Fifteen years of extensive and inadvertent elasmobranch trade in Brazil detected by DNA tools

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Article Impact Statement

In Brazilian elasmobranch trade, 83% of species found are threatened. A broad knowledge of market dynamics is vital to revert this scenario.

Keywords

DNA-based tools, elasmobranchs, conservation genetics, endemic species, Brazilian trade, fisheries genetics, forensics, meta-analysis

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ABSTRACT

The trade of elasmobranchs (sharks and rays) in Brazil raises significant concerns due to the country's rich endemic biodiversity. The present study explores the use of DNA-based tools to monitor the Brazilian elasmobranch trade, focusing on their role in identifying processed products and supporting conservation efforts. A systematic search of literature was carried out and included 35 peer-reviewed articles published between 2008 and 2023. We observed a shift in research focus since 2015 from the development of DNA-based tools to direct trade application. Molecular identification challenges, including costly sequencing and limited resources in national databases, were identified along with proposed solutions, such as protocol optimization and exploration of cost-effective alternatives. Biases in trade analysis papers, particularly a lack of research in the Northeast region of Brazil, as well as issues with sample sizes were evident. Species identified using DNA-based tools included the critically endangered Scalloped Hammerhead Shark (*Sphyrna lewini*), appearing in 46% of the evaluated papers, followed by the Blue Shark (*Prionace glauca*), and several others threatened species, including the critically endangered endemic Brazilian Guitarfish (*Pseudobatos horkelii*) and the recently categorized vulnerable Sharpnose Shark (*Rhizoprionodon porosus*). Other species were reassessed, including previously non-threatened species that are now at risk, emphasizing the need for conservation measures. Our findings highlight the importance of continuing genetic monitoring of shark and ray trade in Brazil, calling for conservation efforts and law enforcement to protect elasmobranch populations, and recommending strategies for accurate species identification, expanded research, and effective management.

KEYWORDS

DNA-based tools, elasmobranchs, conservation genetics, endemic species, trade meta-analysis, fisheries genetics, forensics, meta-analysis

1. INTRODUCTION

The capture of elasmobranchs (sharks and rays) has been increasing worldwide due to industrial, artisanal, and recreational fisheries catering to diverse markets (Pacoureau et al., 2021). There has been a rising demand for shark fins in Asia and an expanding market for their meat in Europe and South America (Dent & Clarke, 2015). Brazil has emerged as the largest importer of shark meat and the eleventh-largest shark fishing country (Barreto et al., 2017; FAO, 2020). High demand for shark meat is often attributed to the regulatory practices surrounding finning worldwide, as a substantial growth in the trade has coincided with the global requirement for landings of both fins and the rest of the body (Dent & Clarke, 2015; Rangel et al., 2021).

A Brazilian "fin laundry" was established due to low meat prices and the umbrella label "cação" extensively used in Brazil for shark meat, leading to a lack of awareness among consumers that this term corresponds to shark species (Bornatowski et al., 2015; FAO, 2020). However, the global shark trade has undergone recent significant market adjustments, with shark meat presenting a higher value than fins, though its value was reported to vary by region and species (Niedermüller et al., 2021). Although less understood, the skate and stingray trade are also concerning. Brazilian markets consider rays an inexpensive product, and they are caught mainly as bycatch (Dent & Clarke, 2015). Nevertheless, Brazil has experienced an increase in ray capture, earning it the status of the third-largest exporter of ray meat to South Korea in 2021, the primary consumer of ray meat worldwide (Niedermüller et al., 2021).

The elasmobranch's capacity to withstand anthropogenic pressures is limited due to their life history characteristics, such as late sexual maturity, with low fecundity and slow growth rates (Ferretti et al., 2010). Unreported and unregulated catches have led to considerable extinction risk for elasmobranchs, being the primary lineage of marine fish with elevated threats, with over a third of chondrichthyan fish species currently categorized as threatened (Dulvy et al., 2021). The Brazilian Guitarfish (*Pseudobatos horkelii*, Pollom et al., 2020a), Sawfish (*Pristis pristis*, Espinoza et al., 2022) and Daggernose Shark (Isogomphodon oxyrhynchus, Pollom et al., 2020c) are among the Brazilian elasmobranchs facing higher risk of extinction. Decreases in biological diversity of elasmobranchs has led to changes in the whole marine community, including ecosystemic imbalance, changing in trophic cascades, and commercial fisheries decline (Bornatowski et al., 2014; Pimiento & Pyenson, 2021; Sherman et al., 2023).

Brazilian legislative measures have been established with the intent of reducing mislabeling and the commercialization of species categorized as Vulnerable (VU), Endangered (EN), or Critically Endangered (CR) according to the Brazilian Ordinance 445/2014 of the Brazilian Ministry of Environment (updated in 2022 and 2023, as ordinances 148/2022 and 354/2023). However, mislabeling and the sale of threatened species continue to occur, and monitoring to curb these practices is still an outstanding challenge (El Bizri et al., 2020; Souza et al., 2021; Wosnick et al., 2023). Elasmobranch meat continues to be sold inadvertently due to their continuous landing without heads and fins and extensive processing when sold as fillets or steaks (Rodrigues-Filho et al., 2012; FAO, 2020). Important diagnostic features of the species are lost in these processes, hampering the morphological identification of the products at landing and commercial points (Domingues et al., 2021).

Molecular techniques have played a fundamental role addressing the illegal commerce of shark and ray products. These techniques enable the accurate identification of species composition at landings, restrain finning practices, and facilitate the identification of processed meat (Clarke et al., 2006; Domingues et al., 2021). Anyway, there is still need to: a) compile all the information available for the Brazilian elasmobranch trade, b) expand the use of molecular techniques to efficiently dissect the traded species composition, and c) assist enforcement inspections with genetic tools to help reverse the current alarming extinction risk of elasmobranch species.

In this context, this study aimed to make a historical assessment of the contribution of molecular tools in analyzing the Brazilian market of shark and ray products by dissecting all peer-reviewed journals published recently. Our overarching objective was to provide a comprehensive overview of the current state of research on the Brazilian elasmobranch market, identify opportunities for further investigation, and improve conservation efforts and law enforcement. To achieve this, five specific goals were established: (i) examine the existing scientific literature in all Brazilian regions, identifying sampling strategy and regions with a shortage of papers, (ii) evaluate the contribution of genetic tools in analyzing the trade of elasmobranchs in Brazil, including an analysis of the most commonly used techniques and the status of genetic databases for Brazilian species, (iii) identify the species composition in Brazilian markets, with a particular focus on the sale of threatened and endemic species over time, (iv) assess the extent to which legislation has contributed to monitoring the fishing and trade of elasmobranchs in Brazil, and (v) identify any gaps or limitations in the research landscape and current regulations. Furthermore, recommendations to address these gaps were provided, ultimately enhancing the effectiveness of conservation efforts.

2. METHODS

We performed an extensive Boolean search (AND, OR, NOT) on Web of Science, Scopus and Google Scholar to collect peer-reviewed papers that applied molecular tools to analyze the catch and trade of elasmobranchs in Brazil. Our literature collection strategy included keywords, titles, and abstract content related to mislabeling and molecular identification. We included papers that met the following criteria: (i) research performed in Brazil, (ii) published until June 31st, 2023, and (iii) research articles. To consolidate data from both databases and remove potential duplicates, we used the R package Bibliometrix (Aria & Cuccurullo, 2017).

To ensure relevancy, we manually inspected the papers for inclusion in our database using *strict consensus* criteria. To uncover any potentially missed papers, we applied the Snowball Method (Wohlin, 2014). We computed inter-rater agreement to infer the consistency and reliability of our assessments by measuring agreement among independent raters evaluating the same set of items (Gisev et al., 2013). To ensure methodological rigor, we followed the protocol from the RepOrting Standards for Systematic Evidence Syntheses (ROSES; Haddaway et al., 2018). For a comprehensive overview of our search process, please refer to Appendix 1.

To provide a historical assessment on the use of molecular tools to analyze the Brazilian market, we categorized the selected peer-reviewed papers into three groups: "Trade Analysis" (papers focusing on the analysis of elasmobranch trade), “Methods” (papers that developed DNA-based tools for species identification), and "Methods/Trade Analysis" (papers combining the development of a DNA-based technique with its applications for trade analysis within the same paper). Initial metadata construction depicts each sample from the respective papers in specific categories to characterize the use of genetics to analyze trade activity in Brazil. For a comprehensive understanding of the metadata and detailed information on the individual categories, please see Appendix 2. We used the metadata information to conduct an exploratory analysis in R. We investigated DNA tool applications in analyzing the elasmobranch trade in the Brazilian market, assessing papers and samples at national and state levels, examining genetic tool development, species composition, threat trends, and mislabeled products.

Two additional datasets were generated based on the species composition found, to obtain information on their extinction risk status, legislation, and genetic availability. The first dataset focused on the extinction risk for each species according to the Brazilian Red Book of Threatened Fauna (ICMBio, 2018), SALVE System (Portuguese for "Biodiversity Extinction Risk Assessment System", available at: https://salve.icmbio.gov.br/) and the International Union for Conservation of Nature (IUCN) Red List of Threatened Species (hereinafter IUCN Red List, available at: https://www.iucnRed_List.org/). Whenever available in the online IUCN Red List assessments, additional specific observations on species populations status for the Southwestern Atlantic Ocean (SWA) were also included. In the same dataset, we depicted key legislations for biodiversity conservation and labeling regulations over time. The second dataset focused on genetic information for each of the identified species, specifically the number of sequences available in the National Center for Biotechnology Information (NCBI) GenBank and in the Barcode of Life Data System (BOLD) database. NCBI GenBank sequences were obtained using the *esearch* command in Entrez Direct (EDirect; Kans, 2013) while BOLD sequences were accessed through the *bold* package (Chamberlain, 2021) in the statistical software R (v.2022.02.3 Team 2021). Both sets of additional data can be found in Appendix 3.

For detailed information on all statistical and visualization analyses conducted, please see the R Markdown script provided in Appendix 4 and the GitHub repository available at (link available upon acceptance).

3. RESULTS

3.1. Research Trends

A total of 35 peer-reviewed papers published between January 2008 and June 2023 were assessed (Figure 1a). Of these, 24 (69%) were categorized as “Trade Analysis”, eight (23%) as “Methods”, and three (9%) as “Methods/Trade Analysis” (Figure 1b). The focus of research has shifted towards trade analysis since 2012, with more “Trade Analysis” studies rather than “Methods” ones (Figure 1a).

Among the 11 “Methods” papers, PCR Multiplex was applied in five papers, PCR-RFLP in four papers, tandem repeats in one paper, and Sanger sequencing in one paper (Table 1; Table S2, in Appendix 2). The most frequently employed molecular marker was COI (n = 6), whereas 16S was the least utilized (n = 1).

The 27 “Trade Analysis” papers collectively involved 3,784 samples (mean = 192; min = 7; max = 747; sd = 194) from 15 Brazilian states, which represent 55% of the total country's federative units, and 88% of those on the coast (Figure 1c). The sampling sizes and strategies for market screening (e.g., market type, labeling, sample location) varied significantly among the papers (Figures 1d,e). Out of the total samples, 85% (n = 3,235) were assigned to specific Brazilian states while the remaining samples were labeled “Unidentified Location” due to incomplete information provided in the papers regarding each sample individually (Table S1, in Appendix 2). The State of Pará had the highest number of papers and samples (n = 10 and 1,022), followed by São Paulo (n = 8 and 998), and Santa Catarina (n = 6 and 194). Notably, there was a scarcity of papers in the Northeast region of Brazil, with only 311 samples collected and four papers across seven of the nine states in this region (Figures 1c-e).

Shark meat was the main target in the Trade Analysis, accounting for 81% of the papers (n = 22) and 1,419 samples, while ray meat was analyzed in six papers and comprised 335 samples (Table S1, in Appendix 2). We observed papers that collected specific groups of elasmobranchs such as guitarfishes (De-Franco et al., 2012; Alvarenga et al., 2021; 342 total samples), angel sharks (Bunholi et al., 2018; 85 samples), and whiptail stingrays (Schmidt et al., 2015; 97 samples). Three papers assessed elasmobranch captured as bycatch (Domingues et al., 2013; 618 total samples; Ferrette et al., 2019a; Guimaraes-Costa et al., 2020), one about fin trade (Ferrette et al., 2019b; 747 samples), and one about sawfish rostra (Faria et al., 2013; 77 samples). In restaurants of education institutions (i.e., school and universities cafeterias), 23 shark samples were served as fish meal, and in commercial restaurants, four out of 10 samples were found to be mislabeled (Alvarenga et al., 2021). An overall of nine papers evaluated the mislabeling in elasmobranch trade, including sharks being sold as salmon and croaker (Staffen et al., 2017) (Table 1 and Table S1, in Appendix 2).

3.2. Molecular Species Identification

Among the molecular tools applied in “Trade Analysis” papers, PCR-RFLP (n =1) was the least applied, followed by Multiplex PCR (n = 5), and Sanger sequencing (n = 24), the only technique applied in all papers published since 2018 (Figure 2a). The COI was the most applied molecular marker for both shark and ray identification, and the most represented in current databases (Figure 2b). Ninety seven percent of the samples (n = 2345) were identified accurately by the COI marker, while the remaining 3% of the samples (n = 69) were identified at the family level (Rajiformes, n = 1), genus level (*Dasyatis* sp., n = 39; *Gymnura* sp., n = 5; *Hypanus* sp., n = 1; *Mustelus* sp,. n = 5; *Narcine* sp.), or at a species complex, in which case it is not possible to assign the sample to a single species (*Carcharhinus obscurus*/*C. galapagensis*, n = 4). Other species complexes were identified through NADH2 (*Squalus brevirostris/S. megalops, n = 1*), and 12S/16S (*C. plumbeus/C. altimus*, n = 4). The region comprising 12S/16S also failed to identify samples at species level (*Rhizoprionodon* sp., n = 35; *Sphyrna* sp., n = 2). Overall, 11 misidentifications were reported (Table S1, in Appendix 2).

By examining the availability of genetic resources in the public sequence databases, we detected a clear bias toward sharks compared to ray species (Figure 2b). Furthermore, it is worth noting that public sequence databases still have a dearth of sequences for a high amount of Brazilian chondrofauna. This is particularly true for those with restricted distribution, such as endemic species of Brazil (Figure 2c). The availability of complete mitochondrial genomes was higher for species with a wide distribution range, whereas the use of shorter mitochondrial markers showed consistent representation across species with both wide and restricted distributions (Figure 2c).

A total of 71 species (44 genera and 28 families) were reported in all "Trade Analysis" papers. Among these species, 36 shark species belong to 16 genera and 27 rays belong to 20 genera (18 marine and two freshwater). Additionally, we detected nine species of bony fish mislabeled as elasmobranch meat (Figure 3). Within the identified families, Carcharhinidae was the most abundant, with three genera leading the ranking: *Carcharhinus* (886 samples in 17 papers), *Prionace* (464 samples and 10 papers), and *Rhizoprionodon* (375 samples in 15 papers) (Figure 3). The top position within paper count was led by both the genus *Sphyrna* (family Sphyrnidae), with a sample count of 345 and *Carcharhinus*, both detected in 17 papers (Figure 3). For rays, guitarfishes (genus *Pseudobatos)* were detected in 298 samples across 8 papers, in which 187 samples belong to the CR *Pseudobatos horkelii* (Figure S2 in Appendix 1), whereas 111 samples to the recently categorized as EN *P. percellens*. The most threatened ray genus, *Pristis*, was also detected in high numbers (113 samples in 3 papers), as well as other genus containing commercially explored and threatened sharks and rays (Figure 3).

3.4. Threatened Species on Trade

We observed an increasing number of threatened species being detected in trade analyses (Table 1) (Figure S3, in Appendix 1). Currently, 83% (n = 54) of the elasmobranch species detected were categorized as threatened (VU, EN or CR) and 12% as Near Threatened (NT) by the IUCN Red List, with only three species categorized as Least Concern (LC) (Table 2). A total of 78% of the elasmobranch samples (n = 2877) detected here belong to threatened categories (Figure S2).

The IUCN Red List extinction risk changed over time (Figure 4, Figure S3 in Appendix 1), with only one species experiencing a decrease in extinction risk (*Rhinoptera brasiliensis*, from EN to VU). Contrastingly, we detected 33 cases where the extinction risk has increased, including 17 species not previously considered threatened, and seven species whose extinction risk worsened by more than one category (Figure 4a). Notably, frequently traded species have become more threatened, such as *Carcharhinus acronotus* (NT to EN), *Rhizoprionodon porosus* (LC to VU), *Sphyrna lewini*, and *S. mokarran* (EN to CR).

Additionally, among the 16 species previously categorized as Data Deficient (DD), 14 have had their extinction risk assessed with nine now considered threatened (Figure 4b), including two of the most commercially traded species: *R. lalandii* (DD to VU), and *C. porosus* (DD to CR). Overall, eight of the species detected are now CR.

Among the reviewed papers only 55% referred to the Brazilian Red Book, while 89% relied only on the IUCN Red List (Figure 5). There were discrepancies in species assessment between the extinction risk in the lists analyzed here (Figure 5), mainly regarding the number of assessed species. All the species found in this study were assessed in the IUCN Red List, while there were Not Evaluated (NE) species in the other two lists: 51% in the Brazilian Red Book, and 8% in the SALVE System. Also, 11% of the species did not have an observation for their SWA populations. Among the categorized species, we noted some differences in the assessment of extinction risk, such as the percentage of CR species, lower in the IUCN Red List (18%), and higher when accounting specific observations for SWA populations (31%). Overall, assessment in a restricted geographical range showed a higher rate of CR species, 25% in the Brazilian Red Book and 31% in the SALVE System (Figure 5a). In contrast, species categorized as EN or CR in the IUCN Red List presented different categories in the published version of the Brazilian Red Book (e.g., *Carcharhinus perezi* - VU) and SALVE System (e.g., *Pseudobatos percellens* and *Zapteryx brevirostris* - VU) (Table S1, in Appendix 3).

3.5. Brazilian Legislation

We examined the historical evolution of Brazilian legislation and species extinction risk assessments. First, we analyzed the extinction risk lists (Figure 5a), including the newly released SALVE System, and the most pertinent regulatory measures based on these assessments (Figure 5b). We identified a discrepancy in the categorization of threatened species (VU, EN, CR) and their protection under Brazilian ordinances. Significant progress were noted from the initial legislation (IN 05/2009), which protected species in Appendices I and II, to subsequent regulations (MMA 445/2014, MMA 148/2022 and MMA 354/2023), which protect VU, EN and CR species. However, the regulations latest regulation (2023) still categorizes a lower percentage (47%) of species as threatened compared to the most recent Brazilian extinction list in the SALVE System (63%). Second, we analyzed labeling regulations, which revealed limited taxonomic classifications and a prevalent use of umbrella labels over time (Table S1, in Appendix 3). Only one regulatory measure mandates species-specific labels, and it applies exclusively to one Brazilian state, Paraná, rather than to the entire country.

3.5. Mislabeling

According to the current national fish labeling (MAPA 570/2023), our study identified mislabeling in 237 samples. This included instances where bony fishes were sold as elasmobranchs in 33 samples, with the swordfish *Xiphias gladius* being the most frequently mislabeled species (n = 13). Furthermore, there were 85 cases where rays were sold as sharks and 22 cases where sharks were sold as rays. Specific instances of mislabeling were also recorded, such as sharks, skates, and stingrays being labeled as guitarfishes (n = 65). Also, a common change in labels was noted among angel sharks and guitarfishes, where angel sharks were labeled as guitarfishes (n = 5), and guitarfishes as angel sharks (n = 16). For other mislabeling appointments, please refer to Table S2, in Appendix 2. Half of the papers analyzing the meat trade included a proper note on the market label, but only 40% of them aimed to evaluate mislabeling activity (Table 1).

4. DISCUSSION

The present study examined the effectiveness and significance of genetic tools in identifying processed products and their role in conservation practices and law enforcement, both in Brazil and potentially worldwide, since Brazil has the second-largest number of endemic elasmobranch species globally (IUCN, 2023). There has been a rise in papers addressing the capture and commercialization of elasmobranchs using genetic tools over the past decade, with a clear temporal trend regarding the type of the research, from papers developing DNA tools to ones applying the tools to inspect the trade (Figure 1). This shift can be attributed to the rapid evolution in price and applicability of molecular techniques, including the use of DNA barcoding for elasmobranchs (Ward et al., 2005).

4.1. Resource Availability Define Paper Design

The DNA-based tools applied in Trade Analysis papers experienced a temporal switch, with Sanger sequencing emerging as the predominant technique (n = 24), and the only one applied since 2018 (Figure 2b). While Sanger sequencing is highly effective for accurate species identification, its continued high-cost for a large sample size poses challenges, particularly in resource-limited regions like Latin America, which can limit frequent market inspections. Several approaches can be pursued to address this such as, optimizing DNA sequencing protocols, including high-throughput techniques like amplicon sequencing, and exploring modern cost-effective methods, like real-time PCR and closed-tube DNA barcoding (Cardenosa et al., 2018; Ballard et al., 2020; Prasetyo et al., 2023; Yeo et al., 2023).

In the economic context of the Global South, it is important to recognize the value of traditional cost-effective techniques, such as PCR-RFLP and PCR Multiplex, which can identify a large set of samples at lower prices (Böhme et al., 2019). Although these techniques require initial development costs, some sets that comprise the most traded species in the Brazilian market are already available (Shivji et al., 2002; Caballero et al., 2012; Ferrito et al., 2019), including those developed nationally (Table S2, in Appendix 2). Among the techniques developed in Brazil, only the PCR Multiplex Method developed by De-Franco et al. (2010) was fully implemented in a Trade Analysis paper (De-Franco et al., 2012). Although other sets were not applied, developed primers have been used to obtain additional genetic information on shark species (Table S1, in Appendix 1). For instance, primers developed by Pinhal et al. (2012) were used by Davis et al. (2019) to assess population genetics of *Rhizoprionodon* in the Northwest Atlantic Ocean.

Fisheries research often relies on opportunistic sampling (Pardo et al., 2016), which reflects other aspects of the limited availability of research funding in environmental sciences and access to required instrumentation. This type of sampling size and strategy can impact forensic analysis, because a lack of statistically calculated sampling campaign may highlight a particular aspect of the trade, such as a seasonal aspect, rather than comprehensively covering the entire trade dynamics, also compromising the accurate comparison across studies (Pardo et al., 2016; Luque & Donlan, 2019). The disparity in sample sizes was indeed observed among the reviewed papers here. This has led to a geographical bias along the Brazilian coast, with a significant number of research papers conducted in the State of Pará (N = 10) in the Northern region, followed by São Paulo (N = 8) in the Southeast and Santa Catarina (N = 6) in the South, while fewer papers were published in the northeastern region (Figure 2). This bias is concerning because certain elasmobranch species endemic to the northeast, such as *Squalus bahiensis* (DD; Pollom et al., 2020d), *Hypanus marianae* (EN; Pollom et al., 2020b), and *Squatina varii* (LC; Rincon et al., 2019) may be subject to overexploitation without public knowledge. Overall, bias in biodiversity research can impact conservation efforts and strategy to avoid should be implemented (Hickisch et al., 2019). The observed pattern of papers published in the North and South/Southeast regions highlights another issue of national funding distribution. The fact that wealthier states (e.g., Rio Grande do Sul, Santa Catarina, Paraná and São Paulo) invest an equivalent amount of funding compared to states with scarce resources (e.g., Pará) indicates a lack of dedication to combating the illegal trade of elasmobranchs and the export of fins to Asian markets, especially considering that São Paulo is an important route for exporting fins collected from the entire Brazilian coast (Frontini & Mano, 2023).

Despite having a greater number of endemic and threatened species (IUCN, 2023), the number of papers analyzing rays is disproportionately low, highlighting a bias in elasmobranch research (Dudgeon et al., 2012; Soares & Petean, 2023). Rays are usually captured as bycatch from trawlers and gillnet fisheries, and their meat has been undervalued in most Brazilian markets, being sold at low prices, exported, or mislabeled (Dulvy et al., 2014; Ferrette et al., 2019a). However, rays can be a valuable fishery resource internationally, which has shifted its industrial fishing to support exportation, with Brazil being nowadays a major exporter of ray meat to South Korea, the largest consumer of ray meat globally (Niedermüller et al., 2021). Anyway, there is a scarcity of papers evaluating products derived from rays fishing industry and bycatch activities. A lack of research was also noted to track illegal finning activity in Brazil, a malpractice banned for over 20 years (IBAMA 121/1998). Thus, there is a pressing need for increased investment in analyzing bycatch and finning activities to comprehensively better understand both domestic and international trade dynamics.

4.2. Unraveling Trade with Molecular Tools: Challenges and Future Directions

DNA barcoding, the primary tool for species identification, faces challenges that can affect its accuracy. Incomplete species representation in databases and insufficient curation can lead to misidentification issues, such as the "Rajiformes" case noted here, a label that encompass 297 species. Additionally, some markers applied to DNA barcoding shows lower discriminatory power for closely related species (Alvarenga et al., 2023). This can impact conservation efforts, especially with threatened and non-threatened species in the same species complex (e.g., *Squalus brevirostris* - EN and *S. megalops* - LC). Integrating phylogenetic analysis can enhance identification reliability by considering factors like branch length and unusual relationship patterns (Felsenstein, 2004). The most applied molecular marker, COI, is one of the markers that has limitations for recently diverged species (Shokralla et al., 2012; Böhme et al., 2019)as observed here for Ca*rcharhinus obscurus an*d C. *galapagensis* (Corrigan et al., 2017). Alternatively, the NADH-ubiquinone oxidoreductase chain 2 (ND2) and the mitochondrial encoded 12S gene (Naylor et al., 2012; Valsecchi et al., 2020; Fernandes et al., 2021) are more effective molecular markers but scarce and underrepresented in current databases (Figure 2b). Broader marker utilization is needed to improve accuracy and expand database entries (Domingues et al., 2021), therefore, establishing a comprehensive national genetic database with regular curation is crucial.

A significant challenge in the fin trade is the genetic diversity among global populations. Shark products derived from worldwide industrial fisheries also share such challenge. The development of a genetic diversity atlas is essential to trace product origins accurately (Domingues et al., 2021). Thus, incorporating fine-scale information and population-level genetic data in sequences databases offers significant potential for tracing wildlife products as demonstrated by Cardeñosa et al. (2020) in Chinese markets, where they found that 84.9% of Thresher Shark (*Alopias pelagicus*) fins belonged to populations from the Eastern Pacific region. In addition, the adoption of high-throughput sequencing techniques, such as DNA metabarcording, is also important to identify species in food mixtures and in bycatch activity (Carvalho et al., 2017b; Cermakova et al., 2023). Despite their potential (Mottola et al., 2022; Albonetti et al., 2023), these techniques are underutilized in elasmobranch detection globally, with no papers published on Brazil yet. Increased research funding for Brazilian research is essential to stay abreast of recent international advancements, and to identify shark and ray presence in processed products like crab dishes, pet food, and cosmetics and contribute to conservation efforts and species regulation (Cardeñosa, 2019; Alvarenga, 2020).

4.3. Threatened Elasmobranchs in Brazilian Trade: A Call to Action for Conservation

The genetic findings highlight the urgent need for conservation measures to protect Brazil's diverse elasmobranch species, especially those already threatened, including CR and endemic species. These species are particularly vulnerable to local fishing practices and market demand, and their loss would significantly impact biodiversity (Pacoureau et al., 2023). Overfishing and bycatch of elasmobranchs poses significant risks, including ecological disruptions within the food chain as these species serve as top- and meso-predators (Myers et al., 2007). The reduced genetic diversity further exacerbates the situation, affecting the overall health and resilience of populations (Jump et al., 2009). This limits their ability to adapt to environmental changes, making them more vulnerable to diseases and climate change impacts (Hampe & Petit, 2005; Domingues et al., 2018; Carvalho et al., 2019). Additionally, low genetic diversity can lead to decreased reproductive success and reduced fitness, ultimately compromising the long-term survival and evolutionary potential of the species (Lande, 1998; Domingues et al., 2018).

Among the elasmobranch species detected here, hammerhead sharks and guitarfishes are particularly vulnerable as they continue to be sold in alarming numbers, despite their high extinction risk. Seven out of eight hammerhead sharks and all guitarfishes occurring in Brazil are categorized as threatened (IUCN, 2023). This situation emphasizes the significant impact of coastal fisheries to the risk of extinction, especially for endemic species like the guitarfishes, as well as other highly threatened and extensively traded elasmobranchs, such as the angel sharks and sawfishes (Faria et al., 2013; Bunholi et al., 2018). Intense fishing pressure on sharpnose sharks (*Rhizoprionodon* spp.) is another conservation concern. Despite their relatively high reproductive rate, relentless fishing has driven two sharpnose sharks with distribution in Brazilian coast (*R. porosus* and *R. lalandii*) to become VU, as previously cautioned by Lessa et al. (2006). The frequent catch of Carcharhinidae species overall is of concern, and the highly traded Blue Shark may face similar fate as sharpnose sharks. Other carcharhinid sharks are prominently sold in Brazil, also experiencing high fishing mortality rates (Bond et al., 2012; Dulvy et al., 2014).

The high rate of threatened species found in this paper emphasize the abovementioned. We illustrated a developing trend in extinction risk status, where a growing number of species, especially the frequently traded ones, are becoming increasingly threatened. Overall, 32 out of the 65 species found in this study were reassessed recently in the IUCN Red List, with 18 experiencing an increase in the extinction risk, including seven species receiving multiple uplisting in their extinction risk. A single species, *Rhinoptera brasiliensis*, previously categorized as EN showed slight improvement, but remains VU (Figure S3). However, this positive change is overshadowed by the overall worsening extinction risk status of many other species. These findings emphasize the detrimental impact of fisheries and trade on elasmobranch populations (Cawthorn & Mariani, 2017), adding further pressure to their declines. The remaining 16 species were previously DD, being 56% now assigned to threatened categories, such as *Rhizoprionodon lalandii* (DD – VU) and *Carcharhinus porosus* (DD – CR). The remaining 44% of reassessed DD species, which were not assigned to threatened categories, had lower detection rates in our trade analysis, such as *Squalus cubensis*, (DD – LC). Two freshwater rays species detected in the papers analyzed remained assessed as DD, emphasizing the need for more research on freshwater elasmobranchs. It is important to note that many DD elasmobranch species have low levels of genetic diversity, which makes them even more vulnerable to stochastic events (Domingues et al., 2018).

Comprehensive conservation strategies are necessary to safeguard and restore genetic diversity among elasmobranch populations in Brazil (Becerril-García et al., 2022). Protecting elasmobranchs is not only vital for biodiversity conservation but also for the livelihoods of local communities dependent on fishing. Implementing effective conservation strategies locally, regionally and globally is vital to prevent the loss of these invaluable species and safeguard marine biodiversity. Given Brazil's extensive coastline, high diversity of endemic species, and its significant role in conservation efforts, the country's actions can have a substantial impact on elasmobranch preservation worldwide (Becerril-García et al., 2022). Conservation strategies crucial for preventing further population declines and genetic erosion should prioritize habitat protection, promote sustainable fishing practices, establish marine protected areas, regulate trade, minimize bycatch, and manage sustainable fishing quotas. Collaboration among the Brazilian government, stakeholders, and international organizations is essential to ensure the success of these conservation efforts.

4.4. Legislative Measures in Brazil: Towards Effective Elasmobranchs Protection?

Fish conservation regulations in Brazil have evolved over the years, reflecting the country's political landscape. Past initiatives, like anti-finning regulations, showcased Brazil's dedication to elasmobranch conservation. The incorporation of Red Book assessments into legislation has addressed the trade and exploitation of threatened elasmobranchs. However, challenges emerged with the substitution or repristination of previous regulations with the introduction of new regulation actions by ordinances MMA No. 148/2022 and No. 354/2023, raising concerns of fisheries interests over conservation priorities. It could be expected that future regulations based on the SALVE System assessments will bring renewed commitment encouraging improved species management and conservation practices. Even though the recent release of the SALVE System improved the quantity of species assessed and updated previous extinction risk status, discrepancies between the IUCN Red List still occur, with a high rate of species still to be evaluated. We also found difference in the extinction rate between the international and national lists analyzed (Figure 5a), including in endemic species, which indicates inconsistent categorization for geographically restricted species. This is attributed to outdated assessments, limited updates in the Brazilian threatened species lists, and factors like data availability, assessment methodologies, political considerations, and regional conservation priorities. To enhance consistency and effectiveness, regular updates, improved integration of data and assessment methodologies, and accurate assessment of extinction risks and conservation needs are crucial. The IUCN Red List provides a global analysis of extinction risk status, however, nationally it is essential that the extinction risk based on the current Brazilian legislation is considered too. Even though the labeling regulations were also updated over time, there were no significant changes for elasmobranch species, that can still be traded under umbrella-labels nationwide. The lack of species-specific labeling and updated scientific names for sharks and rays in fish labeling regulations poses significant challenges for trade monitoring and conservation efforts (Bornatowski et al., 2013). It impedes proper species identification, sustainable fishing practices, and conservation effectiveness. Without accurate identification, tracking and regulating the trade of threatened shark species becomes difficult, potentially hiding and escalating their decline.

4.5. The Mislabeling Risk: Deceptive Practices Beyond Label Swaps

Generic labeling is also a concerning reality for elasmobranchs in Brazil. Besides the use of umbrella labels at markets and restaurants, we also observed generic labeling of fish in education institution restaurants, where elasmobranchs are served without proper species identification, misleading unaware customers to consume shark meat (Alvarenga et al., 2021). Further investigations are essential to evaluate these cases, as there are also concerns about the presence of shark meat in hospitals, posing risks to both conservation efforts and public health due to high mercury levels in shark meat (Barreto et al., 2017). Identifying and exposing such activities is crucial to raise awareness, strengthen regulations, and potentially prohibit these practices. Recently, shark meat purchased by the municipality of São Paulo for distribution in schools was recalled due to public pressure, highlighting the effectiveness of raising awareness and taking action against such practices (Bonin, 2021).

Shark and ray products have also been distributed under mislabeled names such as salmon and croaker (Staffen et al., 2017), while conversely, bony fishes have been frequently sold as elasmobranchs (Almeron-Souza et al., 2018; Calegari et al., 2019; Cruz et al., 2021). Elasmobranchs have been mislabeled not only as bony fish but also as other elasmobranch species (Palmeira et al., 2013; Almeron-Souza et al., 2018; Bunholi et al., 2018; Bernardo et al., 2020; Alvarenga et al., 2021). This deceptive practice not only undermines consumer trust but also poses an especially significant threat to threatened rays, which are sold at low cost (Niedermüller et al., 2021).

We also noted instances of incorrect labeling and incomplete information in the papers analyzed. The misidentifications noted comprised: 1) Failure to differentiate between information related to individual samples, making it impossible to verify the sampling location and label associated with each sample (Table S1, in Appendix 2); 2) Variation on the availability of information on sampling date, market sector, price, and origin of capture among the papers. Some papers provided comprehensive information on all these aspects, while others had missing or incomplete data (Table S2, Appendix 1); 3) Non-standard labeling information, often representing only the regional names rather than the regulated name under legislative measures (Table S1, in Appendix 2). While documenting diverse regional terminology for traded sharks and rays offers valuable insights into market and cultural distinctions within Brazil, it remains important to also incorporate the label of the marketed product. In cases where such designations are absent, explicit acknowledgment should be made in the paper, as the absence could hinder effective identification of mislabeling patterns. Avoiding such mistakes in future papers will be essential to assess the magnitude and dynamics of the Brazilian trade and develop an accurate national-scale analysis of market activity.

5. CURRENT RESEARCH GAPS AND REQUIRED ACTIONS

In conclusion, the present study emphasizes the need to address general research gaps to enhance our ability to assess the trade at a national scale. By doing so, we can develop effective management and conservation measures to protect shark and ray species in Brazil. It is crucial to address these limitations to improve our understanding and take necessary actions to safeguard these vulnerable species.

Research Gaps:

1. Deficiency of papers conducted in the Northeast region of Brazil.
2. Absence of standardized design and sampling protocols, including comprehensive documentation of sampling location, date, price, and market origin.
3. Non-standard or absent labeling information hindering the assessment of mislabeling activity and comprehensive understanding of the dynamics of the shark and ray trade.
4. Concentration of papers primarily focused on analyzing the shark trade, with limited research on ray trade and other types of trades, such as bycatch and illegal finning activities in Brazil.
5. Insufficient application of cost-effective genetic techniques such as PCR Multiplex and PCR-RFLP in Brazilian papers.
6. Lack of utilization of high-throughput technologies to enhance species identification in Brazil, with no research being conducted to explore the potential application of elasmobranch products in mixtures or to track the geographic origin of products.
7. Scarcity of DNA sequences from important Brazilian elasmobranch species, particularly ray species, in genetic databases.

8. Continued trade in threatened and protected species, negatively impacting their conservation status over time.
9. Absence of extinction risk based on the current Brazilian legislation in papers, limiting the comprehensive understanding of legislation compliance.
10. Need for more robust legislation to regulate elasmobranch labeling and restrain mislabeling activities.

Required Actions:

1. Allocate additional funding for research efforts in the Northeast region of Brazil to address regional disparities and enhance understanding of elasmobranch trade and conservation needs.
2. Establish minimal standardized study design and sampling protocols, ensuring detailed documentation of sampling location, date, price, and market origin for accurate and comparable data.
3. Implement standardized labeling notes when sampling elasmobranch products to enable a comprehensive assessment of mislabeling activity and enhance understanding of the dynamics of the shark trade.
4. Expand research to include analysis of ray trade, bycatch, and finning activities in Brazil to obtain a holistic understanding of elasmobranch trade dynamics.
5. Promote the utilization of cost-effective DNA-based tools such as PCR Multiplex and PCR-RFLP in Brazilian papers to improve efficiency and accessibility of species identification, mainly involving the adoption of available tools.
6. Develop and implement new molecular markers based on high-throughput sequencing, such as metabarcoding and single nucleotide polymorphism (SNP).
7. Establish a regularly curated national database with precise species identification and molecular markers, including haplotype differentiation, to effectively support geographical assessments and enhance research and conservation efforts. Foster collaborative initiatives to generate genetic data from significant Brazilian elasmobranch species, especially rays, and contribute to the expansion of molecular markers in the national genetic database.
8. Strengthen enforcement measures and regulations to minimize the trade of threatened and protected species, safeguarding their conservation status through efficient monitoring and frequent inspections.
9. Incorporate both IUCN Red List and current Brazilian legislation extinction risk status into papers to evaluate compliance with legislation.

10. Advocate for new legislation mandating specific and updated species names and discouraging the use of ambiguous labels to enhance transparency and accuracy in elasmobranch trade. Also, implement stricter legislation and monitoring systems to regulate elasmobranch labeling, combat mislabeling activities, and ensure accurate product information throughout the supply chain, enhancing traceability and accountability.

6. APPENDIX

Additional information can be found in the online version of the article at the publisher's website.

Appendix 1 includes detailed information on data acquisition and supplementary results supporting our findings.

Appendix 2 provides the main metadata built to analyze the data from all peer-reviewed papers collected, alongside a detailed explanation of each category.

Appendix 3 offers additional metadata to support our analysis on the availability of genetic resources for Brazilian species and the evolution of Brazilian legislative measures, as well as national and international conservation lists.

Appendix 4 provides detailed R scripts for all analyses performed in this paper. The data used with the R scripts are available at GitHub (link available upon acceptance) to ensure reproducibility.

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8. FIGURES AND TABLES

TABLE 1. Summary of 33 research papers using DNA tools to investigate elasmobranch trade in Brazil from 2008 to 2023, including paper categories, sample sizes, and the number of elasmobranch and endangered species identified (based on the IUCN Red List at publication and current status), dominant species per paper, the Brazilian regions studied, type of DNA-based tool and molecular marker applied, and the determination of whether the paper was mislabeled or not.

Authors	**Paper category**	**Sample size**	**Elasmo branch species**	**IUCN Listed (paper)**	**IUCN Listed (current)**	**Brazilian Region**	**Molecular marker**	**DNA-based tool**	**Most frequent species**	**Mislabel ‡**
Pinhal et al. (2008)	Methods	NA	8	NA	7	All coastal regions	5S rDNA	Tandem Repeats	NA	NO
Mariguela et al. (2009)	Methods	145	3	NA	3	Southeast and South	16S/COI	PCR RFLP	*Pseudobatos percellens*	NO
Mendonca et al. (2009)	Methods	86	2	NA	2	Northeast and Southeast	COI	PCR Multiplex	*Rhizoprionodon porosus*	NO
Pinhal et al. (2009)	Methods	36	2	NA	2	Brazilian	5s rDNA	PCR RFLP	NA	NO
Rodrigues-Filho et al. (2009)	Trade Analysis	122	11	1	10	North	12S-16S	DNA sequencing	*Carcharhinus porosus*	YES
De-Franco et al. (2010)	Methods	145	3	NA	3	Southeast and South	COI	PCR Multiplex	*Pseudobatos percellens*	NO
Mendonca et al. (2010)	Methods	443	25	NA	20	All coastal regions	COI	PCR Multiplex	NA	NO
Pinhal et al. (2012)	Methods/ Trade Analysis	62/90*	7	NA	4	All coastal regions	ITS2	PCR Multiplex	*Rhizoprionodon terranovae*	NO
De-Franco et al. (2012)	Trade Analysis	267	3	NA	3	Northeast, Southeast and Southern	COI	PCR Multiplex	*Pseudobatos horkelii*	NO
Palmeira et al. (2013)	Trade Analysis	44	8	NA	7	Northern	16S/CytB	DNA sequencing	*Pristis perottepeerti*	YES
Ribeiro et al. (2012)	Trade Analysis	41	13	NA	10	Southeast	COI	DNA sequencing	*Atlantoraja castelnaui*	NO
Domingues et al. (2013)	Trade Analysis	317	4	2	4	Southeast	ITS2/COI	PCR Multiplex	*Carcharhinus falciformis*	YES
Faria et al. (2013)	Trade Analysis	77	1	1	1	Northern	CytB	DNA sequencing	NA	NO
Falcão et al. (2016)	Methods	9	3	NA	3	Southern	CytB	PCR RFLP	NA	NO
Schmidt et al. (2015)	Methods/ Trade Analysis	97	4	0	2	Southeast	COI	PCR RFLP	*Dasyatis hypostigma*	NO

Schmidt et al. (2015)	Trade Analysis	1	1	1	1	Southern	COI	DNA sequencing	NA	NO
Nachtigall et al. (2017)	Methods/ Trade Analysis	67/51**	8	1	7	Northern	ITS2	PCR Multiplex	*Rhizoprionodon porosus*	YES
Carvalho et al. (2017a)	Trade Analysis	8	1	NA	0		COI	DNA sequencing	NA	NO
Staffen et al. (2017)	Trade Analysis	14	5	2	3	Southern	COI	DNA sequencing	*Prionace glauca*	YES
Almeron-Souza et al. (2018)	Trade Analysis	63	18	8	14	Southern	COI	DNA sequencing	*Prionace glauca*	NO
Bunholi et al. (2018)	Trade Analysis	85	3	3	3	Southern	COI	DNA sequencing	*Squatina guggenheim*	YES
Feitosa et al. (2018)	Trade Analysis	427	17	4	15	Northern	COI	DNA sequencing	*Sphyrna mokarran*	NO
Marques et al. (2020)	Methods	279	10	5	8	Southeast	COI/CytB	Morphometrics/DNA sequencing	*Gymnura altavela*	NO
Calegari et al. (2019)	Trade Analysis	7	0	NA	NA	Southeast	COI	DNA sequencing	NA	NO
Ferrette et al. (2019a)	Trade Analysis	228	17	5	12	Southeast	COI	DNA sequencing	*Dasyatis sp.*	NO
Ferrette et al. (2019b)	Trade Analysis	747	20	9	18	Northern, Northeast and Southeast	COI	PCR Multiplex and DNA sequencing	*Prionace glauca*	NO
Bernardo et al. (2020)	Trade Analysis	231	16	7	12	Southern	COI	DNA sequencing	*Prionace glauca*	NO
Camacho-Oliveira et al. (2020)	Trade Analysis	52	4	1	2	Southeast	COI	DNA sequencing	*Paratrygon ajereba*	NO
Guimaraes-Costa et al. (2020)	Trade Analysis	73	20	4	13	Northern	COI	DNA sequencing	NA	NO
Rodrigues et al. (2020)	Trade Analysis	118	9	2	5	Northern	COI	DNA sequencing	*Hypanus guttatus*	NO
Alvarenga et al. (2021)	Trade Analysis	220	17	13	15	Southeast	COI	DNA sequencing	*Prionace glauca*	YES
Cruz et al. (2021)	Trade Analysis	56	9	6	8	Southern	COI	DNA sequencing	*Prionace glauca*	YES
Martins et al. (2021)	Trade Analysis	127	20	12	17	Northern	COI	DNA sequencing	*Sphyrna mokarran*	YES

Merten-Cruz et al. (2021)	Trade Analysis	57	17	7	13	All coastal regions	COI	DNA sequencing	*Prionace glauca*	NO
Souza-Araujo et al. (2021)	Trade Analysis	91	13	4	12	Northern	COI	DNA sequencing	*Mustelus higmani*	NO

‡ Mislabel = the paper analyzed used the results provided by DNA analysis to infer if the fish sold was mislabeled. * This paper used 166 samples to design the DNA rapid assay, only 62 of which were collected in Brazil. Also, they applied the technique to 90 other samples collected in Brazilian markets. ** This paper used 67 samples to design the DNA rapid assay, 10 of which were newly collected and 57 from other papers. Also, they applied the technique to 51 other samples collected in Brazilian markets.

TABLE 2. Summary of the extinction risk status and number of shark and ray species found in the papers analyzed. The table includes species categorized CR, EN, VU, NT, LC, DD by the International Union for Conservation of Nature (IUCN) Red List of Threatened Species, and a detailed list of all species found.

Status	**Number of Species**	**Species**
Critically Endangered	12	*Atlantoraja castelnaui, Carcharhinus porosus, Carcharias taurus, Fontitrygon geijskesi, Galeorhinus galeus, Isogomphodon oxyrhynchus, Pristis pristis, Pseudobatos horkelii, Sphyrna lewini, Sphyrna mokarran, Sphyrna tudes, Squatina occulta*
Endangered	20	*Aetobatus narinari, Atlantoraja cyclophora, Carcharhinus acronotus, Carcharhinus obscurus, Carcharhinus perezi, Carcharhinus plumbeus, Carcharhinus signatus, Centrophorus squamosus, Dasyatis hypostigma, Gymnura altavela, Isurus oxyrinchus, Isurus paucus, Mobula thurstoni, Mustelus higmani, Pseudobatos percellens, Sphyrna tiburo, Squalus mitsukurii, Squatina guggenheim, Styracura schmardae, Zapteryx brevirostris*
Vulnerable	20	*Alopias superciliosus, Bathytoshia centroura, Benthobatis kreffti, Carcharhinus brachyurus, Carcharhinus brevipinna, Carcharhinus falciformis, Carcharhinus leucas, Carcharhinus limbatus, Carcharodon carcharias, Ginglymostoma cirratum, Hypanus berthalutzae, Hypanus dipterurus, Myliobatis freminvillei, Myliobatis goodei, Rhinoptera bonasus, Rhinoptera brasiliensis, Rhizoprionodon lalandii, Rhizoprionodon porosus, Rioraja agassizii, Sphyrna zygaena*
Near Threatened	8	*Carcharhinus altimus, Galeocerdo cuvier, Gymnura micrura, Hypanus americanus, Hypanus guttatus, Mustelus canis, Narcine brasiliensis, Prionace glauca*
Least Concern	3	*Carcharhinus galapagensis, Rhizoprionodon terraenovae, Squalus cubensis*
Data Deficient	2	*Paratrygon aiereba, Potamotrygon motoro*

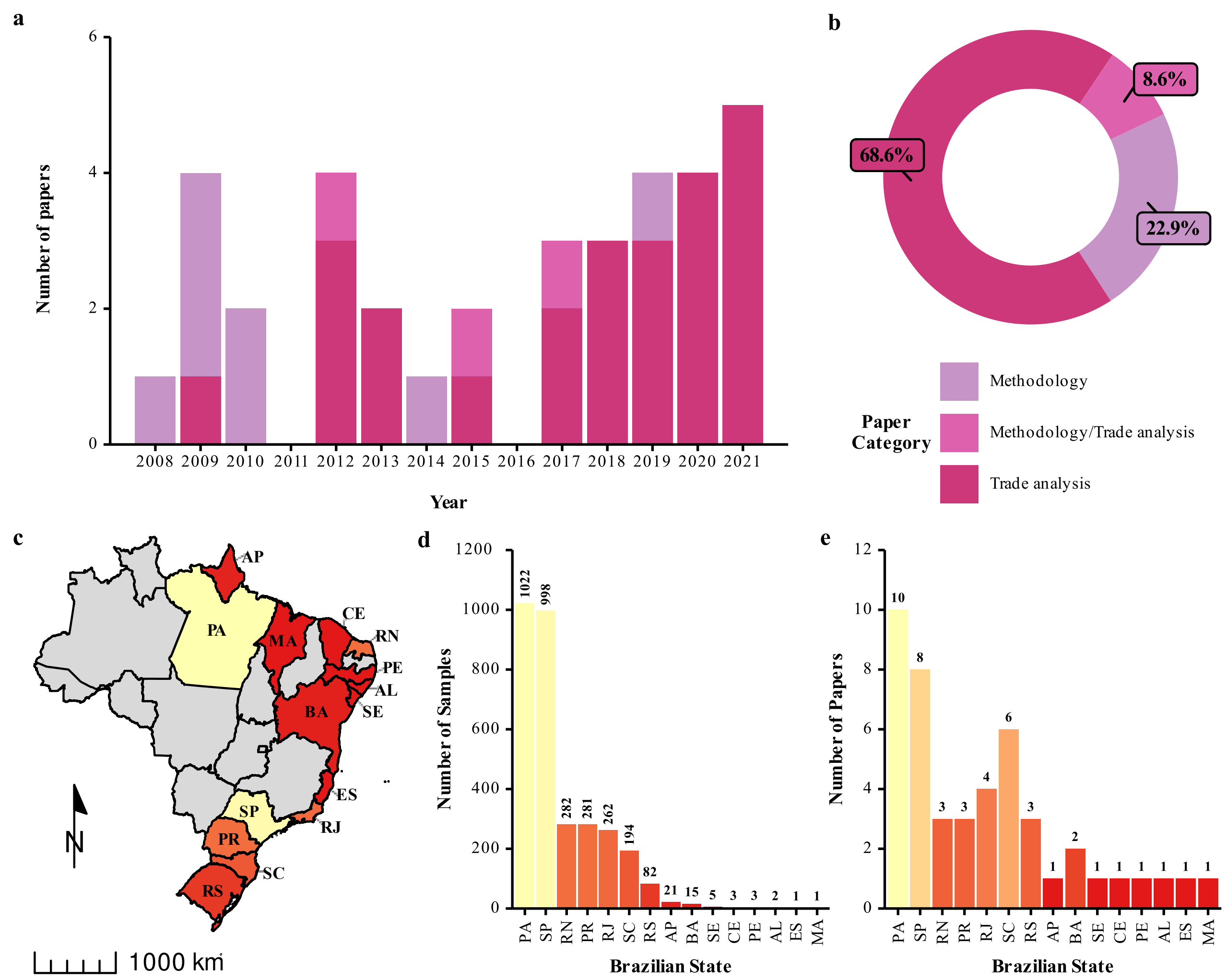

FIGURE 1. Overview of the papers published on the topic of DNA tools to analyze elasmobranchs trade in Brazil between 2008 and 2023, including (A) year of publication, (B) type of research in each paper, and number of samples per (C) Brazilian region, (D) papers and (E) samples. Quantity of paper is inversely proportional to the darkness in shade of colors (i.e., in C the regions in dark shades are the ones with less papers published, in D the dark shades mean less samples available, and in D less papers published).

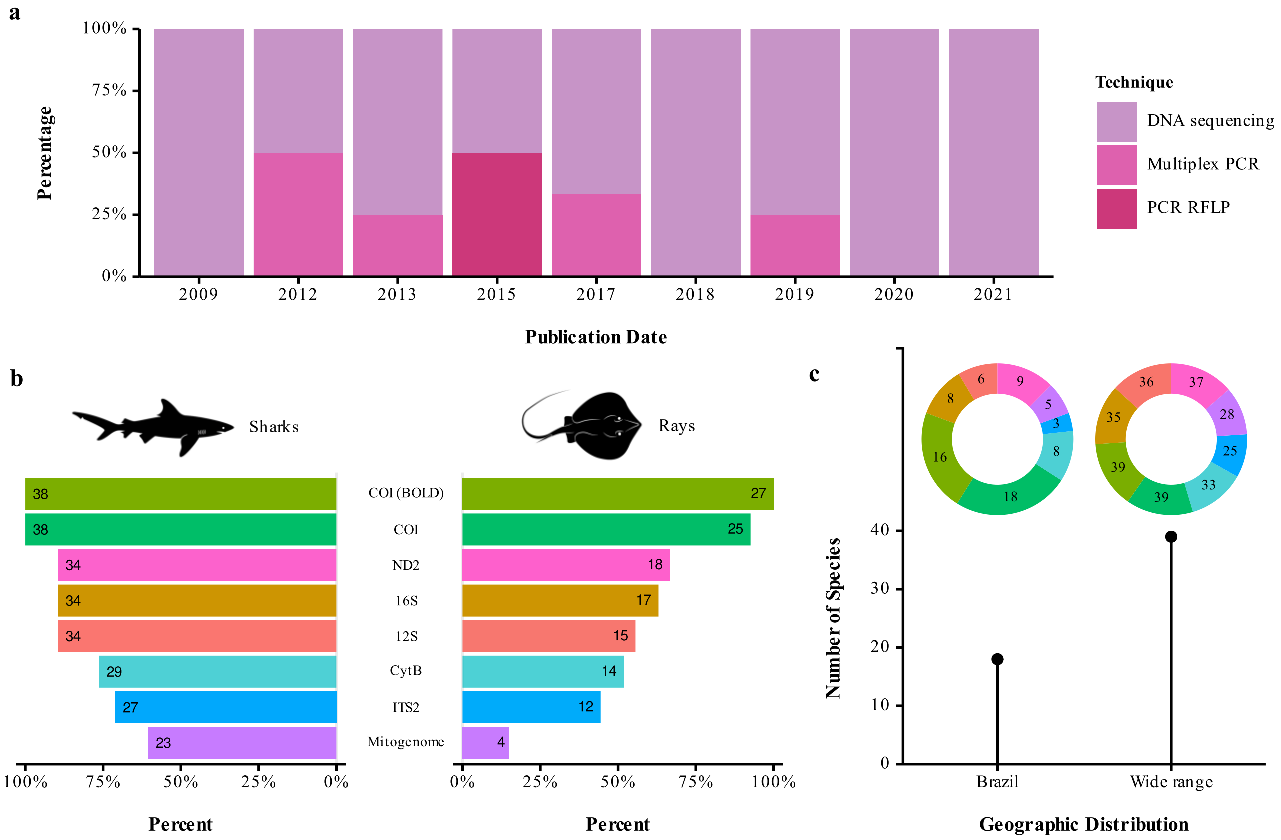

FIGURE 2. DNA tools in elasmobranch trade analysis. (a) Evolution of the DNA techniques applied through time, (b) number of shark and ray species found in all papers (missing this info in this draft), with the availability of sequences in GenBank and BOLD databases per molecular marker for shark (in green) and ray (in orange) species, and (c) number of sequences available in both databases for endemic species of Brazil versus wide-range species.

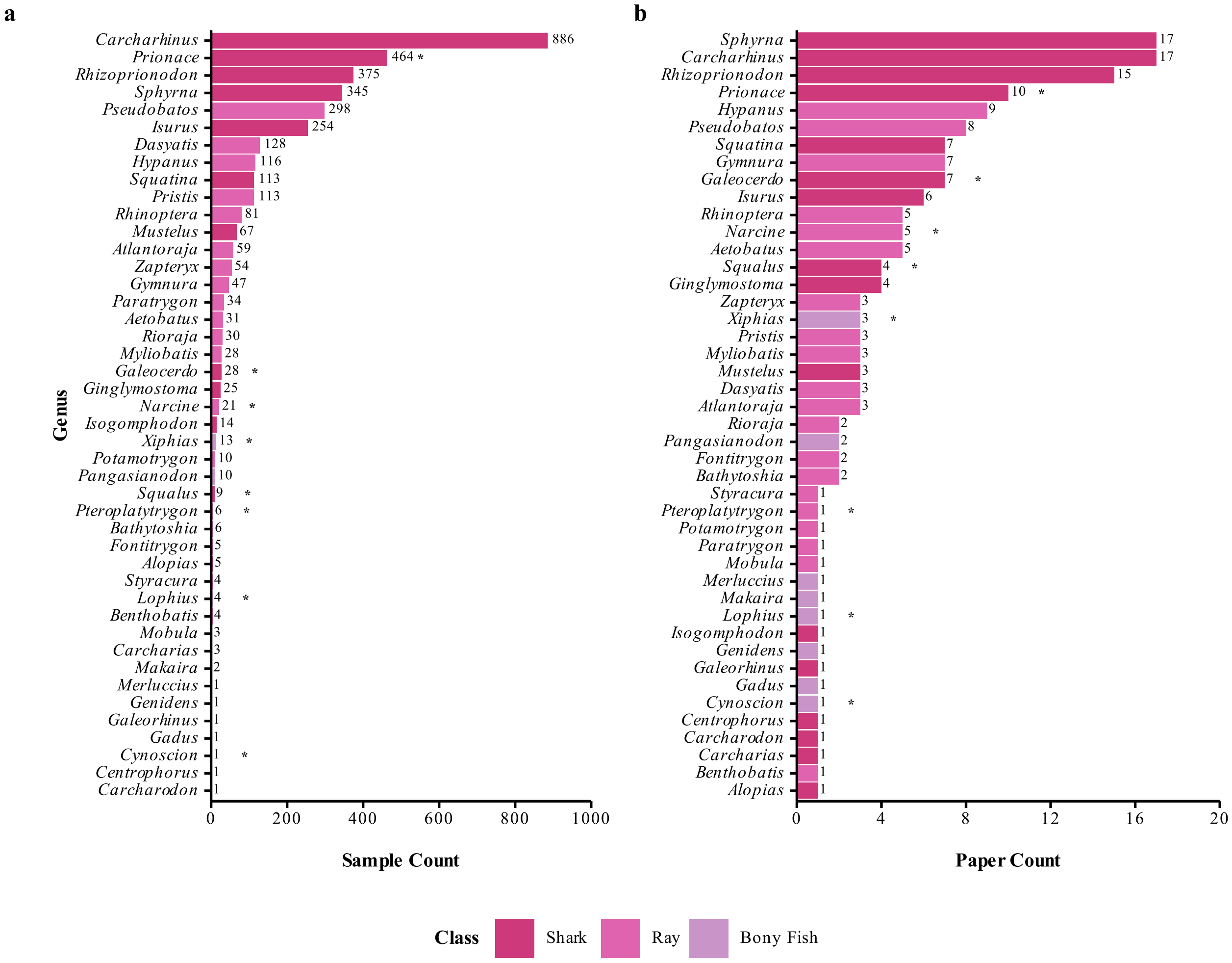

FIGURE 3. The (a) sample count and (b) paper count of genera found in all papers analyzing the elasmobranch trade in Brazil between 2008 and 2023. The class of each genus (shark, ray or bony fish) is defined by the color in the caption. The genera that did not contain endangered species are marked with *.

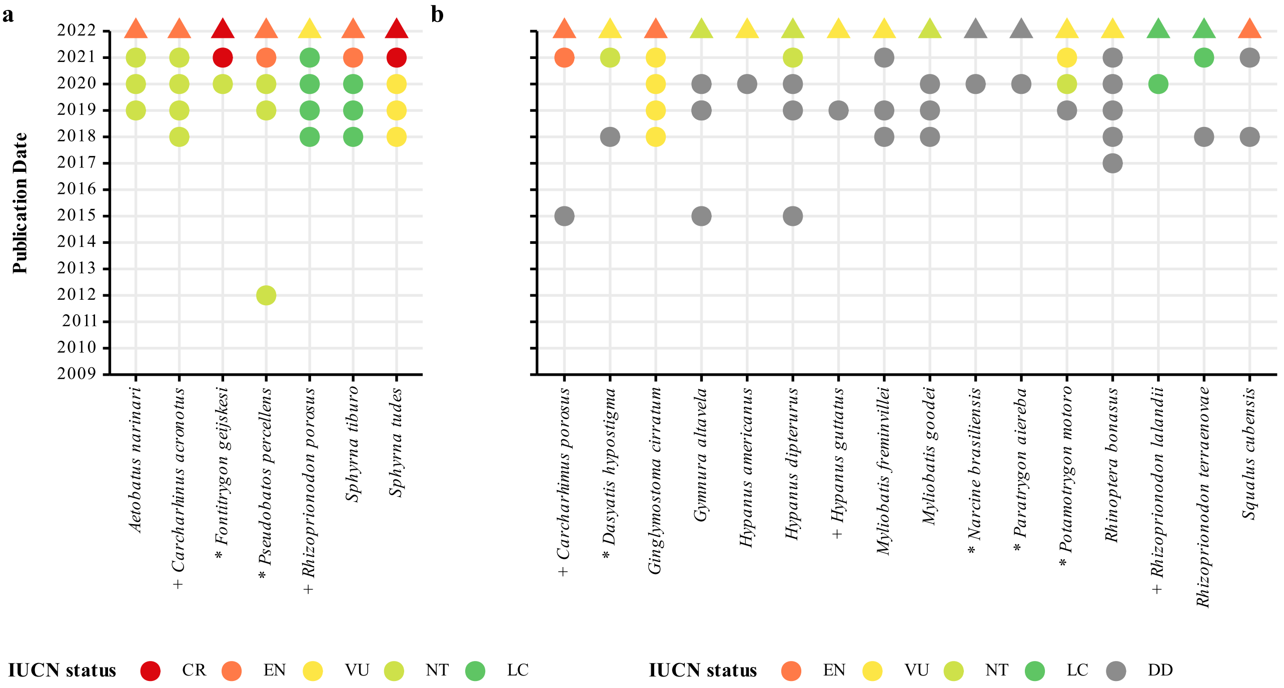

FIGURE 4. Evolution of extinction risk assessment over time for species found in the present study (a) that increased by two statuses and (b) that was previously categorized as data deficient. The category assigned at the time the paper was published is represented by a circle, while the current category is represented by a triangle. The assessment for all elasmobranch species found in the present study can be found in Figure S3.

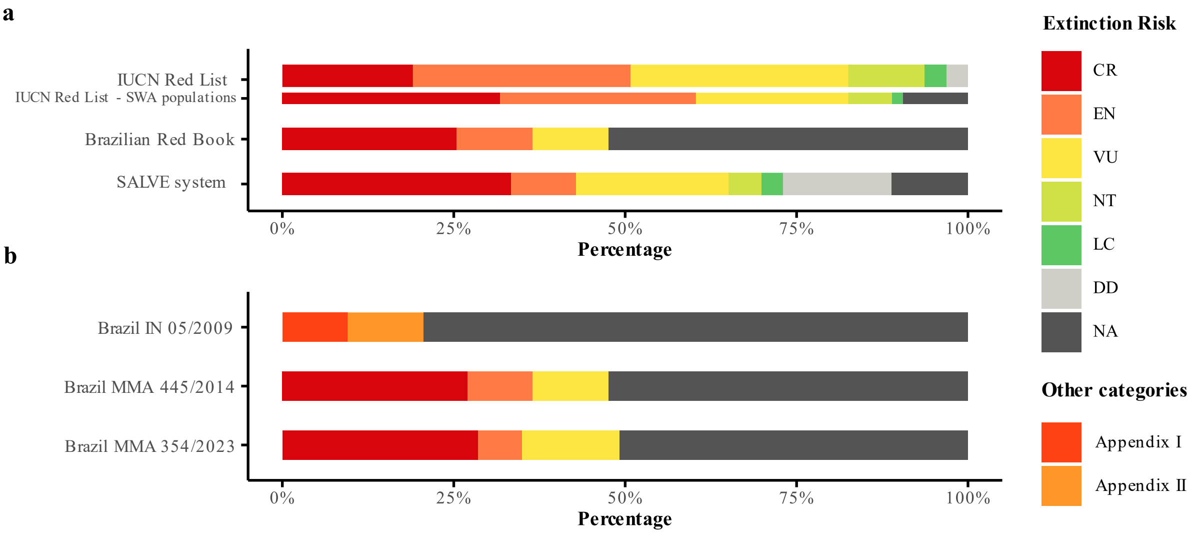

FIGURE 5. Categorization of all elasmobranch species identified in the present study according to (a) international (IUCN Red List considering global assessment and SWA population details) and national assessments of risk of extinction (Brazilian Red Book of Threatened Fauna and SALVE System), and (b) the three main Brazilian ordinances. The Brazilian Ordinance IN05/2004 was the only one that did not follow the standard extinction risk categories (DD, LC, NT, VU, EN, CR), adopting instead two categories (Threatened and Overexploited).

Fifteen years of extensive and inadvertent elasmobranch trade in Brazil detected by DNA tools

APPENDIX 1. Data collection & Analysis | Supplementary Results

Boolean search. To perform the retrieval of studies analyzed here, we followed the ROSES protocol (Haddaway et al. 2018). We performed an extensive Boolean search (AND, OR, NOT) on Web of Science and Scopus to collect papers that have applied DNA data to analyze the catch and trade of sharks and rays in Brazil. Also, papers that developed genetic tools for molecular identification of species. We searched for keywords, titles, and abstract using words for mislabeling, molecular identification and to establish the main group: ("mis*label*" OR "misidentification" OR "molecular tool*" OR "genetics tool*" OR "DNA tool*" OR "DNA barcod*" OR "forens* genetic*" OR "genetic* forens*" OR "molecular identification" OR "genetic* identification" OR "forens* identification") AND ("elasmobranch*" OR "shark*" OR "ray*" OR "guitar*fish*" OR "stingray*" OR "skate*" OR "skateray*"). Since the word ‘ray’ can also be referred to in studies of radiation (ionic rays), taxonomy (as a morphological trait), and diseases (Ray’s fluid, a technique), we limited our search to not retrieve the words: ("x*ray" OR "radiation" OR "radiograph*" OR "Ray* fluid" OR "integrative taxonomy"). We chose not to exclude the word “taxonom*” alone due to their common use to explain the necessity of DNA tools in mislabeling studies. Three filters were also applied: (i) research performed in Brazil, (ii) published until 2022 June 31st, and (iii) research paper. We also performed a search in Scholar using the keywords “Brazil (shark OR ray OR guitarfish OR stingray OR skate) AND mislabel* OR forensic OR genetics OR "molecular identification"”.

Data cleaning. In order to merge data from both databases and remove potential duplicates, we used the R package Bibliometrix (Aria and Cuccurullo 2017). A total of 62 papers were retrieved, and after a manual inspection by three authors (MA, IVB and VPC), we removed 34 studies not relevant for this analysis. We also manually inspected the Scholar search, removing potential duplicates and papers not related to this meta-analysis, retaining four more studies.

Inter-rater agreement. To add a paper to our database, we choose full agreement criteria (i.e. when the paper is selected by the three authors). To calculate the rate of concordance among authors, we utilized the inter-rater agreement metric. A good inter-rater agreement is > 0.8. Here, we had values of 0.871 for the main search in Web of Science and Scopus, 0.916 for the Scholar search and 0.903 when merging all searchers.

Snowball method. Moreover, to search for potential missing papers, we applied the Snowball method (Wohlin 2014) in the 32 papers selected, searching for studies in the list of citations (forward snowballing) and in their cited references (backward snowballing), ending up with three additional papers.

Metadata. From the total 35 peer-reviewed papers, we extracted information for every sample to build metadata divided into "Trade Analysis" (papers that applied DNA methods to inspect the trade and landing of elasmobranchs), and "Methods" (papers that developed a methodology to analyse the trade). The two metadata can be assessed in tables S1 and S2 (Supporting Information 2). The "Trade Analysis" metadata includes 25 subcategories with 5 main categories: 1) study characteristics, 2) market characteristics, (including reported label), 3) product real identity, 4) molecular techniques and 5) conservation status. The "Methods" metadata has 13 subcategories, including the same first 4 main categories. The detailed information of both tables about all the specific subcategories can be assessed in table S2 (Appendix 2).

Data analysis. We conducted an exploratory quantitative analysis in R to investigate the application of DNA tools in analyzing the elasmobranch trade in the Brazilian market. Firstly, we assessed the number of peer-reviewed papers and samples collected at both national and statewide levels. We compared the overall number of papers and samples across different Brazilian states and genera to identify variations in sampling sizes and strategies. Secondly, we examined the development of genetic tools used for species identification in elasmobranch trade. Our primary focus was on trade analysis, where we identified the most employed techniques for analyzing elasmobranch trade. We also determined the species composition of Brazilian markets, including endangered and endemic species. Furthermore, we evaluated s trends in the threat status of species found in Brazilian markets over time and identified cases of mislabeled products. To evaluate the effectiveness of legislation and genetic availability of Brazilian elasmobranch fauna, we reviewed the Brazilian legislation related to endangered species and the labeling of fisheries products and examined the extinction risk status in the main Brazilian lists. Our main goal was to analyze the number of species assessed in the Brazilian lists and covered by Brazilian legislation, also comparing with the IUCN RedList. With this, we analyzed trends in the extinction risk status of these species over time and identified any gaps or limitations in the current regulations. Additionally, we assessed the number of sequences for the species found in genetic databases to evaluate the current coverage of Brazilian species, with a focus on endemic species. For detailed information on all quantitative analyses conducted, please refer to the R Markdown script provided in Supporting Information 4 and to the available repository on GitHub (link available upon acceptance).

Supplementary Tables to support results found in this paper.

Table S1. Displaying method papers with applied resources in subsequent studies: Describing the utilized resource and the research field of the subsequent Study. This table showcases three out of the 11 Method Papers, while the remaining eight studies, as listed (De Franco et al. 2012, Falcão et al. 2016, Mariguela et al. 2009, Mendonça et al. 2009, Mendonça et al. 2010, Nachtigall et al. 2017, Pinhal et al. 2009, Schmidt et al. 2015), did not have their resources applied in other studies.

Paper that provided the resource	Specific resource applied	Paper that applied it	Subject studied with the resource provided
Pinhal et al. 2008	Primers	Pinhal et al. 2011	Evolution of stingrays
De-franco et al 2010	Multiplex PCR set	De-franco et al. 2012	Analysis of elasmobranch trade in Brazil
Pinhal et al. 2012	Primers	Davis et al. 2019	Genetic structure of sharpnose sharks
Pinhal et al. 2012	Primers	Nachtigal et al. 2017	Analysis of elasmobranch trade in Brazil
Pinhal et al. 2012	Primers	Manzanillas Castro & Acosta-López 2022	Analysis of elasmobranch trade in Ecuador

Table S2. Evaluating data completeness in analyzed papers: Column categories and scoring criteria. A score of 0 indicates information absence, while a score of 1 signifies information presence for each category.

Paper	City	State	Region	Market Label	Sampling Date	Market Sector	Capturing Sector	Mislabeling
Rodrigues-Filho_et_al._2009	1	1	1	1 (regional name)	1	1	0	0 (label absent in regulations)
De-Franco_et_al._2012	0	1	1	1	1	0	1	0
Ribeiro_et_al._2012	1	1	1	1 (species name)	0	1	1	0
Pinhal_et_al._2012	1	1	1	1	0	1	0	0
Domingues_et_al._2013	1	1	1	1	1	0	1	0
Faria_et_al._2013	1	1	1	1 (species name)	0	1	0	0
Palmeira_et_al._2013	1	1	1	1	0	1	0	1
Carvalho_et_al._2015	1	1	1	1	1	0	1	0
Schmidit_et_al._2015	1	1	0 (locations are not separately noted for each sample)	0	0	0	1	0
Carvalho_et_al._2017	0	1	1	1	0	1	0	0
Nachtigall_et_al._2017	1	1	1	1	0	0	1	0
Staffen_et_al._2017	1	1	1	1	1	0	1	1
Almeron-Souza_et_al._2018	1	1	1	0 (samples are not	1	1	0	0

				separately noted)				
Bunholi_et_al._2018	1	1	1	1	1	0	1	0
Feitosa_et_al._2018	1	1	0 (locations are not separately noted for each sample)	1	1	1	1	0
Calegari_et_al._2019	0	0	1	1	1	0	1	0
Ferrette_et_al._2019a	1	1	1	1	1	0	1	0
Ferrette_et_al._2019b	1	1	1	1	0	0	1	0
Bernardo_et_al._2020	1	1	1	1	1	1	0	0
Camacho-Oliveira_et_al._2020	1	1	1	1	0	1	0	0
Guimarães-Costa_et_al._2020	1	1	1	1	1	0	1	0
Rodrigues-Filho_et_al._2020	1	1	1	1 (regional name)	0	1	0	0 (label absent in regulations)
Alvarenga_et_al._2021	1	1	1	1	1	1	1	1
Cruz_et_al._2021a	1	1	1	1	1	1	0	0
Cruz_et_al._2021b	1	1	1	1	1	1	0	1
Martins_et_al._2021	1	1	1	1 (regional name)	1	1	1	0
Souza-Araujo_et_al._2021	1	1	1	1	1	1	0	0

Supplementary Figures to support results found in this paper.

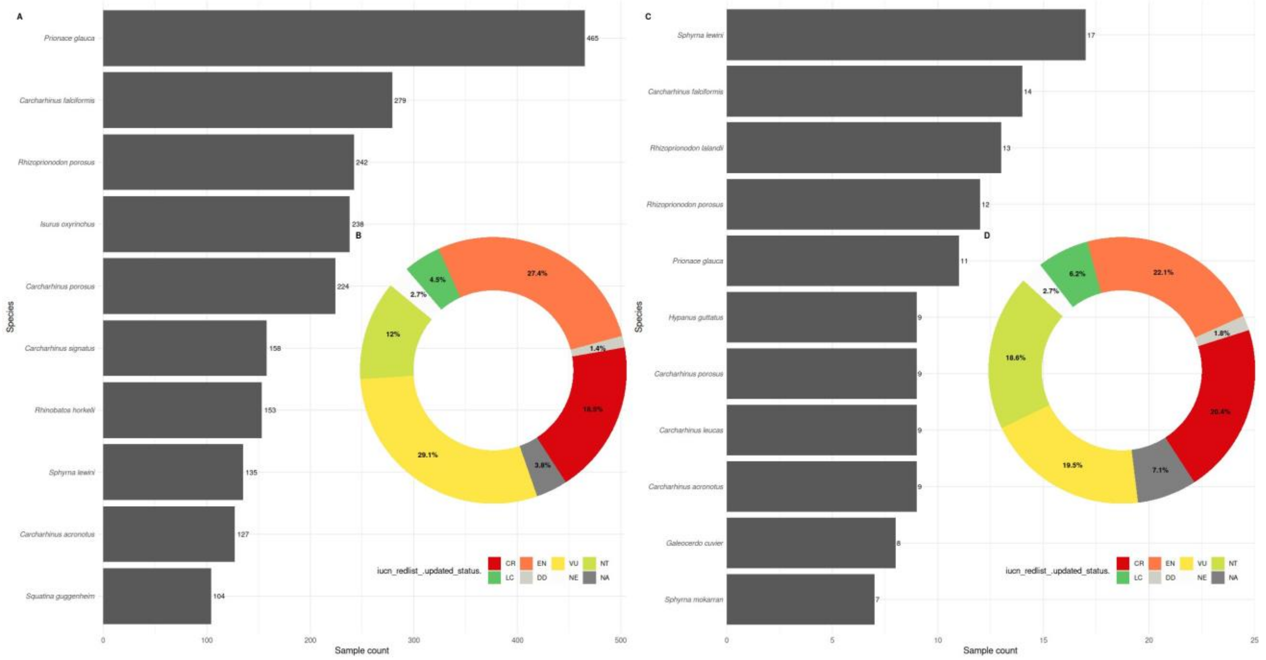

Figure S1. Top species found in all studies analysing the elasmobranchs trade in Brazil, divided by (a) sample count and (c) paper count, including the overall rate of endangered species, divided by (b) sample count and (d) paper count.

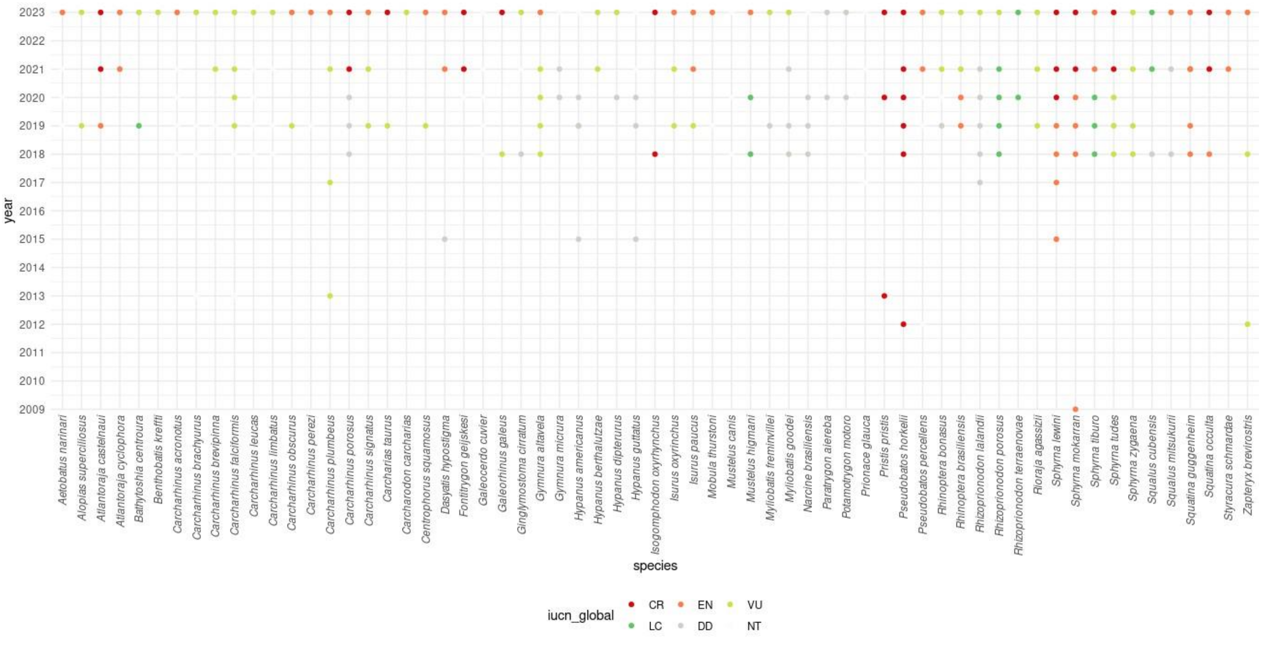

Figure S2. Evolution of extinction risk status over time for all species found in this study.

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