

Morphogenetic Sharks of Genus *Carcharinus* Traded at Southwest Aceh Fish Landing Sites

Morfogenetik Jenis Hiu Genus *Carcharinus* yang Diperdagangkan di Tempat Pendaratan Ikan Aceh Barat Daya

Asri Mursawal^{*1}, Samsul Bahri¹, Sri Wahyuni¹, Hayatun Nufus¹, Rudi Hermi²,
Muhammad Ali Sarong³

¹Department of Marine Science, Faculty of Fisheries and Marine Science,
Teuku Umar University, Aceh Barat, 23681 Indonesia

²Department of Aquatic Resource, Faculty of Fisheries and Marine Science,
Teuku Umar University, Aceh Barat, 23681 Indonesia

³Department of Biology, Faculty of Teacher Training and Education,
Syiah Kuala University, Banda Aceh, 23111 Indonesia

*Correspondent Author: asrimursawal@utu.ac.id

ABSTRACT

Research on the types of sharks traded at the Southwest Aceh fish landing site was conducted in September 2022. The purpose of this study is to analyze the morphology and genetic analysis of sharks. Sampling uses purposive sampling techniques, for morphological analysis refers to shark identification books, while genetic analysis uses the Sanger method used at the sequencing stage. Morphological data analysis using Image-j software, for genetic analysis, using Mega 6 (Molecular Evolutionary Genetic Analysis). Morphological analysis was carried out at the Laboratory of Biosystematics and Marine Genetics, Faculty of Fisheries and Marine Sciences, Teuku Umar University. Genetic analysis was carried out at the Denpasar Biodiversity Laboratory, Bali. The results of this study found two species of sharks with a total of 8 individuals from the genus *Carcharinus*.

Keywords: Shark, Morphology, Genetics, Aceh

ABSTRAK

Penelitian tentang jenis hiu yang diperdagangkan di tempat pendaratan ikan Aceh Barat Daya telah dilakukan pada bulan September tahun 2022. Tujuan penelitian ini adalah untuk menganalisis morfologi dan menganalisis genetik ikan hiu. Pengambilan sampel menggunakan teknik *purposive sampling*, untuk analisis morfologi mengacu pada buku identifikasi ikan hiu, sedangkan analisis genetik menggunakan metode sanger yang digunakan pada tahapan sekuensing. Analisis data morfologi menggunakan software *Image-j*, untuk analisis genetik menggunakan Mega 6 (*Molecular Evolutionary Genetic Analysis*). Analisis morfologi dilakukan pada Laboratorium Biosistematika dan Genetika Kelautan, Fakultas Perikanan dan Ilmu Kelautan, Universitas Teuku Umar. Analisis genetik dilakukan pada Laboratorium Biodiversitas Denpasar, Bali. Hasil dari penelitian ini ditemukan dua spesies ikan hiu dengan total 8 individu dari genus *Carcharinus*.

Kata Kunci: Hiu, Morfologi, Genetik, Aceh

INTRODUCTION

Sharks are a group of cartilaginous fish belonging to the class Chondrichthyes having jaws, paired nostrils, scales, two bear hearts, paired fins, and a frame consisting of cartilage instead of true bones. Sharks are the main component that occupies the food chain as a peak predator starting from coastal ecosystems (Knip et al., 2010). Many shark populations from around the world are experiencing a drastic decline every year (Ward-Paige, 2017). The main cause of the shark's decline is the exploitation of excess both by-catch and as the main target (Worm et al., 2013). Other causes of the decline of sharks such as climate change and habitat destruction (Dulvy et al., 2014). Indonesia is included as one of the countries that utilize the largest shark resources in the world. Characteristic habitat of any region near the coast influences the movement and distribution of each species of shark (White and Potter, 2004).

Genetics is one of the branches of the field of biology that examines the inheritance of traits and genes inherited by their ancestors. In general, genetics examines the science of genes and all aspects that are possessed in them and that are inherited from their parents. DNA (*Deoxyribonucleic Acid*) has a very important role in gene inheritance. DNA itself is a genetic base that can control the nature of living things stored and protected in polypeptides.

This study aims to analyze the morphological characteristics of sharks and analyze the genetics of sharks found at the fish landing site of Southwest Aceh Regency.

MATERIALS AND METHOD

Place and time of research

The study was conducted in September 2022 at the Southwest Aceh fish landing site shown on the location map (Figure 1). Shark sampling consists of 8 individuals from each type of shark found at the fish landing site. Morphological analysis was carried out at the Laboratory of Biosystematics and Marine Genetics, Faculty of Fisheries and Marine Sciences, Teuku Umar University. Genetic analysis was carried out at the Denpasar Biodiversity Laboratory, Bali.

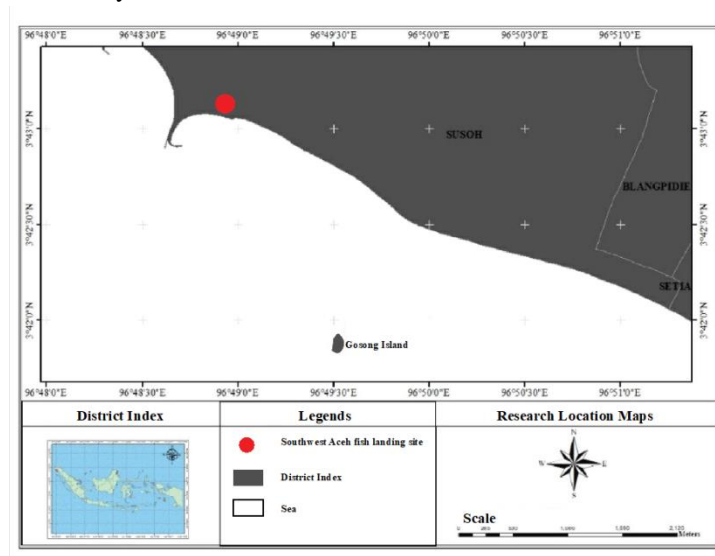


Figure 1. Research location map

Tool and materials

The tools used in this study include cameras, scale boards, coolboxes, sample bottles, logbooks, qiashredder spin column, dneasy spin column and collection tube, bunsen, forceps, centrifuge, glove, vortex, micropipette, tube 1.5 mL and PCR tube, microwave, beker cup, measuring tube, agar mold, PCR machine, electrophoresis device. Materials used to analyze morphology and genetics include 96% ethanol, ddH₂O, dNTP, MgCl₂, agarose, ETBR, and extraction kits.

Morphological analysis

Sampling uses a purposive sampling method where the sampling refers to certain characters to be used in analyzing morphology (Nawawi et al., 2020). Samples obtained from the Fish Landing Site were carried out morphological analysis concerning the shark identification book. The morphological analysis looks at several aspects of the identification key that each type of shark has referring to the identification books "On Board Guide for the Identification of Pelagic Sharks and Rays Western Indian Ocean" (Ebert, 2015) and "Identification Guide to Sharks, Rays, and Skates of the Southeast Asian Region" (Ali et al., 2017). Each individual is taken a fin part 1 cm long for genetic analysis.

Genetic analysis

The genetic analysis goes through several stages starting from extraction, amplification, electrophoresis, and sequencing. At the extraction stage using mtDNA tissue from shark fins. The extraction process uses the

DNeasy kit (QIAGEN Inc.) (Bernard et al., 2016). DNA amplification stages using fish-BCL forward and reverse paired primers: 5'-TCAACYAATCAYAA AGATATYGGCAC-3' and FISH-BCH: 5'-TAAACTTCAGGGTGAC CAAAAAATCA-3'. The stages in DNA amplification go through several processes ranging from heating the PCR (Polymerase Chain Reaction) machine at a temperature of 80°C for 10 seconds, pre-denaturation at a temperature of 94°C for 3 minutes, denaturation of 40 cycles at a temperature of 94°C for 30 seconds, annealing at a temperature of 50°C for 30 seconds and extension at a temperature of 72°C for 45 seconds. The final extension is at a temperature of 72°C for 5 minutes and followed by a cooling process up to a temperature of 37°C (Toha et al., 2020). The electrophoresis stage uses an electric field displacement machine to separate the molecules of chemical compounds contained in DNA (Madduppa, 2013). The composition at the electrophoresis stage includes gel agarose and Ethidium Bromide. The voltage used by the electrophoresis machine is 220V and a current of 400 mA with a duration of 25 minutes (Mursawal, 2020). The next stage uses a sequencing machine to carry out the process of sequencing nucleotide bases on the DNA from each fragment owned in it.

Data analysis

The data analysis process uses MEGA 6 (Molecular Evolutionary Genetic Analysis) software to align nucleotide bases. The nucleotide bases that have been aligned are then matched with the genetic database through the BLAST process which refers to the NCBI (National Center for Biotechnology Information) GenBank. The results of matching data with GenBank are then created as a phylogenetic tree to see the division of groups and types of individuals who have gone through the stages of genetic analysis carried out. Phylogenetic analysis using MEGA 6 software with the Neighbor-Joining statistical method. The phylogenetic analysis used a repeat of 1000 bootstraps and a 2-parameter model to produce a phylogeny tree

RESULT AND DISCUSSION

Based on morphological characteristics by the identification book, it was found that in the Southwest Aceh Fish Landing Site, there are 2 types of sharks landed including *Carcharinus sorrah* and *Carcharinus falciformis*. According to Hall et al. (2012) that *C. falciformis* is one of the most common types of sharks found in Indonesian waters. Morphologically these two species have a body color where the difference in *C. falciformis* is darker than that of *C. sorrah*. The muzzle shape or snout of *C. falciformis* is more rounded than that of *C. sorrah*. In addition, on the tail fin of *C. sorrah*, the lower part of the tip has a black pattern (black tips) while on *C. falciformis* it is slightly black (dusky).



Figure 2. Morphology of *C. sorrah*

The local name *C. sorrah* is a lanjaman cutut, some call it mungsing, feather peacock, or lanyam. Features of *C. sorrah* have a grayish body color in the dorsal part and white in the ventral part. Cutut lanjaman has 2 dorsal (dorsal) fins. The first dorsal fin has a black fringe while the second dorsal fin has a black tip. The tail fin has a black fringe around the tail fin (black margin) and the lower part of the tip has a black pattern (black

tips). This is to the opinion of Mopay et al. (2021) that *C.sorrah* has the characteristics of a long and slightly pointed muzzle, has 2 dorsal fins, and a black pattern on the tip of the second dorsal fin, pectoral fin, and lower caudal fin.

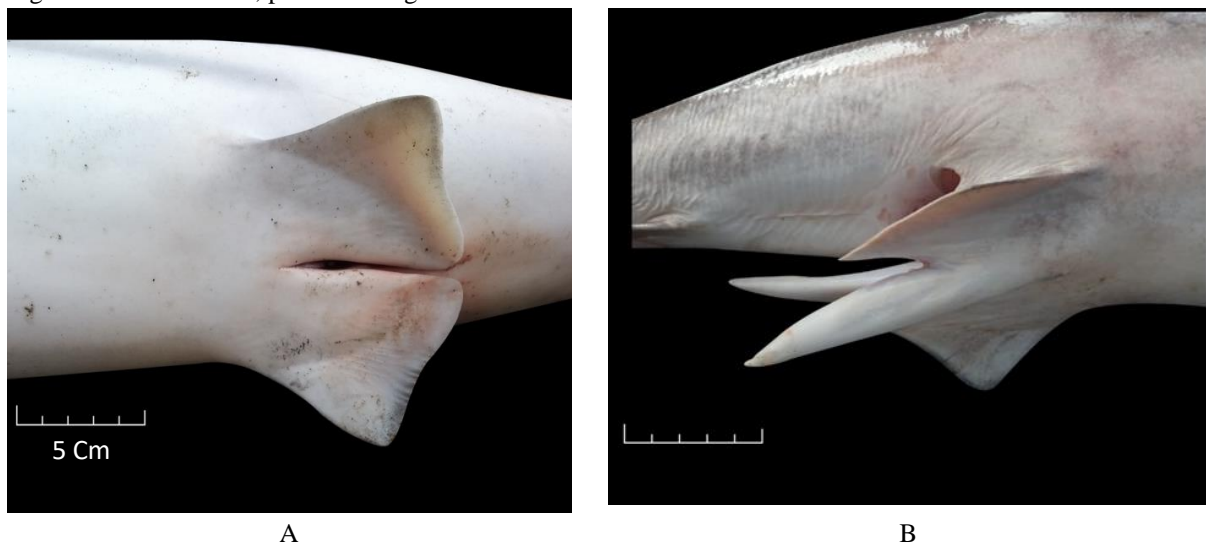


Figure 3. Morphology of *C. falciformis*

Silk shark or *C. falciformis* or some call it the janjam shark. Silk sharks have a blackish-gray body color on the dorsal part and light ash on the ventral part. The muzzle (snout) is long and rounded. The silk shark has 2 dorsal fins; the first dorsal fin has a longer size than the second dorsal fin. The tail fin does not have black tips but has the same dark color as the body color. According to Mopay et al. (2021), silk sharks have no streaks between their backs, rounded and long snouts, and small, slender, and erect lower teeth.

Sex ratio of male to female

The total number of sharks of both species is 8 heads consisting of 3 males and 5 females. Male sharks and female sharks have differences among them female fish have pelvic fins and cloaca while male fish have pelvic fins, cloaca, and clasper. Cloaca is a genital hole for female sharks, while clasper is a male reproductive organ. For more details, please see Figure 4.



A B
Figure 4. Differences between female sharks (A) and male sharks (B)

Sharks that are landed more female are thought to be due to the spawning season. According to Bhagawati et al. (2017) that the female fish will be more actively foraging to support the maturity of its gonads. The total length variable has a high tendency in determining the maturity of the shark's gonads. The result of measuring the total length of *C.sorrah* and *C.falciformis* landed in the Southwest Aceh Fish Landing Site ranged

from 96.91 - 184.83 cm. According to Ebert (2015) that *C.sorrah* mature female gonads range in size from 110 to 118 cm, males 103-128 cm while female *C.falciformis* 213-230 cm and males 183-217 cm. Shark type *C.sorrah* are 2 juveniles (25 %) and adults 6 tails (75%) while type *C. falciformis* is still a juvenile category. This shows that there are still sharks landed in the Southwest Aceh Fish Landing Site that are not yet mature gonads.

Genetic analysis

The results of analysis of the genetic data obtained showed that the base pair length of the DNA of the shark type *C.sorrah* averaged 646 Bp from a total of 3 samples, the type of *C.falciformis* with an average of 647 Bp from a total of 5 samples. The results of this genetic analysis showed the suitability of the same type/species when the morphological analysis was carried out. The level of suitability of the DNA sample data owned by the National Center for Biotechnology Information (NCBI) database is in the percentage range of Query Cover 99-100%, Percentage Identification 99.34-100%, indicating that the samples analyzed are of the same type as those found in databases.

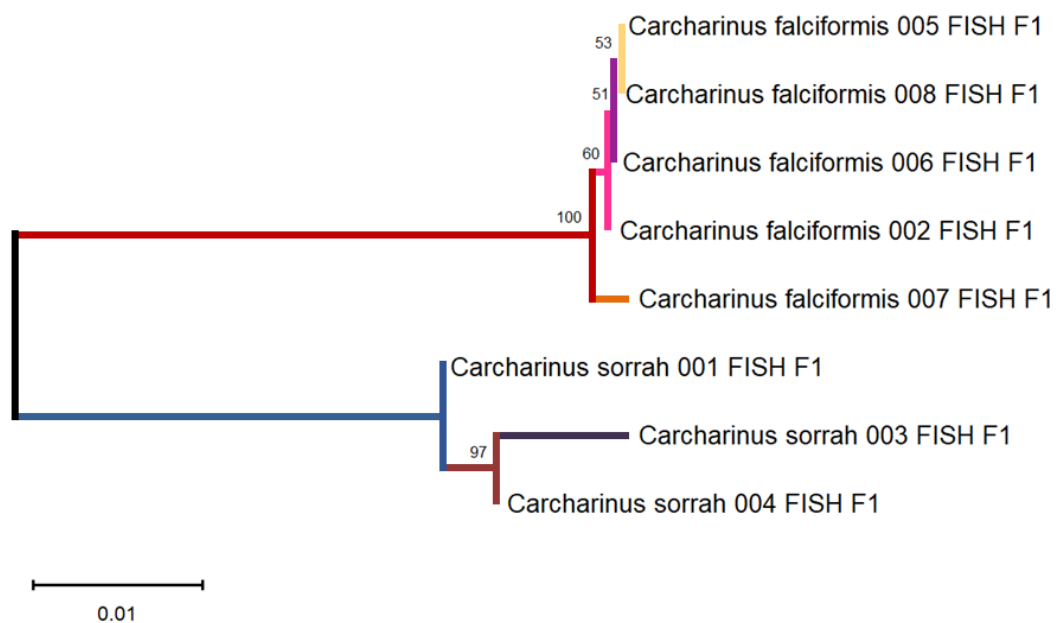


Figure 5. Phylogenetic tree

Genetic analysis that has been carried out using genetic data, obtains a phylogenetic tree. The phylogenetic tree is owned using Neighbor-Joining reconstruction. This reconstruction is used to see the kinship between individuals or species of *C.falciformis* and *C.sorrah*. The phylogenetic tree shows the division of groups between the species studied. From a total of 8 samples obtained from the field, 8 samples of genetic data were generated. The tree reconstruction shows that there is a branching divided into 2 clades which can be seen in Figure 5 which is indicated by the red and blue lines. This clade separates the species *C. falciformis* and *C.sorrah*. In the clades of these two species, there are slight genetic differences between the individuals of each of these species. There is a possibility that the individual genetic differences of the species *C. falciformis* and *C.sorrah* come from several areas or adaptations made to certain areas during a stopover. These sharks include fish that don't settle in one place or fish that migrate during certain seasons.

CONCLUSION

Based on the results of the research that has been done, it can be concluded that: 1) Two shark species were found being traded at the Southwest Aceh Fish Landing Site, namely *C. sorrah* and *C. falciformis*. 2) The total number of sharks of both species is 8 heads consisting of 3 males and 5 females. 3) The genetics of sharks found at the Southwest Aceh Fish Landing Site shows that there are 2 species of sharks, namely *C. sorrah* and *C.falciformis*, which have compatibility or genetic similarities in the database.

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