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Tandel Rutvikkumar P

Department of Fisheries Resource management, College of Fisheries science, Junagadh Agricultural University, Veraval, Gujarat, India

Deepak T Vaghela

College of Fisheries Science, Junagadh Agricultural University, Veraval, Gujarat, India

Sheetal K Bharda

Department of Fisheries Resource management, College of Fisheries science, Junagadh Agricultural University, Veraval, Gujarat, India

DV Bhola

College of Fisheries Science, Junagadh Agricultural University, Veraval, Gujarat, India

MR Patel

College of Fisheries Science, Navsari Agricultural University, Navsari, Gujarat, India

Corresponding Author: Tandel Rutvikkumar P Department of Fisheries Resource

management, College of Fisheries science, Junagadh Agricultural University, Veraval, Gujarat, India

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Tandel Rutvikkumar P, Deepak T Vaghela, Sheetal K Bharda, DV Bhola and MR Patel

Abstract

In present study, scale morphology and microstructure of *Priacanthus hamrur* from Veraval harbour, Gujarat, India was studied with using of Scanning Electron Microscope (SEM). To examine certain features on scale surface, scales from three body regions (Head scale, Belly scale and Caudal scale) were removed, cleaned and prepared for SEM analysis. The spinoid type cycloid scale recorded all over body, which is characteristics of this fish scale. Variation also recorded in scale shape, focus location, circuli shape and width, inter-circular space, minute dentition on circuli, granulation pattern on posterior field, presence of spine on posterior field. Absence of radii on anterior field of scale is considered as additional feature of this fish scale. In addition to qualitative and quantitative characteristics, the J-indices also employed to study relative scale size. The study concluded this scale microstructure can provide useful taxonomic information and had an immense utility in systematic classification of any fish species.

Keywords: Scale morphology, Priacanthus hamrur, veraval harbor

Introduction

Fish scale is a dermal derivatives of fish body, which served as versatile research material. (Esmaeili and Gholami, 2009) ^[9]. Agassiz was the first to divided fishes into four groups (Placoidei, Ganoidei, Ctenoidei and Cycloidei) based on scale morphology (Ansari *et al.*, 2016) ^[4]. From many years, fish scales utilize to understand age determination, growth rate, life span of fishes and past environment experienced by species (Masood *et al.*, 2015; Dapar *et al.*, 2012) ^[23, 6].

More recently, with development of Scanning electron microscope helps to study new features of scale, scale growth and development (Roberts, 1993)^[25] Scale characteristics; ctenii, radii, circuli and lateral canal used effectively in phylogeny, sexual dimorphism, migration period, pathology of fish scale due to water pollution and as bio-indicator of water quality (Esmaeili *et al.*, 2009; Ibanez *et al.*, 2009)^[9, 12, 18]. Many researchers studied variation in the types of scales among different species i.e., *Mugil cephalus, Neolissochilus hexastichus, Garaa Rossica, Channa* genus (Masood *et al.*, 2015; Esmaeili *et al.*, 2007; Ansari *et al.*, 2016; Dey *et al.*, 2014)^[23, 10, 4, 8]. With lot of importance of scale morphology, an attempt made to study scale ultrastructure of *P. hamrur*. The proposed fish species scale morphology has not studied yet, hence this research work was deliberated. The present paper is the first detail investigation on scale morphology of *P. hamrur* along Veraval harbor, Gujarat, India.

In India, *Priacanthidae* family is represented by six species, whereas 21 valid species from five genera recorded along worldwide. The major characteristics of this family are deep body, rough scales, extremely large eyes and bright orange red colour (Mohanty *et al.*, 2019) ^[24]. They are commonly known as Bull' eye or Big eyes (Anjanayappa *et al.*, 2013) ^[1]. The species of this family are epibenthic in habitat and usually found associated with rocks or coral reefs (Starnes, 1988) ^[26].

Methodology

Fish specimens were collected form the Veraval harbor (Lat. N $20^{0}34'613''$ and Long. E $70^{0}12'340''$), Gujarat during the period of September to February 2018. After collection, total length, standard length and weight of each specimen measured. To study the microstructure of scale, the scales were gently removed carefully with forceps from the three regions of the

body: Head scale (HS), Belly scale (BS) and Caudal scale (CS). Instantly, the scales were washed with distilled water and treated with 2% potassium hydroxide solution in order to remove debris and tissue adhering to scale. The cleaned scales were dehydrated in 30, 50, 70 and 90% ethanol for 10 minutes respectively and this scale were dried through kept between two micro-slides for 2-3 days. The cleaned and dried scale mounted on metallic stub and coated with the help of an Ion sputter (EMITEX) by maintaining adequate vacuum pressure for three and half hours. This gold coated scales were viewed under Scanning electron microscope (SEM) at an accelerating voltage of 15 kv at low probe current. Various SEM micrograph taken for further analysis.

Quantitative measurement

Scale width, length and other microstructures such as width of circuli, inter-circular space, spine size, lepidont width were measured. The scale width and length were recorded with using scanning electron micrograph. The j-indices for scale length and width using fish standard length was recorded (Brager and Moritz, 2016; Esmaeili *et al.*, 2014) ^[5, 11]. The arithmetic mean and standard deviation value used to come a variable conclusion.

Result & Discussion

The present study recorded spinoid types cycloid scale in all over body region of *Priacanthus hamrur* (Forsskal, 1775)^[3]. As per Roberts (1993)^[25] the spinoid cycloid type scale can define as spine projecting from the main body of the scale and restricted to the posterior margin or scattered over the posterior field. The pentagonal shaped scale with irregular

rounded in-and edentation recorded in this species. The scale of this species can divide into anterior (rostral), posterior (caudal) and lateral field. The ventral part of a scale is shiny and smooth whereas the dorsal part is rough due to presence of grooves, ridges and tubercles. Focus is the primary part of scale developed during ontogenesis. (Kaur and Dua, 2004; Esmaeili et al., 2007) ^[21, 10]. The focus located at postero-central field (fig.4). Granulation and distinct pre-circuli observed around focus field. The focus shape and position may vary in different species (Helfman *et al.*, 2009) ^[16].

Elevated ridges developed on surface of scale from the focus which known as circuli. The circuli may be convex or concave, continuous to discontinuous towards anterior field (Brager and Moritz, 2016)^[5]. In this species, convex, distinct and discontinuous type circuli recorded on anterior and lateral field. The convex shaped first inter-radial circuli noted in all sales of species (fig.1). The width of the anterior circuli is more than lateral circuli recorded in all scales (fig.2 & fig.3). The space between circuli known as inter-circular space. The inter-circular space is maximum in lateral field and minimum in anterior field, comparable observation recorded by Esmaeili and Gholami, 2009 [9, 12]. The maximum width of circuli and inter-circular space recorded in CS. The structure and arrangement of the circuli accountable for the shape formation of scale and focus circuli play a significant role in species specificity (Kaur and Dua, 2004) ^[21]. Variation also recorded in width of anterior and lateral circuli and intercircular space as mean value shown in Table 1. The distance between circuli denotes fast and slow growth period during different season (Fisher and Pearcy, 2005; Esmaeili et al., 2007) [13, 10]



Fig 1: SEM micrograph of the scales obtained from the three body regions of *P. hamrur* i.e., (a) HS (b) BS and (c) CS. Convex, distinct and discontinuous circuli and grooves on anterior field of all type scales. Circuli (C), 1st Inter-radial circuli (1st IRC), Grooves (G).

Small tooth or denticle like structure seen on circuli under high magnification called lepidonts which varies in shape and size. According to Jawad and Al-Jufaili (2007) ^[19] the characteristics of lepidonts play a significant role in species distinctness. Many authors observed different size and shape of lepidonts in many fish species (Lippitsch, 1990) ^[22]. DeLamater and Courtney (1974) ^[7] concluded that the taxa usually differentiate base on shape, size and orientation of lepidonts on circuli of the scale. There were growing lepidonts observed on anterior circuli (fig.2). The lateral circuli without lepidonts, whereas focus circuli with minute dentition recorded in all scales. However, the first inter-radial circuli without any dentition observed. In the anterior and lateral field, the circuli partitioned by deep and narrow grooves which pointing towards focus called radii, which represents line of scale flexibility. This radial structure absents on anterior radial orientation of all scale of this species during study (fig.1). Brager and Moritz (2016)^[5] similarly recorded spinoid type cycloid scale, distinct circuli and non-presence of radii in various species i.e. *Mora moro, Trichyrincus scabrous, Beryx decadactylus, Scorpaena porcus* along Mediterranean Sea.



Fig 2: SEM micrograph of the scales obtained from the three body regions of *P. hamrur* i.e., shows developing lepidonts (D.L) on circuli (a) HS (b) BS and (c) CS. Inter-circular space (D) between circuli on HS and CS. Anterior circuli (A.C), 1st Inter-circular space (D).



Fig 3: SEM micrograph of the scales obtained from the three body regions of *P. hamrur* i.e., shows lateral circuli of (a) HS (b) BS and (c) CS. Developing lepidont on lateral circuli of BS. Lateral circuli (L.C), Developing lepidonts (D).

The posterior field characterized by the granulation pattern; arrangement of segments and presence of spine or ctenii. It covered with several rows of pigmented tubercles, which formed by the aggregation of epithelial layer of the skin. The whole exposed part of posterior field covered with decomposed tubercles and spine (fig.4). This sub-marginal spine arranged in alternating rows towards the posterior rim. However, the maximum number of spines recorded in caudal scale (CS) and minimum number recorded in head scale (HS) of species. Small and blunt spine recorded in head scale (HS) compare to others (fig.5). The quantitative parameters of spine i.e., length, breadth and number of spines shown in Table 1. Starnes (1988) ^[26] recorded spinoid type cycloid scale in *Priacanthus blochii* and *Priacanthus meeki*. Alkaladi *et al.*, 2013 ^[3] revealed that the characteristics of posterior

(caudal) field of scale is vital in differentiate between species and genera of same family, and also at different families level. The pigmentation (tubercles) on the posterior field of scale is a characteristic feature of the cycloid and ctenoid scale of carps and perches (Johal and Agarwal, 1997; Esmaeili *et al.*, 2007) ^[20, 10]. Many authors recorded that qualitative characters such as circuli shape and arrangement, focus shape and position, lepiodonts shape, tubercles shape and ctenii or spine arrangement are species-specific as they are genetically fixed and more stable (Huges, 1981; Harabawy *et al.*, 2007; Harabawy *et al.*, 2012 and Al-Awadhi *et al.*, 2017) ^[17, 15, 14, 2]. The focal index value, J-indices value for relative scale length (Jsw) and for relative scale width (Jsl) are 0.501 1.145 and 1.420 respective.



Fig 4: SEM micrograph of the scales obtained from the three body regions of *P. hamrur* i.e., shows focus and posterior field with sub-marginal spine of (a) HS (b) BS and (c) CS. Spine (S), Focus (F).



Fig 5: SEM micrograph of the scales obtained from the three body regions of *P. hamrur* i.e., shows sub-marginal spine (S) scattered on posterior field of (a) HS (b) BS and (c) CS.

Table 1: Structural details of some scale microstructu	ire in	Р.	hamrur.
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Sr. no.	Characters	HS
1	Scale width mm	2.65±0.41 mm
2	Scale length mm	2.14±0.49 mm
3	Focus	
	a. Position	Postero-central
	b. Location (Distance from posterior margin) (mm)	1.05 ± 0.02
4	Anterior circuli	
	b. Average width µm	7.63±0.83 μm
	c. Average inter-circular space	0.70±0.67 μm
5	Lateral circuli	
	a. Average width	6.10±2.09 μm
	b. Average inter-circular space	1.16±1.10 μm
6	Lepidonts	Developing
7	Radii	Absent
8	Tubercles	Decomposed
9	Spine	
	a. Shape	Triangular
	b. Length	66.55±13.59 μm
	c. Breadth	18.01±3.15 µm
10	d. No	20

Average and standard deviation value shown in Table.1

Conclusion:

The above study revealed that qualitative characters such as pentagonal scale shape, focus position, convex type circuli, minute lepidonts bearing circuli, spine on caudal field are important criteria for this species characterization and discrimination. In present study, we also tried to evaluate quantitative characters such as JSI, JSW and focal index, which was not investigated earlier. Therefore, investigation prove that scale ultra-structures are promising tools for systematics study of fish species

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