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Molecular and risk-based approaches to the status of Goldbanded Jobfish *Pristipomoides multidens* (Day, 1871) in Kupang, Indonesia

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Abstract. *Wijayanti DP, Indrayanti E, Haryanti D, Wijayanti MK, Elmir ZG, Fachri FR, Bhagooli R, Nozawa Y. 2025. Molecular and risk-based approaches to the status of Goldbanded Jobfish* Pristipomoides multidens *(Day, 1871) in Kupang, Indonesia. Biodiversitas 26: 928-940.* Snappers are economically valuable and important fishing activities in Indonesia. However, the lack of data on the stock status contributes to the uncertainty of the sustainability of the fisheries. We employed the Cytochrome C Oxidase subunit I (COI) mtDNA gene to analyze the genetic diversity of the fish in combination with the Risk-Based Framework (RBF) methods of the Marine Stewardship Council (MSC) to assess the status of the Goldbanded jobfish (*Pristipomoides multidens,* Day, 1871) fishery at Kupang, East Nusa Tenggara. Results indicated that all specimens collected from Kupang District belong to *Pristipomoides multidens* (Day, 1871), forming 2 clades. The clades have a low genetic distance, which likely suggests the fish are caught from the same fishing area. The phylogenetic tree reconstruction indicated potential genetic sub-divisions in the snapper population in the Timor Sea. The Risk-Based Framework approach showed the risk potency of the fishing activity has a low effect on the Goldbanded Jobfish, the baitfishes such as mackerel tuna, scads, Indian mackerel and squids, and most of the by-catch fishes. The final MSC score for the Goldbanded Jobfish fishery was 74, below the MSC ≤80 guide post, suggesting a medium risk potential faced by the fishery. Therefore, catch regulations, such as harvest control rules or open-close fishing should be applied to sustain the fishery.

Keywords: COI gene, demersal fishery, *Pristipomoides multidens*, risk-based framework, Snapper

INTRODUCTION

Indonesian capture fisheries have the characteristics of capturing many species with all non-target species being landed. The composition of fish resources is dominated by 36% of small pelagic fish and 25% of large pelagic fish (Pet et al. 2022). Potential fish resources in Fisheries Management Area (FMA) 573 based on the Ministry of Marine Affairs and Fisheries (MMAF) Decree No.19, 2022 are 1,338,442 tons. The estimation of source potential fish capacity for the demersal fish is 299,600 tons/year (Azizah et al. 2023). Capture fisheries activities are largely unregulated. Control of commercial fishing activities is carried out only based on a sailing vessel license. Most fisheries are "data poor", due to the cost and difficulty of monitoring fish populations and catches (Wibisono et al. 2019). Population biology data of species are inadequate (Dimarchopoulou et al. 2023). National data collection has been based on logbooks, which are frequently inaccurate. As a result, official statistics generally pool various species according to their color, such as Red Snapper (Satria et al. 2019).

The demersal fishery that targets snappers, is considered one of the most valued fish worldwide (Jovanovic and Rafols 2018). Indonesia is the second largest snapper exporter (Cawthorn and Mariani 2017). Snapper fisheries stretch from Sumatra to West Papua. They are taken at depths between 50-500 m using multi-gear dominated by droplines and bottom-longlines with vessel size varying between small boats to large industrial fleets (Wibisono et al. 2021). More than 100 species are landed, with 75% of landed fishes dominated by *Pristipomoides multidens* (Day, 1871), *P. filamentosus* (Valenciennes, 1830), *P. typus* (Bleeker, 1852), *Atrobucca brevis* (Sasaki & Kailola, 1988), *Epinephelus areolatus* (Forsskål, 1775), and *Lutjanus malabaricus* (Bloch & Schneider, 1801) (Wibisono et al. 2019).

The Goldbanded Jobfish (*P. multidens*), also known as Goldband Snapper is widely distributed along the Indo-Pacific region. The fish inhabits reefs on hard bottom areas at depths of 60 to 180 m (Allen 1985). The catch value of the species reached USD 176 million for domestic and international markets (Mous et al. 2021). However, the Goldbanded Jobfish have slow growth rates, mature at a late age, and extended longevities, making them particularly vulnerable to overexploitation (Newman et al. 2016). Despite, the fish is a commercially valuable species, the molecular data on the species is largely unknown (Liu et al. 2023). Not much research has been carried out on the genetic structure of the fish, especially from Indonesian waters (Ovenden et al. 2002, 2004; Wigati et al. 2003). By using restriction fragment length polymorphism and direct sequencing, Ovenden et al. (2004) found a high haplotype diversity among samples collected from northern Australia, central (Tanimbar and Tual), and eastern (Bali, Flores, Sumbawa, West Timor) Indonesia. Meanwhile, Wigati et al. (2003) applied an allozyme marker and reported that samples collected from Moluccas have higher genetic variation than those from Bali and Sumbawa. It was thought that the Bali and Sumbawa samples formed a single population. It was suggested that the genetic connection among samples and the distribution of the fish were affected by the current (Ovenden et al. 2002, 2004). Mitochondrial DNA (mtDNA) which has the characteristics of maternal inheritance, close gene arrangement, and a high evolutionary rate, is an excellent tool to study the phylogeny of the species. A recent study reported that the COI gene of mitochondrial DNA (mtDNA) is an excellent tool for species identification and phylogenetic relationships within the family Lutjanidae (Liu et al. 2023). Sala et al. (2023) characterized the phylogenetic relationship among red snappers from Yapen, Papua using the COI of mtDNA as well.

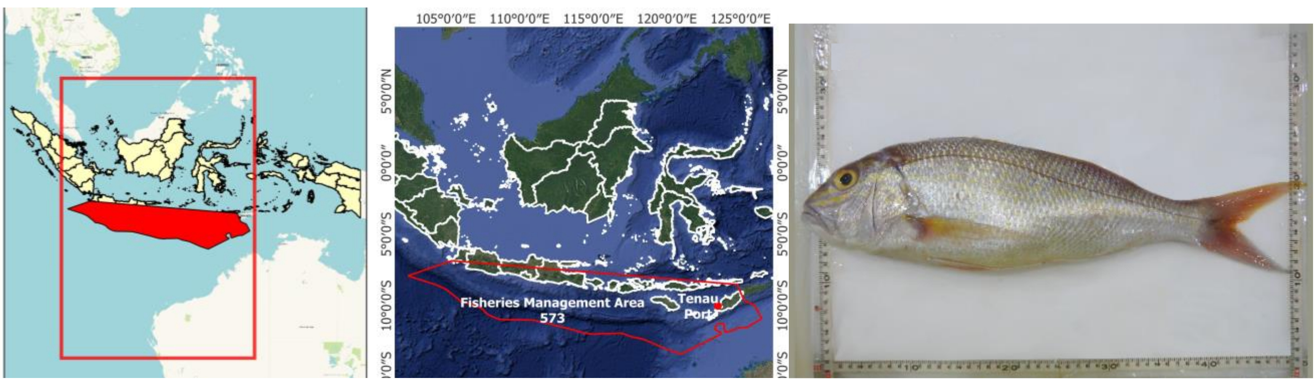
The FMA 573 is located in the Indonesian part of the Timor Sea, near the edge of the Australian continental shelf, is one of the largest fishing areas for snappers, contributing 69% of 81% of the total catch in 2020 (Pet et al. 2022). Total export volume reached 4,172,056 kg with a value reached USD 12,452,211. The fishing grounds are commonly found around West Timor, Rote Island, and the Sawu Sea. The fish are caught mostly by small-scale fishing vessels, with Tenau, Kupang as the main fishing port (Edo and Nalle 2024).

The importance of snapper and grouper fisheries, accounting for 45% of the world's total production, made the fish suitable for the eco-certification scheme. Currently, there are numerous eco-certification schemes (Fletcher 2015) by which the product is assessed against a set of sustainability standards to convince the customer that the product being sold meets the standards of the label (Blandon and Ishihara 2021). In capture fisheries, the Marine Stewardship Council (MSC), remains the organization that shows robust and sustainable principles of certification (Good et al 2024). However, fulfilling the certification procedure is more challenging in developing countries where fisheries are artisanal with poor and limited data (Saldaña-Ruiz et al. 2022).

MSC developed a Risk-based Framework to evaluate data-deficient fishery together with the ecological risk assessment instruments to address the potential risk posed by specific fisheries. Within the framework of ecological risk assessments, there is the semiquantitative Productivity and Susceptibility Analysis (PSA), CA (Consequence Analysis), CSA (Consequence Spatial Analysis), and SICA (Scale Intensity Consequence Analysis), which can be applied to estimate the vulnerability of a particular stock to fishing activity by analyzing the interaction of fishing with the stock, the habitat, and the capacity of the species to face the impacts of fishing (Saldaña-Ruiz et al. 2022). It was reported that Indonesian populations may act as donors of the western Australia coast stock population (Payet et al. 2024). Here, we apply the Risk-based Framework to assess the status of the Goldbanded Jobfish fishery in Kupang District, East Nusa Tenggara together with the genetic diversity of the species to confirm the genetic status of the stock.

MATERIALS AND METHODS

The study was conducted from October to November 2019 and November 2022 at Kupang District, East Nusa Tenggara, Indonesia, which covers the waters around West Timor, Rote Island, Savu Sea, throughout WPP 573 with Tenau Port at Kupang as the main fishing landed base (Figure 1).



A B

Figure 1. A. The fishing ground distribution of vessels that targeted the Red Snapper; B. Goldbanded Jobfish, *Pristipomoides multidens*

Molecular studies

Samples collection

In total 40 pectoral fin samples of the Goldbanded Jobfish snapper were collected on November 14-15 2022 from Kupang; 20 samples were collected from PT MAS (Matsyaraja Arnawa Stambhapura) and 20 samples were obtained from Oeba Fish Market. PT MAS sells high-quality fish with the Goldbanded Jobfish being one of the important products. About 40% of the products are exported to Japan, Australia, the USA, and Taipei. PT MAS obtained the products from 40 fishing vessels measuring 6-29 GT with 300-320 fishers as the crew. The fishery operated a dropline and bottom-longline at the FMA753. The fishers work with companies through a plasma system where the company provides operational costs for fishing. The catch is sold by the fishers to the company by deducting the fishing cost from the whole price of the catch. There is no supplier or wholesaler scheme in the business process. Meanwhile, the seller at the Oeba Fish Market obtained the fish directly from the fishermen or Tenau Port. The fish samples were taken from the muscles near the dorsal fin using a sterilized knife and tweezers. Before collecting the tissue, each sample was photographed and identified according to Mous et al. (2022). The sample was then stored in a 15 mL Falcon tube containing 96% ethanol and preserved at room temperature for further analysis. The fishing grounds of the samples were deduced from an interview with the officer of PT MAS, the seller at the Oeba Fish Market, and fishermen.

DNA extraction, PCR amplification, and DNA barcoding

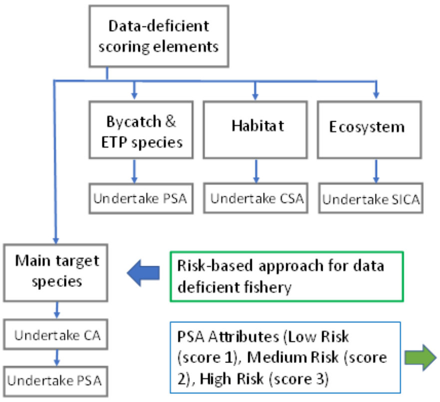
Genomic DNA was obtained following the extraction using a modification of the Chelex 100 method (Pereira et al. 2016). The sample is placed into a 0.6 mL tube containing 10% Bio-Rad BT Chelex at a ratio of 1 : 10 (Kanedi et al. 2023; Tapilatu et al. 2023). All samples were vortexed for 15 seconds and centrifuged for 1 minute at 8000 rpm before being incubated in the heating block for 45 minutes at 95°C. Primer pair of Forward primer (jgHCO2198): 5'-TITCIACIAAYCAYAARGAYATTGG-3' and Reverse primer (jgLCOI490): 5'-TAIACYTCIGGRTGICCRAARAAYCA -3' (Geller et al. 2013) was used for PCR (Polymerase Chain Reaction) amplification of partial gene fragments of Mitochondrial DNA COI (Cytochrome Oxidase subunit I). A 25 μL reaction mixture containing 1.25 μL of DNA template, 12.5 μL of My TaqTM HSRed Mix PCR kit (BIOLINE; 25 μM MgCl2, 5 U/μL Taq Polymerase, 10x Taq Buffer, and 10 μM dNTPs), 1 μL of each primer and 9.25 μL of distilled water. The mixture was run in a thermal cycler using the following PCR cycle: 95°C initial denaturation for 4 minutes; followed by 40 cycles of denaturation at 95°C for 30 seconds, primer annealing at 50°C for 30 seconds, and extension at 72°C for 1 minute; and a final extension step at 72°C for 10 minutes (Saleky and Dailami 2021). All amplified PCR products were then analyzed by electrophoresis in 2% agarose gel (FMC Bioproduct, Rockland, ME, USA) in 1x TAE buffer to assess the yield and run for 20 minutes (10 V cm^{-1}; 40 mA). The gel was then visualized under a UV Transilluminator (GelDoc) after being stained with ethidium bromide. All amplified samples were then sent to PT. Genetika Science for sequencing. Sequences were conducted using Big Dye Terminator v.3.1 according to the company protocols.

Construction of phylogenetic tree and data analysis

The Basic Local Alignment Search Tool (BLAST) of the NCBI (National Centre for Biotechnology Information) (http://blast.ncbi.nlm.nih.gov/), National Institute for Health USA searching was utilized to verify the sequence obtained against the gene database. Alignment of the DNA sequence analysis was performed using MEGA XI software (Kumar et al. 2018). A phylogenetic tree from the sequences was made using the Maximum Likelihood method, Kimura-2-parameter model and 1000 bootstrap replicate to evaluate the degree of confidence at each node. The bootstrap method with 1000 repetitions was applied as well when constructing the genetic distance (Abdussamad et al. 2016). Genetic diversity was examined by determining the numbers of segregating sites (S), haplotype number (h), and haplotype diversity (Hd) using DNAsp (Rozas et al. 2017).

Risk-based framework approach

The risk potency analysis of the Godbanded Jobfish fishery was conducted using the Risk-Based Framework (RBF) referred to MSC (Marine Stewardship Council) guidelines 2014 (Good et al. 2024). Data for the RBF study was collected in October 2019. Data was documented based on several discussions with various stakeholders in Kupang District namely fishermen, representatives of the fishing industry (PT MAS), Non-Government Organizations, the local Fish Quarantine Center, and government officials of Marine and Fisheries Affairs of East Nusa Tenggara Timur. RBF was conducted using CA (Consequence Analysis), PSA (Productivity Susceptibility Analysis), SICA (Scale Intensity Consequence Analysis), and CSA (Consequence Spatial Analysis) approach of the MSC ver. 2.1 (Figure 2). The semi-quantitative data were collected according to the type of assessment. The CA was only used to assess the main target species; the PSA was applied to assess the main target species; bycatch; and ETP (Endangered, Threatened, and Protected) species. While CSA was used to assess the habitat, and the SICA was used to assess the ecosystem. The bait species was also assessed besides the main target species, bycatch, and the ETP species. All assessment type refers to the attributes and scoring in the RBF worksheet available on MSC Guideline 2014. Primary data on the total length of the fish and the weight of the fish were done of the fish caught by the fishermen of PT MAS and the samples obtained from Oeba Fish Market. Data was also provided from secondary data and other relevant literature. Additional data on the sustainable stock status of the Godbanded Jobfish was collected from PT MAS and Oeba Fish Market.



Productivity attribute	High productivity (low risk, score = 1)	Medium productivity (medium risk, score = 2)	Low productivity (high risk, score = 3)
Average age at maturity	<5 years	5-15 ears	>15 years
Average maximum age	<10 years	10-25 years	>25 years
Fecundity	>20.000 eggs per layer	100-20.000 eggs per year	<100 eggs per year
Average maximum size (not to be used when scoring invertebrate species)	<100 cm	100-300 cm	>300 cm
Average size at maturity (not to be used when scoring invertebrate species)	<40 cm	40-200 cm	>200 cm
Reproductive strategy	Broadcasts spawner	Demersal egg layer	Live bearer
Trophic level	<2.75	2.75 – 3.25	>3.25
Density dependence (to be used when scoring invertebrate species only)	Compensatory dynamics at low population size demonstrating or likely	No depensatory or compensatory dynamics demonstrated or likely	Depensatory dynamics at low population sizes (Allee effects) demonstrated or likely

Susceptibility attribute	Low susceptibility (low risk, score = 1)	Medium susceptibility (medium risk, score = 2)	High susceptibility (high risk, score = 3)
Areal overlap (availability): Overlap of the fishing effort with species concentration of the stock	<10% overlap	10-30% overlap	>30% overlap
Encounterability: The position of the stock/species within the water column relative to the fishing gear, and the position of the stock/species within the habitat relative to the position of the gear	Low overlap with fishing gear (low encounterability)	Medium overlap with fishing gear	High overlap with fishing gear (high encounterability). Default score for target species (Principle 1)
Selectivity of the gear type: Potential of the gear to retain species	a. Individuals < size at maturity is rarely caught	a. Individuals < size at maturity are regularly caught	a. Individuals < size at maturity are frequently caught
	b. Individuals < size at maturity can escape or avoid gear	b. Individuals < half the size at maturity can escape or avoid gear	b. Individuals < half the size at maturity are retained by gear
Post-capture mortality (PCM): The chance that, if captured, a species would be released and that it would be in a condition permitting subsequent survival	Evidence of majority released post-capture and survival	Evidence of some released post-capture and survival	Retained species or majority dead when released. Default score for retained species (Principle 1 or Principle 2)

Figure 2. Matrix of Risk-based Framework (RBF) of Marine Stewardship Council (MSC) guidelines 2014

RESULTS AND DISCUSSION

Genetic diversity of the Goldbanded Jobfish

A total of 39 samples were successfully amplified and sequenced. The final sequence results varied in length between 657 and 671 bp. The samples were identified as *Pristipomoides multidens* with Query Cover values ranging from 95-99% and identify values varying from 94-100% (Table 1). The similarity (Identity) results with GenBank data showed that 2 samples out of a total of 39 samples had poor results with query identification ≤90% (AB3 and AB7-1). Meanwhile, 4 other samples had results ranging between 94% and 96% ((AB4 (96.95%), AB11 (94.94%), AB17 (95.99%), and AB18 (96.12%)). One sample has 98.33% percent identification with query covering 98% however, it failed to get deposition code. All the 7 samples were excluded from Table 1. All the sequences generated from the samples successfully deposited in the GenBank database (htttp://www.ncbi.nlm.nih.gov) are shown in Table 1.

Phylogenetic tree, genetic distance, and haplotype diversity

The sequences generated from the samples were analyzed by reconstructing the phylogenetic tree. The tree was built using 32 deposited sequences that were granted Genbank Accession Number. The samples with the codes AB4, AB11, AB17, and AB18 that were not granted an accession number from the Genbank were excluded. The tree was constructed to examine the evolutionary relationship of Goldbanded Jobfish from Kupang District, co-analyzed with *P. multidens* from Australia with accession number (MK092068.1, EF609436.1, KX781861.1, FOAC022-05, FOAC023-05, FOAC024-05, FOAC024-05, FOAC025-05, FOAG373-08, FOA1882-19) and Philippine with accession number (OQ387822.1).

The phylogenetic tree topology of *P. multidens* of Kupang District shows the formation of 2 clades with the same group as *P. multidens* obtained from GenBank. All samples are in a different clade from the outgroup used, namely *P. sieboldii*. This confirms that the sample species studied is *P. multidens* (Figure 3). Most samples are grouped in the first clade, consisting of mixed samples collected from Oeba Fish Market and PT MAS. While the other samples namely A14-2, A37, and A25 collected from PT MAS formed the second clade. Based on Nei's (1972) range of genetic distance showed that all samples have a low genetic distance (0-0.004; the low category ranged from 0.01 to 0.09; moderate 0.1-0.9 and high 1.0-2.0), which means that all samples have a close relationship in the phylogenetic tree.

Haplotype diversity (gene diversity) represents the probability that two randomly sampled alleles differ (Phillips et al. 2019). Haplotype diversity (Hd) results showed a value of 0,7198, while the genetic distance between Clade 1 and 2 is 0.014 (Table 3).

Table 1. Homology analyses of Godbanded Jobfish from Kupang District

Samples code	BLAST identification	Base pair (bp)	Query cover (%)	Ident (%)	Accession code	Deposited accession code
A02	*Pristipomoides multidens*	665	98	100	OQ387822.1	OR625453
A04	*Pristipomoides multidens*	668	98	98,48	OQ387822.1	OR654025
A05	*Pristipomoides multidens*	668	98	100	OQ387822.1	OR625454
A06	*Pristipomoides multidens*	664	98	99,69	OQ387822.1	OR625455
A08	*Pristipomoides multidens*	661	99	100	OQ387822.1	OR625456
A13	*Pristipomoides multidens*	665	98	100	OQ387822.1	OR625457
A14-1	*Pristipomoides multidens*	668	98	100	OQ387822.1	OR625679
A14-2	*Pristipomoides multidens*	668	98	100	MK092068.1	OR625680
A16	*Pristipomoides multidens*	657	99	99,39	OQ387822.1	OR625681
A17	*Pristipomoides multidens*	665	98	99,85	OQ387822.1	OR625682
A18	*Pristipomoides multidens*	661	99	100	OQ387822.1	OR625683
A19	*Pristipomoides multidens*	663	99	99,39	MK092068.1	OR626049
A24	*Pristipomoides multidens*	668	98	100	OQ387822.1	OR626050
A25	*Pristipomoides multidens*	668	98	100	MK092068.1	OR626051
A28	*Pristipomoides multidens*	668	98	99,85	OQ387822.1	OR626052
A29	*Pristipomoides multidens*	665	98	100	OQ387822.1	OR626053
A33	*Pristipomoides multidens*	665	98	99,85	MK092068.1	OR632203
A34	*Pristipomoides multidens*	665	98	99,69	OQ387822.1	OR632204
A35	*Pristipomoides multidens*	668	98	100	OQ387822.1	OR632205
A37	*Pristipomoides multidens*	665	98	100	MK092068.1	OR632206
A38	*Pristipomoides multidens*	665	98	100	OQ387822.1	OR632207
A39	*Pristipomoides multidens*	668	98	100	OQ387822.1	OR632998
A40	*Pristipomoides multidens*	668	98	100	OQ387822.1	OR632999
AB2	*Pristipomoides multidens*	668	98	100	OQ387822.1	OR633000
AB5	*Pristipomoides multidens*	665	98	99,54	OQ387822.1	OR636714
AB6	*Pristipomoides multidens*	671	98	99,85	MK092068.1	OR633001
AB7-2	*Pristipomoides multidens*	668	98	99,69	OQ387822.1	OR633002
AB10	*Pristipomoides multidens*	665	98	100	OQ387822.1	OR633677
AB12	*Pristipomoides multidens*	665	98	100	OQ387822.1	OR633678
AB13	*Pristipomoides multidens*	669	98	98,18	MK092068.1	OR636715
AB16	*Pristipomoides multidens*	665	98	100	OQ387822.1	OR633679
AB20	*Pristipomoides multidens*	665	98	99,08	OQ387822.1	OR633680

Notes: A: Samples obtained from PT MAS, AB: Samples collected from Oeba Fish Market

Table 2. Genetic diversity of Goldbanded Jobfish: n: Number of samples; S: Number of variable sites; h: Number of haplotypes; Hd: Haplotype diversity

n	S	h	Hd
32	27	14	0.7198

Table 3. Genetic distance between clades of Goldbanded Jobfish

Clade	Between distance	Within distance
	Clade 2	
Clade 1	0.014	0.003
Clade 2	-	0.022

Risk-based framework approach

Overview of the fishery

Most of the fishers in FMA 573 fished using dropline and bottom-longline. Drop-line fishers searched for schools of tropical snappers in depths of 80-160 m along reef fronts and on sand flats located near pinnacles. Multiple droplines may be deployed at any one time, each with 30 to 40 hooks (size 11/0 to 13/0) attached to a heavy cord mainline. Hooks are attached by snoods or swivels (short lengths of monofilament fishing line) to a short section of line that is then attached to the mainline with a ‘shark’ (quick release) clip and weight attached. The use of a quick-release clip allows operators to attach pre-baited hooks and continue fishing, while the hauled section with fish attached is cleared. Hooks, generally baited with squid, are set within 25 m of the seabed for 3 to 15 minutes. The bottom-longline fishers usually use 29 GT vessels that are equipped with 5 multiple bottom-longlines (5 blong) comprised of 700 tuna hooks. Length of the line will be about 4 km (1.5-1.8 miles). The length of the tuna hook is about 5.5 cm. The bottom-longline used scads (*Decapterus* spp.), Indian Mackerel (Kembung Lelaki, *Rastrelliger kanagurta* (Jordan & Starks, 1908)), and sardine (Lemuru, *Sardinella lemuru* (Bleeker, 1853)) while the dropline used Tongkol (*Auxis* spp; *Euthynnus* spp.), and squids (*Doryteuthis pealeii* (Lesueur, 1821)) as the fish bait. The vessel brought at least 100-200 kg for one fishing trip (at least 15 days of fishing). The fishery developed lines buoyed and set free of the vessel and later retrieved and hauled aboard by a mechanized winch, however, most vessels have hydraulic or electric reels. Every day did 4-5 settings, with one setting about 5 hours before the line was hauled (soaking time 30 minutes to 1 hour).

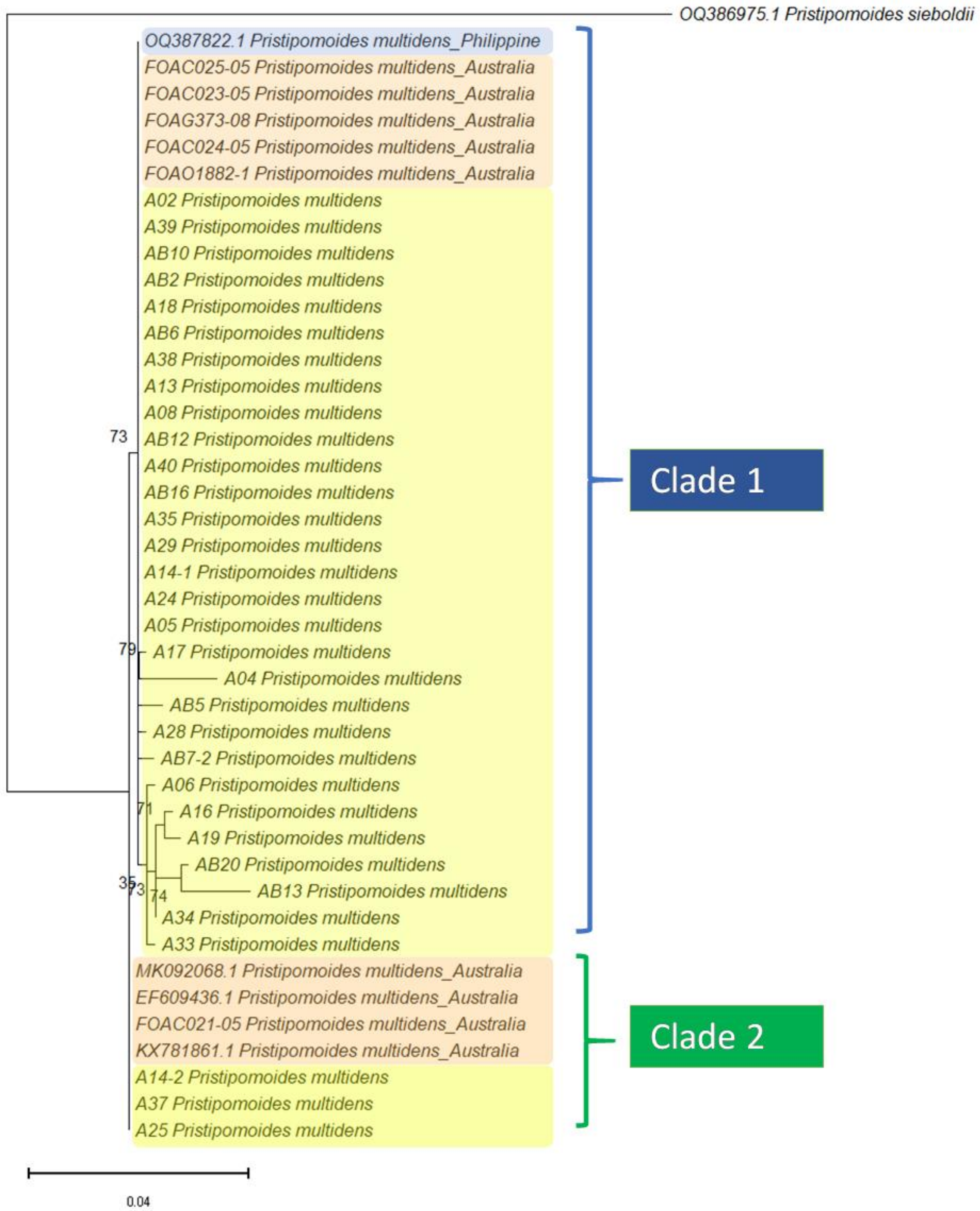


Figure 3. A phylogenetic tree based on the mitochondrial DNA COI of 32 sequence data that were granted an accession number from the Genbank shows the relationships between Goldbanded Jobfish (*Pristipomoides multidens*, Day 1871) and the most closely related Goldbanded Jobfish recorded in the GenBank database. The samples obtained from PT MAS (Matsyaraja Arnawa Stambhapura) and Oeba Fish Market were grouped in Clade 1. Three samples obtained from PT MAS formed the second clade. All samples clustered out of the outgroup sample, *P. sieboldii*. Samples with yellow highlights are our sample from this current research while those with blue and pink highlights were ingroups from the Genbank data collected from the Philippines and Australia, respectively

Life history parameter values of Goldbanded Jobfish

By applying a Crew-Operated Data Recording System (CODRS) method for onboard monitoring of species- and length-composition of catches, Yayasan Konservasi Alam Nusantara (YKAN), successfully obtained data from top 100 species of demersal fishes in the 11 FMA. Participating fishers are equipped with a measuring board, camera, and a GPS tracking device that records vessel coordinates at every hour. In CODRS, fishers are required to take photographs of their entire catch on a measuring board during fishing (Mous et al. 2021; Dimarchopoulou et al. 2023).

Based on the CODRS study, the Goldbanded Jobfish is one of the most important species with a total volume of catch of 18,886 MT or 16% of the total catch. The fish is commonly traded in a mix with the Sharptooth Jobfish (*P. typus*) (Pet et al. 2022). According to the Decree of the MMAF Number 19/2022, the total catch of demersal fishes already exceeded the Total Allowable Catch (TAC) of demersal fishes in FMA 573. The TAC is set at 80% of the Maximum Sustainable Yield (MSY). The TAC value of demersal fish is 269.640 tons/year (Azizah et al. 2023). However, there is no explanation for the TAC of each species.

The Spawning Potential Ratio (SPR) is important for managing fish stocks. SPR is described as a comparison of the spawning ability of stock when subject to fishing to the stock's spawning ability if unfished (Hordyk et al. 2015). The ratio is expressed in percentages between 0-100%. The SPR value can describe the overall egg production of the fish population that may affected by fishing activity. SPR value represents the ability of the stock to maintain its population. For conservation reference points, the SPR commonly is set at 40%. Depending on the fish species, some stocks can maintain the population when the limit reference point of the SPR is kept at 20 to 35%. While less than 10% value of the SPR may lead stock to severe declines (Prince et al. 2015). The SPR value for *P. multidens* based on CODRS study conducted by YKAN is below the reference point of 25% (14%) (Mous et al. 2021).

Risk potency analysis

Productivity Susceptibility Analysis (PSA) was developed to analyze the ecology risk potency of the Goldbanded Jobfish fishery towards target species, secondary species, by-catch species, baitfish, and habitat. Ecological risk variables consist of productivity parameters that cover average age of maturity, average maximum age, fecundity, average maximum size, average size at maturity, reproductive strategy, trophic level and susceptibility, covering availability, encounterability, gear selectivity, and post-capture mortality. The results of Consequence Analysis (CA) can be seen in Table 4 and Productivity Sustainability Analysis (PSA) is presented in Table 5.

Primary and secondary species

Mackerel tuna is classified as a primary species since it is subjected to management under the Fisheries Management Plan of Tuna-Skipjack- Mackerel tuna (MMAF Decree No. 107, 2015) as well as Sardine (*Lemuru*; *Sardinella lemuru*) under the MMAF Decree (No. 68/2016). Under MSC CR v2.0, primary species are defined as the other target species and are subject to management. Indonesia supplies more than 16% of the world's tuna, skipjack, and mackerel tuna production. In addition, Indonesia is the largest production country among 32 Indian Ocean Tuna Commission (IOTC) member countries.

Table 5. Final recapitulation of productivity susceptibility analysis results on Goldbanded Jobfish of dropline and bottom-longline Fishery

Component	Type of gear	
	Dropline	**Bottom-longline**
MSC PSA_derived score	87	87
Risk Category name	Low	Low
MSC scoring guidepost	≥80	≥80
Consequence Score (CA)	60	60
Final MSC score (per scoring element)	74	74

Table 4. Consequence analysis of the Goldbanded Jobfish

	Scoring element	Consequence subcomponents	Consequence score
Stock status outcome	Goldbanded Jobfish (*Pristipomoides multidens* Day, 1871)	Population size	Fish stock is at high risk of overfishing *60
		Reproductive capacity	Gonochoristic. Possible to do intersex (Hassell et al. 2018). Fecundity was between 205.547-3.028.159 eggs (Hukom et al. 2006). *70
		Age/size/sex structure	Max 30 years (Newman and Dunk 2002) *80
		Geographic range	Indo-Pacific from Samoa west to the Red Sea (Allen 1985) and East Africa, and from southern Japan southward to Australia (Allen 1985). It is found to depths between 40 and 245 m.
Rationale for most vulnerable subcomponent	Population size was chosen because there are anecdotal references that catch/trip is decreasing. Only 5 to 6% of the available stock can be harvested sustainably (Newman and Dunk 2002). The percentages of *Pristipomoides multidens* landed in the most recent 365 days, n = 45,469 Immature (<48cm): 43% Small mature (≥48cm, <64cm): 44% Large mature (≥64cm): 12% Mega spawner (≥70.4cm): 3% (a subset of large mature sh) Spawning Potential Ratio: 14 % (Mous et al. 2021). The vast majority of the fish in the catch have not yet achieved their potential growth. The harvest of under-size fish promotes overfishing, and it size distribution]indicates that over-exploitation may already be happening. The risk level is high. Catching small and immature fish needs to be actively avoided and a limit on overall fishing pressure is warranted.		
The rationale for consequence score	**60		

Notes: *: CA score = 60, 80, 100 (depends on the condition of the sub-components, when the condition of the species is sufficient, it will be scored higher. **: The situation is improving; the percentage of immature fish caught over recent years is falling, the percentage of Large Mature, Mega Spawner has risen over recent years; the percentage of SPR has risen over recent years (Mous et al. 2021)

In the third quarter of 2022, mackerel tuna production reached 159.311 tons, which increased by 1.36% from the third quarter of 2021 which reached 157.178 tons (Azizah et al. 2023). Mackerel tuna fishery was regulated under the Fisheries Management Plan (as mentioned previously) covering 11 FMAs including the Indian Ocean, which is part of the Indian Ocean Tuna Commission (IOTC) management area. Most mackerel was caught in multispecies fisheries, mainly by small purse sein The composition production of mackerel FMA 571, 572 and 573 was dominated by Kawakawa (31.2%), Frigate Tuna (29.0%), Longtail Tuna (17.1 %), Narrow-barred soanis mackerel (14.2%), Indo-Pacific King Mackerel (6.2%) and Bullet Tuna (2.4%) MMAF Decree No. 107/2015). The level of mackerel tuna utilization in FMA 573 is 0.99 (yellow indicator), or at the fully exploited level (Damayanti et al. 2023).

Sardinella lemuru contributes significantly to industrial and commercial sardine fisheries throughout most of its range, as well as subsistence fishery in Indonesia. Landings reported to the FAO suggest that the population is declining. Based on an exponential regression, the landings have declined by 32% over the past 10 years. Therefore, *S. lemuru* is assessed as Near Threatened under criterion A2bd (Santos 2018). The increasing uncontrolled number of purse seine vessels in Indonesia has been attributed to the overfishing of this species in the region targeting a large proportion of juveniles and small individuals (~70%) (Santos 2018). In FMA573, lemuru are concentrated in the waters of the Bali Strait. An acoustic survey of the Marine Fisheries Research Institute (BPPL) in the Bali Strait Waters indicates that lemuru are concentrated in parts of Java and Bali, at depths of less than 200 meters, beyond which they were not found. The catch of *S. lemuru* in 2005-2014 in FMA 573 tended to decrease. In the years 2009 to 2012, it experienced a large decline, then began to increase in 2013 and 2014 (MMAF Decree No. 68/2016). A recent study reported that the ecological dimension of lemuru fishery management in the Bali Strait showed a less sustainable index, mostly due to the utilization of the fish, the size of the fish caught, and the total catch obtained (Satyawan et al. 2023).

Secondary species are defined as all species caught, other than the primary species. By-catch composition comprised some members of Lutjanidae, Serranidae, Carangidae, Haemulidae, and one species of Glaucosomatidae, Scomberomorus, Lethrinidae, Scombridae, Sparidae and Loliginidae. The composition of the by-catch of the gear is presented in Table 6. The PSA of the by-catch species and bite fishes can be seen in Table 7.

Table 7. Productivity susceptibility analysis of by-catch species

Scientific name	MSC PSA-derived score	Risk category	MSC scoring guide post
Lutjanus rivulatus	92	Low	≥80
Lutjanus malabaricus	86	Low	≥80
Epiniphelus amblycephalus	91	Low	≥80
Cephalopholis sexmaculata	94	Low	≥80
Plectropomus leopardus	84	Low	≥80
Aprion virescens	89	Low	≥80
Caranx ignobilis	84	Low	≥80
Carangoides gymnostethus	75	Med	60-79
Epinephelus areolatus	92	Low	≥80
Paracesio kusakarii	78	Med	60-79
Plectorhinchus schotaf	75	Med	60-79
Pristipomoides typus	92	Low	≥80
Gymnocranius grandoculis	75	Med	60-79
Glaucosoma buergeri	75	Med	60-79
Pagrus auratus	89	Low	≥80
Lutjanus bohar	89	Low	≥80
Scomberomorus commerson	81	Low	≥80
Epinephelus epistictus	86	Low	≥80
Epinephelus bleekeri	92	Low	≥80
Decapterus sp.	92	Low	≥80
Rastrelliger kanagurta	95	Low	≥80

Table 6. Bycatch species of drop-line and bottom-longline

Family	Species	Common name	Drop-line	Bottom-longline
Lutjanidae	*Lutjanus rivulatus* (Cuvier, 1828)	Blubberlip Snapper		√
	Lutjanus malabaricus (Bloch & Schneider, 1801)	Malabar Blood Snapper		√
	Aprion virescens (Valenciennes, 1830)	Green Jobfish	√	√
	Paracaesio kusakarii (Abe, 1960)	Saddle-Back Grouper	√	√
	Pristipomoides typus (Bleeker, 1852)	Sharptooth Jobfish		√
	Lutjanus bohar (Forsskål, 1775)	Two Spot Red Snapper	√	√
Serranidae	*Epinephelus amblycephalus* (Bleeker, 1857)	Banded Grouper	√	√
	Epinephelus sexmaculatus (Rüppell, 1830)	Sixblotch Hind		√
	Plectropomus leopardus (Lacepède, 1802)	Leopard Coral Grouper		√
	Epinephelus areolatus (Forsskål, 1775)	Areolate Grouper	√	√
Carangidae	*Caranx ignobilis* (Forsskål, 1775)	Giant Trevally		√
	Carangoides gymnostethus (Cuvier, 1833)	Bludge	√	√
Scomberomorus	*Scomberomorus commerson* (Lacepède, 1800)	Narrow-barred Spanish Mackerel		√
Sparidae	*Pagrus auratus* (Forster, 1801)	Australasian Snapper		√
Glaucosomatidae	*Glaucosoma buergeri* (Richardson, 1845)	Deepsea Jewfish	√	√
Haemulidae	*Plectorhincus schotaf* (Forsskål, 1775)	Minstrel Sweetlips		√
Lethrinidae	*Gymnocranius grandoculis* (Valenciennes, 1830)	Blue-lined Large Eye Bream		√
Carangidae	*Decapterus* spp.	Scads		√
Scombridae	*Rastrelliger kanagurta* (Cuvier, 1816)	Indian Mackerel		√
Loliginidae	*Doryteuthis pealeii* (Lesueur, 1821)	Longfin inshore squid	√	

Consequence Spatial Analysis (CSA) is used to describe gear impacts (consequence) and the habitat (spatial) for each habitat that is being affected by fishing gear. The CSA methodology and attributes were developed based on the 'Ecological Risk Assessment for the Effects of Fishing' methodology (Good et al. 2024). The methodology was derived from images, expert opinion, and scientific literature. The result of the CSA score is shown in Table 8. The Scale, Intensity, and Consequence Analysis (SICA) is developed to identify which fishing activities significantly impact the ecosystem. The result of the SICA measurement can be seen in Table 9.

Table 8. CSA rationale table of the habitat

Consequence	Rationale	Score
Regeneration of biota	Regeneration of the biota occurs at deep sea for the target species; the bycatch species all retained	1
Natural disturbance	High in coral reefs area	1
Removability of biota	Erect, medium (<30 cm), moderately rugose, or inflexible biota or moderately robust, shallow-burrowing biota	1
Removability of substratum	The line is hauled not dragged on the bottom	1
Substratum hardness	Unconsolidated sediments	3
Substratum ruggedness	Rocky reefs/sand flats/gravel	3
Seabed slope	In depths of 80-160 m along reef fronts and on sand flats located near pinnacles	1
Spatial	**Rationale**	**Score**
Gear footprint	Demersal longline	2
Spatial overlap	UoA overlap with a habitat is ≤15%	1
Encounterability	Likelihood of encounter- ability is >75%	3
	CSA Score	2. 15
	Final CSA Score	93
	Risk Category	Low

Table 9. SICA scoring for ecosystem

	Spatial scale of fishing activity	Temporal scale of fishing activity	Intensity of fishing activity	Relevant subcomponents	Consequence score
PRINCIPLE TWO: Ecosystem outcome	5	5	3	Species composition	-
				Functional group composition	-
				Distribution of the community	-
				Trophic size/structure	-
Rationale for spatial scale of fishing activity	The ecosystem is deep water with rocky reefs or gravel or sand flats. The overlap with the fishery is 45-60%				
Rationale for temporal scale of fishing activity	Fishing occurs between 201-300 days per year				
Rationale for intensity of fishing activity	Threats to the *P. multidens*, in assessment location mostly are due to limited available habitat (fishing grounds) and predictable locations of fish concentrations, combined with a very high fishing effort on the best-known fishing grounds, as well as the targeting of juveniles. There is a high potential for overfishing in the deep slope hook and line fisheries. The feeding grounds of snappers also are predictable and well-known. Moreover, the market preference for plate size makes the catch impairs the sustainability of the species and increases the risk of the species (Mous et al. 2021). Adjustment upwards of trading limits towards the size at first maturity would be a straightforward improvement in these fisheries. Regulation on minimum size limits to be caught, the number of gear units and fishing season may help to increase the number of *P. multidens* stock in the assessment location.				
Rationale for consequence score	Consequence score is precautionary due to insufficient information.				

Discussion

Molecular studies

The mitochondrial Cytochrome Oxidase subunit 1 (COI) gene is a highly effective marker utilized for the molecular systematics of fish (Bhattacharya et al. 2016; Bingpeng et al. 2018; Liu et al. 2023). Fish DNA barcoding is widely used as an accurate method for species identification, authentication, and phylogenetic analysis of species including Indonesian fish (Toha et al. 2020; Kanedi et al. 2023; Sala et al. 2023; Tapilatu et al. 2023; Ramadhaniaty et al. 2024; Wora et al. 2024). BLAST findings indicated that the Goldbanded Jobfish snappers collected from the Kupang District belonged to a single species *P. multidens* (Table 1). However, samples AB3 and AB7-1 have identification percentages lower than 90%, and samples namely A26B, AB4, AB11, AB17, and AB18, have identification percentages lower than 99%, presumably, there is an error produced by copying the name (and function) of the reference protein to the query sequence (Pearson 2013). The rest of the sample has a similarity level between 94-99%, not as high as reported for *L. gibbus* from Papua Waters (Pranata et al. 2024).

The results of Haplotype diversity (Hd) showed a value of 0,7198, indicating a high value of haplotype (Table 2). The current study found 27 distinct variation sites, designating a high variable of haplotype. Haplotypes are specific combinations of alleles at multiple loci, and their diversity can reflect the genetic variation present. The Hd value suggests a moderate to high level of genetic diversity within the population. The value implies the possibility of a stable population structure, allowing for a mix of genetic material (Phillips et al. 2019). Moreover, the value often correlates with higher reproductive success which may affected by habitat type (Martinez et al. 2018).

Based on the reconstruction of the phylogenetic tree shown in Figure 3, when specimens from the Philippines and Australia were added, the tree analysis formed 2 clades but they did not appear to be grouped by region. Moreover, the tree showed that most of the *P. multidens* obtained from the sampling area have similarities to samples obtained from the Philippines and Australia. Therefore, the tree supports the occurrence of potential genetic sub-divisions in the stocks of snapper in the Timor Sea as well as in the stocks of snapper in the Arafura Sea as reported by Ovenden et al. (2004) and Payet et al. (2024).

The genetic distance between the two clades is also low, ranging from 0.014 (Table 3). This value shows that the Goldband Jobfish snapper has a small genetic distance. According to Devy et al. (2021), a relatively low or small genetic distance value indicates a close relationship. When the genetic distance between or among species is close, the samples observed are morphologically and genetically similar. Genetic distance can measure the level of gene differences between populations and genes (Bramasta et al. 2021).

Moreover, the close genetic distance may explain that the fish collected from PT MAS and Oeba Fish Market were caught in the same fishing area, suggesting one unit of stock. The small genetic distance between the Indonesian Goldband Jobfish snapper and Australia as well as the Philippines can be caused by several factors. Regional connectivity due to currents (Saleky and Daelami 2021), similar habitats and migration patterns (Saleky et al. 2016), as well as genetic mixing or the presence of genetic exchange, may play a role that Goldband Jobfish from different locations have genetic similarities. The fishing area for snapper fish landed at Tenau Port and Oeba Market comes from the same area, namely the FMA 573. This area includes the waters of the Savu Sea, the West Timor Sea, and the Indian Ocean South of Java, and south of Nusa Tenggara (Herwaty et al. 2023). Geographically, this water area is between the southern part of Indonesia and the Australian continent. Indonesian Through Flow (ARLINDO) is suggested to have an important role. The current flows and carries warm water from the western Pacific Ocean to the Indian Ocean through the Timor Sea which is the biggest volume of flow set off northwestern Australia (Gordon et al. 2019). The flow encourages dispersal and causes genetic homogeneity between species in Indonesia and several regions of Australia (Ovenden et al. 2013). The exchange of individuals among marine populations occurs mostly during the pelagic larval stage which is facilitated by sea currents (Cowen and Sponaugle 2009). There is growing evidence of the importance of sea currents in affecting the genetic structure of marine populations (Xuereb et al. 2018), which may control the genetic homogeneity in the fish populations (Damerau et al. 2012) and soldier crab *Mictyris guinotae* populations of the Ryukyu Islands, Japan (Kobayashi et al. 2025). Snead et al. (2023) suggested that the inherent asymmetry of ocean currents may trigger genetic differentiation and directional migration which causes complex distribution of genetic variation in intertidal species. Moreover, additional support was obtained from the research by Sartimbul et al. (2023), which states that Indonesia Through Flow (ARLINDO) and seasonal circulation determine the genetic connectivity of *S. lemuru*.

Risk-based framework approach

The demersal fisheries of snapper are economically important in contributing to the well-being of millions of people in Indonesia. However, the poor data raise uncertainty about the sustainability of the fish. We applied a risk potency analysis to assess the Goldbanded Jobfish (droplines and bottom-longlines) fisheries of FMA 573. Ecological risk variables that incorporate fish productivity were observed. The Consequence Analysis (CA) and the Productivity Sustainability Analysis (PSA) for the risk category of Goldbanded Jobfish fishery show medium to low-risk category (Tables 4 and 5). Table 4 shows that the CA score is 70, while the final MSC score was 74 (Table 4) which was below than MSC scoring guidepost (≤80). This suggests that the potential risk of snapper fishing in Kupang District is medium.

Our study shows, based on measurements of the total length of the samples obtained from PT MAS and Oeba Fish Market, the length of samples ranges between (28±4.35-61±2.78 cm), while the weight ranges between 437-4054 g. It was reported that the length of the *P. multidens* species at first maturity is 38 cm. Most of the samples studied have reached length at first maturity, only 2% of the samples have length <38 cm. The maximum weight of the species is reached at 8664 gr, while the weight of the first maturity is 1356 gr (Pet et al. 2022).

Currently, observation on the Spawning Potential Ratio (SPR) is widely used to describe the probability of the stock maintaining the population under the fishing pressure activity. The SPR value for *P. multidens* based on CODRS study conducted by YKAN is below the reference point of 25% (14%) (Mous et al. 2021). When a comparison was made between the SPR of the CODRS study in 2015 and 2020, there was an improving trend of SPR for a percentage of large mature and mega spawners (Table 4). “Mega spawners” define fish size larger than 1.1 times the optimum harvest size (Alam et al. 2022).

The Goldbanded Jobfish in WPP 573 is fished mostly using dropline and bottom-longline. The dropline utilized mackerel tuna (*Auxis* spp.; *Euthynnus* spp.) and squid, while the bottom longline used Scads *Decapterus* spp., Indian mackerel *R. kanagurta* and sardine *S. lemuru* as the bites. The PSA of the bite fishes and by-catch species show that the MSC-derived score ranges between 75-95, which means that the risk category is medium to low (Table 7). *C. gymnostethus, P. kusakarii, P. schotaf, G. grandoculis, G. buergeri* are the by-catch fishes that the MSC score is categorized as medium risk. Their stock status should be taken into account when exploitation is continued. However, all of the by-catch fish are utilized and sold to the local market or Bali. The bottom longline sometimes catches small sharks. The fishers also reported some sharks were accidentally caught. Most of the shark carcasses (ex. Scalloped hammerhead) were discarded and polluted the sea (personal communication with Fransisko Meo, the officer of the Province Office of Marine and Fisheries Affair). However, there is no adequate information regarding the type of shark species, the qualitative data of the shark, and the treatment of accidentally caught sharks.

The result of the CSA score shows that the gears have a low impact on the habitat (Table 8). The drop line fishery is considered a clean method since the gear is less harmful to the habitat of the target fish. Dropline fisheries are characterized by a very low impact on habitat at the fishing grounds, whereas some impact from entanglement can be expected from bottom-longlines. The application of bottom-longline is suggested to reduce the impact of the gear on the Vulnerable Marine Ecosystem (Pham et al. 2014). As for the SICA score, the temporal scale of fishing activities is likely to put pressure on the fish target since the days of fishing scale is 5, which means that fishing occurs intensively between 200-300 days. During the east or west monsoon, the fishery will not operate. The spatial scale of fishing activity is scored 5 which suggests that the overlap of the ecosystem with the fishery is 45-60%. The rationale for the intensity of fishing could be alarming since most market preferences asked for the plate size of fish. In this situation, the catch impairs the sustainability of the fish and increases the risk of target species (Mous et al. 2021). However, our result showed that less than 2% of the fish were caught below the first maturity length based on measurement of the total length of the samples obtained from PT MAS and Oeba Fish Market.

In conclusion, the use of molecular study combined with the RBF method is rarely applied in Indonesia to describe the status of the snapper fishery. Both methods may help provide more robust data to understand the fishery status. The study found that all samples obtained from Kupang District are identified as *P. multidens*. The samples formed two clades and the genetic distance between the clades is low. The haplotype diversity analysis showed a high value of haplotype. The Goldbanded Jobfish of Kupang District have similarities with GenBank data of the Goldbanded Jobfish of the Philippines and Australia, indicating that they are sourced from the same stock. Our study also showed that the risk potency arising from the fishing activity had a low impact on the Goldbanded Jobfish, the baitfish, namely mackerel tuna, scads, Indian mackerel, and squids, as well as the by-catch fishes. The final MSC score for the Goldbanded Jobfish fishery was 74 (below the MSC ≤80 guide post), which likely suggests the fishery faces a medium risk potential. This alarming situation should be brought into consideration. Management of the area should be applied with caution to avoid the decline of the stock which may have a wider impact on the population network of the fish. The study recommends applying catch regulation of the fish. The fishery needs to avoid catching small fish and stock should be assessed on at more regular basis. Therefore, the sustainability of the fisheries can be warranted.

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