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## Morphological Variations of *Plicofollis* Species (Siluriformes: Ariidae) in Peninsular Malaysia: An Insight into Truss Network Approach

(Variasi Morfologi Spesies *Plicofollis* (Siluriformes: Ariidae) di Semenanjung Malaysia: Pemerhatian Pendekatan Rangkaian Truss)

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### ABSTRACT

*Ariid catfishes, belong to family Ariidae is considered as one of the taxonomically problematic groups, which is still under review by fish taxonomist globally. Species level identification of some ariids often resulted in species misidentification because of their complex characters and very similar morphological characters within genera. A vigilant and detail observation is very important during the species level identification of ariid species. In these contexts, this study was carried out in order to determine the morphological variations of one of the ariid genera, Plicofollis, which have been giving misleading taxonomic information in the south-east Asian countries. A Truss network technique was used throughout the study period. The study was conducted based on 20 truss measurements using 22 to 23 specimens per species, namely P. argyropleuron, P. nella and P. tenuispinis found in Peninsular Malaysian waters. Morphological variations were determined using a multivariate technique of discriminant function analysis (DFA). The results obtained in this study showed that discriminant analysis using truss network measurements has produced very clear separations of all the species in Plicofollis group. Several important morphological characters have been identified, which represent body depth and caudal regions of the fish. The documentary evidences of these variables could be considered as the constructive functional features, which could enable us to assess more accurately to distinguish the species within this complex Ariidae family.*

*Keywords: Ariidae; DFA; morphology; Plicofollis; P. argyropleuron; P. nella; P. tenuispinis; Truss network*

### ABSTRAK

*Ikan ariid yang tergolong dalam keluarga Ariidae merupakan kumpulan ikan yang mempunyai masalah daripada segi pengelasan taksonominya dan masih dikaji di seluruh dunia. Ciri morfologi yang hampir sama sesama spesies dalam genus ariid sering menyebabkan kesalahan dalam mengenalpasti spesies. Pemerhatian yang khusus seringkali diperlukan semasa mengenal pasti spesies ariid ini. Oleh itu, kajian ini dijalankan bertujuan untuk menentukan variasi morfologi salah satu genus ikan ariid, iaitu Plicofollis, yang mempunyai maklumat taksonomi yang mengelirukan di negara-negara Asia Tenggara. Kaedah rangkaian Truss telah diguna dalam kajian ini berdasarkan 20 ukuran truss dengan menggunakan 22 ke 23 spesimen bagi setiap spesies ikan (P. argyropleuron, P. nella dan P. tenuispinis) yang ditemui di perairan Semenanjung Malaysia. Variasi morfologi telah ditentukan menggunakan analisis tinggi diskriminan (DFA). Keputusan menunjukkan bahawa ketiga-tiga spesies Plicofollis telah dipisahkan dengan jelas dalam kumpulan spesies masing-masing. Beberapa ciri morfologi yang berperanan dalam pemisahan spesies Plicofollis telah dikenal pasti, iaitu kawasan yang mewakili kedalaman badan dan ekor ikan. Bukti yang diperoleh ini membolehkan kami menilai dengan lebih tepat dalam proses mengenalpasti spesies daripada keluarga Ariidae yang kompleks ini.*

*Kata kunci: Ariidae; DFA; morfologi; Plicofollis; P. argyropleuron; P. nella; P. tenuispinis; rangkaian Truss*

### INTRODUCTION

The order Siluriformes is composed of 37 recognized families of catfish that are widely distributed and highly diversified in freshwaters (Sullivan et al. 2006). Among these families, only two apomorphic families were adapted in saltwater: Plotosidae from the Indo-West Pacific and Ariidae. Ariids, or sea catfish, comprise approximately 150 species (Betancur-R 2009). Ariid sea catfishes can be considered as the most important group of fishes inhabit tropical estuaries in terms of number of species, density

and biomass (Barletta & Blaber 2007; Barletta et al. 2008; Blaber 2008; McConnell & Lowe-McConnell 1987). These ariids group are well adapted to live in different habitats of estuaries of the tropical and subtropical world (Barletta & Blaber 2007). Their eury-thermohaline capacity and the possession of a Weberian apparatus are considered as the most important characteristics which help this group of fishes to successfully adapt in freshwater, estuarine and marine environments (Ashraf et al. 2011; Dantas et al. 2010).

The family Ariidae is one of the taxonomically problematic group of order Siluriformes. Several taxonomic studies were carried out on ariids. Acero et al. (2007), Acero and Betancur-R (2007) and Betancur-R (2009) reported the systematic and phylogenetic relationships among ariid species. Several new species of ariids were studied and described by some scientists, such as *Arius midgleyi* (Kailola & Pierce 1988), *Notarius biffi* and *Notarius insculptus* (Betancur-R & Acero 2004), *Cathoropsmapale* (Betancur-R & Acero 2005), *Notarius armbrusteri* (Betancur-R & Acero 2006) and *Cathorops raredonae* (Marceniuk & Acero 2009).

The diagnostic characters available for the identification of ariids species are unclear and confusing (Mazlan et al. 2008; Ng 2012). The diagnostic characters described often overlapped between several species. Sometimes the identification characters described could also lost on the specimen due to sampling and handling processes. Since the systematic of ariids is complex, the diagnostic treatments of the species needed to be applied with extensive morphometric and meristic characters. Therefore, truss network technique was applied to provide supplementary taxonomic information to enhance the species identification of *Plicofollis* species and to avoid difficulties on further species identification. The truss network technique, also known as modern morphometric technique (Ashraf et al. 2012; Strauss & Bond 1990; Strauss & Bookstein 1982) was applied by using discriminant

function analysis. The contribution on understanding of the systematic relationship of this technique was also proven by Turan et al. (2011), who carried out a study of Mediterranean grey mullet species.

#### MATERIALS AND METHODS

The samples were collected at different fish landing sites throughout Peninsular Malaysian waters (freshwater, brackish and marine environment) since May 2011 to October 2013. The fish samples were bought at Government Fish Landing Ports (LKIM), local fishermen and also from various fish markets.

The relevant literatures were broadly studied in order to identify the fish samples, such as keys, checklist and description of the fishes. Several fish taxonomy guidelines were used in order to identify the experimental samples (Kailola 1999; Kong 1998; Mansor et al. 1998; Marceniuk & Menezes 2007; Matsunuma et al. 2011; Mazlan et al. 2008).

Overall a total of 68 samples were measured and analysed for morphological variations (Table 1). The measurements were conducted based on truss network anchored at 10 homologous landmarks. This resulted in 20 linear measurements (Figure 1). These measurements were performed to the nearest 0.01 mm using a digital calliper.

The data obtained from truss network technique were firstly standardized for size according to Murta

TABLE 1. Samples collected and analysed for morphological variations

No.	Species	No. of samples analysed
1	<i>P. argyropleuron</i>	23
2	<i>P. nella</i>	22
3	<i>P. tenuispinis</i>	23
	Total	68

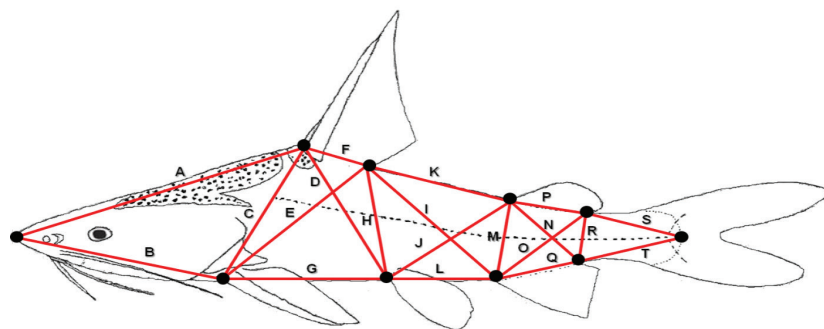


FIGURE 1. Truss network distances of ariids family

(A: Snout to origin of dorsal fin; B: Snout to pectoral fin insertion; C: Pectoral fin insertion to origin of dorsal fin; D: Origin of dorsal fin to pelvic fin insertion; E: Pectoral fin insertion to end of dorsal fin; F: Origin of dorsal fin to end of dorsal fin; G: Pectoral fin insertion to pelvic fin insertion; H: End of dorsal fin to pelvic fin insertion; I: End of dorsal fin to origin of anal fin; J: Pelvic fin to origin of adipose fin; K: End of dorsal fin to origin of adipose fin; L: Pelvic fin insertion to origin of anal fin; M: Origin of adipose fin to origin of anal fin; N: Origin of adipose fin to end of anal fin; O: Origin of anal fin to end of adipose fin; P: Origin of adipose fin to end of adipose fin; Q: Origin of anal fin to end of anal fin; R: End of adipose fin to end of anal fin; S: End of adipose fin to caudal fin; and T: End of anal fin to caudal fin)

et al. (2008). This transformation reduces the effect on different specimen sizes (Reist 1985). It normalizes the individuals in a sample to a single arbitrary size, common to all samples, but maintains the individual variation (Sen et al. 2011). In this study, standard length (SL) was used as a common factor since it strongly correlates with other morphological characters (Ahmed et al. 2015; Jaferian et al. 2010; Reist 1985). An allometric formula by Elliott et al. (1995) was used for the transformation:

$$M_{trans} = \log M - b ((\log SL - \log SL_{mean})),$$

where  $M_{trans}$  is the transformed measurement;  $M$  is the original measurement;  $b$  is the within-group slope regression of the  $\log M$  versus  $\log SL$ ;  $SL$  is the standard length of the fish; and  $SL_{mean}$  is the overall mean of the standard length.

The transformed data were analysed by one-way ANOVA and multivariate analysis of DFA using statistical Package for Social Science (SPSS) version 16.0 software for windows. One-way ANOVA was used in this study in order to identify the significant variables of the truss network measurements. A consequence post-hoc test, namely Bonferroni was also carried out to interpret the results when significant results with  $p$ -value less than 0.05 were obtained from the ANOVA to determine which group were significant from each other.

The relative importance of discriminant functions was determined on the basis of three measures, which were the relative percentage of the eigenvalue and the percent of variance existing in the discriminating values, the associated canonical correlation and Wilk's Lambda and its corresponding chi-square (Muhamad & Mohamad 2012).

The eigenvalue usually indicates the proportion of variance explained, where a large eigenvalue is associated with a strong function. The canonical correlation is a measure of association which summarizes the degree of relatedness between the groups (species) and the discriminant function. The larger value, increase the degree of association, where 1.0 considered as maximum value. While, Wilk's Lambda is an inverse measure of the discriminating power in the original variables, hence, the larger the value, the less discriminating is the function (Borcard et al. 2012; Said et al. 1992). The standardized discriminant function coefficient describes the relative contribution of its associated variables to the functions; the sign indicates positive or negative contribution. The larger the magnitude of the coefficient (disregarding sign), the greater is the variables contribution to the function, while the opposite is true for the lowest coefficients (Said et al. 1992).

## RESULTS AND DISCUSSION

Out of 20 truss network variables measured, 16 variables showed significant differences among the species except variables  $F$  (origin of dorsal fin to end of dorsal fin),  $G$  (pectoral fin insertion to pelvic fin insertion),  $K$  (end of dorsal fin to origin of adipose fin) and  $P$  (origin of adipose fin to end of adipose fin). Table 2 displays the truss network measurements expressed as percent of standard length of all three *Plicofollis* species found in this study.

Table 3 shows the summary of the relevant statistics of the discriminant functions analysis for *P. argyropleuron*, *P. nella* and *P. tenuispinis*. A total of two functions using 16 significant variables were extracted. All the extracted functions explain 100% of the total variability. First function described 62.3% of discriminating power, while second function with 37.7%, respectively.

TABLE 2. Truss network measurements of *P. argyropleuron*, *P. nella* and *P. tenuispinis* expressed as percent of standard length

Variables	Measurements (%SL)		
	<i>P. argyropleuron</i>	<i>P. nella</i>	<i>P. tenuispinis</i>
A	34-49 (43.1, 22)	41-49 (44.1, 22)	39-59 (45.3, 23)
B	22-32 (28.8, 22)	24-34 (30.3, 22)	26-74 (32.2, 23)
C	22-29 (25.3, 22)	23-27 (25.4, 22)	22-32 (26.1, 23)
D	24-33 (27.3, 22)	26-33 (29.7, 22)	22-31 (27.7, 23)
E	22-39 (31.6, 22)	26-33 (31.0, 22)	26-35 (30.6, 23)
H	18-30 (22.9, 22)	22-36 (25.3, 22)	20-28 (23.3, 23)
I	27-41 (33.9, 22)	23-40 (34.8, 22)	31-45 (35.2, 23)
J	19-35 (29.9, 22)	27-32 (29.3, 22)	26-34 (30.3, 23)
L	16-26 (19.8, 22)	16-22 (18.4, 22)	15-24 (19.4, 23)
M	16-23 (19.2, 22)	17-21 (18.9, 22)	16-24 (20.4, 23) <sup>†</sup>
N	13-18 (15.6, 22)	15-19 (17.1, 22)	15-21 (17.8, 23)
O	15-23 (18.7, 22)	17-21 (19.2, 22)	16-24 (20.9, 23)
Q	9-16 (12.1, 22)	8-14 (11.1, 22)	10-16 (13.2, 23)
R	9-15 (11.8, 22)	12-17 (14.8, 22) <sup>†</sup>	10-15 (13.0, 23)
S	15-23 (18.6, 22)	17-24 (20.7, 22)	17-24 (21.1, 23)
T	13-19 (16.7, 22)	16-19 (17.6, 22)	14-21 (17.0, 23)

Figures in parentheses indicate mean values and sample sizes

TABLE 3. Summary of canonical discriminant for *P. argyropleuron*, *P. nella* and *P. tenuispinis*

Function	Eigen-value	Variance (%)	Cumulative (%)	Canonical correlation	Wilks' Lambda	Chi-square	df	Sig.
1	4.043 <sup>a</sup>	62.3	62.3	0.895	0.057	162.804	34	0.000
2	2.449 <sup>a</sup>	37.7	100.0	0.843	0.290	70.578	16	0.000

a. First two canonical discriminant functions were used in the analysis

Table 4 shows the values of functions at group centroids for all the three *Plicofollis* species. The highest value in every function shows the ability of that function to discriminate the respective species according to the variables tested. For *Plicofollis* species, the first function was able to discriminate *P. tenuispinis* from the other two *Plicofollis* species followed by the second function, which was able to discriminate *P. argyropleuron* and *P. nella*.

TABLE 4. Functions at group Centroids for *P. argyropleuron*, *P. nella* and *P. tenuispinis*

Species	Functions	
	F1	F2
<i>P. argyropleuron</i>	-0.279	-2.129
<i>P. nella</i>	-2.316	1.283
<i>P. tenuispinis</i>	2.494 <sup>†</sup>	0.902

The discrimination of the *P. argyropleuron*, *P. nella* and *P. tenuispinis* based on truss network measurements was clearly illustrated in scatter plot as shown in Figure 2. The figure provides an understanding of the relative position of the fish with the axes of the model scaled in relation to the first and second canonical discriminant functions. Each point, which representing a fish specimen in the model is classified according to the centroid to which it is closest. The scatter plot appears reasonably good and useful in pointing the degree of similarity and divergence between species.

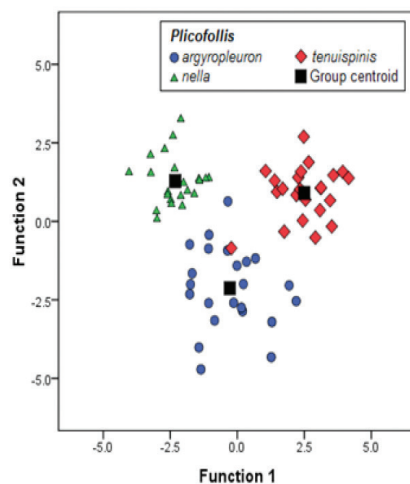
FIGURE 2. Discriminant scatter plot of *P. argyropleuron*, *P. nella* and *P. tenuispinis*

Table 5 shows the structure matrix of canonical discriminant function coefficients for *Plicofollis* species. Variables *M* (origin of adipose fin to origin of anal fin) makes the greatest contribution to the first function with the coefficient value of 0.418. Followed by the second function, with the variable of *R* (end of adipose fin to end of anal fin) with the respective coefficient value of 0.447. Therefore, the results showed that *P. tenuispinis* recorded higher mean value in body depth (variable *M*), while, *P. nella* showed significantly higher caudal region measurement (variable *R*) as in Table 2.

TABLE 5. Standardized canonical discriminant function coefficients

Variables	F1	F2
A	0.311	0.199
B	0.194	0.173
C	0.340	0.136
D	0.071	0.253
E	0.188	-0.031
H	0.056	0.245
I	0.241	0.100
J	0.281	0.027
L	0.249	-0.143
M	0.418 <sup>†</sup>	0.123
N	0.294	0.350
O	0.402	0.241
Q	0.383	0.023
R	-0.016	0.447 <sup>†</sup>
S	0.306	0.432
T	0.122	0.206

† Pooled within-groups correlations between discriminating variables and standardized canonical discriminant functions

Morphometric characters is a quantitative description, which have been successfully used for taxonomic inferences. There are many well-documented morphometric studies that provides evidence for taxonomic dilemmas (Barriga-Sosa et al. 2004; Erguden & Turan 2005). Moreover, a new system of morphometric measurements called the truss system (Bookstein 1982) has been increasingly used for population and taxonomic studies (Ahmed et al. 2014; Cakmak & Alp 2010; Cavalcanti et al. 1999).

In the present study, the discriminant function analysis applied for the *Plicofollis* species using truss network measurements showed clear separation among all the species. The taxonomic description of a species has commonly relied on description of unique sets of morphological characters. The truss measurements which

represent body depth and caudal region were the most discriminate characters in the present classification. The results showed that higher mean values were recorded for *P. tenuispinis* in body depth (origin of adipose fin to origin of anal fin). However, *P. nella* counterparts had significantly higher caudal region measurement (end of adipose fin to end of anal fin).

The superiority exhibited by *P. tenuispinis* and *P. nella* may be as a result of its genetic attributes. As reported by Barton and Turelli (1989), the evolutionary changes in genetic and morphometric characters have been known to be differently affected by many factors including environmental conditions. According to Muchlisin (2013), generally the morphometric approaches showed that the head and caudal region were the major characters for distinguishing the groups. Similar findings have been observed in *Pengasius* and *Tor* sp. while studied along Pahang river, Malaysia (Akbar et al. 2015a, 2015b). Most probably these characters are related to differences in food preferences and swimming activities. Besides, Lim et al. (2014), who carried out a research on the genetic structure of the Japanese threadfin bream (*Nemipterus japonicus*) along the Peninsular Malaysian coast, reported that the morphological characters were changing more rapidly than genetic characters. However, the morphometric method is still considered as a useful tool for fish identification. The method has achieved remarkable success in identification of evolutionary related species and provides a strong foundation and starting point for all current work on fish taxonomy (Muchlisin 2013).

The discriminating variables (the specific measurements of body depth and caudal region) identified for *Plicofollis* species in the current study were considered as supplementary taxonomic information to be used in the field to more accurately identify its species. This is an indication that truss network measurements could be taken into consideration, where it could increase the consistency of individual fish species identification.

#### CONCLUSION

The correct field identification of species is crucial for accurate documentation of biological diversity and its ecological information. Overall, the information provided from discriminant analysis using truss network measurements throughout this study could be used as additional taxonomic evidence for the species description of *Plicofollis* species in order to avoid species misidentification. Furthermore, this technique is very cost effective and could be promoted as a tool for aiding in species identification, particularly for the *Plicofollis* species.

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