Arquivos de Ciências do Mar

COMPARATIVE MORPHOLOGY AND IDENTIFICATION OF EGG CAPSULES OF SKATE SPECIES OF THE GENERA *Atlantoraja* MENNI, 1972, Rioraja WHITLEY, 1939 AND *Sympterygia* MÜLLER & HENLE, 1837

Morfologia comparativa e identificação de cápsulas do ovo das espécies de raias dos gêneros *Atlantoraja* Menni, 1972, Rioraja Whitley, 1939 e *Sympterygia* Müller & Henle, 1837

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ABSTRACT

A comparative study of the morphology of the egg capsule for six species of skates endemic to the southwestern Atlantic Ocean was carried out through literature review and analysis of new data. Egg capsules of Sympterygia acuta *and* S. bonapartii *differ from those of genera* Atlantoraja *and* Rioraja *by their elongated, tendril-like posterior horns and their flat lateral margins. Egg capsules of the two* Sympterygia *species that occurring in the area in question differ from each other in size. In lateral view the egg capsule of* Rioraja agassizi *has convex ventral and dorsal faces, whereas in the three species of* Atlantoraja *the ventral face is flat. Within the genus* Atlantoraja *the most important taxonomical features for the identification of the capsules are the surface texture, the morphology of the velum and the capsule dimensions. The presence and location of attachment fibres is also an important character for capsules identification. Based on the aforementioned identification characteristics, a key to species for egg capsules of the six species is presented.*

Key Words: *Rajidae, egg capsule, taxonomy, phylogeny, batoid.*

RESUMO

Um estudo comparativo da morfologia das cápsulas ovígeras para seis espécies de raias endêmicas do Atlântico Sudocidental através de revisão de literatura e analise de novos dados é apresentado neste trabalho. As cápsulas ovígeras de Sympterygia acuta *e* S. bonapartii *diferem daquelas dos gêneros* Atlantoraja *e* Rioraja *pelos seus chifres posteriores alongados em forma de gavinhas e pelas suas margens laterais planas. As cápsulas ovígeras das duas espécies do gênero* Sympterygia *(que ocorrem na área de estudo) diferem uma da outra pelo tamanho. Em vista lateral, a cápsula ovígera de Rioraja agassizi possui as faces ventral e dorsal convexas, entanto que nas três espécies de* Atlantoraja *a face ventral é achatada. Dentro do gênero* Atlantoraja*, as características taxonômicas mais importantes na identificação das cápsulas são a textura da superfície, a morfologia do velum e as dimensões da cápsula. A presença e disposição das fibras de adesão são também características importantes para a identificação. Em base das características de identificação mencionadas, uma chave de identificação de cápsulas ovigeras para as seis espécies é apresentada.*

Palavras-chave: *Rajidae, cápsulas ovígeras, taxonomia, filogenia, batoideo.*

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INTRODUCTION

Skates are oviparous and deposit their eggs on the sea bottom where development takes place without parental care until hatching (Ishiyama, 1958). The variation in the morphology and external characteristics of the rajid egg capsule may be due to environmental adaptation, in conformity with the functional roles of the capsule (Ishiyama, 1958). This is why elasmobranch egg capsules can be identified at the level of genus and species, while the variation between taxa of the morphology of the egg capsule also reflects phylogenetic relationships in skates (Hubbs & Ishiyama, 1968). The description of the egg capsule is relevant for the study of the distribution and reproductive biology of skates, especially of species that inhabit shallow coastal waters and whose egg capsules are therefore frequently washed ashore (Ishiyama 1958; Hubbs & Ishiyama 1968; Oddone *et al*. 2004).

The genus *Atlantoraja* Menni, 1972 comprises three species, the Spotback skate *A. castelnaui* (Ribeiro, 1907), the Eyespot skate *A. cyclophora* (Regan, 1903) and the La Plata skate *A. platana* (Günther, 1880). The genus *Rioraja* Whitley, 1939, contains a single species, the Rio skate *R. agassizi* (Müller & Henle, 1841). Both genera are endemic to the continental shelf off the southern Atlantic coast of South America. Two species of the genus *Sympterygia* Müller & Henle, 1837 the Bignose fanskate *S. acuta* Garman 1887 and the Smallnose fanskate *S. bonapartii* Müller & Henle, 1841 are also endemic to that shelf area (Compagno, 2005). Egg capsules of skates of the southwestern Atlantic have been described for *Atlantoraja castelnaui*, *A. platana, A. cyclophora, Rioraja agassizi, Sympterygia* acuta, *S. bonapartii* and *Psammobatis* extenta, *P. normani, P. rudis and P. bergi* (Oddone & Vooren, 2002; Braccini & Chiaramonte, 2002; Mabragaña *et al*., 2002; Mabragaña & Cousseau, 2004; Oddone *et al.* 2004; Oddone, 2005; San Martin *et al.*, 2005; Oddone *et al.* 2006; Oddone *et al.*, 2008).

The aim of the present paper is to integrate published descriptions of egg capsules along with new data recorded in order to discuss the comparative morphology of the egg capsules genera *Atlantoraja*, *Rioraja* and *Sympterygia* in the southwest Atlantic in relation with taxonomy and phylogeny. A key to egg capsules is presented.

MATERIAL AND METHODS

Egg-bearing females of *A. cyclophora* and *A. platana* were caught in August-September 2001 and March-April 2002 off South Brazil, between latitudes 30°S and 35°S at depths of 100-300 m (Figure 1).

Of *A. castelnaui* and *R. agassizi,* egg-bearing females were obtained by commercial bottom trawler in April 2005 and April 2006 off the Southeast coast of Brazil, between latitudes 23°37'S and 27°40'S, at depths of 10-146 m (Figure 1).

Of *Sympterygia acuta e S. bonapartii*, only published descriptions of the egg capsules were used (Table 2). Drawings of the egg capsules of *S. bonapartii* were made from samples collected by first author on Cassino Beach (32°10'S; 52°05'W), South Brazil, in 2001.

Freshly collected egg capsules were fixed in 10 % formalin and preserved in 70% ethanol. From each egg capsule were: recorded total length (excluding horns), total width, total length of

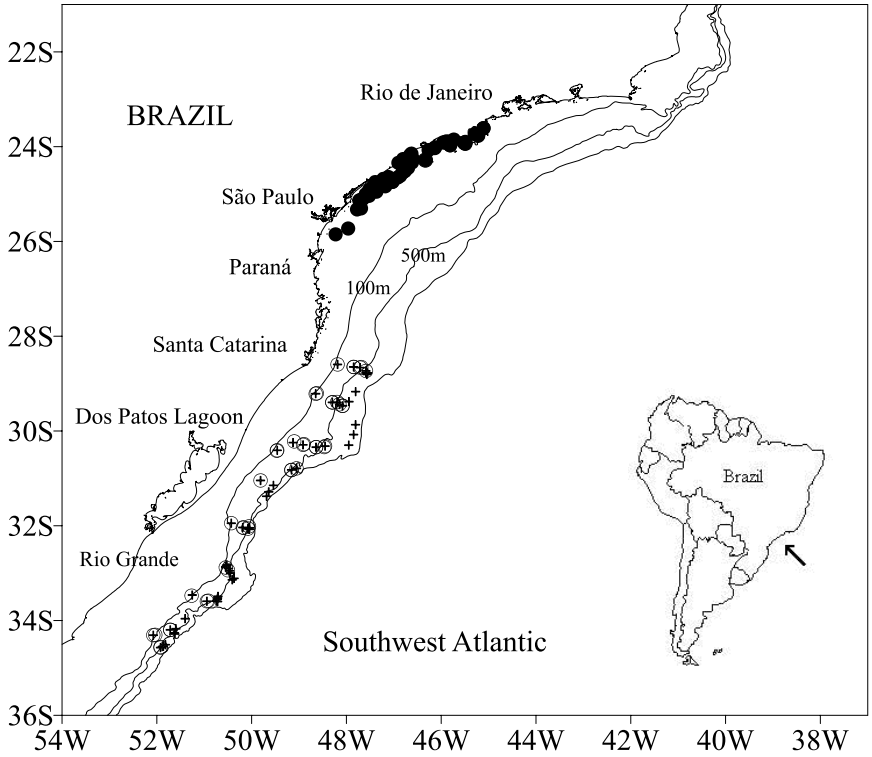

Figure 1 - Map of the study area: South and Southeast Brazil, showing the trawling stations from where samples of *Rioraja agassizi*, *Atlrantoraja castelnaui* (full circles), *A. platana* and *A. cyclophora* (empty circles) came from.

anterior and posterior horns, height, width and thickness of the lateral keel, colour, texture of the surface (texture) and presence of adhesion fibrils. The velum length was measured at it widest central area. Measurements were made as defined by Hubbs & Ishiyama (1968), Templeman (198) and Gomes & Carvalho (1995), with precision of 0.1 mm, with Vernier callipers.

RESULTS

The egg capsules of *Sympterygia acuta* and *S. bonapartii* have the same basic morphology but mean length and width of the capsule of *S. acuta* are 62% of those of *S. bonapartii* (Tables 1 and 2, Figure. 2A and 2C). The main characters that can be used for distinguishing between the genus *Sympterygia* and the genera *Atlantoraja* and *Rioraja* were the form of the posterior extensions and of the lateral margin of the capsule (Table 1, Figure 2A). The posterior extensions of the capsule are rigid and horn-shaped in *A. castelnaui*, *A. platana*, *A. cyclophora* and *Rioraja agassizi*, with length of up 97, 106, 176 and 104% of the capsule length respectively, while in *Sympterygia* those extensions are flexible and have the shape of flexible tendrils, with length of up to 917% of capsule length (Oddone & Vooren, 2001; Figure 2A).

In *Atlantoraja* and *Rioraja* the lateral margins of the capsule have the form of a keel that varies between species in width and thickness (Table 2). In *Sympterygia* the lateral margins of the capsule have the shape of a flat and smooth flange with its surface in the sagittal plane of the capsule (Tables 1 and 2).

Atlantoraja and *Rioraja* differ in the profile of the capsule in lateral view (Figure 3). In *Atlantoraja* the dorsal face of the capsule is convex and the ventral face is flat the latter in *A. castelnaui* and *A. platana*, while in *A. cyclophora* the ventral face is only slightly convex in comparison with the with the dorsal face (Figure 3A-3C). In *Rioraja* the ventral and dorsal faces of the capsule are markedly and similarly convex (3D).

The morphology of the posterior horns in *Atlantoraja* and *Rioraja*, as well as the ratio posterior/anterior horns, is also useful in the species discrimination. In *A. platana* and *A. cyclophora* the anterior horns are curved inwardly and crescent-shaped, in *A. castelnaui* the curvature of the anterior horns is less pronounced (3 A-C), and in *R. agassizi* the anterior horns are almost straight (Figure 3D).

The ratio posterior/anterior horns differ among species, varying from 1.4 in *R. agassizi* to 15.7 in *S. acuta* (Table 2). The egg capsule of *A. platana* can be distinguished from that of *A. cyclophora* by the length of the posterior horns, which are larger longer in *A. platana* (Figure 3A and 3B).

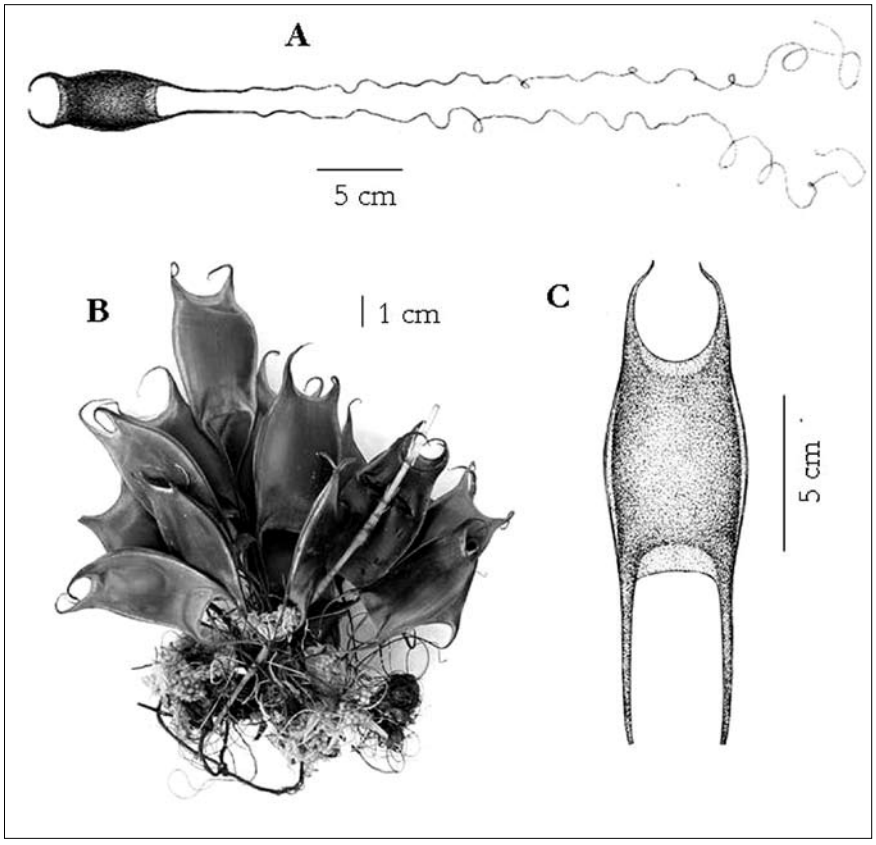

Figure 2 - Drawings of an egg capsule of *Sympterygia acuta* extracted in uterus from a female donated to the authors by fishermen in 2001 (A); photography of a typical deposition of egg capsules of *S. acuta*, reproduced from Oddone & Vooren (2002), part of the substrate to which capsules are attached can be observed (B); and drawing of an empty (hatched) egg capsule of *S. bonapartii* in front view collected in Cassino Beach by the first author in 2001 (C).

Table I - Egg capsule characteristics for *Rioraja agassizi, Atlantoraja castelnaui, A. platana, A. cyclophora, Sympterygia bonapartii* and *S. acuta;* horns morphology, shape in lateral view, lateral margins characteristics, coloration, texture of the surface, location of the attachment fibrils and morphology of the posterior horns. Based in Oddone & Vooren (2002); Mabragaña et al. (2002), Oddone et al. (2004, 2006 and 2008).

Capsule characters	*R. agassizi*	*A. castelnaui*	*A. platana*	*A. cyclophora*	*S. bonapartii*	*S. acuta*
Horns	two pairs of posterior and anterior horns	two pairs of posterior and anterior horns	two pairs of posterior and anterior horns	two pairs of posterior and anterior horns	a pair of anterior horns and a pair of posterior tendrils	a pair of anterior horns and a pair of posterior tendrils
Lateral view	dorsal and ventral faces equally convex	dorsal face convex, ventral face flattened	dorsal face convex, ventral face flattened	dorsal face convex, ventral face slightly convex, almost flattened	dorsal and ventral faces equally convex	dorsal and ventral faces equally convex
Lateral Margins	With lateral keel	with lateral keel	with lateral keel	with lateral keel	smooth and flat	smooth and flat
Colouration	bright brown	shiny medium-brown	shiny medium-brown	shiny medium-brown	amber	metallic-green olive
Surface configuration	smooth	smooth	rough, marked longitudinal striation	rough, marked longitudinal striation	smooth	smooth
Location of the attachment fibrils	ventral face, horn base	ventral face, horn base	ventral face, horn base	ventral face, horn base	tendril-like posterior horns	tendril-like posterior horns
Posterior horns morphology	straight	slightly curved inward	half-moon shape, curved inward	half-moon shape, curved inward	tendril-like	tendril-like

Within the genus *Atlantoraja*, the texture of the surface of the capsule differs between species. The egg capsules of *A. cyclophora* and *A. platana* (Figure 3 A-B and 4 A) have a marked longitudinal striation that makes the capsule rough to the touch and that is absent in *A. castelnaui* (Figure 4 B), whose capsule is smooth to the touch (Figure 3 C) as well as in *Rioraja agassizi* (Figure 4 C).

In addition, egg capsules of *A. castelnaui* can be easily distinguished from the two above congenerics because of their dimensions (Table 2). Capsule length and width are respectively 89-105 mm and 70-76 mm in *A. castelnaui*, while those measurements are 66-74 mm and 41-47 mm in *A. platana* and 62-74 mm and 32-48 mm in *A. cyclophora*.

The egg capsules of *A. platana* and *A. cyclophora* differ in the form of the posterior velum. The margin of the velum is markedly convex in *A. cyclophora*, *and* almost straight or even slightly concave in *A. platana* (Figure 3 A and 3 B).

The disposition of the attachment fibres is also taxonomically important. In the genus *Sympterygia*, attachment fibres occur only along the entire length of the tendril-like posterior horns (Figure 2 A and 2 B). In *Atlantoraja* and *Rioraja*, attachment fibres are situated along the lateral margins and over the entire ventral surface of the capsule and only at the base of the posterior and anterior horns (Figure 3).

According to the aforementioned characteristics, the identification key of egg capsules of species of genera *Atlantoraja*, *Rioraja* and *Sympterygia* (just for the two species endemic to the southwestern Atlantic) is presented below.

1. a. Posterior horns thin, flexible and tendril-like. Lateral margin of the capsule in the form of a smooth flange with its surface in the sagittal plane…….....…….....2
2. a. length of capsule about 50 mm…….....***Sympterygia acuta***
2. b. length of capsule of about 80 mm…….....…… ***Sympterygia bonapartii***

1. b. Posterior horns rigid, not tendril-like. Lateral margin of the capsule in the form of a keel in the frontal plane……..…….....…….3
3. a. Dorsal face convex, ventral face flat or slightly concave…….....….....……..4

4. a. Capsule surface without longitud inal striation, smooth to touch….....….***Atlantoraja castelnaui***
4. b. Surface longitudinally striated, rough to touch…….....……..…….5
5. a. Dorsal face of the capsule asymmetrically convex, with the highest point situated toward the anterior end; edge of velum straight or slightly concave….....… ***Atlantoraja platana***
5. b. Dorsal face of the capsule symmetrically convex with the highest point situated centrally; edge of velum markedly convex.....***Atlantoraja cyclophora***

3. b. Dorsal and ventral face of the capsule equally convex……....……...***Rioraja agassizi***

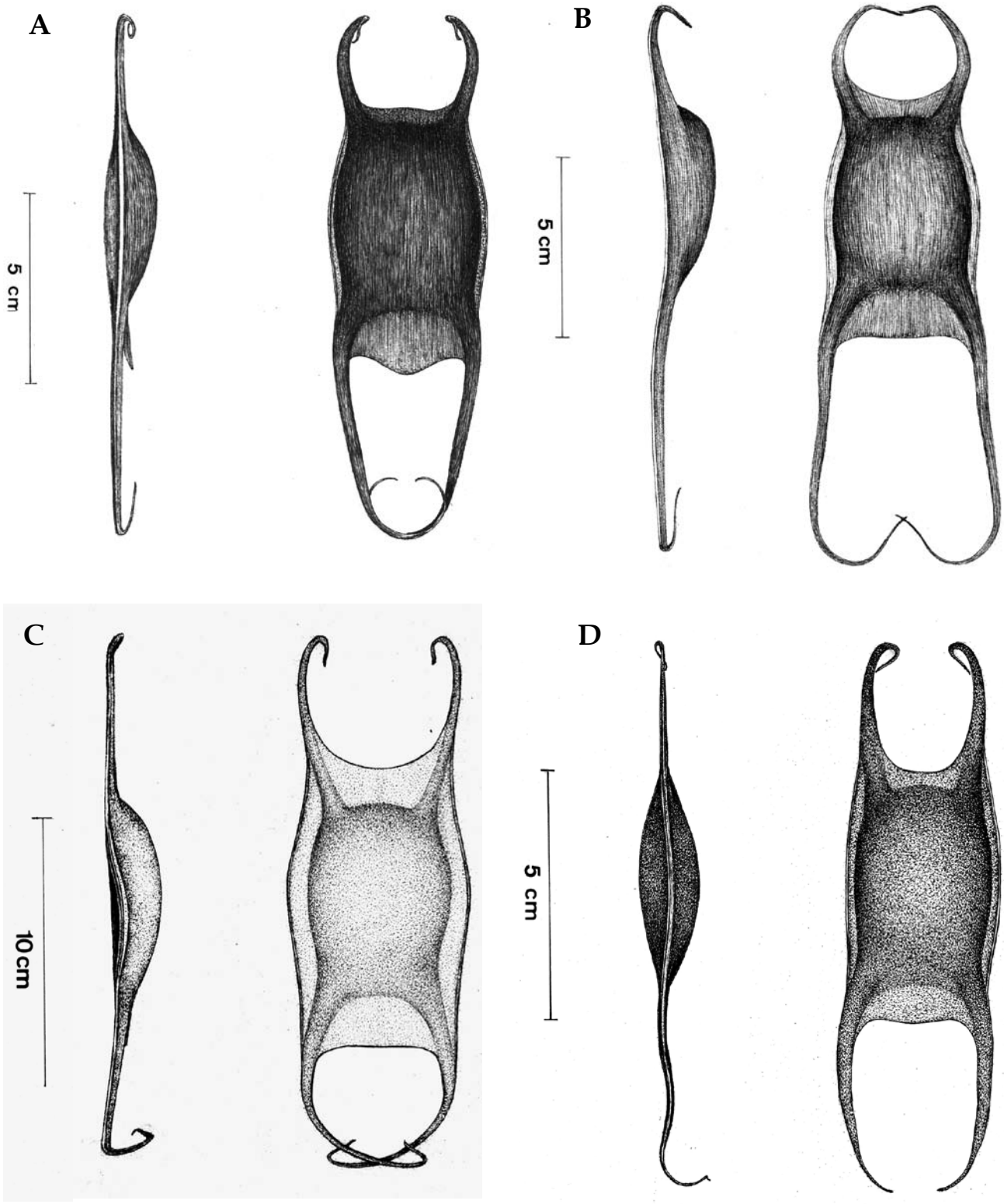

Figure 3 - Drawings of the egg capsules of *Atlantoraja cyclophora* (A); *A. platana* (B); *A. castelnaui* (C); and *Rioraja agassizi* (D) in front view (above) and lateral view (below), based on capsules collected in uterus, fixed, preserved and deposited at the first author's collection.

Table II - Measurements (mm) for egg capsules off *Rioraja agassizi, Atlantoraja castelnaui, A. platana, A. cyclophora, Sympterygia bonapartii* and *S. acuta* from South and Southeast Brazil, Southwest Atlantic Ocean, available in the literature. Characteristics considered: are coloration (after being extracted from uteri), total length (TL), total width (TW), height (H), anterior horns length (AHL), posterior horns length (PHL), ratio posterior horns/anterior horns (Ratio), lateral margin dimensions (Dimension), lateral margin thickness (Thickness), surface texture (Surface), sample size (n), velum, position of adhesion fibres (AF) and literature reference from where data were obtained (Reference).

Species	***R. agassizi***	***A. castelnaui***	***A. platana***	***A. cyclophora***	***S. bonapartii***	***S. acuta***
Coloration	yellow brown	light brown	light brown	light brown	amber	green olive
TL	41-56	89-105	66-74	62-74	76.75±3.92	45-50
TW	22-36	70-77	41-47	32-48	48·37±0·74	27-33
H	7-15	14-19	-	-	-	-
AHL	15-53	57-65	31-49	22-44	-	24-35
PHL	35-68	85-101	85-125	65-98	-	up to 440****
Ratio	1.4	1.6	2.7	2.4	-	~15.7
Lateral Margin	keel	keel	keel	keel	flange	flange
Dimension	1-2	5-9	1-5	1-2	*	~2.0
Thickness	1-3	1.5-2.5	1-3	1-2	*	-
Surface	smooth	smooth	striated	striated	smooth	smooth***
n	119	5	19	19	8	10
Velum	3-10	18-21	-	-	-	-
AF	ventral face, horn base	ventral face, horn base	ventral face, horn base	ventral face, horn base	tendril-like posterior horns**	tendril-like posterior horns
Reference	Oddone *et al.* (2006)	Oddone *et al.* (2008)	Oddone *et al.* (2004)	Oddone *et al.* (2004)	Mabragaña *et al.* (2002)	Oddone & Vooren (2002)

(-)=no data

* truncated, forming a vertical plane, no measurement available

**Oddone, pers. obs.

***though with a delicate striation

****a single complete capsule with complete posterior horns in record

DISCUSSION

Egg capsules equipped with coiled tendril-like posterior horns as observed in the genus *Sympterygia* also occur in cat sharks (Scyliorhinidae), but in those sharks in the anterior horns are also tendril-like. The only possible way to obtain a precise measurement of the length of the posterior tendril-like horns of *Sympterygia* egg cases is by extracting a complete egg capsule from the egg-bearing female (Figure. 2 A), because the posterior horns of the *Sympterygia* egg case are firmly entangled in the substrate on which the egg is laid (Figure. 2 B), so that is difficult to remove the egg case from the substrate without breaking its posterior horns (Oddone & Vooren, 2002). Female cat sharks winds these tendrils around a suitable object during egg laying (Hamlett and Koob 1999) and this egg laying mode may occur in *Sympterygia.* In other skate genera such as *Atlantoraja* and *Rioraja*, the attachment fibres presumably attach the egg case to solid substrates, or particles of sediment may be caught in those fibres, thus anchoring the egg case to a substrate of sand or mud.

The presence of tendril-like posterior horns not only serves for distinguishing *Sympterygia* from *Rioraja* and *Atlantoraja* but from all other skate genera occurring in the area. This is because tendril-like posterior horns are common in egg capsules from cat sharks but among rajids occurs only in the genus *Sympterygia*. Egg capsules of *S. lima* measure about 54 x 47 mm with length of posterior horns about 400 mm, the latter being only valid for *S. brevicaudata* (F. Concha, pers. comm.).

In Chile egg capsules of *S. lima* and *S. brevicaudata* are found in aggregations with egg capsules of cat sharks (Scyliorhinidae) such as

Schroederichthys chilensis (Guichenot, 1848) and that the egg capsules of *S. brevicaudata* are more abundant numerically, being also found entangled in algae as *Durvillaea antarctica* and *Lessonia nigrescens* (F. Concha, pers. comm.[1]). In Brazil, egg capsules of *Sympterigia acuta* e *S. bonapartii* are usually washed ashore or caught by bottom trawl together with the substrate to which they are attached. Typically the substrate is a mass of material composed mainly of stalks of plants from the coastal marshes but also containing tubes of sessile polychaetes and man-made materials such as plastics, pieces of fishing nets and twine. Many egg capsules are attached to a single mass of substrate, with the posterior tendrils of the capsules firmly intertwined with the material of the substrate which thus becomes a consolidated bundle of material that lies on the sandy or muddy sea bottom (Oddone & Vooren, 2002; Figure 4). It is not known how the female makes the tendril of the egg capsule intertwine with the substrate. The long tendril-like extensions of the egg capsule of the *Sympterygia* skates are the factor that enables those fishes to attach the egg capsule to the aforementioned type of substrate. Thus those skates are able to perform their entire life cycle in a habitat of smooth bottom of fine-grained sediment where no other substrate is available for the anchoring of the egg capsule.

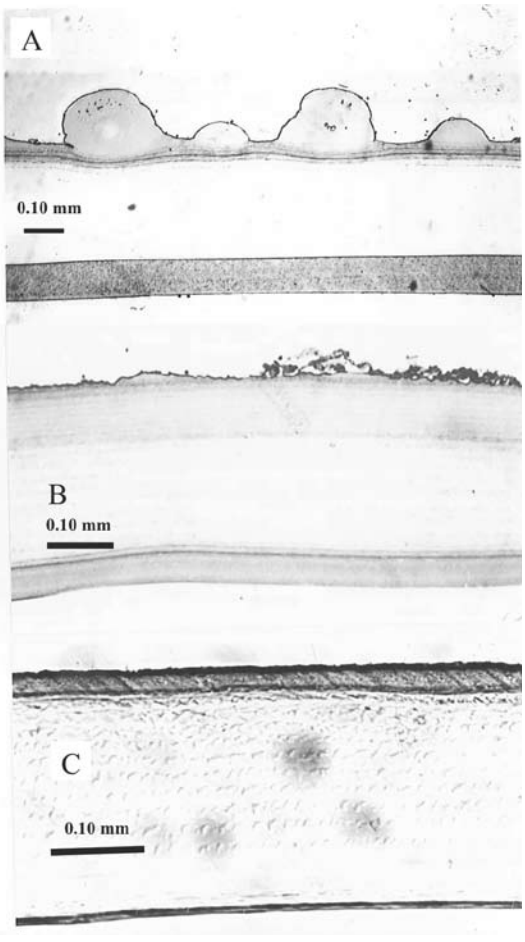

Figure 4 - Comparative microscopical structure (100X) of the dorsal face (transversal section) of two *Atlantoraja* species with different surface texture, i.e., (A) an species with striated surface, *Atlantoraja platana* (A) and another one with soft, non-striated surface, *A. castelnaui* (B) and *Rioraja agassizi* with smooth non-striated surface configuration.

For the Southwest Atlantic, the present key may aid in the identification not only of skate egg capsules collected in utero but also of skate egg capsules found washed ashore on beaches of the southwest Atlantic. On Cassino Beach in south Brazil for instance, egg capsules of *Sympterigia acuta* and *S. bonapartii* containing live embryos are frequently washed ashore (Queiroz, 1986; Brandt, 2005), and the monitoring of seasonal variation of such a phenomenon may aid in the study of the breeding cycle of coastal skates.

For the other genera occurring in the southwest Atlantic, Braccini and Chiaramonte (2002), Mabragaña and Cousseau (2004) and San Martin *et al.* (2005) provided descriptions of the egg capsules of *Psammobatis extenta*, *P. normani*, *P. rudis* and *P. bergi* for the Argentinean sea. In *P. bergii* egg capsule length varied from 25.5 to 29.2 cm and width from 16.2 to 21.1 mm (San Martin *et al.*, 2005). Mean egg capsule length (with standard deviation) and width for *P. rudis* and *P. normani* were 55.75±2.8 and 32.5±1.8; 45.36±5.59 and 31.5±3.93. For *P. extenta* capsule width and length varied between 16.2-21.1 and 25.7-29.2 mm respectively. Egg capsules of *Psammobatis* are smooth to touch though present some striation under magnification and both faces are convex in lateral view, just as in *Sympterygia*, *A. castelnaui* and *R. agassizi*. At least in *P. normani* and *P. rudis* capsule edges are flattened, as in *Sympterygia.* In *P. extenta* capsule wall is so delicate that the fertilized oocyte can bee seen by transparency. For the Brazilian coast descriptions for *Psammobatis* are lacking.

For comparison with skate egg capsules from other species and regions, Bigelow and Shroeder (1953) provided descriptions of egg capsules of *Leucoraja erinacea* for the area of Wood Hole, Massachusetts, sharing this capsules characteristics

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like general morphology and size with those of similar in *R. agassizi* even regarding the two convex faces. Description of the egg capsules of *Raja eglanteria* by these authors indicated that capsules of this species would have more in common with capsules of genus *Bathyraja,* that with the genera in question, as markedly short anterior and posterior horns and rounded sides on upper ventral or dorsal view.

Egg capsules of *Dipturus chilensis* off Uruguay have a length of about 136 mm, presenting longitudinal striation dorsally and ventrally (though to touch are smooth) (Paesch and Oddone, 2008 b). For genus *Bathyraja* in the southwest Atlantic, Paesch and Oddone (2008 a) described the egg capsules of *B. macloviana* and *B. brachyurops*. These egg capsules display different degrees of roughness in the surfaces and capsules of *B. macloviana* are equipped with microscopical prickles situated along the longitudinal striation, which has not been observed in other genera of the Southwest Atlantic. Capsule size ranged from 79 to 91 mm in *B. brachyurops* and from 69 to 75.5 mm in *B. macloviana.* Flat edges were also observed, and a straight and transverse velum was present in both species egg capsules.

In the southwest Atlantic, the egg capsules of the genera *Dipturus* and *Bathyraja* have a groove along their lateral edges, and can from this feature be distinguished from the egg capsules of *Rioraja* and *Atlantoraja.*

In the published keys for the identification of skate capsules from Japan, the North Sea and the North Pacific, the following morphological capsule characters are used: presence and width of the lateral keel; texture of the capsule surface; shape, robustness and size of the horns, position of attachment fibres; capsule size and colour of the egg-chamber (Ishiyama, 1958; Bor, 1998; Ebert, 2005; Ebert and Davis, 2007). Almost all these capsule characters (with exception of robustness of the horns and colour of the egg-chamber) are also used in the present key for *Rioraja*, *Atlantoraja* e *Sympterygia* from the southwest Atlantic, and are thus useful characters for the identification of skate egg capsules in general.

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