



Trophic ecology and bioindicator potential of the North Atlantic tope shark



Paulo Torres^{a,*}, Regina Tristão da Cunha^a, Rodrigo Maia^b, Armindo dos Santos Rodrigues^c

^a CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Pólo dos Açores - Departamento de Biologia, Universidade dos Açores. Rua Mãe de Deus, 58, 9500-801 Ponta Delgada, Açores, Portugal

^b CBA, Centro de Biologia Ambiental - SIIAF, Stable Isotopes and Instrumental Analysis Faculdade de Ciências da Universidade de Lisboa - Departamento de Biologia Vegetal, Edifício C2 - Sala 2.1.16, Campo Grande, 1749-016 Lisboa, Portugal

^c CVARG, Centro de Vulcanologia e Avaliação de Riscos Geológicos - Departamento de Biologia, Universidade dos Açores, Apartado 1422, 9501-801 Ponta Delgada, Açores, Portugal

HIGHLIGHTS

- We analysed *Galeorhinus galeus* muscle tissue, from Mid-Atlantic (Azores).
- We determined trophic ecology, stable isotope and trace metal concentrations.
- Results suggest the occurrence of a Mid-Atlantic tope shark population.
- Results reflect bioaccumulation and suggest biomagnification for As and Hg.
- The species may be used as a Mid-Atlantic bioindicator of environmental quality.

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ABSTRACT

Sharks are top marine predators vital in maintaining ecosystem health and food web structure. In order to assess tope shark (*Galeorhinus galeus*) trophic ecology, stable isotope ratios and trace metal concentrations in muscle tissue were determined, according to size and gender, for 124 individuals caught within the Mid-Atlantic region. Data was complemented and analysed according to previous stomach content information and compared with studies performed in the North East Atlantic. Our results revealed that tope sharks fed at a low trophic level and within a more pelagic-based food web when compared with other North Atlantic regions. MixSIR application reflected its piscivorous diet and study area topography, oligotrophic waters and volcanic nature, suggesting the occurrence of a Mid-Atlantic tope shark population. Considering a non-anthropogenic volcanic source for observed metal contents, the results reflect bioaccumulation and suggest biomagnification processes for As and Hg. These metals exceeded legislated maximum limits for some countries with a maximum of 28.98 ± 1.26 and $0.57 \pm 0.01 \text{ mg kg}^{-1}$ wet weight, respectively, increasing significantly with size for both males and females. Conversely, Cr, Rb and Zn were relatively stable while Cd and Pb were not detected. Hg and Se were strongly correlated, suggesting a Se toxicity mitigation role. Given the tope shark travel capacity and the results obtained, the species may be used as a Mid-Atlantic bioindicator of environmental quality.

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1. Introduction

Sharks are top predators thought to play a significant role in aquatic food webs, generally difficult to study in their natural environment. Two main approaches have been used to determine shark feeding habits: diet composition studies, which use the relative proportions of prey types and respective trophic level (Lucifora et al., 2006), and stable-isotope analyses which estimate assimilated foods based on nitrogen

and carbon stable isotope measurements in the tissues of marine consumers (Hussey et al., 2011).

The use of the naturally occurring isotopes of nitrogen ($\delta^{15}\text{N}$) and carbon ($\delta^{13}\text{C}$) to address ecological issues in marine organisms has been growing over the last decades. Several studies apply stable isotopes to investigate shark trophic levels, diet and diet switching, and isotope turnover rates (see Layman et al., 2012 for a review). This technique assumes that the isotopic composition of animal tissue reflects that of its prey with a trophic enrichment in $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of ~3.4‰ and 1‰, respectively (Post, 2002). While $\delta^{15}\text{N}$ profiles are often used as trophic level predictors (Hussey et al., 2011; Pinnegar et al., 2002), $\delta^{13}\text{C}$ values are useful in evaluating the sources of primary production

* Corresponding author at: Departamento de Biologia, Universidade dos Açores. Rua Mãe de Deus, 58, 9500-801 Ponta Delgada, Açores, Portugal. Tel.: +351 918784502. E-mail address: biol.paulo@gmail.com (P. Torres).

in marine ecosystems and are typically higher (less negative) near coastlines and lower (more negative) in oceanic plankton-based systems (Hussey et al., 2011).

Heavy metal biomagnification throughout the food web has been cited to explain levels much above background values in organisms living in the open ocean (Domi et al., 2005; Magalhães et al., 2007). Given their high trophic level, sharks accumulate significant levels of trace metals in their tissues (Branco et al., 2004; Domi et al., 2005). Additionally, long-lived, slow-growing and highly migratory oceanic fishes, such as tunas, billfishes and pelagic sharks, accumulate high concentrations of trace metals, for example, mercury, often exceeding the limits recommended for human consumption (Branco et al., 2004). The increased captures of elasmobranchs for human consumption in Europe emphasise the need for pollutant monitoring of these species (Storelli et al., 2001).

The tope shark *Galeorhinus galeus* (Linnaeus, 1758) (Carcharhiniformes: Triakidae) is a medium-sized cosmopolitan marine predator that occurs in temperate coastal and shelf waters. Its diet comprises mainly fishes and cephalopods, but their relative contributions vary. In Australia and South Africa, both prey constitute similar proportions (Walker, 1999), while in the Irish Sea (Ellis et al., 1996) and in Argentinean waters (Lucifora et al., 2006), tope sharks are more piscivorous (78% and 98%, respectively), as are those from the Azores (99% fishes; Morato et al., 2003). Some qualitative ontogenetic dietary differences have been reported upon, with juveniles consuming fewer fishes and cephalopods and more small invertebrates (Walker, 1999). Although studies on stomach contents have been performed, few references on their isotopic composition are available for specific regions of the North Atlantic Ocean (Domi et al., 2005; Pinnegar et al., 2002).

The Azorean volcanic islands and surrounding seas are located at a triple junction along the Mid-Atlantic Ridge, separated from the continent by at least 1300 km (Morton et al., 1998). The volcanic origin is reflected by active deep sea (Colaço et al., 2006) and shallow hydrothermal vent activities around the islands (Dionísio et al., 2013; Wallenstein et al., 2009), which release heavy metals into archipelago waters. The three deep sea hydrothermal vent fields, i.e. Rainbow, Menez Gwen and Lucky Strike, are located close to the Azores Triple Junction (ATJ) and are characterised by their different fluid chemical compositions, depths, geological contexts and related biological communities, which impacts all surrounding biodiversity (Desbruyères et al., 2001).

In this study, we examined size-based and gender-specific variations in diet and habitat use by tope shark muscle tissue caught in Mid-Atlantic Ocean waters around the Azores, by combining previous stomach content information, stable isotope profiles ($\delta^{15}\text{N}$ and $\delta^{13}\text{C}$) and heavy metal analyses. *G. galeus* represents a valuable socio-economic marine resource for the Azores and it is thus vital to study its role in the Mid-Atlantic food webs and to investigate its susceptibility to trace metal intake given the volcanic nature of the area.

2. Materials and methods

2.1. Sample collection and preparation

G. galeus individuals were sampled between March and June 2013. These were caught as by-catch in the artisanal long-line commercial fishery within the Mid-Atlantic region (Azores), located between 36–39°N and 25–31°W, ICES division X_{a2}. Individuals were identified, counted, measured (total length [TL]) and sexed. Maturity was determined by macroscopic examination of the gonads and claspers with maturity scales adopted as proposed by Stehmann (2002). Tissue muscle samples for stable isotope and trace metal analyses were excised from the block anterior to the first dorsal fin and stored frozen on arrival at the laboratory (−20 °C). To examine size-based and gender shifts in diet, samples were divided by sex and into three size classes [small (<80 cm); medium (80–100 cm); and large (>100 cm)]. The selected

size classes approximate maturity stages and relate to species-specific life history strategies (Compagno, 1984).

2.2. Stable isotope analyses

Organisms are thought to vary in their concentrations of lipids. As lipids are depleted in ^{13}C relative to the diet (Pinnegar and Polunin, 1999), they were extracted by soaking samples in triplicate 1:1 solutions of chloroform:methanol for 10 min (Beaudoin et al., 2001) and then rinsed with distilled water. The residue obtained was then stored at 50 °C for at least 48 h and reduced to powder. Approximately 1 to 2 mg of ground tissue was used to determine the ^{13}C and ^{15}N isotopic values. Stable isotope ratio analyses were performed at the Stable Isotopes and Instrumental Analysis Facility (SIAF) of the Centre for Environmental Biology (CBA), University of Lisbon (Portugal). $^{13}\text{C}/^{12}\text{C}$ and $^{15}\text{N}/^{14}\text{N}$ ratios in the samples were determined by continuous flow isotope mass spectrometry (CF-IRMS), on a Sercon Hydra 20–22 (Sercon, UK) stable isotope ratio mass spectrometer, coupled to a EuroEA (EuroVector, Italy) elemental analyser for online sample preparation by Dumas-combustion. Stable isotope ratios are expressed in δ notation as parts per thousand (‰) according to the following equation:

$$\delta X = \left[\left(\frac{R_{\text{sample}}}{R_{\text{standard}}} \right) - 1 \right] \times 1000 \quad (1)$$

in which X is ^{13}C or ^{15}N , R_{sample} is the corresponding ratio $^{13}\text{C}/^{12}\text{C}$ or $^{15}\text{N}/^{14}\text{N}$ and R_{standard} represents the ratio for the respective standard. The standards used were IAEA-N1 and USGS-35 for nitrogen isotope ratio, and IAEA-CH6 and IAEA-CH7 for carbon isotope ratio; $\delta^{15}\text{N}$ results referred to Air and $\delta^{13}\text{C}$ to PeeDee Belemnite (PDB). Precision of the isotope ratio analysis, assessed through the standard deviation between 6 and 9 replicates of laboratory standard material interspersed among samples in every batch, was $\leq 0.2\%$.

2.3. Trophic position (TP) estimation

G. galeus diet was quantified using stomach content data accessed from Morato et al. (2003). To summarise stomach content data, prey items were grouped into functional groups according to Cortés (1999). Trophic position was then estimated based on stomach content (TP_{SC}) and $\delta^{15}\text{N}$ (TP_{SI}). Dietary information for *G. galeus* was used in conjunction with the estimated TP of functional prey groups to calculate TP_{SC} using the following equation (Cortés, 1999):

$$\text{TP}_{\text{SC}} = 1 + \left(\sum_{i=1}^n p_i \times \text{TP}_i \right) \quad (2)$$

where TP_{SC} is the trophic position of the species in question, p_i is the proportion of each functional prey group i in the total diet (expressed as % of mass), and TP_i is the trophic position for each functional prey category (n). TP of functional prey groups was taken directly from Cortés (1999). Morato et al. (2003) studied sharks with 58.0–153.0 cm total length and did not find differences in stomach content composition with growth. TP_{SC} was therefore estimated for the overall species and not for each size or sex.

Trophic position using $\delta^{15}\text{N}$ was determined with the following equation:

$$\text{TP}_{\text{SI}} = \lambda + \frac{(\delta^{15}\text{N}_{\text{consumer}} - \delta^{15}\text{N}_{\text{base}})}{\Delta n} \quad (3)$$

where λ is the TP of the organism used to estimate $\delta^{15}\text{N}_{\text{base}}$, Δn is the enrichment in ^{15}N per trophic level, and $\delta^{15}\text{N}_{\text{consumer}}$ is the direct measurement of $\delta^{15}\text{N}$ for the target species (Post, 2002), in this case the tope shark. The species to be used as an estimate for $\delta^{15}\text{N}_{\text{base}}$ should share the same habitat as the target species and integrate the isotopic signature of the food web at a time scale large enough to minimise the effects

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