Molecular Identification of Shark Meat From Local Markets in Southern Brazil Based on DNA Barcoding: Evidence for Mislabeling and Trade of Endangered Species

Fernanda Almerón-Souza1†, Christian Sperb2†, Carolina L. Castilho2, Pedro I. C. Figueiredo1, Leonardo T. Gonçalves1, Rodrigo Machado2, Larissa R. Oliveira3, Victor H. Valiati2* and Nelson J. R. Fagundes1*†

1Laboratório de Genética Médica e Evolução, Departamento de Genética, Universidade Federal do Rio Grande do Sul, Porto Alegre, Brazil, 2Laboratório de Biologia Molecular, Centro de Ciências da Saúde, Universidade do Vale do Rio dos Sinos, São Leopoldo, Brazil, 3Laboratório de Ecologia de Mamíferos, Centro de Ciências da Saúde, Universidade do Vale do Rio dos Sinos, São Leopoldo, Brazil

*Correspondence:
Victor H. Valiati
valiati@unisinos.br
Nelson J. R. Fagundes
nelson.fagundes@ufrgs.br

†These authors have contributed equally to this work.

Specialty section:
This article was submitted to Evolutionary and Population Genetics, a section of the journal Frontiers in Genetics

Received: 01 November 2017
Accepted: 03 April 2018
Published: 27 April 2018

Citation:
Almerón-Souza F, Sperb C, Castilho CL, Figueiredo PICC, Gonçalves LT, Machado R, Oliveira LR, Valiati VH and Fagundes NJR (2018) Molecular Identification of Shark Meat From Local Markets in Southern Brazil Based on DNA Barcoding: Evidence for Mislabeling and Trade of Endangered Species. Front. Genet. 9:138, doi: 10.3389/fgene.2018.00138

Elasmobranchs, the group of cartilaginous fishes that include sharks and rays, are especially vulnerable to overfishing due to low fecundity and late sexual maturation. A significant number of elasmobranch species are currently overexploited or threatened by fisheries activities. Additionally, several recent reports have indicated that there has been a reduction in regional elasmobranch population sizes. Brazil is an important player in elasmobranch fisheries and one of the largest importers of shark meat. However, carcasses entering the shark meat market have usually had their fins and head removed, which poses a challenge to reliable species identification based on the morphology of captured individuals. This is further complicated by the fact that the internal Brazilian market trades several different elasmobranch species under a common popular name: “cação.” The use of such imprecise nomenclature, even among governmental agencies, is problematic for both controlling the negative effects of shark consumption and informing the consumer about the origins of the product. In this study, we used DNA barcoding (mtDNA, COI gene) to identify, at the species level, “cação” samples available in local markets from Southern Brazil. We collected 63 samples traded as “cação,” which we found to correspond to 20 different species. These included two teleost species: Xiphias gladius (n = 1) and Genidens barbus (n = 6), and 18 species from seven elasmobranch orders (Carcharhiniformes, n = 42; Squaliformes, n = 3; Squatiniformes, n = 2; Rhinopristiformes, n = 4; Myliobatiformes, n = 3; Rajiformes, n = 1; and Torpediniformes, n = 1). The most common species in our sample were Prionace glauca (n = 15) and Sphyrna lewini (n = 14), while all other species were represented by four samples or less. Considering IUCN criteria, 47% of the elasmobranch species found are threatened at the global level,
INTRODUCTION

Elasmobranch (subclass Elasmobranchii) is a group of cartilaginous fishes that include sharks (superorder Selachii) and rays (superorder Batoidea). Even though elasmobranchs comprise less than 1% of the world fisheries catch (Food and Agriculture Organization of United Nations, 2014, 2016), these species have biological characteristics that make them particularly vulnerable to overfishing, such as a low fecundity and late sexual maturation (Bornatowski et al., 2014b). Indeed, several recent reports have indicated that there has been a reduction of elasmobranch populations, resulting in demographic collapse at a regional scale (Baum et al., 2003; Barauss et al., 2014). The overfishing of sharks is especially problematic because these top predators play a key role in marine ecosystems, and, therefore, their population dynamics may affect all local marine diversity (van der Elst, 1979; Heithaus et al., 2008; Gallagher et al., 2012; Pauly et al., 2013; Worm et al., 2013; Bornatowski et al., 2014a).

In 1999, FAO (Food and Agriculture Organization) launched an international plan for the conservation and management of sharks and rays, recognizing the high vulnerability of these organisms (Vannucinn, 1999). However, despite this initiative, a significant number of elasmobranch species has remained overexploited or threatened by fisheries activities (Camhi et al., 2009; Cosandey-Godin and Morgan, 2011), which is illustrated by the 42% global increase in the shark meat trade from 2000 to 2011 (Food and Agriculture Organization of United Nations, 2015).

While shark fins are considered to be one of the most valuable products in the ocean (Gallagher and Hammerschlag, 2011), shark meat often attains only 20–60% of the price of tuna and mackerel meat (Bonfil, 1994). As a result, captured individuals usually have their fins removed for the shark fin market, the head is discarded, and the remaining central body part (“cigar”) is then sold for the shark meat market with no special care (Kotas et al., 2008; Ward-Paige et al., 2012). From a taxonomic point of view, the removal of the head and fins represents a challenge to reliable species identification based on morphological features, allowing shark carcasses to be traded fraudulently (Holmes et al., 2009).

Brazil is among the six countries that have the highest capture rate for elasmobranchs (Lack and Sant, 2006), even though a thorough assessment of the impact of industrial fishing is made difficult by inaccurate records (Barreto et al., 2017). Southern Brazil is a region of high elasmobranch diversity (Lucifora et al., 2011), and has a large extractive marine fishing industry, with approximately 160 thousand metric tons of fish caught annually (MPA. Boletim estatístico da pesca e aquicultura, 2011). The two southernmost states, Santa Catarina (SC) and Rio Grande do Sul (RS), are responsible for 98% of the catches (MPA. Boletim estatístico da pesca e aquicultura, 2011). In addition, Brazil is a major player in the meat trade market, acting as the world’s largest importer of shark meat in 2011 (Food and Agriculture Organization of United Nations, 2015). Internally, the Brazilian shark meat market trades several different elasmobranch species under the popular name “cação” (or other related popular terms such as “caçonet e” and “anjo”), which is used to label several species (Figure S1). For example, Neto (2013) found 21 different species traded under the common name “cação”, including hammerhead sharks (Sphyrna spp.), the blue shark (Prionace glauca), the tiger shark (Galeocerdo cuvier), the bull shark (Carcharhinus leucas), the Galapagos shark (C. galapagensis), and the blacktip shark (C. limbatus). Consumers value “cação” meat for its low cost and for being a “thornless fish” (Bornatowski et al., 2007). However, most consumers are not aware that “cação” is a synonym for sharks (or rays), and others believe that “cação” represents a “specific race of sharks” or even “a race of small sharks” (Bornatowski et al., 2015). Supermarkets, fisheries, and restaurants often omit any other information when selling “cação” meat. Indeed, the use of this term is so widespread that even Brazilian regulatory agencies categorize all elasmobranch species as “cação” without any species-specific information (MPA. Boletim estatístico da pesca e aquicultura, 2011).

The imprecise nomenclature of elasmobranchs makes it difficult to mitigate the negative effects of human shark consumption, as it becomes more difficult to inform the consumer if the product comes from a threatened species or from an illegal species trade. Since shark carcasses are sliced before being sold, it is virtually impossible to obtain accurate species diagnosis based on morphological traits for marketed elasmobranchs (Bornatowski et al., 2015). Therefore, there is an increasing need for fast, reliable, and cheap testing for determining the taxonomic identity of commercialized fishes (Rasmussen and Morrissey, 2008). A precise identification of marketed species also assures that the correct information is presented to the consumer, motivating him or her to take part in honest and regulated trade (Moretti et al., 2003; Martinez et al., 2005).

DNA barcoding uses a small fragment from a DNA sequence located within a standardized region of the genome to allow precise species identification (Hebert et al., 2003). In animals, the standard DNA barcode comes from a stretch of 650 base pairs (bp) from the 5′ end of the mitochondrial gene Cytochrome Oxidase Subunit I (COI or Cox 1) (Meyer and Paulay, 2005; Hajibabaei et al., 2007). This technique has been widely used in a range of studies of species identification (e.g., Meyer and
Paulay, 2005; Lowenstein et al., 2010; Carvalho et al., 2011; Rodrigues-Filho et al., 2012; Galimberti et al., 2013). Whilst DNA barcoding is a valuable tool for species identification, especially when the entire organism cannot be accessed for morphology, there are important limitations concerning its accuracy, which depend on the reference database available and on the degree of genetic difference among species (see Frézal and Leblois, 2008 for a review on the pros and cons of DNA barcoding). The aim of this study is to use DNA barcodes to identify, at the species level, samples of “cação” (or similarly labeled) meat available in local markets in Southern Brazil. Finally, we discuss the implications of these findings in the context of elasmobranch conservation in Brazil.

MATERIALS AND METHODS

Sample Collection
We studied samples sold under general names such as “cação,” “caçonete,” and “filé anjo,” which usually refer to elasmobranch species. Between 2008-2013 and in 2016 we acquired filet samples from local fish markets and supermarkets in different cities from the RS and SC states in Southern Brazil (Figure 1, Table 1). We also included in the analysis samples from Sphyrna lewini (n = 4), Pseudobatos horkelii (n = 2), Rhizoprionodon lalandii (n = 1), Narcine brasiliensis (n = 1), Zapteryx brevirostris (n = 2), and Gymnura altavela (n = 1), collected from fishing vessels and morphologically identified according to Figueiredo (1977), to serve as controls for the DNA barcode identification. These samples are identified as E__ in Table 1. All samples were stored in 95% ethanol at −20°C.

Laboratory Procedures
DNA extraction started from a small portion (~100 mg) of the tissue. For most samples we used the Wizard® Genomic DNA Purification Kit (Promega) modified to include an initial digestion step with 200 µg proteinase k (Aljanabi and Martinez, 1997). For the remaining samples, we used a protocol based on the CTAB method (Doyle, 1987). We used the COI primers FishF2 (5′ TCG ACT AAT CAT AAA GAT ATC GGC AC 3′) and FishR2 (5′ ACT TCA GGG TGA CCG AAG AAT CAG AA 3′) (Ward et al., 2005). Amplification reactions were prepared with 0.4 µM of each dNTP, 1.5 mM MgCl2, 0.5 µM of each primer, 1 U Taq Polymerase, and ~40 ng of genomic DNA. Cycling conditions included an initial denaturing step of 94°C for 5′, followed by 10 cycles of 94°C for 1′, 55°C (−0.5°C/cycle) for 1′, and 72°C for 1′30′, and 30 additional cycles of 94°C for 1′, 50°C for 1′, and 72°C for 1′30′, with a final extension step of 72°C for 5′. The amplification products were visualized on a 1% agarose gel stained with GelRed™ (Biotium). PCR products were purified enzymatically using 0.33U SAP (Shrimp Alkaline Phosphatase) and 3.33U ExoI (Exonuclease I). PCR products were sequenced by the Sanger method in Macrogen Inc. (Seoul, South Korea) and Ludwig Biotec (Porto Alegre, Brazil). DNA sequencing was performed on both strands using the primers mentioned above.

Data Analysis
The consensus sequence for each sample was assembled and trimmed in Geneious 9.1 (www.geneious.com). The reliability
### TABLE 1 | Sample information, species identification, average genetic distance, and results from the BLAST search.

| Sample | Candidate species | Avg. distance | % Coverage | % Identity |
|--------|-------------------|---------------|------------|------------|
| IIL04  | Carcharhinus brachyurus | 0.001          | 98         | 99         |
| IIL05  | Carcharhinus brachyurus | 0.001          | 97         | 99         |
| IIL14  | Carcharhinus brachyurus | 0.001          | 98         | 99         |
| IIL04-2| Carcharhinus falciformis | 0.003          | 98         | 99         |
| IIL27  | Carcharhinus falciformis | 0.003          | 99         | 99         |
| FA08   | Galeochirus galesus | 0.001          | 95         | 99         |
| MP60   | Genidens barbus | NC             | 96         | 100        |
| E14*   | Gymnura altavela | 0.020          | 99         | 99         |
| IIL35  | Myliobatis goodei | 0.013          | 96         | 99         |
| E13*   | Nacrine brasiliensis | 0.003          | 99         | 99         |
| IIL15  | Prionace glauca | 0.001          | 99         | 99         |
| IIL20  | Prionace glauca | 0.000          | 99         | 99         |
| IIL31  | Prionace glauca | 0.000          | 100        | 100        |
| IIL34  | Prionace glauca | 0.000          | 100        | 99         |
| IIL35  | Prionace glauca | 0.000          | 100        | 99         |
| O22    | Prionace glauca | 0.000          | 97         | 100        |
| FA02   | Prionace glauca | 0.002          | 99         | 99         |
| FA03   | Prionace glauca | 0.000          | 96         | 100        |
| FA23   | Prionace glauca | 0.000          | 99         | 100        |
| FA24   | Prionace glauca | 0.002          | 100        | 99         |
| FA25   | Prionace glauca | 0.003          | 100        | 99         |
| FA26   | Prionace glauca | 0.000          | 100        | 100        |
| FA27   | Prionace glauca | 0.000          | 100        | 99         |
| FA29   | Prionace glauca | 0.000          | 100        | 99         |
| FA31   | Prionace glauca | 0.000          | 100        | 99         |
| IIL26  | Rajiformes sp. BOLD AABB | 0.000       | 96         | 100        |
| E34*   | Pseudobatos horkelii | 0.003          | 97         | 100        |
| E36*   | Pseudobatos horkelii | 0.002          | 98         | 100        |
| E26*   | Rhizoprionodon lalandi | 0.001          | 94         | 100        |
| IIL13  | Rhizoprionodon lalandi | 0.001          | 99         | 99         |
| FA05   | Rhizoprionodon lalandi | 0.001          | 91         | 99         |
| FA17   | Rhizoprionodon lalandi | 0.001          | 100        | 99         |
| O24    | Rhizoprionodon lalandi | 0.001          | 95         | 100        |
| E07*   | Sphyrna lewini | 0.024          | 100        | 99         |
| E08*   | Sphyrna lewini | 0.021          | 98         | 99         |
| E15*   | Sphyrna lewini | 0.022          | 97         | 100        |
| E44*   | Sphyrna lewini | 0.021          | 97         | 99         |
| M004   | Sphyrna lewini | 0.034          | 97         | 99         |
| MP55   | Sphyrna lewini | 0.024          | 100        | 99         |
| MP57   | Sphyrna lewini | 0.023          | 97         | 100        |
| MP58   | Sphyrna lewini | 0.022          | 97         | 100        |
| O06    | Sphyrna lewini | 0.022          | 97         | 99         |
| O07    | Sphyrna lewini | 0.022          | 97         | 100        |
| O08    | Sphyrna lewini | 0.022          | 98         | 100        |
| O09    | Sphyrna lewini | 0.024          | 100        | 99         |
| O27    | Sphyrna lewini | 0.024          | 100        | 99         |
| O29    | Sphyrna lewini | 0.021          | 99         | 100        |
| O28    | Sphyrna lewina | 0.000          | 100        | 99         |

(Continued)
of each consensus sequence was assessed by a thorough visual inspection of the chromatograms used in the assemblies to check for sequencing errors and artifacts. Low quality regions in the chromatograms, identified as a stretch of five or more contiguous bases having high background noise and uneven spacing, were trimmed and removed before sequence assembly. Because the assembly algorithm gives more weight to better quality reads, cases of sequence heterogeneity between strands are resolved in favor of the best quality read or, if both reads had similar quality for that position, marking it as an ambiguous base (N, R, Y, etc.). The consensus sequence was then used as a query for comparison with the NCBI database (http://www.ncbi.nlm.nih.gov/) using the Basic Local Alignment Search Tool—Nucleotide (BLASTn). In all cases, BLAST matched COI sequences from elasmobranchs (or, in some cases, from teleosts) with good coverage and identity (see Results), suggesting that we generated authentic COI sequences from our samples. We recorded the species representing the top BLAST hit for each query. Following this, we built a dataset of 2,877 COI sequences deposited in the GenBank including all species of all genera represented in the final dataset.

### RESULTS

In total, 63 samples were collected, amplified, sequenced, and compared to GenBank sequences (Table 1). High quality sequences ranged between 204 and 650 bases. There was no sequence heterogeneity between strands involving high quality bases from two or more reads. Overall, our analysis suggests the presence of 20 different species among the samples. Seven samples were identified as belonging to two Actinopterygii (ray-finned fishes) species: *Xiphias gladius* (Perciformes, swordfish; \( n = 6 \)), and *Genidens barbus* (Siluriformes, white sea catfish; \( n = 1 \)). The remaining samples may represent 18 elasmobranch species from three shark orders (Carcharhiniformes, Squaliformes, and Squatiniformes; \( n = 42 \), 3, 2, respectively) and four ray orders (Rhinopristiformes, Myliobatiformes, Rajiformes, and Torpediniformes; \( n = 4, 3, 1, 1 \), respectively). Three ray species (*P. horkelii*, *Z. brevirostris*, and *N. brasiliensis*) were only found in samples from fishing vessels (i.e., they were not purchased in the market).

### Table 1 | Continued

| ID  | Accession no. | Seq. Size | Location     | Type   | Candidate species                      | Avg. distance\(^a\) | % Coverage\(^b\) | % Identity\(^b\) |
|-----|---------------|-----------|--------------|--------|----------------------------------------|---------------------|-----------------|-----------------|
| FA21| MG703561      | 642b      | Porto Alegre, RS | fresh | *Sphyrra zygaena*                      | 0.001               | 100             | 100             |
| MP15| MG703562      | 630b      | Porto Alegre, RS | fresh | *Squalus mitsukurii*                   | 0.000               | 98              | 100             |
| MP18| MG703563      | 603b      | Porto Alegre, RS | fresh | *Squalus mitsukurii*                   | 0.001               | 98              | 100             |
| MP16| MG703564      | 641b      | Porto Alegre, RS | fresh | *Squalus mitsukurii*                   | 0.001               | 96              | 100             |
| FA16| MG703565      | 593b      | Porto Alegre, RS | fresh | *Squatina occulta*                     | 0.000               | 95              | 100             |
| MG08| MG703566      | 204b      | Rio Grande, RS  | fresh | *Squalus mitsukurii*                   | NC                  | 100             | 99              |
| IIL01| MG703567    | 650b      | Itajaí, SC      | frozen| *Xiphias gladius*                      | NC                  | 100             | 99              |
| IIL03| MG703568    | 650b      | Itajaí, SC      | frozen| *Xiphias gladius*                      | NC                  | 100             | 99              |
| IIL16| MG703569    | 650b      | Itajaí, SC      | frozen| *Xiphias gladius*                      | NC                  | 100             | 99              |
| IIL18| MG703570    | 599b      | Itajaí, SC      | frozen| *Xiphias gladius*                      | NC                  | 100             | 99              |
| IIL19| MG703571    | 458b      | Itajaí, SC      | frozen| *Xiphias gladius*                      | NC                  | 100             | 100             |
| IIL25| MG703572    | 622b      | Itajaí, SC      | frozen| *Xiphias gladius*                      | NC                  | 100             | 100             |
| E50*| MG703573     | 650b      | Passo de Torres, SC | fresh | *Zapteryx brevirostris*             | 0.030               | 97              | 99              |
| E54*| MG703574     | 232b      | Passo de Torres, SC | fresh | *Zapteryx brevirostris*             | 0.000               | 100             | 100             |

\(^a\)Average genetic distance against all sequences from the same species in the final dataset.

\(^b\)%Coverage and %Identity values considering the top-BLAST hit for the candidate species.

\(^c\)All samples identified as E__ were obtained directly from fishing vessels, and were not purchased. NC, not computed.
Based on COI sequences, all but one elasmobranch samples were identified at the species level, representing 17 formally described species. One sample was associated with an undescribed or unsequenced species (which occurs in GenBank as “Rajiformes sp. BOLD: AAB1882”). The most common species found among market samples were Prionace glauca (blue shark, n = 15) and S. lewini (scalloped hammerhead shark, n = 14). All other species were far less common, including R. lalandii (Brazilian sharpnose shark, n = 4), Carcharhinus brachyurus (copper shark, n = 3), Carcharhinus falciformis (silk shark, n = 2), Sphyra zygaena (smooth hammerhead shark, n = 2), Galeorhinus galeus (school shark, n = 1), Rhizoprionodon porosus (Caribbean sharpnose shark, n = 1), Squalus cubensis (Cuban dogfish, n = 1). Squalina occulta (hidden angel shark, n = 1), and Squalina guggenheim (spiny angel shark, n = 1). All ray species identified in the study occurred once or twice among the samples: G. altavela (spiny butterfly ray, n = 2), P. horkelli (Brazilian guitarfish, n = 2), Z. brevirostris (shortnose guitarfish, n = 2), Myliobatis goodei (southern eagle ray, n = 1), and N. brasiliensis (Brazilian electric ray, n = 1).

The average genetic distance between each sample and representatives of its most likely candidate species (determined by its clustering in the ML tree) was always lower than 3.50%, and usually lower than 1% (Table 1). The ML tree showed cohesive clusters of conspecific sequences (Figure 2). The few exceptions, which had bootstrap support values lower than 90, included S. mitsukurii, C. brachyurus, S. guggenheim, and S. occulta (Figure 2). In all cases, however, the estimated genetic distance between our samples and reference sequences were used to indicate the most likely candidate species (shown in Table 1).

An interesting case is sample MP16, whose top-hit in BLAST was Squalus montalbani, but clustered with S. mitsukurii in the ML tree (Figure 2). However, both MP16 and MP18 showed a much smaller distance from S. mitsukurii (0.0009) than to any other closely related species (0.0027 vs. S. cf. megalops; 0.0043 vs. S. montalbani; 0.0058 vs. S. elongatus; and 0.0088 vs. S. cf. mitsukurii). Similarly, III04, III05, and III14 were much closer to C. brachyurus (0.0014) than to C. brevifinna (0.0150). MG08 was closer to S. occulta (0.0000) than to S. guggenheim (0.0071), while FA16 was closer to S. guggenheim (0.0008) than to S. occulta (0.0064). The complete distance matrix can be downloaded as Supplementary Material (File S2).

DISCUSSION

We found 18 Elasmobranchii and two Actinopteriigii species among the samples acquired in Southern Brazilian fish markets as “cação,” “caçonete,” or “filé anjo.” This represents 17% of all elasmobranch species registered for Southern Brazil and 13% of the species described for Brazil (Bornatowski et al., 2009). Other studies, based on other molecular markers, that aimed at species identification of shark filets from Northern Brazil have also shown the great number of species being trade without any taxonomic control (Rodrigues-Filho et al., 2009; Palmeira et al., 2013). Unfortunately, our DNA data does not allow us to conclude that these samples represent individuals captured in Southern Brazil. For example, most individuals included in the final dataset did not have location information. Additionally, even if this was available, it is unclear whether COI would have enough resolution to allow unambiguous recognition of regional stocks for these species (Antoniou and Magoulas, 2014). However, the fact that the vast majority of samples collected in this study were purchased fresh is a strong indication that these specimens may have been captured off Southern Brazil or in nearby areas.

The use of the COI DNA barcode allowed us to identify all samples at the specific level even though some cases deserve further discussion. The best match for III26 was an undescribed or unsequenced species, Rajiformes sp. BOLD:AAB1882 (Coverage = 96%, Identity = 100%). The sample MG08 resulted in a short DNA sequence, whose top-result in BLAST was against S. occulta (Coverage = 95%, Identity = 100%), but showed an inconclusive clustering with any Squalina species in the ML tree (Figure 2). Nevertheless, as occurred for other samples (FA16, III04, III05, III14, MP16, MP18), comparing the genetic distance among alternative candidate species allowed the identification of the most likely candidate for each sample. In the case of the samples associated to Squalina, species identification was corroborated by the fact that both S. occulta and S. guggenheim occur off Southern Brazil (Vaz and Vaz and De Carvalho, 2013) and that both samples were acquired as fresh filets, likely indicating a local catch. With a single exception, the species associated with the top-BLAST result also resulted in the lowest average genetic distance. The exception was MG16, whose top-BLAST result was Squalus montalbani (Coverage = 100%, Identity = 99%), but whose lowest average genetic distance was against S. mitsukurii, which also represented the second and third top-BLAST results (Coverage = 96%, Identity = 100%). The low genetic distance among Squalus species and the lack of a clear structure in the ML tree (Figure 2) may indicate that DNA barcoding for this genus may be more complicated than for other genera, and may require other genetic markers. From the taxonomic point of view, it is difficult to discriminate among Squalus species (Haddad and Gadig, 2005), which may be due to a shallow diversification time that is reflected in the low genetic distances among several species. The inherently difficult taxonomy of the genus may favor misnomers in reference databases. In this regard, S. cubensis presents a likely example of database confusion. There are two COI sequences for this species in GenBank. However, while the entry FJ519594 is close to S. mitsukurii (~0.2% genetic distance) the other, FN431670, is distantly related to it (~7.2% genetic distance) and associated with sample MP15 (Figure 2). These issues reinforce the importance of database curation and maintenance, with rigorous taxonomic criteria for the deposition of reference sequences (Ekrem et al., 2007; Teletchea, 2009; Dudgeon et al., 2012). It also highlights that in some cases it may be important to analyze additional genetic markers for a more accurate species identification (Mendonça et al., 2009; Moffah et al., 2011; Pérez-Jiménez et al., 2013).

The most abundant shark species in our samples were Prionace glauca and S. lewini (23.8 and 22.2%, respectively).
FIGURE 2A | (Continued).

Carcharhiniformes

Carcharhiniformes 1

Carcharhiniformes 2

Carcharhiniformes 3

Squaliformes

Squatiniformes

Myliobatiformes

Rajiformes

Torpediniformes

Rhinopristiformes

Prionace glauca

Carcharhinus spp.

Sphyra spp.

Rhizoprionodon spp.

FIGURE 2A | (Continued).
FIGURE 2B | ML tree based on HKY+G+I distance. The miniature on the upper left side shows major groups, displayed in more detail in individual panels. The number of shark and ray symbols represent the number of different species identified in the study for each group. Please note that this is an unrooted tree. Most entries were collapsed and the names were omitted for clarity. Samples from the present study are labeled according to Table 1. The most likely candidate species, together with other closely related species are shown in red. The numbers above the branches represent bootstrap percentage based on 1,000 replicates. Bootstrap values <70 were omitted. Please note the different scale among panels. The full ML tree is available as Supplementary Material (File S3).
TABLE 2 | Conservation status (global, national, and regional) of the species found in this study.

| Species                     | Common namea | IUCNb | ICMBioéc | RSd | SCë |
|-----------------------------|--------------|-------|----------|-----|-----|
| Carcharhinus brachyurus     | cooper shark | NT 2003 | DD*      | –   | –   |
| Carcharhinus falciformis    | silky shark  | NT 2016 | NT*      | –   | –   |
| Galeorhinus galeus          | school shark | VU 2006 | CR       | CR  | –   |
| Gymnura altavela            | butterfly ray| VU 2007 | CR       | EN  | –   |
| Myliobatis goodei           | southern eagle ray | DD 2009 | CR | CR | – |
| Narcine brasiliensis        | Brazilian electric ray | DD 2007 | DD* | – | – |
| Prionace glauca             | blue shark   | NT 2009 | NT*      | VU  | –   |
| Pseudobatos horkelli        | Brazilian guitarfish | CR 2016 | CR       | CR  | CR  |
| Rhizoprionodon lalandii     | Brazilian sharpnose shark | DD 2004 | NT* | – | – |
| Rhizoprionodon porosus      | Caribbean sharpsnose shark | LC 2006 | DD* | – | – |
| Sphyra lewini               | scalloped hammerhead shark | EN 2007 | CR       | CR  | EN  |
| Sphyra zygaena              | smooth hammerhead | VU 2005 | CR       | CR  | EN  |
| Squalus cubensis            | Cuban dogfish | DD 2006 | –       | –   | –   |
| Squalus mitsukurii          | shortspine spurdog | DD 2007 | DD* | – | – |
| Squatina guggenheim         | spiny angel shark | EN 2007 | CR | CR | EN |
| Squatina occulta            | smoothback angel shark | EN 2007 | CR | CR | – |
| Zapteryx breviostris        | shortnose guitarfish | VU 2006 | VU | CR | – |

*Species included in the National List of Species Threatened of Extinction (available at Portaria MMA no. 445 of 2014).
1 IUCN Red List of Threatened Species (IUCN, 2017).
2 Global conservation status according to IUCN (2017) criteria, followed by the year of assessment: (CR) critically endangered; (EN) endangered; (VU) vulnerable; (NT) near threatened; (DD) data deficient; (NE) not evaluated.
3 National conservation status according to the Brazilian Red Book of Threatened Faunal Species (Instituto Chico Mendes de Preservação da Biodiversidade, 2016).
4 Regional conservation status according to the List of Threatened Fauna of the Rio Grande do Sul State (Fundação Zoobotânica e Secretaria do Ambiente Desenvolvimento Sustentável, Decreto n° 51.797).
5 Regional conservation status according to the List of Threatened Fauna of the Santa Catarina State (Fundação de Meio Ambiente – FATMA).

P. glauca is distributed globally and its capture volume has been estimated at approximately 20 million individuals per year (Mendonça et al., 2012). Despite its endangered status (Table 2), P. glauca is the most fished shark in the world, representing 56% of the total catch of pelagic sharks, especially by industrial fisheries in which the target species are tuna or swordfish (Rose, 1996; Dulvy et al., 2003; Camhi et al., 2009). After the increasing market demand for shark fins and the high prices paid for them, these animals began to be targeted for the removal of these parts, with the carcasses being sold worldwide (Domingues, 2011). Indeed, we identified P. glauca in all samples acquired as frozen filets, which may reflect that these individuals were captured in other parts of the world, such as Taiwan and subsequently imported to Brazil (Figure S1). However, we also found P. glauca among fresh samples, which more likely indicates local capture. On the other hand, S. lewini was the most abundant species among fresh samples, which may indicate a higher local impact on this species. Several authors have raised concerns of predatory fishing for this species off Brazil due to the high commercial value of its fins (Amorim et al., 2011). This results in fishing pressures occurring over all phases and life cycles of these animals, including neonates (Mader et al., 2007) both on the continental shelf and in oceanic waters (Kotas, 2004; Kotas et al., 2005; Vooren and Klippel, 2005).

Regarding their conservation status, IUCN estimates that 37% of the elasmobranch species found in this study are considered threatened at the global level, 53% are threatened at the national level, and 47% are critically endangered at the national level (Table 2). It is difficult, however, to present a more regional picture, given that the red list for both Rio Grande do Sul and Santa Catarina states include only 59 and 23.5% of the species identified in this study, even though there are records for most of these species off these Brazilian states (Gadig, 2001). The conservation status for R. lalandii, S. mitsukurii, S. cubensis, M. goodei, and N. brasiliensis is unknown due to data deficiency (DD). In the worst-case scenario, ~50% of the species identified in this study would be threatened to some extent.

Our sampling was restricted to the south of Brazil due to a limited budget, but it would be important to perform similar studies in other Brazilian regions to provide a better picture of the shark fishing and trade in the country. It should be noted, however, that the Southern coast of Brazil is a hotspot for shark diversity, with high species richness, high endemism, and functional richness (Lucifora et al., 2011). Another future direction would be investigating how much of the shark meat market involves individuals fished locally.

Finally, an important issue in the conservation of these species is how local human populations will engage in more sustainable consumption practices. In this sense, labeling the meat of any shark species as “cação” may impose major barriers to conservation measures for this group, allowing the inadvertent consumption of protected species (Jacquet and Pauly, 2008). Indeed, Bornatowski et al. (2015), who interviewed fish meat consumers in Southern Brazil, reported that 61% of respondents...
claimed that they have never tried shark meat, even though they ate “cação.” In addition, 69% of respondents said they did not know that at least 25% of all elasmobranchs are threatened. Given these answers, it is evident that a significant portion of the population buying these products is not aware of the impact of their consumption habits, or of the current conservation status of elasmobranch species. Another issue for consumers is mislabeling of shark products, a common outcome of DNA barcode assessments of seafood products (Barbuto et al., 2010; Filonzi et al., 2010). This is illustrated by the presence of the two teleost species detected among our sample (Table 1). Therefore, it becomes essential and an ethical responsibility for the industry to label their products correctly and allow informed decision-making by the consumers. We suggest that all meat being sold as “cação” should be accompanied by the species common name, followed by its scientific name, and, whenever possible, the species threat categories according to the IUCN Red List. While fishing legislation may also have a positive impact on natural populations by suspending the capture and marketing of endangered elasmobranchs, environmental education measures focusing on the fishing community and on consumers will be fundamental for the effective protection of these species.

**AUTHOR CONTRIBUTIONS**

FA-S, CS, LO, VV, and NF designed the study; FA-S, CS, CC, PF, and RM executed experimental procedures; FA-S, CS, PF, LG, LO, VV, and NF performed data analysis and interpretation; FA-S, CS, VV, and NF wrote the paper.

**FUNDING**

This research was supported by Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq), Fundação Grupo Boticário de Proteção à Natureza (Projeto 0921_20112), Programa de Pós-Graduação em Biologia Animal (PPGBAN-UFRGS), and Programa de Pós-Graduação em Genética e Biologia Molecular (PPGBM-UFRGS) and Programa de Pós-Graduação em Biologia (UNISINOS).

**ACKNOWLEDGMENTS**

The authors would like to thank Walter de Nisa and Castro Neto (Pró-Squalus), Maria João Veloso da Costa Ramos Pereira (UFRGS), Clarissa Fleck (McGill University), Andrea Thomaz (University of Michigan), Daniel Lyons (University of Michigan), Bonnie Armour (University of Adelaide), and two reviewers for contributions in earlier versions of this manuscript.

**SUPPLEMENTARY MATERIAL**

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fgene.2018.00138/full#supplementary-material

---

**REFERENCES**

Aljanabi, S. M., and Martinez, I. (1997). Universal and rapid salt-extraction of high quality genomic DNA for PCR-based techniques. *Nucleic Acids Res.* 25, 4692–4693. doi: 10.1093/nar/25.22.4692

Amorim, A. F., Della-Fina, N., and Piva-Silva, N. (2011). Hammerheads sharks, *Sphyrrus lewini* and *S. zygaena* caught by longliners off Southern Brasil 2007-2008. *Collect. Vol. Sci. Pap. ITCAT 66, 2121–2133.

Antoniou, A., and Magoulas, A. (2014). “Application of mitochondrial DNA in stock identification,” in *Stock Identification Methods: Applications in Fishery Science*, eds S. X. Cadirin, L. A. Kerr, and S. Mariani (Walham, MA: Academic Press). 257–295.

Barausse, A., Correale, V., Cukrovic, A., Finotto, L., Riginella, E., Visentin, E., et al. (2014). The role of fisheries and the environment in driving the decline of elasmobranchs in the northern Adriatic Sea. *ICES J. Mar. Sci.* 71, 1593–1603. doi: 10.1093/icesjms/fst222

Barbuto, M., Galimberti, A., Ferri, E., Labra, M., Malandra, R., Galli, P., et al. (2010). DNA barcoding reveals fraudulent substitutions in shark seafood products: the Italian case of “palombo” (*Mustelus* spp.). *Food Res. Int.* 43, 376–381. doi: 10.1016/j.foodres.2009.10.009

Barreto, R. R., Bornatowski, H., Motta, F. S., Santander-Neto, J., Vianna, G. M. S., and Lessa, R. (2017). Rethinking use and trade of pelagic sharks from Brazil. *Mar. Policy* 85, 114–122. doi: 10.1016/j.marpol.2017.08.016

Baum, J. K., Myers, R. A., Kehler, D. G., Worm, B., Harley, S. J., and Doherty, P. A. (2003). Collapse and conservation of shark populations in the Northwest Atlantic. *Science* 299, 389–392. doi: 10.1126/science.1079777

Bonfil, R. (1994). *Overview of World Elasmobranch Fisheries*. Rome: FAO.

Bornatowski, H., Abilhoa, V., and Charvet-Almeida, P. (2009). Elasmobranchs of the Paraná Coast, southern Brazil, south-western Atlantic. *Mar. Biodivers. Rec.* 2:e158. doi: 10.1017/S1755267209000986

Bornatowski, H., Braga, R. R., Kalinowski, C., and Vitule, J. R. S. (2015). “Buying a pig in a poke”: the problem of elasmobranch meat consumption in Southern Brazil. *Etnobiol. Lett.* 6, 196–202. doi: 10.14237/ebll.6.1.2015.451

Bornatowski, H., Braga, R. R., and Vitule, J. R. S. (2014a). Threats to sharks in a developing country: the need for effective simple conservation measures. *Nat. Conserv.* 12, 11–18. doi: 10.4322/natcon.2014.003

Bosch, H., Costa, L., Robert, M. C., and Pina, J. V. (2007). Hábitos alimentares de tubarões-martelo jovens, *Sphyrrus zygaena* (Carcharhiniformes: *Sphyrynidae*), no litoral sul do Brasil. *Biot. Neotrop.* 7, 213–216. doi: 10.1590/S1676-06320070001000025

Bornatowski, H., Navia, A. F., Braga, R. R., Abilhoa, V., and Corrêa, M. F. M. (2014b). Ecological importance of sharks and rays in a structural foodweb analysis in southern Brazil. *ICES J. Mar. Sci.* 71, 1586–1592. doi: 10.1093/icesjms/fsu025

Camhi, M. D., Valenti, S. V., Fordham, S. V., Fowler, S. L., and Gibson, C. (2009). The Conservation Status of Pelagic Sharks and Rays: Report of the IUCN Shark Specialist Group Pelagic Shark Red List Workshop. Newbury: IUCN Species Survival Commission Shark Specialist Group, 78.

Carvalho, D. C., Neto, D. A., Brasil, B. S., and Oliveira, D. A. (2011). DNA barcoding unveils a high rate of mislabeling in a commercial freshwater catfish from Brazil. *Mitochondrial DNA* 22, 97–105. doi: 10.3109/19401736.2011.588219

Cosandeý-Godin, A., and Morgan, A. (2011). *Fisheries Bycatch of Sharks: Options for Mitigation*. Washington, DC: Ocean Science Division; Pew Environment Group.

Darriba, D., Taboada, G. L., Doallo, R., and Posada, D. (2012). *jModelTest* 2: more models, new heuristics and parallel computing. *Nat. Methods* 9, 772. doi: 10.1038/nmeth.2109

Domingues, R. R. (2011). *Identificação Molecular, Biologia e Pesca de Tubarões do Gênero Carcharhinus (Chondrichthyes – Carcharhiniformes): Uma Contribuição*
Para a Gestão de Pesca do Estado de São Paulo, Brasil. Master’s thesis, Instituto de Pesca, São Paulo.

Doyle, J. J. (1987). A rapid DNA isolation procedure for small quantities of fresh leaf tissue. *Phytochem*. Bull. 19, 11–15.

Dudgeon, C. L., Blower, D. C., Broderick, D., Giles, J. L., Holmes, B. J., Kashiwagi, T., et al. (2012). A review of the application of molecular genetics for fisheries management and conservation of sharks and rays. *J. Fish Biol*. 80, 1789–1843. doi: 10.1111/j.1095-8649.2012.03265.x

Dulvy, N. K., Sadoyv, Y., and Reynolds, J. D. (2003). Extinction vulnerability in marine populations. *Fish. Fish*. 4, 25–64. doi: 10.1046/j.1467-2979.2003.00105.x

Ekrem, T., Willassen, E., and Stur, E. (2007). A comprehensive DNA sequence library is essential for identification with DNA barcodes. *Mol. Phylogenet. Evol*. 43, 530–542. doi: 10.1016/j.ympev.2006.11.021

Food and Agriculture Organization of United Nations (FAO) (2014). The State of World Fisheries and Aquaculture 2014. Rome: FAO.

Food and Agriculture Organization of United Nations (FAO) (2015). State of the Global Market for Shark Products. Rome: FAO.

Food and Agriculture Organization of United Nations (FAO) (2016). The State of World Fisheries and Aquaculture 2016. Contributing to Food Security and Nutrition for All. Rome: FAO.

Figueiredo, J. L. (1977). Manual de Peixes Marinhos do Sudeste do Brasil–

Filonzi, L., Chiesa, S., Vaghi, M., and Marzano, F. N. (2010). Molecular barcoding Tubarões da Costa Brasileira

Hebert, P. D., Cywinska, A., and Ball, S. L. (2003). Biological identifications through DNA barcodes. *Proc. R. Soc. Lond. B Biol. Sci*. 270, 313–321. doi: 10.1098 rspb.2002.2218

Holmes, B. H., Steinke, D., and Ward, R. D. (2009). Identification of shark and ray fins using DNA barcoding. *Fish. Res*. 95, 280–288. doi: 10.1016/j.fishres.2008.09.036

Instituto Chico Mendes de Preservação da Biodiversidade (2016). *Brazil Red Book of Threatened Species of Fauna. Available online at: http://www.icmbio.gov.br/portal/images/stories/comunicacao/publicacoes/publicacoes-diversas/dcom_sumario_executivo_livro_vermelho_da_fauna_brasileira_amecadada_de_extincao_2016.pdf* (Accessed April 11, 2018).

IUCN (2017). The IUCN Red List of Threatened Species. Available online at: http:// www.iucnredlist.org/ (Accessed October 01, 2016).

Jacquet, J. L., and Pauly, D. (2008). Trade secrets: renaming and mislabeling of seafood. *Mar. Policy* 32, 309–318. doi: 10.1016/j.marpol.2007.06.007

Katho, K., and Standley, D. M. (2013). MAFFT multiple sequence alignment software version 7: improvements in performance and usability. *Mol. Biol. Evol*. 30, 772–780. doi: 10.1093/molbev/mst010

Kotas, J. E. (2004). Dinâmica de Populações e pesca do Tubarão-martelo Sphyraena lewini (Griffith & Smith, 1834), Capturado no mar Territorial e zona Econômica Exclusiva do Sudeste-sul do Brasil. Doctoral thesis, Universidade de São Paulo, São Paulo.

Kotas, J. E., Petere, M. Jr., Azvedo, V. D., and Santos, S. D. (2005). A Pesca de Emalhe e de Espinhel-de-Superfície na Região Sudeste-Sul do Brasil. Instituto Oceanográfico-USP.

Kotas, J. E., Petere Junior, M., Fiedler, F., Mastrochirico, V., and Sales, G. (2008). A pesca de emalhe-de-superfície de Santa Catarina direcionada à captura dos tubarões-martelo, *Sphyraena lewini* (Griffith & Smith 1834) e *Sphyraena zygaena* (Linnaeus 1758). Atlântica 30, 113–128. doi: 10.5088/atlantica. v3002.1511

Lack, M., and Sant, G. (2006). *World Shark Catch, Production and Trade 1990-2003*. Department of the Environment and Heritage.

Lowenstein, J. H., Burger, J., Jettein, C. W., Amato, G., Kolokotronis, S. O., and Gochfeld, M. (2010). DNA barcodes reveal species-specific mercury levels in tuna sushis that pose a health risk to consumers. *Biol. Lett*. 6, 692–695. doi: 10.1098/rsbl.2010.0156

Lucifora, L. O., Garcia, V. B., and Worm, B. (2011). Global diversity hotspots and conservation priorities for sharks. *PLoS ONE* 6:e19356. doi: 10.1371/journal.pone.0019356

Moliner, A., Sander, M., Casa, G. E. Jr., Altenhoen, R. J., and dos Anjos, C. S. (2007). Evidências de sobrepesca do tubarão martelo (*Sphyra spp.*) no Rio Grande do Sul. *Biodivers. Pumpeana* 5.

Martínez, I., James, D., and Loréal, H. (2005). Application of Modern Analytical Techniques to Ensure Seafood Safety and Authenticity. FAO Fisheries Technical Paper. Food and Agriculture Organization of United Nations, Rome.

Mendonça, F. F., Hashimoto, D. T., Porto-Foresti, F., Oliveira, C., Gadig, O. B. B., and Foresti, F. (2009). Identification of the shark species *Rhinopropodon lalandii* and *R. porosus* (*Elasmobranchii, Carcharhinidae*) by multiplex PCR and PCR-RFLP techniques. *Mol. Ecol. Resour*. 9, 771–773. doi: 10.1111/j.1755-0998.2009.02524.x

Mendonça, F. F., Usami, L. H., Hashimoto, D. T., Pereira, L. H., Porto-Foresti, F., Oliveira, C., et al. (2012). Identification and characterization of polymorphic microsatellite loci in the blue shark *Prionace glauca*, and cross-amplification in other shark species. *J. Fish Biol*. 80, 2643–2646. doi: 10.1111/j.1095-8649.2012.03291.x

Meyer, C. P., and Paulay, G. (2005). DNA barcoding: error rates based on comprehensive sampling. *PLoS Biol*. 3:e422. doi: 10.1371/journal.pbio.0030422

Moffah, M., Aziz, S. H., Elramah, S., and Favereaux, A. (2011). Classification of sharks in the Egyptian Mediterranean waters using morphological and DNA barcoding approaches. *PLoS ONE* 6:e27001. doi: 10.1371/journal.pone.0027001

Moretti, V. M., Turchini, G. M., Bellagamba, F., and Caprino, F. (2003). Traceability issues in fishery and aquaculture products. *Vet. Res. Commun*. 27, 497–505. doi: 10.1023/B:VERC.0000014207.01900.5c

MPA. Boletim estatístico da pesca e aquicultura (2011). *Relatório do Ministério da Pesca e Aquicultura*. 98. Available online at: http://www.icmbio.gov.br/cepulsel/images/stories/biblioteca/download/estatistica/est_2011_bol__br_a.pdf

Neto, D. A. P. (2013). Detecção de Addulteração de Espécies em Pescado e Derivados por Meio da Técnica de DNA Barcoding, Master’s thesis, Universidade Federal de Minas Gerais, Belo Horizonte.

Palmeira, C. A. M., Rodrigues-Filho, L. F., Sales, J. P. L., Vallinoto, M., Schneider, H., and Sampaio, I. (2013). Commercialization of a critically endangered species (large-toothed sawfish, *Pristis perotteti*) in fish markets of northern Brazil: authenticity by DNA analysis. *Food Control* 34, 249–252. doi: 10.1016/j.foodcont.2013.04.017

Pauly, D., Hilborn, R., and Branch, T. A. (2013). Fisheries: does catch reflect abundance? *Nature* 499, 303–306. doi: 10.1038/494303a

Pérez-Jiménez, J. C., Rocha-Olivares, A., and Sosa-Nishizaki, O. (2013). Morphological and molecular differentiation of smooth-hound sharks (*Genus Mustelus*, Family Triakidae) from the Gulf of California. *J. Appl. Ichthyol*. 29, 268–270. doi: 10.1111/jai.12042
Portaria (2014). MMA N° 443, de 17 de Dezembro de 2014. Available online at: http://www.icmbio.gov.br/cepsul/images/stories/legislacao/Portaria/2014/p_mma_443_2014_lista_esp%C3%A9cies_amea%C3%A7adas_extin%C3%A7%C3%A3o.pdf (Accessed September 03, 16)

Rasmussen, R. S., and Morrissey, M. T. (2008). DNA-based methods for the identification of commercial fish and seafood species. *Compr. Rev. Food Sci. Food Saf.* 7, 280–295. doi: 10.1111/j.1541-4337.2008.00046.x

Rodrigues-Filho, L. F., Pinhal, D., Sodré, D., and Vallinoto, M. (2012). “Shark DNA forensics: Applications and impacts on genetic diversity,” in *Analysis of Genetic Variation in Animals*, ed M. Caliskan (Rijeka: InTech), 269–286.

Rodrigues-Filho, L. F., Rocha, T. C., Rêgo, P. S., Schneider, H., Sampaio, I., and Vallinoto, M. (2009). Identification and phylogenetic inferences on stocks of sharks affected by the fishing industry off the Northern coast of Brazil. *Genet. Mol. Biol.* 32, 405–413. doi: 10.1590/S1415-47572009005000039

Rose, D. A. (1996). *An Overview of World Trade in Sharks and Other Cartilaginous Fishes*. Traffic International.

Stamatakis, A. (2014). RAxML version 8: a tool for phylogenetic analysis and post-analysis of large phylogenies. *Bioinformatics* 30, 1312–1313. doi: 10.1093/bioinformatics/btu033

Swofford, D. L. (2002). *PAUP*: *Phylogenetic Analysis Using Parsimony* (*and Other Methods*). Version 4. Sunderland, MA: Sinauer Associates.

Teletchea, F. (2009). Molecular identification methods of fish species: reassessment and possible applications. *Rev. Fish Biol. Fish.* 19, 265. doi: 10.1007/s11160-009-9107-4

Triant, D. A., and DeWoody, J. A. (2007). The occurrence, detection, and avoidance of mitochondrial DNA translocations in mammalian systematics and phyleogeography. *J. Mammal.* 88, 908–920. doi: 10.1644/06-MAMM-A-204R1.1

van der Elst, R. P. (1979). A proliferation of small sharks in the shore-based Natal sport fishery. *Environ. Biol. Fish.* 4, 349–362. doi: 10.1007/BF0005524

Vannucini, S. (1999). *Shark utilization, Marketing and Trade*. No. 389. Food & Agriculture Org. Rome: FAO.

Vaz, D. F., and De Carvalho, M. R. (2013). Morphological and taxonomic revision of species of *Squatina* from the Southwestern Atlantic Ocean (*Chondrichthyes: Squatiniformes: Squatinidae*). *Zootaxa* 3695, 1–81. doi: 10.11646/zootaxa.3695.1.1

Vooren, C. M., and Klippel, S. (2005). *Ações Para a Conservação de Tubarões e Raias no Sul do Brasil*. Porto Alegre: Igaré.

Ward-Paige, C. A., Keith, D. M., Worm, B., and Lotze, H. K. (2012). Recovery potential and conservation options for elasmobranchs. *J. Fish Biol.* 80, 1844–1869. doi: 10.1111/j.1095-8649.2012.03246.x

Ward, R. D., Zemlak, T. S., Innes, B. H., Last, P. R., and Hebert, P. D. (2005). DNA barcoding Australia’s fish species. *Philos. Trans. R. Soc. Lond. B. Biol. Sci.* 360, 1847–1857. doi: 10.1098/rstb.200 5.1716

Worm, B., Davis, B., Kettemer, L., Ward-Paige, C. A., Chapman, D., Heithaus, M. R., et al. (2013). Global catches, exploitation rates, and rebuilding options for sharks. *Mar. Policy* 40, 194–204. doi: 10.1016/j.marpol.2012.12.034

Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.