ABSTRACT
Dicephalic individuals originate from a dualistic fusion, where the formation of two embryonic bodies occurs by a single blastoderm or by the joining of two distinct blastoderms of the same yolk. The morphometric data are expressed in percentages of total length-TL and were compared with others found in the literature. The dicephalous, newborn dogfish captured, was identified as *Squalus acanthias*, and was caught by a seabob-shrimp throw net, off the coast of Peruibe City e Itanhaém, São Paulo State, Brazil. The specimen was a male with 23.5 cm (right animal) and 22.8 cm (left animal) TL, and 800 g of total weight. Externally, the animal presented a lateral fusion and concerning
the duplication of the head, they were separated until the fifth branchial arch. The exemplar presented: two pairs of first and second dorsal fins with spines; two pairs of pectoral fins; one pair of pelvic fins; one pair of claspers; and one caudal fin, with two superior lobes and one inferior lobe. The internal morphology displayed: two oral cavities; two esophagi; two stomachs (right stomach more developed); two livers (three lobules each); two hearts; one intestine; and one cloaca. The specimen can be considered the first record of dicephaly in S. acanthias.

Keywords: Abnormal, shark, embryonic, diprosopus, captured dogfish, siamesetwins

RESUMO
Os indivíduos dicefálicos se originam de uma fusão dualística, onde a formação de dois corpos embrionários ocorre por um único blastoderme ou pela união de dois blastodermas distintas da mesma gema. Os dados morfométricos são expressos em porcentagens do comprimento total-CT e foram comparados com outros encontrados na literatura. O cação frango bicéfalo recém-capturado foi identificado como Squalus acanthias e foi capturado por uma rede de arrasto de camarões, na costa da cidade de Peruíbe e Itanhaém, Estado de São Paulo, Brasil. O espécime era um macho com 23,5 cm (animal direito) e 22,8 cm (animal esquerdo) LT e 800 g de peso total. Externamente, o animal apresentava fusão lateral e quanto à duplicação da cabeça, foram separados até o quinto arco brânquial. O exemplar apresentou: dois pares de nadadeiras dorsais com espinhos; dois pares de nadadeiras pélvicas; um par de cláspers; e uma nadadeira caudal, com dois lobos superiores e um inferior. A morfologia interna apresentou: duas cavidades orais; dois esôfagos; dois estômagos (estômago direito mais desenvolvido); dois figados (três lóbulos cada); dois corações; um intestino; e uma cloaca. O espécime pode ser considerado o primeiro registro de dicefalia em S. acanthias.

Palavras-chave: Anomalias, tubarão, embrionário, dipróspus, cação frango, neonatos.

1 INTRODUCTION
Congenital anomalies may be modifications or malformations that occur during embryogenesis and may alter structures or functions of the systems. Congenital disorders can occur from minor changes, fundamental changes and aberrations. Apparently, there are no definite reasons, they can be caused by environmental or genetic factors, or both, however, it is considered a rare event (Smith, 2006).

The deformations in elasmobranchs are common, presented by several authors (Bottaro et al., 2005; Saidi et al., 2006). Such as mutations in pectoral fins (Ford, 1930; Blanco-Parra & Niño-Torres, 2011; Mejía-Falla et al., 2011; Ribeiro-Prado et al., 2008) and anomalies in the spine (Ford, 1930; Carrasco & León, 1984; Mancini et al., 2006), in the head (Escobar-Sánchez et al., 2009), and in the mouth (Ford, 1930), diprosopus (Bosinceano, 1934; Hevia-Hormazábal et al., 2011; Muñoz-Ozório et al., 2013) and dicephaly cases (Lozano-Cabo, 1945; Gopalan, 1971; Goto et al., 1981; Masatoshi et al., 1981; Lamilla et al., 1995; Heupel et al., 1999; Mancini et al., 2006; Bejarano-Álvarez et al., 2011; Delpiani et al., 2011; Galván-Magaña et al., 2011; Muñoz-Ozório et al., 2013; Wagner et al., 2013; Santos & Gadig, 2014; Ehemann et al., 2016; Sans-Coma et al., 2017; Capapé & Ali, 2017).
There is considerable difficulty, in relation to characteristics and classification of Siamese individuals. According to Spencer (2000), a designation of the diverse types of union, as well as the identification of individual twins were established. Siamese twins, joined by an ectopic part or accessory attached by their backs, involving or not spinal union, are considered rachipagus (Denardin et al., 2013); or twins united by the ventral region, liver and intestine with separation of the rectum, however, with an independent heart are called omphalopagus (Denardin et al., 2013).

In this group of animals, anomalies of incomplete body duplication have been reported as diprosopus and dicephalous (or bicephalous) which are related to total or partial duplication of the cranial region. The dicephalic (or bicephalic) anomaly occurs due to the total separation of the duplicate structures and joining in the distal region of the skull or beginning of the column, thus, the animal is born with two heads, two encephalon, and a double spine (Sans-Coma et al., 2017).

Dicephalic individuals originate from a dualistic fusion, where the formation of two embryonic bodies occurs by a single blastoderm or by the joining of two distinct blastoderms of the same yolk. As for the origin of these two embryonic bodies, there are two theories: division at the time of embryogenesis, or double formation and consecutive fusion (Bosinceano, 1934). This type of anomaly affects not only the external characteristics of the animals, but also internal organs that can be divided or merge in a fused region (Ford, 1930, Sans-Coma et al., 2017).

According Ehemann et al., (2016) cases of dicephaly and diprosus in sharks have been documented previously in some species. The Diprosopuses reported are as follow: *Squalus acanthias* (Bosinceano, 1934) and *Prionace glauca* (Hevia-Hormazábal et al., 2011). The Dicephali reported are: *Carcharhinus leucas* (Wagner et al., 2013); *C. porosus* (Muñoz-Ozório et al., 2013); *Galeorhinus galeus* (Delpiani et al., 2011); *Galeus atlanticus* (Sans-Coma et al., 2017); *Mustelus higmani* (Ehemann et al., 2016); *Prionace glauca* (Goto et al., 1981; Bejarano-Álvarez et al., 2011; Galván-Magaña et al., 2011; Ehemann et al., 2016); *Rhizoprionodon acutus* (Gopalan, 1971); *R. porosus* (Santos & Gadig, 2014); and *Squalus blainvillei* (Lozano-Cabo, 1945; Capapé & Ali, 2017).

This study intends to report the morphological and anatomical deformities presented in a specimen of *Squalus acanthias*.

### 2 MATERIAL AND METHODS

A newborn dogfish caught with the deformation was captured by a bob-shrimp throw net, off the Peruibe e Itanhaém Coast, São Paulo State, Brazil, observed in Figure 1. The shark was donated to the Institute of Marine and Environmental Biology -IBIMM, in July 2013. The specimen was fixed
in 4% formalin for 24h, and then stored in a 70% alcohol solution. Subsequently, it was deposited in the scientific collection of the Museu do Mar - IBIMM, under the number 005/2013.

**Figure 1** – Location of the catch area of the dogfish dog fish neonate, off the coast of Peruibe City, São Paulo State, Brazil.

The identification was made through the morphological and morphometric characteristics, all the classic measurements were performed with an emphasis on the dermal denticles and dentition (Compagno, 1984; Viana *et al.*, 2016).

The morphometric data was based on Compagno (1984) and they are expressed in percentages of total length. In order to compare our specimen, morphometric data of other *S. acanthias* found in the literature it was included.

To examine the internal anatomy, the animal was positioned with the ventral part resting on the table and computed tomography was used. For the reading of the images, the K-sas diagnostics program was used.

For a better visualization of the organs, a longitudinal cut was made with the aid of a scalpel in the ventral region, exposing the internal anatomy.
3 RESULTS

3.1 IDENTIFICATION

The specimen was a neonate, male with a total length of 23.5 cm (right animal) and 22.8 cm (left animal), 800 g total weight, caught off Peruibe City, São Paulo State, Brazil (24º22’38.78”S and 47º01’12.15”W), seen in Figure 1. It was identified as a Squalidae, *Squalus acanthias* (Linnaeus, 1758), with the following characteristics seen in Figure 2: dorsal view (A); anterior margin of the unilobed nostrils (B); origin of first dorsal-fin spine is behind the free rear tips of the pectoral fins (C); very elongated second dorsal-fin spine, taller than the second dorsal-fin apex (D); unicuspidal dermal denticles (E); unicuspidal teeth in both jaws, flattened labial-lingually overlapped, broad teeth, although short at the crown, upper teeth are smaller than lower, long and thick, oblique and directed laterally cusp (F).

**Figure 2** - Characteristics of *Squalus acanthias* neonate caught off the Peruibe coast: A) dorsal view (red circles: first dorsal-fin spine and second dorsal-fin spine); B) margin of the nostrils; C) dermal denticles; D) teeth.
The measurements of the exemplar are summarized in the Table 1. According to Ebert et al., (2010), 18 exemplars of *S. acanthias* were used from different collections. For morphometric data he used five specimens but did not specify what they are from (Table 1). Also, Viana et al. (2016) analyzed 21 specimens presenting a table with individuals from Southwestern Atlantic Ocean ranging from 20.0 cm to 88.0 cm. However, there is no percentage related to total length of this biometry related to the length making it impossible to compare them.

Table 1. Proportional dimensions as percentages of total length of *Squalus acanthias*, male, neonate from Peruibe City, Sao Paulo, Brazil. Also, measurements from Ebert et al. (2010).

| SPECIMENS                      | RIGHT | LEFT | EBERT ET AL., (2010) |
|--------------------------------|-------|------|---------------------|
| TOTAL LENGTH (TL)              | 23.5  | 22.8 | 42.9-67.8           |
| MEASURES                       | %     | %    | %                   |
| FORK LENGTH (FL)               | 84.7  | 83.3 | -                   |
| PRECAUDAL LENGTH (PRC)         | 79.1  | 79.8 | 77.7-79.1           |
| PRE-SECOND DORSAL LENGTH (PD2) | 63.4  | 64.0 | 58.8-61.0           |
| PRE-FIRST DORSAL LENGTH (PD1)  | 33.6  | 33.3 | 32.8-35.2           |
| HEAD LENGTH (HDL)              | 20.4  | 21.1 | 20.7-22.4           |
| PREBRANCHIAL LENGTH (PG1)      | 17.0  | 16.2 | 17.6-19.2           |
| PRESPIRACULAR LENGTH (PSP)     | 11.1  | 11.0 | 10.8-12.0           |
| PREORBITAL LENGTH (POB)        | 5.5   | 5.3  | 5.6-6.5             |
| PREPECTORAL LENGTH (PP1)       | 20.9  | 20.6 | 20.7-22.9           |
| INTERDORSAL SPACE (IDS)        | 18.7  | 18.9 | 19.8-21.2           |
| DORSAL-CAUDAL SPACE (DCS)      | 7.7   | 6.6  | 10.8-12.0           |
| PRENARIAL LENGTH (PRN)         | 5.1   | 4.4  | 3.5-4.1             |
| PREORAL LENGTH (POR)           | 97.9  | 9.6  | 8.5-9.8             |
| EYE LENGTH (EYL)               | 5.5   | 4.4  | 3.4-3.9             |
| EYE HEIGHT (EYH)               | 2.6   | 2.2  | 1.4-2.2             |
| INTERGILL LENGTH (ING)         | 2.6   | 2.2  | -                   |
| FIRST GILL SLIT HEIGHT (GS1)   | 2.6   | 2.2  | 1.4-1.9             |
| PECTORAL ANTERIOR MARGIN RIGHT (PIAR) | 11.5 | 11.4 | 13.6-16.2           |
| PECTORAL ANTERIOR MARGIN LEFT (PIAL) | 10.6 | 10.5 | 13.6-16.2           |
| PECTORAL BASE RIGHT (PIBR)     | 6.0   | 5.7  | 4.7-6.0             |
| PECTORAL BASE LEFT (PIBL)      | 5.1   | 6.1  | 4.7-6.0             |
| PECTORAL INNER MARGIN RIGHT (PI1R) | 6.4 | 5.3  | 4.4-7.3             |
| PECTORAL INNER MARGIN LEFT (PI1L) | 5.1 | 6.1  | 4.4-7.3             |
| PECTORAL POSTERIOR MARGIN RIGHT (PIPR) | 6.8 | 7.5  | 8.1-11.0            |
| PECTORAL POSTERIOR LEFT (PIPL) | 7.7   | 8.3  | -                   |
| PECTORAL HEIGHT RIGHT (PIHR)   | 8.1   | 7.0  | 9.2-11.0            |
| PECTORAL HEIGHT LEFT (PIHL)    | 8.5   | 7.0  | 9.2-11.0            |
| FIRST DORSAL LENGTH (DIL)      | 12.3  | 11.8 | -                   |
| FIRST DORSAL ANTERIOR MARGIN (D1A) | 11.1 | 10.1 | -                   |
| FIRST DORSAL BASE (DIB)        | 8.9   | 7.9  | -                   |
| FIRST DORSAL HEIGHT (D1H)      | 6.4   | 5.3  | -                   |
| FIRST DORSAL POSTERIOR MARGIN (DIP) | 6.0 | 5.3  | -                   |
| SECOND DORSAL LENGTH (D2L)     | 13.2  | 11.8 | -                   |
| Measurement                              | Value 1 | Value 2 | Range  |
|-----------------------------------------|---------|---------|--------|
| SECOND DORSAL ANTERIOR MARGIN (D2A)    | 8.9     | 7.9     | -      |
| SECOND DORSAL BASE (D2B)               | 7.2     | 6.6     | -      |
| SECOND DORSAL HEIGHT (D2H)             | 3.0     | 3.1     | -      |
| SECOND DORSAL POSTERIOR MARGIN (D2P)   | 5.1     | 4.8     | -      |
| PELVIC LENGTH (P2L)                    | 9.4     | 8.8     | 10.0-11.3 |
| PELVIC ANTERIOR MARGIN (PZA)           | 4.3     | 4.4     | -      |
| PELVIC BASE (P2B)                      | 3.0     | 2.6     | -      |
| PELVIC HEIGHT (P2H)                    | 4.7     | 4.8     | 4.2-5.0 |
| PELVIC INNER MARGIN LENGTH (P2I)       | 2.6     | 2.2     | -      |
| PELVIC POSTERIOR MARGIN LENGTH (P2P)   | 3.0     | 2.6     | -      |
| HEAD HEIGHT (HDH)                      | 8.9     | 8.3     | 8.4-9.6 |
| TRUNK HEIGHT (TRH)                     | 7.7     | 7.9     | 8.8-10.3 |
| MOUTH LENGTH (MOL)                     | 6.4     | 6.1     | -      |
| MOUTH WIDTH (MOW)                      | 7.7     | 7.5     | 7.0-8.7 |
| UPPER LABIAL FURROW LENGTH RIGHT (ULAR)| 3.0   | 2.2     | 1.5-2.6 |
| UPPER LABIAL FURROW LENGTH LEFT (ULAL) | 2.6   | 2.2     | 1.5-2.6 |
| LOWER LABIAL FURROW LENGTH RIGHT (LLAR)| 1.3   | 1.8     | -      |
| LOWER LABIAL FURROW LENGTH LEFT (LLAL) | 1.7   | 2.2     | -      |
| NOSTRIL WIDTH RIGHT (NOWR)             | 1.3     | 1.3     | 1.2-1.3 |
| NOSTRIL WIDTH LEFT (NOWL)              | 1.7     | 1.8     | 1.2-1.3 |
| INTERNARIAL SPACE RIGHT (INWR)         | 4.7     | 4.8     | 3.3-4.0 |

3.2 EXTERNAL MORPHOLOGY

Externally the animal presented the lateral fusion of two almost complete embryos. In relation to the duplication of the head, they are separated until the fifth branchial arch. The exemplar presented the following characteristics in Figure 3: A) two pairs of branchial arches, two pairs of first and second dorsal fins with spines, first pair of pectoral fins, tail and one caudal fin, with two superior lobes and one inferior lobe; B) second pair of pectoral fins; one pair of pelvic fins, one pair of claspers, and one caudal fin, with two superior lobes and one inferior lobe.
Figure 3 – External morphology of *Squalus acanthias*, male neonate caught off the Peruibe City coast, SP: A) dorsal view: gill; first dorsal fin; second dorsal fin; first pair of pectoral fins; caudal fin; two dorsal lobe; and B) ventral view: second pair of pectoral fin; pelvic fin; two clasper; one ventral lobe of caudal fin.

3.3 INTERNAL MORPHOLOGY

The neonate specimen presents the following internal morphology: two oral cavities; two esophagi; two stomachs (right stomach more developed); two livers (three lobules each); two hearts; one intestine; and one cloaca (Figure 4).

Figure 4 - Intern morphology of *Squalus acanthias* neonate caught off the Peruibe coast: mouth; esophagus; stomach; liver; intestine; cloaca and heart.
3.4 COMPUTED TOMOGRAPHY

It was possible to evaluate the skeletal structures through computed tomography and display some of the following characteristics: two heads; one pair of olfactory rosettes; two mouths, two first dorsal fins spine; two second dorsal fin spine and two vertebral columns (Figure 5).

Figure 5 - Computed tomography of Squalus acanthias neonate caught off the Peruibe coast: mouth; olfactory rosette; first dorsal fin spine; second dorsal fin spine.

4 DISCUSSION

The animal presented characteristics of a Squalus acanthias. According Viana et al., (2016), the specimen is characterized by a slender and fusiform body, arched dorsally at the anterior region, lateral keels restricted to the caudal peduncle, short snout that is rounded or obtuse at the tip, lobe-like anterior nasal flaps, yet without forming nasal barbels, the presence of slender dorsal spines without lateral grooves prior to each dorsal fin, extending or not to the apex of corresponding fin, second dorsal fin considerably smaller and more raked than first dorsal fin, caudal peduncle with conspicuous upper and lower precaudal pits, asymmetric caudal fin without subterminal notch, dorsal caudal lobe considerably greater in length than ventral caudal lobe, small, unicuspidal teeth, similar in both jaws with short, slightly oblique cusps and unicuspidal or tricuspidal dermal denticles. In the
A dicephalous specimen demonstrated the presence of the nostril margin, first dorsal-fin spine, second dorsal-fin spine, dermal denticles and teeth.

Comparing the dicephalous specimen with the measurements presented by Ebert et al., (2010) some discrepancies could be observed such as the interdorsal space, dorsal-caudal space, prenarial length, first gill slit height, pectoral anterior margin, pectoral posterior margin, pectoral height, pelvic length, trunk length and internarial space.

These facts show the disadvantage in the use of morphometrics as taxonomic characters (Steffens & D’Aubrey, 1967). Besides the natural factors, the discrepancies in the morphometric characteristics can originate from different measurement techniques and anomaly distortions.

There are two theories about this type of anomaly and its embryonic origin, the dualist theory admits that duplicate animals come from the fusion of two primitively distinct embryos. This bodily division probably occurs during the formation of two embryonic bodies in a single blastoderm or the fusion of two distinct blastoderms developed in the same fetus. There are two theories that admits that double monsters come from the fusion of two originally distinct embryos and, the unicist theory that they originate from the division of a primitively simple embryo (Bosinceano, 1934).

Dicephalous individuals are conjoined at the chest separated (fused side-by-side) with two heads completely separated into one body. Diprosopus, also known as craniofacial duplication, indicates the presence of a single body and head, in which parts or the whole face are duplicated (Sans-Coma et al., 2017). Dicephaly is a case of Siamese twins in which two animals are always derived from a single fertilized egg; both animals are conjugated to varying degrees, presenting two heads (Witschi, 1952).

The occurrence of this abnormality is rare. Bosinceano (1934) described a sample of *S. acanthias* with the cranial region not totally divided, presenting only the region of the tip of the divided face: four eyes, four nostrils and two mouths. Ehemann et al., (2016) described a dicephalous *Prionace glauca* specimen which presented a union where the embryos had two developed heads with almost the same size and shape, and the *S. acanthias* embryo presented the same characteristics with the two heads very evident, clearly demonstrating the presence of the four pairs of gill slits.

Santos & Gadig (2014) described a *Rhizoprionodon porosus* with the same characteristics of two complete heads with two eyes, two pairs of pectoral fins, a pair of pelvic fins, and a single body with four dorsal fins around the body. When opened, the animal had a heart, stomach and duplicate livers and only one intestinal valve, as in the findings of *S. acanthias*, demonstrating that these same animals of different species share the same type of dicephaly.
Another similar case of dicephaly was found by Capapé & Ali (2017) in a fetus at the end of gestation of a longnose spurdog, *Squalus brainvillei*, however presenting a similarity; this animal showed the same characteristics of the *S. acanthias* described in this article.

Delpiani, *et al.*, (2011) also describes the same characteristics found in a *Galeorhinus galeus* shark, similar to the anomaly with the embryo of *Galeus atlanticus* (Sans-Coma *et al.*, 2017). A specimen of *P. glauca* was described with dicephaly but with a deformation in the caudal region of the spine (Galván-Magaña, 2011).

Ventrally the *S. acanthias* presented a pair of pelvic fins as well as a pair of clasps with only one cloacal opening and a lower caudal lobe corroborating the findings of Santos & Gadig (2014), who found the same characteristics in a copy of *Rhizoprionodon porosus*.

According to Sans-Coma *et al.*, (2017) when open, the animal had a heart, duplicate livers and stomachs, the pyloric region of the stomachs was fused, giving rise to a single complete intestine presenting only one intestinal valve, as in the findings of *S. acanthias*, demonstrating that these animals even of different species share the same type of dicephaly.

Muñoz-Ozorio *et al.*, (2013) described dicephaly in *Carcharhinus porosus* but the specimen of the single body showing two dorsal and pectoral fins, an anus and a tail, incomplete division of two heads, but differently from the other works it originated anteriorly to the gills, presenting one mouth for each head, a single branchial opening, a pair of eyes and a shared eye, and presented one vertebral column. This disagrees with the other works cited, where animals with complete dicephaly, presented two completely distinct columns, and the two individuals were joined together only in the ventral region of the animal. Another similar case occurred with *Carcharhinus leucas* where the animal presented only slight division of head in the branchial region, however when submitted to the X-ray it was possible to observe the presence of two distinct columns separating the animal into two heads, merging just after the shoulder girdle. This demonstrated that this animal was dicephalous, but with a joined vertebrae (Wagner *et al.*, 2013). Nevertheless, it appears that no deformation described was exactly the same as another.

5 CONCLUSION

The analyzed specimen can be considered the first record of a dicephalous *S. acanthias*. However, this deformation can be seen in other species. Nevertheless, apparently no deformation was the same as those observed.
ACKNOWLEDGMENT

The Ricardo Henrique, Retired Sergeant of the Environmental Military Police of SP, Animal Care Hospital of Ipiranga-SP. Also, thanks to Fapesp for the feature used in this version (Project 2016/05259-0).

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