UNIVERSIDADE TÉCNICA DO ATLÂNTICO
INSTITUTO DE ENGENHARIA E CIÊNCIAS DO MAR
WEST AFRICAN SCIENCE SERVICE CENTRE ON CLIMATE CHANGE AND ADAPTED LAND USE

Master Thesis

A REGIONAL TAXONOMIC ANALYSIS OF COMMERCIAL FISHES IN CABO VERDE BASED ON GENETIC DATA: A CASE STUDY ON THE HAEMULIDAE FAMILY

SARAH SOFIA DIAS DOS SANTOS

Master Research Program on Climate Change and Marine Sciences

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Sarah Sofia Dias Dos Santos

Master's thesis presented to obtain the master's degree in Climate Change and Marine Sciences, by the Institute of Engineering and Marine Sciences, Atlantic Technical University in the framework of the West African Science Service Centre on Climate Change and Adapted Land Use

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Panel defense

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Examiner 1

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Dedication

I dedicate this thesis to my daughter, Emília Santos, who was the best blessing that happened to me during this master's degree.

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Resumo

Haemulidae é uma das famílias de peixes que tem gerado confusão no que diz respeito à sua taxonomia e distribuição geográfica. Em Cabo Verde, as espécies são amplamente capturadas para consumo e, portanto, é imperativo esclarecer as espécies que têm uma distribuição no arquipélago, bem como fazer a relação filogenética entre as espécies que fazem parte da mesma família. Assim, o principal objetivo deste estudo foi avaliar a relação entre espécies de Haemulidae, com base em dados morfológicos e diversidade genética utilizando um marcador genético padrão mtDNA. Para tal, foram recolhidos espécimes para análise morfológica e foram sequenciadas 35 amostras de tecido de Cabo Verde. No total, 75 sequências de ADN provinham de fontes publicadas e não publicadas. Os dados morfológicos e os tecidos recolhidos mostraram a existência de 4 espécies em Cabo Verde, *Pomadasys incisus*, *Parapristipoma humile*, *Pomadasys jubelini*, e *Parapristipoma macrops*, que foram confirmadas por DNA barcoding. O valor de máxima verosimilhança entre as espécies de Haemulidae foi moderado e as distâncias intra-específicas variaram de 0,005 a 0,301, ajudando a confirmar as 4 espécies. A espécie *P. incisus* demonstrou monofilia absoluta, confirmando o seu estatuto distinto. Enquanto *P. jubelini* apresentou-se como monofilética, ficou agrupado com *Pomadasys perotaei* e *P. rogerii*. Por outro lado, *Brachydeuterus auritus* mostrou ser uma espécie monofilética. Houve a total separação de de *P. humile*, no entanto neste mesmo clado surgiu uma espécie de *Pomadasys rogerii* indicando um potencial erro taxonómico. Os espécimes de *P. macrops* apresentam uma separação dentro do ramo de *P. humile*, revelando uma grande proximidade genética. Na estrutura populacional de *P. rogerii*, observou-se uma variação genética mínima e *P. macrops* e *P. humile* apresentaram uma baixa distância genética, evidente numa rede de haplótipos com passos mutacionais esparsos. Por outro lado, as amostras de *P. incisus* exibiram uma notável variação genética de Cabo Verde em comparação com as contrapartes do Mediterrâneo e do Atlântico Sul. Isto revelou dois grupos distintos: um grupo exclusivo de Cabo Verde e outro que inclui haplótipos de diferentes regiões. Este estudo realça a fusão da morfologia e da genética para uma melhor identificação das espécies da família Haemulidae. A validação genética das espécies de Cabo Verde ajuda a gestão dos recursos. A investigação contribui com novos conhecimentos, formando uma base para futuros estudos moleculares e taxonómicos, servindo como dados de referência de códigos de barras de ADN.

*Palavras-chave*s: Cabo Verde, DNA barcode, Haemulidae, marcadores moleculares, taxonomia.

Abstract

Haemulidae is one of the fish families that has given confusion regarding to its taxonomy and geographical distribution. In Cabo Verde, the species are widely caught for consumption, and it is therefore imperative to clarify the species that have a distribution in the archipelago, as well as to make the phylogenetic relationship between the species that are part of the same family. So, the main objective of this study was to evaluate the relationship between species of Haemulidae based on morphological data and genetic diversity, using a standard mtDNA genetic marker. To do this, specimens were collected for morphological analysis, and 35 tissue samples from Cabo Verde were sequenced. In total, 75 DNA sequences come from both published and unpublished sources. The morphological data and the tissues collected showed the existence of four (4) species in Cape Verde: *Pomadasys incisus*, *Parapristipoma humile*, *Pomadasys jubelini*, and *Parapristipoma macrops*, which were confirmed by DNA barcoding. The maximum likelihood value among Haemulidae species was moderate, and intraspecific distances ranged from 0.005 to 0.301, helping to confirm the 4 species. The species *P. incisus* demonstrated absolute monophyly, confirming its distinct status. While P. jubelini was monophyletic, it was grouped with *Pomadasys perotaei* and *Pomadasys rogerii*. On the other hand, *Brachydeuterus auritus* proved to be a monophyletic species. There was a complete separation of *P. humile*, but in this same clade, a species of *P. rogerii* appeared, indicating a potential taxonomic error. The *P. macrops* specimens display a separation within the *P. humile* branch, revealing close genetic proximity. In *P. rogerii* population structure, minimal genetic variation was noted and *P. macrops* and *P. humile* displayed low genetic distance, evident in a sparse mutational-step haplotype network. Conversely, *P. incisus* samples exhibited notable Cabo Verde genetic variance compared to Mediterranean and South Atlantic counterparts. This revealed two distinct groups: an exclusive Cabo Verde group and another including haplotypes from different regions. This study highlights the fusion of morphology and genetics to better identify the species of the Haemulidae family. Genetic validation of Cape Verdean species helps resource management. The research contributes new knowledge, forming a basis for future molecular and taxonomic studies, and serving as reference data for DNA barcodes.

Keywords: Cabo Verde, DNA barcodes, Haemulidae, molecular markers, taxonomy.

Abbreviations and acronyms

CoxI	Cytochrome C Oxidase I
CytB	Cytochrome B
DNA	Deoxyribonucleic acid
EDTA	Ethylenediaminetetraacetic acid
FMSV	São Vicente fish market
HVS	Hypervariable Sequence
ITCZ	Intertropical Convergence Zone
mtDNA	Mitochondrial DNA
PCR	Polymerase Chain Reaction
rDNA	Ribosomal DNA
RNA	Ribonucleic acid
TBE	Tris/Borate/EDTA
SYBR	Cyanine dye is used as a nucleic acid stain in molecular biology
Fig.	Figure
ML	Maximum Likelihood

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1. Introduction

Over the last 25 years, the ichthyofauna of the Cabo Verde archipelago has been the target of several studies where books, articles, and taxonomic revisions have been published. In research, new species have been described, and the revision of many families, many of which have species of great commercial value Freitas et al. (2018). One fish family well caught in Cabo Verde and with some commercial value is undoubtedly the Haemulidae family, commonly known as the Grunt family, order Perciformes. This family includes over 150 species distributed in tropical and subtropical waters around the world. They are primarily found in the Atlantic, Indian, and Pacific Oceans. They are divided into two subfamilies, *Haemulinae* (Grunts) and *Plectorhynchinae* (Sweetlips), with approximately 17 genera and 145 species (Tavera et al. 2012).

Grunt fish are named for their sound, which resembles a grunting noise produced by grinding their teeth together. They are primarily reef-dwelling species inhabiting coral reefs, rocky areas, seagrass beds, and sandy bottoms. They are usually found in shallow coastal waters, although some species can be found at depths of several hundred meters. Their distribution is considered to be circumglobally and usually found in subtropical, tropical and temperate waters. They are known for forming schools, and these aggregations can include both individuals of the same species or different species of grunts (Appeldoorn et al., 2009).

Adults Grunts are generally more inactive during the day, sheltering near or under submerged objects, rocks, or coral reefs, using the nighttime to feed. Smaller species feed mainly on plankton. However, the majority are carnivorous, feeding on a wide variety of benthic invertebrates, including polychaete worms, crustaceans, and clams, among others. They are pelagic spawners. The maximum recorded size is about 600 mm. Most are considered to be of commercial importance for human consumption (Burkenroad, 1930; Lévêque et al., 1990).

Among the species of the family Hamulidae, known for Cabo Verde, it can be considered that only *Parapristipoma humile* Bowdich, (1825) is endemic Freitas, (2014). Different methods have been adopted for the study of Cabo Verde ichthyofaunal. By genetic variability of species and populations on oceanic islands, is possible to characterize local genetic variability, translating into altered adaptive capacity or response to habitat change. In this case, we want to base ourselves on genetic data due to the accuracy of molecular identification systems that have been improved with the advances in molecular biology methods (Hebert et al., 2003).

Reiner (1996) is one of the first and most important fish publications, according to Hanel & John, (2015) This author listed 520 species, most probably because he included species he thought likely to be found in this area. Unfortunately, it has caused confusion and taxonomic errors. Soon, to make a checklist with only the fish species of this region, studies have been made recently, such as Wirtz et al., (2013) who listed 315 coastal fish species, and Hanel & John, (2015) reviewed ignored literature and included mesopelagic species in their inventory (Freitas, 2014)

To describe species, taxonomists always use morphological analyses that generally have a fundamental role in the recognition of the species or higher categories and often require the use of equipment (optical or electron microscopy) or special techniques (e.g., diaphanization and staining) Becker, (2005). Taxonomic studies and descriptions of new species for science in this area all bring updates to the checklist of the Cabo Verde ichthyofauna. Since Reiner (1996), the Cabo Verde fish listing has been subject to several revisions and some families still need special attention. Freitas (2014), believes that it has been creating a lot of confusion about this family classification and not solving the problem, listing the doubtful species or putting some species lacking information. In this study, in addition to the phylogenetic study, the target species chosen were based on the need to confirm doubtful species, namely Wirtz et al. (2013) list, the species *Brachydeuterus auritus* (Valenciennes in Cuvier & Valenciennes, 1832), *Parapristipoma macrops* (Pellegrin, 1912), *Plectorhinchus mediterraneus* (Guichenot, 1850), *Pomadasys suillus* (Cuvier in Cuvier & Valenciennes, 1830) and *Pomadasys rogerii* (Cuvier in Cuvier & Valenciennes, 1830).

Using genetic data to collate the occurrence or absence of Haemulidae species may be the way forward for the Cape Verdean species question. When classical taxonomy does not help or morphology is compromised, genetic species identification helps match an unknown sample to a known reference sample in an online database by comparing gene sequences, usually mitochondrial DNA (mtDNA). The recent introduction of DNA barcoding has suggested that the mitochondrial DNA gene cytochrome c oxidase I (CoxI) be used as a 'barcode' for most animals. This is a rapidly expanding area of research supported by an international consortium of major natural history museums, herbaria, and other organizations (Dawnay et al., 2007). Although much controversy exists regarding the choice of Mitochondrial DNA (mtDNA) markers for taxon identification, it is widely used in many studies. The control region (which includes D-loop and the hypervariable sequence - HVS), the cytochrome B (CytB) and cytochrome C oxidase I (CoxI) have been the most widely used

sequences to identify and catalogue species, discriminate subspecies and study evolution. Hebert et al. (2003) believe that the information contained in the Cytochrome Oxidase subunit I (CoxI) gene alone in mtDNA is sufficient to allow the identification of animal taxa to species level, as well as the critical discovery of new species and also critical species. It should be noted that due to its speed, reliability, and accessibility, mtDNA has been used in several studies. Since then, large amounts of DNA barcoding data have accumulated in publicly available databases (Costa & Antunes, 2012).

Due to controversial Hamulidae relationships, there is a need to re-evaluate phylogenetic relationships, since neither morphological nor molecular studies have been performed focusing on the extant species in Cabo Verde. The family is raising questions about new species, doubtful, erroneous IDs, and cryptic species. Could DNA barcoding be a more accurate method? Do we already have genetic data on these dubious species? These and other questions can be raised in this study.

1.1. Objectives of the work

This study aimed at evaluating the relationship between the Haemulidae species (Teleostei) in Cabo Verde waters based on morphological characters and genetic diversity using a standard mtDNA genetic marker. Within this context, the specific objectives are:

a) To validate the presence of grunt species in the Cabo Verde archipelago;

b) To contribute to the knowledge of the systematic relationships across the family Haemulidae based on mitochondrial DNA (mtDNA);

c) To understand whether the observed genetic divergence is accompanied by morphological differentiation;

d) To perform a phylogeographical study comparing Cabo Verdean species with other regions.

2. Literature review

Taxonomy is known as the science where the identification of organisms is made based on the comparison of their characteristics, which aims to group organisms in an organized and systematic way, allowing the identification, naming, and study of the different groups of living beings, that is, it makes it possible to describe diversity, find an organization between groups and understand the processes that generate this biological variety (Bonisson, 2014). In this way, the branch of biology that studies the classification of organisms is called Systematics. It comprised three evolutionary schools: the gradist, pheneticist, and phylogeneticist. The gradient school is based on observable morphological characteristics and seeks to create a linear classification (the idea that species are grouped based on their general similarity). The pheneticist school uses quantitative methods to assess the distance or similarity between organisms, involving comparing many morphological or molecular characters to quantify the differences between taxa (Araújo-De-Almeida & Santos, 2014). The phylogeneticist school is concerned with reconstructing the phylogenetic relationships between organisms, i.e., their evolutionary history. It uses derived shared characteristics, called synapomorphies, to infer the existence of clades, which are monophyletic groups containing a common ancestor and all its descendants, creating a classification that reflects the phylogenetic tree of organisms, representing their real evolutionary relationships (Soares & Nakamura, 2021)

Phylogenetic analysis can be based on morphological or molecular characters or a combination A phylogenetic analysis based on DNA/RNA/protein sequences aims to establish a relationship/unrelatedness between various groups of organisms or between various regions where organisms are distributed. In principle, everything can be compared, from whole genomes to individual genes. In the case of individual genes, phylogeneticists can compare the whole gene or just the coding regions. The choice of marker to be used may be based on several criteria depending on the objectives of the study (Avise & Wollenberg, 1997) Nowadays, the combined analyses of genetic data and morphological data allow us to renew the concept of "species" and to create lineages that would otherwise not be possible. The geographical separation of an ancestral species into separate lineages is an important part of allopatric speciation in the most common species concepts, even if different authors disagree on the characteristics that these lineages must eventually achieve for the speciation process to be considered complete (Wiens, 2004).

Several phylogenetic and morphological studies have been carried out using the islands of the Macaronesian region as a study area. The region is made up of four archipelagos: Açores,

Madeira, Canary Islands, and Cabo Verde Islands. Regarding marine species composition, there is a high degree of variation between Cabo Verde and the other archipelagos, which usually occurs due to mechanisms such as dispersal ability, environmental conditions, type of reproduction and geological age of the islands, identified as important in structuring communities on oceanic islands (Pablo et al., 2013). In the case of ichthyological fauna for example, Cabo Verde contains many species of tropical coastal fish, which cannot be found in Senegal and the other archipelagos, probably due to cold sea surface temperature of around 14 ºC caused by the upwelling system (Wirtz et al., 2013). Several factors influence the oceanic water circulation around Cabo Verde islands: we have a large-scale surface circulation station through the meridional displacement of the Intertropical Convergence Zone (ITCZ) and the seasonal variability of trade winds.

Regarding Brito et al. (2007), the archipelago of Cabo Verde shows species predominantly from Guinea, followed by tropical-subtropical fishes, denominated Amphi-Atlantic species, and several endemic coastal species. It is necessary to constantly review the art of research into its ichthyofauna, due to recent studies (taxonomy and description of new species, intra-specific fish relationships, among others) carried out on the seamounts. Concerning Freitas, 2014, since the mid-1980s, there have been several studies that have brought new data for a review of Cabo Verde's ichthyofauna (e.g., (Brito & Miller, 2001; Freitas, 2014; J. A. P. González et al., 2009; Wirtz et al., 2013).

Another way to approach the ichthyofauna is the fish larvae study, so John & Hanel (2008) made the collection based on neuston stations and trawl within the Cabo Verde archipelago, and identification was based on meristic and morphometric data specific to late flexion and post-flexion larvae and found that *Chromis lubbocki* was similar in morphometry and pigmentation to the Mediterranean and also suggested the occurrence of *Chromis* cyanide in Cabo Verde waters. After this, Hanel et al., (2010) in their study on the distribution, composition and abundance of fish larvae in the Senghor seamount, northeast of the Cape Verde Archipelago, and the results showed a Cape Verde, obtained results that showed a diverse fish community, where they caught 68 specimens of 37 species.

Hanel & John (2015), revised the Caboverdean fish checklist based on almost all existing inventories (both literature and digital), and new unpublished data, both own and foreign. They have achieved a total number of 1046 named species listed where 779 valid species as supported by at least one credible record (‘native’), built on Clofeta 1990. Of these, 91 species represent new records in the listings. A commentary on 64 West African mainland species

erroneously attributed to the archipelago. 12 records are Uncertainties (unclear taxonomic status according to identification), which can maybe be proven in the future to be present, and 116 species present unsatisfactory zoogeographic knowledge. They noted that the Fishbase compilation[1] is mostly based on studies by Reiner in 2005 and 1996, i.e., they have not added more recent studies (e.g., Brito et al., 2007). The checklist included many widely distributed species (mostly mesopelagic to bathypelagic species), demonstrating a certain peculiarity for the coastal fishes of the Cabo Verde Archipelago.

In Wirtz et al (2013), the coastal fish species documented for the Cabo Verde waters, we found ten valid species for the Haemulidae family, five of which need confirmation. Also, it is believed that there was some taxonomic confusion with some other species, not found in Cabo Verde waters.

One of the subjects covered in Freitas, (2014), is fish species considered endemic to Cabo Verde. Among the Hamulidae species known from Cabo Verde, it can be considered that *Parapristipoma humile* is possibly an endemic species. This is because it can often be confused with *Parapristipoma octolineatum* (Valenciennes in Cuvier & Valenciennes, 1833), which raises doubts about its distribution. Doubts about this species being endemic or not, come from many years ago, Brito et al. (2007), confuse *P. humile* with its congener *P. octolineatum* (Valenciennes, 1833), a species described from Senegal and also present in Cabo Verde, although less abundant. This may be related to the loss of color of the fish after fishing, as *P. humile* shows more clear lines. However, they are thin and not always visible, disappearing quickly after death, while in *P. octolineatum* the clear lines are visible and they are preserved in the fresh animal.

The problems of the classic taxonomy of Cabo Verde ichthyofauna can be solved by genetic analysis. Since the early 2000s Summerer et al. (2001) performed phylogenetic analyses (using 482 *bp.* of the mitochondrial 16S rDNA and 461 *bp.* of the control region) of 16 species of *Diplodus* genera, plus *Oblada melanura*, *Pagellus bogaraveo*, and *Pagellus acarne*, all close of *Diplodus.* The main objective was to resolve the phylogenetic relationships within the *Diplodus* radiation using a more rapidly evolving mitochondrial gene segment and to trace the eco-morphological diversification and biogeographical spread of the genus. These authors recognize that two representatives of *Pagellus* are a sister group to *Diplodus*. They noted that previous studies by Hanel & Sturmbauer, (2000), which were based on a 486 bp segment of the mitochondrial 16S rDNA of all 24 seabream species from the Northeast Atlantic

[1] www.fishbase.se

and the Mediterranean, did not agree with the morphology of missions from previous studies, due to the young evolutionary age and rapid diversification of this group. *D. sargus lineatus*, which is endemic to Cabo Verde islands, was considered the most ancestral division within this clade. This means that the methodology depending on the study case, for better results, has to be well chosen.

We are constantly seeing changes and evolutions in the study methods used and, when it comes to species identification. Recently in genetic studies, the DNA barcode has been used a lot. In these types of studies, a marker is used, which is mitochondrial DNA (mtDNA), the DNA sequence of it is a segment of the cytochrome oxidase I (CoxI) gene. (Hebert et al., 2003). Although the use of this marker shows a high success rate in the rapid diagnosis of various taxonomic groups, DNA barcodes can elicit both enthusiastic and skeptical reactions (Costa & Antunes, 2012).

Moritz & Cicero, (2004) point to the possibility of overlapping levels of intra- and interspecific variation that can be obtained from this marker, which would make it difficult to accurately identify the species of a sample. They highlight in their study of Neotropical species (in this case birds) as they were considered a challenge for DNA barcoding due to the high diversity observed in this region. However, studies with some groups of Neotropical avifauna have shown that DNA barcoding can diagnose a large number of species and can contribute to information about the divergence patterns of the study region.

Costa & Antunes (2012) emphasize that the success of many anticipated applications of DNA barcoding depends on rigorous taxonomic expertise and take this opportunity to highlight applications that they believe will improve our ability to monitor and understand biological diversity and ecosystem functioning. However, studies with some groups of Neotropical avifauna have shown that DNA barcoding can diagnose a large number of species and can contribute to information about the divergence patterns of the study region.

A recent investigation was conducted by Lopes et al., (2021)Lopes et al., (2021)Lopes et al., (2021) to validate the occurrence of the roughear scad, *Decapterus tabl* in Cabo Verde, using both morphological and genetic information, with focus on the gene CoxI.

In 2015 and 2020, several Makarel specimens were captured that proved to be very different from the species reported for this region *(Decapterus macarellus* and *Decapterus punctatus)* to analyze their difference. Morphological data that distinguish it from the other *D. tabl*: larger size (Standard length in mm, 313-348), reddish caudal fin, the serrated margin on the operculum membrane, 10 to 12 branchial fins (first branchial arch) on the upper portion

and 30 to 33 on the lower portion (including rudimentary ones). Genetic barcoding based on mitochondrial cytochrome oxidase I with Bayesian and Maximum Likelihood analysis yielded identical results, revealing well-supported clades for most of the species described for this genus, and the haplotype network of *D. tabl* showed two distinct groups separated by several mutational steps. One of the theories examined regarding rare and new fish species was rooted in the tropicalization phenomena observed in the Canary Islands, which have been recognized as significant factors that enhance the richness of fish species.

Considering the Haemulidae family, Tavera et al. (2012) did a phylogenetic study to understand the evolutionary history and dynamics of habitats and geographic regions. Their diversity can manifest in various ways, whether in physical form (elongated or robust), ecological roles (distinct feeding behaviors and prey preferences), and habitats (from temperate reefs and coral reefs to muddy or sandy seabed).

When Haemulidae fish are identified based on their morphology, one of the outstanding features is the presence of enlarged sensory pores on the chin and the attachment of the sixth infraorbital to the skull (Figure 1), which are considered unusual characteristics in percoids. Within the subfamilies and genera of Haemulidae, the number, shape and position of the pores on the chin are crucial to their identification. Haemulines, have two pores on the chin, a median groove on the chin, or both. Plectorhinchines, on the other hand, have four to six chin pores. (Tavera et al., 2018).

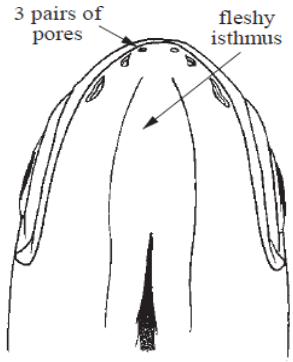


Figure 1: Underside of the head, representing pores at the chin, on a Haemulidae specimen.

This study uses molecular data to provide the most comprehensive phylogeny of the Haemulidae and new world grunts. The monophyly of the Haemulinae and Plectorhinchinae subfamilies proposed earlier is strongly supported by the results. However, some questions remain unresolved, even if the phylogenetic hypotheses are robust at the subfamily and genus

level. Although their phylogenetic hypotheses are robust at the subfamily and the generic level, some questions remain unresolved.

Genetics tools can be the key to solve taxonomic problems (species identification) and also evolutionary history. The rate of molecular evolution of the CoxI gene allows us to distinguish nearby species and phylogeographic groups within the same species. This allows us to highlight some of the main applications that this method can be used: for the identification of cryptic species, species identification from fragments of biological material, the discovery of new species, identification of juvenile and adult forms of the same species, etc. That being said we can say that DNA barcoding can be used as an integrative complement in the taxonomic approach to identifying marine fish and this means that can be beneficial in the management and conservation of marine fish in the coastal region. Which aims at the preservation and management of global biodiversity, it is extremely necessary to have a DNA barcode database representative of the fish diversity in Cabo Verde. Such species identification will allow the addition of one more piece of data that can be useful in combating overfishing, habitat degradation and pollution among others (de Carvalho et al., 2007; Goldstein & DeSalle, 2011; Valentini et al., 2009).

2.1. Morphological examination

All descriptions and other information about the species that were made in this study were based on various bibliographies and databases that are controlled by taxonomic and thematic experts (e.g., Carpenter & De Angelis (2016); multiple databases like WoRMS, FishBase, BOLD Systems; Appeldoorn et al., 2009; Dupérier & Brygoo, 1983; Lévêque et al., 1990).

Systematic account

Order Eupercaria incertae sedis

Family Haemulidae (Gill, 1885)

Description: Haemulids typically have a moderately deep and laterally compressed body shape. Their bodies are oblong, with a distinct head and a slightly arched dorsal profile. Head profile is more or less convex in most species with a terminal or slightly inferior mouth that normally is small to moderate, lips sometimes thick, and they often have robust teeth (teeth

conical, in narrow bands in each jaw, the outer series enlarged, but no strong canines, roof of mouth toothless and posterior margin of suborbital not exposed), which are used for feeding on a variety of prey including crustaceans and small fishes. Chin with two (2) to six (6) pores anteriorly, preopercle with posterior margin slightly concave and serrated, and the opercula can be with or without a spine. The dorsal fin is typically continuous and has spines ten (10) to fourteen (14) strong followed by fourteen (14) soft rays. Pelvic fins below pectoral-fin bases, with one (1) spine and five (5) soft rays; anal fin with tree (3) spines, the second often very prominent, and six (6) to thirteen (13) soft rays and caudal fin emarginate to forked. They possess a well-developed swim bladder. The lateral line is a well-developed system, which is a sensory system consisting of a series of small pores and canals along the sides of the body(Carpenter & De Angelis, 2016).

Coloration: highly variable, displays vibrant coloration often in combination with various shades of yellow, silver, or brown and ranging from uniformly colored to striped, banded, blotched, and spotted(Carpenter & De Angelis, 2016).

Biology and Habitat and Distribution: The fish are usually small to medium-sized, almost all from shallow coastal waters of tropical and subtropical regions, i.e., they can be found in the Atlantic, Indian, and Pacific. Chiefly marine, some brackish, and rarely freshwater(Carpenter & De Angelis, 2016).

Subfamily Haemulinae (Gill, 1885)

The subfamily Haemulinae was first described by Theodore Gill in 1885. Includes a diverse range of marine fishes in various tropical and subtropical regions worldwide.

Description: Most species within the subfamily Haemulinae have a moderately deep and laterally compressed body shape, typically oval or elongated, with a distinct head and a slightly arched dorsal profile. Generally, have a terminal or slightly inferior mouth, positioned towards the front of the head. The dorsal fin of Haemulinae species is typically continuous and contains spines followed by thirteen to sixteen (13-16) soft rays.

Coloration: Many species exhibit vibrant patterns of stripes, bars, or spots on their bodies, often in combination with various shades of yellow, silver, or brown.

Biology and Habitat and Distribution: Primarily found in marine habitats, including

coral reefs, rocky bottoms, and seagrass beds. Can inhabit tropical and subtropical regions around the world, including the Atlantic, Pacific, and Indian Oceans. They are often considered important members of reef fish communities, contributing to ecosystem dynamics and functioning.

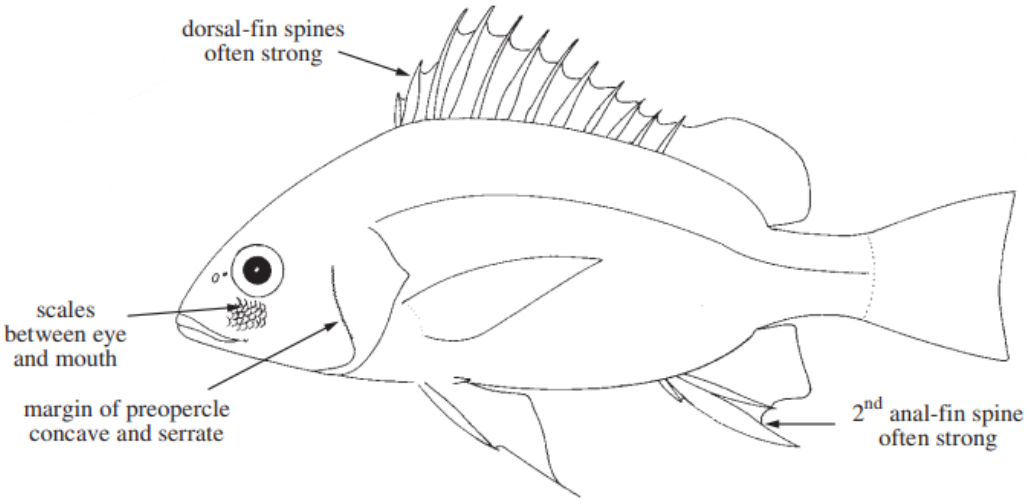


Figure 2: Haemulinae shape (Source: second the catalogue of FAO, 2016).

Genus *Pomadasys* (Lacepède, 1802)

Description: The species typically have a moderately deep and laterally compressed body shape, elongated and slightly oval, with a distinct head and a slightly arched dorsal profile. The dorsal fin is single, separated by a large notch, with twelve to fourteen (12-14) strong spines and twelve to sixteen (12-16) soft rays, and at the base is a row of scales. The pectoral fins have one unsegmented and fourteen to fifteen (14-15) soft rays (Table 1). The anal fin with three (3) spines and seven (7) rays, and the second anal fin spine is thicker than the others. Caudal rays emarginate, with small interradial scales covering at least one-third of the rays. The lateral line is located in the upper third of the body, arched up to the peduncle. This genus has a fleshy mouth with a small opening and jaws of the same size. Teeth aciculate, small, and in more than one series; nostrils broad with two pores situated near the orbit. Mentonian region with two anterior pores and a posterior median sulcus posterior median groove. Denticulations are evident on the preopercle, with exposed scapular bone with its denticulated end. Single, broad, silver-colored swim bladder(Lévêque et al., 1990).

Coloration: This can be quite diverse and vary significantly among different species. They may have brown, grey, and silver tones; stripes and bars are common patterns seen as vertical or diagonal and are often more prominent in juveniles than adults. Some *Pomadasys* species have a distinct dark spot (circular or oval) or ocellus on the caudal peduncle. They can present sexual dimorphism (differences in coloration between males and females)(Carpenter & De Angelis, 2016).

Biology and Habitat: They are usually associated with shallow coastal waters, including coral reefs, rocky bottoms, seagrass beds, estuaries, and mangrove areas. are widely distributed in tropical and subtropical waters of the Atlantic, Indian, and Pacific Oceans(Appeldoorn et al., 2009).

Distribution: The distribution of *Pomadasys* species is influenced by factors such as water temperature, salinity, food availability, and others. So, they have a widespread distribution in tropical and subtropical waters of the Atlantic, Indian, and Pacific Oceans(Appeldoorn et al., 2009).

Table 1: Main morphological characteristics of Pomadasys Genus.

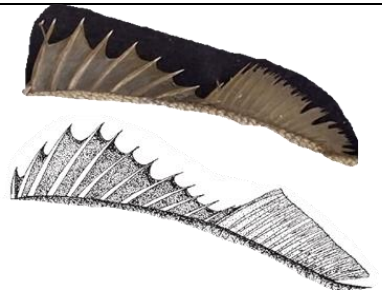
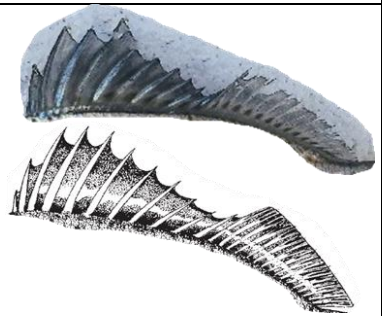
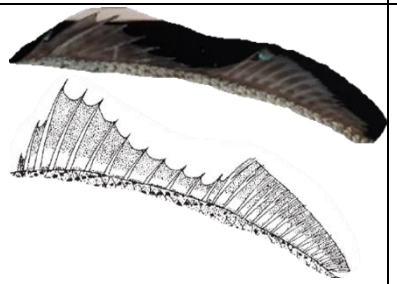
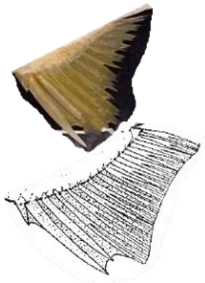
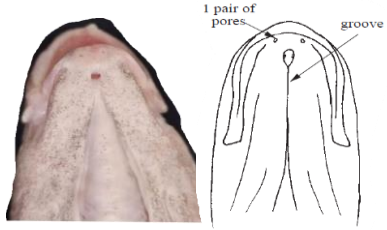
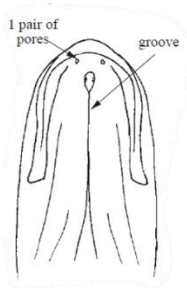
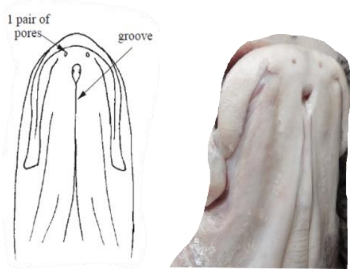
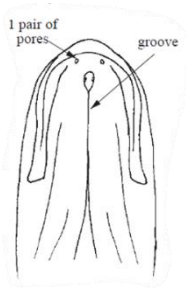
Genus *Pomadasys* (Lacepède, 1802)				
Species **Morphological characteristics**	*Pomadasys incisus* (Bowdich, 1825)	*Pomadasys perotaei* (Cuvier, 1830)	*Pomadasys jubelini* (Cuvier, 1830)	*Pomadasys rogerii* (Cuvier, 1830)
Dorsal-fin rays				
	2 dorsal fins, in which dorsal spines (total): 12 and soft dorsal rays (total): 15-18.	Dorsal fin with 10 to 12 spines and 15 or 17 soft rays.	Dorsal spines (total): 11 – 12 and dorsal soft rays (total): 15-17.	Dorsal fin with 12 spines and 14 to 16 soft rays; the first soft ray is longer than the last spine.
Anal-fin rays				
	3 Anal spines and 11 -13 soft anal rays.	Anal fin with 3 spines and typically 10 soft rays; the first spine is very short, the second long.	Anal fin with 3 spines and 8 – 10 anal soft rays.	Anal fin with 3 spines, and typically 9 or 10 soft rays; the first spine very short, the second long.

Table 4: (continued)

Species **Morphological characteristics**	*Pomadasys incisus* (Bowdich, 1825)	*Pomadasys perotaei* (Cuvier, 1830)	*Pomadasys jubelini* (Cuvier, 1830)	*Pomadasys rogerii* (Cuvier, 1830)
Snout, Mouth and Eye				
	Snout length 0.9 to 1.4 times the orbit diameter; orbit diameter 3.2 to 3.8 times in head length; mouth slightly oblique.	Snout length less than orbit diameter; eye large, orbit diameter 4.0 to 4.3 times in head length; mouth moderately small, barely reaching to anterior eye margin.	Snout long in large individuals and pointed, its length 0.8 to 1.1 times in orbit diameter; eye moderately small, orbit diameter 3.0 to 3.6 times in head length; mouth slightly oblique.	Snout length 0.6 to 1.1 times in orbit diameter; eye moderately small, orbit diameter 2.9 to 4.2 times in head length; mouth slightly oblique.
Pores				
	1 pair of small chin pores at symphysis of low lip and a single pit opening to a pair of pores at symphysis of the lower jaw.	1 pair of small chin pores at symphysis of low lip and a single pit opening to a pair of pores at symphysis of the lower jaw.	1 pair of small chin pores at symphysis of low lip and a single pit opening to a pair of pores at symphysis of the lower jaw.	1 pair of small chin pores at symphysis of low lip and a single pit opening to a pair of pores at symphysis of the lower jaw.

***Pomadasys incisus* (Bowdich, 1825)**

(Fig. 3a))

Synonyms: *Orthopristis bennetti* (Lowe, 1838); *Pomadasis bennettii* (Lowe, 1838); *Pomadasys benetti* (Lowe, 1838); *Pomadasys bennetti* (Lowe, 1838); *Pomadasys bennettii* (Lowe, 1838); *Pristipoma bennettii* Lowe, 1838; *Pristipoma ronchus* Valenciennes, 1838.

Description: The body is oblong. Have a short snout, about equal to eye diameter. A small, with the maxilla not reaching the anterior edge of the eye. This species has two (2) anterior pores on the chin, followed by a median pit. Have two fins where the first dorsal fin with twelve (12) spines and the second dorsal fin with fifteen to sixteen (15-16) soft rays; the anal fin has tree (3) spines and eleven to thirteen (11-13) soft rays and normally the third anal fin spine is equal to or longer than the second one. Twenty to seven (27) Vertebrae. Caudal fin forked; scales slightly ctenoid; seven to eight (7-8) scale rows between lateral line and middle of spinous dorsal-fin base(Carpenter & De Angelis, 2016).

Coloration: black dark brown, silvery white on the belly. Sometimes large dark spots on the back and flanks, but never small spots or bands; a black spot at the upper angle of the opercle(Carpenter & De Angelis, 2016).

Distribution: Eastern Atlantic: Straits of Gibraltar to Angola. Also, Madeira, the Canaries, and Cabo Verde Islands. Northward extending into the western Mediterranean(Carpenter & De Angelis, 2016).

Biology and Habitat: Feeds on bottom and near-bottom invertebrates. Found over hard bottoms and sand and also can enter estuaries and lagoons at sexual maturation(Carpenter & De Angelis, 2016).

***Pomadasys perotaei* (Cuvier, 1830)**

(Fig. 5b))

Synonym: *Pomadasys peroteti* (mispelling of Cuvier 1830); *P. perotoei* (Cuvier, 1830) *P. jubelini* and *P. rogerii* as misidentification.

Description: Body oblong and compressed. The snout length is less than the orbit diameter. Large eye, with an orbit diameter 4.0 to 4.3 times in head length rather short, about equal to eye diameter. Has 1 pair of small chin pores at the symphysis of the lower lip and a single pit opening to a pair of pores at the symphysis of the lower jaw. Fifteen to seventeen (15 -17) gill rakers on the lower limb of the first arch. Have two fins where we have the first dorsal fin with eleven to thirteen (11-13) spines, where the last dorsal spine is about the same length or even shorter, exceptionally somewhat longer, than the penultimate one. The second fin has fifteen to seventeen (15-17) soft rays. The anal fin has tree (3) spines (the first spine is very short, the second long, third anal spine is mostly much thinner) and nine to ten (9-10) soft rays. The pectoral fin is very long, almost reaching the level of the anus. Caudal fin forked. The scales are slightly ctenoid. Has six to eight (6-8) scale rows between the lateral line and the middle of the spinous dorsal-fin base(Carpenter & De Angelis, 2016).

Coloration: Back silvery grey with a bluish tinge, belly silvery. On the back and flanks it has irregularly distributed light brown spots. Spots are sometimes arranged in oblique, curved rows anteriorly above the lateral line. A dark or yellow spot is always present on the upper angle of the operculum(Carpenter & De Angelis, 2016).

Distribution: Eastern Atlantic: West coast of Africa (from Senegal to Angola) and also Mauritania(Carpenter & De Angelis, 2016).

Biology and Habitat: Feeds on other fish, shrimps, crabs, mollusks, annelids, zooplankton, and detritus. Found over sand and mud in coastal waters throughout its range, including brackish water habitats(Carpenter & De Angelis, 2016).

***Pomadasys jubelini* (Cuvier, 1830)**

(Fig. 3c))

Synonym: *Pomadasis jubelini* (Cuvier, 1830), *Pristipoma jubelini* (Cuvier, 1830).

Description: Body oblong and compressed. The snout is large and pointed, in large individuals, obtuse, distinctly longer than the eye diameter. The eye is moderately small and the mouth is slightly oblique. Have 1 pair of small chin pores at the symphysis of the low lip and a single pit opening to a pair of pores at the symphysis of the lower jaw. Twelve to fifteen (12-15) gill rakers on the lower limb of the first arch. The first dorsal fin has eleven to twelve

(11-12) spines and the second one has fifteen to seventeen (15-17) soft rays. The anal fin has tree (3) spines and eight to ten (8-10) soft rays, where the first spine is very short and the second is long. The caudal fin is strongly emarginate(Carpenter & De Angelis, 2016).

Coloration: Background silvery, back and sides with small dark spots arranged in sinuous oblique or horizontal lines. Fins grey, the dorsal with a light longitudinal band. Normally has a golden yellow blotch on the snout and a yellow-golden to darkish blotch on the upper angle of the opercle(Carpenter & De Angelis, 2016).

Distribution: West African coast from Mauritania to southern Angola(Carpenter & De Angelis, 2016).

Biology and Habitat: Feeds on fish and benthic crustaceans as well as on mollusks and worms. Inhabit sandy and muddy bottoms of coastal waters and estuaries. Sometimes found in freshwater(Carpenter & De Angelis, 2016).

Remarks: Second, Wirtz et al., (2013), couldn't find any evidence for the presence of this species in the Cabo Verde Islands; not a valid record. However, in the catalogue of FAO 2016, they say that Previous FAO and other publications have illustrated *Pomadasys jubelini* as *P. rogerii* and vice versa.

***Pomadasys rogerii* (Cuvier, 1830)**

(Fig. 3d))

Synonym: *Pomadasys rogeri* (Cuvier, 1830), *Pristipoma rogerii* (Cuvier, 1830).

Description: Body oblong and compressed. Snout length 0.6 to 1.1 times in orbit diameter. The eye is moderately small and the mouth is slightly oblique. Has one (1) pair of small chin pores at the symphysis of the low lip and a single pit opening to a pair of pores at the symphysis of the lower jaw. Eleven to fifteen (11-15) gill rakers on the lower limb of the first arch. The dorsal fin with twelve (12) spines and fourteen to sixteen (14-16) soft rays and the first soft ray is longer than the last spine. The anal fin has tree (3) spines, typically nine or ten (9 / 10) soft rays with the first spine very short and the second long. The caudal fin emarginate(Carpenter & De Angelis, 2016).

Coloration: Background silvery, lighter ventrally, with blackish or dark brown rounded spots irregularly spread on back and sides; upper back anterior to line from origin of dorsal fin to origin of lateral line typically without spots or with a few faint spots; typically, faint spots or no spots present in scale rows above, below and on anterior scales of lateral line; fins whitish to blackish; tip of lower lobe of caudal fin sometimes yellowish(Carpenter & De Angelis, 2016).

Distribution: West African coast from Mauritania to Angola(Carpenter & De Angelis, 2016).

Biology and Habitat: Inhabits coastal waters close to the bottom Occasionally enters estuaries and other brackish water areas(Carpenter & De Angelis, 2016).

Remarks: Second, the catalog of FAO 2016, *Pomadasys jubelini* was mistakenly keyed as *P. rogerii* in the 1985 version of this guide, resulting in some confusion(Carpenter & De Angelis, 2016).

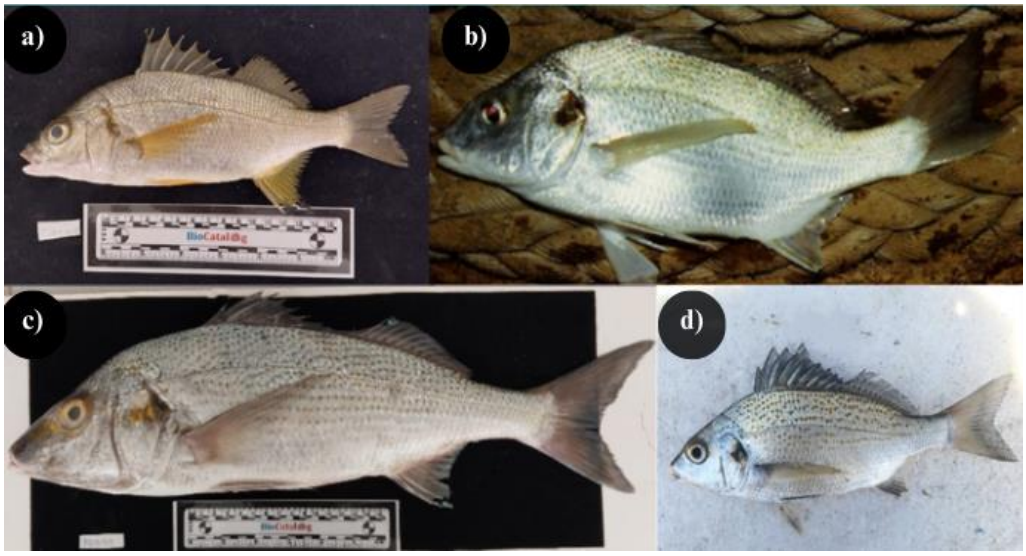


Figure 3: *Pomadasys* species targeted in this study: **a**) *P. incisus*; **b**) *P. perotaei*; **c**) *P. jubelini*; and **d**) *P. rogerii.* Sources: In this study, FishBase[2] and BOLDSystems[3].

Genus *Brachydeuterus* (Gill, 1862)

Description: The species typically have a moderately deep and laterally compressed

[2] www.fishbase.se
[3] https://www.boldsystems.org/index.php/databases

body shape, elongated and slightly oval, with a distinct head and a slightly arched dorsal profile. Eighteen to twenty-two (18-22) gill rakers on the first gill arch. The snout is shorter than the eye diameter. Thin lips. Two (2) chin pores followed by a median dimple. Twenty-six (26) vertebrae. The dorsal fin is typically continuous but can be also two (2). The spines are usually stout and may be serrated or rough. Have a terminal or slightly inferior mouth towards the front of the head. Monotypic genus(Lévêque et al., 1990).

Coloration: Common colors include various shades of brown, grey, and silver. Some species may have distinct patterns of stripes, bars, or spots on their bodies(Lévêque et al., 1990).

Habitat: Found in coastal marine environments. They may inhabit various habitats, such as rocky reefs, seagrass beds, and coral reefs(Lévêque et al., 1990).

Distribution: Often occurring in tropical and subtropical regions(Lévêque et al., 1990).

***Brachydeuterus auritus* (Valenciennes, 1832)**

(Fig. 4)

Synonym: *Otoperca aurita* (Valenciennes, 1832)

Description: Oblong, compressed body with a slightly and evenly convex dorsal profile. Snout shorter than eye diameter. Large, obliquely slit, protractile mouth; lower jaw extremely prominent. Chin with a pair of small pores near lips and another pair of pores, very close to each other, at symphysis of the lower jaw. Dorsal fin with twelve (12) moderately strong spines and eleven to thirteen (11-13) soft rays; anal fin with tree (3) spines and none or ten (9 / 10) (rarely 8) soft rays. The caudal fin is deeply emarginate. Lateral-line scales forty-eight to fifty-two (48-52); four or five (4 / 5) scale rows above and eleven or twelve (11 / 12) below the lateral line(Carpenter & De Angelis, 2016).

Coloration: Olive back, silvery to white sides. A dark spot on the upper edge of the operculum. Sometimes a few small dark spots on the base of the dorsal fin(Carpenter & De Angelis, 2016).

Habitat and Biology: This species is semi-pelagic in coastal waters ten to one hundred meters (10-100 m) deep. Brackish waters and estuaries. Occurs over sandy and muddy bottoms. Feeds on invertebrates and small fishes(Carpenter & De Angelis, 2016).

Distribution: West African coasts, from Mauritania to Angola (exceptionally Morocco)(Carpenter & De Angelis, 2016).

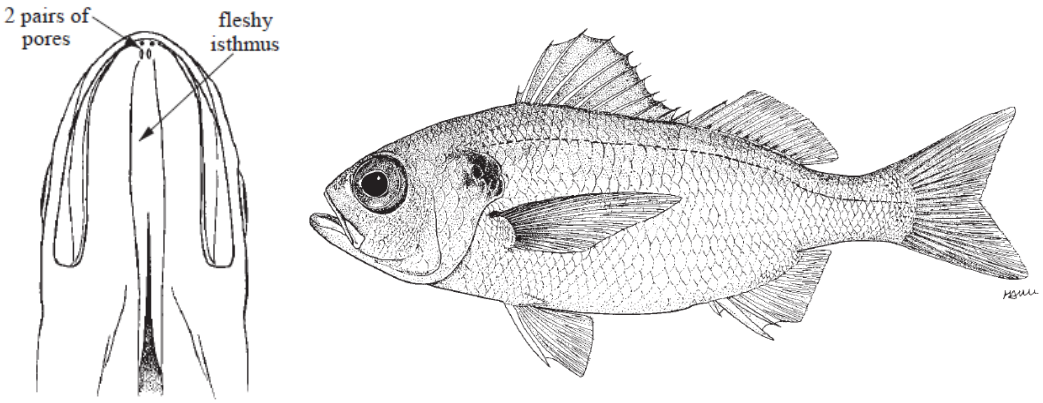


Figure 4*:* *Brachydeuterus auritus* (Valenciennes, 1832) shape, second the catalogue of FAO (2016), highlighting the main morphological features.

Subfamily Plectorhinchinae (Jordan & Thompson, 1912)

Description: They are relatively large fish, growing from thirty-five (35) centimeters to over a meter long, with large heads and large, bulging lips. Their teeth are small and conical, gullet teeth are present. The caudal fin is only slightly notched or rounded(Lévêque et al., 1990).

Coloration: Strikingly colored with white and yellow base and fin colors and yellow and black longitudinal stripes and spots. Juveniles are completely different in coloration in many species(Lévêque et al., 1990).

Biology and Habitat: They will often associate with other fishes of similar species; several species of sweetlips sometimes swim together. They are usually seen in clusters in nooks and crannies or under overhangs. At nightfall, they venture from their shelters to seek out their bottom-dwelling invertebrate prey, such as bristle worms, shrimps, and small crabs. Inhabits sandy and muddy bottoms(Lévêque et al., 1990).

Distribution: Most of the species are found in oceanic waters of tropical, subtropical water along the Indo-Western Pacific, but also some of them can be found in East Atlantic and Mediterranean(Lévêque et al., 1990).

Genus *Parapristipoma* (Bleeker, 1873)

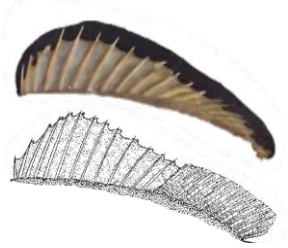
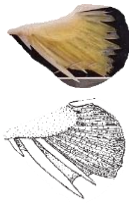
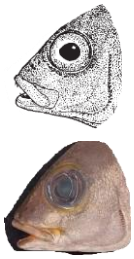
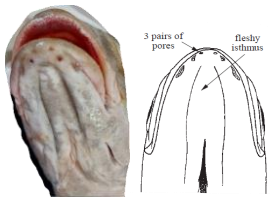
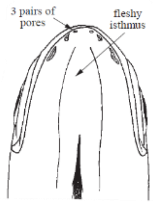
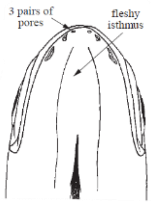
Description: The oblong body is compressed, covered with small ctenoid scales. The head is covered with scales on the upper part, front, suborbital region, opercula, and posterior part of the lower jaw. The upper jaw is scaly. The maxillae are nearly equal, with the upper one moderately protrusible, and the lower one slightly prominent. Mouth oblique. Teeth in several rows on the jaws and pharynx, small, slender, and sharp; no teeth on the vomer or palate. The anterior part of the suborbital bone is edentulous. Thin lips. The lower jaw has two small pores at the front and no median fossa. Serrated preoperculum. No true opercular spine. Pectoral fins are highly acute and falciform. Dorsal and anal fins with a scaly sheath at the base, no spine on the dorsal fin and the rayed part twice to much more than twice as long as high. Pseudobranchiae present. Simple swim bladder. Dorsal spines thirteen to fifteen (13-15), anal spines tree (3)(Dupérier & Brygoo, 1983).

Coloration: Parapristipoma species may exhibit a variety of colors and patterns. Common colors include various shades of brown, grey, silver, and sometimes yellow. Some species may have distinct patterns of stripes, bars, or spots on their bodies(Dupérier & Brygoo, 1983).

Biology and Habitat: Typically found in coastal marine environments. They may inhabit various habitats, such as rocky reefs, seagrass beds, and coral reefs, often occurring in tropical and subtropical regions(Dupérier & Brygoo, 1983).

Distribution: West African coast from Mauritania to Senegal, North of Europe (Portugal, Spain), Asia (Japan)(Carpenter & De Angelis, 2016).

Table 2: Main morphological characteristics of the *Parapristipoma* Genus.

Species **Morphological characteristics**	*Parapristipoma humile* (Bowdich, 1825)	*Parapristipoma octolineatum* (Valenciennes, 1833)	*Parapristipoma macrops* (Pellegrin, 1912)
Dorsal-fin rays			
	1 dorsal fin, with 13 spines, and 15-16 soft dorsal rays.	1 dorsal fin with 13 spines and 14 or 15 soft rays.	1 dorsal fin with 13 spines and 15 or 16 soft rays.
Anal-fin rays			
	3 anal spines and 7-8 soft anal rays.	Anal fin with 3 spines and 7 soft rays.	The anal has 3 spines and 8 soft rays; the second spine is much stronger but does not exceed the third.
Snout, Mouth and Eye			
	Short snout, approximately equal to the diameter of the eye; mouth slightly oblique, nearly terminal.	Snout rounded, shorter than eye diameter; mouth slightly oblique.	The snout is conical and its length is less than the diameter of the eye; ; mouth oblique.
Pores			
	Chin with 3 pairs of pores (anterior pair smaller than the others).	Chin with 3 pairs of pores (anterior pair smaller than the others).	Chin with 3 pairs of pores (anterior pair smaller than the others).

Parapristipoma humile (Bowdich, 1825)

(Fig. 5c))

Synonym: *Pristipoma humilis* (Bowdich, 1825) and *Parapristoma macrops* (Pelegrin, 1912).

Description: The body is elongate and compressed. Snout nearly as long as orbit diameter. Normally the mouth is slightly oblique, nearly terminal and the lips moderately thin. Has tree (3) pairs of pores (the anterior pair smaller than the others) on the chin. Four (4) pores in the nasal area. Twenty-one to twenty-tree (21-23) gill rakers on the lower limb of the first arch. The teeth are conical, in several bands. Preopercle serrated. One (1) dorsal fin, with thirteen (13) spines and fifteen to sixteen (15-16) soft dorsal rays. The anal fin has tree (3) spines and seven to eight (7 -8) soft anal rays. Fifty-two to fifty-nine (52-59) in the lateral line(Carpenter & De Angelis, 2016).

Coloration: Body mostly brownish to greyish. The caudal peduncle and caudal fin are yellow and other fins yellowish to orangish, the pelvic-fin spine and anterior anal-fin spine are whitish(Carpenter & De Angelis, 2016).

Distribution: From the Straits of Gibraltar to Angola, northward extending into the Mediterranean. Very common in the Cabo Verde Islands(Carpenter & De Angelis, 2016).

Biology and Habitat: Found from littoral to one hundred (100) m, over sand and rocks(Carpenter & De Angelis, 2016).

Remarks: Second Wirtz et al., (2013) and FishBase, because of the frequent confusion between this species and *Parapristipoma octolineatum*, its distribution is unclear. It could be endemic to the Cabo Verde Islands.

Parapristipoma octolineatum (Valenciennes, 1833)

(Fig. 5b))

Synonym: *Pristipoma octolineatum* (Valenciennes, 1833) and *Diagramma octolineatum* (Valenciennes, 1833).

Description: The body is elongated and compressed. The snout is rounded, shorter than the eye diameter and the mouth is slightly oblique with the lips moderately thin. Has the chin with tree (3) pairs of pores (the anterior pair smaller than the others) and the preopercle is serrated. Twenty-one to twenty-tree (21-23) gill rakers on the lower limb of the first arch. The first dorsal fin with thirteen (13) spines and the second one with fourteen or fifteen (14 / 15) soft rays. The anal fin with tree (3) spines and seven (7) soft rays. Fifty-tree to fifty-eight (53-58) in lateral line(Carpenter & De Angelis, 2016).

Distribution: From Spain and Portugal to Angola. Also found in the western Mediterranean (uncommon)(Carpenter & De Angelis, 2016).

Biology and Habitat: Feeds on crustaceans and molluscs and occurs in shallow waters from the shoreline to about 50 m on sand or rocky bottom(Carpenter & De Angelis, 2016).

Remarks: Because *P. humile* also can have stripes, it is frequently confused with *P. octolineatum*. However, the photo in Brito et al., (2007) and other photos we have clearly show *P. octolineatum.*

***Parapristipoma macrops* (Pellegrin, 1912)**

(Fig. 5a))

Synonym: *Diagramma macrops* (Pellegrin, 1912) and *Diagrammella macrops* (Pellegrin, 1912).

Description: The height of the body is about the same as the length of the head. The snout is conical and its length is less than the diameter of the eye. The upper profile of the head is almost straight and only slightly convex. The lips are quite thick. The chin has six pores, the two median punctiform, the lateral elongated, ovoid. The teeth are small, conical, in bands, where those of the outer row are more developed, but there are no canines. The scales extend across the top of the head up to the level of the nostrils. There are seven gill rays and twenty to twenty-tree (20-23) elongated gills at the base of the first branchial arch. The lateral line pierces fifty-five to fifty-nine (55-59) scales. The dorsal fin begins just anterior to the beginning of the pectoral and comprises thirteen (13) spines and fifteen or sixteen (15 / 16) soft rays. There is

no notch between the rayed part of the dorsal and the soft part. The anal has tree (3) spines and eight (8) soft rays and the second spine is much stronger but does not exceed the third. The caudal section is emarginate(Dupérier & Brygoo, 1983).

Coloration: The coloring is violet on the head and body, orange-yellow on the belly. The fins are purplish(Dupérier & Brygoo, 1983).

Distribution: Eastern Atlantic (Angola, probably ranging northwards)(Dupérier & Brygoo, 1983).

Biology and Habitat: Marine fish, demersal and of tropical climate, that lives between 10 and 100 meters deep on the continental shelf(Dupérier & Brygoo, 1983).

Remarks: The only picture that was found from this species belongs to the original description of Pellegrin, 1912.

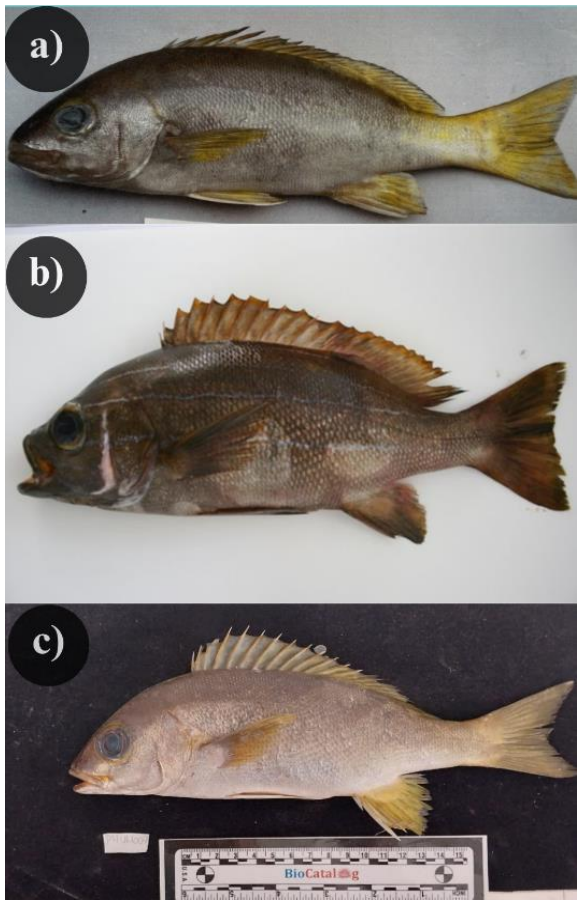


Figure 5: *Parapristipoma* species targeted in this study: a*) P. macrops*; b) *P. octolineatum* and c) *P. humile*. Sources: In this study, FishBase and (González et al., 2009).

Genus *Plectorhinchus* (Lacepède, 1801)

Description: Body compressed, oblong. Head strongly obtuse, upper profile well convex. Eye moderate. Mouth small, horizontal, upper jaw protractile. Maxillary slips below preorbital. Lips fleshy. Jaws with bands of villiform teeth, pointed, in about four to six (4-6) rows. Chin with pores, without central grooves or barbels. Preopercle serrate. Suborbitals without spines or serrate. No opercular spine. Branchintegrals six or seven (6-7). Pseudobranchiae large. Air bladder simple. Twenty-six or twenty-seven (26 / 27) vertebrae, of which fifteen or sixteen (15 / 16) caudal. Scales small or moderate, fifty to one hundred twenty-five (50-125) in lateral series, ctenoid. Soft vertical fins are scaly basally. Ventral base with scaly flap. Lateral line continuous. One dorsal, depressible in the groove, with nine to fourteen (9-14) stout spines, rays fifteen to twenty-tree (15-23). Anal spines tree (3), rays six to nine (6 -9). Pectoral pointed. The Ventral is inserted a little behind the pectoral base(Lévêque et al., 1990).

Coloration: Common colors include various shades of brown, grey, silver, and sometimes yellow. Some species may have distinct patterns of stripes, bars, or spots on their bodies(Lévêque et al., 1990).

Distribution: From the Straits of Gibraltar to Namibia; northward extending into the Mediterranean and along the coasts of Spain and Portugal. Also, the East coast of the African continent, India, and Australia(Carpenter & De Angelis, 2016).

Biology and Habitat: *Plectorhinchus* species are typically found in coastal marine environments. They may inhabit various habitats, such as rocky reefs, coral reefs, and sandy bottoms, often occurring in tropical and subtropical regions.

Plectorhinchus mediterraneus (Guichenot, 1850)

(Fig. 6)

Synonym: *Diagramma mediterraneum* (Guichenot, 1850) and *Parapristipoma mediterraneum* (Guichenot, 1850).

Description: Body oblong and compressed. Snout 1.3 to 1.8 times the eye diameter; eye medium-sized (3.5 to 5 times in head length). The mouth is oblique, the maxilla reaches to anterior eye margin and the lips are relatively thick. Teeth conical, set in several bands in jaws. In the chin has tree (3) pairs of pores (anterior pair smaller than the others) and the preopercle is serrated. Nineteen or twenty (19 / 20) gill rakers on the first arch. Dorsal fin with ten to thirteen (10-13) spines and seventeen to twenty (17-20) soft rays: The anal fin with tree (3) spines and eight or nine (8 / 9) soft rays. The caudal fin emarginate with pointed lobes(Carpenter & De Angelis, 2016).

Coloration: Body greyish to brownish, lighter stripes, another indistinct dark stripe just below soft dorsal fin and curving onto upper caudal peduncle; fins greyish to brownish, the tips generally darker, especially the pectoral fins(Carpenter & De Angelis, 2016).

Distribution: West African coast, from the Straits of Gibraltar to Namibia; northward extending into the Mediterranean and along the coasts of Spain and Portugal(Carpenter & De Angelis, 2016).

Biology and Habitat: Inhabits sand and muddy sand bottoms from the coastline to about 180 m depth. Feeds on benthic and planktonic crustaceans and mollusks. Coastal waters throughout its range(Carpenter & De Angelis, 2016).

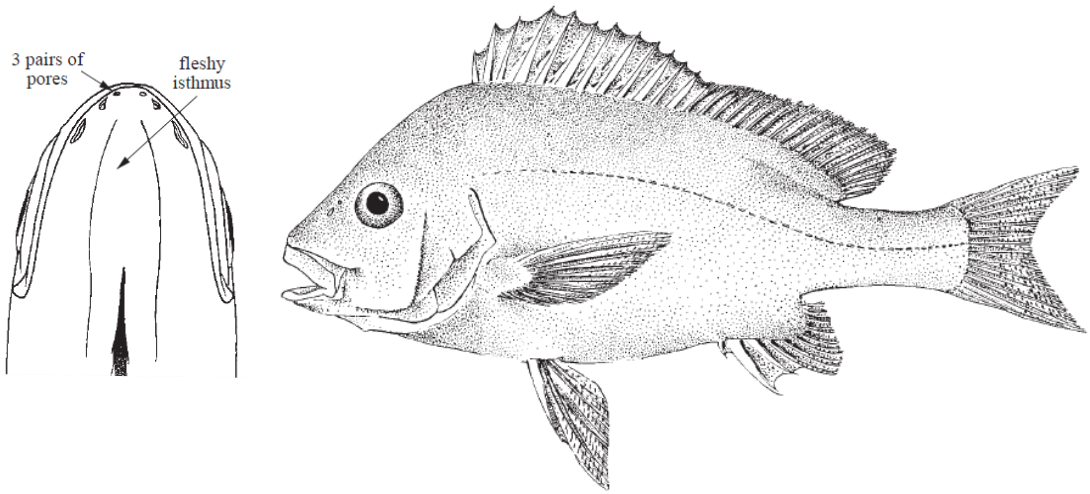


Figure 6: *Plectorhinchus mediterraneus* (Guichenot, 1850) shape, second the catalogue of FAO (2016), highlighting the main morphological features.

3. Materials and Methods

3.1 Study Area

Cabo Verde consists of ten (10) islands (only nine (9) are inhabited) and eight (8) islets located on the Northwest African coast, approximately 600 km west of Senegal, with a total area of 4,033 km^2 (Figure 2). The prevailing winds hold a significant influence on the islands, leading to their geographical division into two distinct groups: Barlavento (consisting of Santo Antão, São Vicente, Santa Luzia, Islets Raso and Branco, São Nicolau, Sal, and Boavista), situated upwind of the northeasterly trade winds; and Sotavento (encompassing Maio, Santiago, Fogo, Brava, Islets Grande, and De Cima), located downwind from these prevailing winds. Very close to the water's surface, we can find Seamounts (Noroeste, Nova Holanda, Bancona, and João Valente) (Rolan, 2005). Of volcanic origin, the archipelago was formed from rocks resulting from oceanic eruptions. The coastline is about 965 kilometres long, while the slope of the island platform maintains a steep incline up to a depth of 200 meters, gradually decreasing beyond that point to 1000 meters. This covers a region of 5,934 square kilometres. Biogeographically, Cabo Verde belongs to the Macaronesia region, including Madeira, the Azores, and the Canary Islands in the eastern Atlantic Ocean (Pelegrí & Peña-Izquierdo, 2005).

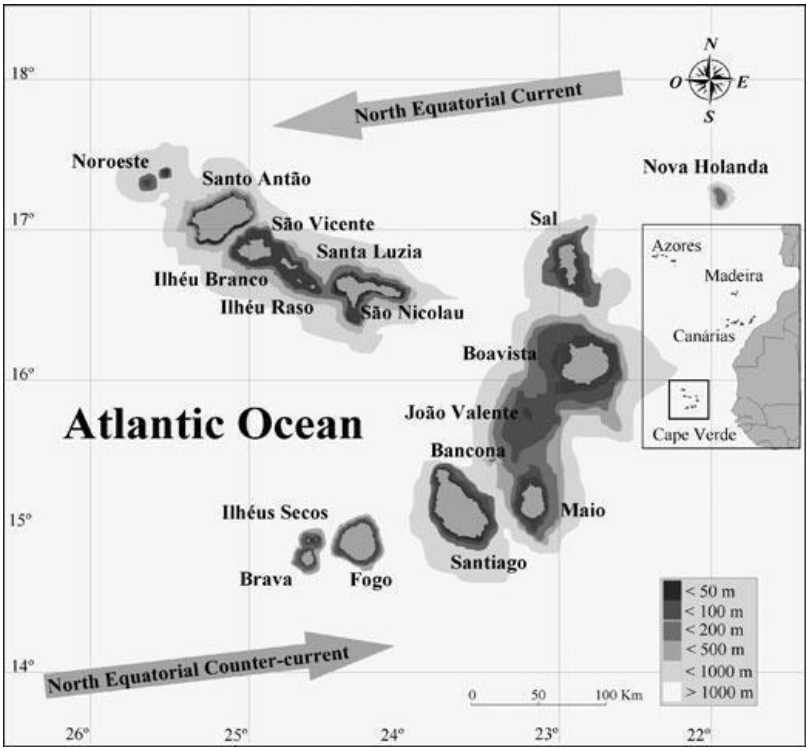


Figure 7: Geographic location of the Cabo Verde archipelago in the biogeographic region of Macaronesia (Source: Medina, 2007).

3.2. Laboratory Analysis

3.2.1. Morphological Analysis

Morphological analysis of fish is a process that involves the observation and description of the physical and structural characteristics of fish for identification and taxonomic classification. The fish analysis is based on directly observing fish's external and internal features, allowing the distinction between different species and understanding their evolutionary relationships. In the case of this study, there was only a description of the external characteristics of the fish samples to help the identification, which was confirmed later by phylogenetic systematics. So, after purchasing the samples at the fish market, they were adequately identified using systematic taxonomy. First, they were photographed, and then they were identified with the help of taxonomic identification keys.

Most of the specimens used in this study were collected at the São Vicente fish market and the locations where the specimens originated were noted. The following characteristics of the fish were observed: coloration, body shape, number and arrangement of fins, number of rays and spines on the fins, and number of pores on the chin. All the description made to identify the samples was based on various taxonomic keys (e.g., Fernández-Gil et al., 2013; Saldanha (2003), also various FAO species identification guide).

The sampling was done in order to obtain at least five individuals of each species (Table 1), the target of the study, which is listed in the table below, according to research done (e.g., Freitas, 2014; J. A. González et al., 2010; Wirtz et al., 2013).

Table 3: Validation of the species belonging to the Haemulidae family targeted in the study.

Genera	Scientific name	Status	Authority
Pomadasys	*Pomadasys incisus*	Valid	Bowdich, 1825
	Pomadasys perotaei	Valid	Cuvier in Cuvier & Valenciennes, 1830
	Pomadasys jubelini	Valid	Cuvier in Cuvier & Valenciennes, 1830
	Pomadasys suillus	Doubtful	Cuvier in Cuvier & Valenciennes, 1830
	Pomadasys rogerii	Not Valid	Cuvier in Cuvier & Valenciennes, 1830
Parapristipoma	*Parapristipoma humile*	Endemic	Bowdich, 1825
	Parapristipoma octolineatum	Valid	Valenciennes in Cuvier & Valenciennes, 1833
	Parapristipoma macrops	Need Confirmation	Pellegrin, 1912
Brachydeuterus	*Brachydeuterus auritus*	Need Confirmation	Valenciennes in Cuvier & Valenciennes, 1833
Plectorhinchus	*Plectorhinchus mediterraneus*	Not Valid	Guichenot, 1850

3.2.2. Molecular Analyses

a) Sampling

The specimens used in this study were obtained and later identified in the laboratory and acquired through weeks of visits to the São Vicente fish market (FMSV)**.** Once correctly identified, all information was noted for this study's database, and finally, the specimens were photographed. Through the network formed during this study, it was possible to obtain samples from other sources, mainly because the same species cannot be found at the FMSV. Many tissue samples that were integrated into the study come from projects previously developed in the archipelagos of Cabo Verde and the Canary Islands, information that has not yet been published (Table 2).

Table 4: Contains information on samples according to the sampling site and project to which they belong.

Species	Region/Country	Site 1	Site 2	Type material	Scientific Project
Pomadasys incisus	Cabo Verde	FMSV	Porto Novo	**specimen**	**Present study**
	Cabo Verde	FMSV	São Pedro	tissue	BIOTECMAR
Pomadasys jubelini	Cabo Verde	FMSV	São Pedro	**specimen**	**Present study**
Pomadasys rogerii	Cabo Verde	FMSV	Santa Luzia	tissue	BIOTECMAR
	Cabo Verde	--	--	tissue	PROACTIVA / MARPROF-
Parapristipoma humile	Cabo Verde	FMSV	São Pedro	**specimen**	**Present study**
	Cabo Verde	FMSV	Porto Novo	**specimen**	**Present study**
	Cabo Verde	FMSV	São Pedro	tissue	BIOTECMAR
Parapristipoma octolineatum	Canarias	--	--	tissue	PESCANARIAS
Parapristipoma macrops	Cabo Verde	--	--	tissue	PROACTIVA

b) DNA extraction

For each sample, a tissue sample was collected and stored in a 1.5 ml tube with 96% alcohol in a refrigerator at -20 ºC. The protocol E.Z.N.A. Tissue DNA Kit (Omega Bio-Tek) was used to perform the extraction following the kit protocol: Start by transferring approximately 20 mg of the sample into a sterile container, the microcentrifuge tube, adequately labelled. These were left to dry overnight. Next, lysis buffer was added which helps break down the cell membranes and release the DNA, consisting of 200 µl of TL Buffer and 20 µl of Proteinase K and was mixed gently to ensure that the solution was homogenized. After this, the samples were incubated in the oven at 60°C for about 3 hours and were shaken every half hour during this period. At the end of the incubation period, the samples were centrifuged at maximum speed (12. 300 rpm) for 5 minutes, and then the liquid portion (supernatant) of the sample was carefully removed, avoiding any precipitates or debris and transferred to a sterile 1.5 ml microcentrifuge tube (this was done carefully to disturb or transfer any of the

insoluble pellets. Next, the DNA was purified by adding BL buffer, shaking the samples to mix them, and then placing them in an oven at 70°C for 10 minutes. 220 µl of 100 % ethanol was added and then inserted a HiBind DNA Mini Column into a collection tube (2ml). After transferring the entire sample into these new tubes, including the precipitates, they were centrifuged at maximum speed for 1 minute. The filtrate was discarded and the collection tubes were reused. 500 µl of HBC Buffer diluted with 100 % isopropanol was added and then centrifuged at maximum speed for 30 seconds and the filtrate was discarded. After this, the HiBind Mini Column was inserted into a new collection tube. After this, 700 µl of DNA Wash Buffer diluted with 100% ethanol was added, and centrifuged at maximum speed for 30 seconds and the filtrate was discarded and the collection tube reused. This step was repeated and after that, the empty HiBind DNA Mini Column was centrifuged at maximum speed for 2 minutes to dry the column (to remove any alcohol residue). Next, the HiBind Mini Column in a 1.5 ml microcentrifuge tube. Into this was added 50 µl of Elution Buffer previously heated to 70°C and left at room temperature for 2 minutes and then centrifuged at maximum speed for 1 minute. This step was repeated once more and after finishing the procedure, the DNA was stored in the refrigerator at a temperature of 6ºC.

d) DNA amplification by PCR technique and Sequencing

The polymerase chain reaction (PCR) is an in vitro technique that allows the amplification of specific DNA/RNA sequences. This step consists of, Denaturation: This involves raising the temperature of the reaction mixture to 95°C, causing the double-stranded DNA to split into two single strands; Annealing: In order for the specific DNA primers to bind to the target DNA sequence, after denaturation, the reaction temperature is reduced to a range of 50°C to 65°C; and Extension: After the primers bind to the target DNA, the temperature is raised to the optimum operating range of the DNA polymerase enzyme, usually around 72°C. DNA polymerase synthesizes new DNA strands by adding complementary nucleotides to the primers.

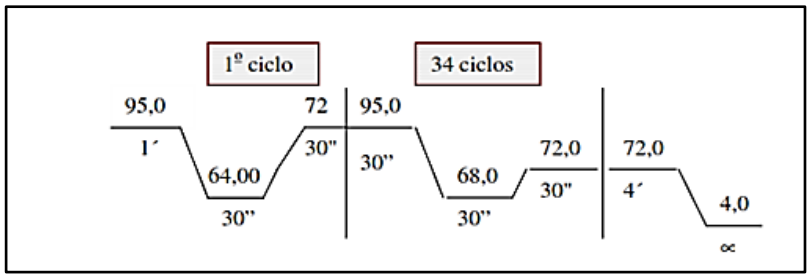


Figure 8: Schematic demonstration of the programming of the PCR cycles with temperature (ºC) and time.

For PCR, a universal mitochondrial marker, a 650 bp segment of Cytochrome Oxidase one (CoxI), was selected. The amplification utilized a primer pair specified as CoxI FishF2 Forward (5'-TCG ACT AAT CAT AAA GAT ATC GGC AC-3') and FishR2 Reverse (5'-ACT TCA GGG TGA CCG AAG AAT CAG AA-3') for this study. PCR reactions were performed using Phusion High-Fidelity PCR Master Mix, as it offers high-speed, high-performance PCR, with 2X mix containing Phusion DNA Polymerase. Together with the samples, a negative control was included in the amplification to test for contamination. In this research, it was used a 2720 thermal cycler (Applied Biosystems). The selected marker, CoxI, was used, followed by 35 cycles consisting of denaturation at 94°C for 30 seconds, primer binding at 52°C for 40 seconds, annealing at 42°C for 30 seconds and extension using Taq DNA polymerase at 72°C for 1 minute.For each step of the laboratory procedure, whether DNA extraction or DNA amplification (PCR), electrophoresis was performed to quantify and visualize the results that were obtained. Gel electrophoresis is a widely used laboratory technique that separates molecules, such as DNA, RNA, and proteins, based on their size and charge.

It was prepared a 1.5% agarose gel to assess the quality of the PCR-amplified products. These products were then stained with SYBR Safe, and a buffer solution of TBE (or EDTA) at a 1X concentration was employed.After placing the DNA samples into the wells, a *100bp Ladder* (molecular weight standard) was added. After all this was done the electric field was turned on at 50 V for 20 minutes. The gels were photographed and edited. The size of the DNA fragment was estimated by comparison with the standard *Ladder 100 bp*.

The samples were then sent to continue the laboratory procedure with the purification phase of the PCR products for sequencing, which was done in Caparica, Portugal, by the company STAB vida.

3.3. Statistic Analyses

Different datasets were used to address the different objectives of this research to obtain the best possible representativeness and to clarify any taxonomic doubts (the sequences were compared with the sequences available in BoldSystems database[4] and GenBank[5] to taxonomically identify the specimens. During this phase, we executed validation using BLAST (Basic Local Alignment Search Tool) to determine the similarity percentages between our sequences and those stored in the database. DNA sequences from published species, private databases of collections and museums, as well as regional species cataloguing projects, were integrated into the study.

The CoxI sequences were manually edited and corrected using BioEdit, Hall (1991), followed by GENEIOUS 5.4 Drummond et al. (2010), thorough observation of the chromatograms was made and several alignments were performed. Haplotypes networks were assembled based on the 95% maximum parsimony criterion in the TCS 1.2 program Clement et al. (2000), which was later refined in the TCSbU program (Múrias Dos Santos et al., 2016). Using DNASP 5.00.07 Rozas et al. (2003) the haplotypic diversity index (h), the nucleotide diversity index (π), and the mean differences between nucleotides (k) were calculated.

To test the phylogenetic position of the Haemulidae species for Cabo Verde concerning other Atlantic regions, the sequences from the archipelago were further aligned with sequences made available from another place (Table 3). From this resulting alignment, a Neighbor-joining tree was constructed with Kimura-two-parameter (K2+G+I) using a discrete Gamma distribution (+G) with 4 rate categories and by assuming that a certain fraction of sites are evolutionarily invariable (+I) distance model in MEGA 7.0.18 for the COI gene (Kumar et al., 2018). Maximum Likelihood value (ML) method was employed to evaluate phylogenetic relationships among Haemulidae species of COI data based on the K2+G+I genetic distance using MEGA 7.0.18 with bootstrap tests of 1000 replicates to verify the robustness of the tree and to estimate the interspecific genetic distances (Kumar et al., 2018) with all sequences obtained for Cabo Verde, Atlantic, and Mediterranean.

[4] https://www.boldsystems.org/index.php/databases
[5] https://www.ncbi.nlm.nih.gov/genbank/

Table 5: List of samples and DNA sequences included in the genetic analyses. Their code and taxonomic identification are given, as well as their distribution range, climatic zone, location, and GenBank accession number.

Species	Location 1		Location 2	Collection Date	N	Specimen ID	Database Code	OBS
Haemilidae Family								
Pomadasys incisus (Bowdich, 1825)	Cabo Verde	Tarrafal Monte Trigo	Santo Antão	2023	9	PINC (001 – 009)	_____	In this study
	Cabo Verde	São Pedro	São Vicente	2015	5	PINC VV1	_____	BIOTECMAR
	Israel	Haifa	Mediterranean	2012	1	-	KF564312	GenBank
	Turkey	Adana	Mediterranean	2013	1	S239	KY176578	GenBank
	Spain	Cadiz	Mediterranean	2009	1	MLFPI7	KJ768284	GenBank
	Angola	Luanda	West Africa Coast	2002	1	A314	A314-COI	AquaGene
Pomadasys jubelini (Cuvier, 1830)	Cabo Verde	São Pedro	São Vicente	2023	8	PJUB (001 – 008)	_____	In this study
	Angola	Catanha	São Vicente	2003	1	B284	B284-COI	AquaGene
Parapristipoma humile (Bowdich, 1825)	Cabo Verde	São Pedro	São Vicente	2023	9	PHUM (001-009)	_____	In this study
	Cabo Verde	Baia de Fateja	São Vicente	2015	7	PHUM VV	_____	BIOTECMAR
	Cabo Verde	_____	Santa Luzia	2015	4	PROG (6 – 17) VL	_____	BIOTECMAR
Parapristipoma macrops (Pellegrin, 1912)	Cabo Verde	_____	_____	2007-2013	2	ParMac (09; 07) CV	_____	PROACTIVA
Pomadasys rogerii (Cuvier, 1830)	Cabo Verde	_____	_____	2007-2013	2	PomRog (01; 02) CV	_____	PROACTIVA / MARPROF-CV
	Cabo Verde	_____	Boavista	2005	1	CVRD171-13	_____	Smithsonian museum
	Angola	Luanda	_____	2003	1	B227	B227-COI	AquaGene
Plectorhinchus mediterraneus (Guichenot, 1850)	Noroeste África (Marrocos- Senegal)	_____	_____	2002-2006	2	PleMed (03; 04) EE-c	_____	FISHTRACE
	Angola	Luanda	_____	2003	1	B209	B209-COI	AquaGene
Parapristipoma octolineatum (Valenciennes, 1833)	Spain	Vigo	_____	2018	1	GAL011	MH980024	GenBank
	_____	_____	_____	2010	1	ODU 3220	HQ676781	
	Angola	Luanda	_____	2002	1	A209	A209-COI	AquaGene
	Canarias	Canarias	_____	-	2	Poct (3;4) Reg72; Reg73	_____	PESCANARIAS
Pomadasys perotaei (Cuvier, 1830)	Senegal	_____	_____	2002-2006	2	PomPer (02; 03) EE-c	_____	FISHTRACE
	_____	_____	_____	2010		ODU 3260	HQ676800	GenBank
	Angola	Luanda	_____	2003		B066	B066-COI	AquaGene
Brachydeuterus auritus **(**Valenciennes, 1832)	_____	_____	_____	2010	1	ODU 3238	HQ676755.1	GenBank
	Nigeria	Bayelsa	_____	2015	1	NMFS0021	KY442719.1	
	Nigeria	Olso	_____	2015	1	NMFS0015	KY442713.1	

	Angola	Luanda	_____	2002	1	A355	A355-COI	AquaGene
Species **Outgroup**	**Location 1**		**Location 2**	**Collection Date**	**N**	**Specimen ID**	**Database Code**	**OBS**
Lutjanus agennes (Bleeker, 1863)	Nigeria	Ogun State	_____	2017	1	NMFS0028	KY442726.1	GenBank
Lutjanus fulgens (Valenciennes, 1830)	Ghana	Denu	_____	2020	1	LFV4	MN986448.1	GenBank
	Ghana	Denu	_____	2020	1	LFV5	MN986449.1	GenBank
	Ghana	Denu	_____	2020	1	LFV6	MN986450.1	GenBank
Ocyurus chrysurus (Bloch, 1791)	Mexico	Quintana Roo	_____	2010	1	MFLS4464	JN311992.1	GenBank
	Anguilla	Anegada Passage	_____	2007	1	MFLV2326	HM389791.1	GenBank
	British Virgin Islands	South of Virgin G	_____	2007	1	MFLV2322	HM389789.1	GenBank

4. Results

4.1. Molecular Barcoding

In this study, a total of 35 tissue samples collected from the fish market of São Vicente, and from the research projects illustrated above, were processed. Due to the difficulties encountered, only 3 species of the Haemulidae family were sampled in the FMSV. In contrast, the other samples that joined these for DNA extraction were collected by other projects (Table 4). For the molecular analyses, 48 sequences from various sources were used and added to the extracted DNA samples. The number of sampled individuals that were sent for sequencing totalled 35 specimens, ranging from a minimum of 2 and a maximum of 9 per species.

The sequencing result was obtained with 640 bp of the gene CoxI for all sampled individuals. From the alignment, it was possible the molecular identification of four species of the genus Pomadasys and the tree of the genus *Parapristipoma*. The results show 153 substitutions and 147 parsimoniously informative sites in the alignment. The rate variation model allowed for some sites to be evolutionarily invariable 60.9549% sites (+I). The nucleotide frequencies are 23.37% (A), 28.13% (T), 27.55% (C) and 20.95% (G). The average nucleotide frequencies obtained show high percentages of G+C, around 0.48.5%. The transition/transversion rate ratios are k1 = 10.986 (purines) and k2 = 7.74 (pyrimidines). A total of 39 haplotypes were obtained, with haplotypic diversity (Hd- 0.942, SD 0.014) and (pi= 0.130, SD 0.00599) and the average number of nucleotide differences (k: 57.167).

4.2. Phylogenetic Study

The intraspecific distances ranged from 0.005 for *P. humile* and *P. macrops* to 0.301 for *P. macrops* and *P. rogerii* (Table 6).

Table 6*:* Genetic distances calculated for the CoxI marker, using the model (K2P - Kimura two parameters), among Haemulidae species and the outgroup species (Lutjanidae species).

	Species	*PR*	*PH*	*PP*	*PO*	*PME*	*PMA*	*PJ*	*PI*	*BA*	*LA*	*LF*
PR	*P. rogerii*											
PH	*P. humile*	0,297										
PP	*P. perotaei*	0,211	0,256									
PO	*P. octolineatum*	0,299	0,049	0,276								
PME	*P. mediterraneus*	0,270	0,135	0,246	0,113							
PMA	*P. macrops*	0,301	0,005	0,260	0,048	0,129						
PJ	*P. jubelini*	0,033	0,295	0,190	0,299	0,270	0,299					
PI	*P. incisus*	0,190	0,208	0,208	0,222	0,238	0,212	0,188				
BA	*B. auritus*	0,273	0,255	0,252	0,225	0,248	0,256	0,268	0,216			
LA	*L. agennes*	0,312	0,232	0,308	0,230	0,248	0,230	0,308	0,237	0,283		
LF	*L. fulgens*	0,313	0,222	0,244	0,215	0,218	0,220	0,302	0,235	0,260	0,131	
OC	*O. chrysurus*	0,307	0,222	0,255	0,198	0,209	0,220	0,297	0,225	0,252	0,125	0,049

The COI marker evidenced the monophyly of the four genera tested in this study: *Pomadasys*, *Parapristipoma*, *Brachydeuterus and Plectorhinchus* (Fig. 9).

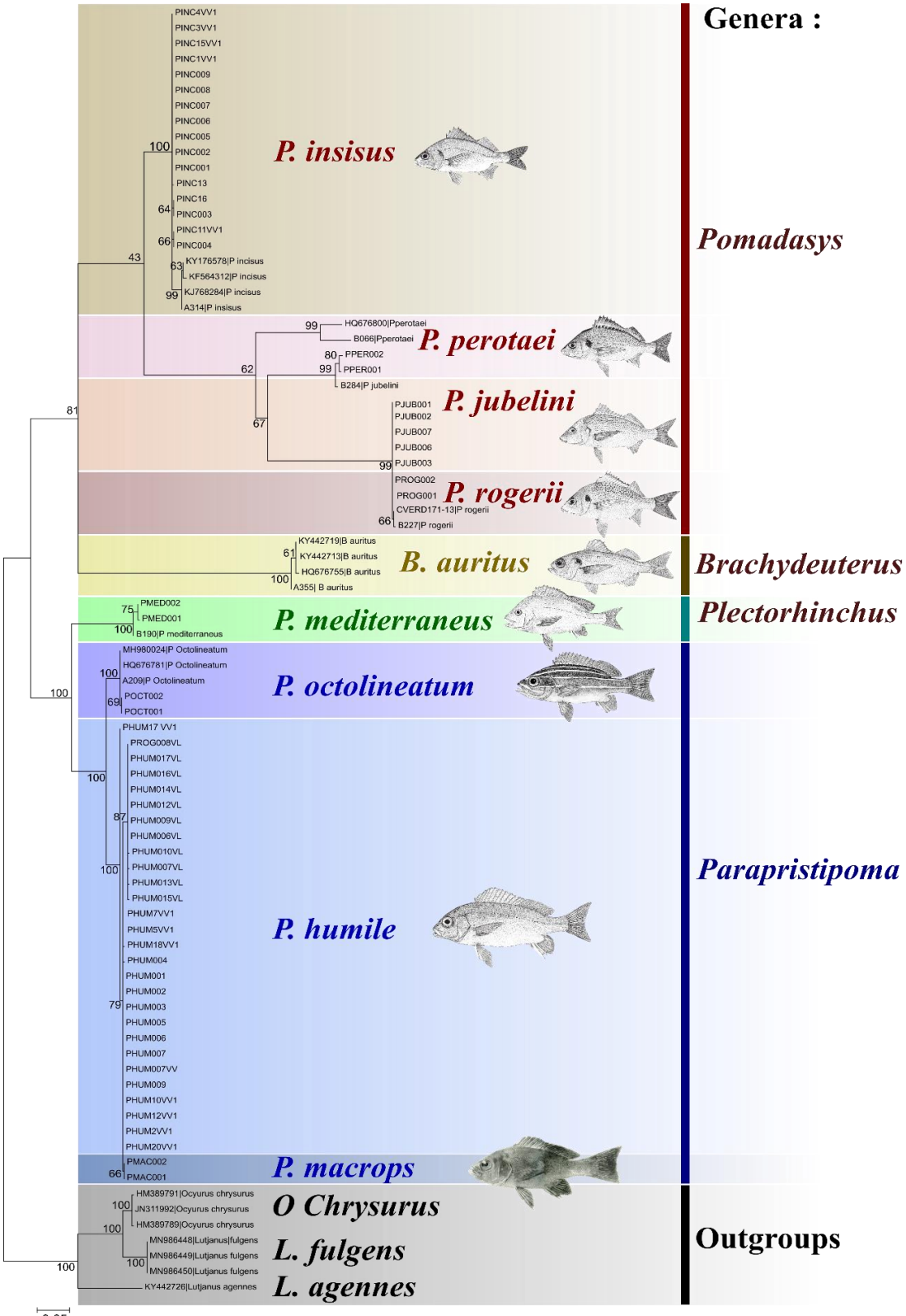


Figure 9: Phylogenetic tree for the COI marker inferred by ML of the Haemulidae family. *This tree was* constructed together with sequences obtained from GENBANK, BIOTECMAR, and AquaGene, *and included* 3 species of the Lutjanidae family as outgroups. Bootstrap proportions, correspond to the values at each node of the tree. The size of the branches represents the substitution rate.

With a 100 % confidence value, *P. incisus* formed a clade alone, therefore it is possible to say that the monophyly of this species was confirmed with maximum support in this analysis. On a confidence level below 70 %, *P. perotaei* and *P. jubelini* are almost in the same clade. It is noted that *P. jubelini* is a monophyletic species however, it shares its clade with *P. perotaei* (*P. perotaei* and *P. jubelini*, B284, almost separated by a mutation) and *P. rogerii* (CVERD171), showing a separation of few mutational steps.

In the case of *B. auritus*, obtaining samples from Cabo Verde was impossible. Hence, the sequences used in the alignment belong to online databases that are from different geographical regions. It is noted that this is a monophyletic species and that by a mutational step, one of the specimens (A355) is separated from the others by a 66% confidence.

P. humile separates from the other species, forming a 100% confidence node. However, it has a *P. rogerii* (PROG008VL) species and other *P. humile* species in an 88% confidence node. This appears to be a taxonomic error. Within the same branch of *P. humile*, separated with 66% confidence, we have two specimens of *P. macrops*, i.e., this shows a short genetic distance between these two species.

4.3. Population Structure

Due to the difficulty in finding or even existing sequences in online databases, only a few species were chosen to make haplotype networks. In the case of *P. rogerii*, we observed Little genetic variation, as there is no separation by mutagenic steps, between the samples from Cabo Verde and the other regions (Fig. 9 - haplotype network A). Even so, there are no shared haplotypes, so we can say that there is the presence of unique haplotypes or exclusive for each region.

Comparing the two species *P. macrops* and *P. humile*, they showed a low genetic distance with a haplotype network with few mutational steps (Fig. 9 - haplotype network B). In this, it is observed that despite little genetic variation, there is no presence of shared haplotypes.

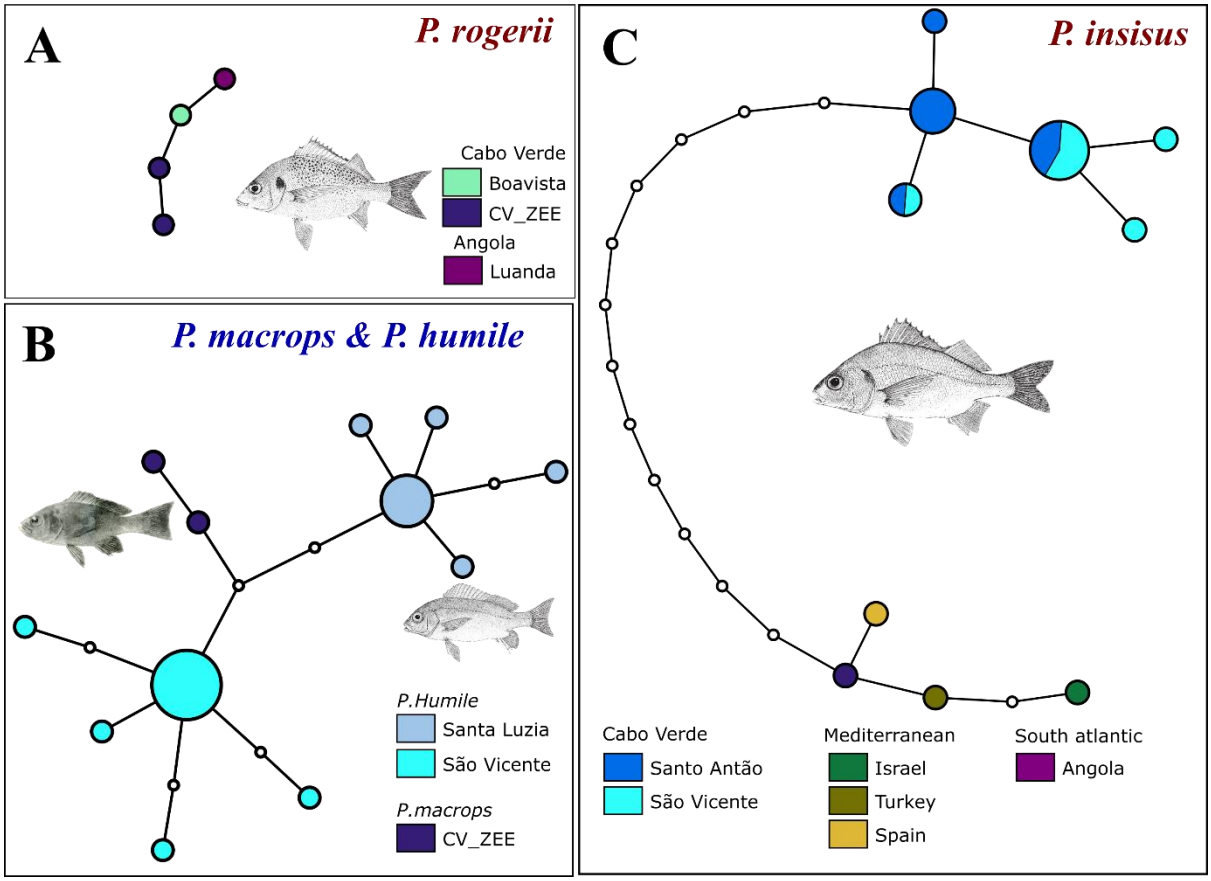


Figure 10: Haplotype network obtained from CoxI sequences for populations of some Haemulidae species from Cape Verde + Databases, where: *P. rogerii* - haplotype network A, P. macrops and *P. humile* - haplotype network B and *P. incisus* - haplotype network C. Each color represents a sampling location, and each dot represents a mutation. Frequency of the haplotypes corresponds to the circle's diameter.

The *P. incisus* samples showed a high genetic variation concerning Cabo Verde specimens, compared to Mediterranean and South Atlantic samples (Fig. 10 - haplotype network C). This shows the formation of two highly differentiated groups (differing in 12 mutational steps), one group formed exclusively from Cabo Verde, while the other comprises the remaining haplotypes from the other regions.

5. Discussion

Our methodology involved a two-pronged approach. First, we undertook a comprehensive taxonomic analysis, meticulously examining morphological features. Secondly, we conducted a genetic analysis using our samples and DNA barcodes from online databases. Integrating these insights, we reconstructed Table 1. Thus, we present the results of the taxonomic and genetic investigations (Table 7).

Table 7: Update of the status of the species addressed in this study based on morphological and genetic analysis.

Genera	Scientific name	Status	Authority
Pomadasys	*Pomadasys incisus*	Valid	Bowdich, 1825
	Pomadasys perotaei	Need confirmation	Cuvier in Cuvier & Valenciennes, 1830
	Pomadasys jubelini	Valid	Cuvier in Cuvier & Valenciennes, 1830
	Pomadasys suillus	Not Valid	Cuvier in Cuvier & Valenciennes, 1830
	Pomadasys rogerii	Doubtful	Cuvier in Cuvier & Valenciennes, 1830
Parapristipoma	*Parapristipoma humile*	Endemic	Bowdich, 1825
	Parapristipoma octolineatum	Doubtful	Valenciennes in Cuvier & Valenciennes, 1833
	Parapristipoma macrops	Valid	Pellegrin, 1912
Brachydeuterus	*Brachydeuterus auritus*	Need Confirmation	Valenciennes in Cuvier & Valenciennes, 1833
Plectorhinchus	*Plectorhinchus mediterraneus*	Not Valid	Guichenot, 1850

The species that remained valid were those that could be sampled during this study and that were caught in Cabo Verde. In this case, we have *P. incisus, P. jubelini, P. macrops e P. humile.* The status of valid previously in the *P. perotaei* goes to the status of, needs confirmation because it was not possible to collect samples of these in Cabo Verde, as well as the difficulty in finding sequences in the online databases acquired from samples collected in the geographical region of Cabo Verde. The only *P. perotaei* samples that we had, one of them is not known were collected and another was collected in Angola. However, the existence of the species in Cape Verde remains in doubt, something that should be reviewed in new studies.

P. suillus, this species has never been documented in Cabo Verde, and there is a notable scarcity of information about it in online databases, scarce or practically non-existent findings. In addition, no scientific papers on this species could be located. The substantial lack of information gives rise to uncertainty as to the existence of the species, regardless of its geographical distribution, causing it to maintain a non-valid status.

It was possible to obtain samples of *P. rogerii* that were caught in Cabo Verde, as well

as an online DNA sequence available in BoldSystems database[6], so it has gone from not valid to doubtful status. However, genetically we were not able to determine if it is this species, or if it was a taxonomic error. The following species, *P. mediterraneus, B. auritus* and *P. octolineatum*, retained their status (Table 7), not only because it was not possible to obtain samples from Cabo Verde, as well as the lack of information in the online databases.

5.1. Morphological Considerations

Considering that one of the objectives of this study was to gather all information regarding the species, whether taxonomic or genetic, published or unpublished, certain species stood out due to the ease with which their information was obtained. This highlights that they are more extensively researched species, particularly when they hold commercial value. One species that stands out most in this area is *P. incisus*. This species is mainly found along the African coast, Canary Islands and in the Mediterranean Sea and has been the subject of numerous studies (e.g., Kapiris et al., 2008; Pajuelo et al., 2003; Menezes et al., 2004) and has significant commercial value in the countries where it occurs. In the remarks of Wirtz et al., 2013, they mention that in the study by Monteiro 1998, *P. incisus* was mistaken for *P. humile* in a photo. However, with the advance of meristic and DNA Barcoding studies, nowadays, this species is hardly confused, as there is enough material to clarify the taxonomic doubts that may arise.

Of the species belonging to the genus *Pomadasys*, the ones that raise great taxonomic doubts and are often confused are *P. rogerii* and *P. jubelini*. During the research, it was noted that most of the studies carried out on *P. jubelini* are ecological and morphometric studies, with a great lack of taxonomic studies on this species. It was found a study, by Dorairajl (1970), about the distribution of this species, but in the images of the article, it is noted that it does not correspond to the current shape, which is in the FAO 2016 catalogue. Even more difficult was to find studies on *P. rogerii*, either ecological, genetic, or taxonomic studies. One of the databases used during the search was FishBase[7], where it was found photos and descriptions of the species. But clearly, one can see that the morphological features found in the description are confused between these two species and the same thing happens with the published photos.

[6] https://www.boldsystems.org/index.php/databases

[7] Fishbase.se

Making a brief morphological comparison of these two species, according to Carpenter & De Angelis (2016), we can highlight the main characteristics that differentiate them: *P. jubelini* - Body oblong and compressed, its depth contained 2.7 to 3.1 times in standard length; snout long in large individuals and pointed, its length 0.8 to 1.1 times in orbit diameter, eye moderately small, orbit diameter 3.0 to 3.6 times in head length; back and sides with small dark spots arranged in sinuous oblique or horizontal lines; a golden yellow blotch on the snout and a yellow golden to darkish blotch on the upper angle of opercle. *P. rogerii* – has a body oblong and compressed, its depth is 2.6 to 2.9 times in standard length; its Snout length is 0.6 to 1.1 times in orbit diameter; its eye is moderately small, orbit diameter of 2.9 to 4.2 times in head length; blackish or dark brown rounded spots irregularly spread on back and sides and tip of the lower lobe of caudal fin sometimes yellowish.

In the same genus mentioned above, one species that can also be confused with *P. jubelini* is *P. perotaei*. This is because, like *P. rogerii*, this species also has some morphological characteristics very similar to *P. jubelini*. However, its taxonomic status is well clarified, as no remarking was found during this study. As for its status, it remained the same as before, as it was not possible to obtain data on its presence in Cabo Verde waters, evidencing a lack of studies focused on this species.

The *P. octolineatum* specimen obtained during this study was not native to the Cabo Verde islands but rather from other regions, rendering its presence in this study unverifiable. This species has been formally described in Senegal, and given the typical geographical proximity of these countries, it is plausible that it could inhabit Caboverdean waters. Our research found that studies (e.g., Oliveira et al., 2015; Freitas et al., 2019) were identified in which *P. octolineatum* had been captured. However, due to the absence of a thorough taxonomic analysis (study based on visual census), certainty regarding its true identity remains elusive. Brito et al. (2007) highlight the confusion surrounding these species likely stems from the color loss that occurs when fish are preserved in cans. Notably, the coloration of these two species significantly differs, as illustrated in (Figure 6). In live *P. humile*, lighter lines exist, albeit sometimes faint and not consistently visible, with their prominence influenced by the observer's perspective. However, postmortem, these light lines fade rapidly. In contrast, *P. octolineatum* maintains the integrity of these lines in freshly deceased specimens.

Besides *P. incisus*, as one of the main species of the Haemulidae family in Cabo Verde, we have *P. humile*, which was sampled and identified in this study. Since it was identified and made its original description (e.g., Dupérier & Brygoo, 1983), it has always been remembered

in Cabo Verde, so it is considered an endemic species of the archipelago. More recent studies, like, Brito et al. (2007), also confirm this information.

The species *P. macrops* is also one of the species in which there was great difficulty in finding information about its taxonomy, as well as published data. The online databases do not have updated species descriptions, so the only one found was the original. Also, the photo found in the original description, in addition to not being in good condition, did not correspond to the morphological characteristics of the original description. So, the only photo that was found (Figure 6) belongs to a recent study, González et al. 2009, where this species was mentioned for the first time in Cabo Verde waters. The doubt about whether or not this species is valid for Cabo Verde is because, in the original description, there was already talk of the similarities concerning *P. humile*. Wirtz et al. (2013), also raise this same question.

Within the Plectorhinchus genus, *P. mediterraneus* maintains an invalid status, given its absence from Cabo Verdean waters. Notably, this species has accumulated significant information owing to its commercial significance in Mediterranean countries, driven by substantial consumer demand (Merlo et al., 2012). Consequently, this commercial interest has catalyzed numerous studies, underscoring its central role as a research focal point.

Like the previous species, *B. auritus* also has a lot of information available, as there are several studies (e.g., Adenike Adebiyi, 2013; Konan et al., 2015) which emphasize its important commercial value, mainly on the West African coast, focus in fishing management. Their status remained invalid as no evidence of their presence in Cabo Verde was found.

5.2. Genetic analyses

5.2.1. Phylogenetic Study

The morphological basis of our tree was corroborated and recovered as in previous studies (e.g., Liang et al., 2012; Tavera et al., 2012) showing strong support for the monophyly of the two subfamilies *Haemulinae* and *Plectorhinchinae*, which once again confirmed their status as sister groups. At the base of the tree, we have the subfamily *Haemulinae*, represented by two well-supported genera, the *Pomadasys*, and the *Brachydeuterus*. The *Pomadasys* in this study were presented as monophyletic, divided into 3 clades, corresponding to each species, respectively, *P. incisus*, *P. jubelini* + *P. rogerii*, and *P. perotaei*.

Different from the results in Tavera et al. (2018), in which *P. incisus* is found in a nested clade, we have the total separation of this species in this study. This species is divided into 2

subclades, a larger one (species from Cabo Verde) and a smaller one (sequences from online databases), this division is probably due to geographical differences.

Concerning *P. perotaei*, its clade exhibited a notable division into two segments, a distinction we again attribute to the apparent geographical divergence of these sequences. Intriguingly, one of the subclades coexists alongside a species of *P. jubelini* (B284), which itself resides in a separate, robustly supported clade along with other specimens. It is likely that was confronted in another instance of misidentification, potentially arising from the morphological resemblance between these species, as discussed in the preceding section. It is possible to see in the tree that these two species are very close genetically, being separated by a few mutations. During the alignment, some mutations of *P. perotaei* that corresponded to *P. jubelini* were observed, thus reinforcing the possibility of having been misidentified.

Based on different research sources (FishBase; Carpenter & De Angelis, 2016), *P. jubelini* and *P. rogerii* are confused, and their morphological descriptions are mixed up, more time is possible to show this in our phylogenetic tree. Bearing in mind that the only way to share a clade, with a node of 99% confidentiality, would be if the specimens belong to the same species. And as *P. jubelini* was sampled and identified by us, based on the updated FAO catalogue, unlike *P. rogerii* which was not identified in this study, we can say that there is a greater probability of *P. rogerii* (PROG001, CVERD171| *P. rogerii* and B227|*P. rogerii*) has been misidentified. During this study, this whole situation of misidentification, due to the morphological similarities of these two species made us hypothesize that perhaps, this all happens when *P. jubelini* is captured as a juvenile, as we believed that at this stage it is very similar to *P. rogerii*. In Cabo Verde, specimens of *P. jubelini* are usually caught in or near the adult stage. According to the FAO description, one of the characteristics that strongly distinguishes these two species is the size and shape of the nose (Table 4), and this is emphasized only in the adult stage. However, another hypothesis we can raise is that it may be a very recent divergence. The marker used in this study, CoxI, being a mitochondrial gene, is translated into an evolutionarily conserved protein and did not show a high separation of the species Prosser et al. (2013), meaning that this gene tends to remain relatively stable and unchanged over time, especially compared to other genes. So, we need to use a conjugate of nuclear and mitochondrial markers to distinguish the two species.

The other part of the *Haemulidae* tree has the subfamily *Plectorhinchinae*, which is divided by a well-supported (100%) confidence node, into three clades where we have, *P. mediterraneus*, *P. octolineatum*, and *P. humile* + *P. macrops* + *P. rogerii*. Two strongly

supported clades of two species, *P. mediterraneus* and *P. octolineatum. P. mediterraneus* in this study presents itself as a monophyletic species concerning the other species of its subfamily. Unlike other studies where it showed to be paraphyletic, according to Tavera et al. (2018) he was facing a synonymous species.

Most species belonging to *P. humile,* it is in a nested clade with *P. rogerii* and *P. macrops*. *P. rogerii* (PROG008VL) in this clade was probably a typo or a case of misidentification, as morphologically they do not share characteristics as they belong to different genera and subfamilies (Dupérier & Brygoo, 1983).

P. humile is considered to be an endemic species of Cabo Verde (Brito et al., 2007; Freitas, 2014), so they underscored that the Cabo Verde islands stand out due to their significantly greater abundance of coastal fish endemism and compared to the other Macaronesia archipelagos. Additionally, Cabo Verde exhibits disparities from these archipelagos in marine life, community composition, and ichthyogeography. Because it was sampled and identified in this study according to the original description and FAO catalogues, there is no doubt that the identification was well done.

It had been mentioned in the taxonomic analysis that since the original description *P. macrops* can be confused with *P. humile* and vice versa. This can be seen in the tree, that these two species are separated practically by a mutational step. As *P. macrops* was not identified in this study, only the tissue and the photo of the specimen were provided, we cannot confirm, if there is a misdetection. The ultimate arrangement of these species within the clade encompassing all *P. humile*, including *P. macrops*, must await a more exhaustive sampling of these species and renewed scrutiny of their morphological traits.

5.2.2. Population Structure

In the population structure of *P. rogerii* (Fig. 9 - haplotype network A), a slight genetic variation can be observed among specimens from Angola and Cabo Verde. These two regions are not geographically close, yet they could potentially harbor genetically similar species, just as Freitas et al. (2018) mention the occurrence of certain species of Cabo Verde, West African coast. However, the outcome of the phylogenetic analysis does not definitively confirm whether these specimens belong to *P. rogerii*.

The population of *P. humile* was presented with little variation with *P. macrops* (Fig. 9 - haplotype network B). Probably the *P. macrops* identified were *P. humile.* So, it may have

errors in the taxonomic identification and this does not allow us to identify these two species, for reasons explained above.

Due to the pronounced endemism in Cabo Verde, certain species may be gradually diverging genetically over time, influenced by factors like the isolation of the islands, substantial diversity of habitats, and the persistence of warm tropical waters during glacial periods (Freitas, 2014). Therefore, these and other reasons may explain the population structure of *P. incisus* (Fig. 9 - haplotype network C). Perhaps it is possible to raise the hypothesis that it is allopatric speciation. From the tree, it can be seen with great support and in the haplotype network we have the specimens from Cabo Verde, separated by many mutations from those from the Mediterranean. Therefore, more work should be invested to find out if we are facing a new species.

6. Conclusions

The results obtained from the taxonomic study, which involved morphological analysis of the species, proved challenging. This was due to the presence of closely resembling characteristics among some species. However, a more significant challenge emerged when working with certain species that could only be examined based on their original descriptions, which often remain outdated. Consequently, this outdated information gives rise to numerous uncertainties during the identification process. Furthermore, more available material is needed for certain species. This scarcity encompasses taxonomic information and genetic data, such as DNA barcoding, which could play a crucial role in substantiating the obtained results.

Regarding the question related to the difficulty in sampling the species necessary to carry out this study, we think it is since some species of Haemulidae can only be captured at greater depths, for example, *P. macrops* in González et al., (2009) which was caught between 120 and 150 m depth. Considering that in Cabo Verde the species of this family are caught by artisanal fishing, using nets (which allow a depth catch of up to 15 m), it is quite difficult to know if we can catch these species.

In the case of the presence or absence of *P. rogerii*, we noticed that when asked about this species in the FMSV, the fishmongers recognized it immediately as *P. jubelini*. So even if we have this species, distinguishing will be challenging due to the similar characteristics.

Surveys have noted that there needs to be more studies for some fish families in Cabo Verde. Usually, the most studied families are the fish with the highest commercial value, such as the Serranids and Tunids.

In general, the species of the family Haemulidae, distributed in the West African zone, need more taxonomic and genetic studies. This is because most of the species in the checklist of Haemulidae family for Cabo Verde were not found in genetics data in the online database (e.g., *P. jubelini*, *P rogerii*, *P. suillus*, *P. humile*), as well as a description of morphological characteristics (B. auritus, P. suillus, P. macrops).

The Haemulidae family for Cabo Verde has species from two subfamilies, Haemulinae and Plectorhinchinae. The subfamily Haemulinae is represented by the genera *Pomadasys* and *Brachydeuterus* and the Plectorhinchinae is represented by *Parapristipoma* and *Plectorhinchus*. The unity of the family and its subfamilies, along with clearly distinguishable branches within those subfamilies, receive strong confirmation across all analyses (with bootstrap values exceeding 80%). However, this phylogeny calls into question the validity of some of the species and leaves several other questions unanswered. As for the position of *P.*

rogerii remains unresolved until specimens are available. Defining boundaries and relationships of species such as *P. jubelini* and *P. rogerii*, as well as *P. macrops* and *P. humile* an intricate morphological examination will be necessary. This will serve to scrutinize and enhance the existing phylogenetic hypothesis.

One of the conclusions reached during this study is proof of the importance of DNA barcoding. Although it is a powerful tool for rapid and accurate species identification, it has advantages and disadvantages. Advantages include identifying of non-visible species (cases where species are difficult to distinguish based on external characteristics), universality across species (the CoxI marker is conserved in most species), applicability in conservation and forensics, and standardized methodology. The disadvantages noted in this study were limitations in differentiating closely related species (very genetically close species may have similar barcodes, making it difficult to differentiate them), limited scope (provides information only on the genetic diversity of a specific region of the genome), failures in poorly characterized species (when the underlying taxonomy is not well defined, DNA barcoding may result in incorrect identifications). Therefore, it is recommended that as remains a challenge in barcoding initiatives to substantiate results with taxonomics, it is up to the systematic community to accommodate the molecular data and decide on the levels of variation worthy of formal taxonomic treatment.

7. Recommendations

Although the results of this study clear up some of the existing taxonomic doubts, it will be crucial to complement them with a larger sample in terms of number, geographical area, and depth so that the presence and absence of Haemulidae species in Cabo Verde can be confirmed more precisely.

As for genetic analysis, more molecular markers need to be used, particularly in the mitochondrial and nuclear genomes to clarify the evidence further. It will be essential to carry out morphological and anatomical studies of the species, such as *P. macrops*, as it was not possible to find photos or morphological descriptions in the updated online databases. In general, it is recommended that this species be redescribed.

According to the results obtained in the phylogeographic analysis of *P. incisus*, a more exhaustive study is needed to understand better why there is so much genetic divergence concerning species from other geographical regions, which could raise the possibility of allopatric speciation. Also, it must be important to clarify the taxonomic errors that exist between *P. jubelini* and *P. rogerii* by conducting exhaustive sampling of these species, re-examining morphological characteristics, and carrying out genetic analyses using more molecular markers.

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Appendix

Pomadasys incisus (Bowdich, 1825)

Pomadasys perotaei (Cuvier, 1830)

Pomadasys jubelini (Cuvier, 1830)

Pomadasys rogerii (Cuvier, 1830)

Brachydeuterus auritus (Valenciennes, 1832)

Parapristipoma humile (Bowdich, 1825)

Parapristipoma octolineatum (Valenciennes, 1833)

Parapristipoma macrops (Pellegrin, 1912)

Plectorhinchus mediterraneus (Guichenot, 1850)