted weight–length relationships estimated for *P. senegalensis* (PSS) estimated in Côte d’Ivoire and Ghana, by area (West and East) and combined sexes.

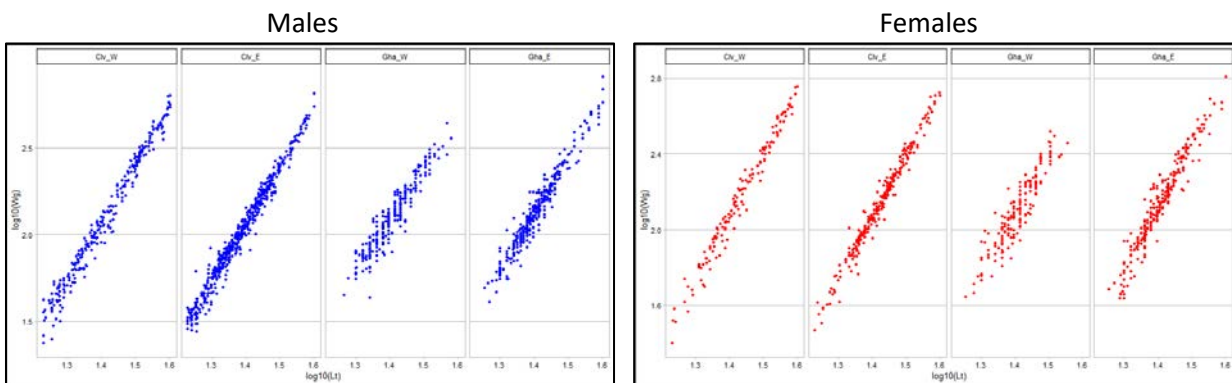


Figure 3.1.31.- Gutted weight–length relationships (log values) estimated for *P. senegalensis* (PSS) in d’Ivoire and Ghana, by area and by sex.

The regression lines indicate similar relationships between the two areas in Côte d’Ivoire (West and East), and Ghana East, with slopes between 3.2-3.3 for sex combined, while the slope in Ghana West is lower (2.96). The same differences among the three areas and Ghana West occur when comparing males and females, separately (Table 3.1.21 and Figure 3.1.31). In fact, significant differences are found between Ghana West and the other three areas, by the pairwise comparisons, both for males and females and for combined sexes (Section 4.5.1 in Annex 4). These differences might be partially due to

a smaller length range sampled in this area. It should be noted that a problem in sampling conducted in Ghana West almost systematically occurred, involving a lack of accuracy in the measures taken (individuals were measured in 0.5 or 1 cm instead in mm).

Weight conversion factor

The conversion factors, sample sizes for total weight (Wt)–gutted weight (Wg) linear relationships of *P. senegalensis* in each country-area (sex combined) are shown in Table 3.1.22.

Similar Wt–Wg conversion factors were obtained in the analyzed country-areas (0.99-1.01). No significant differences ($p > 0.05$) were found among the slopes in the different areas by the pairwise comparisons performed (section 4.5.2 of Annex 4).

Table 3.1.22.- Parameters of the total weight (Wt)–gutted weight (Wg) linear relationships for *P. senegalensis* (PSS) estimated in Côte d’Ivoire and Ghana, by area (West and East) and combined sexes.

<i>P. senegalensis</i> (PSS)	C.Ivoire_W	C.Ivoire_E	Ghana_W	Ghana_E
b	1.003	0.996	1.009	0.995
N	410	847	521	684
R ²	0.998	0.994	0.985	0.988

Condition factor

The Le Cren’s condition factor (K) was estimated for mature females of *P. senegalensis*, by quarter and country-area (Figure 3.1.32). No information was available for the second quarter in Côte d’Ivoire West and for the first quarter in Ghana West. The trend of the median values of K was similar for the eastern areas of both countries, but different to those partially available in the western areas. The lowest and highest mean values of K in the eastern areas were estimated for the second and third quarter, respectively. Lowest available mean values of K in the western areas were obtained during the third quarter. When comparing the median values by quarter, significant differences were found between Ghana East and the two areas in Côte d’Ivoire during the first quarter, between the two eastern areas in the third quarter, and between Ghana East and the other three areas during the fourth quarter (section 4.5.3 in Annex 4).

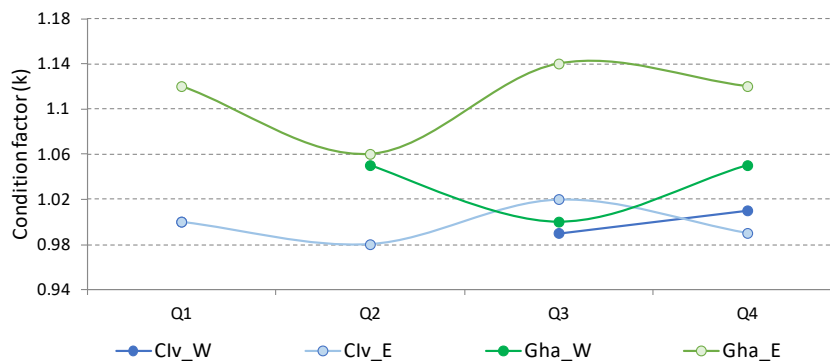


Figure 3.1.32.- Quarterly variation of the median values of the Le Cren’s condition factor (K) in mature females of *P. senegalensis* (PSS) in Côte d’Ivoire and Ghana, by area (West and East).

Reproduction parameters and features

Sex-ratio

Sex-ratio (male:female) of *P. senegalensis* ranged between 1:0.4 (31% of females) in Côte d'Ivoire East and 1:0.7 (42% of females) in Ghana East (Table 3.1.23). The proportion of males was higher in all country-areas.

Table 3.1.23.- Sex ratio values estimated for *P. senegalensis* (PSS) in Côte d'Ivoire and Ghana, by area (West and East).

Sex-ratio	CIV_W	CIV_E	Gha_W	Gha_E
% Females	36%	31%	38%	42%
M:F	1:0.6	1:0.4	1:0.6	1:0.7

The sex ratio of *P. senegalensis* was analyzed by length (Figure 3.1.33). Length range for females was smaller in Ghana (20-38 cm) than in Côte d'Ivoire (14-46 cm). A similar overall trend was observed in most areas, with a proportion of males between 50-75% from the 22 cm range onwards. A higher proportion of males (75-90%) is observed at sizes below 20 cm.

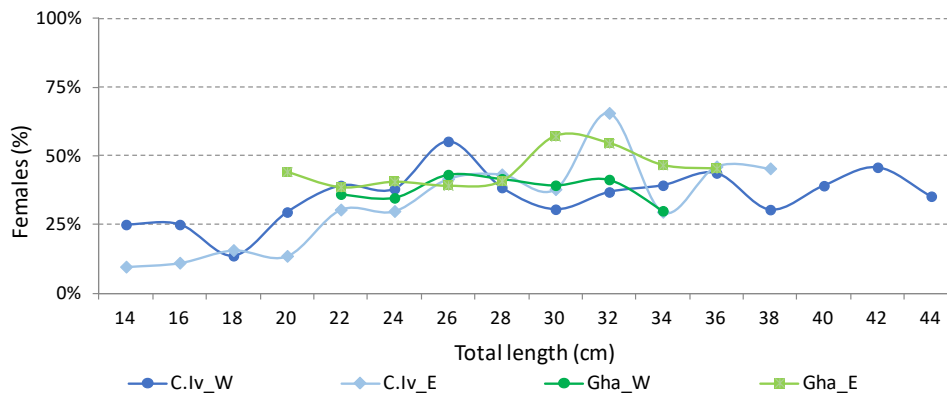


Figure 3.1.33.- Female percentage of *P. senegalensis* (PSS) by length class (2 cm) in Côte d'Ivoire and Ghana, by area (West and East).

Spawning period

The monthly percentage of mature and immature females of *P. senegalensis* and the monthly mean of their GSI is shown in Figure 3.1.34. This GSI was estimated only for females higher than 24.6 cm, which is the size at first maturity estimated for females of this species in neighbour waters off Benin (Sossoukpe et al., 2013).

It should be taken into account the lack of biological samplings of *P. senegalensis* during five months in the western areas of both Côte d'Ivoire and Ghana, and during two-three months in the eastern areas of both countries. This makes difficult the identification of clear spawning seasons in these areas. In addition, the monthly percentage of mature females was low or null in many of the sampled months in Ghana, probably linked to the small size of the sampled individuals (Table 2.5 of Annex 2).

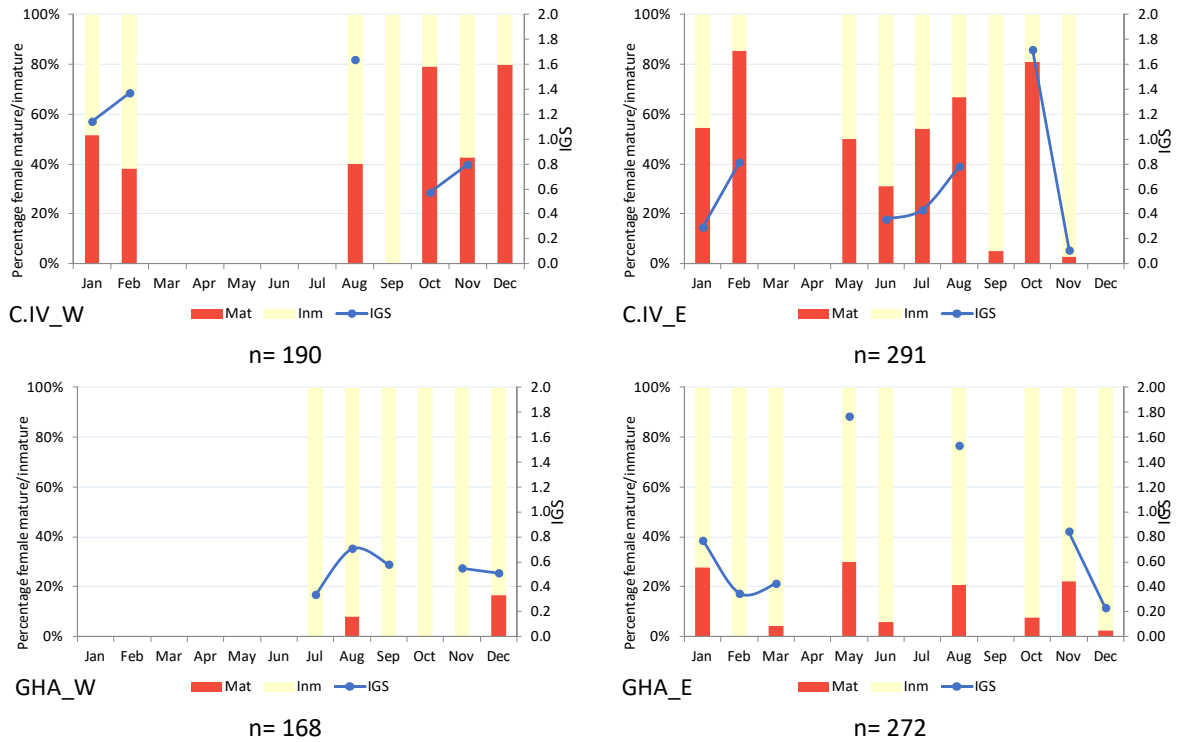


Figure 3.1.34.- Monthly percentage of mature/immature females and mean GSI of for *P. senegalensis* (PSS) in Côte d'Ivoire (top) and Ghana (bottom), by area West (left) and East (right).

Maturity ogives

The maturity ogives of females in Côte d'Ivoire and Ghana, are shown, by area, in Figure 3.1.35.

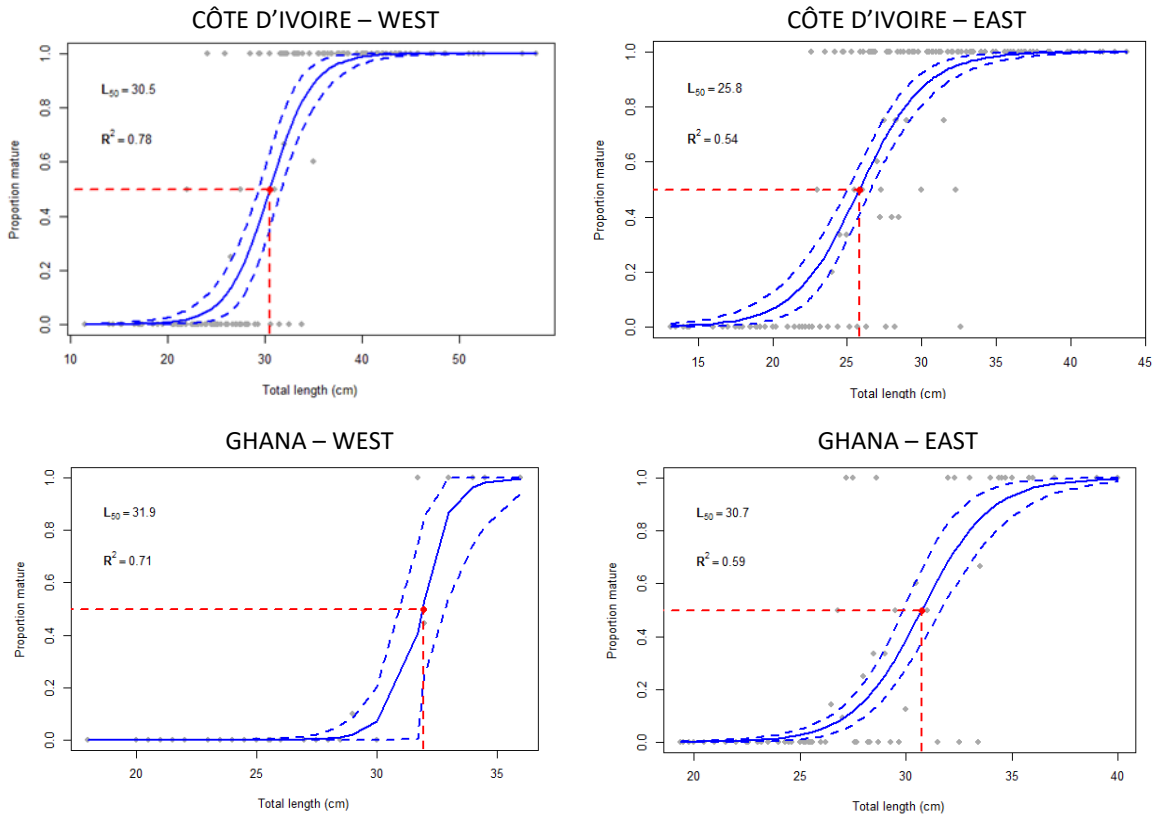


Figure 3.1.35.- Maturity ogives of females of *P. senegalensis* (PSS) in Côte d'Ivoire (top) and Ghana (bottom), by area West (left) and East (right).

Length at first maturity

The estimated values of the length at first maturity (L50) of *P. senegalensis*, their confidence intervals and the number of females used for the estimations in their correspondent spawning peaks are shown in Table 3.1.24. Lengths at first maturity (L50) of females are consistent between three of the four studied countries-areas, showing close values, between 30.5 and 31.9 cm TL in Côte d'Ivoire West and Ghana (West and East) and overlapped confidence intervals among the three areas. The L50 estimated for Côte d'Ivoire East was smaller (25.8 cm) and more in agreement with the one previously estimated in waters off Benin (Sossoukpe et al., 2013).

Table 3.1.24.- Spawning period and peaks, L50s and their confidence intervals, number of individuals used for their estimation and their length range (CL in mm). *P. senegalensis* (PSS) in Côte d'Ivoire and Ghana by area (West and East).

FEMALES	C. IVOIRE_W	C. IVOIRE_E	GHANA_W	GHANA_E
Spawning period	6 of 7 sampled months ^{*1}	All sampled months	2 of 6 sampled months	8 of 9 sampled months ^{*4}
Spawning peaks	Unknown	Unknown ^{*2}	Unknown ^{*3}	Unknown
L50 (Confidence intervals), cm	30.5 (29.4 – 31.7)	25.8 (25.0 – 26.6)	31.9 (31.1 – 32.8)	30.7 (29.9 – 31.6)
N	171	231	135	252
Length range (median), cm	11.5 – 58.0 (29.5)	13.2 – 43.8 (28.0)	18.0 – 36.0 (26.0)	19.4 – 40.0 (25.8)

^{*1}The six sampled months considered for L50 estimation.

^{*2}Months considered for L50 estimation: all except September and November.

^{*3}Months considered for L50 estimation: August and December.

^{*4}The eight sampled months considered for L50 estimation.

3. 1. 6. *Pagellus bellottii* (PAR)

A total of 2764 individuals of red pandora *P. bellottii* were sampled for obtaining life history traits during the period October 2019-March 2021. The number of individuals sampled by month and country/area, for females and combined sexes is shown in Figure 3.1.36. As the biological sampling started later in Côte d’Ivoire, the synchronized sampling period among the two countries was May 2020- February 2021 (Figure 3.1.36). The total number of individuals sampled for biology, by sex, length range (cm of TL) and country/area is shown in Annex 2-Table 2.6.

Table 3.1.25 shows the number of individuals sampled by quarter and country-area. Côte d’Ivoire West lacks sampling during the second quarter, due to the COVID pandemic lockdown that affect for a longer period to the industrial fleet, which is the one targeting this species in the area (Tapé, *pers. comm.*). Samples from Ghana West are also lacking from January to April, due to COVID and to other reasons.

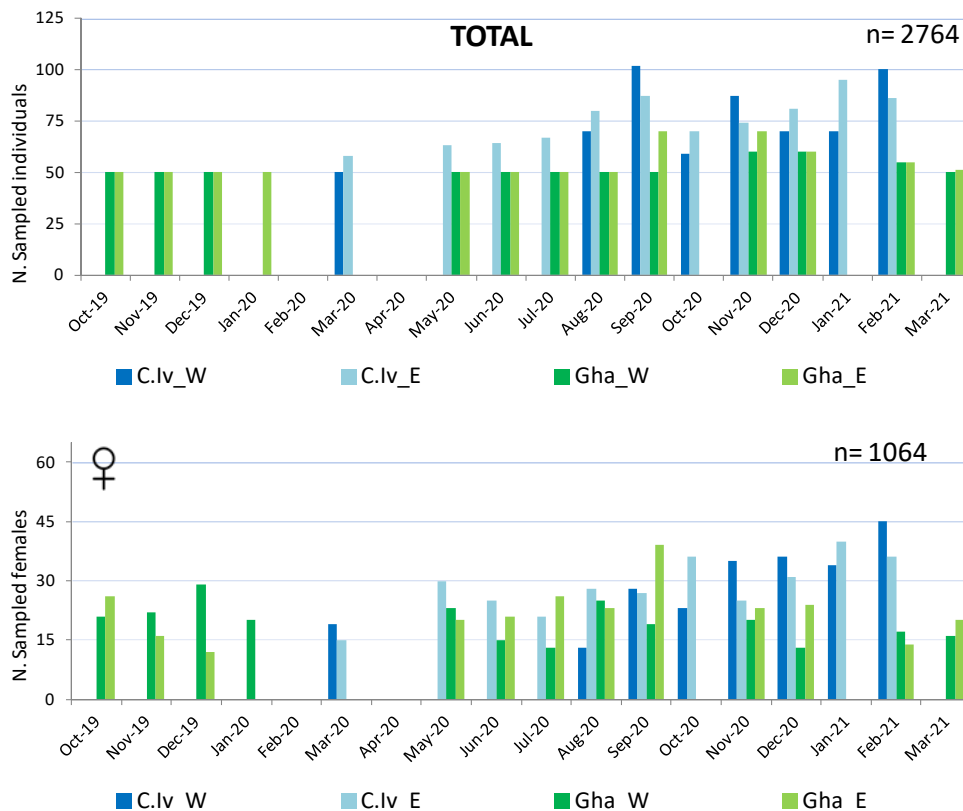


Figure 3.1.36.- Number of *P. bellottii* (PAR) sampled by month and country-area, for combined sexes (top) and for females (bottom).

Table 3.1.25.- Number of individuals of *P. bellottii* (PAR) sampled by quarter in Côte d’Ivoire and Ghana, by area (West and East).

QUARTER	C.Iv_W	C.Iv_E	Gha_W	Gha_E
Quarter 1	220	239	105	156
Quarter 2	–	127	100	100
Quarter 3	172	234	150	170
Quarter 4	216	225	270	280
Total	608	825	625	706

Minimum, maximum, mean and median values of length are summarized and plotted by zone in Table 3.6.1. and Figure 3.6.1. of Annex 3.

Bigger individuals were sampled in West Côte d'Ivoire (median value of 23 cm TL) than in the other three areas (median values of 19.5-20 cm TL). As explained for *P. senegalensis*, this is due to the fact that this species is also mainly fished by industrial coastal trawlers, at deeper waters, while smaller individuals in shallower waters near or into the estuaries are fished by the artisanal fleet (Tapé, *pers. comm.*).

Weight-related parameters

To make possible any comparison of weight related parameters of *P. bellottii*, a common length range of 13-25 cm between the four country-areas was selected.

Weight–length relationships

The results of the gutted weight (Wg)–total length (Lt) relationship of *P. bellottii*, their coefficients of determination, sample size and length and weight ranges for each country-area are shown by sex and for sex combined in Table 3.1.26 and plotted for sex combined in Figure 3.1.37.

Table 3.1.26.- Parameters of the gutted weight–length relationships for *P. bellottii* (PAR) estimated in Côte d'Ivoire and Ghana, by area (West and East).

MALES				
<i>P. bellottii</i> (PAR)	C.Ivoire_W	C.Ivoire_E	Ghana_W	Ghana_E
a	0.007	0.011	0.016	0.018
b	3.19	3.05	2.93	2.91
n	290	469	320	286
R ²	0.96	0.98	0.85	0.90
Length range (TL, cm)	13.6-25.0	13.0-25.0	15.0-25.0	13.0-24.5
Gutted weight range (g)	24-223	28-216	44-229	31-212
Weight range (g)	25-238	28-229	46-241	32-217
FEMALES				
<i>P. bellottii</i> (PAR)	C.Ivoire_W	C.Ivoire_E	Ghana_W	Ghana_E
a	0.012	0.011	0.014	0.024
b	3.02	3.04	2.98	2.82
n	201	292	227	276
R ²	0.95	0.97	0.86	0.89
Length range (TL, cm)	13.6-25.0	13.0-25.0	15.0-25.0	13.0-24.5
Gutted weight range (g)	36-221	27-225	46-213	33-223
Weight range (g)	37-236	30-237	50-224	33-228
SEX COMBINED				
<i>P. bellottii</i> (PAR)	C.Ivoire_W	C.Ivoire_E	Ghana_W	Ghana_E
a	0.009	0.011	0.016	0.021
b	3.05	3.13	2.93	2.87
n	492	763	609	687
R ²	0.98	0.96	0.85	0.90
Length range (TL, cm)	13.4-25.0	13.0-25.0	15.0-25.0	13.0-24.5
Gutted weight range (g)	24-223	24-225	44-229	31-222
Weight range (g)	25-238	28-237	46-241	32-228

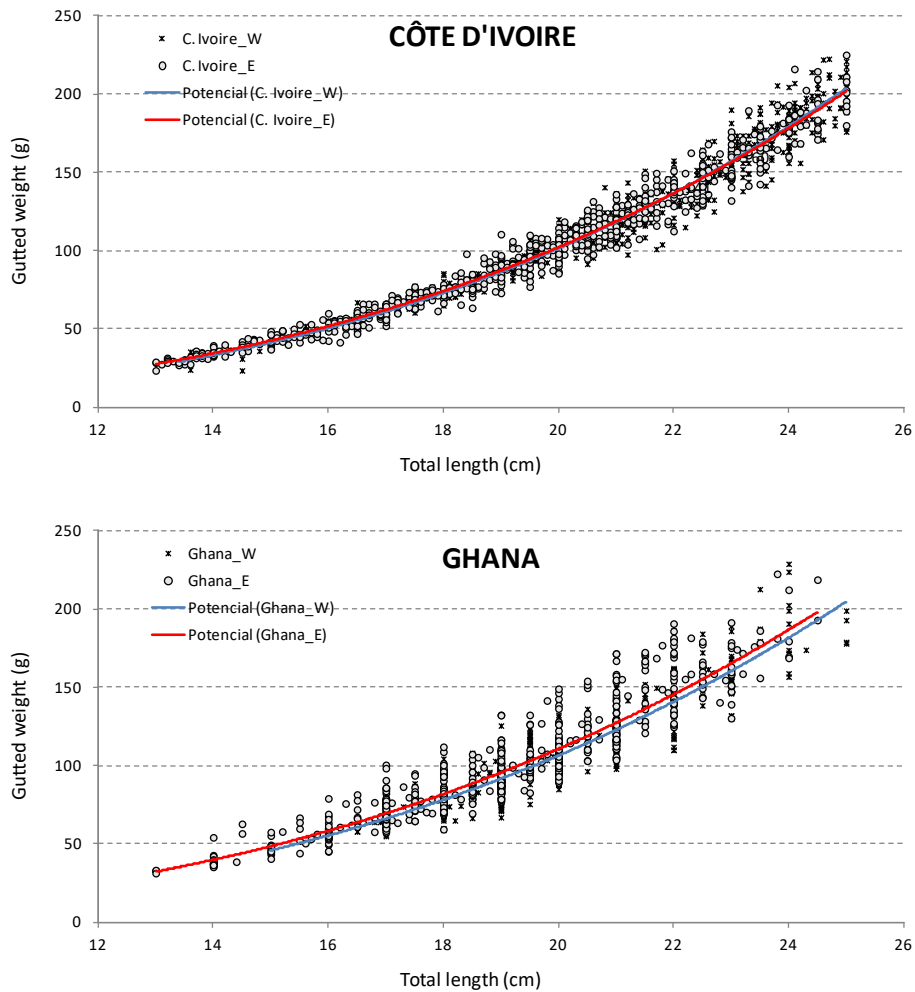


Figure 3.1.37.- Gutted weight–length relationships estimated for *P. bellottii* (PAR) estimated in Côte d’Ivoire and Ghana, by area (West and East) and for combined sexes.

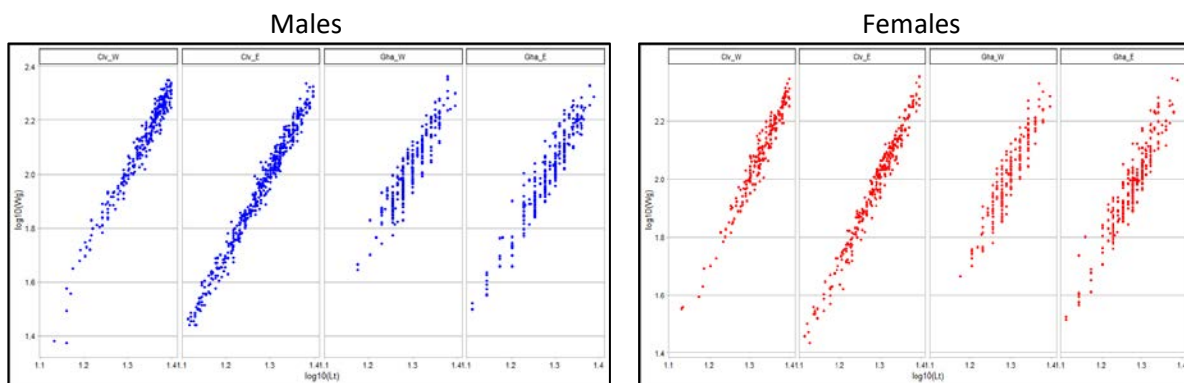


Figure 3.1.38.- Gutted weight–length relationships (log values) estimated for *P. bellottii* (PAR) in Côte d’Ivoire and Ghana, by sex and by area.

The regression lines of *P. bellottii* indicate similar slope values, (around 3.1) between the two areas in Côte d’Ivoire and between two areas of Ghana (around 2.9), both for sex combined. The differences found between both countries might be partially due to the small number of large specimens sampled in Ghana (Table 3.1.26 and Figure 3.1.38). Significant differences ($p < 0.05$) between the two countries for combined sexes were obtained in the pairwise comparison, although when analysing by sex, different patterns are found (Section 4.6.1 in Annex 4). For females, significant differences are only

found between the eastern areas of the two countries while males show significant differences between Côte d'Ivoire West and the other three studied areas. It should be noted that a systematic problem in sampling occurred in Ghana, which involved a lack of accuracy in the measures, as most lengths were taken at 0.5-1 cm level instead of at mm.

Weight conversion factor

The conversion factors, sample sizes for total weight (Wt)–gutted weight (Wg) linear relationships of *P. bellottii* in each country-area (sex combined) are shown in Table 3.1.27.

Similar slopes of the four sampled areas are observed (ranging from 0.99 to 1.01), although significant differences ($p < 0.05$) were found among them except between those from Côte d'Ivoire East and Ghana West (Section 4.6.2 in Annex 4).

Table 3.1.27.- Parameters of the total weight (Wt)–gutted weight (Wg) linear relationships for *P. bellottii* (PAR) estimated in Côte d'Ivoire and Ghana, by area (West and East). Sex combined.

<i>P. bellottii</i> (PAR)	C.Ivoire_W	C.Ivoire_E	Ghana_W	Ghana_E
b	0.986	1.009	1.013	1.000
N	492	763	609	687
R ²	0.995	0.996	0.993	0.997

Condition factor

The Le Cren's condition factor (K) was estimated for mature females of *P. bellottii*, by quarter and country-area (Figure 3.1.39). No information was available for the second quarter in Côte d'Ivoire West. The trend of the median values of K was similar in the two areas of Ghana, but different to those available in Côte d'Ivoire. The lowest and highest mean values of K in the North and South were estimated for the second and fourth quarter, respectively. This patten seem to be delayed in Côte d'Ivoire East, with the lowest K in the third quarter. When comparing the median K by quarter, some significant differences were found, between different areas during the four quarters (section 4.6.3 in Annex 4).

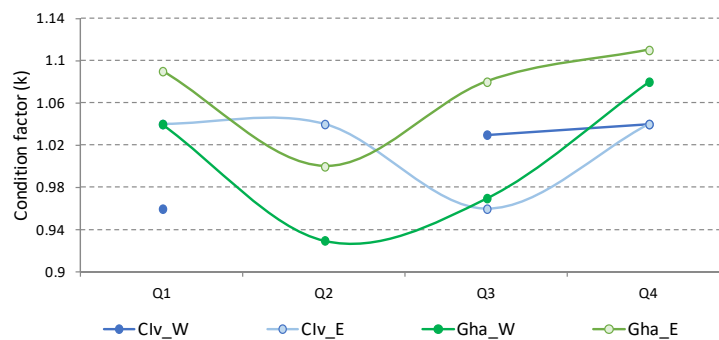


Figure 3.1.39.- Quarterly variation of the median values of the Le Cren's condition factor (K) in mature females of *P. bellottii* (PAR) in Côte d'Ivoire and Ghana, by area (West and East)

Reproduction parameters and features

Sex-ratio

Sex-ratio (male:female) of *P. bellottii* ranged around 1:0.6-0.7 (38-42 % of females) in Côte d'Ivoire East and West and 1:1 (49% of females) in Ghana East (Table 3.1.28). Thus, the proportion of males was higher in the sampled areas of Côte d'Ivoire and Ghana-West, increasing progressively towards the East.

Different trends in the female proportions were observed between the two countries, being more similar within them (Figure 3.1.40). The sex ratio was analyzed by length. Length range for females was smaller in Ghana (14-26 cm) than in Côte d'Ivoire (14-30 cm). The proportion of males generally was higher in all length ranges in Côte d'Ivoire and Ghana West, with the exception of individuals between 20-22 cm in Côte d'Ivoire-West. In Ghana East, females were slightly more abundant than males at sizes between 16 and 20 cm.

Table 3.1.28.- Sex ratio values estimated for *P. bellottii* (PAR) in Côte d'Ivoire and Ghana, by area (West and East).

Sex-ratio	C.Iv_W	C.Iv_E	Gha_W	Gha_E
% Females	38%	38%	42%	49%
M:F	1:0.6	1:0.6	1:0.7	1:1

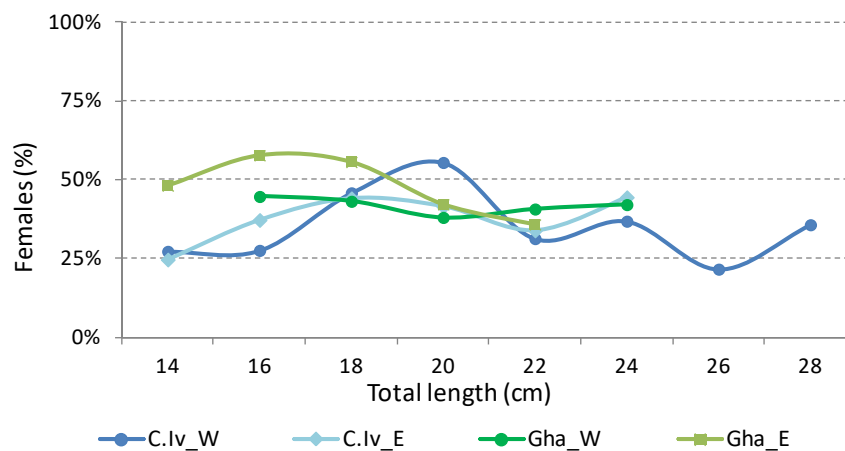


Figure 3.1.40.- Female percentage of *P. bellottii* (PAR) by length class (2 cm) in Côte d'Ivoire and Ghana, by area (West and East).

Spawning period

The monthly percentage of mature and immature females and the monthly mean of their GSI of *P. bellottii* is shown in Figure 3.1.41. This GSI was estimated only for females bigger than 12.4 cm, which is the size at first maturity estimated for females of this species in waters off Côte d'Ivoire (Kouame et al., 2018). It should be noted that this is L50 is fork length (FL) while total length (TL) is used in our work.

In Côte d'Ivoire, mature females were found in all sampled months, indicating that the species might reproduce all year around. However, no mature females were found at least during one month in Ghana (both West and East). The combination of the available information on the proportions of mature females and GSI shows two potential spawning peaks in Côte d'Ivoire West, that cannot be exactly located due to the lack of samplings during four months. One single and extended peak is observed in the other three areas, that starts, at the latest, on May and lasts until August (in Ghana East, the easternmost area studied) or September (in Côte d'Ivoire West, East and Ghana West) (Table 3.1.29).

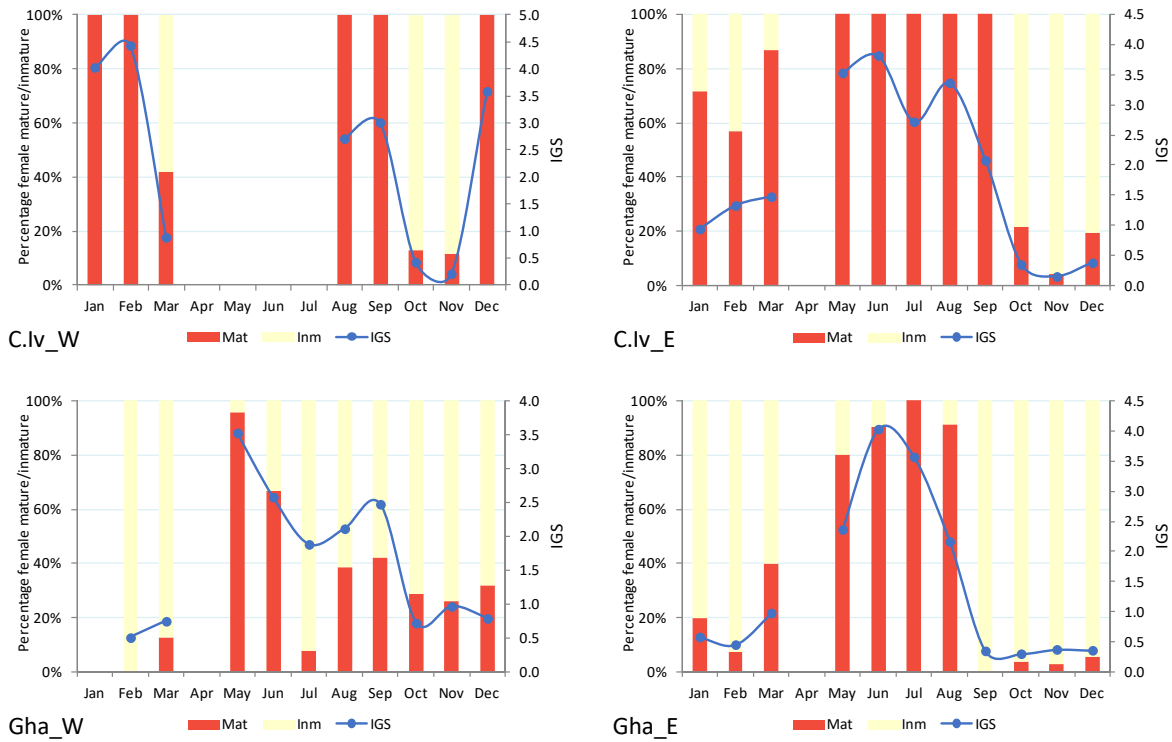


Figure 3.1.41.- Monthly percentage of mature/immature females and mean GSI of *P. bellottii* (PAR) in Côte d'Ivoire (top) and Ghana (bottom), by area West (left) and East (right).

Maturity ogives

The maturity ogives of *P. bellottii* females in Côte d'Ivoire and Ghana East are shown in Figure 3.1.42.

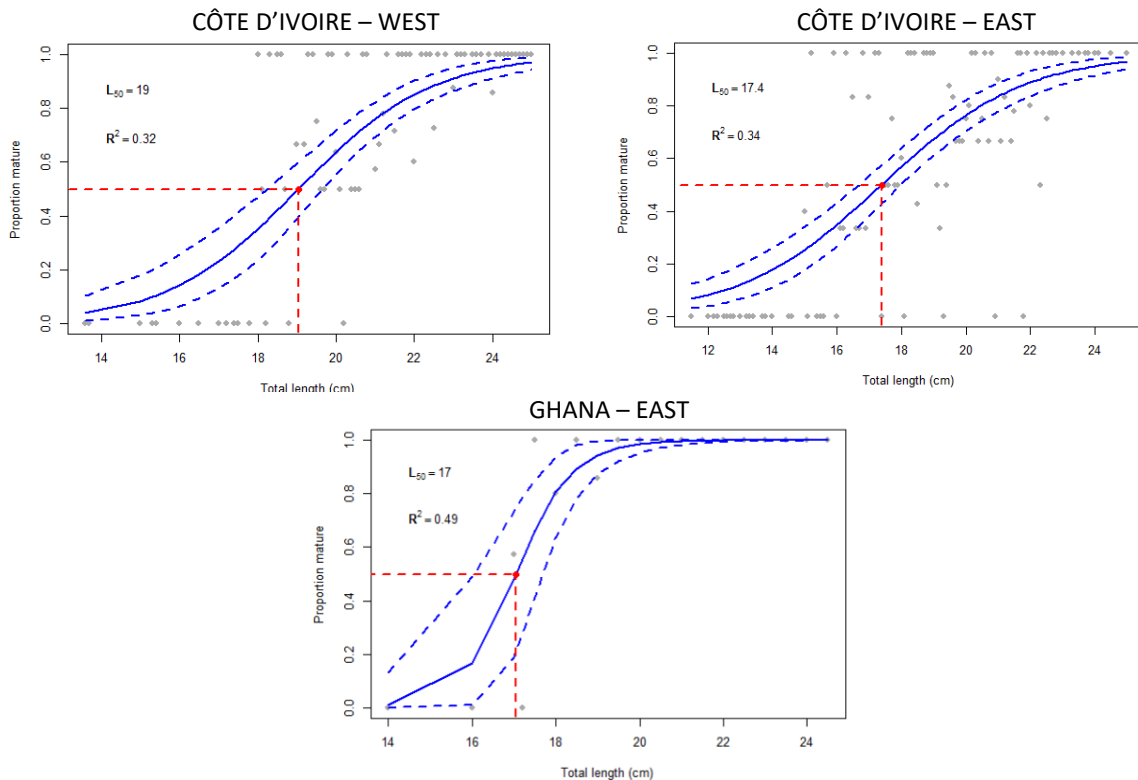


Figure 3.1.42.- Maturity ogives of females of *P. bellottii* (PAR) in Côte d'Ivoire (top) and Ghana (bottom), by area West (left) and East (right).

All the sampled period was used for estimating the ogives in Côte d'Ivoire considering that no immature individuals were registered during the spawning peaks. In fact, the fitting of the maturity at length observations to the model predictions in both areas of Côte d'Ivoire was quite poor, this probably related to the low number of immature individuals. Not quality or enough information was available to estimate a representative maturity ogive for the species in Ghana West. It should be noted that measures of total length in Ghana were taken in cm instead of mm, reducing the accuracy of the observations.

Length at first maturity

The estimated values of the length at first maturity (L50) of *P. bellottii* with their confidence interval and the number of females used for the estimations are shown in Table 3.1.29. It should be noted that although spawning peaks were identified for both areas in Côte d'Ivoire, data from all sampled months were used for the estimation of L50 as the totality of the females were mature during the peaks detected. The confidence intervals of the L50 estimated for the eastern areas of Côte d'Ivoire and Ghana (17.4 and 17.0 cm, respectively) overlapped, and thus not significance differences are considered for both areas. However, results show that mature females are bigger (L50 of 19 cm) in Côte d'Ivoire West, where sampled individuals were also larger (median value of 21.4 cm against median values of 19 and 19.7 cm in the other two areas).

Table 3.1.29.- Spawning periods and peaks, L50s and their confidence intervals, number of individuals and their length range (LT in cm) of females of *P. bellottii* (PAR) in Côte d'Ivoire and Ghana by area (West and East).

FEMALES	C.Iv_W	C.Iv_E	Gha_W	Gha_E
Spawning period	All year	All year	All year	All year
Spawning peak	- Sept - Dec-Feb....*	May-Sept*	May-Sept	May-Aug
L50 (Confidence intervals), cm	19.0 (18.2 - 19.7)	17.4 (16.7 – 18.0)	–	17.0 (16.0 – 17.7)
N	204	301	95	88
Length range (median), cm	13.6 – 25.0 (21.4)	11.5 – 25.0 (19.7)	16.0 – 25.0 (19.0)	14.0 – 24.5 (19.5)

*Data from all sampled months are used for the estimation of L50

3.2. MORPHOMETRY

3.2.1. *Epinephelus aeneus* (GPW)

3.2.1.a Body shape

A total of 329 specimens of thiof *E. aeneus* were analyzed for body shape. The number and lengths of the specimens studied from each area are shown in Table 3.2.1.

Table 3.2.1.- Number of specimens of *E. aeneus* (GPW) analyzed for body shape and total length (Lt) ranges by studied area.

Area	Acronym	Number	Mean Lt (cm)	Lt range (cm)
Northern Mauritanian waters	Mau_N	44	37	28-41
Southern Mauritanian waters	Mau_S	37	9	8-9
Northern Senegal waters	Sen_N	110	30	8-40
Southern Senegal waters	Sen_S	138	30	8-40

Multivariate analysis

Principal Component Analysis (PCA)

Bartlett's sphericity was statistically significant ($p < 0.001$), and therefore the factor analysis was feasible. The results indicated that the first three factors together explained 81.01% of the total morphometric variation. Table 3.2.2 shows the proportion of the total variance explained by each factor, the morphometric distances obtained with significant loadings and the parts of the body corresponding to these distances.

Table 3.2.2.- Factors obtained by the PCA that explained most of the morphometric variation. *E. aeneus* (GPW)

FACTOR	% of total variance	Morphometric distances	Part of the body	Colour in Figure 3.2.1
1	53.08%	m5, m12, m10, m9	Central part and body depth	Red
2	17.30%	m8, m6, m1, m16	Head length	Green
3	10.63%	m3, m4	Caudal peduncle depth and lower rear length	Yellow

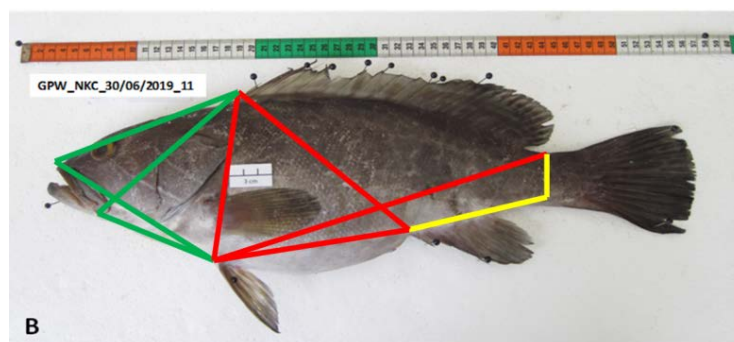


Figure 3.2.1.- Morphometric variables of *E. aeneus* (GPW) with meaningful loadings in the PCA, corresponding to the first (red), second (green) and third factor (yellow).

The scatter plot of factor 1 and factor 2 shows considerable overlap among areas (Figure 3.2.2).

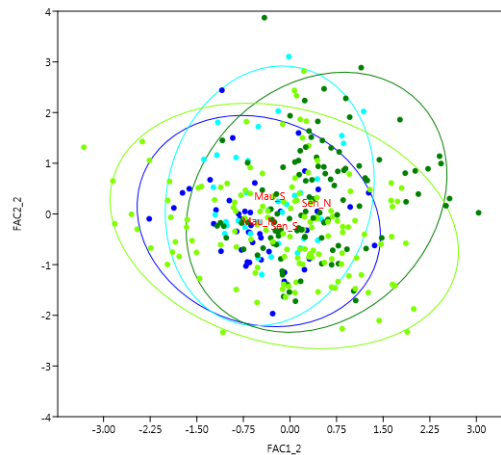


Figure 3.2.2.- Scatter plot of scores on the two factors extracted from morphometric characters of *E. aeneus* (GPW), in Mauritania and Senegal, by areas (North and South).

Linear discriminant analysis (LDA)

A univariate ANOVA showed significant differences ($P < 0.001$) in 10 of the 14 morphometric measurements of the fish sampled from the four areas (Annex 5 and Figure 3.2.3). The Wilks' lambda test of discriminant function analysis showed significant differences in morphometric measurements of all the areas ($P < 0.001$) (Annex 5).

The discriminant function analysis (DFA) showed three discriminant functions that significantly ($P < 0.001$) classify the subjects into three groups (see Table 3.2.4). Table 3.2.3 shows the proportion of the between-group variability explained by each DF, the contribution of the morphometric measurements to them (detailed in Table 3.2.4) and the part of the body corresponding to these measures.

Table 3.2.3.- Discriminant functions from DFA. *E. aeneus* (GPW)

DF	% of between-group variability	Morphometric measurements	Part of the body	Colour in Figure 3.2.3
1	68.7%	m3, m11, m9	Caudal peduncle depth, rear length and body depth	Blue
2	23.8%	m1	Upper head length	Black
3	7.5%	m7, m4	Mouth size and lower rear length	Pink



Figure 3.2.3.- Morphometric variables of *E. aeneus* (GPW) with meaningful loadings in the LDA, corresponding to the first (blue), second (black) and third factor (pink).

Table 3.2.4.- Contribution of morphometric measurements to the discriminant functions for *E. aeneus* (GPW).

	Structure Matrix		
	Function		
	1	2	3
m3	,847*	,292	-,005
m12 ^a	,554*	,154	-,036
m11	,512*	-,424	,172
m9	,459*	,375	,233
m2 ^a	,451*	-,364	,000
m14 ^a	,436*	,047	,024
m10 ^a	,363*	,303	,098
m1	-,171	,675*	,443
m16 ^a	,096	,590*	,155
m6 ^a	,086	,488*	-,042
m5 ^a	,243	,368*	-,078
m7	-,106	-,200	,768*
m8 ^a	-,090	,296	,468*
m4	,119	-,158	,177*

Pooled within-groups correlations between discriminating variables and standardized canonical discriminant functions
Variables ordered by absolute size of correlation within function.

*. Largest absolute correlation between each variable and any discriminant function

a. This variable not used in the analysis.

The LDA results show overlapping of the individuals from the four areas, with even greater closeness between those from more distant areas (northern Mauritanian and southern Senegal) (Figure 3.2.4).

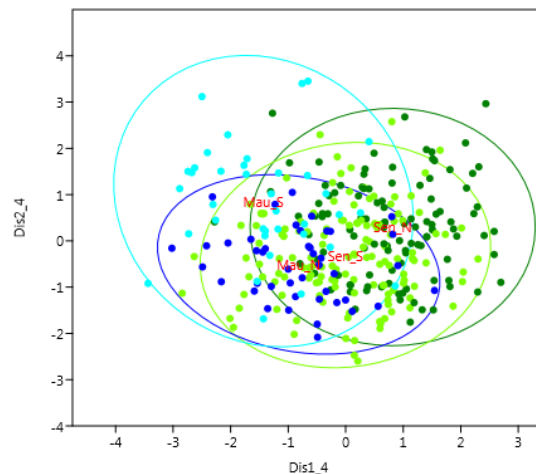


Figure 3.2.4.- Scatterplot of the first two canonical discriminant scores from the DFA for morphometric characters of *E. aeneus* (GPW) in Mauritanian and Senegal, by areas (North and South).

LDA using cross-validation classification showed 55.0% correct classification of the *E. aeneus* individuals into their original populations. The proportion of correctly classified varied between 22.7% and 62.3%, being able to be more clearly classified the *E. aeneus* individuals from Senegal North and South (60.9-62.3%) (Table 3.2.5; Figure 3.2.5).

The Senegalese individuals of *E. aeneus* as a whole (North and South), show high values of correct classification, being minimum the proportion of them belonging to Mauritania. However, Mauritanian individuals seem to show as much belonging to Mauritania as they do to Senegal (Table 3.2.5; Figure 3.2.5).

Table 3.2.5.- Classification of *E. aeneus* (GPW) individuals, in number (top) and proportion (bottom) into their original population using classification matrix (confusion matrix) of the LDA based on truss morphometry: Mauritania and Senegal, by area (North and South).

		Country area	Predicted Group Membership				Total
			Mau_N	Mau_S	Sen_N	Sen_S	
Original	Count	Mau_N	10	5	5	24	44
		Mau_S	1	18	3	15	37
		Sen_N	1	4	67	38	110
		Sen_S	12	7	33	86	138
	%	Mau_N	22.7	11.4	11.4	54.5	100
		Mau_S	2.7	48.6	8.1	40.5	100
		Sen_N	0.9	3.6	60.9	34.5	100
		Sen_S	8.7	5.1	23.9	62.3	100

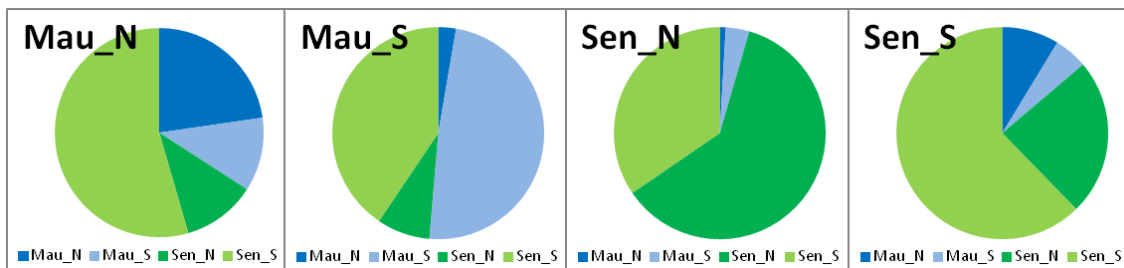


Figure 3.2.5.- Classification of *E. aeneus* (GPW) individuals (in %) into their original population using classification matrix of the LDA based on truss morphometry: Mauritania and Senegal, by area (North and South).

Multiple comparison tests using ANOVA on factor 1 (accounting the 69% of the variance), showed no significant differences ($P > 0.001$) between the individuals from North and South Mauritania. The two Senegalese areas showed close values, but significantly different from each other (Figure 3.2.6).

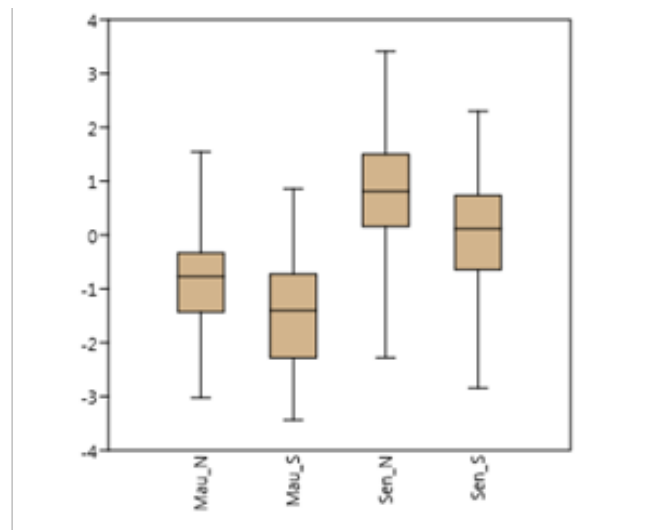


Figure 3.2.6.- Multiple comparisons of scores from factor 1 for areas obtained from the LDA based on truss morphometry of *E. aeneus* (GPW) in Mauritania and Senegal, by area (North and South).

3.2.1.b- Otolith shape

Of the 354 photographs of *E. aeneus* taken in the four areas, 230 were used for the otolith shape analysis. They correspond to the left otoliths and were correctly identified with the sampled individual.

For *E. aeneus*, 16 harmonics were used to convert the otoliths contours to Fourier coefficients (A, B, C, D), resulting in 64 Fourier coefficients for each otolith (Figure 3.2.7). To check that the choice of harmonics is correct, a random reconstruction of an *E. aeneus* otolith was performed (see example in Figure 3.2.8).

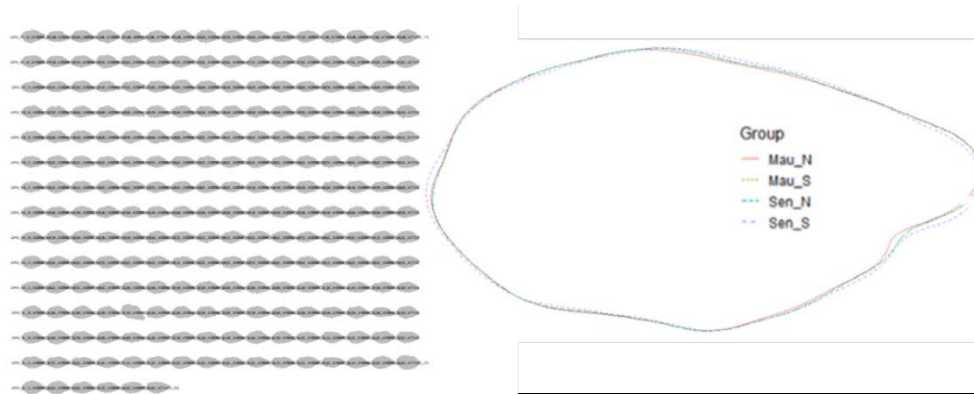


Figure 1.2.7.- Panel of otoliths shapes and mean shapes of *E.aeneus* (GPW) otoliths from Mauritania and Senegal, by area (North and South), calculated by invert transformation of Fourier coefficients.

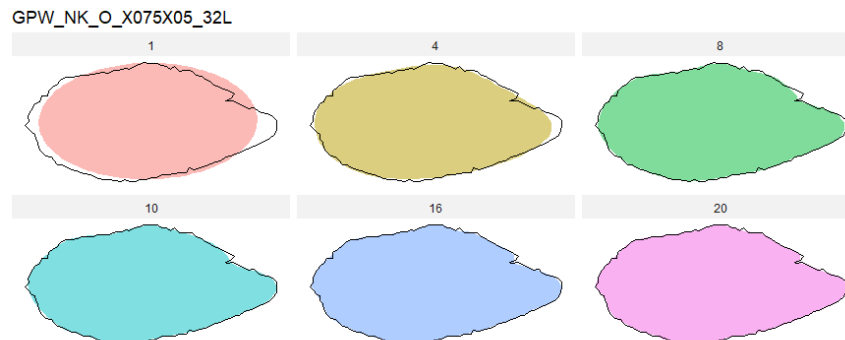


Figure 3.2.8.- Random reconstruction of *E. aeneus* (GPW) otolith. Example from Nouakchott (Mauritania) using 1, 4, 8, 10, 16 and 20 harmonics.

Multivariate analysis: Principal Component Analysis (PCA), Linear discriminant analysis (LDA) and Covariate Assisted Principal (CAP)

Multivariate analyses were performed with the obtained data from otolith shape. First, PCA and LDA were performed, followed by a CAP. In order to detect any potential possible allometric effect, the CAP1 function obtained was graphically represented in relations to the total length (TL) (Figure 3.2.9). The lack of parallelism of the graph respect to the x axis indicated an influence of the TL on the results, indicating the need to correct the allometric effect.

Figure 3.2.10 shows the results of the PCA of otolith shape for *E. aeneus* after correcting the allometric effect. There is an overlap in all the four zones of study. The two principal components

explained 55.9% of the total variance in the otolith shapes of *E. aeneus*, the first component accounting for 35.7% and the second for 20.2%.

The overlap among country-zones was also shown by the results of the LDA analysis. In this case, the DF1 explained the 66.21% of the total variance, DF2 explained 21.25%, and DF3 explained 12.53%. The confusion matrix from this analysis revealed that only the 39.3 % of otoliths were correctly classified. It should be noted that this is the lowest percentage of all the studied species.

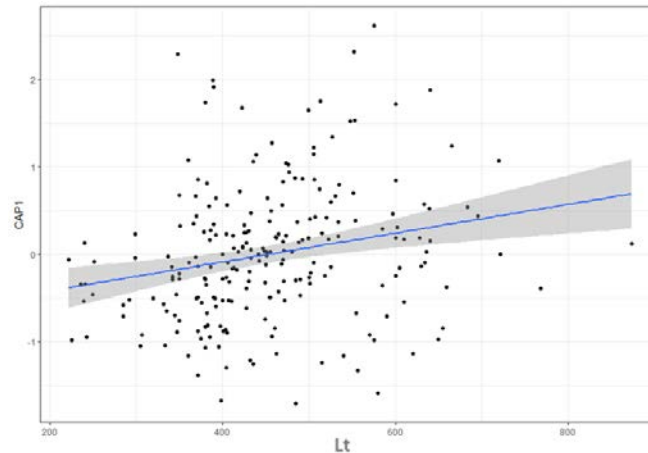


Figure 3.2.9.- Relationship between the total length and the first dimension resulting from CAP in *E. aeneus* (GPW).

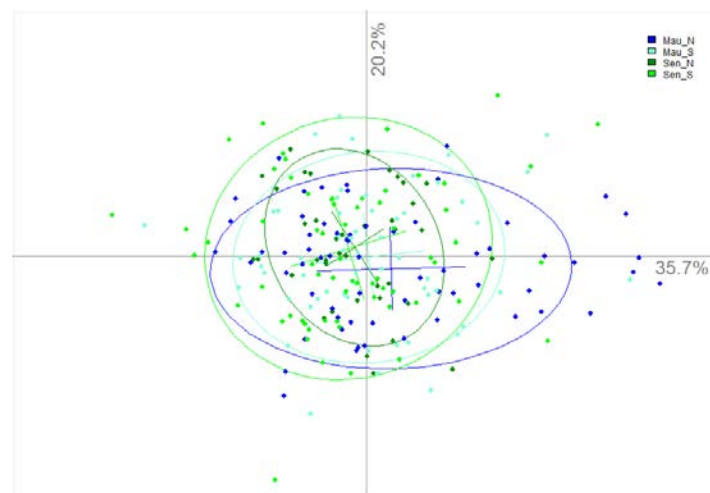


Figure 3.2.10.- Scatter plot of scores on the two principal components of PCA of *E. aeneus* (GPW) in Mauritania and Senegal, by area (North and South).

Figure 3.2.11 shows the proportions of the individual's distribution by country-areas, obtained from the confusion matrix (Table 3.2.6). A mixture of the sampled individuals from different country-areas in each studied area, according to their otolith shape can be clearly observed.

Table 3.2.6.- Classification of *E. aeneus* (GPW) individuals in number (top) and proportion (bottom) into their original population using classification matrix (confusion matrix) of the LDA based on otolith shape: Mauritania and Senegal, by area (North and South).

		Country area	Predicted Group Membership				Total
			Mau_N	Mau_S	Sen_N	Sen_S	
Original	Count	Mau_N	23	25	8	10	66
		Mau_S	21	22	10	6	59
		Sen_N	9	13	8	10	39
		Sen_S	10	9	9	38	66
	%	Mau_N	34.8	38.0	11.8	15.3	100
		Mau_S	36.3	36.6	16.4	10.5	100
		Sen_N	21.8	32.4	20.1	25.9	100
		Sen_S	15.2	13.6	13.6	57.6	100

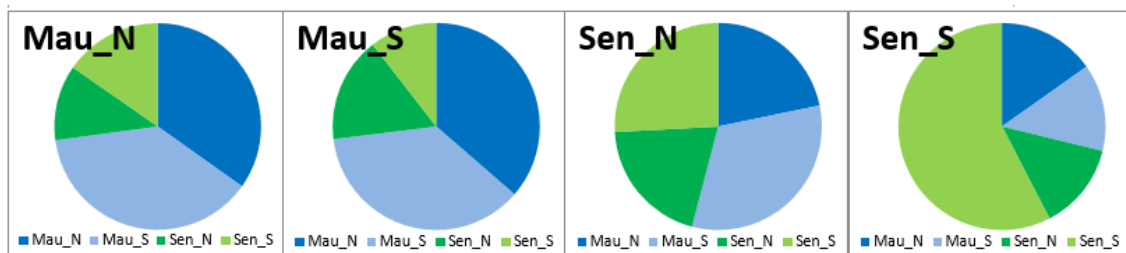


Figure 3.2.11.- Classification of *E. aeneus* (GPW) individuals (in %) into their original population using classification matrix of the LDA based on otolith shape: Mauritania and Senegal, by area (North and South).

The CAP combining elements of PCA and LDA, also shows an overlap of the four study areas. CAP1 explained the largest amount of variation in the dataset (Figure 3.2.12).

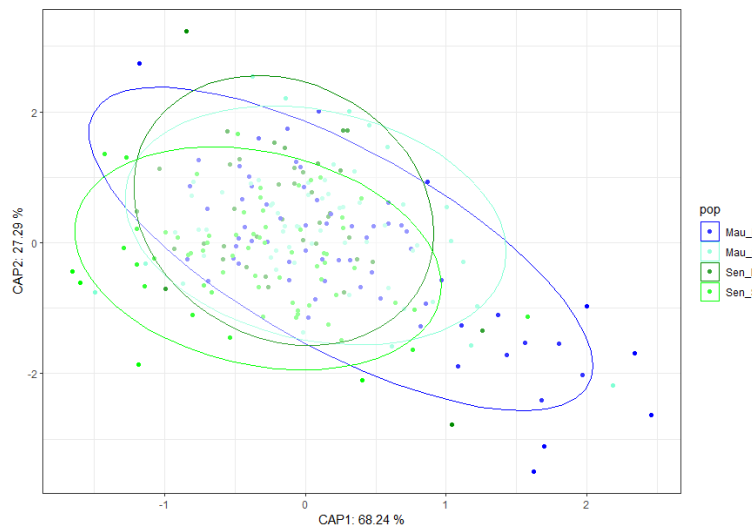


Figure 3.2.122.- Scatterplot of CAP results for *E. aeneus* (GPW) in Mauritania and Senegal, by area (North and South).

3.2.2. *Penaeus notialis* (SOP)

3.2.2.a- Body shape

Body shape analysis using truss network from 228 specimens was performed. Table 3.2.7 shows the number and lengths of the specimens studied from each area.

Table 3.2.7.- Number of specimens of *P. notialis* (SOP) analyzed and carapace (or cephalotorax) length (CL) ranges by studied area.

Area	Acronym	Number	Mean CL (mm)	CL range (mm)
Northern Mauritanian waters	Mau_N	51	40	28-58
Southern Mauritanian waters	Mau_S	35	36	28-45
Northern Senegal waters	Sen_N	39	33	22-56
Southern Senegal waters	Sen_S	31	47	27-58
Gambian waters	Cam	37	36	23-57
Northern Guinea-Bissau waters	G.Biss_N	35	33	24-49

Multivariate analysis

Principal Component Analysis (PCA)

The factor analysis was feasible as the Bartlett's sphericity was statistically significant ($p < 0.001$). The results indicated that the first three factors together explained 64.09% of the total morphometric variation. Table 3.2.8 shows the proportion of the total variance explained by each factor, number of morphometric distances with significant loadings obtained and the correspondent part of the body. The scatter plot of factor 1 and factor 2 shows considerable overlap among areas (Figure 3.2.14).

Table 3.2.8.- Factors obtained by the PCA that explained most of the morphometric variation. *P. notialis* (SOP).

FACTOR	% of total variance	Morphometric distances	Part of the body	Colour in Fig. 3.2.13
1	47.04%	m12, m17, m13, m23, m18, m22, m8, m27, m39, m36, m21, m16, m28, m50, m31, m11	depths of abdominal segments	Red
2	11.15%	m1, m46, m5, m4, m2, m3, m10, m53, m7, m33, m24, m19, m37, m14, m6	lengths and depths of the carapace and telson length	Green
3	5.89%	m48, m47, m34, m41, m32, m42, m44, m26, m29, m52, m38, m43, m40, m45	lengths of rostrum and of abdominal segment	Yellow

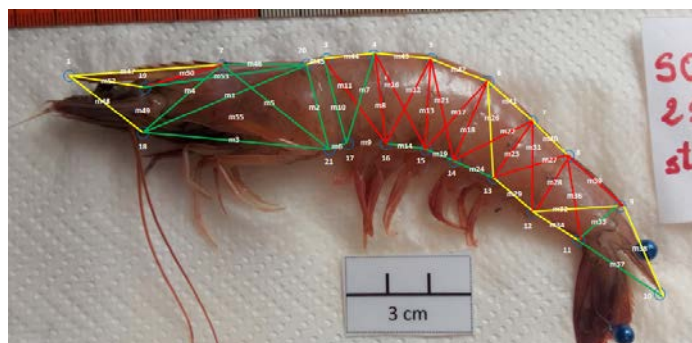


Figure 3.2.13.- Morphometric variables of *P. notialis* (SOP) with meaningful loadings in the PCA, corresponding to the first (red), second (green) and third factor (yellow).

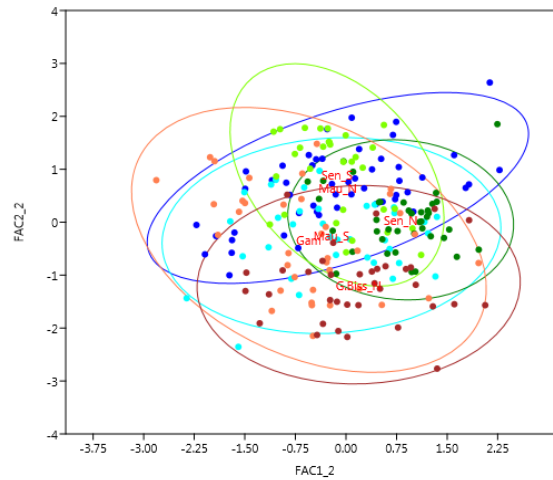


Figure 3.2.14.- Scatter plot of scores on the two factors extracted from morphometric characters of *P. notialis* (SOP) in Mauritania, Senegal, Gambia and Guinea-Bissau, by areas.

Linear discriminant analysis (LDA)

A univariate ANOVA showed significant differences ($P < 0.001$) in 45 of the 47 morphometric measurements of the shrimp sampled from the six areas (Annex 5; Figure 3.2.15). The Wilks' lambda test of discriminant function analysis showed significant differences in morphometric measurements of all the areas ($P < 0.001$) (Annex 5).

The discriminant function analysis showed three discriminant functions (DF) that allowed to significantly ($P < 0.001$) classify the subjects into five groups. Table 3.2.9 shows the morphometric measurements contributing to each DF, being all of them the most relevant for population discrimination in *P. notialis* (Table 3.2.10), and the part of the body corresponding to each DF (Figure 3.2.15).

Table 3.2.9.- Discriminant functions from DFA. *P. notialis* (SOP)

DF	% of between-group variability	Morphometric measurements	Part of the body	Colour in Fig. 3.2.15
1	37.6%,	m34, m23, m6, m28	Length and depth of the 5 th and 6 th abdominal segments	Blue
2	30.9%,	m1,m19, m47, m32, m9, m33, m2	Length and depth of carapace and rostrum, lower length of 1 st and 3 rd abdominal segments and depth of the 6 th abdominal segment	Black
3	14.6%	m45, m40, m49	Depth of the rostrum area	Pink
4	6.3%	–	–	–
5	10.5%	m41, m43, m44	Upper length of 1 st , 2 nd and 4th abdominal segments	Green

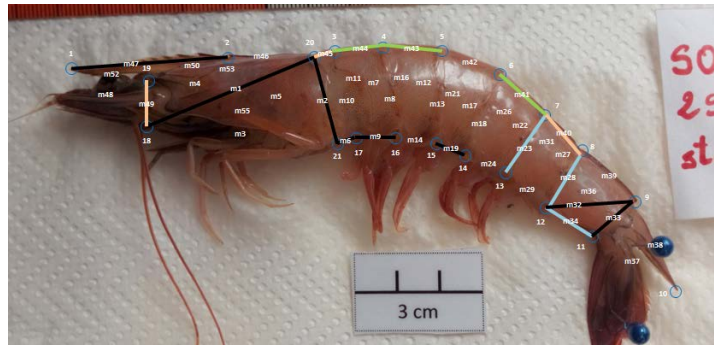


Figure 3.2.15.- Morphometric variables of *P. notialis* (SOP) with meaningful loadings in the LDA, corresponding to the first (blue), second (black), third (pink) and fifth factor (green).

Table 3.2.10.- Contribution of morphometric measurements to the discriminant functions for *P. notialis* (SOP).

	Structure Matrix				
	Function				
	1	2	3	4	5
m26 ^a	.443 [*]	.144	-.042	-.165	.410
m52 ^a	.398 [*]	.320	-.230	-.038	.147
m34	.397 [*]	.352	.162	-.393	.112
m8 ^a	.377 [*]	.238	-.037	-.096	.177
m21 ^a	.367 [*]	.286	.007	-.219	.264
m36 ^a	.358 [*]	.221	.224	-.091	.118
m31 ^a	.353 [*]	.195	-.057	.049	.233
m23	.350 [*]	.102	.089	.074	.200
m16 ^a	.323 [*]	.286	-.004	-.182	.298
m6	.302 [*]	.298	-.071	.194	.012
m28	.294 [*]	.241	.139	.156	.252
m22 ^a	.294 [*]	.167	.134	-.215	.226
m18 ^a	.281 [*]	.118	.051	-.249	.216
m12 ^a	.273 [*]	.062	.084	-.070	.138
m13 ^a	.259 [*]	.129	.075	-.133	.189
m27 ^a	.242 [*]	.184	.106	-.121	.225
m39 ^a	.222 [*]	.119	.166	-.184	.137
m1	.158	.476 [*]	-.278	.421	.246
m19	.021	.470 [*]	-.056	.053	.401
m47	.409	.426 [*]	-.360	-.293	-.043
m11 ^a	.276	.414 [*]	-.105	-.171	.302
m4 ^a	.102	.413 [*]	-.333	.236	.099
m32	.267	.407 [*]	.160	-.385	.228
m9	-.150	.400 [*]	.000	.054	.312
m10 ^a	.219	.396 [*]	-.122	.078	.317
m7 ^a	.207	.396 [*]	-.070	-.046	.360
m33	.046	.355 [*]	.152	.191	.293
m3 ^a	.088	.343 [*]	-.145	.293	.262
m2	.003	.340 [*]	-.145	.095	.082
m38 ^a	.207	.332 [*]	-.033	-.134	.221
m53 ^a	.008	.308 [*]	-.229	.302	.119
m42 ^a	.269	.298 [*]	-.012	-.248	.132
m45	.233	.175	-.320 [*]	.312	.188
m40	.228	.266	-.308 [*]	-.204	.186
m50 ^a	-.002	.019	-.214 [*]	-.037	-.096
m49	.015	-.134	-.150 [*]	-.009	.104
m46 ^a	.096	.337	-.182	.408 [*]	.378
m48 ^a	.307	.127	-.148	-.351 [*]	.011
m17 ^a	.165	.158	.099	-.219 [*]	.155
m41	.314	.099	-.094	-.190	.544 [*]
m43	.186	.214	-.075	-.233	.525 [*]
m44	.212	.375	-.263	-.280	.500 [*]
m29 ^a	.139	.189	-.092	-.158	.323 [*]
m5 ^a	.027	.295	-.077	.223	.301 [*]
m37 ^a	.086	.255	-.104	.016	.299 [*]
m14 ^a	.083	.211	-.046	.115	.263 [*]
m24 ^a	.226	.243	-.093	.050	.260 [*]

Pooled within-groups correlations between discriminating variables and standardized canonical discriminant functions. Variables ordered by absolute size of correlation within function.

a. This variable not used in the analysis.

*. Largest absolute correlation between each variable and any discriminant function

These five groups classified by the LDA show greater overlap between the two Mauritanian areas (North and South), with individuals from the other areas being differentiated from Mauritania and also from each other. Individuals from Guinea-Bissau show the greatest distance from Mauritania, which are the most distant areas in latitude. It should be noted that Gambian individuals also show differentiation with respect to those from the two areas of Senegal, despite being geographically located between them (Figure 3.2.16).

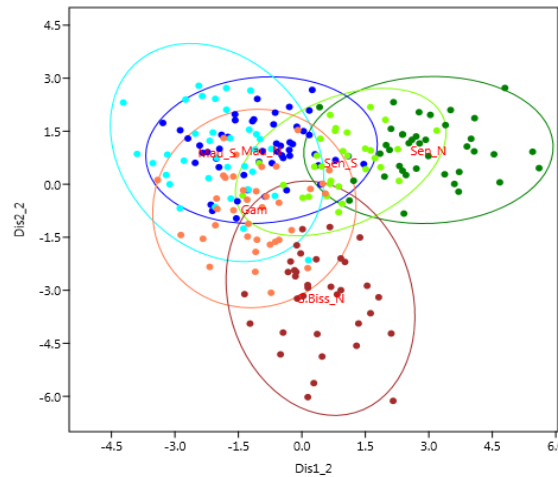


Figure 3.2.16.- Scatterplot of the first two canonical discriminant scores from the DFA for morphometric characters of *P. notialis* (SOP) in Mauritania, Senegal, Gambia and Guinea-Bissau, by areas.

LDA using cross-validation classification showed 88.2% correct classification of the *P. notialis* individuals into their original populations. The proportion of correctly classified varied between 80.4% and 97.1%. The individuals from northern Senegal and northern Guinea-Bissau waters were the most clearly classified (94.9% and 97.1%, respectively) (Table 3.2.11; Figure 3.2.17). Individuals of *P. notialis* from all areas studied show a high percentage of correct classification. Therefore, six independent groups corresponding to each studied area could be considered. As an alternative option to consider a more correct classification, four groups corresponding to each of the countries studied could be also established: Mauritanian waters (N and S); Senegal waters (N and S); Gambian waters; northern Guinea-Bissau waters.

Table 3.2.11.- Classification of *P. notialis* (SOP) individuals (in number and %) into their original population using classification matrix of the LDA based on truss morphometry. Mauritania, Senegal, Gambia and Guinea-Bissau, by areas.

		Country area	Predicted Group Membership					Total	
			Mau_N	Mau_S	Sen_N	Gam	Sen_S		G.Biss_N
Original	Number	Mau_N	41	3	0	5	2	0	51
		Mau_S	2	31	0	1	0	1	35
		Sen_N	1	0	37	1	0	0	39
		Gam	2	0	0	33	1	1	37
		Sen_S	0	0	6	0	25	0	31
		G.Biss_N	0	0	0	1	0	34	35
	%	Mau_N	80,4	5,9	0	9,8	3,9	0	100
		Mau_S	5,7	88,6	0	2,9	0	2,9	100
		Sen_N	2,6	0	94,9	2,6	0	0	100
		Gam	5,4	0	0	89,2	2,7	2,7	100
		Sen_S	0	0	19,4	0	80,6	0	100
		G.Biss_N	0	0	0	2,9	0	97,1	100

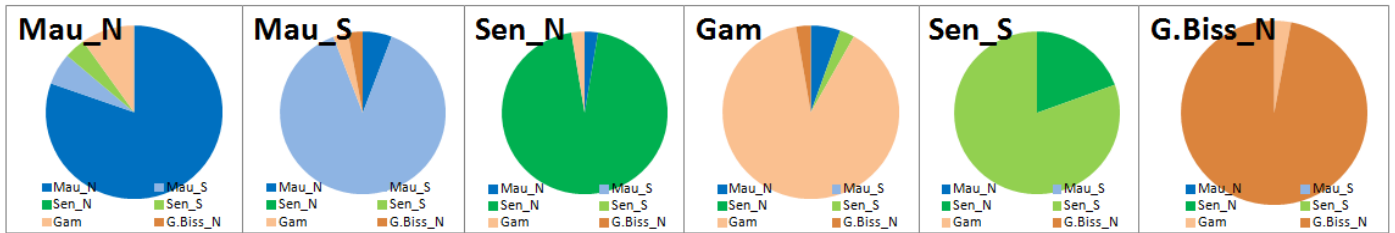


Figure 3.2.17.- Classification of *P. notialis* (SOP) individuals (in %) into their original population using classification matrix of the LDA based on truss morphometry: Mauritania, Senegal, Gambia and Guinea-Bissau, by areas.

Multiple comparison tests using ANOVA on factor 1 (accounting the 38% of the variance), showed significant differences ($P \leq 0.001$) among the individuals from all areas except between those from Mauritania North and Gambia, and between those from Senegal South and Guinea-Bissau (Figure 3.2.18). The factor 2, accounting 31% of the variance, showed no significant differences between the individuals from the four northern areas Mauritania (North first four areas studied of Mauritanian and Senegal waters), but significant differences ($P \leq 0.001$) respect to those from Gambian and Guinea-Bissau waters were found (Figure 3.2.18).

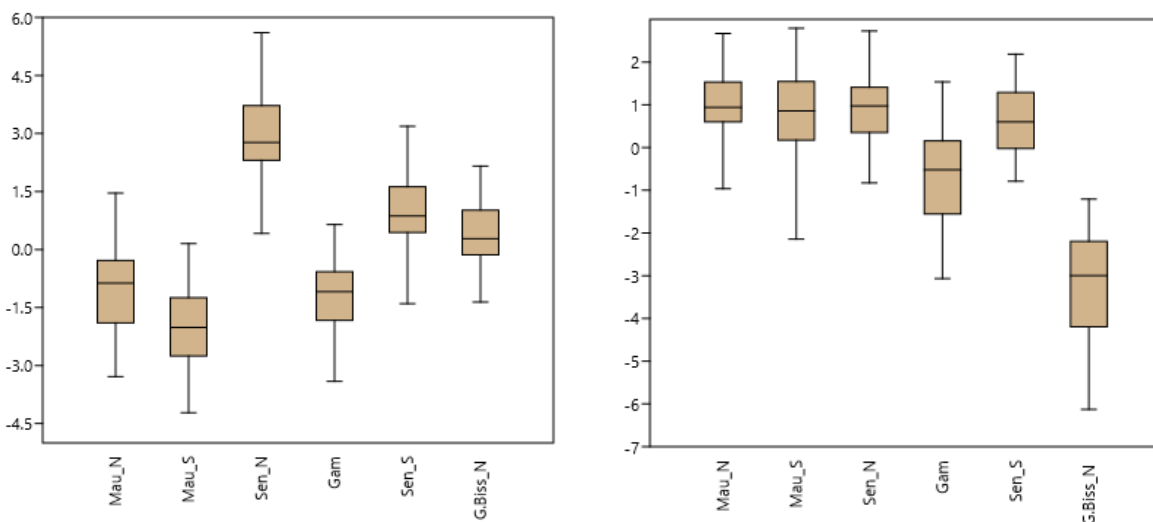


Figure 3.2.18.- Multiple comparison of scores from factor 1 (left) and factor 2 (right) for areas obtained from the LDA based on truss morphometry of *P. notialis* (SOP), in Mauritania, Senegal, Gambia and Guinea-Bissau, by areas.

3.2.3. *Pagrus caeruleostictus* (BSC)

3.2.3.a- Body shape

Body shape analysis using truss network from 249 specimens was performed. The number and lengths of the specimens collected from each area are shown in Table 3.2.12.

Table 3.2.12.- Number of specimens of *P. caeruleostictus* (BSC) analyzed and total length (Lt) ranges by studied area.

Area	Acronym	Number	Mean Lt (cm)	Lt range (cm)
Southern Guinea-Bissau waters	G.Biss_S	86	19	14-28
Northern Guinea waters	Gui_N	89	24	15-29
Southern Guinea waters	Gui_S	74	19	14-29

Multivariate analysis

Principal Component Analysis (PCA)

Bartlett's sphericity was statistically significant ($p < 0.001$), and therefore the factor analysis was feasible. The results of factor analysis indicated that the first three factors together explained 80.63% of the total morphometric variation. Table 3.2.13 shows the proportion of the total variance explained by each factor, number of morphometric distances obtained with significant loadings and the correspondent part of the body.

Table 3.2.13- Factors obtained by the PCA that explained most of the morphometric variation. *P. caeruleostictus* (BSC).

FACTOR	% of total variance	Morphometric distances	Part of the body	Colour in Fig. 3.2.19
1	43.63%	m11, m4, m12, m3	Lower and rear part of the fish body	Red
2	25.94%	m1, m16, m8, m6, m9	Head length and depth	Green
3	11.06%	m10, m2 and m14	Straight and oblique upper measurements of the fish body	Yellow

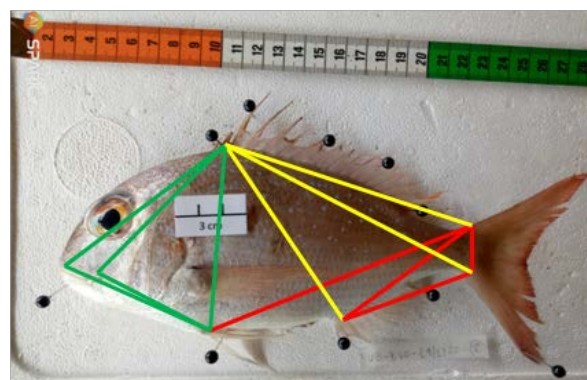


Figure 3.2.19.- Morphometric variables of *P. caeruleostictus* (BSC) with meaningful loadings in the PCA, corresponding to the first (red), second (green) and third factor (yellow).

The scatter plot of factor 1 and factor 2 shows overlap, being northern and southern Guinea more closely overlapped (3.2.20).

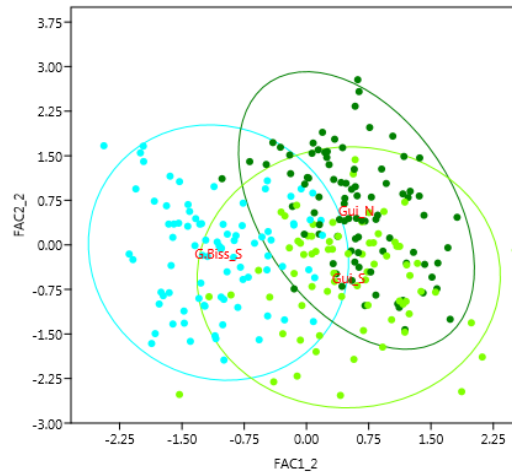


Figure 3.2.20. Scatter plot of scores on the two factors extracted from morphometric characters of *P. caeruleostictus* (BSC): Guinea-Bissau South and Guinea (North and South).

Linear discriminant analysis (LDA)

A univariate ANOVA showed significant differences ($P < 0.001$) in 11 of the 14 morphometric measurements of the fish sampled from three areas (Annex 5; Figure 3.2.21). The Wilks' lambda test of discriminant function analysis showed significant differences in morphometric measurements of all the areas ($P < 0.001$) (Annex 5).

The discriminant function analysis showed three discriminant functions (DF) that allowed to significantly ($P < 0.001$) classify the subjects into two groups (see Table 3.2.15), differentiated by a number of morphometric measures, which define different parts of the body. The contribution of the morphometric measurements to the discriminant functions is showed in Table 3.2.14. These measurements are the most relevant for population discrimination in *P. caeruleostictus* (Table 3.2.14; Figure 3.2.21).

Table 3.2.14.- Discriminant functions from DFA. *P. caeruleostictus* (BSC).

DF	% of between-group variability	Morphometric measurements	Part of the body	Colour in Fig. 3.2.21
1	88.3%	m3, m4, m12, m5, m10	Lower and rear lengths of the fish body	Blue
2	11.7%	m7, m16, m9	Head and mouth size and body depth	Black

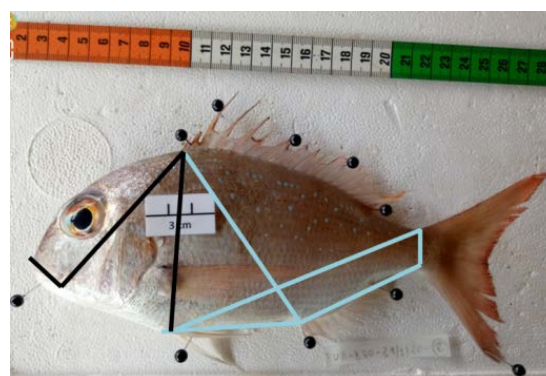


Figure 3.2.21.- Morphometric variables of *P. caeruleostictus* (BSC) with meaningful loadings in the LDA, corresponding to the first (blue) and second (black).

Table 3.2.15.- Contribution of morphometric measurements to the discriminant functions for *P. caeruleostictus* (BSC).

	Structure Matrix	
	Function	
	1	2
m3	.638*	-.148
m4	.575*	.413
m11 ^a	.570*	.391
m12	.488*	.415
m14 ^a	.377*	.325
m5	.118*	-.109
m10	.064*	.056
m1 ^a	.269	-.647*
m8 ^a	-.042	-.601*
m7	.089	-.559*
m16	.326	-.530*
m2 ^a	.218	.332*
m6 ^a	.009	-.328*
m9	.077	-.189*

Pooled within-groups correlations between discriminating variables and standardized canonical discriminant functions
Variables ordered by absolute size of correlation within function.

*. Largest absolute correlation between each variable and any discriminant function

a. This variable not used in the analysis.

The DFA show results represented in scatterplot show two groups, one with closeness and overlapping individuals, from the two Guinean areas, and another one with those individuals from Guinea-Bissau South (Figure 3.2.22).

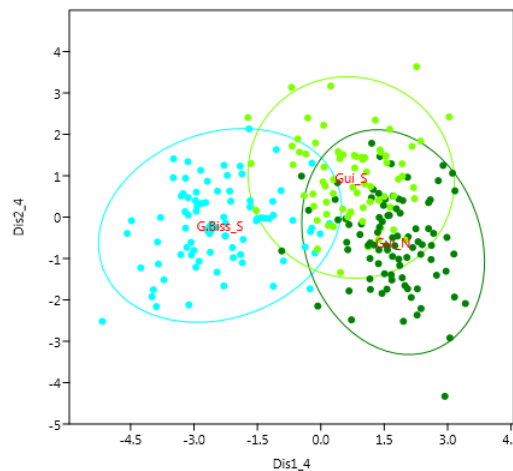


Figure 3.2.22.- Scatterplot of the first two canonical discriminant scores from the discriminant function analysis (DFA) for morphometric characters of *P. caeruleostictus* (BSC). Guinea-Bissau South and Guinea (North and South).

LDA using cross-validation classification showed 84.7% correct classification of the *P. caeruleostictus* individuals into their original populations. The proportion of correctly classified varied between 79.7% and 89.5%, being able to be more clearly classified the *P. caeruleostictus* individuals from Guinea-Bissau South (Table 3.2.16; Figure 3.2.23).

The Guinea individuals of *P. caeruleostictus* as a whole (North and South), show high values of correct classification, being minimum the proportion of them belonging to Guinea-Bissau (Figure 3.2.23). Therefore, the cross-validation classification shows at least two groups: on the one hand, Guinea-Bissau individuals of *P. caeruleostictus* and, on the other hand, Guinea individuals (North and South). In addition, the samples from North and South Guinea could also be considered as differentiated from each other with a correct classification of around 80%, each one.

Table 3.2.16.- Classification of *P. caeruleostictus* (BSC) individuals (in number and %) into their original population using classification matrix of the LDA based on truss morphometry. Guinea-Bissau South and Guinea (North and South).

		Country area	Predicted Group Membership			Total
			G.Biss_S	Gui_N	Gui_S	
Original	Count	G.Biss_S	77	3	6	86
		Gui_N	1	75	13	96
		Gui_S	3	12	59	74
	%	G.Biss_S	89.5	3.5	7.0	100
		Gui_N	1.1	84.3	14.6	100
		Gui_S	4.1	16.2	79.7	100

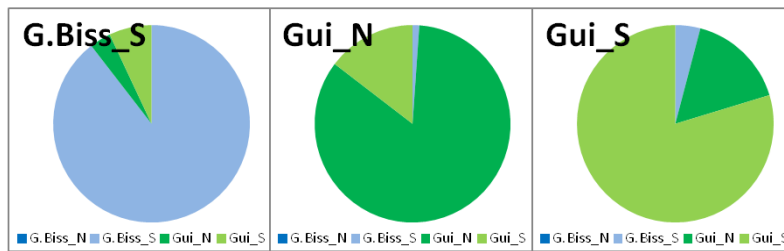


Figure 3.2.23.- Classification of *P. caeruleostictus* (BSC) individuals (in %) into their original population using classification matrix of the LDA based on truss morphometry. Guinea-Bissau South and Guinea (North and South).

Multiple comparison tests using ANOVA on factor 1 (accounting the 88% of the variance), showed significant differences ($P \leq 0.001$) among the individuals from the three areas, although closer values between individuals from the both northern and southern areas of Guinea are observed on the one hand, and the individuals of Guinea-Bissau, on the other hand (Figure 3.2.24).

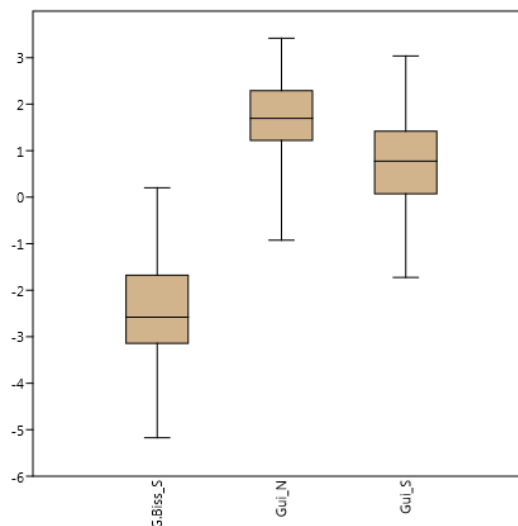


Figure 3.2.24.- Multiple comparison of scores from factor 1 for areas obtained from the LDA based on truss morphometry of *P. caeruleostictus* (BSC). Box plots with same superscript in Guinea-Bissau South and Guinea (North and South).

3.2.3.b Otolith shape

A total of 338 otoliths of *P. caeruleostictus* of 369 photographed otoliths from Guinea-Bissau South and Guinea North and South were analysed for otolith shape.

To properly summarize otolith shapes, 99% of the cumulative Fourier power required the use of 17 harmonics resulting in 68 Fourier coefficients for each otolith, being 65 Fourier coefficients after normalization (Figure 3.2.25).

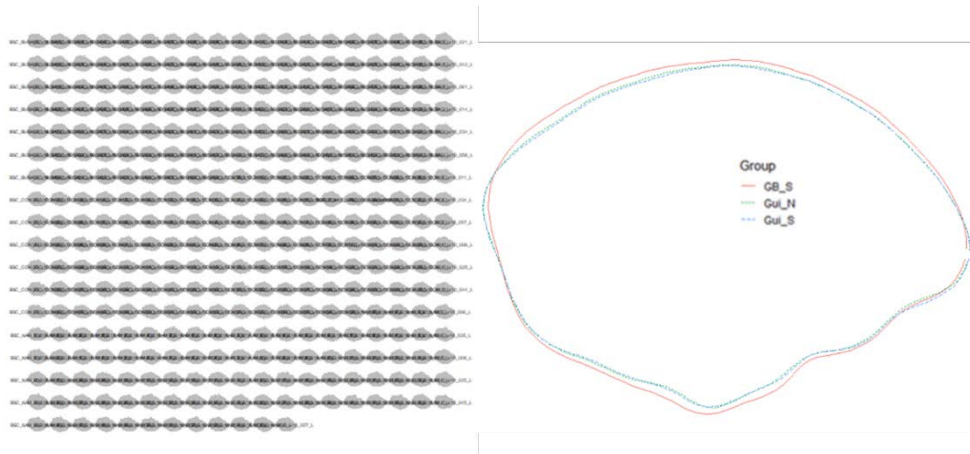


Figure 3.2.25.- Panel of otoliths shapes and mean shapes of *P. caeruleostictus* (BSC) otoliths from Guinea-Bissau South and Guinea (North and South).

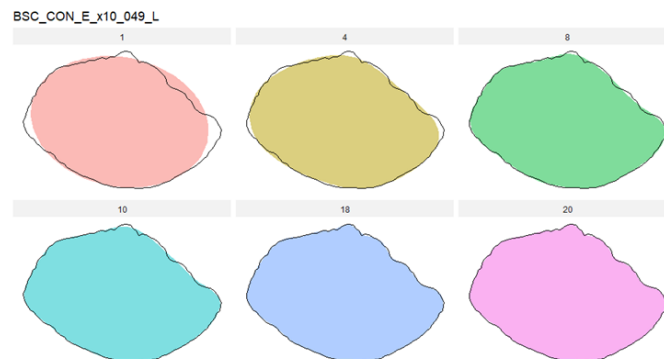


Figure 3.2.26.- Random reconstruction of *P. caeruleostictus* (BSC) otoliths. Example from Conakry (Guinea) using 1, 4, 8, 10, 18 and 20 harmonics.

Multivariate analysis: Principal Component Analysis (PCA), Linear discriminant analysis (LDA) and Covariate Assisted Principal (CAP)

Once the otolith contours were obtained and after their transformation to Fourier coefficients, the multivariate analyses of PCA, LDA and CAP were performed. However, the graphical representations of the CAP1 and total length showed an influence of some of the descriptors used in multivariate analysis by length of fish (Figure 3.2.27).

After the elimination of allometric effect through an ANCOVA test, all statistical analysis was repeated.

PCA results showed a clear overlap in the three studied areas, especially in the two areas of Guinea (North and South). Likewise, the two principal components explained 42.70 % of the total variance in the otolith shapes: the first component accounted for 27.5% and the second component for 15.2% (Figure 3.2.28).

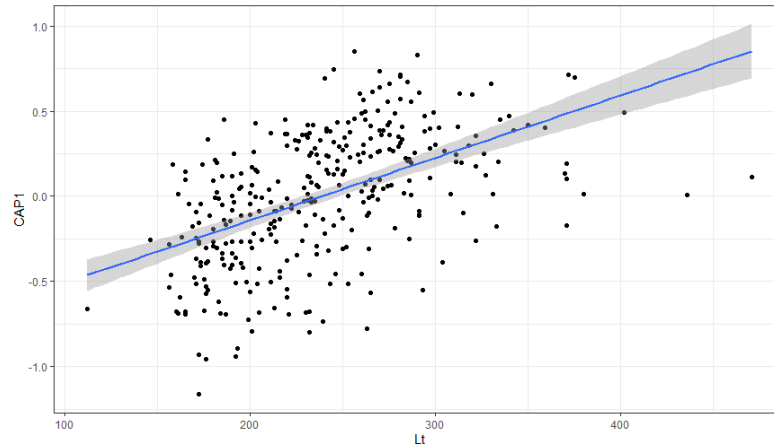


Figure 3.2.27.- Relationship between the total length variable and the first CAP dimension resulting from CAP for *P. caeruleostictus* (BSC).

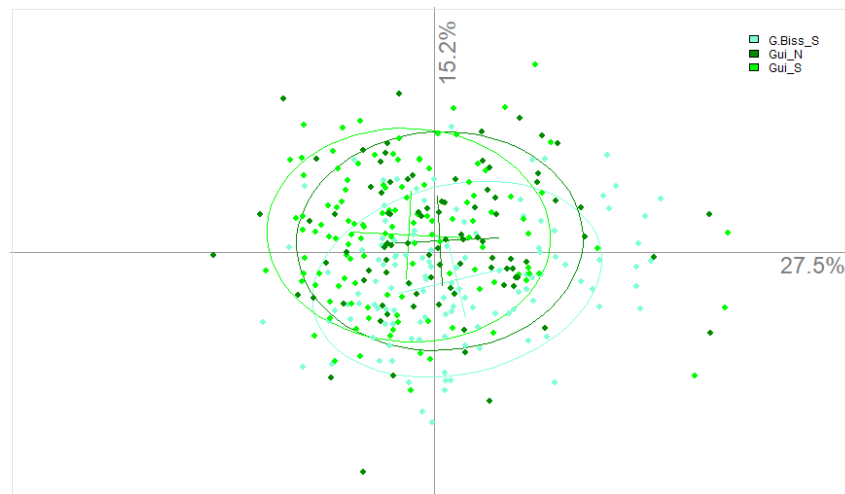


Figure 3.2.28.- Scatter plot of scores on the two principal components of PCA of *P. caeruleostictus* (BSC) in Guinea-Bissau South and Guinea (North and South).

The different contributions of the two discriminant functions of the LDA were 77.32% and 22.68%, respectively. Results obtained from the LDA, presented in a confusion matrix, showed a mixture of the three studied areas, with an average percentage of correct classification of 60.30%. The values of correct classification ranged between 44.1% in North Guinea and 72.4% in South Guinea-Bissau (Table 3.2.17 and Figure 3.2.29).

Table 1.2.17.- Classification of *P. caeruleostictus* (BSC) individuals (in number and %) into their original population using classification matrix (confusion matrix) of the LDA based on otolith shape. Guinea-Bissau South and Guinea (North and South).

		Country area	Predicted Group Membership			Total
			G.Biss_S	Gui_N	Gui_S	
Original	Count	G.Biss_S	89	14	20	123
		Gui_N	17	41	35	92
		Gui_S	18	29	72	120
	%	G.Biss_S	72.4	11.4	16.1	100
		Gui_N	18.2	44.1	37.9	100
		Gui_S	15.1	24.6	60.3	100

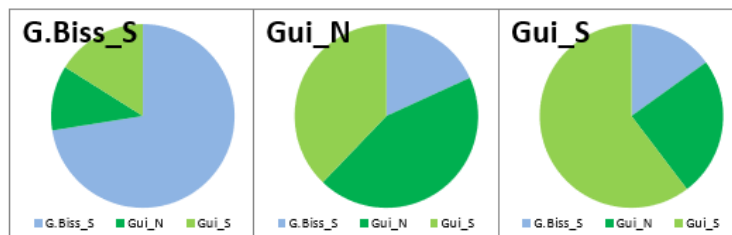


Figure 3.2.29.- Classification of *P. caeruleostictus* (BSC) individuals (in %) into their original population using classification matrix of the LDA based on otolith shape. Guinea-Bissau South and Guinea (North and South).

The CAP analysis shows an overlap of the three studied areas with some differentiation between countries (Figure 3.2.30).

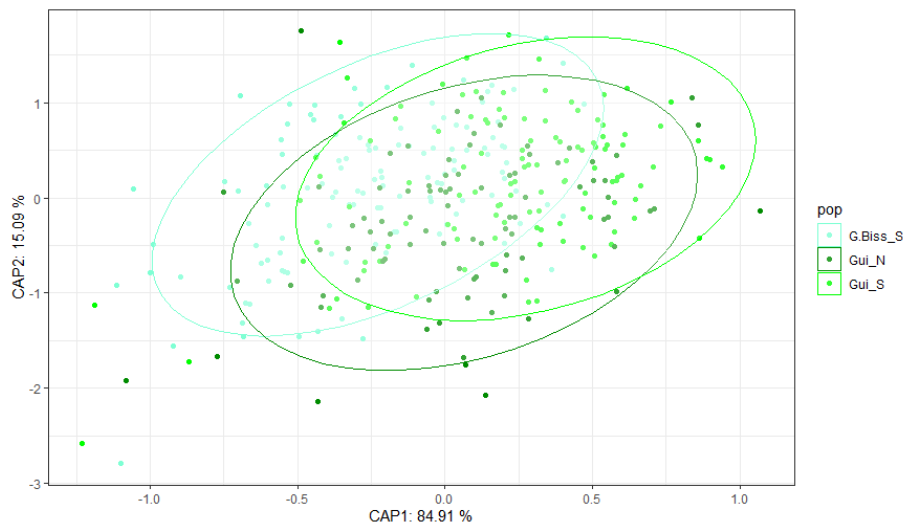


Figure 3.2.30.- Scatterplot of CAP results for *P. caeruleostictus* (BSC) in Guinea-Bissau South and Guinea (North and South).

3.2.4. *Pseudotolithus elongatus*

3.2.4.a- Body shape

Body shape analysis using truss network from 242 specimens was performed. The number and lengths of the specimens collected from each area are shown in Table 3.2.18.

Table 3.2.18.- Number of specimens of *P. elongatus* (PSE) analyzed and total length (Lt) ranges by studied area.

Area	Acronym	Number	Mean Lt (cm)	Lt range (cm)
Northern Guinea-Bissau waters	G.Biss_N	58	22	18-28
Southern Guinea-Bissau waters	G.Biss_S	58	25	17-33
Northern Guinea waters	Gui_N	63	24	17-33
Southern Guinea waters	Gui_S	63	23	19-31

Multivariate analysis

Principal Component Analysis (PCA)

A factor analysis was feasible as the Bartlett's sphericity was statistically significant ($p < 0.001$). The results of factor analysis indicated that the first three factors together explained 71.07% of the total morphometric variation. The variance explained by each factor, together with the morphometric distances with significant loadings and the corresponding parts of the body are shown in Table 3.2.19 and Figure 3.2.31.

Table 3.2.19.- Factors obtained by the PCA that explained most of the morphometric variation. *P. elongatus* (PSE).

FACTOR	% of total variance	Morphometric distances	Part of the body	Colour in Fig. 3.2.31
1	44.66%	m17, m2, m10, m19, m11, m6, m1	Straight and oblique measurements and depth of the central part of the body and head	Red
2	16.85 %	m13, m3, m15, m4, m12	Lengths and depth of the rear part of the body	Green
3	9.56%	m9, m7	Two lower head length measurements	Yellow

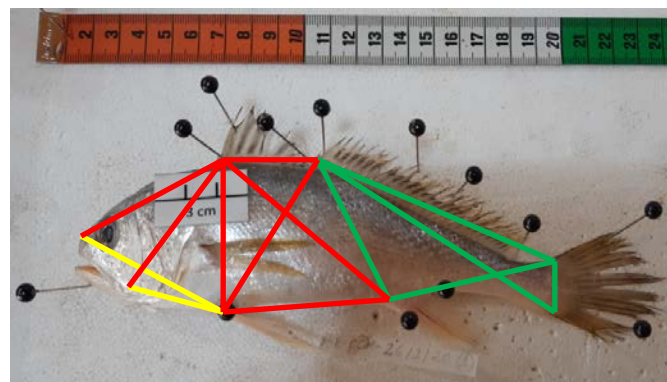


Figure 3.2.31.- Morphometric variables of *P. elongatus* (PSE) with meaningful loadings in the PCA, corresponding to the first (red), second (green) and third factor (yellow).

The scatter plot of factor 1 and factor 2 shows overlap, being the two areas of Guinea (North and South) more closely overlapping (Figure 3.2.32).

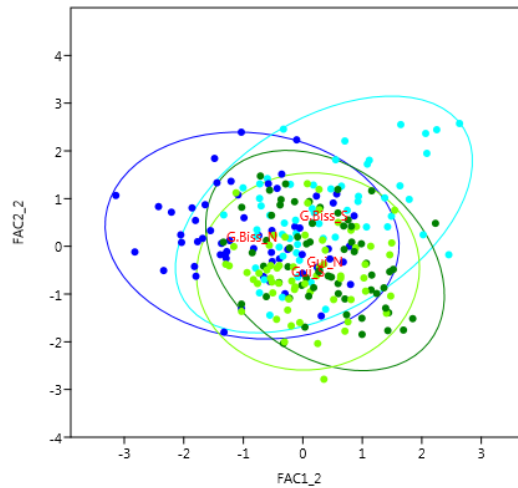


Figure 3.2.32.- Scatter plot of scores on the two factors extracted from morphometric characters of *P. elongatus* (PSE): Guinea-Bissau and Guinea, by areas (North and South).

Linear discriminant analysis (LDA)

A univariate ANOVA showed significant differences ($P < 0.001$) in 13 of the 16 morphometric measurements of the fish sampled from four areas (Annex 5; Figure 2). The Wilks' lambda test of discriminant function analysis showed significant differences in morphometric measurements of all the areas ($P < 0.001$) (Annex 5).

The discriminant function analysis showed three discriminant functions (DF) that allowed to significantly ($P < 0.001$) classify the subjects into three groups. Table 3.2.20 shows the three DF obtained by the DFA, with contribution of the morphometric measurements to each one (Table 3.2.21), being the most relevant for population discrimination in *P. elongatus*. The part of the body of each DF is also indicated Table 3.2.20 and Figure 3.2.33.

Table 3.2.20.- Discriminant functions from DFA. *P. elongatus* (PSE).

DF	% of between-group variability	Morphometric measurements	Part of the body	Colour in Fig.3.2.23
1	72.7%	m6	Central length of the lower part of the body	Blue
2	16.1%	m12, m1, m8	Head and mouth lengths and central depth of the body	Black
3	11.1%	m17, m15, m4	Central depth, rear length and caudal peduncle depth	Pink

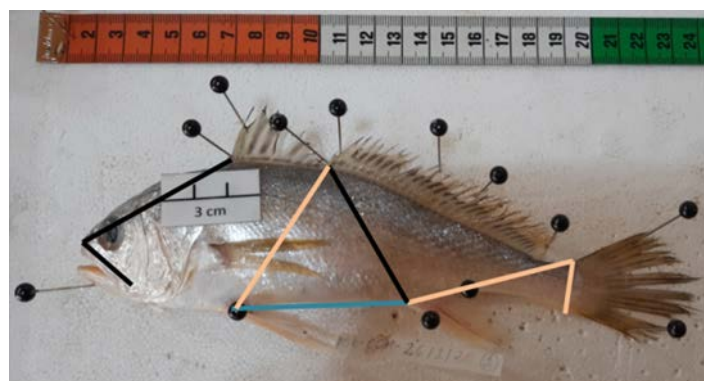


Figure 3.2.33.- Morphometric variables of *P. elongatus* (PSE) with meaningful loadings in the LDA, corresponding to the first (blue), second (black) and third factor (pink).

Table 3.2.21.- Contribution of morphometric measurements to the discriminant functions for *P. elongatus* (PSE).

	Structure Matrix		
	1	2	3
m6	,363*	,280	,261
m12	-,167	,865*	,005
m13 ^a	-,354	,676*	,118
m3 ^a	-,231	,626*	,076
m11 ^a	-,075	,625*	,174
m1	,159	,467*	,275
m9 ^a	-,154	,414*	,154
m7 ^a	,000	,355*	,076
m8	-,259	,288*	,284
m17	,195	,282	,622*
m15	-,058	,150	,538*
m4	-,487	,208	,503*
m10 ^a	-,019	,438	,474*
m5 ^a	-,086	,146	,434*
m2 ^a	-,003	,275	,408*

Pooled within-groups correlations between discriminating variables and standardized canonical discriminant functions
 Variables ordered by absolute size of correlation within function.

*. Largest absolute correlation between each variable and any discriminant function

a. This variable not used in the analysis.

Among the three groups discriminated by the DFA, individuals from the two areas in Guinea-Bissau (North and South) show more closeness and overlapping than those from the two Guinean areas (Figure 3.2.34).

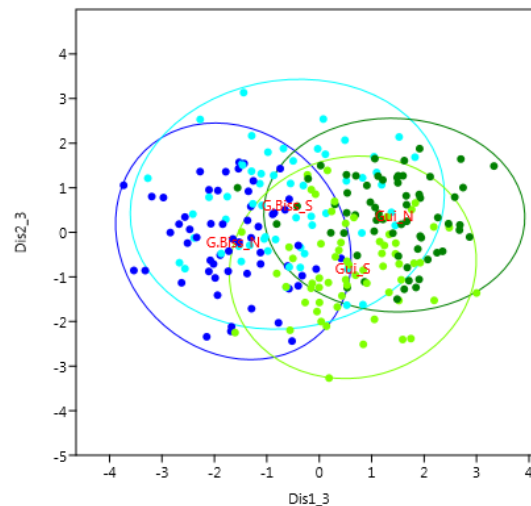


Figure 3.2.34.- Scatterplot of the first two canonical discriminant scores from the discriminant function analysis (DFA) for morphometric characters of *P. elongatus* (PSE). Guinea-Bissau and Guinea, by area (North and South).

LDA using cross-validation classification showed 68.2% correct classification of the *P. elongatus* individuals into their original populations. The proportion of correctly classified varied between 56.9% and 74.1%, being able to be more clearly classified (68.3-74.1%) the *P. elongatus* individuals from North Guinea-Bissau (74.1%), North Guinea (73%) and South Guinea (68.3%) than those of South Guinea-Bissau (56.9 %) (Table 3.2.22; Figure 3.2.35).

The Guinea-Bissau individuals of *P. elongatus* as a whole (North and South), show high values of correct classification, being limited the proportion of them belonging to Guinea. On the other hand, Guinea individuals (together North and South), are also highly classified as individuals from Guinea, with a limited proportion of them belonging to Guinea-Bissau (Table 3.2.22; Figure 3.2.35). Therefore, the cross-validation classification shows two groups of *P. elongatus*: on the one hand,

Guinea-Bissau individuals (North and South) and on the other hand, Guinea individuals (North and South).

Table 3.2.22.- Classification of *P. elongatus* (PSE) individuals (in number and %) into their original population using classification matrix of the LDA based on truss morphometry: Guinea-Bissau and Guinea, by area (North and South).

	Country area	Predicted Group Membership				Total	
		G.Biss_N	G.Biss_S	Gui_N	Gui_S		
Original	Count	G.Biss_N	43	9	0	6	58
		G.Biss_S	11	33	9	5	58
		Gui_N	1	6	46	10	63
		Gui_S	2	7	11	43	63
	%	G.Biss_N	74.1	15.5	0.0	10.3	100
		G.Biss_S	19.0	56.9	15.5	8.6	100
		Gui_N	1.6	9.5	73.0	15.9	100
		Gui_S	3.2	11.1	17.5	68.3	100

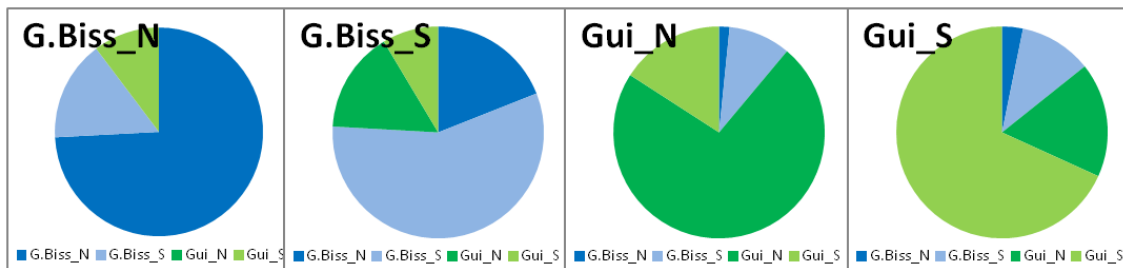


Figure 3.2.35.- Classification of *P. elongatus* (PSE) individuals (in %) into their original population using classification matrix of the LDA based on truss morphometry: Guinea-Bissau and Guinea, by area (North and South).

Multiple comparison tests using ANOVA on factor 1 (accounting the 73% of the variance), showed significant differences ($P \leq 0.001$) between the individuals from the four areas, although closer values between individuals from the two areas of each country are observed, northern and southern of Guinea-Bissau on the one hand, and northern and southern of Guinea, on the other hand (Figure 3.2.36).

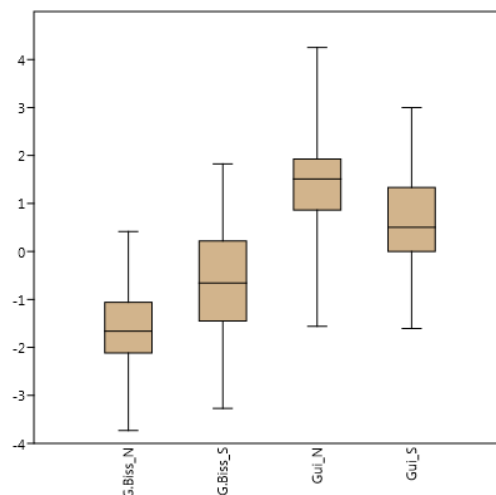


Figure 3.2.36.- Multiple comparison of scores from factor 1 for areas obtained from the LDA based on truss morphometry of *P. elongatus* (PSE) in Guinea-Bissau and Guinea, by area (North and South).

3.2.4.b- Otolith shape

A total number of 521 otoliths of *Pseudotolithus elongatus* were analysed for stock identification in Guinea-Bissau and Guinea.

The first 12 harmonics were used for the analysis as they reconstruct the otolith shape with 99% precision, that is explaining 99% of the otolith contour variation for this species. Each harmonic is composed by four coefficients (A, B, C, D) resulting in 48 Fourier coefficients for each otolith, 45 after normalization (Figure 3.2.37).



Figure 3.2.37.- Panel of otoliths shapes and mean shapes of *P. elongatus* (PSE) otoliths from Guinea-Bissau and Guinea, by area (North and South).

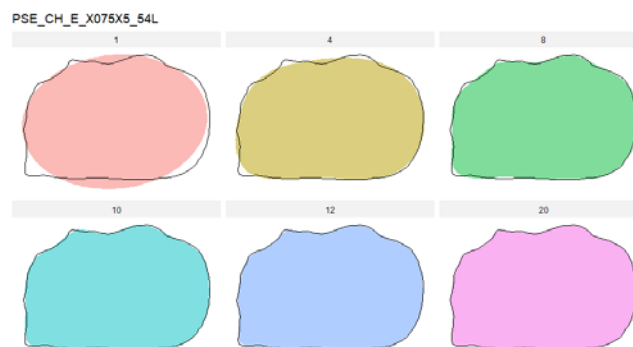


Figure 3.2.38.- Random reconstruction of *P. elongatus* (PSE) otoliths. Example from Cacheu (Guinea-Bissau) using 1, 4, 8, 10, 12 and 20 harmonics.

Multivariate analysis: Principal Component Analysis (PCA), Linear discriminant analysis (LDA) and Covariate Assisted Principal (CAP)

Once the otoliths were transformed to numbers (Fourier coefficients), the multivariate analysis was performed through PCA, LDA, the confusion matrix and CAP.

The CAP analysis showed certain influence of the fish length on the studied variables (Figure 3.2.39). Therefore, the allometric effect was corrected by eliminating those variables that could interfere with the classification. Once the correction was done, the analyses were repeated and new and more robust results were obtained.

The first two PCA components explained 56.5% of the variance in shape otoliths. The scatter plot of these two components showed a higher overlap in both Guinean areas (North and South) (Figure 3.2.40). This potential overlap was confirmed with the results of the LDA, as it classified the otoliths from each zone, with 51.56 % precision: DF1 contributed 66.01%, DF2 26.72% and DF3 7.27%.

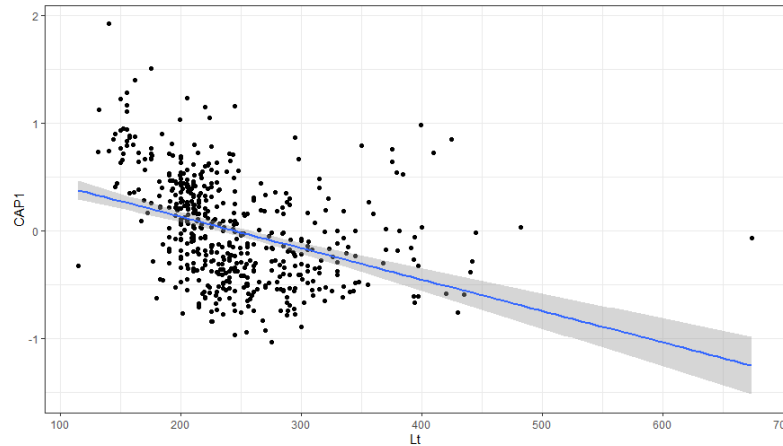


Figure 3.2.39.- Relationship between the total length variable and the first CAP dimension resulting from CAP for *P. elongatus* (PSE).

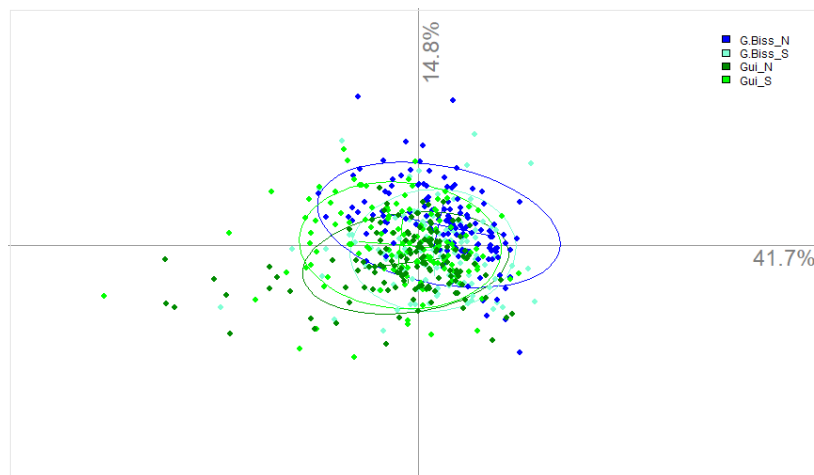


Figure 3.2.40.- Scatter plot of scores on the two principal components of PCA of *P. elongatus* (PSE) in Guinea-Bissau and Guinea, by area (North and South).

The confusion matrix (Table 3.2.23) showed 51.56% correct classification of *P. elongatus* into their original zones. The proportion of correct classification varied from 40.5% (in Guinea South) to 65.7% (in Guinea-Bissau North). At country level, individuals from Guinea-Bissau were better classified in their original area than individuals from Guinea, although a mixture of individuals was observed in all areas (see Figure 3.2.41).

Table 3.2.23.- Classification of *P. elongatus* (PSE) individuals (in number and %) into their original population using classification matrix (confusion matrix) of the LDA based on otolith shape: Guinea-Bissau and Guinea, by area (North and South).

		Country area	Predicted Group Membership				Total
			G.Biss_N	G.Biss_S	Gui_N	Gui_S	
Original	Count	G.Biss_N	88	19	10	17	134
		G.Biss_S	22	60	20	15	117
		Gui_N	15	24	68	33	140
		Gui_S	22	16	39	53	130
	%	G.Biss_N	65.7	14.0	7.4	12.8	100
		G.Biss_S	18.7	51.2	17.4	12.5	100
		Gui_N	10.8	17.5	48.4	23.4	100
		Gui_S	17.2	12.4	29.7	40.5	100

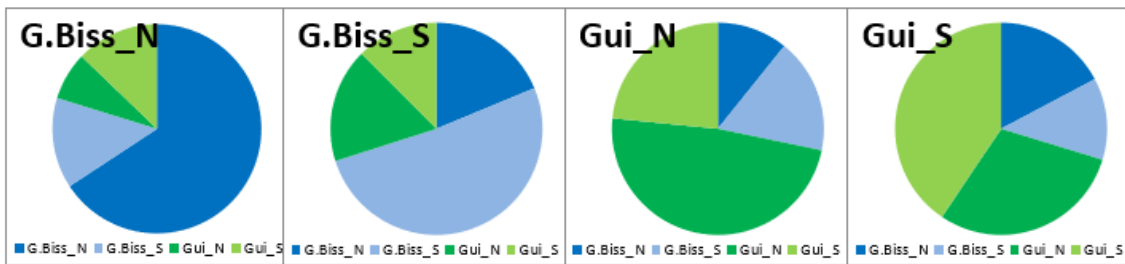


Figure 3.2.41.- Classification of *P. elongatus* (PSE) individuals (in %) into their original population using classification matrix of the LDA based on otolith shape: Guinea-Bissau and Guinea, by area (North and South).

The CAP analysis showed overlap in the four study areas, although differences at country level could be observed (Figure 3.2.42).

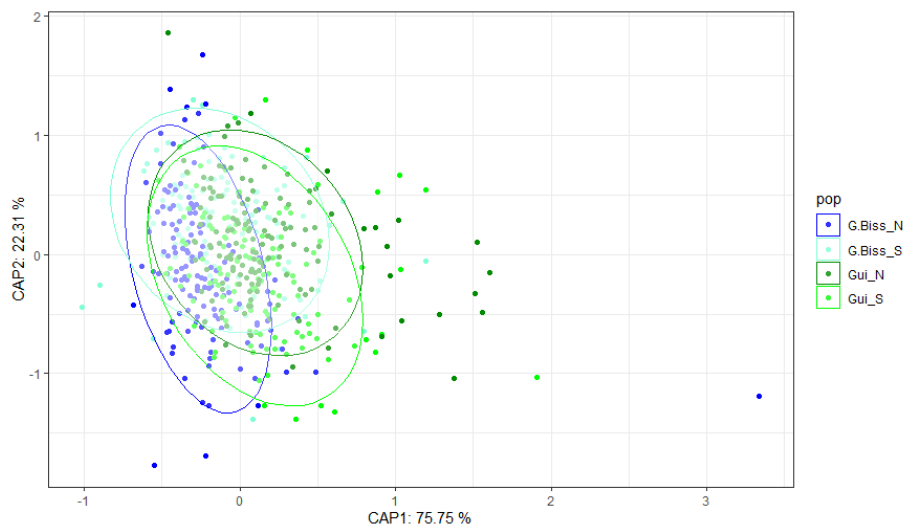


Figure 3.2.42.- Scatterplot of CAP results for *P. elongatus* (PSE) in Guinea-Bissau and Guinea, by area (North and South).

3.2.5. *Pseudotolithus senegalensis* (PSS)

3.2.5.a- Body shape

Body shape analysis using truss network from 190 specimens was performed. The number and lengths of the specimens collected from each area are shown in Table 3.2.24.

Table 3.2.24.- Number of specimens of *P. senegalensis* (PSS) analyzed and total length (Lt) ranges by studied area.

Area	Acronym	Number	Mean Lt (cm)	Lt range (cm)
Western Côte d'Ivoire waters	C.Iv_W	33	25	15-33
Eastern Côte d'Ivoire waters	C.Iv_E	51	22	15-34
Western Ghanaian waters	Gha_W	38	25	17-31
Eastern Ghanaian waters	Gha_E	68	27	19-33

Multivariate analysis

Principal Component Analysis (PCA)

Bartlett's sphericity was statistically significant ($p < 0.001$), and therefore the factor analysis was feasible. The results indicated that the first two factors together explained 78.85% of the total morphometric variation. The percentage of the total variance explained by each factor, together with the morphometric distances with significant loadings, and the body parts corresponding to them are included in Table 3.2.25.

Table 3.2.25.- Factors obtained by the PCA that explained most of the morphometric variation. *P. senegalensis* (PSS).

FACTOR	% of total variance	Morphometric distances	Part of the body	Colour in Fig.3.2.43
1	52.39%	m17, m10, m13, m12, m11, m3, m4, m6	Straight and oblique measurements and depth of the central and posterior part of the body	Red
2	26.46 %	m1, m19, m7, m15	Head lengths	Green

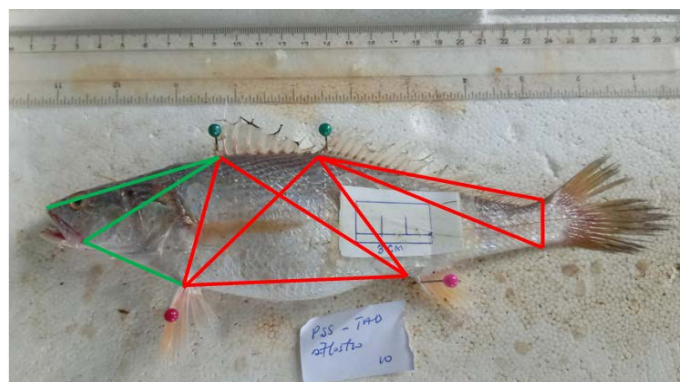


Figure 3.2.43.- Morphometric variables of *P. senegalensis* (PSS) with meaningful loadings in the PCA, corresponding to the first (red) and second factor (green).

The scatter plot of factor 1 and factor 2 shows overlap, being the western and eastern waters of Côte d'Ivoire more closely overlapping on one side, and the western and eastern waters of Ghana, on the other side (Figure 3.2.44).

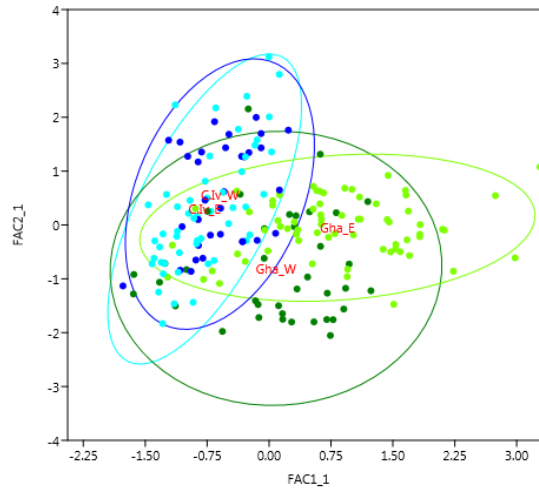


Figure 3.2.44.- Scatter plot of scores on the two factors extracted from morphometric characters of *P. senegalensis* (PSS): Côte d'Ivoire and Ghana (West and East).

Linear discriminant analysis (LDA)

A univariate ANOVA showed significant differences ($P < 0.001$) in 15 out of 16 morphometric measurements of the fish sampled from the four areas (Annex 5; Figure 3.2.45). The Wilks' lambda test of discriminant function analysis showed significant differences in morphometric measurements of all the areas ($P < 0.001$) (Annex 5).

The DFA showed three discriminant functions (DF) that allowed to significantly ($P < 0.001$) classify the subjects into two main groups. Table 3.2.26 shows the two main groups classified by the DFA, together with those measurements that are the most relevant for population discrimination in *P. senegalensis* (Table 3.2.27) and the parts of the body corresponding to them are shown (Table 3.2.27 and Figure 3.2.45).

Table 3.2.26.- Discriminant functions from DFA. *P. senegalensis* (PSS).

DF	% of between-group variability	Morphometric measurements	Part of the body	Colour in Fig.3.2.45
1	73.8%	m12, m11, m5	Lengths of the fish body	Blue
2	24.9%	m1, m7	Head lengths	Black



Figure 3.2.45.- Morphometric variables of *P. senegalensis* (PSS) with meaningful loadings in the LDA, corresponding to the first (blue) and second factor (black).

Table 3.2.27.- Contribution of morphometric measurements to the discriminant functions for *P. senegalensis* (PSS).

	Structure Matrix		
	Function		
	1	2	3
m13 ^a	,678*	,362	,261
m3 ^a	,647*	,172	,146
m17 ^a	,624*	,257	,028
m12	,557*	,313	,362
m11	,552*	,482	,014
m10 ^a	,525*	,257	,127
m4 ^a	,516*	,279	,146
m15 ^a	,492*	,090	-,036
m2 ^a	,461*	,428	-,403
m5	,423*	,332	,074
m6 ^a	,410*	,333	,098
m1	-,314	,617*	,485
m19 ^a	-,119	,611*	,306
m7	-,306	,609*	-,232
m9 ^a	-,287	,604*	-,016
m8 ^a	,123	,135	,324*

Pooled within-groups correlations between discriminating variables and standardized canonical discriminant functions
Variables ordered by absolute size of correlation within function.

a. This variable not used in the analysis.

*. Largest absolute correlation between each variable and any discriminant function

The two groups classified by the DFA show differentiation of the two Ivorian areas from the two Ghanaian areas (Figure 3.2.46). Both *P. senegalensis* samples from Côte d'Ivoire showed closeness, constituting a group, while the Ghanaian samples constituted a more heterogeneous group.

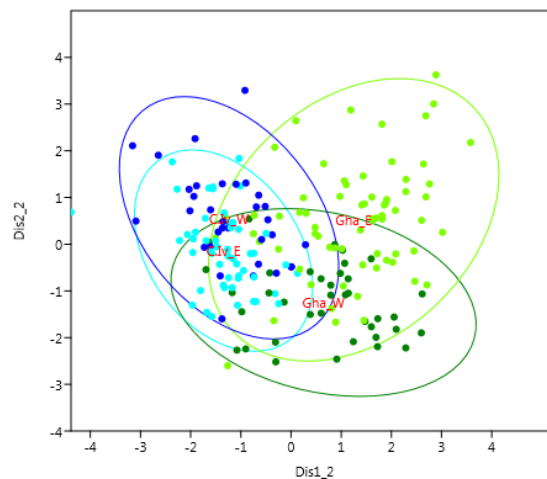


Figure 3.2.46.- Scatterplot of the first two canonical discriminant scores from the discriminant DFA for morphometric characters of *P. senegalensis* (PSS): Côte d'Ivoire and Ghana, by area (West and East).

LDA using cross-validation classification showed 67.9% correct classification of the *P. senegalensis* individuals into their original populations. The proportion of correctly classified varied between 36.4% and 80.4%, being able to be more clearly classified the *P. senegalensis* individuals from Ghana West and East (63.2% and 76.5%, respectively) and Côte d'Ivoire East (80.4%) than those from Côte d'Ivoire West (36.4 %) (Table 3.2.28; Figure 3.2.47).

The Côte d'Ivoire individuals of *P. senegalensis* as a whole (W and E), show high values of correct classification as Côte d'Ivoire individuals, being minimal the proportion of them belonging to Ghana (Figure 3.2.47). On the other hand, Ghanaian individuals (together W and E), are also highly classified as individuals from Ghana, with a minimum of Côte d'Ivoire individuals (Table 3.2.28; Figure 3.2.47). Therefore, the cross-validation classification shows two groups of *P. senegalensis*: on the one hand, Côte d'Ivoire individuals (West and East) and, on the other hand, Ghanaian individuals (West and East).

Table 3.2.28.- Classification of *P. senegalensis* (PSS) individuals (in number and %) into their original population using classification matrix of the LDA based on truss morphometry: Côte d'Ivoire and Ghana, by area (West and East).

		Country area	Predicted Group Membership				Total
			C.Iv_W	C.Iv_E	Gha_W	Gha_E	
Original	Count	C.Iv_W	12	18	2	1	33
		C.Iv_E	8	41	2	0	51
		Gha_W	0	7	24	7	38
		Gha_E	3	4	9	52	68
	%	C.Iv_W	36.4	54.5	6.1	3.0	100
		C.Iv_E	15.7	80.4	3.9	0.0	100
		Gha_W	0.0	18.4	63.2	18.4	100
		Gha_E	4.4	5.9	13.2	76.5	100

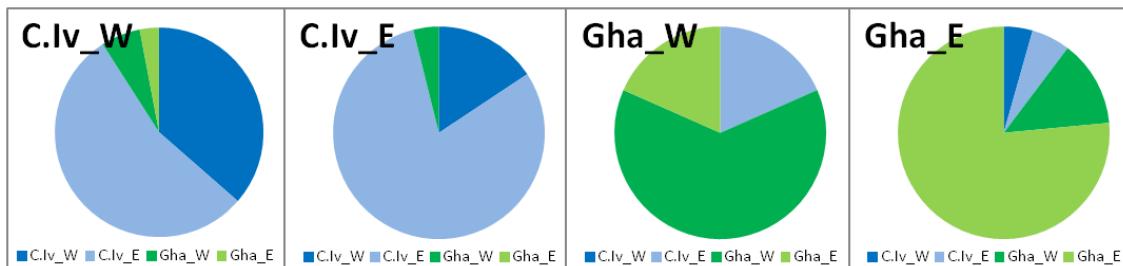


Figure 3.2.47.- Classification of *P. senegalensis* (PSS) individuals (in %) into their original population using classification matrix of the LDA based on truss morphometry: Côte d'Ivoire and Ghana, by area (West and East).

Multiple comparison tests using ANOVA on factor 1 (accounting the 74% of the variance), showed no significant differences ($P > 0.001$) between the individuals from western and eastern Côte d'Ivoire, that can belong to a single stock (Figure 3.2.48). The two Ghanaian areas showed values close to, but significantly different ($P \leq 0.001$) from each other, and markedly different from those of Côte d'Ivoire. The two groups, corresponding to each country, are thus observed.

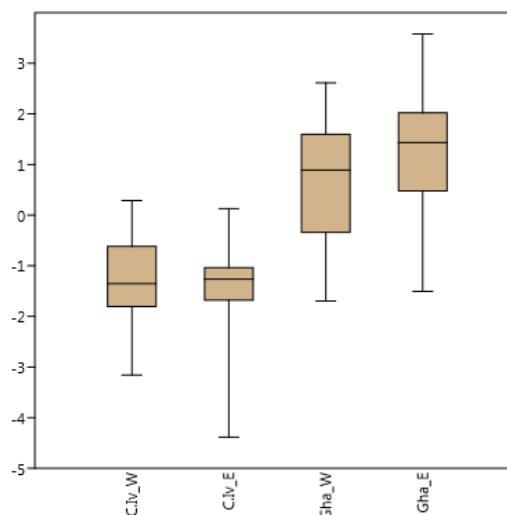


Figure 3.2.48.- Multiple comparison of scores from factor 1 for areas obtained from the LDA based on truss morphometry of *P. senegalensis* (PSS) in Côte d'Ivoire and Ghana, by area (West and East).

3.2.5.b- Otolith shape

A number of 339 otoliths of *P. senegalensis* from Côte d'Ivoire and Ghana were photographed for otolith shape analysis. However, only 187 otoliths could be analysed, as the others could not be correctly linked to their correspondent individual.

The first 15 harmonics were considered to perform the analyses, as they are able of reproducing the otolith shape of *P. senegalensis* with 99% accuracy (Figure 3.2.49).

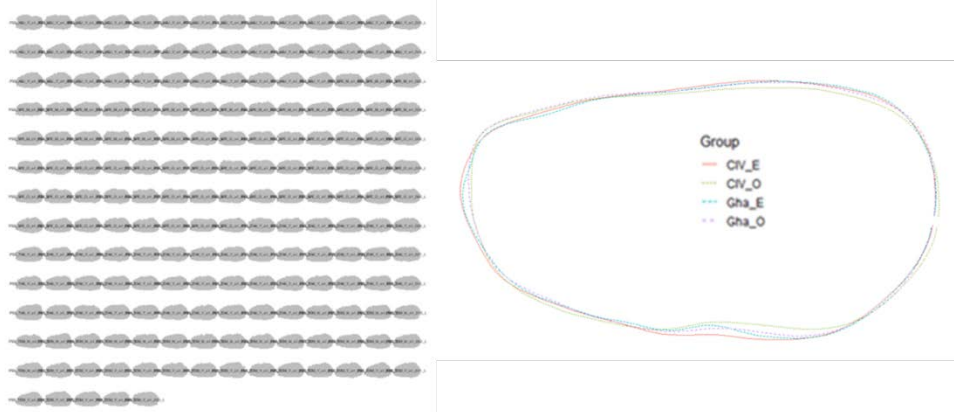


Figure 3.2.49- Panel of otoliths shapes and mean shapes of *P. senegalensis* (PSS) otoliths from Côte d'Ivoire and Ghana, by area (West and East).

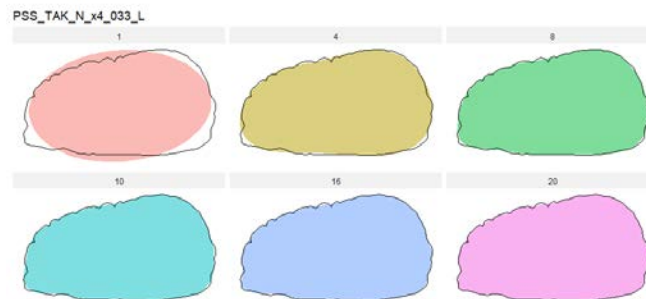


Figure 3.2.50.- Random reconstruction of *P. senegalensis* (PSS) otoliths. Example from Takoradi (Ghana) using 1, 4, 8, 10, 16 and 20 harmonics.

Multivariate analysis: Principal Component Analysis (PCA), Linear discriminant analysis (LDA) and Covariate Assisted Principal (CAP)

Once the contours of all the otoliths were transformed by Fourier analysis, different multivariate analyses were performed. The Covariate Assisted Principal (CAP) analysis (figure 3.2.51) indicated a possible influence of the length of the individuals in the otolith shape analysis. Following this observation, the allometric effect was removed by eliminating the descriptors influenced by length, by applying an ANCOVA test. After the PCA, LDA and CAP analyses were repeated.

The PCA results showed two principal components explaining 51.7% of the overall variance of the otolith shape in the overlapping areas (figure 3.2.52). The first component contributed 38.8% and the second 12.9%. LDA's results showed three DFs: the first, second and third DFs accounting for 47.96%, 30.80% and 21.24% of the variability, respectively.

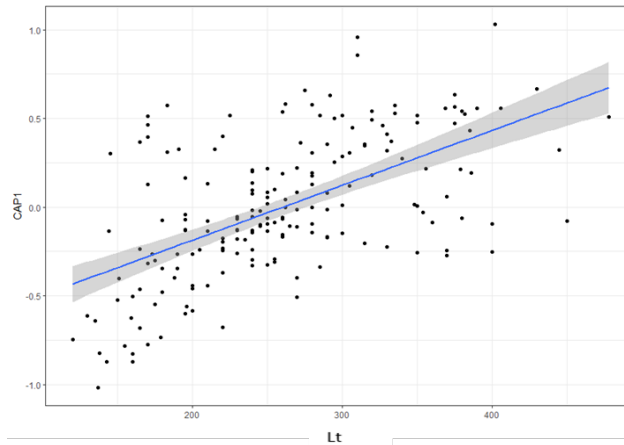


Figure 3.2.51.- Relationship between the total length variable and the first CAP dimension resulting from CAP for *P. senegalensis* (PSS).

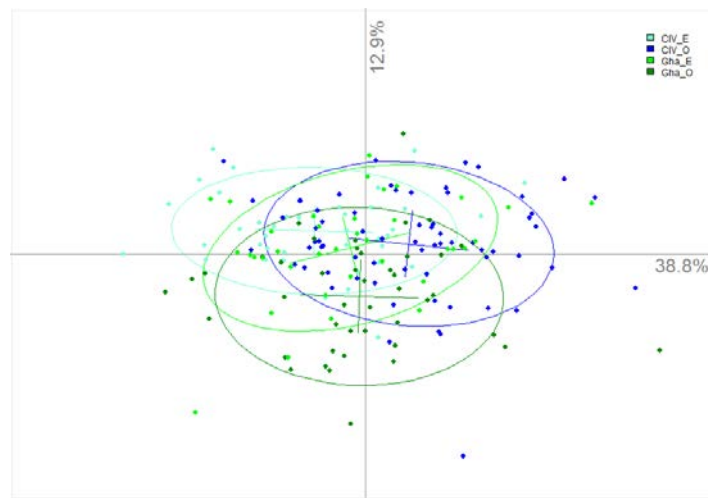


Figure 3.2.52.- Scatter plot of scores on the two principal components of PCA of *P. senegalensis* (PSS) in Côte d'Ivoire and Ghana, by area (West and East).

The classification matrix obtained from the LDA (Table 3.2.29) indicated that the value of correct classification for *P. senegalensis* otoliths was 61.25% with a range of variation between 51.4% (individuals from Ghana West) and 65.2% (individuals from Ghana East) (see Figure 3.2.53). The scatter plot CAP showed an overlapping of all the zones (Figure 3.2.53).

Table 3.2.29.- Classification of *P. senegalensis* (PSS) individuals (in number and %) into their original population using classification matrix (confusion matrix) of the LDA based on otolith shape: Côte d'Ivoire and Ghana, by area (West and East).

		Country area	Predicted Group Membership				Total
			C.Iv_W	C.Iv_E	Gha_W	Gha_E	
Original	Count	C.Iv_W	44	7	12	5	67
		C.Iv_E	9	24	4	2	39
		Gha_W	12	5	23	4	44
		Gha_E	6	4	3	24	37
	%	C.Iv_W	65.0	9.8	17.6	7.5	100
		C.Iv_E	23.5	62.3	9.6	5.1	100
		Gha_W	28.0	10.6	51.4	9.8	100
		Gha_E	15.2	11.6	8.1	65.2	100

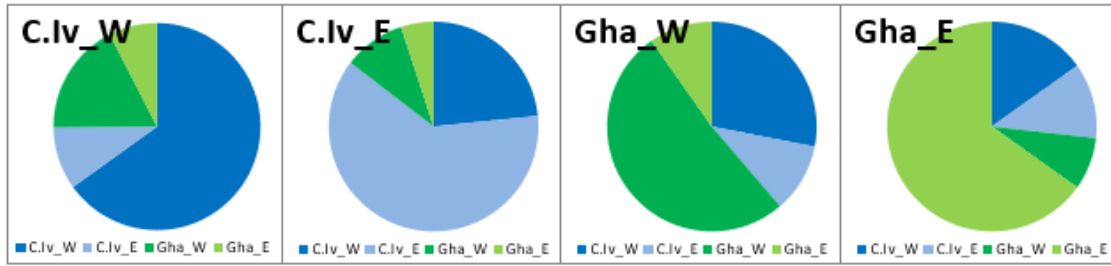


Figure 3.2.53.- Classification of *P. senegalensis* (PSS) individuals (in %) into their original population using classification matrix of the LDA based on otolith shape: Côte d'Ivoire and Ghana, by area (West and East).

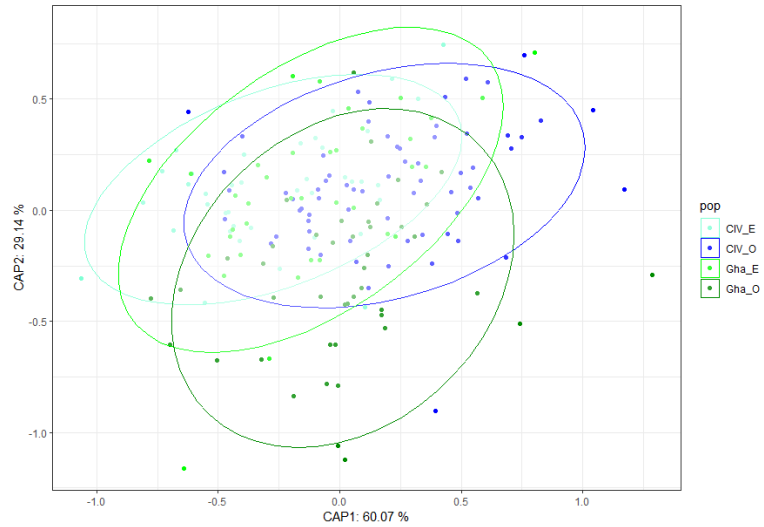


Figure 3.2.54.- Scatterplot of CAP results for *P. senegalensis* (PSS) in Côte d'Ivoire and Ghana, by area (West and East).

3.2.6. *Pagellus bellottii* (PAR)

3.2.1.a- Body shape

Body shape analysis using truss network from 212 specimens was performed. The number and lengths of the specimens collected from each area are shown in Table 3.2.30.

Table 3.2.30.- Number of specimens of *P. bellottii* (PAR) analyzed and total length (Lt) ranges by studied area.

Area	Acronym	Number	Mean Lt (cm)	Lt range (cm)
Western Côte d'Ivoire waters	C.Iv_W	30	15	9-18
Eastern Côte d'Ivoire waters	C.Iv_E	63	16	10-20
Western Ghanaian waters	Gha_W	60	16	14-20
Eastern Ghanaian waters	Gha_E	59	16	13-20

Multivariate analysis

Principal Component Analysis (PCA)

The factor analysis was feasible as Bartlett's sphericity was statistically significant ($p < 0.001$). Results indicated that the first three factors together explained 74.82% of the total morphometric variation. The proportion of the total variance explained by each factor, together with the morphometric distances with significant loadings and their corresponding parts of the body (Figure 3.2.55) are summarized in Table 3.2.31.

Table 3.2.31.- Factors obtained by the PCA that explained most of the morphometric variation. *P. bellottii* (PAR).

FACTOR	% of total variance	Morphometric distances	Part of the body	Colour in Fig.3.2.55
1	38.35%	m14, m2, m10, m12	Straight and oblique measurements of the posterior half of the fish body	Red
2	22.61%	m3, m11, m9	Depth of the fish body and the caudal peduncle	Green
3	13.86%	m16, m1	Two head measurements	Yellow

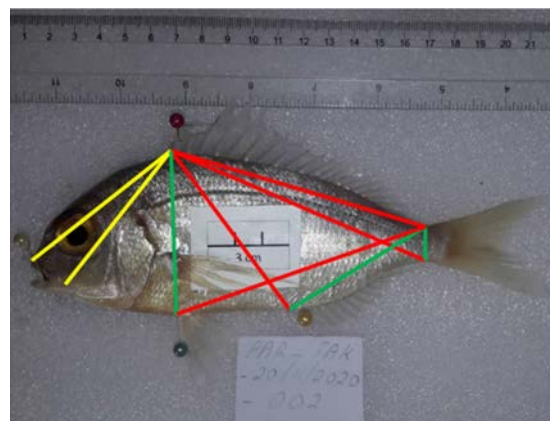


Figure 3.2.55.- Morphometric variables of *P. bellottii* (PAR) with meaningful loadings in the PCA, corresponding to the first (red), second (green) and third factor (yellow).

The scatter plot of factor 1 and factor 2 shows overlap among the four areas with closer overlapping between the two areas of Côte d'Ivoire (West and East) on one side, and between the two areas of Ghana (West and East) on the other side (Figure 3.5.56).

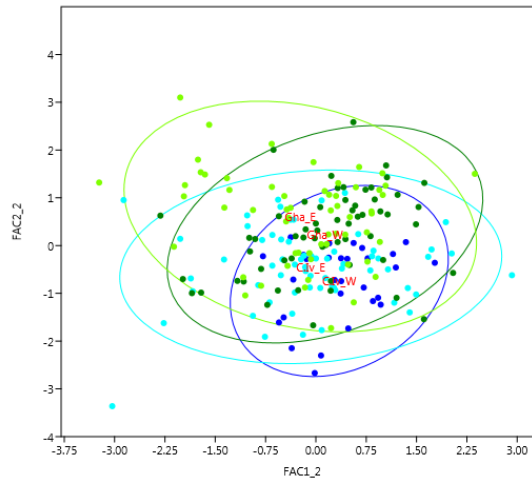


Figure 3.5.56.- Scatter plot of scores on the two factors extracted from morphometric characters of *P. bellottii* (PAR): Côte d'Ivoire and Ghana, by area (West and East).

Linear discriminant analysis (LDA)

A univariate ANOVA showed significant differences ($P < 0.001$) in 10 out of 14 morphometric measurements of the fish sampled from four areas (Annex 5; Figure 3.2.57). The Wilks' lambda test of discriminant function analysis showed significant differences in morphometric measurements of all the areas ($P < 0.001$) (Annex 5).

The discriminant function analysis (DFA) showed three discriminant functions (DF) that allowed to significantly ($P < 0.001$) classify the subjects into three groups. Table 3.2.32 shows the proportion of the between-group variability explained by each DF, the contribution of the morphometric measurements to them (detailed in Table 3.2.33) and the correspondent part of the body.

The DFA results showed differentiation of the two Ivorian areas from the two Ghanaian areas (Figure 3.2.58). Both *P. bellottii* samples from Côte d'Ivoire showed closeness, constituting a group, while both Ghanaian samples were grouped together in another group.

Table 3.2.32.- Discriminant functions from DFA. *P. bellottii* (PAR).

DF	% of between-group variability	Morphometric measurements	Part of the body	Colour in Fig. 3.2.57
1	86.2%	m6, m5	Lengths of lower head and fish body	Blue
2	11.2%	m1, m9, m4	Upper head length, body depth and lower rear body length	Black
3	2.6%	m7, m8	Mouth size and length of lower head	Pink

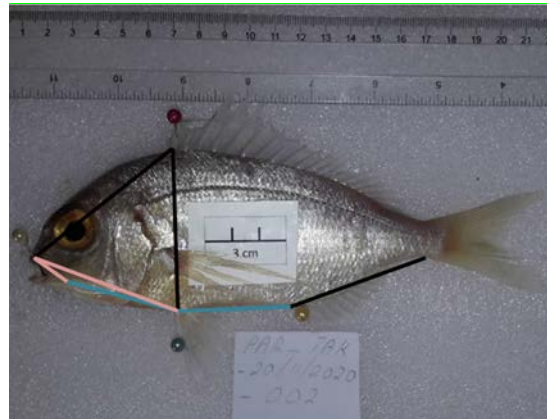


Figure 3.2.57.- Morphometric variables of *P. bellottii* (PAR) with meaningful loadings in the LDA, corresponding to the first (blue), second (black) and third factor (pink).

Table 3.2.33.- Contribution of morphometric measurements to the discriminant functions for *P. bellottii* (PAR).

	Function		
	1	2	3
m6	,385*	,060	-,002
m5	,283*	,234	-,102
m11 ^a	-,186*	-,086	-,004
m3 ^a	-,124*	,086	,123
m16 ^a	,239	,683*	,031
m1	,341	,642*	,059
m9	-,308	,514*	-,292
m10 ^a	,101	,342*	-,266
m4	-,190	-,207*	,018
m7	-,370	,106	,793*
m12 ^a	,077	,116	-,369
m8	,203	-,006	,308*
m14 ^a	,032	-,043	-,282*
m2 ^a	-,027	-,002	-,222*

Pooled within-groups correlations between discriminating variables and standardized canonical discriminant functions
Variables ordered by absolute size of correlation within function.

*. Largest absolute correlation between each variable and any discriminant function

a. This variable not used in the analysis.

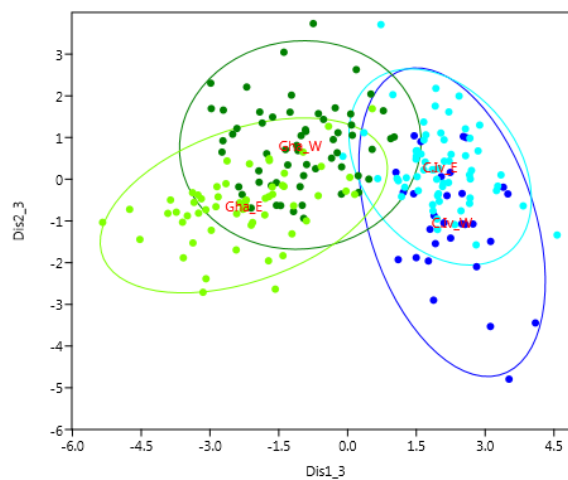


Figure 3.2.58- Scatterplot of the first two canonical discriminant scores from the DFA for morphometric characters of *P. bellottii* (PAR): Côte d'Ivoire and Ghana, by areas (West and East).

LDA using cross-validation classification showed 75.9% correct classification of the *P. bellottii* individuals into their original populations. The proportion of correctly classified varied between 53.3% and 84.1%. The *P. bellottii* individuals more clearly classified were those from Ghana West (75%), Ghana East (79.7%) and Côte d'Ivoire East (84.1%) (Table 3.2.34; Figure 3.2.59).

The Côte d'Ivoire individuals of *P. bellottii* as a whole (West and East), show high values of correct classification as Ivorian individuals, being minimal the proportion of them belonging to Ghana (see Figure 3.2.59). On the other hand, Ghanaian individuals (together West and East), are also highly classified as individuals from Ghana, with a minimum of Côte d'Ivoire individuals, higher in Ghana West (Table 3.2.34; Figure 3.2.59). Therefore, the cross-validation classification shows two groups of *P. bellottii*: on the one hand, Ivorian individuals (West and East) and, on the other hand, Ghanaian individuals (West and East).

Table 3.2.34.- Classification of *P. bellottii* (PAR) individuals (in number and %) into their original population using classification matrix of the LDA based on truss morphometry: Côte d'Ivoire and Ghana, by areas (West and East).

		Country area	Predicted Group Membership				Total
			C.Iv_W	C.Iv_E	Gha_W	Gha_E	
Original	Number	C.Iv_W	16	14	0	0	30
		C.Iv_E	8	53	2	0	63
		Gha_W	0	5	45	10	60
		Gha_E	0	2	10	47	59
	%	C.Iv_W	53.3	46.7	0.0	0.0	100
		C.Iv_E	12.7	84.1	3.2	0.0	100
		Gha_W	0.0	8.3	75.0	16.7	100
		Gha_E	0.0	3.4	16.9	79.7	100

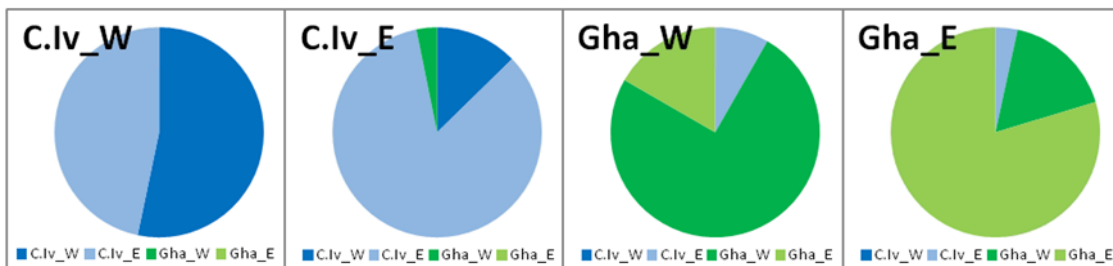


Figure 3.2.59.- Classification of *P. bellottii* (PAR) individuals (in %) into their original population using classification matrix of the LDA based on truss morphometry: Côte d'Ivoire and Ghana, by areas (West and East).

Multiple comparison tests using ANOVA on factor 1 (accounting the 87% of the variance), showed no significant differences ($P > 0.001$) between the individuals from West Côte d'Ivoire and East Côte d'Ivoire), that can belong to a single stock (Figure 3.2.60). The two Ghanaian areas showed values close to, but significantly different ($P \leq 0.001$) from each other, and markedly different from those of Côte d'Ivoire. The two groups, corresponding to each country, are thus observed.

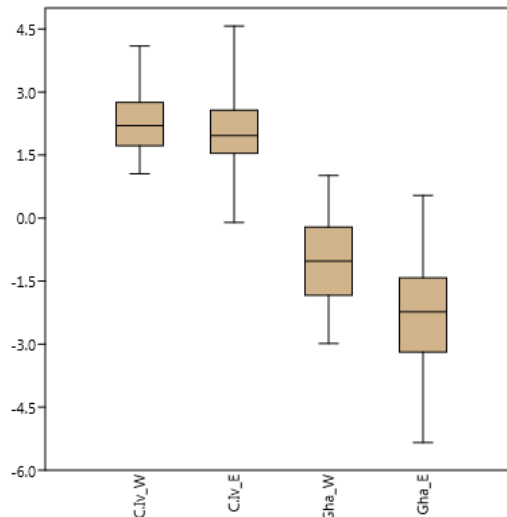


Figure 3.2.60.- Multiple comparison of scores from factor 1 for areas obtained from the LDA based on truss morphometry of *P. bellottii* (PAR) in Côte d'Ivoire and Ghana, by areas (West and East).

3.2.6.b- Otolith shape

Unfortunately the otolith shape analysis was hampered by the lost and/or misidentification of some otoliths coming from different sampled areas. This is the case of the otoliths corresponding to the individuals sampled and in May in Takoradi and in November in Tema (Ghana West and East, respectively), that were not received at the IEO. In addition, Ghana West could not be considered in the analysis, since it was not possible to correlate the otoliths sampled in Takoradi with the individuals of the sampling available (in November 2020). Thus, only 216 right otoliths could be included in the analysis from a total of 347 left and right otoliths photographed.

As for *P. caeruleostictus*, 17 harmonics were needed for *P. bellottii* to reconstruct the shape of the otoliths with 99% accuracy (Figure 3.2.61).

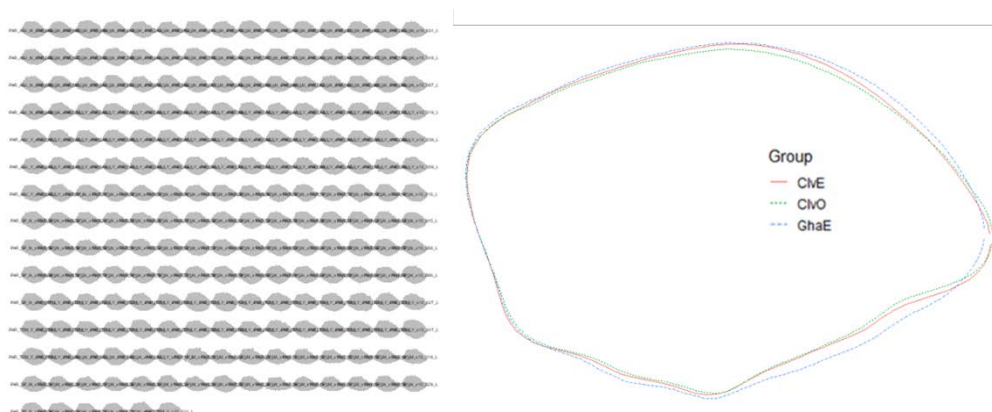


Figure 3.2.61.- Panel of otoliths shapes and mean shapes of *P. bellottii* (PAR) otoliths from Côte d'Ivoire and Ghana, by zone (West and East).

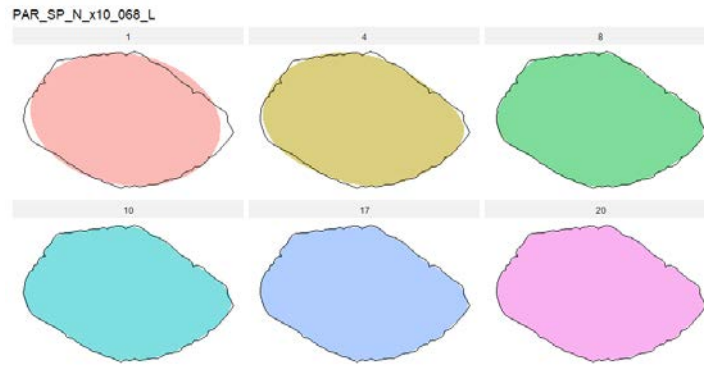


Figure 3.2.62.- Random reconstruction of *P. bellottii* (PAR) otoliths. Example from San Pedro (Côte d’Ivoire) using 1, 4, 8, 10, 17 and 20 harmonics.

Multivariate analysis: Principal Component Analysis (PCA), Linear discriminant analysis (LDA) and Covariate Assisted Principal (CAP)

After multivariate analyses of the otolith shape of *P. bellottii* taking into consideration all the Fourier descriptors calculated from the contours, the potential influence of the allometric effect was checked by the CAP analysis (Figure 3.2.63). The results of the CAP conformed this influence and thus, the descriptors that are influenced by the length by applying an ANCOVA test were eliminate from the data set the and the multivariate analyses consequently resumed.

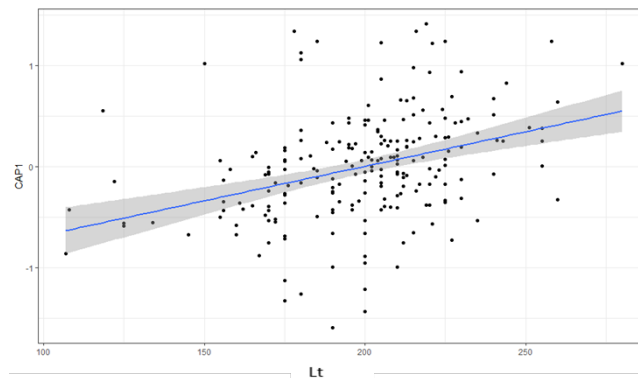


Figure 3.2.63.- Relationship between the total length variable and the first CAP dimension resulting from CAP for *P. bellottii* (PAR).

The results of the PCA showed that two principal components explain 50.50% of the total otolith shape variation, the first component explaining 37.40% and the second 13.1%. The scatter plot indicated overlap between the three analysed areas (Figure 3.2.64).

Two discriminant functions were obtained from the LDA analysis: DF1 and DF2, explaining 61.50% and 38.5% (respectively) of the classification of individuals of *P. bellottii* in their original zone. The matrix confusion obtained from the LDA (Table 3.2.35) showed a 53.36% of correct classification, varying between 47.9% (in Ghana East) and 57.8% (in Côte d’Ivoire East). The proportion of correct classification in the three areas (around 50%) indicates that half of the individuals comes from other area zones, this indicating a mixture of the population (see Figure 3.2.65).

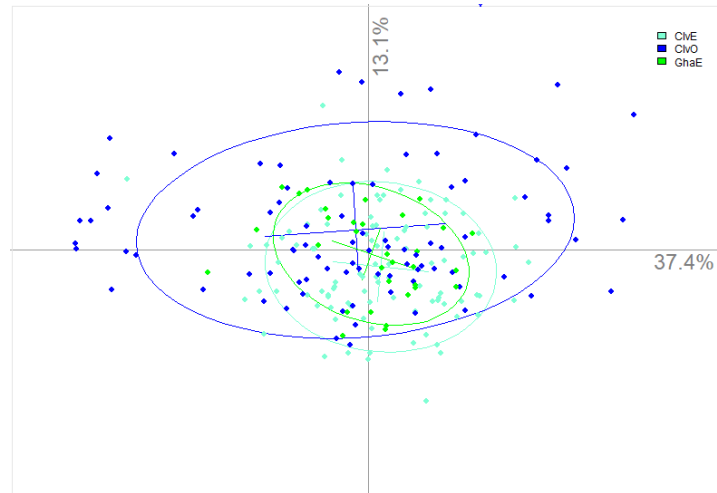


Figure 3.2.643.- Scatter plot of scores on the two principal components of PCA of *P. bellottii* (PAR) in Côte d'Ivoire and Ghana, by area (West and East).

Table 3.2.35.- Classification of *P. bellottii* (PAR) individuals (in number and %) into their original population using classification matrix (confusion matrix) of the LDA based on otolith shape: Côte d'Ivoire and Ghana, by area (West and East).

		Country area	dicted Group Members			Total
			C.Iv_W	C.Iv_E	Gha_E	
Original	Number	C.Iv_W	44	33	10	30
		C.Iv_E	29	56	11	63
		Gha_E	6	11	15	59
	%	C.Iv_W	50.6	37.8	11.8	100
		C.Iv_E	30.6	57.8	11.7	100
		Gha_E	17.6	33.8	47.9	100

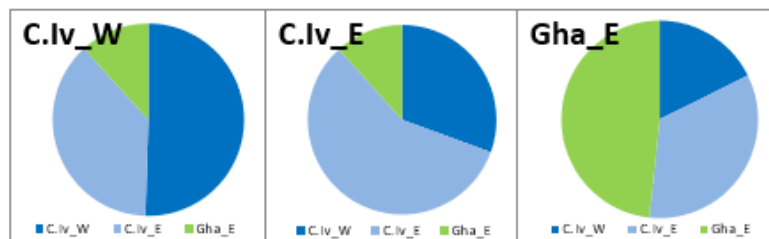


Figure 3.2.65.- Classification of *P. bellottii* (PAR) individuals (in %) into their original population using classification matrix of the LDA based on otolith shape: Côte d'Ivoire and Ghana, by area (West and East).

An overlap between the three areas, similar to that showed by the PCA is observed in the scatter plot of the two main CAP functions (Figure 3.2.66).

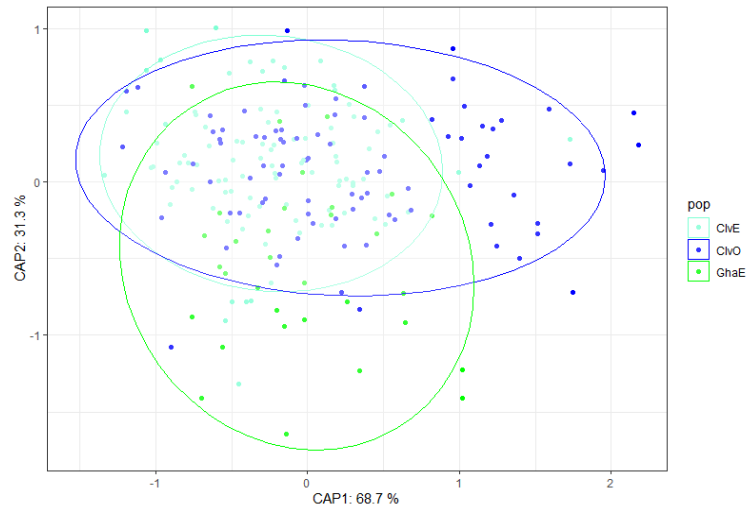


Figure 3.2.66- Scatterplot of CAP results for *P. bellottii* (PAR) in Côte d'Ivoire and Ghana, by area (West and East).

It should be noted that these results may vary since, as mentioned above, West Ghana could not be considered in the analysis.

3.2.7 Summary table- Morphometry

In order to summarize all results obtained by all methods used for stock identification by morphometry (body and otolith shape), Table 3.2.36 is presented. It includes the most relevant results of the analysis carried out in the five fish species for otolith shapes and in the six species (including the southern pink shrimp *P. notialis*) for body shape.

Table 3.2.36.- Summary table of the results of the different multivariate analyses on body and otolith shapes performed on all target species.

Species		GPW		SOP	BSC		PSE		PSS		PAR	
Method		Body shape	Otolith shape	Body shape	Body shape	Otolith shape	Body shape	Otolith shape	Body shape	Otolith shape	Body shape	Otolith shape
n		329	230	228	249	335	242	521	190	187	212	216
n° harmonics		–	16	–	–	17	–	12	–	15	–	17
PCA	PCA C1	53.0	35.8	47.0	43.6	27.5	44.7	41.7	52.4	38.8	38.4	37.4
	PCA C2	17.3	20.2	11.2	25.9	15.2	16.8	14.8	26.5	12.9	22.6	13.1
	C1 + C2	70.3	56.0	58.2	69.5	42.7	61.5	56.5	78.9	51.7	61.0	50.5
LDA	LD1	68.7	66.2	37.6	88.3	77.3	72.7	66.0	73.8	48.0	86.2	61.5
	LD2	23.8	21.2	30.9	11.7	22.7	16.1	26.7	24.9	30.8	11.2	38.5
	LD3	7.5	12.5	14.6	–	–	11.1	7.3	–	21.2	2.6	–
	% CC (accuracy)	55.0	39.3	88.2	84.7	60.3	68.2	51.6	67.9	61.3	75.9	53.4
CAP	CAPRESULT 1	–	76.8	–	–	84.9	–	75.8	–	60.1	–	68.7
	CAPRESULT 2	–	20.5	–	–	15.1	–	22.3	–	29.1	–	31.3

3.3. GENETICS

3.3.1. Study of the genetic structure of populations to identify the geographical limits of stocks using microsatellite markers

3.3.1.a *Epinephelus aeneus* (GPW)

Characterization of GPW genetic diversity

Genomic DNA was obtained from 509 individuals of *E. aeneus* (Figure 3.3.1). The quality of the DNA obtained was not homogeneous among all samples (data not shown).

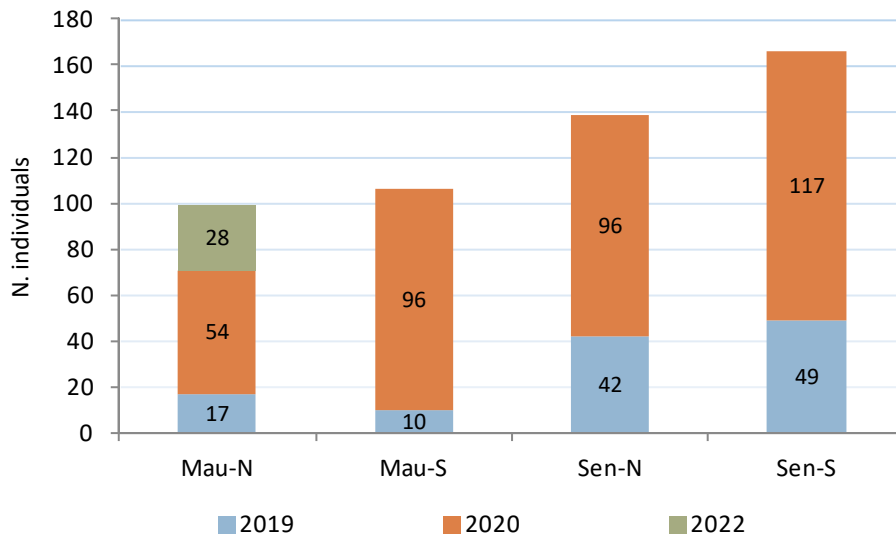


Figure 3.3.1.- Number of *E. aeneus* (GPW) individuals sampled for genetics by country-area and year.

The final design allowed the amplification of eight microsatellite loci (Dor et al., 2014) in two multiplex PCR reactions; PCR M1 (ARO1105, ARO1045, ARO1084 and ARO1083) and PCR M2 (ARO1003, ARO1120 and ARO 1137), and a single PCR M3 (ARO 1131) (Table 3.3.1). Amplification was only obtained in 461 individuals due to variable quality in DNA.

Fragment analysis of the PCR products was performed using GeneScan™ 500 LIZ (Applied Biosystems, Foster City, CA, USA) as size standard. Alleles were called at 8 loci in 461 GPW (Figure 3.3.2) with software GeneMarker v2.7.0 (SoftGenetics). The established internal threshold for missing data per individual was 25% (two loci with missing data), resulting in genetic data for 354 thiof individuals. Table 3.3.2 shows the number (N) of individuals of *E. aeneus* genotyped at each loci, and the total number of alleles (Na) counted.

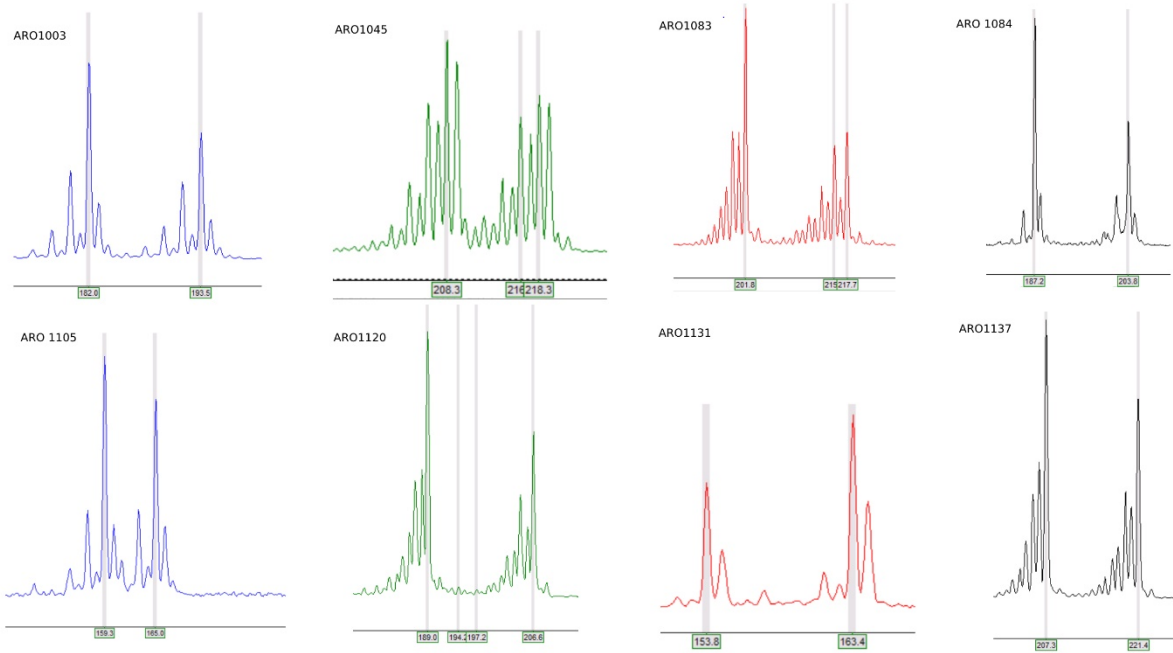


Figure 3.3.2.- Allele calling for eight *E. aeneus* (GPW) microsatellite markers.

Table 3.3.1.- Characterization of genetic diversity at *E. aeneus* (GPW) microsatellite loci

Pop ^a	ARO1105	ARO1045	ARO1084	ARO1083	ARO1003	ARO1120	ARO 1137	ARO 1131
MAU_N	57/23	58/21	65/22	65/30	65/27	65/26	66/24	66/19
MAU_S	35/23	41/18	50/27	49/26	50/30	50/28	50/24	47/19
SEN_N	72/26	71/22	92/31	93/30	93/33	93/35	93/26	91/22
SEN_S	120/28	120/24	143/35	140/33	145/33	145/24	144/28	141/24

^a Pop refers to each area where GPW were collected. Number of GPW individuals (N) and number of alleles (Na) characterized for each microsatellite loci. Each cell indicates data for N/Na.

Table 3.3.2.- Estimation of *E. aeneus* (GPW) genetic diversity per country-area: Mauritania and Senegal (North and South).

Pop ^a	N	Na	Ne	H _O	H _E	uH _E
MAU_N	63	24	13	0.871	0.903	0.910
MAU_S	47	24	13	0.893	0.909	0.919
SEN_N	88	28	14	0.861	0.913	0.919
SEN_S	137	90	14	0.866	0.912	0.915

^aPop refers to each area where GPW were collected. Each cell indicates the average value per area.

- N= number of individuals analyzed;
- Na= number of different counted alleles;
- Ne= number of effective alleles according to allele frequencies;
- H_O= observed heterozygosity;
- H_E= expected heterozygosity according to allele frequencies;
- uH_E= unbiased expected heterozygosity according to sample size.

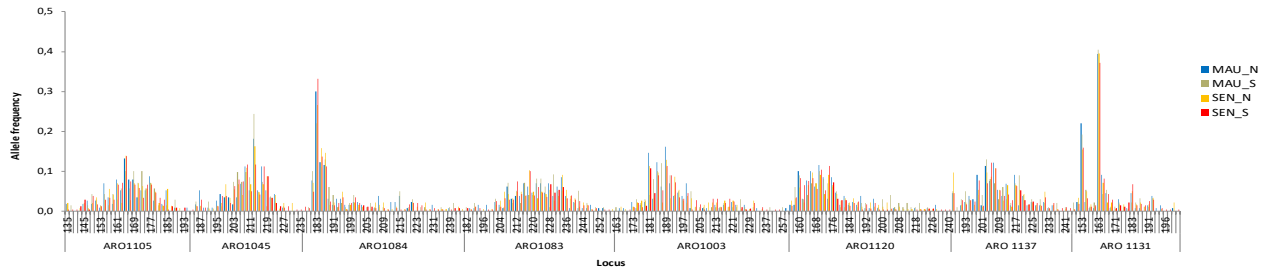


Figure 3.3.3.- Allele frequency for *E. aeneus* (GPW) per microsatellite locus and country-area: Mauritania and Senegal (North and South).

The inbreeding coefficient (F_{IS}) is the proportion of the variance in the subpopulation contained in an individual. High F_{IS} implies a considerable degree of inbreeding. The values obtained for F_{IS} and the probabilities associated to HW equilibrium are shown in Table 3.3.3 In all four areas significant deviations from HW proportions and homozygote excess (positive F_{IS}) were obtained for loci ARO1003, indicating the possible presence of null alleles.

Table 3.3.3.- Inbreeding coefficient F_{IS}

Pop ^a	ARO1105	ARO1045	ARO1084	ARO1083	ARO1003	ARO1120	ARO 1137	ARO 1131
MAU_N	0.0508	-0.0259	0.0482	0.0185	0.2504***	0.0508	-0.0329	-0.0221
MAU_S	0.0084	0.0537**	-0.0128	-0.0047	0.1752***	0.0131	-0.0129	0.0105
SEN_N	0.0302	0.0263	0.1438*	0.0771***	0.1345*	0.0161	0.0643	0.0078
SEN_S	0.0092	0.0682	0.0730	0.0495	0.1659***	0.0304*	0.0218	0.0144

^aPop refers to each area where GPW were collected. Each cell indicates the value of the inbreeding coefficient F_{IS} calculated according to (Weir and Cockerham, 1984), followed by the significance level (* $p < 0.05$, ** $p < 0.01$, or *** $p < 0.001$) of the probability p value obtained with the exact probability test for HW equilibrium calculated by the Markov chain method (10,000 dememorization, 1,000 batches, 1,0000 iterations per batch).

Structuring of GPW genetic diversity

F_{ST} is the proportion of the total genetic variance contained in a subpopulation relative to the total genetic variance. Values can range from 0 to 1. High F_{ST} implies a considerable degree of differentiation among populations. Structuring of genetic diversity of *E. aeneus* was assessed through estimation of F_{ST} from multilocus genotypes of the species grouped into 4 populations or sampling areas (Table 3.3.4), either considering all eight microsatellite loci characterized, or from the data for seven loci, excluding ARO1003. The results were identical in both cases.

Table 3.3.4.- F_{ST} pairwise comparison of GPW genetic diversity at 8 loci in 4 populations.

	MAU_N	MAU_S	SEN_N	SEN_S
MAU_N				
MAU_S	0.004			
SEN_N	0.003	0.004		
SEN_S	0.003	0.005	0.002	

All F_{ST} values were non-significant after Bonferroni correction (α 0.05) for multiple comparisons. Non-significant F_{ST} pairwise comparisons indicate genetic connectivity between the sampling areas. Estimation of migrants per microsatellite locus between areas is shown in Table 3.3.5, supporting a high level of migration with an average number of migrants of 51, and thus high levels of gene flow between sampling areas.

Table 3.3.5.- Number of migrants per microsatellite locus.

Locus	All Pops
ARO1105	52
ARO1045	37
ARO1084	47
ARO1083	48
ARO1003	35
ARO1120	55
ARO1137	54
ARO1131	84
Mean \pm SE	51 \pm 5

To assess if weak barriers to gene flow exist between the highly connected *E. aeneus* populations under study, clustering of genetic diversity was performed through Discriminant Analysis of Principal Components (DAPC) (Figure 3.3.4). This analysis performed with genetic data for 354 *E. aeneus* genotyped at 8 microsatellite loci and grouped in 4 sampling areas. The proportion of conserved variance is 96.6%. The eigenvalues for the first two vectors (Discriminant functions 1 and 2) are 113.8 and 77.6 (Figure 3.3.4).

The DAPC illustrates high levels of gene flow between Senegalese populations of *E. aeneus*.

Clustering of genetic diversity was studied through Bayesian algorithms (Figure 3.3.5) that assign individuals to a number of K clusters assuming an admixture model and considering sampling location of 354 *E. aeneus* individuals genotyped at 8 loci. STRUCTURE software was used considering an ancestry admixture correlated allele frequency model, and prior sampling location information. Structure Harvester exploration of results indicated the best K was 2, and Clumpak produced the most frequent consensus solution for $K = 2$, proportionally assigning each of the *E. aeneus* individuals to two

populations (blue or orange in Figure 3.3.5). The consensus solution for $K = 2$ again supports high levels of genetic connectivity in the studied region and gene flow due to migration between areas.

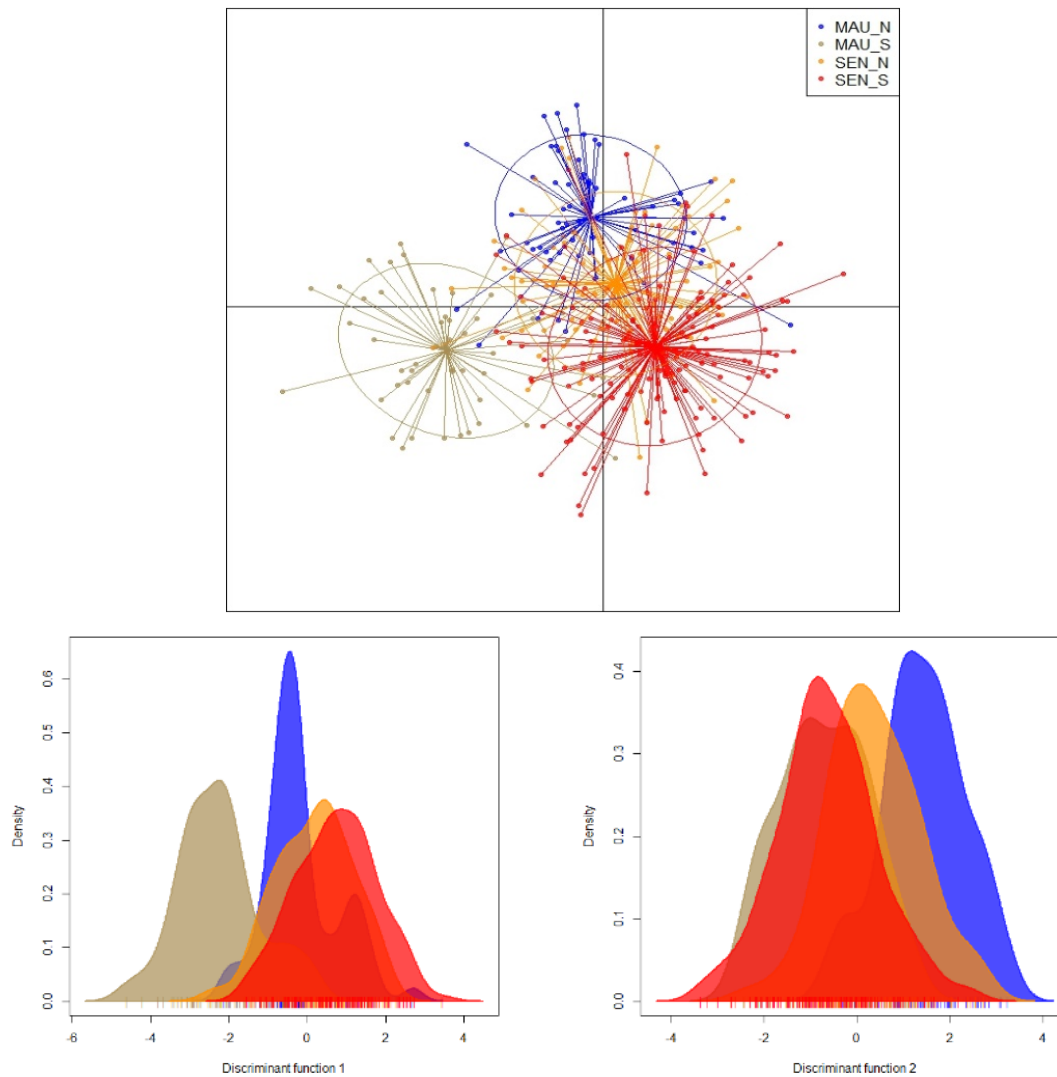


Figure 3.3.4.- Discriminant Analysis of Principal Components (DAPC) of *E. aeneus* genetic data in the studied country-areas: Mauritania and Senegal (North and South).

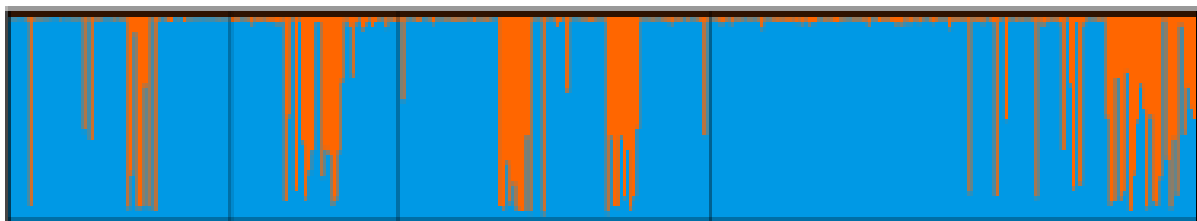


Figure 3.3.5.- Consensus solution for Bayesian approach to cluster genetic diversity of 354 *E. aeneus* (GPW) genotyped at 8 microsatellite loci and grouped in 4 country-areas: Mauritania and Senegal (North and South).

3.3.1.b *Penaes notialis* (SOP)

Characterization of SOP genetic diversity

Genomic DNA was obtained from 802 individuals of *P. notialis* (Figure 3.3.6). The quality of the DNA obtained was not homogeneous among all samples. For this reason, amplification was only obtained in 536 individuals.

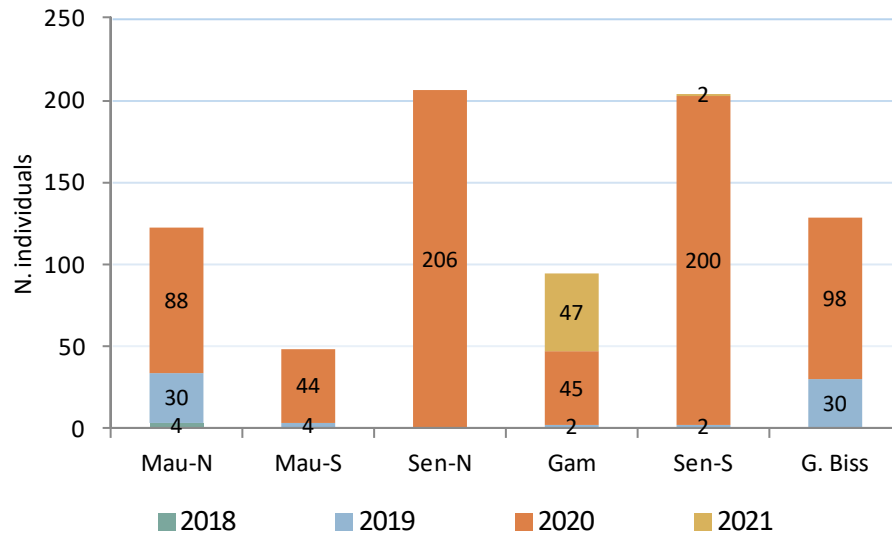


Figure 3.3.6.- Number of *P. notialis* (SOP) individuals sampled for genetics by country-area and year.

Molecular markers assessment

The final design allowed amplification of five microsatellite loci of *P. notialis* (Robainas-Barcia et al., 2002 and 2008) in one multiplex PCR (M1) and single PCRs M2 and M3 (PnS01 and PnS20).

Fragment analysis of the PCR products was performed using GeneScan™ 500 LIZ (Applied Biosystems, Foster City, CA, USA) as size standard. Alleles were called at 5 loci in 536 individuals of *P. notialis* (Figure 3.3.7) with software GeneMarker v2.7.0 (SoftGenetics). Table 3.3.6 shows the number (N) of individuals genotyped at each loci, and the total number of alleles (Na) counted.

Figure 3.3.7.- Allele calling for five microsatellite markers of *P. notialis* (SOP).

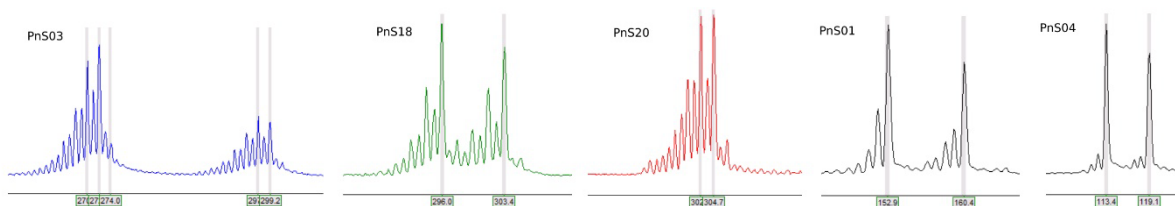


Table 3.3.6.- Characterization of genetic diversity by microsatellite and population (country-area).

Pop ^a	PnS03	PnS18	PnS01	PnS04	PnS20
MAU-N	55/25	94/36	96/15	97/12	93/93
MAU-S	27/17	44/28	45/12	43/10	44/65
SEN-N	102/40	154/43	157/16	150/13	147/114
GAM	24/15	36/32	46/13	47/11	42/56
SEN-S	79/29	121/39	128/15	123/13	127/103
G.BISS	38/25	52/32	59/13	54/11	55/58

^aPop refers to each area where SOP individuals were collected. Number of SOP individuals (N) and number of alleles (Na) characterized for each microsatellite loci. Each cell indicates N/Na. In red numbers, where Na is greater than N.

Microsatellite Pns20 exhibits a number of alleles higher than the number of individuals in four out of six study areas. This high variability exhibited by the genetic marker makes it difficult to interpret the results correctly, considering that 50 individuals per area were sampled in each of the sampling sessions carried out.

Two alternatives are proposed to solve this problem:

- The first is to replace the microsatellite marker with a less variable one, so that the total variability of the species could be recovered with the sample size available. So far, no another suitable marker has been found among those available in the scientific literature, that meets this condition, not presenting null alleles and with relatively simple allele calling.
- The second option is to increase the number of samples analysed. More than 100 individuals per sample should be analysed to ensure that all possible alleles are recovered, while samples for genetic were collected from 50 individuals per samples, in most cases.

That said, it would be necessary to design new markers or rethink a larger sampling design to obtain reliable data for this species. Nevertheless, basic diversity analyses were carried out with the results obtained and results are presented below (Table 3.3.7).

Table 3.3.7.- Estimation of genetic diversity of *P. notialis* by country-area.

Pop ^a	N	Na	Ne	H _O	H _E	uH _E
MAU-N	87	36	24	0.679	0.916	0.921
MAU-S	41	26	19	0.702	0.903	0.915
SEN-N	142	45	26	0.706	0.922	0.925
GAM	39	25	18	0.626	0.909	0.922
SEN-S	116	40	25	0.698	0.912	0.916
G.BISS	51	28	19	0.652	0.913	0.922

^aPop refers to each area where SOP were collected. Each cell indicates the average value per country-area.

- N=number of individuals analyzed;
- Na= number of different alleles;
- Ne= number of effective or equally frequent alleles;
- H_O=observed heterozygosity;
- H_E= expected heterozygosity according to allele frequencies;
- uH_E= Unbiased expected heterozygosity according to sample size.

The values obtained for F_{IS} and the probabilities associated to HW equilibrium are shown in Table 3.3.8. In all six areas significant deviations from HW proportions and a deficiency in heterozygotes (positive F_{IS}) were observed for locus PnS03, PnS18 and PnS20, meaning inbreeding. In nature, a deficiency of heterozygotes can be caused by the Wahlund effect. This effect is due to a mix of two sub-populations that mate mostly among themselves but overlap. Also, this can be explained by the presence of null alleles.

In this particular case, as we mentioned earlier is not possible to assess the actual cause of this deficiency.

Table 3.3.8.- Inbreeding coefficient F_{IS} for *P. notialis* by country-area.

Pop ^a	PnS03	PnS18	PnS01	PnS04	PnS20
MAU-N	0.8850***	0.1172***	0.0192*	0.1025	0.1610**
MAU-S	0.8452***	0.0170	0.0100	0.0840	0.1728**
SEN-N	0.8266***	0.0534**	-0.0592	0.1156**	0.2030***
GAM	0.9125***	0.1376**	0.1208	0.0587	0.3524***
SEN-S	0.7449***	0.0695*	0.0747	0.0785*	0.1977***
G.BISS	0.9183***	0.0984**	0.0126	0.0035	0.3748***

^aPop refers to each area where SOP were collected. Each cell indicates the value of the inbreeding coefficient F_{IS} calculated according to (Weir and Cockerham. 1984), followed by the significance level (* $p < 0.05$. ** $p < 0.01$. or *** $p < 0.001$) of the probability p value obtained with the exact probability test for Hardy–Weinberg equilibrium calculated by the Markov chain method (10.000 dememorization. 1.000 batches. 1.0000 iterations per batch).

Structuring of SOP genetic diversity

Structuring of genetic diversity of *P. notialis* was assessed through estimation of F_{ST} from multilocus genotypes of the species grouped into 6 populations or country-areas (Table 3.3.9) considering all five microsatellite loci.

All significant F_{ST} pairwise comparisons involved SAL population (SAL=SALOUM-Casamance. Senegal-South). As already mentioned, it is necessary to be cautious with the results obtained and to corroborate them with adequate sampling and more appropriate genetic markers.

The number of migrants estimated per locus is shown in Table 3.3.10. The average number of migrants is 27 implying high levels of gene flow and connectivity between sampling areas.

Table 3.3.9.- F_{ST} pairwise comparison of *P. notialis* genetic diversity at 5 loci in the six country-areas.

Pop ^a	MAU-N	MAU-S	SEN-N	GAM	SEN-S	G.BISS
MAU-N		0.569	0.193	0.006	0.000	0.004
MAU-S	0.006		0.005	0.007	0.011	0.006
SEN-N	0.004	0.565		0.004	0.007	0.004
GAM	0.626	0.947	0.839		0.010	0.007
SEN-S	0.008	0.000	0.000	0.009		0.010
G.BISS	0.848	0.898	0.478	0.883	0.000	

F_{ST} values below the diagonal. Probability. P (rand \geq data) based on 9999. Significant F_{ST} in red.

Table 3.3.10.- Characterization of number of migrants (Nm)

Locus	Nm
	All Pops
PnS03	20
PnS18	25
PnS01	48
PnS04	13
PnS20	31
Mean \pm SE	27 \pm 6

To assess the existence of putative barriers to gene flow, such as that detected in Senegal South (Table X34), clustering of genetic diversity was performed through Discriminant Analysis of Principal Components (DAPC) Figure 3.3.8.

Clustering of genetic diversity was studied through Bayesian algorithms (Figure 3.3.9) that assign individuals to a number of K clusters assuming an admixture model and considering sampling location of 536 *P. notialis* genotyped at 5 loci. The consensus solution for $K = 2$ again supports high levels of genetic connectivity in the studied region (blue colour) and gene flow due to migration between areas (orange).

Structure Harvester exploration of results indicated the best K was 2 and Clumpak produced the most frequent consensus solution for $K = 2$ proportionally assigning each of the *P. notialis* individuals to two “populations” (blue or orange). STRUCTURE software was used considering an ancestry admixture correlated allele frequency model and prior sampling location information. The image recovered from the Structure analysis shows some gradation in the presence of individuals with genotypes highlighted in orange. These individuals show introgression of genes from other populations. That is the longer the length of the bar, the higher the proportion of introgressed genes (the result of the presence of foreign individuals that have reproduced with the local ones). The most recent introgression is observed in the samples from Gambia and Guinea-Bissau.

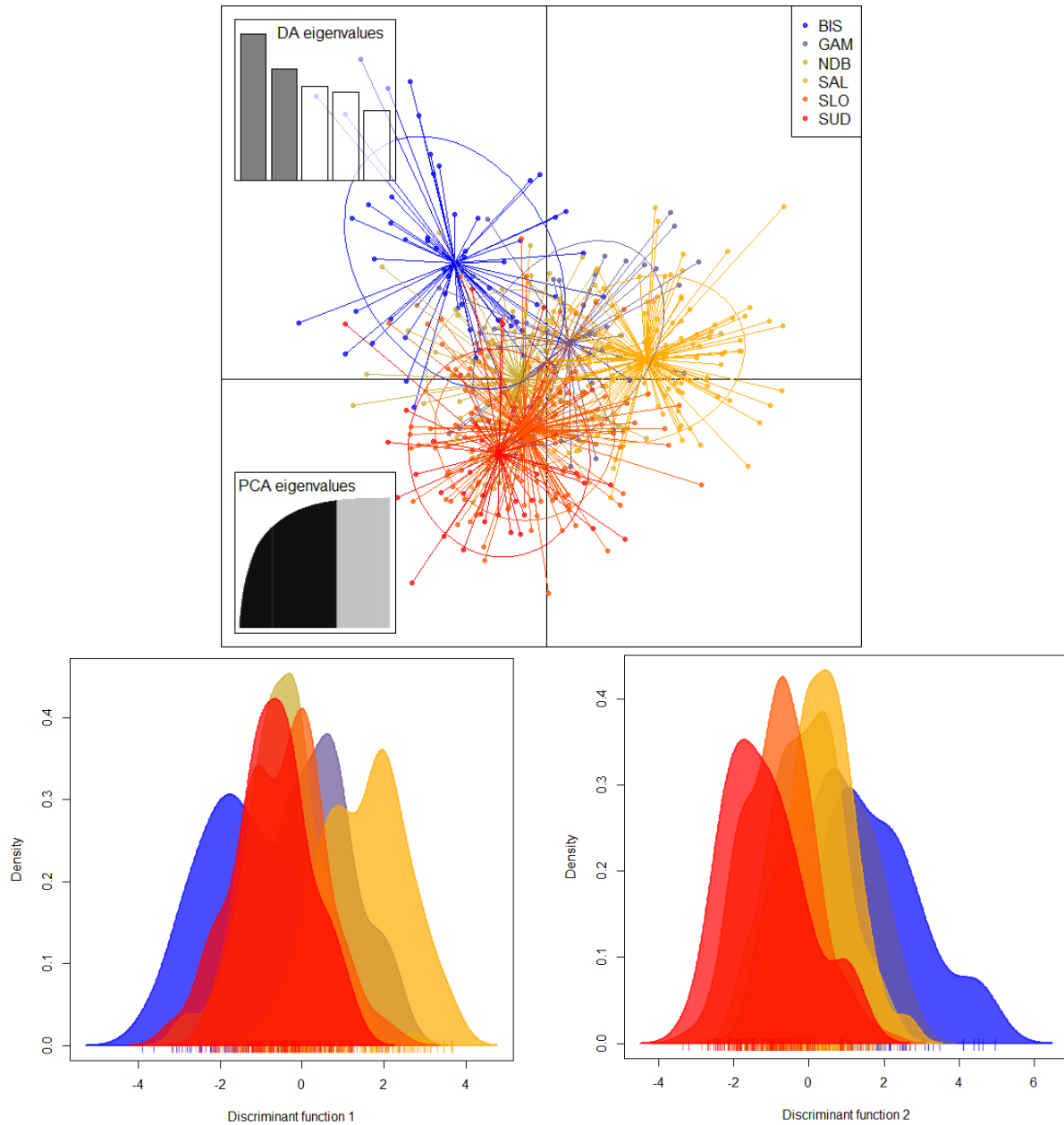
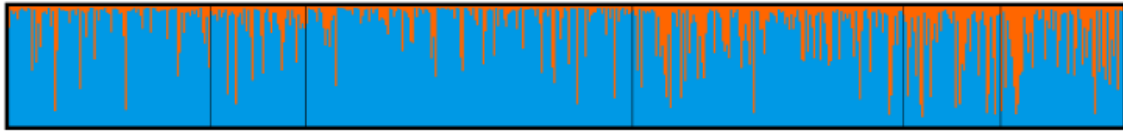
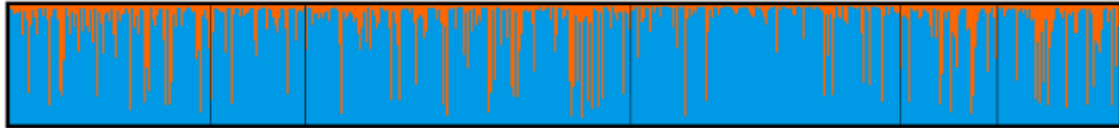


Figure 3.3.8.- Discriminant Analysis of Principal Components (DAPC) of *P. notialis* (SOP) genetic data in the studied country-areas: NDB (Mauritania-North), SUD (Mauritania South), SLO (Senegal-North), GAM (Gambia), SAL (Senegal-South), BIS (Guinea-Bissau).

K=2



K=2 MinorCluster1



K=2 MinorCluster2



MAU-N MAU-S SEN_N GAM SEN_S G.BISS

Figure 3.3.9.- Consensus solution for Bayesian approach to cluster genetic diversity of 536 *P. notialis* (SOP) genotyped at 5 microsatellite loci and grouped in 6 sampling areas.

3.3.2 Genetic information of new species

3.3.2.a *Epinephelus aeneus* (GPW)

For *Epinephelus aeneus* a 596 bp fragment of the cytochrome oxidase I (COI) gene was amplified and sequenced. The sequences were checked with the BLAST algorithm and exhibited 100% similarity and an e-value with a statistical significance of 0 suggesting evidence of homology. Of the 44 sequenced samples of *E. aeneus* a total of 27 were obtained. A total of 594 conserved sites and 2 parsimoniously informative variable sites were found. A translation (A-G) was observed in two samples from Senegal South at position 33 and a transversion (A-C) in two samples from Senegal South and 2 samples from Mauritania North at position 39 (Figure 3.3.10). The analysis revealed a nucleotide variation of A = 25.1 %; T = 29.9 %; C = 28 % and G = 17 %. Mean diversity was 0.0006 with an error of 0.0005 while evolutionary diversity was 0.0097 with an error of 0.0012.

	10 20 30 40 50
SAL_46_22	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
SAL_42_22	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
SAL_10_14	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
SAL_5_25_0	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
SAL_7_25_0	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
SAL_4_25_0	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
SAL_6_12_1	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
SAL_10_12	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
SAL_5_12_1	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
KAY_10_25	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
KAY_14_25	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
KAY_35_25	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
KAY_12_10	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
KAY_11_12	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
KAY_6_01_0	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
KAY_1_01_0	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
KAY_16_25	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
NRC_9_30_1	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
NRC_2_30_0	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
NRC_3_30_0	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
NRC_6_23_0	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
NRC_7_23_0	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
NRC_5_23_0	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
NDB_7_21_0	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
NDB_6_21_0	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
NDB_4_21_0	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
NDB_41_12	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
NDB-50_12	CGGGGGCTCT CCTTGGAGAC GACCAGATCT ATAACGTAAT TGTTACAGCA
Epinephelu	cggggagctct actaggogac gaccagatct ataatgtaat tgttacagca

Figure 3.3.10.- Partial sequence of the COI gene of *E. aeneus* (GPW). At positions 33 and 39 (red box) the variable sites of the sequences can be observed.

To infer the evolutionary history of *E. aeneus*, different phylogenetic trees were constructed. As an outgroup, a species close to this one, *Epinephelus coioides* (Hamilton, 1822) was chosen and the sequence was extracted from GenBank with the accession number KU722929. Models that best fit the nucleotide substitutions in the data were chosen and analyses were performed based on these models. First, we inferred using the ML method and the HKY model (Figure 3.3.11) with the highest log likelihood - 1112.16 and bootstrap value of 1000 replicates. Next, an analysis was performed with the NJ method and the p - distance model (Figure 3.3.12) in which the arm length was 0.12416107. Finally, a phylogenetic tree was calculated with the maximum parsimony method (Figure 3.3.13) in which the coherence index is 0.666667, the retention index is 0.750000 and the composite index is 0.740000. It can be seen that a polytomy is formed in all cases. By ML no significant differences are found between localities while with NJ and MP some kinship is observed between the populations of Mauritania North and Senegal South.

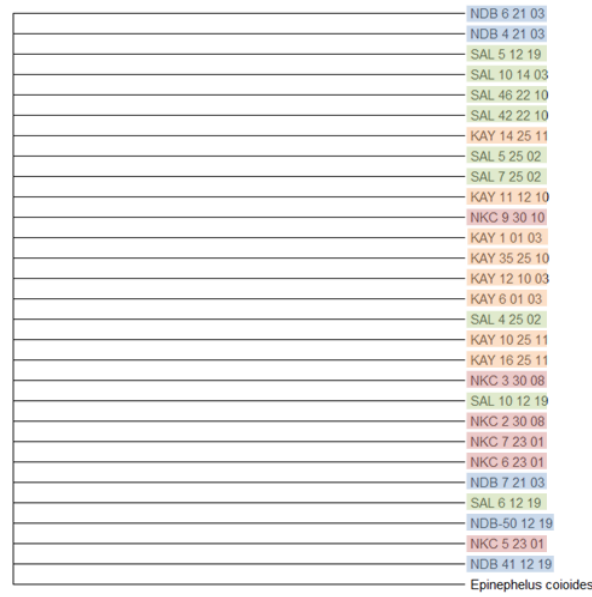


Figure 3.3.11.- ML phylogenetic tree based on the COI gene for 28 individuals of *E. aeneus* (GPW). Mau-N (blue), Mau_S (pink), Sen_N (orange), Sen-S (green). Constructed with the HKY model.

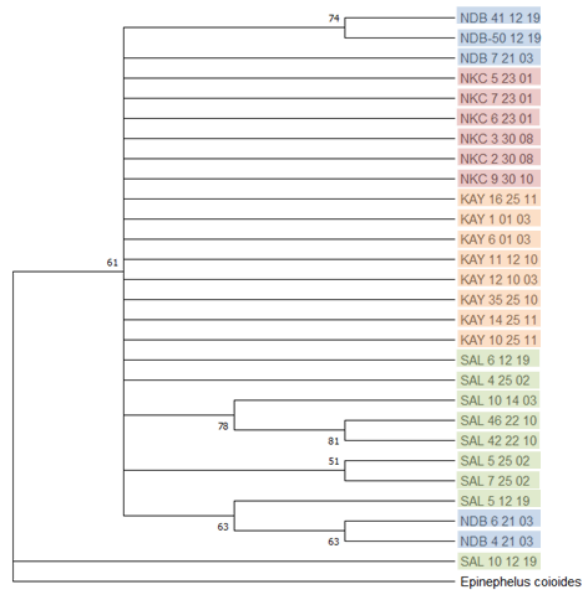


Figure 3.3.12.- NJ phylogenetic tree based on the COI gene for 28 individuals of *E. aeneus* (GPW). Mau-N (blue), Mau_S (pink), Sen-N (orange), Sen-S (green). Built with the p-distance model. The numbers of the branches are the bootstrap values.

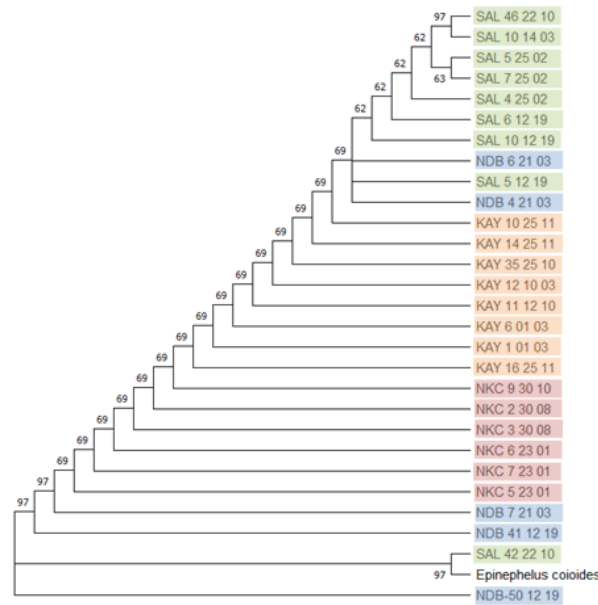


Figure 3.3.13.- Phylogenetic tree of MP based on the COI gene for 28 individuals of *E. aeneus* (GPW). Mau-N (blue), Mau-S (pink), Sen-N (orange), Sen-S (green). Bootstrap values are next to the branch. Performed by MEGA X software.

3.3.2.b *Penaeus notialis* (SOP)

For *Penaeus notialis* a 490 bp fragment of the COI gene was amplified and sequenced. The sequences were checked with BLAST showing a similarity between 98- 100 %. Of the 51 amplified sequences of *P. notialis*, reliable results were obtained from only 25 individuals. In this case, there were no variable sites, all 490 were conservable sites. The nucleotide variation frequency is A = 30.7 %; T = 30.7 %; C = 19.3 % and G = 19.3 %. Both evolutionary distance and mean and between-population diversity were 0.

Two different phylogenetic trees were inferred. *Penaeus vannamei* (Boone, 1931) was chosen as the outgroup. The sequence was obtained from GenBank with accession number MT607592.

An ML tree was constructed using the T92 model and the MEGA program. The highest log likelihood value was - 934.15 (Figure 3.3.14). An NJ tree was also constructed applying the T92 model. The sum of the branch value was 0.16113947 (Figure 3.3.15). In both cases, since so little variation was found between the sequences, it can be observed that a polytomy is generated.

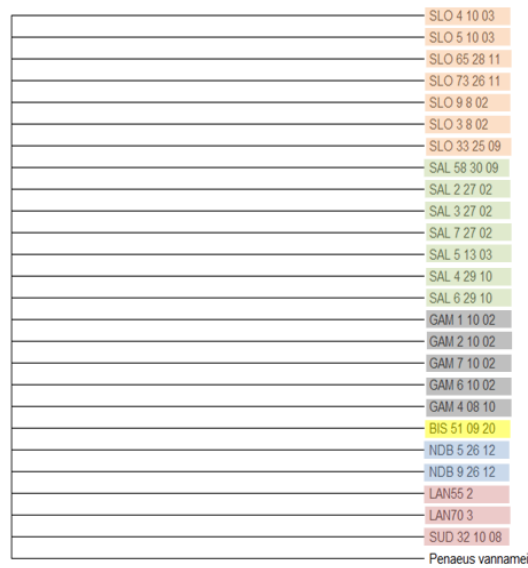


Figure 3.3.14.- ML phylogenetic tree based on the COI gene for 25 individuals of *P. notialis* (SOP), performed with the T92 model. Mau-N (blue), Mau-S (pink), Sen-N (orange), Sen-S (green), Gambia (gray) and G. Biss (yellow).

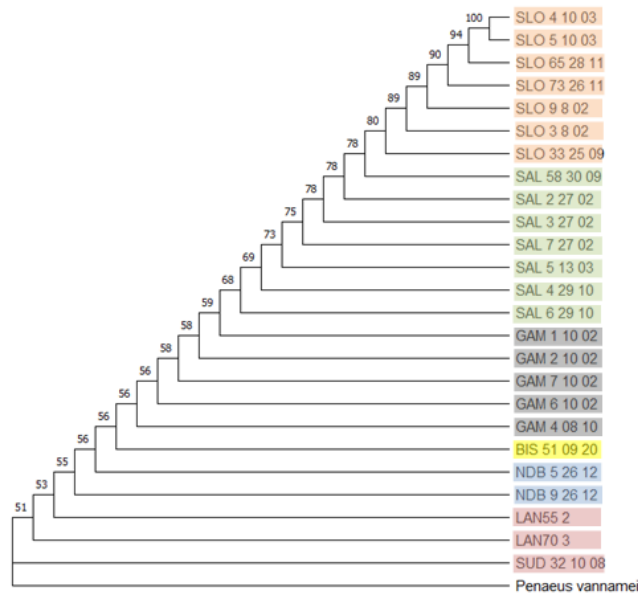


Figure 3.3.15.- NJ phylogenetic tree based on the COI gene for 25 individuals of *P. notialis* (SOP). In blue is represented Mauritania north. Mau-N (blue), Mau-S (pink), Sen-N (orange), Sen-S (green), Gambia (gray) and G. Biss (yellow). The bootstrap value is next to the branch.

3.3.2.c *Pseudotolithus senegalensis* (PSS)

Mitochondrial genome. Raw data mtDNA from the two samples sequenced generated about 2.2 Gb of reads length 150 bp. Samples 23 and 42 generated 14.119.178 and 15.285.286 reads respectively. The mtDNA of *P. senegalensis* is a closed-circular DNA molecule of 16.502 bp length (Figure 3.3.16). The nucleotide composition of the mtDNA is biased toward A+T nucleotides (52.1 %).

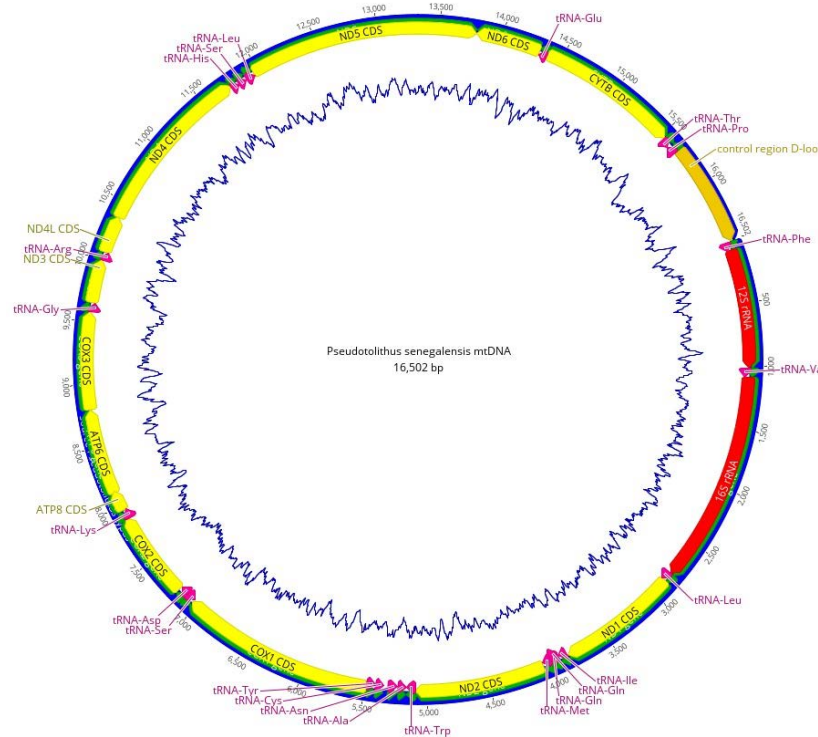


Figure 3.3.16.- Map of the *P. senegalensis* (PSS) mitochondrial genome. The inner ring indicates GC content of the genome.

The mtDNA of *P. senegalensis* contained 37 genes: 2 rRNA genes, 13 protein coding genes (PCGs), 22 tRNA genes and one control region D-loop (Figure 3.3.16). Characteristics of the *P. senegalensis* mitochondrial genome are shown in Table 3.3.11. All the proteins genes are coded in the heavy (H) DNA strand except the ND6, which is consistent with the arrangement of the mitochondrial genomes of vertebrates and especially mitochondrial genomes of fishes (Satoh et al., 2016).

Regarding the start codons in the 13 PCG, all of them are ATG, the most predominant among vertebrates. The inferred stop codons present in the *P. senegalensis* mitogenome are: TAA (ND1. ND2. ATP8. ATP6. COX3. ND4L and ND5). AGA (COX1. COX2 and ND4). TAG (ND3 and ND6). AGA (ND4) and GCT (CYTB). According to Satoh et al. (2016), except for the stop codons of the CYTB (GCT) and COX2 (AGA) genes, the rest are present in the mitochondrial genomes of fish. This point needs further study by comparing all fish mitochondrial genomes that have been sequenced up to the time of writing this report.

MITOS2 annotated several peculiarities: PSS_TAD_23: Split/duplicated features: OH and Overlaps: (atp6.atp8):10; (nad4.nad4l):7; (trnS2.cox1):5; (nad6.nad5):4; (trnR.nad3):2; (trnG.cox3):1; (cox3.atp6):1; (trnW.nad2):1; (trnM.trnQ):1; (trnQ.trnI):1 and in PSS_TAD_42: Overlaps: (atp8.atp6):10; (nad4l.nad4):7; (cox1.trnS2):5; (nad5.nad6):4; (nad3.trnR):2; (atp6.cox3):1; (cox3.trnG):1; (trnI.trnQ):1; (trnQ.trnM):1; (nad2.trnW):1.

Table 3.3.11.- Sequence characteristics of *Pseudotolithus senegalensis* (PSS) mitochondrial genome.

Locus name	From	To	Size	Strand	Number of aminoacids	Inferred init.codon	Inferred termin.codon	% GC	Intergenic nucleotides
tRNA-Phe	1	68	68	H				42.6	
12S-rRNA	69	1017	949	H				48.9	
tRNA-Val	1018	1089	72	H				50.0	
16S-rRNA	1090	2790	1701	H				46.8	
tRNA-Leu2	2791	2864	74	H				47.3	
ND1	2865	3839	975	H	324	ATG	TAA	49.2	
tRNA-Ile	3844	3913	80	H				45.7	1
tRNA-Gln	3913	1983	71	L				46.5	1 1
tRNA-Met	3983	4051	69	H				40.6	1
ND2	4052	5098	1047	H	348	ATG	TAA	49.3	1
tRNA-Trp	5098	5168	71	H				53.5	1
tRNA-Ala	5170	5238	69	L				38.2	
tRNA-Asn	5242	5314	73	L				52.1	
tRNA-Cys	5352	5417	66	L				48.5	
tRNA-Tyr	5418	5487	70	L				48.6	
COX1	5489	7045	1557	H	518	ATG	AGA	48.3	5
tRNA-Ser2	7041	7111	71	L				52.1	5
tRNA-Asp	7115	7183	69	H				49.3	
COX2	7192	7890	699	H	230	ATG	AGA	47.8	
tRNA-Lys	7883	7956	74	H				48.0	
ATP8	7958	8125	168	H	55	ATG	TAA	44.0	10
ATP6	8116	8799	684	H	227	ATG	TAA	49.3	10
COX3	8799	9584	786	H	262	ATG	TAA	48.6	1 1
tRNA-Gly	9584	9654	71	H				32.4	1 1
ND3	9655	10005	351	H	116	ATG	TAG	52.3	2
tRNA-Arg	10004	10072	69	H				34.8	2
ND4L	10073	10369	297	H	98	ATG	TAA	52.2	7
ND4	10363	11748	1386	H	461	ATG	AGA	50.6	7
tRNA-His	11744	11812	69	H				39.7	
tRNA-Ser1	11813	11880	68	H				58.8	
tRNA-Leu1	11886	11958	73	H				43.8	
ND5	11959	13797	1839	H	612	ATG	TAA	48.4	4
ND6	13794	14315	522	L	173	ATG	TAG	47.5	4
tRNA-Glu	14316	14384	69	L				40.6	
CYTB	14389	15529	1141	H	379	ATG	GCT	49.6	
tRNA-Thr	15530	15601	72	H				58.3	
tRNA-Pro	15606	15675	70	L				34.3	
D-loop	15676	16502	827	H				37.3	

- COX1, COX2 and COX3: cytochrome oxidase subunits I, II and III;
- CYTB: cytochrome b apoenzyme;
- ND 1-6. 4L: NADH dehydrogenase subunit 1-6. 4L;
- ATP6 and AATP8: ATP synthase subunits 6 and 8;
- 12S-rRNA: small ribosomal subunit RNA;
- 16S-rRNA: large ribosomal subunit RNA;
- tRNA: transfer RNAs specific for a single aminoacid;
- tRNA-Leu1 y L2: transfer RNA specific for Leucine differentiate codon recognized: CUN or UUR;
- tRNA-Ser1 and S2: transfer RNA specific for Serine differentiate codon recognized: AGN or UCN;
- D-loop: single large non-coding region

In a nutshell, ten pairs of protein-coding genes located directly adjacent to each other: ATP6-ATP8, ND6-ND5, tRNA-Gly-COX3, tRNA-Trp-ND2, ND4-ND4L, tRNA-Arg-ND3, tRNA-Gln-tRNA-Ile, trn-Ser2-COX1. COX3-ATP6 and tRNA-Met-tRNA-Gln present some overlap between adjacent genes (Table 3.3.11). Such overlaps have been observed in other mitochondrial genomes of fishes (Satoh et al. 2016) but until now this is the genome in which there is the highest number of genes with these overlaps.

A control region (D-loop) was found in the heavy strand (15676-16502 bp) with a length of 837 bp. This region possesses the main characteristics present in other mitochondrial control regions of fishes (Figure 3.3.17). Locations of the conserved sequence blocks domains and variable regions are mapped. The location of the T-homopolymer region is represented in broken lines (from Satoh et al. 2016).

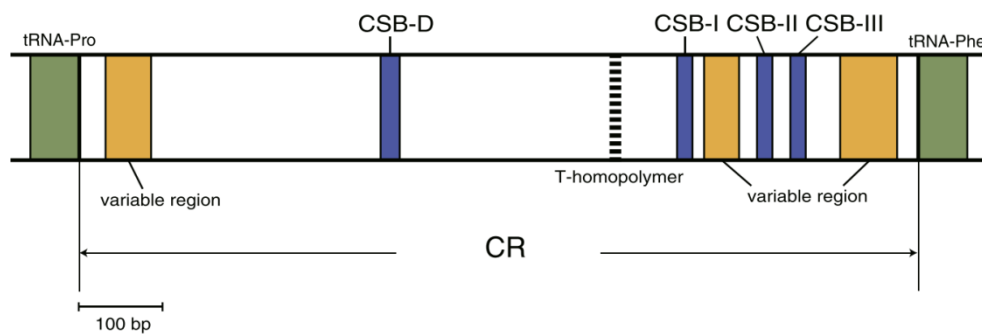


Figure 3.3.17.- Diagram of the control region of the fish mitochondrial genomes (CR).

Sequences alignment of mtDNA of different species of the Scianidae family were analyzed using the Geneious Prime 2023 software. *Conger erebennus* was chosen as outgroup. The phylogenetic tree of Sciaenidae (Figure 3.3.18) shows that *P. senegalensis* is phylogenetically closer to *P. typus* than to *P. elongatus*. For this reason, the identification of the individuals is considered correct. A more in-depth study of the relationships between the species of this genus in the area would be advisable.

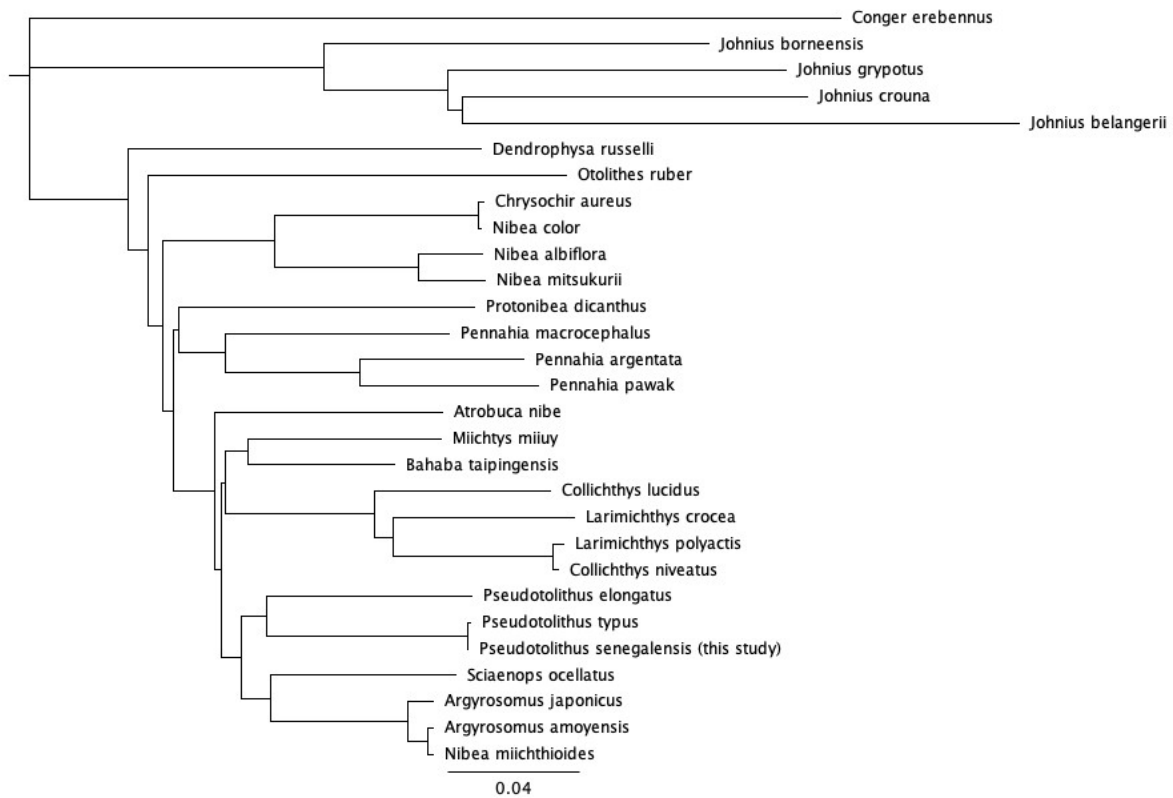


Figure 3.3.18.- Phylogenetic tree of *P. senegalensis* (PSS) mitochondrial genome relationship with other fish species of the Sciaenidae family.

3.3.2.d *Pagellus bellottii* (PAR)

Twenty-nine sequences from 40 samples from Ghana (18 from Takoradi in the West and 11 from Tema in the East) were obtained. The alignment comprises three other *P. bellottii* sequences (KJ012386.1, KY802044.1 and JN900485.1) and one *Pagellus natalensis* sequence (JF494050.1) retrieved from GenBank. The best model according to the Akaike criteria AICc was HKY (Hasegawa- Kishino-Yano, 1985).

An ML tree was inferred using the Hasegawa-Kishino-Yano model. The highest log likelihood value was - 947.73 (Figure 3.3.19). The bootstrap consensus tree inferred from 500 replicates (Felsenstein, 1985) is taken to represent the evolutionary history of the taxa analyzed. Initial trees for the heuristic search were obtained automatically by applying Neighbor-Join and BioNJ algorithms to a matrix of pairwise distances estimated using the Maximum Composite Likelihood (MCL) approach and then selecting the topology with superior log likelihood value. A discrete Gamma distribution was used to model evolutionary rate differences among sites (5 categories (+G. parameter = 200.0000)). This analysis involved 33 nucleotide sequences. All positions containing gaps and missing data were eliminated (complete deletion option). There were a total of 595 positions in the final dataset.

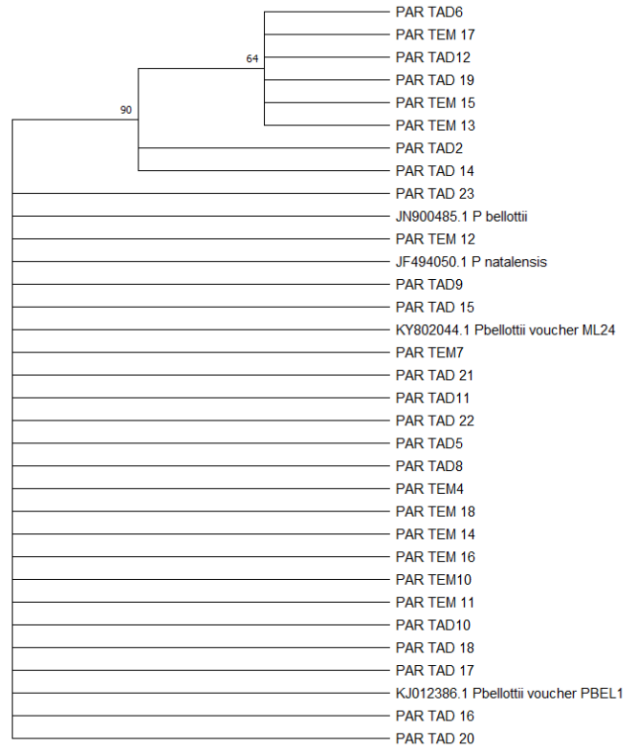


Figure 3.3.19.- Phylogenetic tree of *P. bellottii* (PAR) by ML method and Kimura 2-parameter model.

Although there are certain variable positions whereby eight of the sequences obtained are grouped into a cluster, no differentiation is observed between the two sampling areas of the species.

Even the sequence of *Pagellus natalensis*, used as an outgroup, is not differentiated from the rest and forms a polytomy with the rest. Further analysis by analysing a larger number of individuals is necessary to conclude anything. The objective of this section which was to obtain genetic information on this species in the area has been achieved.

4. DISCUSSION

STOCK IDENTIFICATION

A holistic approach has been used for stock identification of six target species of the three regional cases of studies: *Epinephelus aeneus* and *Penaeus notialis* (Mauritania and Senegal, including Gambia and Guinea-Bissau for *P. notialis*), *Pagrus caeruleostictus* and *Pseudotolithus elongatus* (Guinea-Bissau and Guinea) and *Pseudotolithus senegalensis* and *Pagellus bellottii* (Côte d'Ivoire and Ghana). Three main groups of techniques have been used: life-history traits, morphometry (body and otolith shape, the last for the five fish species) and genetics (for *E. aeneus* and *P. notialis*).

In general, life history traits techniques are complementary and provide basic information with different perspectives for a biological-based stock identification. Life history traits for certain species are analysed for the first time, while for others previous information available is updated.

Through the DEMERSTEM project, we have studied the stock structure of each of the five fish target species using truss network morphometric and otolith shape techniques, and of one shrimp species using truss network morphometrics. This is the first time this has been performed for these species and areas. Regarding body morphometrics, the availability of images of the individuals has allowed us to check in detail the situation of the landmarks in each specimen and to confirm some supposed anomalies and outliers (Cadrin and Friedland, 1999).

Genetics tools provide a direct basis for stock structuring and to interpret phenotypic-based patterns. It has the inconvenient of being expensive and the lack of microsatellite markers of four of the six target species.

“Thiof” *Epinephelus aeneus*

The studied life history traits do not show conclusive results in relation to stock identity for *E. aeneus*. Weight-related parameters, only available for females and sex combined, show differences scenarios (similarities or dissimilarities between Mauritania and Senegal), depending on the parameter considered. Thus, weight-length relationships are different between the two countries, being slightly but significantly higher in Mauritania than in Senegal. However, weight conversion factor for sex combined and the quarterly trends of the condition factor of mature females show similarities among the four studied areas, although some significant differences are found when we compare them among areas by quarters (Table 4.1). Thus, potential differences in weight-related parameters are not robust enough to assume the same or different stocks, although most of them show similar patterns for the two countries.

In relation to reproduction parameters, the spawning peaks in Mauritania and Senegal occur in a similar period, although they are more extended in Senegal. The length at first maturity is similar in the two Senegalese areas, although unfortunately it could not be estimated for Mauritania.

Regarding morphometry, LDA using cross-validation classification showed 55.0% and 39.3% correct classification of the *E. aeneus* individuals into their original populations in Mauritania and Senegal (North and South) using body morphometrics (truss network) and otolith shape respectively. The body morphometrics was more discriminating in identifying groups than the otolith shape, showing a difference of correct classification between both approaches of 15.7%. These percentages of correct classification are the lowest of all the species studied in the project. Taking into account the results from both approaches, relevant percentages of *E. aeneus* individuals from the four areas can be found in all of them, north and south Mauritania, north and south Senegal, indicating overlap of the study areas. This would indicate that the same phenotypic group of *E. aeneus* is occupying the four areas of both countries, Mauritania and Senegal. It could be due to a certain degree of interaction,

movements or possible migratory phenomena in individuals of the species in the studied area. A higher connectivity is found between both areas of Mauritania than between those of Senegal, with extensive overlap of the individuals from two Mauritanian areas.

For the first time, molecular markers were used to decipher the genetic structure of *E. aeneus* in the area. The combination of microsatellites and mitochondrial sequences has been useful in close species such as the wreckfish *Polyprion americanus* (Presa et al., 2023) to assess genetic divergence and connectivity among gene pools. There are many examples of the use of microsatellite markers applied to fisheries assessment, such as in European hake stocks (see, for instance, Pita et al., 2014, and 2017; Maggini et al., 2022) or the relationship among hake species of the genus *Merluccius* (Pérez et al., 2021).

The selected microsatellites used in the genetic study for *E. aeneus* have a high variability. The designed genetic tool (3 PCR reactions, 8 loci) is suitable for the assessment of connectivity in the species. All genetic analyses show high gene flow and homogeneity in the study area. Introgression detected with the Bayesian analysis performed with Structure suggests the presence of migrants from other regions in the area. In future studies it would be advisable to extend sampling to the areas surrounding our study area to determine the real impact of migration on the genetic diversity of the stock.

In addition to microsatellite analysis, representative of the nuclear genome, a 596 bp fragment of the mitochondrial cytochrome oxidase I (COI) gene was amplified and sequenced. The sequences were compared with those deposited in the GenBank database with the BLAST algorithm and showed 100% similarity. Phylogenetic analysis of 27 representative sequences from the study area has shown a total of 594 conserved sites and 2 parsimoniously informative variable sites. This gene has proven useful in closely related species to detect variants, subspecies or distinct species (Presa et al., 2023). In our case it confirms the homogeneity detected with microsatellites. All sequences obtained will be deposited in databases.

In view of these results (Table 4.1), both from the morphometry and genetic studies, a common stock of *E. aeneus* should be considered for Mauritania and Senegal. Thus, our study rejects the hypothesis of the existence of two stocks of thiof in Senegal, one north of the Cap Vert peninsula which performs a North-South migration and another in the south which would be sedentary (Cury and Worms, 1982; Cury, and Roy, 1988). However, it confirms the theory of one single stock between both countries, performing a great North-South migration along the coast (Champagnat and Domain, 1978; Laurans et al., 2003; Laurans, 2005), based on fishery information.

Thus, the consideration of a single stock of *E. aeneus* in Mauritania and Senegal is advised for stock assessment and management purpose. The thiof was already assumed as a single management unit for Mauritania, Senegal and the Gambia by CECAF, but with no biological basis for that. This study provides the biological phenotypic and genetic basis needed to confirm the existence of a single population. The extension of this study to longer periods and to northern and southern areas is highly recommended to determining appropriate geographic boundaries, needed to define the stock structure and distribution of *E. aeneus* in West African waters.

Southern pink shrimp *Penaeus notialis*

The studied life history traits do not show conclusive results in relation to stock identity for *P. notialis*, as both weight-related and reproduction parameters and features show different scenarios, this meaning similarities or differences among country-areas, depending on the parameter or feature considered (Table 4.1). It should be noted that in general life history traits of Guinea-Bissau are quite different than those of other areas. This might be due to the fact that the fraction of the population sampled in this country only corresponded to juveniles, while both juveniles and adults were

sampled in the other areas. In fact, no mature females were sampled in Guinea-Bissau commercial landings and the only ones analyzed in this study come from a scientific survey.

Considering the results obtained from morphometry, LDA using cross-validation classification showed 88.2% correct classification of the *P. notialis* individuals into their original populations using body shape (truss network). This percentage of correct classification is the highest obtained for body shape of all the species studied. Body morphometrics revealed that although the individuals from the studied areas show some overlapping, heterogeneity and spatial variations among sampling areas exist, with the presence of six groups, corresponding to each of the country-areas analyzed.

The overall morphometry results suggest the existence of six population units of *P. notialis* in the study area, northern Mauritania, southern Mauritania, northern Senegal, Gambia, southern Senegal and northern Guinea-Bissau. Our results imply an extension of the number of stock units considered so far in the area (three), without any known biological basis: one in the Banc d'Arguin (Mauritania); another in North Senegal (Saint Louis stock) located North Cape Verde and extending to South Mauritania (L'homme & Garcia, 1984); and the third shared between South Senegal and Guinea-Bissau (Roxo-Bijagos stock) (L'homme & Garcia, 1984). The Saint Louis stock is related to the Senegal River, while the Roxo-Bijagos stock is linked to Sine Saloum, Gambia, Casamance and Cacheu rivers. Juveniles would develop in these rivers mouths, migrating to the open water for reproduction. The main difference between the two Senegalese stocks is explained by lower freshwater inputs, weaker artisanal fishing and more limited nurseries, in the North (L'homme & Garcia, 1984). However, these reasons are not enough to biologically support the identity of the stocks. In our study, the overlap between these areas is very low, from the morphological point of view. It is also to be noted the lack of mixing between individuals from Gambia and Senegal-North, although it is a very small proportion of overlap between Gambia and South Senegal, which is logical considering that two sampling places of Senegal South (Saloum and Casamance) are located north and south Gambia, respectively. The existence of three independent populations in Senegal-Gambia might be explained by the different types of estuaries in the sampling areas. The Gambia estuary matches all the criteria of a "normal" estuary, characterized by Albaret (1999) as an estuary widely open to marine and freshwater domains, with a salinity range from freshwater to 39. Oppositely, the Sine Saloum delta, in South Senegal is considered an "inverse" estuary with nearly no freshwater input, relatively high transparency and salinity higher or equal to that of seawater (Simier et al., 2004; 2006). Apart from morphometric differences, some life history traits clearly differ from Gambia to North and South Senegal, that might be linked to the special characteristics of the Gambia estuary. The Gambia estuary is one of the last aquatic ecosystems of West Africa that has not yet been affected by strong environmental changes and human disturbances. It is moderately exploited by small-scale fisheries and does not receive any severe pollution from human activities. The Gambia River still has a natural flood regime with no dams or weirs. In contrast to the neighbouring Casamance and Sine Saloum estuaries, it has been free of major climatic perturbation and was less affected by the succession of drought periods experienced by the Sahel region in the last decades. Thus, ecological habitats for *P. notialis* in these estuaries explain the development of different populations, which might have adapted to different habitats.

Taking into account that there is a certain degree of mixture of individuals from Mauritania (North and South) and Senegal (North and South), four groups corresponding to each of the countries studied could be also considered for assessment and management purposes: Mauritania, Senegal, Gambia and Guinea-Bissau.

Microsatellite markers had been successfully applied to examine genetic diversity, population structure and connectivity of *P. notialis* in five localities distributed in the Caribbean Sea of Colombia, in the Western Atlantic Ocean (Atencia-Galindo et al., 2021). The authors interpret the genetic results obtained in relation to the life history of the species to determine how geomorphological variations on the continental shelf and marine currents influence the spatial distribution of the size

structure and genetic lineages of pink shrimp, as well as the implications of this type of study for the sustainable management of this valuable fishery resource.

The selected microsatellites applied to our samples of *P. notialis* for genetic analysis have shown extremely high variability. The genetic tool designed (3 PCR reactions, 5 loci) is neither suitable nor sufficient for the assessment of connectivity in the species. It would be very relevant to redesign sampling according to these results and to obtain new genetic markers appropriate to the observed variability. Our results are consistent with the findings of Atencia-Galindo et al. (2021). These authors state that a more precise methodological design is recommended for the development of genetic monitoring, using a greater number of specific molecular markers for this species, and increasing the sample size per locality. Due to the high structuring of the populations in this species and their great variability, as shown by the markers used in this project, it would be advisable to obtain new markers to complete the study or to use another type of marker, less variable and highly informative, such as SNPs (Single Nucleotide Polymorphisms).

For *Penaeus notialis* a 490 bp fragment of the COI gene was amplified and sequenced in 25 individual DNA samples. The sequences were checked with BLAST showing a similarity between 98- 100 %. All positions were identical.

On the basis of the results of morphometry, which are conclusive enough to take into account, stock assessment and management could be based on a more detailed resolution level than the current allocation of a single stock unit by country. This means considering these components of *P. notialis* as different populations or functional units (as e.g. those considered for other species of crustaceans, such as the Norway lobster (*Nephrops norvegicus*) in European waters separately (ICES, 2022). This might contribute to a more accurate knowledge of the stock status in order to achieve its sustainable exploitation. Ideally, these results should have been linked to those obtained from the genetic analysis, in order to confirm if the units identified are in fact independent stocks or subpopulations of the same stocks. Thus, completing the number of samples for genetics or looking for new genetic markers for *P. notialis* is highly recommended.

In addition, results show that the alternative consideration of one stock by each country is also feasible as the small mixture of individuals existing among areas mostly occurs within the same country: Mauritania, (North and South) and Senegal (North and South). It should be noted that Gambia constitutes an independent unit in relation to Senegal, as there is only a small overlap of the pink shrimps morphometry between Gambia and Senegal-South. Thus, taking into account the difficulties of obtaining fisheries statistics by areas in each country, the assessment and management by separate countries (Mauritania, Senegal, Gambia and Guinea-Bissau) seems feasible from the biological point of view.

Bluespotted seabream *Pagrus caeruleostictus*

Although the results of life history traits cannot be taken into account individually to assess the stock identity of *P. caeruleostictus* in Guinea and Guinea-Bissau, most parameters considered show significant differences, at least between Guinea and South Guinea-Bissau (Table 4.1), mainly those weight-related. However, there were also found similarities in the length of first maturity of the species in the southern areas of both countries. The spatial and temporal limitation of reproduction parameters and features involve the need to interpret these results with caution.

Results from morphometry are accurate and both methods used provide the same scenario (Table 4.1). LDA using cross-validation classification showed 84.7% and 60.36% correct classification of the *P. caeruleostictus* individuals into their original populations using body morphometrics (truss network) and otolith shape respectively. The body morphometrics was more discriminating in identifying groups than the otolith shape, showing a difference of correct classification between both approaches of 24.4%. Body morphometrics and otolith shape reveal that although the individuals

from the studied areas show some overlapping, heterogeneity and spatial variations among sampling areas exist, with the presence of two main groups, on the one hand the Guinea-Bissau individuals and on the other hand the Guinean individuals. In addition, the results of body shape also show that northern and southern individuals from Guinea could also be differentiated from each other. The small overlapping of individuals in the case of South Guinea-Bissau and Guinea might be explained by the fact that fishermen from the Tristan Island in North Guinea, where some samples were bought, sometimes develop their activity in South Guinea-Bissau (Diallo & Camara, *pers. comm.*).

The overall results from morphometry suggest the existence of two population units of *P. caeruleostictus* in the study area, Guinea-Bissau and Guinea, and connectivity at a short spatial scale evidenced by the higher overlap of the individuals from two areas of Guinea. The existence of two different populations seems to reveal limited movements of the species among areas, which differs to the reproductive long migration along the continental shelf of Mauritania and Senegal, described by Champagnat & Domain (1978) for this and other demersal species defined as “species with Saharan affinities”. These authors described a reproductive migration northwards during the warm season with return to the south in December. Their study was based on the seasonal variations in abundance and size structure of several demersal species in the Senegal artisanal fishery and along seven trawl transects between Cape Timiris in Mauritania and Cape Roxo in Senegal. In our study, the reproduction of *P. caeruleostictus* occurs all year around, at least in Guinean waters and thus, no migration processes linked to reproduction are observed, showing that the species might have a sedentary behavior, at least in waters off Guinea and Guinea-Bissau.

Based on the stock identity study of *P. caeruleostictus* in Guinea-Bissau and Guinea, we could advice on the stock assessment and management based on a more detailed resolution than the current consideration of one single stock of “seabreams” (Sparidae) for Guinea-Bissau, Guinea, Sierra Leone and Liberia, that follows practical reasons and has no any biological basis. At least, there is biological basis for considering the populations of *P. caeruleostictus* from Guinea-Bissau and Guinea, separately. This could contribute to a more accurate knowledge of the stock status in order to achieve its sustainable exploitation.

The extension of this study to longer periods and to the southern areas considered as the same stock that Guinea Bissau-Guinea by CECAF (Sierra Leona and Liberia) is highly recommended. In addition, improving the landing reporting at species level is a must to produce reliable assessments of the stocks.

Bobo croaker *Pseudotolithus elongatus*

The results of life history traits show inconsistencies in relation to the consideration of similar or independent biological units of *P. elongatus* in Guinea-Bissau and Guinea (Table 4.1). This might be partly due to the spatial and temporal limitations of biological sampling. Thus, although new biological information on the species is provided, this is not consistent enough to be used for stock identity.

However, results from the two morphometric techniques have revealed to be consistent for stock identification of this species in the considered area. LDA using cross-validation classification showed 68.2% and 51.6% correct classification of the *P. elongatus* individuals into their original populations using body morphometrics (truss network) and otolith shape, respectively. The body morphometrics was more discriminating in identifying groups than the otolith shape, showing a difference of correct classification between both approaches of 16.6%. Body and otolith shape reveal heterogeneity and spatial variations among sampling areas, with the presence of two main groups, on the one hand the Guinea-Bissau individuals and on the other hand the Guinea individuals. This heterogeneity is clear in spite that individuals from the studied areas show some overlapping, which in the case of North Guinea and South Guinea-Bissau can be explained by the potential fishery activity of Guinean

fishermen from the Tristan Island in waters off Guinea-Bissau (Diallo & Camara, *pers. comm.*), as explained above for *P. elongatus*.

Thus, the overall results from morphometry suggest the existence of two population units of *P. elongatus* in the study area: Guinea-Bissau and Guinea. In addition, connectivity at a short spatial scale was evidenced by the higher overlap of the individuals from the two areas of Guinea-Bissau on the one hand, and of those from two areas of Guinea, on the other hand. It should be noted that *P. elongatus* is an estuarine species, that is caught in the open continental shelf only during the rainy season except in areas where estuarine conditions occur throughout the year (Longhurst, 1969). In general, there is very little evidence to show the existence of movements or migrations of *P. elongatus*, although spawning movements of limited extent may occur (Longhurst, 1969), as it happens with similar croakers in the western tropical Atlantic (Lowe, 1962). Spawning only occurs in estuaries and occasionally very close inshore, generally in untrawlable shallow waters. This might explain the existence of independent stocks, linked to different river systems. In fact, Domain (1999), described concentration areas of this species near the main river outflows in the Guinean coastal zone: Compony (Kogon) and Nunez Rivers in the north, Konkouré River in the north of Conakry and Mellacorée River in the south.

Therefore, based on morphometry results it is recommended the stock assessment and management of *P. elongatus* in this area based on a more detailed resolution than the current consideration of one single stock unit for Guinea-Bissau, Guinea, Sierra Leone and Liberia, as it has been assessed by CECAF so far, following practical reasons. Now there is a biological basis which allows considering two components of *P. elongatus* population, one in Guinea-Bissau and another in Guinea, separately. This could contribute to a more accurate knowledge of the stock status in order to achieve its sustainable exploitation. In addition, we recommend the extension of this study to longer periods and to the southern countries considered as the same stock that Guinea Bissau-Guinea by CECAF (Sierra Leona and Liberia).

Cassava croaker *Pseudotolithus senegalensis*

The results of life history traits of *P. senegalensis* in Côte d'Ivoire and Ghana cannot be considered consistent for stock identification of this species in the area. While similarities were found between the two countries for some weight-related parameters, others show significant differences or limited information due to the sampling issues detected (Table 4.1). Although the information was not enough or consistent for the identification of spawning peaks that could be compared among country-areas, similar lengths at first maturity were estimated for the species in the eastern areas of both countries, this suggesting certain similarities in relation to reproduction parameters.

Oppositely, morphometry results provided very useful information in relation to the stock identity of the cassava croaker in the two countries of the Gulf of Guinea. LDA using cross-validation classification showed 67.9% and 61.3% correct classification of the *P. senegalensis* individuals into their original populations using body shape (truss network) and otolith shape respectively. The body morphometrics was more discriminating in identifying groups than the otolith shape, showing a difference of correct classification between both approaches of 6.7%. Body and otolith shape reveal that although the individuals from the studied areas show some overlapping, heterogeneity and spatial variations among sampling areas exist, with the presence of two main groups, on the one hand the Ivorian individuals and on the other hand the Ghanaian individuals.

Thus, the overall morphometry results suggest the existence of two population units of *P. senegalensis* in the study area, Côte d'Ivoire and Ghana. In addition, connectivity at short spatial scale is evidenced by the higher overlap of the individuals from the two areas of Côte d'Ivoire on the one hand, and the higher overlap of those from the two areas of Ghana on the other hand. As explained for *P. elongatus*, there is no evidence of seasonal movements or migrations apart from potential and limited extend spawning movements to deeper waters (Longhurst, 1969; Troadec,

1971; Okyere & Blay, 2020), this explaining the existence of independent stocks in each country. However, oppositely to *P. elongatus* that inhabits estuarine areas, *P. senegalensis* lives in the open shelf, at depths less than 50 m, hardly ever enters estuarine systems (Longhurst, 1969), and thus, it is more affected by coastal processes. It should be noted that the productivity in coastal areas off Côte d'Ivoire and Ghana is very much influenced by two coastal seasonal upwellings: one major upwelling during the boreal summer and one minor upwelling during the boreal winter (Roy, 1995). However, the influence of both upwelling differs among the two countries. The intensity of the minor upwelling has its maximum in the vicinity of Cape Palmas, at eastern waters than Côte d'Ivoire and sharply decreases toward the east to become almost unnoticeable in Ghanaian coastal waters (Cury & Roy, 2002). Oppositely, the extension over the continental shelf of the winter upwelling is much more pronounced off Ghana than off Côte d'Ivoire (Cury & Roy, 2002). In addition, the productivity linked to river outflows should be considered. Main rivers outflows in Côte d'Ivoire are from the Sassandra River in the west and from Bandama and Komoe rivers in the east, while the Volta River system, in east Ghana, is the main in this country. Differences in the upwelling processes and in the rivers outputs (mainly in the eastern areas) might have influenced in the development of different ecological conditions in both countries, to which the species should have adapted, resulting in two different populations.

One single stock of croakers (*Pseudotolithus* spp.) for Côte d'Ivoire, Ghana, Togo and Benin is currently considered by CECAF for assessment purposes, this following practical reason and with no biological basis so far. Therefore, based on the results from both morphometric techniques, stock assessment and management of *P. senegalensis* might be based on a more detailed resolution than the current one that is at least considering these components of *P. senegalensis* population, Côte d'Ivoire and Ghana, separately. This could contribute to a more accurate knowledge of the stock status in order to achieve its sustainable exploitation. The extension of this study to longer periods and areas, especially to those that are considered by CECAF as sharing the same stock that Côte d'Ivoire-Ghana (Togo and Benin) is highly recommended. In addition, improving the landing reporting at species level is a must to produce reliable assessments of the stocks.

Red pandora *Pagellus bellottii*

As occurring for other studied species, results from life history traits are not conclusive for stock identification. While similarities were found between the Ghana and Côte d'Ivoire for some reproductive parameters (i.e. length at first maturity of females in the two eastern areas of both countries), most weight related parameters show significant differences between the two countries (Table 4.1).

However, morphometry shows conclusive results. LDA using cross-validation classification showed 75.9% and 53.4% correct classification of the *P. bellottii* individuals into their original populations using body morphometrics (truss network) and otolith shape respectively. The body morphometrics was more discriminating in identifying groups than the otolith shape, showing a difference of correct classification between both approaches: 22.5%. Body morphometrics and otolith shape reveal that although the individuals from the studied areas show some overlapping, heterogeneity and spatial variations among sampling areas exist, with the presence of two main groups, on the one hand the Côte d'Ivoire individuals and on the other hand the Ghanaian individuals.

The overall results from morphometric studies suggest the existence of two population units of *P. bellottii* in the study area, Côte d'Ivoire and Ghana, and connectivity at short spatial scale evidenced by the higher overlap of the individuals from two areas of Côte d'Ivoire on the one hand, and the higher overlap of those from two areas of Ghana on the other hand. The existence of independent stocks might be related to the sedentary character of the species in this area, in which only seasonal migration to inshore waters (around 25 m depth) during the upwelling months are reported, while they become restricted to depths around 45-55 m during the period with hydrographic stable waters

and a sharp thermocline (Rijavec, 1973). A sedentary behaviour with stock differentiation by country (Guinea-Bissau and Guinea) has also been evidenced in the other sparid species studied here (*P. caeruleostictus*). In addition, the above mentioned ecological differences for *P. bellottii* existing between Côte d'Ivoire, linked to the two seasonal upwellings and the main river outflows in the eastern areas of the two countries, remain valid reasons to consider the adaptation of the species to the particularity of each country and thus, resulting in two different populations.

Therefore, based in these results, we recommend assessing and managing *P. bellottii* considering separately two population units of *P. bellottii*, one in Côte d'Ivoire and another in Ghana. This means that assessment and management should be based on a more detailed resolution level than the current single stock unit considered by CECAF for Côte d'Ivoire, Ghana, Togo and Benin, for assessment purposes. The assumption of one single stock for these four countries has followed practical reasons and had no any biological basis. This consideration of independent stocks by countries could contribute to a more accurate knowledge of the stock status in order to achieve its sustainable exploitation. We also recommend the extension of this study to longer periods and areas, especially to those that are considered by CECAF as sharing the same stock than Côte d'Ivoire-Ghana (Togo and Benin).

New genetic information on coastal demersal species from West Africa

In addition to the information on stock identification, this study has contributed to provide new genetic information of four of the studied species in West Africa. Some general inputs are given below although detailed information will be provided in articles that are expected to be produced in a short term.

In the case of *P. senegalensis*, none of the amplification conditions tested nor the primer pairs produced an amplicon amenable to sequencing. Visualization on agarose gel gave the image of an asymmetric PCR, which led us to suspect genome rearrangement. To identify the problem, it was decided to sequence the entire mitochondrial genome. As part of the DEMERSTEM project, the mitochondrial genome of *Pseudotolithus senegalensis* has been sequenced for the first time. Some peculiarities were detected, consistent with the amplification problems mentioned above. The complete mitochondrial genome will be deposited in the NCBI database and data will be released at the same time as the scientific publication. The information obtained in this project will be very useful in future projects in order to be able to unequivocally identify samples of this species.

For the red pandora *P. bellottii*, a fragment of the Cytochrome Oxidase I (COI) mitochondrial gene was sequenced to both, obtain genetic information and for its inclusion in the BOLD database of the Barcode of Life. Only one haplotype was detected, common to both *P. bellottii* and *Pagellus natalens*, used as an outgroup. The phylogenetic analysis recovers a polytomy. It would be advisable to sequence other markers, both mitochondrial and nuclear, with greater variation and to infer a new phylogeny with both types. The objective for *P. bellottii* which was to obtain genetic information on this species in the area has been achieved.

Conclusions

In summary, the use of holistic approach, using at least a genetic procedure and at least one phenotypic-based approach is very useful for integrated stock identification. Phenotypical characters may be more useful than only genetic ones, because a gene flow of small magnitude may prevent the detection of significant genetic differences. However, consistent morphometric differences between locations may indicate a population separation, and thus, the existence of different stock units as even if it was the case that those morphometric differences were caused by environmental influence, an extensive mixing of individuals from different locations would make them undetectable.

The two morphometric techniques used in our study (body shape-truss network and otolith shape) show reliable information for stock identification. Among the two morphometric methods, body shape has provided better results for differentiation among areas than otolith shape. Data from life history traits do not show conclusive results, as different sceneries could be provided for the same species depending on the selected parameter or feature. In addition, some differences in weight related parameters and/or reproductive parameters or features, found among areas could be attributed to sampling issues. However, this study has contributed to improve the biological knowledge of the species in the study area.

Morphometry and genetic results are consistent for the thiof *E. aeneus*, both showing one single stock in Mauritania and Senegal. This confirms the theory of the existence of one single stock for both countries, and provides the biological basis that was lacking. In addition, genetics show introgression from other areas. Oppositely, low estimated morphologic variability within samples of *P. notialis* suggests that each sampled country-zone forms a phenotypically homogeneous group, with clear differences to the others. However, we have not been able to confirm this genetically yet, as the number of genetic samples used is insufficient to recover the genetic diversity of the species. More individuals should be sampled and other markers need to be tried.

For the rest of species, the following phenotypically homogeneous groups can be distinguished by morphometry, at least at country level: *P. caeruleostictus* and *P. elongatus* (one stock in Guinea-Bissau and another in Guinea, for each species); *P. senegalensis* and *P. bellottii* (one stock in Côte d'Ivoire and one in Ghana, for each species).

In our study we have found evidences of several stocks which had previously been considered as one single stock. This has also been found in other species in northernmost Atlantic waters by using body shape and otolith shape analysis (Vasconcelos et al., 2018; Muniz et al., 2020). A finer resolution than a single stock model in its assessment and a precautionary approach in its management strategy were also recommended for those species.

The extension of this study to longer periods and to neighboring areas is highly recommended to determining appropriate geographic boundaries, needed to define the structure and distribution of these West African stocks.

Table 4.1.- Summary results obtained by species and technique for stock identity.

SPECIES	LIFE HISTORY TRAITS				MORPHOMETRY		GENETICS	
	Weight-related parameters		Reproduction		Body Shape	Otolith shape		
	Weight-length relationships	Weight conversion factor (sex combined)	Condition factor (quarterly trends of mature females)	Spawning peak (females)				L50 (females)
GPW	<p><u>Similarities between:</u> Two areas among countries. <u>Differences between:</u> Mauritania and Senegal. (For females and sex combined). Not enough data for males</p>	<p><u>Similarities between:</u> Two areas among countries. Mauritania and Senegal.</p>	<p><u>Similarities between:</u> Two areas among countries. Mauritania and Senegal.</p>	<p><u>Similarities between:</u> Two areas among countries. Mauritania and Senegal (more extended in Senegal)</p>	<p><u>Similarities between:</u> Two areas in Senegal. No estimations in Mauritania.</p>	<p>Same phenotypic group in the four country- areas (Mauritania and Senegal, both N and S).</p>	<p>Same phenotypic group in the four country- areas (Mauritania and Senegal, both N and S).</p>	<p>High gene flow and homogeneity in the the four country-areas (Mauritania and Senegal, both N and S).</p>
SOP	<p><u>Similarities between :</u> Two areas among countries (considering Senegal-Gambia). G. Bissau and Mauritania N, Senegal (N & S) (sex combined) <u>Differences between:</u> Mauritania N and Senegal, Gambia (for sex combined). Guinea-Bissau and Mauritania N, Gambia and Senegal S ; Senegal N and Gambia (for males) Mauritania N and Gambia (females)</p>	<p>No data on gutted weighth for this species</p>	<p><u>Similarities between:</u> Mauritania N and Senegal (incompleted data from Mauritania S) <u>Differences between:</u> G. Bissau and the other areas</p>	<p><u>Differences</u> among country-areas in time and extension of two spawning peaks. Lacking information from Mauritania S and G. Bissau.</p>	<p><u>Similarities between:</u> Mauritania-North, Gambia and Senegal-South. <u>Differences between:</u> Senegal North and Mauritania-North, Gambia, Senegal-South. No estimations in Mauritania S and G. Bissau.</p>	<p>Six morphologically different groups: _Mauritania N _Mauritania S _Senegal N _Gambia _Senegal S _G.Bissau</p>	<p>-</p>	<p>No results</p>
BSC	<p><u>Similarities between:</u> Two areas in Guinea (males and females). <u>Differences between:</u> G. Bissau S and Guinea (males, females and sex combined)</p>	<p><u>Similarities between:</u> Two areas in Guinea. <u>Differences between:</u> G. Bissau S and Guinea.</p>	<p><u>Similarities between:</u> G. Bissau S and Guinea N (trends) <u>Differences between:</u> Lower values in G. Bissau.</p>	<p><u>Similarities between:</u> Two areas in Guinea and G. Bissau. Lacking information from G. Bissau S.</p>	<p><u>Similarities between:</u> G. Bissau S and Guinea S. Lacking information from Guinea N.</p>	<p>Three morphologically different groups: _G.-Bissau _Guinea-North _Guinea-South</p>	<p>Two morphologically different groups: _G.Bissau _Guinea</p>	<p>-</p>
PSE	<p><u>Similarities between:</u> All areas (sex combined) G. Bissau S and Guinea (males and females, separately) G. Bissau N and Guinea (females) <u>Differences between:</u> G. Bissau N and S (females and males) G. Bissau N and Guinea S (females and males) G. Bissau N and Guinea N (males)</p>	<p><u>Similarities between:</u> Two areas in G. Bissau. G. Bissau and Guinea. <u>Differences between:</u> Two areas in Guinea</p>	<p><u>Similarities between:</u> Two areas in Guinea. <u>Differences between G. Bissau and Guinea</u> (incompleted data from G. Bissau N)</p>	<p><u>Similarities between:</u> Two areas in Guinea and G. Bissau S. Lacking information from G. Bissau N.</p>	<p><u>Similarities between:</u> G. Bissau N and Guinea N No estimations from G. Bissau S and Guinea S.</p>	<p>Two morphologically different groups: _G.Bissau _Guinea</p>	<p>Two morphologically different groups: _G.Bissau _Guinea</p>	<p>-</p>
PSS	<p><u>Similarities between:</u> C. Ivoire and Ghana E (males, females and sex combined) <u>Differences between:</u> Ghana W and other 3 areas (for males, females and sex combined) --> Attributable to sampling issues.</p>	<p><u>Similarities between:</u> Two areas among countries. C. Ivoire and Ghana</p>	<p><u>Similarities between:</u> C. Ivoire E and Ghana E (incompleted data from the W areas)</p>	<p>Not enough or consistent information.</p>	<p><u>Similarities between:</u> C. Ivoire W and Ghana</p>	<p>Two morphologically different groups: _C. Ivoire _Ghana</p>	<p>Two morphologically different groups: _C. Ivoire _Ghana</p>	<p>-</p>
PAR	<p><u>Similarities between:</u> Two areas among countries (females and sex combined) C. Ivoire E and Ghana W (males and sex combined) C. Ivoire W and Ghana & C. Ivoire E and Ghana W (females) <u>Differences between:</u> C. Ivoire W and Ghana (males and sex combined) C. Ivoire E and Ghana E (males, females and sex combined) Two areas in C. Ivoire (males)</p>	<p><u>Similarities between:</u> Two areas in Ghana. C. Ivoire E and Ghana <u>Differences between:</u> Two areas in C. Ivoire. C. Ivoire W and Ghana.</p>	<p><u>Differences between:</u> C. Ivoire E and Ghana (incompleted data from C. Ivoire W)</p>	<p><u>Similarities between:</u> C. Ivoire E and Ghana Lacking information from C.Ivoire W</p>	<p><u>Similarities between:</u> C. Ivoire E and Ghana E No estimations from Ghana W.</p>	<p>Two morphologically different groups: _C. Ivoire _Ghana</p>	<p>Two morphologically different groups: _C. Ivoire _Ghana</p>	<p>-</p>

Similar between country-areas
Different between country-areas
Similarities and differences between country-areas
No conclusive findings

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6.- ANNEXES

ANNEX 1- Sampling carried out by species, country, area and month/year. BIO=biological sampling, GEN= genetic sampling, MOR= body pictures for morphometry. NP= not planned.

Table 1.1.- *Epinephelus aeneus* (GPW)

COUNTRY	Area/Month	Period 2					Period 1					Period 2							
		Aug 19	Sep 19	Oct 19	Nov 19	Dec 19	Jan 20	Feb 20	Mar 20	Apr 20	May 20	Jun 20	Jul 20	Aug 20	Sep 20	Oct 20	Nov 20	Dec 20	Jan 21
MAURITANIA	NORTH (Nouadhibou)	-	BIO	-	-	BIO-GEN	BIO-GEN	BIO	BIO-MOR-GEN	BIO	BIO	BIO	BIO	BIO	BIO	BIO-MOR-GEN	BIO	-	-
	SOUTH (Nouakchott)	BIO-GEN	-	-	-	-	BIO-GEN	-	BIO-MOR-GEN	-	-	BIO	BIO	BIO	BIO	BIO-MOR-GEN	-	-	-
SENEGAL	NORTH (Kayar)	-	-	BIO	BIO	BIO-MOR-GEN	BIO	BIO-GEN	BIO-MOR-GEN	-	BIO	BIO	BIO	BIO	BIO	BIO-GEN	BIO-GEN	BIO	BIO
	SOUTH (Saloum)	-	BIO	BIO	BIO	BIO-MOR-GEN	BIO	BIO-MOR-GEN	BIO-MOR-GEN	-	BIO	BIO	BIO	BIO	BIO	BIO-MOR-GEN	BIO	BIO	BIO

Table 1.2.- *Penaeus notialis* (SOP)

COUNTRY	Area/Month	Period 1					Period 2													
		Sep 19	Oct 19	Nov 19	Dec 19	Jan 20	Feb 20	Mar 20	Apr 20	May 20	Jun 20	Jul 20	Aug 20	Sep 20	Oct 20	Nov 20	Dec 20	Jan 21	Feb 21	Mar 21
MAURITANIA	NORTH (Nouadhibou)	-	-	-	BIO-GEN	BIO	BIO-MOR-GEN	BIO-MOR-GEN	BIO	BIO	BIO	BIO	BIO-MOR-GEN	BIO	BIO	BIO	-	-	NP	NP
	SOUTH (Nouakchott)	-	-	-	-	-	-	-	-	-	-	-	BIO-MOR-GEN	-	-	-	-	-	NP	NP
SENEGAL-GAMBIA	NORTH (Saint Louis)	-	BIO-MOR-GEN	BIO	BIO	BIO	BIO-MOR-GEN	BIO-MOR-GEN	-	BIO	BIO	BIO	BIO	BIO-MOR-GEN	BIO-MOR-GEN	BIO-GEN	BIO	-	NP	NP
	NORTH (Kayar + Dakar in Dec19-Feb 20)	-	BIO	BIO	BIO	BIO	BIO	BIO	BIO	BIO	BIO	BIO	BIO	BIO	BIO	BIO	BIO	BIO	NP	NP
	GAMBIA	-	-	-	-	-	-	-	-	-	-	BIO	BIO-MOR-GEN	BIO	BIO-GEN	BIO-GEN	BIO	BIO	BIO-MOR-GEN	BIO
	SOUTH (Saloum + Casamance in Sept-Oct 19)	BIO-MOR-GEN	BIO	BIO	BIO	BIO	BIO-MOR-GEN	BIO-GEN	-	-	BIO	BIO	BIO	BIO-MOR-GEN	BIO-MOR-GEN	BIO	BIO-GEN	BIO	NP	NP
GUINEA-BISSAU	NORTH (Cacheu)	-	-	-	-	-	-	BIO-MOR-GEN	BIO	BIO	BIO	BIO	BIO	BIO-MOR-GEN	BIO	BIO	BIO	BIO	BIO	NP

Table 1.3.- *Pagrus caeruleostictus* (BSC)

COUNTRY	Area/Month	Period 1					Period 2													
		Sep 19	Oct 19	Nov 19	Dec 19	Jan 20	Feb 20	Mar 20	Apr 20	May 20	Jun 20	Jul 20	Aug 20	Sep 20	Oct 20	Nov 20	Dec 20	Jan 21	Feb 21	
GUINEA-BISSAU	NORTH (Bissau)	-	-	BIO	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	NP
	SOUTH (Buba)	BIO	BIO	BIO	BIO	BIO-MOR-GEN	BIO	BIO	BIO	BIO	BIO	BIO	BIO-MOR-GEN	BIO	-	BIO	BIO	BIO	BIO	BIO
GUINEA	NORTH (Kamsar)	-	BIO	BIO	BIO	BIO-MOR-GEN	BIO	BIO-MOR-GEN	BIO	BIO	BIO	BIO	BIO	BIO-MOR-GEN	BIO-MOR-GEN	BIO-MOR-GEN	-	BIO-MOR-GEN	NP	
	SOUTH (Conakry)	-	BIO	BIO	BIO	BIO-MOR-GEN	BIO-MOR-GEN	BIO-MOR-GEN	BIO	BIO	BIO	BIO	BIO	BIO-MOR-GEN	BIO-MOR-GEN	BIO-MOR-GEN	-	BIO-MOR-GEN	NP	

Table 1.4.- *Pseudotolithus elongatus* (PSE)

COUNTRY	Area/Month	Period 1												Period 2					
		Sep 19	Oct 19	Nov 19	Dec 19	Jan 20	Feb 20	Mar 20	Apr 20	May 20	Jun 20	Jul 20	Aug 20	Sep 20	Oct 20	Nov 20	Dec 20	Jan 21	Feb 21
GUINEA-BISSAU	NORTH (Cacheu)	BIO	BIO	BIO	BIO	BIO-MOR	BIO	-	-	-	-	BIO	-	BIO	BIO	BIO-MOR	BIO	BIO	BIO
	SOUTH (Cacine, Buba in Sept. 20)	BIO	BIO	BIO	BIO	BIO-MOR	BIO	BIO	BIO	BIO	BIO	BIO	BIO	BIO	BIO	BIO-MOR	BIO	BIO	BIO
GUINEA	NORTH (Kamsar)	-	BIO	BIO	BIO	BIO	BIO	BIO-MOR	BIO	BIO	BIO	BIO	BIO	BIO	BIO-MOR	BIO-MOR	NP	NP	NP
	SOUTH (Conakry)	BIO	BIO	BIO	BIO	BIO	BIO	BIO-MOR	BIO	BIO	BIO	BIO	BIO	BIO-MOR	BIO-MOR	BIO-MOR	NP	NP	NP

Table 1.5.- *Pseudotolithus senegalensis* (PSS)

COUNTRY	Area/Month	Period 2					Period 1							Period 2					
		Oct 19	Nov 19	Dec 19	Jan 20	Feb 20	Mar 20	Apr 20	May 20	Jun 20	Jul 20	Aug 20	Sep 20	Oct 20	Nov 20	Dec 20	Jan 21	Feb 21	Mar 21
CÔTE D'IVOIRE	WEST (San Pedro)	-	-	-	-	-	BIO	-	MOR	-	-	BIO	BIO	BIO-MOR	BIO	BIO	BIO	BIO	-
	EAST (Abidjan)	-	-	-	-	-	BIO	-	BIO-MOR	BIO	BIO	BIO	BIO	BIO-MOR	BIO	BIO	BIO	BIO	-
GHANA	WEST (Takoradi)	BIO-MOR-GEN	BIO	BIO	-	-	-	-	BIO-MOR	-	BIO	BIO	BIO	GEN	BIO	BIO	-	BIO-MOR	-
	EAST (Tema)	BIO-MOR-GEN	BIO	BIO	BIO	-	-	-	BIO-MOR	BIO	BIO	BIO	-	-	BIO-MOR	BIO	-	BIO-MOR	BIO

Table 1.6.- *Pagellus bellottii* (PAR)

COUNTRY	Area/Month	Period 1												Period 2					Period 1	
		Oct 19	Nov 19	Dec 19	Jan 20	Feb 20	Mar 20	Apr 20	May 20	Jun 20	Jul 20	Aug 20	Sep 20	Oct 20	Nov 20	Dec 20	Jan 21	Feb 21	Mar 21	
CÔTE D'IVOIRE	WEST (San Pedro)	-	-	-	-	-	BIO	-	-	-	-	BIO	BIO-MOR	BIO	BIO-MOR	BIO	BIO	BIO	-	
	EAST (Abidjan)	-	-	-	-	-	BIO	-	BIO-MOR	BIO	BIO	BIO	BIO	BIO	BIO-MOR	BIO	BIO	BIO	-	
GHANA	WEST (Takoradi)	BIO	BIO	BIO-GEN	-	-	-	-	BIO-MOR	BIO	BIO	BIO	BIO	-	BIO-MOR	BIO	-	BIO	BIO	
	EAST (Tema)	BIO	BIO	BIO-GEN	BIO	-	-	-	BIO-MOR	BIO	BIO	BIO	BIO	-	BIO-MOR	BIO	-	BIO-MOR	BIO	

ANNEX 2- Number of individuals sampled for life history traits, by species, length range, sex and country-zone.

Table 2.1.- *Epinephelus aeneus* (GPW)

MALES					
Length range (cm)	Mau_N	Mau_S	Sen_N	Sen_S	Total
<28			14	20	34
28-32			10	14	24
32-36			3	7	10
36-40			8	8	16
40-44		1	2	6	9
44-48		2	2	3	7
48-52			4	2	6
52-56					0
56-60	1		3	1	5
60-64		1			1
64-68		2			2
68-72					
72-76	2				2
76-80	2				2
>80	1				1
Total	6	6	46	61	119
FEMALES					
Length range (cm)	Mau_N	Mau_S	Sen_N	Sen_S	Total
<28		3	11	29	43
28-32	5	4	15	51	75
32-36	14	6	32	42	94
36-40	31	22	72	47	172
40-44	33	29	49	63	174
44-48	61	27	46	54	188
48-52	60	20	52	50	182
52-56	57	21	70	46	194
56-60	51	14	57	29	151
60-64	57	7	47	31	142
64-68	61	12	33	22	128
68-72	72	8	15	16	111
72-76	75	6	4	14	99
76-80	52	4	3	11	70
>80	30	3	0	6	39
Total	659	186	506	511	1862
SEX-COMBINED					
Length range (cm)	Mau_N	Mau_S	Sen_N	Sen_S	Total
<28		3	26	80	109
28-32	5	4	25	78	112
32-36	14	11	35	58	118
36-40	32	23	81	57	193
40-44	33	30	51	69	183
44-48	64	29	48	57	198
48-52	61	22	56	52	191
52-56	61	23	70	46	200
56-60	54	14	60	30	158
60-64	63	8	47	31	149
64-68	66	14	33	22	135
68-72	79	8	15	16	118
72-76	80	6	4	14	104
76-80	54	4	3	11	72
>80	31	3	0	6	40
Total	697	202	554	627	2080

Table 2.2.- *Penaeus notialis* (SOP)

MALES							
Length range (LC, mm)	Mau_N	Mau_S	Sen_N	Gam	Sen_S	G.Biss	Total
10-14			2		2	14	18
14-18	1		18	9	22	215	265
18-22	26	7	126	65	157	180	561
22-26	125	50	167	84	130	160	716
26-30	227	21	129	118	72	91	658
30-34	135	6	32	49	45	11	278
34-38	30	1	11	7	12	3	64
38-42	1	1					2
42-46	1				1		2
Total	546	86	485	332	441	674	2564
FEMALES							
Length range (LC, mm)	Mau_N	Mau_S	Sen_N	Gam	Sen_S	G.Biss	Total
10-14			2		2	47	51
14-18			42	11	30	303	386
18-22	4	3	91	51	76	206	431
22-26	69	13	137	44	124	126	513
26-30	154	19	196	56	151	146	722
30-34	135	24	185	105	134	62	645
34-38	139	31	160	117	139	30	616
38-42	84	15	132	50	119	1	401
42-46	49	7	58	6	79		199
46-50	46	2	36		62		146
50-54	21		12		28		61
54-58	2		2		3		7
58-62					1		1
Total	703	114	1053	440	948	921	4179
SEX-COMBINED							
Length range (LC, mm)	Mau_N	Mau_S	Sen_N	Gam	Sen_S	G.Biss	Total
10-14			4		4	61	69
14-18	1		65	21	53	518	658
18-22	30	10	224	116	234	386	1000
22-26	194	63	304	128	255	286	1230
26-30	381	40	325	174	223	237	1380
30-34	270	30	217	154	179	73	923
34-38	169	32	171	124	151	33	680
38-42	85	16	132	50	119	1	403
42-46	50	7	58	6	80		201
46-50	46	2	36		62		146
50-54	21		12		28		61
54-58	2		2		3		7
58-62					1		1
Total	1249	200	1550	773	1392	1595	6759

Table 2.3.- *Pagrus caeruleostictus* (BSC)

MALES				
Length range (cm)	G.Biss_S	Gui_N	Gui_S	Total
9-12	3			3
12-15	4		1	5
15-18	97	7	19	123
18-21	133	69	101	303
21-24	121	63	169	353
24-27	56	85	232	373
27-30	20	95	77	192
30-33	7	74	53	134
33-36	3	45	55	103
36-39		20	18	38
39-42		10	11	21
42-45			1	1
Total	444	468	737	1649
FEMALES				
Length range (cm)	G.Biss_S	Gui_N	Gui_S	Total
9-12	2			2
12-15	6		4	10
15-18	72	15	24	111
18-21	170	90	135	395
21-24	136	102	189	427
24-27	38	127	113	278
27-30	19	82	49	150
30-33	5	56	42	103
33-36	1	25	34	60
36-39		13	13	26
39-42		7	7	14
42-45		2	4	6
45-48		2		2
48-51		3		3
54-57			1	1
Total	449	524	615	1588
SEX-COMBINED				
Length range (cm)	G.Biss_S	Gui_N	Gui_S	Total
6-9		2		2
9-12	8	13	5	26
12-15	15	41	58	114
15-18	200	84	142	426
18-21	327	204	285	816
21-24	271	170	370	811
24-27	97	216	350	663
27-30	39	183	126	348
30-33	14	132	95	241
33-36	4	73	89	166
36-39		33	33	66
39-42		17	18	35
42-45		2	5	7
45-48		2		2
48-51		3		3
54-57			1	1
Total	975	1175	1577	3727

Table 2.4.- *Pseudotolithus elongatus* (PSE)

MALES					
Length range (cm)	G.Biss_N	G.Biss_S	Gui_N	Gui_S	Total
10-15	7	18	1	1	27
15-20	62	97	25	22	206
20-25	279	245	151	200	875
25-30	62	72	90	56	280
30-35	6	23	79	29	137
35-40		5	17	15	37
40-45		2	2		4
45-50			1		1
50-55					
55-60				3	3
60-65				2	2
65-70				1	1
Total	416	462	366	329	1573
FEMALES					
Length range (cm)	G.Biss_N	G.Biss_S	Gui_N	Gui_S	Total
10-15	6	10	1		17
15-20	29	51	38	45	163
20-25	304	165	125	316	910
25-30	68	247	94	142	551
30-35	7	160	123	73	363
35-40		17	39	49	105
40-45		1	16	24	41
45-50			8	14	22
50-55				5	5
55-60	1	1	2	5	9
60-65			1	4	5
65-70					
70-75				1	1
Total	415	652	447	678	2192
SEX-COMBINED					
Length range (cm)	G.Biss_N	G.Biss_S	Gui_N	Gui_S	Total
10-15	13	38	89	49	189
15-20	98	170	199	171	638
20-25	600	426	349	581	1956
25-30	131	320	198	201	850
30-35	13	183	211	104	511
35-40		22	60	64	146
40-45		3	18	24	45
45-50			9	14	23
50-55				5	5
55-60	1	1	2	8	12
60-65			1	6	7
65-70				1	1
70-75				1	1
Total	856	1163	1136	1229	4384

Table 2.5.- *Pseudotolithus senegalensis* (PSS)

MALES					
Length range (cm)	C.Iv_W	C.Iv_E	Gha_W	Gha_E	Total
10-14	11	7			18
14-18	36	76			112
18-22	69	167	35	41	312
22-26	62	194	119	173	548
26-30	33	122	85	118	358
30-34	49	52	34	25	160
34-38	26	33	13	14	86
38-42	30	9	1	7	47
42-46	24	1		2	27
46-50	6	1			7
50-54		1			1
Total	346	663	287	380	1676
FEMALES					
Length range (cm)	C.Iv_W	C.Iv_E	Gha_W	Gha_E	Total
10-14	2	2			4
14-18	12	9			21
18-22	19	28	18	37	102
22-26	39	84	65	114	302
26-30	31	90	63	78	262
30-34	25	46	23	32	126
34-38	18	20	5	12	55
38-42	16	8		2	26
42-46	17	4		1	22
46-50	5				5
50-54	5	1			6
54-58	1	1			2
58-62	1	1			2
>62	1				1
Total	192	294	174	276	936
SEX-COMBINED					
Length range (cm)	C.Iv_W	C.Iv_E	Gha_W	Gha_E	Total
10-14	15	9			24
14-18	52	94	1	1	148
18-22	88	198	81	92	459
22-26	101	279	213	303	896
26-30	64	212	151	210	637
30-34	74	98	57	60	289
34-38	44	54	19	28	145
38-42	46	17	1	9	73
42-46	41	5		3	49
46-50	11	1			12
50-54	5	2			7
54-58	1	1			2
58-62	1	1			2
>62	1				1
Total	544	971	523	706	2744

Table 2.6.- *Pagellus bellottii* (PAR)

MALES					
Length range (cm)	CIV_W	CIV_E	Gha_W	Gha_E	Total
10-12	5	6			11
12-14	5	35		4	44
14-16	8	58	2	14	82
16-18	21	86	26	41	174
18-20	39	90	137	87	353
20-22	57	121	101	100	379
22-24	114	76	51	43	284
24-26	84	30	11	3	128
26-28	29	6			35
28-30	9	1		1	11
30-32	2				2
32-34	1				1
34-36				1	1
Total	374	509	328	294	1505
FEMALES					
Length range (cm)	CIV_W	CIV_E	Gha_W	Gha_E	Total
10-12	1	1			2
12-14	2	19		2	23
14-16	3	19	1	13	36
16-18	8	51	21	56	136
18-20	33	71	104	109	317
20-22	71	86	62	73	292
22-24	52	39	35	24	150
24-26	49	24	8	4	85
26-28	8	2			10
28-30	5	1			6
30-32	1				1
32-34			1	1	2
34-36			1	1	2
>36		1		1	2
Total	233	314	233	284	1064
SEX-COMBINED					
Length range (cm)	CIV_W	CIV_E	Gha_W	Gha_E	Total
10-12	6	7	1		14
12-14	7	55		6	68
14-16	11	78	4	42	135
16-18	29	137	52	132	350
18-20	72	161	273	251	757
20-22	128	207	182	190	707
22-24	167	115	92	73	447
24-26	133	54	19	7	213
26-28	37	8			45
28-30	14	2		1	17
30-32	3				3
32-34	1		1	1	3
34-36			1	2	3
>36		1		1	2
Total general	608	825	625	706	2764

ANNEX 3- Number of individuals and mean lengths by species and country-zone used for life history traits.

Table 3.1.1.- Summary statistics of *Epinephelus aeneus* (GPW) by zone

Stat/Zone	Mau_N	Mau_S	Sen_N	Sen_S
N	697	202	554	627
Min (cm)	28.4	25.3	22.7	20.7
Max (cm)	100.0	85.0	79.5	87.3
Mean (cm)	60.3	50.2	48.1	44.1
Median (cm)	61.5	48.1	48.5	42.2

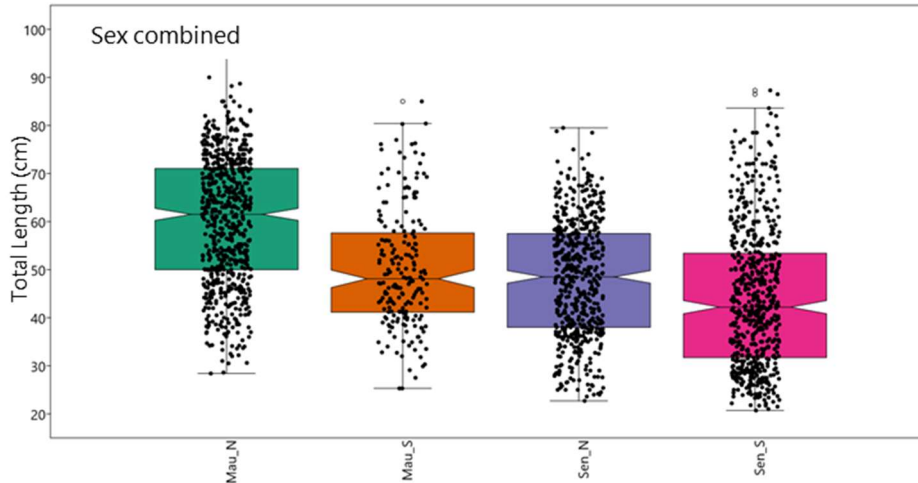


Figure 3.1.1.- Boxplot of *Epinephelus aeneus* (GPW) total lengths (cm) by zone

Table 3.1.2.- Summary statistics of *Epinephelus aeneus* (GPW) females by zone

Stat/Zone	Mau_N	Mau_S	Sen_N	Sen_S
N	659	186	506	511
Min (cm)	28.4	25.3	22.7	20.7
Max (cm)	100.0	85.0	79.5	87.3
Mean (cm)	60.1	50.5	49.4	47.1
Median (cm)	61.0	48.2	49.7	45.8

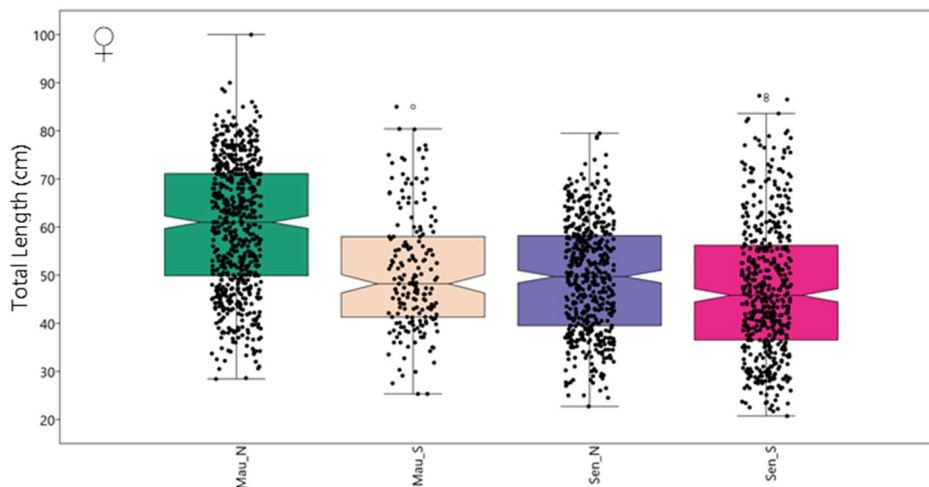


Figure 3.1.2.- Boxplot of *Epinephelus aeneus* (GPW) females total lengths (cm) by zone

Table 3.2.1.- Summary statistics of *Penaeus notialis* (SOP) by zone

Stat/Zone	Mau_N	Mau_S	Sen_N	Gam	Sen_S	GBiss
N	1267	100	1533	769	1386	1588
Min (mm)	17.6	20.9	14.0	14.8	12.5	10.4
Max (mm)	50.0	37.2	50.0	45.0	55.8	38.2
Mean (mm)	31.7	25.2	29.1	28.5	30.1	21.1
Median (mm)	30.2	24.6	28.15	28.5	28.6	19.5

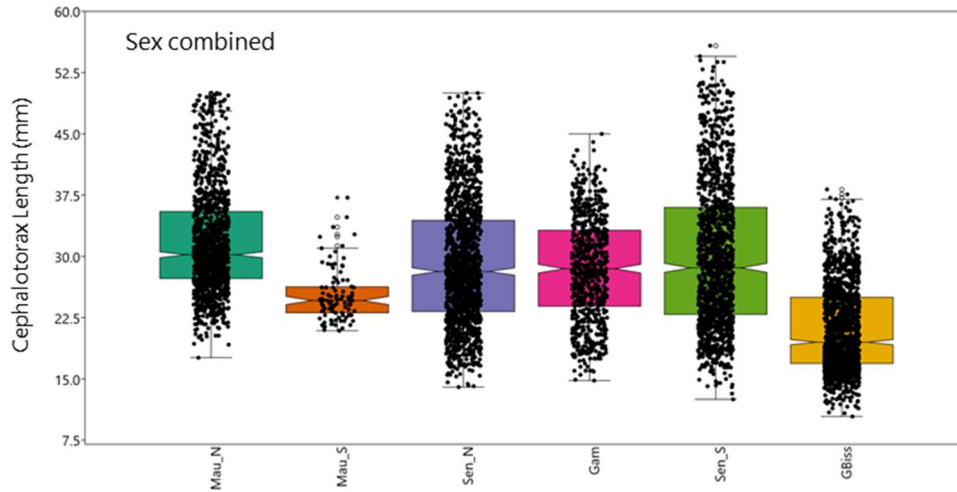


Figure 3.2.1.- Boxplot of *Penaeus notialis* (SOP) cephalotorax lengths (mm) by zone

Table 3.2.2.- Summary statistics of *Penaeus notialis* (SOP) males by zone

Stat/Zone	Mau_N	Mau_S	Sen_N	Gam	Sen_S	GBiss
N	528	64	483	329	438	668
Min (mm)	17.6	20.9	14.4	14.8	14.5	11.9
Max (mm)	37.3	31.3	36.9	35.5	43.8	35.2
Mean (mm)	28.1	24.1	24.5	25.5	23.9	20.8
Median (mm)	28.6	24.1	24.48	26.0	22.9	19.7

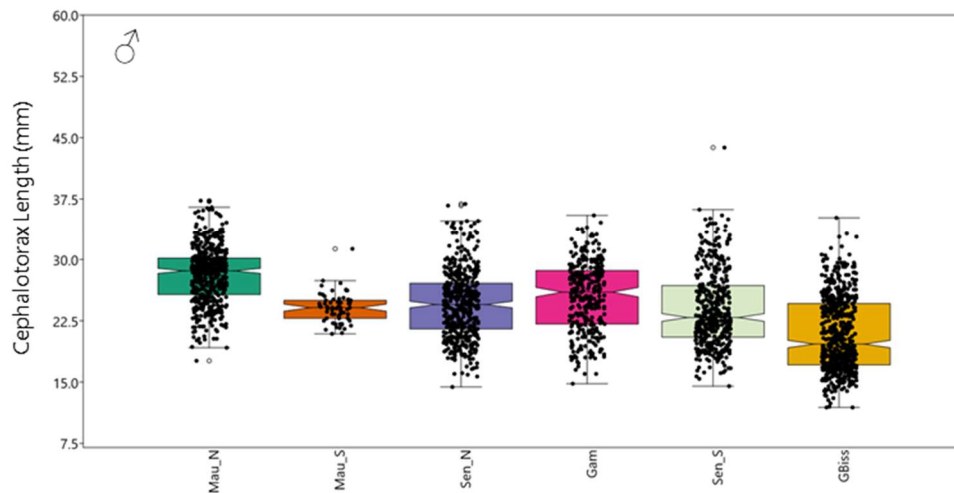


Figure 3.2.2.- Boxplot of *Penaeus notialis* (SOP) males cephalotorax lengths (mm) by zone

Table 3.2.3.- Summary statistics of *Penaeus notialis* (SOP) females by zone

Stat/Zone	Mau_N	Mau_S	Sen_N	Gam	Sen_S	GBiss
N	739	36	1038	439	945	920
Min (mm)	21.0	21.2	14.0	14.9	12.5	10.4
Max (mm)	50.0	37.2	50.0	45.0	55.8	38.2
Mean (mm)	34.2	27.3	31.4	30.8	32.9	21.3
Median (mm)	33.8	27.1	31.1	32.2	32.5	19.4

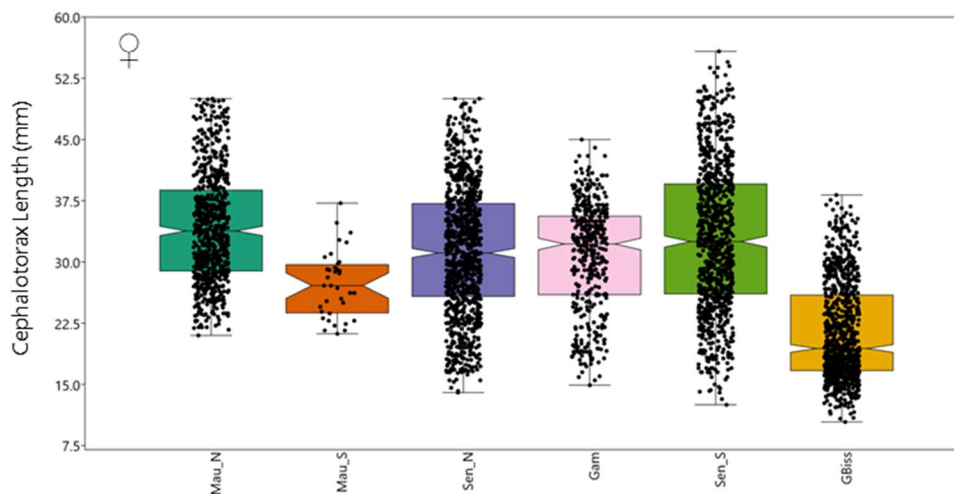


Figure 3.2.3.- Boxplot of *Penaeus notialis* (SOP) females cephalotorax lengths (mm) by zone

Table 3.3.1.- Summary statistics of *Pagrus caeruleostictus* (BSC) by zone

Stat/Zone	G.Biss_S	Gui_N	Gui_S
N	980	1282	1413
Min (cm)	11.0	6.8	10.2
Max (cm)	33.7	50.4	55.0
Mean (cm)	20.8	24.9	24.0
Median (cm)	20.5	24.5	23.5

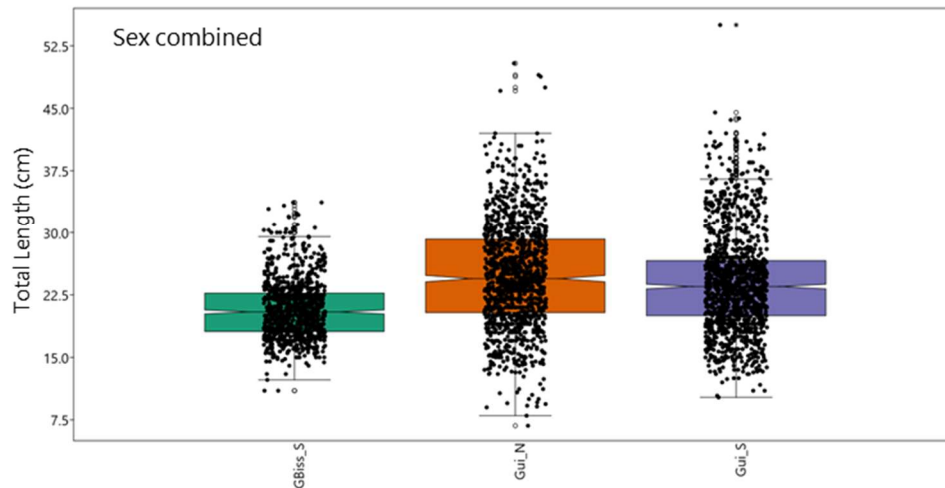


Figure 3.3.1.- Boxplot of *Pagrus caeruleostictus* (BSC) total lengths (cm) by zone

Table 3.3.2.- Summary statistics of *Pagrus caeruleostictus* (BSC) males by zone

Stat/Zone	G.Biss_S	Gui_N	Gui_S
N	448	460	609
Min (cm)	11.0	16.0	16.0
Max (cm)	33.7	34.0	34.0
Mean (cm)	21.1	25.8	24.7
Median (cm)	20.6	25.7	24.7

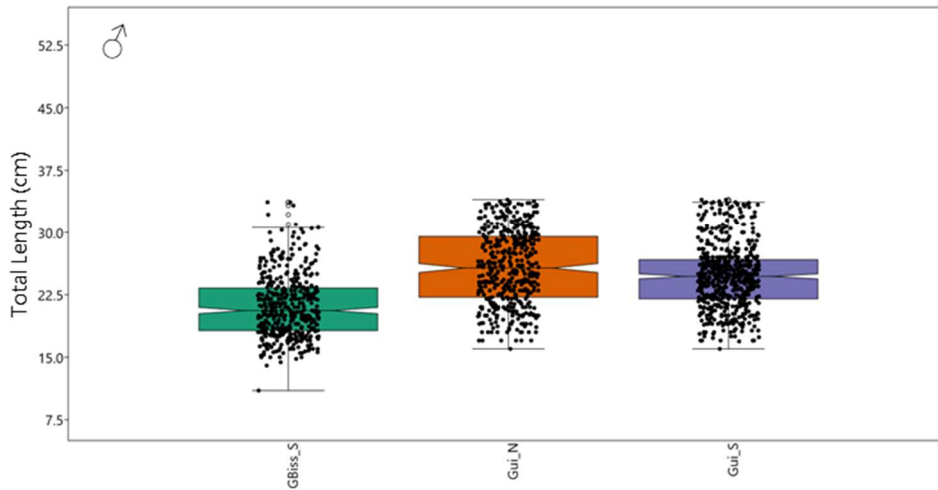


Figure 3.3.2.- Boxplot of *Pagrus caeruleostictus* (BSC) males total lengths (cm) by zone

Table 3.3.3.- Summary statistics of *Pagrus caeruleostictus* (BSC) females by zone

Stat/Zone	G.Biss_S	Gui_N	Gui_S
N	448	460	609
Min (cm)	11.0	16.0	16.0
Max (cm)	33.7	34.0	34.0
Mean (cm)	21.1	25.8	24.7
Median (cm)	20.6	25.7	24.7

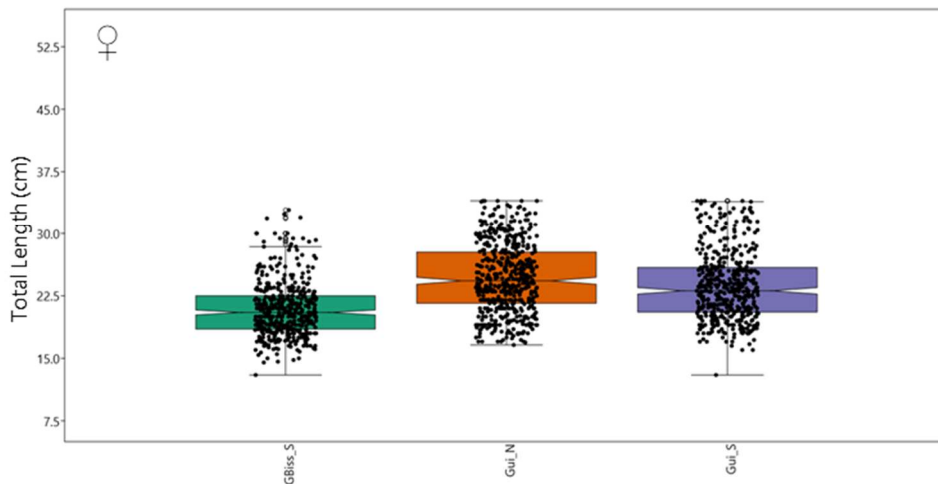


Figure 3.3.3.- Boxplot of *Pagrus caeruleostictus* (BSC) females total lengths (cm) by zone

Table 3.4.1.- Summary statistics of *Pseudotolithus elongatus* (PSE) by zone

Stat/Zone	G.Biss_N	G.Biss_S	Gui_N	Gui_S
N	654	1034	1130	1210
Min (cm)	14.3	10.9	11.0	10.5
Max (cm)	32.7	43.0	64.5	60.5
Mean (cm)	22.3	24.7	24.7	24.9
Median (cm)	21.6	24.2	23.5	23.0

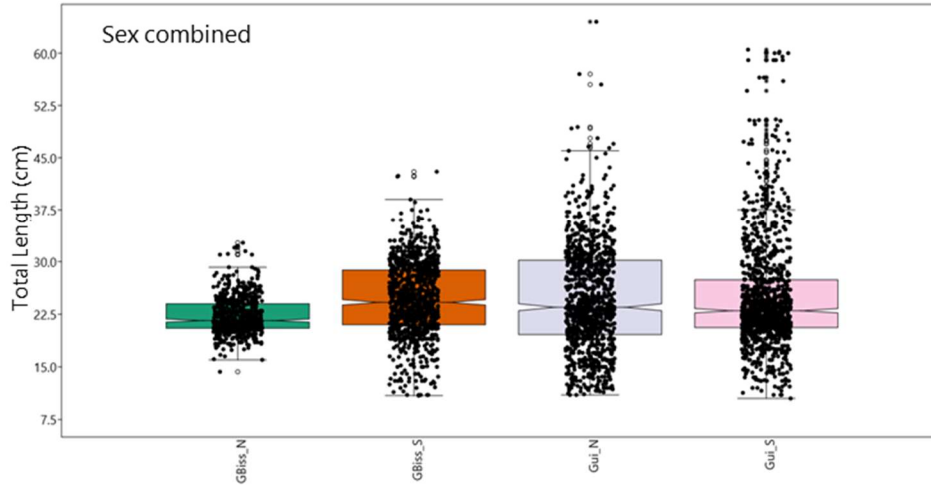


Figure 3.4.1.- Boxplot of *Pseudotolithus elongatus* (PSE) total lengths (cm) by zone

Table 3.4.2.- Summary statistics of *Pseudotolithus elongatus* (PSE) males by zone

Stat/Zone	G.Biss_N	G.Biss_S	Gui_N	Gui_S
N	313	420	363	326
Min (cm)	14.3	10.9	15.5	17.0
Max (cm)	32.7	43.0	46.4	60.0
Mean (cm)	22.2	22.3	26.1	25.0
Median (cm)	21.9	22.0	25.0	23.5

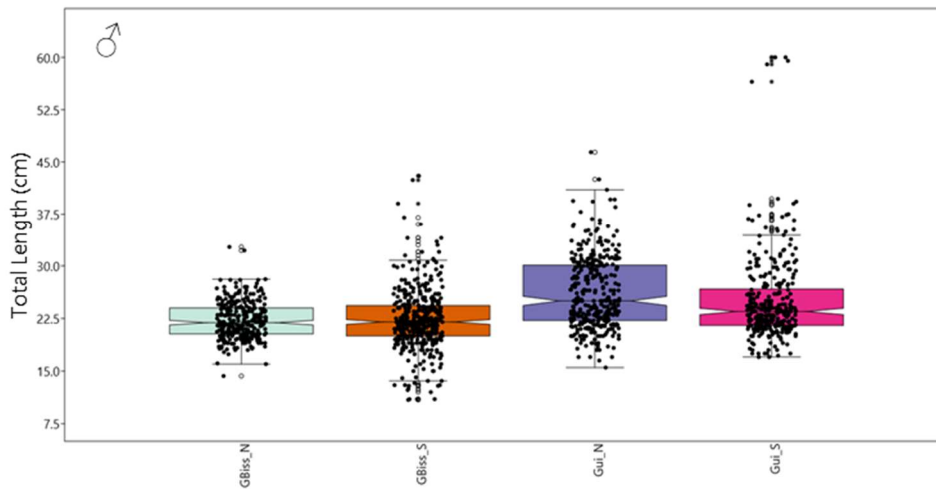


Figure 3.4.2.- Boxplot of *Pseudotolithus elongatus* (PSE) males total lengths (cm) by zone

Table 3.4.3.- Summary statistics of *Pseudotolithus elongatus* (PSE) females by zone

Stat/Zone	G.Biss_N	G.Biss_S	Gui_N	Gui_S
N	319	579	446	668
Min (cm)	16.5	11.0	14.5	16.0
Max (cm)	32.7	42.3	64.5	60.5
Mean (cm)	22.5	26.8	28.4	27.2
Median (cm)	21.5	27.5	28.5	24.5

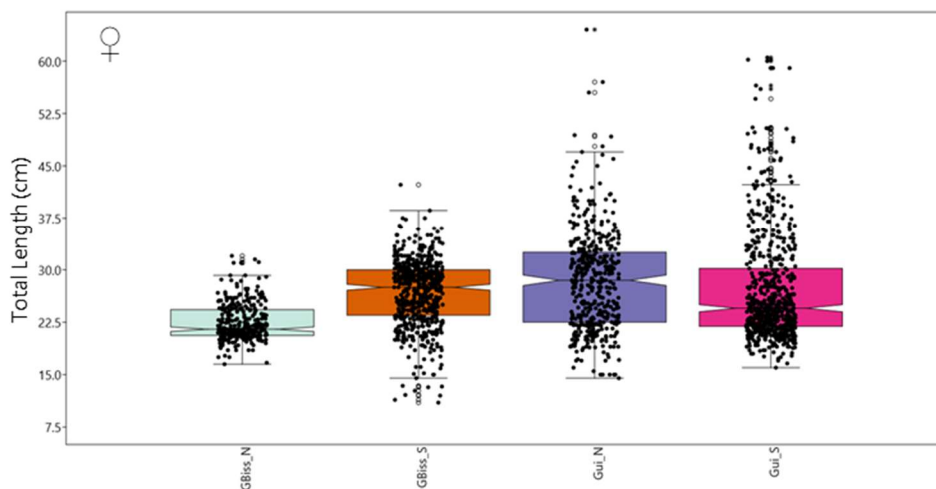


Figure 3.4.3.- Boxplot of *Pseudotolithus elongatus* (PSE) females total lengths (cm) by zone

Table 3.5.1.- Summary statistics of *Pseudotolithus senegalensis* (PSS) by zone

Stat/Zone	Civ_W	Civ_E	Gha_W	Gha_E
N	541	966	521	688
Min (cm)	11.5	12.0	17.4	17.0
Max (cm)	58.0	60.5	38.0	45.0
Mean (cm)	28.4	25.1	25.3	25.9
Median (cm)	26.7	24.4	25.0	25.0

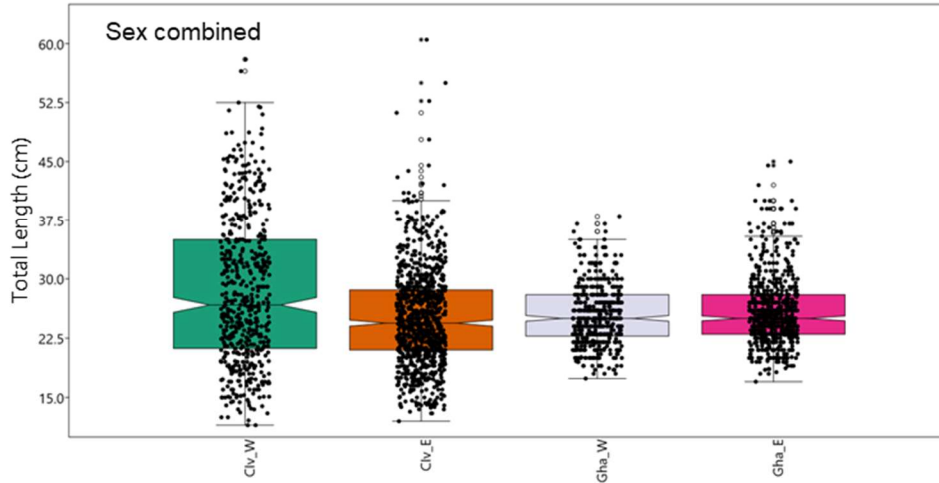


Figure 3.2.1.- Boxplot of *Pseudotolithus senegalensis* (PSS) total lengths (cm) by zone

Table 3.5.2.- Summary statistics of *Pseudotolithus senegalensis* (PSS) males by zone

Stat/Zone	Civ_W	Civ_E	Gha_W	Gha_E
N	345	661	286	367
Min (cm)	11.5	12.0	18.5	18.0
Max (cm)	49.2	51.2	38.0	45.0
Mean (cm)	27.4	24.2	25.8	25.9
Median (cm)	25.5	23.5	25.0	25.0

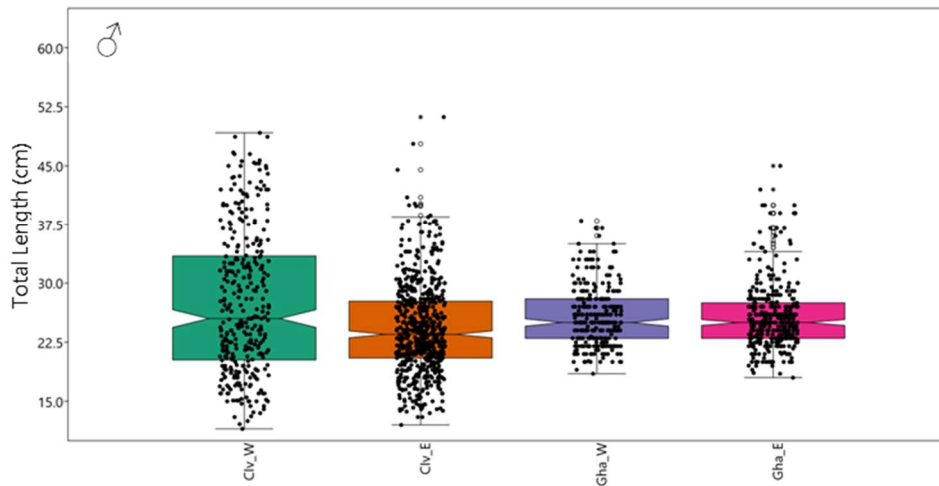


Figure 3.5.2.- Boxplot of *Pseudotolithus senegalensis* (PSS) males total lengths (cm) by zone

Table 3.5.3.- Summary statistics of *Pseudotolithus senegalensis* (PSS) females by zone

Stat/Zone	Civ_W	Civ_E	Gha_W	Gha_E
N	190	292	173	272
Min (cm)	11.5	13.2	18.0	18.0
Max (cm)	58.0	60.5	36.0	44.5
Mean (cm)	30.6	27.5	25.8	26.0
Median (cm)	28.3	27.2	26.0	25.4

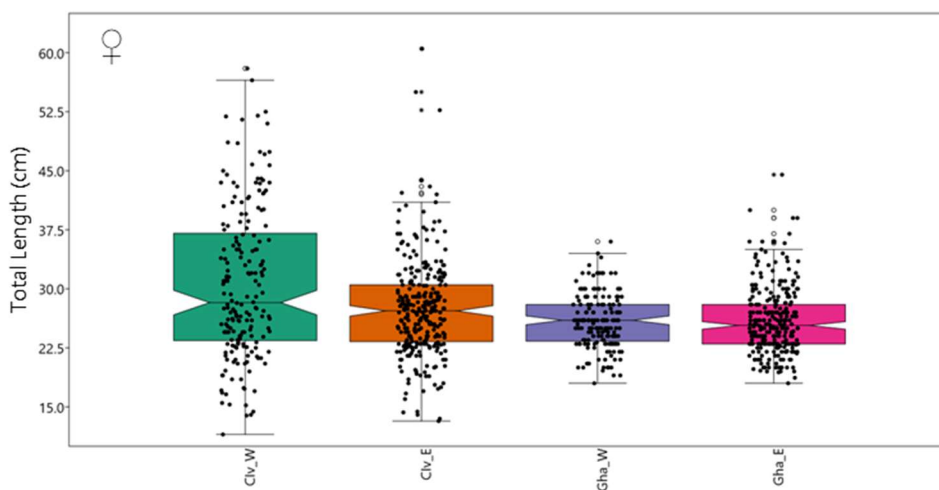


Figure 3.5.3.- Boxplot of *Pseudotolithus senegalensis* (PSS) females total lengths (cm) by zone

Tabla 3.6.1.- Summary statistics of *Pagellus bellottii* (PAR) by zone

Stat/Zone	Civ_W	Civ_E	Gha_W	Gha_E
N	608	824	625	705
Min (cm)	10.7	11.0	10.0	12.0
Max (cm)	32.5	28.0	34.0	34.5
Mean (cm)	22.2	19.3	19.8	19.1
Median (cm)	22.5	19.6	19.5	19.0

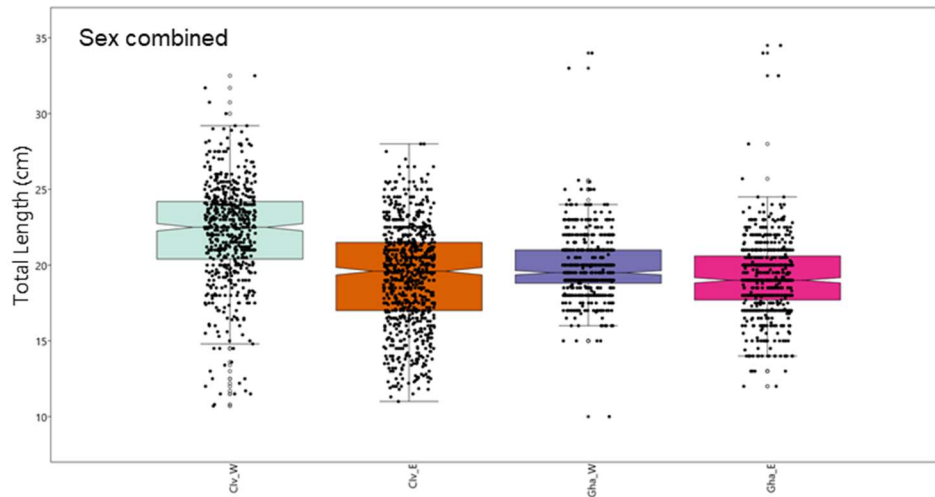


Figure 3.6.1.- Boxplot of *Pagellus bellottii* (PAR) total lengths (cm) by zone

Table 3.6.2.- Summary statistics of *Pagellus bellottii* (PAR) males by zone

Stat/Zone	Civ_W	Civ_E	Gha_W	Gha_E
N	374	509	328	294
Min (cm)	10.7	11.0	15.0	12.0
Max (cm)	32.5	28.0	25.6	34.5
Mean (cm)	22.3	19.1	19.9	19.5
Median (cm)	23.0	19.5	19.7	20.0

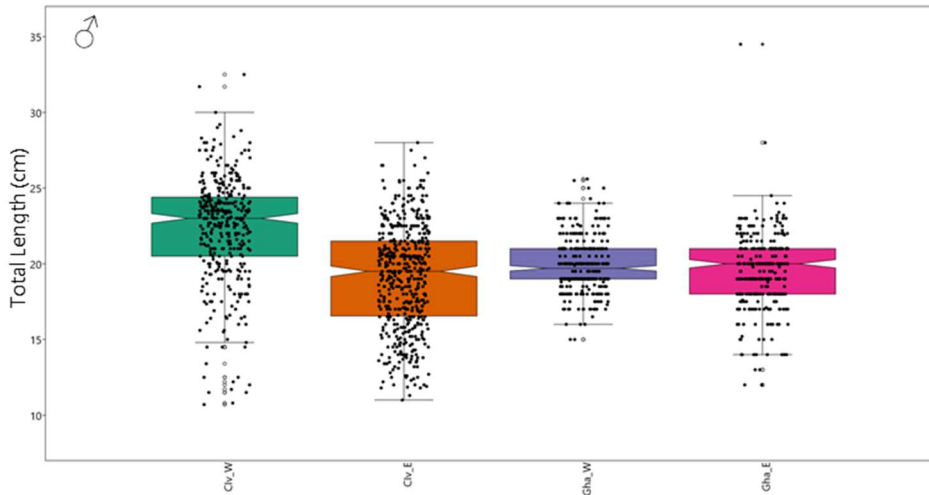


Figure 3.6.2.- Boxplot of *Pagellus bellottii* (PAR) males total lengths (cm) by zone

Table 3.6.3.- Summary statistics of *Pagellus bellottii* (PAR) females by zone

Stat/Zone	Civ_W	Civ_E	Gha_W	Gha_E
N	233	313	233	283
Min (cm)	11.5	11.5	15.0	13.0
Max (cm)	30.8	28.0	34.0	34.0
Mean (cm)	21.9	19.5	19.9	19.1
Median (cm)	21.9	19.8	19.5	19.0

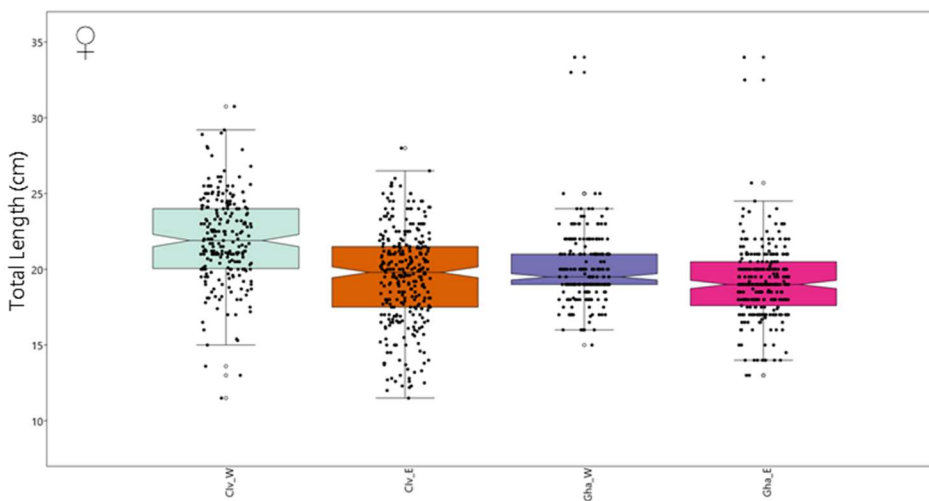
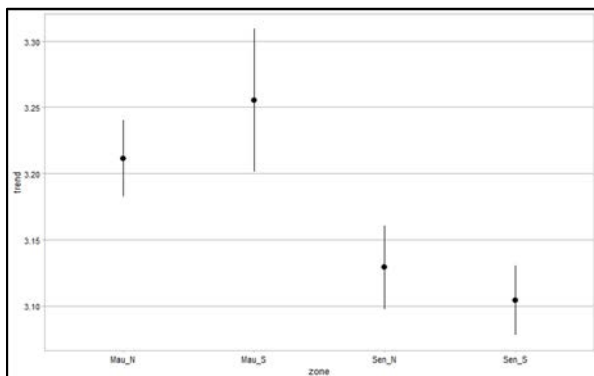


Figure 3.6.3.- Boxplot of *Pagellus bellottii* (PAR) females total lengths (cm) by zone

ANNEX 4- Weight related parameters. By species: 1) Weight- Length Relationship: pair-wise comparison analysis. Figures of median values and Standard errors (SE) (right) and tables of slope pair-wise comparisons (**significant difference at $p > 0.05$) (left), by sex and for sex combined; 2) Conversion factor: pair-wise comparison analysis. Figures of median values and Standard errors (SE) (right) and tables of slope pair-wise comparisons (**significant difference at $p > 0.05$) (left), for sex combined; 3) Figures of quarterly condition factors of mature females by country-zone: median values and standard errors (SE). All by species.

4.1. *Epinephelus aeneus* (GPW)

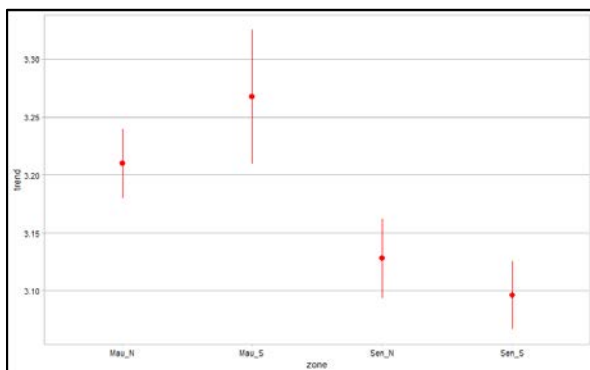
4.1.1. Gutted Weight- Length Relationship



SEX COMBINED

Slope comparisons:

contrast	p.value
Mau_N - Mau_S	0.51
Mau_N - Sen_N	0 **
Mau_N - Sen_S	0 **
Mau_S - Sen_N	0 **
Mau_S - Sen_S	0 **
Sen_N - Sen_S	0.64

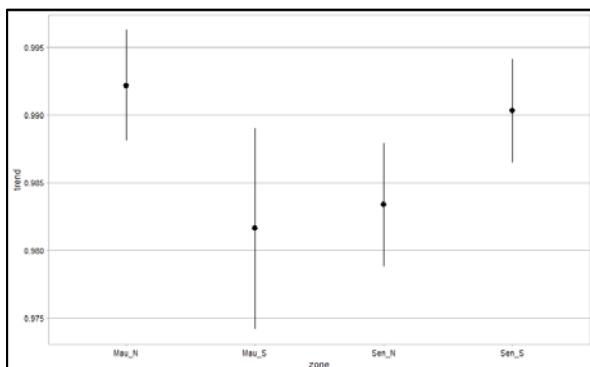


FEMALES

Slope comparisons:

contrast	p.value
Mau_N - Mau_S	0.31
Mau_N - Sen_N	0 **
Mau_N - Sen_S	0 **
Mau_S - Sen_N	0 **
Mau_S - Sen_S	0 **
Sen_N - Sen_S	0.51

4.1.2. Conversion factor

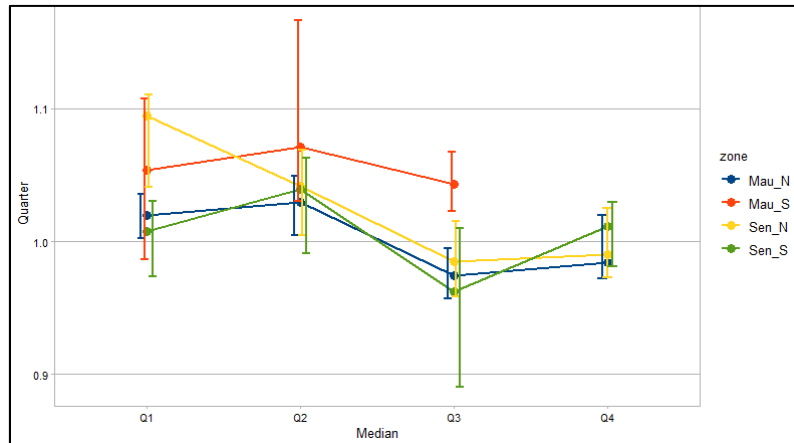


SEX COMBINED

Slope comparisons:

contrast	p.value
Mau_N - Mau_S	0.07
Mau_N - Sen_N	0.02 **
Mau_N - Sen_S	0.91
Mau_S - Sen_N	0.98
Mau_S - Sen_S	0.17
Sen_N - Sen_S	0.1

Figure 4.1.3.- Quarterly condition factors (mature females)

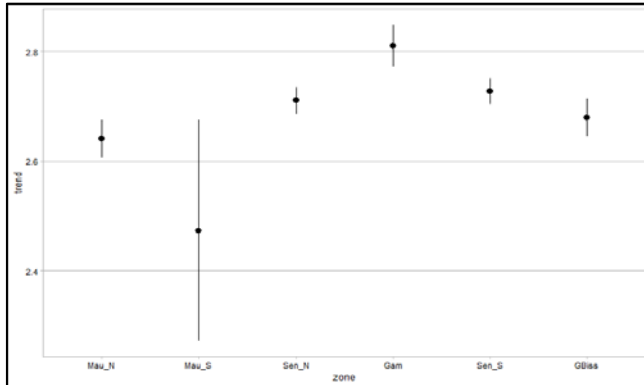


Median comparisons:

group1	group2	p.value
Mau_SQ1	Mau_NQ1	1
Sen_NQ1	Mau_NQ1	0.09
Sen_NQ1	Mau_SQ1	1
Sen_SQ1	Mau_NQ1	1
Sen_SQ1	Mau_SQ1	1
Sen_SQ1	Sen_NQ1	0.01 **
Mau_SQ2	Mau_NQ2	1
Sen_NQ2	Mau_NQ2	1
Sen_NQ2	Mau_SQ2	1
Sen_SQ2	Mau_NQ2	1
Sen_SQ2	Mau_SQ2	1
Sen_SQ2	Sen_NQ2	1
Mau_SQ3	Mau_NQ3	0 **
Sen_NQ3	Mau_NQ3	1
Sen_NQ3	Mau_SQ3	0.03 **
Sen_SQ3	Mau_NQ3	1
Sen_SQ3	Mau_SQ3	0 **
Sen_SQ3	Sen_NQ3	1
Mau_SQ4	Mau_NQ4	1
Sen_NQ4	Mau_NQ4	1
Sen_NQ4	Mau_SQ4	1
Sen_SQ4	Mau_NQ4	1
Sen_SQ4	Mau_SQ4	1
Sen_SQ4	Sen_NQ4	1

4.2. *Penaeus notialis* (SOP)

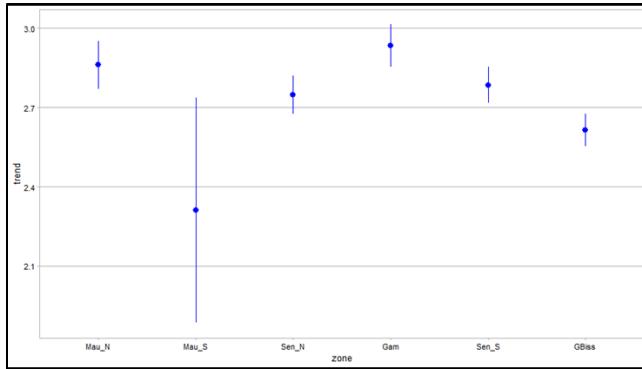
4.2.1. Total Weight- Length Relationship



SEX COMBINED

Slope comparisons:

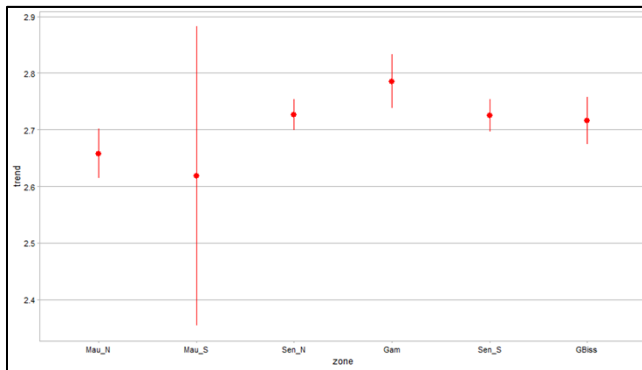
contrast	p.value
Mau_N - Mau_S	0.61
Mau_N - Sen_N	0.01 **
Mau_N - Gam	0 **
Mau_N - Sen_S	0 **
Mau_N - GBiss	0.63
Mau_S - Sen_N	0.2
Mau_S - Gam	0.02 **
Mau_S - Sen_S	0.14
Mau_S - GBiss	0.36
Sen_N - Gam	0 **
Sen_N - Sen_S	0.92
Sen_N - GBiss	0.7
Gam - Sen_S	0 **
Gam - GBiss	0 **
Sen_S - GBiss	0.23



MALES

Slope comparisons:

contrast	p.value
Mau_N - Mau_S	0.13
Mau_N - Sen_N	0.39
Mau_N - Gam	0.84
Mau_N - Sen_S	0.78
Mau_N - G.Biss	0 **
Mau_S - Sen_N	0.35
Mau_S - Gam	0.05 **
Mau_S - Sen_S	0.26
Mau_S - GBiss	0.73
Sen_N - Gam	0.01 **
Sen_N - Sen_S	0.98
Sen_N - GBiss	0.07
Gam - Sen_S	0.06
Gam - GBiss	0 **
Sen_S - GBiss	0 **

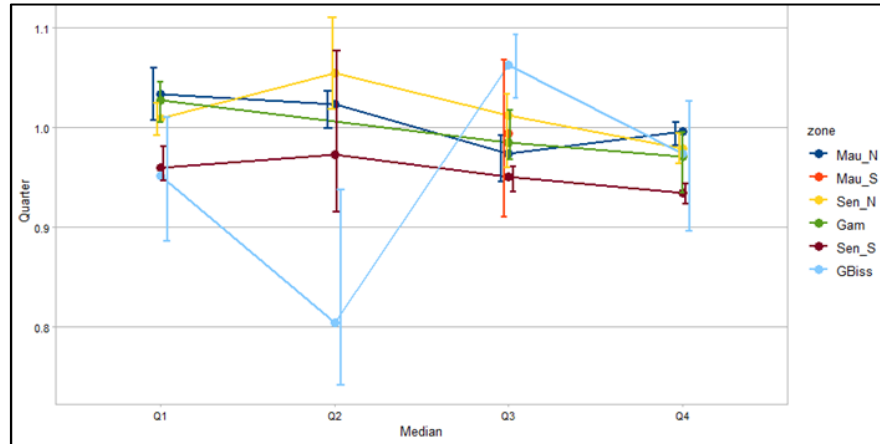


FEMALES

Slope comparisons:

contrast	p.value
Mau_N - Mau_S	1
Mau_N - Sen_N	0.09
Mau_N - Gam	0 **
Mau_N - Sen_S	0.12
Mau_N - GBiss	0.41
Mau_S - Sen_N	0.97
Mau_S - Gam	0.83
Mau_S - Sen_S	0.97
Mau_S - GBiss	0.98
Sen_N - Gam	0.29
Sen_N - Sen_S	1
Sen_N - GBiss	1
Gam - Sen_S	0.28
Gam - GBiss	0.27
Sen_S - GBiss	1

4.2.2.- Quarterly condition factors (mature females)

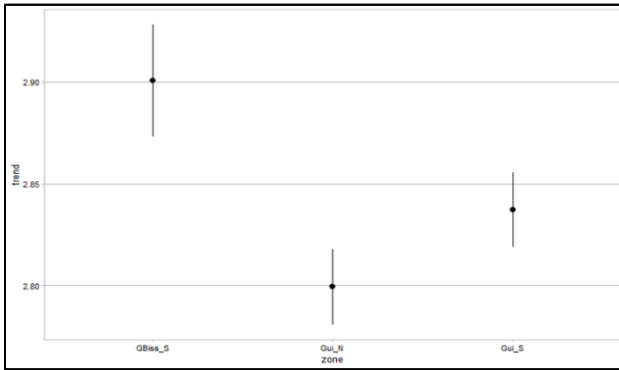


Mean comparisons:

group1	group2	p.value	group1	group2	p.value
Sen_NQ1	Mau_NQ1	1	GBissQ3	GamQ3	1
GamQ1	Mau_NQ1	1	GBissQ3	Sen_SQ3	0 **
GamQ1	Sen_NQ1	1	Sen_NQ4	Mau_NQ4	1
Sen_SQ1	Mau_NQ1	0 **	GamQ4	Mau_NQ4	0.61
Sen_SQ1	Sen_NQ1	0 **	GamQ4	Sen_NQ4	1
Sen_SQ1	GamQ1	0 **	Sen_SQ4	Mau_NQ4	0 **
GBissQ1	Mau_NQ1	1	Sen_SQ4	Sen_NQ4	0 **
GBissQ1	Sen_NQ1	1	Sen_SQ4	GamQ4	0.25
GBissQ1	GamQ1	0.53	GBissQ4	Mau_NQ4	1
GBissQ1	Sen_SQ1	1	GBissQ4	Sen_NQ4	1
Sen_NQ2	Mau_NQ2	1	GBissQ4	GamQ4	1
Sen_SQ2	Mau_NQ2	1	GBissQ4	Sen_SQ4	1
Sen_SQ2	Sen_NQ2	1			
GBissQ2	Mau_NQ2	0 **			
GBissQ2	Sen_NQ2	0 **			
GBissQ2	Sen_SQ2	0.04 **			
Mau_SQ3	Mau_NQ3	1			
Sen_NQ3	Mau_NQ3	1			
Sen_NQ3	Mau_SQ3	1			
GamQ3	Mau_NQ3	1			
GamQ3	Mau_SQ3	1			
GamQ3	Sen_NQ3	1			
Sen_SQ3	Mau_NQ3	1			
Sen_SQ3	Mau_SQ3	1			
Sen_SQ3	Sen_NQ3	0.31			
Sen_SQ3	GamQ3	0.02 **			
GBissQ3	Mau_NQ3	0.16			
GBissQ3	Mau_SQ3	1			
GBissQ3	Sen_NQ3	1			

4.3.- Pagrus caeruleostictus (BSC)

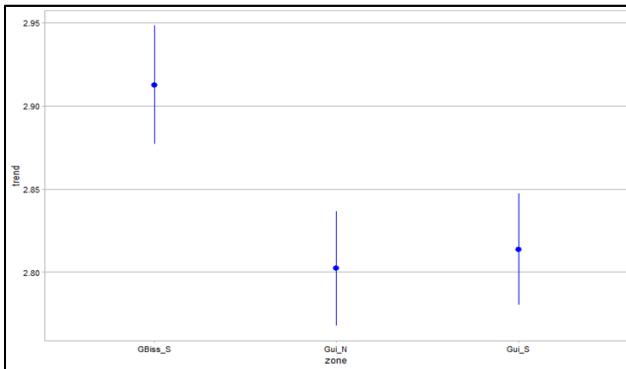
4.3.1. Gutted Weight- Length Relationship



SEX COMBINED

Slope comparisons:

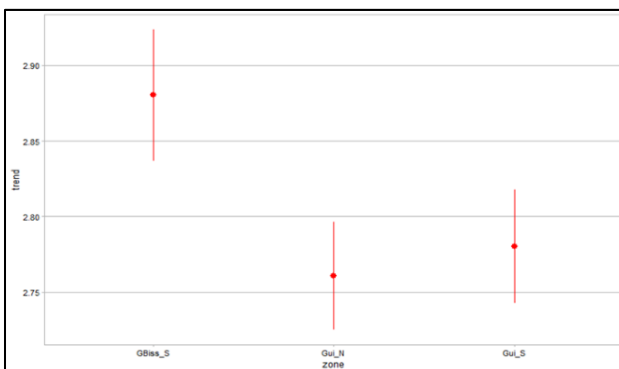
contrast	p.value
G.Biss_S - Gui_N	0 **
G.Biss_S - Gui_S	0 **
Gui_N - Gui_S	0.01 **



MALES

Slope comparisons:

contrast	p.value
G.Biss_S - Gui_N	0 **
G.Biss_S - Gui_S	0 **
Gui_N - Gui_S	0.88

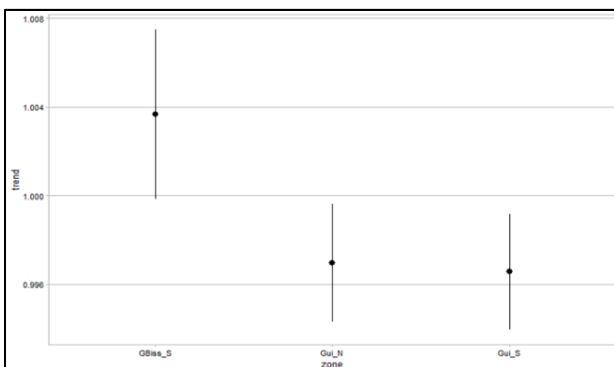


FEMALES

Slope comparisons:

contrast	p.value
G.Biss_S - Gui_N	0 **
G.Biss_S - Gui_S	0 **
Gui_N - Gui_S	0.74

4.3.2. Conversion factor

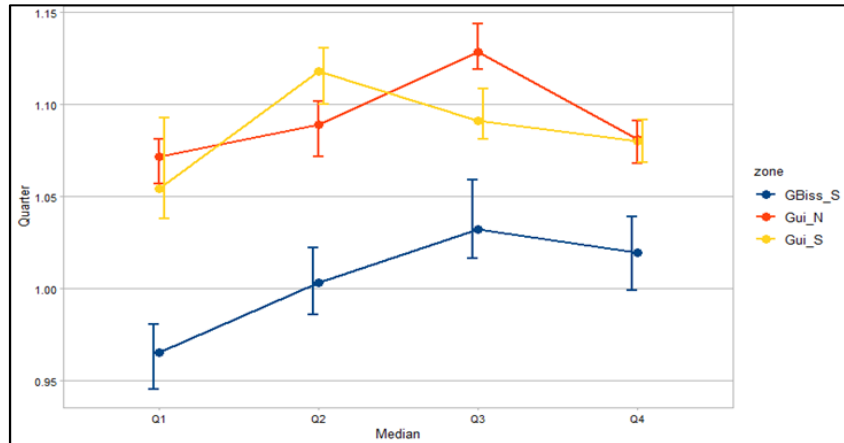


SEX COMBINED

Slope comparisons:

contrast	p.value
G.Biss_S - Gui_N	0.01 **
GBiss_S - Gui_S	0.01 **
Gui_N - Gui_S	0.98

Table 4.3.3.- Quarterly condition factors (mature females)

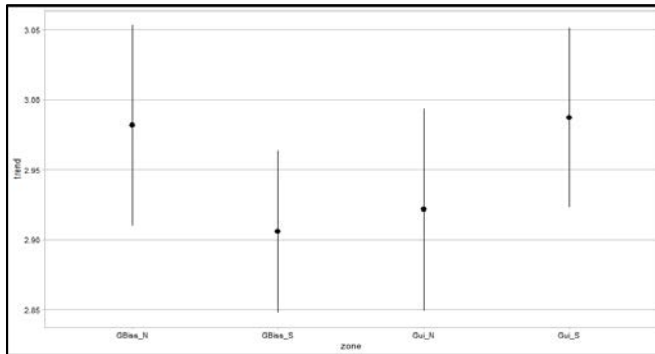


Median comparisons:

group1	group2	p.value	
Gui_NQ1	GBiss_SQ1	0	**
Gui_SQ1	GBiss_SQ1	0	**
Gui_SQ1	Gui_NQ1	1	
Gui_NQ2	GBiss_SQ2	0	**
Gui_SQ2	GBiss_SQ2	0	**
Gui_SQ2	Gui_NQ2	0.09	
Gui_NQ3	GBiss_SQ3	0	**
Gui_SQ3	GBiss_SQ3	0	**
Gui_SQ3	Gui_NQ3	0	**
Gui_NQ4	GBiss_SQ4	0	**
Gui_SQ4	GBiss_SQ4	0	**
Gui_SQ4	Gui_NQ4	1	

4. 4. *Pseudotolithus elongatus* (PSE)

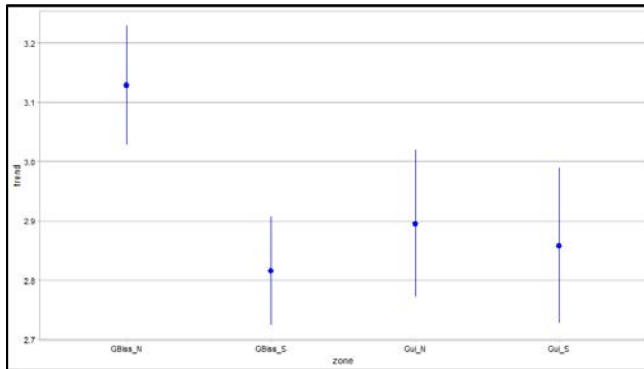
4.4.1. Gutted Weight- Length Relationship



SEX COMBINED

Slope comparisons:

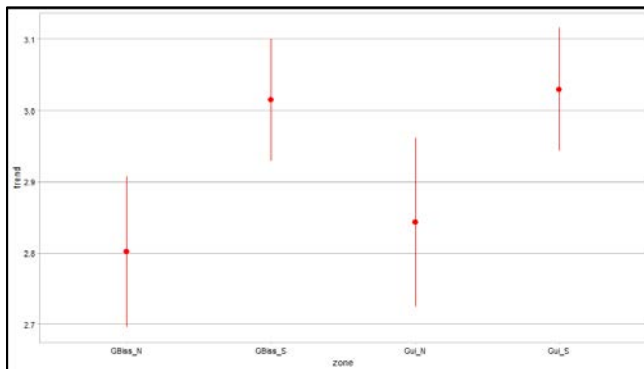
contrast	p.value
GBiss_N - GBiss_S	0.37
GBiss_N - Gui_N	0.65
GBiss_N - Gui_S	1
GBiss_S - Gui_N	0.99
GBiss_S - Gui_S	0.25
Gui_N - Gui_S	0.54



MALES

Slope comparisons:

contrast	p.value
GBiss_N - GBiss_S	0 **
GBiss_N - Gui_N	0.02 **
GBiss_N - Gui_S	0.01 **
GBiss_S - Gui_N	0.74
GBiss_S - Gui_S	0.95
Gui_N - Gui_S	0.98

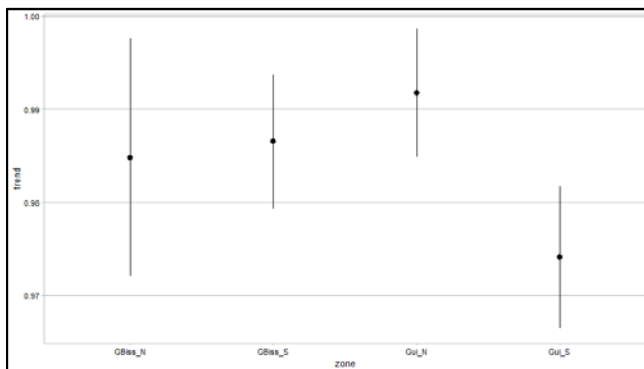


FEMALES

Slope comparisons:

contrast	p.value
GBiss_N - GBiss_S	0.01 **
GBiss_N - Gui_N	0.95
GBiss_N - Gui_S	0.01 **
GBiss_S - Gui_N	0.1
GBiss_S - Gui_S	0.99
Gui_N - Gui_S	0.06

4.4.2. Conversion factor

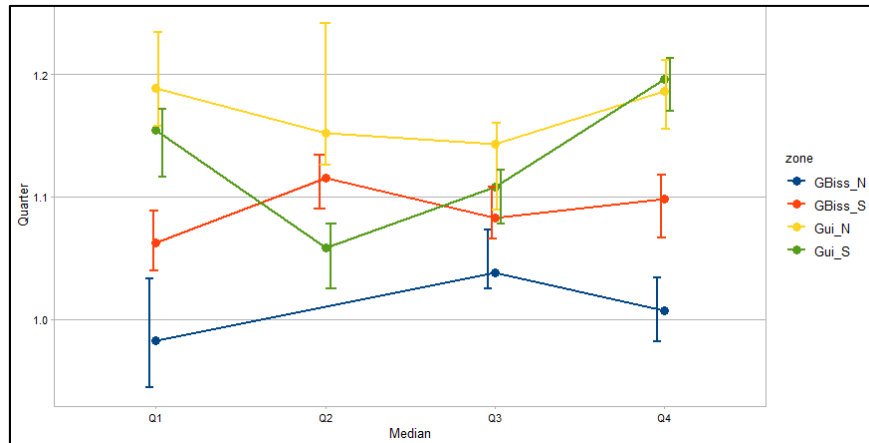


SEX COMBINED

Slope comparisons:

contrast	p.value
GBiss_N - GBiss_S	1
GBiss_N - Gui_N	0.79
GBiss_N - Gui_S	0.49
GBiss_S - Gui_N	0.73
GBiss_S - Gui_S	0.09
Gui_N - Gui_S	0 **

Figure 4.4.3.- Quarterly condition factors (mature females)

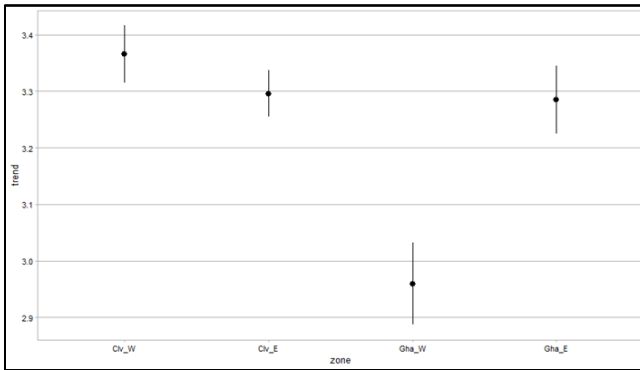


Median comparisons:

group1	group2	p.value
GBiss_SQ1	GBiss_NQ1	0.02 **
Gui_NQ1	GBiss_NQ1	0 **
Gui_NQ1	GBiss_SQ1	0 **
Gui_SQ1	GBiss_NQ1	0 **
Gui_SQ1	GBiss_SQ1	0 **
Gui_SQ1	Gui_NQ1	0.11
GBiss_SQ2	GBiss_NQ2	1
Gui_NQ2	GBiss_NQ2	1
Gui_SQ2	GBiss_NQ2	1
Gui_SQ2	GBiss_SQ2	0.5
Gui_SQ2	Gui_NQ2	0 **
GBiss_SQ3	GBiss_NQ3	1
Gui_NQ3	GBiss_NQ3	0 **
Gui_NQ3	GBiss_SQ3	0.4
Gui_SQ3	GBiss_NQ3	0.03 **
Gui_SQ3	GBiss_SQ3	1
Gui_SQ3	Gui_NQ3	1
GBiss_SQ4	GBiss_NQ4	0 **
Gui_NQ4	GBiss_NQ4	0 **
Gui_NQ4	GBiss_SQ4	0 **
Gui_SQ4	GBiss_NQ4	0 **
Gui_SQ4	GBiss_SQ4	0 **
Gui_SQ4	Gui_NQ4	1

4.5. *Pseudotolithus senegalensis* (PSS)

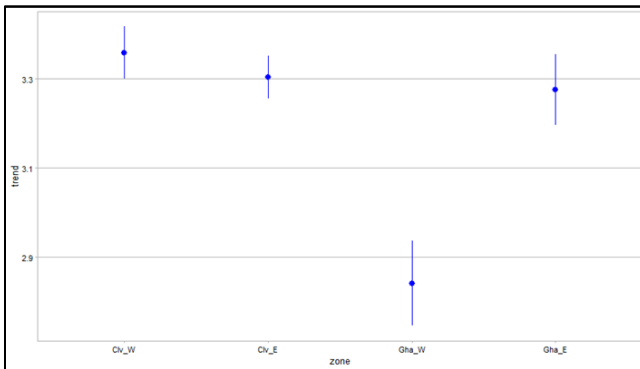
4.5.1. Gutted Weight- Length Relationship



SEX COMBINED

Slope comparisons:

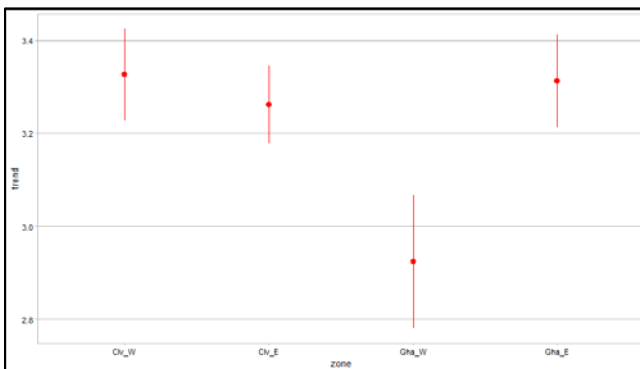
contrast	p.value
Clv_W - Clv_E	0.16
Clv_W - Gha_W	0 **
Clv_W - Gha_E	0.19
Clv_E - Gha_W	0 **
Clv_E - Gha_E	0.99
Gha_W - Gha_E	0 **



MALES

Slope comparisons:

contrast	p.value
Clv_W - Clv_E	0.48
Clv_W - Gha_W	0 **
Clv_W - Gha_E	0.35
Clv_E - Gha_W	0 **
Clv_E - Gha_E	0.94
Gha_W - Gha_E	0 **

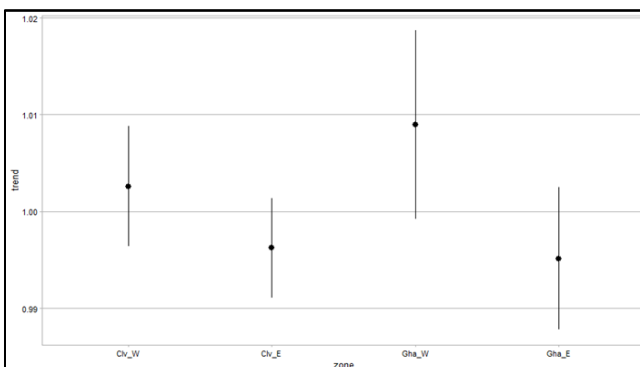


FEMALES

Slope comparisons:

contrast	p.value
Clv_W - Clv_E	0.76
Clv_W - Gha_W	0 **
Clv_W - Gha_E	1
Clv_E - Gha_W	0 **
Clv_E - Gha_E	0.87
Gha_W - Gha_E	0 **

4.5.2. Conversion factor

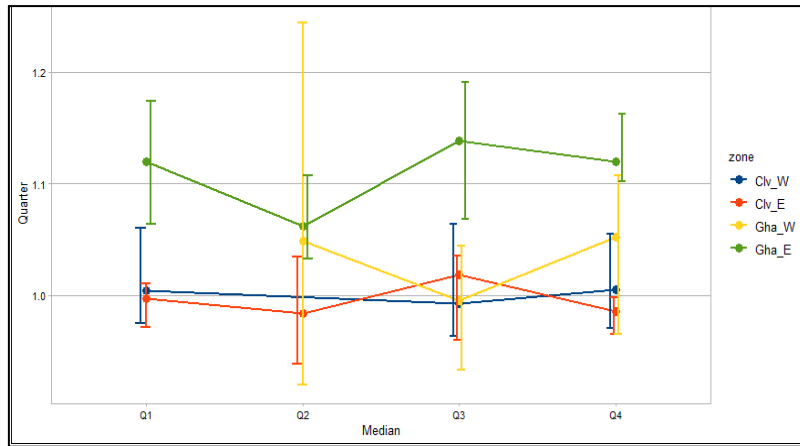


SEX COMBINED

Slope comparisons:

contrast	p.value
Clv_W - Clv_E	0.41
Clv_W - Gha_W	0.7
Clv_W - Gha_E	0.43
Clv_E - Gha_W	0.11
Clv_E - Gha_E	1
Gha_W - Gha_E	0.12

Figure 4.5.3.- Quarterly condition factors (mature females)

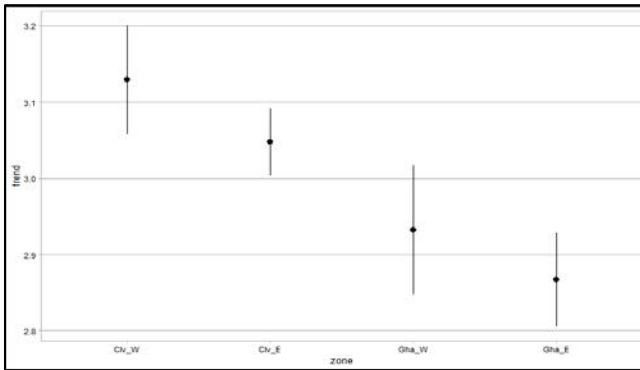


Median comparisons:

group1	group2	p.value
Clv_EQ1	Clv_WQ1	1
Gha_WQ1	Clv_WQ1	1
Gha_WQ1	Clv_EQ1	1
Gha_EQ1	Clv_WQ1	0 **
Gha_EQ1	Clv_EQ1	0 **
Gha_EQ1	Gha_WQ1	1
Gha_WQ2	Clv_EQ2	1
Gha_EQ2	Clv_EQ2	0.52
Gha_EQ2	Gha_WQ2	1
Clv_EQ3	Clv_WQ3	1
Gha_WQ3	Clv_WQ3	1
Gha_WQ3	Clv_EQ3	1
Gha_EQ3	Clv_WQ3	0.61
Gha_EQ3	Clv_EQ3	0.03 **
Gha_EQ3	Gha_WQ3	0.35
Clv_EQ4	Clv_WQ4	1
Gha_WQ4	Clv_WQ4	1
Gha_WQ4	Clv_EQ4	1
Gha_EQ4	Clv_WQ4	0 **
Gha_EQ4	Clv_EQ4	0 **
Gha_EQ4	Gha_WQ4	0.03 **

4.6. *Pagellus bellottii* (PAR)

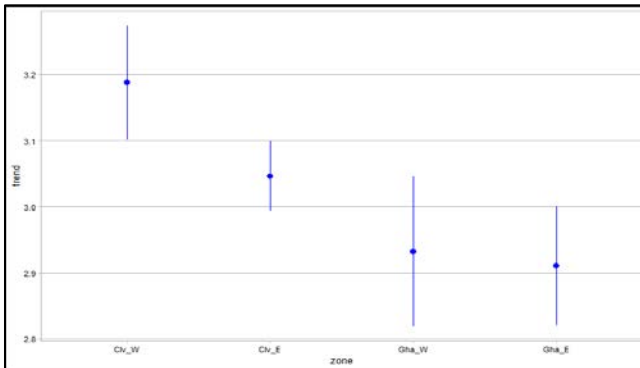
4.6.1. Gutted Weight- Length Relationship



SEX COMBINED

Slope comparisons:

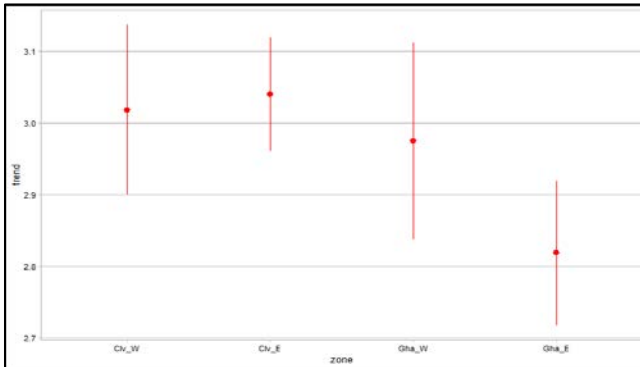
contrast	p.value
Clv_W - Clv_E	0.22
Clv_W - Gha_W	0 **
Clv_W - Gha_E	0 **
Clv_E - Gha_W	0.08
Clv_E - Gha_E	0 **
Gha_W - Gha_E	0.62



MALES

Slope comparisons:

contrast	p.value
Clv_W - Clv_E	0.03 **
Clv_W - Gha_W	0 **
Clv_W - Gha_E	0 **
Clv_E - Gha_W	0.28
Clv_E - Gha_E	0.05 **
Gha_W - Gha_E	0.99

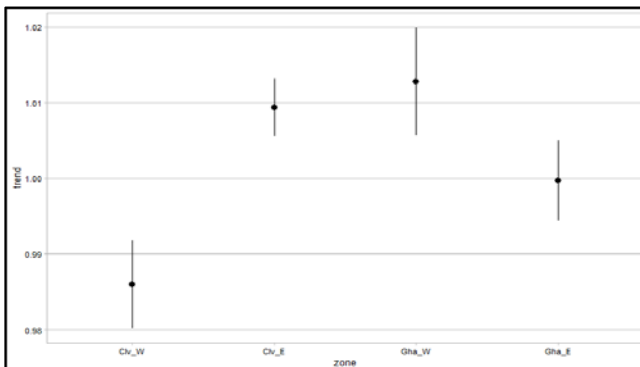


FEMALES

Slope comparisons:

contrast	p.value
Clv_W - Clv_E	0.99
Clv_W - Gha_W	0.97
Clv_W - Gha_E	0.06
Clv_E - Gha_W	0.85
Clv_E - Gha_E	0 **
Gha_W - Gha_E	0.27

4.6.2. Conversion factor

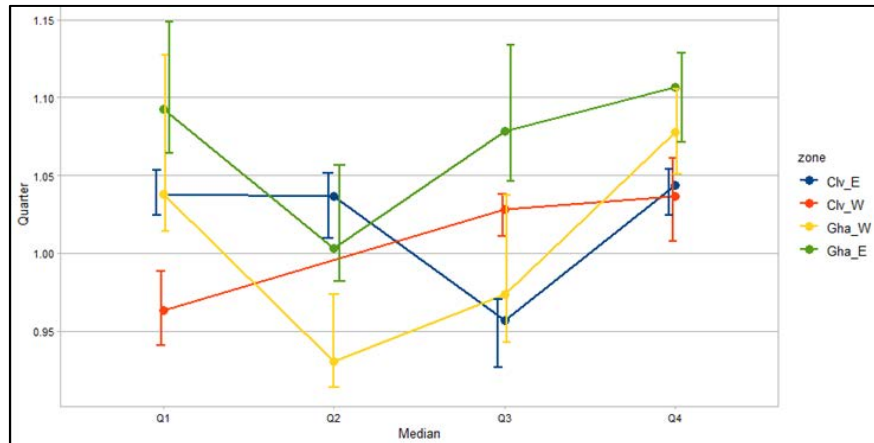


SEX COMBINED

Slope comparisons:

contrast	p.value
Clv_W - Clv_E	0 **
Clv_W - Gha_W	0 **
Clv_W - Gha_E	0 **
Clv_E - Gha_W	0.84
Clv_E - Gha_E	0.02 **
Gha_W - Gha_E	0.02 **

4.6.3. Quarterly condition factors (mature females)



Median comparisons:

group1	group2	p.value
Clv_EQ1	Clv_WQ1	0 **
Gha_WQ1	Clv_WQ1	0 **
Gha_WQ1	Clv_EQ1	1
Gha_EQ1	Clv_WQ1	0 **
Gha_EQ1	Clv_EQ1	0.01 **
Gha_EQ1	Gha_WQ1	1
Gha_WQ2	Clv_EQ2	0 **
Gha_EQ2	Clv_EQ2	1
Gha_EQ2	Gha_WQ2	0 **
Clv_EQ3	Clv_WQ3	0 **
Gha_WQ3	Clv_WQ3	1
Gha_WQ3	Clv_EQ3	1
Gha_EQ3	Clv_WQ3	0.26
Gha_EQ3	Clv_EQ3	0 **
Gha_EQ3	Gha_WQ3	0 **
Clv_EQ4	Clv_WQ4	1
Gha_WQ4	Clv_WQ4	0.09
Gha_WQ4	Clv_EQ4	0.9
Gha_EQ4	Clv_WQ4	0 **
Gha_EQ4	Clv_EQ4	0 **
Gha_EQ4	Gha_WQ4	1

ANNEX 5- Results of the ANOVA for the morphometric characters of the specimens collected (left) and of the Wilks' lambda test for verifying differences among areas with morphometric measurements using DFA (right).

5.1. *Epinephelus aeneus* (GPW)

Tests of Equality of Group Means

	Wilks' Lambda	F	df1	df2	Sig.
m1	,897	12,383	3	325	,000
m2	,909	10,880	3	325	,000
m3	,710	44,338	3	325	,000
m4	,986	1,563	3	325	,198
m5	,919	9,506	3	325	,000
m6	,911	10,575	3	325	,000
m7	,953	5,331	3	325	,001
m8	,963	4,161	3	325	,007
m9	,873	15,774	3	325	,000
m10	,910	10,715	3	325	,000
m11	,848	19,465	3	325	,000
m12	,865	16,922	3	325	,000
m14	,960	4,533	3	325	,004
m16	,922	9,206	3	325	,000

Wilks' Lambda

Test of Function(s)	Wilks' Lambda	Chi-square	df	Sig.
1 through 3	,512	216,209	18	,000
2 through 3	,793	75,017	10	,000
3	,943	18,881	4	,001

5.2. *Penaeus notialis* (SOP)

Tests of Equality of Group Means

	Wilks' Lambda	F	df1	df2	Sig.
m1	,546	36,988	5	222	,000
m2	,771	13,165	5	222	,000
m3	,634	25,681	5	222	,000
m4	,595	30,228	5	222	,000
m5	,719	17,338	5	222	,000
m6	,671	21,781	5	222	,000
m7	,695	19,520	5	222	,000
m8	,693	19,633	5	222	,000
m9	,678	21,116	5	222	,000
m10	,643	24,643	5	222	,000
m11	,591	30,790	5	222	,000
m12	,734	16,077	5	222	,000
m13	,715	17,735	5	222	,000
m14	,720	17,271	5	222	,000
m16	,661	22,749	5	222	,000
m17	,802	10,928	5	222	,000
m18	,665	22,376	5	222	,000
m19	,631	26,008	5	222	,000
m21	,607	28,747	5	222	,000
m22	,672	21,668	5	222	,000
m23	,717	17,530	5	222	,000
m24	,722	17,077	5	222	,000
m26	,621	27,091	5	222	,000
m27	,750	14,809	5	222	,000
m28	,694	19,601	5	222	,000
m29	,730	16,426	5	222	,000
m31	,695	19,487	5	222	,000
m32	,574	32,901	5	222	,000
m33	,721	17,154	5	222	,000
m34	,534	38,764	5	222	,000
m36	,622	27,004	5	222	,000
m37	,659	22,939	5	222	,000
m38	,556	35,442	5	222	,000
m39	,710	18,094	5	222	,000
m40	,687	20,217	5	222	,000
m41	,680	20,922	5	222	,000
m42	,668	22,108	5	222	,000
m43	,726	16,751	5	222	,000
m44	,590	30,905	5	222	,000
m45	,703	18,766	5	222	,000
m46	,663	22,581	5	222	,000
m47	,480	48,187	5	222	,000
m48	,619	27,320	5	222	,000
m49	,934	3,134	5	222	,009
m50	,975	1,149	5	222	,335
m52	,519	41,171	5	222	,000
m53	,830	9,124	5	222	,000

Wilks' Lambda

Test of Function(s)	Wilks' Lambda	Chi-square	df	Sig.
1 through 5	,015	905,345	85	,000
2 through 5	,057	619,152	64	,000
3 through 5	,185	363,219	45	,000
4 through 5	,385	205,843	28	,000
5	,682	82,529	13	,000

5.3.- *Pagrus caeruleostictus* (BSC)

Tests of Equality of Group Means

	Wilks' Lambda	F	df1	df2	Sig.
m1	,752	40,597	2	246	,000
m2	,833	24,659	2	246	,000
m3	,432	161,662	2	246	,000
m4	,469	138,996	2	246	,000
m5	,953	6,087	2	246	,003
m6	,889	15,316	2	246	,000
m7	,864	19,356	2	246	,000
m8	,854	20,971	2	246	,000
m9	,967	4,179	2	246	,016
m10	,986	1,800	2	246	,167
m11	,468	139,994	2	246	,000
m12	,545	102,769	2	246	,000
m14	,642	68,520	2	246	,000
m16	,686	56,413	2	246	,000

Wilks' Lambda

Test of Function(s)	Wilks' Lambda	Chi-square	df	Sig.
1 through 2	,167	433,709	16	,000
2	,703	85,517	7	,000

5.4. *Pseudolithus elongatus* (PSE)

Tests of Equality of Group Means

	Lambda de Wilks	F	gl1	gl2	Sig.
m1	,902	9,338	3	257	,000
m2	,893	10,289	3	257	,000
m3	,869	12,884	3	257	,000
m4	,730	31,653	3	257	,000
m5	,957	3,877	3	257	,010
m6	,831	17,446	3	257	,000
m7	,965	3,095	3	257	,027
m8	,872	12,587	3	257	,000
m9	,918	7,680	3	257	,000
m10	,930	6,465	3	257	,000
m11	,932	6,254	3	257	,000
m12	,906	8,926	3	257	,000
m13	,848	15,312	3	257	,000
m15	,943	5,193	3	257	,002
m17	,871	12,721	3	257	,000
m19	,917	7,783	3	257	,000

Wilks' Lambda

Test of Function(s)	Wilks' Lambda	Chi-square	df	Sig.
1 through 3	,263	314,977	21	,000
2 through 3	,629	109,158	12	,000
3	,824	45,642	5	,000

5.5. *Pseudolithus senegalensis* (PSS)

Tests of Equality of Group Means

	Wilks' Lambda	F	df1	df2	Sig.
m1	,752	20,432	3	186	,000
m2	,708	25,595	3	186	,000
m3	,694	27,360	3	186	,000
m4	,632	36,129	3	186	,000
m5	,764	19,105	3	186	,000
m6	,784	17,123	3	186	,000
m7	,762	19,361	3	186	,000
m8	,938	4,089	3	186	,008
m9	,803	15,184	3	186	,000
m10	,572	46,420	3	186	,000
m11	,647	33,823	3	186	,000
m12	,670	30,532	3	186	,000
m13	,663	31,567	3	186	,000
m15	,708	25,626	3	186	,000
m17	,567	47,264	3	186	,000
m19	,835	12,240	3	186	,000

Wilks' Lambda

Test of Function(s)	Wilks' Lambda	Chi-square	df	Sig.
1 through 3	,272	240,367	15	,000
2 through 3	,658	77,100	8	,000
3	,975	4,760	3	,190

5.6. *Pagellus bellottii* (PAR)

Tests of Equality of Group Means

	Wilks' Lambda	F	df1	df2	Sig.
m1	,611	44,138	3	208	,000
m2	,959	2,938	3	208	,034
m3	,846	12,648	3	208	,000
m4	,865	10,798	3	208	,000
m5	,753	22,770	3	208	,000
m6	,642	38,638	3	208	,000
m7	,629	40,828	3	208	,000
m8	,858	11,483	3	208	,000
m9	,670	34,180	3	208	,000
m10	,930	5,212	3	208	,002
m11	,883	9,163	3	208	,000
m12	,970	2,180	3	208	,091
m14	,971	2,037	3	208	,110
m16	,714	27,790	3	208	,000

Wilks' Lambda

Test of Function(s)	Wilks' Lambda	Chi-square	df	Sig.
1 through 3	,128	423,081	21	,000
2 through 3	,605	103,204	12	,000
3	,899	21,845	5	,001