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## FLUCTUATING ASYMMETRY AND LENGTH-WEIGHT RELATIONSHIP OF THE THREE POPULATIONS OF *SARDINELLA LEMURU* (CLUPEIFORMES, DOROSOMATIDAE) FROM SURIGAO DEL NORTE, PHILIPPINES

J. G. Cinco, J. Farma, J. D. Silvosa, C. B. Weling & J. T. Cuadrado\*

College of Arts and Sciences, Surigao del Norte State University,  
Narciso St., Surigao City, 8400 Caraga, Philippines

\*Corresponding author

E-mail: jerrycuadrado90@gmail.com

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**Fluctuating Asymmetry and Length-Weight Relationship of the Three Populations of *Sardinella lemuru* (Clupeiformes, Dorosomatidae) from Surigao del Norte, Philippines.** Cinco, J. G., Farma, J., Silvosa, J. D., Weling, C. B. & Cuadrado, J. T. — Despite its economic importance and current conservation status, studies on *Sardinella lemuru* (Bleeker, 1853) are very scarce in the province of Surigao del Norte, Philippines, so this study was conducted to assess the body shape and health status of fish populations in the area using fluctuating asymmetry (FA) and length-weight ratio (LWR) analyses. 258 individuals were collected from the coastal waters of Malimono ( $n = 6$ ), San Francisco ( $n = 101$ ) and Surigao (100) from August to October 2021 using fishnets and gillnets. Among the three populations, fish samples from Malimono have the highest cumulative variation of 63.07 % followed by San Francisco and Surigao City (60.85 % and 55.51 %). The high percentage of FA among the three populations (57.66 % in Malimono; 54.43 % in San Francisco; and 49.78 % in Surigao City) also suggests that individuals had deformities particularly, especially in the fins and head region of the fish. The LWR of the fish samples ( $b = 2.6398$  in Malimono;  $b = 2.7541$  in San Francisco; and  $b = 2.8377$  in Surigao City) indicates a negative allometric growth pattern, suggesting that the fish samples do not grow symmetrically or become thinner with increasing length. The results of FA and LWR analyses suggest morphological abnormalities in the fish samples that may be attributed to the different environmental stressors in the area. Studies on sex differences, water quality and fish ecology are suggested to better understand the current status of fish in the area.

**Key words:** Environmental stressors, fins, head region, negative allometric growth pattern.

## Introduction

Marine pollution and coastal degradation have become serious threats in the world affecting marine life and the ecosystem (Kylili et al., 2021). This pollution and degradation weaken or destroy the normal ecosystem which supports human health, the production of food, and biodiversity (Tare & Bhojwani, 2012). Fishes are among the resources provided by the ocean that are not exempt from the harm brought by these anthropogenic activities (Davies & Baum, 2012).

On the other hand, *Sardinella lemuru* (Bleeker, 1853) is a native fish species widely distributed in the Philippines and contributes significantly to industrial sardine fisheries (Willette et al., 2013; Santos, 2018). However, this sardine species was listed as Vulnerable by IUCN (2018). Overfishing and increased use of purse seine are among the threats to this sardine species (Giducos et al., 2015). The anthropogenic activities, as well as the excessive use of fish resources, will have an adverse impact on the ecosystem and these impacts can cause changes in abundance, productivity, community structure, and even changes in species size spectra, especially during sensitive developmental stage (Pertami et al., 2018).

Fluctuating Asymmetry (FA) is used to determine the symmetrical and asymmetrical differences of individuals and is identified to be an efficient biomarker of environmental condition and stress, and developmental instability (Lecera et al., 2015; Beasley et al., 2013; Arreglado et al., 2013). In addition, the length-weight relationship (LWR) of fish is another tool used to assess the health and physiological conditions influenced by environmental conditions (Blackweel et al., 2000; Saygin et al., 2016). Surigao del Norte is one of the coastal provinces of the Philippines which is known to be rich in biodiversity and considered a natural fishing ground. However, the coastal areas and waters of this province are exposed to many human activities, population growth, water pollution, and pollutants from households. At present, published studies assessing the condition of *Sardinella lemuru* in the area are very scarce. Thus, this study was conducted to investigate the body shape differences and health of this sardine species using FA and LWR which could be a basis to assess the condition of the fish in the area.

## Material and Methods

### Fish Sample Collection

Surigao del Norte is located along the Northeastern (Caraga Region) coast of Mindanao with a total population of 485,088 (PSA, 2021). The province is composed of mainland Surigao, the major islands of Siargao, and other small adjacent islands. The human populations of this province rely mainly on agricul-

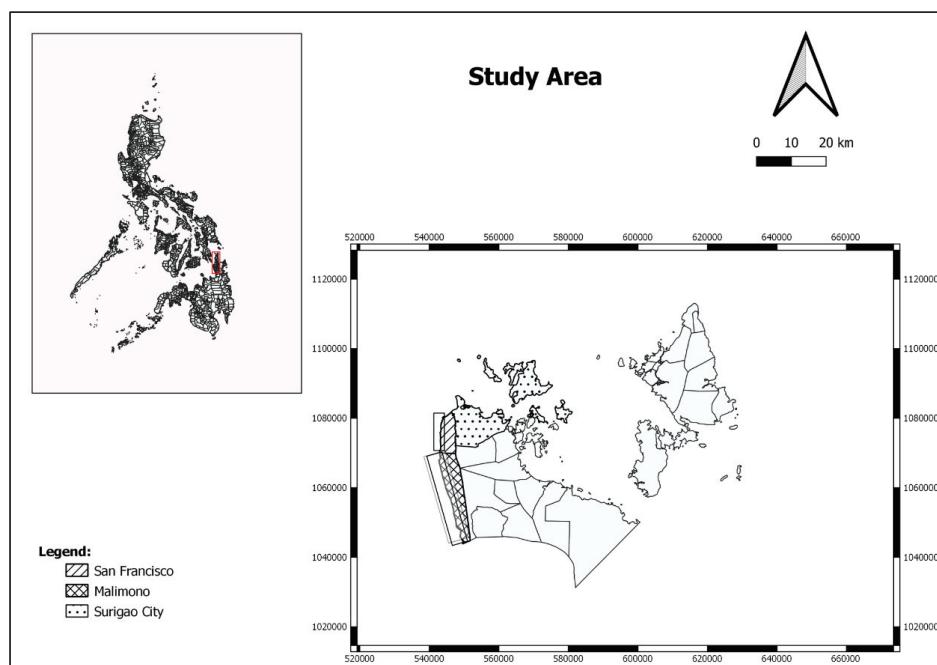


Fig. 1. The Philippine map showing the location of Caraga Region in Mindanao Island and the three sampling areas in the province of Surigao del Norte. Map credited to Engr. Medielyn M. Odtojan.

**Table 1.** Description of the landmark points according to (Luceño et al., 2014)

Coordinates	Location
1	Anterior tip of snout at upper jaw
2	Most posterior aspect of neurocranium (beginning of scales nape
3	Origin of dorsal fin
4	Insertion of dorsal fin
5	Anterior attachment of dorsal membrane from caudal fin
6	Posterior end of vertebrae column
7	Anterior attachment of ventral membrane from caudal fin
8	Insertion of anal fin
9	Origin of anal fin
10	Insertion of pelvic fin
11	Origin of pectoral fin
12–16	Contour of the gill cover
17	Posterior most portion of maxillary
18	Center of the eye

ture and fisheries. The fish samples were collected in the coastal waters of Surigao City and the municipalities of San Francisco and Malimono (fig. 1) using fishnets or gill nets. The collection of fish samples was done twice a month for three months starting August to October 2021. A total of 258 mature individuals (100 from Surigao City; 101 from San Francisco; and 86 from Malimono) were collected for the entire area. Fish samples collected were placed in a box filled with iced water and brought to the laboratory for further processing.

#### Measurement of Length and Weight

Fish samples were blot-dried using a paper towel before the length and weight were obtained. The length of individuals was taken with a 0.1 cm accuracy level using a vernier caliper (Natividad et al., 2015). The weight of individuals was measured using a digital weighing scale with 0.1g precision.

#### Digital Image Preparation, Landmark Selection, and Digitization

After the length and weight of individuals were obtained, the fish samples were placed in a flat Styrofoam for pinning of fins and tail to make it wider and to clearly observe the sample site of origin. Using a tiny brush, 10 % formalin was applied to the entire fish sample to harden it. The left and right portions of the fish were photographed three times using a mobile camera. A total of 18 landmarks were digitized using the tpsDig2 program version 2.0 (Rohlf, 2004) and saved as a tps file. The location of the landmarks and the anatomical descriptions was adapted from Luceño et al. (2014) and presented in table 1.

The generated data coordinates serve as the baseline data in analyzing the FA of the fish. The left and right landmark coordinates of the TPS were processed in Symmetry and Asymmetry in Geometric Data (SAGE) software to get the principal components that imply the deformation grid of the individual asymmetry (Natividad et al., 2015). SAGE is useful in shape conformation of individuals variation (asymmetric, symmetric, and error including the probable of co-variance condition).

#### Fluctuating Asymmetry and Length-Weight Relationship Analyses

The overall and localized fluctuating asymmetries were determined by subjecting the paired landmark coordinates to Procrustes ANOVA (Klingenberg & McIntyre, 1998) using the SAGE software version 1.0 (Rohlf, 2004). The Principal Component Analysis (PCA) was used to compare variability among individual symmetry and to determine the affected landmarks using symmetry and asymmetry scores. In addition, to clearly visualize this variation, a histogram was generated using PAST (Paleontological Statistics) software (Hammer et al., 2001).

The length-weight equation  $W = aL^b$  (Pauly, 1984) was used to estimate the relationship between the weight (g) of the fish and its total length (cm). Using the linear regression of the log-transformed equation:  $\log(W) = \log(a) + b \log(L)$ , the parameters  $a$  and  $b$  were calculated with ' $a$ ' representing the intercept and ' $b$ ' the slope of the relationship. Values of the exponent  $b$  provide information on fish growth. When  $b = 3$ , the increase in weight is isometric. When the value of  $b$  is other than 3, the weight increase is allometric; positive allometric if  $b > 3$  and negative allometric if  $b < 3$  (Levent et al., 2007).

## Results and Discussion

In this study, the results for Procrustes ANOVA for the shape variation of *S. lemuru* from the three locations; Malimono, San Francisco, and Surigao City, Philippines are shown in table 2. The left and right sides were analyzed to compare the fluctuating asymmetry. Three factors were analyzed for FA including the individuals, sides, and individuals x sides. The resulting factors show high significance ( $p < 0.0001$ ) in Malimono and Surigao City, the same with the sides and interaction of individuals and sides in San Francisco. However, the results in individual symmetry of the latter showed no significant differences ( $p < 0.3205$ ). The asymmetry in the morphology of the three factors of *S. lemuru* from the three locations may indicate that the individuals in the study area are exposed to environmental stress as Natividad et al. (2015) stated that fishes showing morphological changes are caused by high exposure to environmental stressors.

**Table 2.** Procrustes ANOVA Result on the body shape of *S. lemuru*

Factors	SS	DF	MS	F	P-value
Malimono					
Individuals	0.3956	2720	0.0001	1.2567	< 0.0001 **
Sides	0.0167	32	0.0005	4.5007	< 0.0001 **
Individual x Sides	0.3148	2720	0.0001	14.6808	< 0.0001 **
Measurement Error	0.0868	11 008	0	–	–
San Francisco					
Individuals	0.4102	2720	0.0002	1.018	< 0.3205 <sup>ns</sup>
Sides	0.0206	32	0.0006	4.3512	< 0.0001**
Individual x Sides	0.4029	2720	0.0001	13.3342	< 0.0001**
Measurement Error	0.1223	11 008	0	–	–
Surigao City					
Individuals	0.3999	2720	0.0001	1.166	< 0.0001**
Sides	0.033	32	0.001	8.1734	< 0.0001**
Individual x Sides	0.343	2720	0.0001	18.2707	< 0.0001**
Measurement Error	0.076	11 008	0	–	–

\*\* Highly significant.

**Table 3.** Principal component analysis showing the values of symmetry and asymmetry scores in *S. lemuru* with the summary of the affected landmarks from three location

PCA	Individual (Symmetry), %	Sides (Directional Asymmetry), %	Interaction (Fluctuating Asymmetry), %	Affected Landmarks
Malimono				
PC1	36.11	100	38.28	1,2,3,4,5,6,7,11,12,13,14,15,17,18
PC2	26.96		19.38 %	1,3,4,5,6,7,9,10,11,13,14,15,16,17,18
	63.07		57.66	
San Francisco				
PC1	37.26	100	36.53	1,2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19
PC2	23.59		17.90	1,3,4,5,6,7,9,10,11,13,14,15,16,17,18
	60.85		54.43	
Surigao City				
PC1	32.97	100	29.24	1,2,3,4,5,6,11,12,13,14,15,17,18
PC2	22.54		20.54	1,3,4,5,6,7,8,9,10,11,13,14,15,16,17,18
	55.51		49.78	

There were two principal components (PC) considered per sampling locations (table 3). The skewness of the histogram was reflected in every PC score along with the deformation grid to determine affected landmarks of the fish collected from the three locations (figs 2–4). Among the three populations, fish samples from Malimono have the highest cumulative variation of 63.07 % wherein PC 1 has the highest variation accounted (36.11 %). On the other hand, the fish samples in San Francisco have a cumulative variation of 60.85 % while samples collected from Surigao City have a cumulative variation of 55.51 %. In both locations, PC 1 still has the highest variation accounted for (37.26 % and 32.97 %). The percentage of overall variation exhibited by PC1 and PC2 of the three locations with Malimono as the highest suggest that the population of *S. lemuru* in this area un-

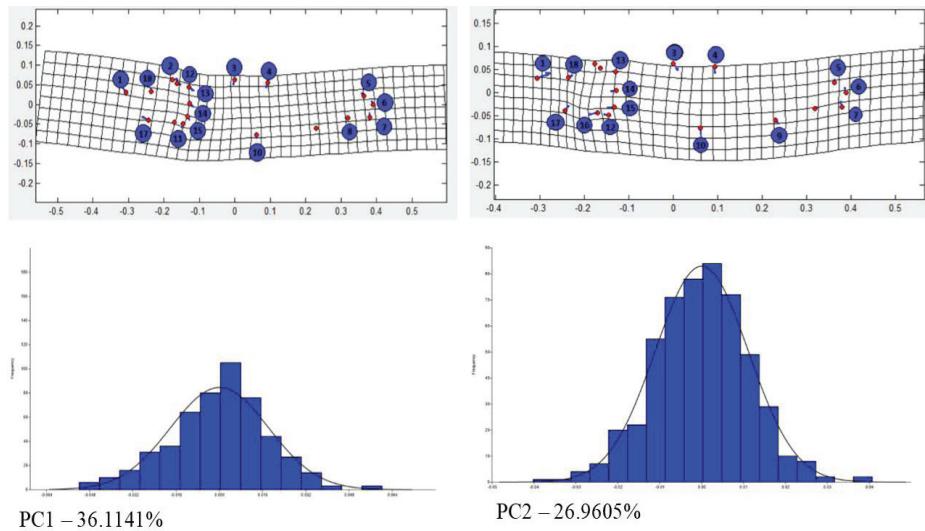


Fig. 2. Principal components (PC) inferred deformation grid and histogram of individual (symmetry) in Malimono, Surigao del Norte.

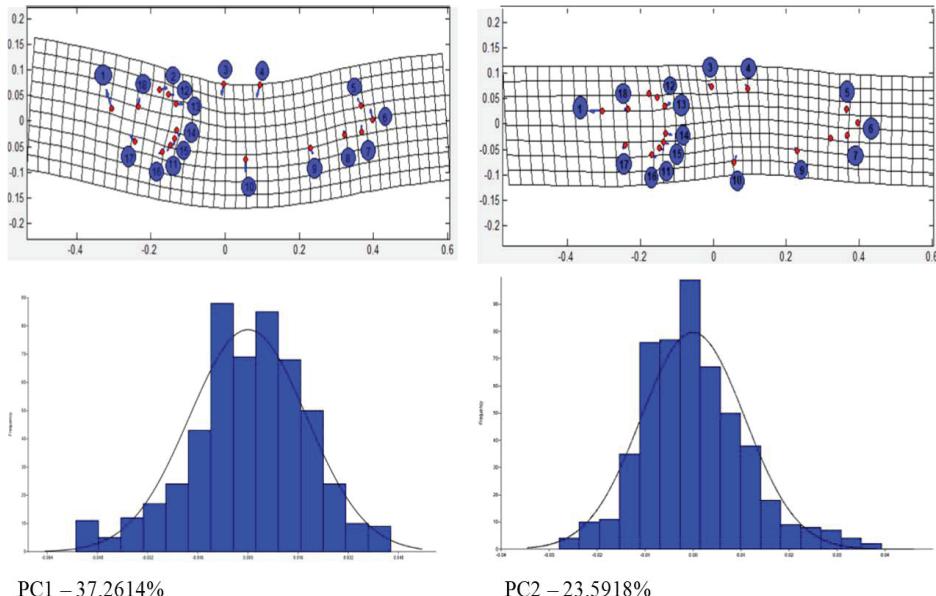


Fig. 3. Principal components (PC) inferred deformation grid and histogram of individual (symmetry) in San Francisco, Surigao del Norte.

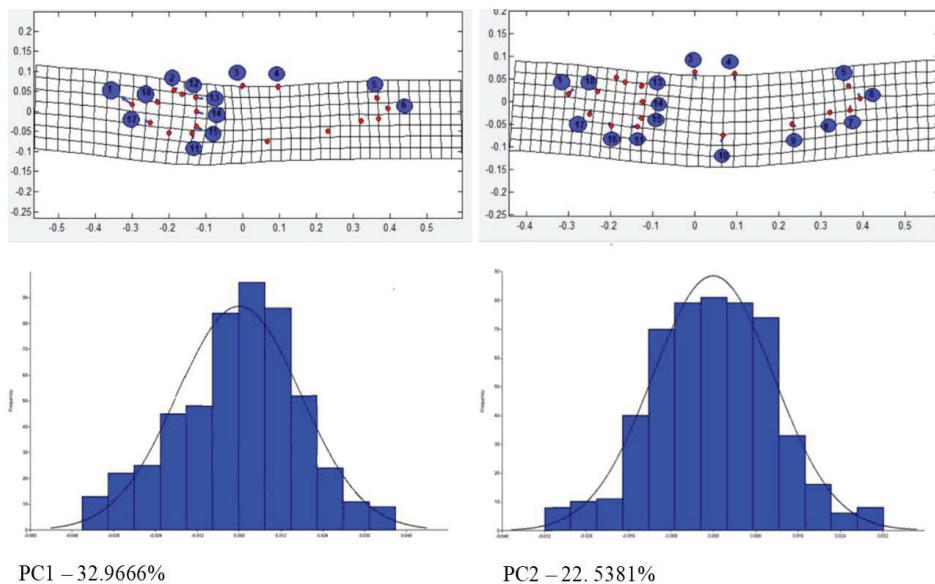


Fig. 4. Principal components (PC) inferred deformation grid and histogram of individual (symmetry) in Surigao City, Surigao del Norte.

dergoes developmental instability and might be due to unstable environmental condition and anthropogenic activities such as pollution and exploitation.

Figures 3, 4, and 5 show the actual digitized image of fishes showing the affected landmarks together with the PCA deformation grid which displayed the asymmetrical shape of *S. Lemuru*. Generally, landmarks 1, 3, 4, 5, 6, 11, 13, 14, 15, 17, and 18 are the commonly affected landmarks of the three populations of *S. lemuru* in Surigao del Norte. These landmarks were mostly of the head region such as the anterior tip of the snout at the upper jaw, the contour of the gill cover, the posterior-most portion of the maxillary, and the center of the eye as well as the fins of the fish samples (dorsal fin, caudal fin, and pectoral fin). The high fluctuating asymmetry (FA) among the three populations (57.66 %; 54.43 %; and 49.78 %) also suggests that *S. lemuru* in these locations exhibited morphological deformation.

These deformations such as in the head region could be attributed to the feeding adaptation of fish such as maximizing buccal volume and suction velocity as well as maneuverability when foraging (Caldecott & Adams, 1998; Webb et al., 2008). The shifts in the type of diet and availability of food may also contribute to morphological differences such as head size and mouth position (Haas et al., 2010) which are apparent from the collected fish samples. While deformities in fins can be attributed to their functions such as swimming or locomotion, propelling, stability, and even protection from predators. Thus, the mobility of the fish is considered a factor in determining the direct effect of stressors on the fish, especially in the dorsal, caudal, anal, and pelvic fin or pectoral fin (Natividad et al., 2015).

The length-weight relationship is important since it can provide valuable information on the habitat where the fish lives, modeling aquatic ecosystems, and fish stocks (Pauly, 1993; Kulbicki et al., 2005; King, 2007). *S. lemuru* collected from Malimono has an average length and weight of  $15.06 \pm 1.06$  cm and  $27.04 \pm 5.41$  g respectively while individuals collected in San Francisco have an average length of  $15.15 \pm 0.93$  cm and an average weight of  $31.39 \pm 6.0$  g, and an average length of  $15.82 \pm 0.1$  cm and an average weight of  $33.35 \pm 6.63$  g in Surigao City (table 4). The length and weight of the fish samples have no significant difference ( $p = 0.9937$ ) among sampling locations.

**Table 4.** Length, Weight & b value of *S. lemuru* in three sampling stations

Site	Average ± SD				
	Length, cm	Weight, g	a	b	R <sup>2</sup>
Malimono	15.06 ± 1.06	27.04 ± 5.41	0.0208	2.6398	0.9118
San Francisco	15.15 ± 0.93	31.39 ± 6.01	0.0174	2.7541	0.8207
Surigao City	15.82 ± 0.97	33.35 ± 6.63	0.013	2.8377	0.7861

The “b” values of the LWR obtained were 2.6398 in Malimono, 2.7541 in San Francisco, and 2.8377 in Surigao City, indicating a negative allometric growth pattern. These results suggest that fish samples do not grow symmetrically or it becomes thinner with increasing length (Tesch, 1968; King, 1996). Habitat, fish activities, food habits, seasonal, growth rates, temperature, trophic level, and food availability in the community (Lowe-McConnell, 1987; Mizuno & Furtado, 1982 in Isa et al., 2010) are among the factors that contribute to the differences in the growth of fish.

## Conclusion and Recommendations

This study demonstrates that *Sardinella lemuru* in the coastal waters of Surigao del Norte showed fluctuating asymmetry and deformities observed in the head region and fins. While the length-weight relationship suggests that fish becomes thinner with increasing length. The present study provides pioneering data and essential information with regard to the current condition of *S. lemuru* in the coastal waters of Surigao del Norte, Philippines. Hence, studies in a much longer period of time with seasonal variation, including physico-chemical parameters of water, sex of fishes, and its ecology are suggested to better assess the status of this fish population in the area.

## Acknowledgment

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## Conflict of Interest

The authors declare that there is no conflict of interest regarding the publication of this manuscript.

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