Physicochemical Properties and Microstructure of Mixed-species Surimi Made from *Decapteru***s sp. and** *Priacanthus* **sp.**

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Abstract

The surimi industry plays a substantial role in bolstering Indonesia's fishing sector. The collapse of the majority of surimi companies can be attributed to the government limitation on trawling, which served as the primary source of demersal fish utilized in surimi processing. This restriction resulted in a significant shortage of raw material for industry. Therefore, it is imperative to explore alternate fish species that can serve as viable raw materials for the production of surimi. Alternative fish species are needed as raw materials for surimi. The objective of this work was to investigate the physicochemical characteristics and microstructure of surimi produced from two distinct Indonesian fish species: *Decapterus* sp., a pelagic species, and *Priacanthus* sp., a demersal species. Five ratios of *Decapterus* sp and *Priacanthus* sp; 1:0 (F1: control 1), 3:1 (F2), 1:1 (F3), 1:3 (F4), and 0:1 (F5: control 2) were tested. The proximate composition showed that the addition of *Decapterus* sp. to the formula increased the protein and fat content of mixed surimi. The formula F4 showed the highest value for almost all textural profile parameters; there were springiness, cohesiveness, gumminess, chewiness, and resilience. The same result also presented in the gel strength, F4 also has the highest value. The Water Holding Capacity value was low, and it showed no difference between all formulas. The whiteness value was in the range of 66-68, and it showed no difference between all formulas. Protein patterns showed no myosin heavy chain in all formulas. Microstructure analysis showed that only F5 (control 2) was compact and denser, but the other formulas (F1, F2, F3, and F4) showed a large number of loose and porous structures. In the microbial analysis, all formulas showed the Total Plate Count value (< 6 x 10⁵ cfu/g) was under the maximum acceptable value (10⁷ cfu/g). The findings of the study indicated that the mixed-surimi, consisting of *Decapterus* sp and *Priacanthus* sp at a ratio of 1:3, exhibited the highest level of gel strength. In conclusion, *Decapterus* sp has the potential to serve as a viable raw material when combined with *Priacanthus* sp.

Keywords: microstructure, mixed species, physicochemical, surimi

Introduction

Surimi is an intermediate fish product, and a wide range of products can be developed from it depending on their functional and nutritional properties. Due to the growing population and consumer's awareness of nutritious food, there has been an increase in the demand for surimi-based products in recent decades. The world surimi production in 2020 reached over one million MT, including 650,000 MT of tropical fish surimi, 250,000 MT of Alaska pollock surimi, 60,000 MT of freshwater fish surimi, 40,000 MT of other cold-water surimi and 10,000 MT of other fish species surimi. Approximately 60% of the world production is from Southeast Asia (tropical fish), 28.9% from the US (Alaska pollock and Pacific whiting), 6.1% from China (silver carp), 3.7% from Japan (pollock), 1.0% from Argentina and Chile (southern blue whiting), and 0.4% from France (blue whiting). (Park, 2020; Sihono et al., 2021, Yin & Park, 2023). According to them, the surimi from Southeast Asia was produced from tropical fish such as lizard fish, big eye snapper, threadfin bream, ribbon fish, croaker, and other demersal fish species. Since Indonesia's coastline and fishing grounds are the longest in Southeast Asia countries, Indonesia has the largest fish resource to produce surimi in this region.

Over 50% of the world's fish is produced in the Asia-Pacific region, with China leading the way, followed by Indonesia. The quantity of raw materials to produce surimi in Indonesia rose from 157,000 tons in 2004 to 196,000 tons in 2008, decreased in 2009, then rose to 350,000 tons in 2014 and 463,000 tons in 2018. Indonesia contributed approximately 28% of the production of the demersal fish used as raw materials to produce surimi, including *Mullidae, Nemipteridae, Priacanthidae, Sciaenidae*, and *Synodontid*. Unfortunately, Indonesia produced only 40,000 tons of tropical fish surimi in 2019, lagging behind China (230,000 tons), Vietnam (180,000 tons), India (90,000 tons), and Thailand (60,000 tons). The low productivity of surimi industry might be due to the fact that most potential fishing grounds in Indonesia are located in remote areas at the East part of the country with poor infrastructure, electricity, water supply and some other logistics, whereas the Western part of fishing grounds which are closer to the populated and industrialized areas have been depleted (Guenneugues & Ianelli, 2013; Pangsorn et al., 2007; Park, 2020; SEAFDEC, 2022). New investments in surimi factories and infrastructure in the potential fishing grounds areas are expected to be able to lower the production cost and increase productivity.

The surimi industry supports a significant contribution to Indonesia's fishing industry. In 1995, it was marked as the beginning of the surimi industry, and as of 2017, sixteen surimi industries were in operation. The total investment reached US\$ 188.6 million, labor absorption reached 7,725 individuals, and 90% of surimi production was exported in 2015 with the value of US\$ 142 million (Hikmayani et al., 2017). However, since 2017 the production of raw materials for surimi industry dropped significantly resulted from the regulation enforcement of the government banning trawl (known locally as *cantrang*) operation in most fishing grounds in Indonesia, since the demersal fish as raw material for surimi processing mainly came from the trawl by catch. Due to a lack of raw material, fish price increased and consequently many surimi producers collapsed (Hikmayani et al., 2017). To overcome this problem, alternative solutions must be formulated immediately to help the continuation of surimi industry, one of which is finding a new formula of surimi using potential raw materials available locally.

Numerous studies in surimi formulation have been done to investigate the effects of using different fish species (Suryaningrum et al., 2015), treatments (Cao et al., 2018; Núñez-Flores et al., 2018; Wang et al., 2019; Zhang et al., 2018) and natural products (Dong et al., 2019; Zhou et al., 2019; Zilda, 2014) resulted unique functional characteristics of surimi. *Decapterus*

sp. is an important fisheries commodity in Indonesia, and its production was 117,264 tons (13.95% of capture fisheries production) in 2020 (BPS, 2020). It has the potential to become a raw material for surimi production. Due to the portion of red flesh greater than white flesh, *Decapterus* sp. was producing surimi mixed with *Priacanthus* sp., a white flesh fish that is common to surimi material. However, research on using mixed species to produce surimi was limited. For this reason, the mixed surimi from *Decapterus* sp. and *Priacanthus* sp. was formulated in this study. Therefore, the effects of surimi made from a combination between a pelagic spesies (*Decapteru*s sp.) and a demersal (*Priacanthus* sp.) on physicochemical properties and microstructure of the surimi were investigated.

Material and Methods

Material and Surimi Preparation

Fresh bigeyes fish (*Priacanthus* sp.) was obtained from fish landing site at Cituis, Banten, Indonesia. The fish was supplied by one-day fishing fisherman. The frozen *Decapteru*s sp. was obtained from Nizam Zachman Ocean Fishing Harbour, Jakarta, Indonesia. *Decapteru*s sp. was gutted and filleted. The fillet was then minced with meat mincer (Bibun, Japan). While *Priacanthus* sp was gutted and beheaded. The fish was put into meat bone separator machine (0.2 mm diameter of holes) (Bibun, Japan) and the minced meat was then collected.

The surimi was made following the method used by Suryaningrum et al. (2015) with some modification. Surimi was processed by washing the minced meat twice using 0.5% (w/w) NaHCO₃ in fresh water for 30 min at 4–6 °C. The ratio of the water and minced meat was 1: 5. After the second washing, the third washing was done using 0.5% w/w of salt with the same ratio of water and meat, same temperature and duration time were applied as in the first and second washing. The meat was then put into meat dehydrator (Bibun, Japan) to reduce its moisture content. To remove scales and spines, the meat was filled into strainer machine (Bibun, Japan) then the meat was collected. A cryoprotectant (4% w/w of sorbitol, 4% w/w of sucrose and 0.2% w/w of polyphosphate) was added to the meat and mixed using a mixer (Bibun, Japan). The minced meat of *Decapteru*s sp. and *Priacanthus* sp were processed separately. The ratios of mixed surimi from *Priacanthus* sp and *Decapterus* sp. were following Yi et al. (2020) with a slight modification. The minced meat was then mixed using five different ratios of *Decapteru*s sp. and *Priacanthus*

sp. (1:3; 1:1; and 3:1; with 1:0 and 0:1 as the controls). As much as 500 g of each portion of the mixed meats were filled into a stainless pan measuring 20 cm x 20 cm x 7 cm, then frozen and preserved in a cold storage with the temperature of -18 °C until used. Two identical processes were done separately as replications.

Surimi Gel Preparation

Surimi gel preparation referred to Zhou & Yang (2019) with a slight modification. Thawed surimi was chopped for 1 min using meat chopper (Philips, series 5000) then 2.5% of NaCl (w/w) was added and chopped for another 3 min at 4 $^{\circ}$ C. The paste was stuffed into collagen casing with diameter of 27 mm and both ends were tightly sealed. The surimi paste was incubated at 80–90 °C for 15 min and immediately transferred into cold water with ice (4 °C) for 20 min. The cooked surimi gels were stored at 4 °C overnight prior to analysis.

Proximate Analysis

The proximate analysis in this study refers to the Indonesia National Standard (SNI). The samples were examined for moisture content (AOAC, 2000), crude protein (AOAC, 2005), crude fat (AOAC, 2005), ash (AOAC, 2000), and total carbohydrate (by different). The moisture content was determined using air oven drying at 105 °C until a constant weight was reached. The crude protein content $(N \times 6.25)$ was calculated using the macro-Kjeldahl method. The crude fat content was analyzed using the Soxhlet extraction method with petroleum ether as a solvent, and then the extract was evaporated and dried until it reached a constant weight. The ash content was determined by incineration at 550 °C using a muffle furnace.

Yield and Textural Profile Analysis (TPA)

The yield calculation was done by comparing the weight of raw material with the weight of minced meat before mixing the two kinds of minced meat. The distribution results were mixed and applied by 100% to get a percentage of the yield of surimi. The TPA was done following (Zhou et al., 2020; Zhou & Yang, 2019) method with a slight modification. The surimi gel was equilibrated at room temperature for 30 min prior to measurement. The conditions for TPA test were as follows: pre-test speed 2.00 mm/s; test speed 1.00 mm/s; post-test speed 2.00 mm/s; trigger force 10.0 g; deformation 40%. With spherical plunger 75 plunger (50 mm diameter) (TA. XT Plus; Stable Micro System, London, England).

Gel Strength

The prepared surimi gel was cut into a cylinder of φ 27 mm × 25 mm and equilibrated at room temperature for 30 min. The values of the breaking force (force/g) and deformation of the surimi gel were determined by a TA.XT Plus (Stable MicroSystem, London, England) equipped with a P/5s spherical plunger. The test conditions were as follows, pre-test speed: 1.00 mm/ s; test speed: 1.10 mm/s; speed after test: 10.00 mm/ s; trigger force 10.0 g; displacement: 15 mm (Zhou et al., 2020)

Water Holding Capacity (WHC)

The water-holding capacity (WHC) of gel samples was evaluated according to the method described by Dong et al. (2019) with slight modifications. Gel samples (3 g) were transferred to centrifuge tubes (80 mL) and then centrifuged at 8,000g for 30 min at 4 °C. The WHC was expressed as the final weight of the centrifuge as a percentage of weight before centrifugation.

Whiteness

Colour analysis was done on surimi gel by crushing it and placed on a special beaker glass for colour testing on the ColorFlex EZ tool (HunterLab, Virgina USA) with the calculation formula can be seen in Equation 1 of User's Manual Version 1.

Whiteness = 100 -
$$
\{(100 - L^2) + a *^2 + b *^2\}^{\frac{1}{2}}
$$
 (1)

Sodium Dodecyl sulfate - Polyacrylamide Gel Electrophoresis (SDS-PAGE) Analysis

The protein patterns of surimi gels were analyzed by SDS-PAGE according to the method of Cao et al. (2018). Preparing the protein sample, 27 ml of 5% (w/v) SDS solution heated to 85 \degree C was added to the sample (3 g). The mixture was then vortexed (MamimixPlus, ThermoFisher, Massachusetts, USA) for 2 min. The homogenate was incubated at 85 \degree C for 1 h to dissolve total proteins. The samples were centrifuged (K3 Series, Centurion Scientific Ltd, Indiana, USA) at 3,500 g for 20 min to remove undissolved debris. The protein concentration of the supernatant was determined by the Bradford method using bovine serum albumin as the standard. The samples (20 μ g protein) were loaded into the polyacrylamide gel made of 12% running gel and 5% stacking gel and subjected to electrophoresis at constant current. After separation, the proteins were stained with

0.02% (w/v) Coomassie Brilliant Blue R-250 in 50% (v/v) methanol and 7.5% (v/v) acetic acid.

Scanning Electron Microcopy (SEM) Analysis

The surimi gel samples were cut into small pieces (0.25 x 0.25 x 0.25 cm) and fixed with 2.5% glutaraldehyde in phosphate buffer saline (Oxoid, BR0014G) at pH 7.2 for 3 h at room temperature. The fixed samples were then rinsed twice with distilled water. The specimen then was dehydrated in graded ethanol solution with serial concentration of 50%, 70%, 90% and 100% for 15 min each (Chen et al., 2023; Nyaisaba et al., 2019). Subsequently, the desiccated samples underwent a drying process at ambient temperature for a duration of 48 hours. The dried samples were mounted on a bronze stub and sputtercoated with gold. The specimens were observed with a scanning electron microscope (Neoscope 6000, JEOL Ltd, Japan) at an acceleration voltage of 10 kV.

Microbial Analysis

Microbiological analysis was performed on surimi gel. As much as 20 g of surimi gel along with 80 ml of peptone water was crushed using a stomacher for 2 min. The dilution series were made, and 1 ml was taken from the dilution series and spread on the aerobic petrifilmTM. Petrifilm was then incubated for 48 h at 36±1°C (Dewi et al., 2021).

Statistical Analysis

Statistical analysis was used to see the significant rate between treatments expressed when $p<0.05$ in a one-way ANOVA analysis. Prism8 (GraphPad Prism software Version 8.0.2, CA USA) for the analysis.

Results and Discussion

Proximate Composition

Fresh fish meat primarily consists of water (65– 85%), protein (15–24%), fat (0.1–22%), carbohydrates (1-3%), and inorganics (0.8–2%) (Balange, 2009). The quantity of fish meat varies depending on the species, age, pre- or post-spawning season, the feeding conditions and body part (Suzuki, 1981). The proximate composition of mixed-species surimi is presented in Table 1. The statistical analysis of the moisture content showed in Table 1 indicated significantly different $(p<0.05)$ between the mixed-species surimi formulation (F2, F3, F4) and control (F1, F5), but ash, protein and fat content showed no significantly different $(p>0.05)$.

Water, protein, and lipids make up 98% of the total weight of fish muscle, making them the three primary components of fish. These elements are essential to the fish's functional qualities and nutritional value. The final 2% of the mince is made up of carbohydrates, vitamins, and minerals (Ofstad et al., 1996). Overall, the proximate composition of all blended surimi formulas closely resembled that of the raw materials (*Decapterus* sp and *Priacantus* sp) (Table 1). Nevertheless, the fat levels in blended surimi were reduced compared to the fat content of the raw material. The Bigeye snapper is categorized as lean fish because their fat content is less than 2 g/ 100 g mince. The fat level of mixed surimi was found to be higher compared to the findings of Chinabhark in 2007. They use *Priacanthus tayenus* as the raw material and the fat content was 0.04%. The moisture content of around 78.8% and a protein content of 16.7% (Dileep et al., 2012), in line with the result of Jaziri et al. (2022) that in their research found that bigeye snapper's contents

Note: $n = 3$, The superscripts label denotes significant difference across formulas

F1: *Decapteru*s sp : *Priacanthus* sp = 1:0

F2: *Decapteru*s sp : *Priacanthus* sp = 3:1

F3: *Decapteru*s sp : *Priacanthus* sp = 1:1

F4: *Decapteru*s sp : *Priacanthus* sp = 1:3

F5: *Decapteru*s sp : *Priacanthus* sp = 0:1

of moisture, protein, fat, ash and carbohydrate ranged from 56.22-79.26%, 12.46-31.14%, 0.24-1.29%, 1.27-22.86% and 0.42-0.98%, respectively. *Decapterus* sp, on the other side, has greater proportion of dark flesh than white flesh with moisture content 51.69-62.32%; ash content 0.07-1.28%; protein content 25.94-30.73%; fat content 1.37-1.82% (Hadinoto et al., 2017).

Textural Profile

Texture profile analysis (TPA) is a technique for evaluating food texture that stimulates two cycles of compression and decompression during mastication (Fang et al., 2021). TPA is an empirical method for analyzing the texture of surimi. TPA measures six parameters: hardness, springiness, cohesiveness, chewiness, gumminess, and resilience (Figure 1). These six parameters are important attributes to surimi gel (Dong et al., 2019). Figure 1 showed that the highest of all parameters value of mixed-surimi was produced by F4 *(p*<0.05). The *Decapterus* sp content in the surimi formula was indicated to reduce the TPA value. Instead, the *Priacanthus* sp increased the TPA value. *Decapterus* sp is a pelagic fish that contains dark muscle, as noted by Panpipat et al (2023) dark muscle is less effective for gelation compared to light muscle, primarily due to its higher content of sarcoplasmic proteins and lipids. Sarcoplasmic proteins negatively influenced the strength, deformability, and colour of gels made from fish myofibril proteins. The result showed that F4 is the best mixed-species formula for the raw material of surimi. Higher hardness and chewiness is attributed to the higher protein and lower moisture content (Massingue et al., 2021). Gel strength of the surimi is directly correlated with hardness (Tanuja et al., 2014). The higher value in hardness, cohesiveness, gumminess, and chewiness due to their higher water holding capacity and water retention capacity (Kim et al., 2018). However, compared to the control, the values of F4 were lower than F5 (100% *Priacanthus* sp)*.* This indicates that a greater concentration of *Priacanthus* sp resulted in a higher TPA value. It can find that *Priacanthus* sp had a significant effect on hardness, springiness, cohesiveness, gumminess, chewiness, and resilience $(P < 0.05)$. The TPA profile of mixed surimi exhibited greater values in comparison to the surimi produced by Liu et al. in 2014. This indicates that the mixed surimi has the potential to be implemented. The industry demands surimi with a high TPA value, and it has been verified that *Priacanthus* sp possesses this characteristic

(Julavittayanukul 2006). However, *Decapterus* sp is known as a pelagic fish species that require additional methods and treatment to enhance its gel quality (Li et al., 2022 & Yi et al., 2022). The high value of TPA indicates that the gel surimi formed has the ability not to be easily destroyed. The gel texture is affected by the type of fish used to prepare the surimi, the salt concentration used to solubilize the proteins, the heat treatment temperature and time, and the moisture content (Medina & Garrote, 2002). In this study, we used sucrose and sorbitol as cryoprotectants, according to Medina & Garrote, (2002) the sucrose and sorbitol mixtures displayed excellent behavior throughout frozen storage, particularly at 45 and 90 days of storage. Low TPA values can be improved by the addition of other materials as reported by Dong et al. (2019), Zhou et al. (2019) and Yi et al. (2022) added potato flour and egg white flour to improve the texture of surimi and proved successful. The additional materials to mixedspecies surimi may be considered to improve the surimi texture of mixed-species fish.

Gel Strength

Gel strength is one of the most important functional properties of surimi gel. Gel strength, which serves as the main determinant of surimi quality and price, is commonly used to describe the textural characteristics developed during gelation (Xiong et al., 2021). Figure 2 shows that the surimi of 100% *Priacanthus* (F5) has the highest gel strength and is very different *(p*<0.05) than other formulas. Mixed-species surimi with a comparison formula 1:3 $(F4)$ and 1:1 $(F3)$ has better strength than mixed-species surimi with a ratio of 3:1 (F2). During the thermal gelation process, disulfide bonds were discovered in the actomyosin of bigeye snappers (Benjakul et al., 2001).

The low strength of the gel in mixed-species surimi may be due to the presence of a mixture of *Decapteru*s sp known to be pelagic fish with low gel strength. The surimi gel strength depends on the fish species used. It is well known that dark muscle fish meat has a lower gel-forming capacity than ordinary muscle (Chen, 2002).The addition of egg white (Zhou et al., 2019), potato flour (Dong et al., 2019), lecithin (Zhou et al., 2020) soy protein isolates and transglutaminase (Zilda, 2014) can increase gel strength in surimi. Research on the use of additional materials above is feasible to see the extent to which the additional material can increase the strength of the gel in surimi with the raw materials of mixed-species fish.

- F4: *Decapteru*s sp : *Priacanthus* sp = 1:3
	- F5: *Decapteru*s sp : *Priacanthus* sp = 0:1

Figure 1. Box plots of textural profile of mixed-species surimi.

F1: *Decapteru*s sp : *Priacanthus* sp = 1:0 F2: *Decapteru*s sp : *Priacanthus* sp = 3:1 F3: *Decapteru*s sp : *Priacanthus* sp = 1:1 F4: *Decapteru*s sp : *Priacanthus* sp = 1:3 F5: *Decapteru*s sp : *Priacanthus* sp = 0:1 Figure 2. Box plots of gel strength of mixed-species surimi.

Water Holding Capacity (WHC)

Water Holding Capacity can be defined as a gel's capacity to combine with water or the ability of the protein to form a gel to retain water, and it is often based on interactions between proteins-water, water distribution and the structure of gel. The higher WHC values indicate that the surimi gel retains more water because there is less internal water pressure. The quality of surimi gels is tightly related with the WHC (Marín et al., 2018; Zhang et al., 2018). The statistical analysis of the WHC shown in figure 3 indicates no interactions $(p>0.05)$ between the mixed-species surimi formulation (F2, F3, F4) and control (F1, F5), but all samples show the low WHC. The F5 formula had the lower WHC value, however the value was not significantly different from other formulas. The low WHC samples show that the gel networks were weak, which reduced their capacity to hold water. More water is bound into a dense gel network, increasing its WHC, whereas a poor gel network results in a lower WHC (Tang et al., 2019). Singh and Benjakul (2018) reported that the worse WHC of the gel network was brought on by degradation of muscle proteins in modori gel. The different handling of raw material, frozen *Decapterus* and fresh *Priacanthus* in surimi formulation did not affect the WHC. The quality of the surimi can vary depending on the time and temperature between capture and processing. Fish that has been kept on ice for a long time will have a lower gel quality, because longer periods of frozen storage, water was more readily released from muscle tissue. However, it appears that different species exhibit varying rates of gel strength reduction (Benjakul et al., 2002). Some factors such as pH and ionic strength determine the formation of protein gel and WHC. The protein denaturation was reflected by the lower WHC of protein gel (Shaviklo, 2006). Kudre and Benjakul (2013) used trypsin inhibitor to improve water holding capacity and gel strength.

Whiteness

The quality of surimi gel was also determined by whiteness value. Whiteness is an important feature of gel that directly influences consumer preference and decision. Fish freshness, kind of raw material used and muscle colour affect the colour and whiteness of surimi (Benjakul et al., 2002; Shaviklo, 2006). The statistical analysis shown in figure 4 indicates there was no significant effect (*p*>0.05) on whiteness between the mixed-species surimi formulation (F2, F3, F4) and control (F1, F5). The whiteness of mixedspecies surimi formulations was in the range of 66- 68. The result was similar to Benjakul et al., (2002), who reported that the whiteness of surimi from

F1: *Decapteru*s sp : *Priacanthus* sp = 1:0 F2: *Decapteru*s sp : *Priacanthus* sp = 3:1 F3: *Decapteru*s sp : *Priacanthus* sp = 1:1 F4: *Decapteru*s sp : *Priacanthus* sp = 1:3 F5: *Decapteru*s sp : *Priacanthus* sp = 0:1 Figure 3. Box plots of gel WHC of mixed-species surimi.

Priacanthus was 67–79 and it decreased when the storage time increased.

In this study, the mixed-species surimi used the frozen *Decapteru*s in the formulation. The whiteness of mixed-species surimi could be affected by many factors, including raw material frozen storage, removal, washing and heating process. According to Benjakul et al. (2002) during frozen storage, fish muscle pigments such as metmyoglobin or methaemoglobin was oxidized to produce discolouration of muscle. The material treatment by removing the dark muscle of *Decapterus* before being processed to be surimi gel could affect the whiteness because if the dark muscle is not removed from the flesh, it will make the colour of minced fish darker. Washing improved the surimi's whiteness by removing most of the pigment, blood, and impurities (Hassan et al., 2017). A similar result of the heating effect on the whiteness of mixed-species surimi was also reported by Yi et al. (2020). Heating at a temperature of 90 °C for 15 minutes did not significantly affect the whiteness between mixedspecies formulas. The factors previously stated did not affect the whiteness between formulas of mixedspecies surimi because all formulations were processed under the same conditions, and there was no variation

F2: *Decapteru*s sp : *Priacanthus* sp = 3:1 F3: *Decapteru*s sp : *Priacanthus* sp = 1:1

F4: *Decapteru*s sp : *Priacanthus* sp = 1:3

F5: *Decapteru*s sp : *Priacanthus* sp = 0:1

Figure 4. Box plots of gel whiteness of mixed-species surimi.

in factors such as washing cycle and time, heating time, and temperature.

The mixed-species surimi in this research uses *Decapterus* sp., a pelagic fish that contains dark muscle. The filleting process to remove dark muscle from the flesh before it was minced was very crucial because it determined the whiteness of surimi. If part of the dark muscle is not removed from the flesh, it will produce darker minced fish, and finally, the surimi as a final product will also have a darker colour and a lower whiteness value. According to Figure 4, the whiteness value was not significantly different between the formulations, indicating that the filleting process was effective in removing the dark muscle from the *Decapterus* flesh.

Protein Pattern

The two major proteins in surimi paste were myosin heavy chain (MHC) and actin. SDS-PAGE protein patterns of mixed-species surimi are shown in Figure 5. MHC was not found in all sample's surimi. Kamaboko gel's decreased MHC band intensity was most likely caused by endogenous transglutaminasemediated polymerization. Despite the decreased proteolysis, it was shown that MHC was less preserved in kamaboko gel. Bigeye snapper surimi gel's decreased MHC band intensity was most likely caused by MHC polymerization, especially during setting at high temperature was implemented. However, kamaboko also revealed the presence of breakdown proteins, whose molecular weights (MW) range from 70 to 190 kDa. *Decapterus* sp is a dark-fleshed fish have been reported to have poorer gel properties because they contain high level protease, which induce the degradation of protein (Kudre & Benjakul, 2013).

Decreased MHC was linked to increased gel strength, which was accompanied by an increase in non-disulfide covalent cross-linking. Decreased MHC and lower solubility occurred simultaneously (Benjakul & Visessanguan, 2003). In comparison to other muscle proteins including actin, troponin, and tropomyosin, MHC was also more susceptible to proteolytic destruction. The removal of MHC in the modori gel together with the formation of new protein bands with MWs of around 110 to 120 kDa revealed that MHC had degraded during incubation at 60°C (Arfat & Benjakul, 2012).

Note: SDS page result for F4 and F5 was separated due to limited well in the agar to accommodate all samples at once.

- F1: Decapterus sp : Priacanthus sp = $1:0$
- F2: *Decapteru*s sp : *Priacanthus* sp = 3:1
- F3: *Decapteru*s sp : *Priacanthus* sp = 1:1
- F4: *Decapteru*s sp : *Priacanthus* sp = 1:3
- F5: *Decapteru*s sp : *Priacanthus* sp = 0:1

Figure 5. SDS-Page of surimi gels of mixed-species surimi

F1: *Decapteru*s sp : *Priacanthus* sp = 1:0 F2: *Decapteru*s sp : *Priacanthus* sp = 3:1 F3: *Decapteru*s sp : *Priacanthus* sp = 1:1 F4: *Decapteru*s sp : *Priacanthus* sp = 1:3 F5: *Decapteru*s sp : *Priacanthus* sp = 0:1

Figure 6. Scanning electron micrograph of mixed-species surimi

Microstructure of The Surimi Gel

The microstructure of mixed-species surimi is shown in Figure 6. Surimi gel F5 shows more compact and denser than the other samples. The kamaboko from surimi formulation using *Decapterus* sp F1, F2, F3, and F4 resulting three-dimensional protein structure with poorer gel property consisted of a large number of loose and porous structure. The larger voids or grooves in the coarser microstructure, which was consistent with the low WHC. Due to the slow heating rate and non-uniform mass transfer, boiling or heating of surimi typically resulted in the network pore size of

surimi gels being large and non-uniform (Zheng et al., 2022).

Surimi gel from Bigeye snapper was finer and denser than others and have high deformation and breaking force (Rawdkuen & Benjakul, 2008). Crosslinking between myofibrillar and sarcoplasmic protein resulted in polygonal structures of kamaboko gel (Jafarpour & Gorczyca, 2009). Improving kamaboko gel from dark fleshed fish*,* Kudre & Benjakul (2013) used trypsin inhibitor in the surimi formulation resulted kamaboko gel that was more compact and finer with smaller voids.

- F1: *Decapteru*s sp : *Priacanthus* sp = 1:0
- F2: *Decapteru*s sp : *Priacanthus* sp = 3:1
- F3: *Decapteru*s sp : *Priacanthus* sp = 1:1
- F4: *Decapteru*s sp : *Priacanthus* sp = 1:3

F5: *Decapteru*s sp : *Priacanthus* sp = 0:1

Figure 7. Box plots of total plate count of mixed-species surimi

Microbial Analysis

There were no significant differences ($P > 0.05$) with respect to TPC between the samples ($< 6 \times 10^5$) CFU/ml) (Figure 7). The maximum acceptable bacterial load for a suitable shelf life is 10⁷ CFU/g (Al-Bulushi et al., 2005). TPC is one of the microbiological parameters to determine the level of deterioration. The kind of fish as surimi materials did not affect the TPC. The initial microbial count of the samples significantly influenced by the cleanliness of the fish handling and surimi preparation (Moosavi-Nasab et al., 2019).

Conclusion

Formula of F4 with ratio of *Decapterus* sp. and *Priacanthus* sp (1:3) can improve the water binding (WHC), texture profile (TPA), protein pattern, and color of mixed surimi, while the microstructure (SEM) is quite delicate. However, mixed surimi with ratio of *Decapterus* sp. and *Priacanthu*s sp (1:3) still requires refinement in order to approach the gel strength of 100% *Priacanthus* sp. surimi. According to the results, the mixed-species surimi of *Decapterus* sp and *Priacanthus* sp indicates the potential formula to overcome the raw material scarcity caused by the government's regulation enforcement banning trawling. The results of this study will support the feasibility of utilizing *Decapterus* sp, a dark-fleshed pelagic fish species, for surimi production. This could address the shortage of raw materials and promote sustainability within the surimi industry. In the future, improvement methods to reduce the dark muscle of pelagic fish will be able to produce surimi of the same quality as light flesh surimi.

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References

- Al-Bulushi, I. M., Kasapis, S., Al-Oufi, H., & Al-Mamari, S. (2005). Evaluating the quality and storage stability of fish burgers during frozen storage. *Fisheries Science*, *71*(3), 648– 654. <https://doi.org/10.1111/j.1444-2906.2005.01011.x.>
- Association of Official Analytical Chemists (AOAC). (2000). Official Methods of Analysis of AOAC, International. *17th ed. AOAC International*, Gaithersburg, MD, USA 2000.
- Association of Official Analytical Chemists (AOAC). (2005). Official Methods of Analysis of AOACInternational.*18th ed. Association of Official Analytical Chemists*, Washington DC.
- Arfat, Y. A., & Benjakul, S. (2012). Gelling characteristics of surimi from yellow stripe trevally (Selaroides leptolepis). *International Aquatic Research*, *4*(1), 1–13.<https://doi.org/> 10.1186/2008-6970-4-5.
- Balange, K. A. (2009). Enhancement of gel strength of surimi using oxidized phenolic compounds [Doctor of Philosophy's thesis, Prince of Songkla University Copyri].
- Benjakul, S., & Visessanguan, W. (2003). Transglutaminasemediated setting in bigeye snapper Surimi. *Food Research International*, *36*(3), 253–266. <https://doi.org/10.1016/> S0963-9969(02)00167-9.
- Benjakul, S., Visessanguan, W., Ishizaki, S., & Tanaka, M. (2001). Differences in gelation characteristics of natural actomyosin from two species of bigeye snapper, *Priacanthus tayenus* and *Priacanthus macracanthus*. *Journal of Food Science*, *66*(9), 1311–1318. https://doi.org/10.1111/j.1365- 2621.2001.tb15207.x.
- Benjakul, S., Visessanguan, W., Riebroy, S., Ishizaki, S., & Tanaka, M. (2002). Gel-forming properties of surimi produced from bigeye snapper, *Priacanthus tayenus* and *P macracanthus*, stored in ice. *Journal of the Science of Food and Agriculture*, 82(13), 1442–1451. <https://doi.org/> 10.1002/jsfa.1207.
- Bhatta, B. U., Prabhu, R. M., Reddy, A. M., & Elavarasan, K. (2015). Biochemical Changes in Dressed *Priacanthus hamrur* (Bull's Eye) During Frozen Storage and Its Effect on

Physical and Sensory Quality of Fish Sausage. *International Journal of Food Properties*, 18:897–908. DOI: 10.1080/ 10942912.2013.837062.

- BPS-Statistics Indonesia. (2020). Statistic of fishing port 2020. BPS-Statistics Indonesia, Directorate of Livestock, *Fisheries and Forestry statistics.*
- Cao, H., Fan, D., Jiao, X., Huang, J., Zhao, J., Yan, B., Zhou, W., Zhang, W., & Zhang, H. (2018). Effects of microwave combined with conduction heating on surimi quality and morphology. *Journal of Food Engineering*, *228*, 1–11. https:/ /doi.org/10.1016/j.jfoodeng.2018.01.021.
- Chen, H. H. (2002). Decoloration and gel-forming ability of horse mackerel mince by air-flotation washing. *Journal of Food Science*, 67(8), 2970–2975. <https://doi.org/10.1111/> j.1365-2621.2002.tb08847.x.
- Chen, J., Fan, Y., Zhang, X., Yuan, Z., Zhang, H., Xu, X., Qi, J., Xiong, G., Mei, L., Zhu, Y., Yang, L., & Li, C. (2023). Effect of antifreeze protein on the quality and microstructure of frozen chicken breasts. *Food Chemistry*, 404(PA), 134555. <https://doi.org/10.1016/j.foodchem.2022.134555.>
- Dewi, F. R., Powell, S. M., & Stanley, R. A. (2021). The effects of pre-processing sanitation and modified atmosphere packaging on microbial growth in bulk packs of Atlantic salmon (Salmo salar) fillets. *IOP Conference Series: Earth and Environmental Science*, *733*(1).<https://doi.org/10.1088/> 1755-1315/733/1/012081
- Dileep, A. O., Shamasundar, B. A., Binsi, P. K., Badii, F., & Howell, N. K. (2011). Composition, physicochemical and rheological properties of fresh bigeye snapper fish (*Priacanthus hamrur*) mince. *Journal of Food Biochemistry*, 36(5), 577–586. https://doi.org/10.1111/j.1745- 4514.2011.00592.x.
- Dong, X., Huang, Y., Pan, Y., Wang, K., Prakash, S., & Zhu, B. (2019). Investigation of sweet potato starch as a structural enhancer for three-dimensional printing of S*comberomorus niphonius* surimi. *Journal of Texture Studies*, 50(4), 316– 324. <https://doi.org/10.1111/jtxs.12398.>
- Fatin, N. S., Huda, N., & David, W. (2015). Physicochemical properties of Japanese Scad (*Decapterus Maruadsi*) surimi prepared using the acid and alkaline solubilization methods. *International Journal of Scientific & Engineering Research*, Vol. 6 (4):141-147.
- Guenneugues, P., & Ianelli, J. (2013). Surimi resources and market. in J. W, Park, *Surimi and surimi seafood* (3th ed.) (pp. 25–53). CRC Press. <https://doi.org/10.1201/b16009-> 7.
- Hadinoto, S., & Kolanus, J. P. M. 2017. Evaluation of nutritional value and quality of round scad *(Decapterus sp)* presto with addition liquid smoke and yeast*. Majalah BIAM* 13 (01):22-30.
- Hikmayani, Y., Apriliani, T., & Adi, T. R. (2017). Alternatif solusi bagi keberlanjutan industri surimi di Indonesia. *Buletin Ilmiah Marina Sosial Ekonomi Kelautan dan Perikanan*, 3(1), 41. <https://doi.org/10.15578/marina.v3i1.6100.>
- Jafarpour, A., & Gorczyca, E. M. (2009). Characteristics of sarcoplasmic proteins and their interaction with surimi and kamaboko gel. *Journal of Food Science*, *74*(1), 16–22. https:/ /doi.org/10.1111/j.1750-3841.2008.01009.x.
- Jaziri, A. A., Hasanuddin, H., Shapawi, R., Mokhtar, R. A. M., Noordin, W. N. M., & Huda, N. (2022). Nutritional

composition and mineral analysis of the by-products from tropical marine fish, purple-spotted bigeye (*Priacanthus tayenus* Richardson, 1846) and barracuda (*Sphyraena obtusata Cuvier*, 1829). *OP Conf. Series: Earth and Environmental Science* 967. 012051. doi:10.1088/1755- 1315/967/1/012051.

- Kim, T. K.; Shim, J. Y.; Hwang, K. E.; Kim, Y. B.; Sung, J. M.; Paik, H. D.; Choi, Y. S. (2018). Effect of Hydrocolloids on the Quality of Restructured Hams with Duck Skin. *Poultr. Sci*, 97(12), 4442–4449. DOI: 10.3382/ps/pey309.
- Kudre, T. G., & Benjakul, S. (2013). Combining effect of microbial *transglutaminase* and bambara groundnut protein isolate on gel properties of surimi from sardine (*Sardinella albella*). *Food Biophysics,* 8(4), 240–249. <https://doi.org/> 10.1007/s11483-013-9292-5.
- Marín, D., Alemán, A., Sánchez-Faure, A., Montero, P., & Gómez-Guillén, M. C. (2018). Freeze-dried phosphatidylcholine liposomes encapsulating various antioxidant extracts from natural waste as functional ingredients in surimi gels. *Food Chemistry*, *245*, 525–535. <https://doi.org/10.1016/j.foodchem.2017.10.141.>
- Massingue, A. A., Paula, M. M. O., Rocha, A. P., Haddad, G. B. S., Carmo, E. L., Ramos, A. L. S., & Ramos, E. M. (2021). Texture profile of surimi-like material from mechanically deboned turkey meat. *Brazilian Journal of Food Technology*, 24. <https://doi.org/10.1590/1981-6723.34519.>
- Medina, J. R., & Garrote, R. L. (2002). The effect of two cryoprotectant mixtures on frozen surubi surimi. *Brazilian Journal of Chemical Engineering*, 19(4), 419–424. <https://> doi.org/10.1590/S0104-66322002000400010.
- Moosavi-Nasab, M., Asgari, F., & Oliyaei, N. (2019). Quality evaluation of surimi and fish nuggets from Queen fish (*Scomberoides commersonnianus)*. *Food Science and Nutrition*, 7(10), 3206–3215. <https://doi.org/10.1002/> fsn3.1172.
- Núñez-Flores, R., Cando, D., Borderías, A. J., & Moreno, H. M. (2018). Importance of salt and temperature in myosin polymerization during surimi gelation. *Food Chemistry*, 239, 1226–1234. https://doi.org/10.1016/ j.foodchem.2017.07.028.
- Nyaisaba, B. M., Hatab, S., Liu, X., Chen, Y., Chen, X., Miao, W., Chen, M., & Deng, S. (2019). Physicochemical changes of myofibrillar proteins of squid (*Argentinus ilex*) induced by hydroxyl radical generating system. *Food Chemistry*, 297(December 2018), 124941. <https://doi.org/10.1016/> j.foodchem.2019.06.008.
- Ohoiwutun, M. K., Tumiwa, B. B., Serpara, S. A., Nara, S. M., & Tuarita, M. Z. (2020). Investigation on quality of smoked Scad Mackerel (*Decapterus ruselli*) processed using different fuels. *IOP Conf. Series: Earth and Environmental Science* 517 (2020) 012023. doi:10.1088/1755-1315/517/1/012023.
- Pangsorn, S., Laong-manee, P., & Siriraksophon, S. (2007). Status of surimi industry in the Southeast Asia. *PG*-*1*-*17*, 1–28. http://repository.seafdec.or.th/handle/20.500.12067/ 611?show=full
- Panpipat, W., Thongkam, P., Boonmalee, S., Cavdar, H.K., & Chaijan, M. (2023). Surimi Production from Tropical Mackerel: A Simple Washing Strategy for Better Utilization of Dark-Fleshed Fish. *Resources*, 12 (126). <https://doi.org/> 10.3390/ resources12100126
- Park, J. (2020, December). The production of surimi and surimi seafood from tropical fish – a landscape view of the industry. <https://certificationandratings.org/wp-content/uploads/> 2023/04/Surimi-Landscape-Report-Final-2.pdf .
- Rawdkuen, S., & Benjakul, S. (2008). Whey protein concentrate: Autolysis inhibition and effects on the gel properties of surimi prepared from tropical fish. *Food Chemistry*, 106(3), 1077–1084.https://doi.org/10.1016/j.foodchem.2007. 07.028.
- Shaviklo, G. R. (2006). Quality assessment of fish protein isolates. Gholam Reza Shaviklo UNU - Fisheries Training Programme. *Fisheries (Bethesda)*.
- South East Asian Fisheries Development Center. (2022). Fisheries country profile: Indonesia (2022).
- Sihono, Purnomo, A. H., Wibowo, S., & Dewi, F. R. (2021). Current (2021) status of surimi industry in Indonesia and possible solutions: A review. *IOP Conference Series: Earth and Environmental Science*, 919(1).<https://doi.org/10.1088/> 1755-1315/919/1/012036
- Suzuki, T. (1981). Fish and Krill Protein: Processing Technology. (pp. 260), Applied science publishers, London.
- Suryaningrum, T. D., Irianto, H. E., & Ikasari, D. (2015). Characteristics of kamaboko from catfish (*Clarias Gariepinus*) surimi processed with carrot and beet root as filler and natural food colorants. *Squalen Bulletin of Marine and Fisheries Postharvest and Biotechnology,* 10(3), 99. <https://doi.org/10.15578/squalen.v10i3.169.>
- Tanuja. S., Viji, P., Zynudheen, A. A., Ninan, G., & Joshy, C. G. (2014). Composition, Textural Quality and Gel Strength of Surimi Prepared from Striped Catfish (*Pangasianodon hypophthalmus, Souvage*, 1878). *Fishery Technology* 51:1 - 6.
- Wang, J., Tang, J., Park, J. W., Rasco, B., Tang, Z., & Qu, Z. (2019). Thermal gelation of Pacific whiting surimi in microwave assisted pasteurization. *Journal of Food Engineering*, 258(February), 18–26. <https://doi.org/> 10.1016/j.jfoodeng.2019.04.001.
- Yi, S., Li, Q., Qiao, C., Zhang, C., Wang, W., Xu, Y., Mi, H., Li, X., & Li, J. (2020). Myofibrillar protein conformation enhance gel properties of mixed surimi gels with *Nemipterus virgatus* and *Hypophthalmichthys molitrix. Food Hydrocolloids*, 106: 1-11. https://doi.org/10.1016/ j.foodhyd.2020.105924
- Yin, T., & Park, J.W. (2023). Comprehensive review: byproducts from surimi production and better utilization. *Food Science and Biotechnology*, 32: 1957–1980. <https://doi.org/> 10.1007/s10068-023-01360-8.
- Zhang, L., Li, Q., Shi, J., Zhu, B., & Luo, Y. (2018). Changes in chemical interactions and gel properties of heat-induced surimi gels from silver carp (*Hypophthalmichthys molitrix*) fillets during setting and heating: Effects of different washing solutions. *Food Hydrocolloids*, *75*, 116–124.<https://doi.org/> 10.1016/j.foodhyd.2017.09.007
- Zhou, X., Chen, T., Lin, H., Chen, H., Liu, J., Lyu, F., & Ding, Y. (2019). Physicochemical properties and microstructure of surimi treated with egg white modified by tea polyphenols. *Food Hydrocolloids*, 90(October), 82–89. <https://doi.org/> 10.1016/j.foodhyd.2018.07.031
- Zhou, X., Lin, H., Zhu, S., Xu, X., Lyu, F., & Ding, Y. (2020). Textural, rheological and chemical properties of surimi nutritionally-enhanced with lecithin. *LWT*, 122(September 2019), 108984. <https://doi.org/10.1016/j.lwt.2019.108984>
- Zhou, Y., & Yang, H. (2019). Effects of calcium ion on gel properties and gelation of tilapia (*Oreochromis niloticus*) protein isolates processed with pH shift method*. Food Chemistry*, *277*: 327–335. https://doi.org/10.1016/ j.foodchem.2018.10.110
- Zilda, D. Z. (2014). Microbial *transglutaminase*: source, production and its role to improve surimi properties. *Squalen Bulletin of Marine and Fisheries Postharvest and Biotechnology*, 9(1):35. https://doi.org/10.15578/ squalen.v9i1.82.