Structure and dental sexual dimorphism in *Dasyatis hypostigma* (Santos & Carvalho, 2004) (Myliobatiformes, Dasyadae)

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In elasmobranchs sexual dimorphism is quite widespread, having differences such as variation in body size and on your teeth and spines, which are exclusive to mature males in some species. Dental sexual dimorphism is related to breeding and mating, where males held the females with their mouth. The importance of this work is to highlight the sexual heterodontia in lane *Dasyatis hypostigma*, stressing their structures by scanning electron microscopy. Dental arches of males and females were fixed in buffered 10% formal solution, processed and documented photo. The study confirmed the sexual heterodontia in this species; the females having their teeth are of type crusher, with rounded crowns and teeth in males where observed grabber type teeth, which are characterized by a sharp monocuspidate in the central region and with rounded crowns in the distal region. Both in females as in males have ornamentations filling the entire surface of the vestibular side of the crown. Structural and taxonomic information about the species are important for future comparative studies.

Keywords: teeth; mating; reproduction; feeding; scanning electron microscopy

1. Introduction

The evolutionary history of the Chondrichthyes (cartilaginous fish) date of at least 400 million years, with appearance in the Devonian period and diversification in two subclasses in the transition to the Carboniferous: the Holocephalii (chimeras) and Elasmobranchii (sharks and rays) [1]. The Batoidea (rays) diverged from its relatives sharks for nearly 180 million years ago, during the Jurassic period [2].

In elasmobranchs sexual dimorphism is quite widespread and common in batoid, having differences such as variation in body size and on your teeth and spines, which are unique to males mature [3-8].

Sexual Heterodontia in many cases, the teeth of mature males are different in their form of teeth of immature males and females [9-10], where the males are of type grabber and females are of type crusher [3.11]. The dimorphism is often found in the anterior teeth [10], related to breeding and mating, where males hold females with your mouth [4,6,8,13].

The stingrays of the family Dasyatidae, the order Myliobatiformes, comprise a total of six genera and more than 80 species, with several species not yet described, being most abundant in tropical waters [14]. Six species of *Dasyatis* have been reported from the southwestern Atlantic Ocean off the coast of Brazil: the southern stingray, *D. hypostigma* (Santos & Carvalho, 2004); *D. americana* (Hildebrand & Schroeder, 1928); the roughtail stingray, *D. centroura* (Mitchill, 1815); the wingfin stingray, *D. geijskesi* (Boeseman, 1948); the longnose stingray *D. guttata* (Bloch & Schneider, 1801); the Brazilian large-eyed stingray, *D. marianae* (Gomes, Rosa & Gadig, 2000); and the bluntnose stingray, *D. say* (Lesueur, 1817) [15].

The Groovebelly Stingray, *D. hypostigma* is a small species (~ 58 cm wide) of coastal and estuarine habit, previously confused with other species. They are found in the Southeast and South of Brazil, varying between the States of Espírito Santo to Rio Grande do Sul. Its northernmost limit can extend to the coast of Bahia, being captured in shallow waters, at depths ranging from 5 to 80 m, but typically within 5 to 40 m [15-16]. This species is often caught as bycatch in bottom trawls of shrimp and is used as a food source. These captures, along with the destruction and pollution of the coastal habitat, seem to be the main threat, but due to lack of studies the species is evaluated as "Data Deficient" by the red list of threatened species the IUCN [16].

The importance of this work is to highlight the sexual heterodontia in lane d. hypostigma, providing information for future comparative studies and mechanisms used in the feed.

2. Material and Methods

Dental arches were obtained from three males and four females (mature) and three males and three females (immature) of *Dasyatis hypostigma* species (Fig. 1). The specimens were captured incidentally in fishing pink shrimp, in the period from July to September 2013 in the Southeast and South of Brazil (23°-26°S e 42°-47°W). The specimens were donated by fishermen from trawling for pink shrimp to the fishing Institute/Santos, in partnership with the Department of Anatomy of domestic and wild animals of the Faculty of veterinary medicine and Zootechny of the University of São

Paulo (FMVZ-USP). This study was approved by the Ethics Committee on the use of Animals (CEUA) nº 5785050214, FMVZ-USP.

For scanning electron microscopy (SEM), the arcades were immersed in fixing solution of formaldehyde 10%. Then were washed in distilled water, subject to dehydration in growing series of alcohols and kiln-dried at 37°c. In the sequel the samples were positioned and mounted on aluminum metal bases and subjected to the metallic finish "sputting" with gold ions in the unit being analyzed EMITECH-K550 and photographed in scanning electron microscope LEO 435VP (FMVZ-USP).

The description of the teeth of the rays was based on nomenclature of Moss [17] e Cappetta [11].



Fig. 1 Dasyatis hypostigma streak male scheme. (A) dorsal view and (B) ventral view.

3. Results

3.1 Females

In immature females (Fig. 2A, B and C) and in the mature females (Fig. 2D, E and F) morphological structure and arrangement of teeth is the same and there is only an increase of size of the teeth in mature females.

In the central region of the lower jaw there is an increase in the amount of teeth, being a wider region (Fig. 2A). The teeth are of type crusher, with rounded crowns. On transverse depression of the teeth, on the face of the Crown vestibular, ornamentation of rounded shape (Fig. 2C and F), which fill the entire surface irregular. No differences were



Fig. 2 Scanning electron microscopy. Teeth of *Dasyatis hypostigma* streak females. (A, B and C) lower jaw of the juvenile female. (A) the central region of the lower jaw is wider, with an increase in the amount of teeth (ellipse). (D, E and F) adult female upper jaw. Teeth crusher type, with rounded crowns. Arrows indicate some ornamentation on the vestibular crown. Scale bar: (A e D): 1 mm; (B e E): 300μ m; (C e F): 100μ m.



Fig. 3 Scanning electron microscopy. Teeth of males of *Dasyatis hypostigma* streak. (A-C) lower jaw teeth of juvenile male, of type crusher, with rounded crowns. (D-F) and lower jaw (G-I) mature male upper jaw, teeth, grabber type characterized by a sharp, monocuspidate and with rounded crowns. Teeth with cusp becomes milder in the lateral teeth, until the distal region (teeth with rounded crowns). Ornamentations in the vestibular surface of the crown (arrows); a groove (g) dividing the cusp to the middle. Scale bar: (A, B, D, E, F, G e H): 1 mm; (C): 300µm; (I): 100µm.

3.2 Males

In males as in females, in the central region of the lower jaw there is presence of more teeth (Fig. 3A and E), a wider region. In young males (Fig. 3A, B and C) are similar to teeth found in females: the type crusher, with rounded crowns with adornments on the vestibular side (Fig. 3C) by completing all the uneven surface.

In mature males the teeth are of the type with a grabber cusp and with rounded crowns. In the sinfisial region of the jaws (Fig. 3E and H), the teeth are more prominent and pointed cusps becoming smoother in the lateral teeth (Fig. 3D and F), until the distal region (Fig. 3H), where the teeth have rounded crowns.

The lower teeth with a cusp are triangular format (Fig. 3E), with more rounded tips, his superiors have sharp tips (Fig. 3H). The apexes are geared toward the side opposite to the symphysis and lingual region (Fig. 3D, F, G and H). As well as in females, all teeth have vestibular surface ornamentation of the crown. In the center of the teeth with a cusp, a groove dividing the cusp to the middle (Fig. 3I).

4. Discussion

The streak *D. hypostigma* has dental pronounced sexual dimorphism, as noted by Sanots and De Carvalho [15] and in other stingrays of the family Dasyatidae already parsed [5, 6, 8.18]. In this study noted that the teeth in mature females and juvenile males have rounded crowns and in sexually mature males are with cusp and pointed. In males the teeth of the first lines of each side of the series are rounded as in juvenile males and females, but the centerlines of the maxilla are elongated, later projecting cusps. The dental formula for juveniles (over 300 mm wide) and adults is 37-46/43-50 [15].

Dental sexual dimorphism is suggested by two leading theories based on the difference in feeding and reproduction. Feduccia and Slaughter [3] suggested that sexual dimorphism is due to differences in feeding preference and use of niche, preventing intraspecific competition for food. However, this was disputed by McEachran [19] after examining samples of the stomach contents of more than 1600 rays, known to have sexually dimorphic dentition, where did not find significant differences in the foods consumed between sexes for immature or mature specimens.

The most plausible explanation and used in the literature is that sexual dimorphism is important dental in reproductive behavior. To have internal fertilization, pre-copulatory activities involve actions that result in male grasping the female in order to provide a proper alignment for inserting a clasper (copulatory organ) into the female [20]. In many sharks and rays males hold orally female fins, bite marks that are less tenacious than used for food and generally do not employ full power or complete closure of the jaw [20].

Taniuchi and Shimizu [5] and Nordell [6] present studies showing that in the teeth of male dimorphism of Dasyatis akajei and Urolophus halleri feature a static form. Second Kajiura and Tricas [8], the males of *D. sabina* feature an adaptation on dentition to increase reproductive efficiency during the mating season, where the mature males dentition is modified to more pronounced cusps seasonally, and returning to the state after that period, crusher since they are more functional in feeding than the dentition of type, grabber with cusps.

5. Conclusion

The study of the teeth of *Dasyatis hypostigma* streak by scanning electronic microscopy allowed analyze structures and variations of the dentition in female and juvenile and mature males, in addition to the attestation of sexual heterodont in this species. Only the anatomical study is not sufficient to determine the mechanisms used in the feed, but back important structural information for future comparative studies. Seasonal analyses are necessary to establish that the male of this species develops teeth only during the mating season, since this dentition is not best suited for their food, according to Kajiura and Tricas [8], composed mainly of polychaetes, molluses, fish and crustaceans [21].

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