



Meristic and morphometric variation in two emperors (Actinopterygii: Perciformes: *Lethrinus*) in Sulawesi waters

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Abstract. The aim of this research was to determine the differences in meristic and morphometric characteristics between *Lethrinus lentjan* (Lacepède, 1802), and *Lethrinus rubrioperculatus* (Sato, 1978), from two different areas: the waters off Makassar (South Sulawesi) and Manado (North Sulawesi). A total of 120 specimens were used in this research (30 specimens/species/site). Analysis of variance yielded six meristic and 21 morphometric characters selected to be transformed and used in further analysis. The meristic variables making the highest contribution towards between-group discrimination were the number of scales within the lateral line, scale rows between lateral line and base of middle dorsal fin spines, and scale rows in transverse series between origin of anal fin and lateral line. The morphometric data reveal between-population differences in 18 variables for *L. lentjan* and 17 variables for *L. rubrioperculatus*. Both species had higher mean values for morphometric characters in the Makassar population compared to the Manado population. The phenotypic variation observed in this research can be linked to environmental influences, however further research is needed on spatial and temporal segregation or populations or stocks, and whether there is a link between phenotype and genotype in these species.

Key Words: Lethrinid, truss morphometric, discrimination.

Introduction. The pink ear emperor, *Lethrinus lentjan* (Lacepède, 1802), and spotcheek emperor, *Lethrinus rubrioperculatus* (Sato, 1978), are marine demersal fish with widespread tropical and subtropical distributions which include the waters around Sulawesi (Carpenter & Allen 1989; Ezzat et al 1992; Burhanuddin & Iwatsuki 2018). These emperors both belong to the order Perciformes, but differ in body form and dentition type. *L. lentjan* has a deeper body, and is a slow swimmer; it is a carnivorous fish with molariform teeth that can crush benthic organisms. *L. rubrioperculatus* has a more elongated body form and is a somewhat faster swimmer; it is a predatory fish with conical teeth (Lo Galbo et al 2002; Carpenter 1996).

In the waters around Sulawesi, most fishes belonging to the family Lethrinidae are fisheries target species, highly sought after by local fishing communities including those living in and around Makassar City, South Sulawesi and Manado City in North Sulawesi. The official capture fisheries production figures for emperors in 2011 totalled 5,443.3 t from Makassar and 660.0 t from Manado (KKP 2018). The level of destructive fishing varies between these two fishing areas, being classified as high in waters around Makassar and low in waters around Manado (KKP 2019).

These regional differences in fisheries production and fishing pressure have given rise to fears that *Lethrinus* population fitness could be affected, including imbalance in the sex ratio of certain species or populations. Such imbalance could trigger the

emergence of cryptic species, which could affect population structure and lead to changes in morphology (Afrisal et al 2018; Healey et al 2018a).

According to the IUCN Red List, several species in the genus *Lethrinus* are classified as Least Concern, meaning that they are not currently considered at risk of extinction, including *L. lentjan* (Carpenter et al 2016a) and *L. rubrioperculatus* (Carpenter et al 2016b). The assessments note that these fishes are relatively long-lived (at least 19 years for *L. lentjan*) sequential protogynous hermaphrodites with wide Indo-Pacific distributions. Although globally the species is considered at low risk, they note that some populations have experienced localized overfishing, with monitoring and regulation recommended in areas of heavy fishing pressure (Carpenter et al 2016a, b). None of the regional studies cited in these assessments covers Indonesian populations.

One type of research which can inform sustainable and science-based management of *Lethrinus* stocks is the analysis of morphometric, meristic and genetic characteristics. Such data can be used to detect differences between individuals and populations, grouping them based on indices of similarity or through genetic distance and cluster analyses (Burhanuddin 2015). The morphometric truss method has been recommended as superior to conventional morphometric methods because it can accurately and efficiently describe body shape based on a limited number of measurements (Turan et al (2004)

There is a substantial body of published research on *L. lentjan*, in particular on the reproductive biology of this species (Budimawan et al 2013; Currey et al 2013). However, research on *L. rubrioperculatus* is much more limited, especially for Indonesian populations of this species (Young & Martin 1982). However, there is a lack of research on the morphometric and meristic characters of these two species in general, and in particular on populations from environments with different characteristics. The aim of this research was to differentiate the morphometric and meristic characters of *L. lentjan* and *L. rubrioperculatus* from two major fishing areas in the waters around Sulawesi and identify diagnostic characters for each of these *Lethrinus* species. The data and information generated by this research should be of benefit for fisheries development and the sustainable management of lethrinid stocks in the seas around Sulawesi.

Material and Method. *Lethrinus lentjan* and *L. rubrioperculatus* were collected from the waters around Manado City in North Sulawesi and Makassar City in South Sulawesi (Figure 1). Samples of 30 specimens were collected at each site for each species in June and November 2019 (120 specimens in total). Species identification followed Carpenter & Allen (1989).

Observation of morphometric and meristic characters and data analysis were carried out at the Marine Biology Laboratory, Hasanuddin University, Makassar, Indonesia. The morphometric truss design (Figure 2) comprised 21 variables to be measured on each *Lethrinus* specimen.

These characters were based on 10 landmark points, with each character joining two points in horizontal, vertical or diagonal directions. This truss enabled a detailed and specific analysis of fish body shape (Ballesteros-Córdova et al 2016). A description of each character is provided in Table 1.

Meristic characters counted in this study followed Hubbs & Lagler (1958). These characters comprised: lateral-lines scales; scale rows between lateral line and base of middle dorsal fin spines, scale rows in transverse series between origin of anal fin and lateral line; spines and rays in the dorsal, anal, ventral and pectoral fins, and caudal fin rays. The number of gill-rakers was also counted (Burhanuddin et al 2003).

The samples collected differed in terms of size and age; therefore, the morphometric data were standardised using the regression transformation in Elliot et al (1995) before further analysis. This model can be used to remove bias due to size-related effects (Jolicoeur 1963). The regression formula from Elliot et al (1995) is as follows:

$$M_s = M_o \left(\frac{L_s}{L_o} \right)^b$$

where: M_s is the transformed measurement of a given character, M_0 is the untransformed morphometric measurement (in mm), L_s = standard length of the individual measured, and L_t = mean standard length of all individuals measured. The exponent b is the slope of the regression of $\log M$ against $\log L$, where for each individual specimen $\log L$ is the logarithm of the standard length and $\log M$ is the logarithm of the variable of interest.

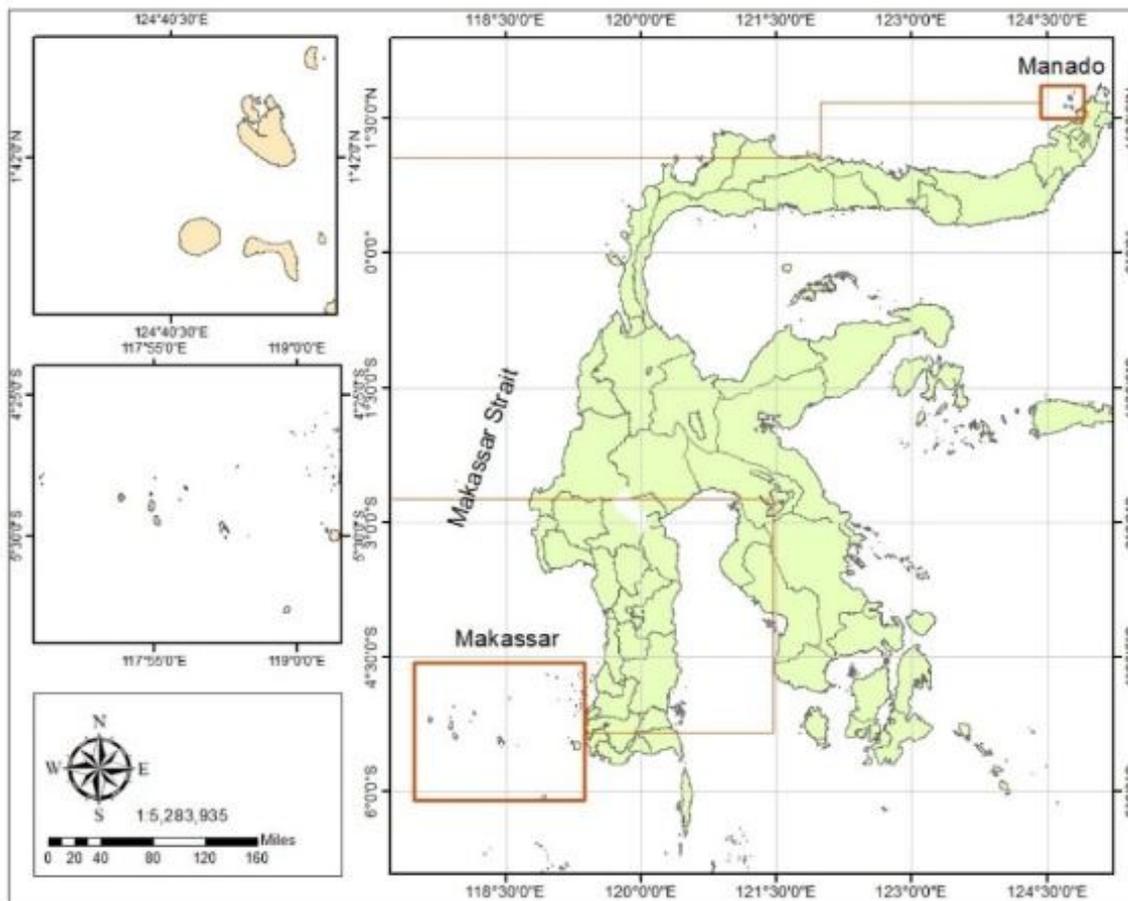


Figure 1. Sampling locations for Lethrinids around Sulawesi: Manado, North Sulawesi, and Makassar, South Sulawesi.

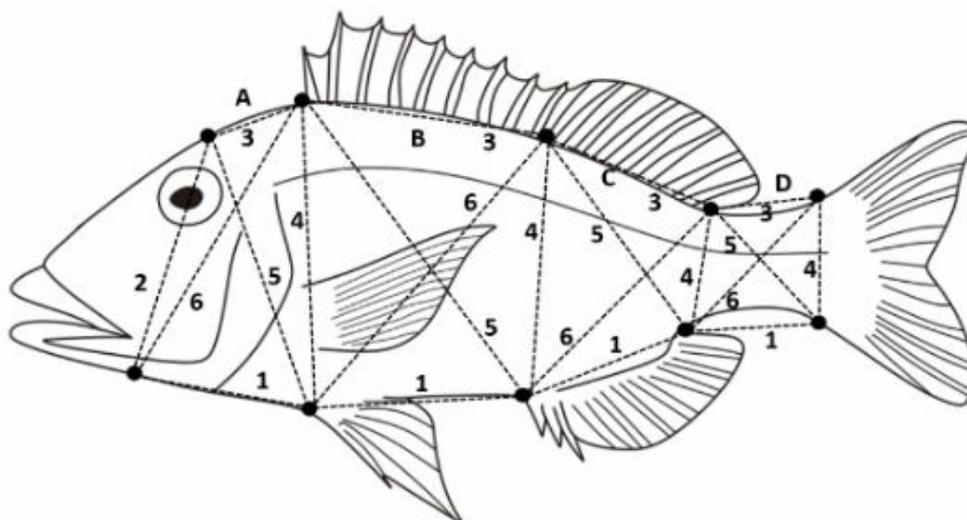


Figure 2. Outline drawing of the *Lethrinus* morphometric truss used, showing the anatomical landmark points (numbered 1-6) and the 21 morphometric distance measures recorded on each individual, following Brzesky & Doyle (1988).

Table 1

Lethrinus morphometric characters measured, based on Brzesky & Doyle (1988)

Body section	Code	Character description
Head	A1	The posteriormost point of the maxilla to the origin of the pelvic fin
	A2	The posteriormost point of the maxilla to the posteriormost point of the eye
	A3	The posteriormost point of the eye to the origin of the dorsal fin
	A4	The origin of the pelvic fin to the origin of the dorsal fin
	A5	The posteriormost point of the eye to the origin of the pelvic fin
	A6	The posteriormost point of the maxilla to the origin of the dorsal fin
Anterior body	B1	The origin of the pelvic fin to the origin of the anal fin
	B3	The origin of the dorsal fin to the point between the spinous and soft portion of the dorsal fin
	B4	The origin of the anal fin to the point between the spinous and soft portion of the dorsal fin
	B5	The origin of the dorsal fin to the origin of the anal fin
	B6	The origin of the pelvic fin to the point between the spinous and soft portions of the dorsal fin
	Posterior body	C1
C3		The point between the spinous and soft portions of the dorsal fin to the insertion of the dorsal fin
C4		The insertion of the anal fin to the insertion of the dorsal fin
C5		The point between the spinous and soft portions of the dorsal fin to the insertion of the anal fin
C6		The origin of the anal fin to the insertion of the anal fin
Tail		D1
	D3	The insertion of the dorsal fin to the anterior attachment of the dorsal membrane from caudal fin
	D4	The anterior attachment of the ventral membrane from caudal fin to the anterior attachment of the dorsal membrane from caudal fin
	D5	The insertion of the dorsal fin to the anterior attachment of the ventral membrane from caudal fin
	D6	The insertion of the anal fin to the anterior attachment of the dorsal membrane from caudal fin

Statistical analyses were conducted in SPSS version 22.0. The standardised morphometric data were tested for normality and homogeneity. As the data obtained were not distributed normally ($p > 0.05$), a square root transformation was applied using the "sqrt" function in SPSS prior to implementing further statistical analyses. In order to compare the mean values of each morphometric parameter and detect site-specific diagnostic characters, for *L. lentjan* t-tests were performed on the transformed data. The transformed data for *L. rubrioperculatus* were not normally distributed, and the original (untransformed) data were therefore analysed using the Mann-Whitney non-parametric test in SPSS.

Results. Based on morphological characters, two samples were identified as *L. lentjan* (Figure 3A) and *L. rubrioperculatus* (Figure 3B). Characters shared by the two species include a fully scaly posterior angle of the operculum, and a single dorsal fin. Specific traits of *L. lentjan* include a moderately deep body, red colouration around the margin of the operculum and the pectoral axil, body colour greenish or grey, shading to white below, and centres of scales on upper sides often white. Characteristics of *L. rubrioperculatus* include a moderately elongate body, brownish body colour with scattered irregular small black blotches, and red colouration of the upper margin of the operculum.

The mean meristic counts of 6 characters were the same for both species. These were: dorsal fin spine and ray counts (DX, 10), anal fin spine and ray counts (AIII, 9), ventral fin spine and ray counts (V, 5), pectoral fin spine and ray counts (P, 12), caudal fin ray count (16), and number of gill-rakers (4).

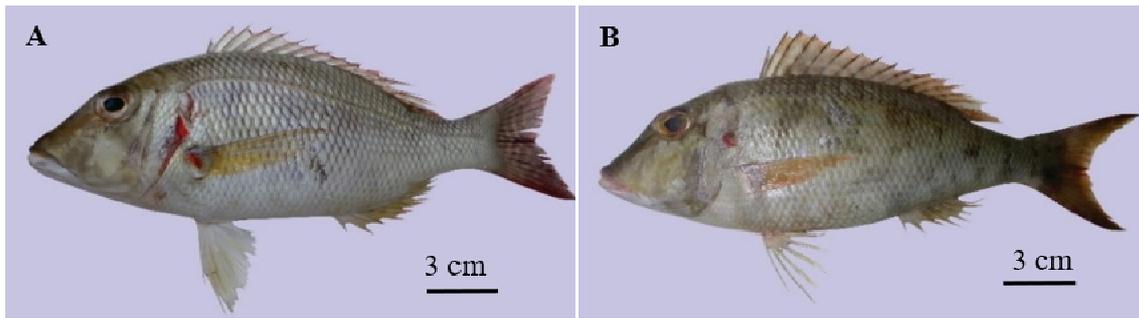


Figure 3. Morphology of two lethrins: *L. lentjan* (A), and *L. rubrioperculatus* (B).

Three meristic characters differed between species and site. The variation in these three counts was higher in *L. lentjan* from Manado than from Makassar. The lateral line scale count varied from 44-49 in Manado, and 45-50 in Makassar. The number of scale rows above the lateral line was more varied in Manado (5, 5.5, 6) than in Makassar (5.5, 6), while the number of scale rows below the lateral line ranged from 13 to 15 in Manado but was 14 in all specimens from Makassar.

A similar but opposite result was found for *L. rubrioperculatus*, with greater variation in specimens from Makassar compared to those from Manado. The lateral line scale count varied from 43 to 49 in Manado and 46-50 in Makassar. The number of scale rows above the lateral line ranged from 4 to 6 in Manado and from 5 to 6 in Makassar. However, the number of scale rows below the lateral line ranged from 14 to 16 at both study sites.

The mean values (\pm standard deviation) of the 21 morphometric characters measured for the two lethrins species from Manado and Makassar are shown in Table 2, together with the level of significance of between-site differences. The t-test showed that 18 out of 21 ratios differed significantly for *L. lentjan*, with mean values higher for the Makassar population than the Manado population.

Table 2
Variation in morphometric characters (mean \pm standard deviation) and significance level of between-site difference for two lethrins species

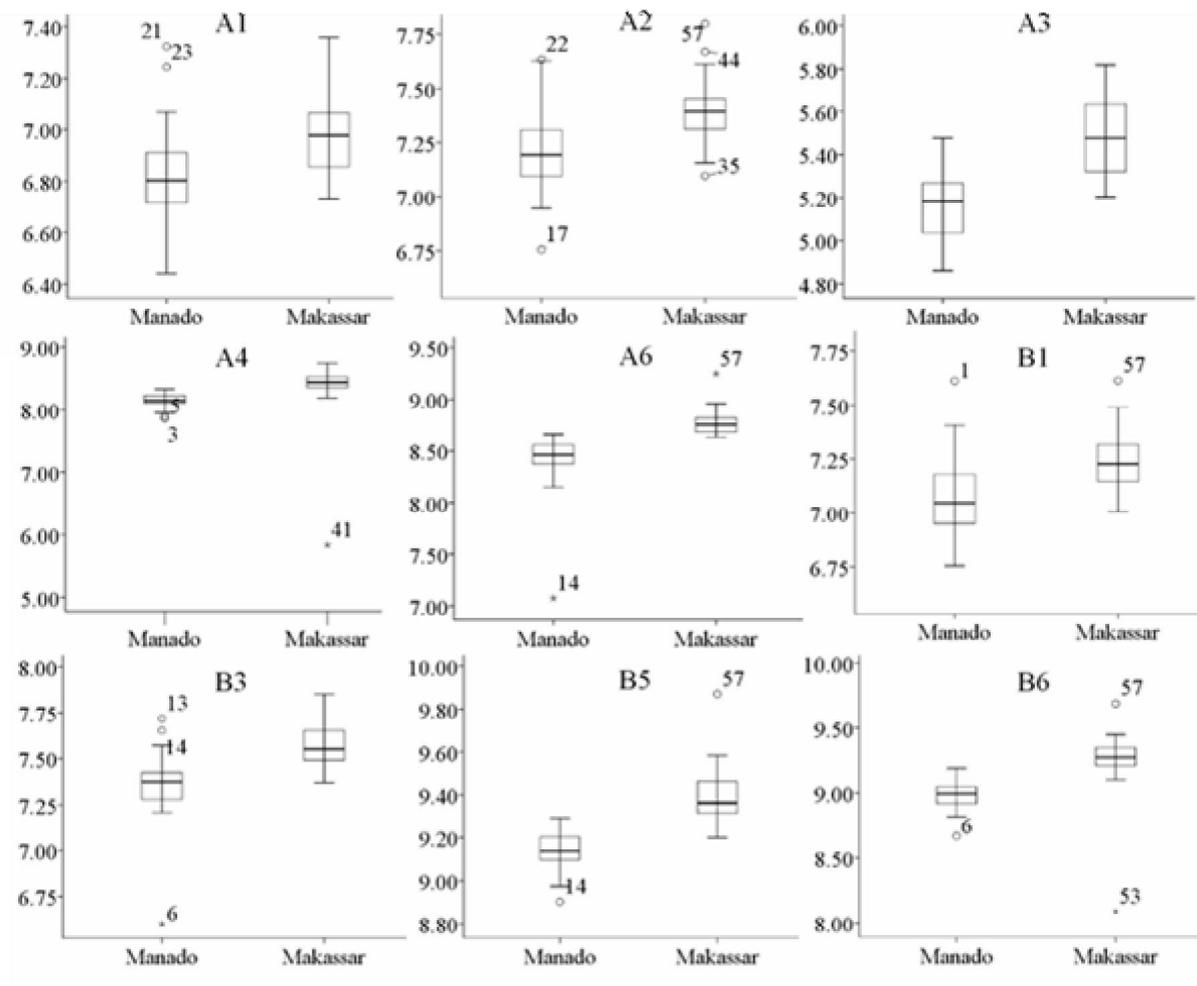
Code	<i>L. lentjan</i>		Sig*	<i>L. rubrioperculatus</i>		Sig*
	Manado	Makassar		Manado	Makassar	
	Mean \pm SD			Mean \pm SD		
A1	6.829 \pm 0.181	6.968 \pm 0.144	***	47.811 \pm 1.408	45.587 \pm 6.277	NS
A2	7.205 \pm 0.188	7.384 \pm 0.147	***	48.292 \pm 1.506	57.048 \pm 9.772	NS
A3	5.170 \pm 0.172	5.481 \pm 0.187	***	24.260 \pm 1.506	25.763 \pm 3.838	***
A4	8.141 \pm 0.118	8.375 \pm 0.501	***	58.107 \pm 1.885	71.740 \pm 11.800	***
A5	7.986 \pm 0.176	8.144 \pm 0.557	NS	57.155 \pm 1.842	68.552 \pm 10.451	NS
A6	8.423 \pm 0.278	8.778 \pm 0.125	***	66.019 \pm 1.996	75.472 \pm 11.854	***
B1	7.070 \pm 0.178	7.237 \pm 0.136	***	49.147 \pm 1.924	49.222 \pm 7.184	NS
B3	7.360 \pm 0.186	7.582 \pm 0.117	NS	55.517 \pm 1.236	56.365 \pm 7.960	NS
B4	7.937 \pm 0.324	8.050 \pm 0.149	***	53.038 \pm 1.520	59.419 \pm 4.767	***
B5	9.143 \pm 0.096	9.399 \pm 0.132	***	76.477 \pm 1.473	84.751 \pm 7.098	***
B6	8.985 \pm 0.107	9.257 \pm 0.248	***	75.728 \pm 2.216	83.944 \pm 6.903	***
C1	5.988 \pm 0.621	5.984 \pm 0.109	NS	32.869 \pm 1.097	35.692 \pm 3.334	***
C3	5.825 \pm 0.138	6.021 \pm 0.120	***	32.301 \pm 1.977	36.169 \pm 1.424	***
C4	5.559 \pm 0.069	5.649 \pm 0.109	***	28.798 \pm 0.777	32.180 \pm 2.397	***
C5	7.519 \pm 0.129	7.704 \pm 0.135	***	51.500 \pm 1.234	56.680 \pm 2.044	***
C6	7.427 \pm 0.081	7.548 \pm 0.105	***	51.801 \pm 5.099	56.870 \pm 1.964	***
D1	5.414 \pm 0.188	5.547 \pm 0.210	***	30.743 \pm 1.156	36.131 \pm 3.295	***
D3	5.307 \pm 0.189	5.447 \pm 0.174	***	29.609 \pm 1.855	33.933 \pm 4.849	***
D4	4.832 \pm 0.123	4.925 \pm 0.107	***	20.525 \pm 1.850	23.001 \pm 1.403	***
D5	6.183 \pm 0.131	6.399 \pm 0.124	***	37.059 \pm 2.615	42.414 \pm 3.211	***
D6	6.293 \pm 0.219	6.401 \pm 0.180	***	39.424 \pm 1.325	46.147 \pm 2.918	***

*t-test and non-parametric Mann-Whitney test: *** indicates a significant difference and NS = no significant difference (at $\alpha = 0.05$).

The three characters which did not differ significantly between the two *L. lentjan* populations were situated in the head section, anterior body section, and posterior body section. These were, respectively: the posteriormost point of the eye to the origin of the pelvic fin (A5); the origin of the dorsal fin to the point between the spinous and soft portion of the dorsal fin (B3); and the origin of the anal fin to the insertion of the anal fin (C1).

For *L. rubrioperculatus*, the Mann-Whitney non-parametric tests showed significant between-site differences ($p < 0.05$) for 17 out of the 21 characters measured. The four characters which did not differ significantly between the two research sites were A1, A3, B1, and B3. Two of these were in the head section: A1 (posteriormost point of the maxilla to the origin of the pelvic fin) and A3 (posteriormost point of the maxilla to the posteriormost point of the eye). The other two were in the anterior body section: B1 (origin of the pelvic fin to the origin of the anal fin) and B3 (origin of the dorsal fin to the point between the spinous and soft portion of the dorsal fin).

Box plots (Figure 4) demonstrate the between-site difference in morphometric characters for *L. lentjan*. The highest number of outlier values (*L. lentjan* specimens from Manado and Makassar) was observed in the A2 character (posteriormost point of the maxilla to the posteriormost point of the eye). The D6 character (insertion of the anal fin to the anterior attachment of the dorsal membrane from caudal fin) had the lowest number of outlier values (1 specimen). There were no outlier values for two characters: A3 (posteriormost point of the eye to the origin of the dorsal fin), D3 (insertion of the dorsal fin to the anterior attachment of the dorsal membrane from caudal fin).



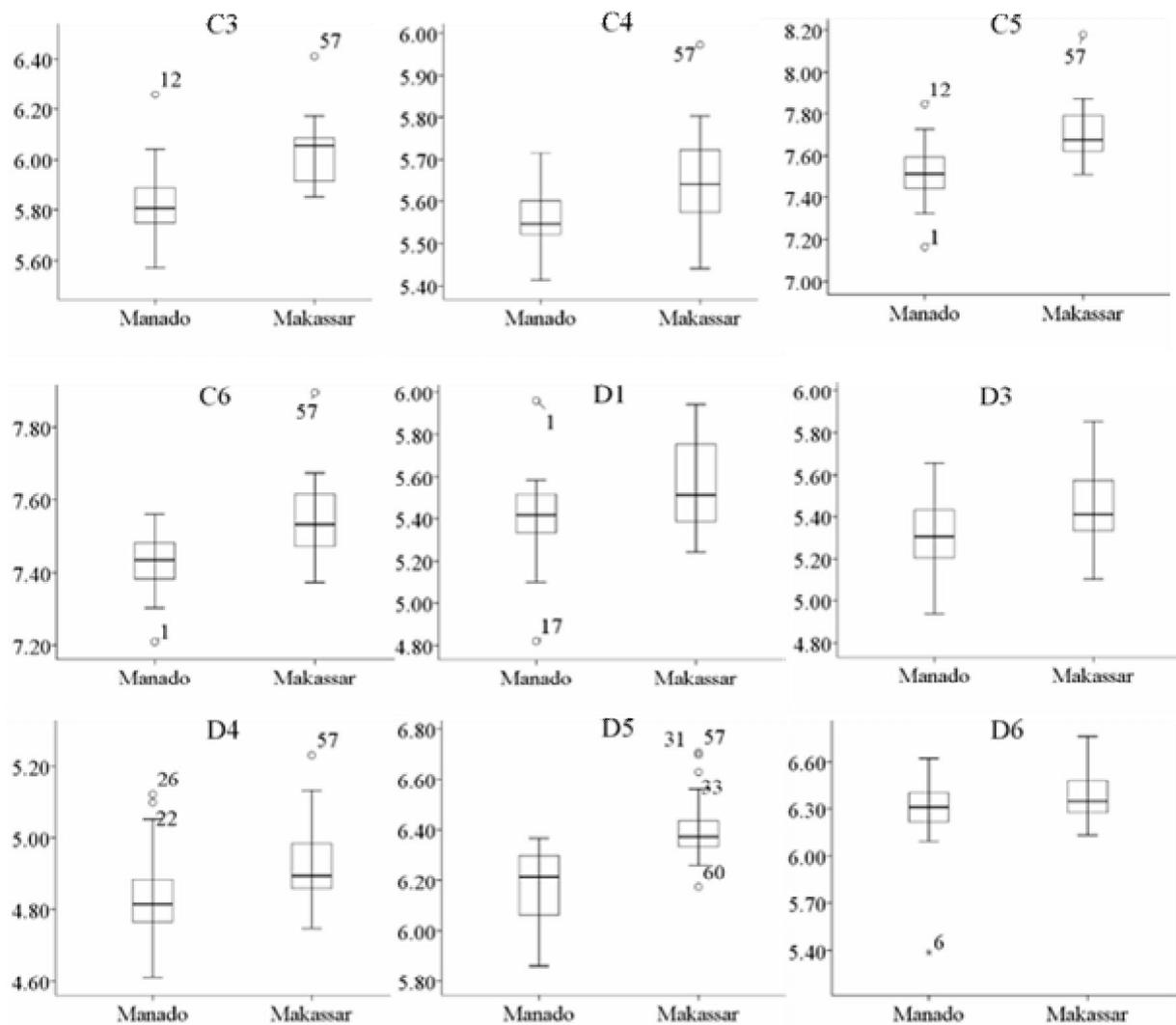
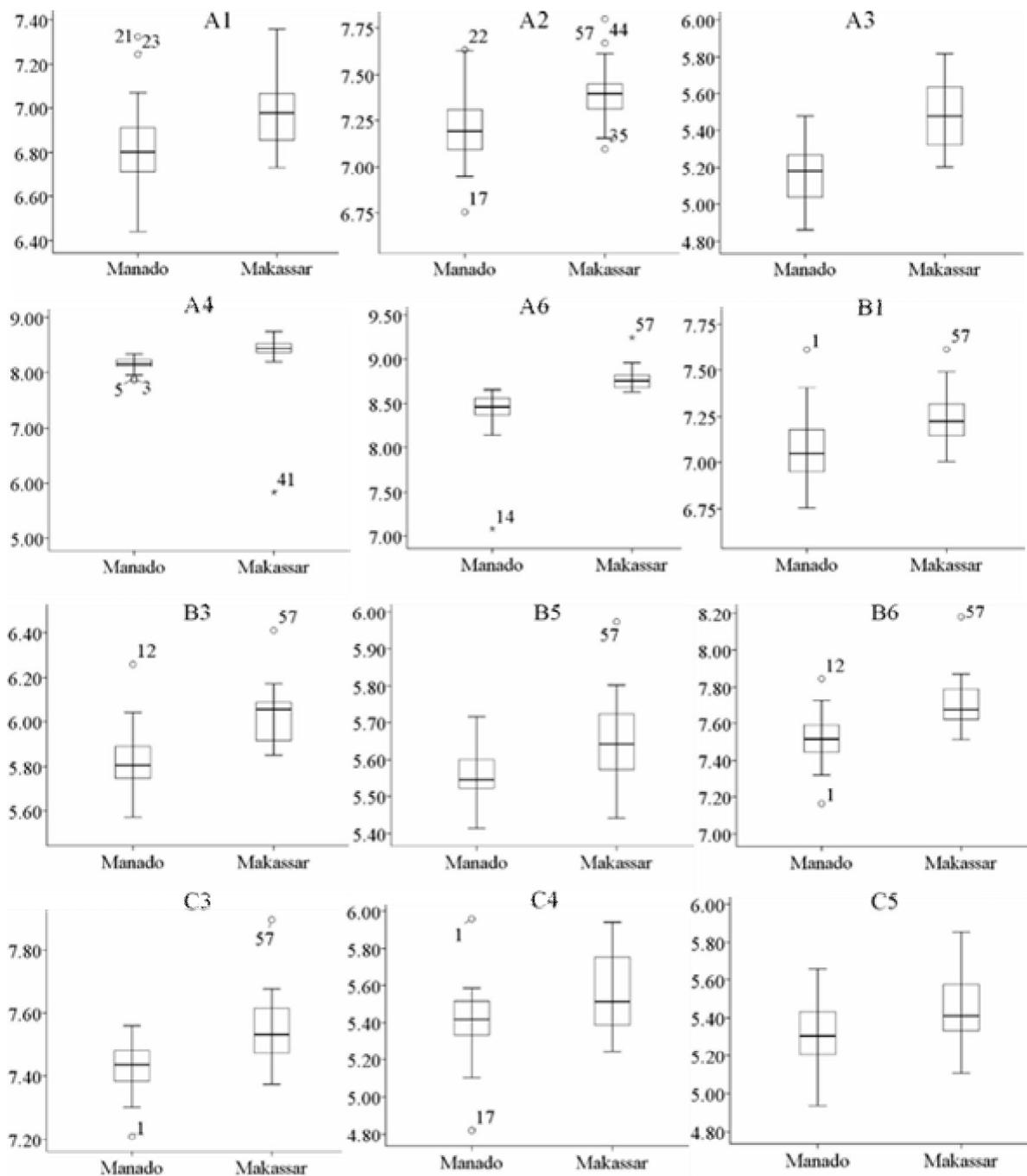


Figure 4. Box plot (bold line = mean value; box = interquartile range; whiskers denote 0.95 confidence interval; circles denote outliers) of the 18 most notable morphometric characters differentiating *L. lentjan* specimens collected from Manado (n = 30) and Makassar (n = 30). The numbers beside each outlier denote the serial number allocated to the specimen in question.

Similar box plots (Figure 5) show the spread of morphometric characteristics for the *L. rubrioperculatus* collected from the two study sites. The highest number of outlier values (5 specimens) observed was for the A2 character (posteriormost point of the maxilla to the posteriormost point of the eye). There was one outlier specimen each for characters C4 (insertion of the anal fin to the insertion of the dorsal fin) and D6 (insertion of the anal fin to the anterior attachment of the dorsal membrane from caudal fin). There were no outliers for the characters A3 (posteriormost point of the eye to the origin of the dorsal fin) and D3 (insertion of the dorsal fin to the anterior attachment of the dorsal membrane from caudal fin).



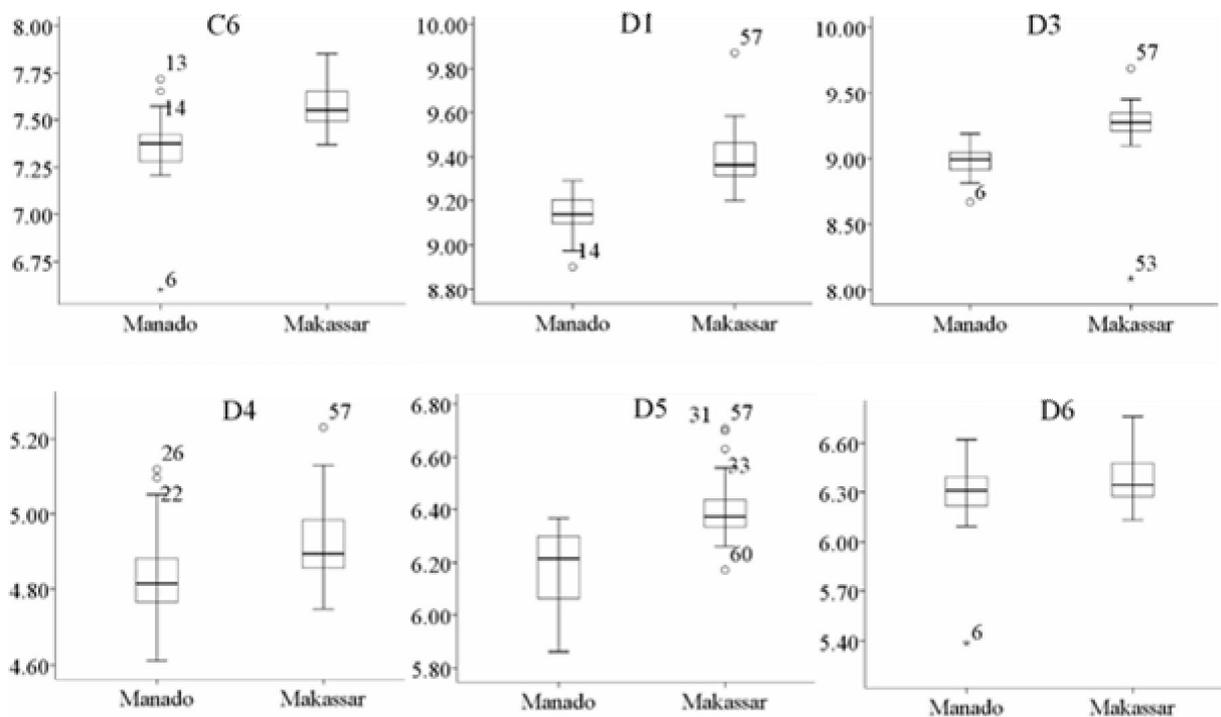


Figure. 5. Box plot (bold line = mean value; box = interquartile range; whiskers denote 0.95 confidence interval; circles denote outliers) of the 18 most notable morphometric characters differentiating *L. rubrioperculatus* specimens collected from Manado (n = 30) and Makassar (n = 30).

Discussion. Taxonomically, lethrins are considered one of the most problematic groups of tropical fishes (Carpenter & Allen 1989; Mekkawy 2017). Although molecular biology methods such as DNA barcoding are gaining in popularity, and have contributed to the elucidation of lethrins phylogeny and taxonomy (Mekkawy 2017), traditional morphological methods are still important, especially in a field identification context. Commonly used methods for species identification include the use of external morphological characters such as fin spines and rays, dentition, body shape, and colour or pigmentation patterns (Burhanuddin & Iwatsuki 2003). Identification keys based on meristic characters of fish in the genus *Lethrinus* are practical and relatively easy to use on adult specimens, but can often fail to resolve the taxonomy of juvenile specimens (Carpenter & Allen 1989). Furthermore, according to Lo Galbo et al (2002), variation in morphological characters can be strongly influenced by feeding habits and the adaptation of individuals to their environment, and can also be affected by homoplasy.

It has been suggested that higher variation in meristic counts may be related to differences in environmental conditions (Turan et al 2004). Many fish exhibit phenotypic and behavioural plasticity which enables them to adapt to their environment and changes in environmental conditions through modifications in physiology and behaviour (Clayton 1981; Stearns 1983; Meyer 1987). Such adaptations can lead to changes in morphology, reproduction and survival capacity due to environmental drivers (Jawad 2001). The meristic counts in this study tended to be higher than those reported from previous studies. For *L. lentjan*, Carpenter & Allen (1989) report 46 or 47 lateral line scales, with 5 1/2 scale rows between the lateral line and base of middle dorsal fin spines and 15 or 16 scale rows in transverse series between origin of the anal fin and the lateral line. For *L. rubrioperculatus* they note an absence of scales on the cheek and report 47 to 49 lateral-line scales; 4.5 scale rows between lateral line and base of middle dorsal fin spines; and 15 or 16 scale rows in transverse series between the origin of anal fin and lateral line. These differences indicate that both Sulawesi populations of the two lethrins species studied have undergone morphological differentiation, although in general the meristic

counts were higher for specimens from Manado compared to those from the Makassar site.

Fishing pressure and destructive fishing do not appear to be influencing factors with respect to morphometric characters. This evaluation is based on the higher values of morphometric characters recorded for the specimens collected from Makassar compared to those from Manado. One factor which can have a highly significant influence is the availability of food. Wimberger (1992) states that fish body shape is affected by the genetic makeup of an individual as well as by environmental influences, such as diet, development, growth rate and nutrition. The generally larger values of morphometric characters in the fish from Makassar could be due to a greater abundance of food and/or reduced competition for food, thus influencing condition, as well as adaptation to the local environment, in line with the findings of Wimberger (1992). Intraspecies variations in morphology can also be related to condition factor, which reflects the fatness of an individual based on its length and weight (Lagler et al 1977). Studies on the length-weight relation of *L. lentjan* from around the world generally report an allometric growth pattern, with the exception of the Egyptian Red Sea where the growth pattern was isometric (Currey et al 2013; Zaahkouk et al 2017). Furthermore, Norau (2010) reported variation in the length-frequency composition of *L. lentjan* caught in the coral reef areas of the Gurraici Islands, in Halmahera Selatan Sub-District based on coral condition. The larger length classes were more common where the reefs were in good condition compared to degraded reefs.

In addition to the length-weight relationship and growth pattern, reproductive characteristics are also an important consideration. Lethrinids are protogynous hermaphrodites (Marriott et al 2010), however the two species in this study change sex from female to male at different ages and sizes. While sex change can occur in *L. lentjan* at ages of five years or more at a total length of around 33 cm (Wassef 1991), *L. rubrioperculatus* tend to become males at 3-4 years of age at a fork length (FL) of around 28-29 cm (Ebisawa 1997). However, Ebisawa (1997) reports sex change over a range of 26-42 cm FL; furthermore, research on the biological characteristics of *L. rubrioperculatus* in the Northern Mariana Islands (Trianni 2011) and Ryukyu Islands (Ebisawa 1997) reveals that the size range at which sex change takes place varies between populations. Most *Lethrinus* specimens in this study were below the reported size range in which sex change takes place, indicating a possible imbalance in the sex ratio. It can be assumed that the majority of the fish in this study were still in the female stage. According to Effendie (2002), the relative numbers of female and male fish caught can be affected by differential patterns of movement or migration (for feeding and spawning), growth patterns, mortality and age at first maturity for each sex. Furthermore, regional or site-based differences in topography, habitat typology and environmental gradients can influence genetic characteristics, potentially leading to the evolution of cryptic species; such processes have been reported in the genus *Lethrinus* (Healey et al 2018b).

Conclusions. The meristic counts were more varied in *Lethrinus lentjan* from Manado than in those from Makassar; conversely, meristic counts of *L. rubrioperculatus* from Makassar varied more than those from Manado. The mean values of 21 morphometric characters for these two lethrinids were higher in Makassar than Manado.

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