Population Analysis of Bali Sardinella (*Sardinella lemuru*, Bleeker 1853) Landed in PPI Kedonganan using Length-Weight Data and Digital Analysis (ImageJ)

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Abstract

Sardinella lemuru, one of the important pelagic fish in Bali Strait waters, has been reported to be in the overfishing condition. Various studies have been conducted to understand its population condition. Length-weight data has been also widely used to understand the population dynamic of S. lemuru, although data collection generally using manual direct measurement methods, which has the disadvantages if used on the enormous number of samples. Therefore, this research aims to understand the significance of S. lemuru length measurement using digital image analysis (ImageJ) compared with the manual measurement. Moreover, this research also aims to understand the population condition of S. lemuru landed in PPI Kedonganan analyzed using its length-weight data. The result indicated no significance difference between length measurement using manual methods and digital image methods, thus conclude that digital image data can be used to measure the fish length and represent the accurate measurement. Population condition analysis showed that S. lemuru population is in isometric condition (b=3); with indication of declining in the length growth pattern compared with previous research ($L\infty$= 20.75 cm with K= 1.1); the mortality estimation showed that fishing mortality (F=1.27) was lower than natural mortality (M=2.22), with the survival rate higher on smaller size fish. This research result can be used as an addition to the S. lemuru fisheries data and can also be used as reference methods to help conduct sampling and data collection more efficiently.

Keywords: *Sardinella lemuru, digital image, ImageJ, length-weight, analysis*

Introduction

S. lemuru widely known as its local name, lemuru fish or "ikan kucing" is one of the fish from Clupeidae family. This fish is the main catch for the fisherman within the area of Bali Strait waters (Wujdi *et al*., 2013; Hendiari *et al*., 2020). As one of the important small pelagic fisheries commodity products, *S. lemuru* has its advantage compared to other small pelagic fish due to its cheap price, easy to catch, and has a high content of omega-3 nutrients. Due to the high demand of this fish commodity, the fisheries catch data of *S. lemuru* has indicated an increasing catch rate over the years within Bali Strait waters (Wujdi *et al*., 2013). This fisheries condition, if prolonged for a certain time will lead to the decreasing number of *S. lemuru* population. In the worst case, it will lead to extinction (Ambariyanto, 2017). This was supported by the fisheries data that showed the decreasing catch number of *S. lemuru* from 42 thousand tons in 2009, to 38 thousand tons in 2010 (Himelda *et al*., 2011). Various studies had been conducted to collect information that can help sustainable lemuru fisheries management in Indonesia, including research on fisheries aspect (Listiyani *et al*., 2017), biology (Wujdi *et al*., 2013; Sartimbul *et al*., 2018), ecology (Pertami *et al*., 2019; Wijaya *et al*., 2021), and genetic (Hendiari *et al*., 2020).

Information of the population condition of fish species can be understand using fishery stock analysis (Rago, 2005), fish length-weight and growth data (Famoofo and Abdul, 2020; Indarjo *et al*., 2020; Olopade *et al*., 2020; Annisa *et al*., 2021), and genetic data (Pertiwi *et al*., 2017; Sartimbul *et al*., 2018; Malik *et al*., 2020). Length-weight data has been also widely used to understand the population

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dynamic of *S. lemuru* (Wujdi *et al*., 2012; Pertami *et al*., 2018; Annisa *et al*., 2021), especially within Bali Strait waters. The collection of length-weight data wascarried out using direct measurement methods. Generally, data collection of fish length usually conducted using contact method or non-contact method. (Mackvandi *et al*., 2015). Contact methods usually done manually through survey and field sampling. However, this method considered ineffective for sample with an enormous number, due to the high cost and time consuming. Meanwhile, the non-contact method usually conducted with the help of technology and digital image analysis. Fish morphology and biometry measurement analysis using digital image can provide the objective, fast, easy, and cost effective (Islamadine *et al*., 2018). Several studies has been conducted on the fish measurement using non-contact methods, with the help of program for digital image analysis, including fish measurement using ImageJ (Andrialovanirina *et al*., 2020); IMAFISH_ML: automatic software for digital analysis of morphometric of aquaculture fish (Navarro *et al*., 2015); biometric measurement of fish inside aquarium (Mackvandi *et al*., 2015); measurement using FileDI framework (Shafry *et al*., 2011); fish measurement using gray scale images (Islamadina *et al*., 2018), and many other various digital image analysis programs.

Due to the ineffectiveness of the manual measurement, in terms of time and budget, especially for the high number of samples, therefore, this research aims to understand the significance of *S. lemuru* length measurement using one of the digital image analysis programs known as ImageJ (Schenider *et al*., 2012) compared with the manual measurement. Moreover, this research also aims to understand the population condition of *S. lemuru* landed in PPI Kedonganan analyzed using its length-weight data. This research is expected to be an initial study for the use of digital image to measure *S. lemuru* length. Furthermore, it expected to help to make the sampling activity more effective and easier to do; and can be used to measure other species of fish. The result of this study can also act as the additional information on the weight-length structure and population condition of *S. lemuru* in Bali Strait waters.

Materials and Methods

Bali Sardinella (*S. lemuru*) samples were collected from Pusat Pendaratan Ikan (PPI) Kedonganan, Bali (Figure 1.). Samples were collected for 4 months during May–August 2022, and conducted through six times of sampling periods. The numbers of samples collected in each sampling were not equal but based on the similar weight (around 1-2 kg samples on each sampling). The samples collected within the range of this weight because it already represents the minimum individual number of samples needed for population study (minimum of 20 individual). Interview with the fishermen were done, to ensure the fish were caught around Bali Strait waters.

Collected samples were identified using genetic analysis to ensure the sample is the species of *S. lemuru*. Genetic identification was conducted using six (6) samples, which were the proxy of each sampling period. The DNA sample was extracted and then amplified using polymerase chain reaction (PCR) methods at the control region mitochondrial DNA loci, using the primer CRK and CRE (Lee *et al*., 1995). Phylogenetic analysis was then used to analyze the sequence data.

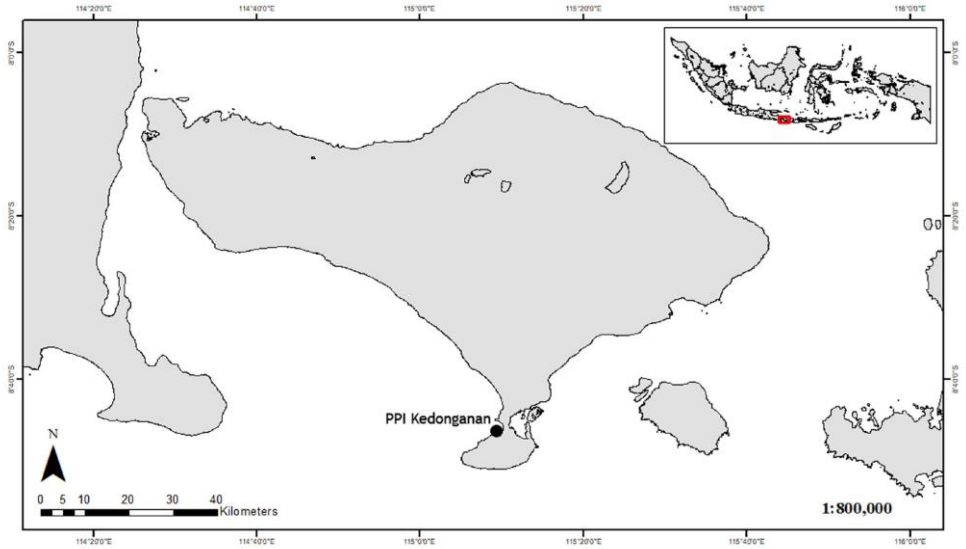

Figure 1. Bali Sardinella' sampling location at the southern part of Bali (PPI Kedonganan) indicated with the black dot.

Length and weight data were measured using manual and digital image. Manual measurement was done using a 1 mm scale ruler and 0.1 mm of weight scales. The data measured were total length (TL), standard length (SL), fork length (FL) in centimeter (cm), and weight in gram (g). Meanwhile, for measurement using digital image, each of the individual sample was photographed using digital camera on the lateral side. The digital image of each sample was the analyzed using ImageJ program (Schneider *et al*., 2012) to measure the TL, SL, and FL length. The sample's image and illustration of the length measurement was shown in Figure 2.

Length-weight relationship was analyzed using the FL data, using the analysis in Microsoft Excel. The length-weight relationship was estimated using the equation as given by Ricker, 1970: $W=aL^b$. Where "W" is body weight (gram), "L" is length; "a" is intercept and "b" is slope of regression line (Tesch, 1971). Logarithmic forms of the Tesch equations are as follows: log W = log a + b log L. Where log "L" is the independent and log "W" is dependent variables. The coefficient of correlation (r) was estimated to determine the quality of linear regression. The t-test analysis was carried out to calculate the value of "b". ELEFAN methods used to estimate the growth length parameters of asymptotic length ($L\infty$) and kurvatur parameter (K). Total mortality (Z), natural mortality (M), catch mortality (F) and exploitation rate (E) was also analyzed using ELEFAN methods.

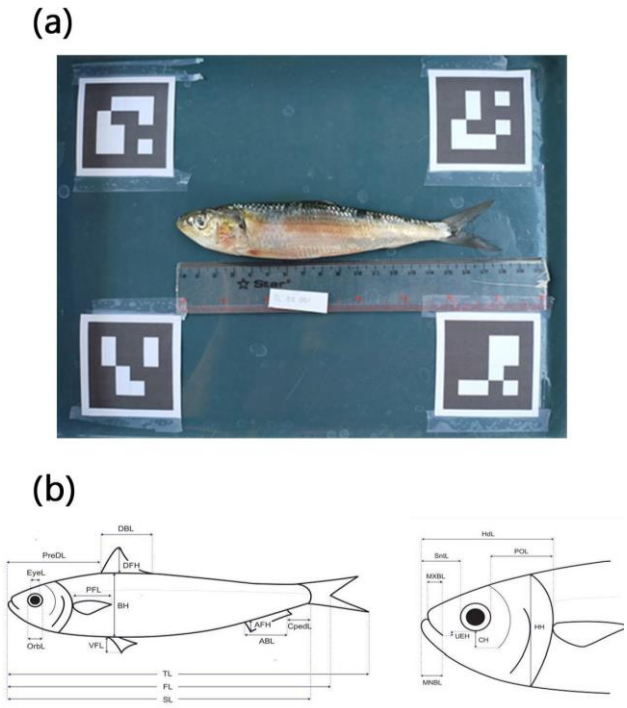

Figure 2. (a) One of the *S. lemuru* sample, (b) Illustration of length measurement on *S. lemuru* according to Sartimbul *et al*. (2018).

Mortality rate estimation was also analyzed using Virtual Population Analysis (VPA). Both ELEFAN and VPA were conducted using FISAT II Program (Gayanillo *et al*., 2005). Meanwhile, comparison analysis between two different measurement methods, i.e., manual and digital image (ImageJ) was done using ANOVA (Analysis of Variance) two-way with replication using Microsoft Excel.

Result and Discussion

The total of 339 individual samples of *S. lemuru* were collected from PPI Kedonganan during six times sampling period of May – August 2022. Genetic identification confirms the samples as the species of *S. lemuru* (unpublished data). Interview with the fisherman also confirm that the Bali Sardinella sold at the PPI Kedonganan was caught at the Bali Strait waters. The samples were collected based on the similar weight total (around 1 to 2 kg for each sampling period), thus made the difference in the number of individual samples collected on each sampling period. Based on the description of the length and weight measurement, the total length (TL) of the overall samples range between 7.3–21 cm with the average of 13.78 cm; while the weight range between 3–64 gram with the average of 24.04 gram.

In this research, population condition analysis was based on the fish length-weight relationship; length growth patter estimated using asymptotic length ($L\infty$) and growth coefficient (K) per year; and mortality estimation analysis.

Length-weight relationship

Length-weight relationship was calculated using the fork length (FL) data. This type of length was choose in this study, because total length (TL) and fork length (FL) are usually utilised in studies of fish growth, whereas standard length (SL) is mainly used in systematic studies (Arslan *et al*., 2004). In several cases, the fish tail was sometime broken, thus the total length measurement cannot be performed. Therefore, the fork length (FL) data can also be used to estimate the total length (TL) of Bali sardinella using the equation mentioned in the length-length relationship study of Bali sardinella by Pertami *et al*. (2018).

There were three possible values of "b" that appear in the calculation of the length-weight relationship of fish, which is b<3, b=3, and b>3. The value of b=3 showed an isometric growth, which means a balance condition between the length and weight of the fish. Meanwhile, the value of b<3 showed negative allometric, which is the conditions where the growth on the weight is slower than the

length (the fish is bony). On the contrary, the value of b>3 showed positive allometric, which is the conditions where the growth of the weight is faster than the length (the fish is stout) (Effendie, 2002). The result of the length-weight relationship showed the "b" value of 3 (b=3), which indicated that the *S. lemuru* population was in the isometric growth condition, where it appears that the length and the weight of the fish is in the balance condition. Moreover, determination coefficient showed the number of 0.984. This value indicated that 98% of the weight gains of the samples; during the 4 months sampling periods (May–August 2022) was influenced by the length of the fish. This result is inline with the research conducted by Setyohadi (2010), which mentioned that isometric growth was found in male and female *S. lemuru* from Bali Strait waters. However, different result showed by Annisa *et al*. (2021) in their research on *S. lemuru* landed in PPN Pengambengan-Bali, and Wujdi *et al*. (2012) from Bali Strait waters that indicated the length-weight relationship of *S. lemuru* is in the positive allometric condition (b>3).

The difference in the fish growth pattern can be affected by several factors, including gonad maturity stage, spawning condition, food, sex, and age (Radongkir *et al.*, 2018). Aprilianty (2000) also mentioned that the difference in the value of "b" can occur due to the influence of time, place, and environmental conditions during research or sampling period. This is inline with the time difference between the present study, which was conducted during May until August; while the research conducted by Annisa *et al*. (2021) was conducted on October to December.

Length growth pattern

Length growth pattern was analyzed using ELEFAN model in FISAT II Program. The result of this analysis showed the asymptotic length ($L\infty$) of *S. lemuru* is in the value of 20.75 cm, with the growth coefficient (K) per year reached 1.1. Asymptotic length is the estimation of theoretical maximum length that the fish can reach, while the growth coefficient is the value that showed the growth rate of the fish until it reaches the asymptotic stage. Regarding similar research conducting on *S. lemuru*, it is known that asymptotic length of *S. lemuru* from the research conducted by Wudianto *et al*. (2002) showed the value of 20.99 cm with the growth coefficient of 1.23 per year. Meanwhile, other similar research conducted by Wujdi *et al*. (2012) indicated the declining in the asymptotic length with the value of 20.75 cm, with the growth coefficient of 1.2 per year. Compared to the result of this project, it is showed a declining pattern in the growth of *S. lemuru* over the year. Wujdi *et al*. (2012) also mentioned that *S. lemuru* caught within Bali Strait waters indicated overfishing. Declining in the fish length size can also be the results of overfishing.

Mortality estimation analysis

Mortality estimation analysis showed the total mortality value (Z) of 3.38. After incorporating the average sea temperature information during May – August 2022 in Bali Strait waters (28.2^{o}C) (based on https://seatemperature.info) into the analysis, the natural mortality (M) estimation indicated the value of 2.22; while the fishing mortality (F) estimation indicated the value of 1.27. This result showed that the fishing mortality (F) is lower than the natural mortality (M). Different result showed by Setyohadi *et al*. (2010) and Wujdi *et al*. (2012) in their research on *S. lemuru* in Bali Strait waters, which indicated the higher fishing mortality (F) compared to natural mortality (M). The difference in result indicated that sampling duration and the period of sampling conducted might affect the mortality value, which was in this research, the sampling was only done for 4 months during May until August. Previous study also mentioned that *S. lemuru* in Bali Strait waters has undergone two recruitment patterns within one year, which was in February and July (Wujdi *et al*., 2012). Therefore, it is potentially that the sampling period within this research, was only represent the recruitment pattern, and not represent the data of the non-recruitment phase of *S. lemuru* within Bali Strait waters.

Furthermore, the normal distribution pattern of *S. lemuru* according to the temperature in Bali Strait waters is within the range of 25-31^{o}C (Setyohadi, 2012). According to Wudianto (2001), it is mentioned that the water temperature does not have much effect on the abundance of *S. lemuru*, but rather the affected of chlorophyll-a content to the *S. lemuru* food distribution pattern. For this reason, it can be stated that the natural mortality value (M) generated from FISAT II with the sea temperature data of 4 months incorporated into the analysis, may not represent the actual mortality of *S. lemuru*.

Mortality estimation analysis was also conducted based on the length structure using Virtual Population Analysis (VPA). Based on the VPA for four months sampling periods, the result showed that *S. lemuru* with the smaller size would have higher chance of survival compare to the *S. lemuru* with bigger size. Moreover, for the fish with the size > 15.8 cm has higher mortality rate. Although *S. lemuru* in Bali Strait waters has undergone overfishing stage (Wujdi *et al*., 2012), according to this result, it could indicate that the survival rate of smaller size *S. lemuru* is higher, which can give the opportunity for small size *S. lemuru* to breed or spawn before they

were caught by fishing activities. Despite this possibility, the result could also be because the sampling period was in the recruitment phase, thus affect the size of the *S. lemuru* caught, which was still in the stage of smaller size fish.

Comparison of manual and digital image (ImageJ) measurement

Comparison between the two methods, using manual and digital image (ImageJ) measurement was done using the length data. The length data (interaction of SL, interaction FL, and interaction TL) was analyzed using ANOVA two-way with replication and showed the P-value for all length data interaction are above 0.99. This result indicated that there is no significant difference between the length measurement using both methods, with the p-value >0.05. From this result, it can be inferred that there is no different measurement between the two methods, therefore the digital image using ImageJ program can be used accurately to measure the fish length. This program is one of the open-source software for processing and analyzing scientific images, and can be used to display, annotate, edit, calibrate, measure, analyze, process, print and save raster of image data (Schneider *et al*., 2012). According to this result, digital image can be used to help measure the length of fish, effectively in terms of time and budget, especially for sampling activity with an enormous number of samples that needs to be collected.

Conclusion

There is no significance difference between length measurement using manual methods and digital image methods, thus conclude that digital image data can be used to measure the fish length and represent the accurate measurement. Population condition analysis showed that *Sardinella lemuru* population is in isometric condition (b=3); with indication of declining in the length growth pattern compared with previous research ($L\infty$= 20.75 cm with K= 1.1); and the mortality estimation showed that fishing mortality (F=1.27) was lower than natural mortality (M=2.22), with the survival rate higher on smaller size fish which based on Virtual Population Analysis for 4 months. The result of this study can be used as the recent update on *S. lemuru* conditions in Bali strait, while also provide an efficient reference method to conduct sampling and data collection in fisheries study.

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