Marine Biology (2025) 172:155
https://doi.org/10.1007/s00227-025-04697-2

ORIGINAL PAPER

Trophic ecology of the longnose lancetfish based on stable isotope values ($\delta^{13}C$ and $\delta^{15}N$), mercury concentrations and stomach contents in the tropical western Pacific Ocean

Yu Ding[1] · Zezheng Li[1] · David Mboglen[1,2] · Yunkai Li[1,3,4]

Received: 31 July 2024 / Accepted: 20 June 2025

Abstract

Intensifying fishing pressure has profoundly altered marine food web structures across global ocean ecosystems. The longnose lancetfish (*Alepisaurus ferox*) is a widespread deep-sea predator that occupies a mid-trophic position in oceanic ecosystems. Acting both as a predator and as prey for apex predators, it plays a key role in the energy transfer across trophic levels in the open ocean. Changes in its trophic ecology may disrupt the efficiency of this energy transfer, with potential cascading affects higher trophic levels. In this study, we employed a combination of stomach content analysis (SCA), stable isotope analysis (SIA), and total mercury (THg) concentrations to examine the feeding ecology and dietary patterns of individual longnose lancetfish in the tropical western Pacific. Findings from the SCA revealed that longnose lancetfish consume a diverse array of prey, including fish, crustaceans, and other organisms, with notable ontogenetic dietary shifts. Smaller individuals demonstrated a preference for crustaceans, whereas larger individuals favored fish. SIA corroborated these observations, indicating dietary variation associated with growth. Positive correlations were identified between body size and stable isotope values in muscle tissue, suggesting dietary shifts and/or migratory behaviors between the epi/upper mesopelagic layers. Total mercury (THg) analysis revealed clear size-dependent bioaccumulation in longnose lancetfish, with THg levels in muscle and liver strongly correlated with fork length. In muscle, the $\delta^{15}N$-THg relationship indicated that trophic position drove mercury accumulation, while contrasting patterns in the liver reflected its role in detoxification and short-term diet integration. The greater niche width and overlap rates in liver tissue compared to muscle tissue in both small and large individuals suggested that short-term feeding habits are more diverse, with increased overlap and competition for resources within the same environment. These findings enhanced our understanding of the feeding strategies and ecological roles of the longnose lancetfish in mesopelagic ecosystems, with potential implications for comprehending long-term changes in these under-researched populations, particularly concerning shifts in dietary patterns and predator–prey dynamics.

Keywords *Alepisaurus ferox* · Feeding habits · Trophic position · Elemental tracers · Bioaccumulation · Niche width

Responsible Editor: C. Harrod.

✉ Yunkai Li
ykli@shou.edu.cn

[1] College of Marine Living Resource Sciences and Management, Shanghai Ocean University, Shanghai 201306, China

[2] Specialized Research Station On Marine Ecosystems, Institute of Agricultural Research for Development (IRAD), 219 Kribi, Cameroon

[3] The Key Laboratory of Sustainable Exploitation of Oceanic Fisheries Resources, Ministry of Education, Shanghai 201306, China

[4] National Engineering Research Centre for Oceanic Fisheries, Shanghai Ocean University, Shanghai 201306, China

Published online: 12 September 2025

Introduction

The intensification of global fishing activities has resulted in marked reductions in apex pelagic predators in the open ocean, such as sharks, tunas, and billfishes, thereby undermining their role in regulating mesopelagic ecosystems through top-down control (Casini et al. 2009; Worm and Tittensor 2011; Worm et al. 2013; Pacoureau et al. 2021). These apex predators facilitate energy transfer across various oceanic strata via diel vertical migrations. For instance, species like yellowfin tuna (*Thunnus albacares*) and swordfish (*Xiphias gladius*) prey on mesopelagic fish, while blue sharks (*Prionace glauca*) and pelagic threshers (*Alopias pelagicus*) target cephalopods at depths ranging from 200 to 1000 m (Polo-Silva et al. 2013; Weng et al. 2015; Trujillo-Olvera et al. 2018; Fujinami et al. 2018). The decline of these predators triggered trophic cascades, where diminished predation leads to increased populations of mesopelagic species, which in turn suppress the abundance of zooplankton and smaller fish (Spiers et al. 2016; Grubbs et al. 2016; Rupp and Bornatowski 2021). Furthermore, the overfishing of high-level predators such as tuna, may lead to an increase in the population of lanternfish, which subsequently suppress zooplankton (Ward and Myers 2005; Heithaus et al. 2012; Ortuño Crespo and Dunn 2017). Such ecological disturbances destabilize prey assemblages and hinder carbon export processes that are essential for oceanic biogeochemical cycles.

Mesopelagic fishes, recognized as one of the most abundant vertebrates on the planet, are integral to the transfer of energy within open-ocean food webs, linking primary consumers to apex predators (Davison et al. 2013; Olivar et al. 2019; Clarke et al. 2020; Woods et al. 2023). However, significant knowledge gaps persist regarding trophodynamics and interspecific interactions within mesopelagic assemblages (Iglesias et al. 2023). Accurately quantifying these interactions is essential for refining ecosystem models and informing fisheries management strategies aimed at mitigating cascading effects on marine ecosystems (Young et al. 2015; Woods et al. 2022).

The longnose lancetfish (*Alepisaurus ferox*), a mid-trophic deep-sea predator with a global distribution, excluding polar regions, is often captured as bycatch in pelagic longline fishing operations targeting swordfish and tuna (Carruthers et al. 2009; Pan et al. 2024). Similar to other members of the Alepisauridae, it possesses a large stomach and low digestive capacity, which allows prey items to remain intact for extended durations, facilitating high taxonomic resolution of stomach contents (Wassersug and Johnson 1976). In the Pacific and Indian Oceans, the longnose lancetfish exhibits generalist feeding behaviors, primarily consuming fish and crustaceans, with dietary preferences shifting to include fish and cephalopods as individuals mature (Potier et al. 2007b; Portner et al. 2017, 2023; Liu et al. 2019). Its substantial stomach capacity and wide distribution enable it to accumulate prey over various spatial and temporal scales, effectively sampling ecosystems that are challenging to survey using conventional methods, such as trawling (Varghese et al. 2010). These characteristic positions the longnose lancetfish as an effective biological sampler for understudied pelagic communities (Portner et al. 2023). Additionally, it functions as both a predator of mesopelagic micronekton and a prey species for apex predators, thereby serving as a crucial energy conduit between mesopelagic and epipelagic ecosystems (Potier et al. 2007a; Young et al. 2010). Alterations in its trophic ecology could disrupt the efficiency of energy transfer, consequently affecting higher trophic levels.

A further understanding of predator–prey dynamics within the mesopelagic zone necessitates methodologies that effectively capture both short-term and long-term dietary patterns. Stomach content analysis (SCA) offers direct insights into the dietary habits of species at the taxonomic level, capturing the most recent feeding event (Hyslop 1980). In contrast, biochemical tracers, including stable isotopes and mercury, provide a more comprehensive understanding of long-term dietary patterns and help to reduce biases associated with the rapid digestion of certain prey items (Brodeur et al. 2021; Li et al. 2022). The stable isotope ratios of carbon and nitrogen ($\delta^{13}C$ and $\delta^{15}N$, respectively) serve as trophic biomarkers over timeframes ranging from weeks to years, contingent upon the specific tissue analyzed, thereby elucidating long-term dietary trends across various species (Post 2002; Newsome et al. 2007). For example, the turnover rates of liver tissue reflect dietary intake over periods of weeks to months, whereas muscle tissue can integrate dietary signals over the course of a year in long-lived species (Vander Zanden et al. 2015). It is reported that the $\delta^{13}C$ values in marine fish show moderate variability ($2.1 \pm 1.4‰$) from prey to consumer, indicating the source of primary productivity, while $\delta^{15}N$ values exhibit predictable trophic enrichment ($3.1 \pm 1.6‰$), allowing for a direct inference of a species' trophic position (Dalerum and Angerbjorn 2005, Ishikawa et al. 2013, Canseco et al. 2022). When combined, these two isotope ratios ($\delta^{15}N$ and $\delta^{13}C$) facilitate the quantification of niche breadth, thereby reflecting the diversity of resources utilized or the range of environments adapted by the species in question (Post 2002). The SIA method is limited in its ability to identify specific prey taxa, which necessitates the use of complementary SCA to correlate isotopic signatures with particular prey species (Hyslop 1980; Newsome et al. 2007). Mercury (Hg), a widespread environmental contaminant, exhibits distinct vertical stratification within marine food webs, with mesopelagic fish demonstrating higher concentrations than their epipelagic counterparts (Monteiro

et al. 1996; Bowman et al.2020; Hilgendag et al. 2022; Zhang et al. 2024). This phenomenon is attributed to microbial methylation processes and trophic compression effects (Choy et al. 2009; Lamborg et al. 2014). The analysis of Hg concentration can provide valuable insights into biological effect thresholds, which are defined as the critical levels at which Hg begins to exert harmful physiological or ecological effects on organisms. Hg concentrations alone are insufficient for accurately determining trophic levels or dietary preferences due to various confounding factors, such as the bioavailability of Hg in the local environment and growth dilution effects. Changes in the local environmental conditions further influence foraging habitats and drives bioaccumulation (Bezerra et al. 2019). Additionally, growth dilution effects complicate interpretations, as rapid growth in species such as mesopelagic fishes can reduce tissue Hg concentrations regardless of dietary intake (Wang and Wang 2018; Chételat et al. 2021). Detoxification processes, including selenium-mercury antagonism and metallothionein protein binding, affect Hg retention efficiency among different taxa, obscuring true trophic relationships (Onsanit and Wang 2011; Kong et al. 2023). Furthermore, age- and size-dependent bioaccumulation results in higher Hg levels in older or larger individuals due to prolonged exposure, while variations in metabolic rates—such as greater Hg elimination in migratory species compared to sedentary ones—introduce additional uncertainty (Sardenne et al. 2017). These factors underscore the necessity for supplementary methods (e.g., stable isotopes) to elucidate Hg accumulation pathways in relation to trophic ecology.

These methodologies have been differentially applied differently across various ocean regions. In the North Atlantic, combined SCA and stable isotope analysis (SIA) have characterized the dietary habits and trophic positions of lancetfish (*Alepisaurus* spp.) (Keller et al. 2016), while in the central and eastern North Pacific, investigations into the distribution of Hg across various tissue types in the longnose lancetfish have elucidated the mechanisms of Hg accumulation within the organism (Chen et al. 2025). Furthermore, these studies have demonstrated that ontogenetic dietary shifts contributed to Hg bioaccumulation in pelagic predators that inhabited similar environments as the lancetfish, thereby positioning the lancetfish as a viable candidate for monitoring spatial and temporal variations in Hg levels within deep-sea pelagic ecosystems (Chen et al. 2025). In contrast, research in the tropical western Pacific has been predominantly restricted to SCA (Liu et al. 2019), which offers only transient insights into recent feeding behaviors and fails to elucidate long-term dietary trends, nutrient assimilation rates, or baseline energy sources (Young et al. 2015). Combining biochemical tracers with conventional stomach content analysis may help overcome these challenges by measuring the diversity of trophic niches and clarifying nutritional reliance on specific prey types. This approach is essential for addressing knowledge gaps in the trophic ecology of lancetfish, particularly in underresearched regions such as the tropical western Pacific, where detailed data are necessary to predict the impacts of fisheries and climate change on pelagic ecosystems.

This study presents an in-depth assessment of the trophic ecology of the longnose lancetfish through the integration of SIA, SCA, and total mercury (THg) concentration measurements. By examining ontogenetic dietary shifts and foraging preferences, we seek to reconcile the differences between short-term prey consumption and the energy sources that are assimilated. These results yield high-resolution data that are essential for forecasting the cascading effects of anthropogenic influences on the dynamics of open ocean food webs.

Materials and methods

Sample collection

A total of 105 longnose lancetfish were collected as bycatch from tuna longline fishing operations in the tropical western Pacific Ocean (11°N–17°N and 130°E–139°E) during the months of August and September from 2021 to 2023 (Fig. 1). The fork length (FL) of each specimen was measured to the nearest centimeter, and the total weight was recorded to the nearest gram in a laboratory setting. In accordance with the methodology established by Jantz et al. (2013), a superficial incision was made from the anterior region of the abdomen to the central pelvic girdle at the anus to access the peritoneal cavity. Additional incisions were performed in the stomach and pyloric sphincter to facilitate the extraction of the stomach, resulting in the successful retrieval of 98 stomachs for analysis. Approximately 2 g of tissue samples were collected from the dorsal white muscle and liver of each fish for long-term and short-term dietary analysis, respectively. All samples were placed in sterile tubes and subsequently stored at −80°C for future analysis.

Stomach content analysis

SCA was performed in accordance with the methodology outlined by Jackson et al. (2000). Initially, the stomach contents were extracted, and the gastric lining was thoroughly rinsed with fresh water to eliminate any residual material. Subsequently, the contents were spread on paper towels to remove excess debris and moisture. Each prey sample was documented through photography, and its weight and total length were recorded with precision to the nearest 0.1 mg and 0.1 mm, respectively. Additionally, muscle tissue from

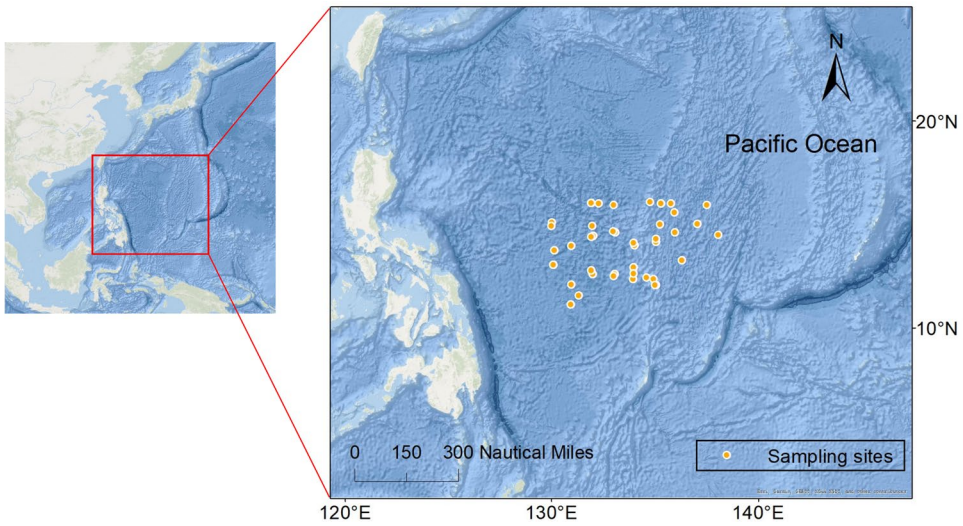

Fig. 1 Sampling sites in the tropical western Pacific in August and September from 2021 to 2023

Table 1 PCR primer information for stomach contents from longnose lancetfish

Primer	Sequence (5'—3')	References
FishF1	TCAACCAACCACAAAGACATTGG CAC	Ward et al. (2005)
FishR1	TAGACTTCTGGGTGGCCAAAGAA TCA	
LCO1490	GGTCAACAAATCATAAAGATATTGG	Ivanova et al. (2007)
HCO2198	TAAACTTCAGGGTGACCAAAAAA TCA	

the prey was sampled. Total length for fishes, mantle length for squids, distance from eye to end of abdomen for crustaceans, and total length for gastropods and polychaetes were obtained for all prey samples to ensure uniformity across taxa, thereby facilitating the generation of comparable data for stable isotope mixing models (SIMMs) and reducing the potential for bias. The longline operations utilized Argentine shortfin squid (*Illex argentinus*) and Chub mackerel (*Scomber japonicus*) as bait. Notably, these bait species were not native to the study area, thereby eliminating the potential for bias in the SCA due to bait consumption.

Species were identified to the lowest possible taxonomic level by collecting reference data based on the morphological characteristics and other identifiable features of the forage organisms. For organisms that could not be identified through observation, small tissue samples were collected for DNA extraction using the Marine Animal Tissue Genomic DNA Extraction Kit provided by Tiangen. The mitochondrial gene cytochrome oxidase subunit I (COI) was selected as the molecular marker for polymerase chain reaction (PCR) amplification, utilizing two sets of universal primers (Table 1). The PCR reaction was performed in a 10 μL mixture, which included 1 μL of DNA template, 3 μL of double-distilled water (ddH_2O), 5 μL of Premix Ex Taq enzyme (TaKaRa), and 0.5 μM of each forward and reverse primer.

The PCR reaction conditions were as follows: an initial denaturation at 94°C for 3 min, followed by 34 cycles consisting of denaturation at 94°C for 30 s, annealing at 55°C for 30 s, and extension at 72°C for 1 min. A final extension was performed at 72°C for 10 min, after which the reaction was maintained at 4°C. The PCR amplification products were sent to Biotech Sequencing in Shanghai, China, for DNA sequencing. Subsequently, the sequencing results were aligned and compared with nucleic acid sequences available in the NCBI (National Center for Biotechnology Information) database for species identification.

Stable isotope analysis and THg analysis

Samples of muscle (n = 105), liver (n = 85), and prey muscle (n = 260) from longnose lancetfish were prepared for SIA and THg analysis. To evaluate temporal variations in trophic interactions, we conducted an analysis of stable isotopes in both muscle and liver tissues of the lancetfish. Muscle tissue, characterized by slower isotopic turnover rates (half-life: weeks to months), integrates dietary signals over extended temporal scales, thereby reflecting assimilated energy sources and habitat utilization (Tieszen et al. 1983; Hesslein et al. 1993). Conversely, liver tissue demonstrates a more rapid isotopic turnover (half-life: days to weeks), which offers insights into recent foraging behavior and short-term dietary changes (Logan and Lutcavage 2010; Boecklen et al. 2011). All tissue samples were thoroughly cleaned with Milli-Q water to eliminate surface contaminants, subsequently placed in 2 ml centrifuge tubes, and freeze-dried at −55°C for a duration of 48 h.

The C:N ratios for longnose lancetfish muscle tissue were previously reported to be < 3.5 (Gao et al. 2024). Therefore, samples were directly used for isotopic analysis without lipid extraction (Post et al. 2007). In contrast,

we performed chemical lipid extractions on both the liver of longnose lancetfish and muscle tissues of their prey. Lipids were extracted by adding 12 ml of a dichloromethane–methanol solution (2:1, v/v) to a 15 ml centrifuge tube containing 1.5 mg of sample. The mixture was thoroughly agitated and allowed to rest for 24 h prior to centrifugation. Following the extraction of the supernatant, the sample was subsequently dried in an oven at 40°C.

The dried samples were ground into a fine powder, and approximately 1.5 mg was transferred into a tin capsule. The $\delta^{13}C$ and $\delta^{15}N$ values were determined using a stable isotope ratio mass spectrometer (IsoPrime 100, UK) in conjunction with an elemental analyzer (Vario ISOTOPE Cube, Germany). The $\delta^{13}C$ and $\delta^{15}N$ values were calculated using the following equations (Brand 2011):

$$\delta(X) = (R_{sample}/R_{standard} - 1) \quad (1)$$

where X represents either ^{13}C or ^{15}N; R_{sa} and R_{st} denote the isotopic ratios of the sample and the standard sample, respectively. The international standard for $\delta^{13}C$ is vPDB, produced by the International Atomic Energy Center in Vienna, while the international standard for $\delta^{15}N$ is atmospheric nitrogen. To ensure the reliability and accuracy of the test results, three laboratory standards (protein: $\delta^{13}C = -26.98‰$, $\delta^{15}N = 5.96‰$) were included in each set of 15 samples. The $\delta^{13}C$ and $\delta^{15}N$ values were calibrated using the international standard samples USGS 24 (−16.5‰ vPDB) and USGS 26 (53.7‰ vN2), achieving analytical accuracies of 0.15‰ for $\delta^{13}C$ and 0.20‰ for $\delta^{15}N$.

THg concentrations were quantified utilizing thermal decomposition (combustion), amalgamation, and atomic absorption spectroscopy, employing a calibrated Direct Mercury Analyzer (DMA-80, Milestone, Italy). Approximately 0.1 g of powdered samples were introduced into the DMA-80, where they were subjected to drying and subsequently combusted at a temperature of 650°C in an oxygen-rich environment. The analytical procedure for the tissue samples involved a drying duration of 100 s, a decomposition period of 150 s, and a waiting interval of 10 s. Quality control measures encompassed the continuous analysis of laboratory method blanks (one blank per sample), duplicate analyses of tissue samples (10% of the total samples), and the evaluation of certified reference material (DORM-4) (O'Bryhim et al. 2017; Li et al. 2022). Additionally, three laboratory standards were incorporated for every 20 samples to ensure that recovery data remained within acceptable thresholds. The precision of the duplicate samples yielded an average of ± 6.56%, while the recovery rates for the certified reference material varied between 95 and 108%.

Data analysis

To delineate distinct growth stages for investigating dietary shifts, the allometric relationship between weight (W) and length (L) was initially characterized using a linear model: logW = loga + blogL (where a and b are model parameters), fitted via ordinary least squares regression. Subsequently, potential inflection points in the growth trajectory were identified using a segmented linear regression model, implemented with the "segmented" package in R (Segura et al. 2013). This approach partitions the dataset, fitting distinct linear models to each segment, thereby better accommodating non-linear variations in the overall growth pattern. The model can be expressed in the following general form:

$$log\ W = \begin{cases} log\ a_1 + b_1\ log\ L & \text{if L } \leq \text{turning point} \\ log\ a_2 + b_2\ log\ L & \text{if L } > \text{turning point} \end{cases} \quad (2)$$

where a_1, b_1, a_2, b_2 are the parameters of the model, the turning point signifies the transition from one growth phase to another.

This turning point is identified using a function from the "segmented" package, indicating a change in the growth phase-where mass increases until the onset of maturation, after which it may begin to decline. Based on this turning point, samples are categorized into two fork length groups: those before the turning point (small group) and those after (large group).

Prey species were systematically classified into three distinct categories based on their taxonomic classification (fish, cephalopod, crustacean, gastropoda, and polychaetes), depth layer (epipelagic, epi/upper mesopelagic, mesopelagic, and bathypelagic), and size [0–2 cm (micro prey), 2–5 cm (small prey), and > 5 cm (large prey)]. To determine the depth range of these prey species, we conducted a comprehensive review of peer-reviewed literature, trawl records, and taxonomic guides, in addition to utilizing the most reliable distribution data (refer to Appendix 1, Table S1). Depth information was accessible for all species analyzed, with 25 species corroborated by site-specific trawl records and published studies on vertical distribution. The remaining species were sourced from global marine organism databases, such as FishBase (www.fishbase.org) and SeaLifeBase (www.sealifebase.org).

The SCA calculations incorporated the percentage of number (N%), percentage of mass (W%), and frequency of occurrence (F%), alongside the index of relative importance (IRI) and the percent relative importance index (IRI%) for each prey item. Each metric addresses specific biases inherent in dietary interpretation: N% may disproportionately highlight small-bodied prey that are numerically abundant, while W% may skew the representation towards larger,

albeit less frequently consumed, prey. Furthermore, F% alone does not account for the quantity of prey present in each stomach (Baker et al. 2014). Though the N%, W%, and F% directly describe diet composition, to compare with the SIMMs results, IRI% was used to provide a more balanced evaluation (Hyslop 1980; Cortés 1997). The calculations were conducted using Eqs. (2) and (3) as outlined by Fonteles-Filho (2011).

$$\mathrm{IRI} = (\mathrm{N\%} + \mathrm{W\%}) \times \mathrm{F\%} \tag{3}$$

$$\mathrm{IRI\%} = \left[\mathrm{IRI} / \sum \mathrm{IRI} \right] \times 100\% \tag{4}$$

N% signifies the ratio of a particular prey item to the overall count of all prey items, while W% reflects the ratio of the total mass of a specific prey item to the cumulative mass of all prey items. F% represents the frequency of occurrence of a specific prey item within non-empty stomachs. The IRI serves as a holistic measure for evaluating the significance of a specific prey item. The IRI% is expressed as a percentage of the total IRI for all prey items.

The niche breadth derived from stomach contents was assessed utilizing the Shannon–Wiener index (H'), the Pielou's evenness index (J'), and the Simpson dominance index (D). A higher H' value indicates a greater feeding width in longnose lancetfish, while J' normalizes the H' value to a scale of 0 to 1, reflecting the diversity of forage organisms. The calculation formula is as follows (Krebs 1989):

$$H' = -(\sum P_i \times \ln P_i) \tag{5}$$

$$J' = H'/H'_{\max} = H'/\ln S \tag{6}$$

$$\mathrm{D} = \sum P_i^2 \tag{7}$$

where S represents the number of prey species, and P_i denotes the proportion of prey species i.

To assess differences in diet composition between groups (small and large longnose lancetfish specimens), a one-way Analysis of Similarities (ANOSIM) was performed based on a Bray–Curtis dissimilarity matrix, calculated from the numerical abundance of prey (species taxonomic level), and significance was tested using with 9999 permutations. If the results were found to be significant ($P<0.05$), post hoc analyses (pairwise comparisons) were applied to identify specific differences between groups (small and large specimens). Dietary patterns and group separation were then graphically visualized using non-metric multidimensional scaling (NMDS). To quantify niche space, standard ellipsoid volumes (SEV) were calculated for each group using both stable isotope and THg data and corrected for small sample sizes using the Stable Isotope Bayesian Ellipses in R (SIBER) package (Jackson et al. 2011; Skinner et al. 2019). The SEV serves as an indicator of the core isotopic niche breadth of a population within a three-dimensional framework. An increased SEV signifies greater variability among individuals across these dimensions, which may indicate a broader range of resource utilization, such as feeding across diverse habitats, trophic levels, or prey exhibiting varying levels of Hg bioaccumulation. Furthermore, overlapping SEVs between groups may imply niche competition or partitioning (Skinner et al. 2019).

Given that the $\delta^{13}C$, $\delta^{15}N$ values, and THg concentration data for the longnose lancetfish did not conform to the assumptions of normality, Spearman's rank correlation analysis was performed to assess the relationships between $\delta^{13}C$ and $\delta^{15}N$ values in muscle and liver tissues, fork length, and THg concentrations. The non-parametric Mann–Whitney U test was utilized to compare the differences in $\delta^{13}C$, $\delta^{15}N$ values, and THg concentrations between (1) muscle and liver tissues of longnose lancetfish and (2) large versus small individuals. Additionally, the Kruskal–Wallis test was employed to investigate potential differences in $\delta^{13}C$, $\delta^{15}N$ values, and THg concentrations across five taxonomic groups, as well as among four depth layer groups and three size groups ($\alpha=0.05$).

To estimate the proportion of each dietary component, we employed SIMMs implemented in the MixSIAR package in R (Stock et al. 2018). We performed Kruskal–Wallis tests to evaluate the differences in isotope values among various prey groups. The $\delta^{13}C$ and $\delta^{15}N$ values exhibited significant differences across the four depth layer groups ($\delta^{13}C$: $\chi^2(3)=21.7$, $P<0.01$; $\delta^{15}N$: $\chi^2(3)=29.0$, $P<0.01$) and among the three prey size categories ($\delta^{13}C$: $\chi^2(2)=16.0$, $P<0.01$; $\delta^{15}N$: $\chi^2(2)=14.9$, $P<0.01$). However, when analyzed by taxonomic group, the $\delta^{13}C$ and $\delta^{15}N$ values did not reveal significant differences between fish and cephalopods ($P>0.05$). Consequently, these two taxa were combined for analytical purposes. Furthermore, the interpretation of stable isotope data necessitates the use of a trophic discrimination factor (TDF), given that the isotopic discrepancies between prey and consumer can differ among species. While direct observations can provide these values, they often necessitate prolonged periods of confinement for live specimens. In instances where such analyses are not feasible, TDF values for consumers within the same feeding guild (e.g., carnivores, herbivores) may be sourced from existing literature (Vanderklift and Ponsard 2003; Logan et al. 2008; Caut et al. 2009; Parnell et al. 2013). Previous studies indicated that SIMMs outputs may be sensitive to the selected TDF values due to uncertainty surrounding TDFs (Bond and

Table 2 Dietary composition for longnose lancetfish by percent number (N%), percent mass (W%), percent frequency of occurrence (F%), and percent relative importance index (IRI%)

Prey item	N%	W%	F%	IRI%
Fish	**22%**	**80%**	**47%**	**52%**
Lepidocybium flavobrunneum	4%	8%	7%	4%
Sternoptyx pseudobscura	4%	5%	16%	6%
Omosudis lowii	2%	3%	9%	2%
Nesiarchus nasutus	2%	7%	4%	1%
Encrasicholina punctifer	2%	1%	4%	<1%
Sternoptyx diaphana	2%	1%	5%	1%
Diodon hystrix	1%	24%	4%	6%
Brama japonica	1%	3%	4%	1%
Melichthys vidua	1%	8%	4%	1%
Scopelarchus michaelsarsi	<1%	<1%	2%	<1%
Magnisudis atlantica	<1%	1%	2%	<1%
Paralepis brevirostris	<1%	1%	2%	<1%
Lestidium orientale	<1%	<1%	2%	<1%
Gempylus serpens	<1%	3%	2%	<1%
Dysalotus alcocki	<1%	1%	2%	<1%
Heteropriacanthus cruentatus	<1%	5%	2%	<1%
Ariosoma meeki	<1%	<1%	2%	<1%
Naso vlamingii	<1%	<1%	2%	<1%
Xenolepidichthys dalgleishi	<1%	1%	2%	<1%
Alepisaurus ferox	<1%	<1%	2%	<1%
Cephalopod	**6%**	**12%**	**19%**	**4%**
Walvisteuthis jeremiahi	2%	4%	5%	2%
Enoploteuthis reticulata	1%	4%	2%	<1%
Japetella diaphana	1%	2%	4%	<1%
Onychoteuthis compacta	1%	<1%	4%	<1%
Argonauta argo	<1%	1%	2%	<1%
Bolitaena pygmaea	<1%	<1%	2%	<1%
Octopoteuthis megaptera	<1%	<1%	2%	<1%
Crustacean	**51%**	**7%**	**63%**	**40%**
Parascelidae	17%	2%	40%	35%
Parhyale hawaiensis	8%	1%	16%	7%
Phrosina semilunata	6%	<1%	19%	6%
Platyscelus armatus	5%	1%	16%	4%
Stenothoidae	5%	<1%	7%	2%
Galatheocaris abyssalis	3%	2%	7%	2%
Phronima sedentaria	2%	<1%	5%	<1%
Euphausia pacifica	1%	<1%	2%	<1%
Platyscelus ovoides	1%	<1%	2%	<1%
Euryozius camachoi	1%	<1%	2%	<1%
Lanceola sayana	<1%	<1%	2%	<1%
Unidentified shrimp	2%	<1%	5%	5%
Gastropoda	**4%**	**<1%**	**14%**	**1%**
Cavolinia globulosa	2%	<1%	9%	1%
Cavolinia gibbosa	2%	<1%	7%	1%
Unidentified gastropoda	<1%	<1%	2%	<1%
Polychaetes	**16%**	**<1%**	**21%**	**4%**
Other Unidentified organisms	**2%**	**<1%**	**9%**	**<1%**

Diamond 2011). To address this issue, sensitivity analyses were conducted on the results obtained. Two SIMMs were run by species and by tissue type using different TDF values from prior research: general values from Post (2002) (TDFs: 0.4±1.3 ‰ for $\delta^{13}C$ and 3.4±1.0 ‰ for $\delta^{15}N$ in both muscle and liver) and specific values from Sweeting et al. (2007a, b) (TDFs: 1.7±1.1 ‰ for $\delta^{13}C$ and 3.2±1.3 ‰ for $\delta^{15}N$ in muscle; 0.9±1.3 ‰ for $\delta^{13}C$ and 2.3±0.9 ‰ for $\delta^{15}N$ in liver). An average value of the estimated contribution of each group was calculated. As recommended by Vander Zanden and Rasmusen (1999), we tested models with both sets of TDF values for goodness of fit, based on Gelman-Rubin test values below 1.1 (Gelman et al. 1995). For model inputs, the Markov Chain Monte Carlo (MCMC) method was set to test length, the error structure was defined as the 'resid' process, and we employed both Gelman-Rubin and Geweke diagnostics to assess model convergence. All SIMMs were run for 10^5 iterations.

Results

Stomach content analysis

In this study, the fork length of longnose lancetfish ranged from 55 to 170 cm, with a mean length of 96±23 cm. The individual weights of the longnose lancetfish varied between 400 and 11,000 g, resulting in an average mass of 2999±2506 g. Out of the 98 stomachs examined from longnose lancetfish, 45 (46%) were classified as either empty or containing items that were not considered prey, and 53 of the stomachs (54%) contained prey. A total of 260 prey items were identified; 83 of these were recognized based on morphological characteristics, while the remaining samples were analyzed using DNA barcoding (Table 2).

The identified organisms were categorized into five primary taxonomic groups: fish, cephalopods, crustaceans, gastropods, and polychaetes. A total of 45 species from 34 families were identified, excluding unidentified organisms, based on SCA and DNA barcoding results. These taxonomic classifications were confirmed through both morphological (visual) and molecular (DNA barcoding) methods. Within these groups, 20 fish species were identified. Additionally, there were 14 species of crustaceans, 7 species of cephalopods, 3 species of gastropods, and 1 species of polychaete. According to the index analysis, crustaceans, fish, and polychaetes were quantitatively dominant (N%) in the stomach contents of longnose lancetfish, representing 51%, 22%, and 16%, respectively. By mass (W%), fish are highest (80%), followed by cephalopods (12%) and crustaceans (7%) (Table 2). In terms of the IRI%, fish and crustaceans were the most significant prey items for longnose

lancetfish, collectively accounting for over 90% of the total IRI%. Among fish, the primary prey was identified as *S. pseudobscura* (IRI%=6%), while for crustaceans, it was Parascelidae (IRI%=35%). When prey was categorized by their respective water layers and size, epi/upper mesopelagic taxa constituted 60% of the stomach contents IRI%, whereas large prey accounted for more than 50% of the IRI% (Fig. 2). The results of the stomach contents diversity analysis indicated a Shannon–Wiener index (H') of 3.1, a Pielou's evenness index (J') of 0.8, and a Simpson dominance index (D) of 0.08.

The analysis of segmented relationships between fork length and weight in lancetfish identified a critical threshold at a fork length of 98 cm, which serves to differentiate between smaller (FL<98 cm) and larger (FL≥98 cm) groups. The stomach contents of smaller individuals (n=37) comprised 188 prey items, which included 16 fish, 6 cephalopods, 3 gastropods, 12 crustaceans, and 1 polychaete species. Conversely, larger individuals (n=16) contained 72 prey items, consisting of 9 fish, 4 cephalopods, 1 gastropod, 8 crustaceans, and 1 polychaete species. For the smaller individuals, crustaceans were the predominant forage group, representing 52% of the IRI%, and exhibited H' of 3.0, J' of 0.8, and D of 0.08. In contrast, fish constituted the primary dietary component for larger individuals, with an IRI% of 69% and diversity indices of H'=2.7, J'=0.9, and D=0.1 (Table 3). Both size categories of longnose lancetfish predominantly foraged on prey from the epi/upper mesopelagic zones, with small individuals exhibiting an IRI% of 47% and large individuals at 67% for this specific forage group. Notably, both size classes primarily consumed large prey items, although larger individuals exhibited a greater tendency to target higher-bodied prey compared to smaller conspecifics. The NMDS (Fig. 3) resulting from ANOSIM and corrected ellipse areas for small sample sizes (hereafter 'niche space') revealed different sized niches among groups, indicating a difference between the feeding habits of small and large lancetfish and the largest prey niche space was observed in large individuals (F=0.128, P=0.031). This observation suggests a higher degree of dispersion among larger individuals in multivariate dietary space, indicating

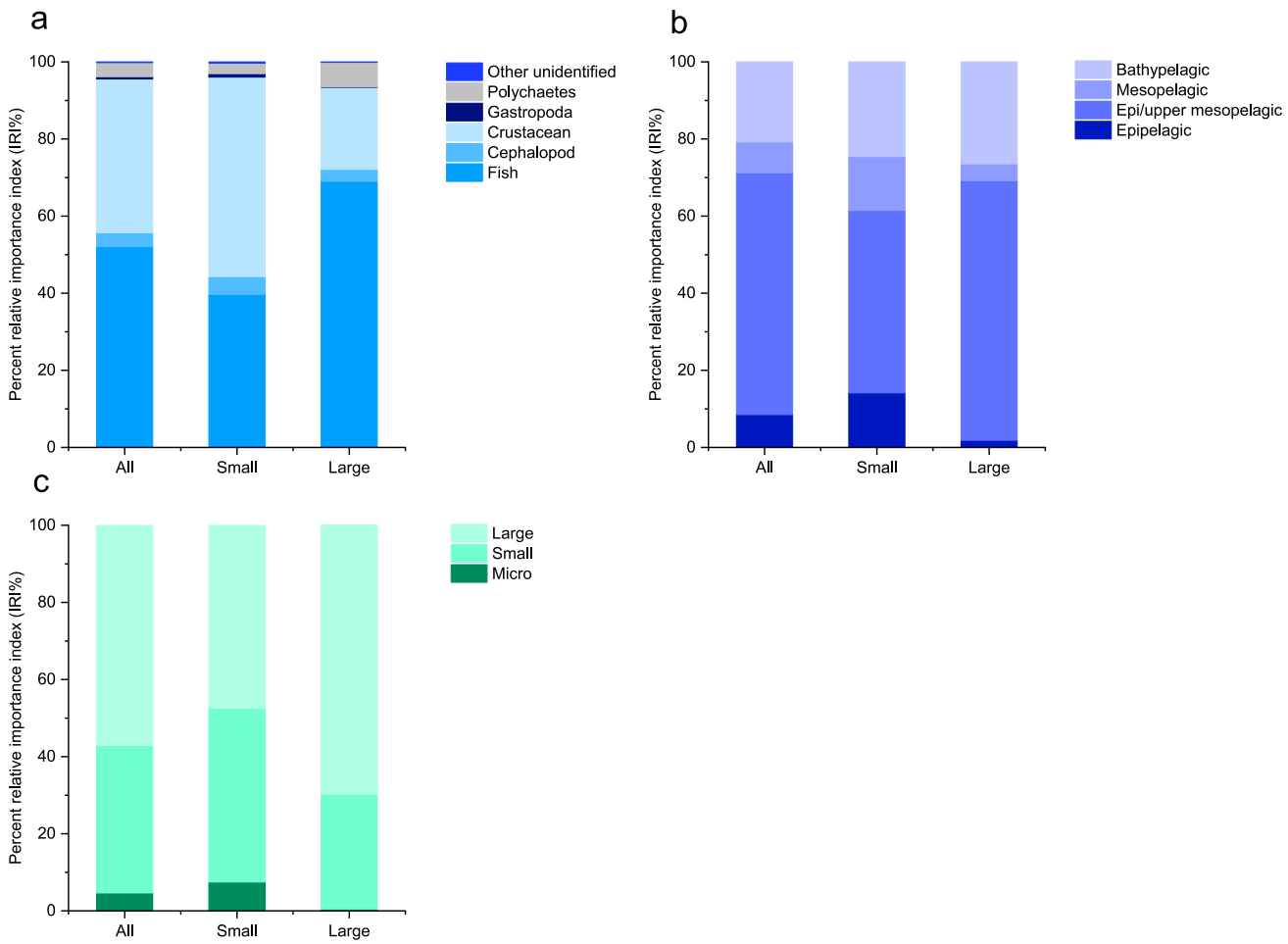

Fig. 2 Classification of stomach contents (IRI%) of longnose lancetfish according to the prey. Note: **a** taxonomic, **b** depth layer, **c** size

Table 3 Stomach contents (IRI%) of small and large longnose lancetfish from the tropical western Pacific

Taxa	IRI% (small)	IRI% (large)
Fish	**40%**	**69%**
Omosudis lowii	3%	<1%
Nesiarchus nasutus	1%	1%
Encrasicholina punctifer	1%	–
Dysalotus alcocki	<1%	–
Magnisudis atlantica	<1%	–
Naso vlamingii	<1%	–
Lepidocybium flavobrunneum	6%	2%
Paralepis brevirostris	<1%	–
Sternoptyx pseudobscura	4%	10%
Scopelarchus michaelsarsi	<1%	–
Sternoptyx diaphana	1%	–
Alepisaurus ferox	<1%	–
Brama japonica	2%	–
Xenolepidichthys dalgleishi	<1%	–
Lestidium orientale	<1%	–
Melichthys vidua	<1%	3%
Ariosoma meeki	–	<1%
Heteropriacanthus cruentatus	–	2%
Gempylus serpens	–	2%
Diodon hystrix	–	30%
Cephalopod	**5%**	**3%**
Argonauta argo	<1%	–
Enoploteuthis reticulata	1%	–
Octopoteuthis megaptera	<1%	–
Walvisteuthis jeremiahi	2%	1%
Onychoteuthis compacta	<1%	<1%
Japetella diaphana	<1%	1%
Bolitaena pygmaea	–	<1%
Crustacean	**52%**	**21%**
Stenothoidae	1%	3%
Phrosina semilunata	9%	–
Euphausia pacifica	<1%	–
Parascelidae	39%	13%
Phronima sedentaria	<1%	<1%
Platyscelus armatus	5%	2%
Platyscelus ovoides	<1%	–
Galatheocaris abyssalis	1%	2%
Unidentified shrimp	<1%	1%
Parhyale hawaiensis	7%	2%
Euryozius camachoi	<1%	–
Lanceola sayana	–	<1%
Gastropoda	**1%**	**<1%**
Cavolinia gibbosa	1%	–
Cavolinia globulosa	1%	<1%
Unidentified gastropoda	<1%	–
Polychaetes	**3%**	**6%**
Other Unidentified organisms	**<1%**	**<1%**

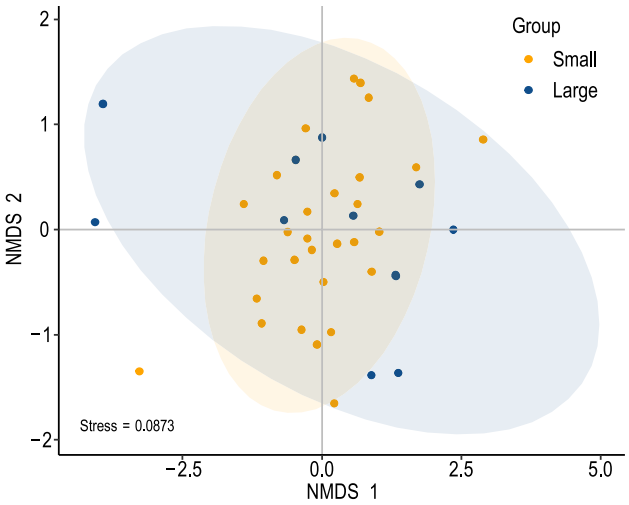

Fig. 3 Non-metric multidimensional scaling (NMDS) from the diet of small (FL<98 cm) and large (FL≥98 cm) longnose lancetfish from the tropical western Pacific. NMDS was performed by the taxonomic composition (species level) and abundance (N) of stomach contents

Table 4 Values (Mean±SD) and ranges of $\delta^{13}C$, $\delta^{15}N$ values (‰) and THg concentrations ($\mu g \cdot g^{-1}$, dry-weight) in muscle and liver of small and large longnose lancetfish

Tissues	n	$\delta^{13}C$ (‰)	$\delta^{15}N$ (‰)	THg ($\mu g \cdot g^{-1}$)
Muscle	105	−17.8±0.4	10.6±1.0	0.129±0.085
		−18.7 to −16.5	7.2 to 13.5	0.009 to 0.562
Liver	85	−17.7±0.4	10.2±1.3	0.220±0.146
		−19.0 to −16.1	5.4 to 13.4	0.063 to 0.705
Small, muscle	68	−17.9±0.4	10.4±1.0	0.109±0.075
		−18.7 to −16.5	7.2 to 12.3	0.009 to 0.562
Large, muscle	37	−17.6±0.4	11.0±1.0	0.166±0.092
		−18.3 to −16.6	8.8 to 13.5	0.072 to 0.535
Small, liver	50	−17.8±0.4	10.1±1.2	0.191±0.112
		−18.7 to −16.8	5.4 to 10.1	0.063 to 0.341
Large, liver	35	−17.6±0.5	10.4±1.4	0.245±0.138
		−19.0 to −16.1	6.5 to 13.4	0.075 to 0.705

greater inter-individual variability in diet composition relative to smaller individuals (stress=0.09).

Stable isotope analysis and THg concentrations

In longnose lancetfish, the $\delta^{13}C$ values in muscle tissues ranged from −18.7‰ to −16.5‰, while in liver tissues, they varied from −19.0‰ to −16.1‰. The $\delta^{15}N$ values for muscle tissues ranged from 7.2‰ to 13.5‰, and for liver tissues, from 5.4‰ to 13.4‰ (Table 4). The isotopic values of the prey species exhibited a range of −20.2‰ to −16.9‰ for $\delta^{13}C$ and from 2.5‰ to 9.1‰ for $\delta^{15}N$ (Fig. 4). Spearman's rank correlation analysis indicated a significant relationship between the fork length of the lancetfish and $\delta^{13}C$ ($r=0.34$, $P<0.01$) and $\delta^{15}N$ ($r=0.37$, $P<0.01$) values in muscle tissues; however, no significant correlation was observed in liver tissues ($\delta^{13}C$: $r=0.12$, $P>0.05$; $\delta^{15}N$: $r=0.18$, $P>0.05$)

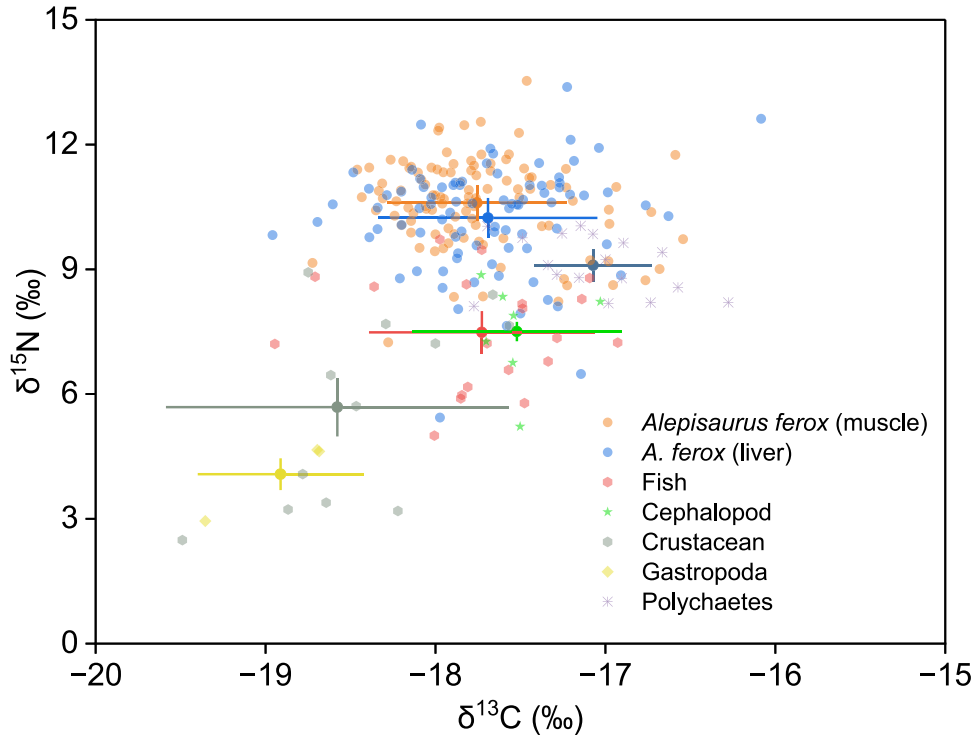

Fig. 4 Stable isotope biplot of longnose lancetfish and their prey in the tropical western Pacific

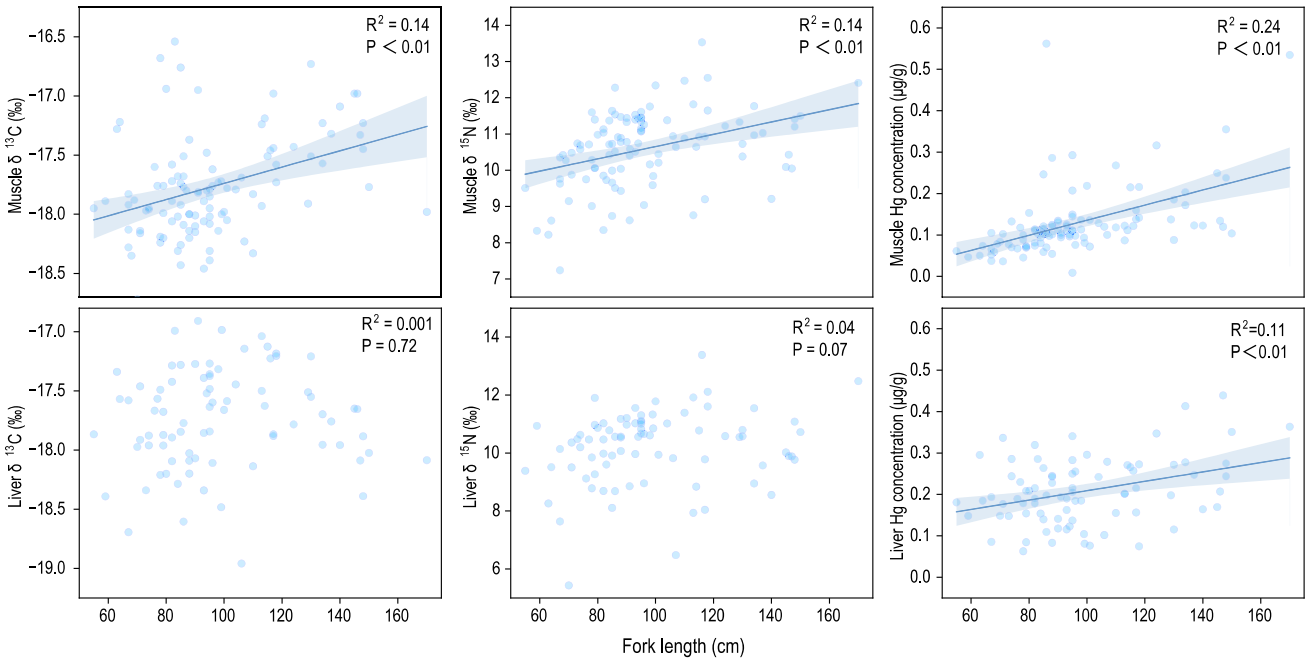

Fig. 5 Relationship between fork length, $\delta^{13}C$, $\delta^{15}N$ values, and THg concentrations in muscle and liver of longnose lancetfish from tropical western Pacific

(Fig. 5). Mann–Whitney *U* tests further demonstrated significant differences in $\delta^{13}C$ and $\delta^{15}N$ values between muscle and liver tissues ($\delta^{13}C$: $U=726$, $P<0.001$; $\delta^{15}N$: $U=698$, $P<0.001$) and between smaller and larger individuals in muscle tissues ($\delta^{13}C$: $U=686$, $P<0.001$; $\delta^{15}N$: $U=896$, $P=0.02$). Conversely, no significant differences were detected in liver tissues between small and large individuals ($\delta^{13}C$: $U=702$, $P=0.32$; $\delta^{15}N$: $U=698$, $P=0.30$). The results from the SEV indicated the isotopic niche volumes, revealing lower niche overlap in muscle tissues (small: 16.3%; large: 12.2%) and greater overlap in liver tissues (small:85.9%; large:46.1%) between small and large

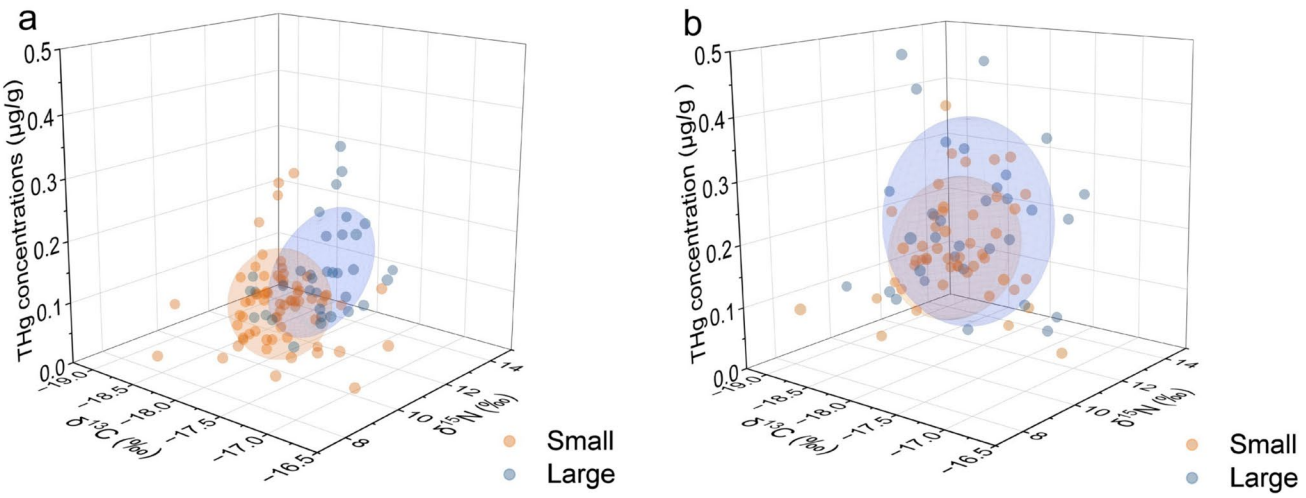

Fig. 6 Isotopic-THg niche in muscle (**a**) and liver (**b**) of small and large longnose lancetfish from the tropical western Pacific

Table 5 Isotopic-THg niche volume (standard ellipsoid volume, SEV, $‰^2 \cdot \mu g \cdot g^{-1}$) and the overlap in muscle and liver of small and large longnose lancetfish from the tropical western Pacific

	Muscle	Liver
SEV		
Small	0.196	0.457
Large	0.261	0.852
Overlap	0.032	0.393
Overlap rate		
Small	0.163	0.859
Large	0.122	0.461

Table 6 THg concentrations ($\mu g \cdot g^{-1}$, dry-weight) in different prey taxa of longnose lancetfish

Prey groups	THg ($\mu g \cdot g^{-1}$)	Prey groups	THg ($\mu g \cdot g^{-1}$)	Prey groups	THg ($\mu g \cdot g^{-1}$)
Fish	0.082^{ab}	Epipelagic	0.097^{ab}	0–2 cm (micro)	0.104^{a}
Cephalopod	0.133^{b}	Epi/upper mesopelagic	0.077^{ab}	2–5 cm (small)	0.101^{a}
Crustacean	0.076^{a}	Mesopelagic	0.047^{a}	>5 cm (large)	0.087^{a}
Gastropoda	0.144^{b}	Bathypelagic	0.141^{b}		
Polychaetes	0.219^{bc}				

Alphabetic characters infer statistical differences in individual THg concentrations between species (α=0.05). The absence of same alphabetic characters following values indicates no statistical difference

longnose lancetfish (Fig. 6). Importantly, the niche volumes for liver tissues were found to exceed those for muscle tissues in both size categories of lancetfish (Table 5).

Tables 4 and 6 provide data on the THg concentrations in longnose lancetfish and their associated prey species, respectively. The longnose lancetfish displayed higher THg concentrations in their liver compared to their muscle tissues. Spearman's rank correlation analysis revealed a significant correlation between fork length and THg concentrations in both muscle (r=0.61, $P<0.01$) and liver tissues (r=0.25, $P<0.05$) (Fig. 5). Additionally, $\delta^{15}N$ values in muscle tissues were significantly positively correlated with THg concentrations (r=0.59, $P<0.01$), whereas no significant correlation was observed in liver tissues (r=0.15, $P>0.05$). Larger individuals exhibited significantly elevated THg levels in muscle tissues compared to their smaller counterparts ($U=527$, $P<0.01$). Among the various prey types, polychaetes were found to have the highest THg concentrations.

Stable isotope mixing models

Diets derived from SIMMs that integrated both SCA and SIA exhibited some divergence from those determined exclusively through SCA. The five taxa identified with the highest W% as food sources included fishes/cephalopods, crustaceans, polychaetes, and gastropods. Analyses of muscle and liver tissue via SIMMs indicated that crustaceans were the predominant dietary component for longnose lancetfish, with mean contributions of 58.9±9.2% and 55.9±8.7%, respectively, followed by fishes/cephalopods, which contributed 20.3±9.7% and 21.5±9.5%. When prey was categorized by depth layer, the SIMMs results for both muscle and liver tissues suggested that the epi/upper mesopelagic zone served as the primary food resource for longnose lancetfish, with contributions of 34.1±14.9% and 56.5±11.5%. Furthermore, when prey was classified by size, the SIMMs results indicated that large prey constituted the main dietary sources for longnose lancetfish, with contributions of 49.3±8.9% and 41.7±11.3% for muscle and liver tissues, respectively (Fig. 7).

The SIMMs results for muscle and liver tissues revealed that crustaceans (58.5±9.5% and 51.8±9.3%) and fishes/cephalopods (18.5±8.9% and 20.6±9.5%) are significant dietary components for smaller longnose lancetfish. In

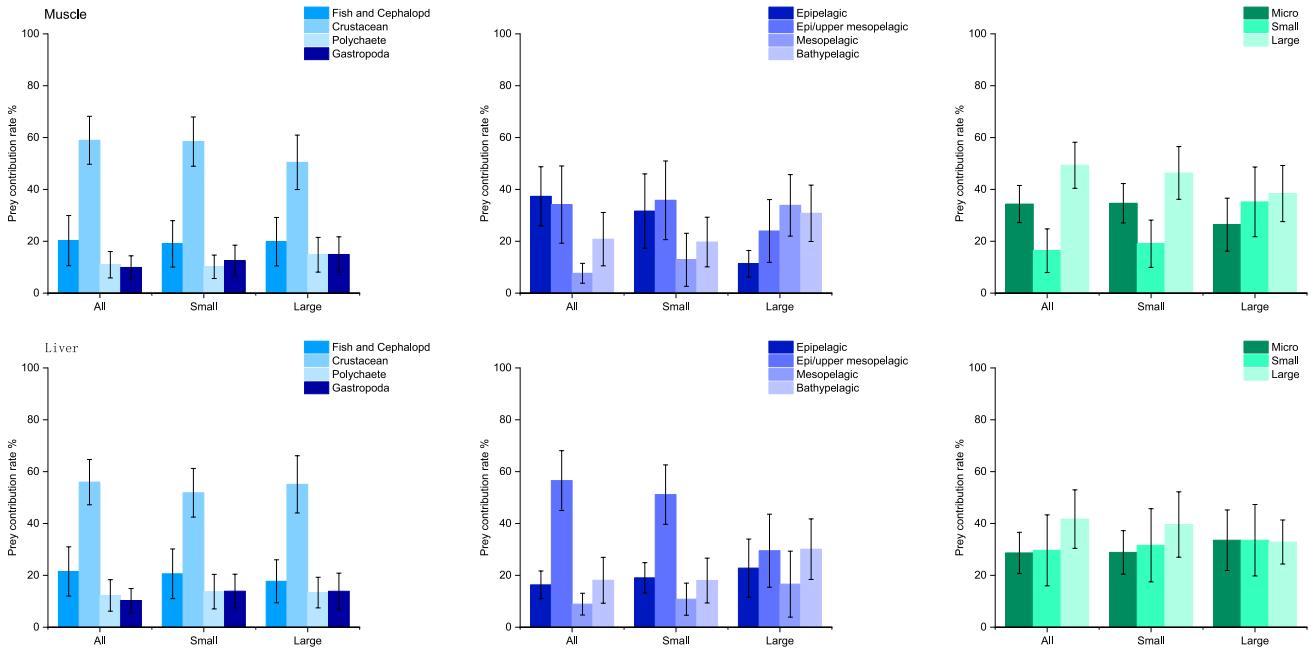

Fig. 7 Contributions of different food source groups in the muscle and liver of the longnose lancetfish based on SIMMs. Mean values and lower and upper 95% confidence intervals of contributions are showed in the figure

contrast, for larger specimens, the contributions of crustaceans were 50.4 ± 10.5% and 55.1 ± 11.1%, while those for fishes/cephalopods were 19.9 ± 9.4% and 17.7 ± 8.2%. The SIMMs results further indicated that the epi/upper mesopelagic zone is a crucial food source for small longnose lancetfish (35.8 ± 15.2% for muscle and 51.1 ± 11.5% for liver), whereas deeper prey from the mesopelagic and bathypelagic zones serve as the primary food source for larger longnose lancetfish. When examining prey size, the SIMMs results for muscle and liver tissues suggested a comparable contribution ratio of prey size to feeding between large and small individuals (Fig. 7).

Discussion

Recent developments in the field of predator foraging ecology have adopted a comprehensive approach that integrates various methodologies, including SCA, SIA, THg concentrations, and DNA barcoding. In the present study, we applied this integrated framework to enhance our understanding of the trophic ecology of the longnose lancetfish. Our results indicate that the longnose lancetfish predominantly preys on fish and crustaceans, with a notable shift in dietary preferences correlating with growth. Specifically, as individuals increase in size, there is a discernible trend towards the consumption of prey from deeper oceanic layers, suggesting a potential relationship between body size and foraging depth.

Diet composition

The dietary preferences of longnose lancetfish are intricate and shaped by a multitude of factors, including prey availability and mobility, prey abundance, ontogenetic changes, energy content of prey, prey size selection, as well as seasonal and spatial variations (Portner et al. 2017; Chen et al. 2022). The SCA conducted in this study reveals a diverse array of prey, corroborating previous research that identifies fish and crustaceans as primary dietary components (Choy et al. 2013). However, the composition of their diet varies across different habitats. For example, in the central North Pacific, longnose lancetfish primarily consume crustaceans and mesopelagic fish (Portner et al. 2017), while in the equatorial Pacific, their diet is predominantly composed of mesopelagic fish, with mesopelagic crustaceans being less prevalent (Moteki 1993). The study by Liu et al. (2019) reported that mollusks were the most abundant and nutritionally significant prey in the western tropical Pacific, which contrasts sharply with the findings of the current study. This discrepancy may be indicative of potential limitations in single-SCA methodologies, highlighting the necessity for validation through alternative analytical techniques.

Traditionally, longnose lancetfish have been characterized as opportunistic predators with a broad dietary range (Potier et al. 2007a). However, the findings of this study suggest that longnose lancetfish exhibit a more selective predation strategy. The most significant prey at both the genus and species levels, as measured by N%, W%, and F%, closely

align with those identified in other regional studies (Choy et al. 2013; Liu et al. 2019), with Sternoptychid fishes constituting a significant component of the diet. This contrasts sharply with mesopelagic micronekton communities, where Myctophidae typically dominate the biomass in midwater trawl surveys (Gjøsæter and Kawaguchi 1980; Sassa et al. 2004; Sassa 2019). If the predator feeds in a passive manner (the prey selection is based on the prey's availability in the environment), then Myctophidae should make up a significant part of its diet. However, their near absence in this study suggests a preference for active prey selection. This selective feeding behavior aligns with the findings of Choy et al. (2013) and Portner et al. (2017), further demonstrating a specialized feeding strategy for this species.

The analysis conducted using SIMMs offers a detailed examination of the dietary habits of lancetfish, highlighting the distinctions between their long-term feeding behaviors and the immediate dietary composition identified through SCA. Notably, while SCA identified large prey as significant, the influence of micro prey on predator isotopic signatures suggests a contradiction to SCA's assertion that small prey constituted a more substantial component of the diet. Although SCA provides a high level of taxonomic resolution for reconstructing trophic interactions (Santander-Neto et al. 2021), its reliance on stomach contents introduces potential biases. Specifically, bigger prey items may be disproportionately represented in the diet of lancetfish, attributable to their slower digestion rates, whereas gelatinous or smaller prey may be underestimated due to they are more easily split into smaller pieces to be digested quickly. Conversely, SIA mitigates such temporal and digestibility biases (Keller et al. 2016), yielding biomass-weighted dietary estimates (Olson et al. 2010). However, the precision of SIMMs is limited by factors such as variability in tissue-specific TDF, the phylogenetic grouping of prey, and isotopic overlap among prey types (Stephens et al. 2023). In this study, the absence of distinct stable isotope signatures between fish and cephalopod prey further constrains the resolution of SIMMs, necessitating careful interpretation.

The integration of SCA and SIMMs creates a robust framework that addresses the limitations inherent in each method, thereby providing a comprehensive understanding of lancetfish trophic ecology. While SCA captures short-term, taxon-specific prey consumption, SIMMs reflect assimilated diets over extended periods, incorporating a variety of prey types (Diaz et al. 2017; Brodeur et al. 2021). The high taxonomic resolution afforded by SCA enhances the source definitions utilized in SIMMs, thereby reducing uncertainty in prey isotopic baselines. This synergistic approach is particularly vital for lancetfish, which inhabit dynamic pelagic environments where prey availability varies with depth and time. By merging the SCA's snapshot of recent foraging with the integrated dietary signals derived from SIMMs, researchers can differentiate between opportunistic feeding events and sustained trophic strategies. The findings categorized by water layer indicate that the results obtained from the SIMMs are more generalized in comparison to those derived from the SCA. Ultimately, this dual methodology mitigates the biases associated with singular approaches: SCA compensates for SIMMs potential overestimation of isotopically similar prey, while SIMMs identify prey that may not be represented in stomach contents. As highlighted by Grey et al. (2002), it is advisable to employ SIA as a complementary tool rather than a standalone substitute for traditional dietary analysis. Furthermore, the concurrent application of stable isotope and THg analysis has become a prevalent practice for elucidating the relationship between feeding ecology and Hg exposure. Contaminant data can further enhance this approach by providing additional insights into feeding composition or habitat shifts (Kiszka et al. 2015). This integration enriches isotope data from trophic ecology studies, thereby deepening our understanding of fish feeding behaviors and trophic status.

Analysis of liver and muscle tissues revealed slight differences in diet over both short (days to weeks) and long (months or longer) periods (Perga and Gerdeaux 2005). Differences in stable isotopes among tissues may result from variations in metabolic rates, which are indicative of differing feeding periods (Fry and Arnold 1982; Herzka and Holt 2000). Liver tissues, which are characterized by elevated metabolic rates, demonstrated shorter dietary intake periods as evidenced by $\delta^{13}C$ and $\delta^{15}N$ values (Martínez del Rio et al. 2009). Furthermore, the affinity of metals for specific organs appears to vary; THg concentrations were significantly higher in the liver compared to muscle, suggesting that the liver possesses a greater capacity for Hg storage. This phenomenon may be linked to the liver's function as a primary metabolic organ responsible for processing and eliminating a greater volume of toxins (Nicula et al. 2009). This finding aligns with the short-term intake of large prey and is consistent with previous studies linking THg concentrations to fish size within marine food webs (Burger and Gochfeld 2007, 2011).

While SIA is effective in tracking assimilated prey, both SCA and SIMMs exhibit inherent limitations in detecting gelatinous prey taxa. The efficacy of SIMMs is contingent upon the sampled prey sources, which may result in an underrepresentation of cryptic taxa (Drazen and Sutton 2017). Gelatinous taxa, including salps and ctenophores, are frequently omitted from SCA and SIMMs source pools due to their rapid digestion and the lack of enduring morphological remnants (Diaz et al. 2017). The underrepresentation of fully digested prey constitutes a common limitation of both SIMMs and SCA, highlighting the challenges associated

with reconstructing predator diets. To mitigate these deficiencies, future research should implement a comprehensive framework grounded in a Bayesian prior for the SIMMs of regional gelatinous zooplankton biomass (Diaz et al. 2017; Choy et al. 2017) and employ techniques such as environmental DNA (eDNA) metabarcoding or fatty acid analysis to identify gelatinous prey DNA and trace lipid-driven trophic pathways (Taguchi et al. 2014; Pitt et al. 2009). Such approaches would enhance the framework and provide a multidimensional perspective on the feeding ecology of longnose lancetfish in the data-scarce tropical Pacific.

Ontogenetic dietary shifts

Variation in the body size of predators are correlated with differences in resource utilization and habitat preferences. Segmented linear regression modeling of fork length and body weight revealed a growth inflection point at a fork length of 98 cm, allowing for the classification of individuals into small and large categories. The results of SCA indicated that as the size of longnose lancetfish increased, there was a corresponding decline in the consumption of small and micro prey. The greater exploitation of deep-water resources by larger individuals suggests ontogenetic shifts in vertical foraging strategy and trophic niche breadth. Additionally, muscle SIMMs results demonstrated a reduced significance of micro prey among larger longnose lancetfish (Fig. 7). In a related study, Portner et al. (2017) documented dietary variation in longnose lancetfish within the North Pacific Subtropical Gyre (NPSG) at a fork length of 97 cm, noting that larger individuals consumed a higher proportion of fish and octopods, while exhibiting a decreased consumption of crustaceans and a greater propensity for cannibalism compared to their smaller counterparts. In the central North Pacific Ocean, the abundance of cephalopods and the presumed foraging depth of longnose lancetfish prey were found to significantly increase with the growth of the fish (Chen et al. 2022). Smaller longnose lancetfish primarily consumed smaller, muscular cephalopods in shallow habitats, whereas larger individuals targeted bigger, gelatinous cephalopods in deeper waters. These observations are consistent with behaviors documented in various marine migratory species, where larger individuals tend to access bigger prey (Tsai et al. 2015).

In marine ecosystems, the isotopic values of organic matter with depth typically exhibit distinct trends influenced by multiple factors, including the source of organic material, photosynthetic activity, sediment decomposition, and mixing processes (Tuerena et al. 2019). The $\delta^{13}C$ values of organic carbon may increase with depth due to complex physical processes associated with upwelling currents, which may involve sediment decomposition and mixing, as well as varying sources of organic carbon from deeper waters (Meyers 1994; Schouten et al. 2000; Fry 2006). $\delta^{15}N$ values of organic particles tend to increase with depth as a result of microbial degradation, which may have impacted our findings (Mintenbeck et al. 2007). Elevated $\delta^{13}C$ and $\delta^{15}N$ values in the muscle tissue of larger individuals suggest heightened feeding activities in deeper or benthic layers. Furthermore, increased $\delta^{15}N$ values in larger lancetfish may indicate shifts in trophic position with ontogeny, as larger individuals may transition to consuming higher-trophic-level prey (e.g. piscivory or cephalopod predation) compared to smaller conspecifics that primarily feed on zooplankton or micronekton. This potential dietary shift is consistent with the established trophic enrichment factor of 3 to 4‰ for $\delta^{15}N$ per trophic level (Post 2002), suggesting size-dependent niche partitioning within the species. Future research that integrates SCA with isotopic mixing models would be beneficial in disentangling the effects of depth-related foraging from those associated with trophic levels.

The NMDS analysis indicated that larger longnose lancetfish exhibit more pronounced differences in feeding behaviors compared to their smaller counterparts. While larger individuals primarily focused on energetically advantageous prey such as squid and fish, their diets exhibited less consistency among individuals, implying potential niche partitioning or opportunistic feeding behaviors. In contrast, smaller individuals displayed a tighter clustering, indicative of a consistent dietary reliance on a diverse range of smaller prey, including crustaceans and gastropod organisms. This observation is consistent with findings from the SEV analysis of muscle and liver tissues. However, the increased variability observed in larger individuals may also be attributed to a smaller sample size. Notably, larger individuals demonstrated a preference for consuming bigger prey, such as fish, while still incorporating smaller prey, such as crustaceans, into their diet. This ontogenetic shift in prey size selection is likely influenced by energetic requirements and gape limitations, rather than solely by prey diversity. Such dietary transitions with growth are frequently observed across various fish species and are associated with the optimization of feeding efficiency and the reduction of intraspecific competition as individuals increase in size (Gerking 2014). Though there is no reproduction information for the longnose lancetfish, it is well-documented that a fish's diet evolves as it matures, which can significantly influence its performance and population dynamics, particularly concerning maturity stages and fluctuating energy demands (Wootton 2012; Gerking 2014). Nikolsky and Birkett (1963) posited that morphological changes could drive alterations in fish feeding behavior, with juveniles primarily targeting smaller prey that require less energy expenditure. Similar to many marine predators, longnose lancetfish exhibit a limited feeding capacity during their early developmental stages, with juveniles mainly consuming

smaller prey. Improvements in feeding mechanisms, including improved swimming capabilities, refined visual acuity, and increased gape size, broaden prey availability as they grow, allowed larger individuals to access a broader diversity of prey types (Meynier et al. 2008). This size-dependent foraging behavior aligns with the gape-limitation constraints observed in fish that consume other fish (Noakes and Godin 1988; Mittelbach and Persson 1998) and reflects the ontogenetic shifts in prey preferences noted in this species (Portner et al. 2017; Chen et al. 2022).The size-dependent dietary shifts in the ecological role of longnose lancetfish are crucial for satisfying their energy needs and adapting to the deep-sea environment within the pelagic ecosystems of the tropical western Pacific Ocean. These ontogenetic dietary transitions optimize energy acquisition for longnose lancetfish, allowing larger individuals to exploit deeper, energy-rich prey while minimizing niche overlap with smaller conspecifics. This adaptive strategy enhances their resilience to the oligotrophic conditions prevalent in the tropical western Pacific pelagic zone.

The results of our investigation into THg concentrations revealed a growth-related accumulation in longnose lancetfish, with larger specimens exhibiting elevated THg levels in both muscle and liver tissues. This observation is consistent with established principles of bioaccumulation and biomagnification in pelagic fish, wherein Hg concentrations increase with the size of the predator due to extended exposure and trophic position (Soto-Jiménez et al. 2010; Lavoie et al. 2013). Larger and older individuals demonstrated higher Hg accumulation, a trend that has been extensively documented in various fish species (Burger and Gochfeld 2011; Bradley et al. 2017). The correlation observed between THg concentrations and depth is likely influenced by environmental and habitat-related factors. In deeper mesopelagic zones, hypoxic conditions facilitate the microbial conversion of inorganic Hg to bioavailable methylmercury (MeHg) (Sunderland et al. 2006), while the longer food chains present in these environments enhance biomagnification rates (Chen et al. 2008). For example, deep-dwelling prey such as myctophids and cephalopods typically occupy higher trophic levels and possess greater Hg concentrations than shallow-water zooplankton, thereby transferring larger quantities of Hg to their predators (Le Bourg et al. 2019). Although physiological characteristics (e.g. growth dilution, detoxification) and local Hg inputs (e.g. anthropogenic pollution) can influence Hg bioaccumulation rates (Rhind 2009; Newman 2014), trophic position and habitat-specific Hg dynamics remain the predominant factors. Future research should aim to quantify mercury isotopic signatures to differentiate between dietary and environmental uptake pathways, as well as integrate SCA with vertical migration patterns to clarify the impact of depth on Hg burdens.

Conclusion

The findings of this study indicate that the diet of longnose lancetfish in the tropical western Pacific Ocean predominantly comprises crustaceans and fish, with a notable focus on epi/upper mesopelagic prey. Significant variability in feeding ecology among individual longnose lancetfish was noted, with a growing preference for fish and deeper-water prey as the fish mature. This pattern suggests niche partitioning, which may serve to mitigate intraspecific competition—a vital adaptation in the oligotrophic open ocean. Longnose lancetfish play a crucial role in mediating energy transfer between the mesopelagic and epipelagic zones. Further investigation into the feeding ecology of this pelagic predator is essential for comprehending the strategies that facilitate the survival of longnose lancetfish and other mesopelagic fish species. Although high bycatch rates in Pacific longline fisheries have not yet led to observable population declines, the ecological adaptability of longnose lancetfish may confer resilience against future disruptions in prey availability due to climate change. In long-term feeding ecology studies, SCA is subject to various influencing factors, and fluctuations in stable isotope values can obscure the effects of migratory cycles on feeding, potentially resulting in discrepancies between SCA and SIA findings. These inconsistencies underscore the necessity for multi-tracer frameworks to distinguish between short-term foraging events and long-term trophic strategies. The integration of SCA with biochemical techniques has proven to be an effective approach for exploring the feeding ecology of longnose lancetfish.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s00227-025-04697-2.

Acknowledgements This work was supported by the National Natural Science Foundation of China (#42276092) and the Program for Professor of Special Appointment (Eastern Scholar) at Shanghai Institutions of Higher Learning.

Authors Contributions YD and YL conceived and designed the experiments. YD performed the experiments and analyzed the data with the help of ZL and YL. YD wrote the manuscript with the advice of DM, YL and ZL. All authors provided editorial feedback and agreed that the manuscript should be submitted in this form.

Funding The National Natural Science Foundation of China, 42276092, Yunkai Li, Program for Professor of Special Appointment (Eastern Scholar) at Shanghai Institutions of Higher Learning.

Data Availability The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Declarations

Conflict of interest The authors declare that they have no known competing financial interests or personal relationships.

Ethical approval We followed all applicable institutional or national guidelines for the care and use of animals. All samples in this study were dead bycatch that did not engage in illegal commercial activities. This scientific research activity complies with the relevant requirements of the Wildlife Protection Law of the People's Republic of China.

References

Baker R, Buckland A, Sheaves M (2014) Fish gut content analysis: robust measures of diet composition. Fish Fish 15(1):170–177. https://doi.org/10.1111/faf.12026

Bezerra MF, Lacerda LD, Lai C (2019) Trace metals and persistent organic pollutants contamination in batoids (Chondrichthyes: Batoidea): A systematic review. Environ Pollut 248:684–695. https://doi.org/10.1016/j.envpol.2019.02.070

Boecklen WJ, Yarnes CT, Cook BA, James AC (2011) On the use of stable isotopes in trophic ecology. Annu Rev Ecol Evol Syst 42(1):411–440. https://doi.org/10.1146/annurev-ecolsys-102209-144726

Bond AL, Diamond AW (2011) Recent Bayesian stable-isotope mixing models are highly sensitive to variation in discrimination factors. Ecol Appl 21(4):1017–1023. https://doi.org/10.1890/09-2409.1

Bowman KL, Lamborg CH, Agather AM (2020) A global perspective on mercury cycling in the ocean. Sci Total Environ 710:136116. https://doi.org/10.1016/j.scitotenv.2019.136166

Bradley MA, Barst BD, Basu N (2017) A review of mercury bioavailability in humans and fish. Int J Environ Res Public Health 14(2):169. https://doi.org/10.3390/ijerph14020169

Brand WA (2011) New reporting guidelines for stable isotopes–an announcement to isotope users. Isot Environ Health Stud 47(4):535. https://doi.org/10.1080/10256016.2011.645702

Brodeur RD, Buckley TW, Lang GM, Draper DL, Buchanan JC, Hibpshman RE (2021) Demersal fish predators of gelatinous zooplankton in the northeast Pacific Ocean. Mar Ecol Prog Ser 658:89–104. https://doi.org/10.3354/meps13489

Burger J, Gochfeld M (2007) Risk to consumers from mercury in Pacific cod (*Gadus macrocephalus*) from the Aleutians: Fish age and size effects. Environ Res 105(2):276–284. https://doi.org/10.1016/j.envres.2007.05.004

Burger J, Gochfeld M (2011) Mercury and selenium levels in 19 species of saltwater fish from New Jersey as a function of species, size, and season. Sci Total Environ 409(8):1418–1429. https://doi.org/10.1016/j.scitotenv.2010.12.034

Canseco JA, Niklitschek EJ, Harrod C (2022) Variability in $\delta^{13}C$ and $\delta^{15}N$ trophic discrimination factors for teleost fishes: a meta-analysis of temperature and dietary effects. Rev Fish Biol Fish 32:313–329. https://doi.org/10.1007/s11160-021-09689-1

Carruthers EH, Schneider DC, Neilson JD (2009) Estimating the odds of survival and identifying mitigation opportunities for common bycatch in pelagic longline fisheries. Bio Conserv 142(11):2620–2630. https://doi.org/10.1016/j.biocon.2009.06.010

Casini M, Hjelm J, Molinero JC, Lövgren J, Cardinale M, Bartolino V, Belgrano A, Kornilovs G (2009) Trophic cascades promote threshold-like shifts in pelagic marine ecosystems. Proc Natl Acad Sci 106(1):197–202. https://doi.org/10.1073/pnas.0806649105

Caut S, Angulo E, Courchamp F (2009) Variation in discrimination factors ($\Delta^{15}N$ and $\Delta^{13}C$): the effect of diet isotopic values and applications for diet reconstruction. J Appl Ecol 46(2):443–453. https://doi.org/10.1111/j.1365-2664.2009.01620.x

Chen C, Amirbahman A, Fisher N, Harding G, Lamborg C, Nacci D, Taylor D (2008) Methylmercury in marine ecosystems: spatial patterns and processes of production, bioaccumulation, and biomagnification. EcoHealth 5:399–408. https://doi.org/10.1007/s10393-008-0201-1

Chen RS, Portner EJ, Choy CA (2022) Gelatinous cephalopods as important prey for a deep-sea fish predator. Mar Biol 169(12):155. https://doi.org/10.1007/s00227-022-04116-w

Chen RS, Paulson ET, Schartup AT, Choy CA (2025) Diet shifts drive mercury bioaccumulation and distribution in tissues of the longnose lancetfish (*Alepisaurus ferox*). Mar Pollut Bull 213:117590. https://doi.org/10.1016/j.marpolbul.2025.117590

Chételat J, Shao Y, Richardson MC, MacMillan GA, Amyot M, Drevnick PE, Gill H, Köck G, Muir DC (2021) Diet influences on growth and mercury concentrations of two salmonid species from lakes in the eastern Canadian Arctic. Environ Pollut 268:115820. https://doi.org/10.1016/j.envpol.2020.115820

Choy CA, Popp BN, Kaneko JJ, Drazen JC (2009) The influence of depth on mercury levels in pelagic fishes and their prey. Proc Natl Acad Sci 106(33):13865–13869. https://doi.org/10.1073/pnas.0900711106

Choy CA, Portner E, Iwane M, Drazen JC (2013) Diets of five important predatory mesopelagic fishes of the central North Pacific. Mar Ecol Prog Ser 492:169–184. https://doi.org/10.3354/meps10518

Choy CA, Haddock SH, Robison BH (2017) Deep pelagic food web structure as revealed by *in situ* feeding observations. Proc R Soc B Biol Sci 284(1868):20172116. https://doi.org/10.1098/rspb.2017.2116

Clarke LJ, Trebilco R, Walters A, Polanowski AM, Deagle BE (2020) DNA-based diet analysis of mesopelagic fish from the southern *Kerguelen Axis*. Deep Sea Res Part II: Top Stud Oceanogr 174. https://doi.org/10.1016/j.dsr2.2018.09.001

Cortés E (1997) A critical review of methods of studying fish feeding based on analysis of stomach contents: application to elasmobranch fishes. Can J Fish Aquat Sci 54(3):726–738. https://doi.org/10.1139/cjfas-54-3-726

Davison PC, Checkley DM Jr, Koslow JA, Barlow J (2013) Carbon export mediated by mesopelagic fishes in the northeast Pacific Ocean. Prog Oceanogr 116:14–30. https://doi.org/10.1016/j.pocean.2013.05.013

Dalerum F, Angerbjörn A (2005) Resolving temporal variation in vertebrate diets using naturally occurring stable isotopes. Oecologia 144:647–658. https://doi.org/10.1007/s00442-005-0118-0

Diaz Briz L, Sánchez F, Marí N, Mianzan H, Genzano G (2017) Gelatinous zooplankton (ctenophores, salps and medusae): an important food resource of fishes in the temperate SW Atlantic Ocean. Mar Biol Res 13(6):630–644. https://doi.org/10.1080/17451000.2016.1274403

Drazen JC, Sutton TT (2017) Dining in the deep: the feeding ecology of deep-sea fishes. Annu Rev Mar Sci 9(1):337–366. https://doi.org/10.1146/annurev-marine-010816-060543

Fonteles-Filho AA (2011) Oceanografia, biologia, dinâmica populacional de recurso pesqueiros. 1 Edição. Expressão Gráfica e Editora. 464p.

Fry B, Arnold C (1982) Rapid $^{13}C/^{12}C$ turnover during growth of brown shrimp (*Penaeus aztecus*). Oecologia 54:200–204. https://doi.org/10.1007/BF00378393

Fry, B (2006) Stable isotope ecology (Vol.521, p318) New York: Springer.

Fujinami Y, Nakatsuka S, Ohshimo S (2018) Feeding Habits of the Blue Shark (*Prionace glauca*) in the Northwestern Pacific Based on Stomach Contents and Stable Isotope Ratios. Pac Sci 72(1):21–39. https://doi.org/10.2984/72.1.2

Gao H, Yu R, Gong Y, Chen X, Li Y, Liu B, Gan Q (2024) Diversity but not abundance of ingested plastics changes with ontogenetic dietary shift: Stable isotope insights into plastic contamination in a mesopelagic predator longnose lancetfish *Alepisaurus ferox*. Mar Pollut Bull 208:116910. https://doi.org/10.1016/j.marpolbul.2024.116910

Gelman A, Carlin JB, Stern HS, Rubin DB (1995) Bayesian data analysis Chapman and Hall/CRC.

Gerking SD (2014) Feeding ecology of fish. Elsevier

Gjøsæter J, Kawaguchi K (1980) A review of the world resources of mesopelagic fish.

Grey J, Thackeray SJ, Jones RI, Shine A (2002) Ferox Trout (*Salmo trutta*) as "Russian dolls": complementary gut content and stable isotope analyses of the Loch Ness foodweb. Freshw Biol 47(7):1235–1243. https://doi.org/10.1046/j.1365-2427.2002.00838.x

Grubbs RD, Carlson JK, Romine JG, Curtis TH, McElroy WD, McCandless CT, Musick JA (2016) Critical assessment and ramifications of a purported marine trophic cascade. Sci Rep 6(1):20970. https://doi.org/10.1038/srep20970

Heithaus MR, Wirsing AJ, Dill LM (2012) The ecological importance of intact top-predator populations: a synthesis of 15 years of research in a seagrass ecosystem. Mar Freshw Res 63(11):1039–1050. https://doi.org/10.1071/MF12024

Herzka SZ, Holt GJ (2000) Changes in isotopic composition of red drum (*Sciaenops ocellatus*) larvae in response to dietary shifts: potential applications to settlement studies. Can J Fish Aquat Sci 57(1):137–147. https://doi.org/10.1139/f99-174

Hesslein RH, Hallard KA, Ramlal P (1993) Replacement of sulfur, carbon, and nitrogen in tissue of growing broad whitefish (*Coregonus nasus*) in response to a change in diet traced by $\delta^{34}S$, $\delta^{13}C$, and $\delta^{15}N$. Can J Fish Aquat Sci 50(10):2071–2076. https://doi.org/10.1139/f93-230

Hilgendag IR, Swanson HK, Lewis CW, Ehrman AD, Power M (2022) Mercury biomagnification in benthic, pelagic, and benthopelagic food webs in an arctic marine ecosystem. Sci Total Environ 841:156424. https://doi.org/10.1016/j.scitotenv.2022.156424

Hyslop EJ (1980) Stomach contents analysis—a review of methods and their application. J Fish Biol 17(4):411–429. https://doi.org/10.1111/j.1095-8649.1980.tb02775.x

Iglesias IS, Santora JA, Fiechter J, Field JC (2023) Mesopelagic fishes are important prey for a diversity of predators. Front Mar Sci 10:1220088. https://doi.org/10.3389/fmars.2023.1220088

Ishikawa NF, Hyodo F, Tayasu I (2013) Use of carbon-13 and carbon-14 natural abundances for stream food web studies. Ecol Res 28:759–769. https://doi.org/10.1007/s11284-012-1003-z

Ivanova NV, Zemlak TS, Hanner RH, Hebert PD (2007) Universal primer cocktails for fish DNA barcoding. Mol Ecol Notes 7(4):544–548. https://doi.org/10.1111/j.1471-8286.2007.01748.x

Jackson GD, Buxton NG, George MJ (2000) Diet of the southern opah *Lampris immaculatus* on the Patagonian Shelf; the significance of the squid *Moroteuthis ingens* and anthropogenic plastic. Mar Ecol Prog Ser 206:261–271. https://doi.org/10.3354/meps206261

Jackson AL, Inger R, Parnell AC, Bearhop S (2011) Comparing isotopic niche widths among and within communities: SIBER-Stable Isotope Bayesian Ellipses in R. J Anim Ecol 80(3):595–602. https://doi.org/10.1111/j.1365-2656.2011.01806.x

Jantz LA, Morishige CL, Bruland GL, Lepczyk CA (2013) Ingestion of plastic marine debris by longnose lancetfish (*Alepisaurus ferox*) in the North Pacific Ocean. Mar Pollut Bull 69(1–2):97–104. https://doi.org/10.1016/j.marpolbul.2013.01.019

Keller HR, Hirons AC, Kerstetter DW (2016) Combined stomach content and $\delta^{13}C/\delta^{15}N$ analyses of oilfish, escolar, snake mackerel and lancetfish in the western North Atlantic. Mar Ecol 37(4):727–736. https://doi.org/10.1111/maec.12317

Kiszka JJ, Aubail A, Hussey EN, Heithaus MR, Caurant F, Bustamante P (2015) Plasticity of trophic interactions among sharks from the oceanic south-western Indian Ocean revealed by stable isotope and mercury analyses. Deep-Sea Res Part i: Oceanogr Res Pap 96:49–58. https://doi.org/10.1016/j.dsr.2014.11.006

Kong X, Zhang J, Li Y, Otsuka S, Liu Q, He Q (2023) Selenium in the liver facilitates the biodilution of mercury in the muscle of *Planiliza haematocheilus* in the Jiaozhou Bay. China Ecotoxicol Environ Saf 258:114981. https://doi.org/10.1016/j.ecoenv.2023.114981

Krebs CJ (1989) Ecological methodology, vol 654. Harper Colins, New York

Lamborg CH, Hammerschmidt CR, Bowman KL, Swarr GJ, Munson KM, Ohnemus DC, Lam PJ, Heimbürger LE, Rijkenberg MJ, Saito MA (2014) A global ocean inventory of anthropogenic mercury based on water column measurements. Nature 512(7152):65–68. https://doi.org/10.1038/nature13563

Lavoie RA, Jardine TD, Chumchal MM, Kidd KA, Campbell LM (2013) Biomagnification of mercury in aquatic food webs: a worldwide meta-analysis. Environ Sci Technol 47(23):13385–13394. https://doi.org/10.1021/es403103t

Le Bourg B, Kiszka JJ, Bustamante P, Heithaus MR, Jaquemet S, Humber F (2019) Effect of body length, trophic position and habitat use on mercury concentrations of sharks from contrasted ecosystems in the southwestern Indian Ocean. Environ Res 169:387–395. https://doi.org/10.1016/j.envres.2018.11.024

Li Z, Pethybridge HR, Gong Y, Wu F, Dai X, Li Y (2022) Effect of body size, feeding ecology and maternal transfer on mercury accumulation of vulnerable silky shark *Carcharhinus falciformis* in the eastern tropical pacific. Environ Pollut 309:119751. https://doi.org/10.1016/j.envpol.2022.119751

Liu P, Dai X, Wang J, Gao C, Wu F, Wang X (2019) Composition of stomach contents of longnose lancetfish (*Alepisaurus ferox*) in western tropical Pacific Ocean. South China Fish Sci 15(1):20–30

Logan JM, Jardine TD, Miller TJ, Bunn SE, Cunjak RA, Lutcavage ME (2008) Lipid corrections in carbon and nitrogen stable isotope analyses: comparison of chemical extraction and modelling methods. J Anim Ecol:838–846. https://doi.org/10.1111/J.1365-2656.2008.01394.x

Logan JM, Lutcavage ME (2010) Stable isotope dynamics in elasmobranch fishes. Hydrobiologia 644:231–244. https://doi.org/10.1007/s10750-010-0120-3

Martínez del Rio C, Wolf N, Carleton SA, Gannes LZ (2009) Isotopic ecology ten years after a call for more laboratory experiments. Biol Rev 84(1):91–111. https://doi.org/10.1111/j.1469-185X.2008.00064.x

Meyers PA (1994) Preservation of elemental and isotopic source identification of sedimentary organic matter. Chem Geol 114(3–4):289–302. https://doi.org/10.1016/0009-2541(94)90059-0

Meynier L, Pusineri C, Spitz J, Santos MB, Pierce GJ, Ridoux V (2008) Intraspecific dietary variation in the short-beaked common dolphin *Delphinus delphis* in the Bay of Biscay: importance of fat fish. Mar Ecol Prog 354:277–287. https://doi.org/10.3354/meps07246

Mintenbeck K, Jacob U, Kunst R, Arntz WE, Brey T (2007) Depth-dependence in stable isotope ratio $\delta^{15}N$ of benthic POM consumers: the role of particle dynamics and organism trophic guild. Deep Sea Res Part I Oceanogr Res Pap 54(6):1015–1023. https://doi.org/10.1016/j.dsr.2007.03.005

Mittelbach GG, Persson L (1998) The ontogeny of piscivory and its ecological consequences. Can J Fish Aquat Sci 55(6):1454–1465. https://doi.org/10.1139/f98-041

Monteiro LR, Costa V, Furness RW, Santos RS (1996) Mercury concentrations in prey fish indicate enhanced bioaccumulation in mesopelagic environments. Mar Ecol Prog Ser 141:21–25. https://doi.org/10.3354/meps141021

Moteki M (1993) Stomach contents of longnose lancetfish, *Alepisaurus ferox*, in Hawaiian and central equatorial Pacific waters. J Tokyo Univ Fish 80:121–137

Newman MC (2014) Fundamentals of ecotoxicology: the Science of Pollution CRC press.

Newsome SD, Martinez del Rio C, Bearhop S, Phillips DL (2007) A niche for isotopic ecology. Front Ecol Environ 5(8):429–436. https://doi.org/10.1890/060150.1

Nicula M, Negrea P, Gergen I, Harmanescu M, Gogoasa I, Lunca M (2009) Mercury bioaccumulation in tissues of fresh water fish *Carassius auratus gibelio* (Silver Crucian Carp) after chronic mercury intoxication. Univ Stiinte Agric Med Vet Lasi Lucr Stiint 5:676–679

Nikolsky GV, Birkett L (1963) The ecology of fishes (Trans Birkett L) Academic Press. London:p352.

Noakes DLG, Godin JGJ (1988) 5 ontogeny of behavior and concurrent developmental changes in sensory systems in teleost fishes. Fish Physiol 11:345–395. https://doi.org/10.1016/S1546-5098(08)60217-4

O'Bryhim JR, Adams DH, Spaet JL, Mills G, Lance SL (2017) Relationships of mercury concentrations across tissue types, muscle regions and fins for two shark species. Environ Pollut 223:323–333. https://doi.org/10.1016/j.envpol.2017.01.029

Olivar MP, Bode A, López-Pérez C, Hulley PA, Hernández-León S (2019) Trophic position of lanternfishes (Pisces: Myctophidae) of the tropical and equatorial Atlantic estimated using stable isotopes. ICES J Mar Sci 76(3):649–661. https://doi.org/10.1093/icesjms/fsx243

Olson RJ, Popp BN, Graham BS, López-Ibarra GA, Galván-Magaña F, Lennert-Cody CE, Fry B (2010) Food-web inferences of stable isotope spatial patterns in copepods and yellowfin tuna in the pelagic eastern Pacific Ocean. Prog Oceanogr 86:124–138. https://doi.org/10.1016/j.pocean.2010.04.026

Onsanit S, Wang WX (2011) Sequestration of total and methyl mercury in different subcellular pools in marine caged fish. J Hazard Mater 198:113–122. https://doi.org/10.1016/j.jhazmat.2011.10.020

Ortuño Crespo G, Dunn DC (2017) A review of the impacts of fisheries on open-ocean ecosystems. ICES J Mar Sci 74(9):2283–2297. https://doi.org/10.1093/icesjms/fsx084

Pacoureau N, Rigby CL, Kyne PM, Sherley RB, Winker H, Carlson JK, Fordham SV, Barreto R, Fernando D, Francis MP, Jabado RW, Herman KB, Liu KM, Marshall AD, Pollom RA, Romanov EV, Simpfendorfer CA, Yin JS, Kindsvater HK, Dulvy NK (2021) Half a century of global decline in oceanic sharks and rays. Nature 589(7843):567–571. https://doi.org/10.1038/s41586-020-03173-9

Pan B, Zhu J, Lin Q, Geng Z, Wu F, Zhang Y (2024) Study on the catch, bycatch and discard of Chinese pelagic longline fisheries in the Atlantic Ocean. Aquac Fish 9(2):280–286. https://doi.org/10.1016/j.aaf.2022.03.002

Parnell AC, Phillips DL, Bearhop S, Semmens BX, Ward EJ, Moore JW, Inger R (2013) Bayesian stable isotope mixing models. Environmetrics 24(6):387–399. https://doi.org/10.1002/env.2221

Perga ME, Gerdeaux D (2005) 'Are fish what they eat' all year round? Oecologia 144(4):598–606. https://doi.org/10.1007/s00442-005-0069-5

Pitt KA, Connolly RM, Meziane T (2009) Stable isotope and fatty acid tracers in energy and nutrient studies of jellyfish: a review. Hydrobiologia 616(1):119–132. https://doi.org/10.1007/978-1-4020-9749-2_9

Polo-Silva C, Newsome SD, Galván-Magaña F, Grijalba-Bendeck M, Sanjuan-Muñoz A (2013) Trophic shift in the diet of the pelagic thresher shark based on stomach contents and stable isotope analyses. Mar Biol Res 9(10):958–971. https://doi.org/10.1080/17451000.2013.793802

Portner EJ, Polovina JJ, Choy CA (2017) Patterns in micronekton diversity across the North Pacific Subtropical Gyre observed from the diet of longnose lancetfish (*Alepisaurus ferox*). Deep Sea Res Part i: Oceanogr Res Pap 125:40–51. https://doi.org/10.1016/j.dsr.2017.04.013

Portner EJ, Mowatt-Larssen T, Carretero ACL, Contreras EA, Woodworth-Jefcoats PA, Frable BW, Choy CA (2023) Harnessing a mesopelagic predator as a biological sampler reveals taxonomic and vertical resource partitioning among three poorly known deep-sea fishes. Sci Rep 13(1):16078. https://doi.org/10.1038/s41598-023-41298-9

Post DM (2002) Using stable isotopes to estimate trophic position: models, methods, and assumptions. Ecology 83(3):703–718. https://doi.org/10.2307/3071875

Post DM, Layman CA, Arrington DA, Takimoto G, Quattrochi J, Montana CG (2007) Getting to the fat of the matter: models, methods and assumptions for dealing with lipids in stable-isotope analyses. Oecologia 152:179–189. https://doi.org/10.1007/s00442-006-0630-x

Potier M, Marsac F, Cherel Y, Lucas V, Sabatié R, Maury O, Ménard F (2007a) Forage fauna in the diet of three large pelagic fishes (lancetfish, swordfish and yellowfin tuna) in the western equatorial Indian Ocean. Fish Res 83(1):60–72. https://doi.org/10.1016/j.fishres.2006.08.020

Potier M, Ménard F, Cherel Y, Lorrain A, Sabatié R, Marsac F (2007b) Role of pelagic crustaceans in the diet of the longnose lancetfish *Alepisaurus ferox* in the Seychelles waters. Afr J Mar Sci 29(1):113–122. https://doi.org/10.2989/AJMS.2007.29.1.10.75

Rhind S (2009) Anthropogenic pollutants: a threat to ecosystem sustainability? Philos Trans R Soc b: Biol Sci 364(1534):3391–3401. https://doi.org/10.1098/rstb.2009.0122

Rupp A, Bornatowski H (2021) Food web model to assess the fishing impacts and ecological role of elasmobranchs in a coastal ecosystem of Southern Brazil. Environ Biol Fish 104(8):15. https://doi.org/10.1007/s10641-021-01120-9

Santander-Neto J, Freitas DJ, Bornatowski H, Lessa R (2021) Feeding habits of *Urotrygon microphthalmum* (Myliobatiformes: Urotrygonidae) caught off northeastern Brazil. Neotrop Ichthyol 19:e210039. https://doi.org/10.1590/1982-0224-2021-0039

Sardenne F, Hollanda S, Lawrence S, Albert-Arrisol R, Degroote M, Bodin N (2017) Trophic structures in tropical marine ecosystems: a comparative investigation using three different ecological tracers. Ecol Indic 81:315–324. https://doi.org/10.1016/j.ecolind.2017.06.001

Sassa C, Kawaguchi K, Hirota Y, Ishida M (2004) Distribution patterns of larval myctophid fish assemblages in the subtropical–tropical waters of the western North Pacific. Fish Oceanogr 13(14):267–282. https://doi.org/10.1111/j.1365-2419.2004.00289.x

Sassa C (2019) Estimation of the spawning biomass of myctophids based on larval production and reproductive parameters: the case study of *Benthosema pterotum* in the East China Sea. ICES J Mar Sci 76(3):743–754. https://doi.org/10.1093/icesjms/fsy051

Schouten S, Hopmans EC, Pancost RD, Damsté JSS (2000) Widespread occurrence of structurally diverse tetraether membrane lipids: evidence for the ubiquitous presence of low-temperature relatives of hyperthermophiles. Proc Natl Acad Sci 97(26):14421–14426. https://doi.org/10.1073/pnas.97.26.14421

Segura AM, Milessi AC, Vögler R, Galvan-Magaña F, Muggeo VMR (2013) The determination of maturity stages in male elasmobranchs (Chondrichthyes) using a segmented regression of clasper length on total length. Can J Fish Aquat Sci 70(6):830–833. https://doi.org/10.1139/cjfas-2012-0545

Skinner C, Mill AC, Newman SP, Newton J, Cobain MR, Polunin NV (2019) Novel tri-isotope ellipsoid approach reveals dietary variation in sympatric predators. Ecol Evol 9(23):13267–13277. https://doi.org/10.1002/ece3.5779

Soto-Jiménez MF, Amezcua F, González-Ledesma R (2010) Nonessential metals in striped marlin and Indo-Pacific sailfish in the southeast Gulf of California, Mexico: concentration and assessment of human health risk. Arch Environ Contam Toxicol 58:810–818. https://doi.org/10.1007/s00244-009-9452-2

Spiers EK, Stafford R, Ramirez M, Izurieta DFV, Cornejo M, Chavarria J (2016) Potential role of predators on carbon dynamics of marine ecosystems as assessed by a Bayesian belief network. Ecol Inform 36:77–83. https://doi.org/10.1016/j.ecoinf.2016.10.003

Stephens RB, Shipley ON, Moll RJ (2023) Meta-analysis and critical review of trophic discrimination factors ($\Delta^{13}C$ and $\Delta^{15}N$): Importance of tissue, trophic level and diet source. Funct Ecol 37(9):2535–2548. https://doi.org/10.1111/1365-2435.14403

Stock BC, Jackson AL, Ward EJ, Parnell AC, Phillips DL, Semmens BX (2018) Analyzing mixing systems using a new generation of Bayesian tracer mixing models. PeerJ 6:e5096. https://doi.org/10.7717/peerj.5096

Sunderland EM, Gobas FA, Branfireun BA, Heyes A (2006) Environmental controls on the speciation and distribution of mercury in coastal sediments. Mar Chem 102(1–2):111–123. https://doi.org/10.1016/j.marchem.2005.09.019

Sweeting CJ, Barry JT, Polunin NVC, Jennings S (2007a) Effects of body size and environment on diet-tissue $\delta^{15}N$ fractionation in fishes. J Exp Mar Biol Ecol 340:1–10

Sweeting CJ, Barry JT, Polunin NVC, Jennings S (2007b) Effects of body size and environment on diet-tissue $\delta^{13}C$ fractionation in fishes. J Exp Mar Biol Ecol 352:165–176. https://doi.org/10.1016/j.jembe.2007.07.007

Taguchi T, Miura Y, Krueger D, Sugiura S (2014) Utilizing stomach content and faecal DNA analysis techniques to assess the feeding behaviour of largemouth bass *Micropterus Salmoides* and bluegill lepomis macrochirus. J Fish Biol 84(5):1271–1288. https://doi.org/10.1111/jfb.12341

Tieszen LL, Boutton TW, Tesdahl KG, Slade NA (1983) Fractionation and turnover of stable carbon isotopes in animal tissues: implications for $\delta^{13}C$ analysis of diet. Oecologia 57:32–37. https://doi.org/10.1111/jfb.12341

Tsai CN, Chiang WC, Sun CL, Shao KT, Chen SY, Yeh SZ (2015) Stomach content and stable isotope analysis of sailfish (*Istiophorus platypterus*) diet in eastern Taiwan waters. Fish Res 166:39–46. https://doi.org/10.1016/j.fishres.2014.10.021

Trujillo-Olvera A, Ortega-García S, Tripp-Valdez A, Escobar-Sánchez O, Acosta-Pachón TA (2018) Feeding habits of the swordfish (*Xiphias gladius* Linnaeus, 1758) in the subtropical northeast Pacific. Hydrobiologia 822:173–188. https://doi.org/10.1007/s10750-018-3680-2

Tuerena RE, Ganeshram RS, Humphreys MP, Browning TJ, Bouman H, Piotrowski AP (2019) Isotopic fractionation of carbon during uptake by phytoplankton across the South Atlantic subtropical convergence. Biogeosciences 16(18):3621–3635. https://doi.org/10.5194/BG-16-3621-2019

Vanderklift MA, Ponsard S (2003) Sources of variation in consumer-diet $\delta^{15}N$ enrichment: a meta-analysis. Oecologia 136:169–182. https://doi.org/10.1007/s00442-003-1270-z

Vander Zanden MJ, Rasmussen JB (1999) Primary consumer $\delta^{13}C$ and $\delta^{15}N$ and the trophic position of aquatic consumers. Ecology:1395–1404.

Vander Zanden MJ, Clayton MK, Moody EK, Solomon CT, Weidel BC (2015) Stable isotope turnover and half-life in animal tissues: a literature synthesis. PLoS ONE 10(1):e0116182. https://doi.org/10.1371/journal.pone.0116182

Varghese SP, Somvanshi VS, Varghese S (2010) Discontinuous distribution of *Alepisaurus ferox* Lowe, 1833 (Alepisauridae, Teleostei) in the Indian EEZ as revealed by the tuna longline survey. Indian J Geo-Mar Sci 39(3):406–414. https://doi.org/10.1007/s13131-010-0069-0

Wang R, Wang WX (2018) Diet-specific trophic transfer of mercury in tilapia (*Oreochromis niloticus*): biodynamic perspective. Environ Pollut 234:288–296. https://doi.org/10.1016/j.envpol.2017.11.071

Ward P, Myers RA (2005) Shifts in open-ocean fish communities coinciding with the commencement of commercial fishing. Ecol 86(4):835–847. https://doi.org/10.1890/03-0746

Ward RD, Zemlak TS, Innes BH, Last PR, Hebert PD (2005) DNA barcoding Australia's fish species. Philos Trans R Soc b: Biol Sci 360(1462):1847–1857. https://doi.org/10.2307/30040931

Wassersug RJ, Johnson RK (1976) A remarkable pyloric caecum in the evermannellid genus *Coccorella* with notes on gut structure and function in alepisauroid fishes (Pisces, Myctophiformes). J Zool 179(2):273–289. https://doi.org/10.1111/j.1469-7998.1976.tb02296.x

Weng JS, Lee MA, Liu KM, Hsu MS, Hung MK, Wu LJ (2015) Feeding ecology of juvenile yellowfin tuna from waters southwest of Taiwan inferred from stomach contents and stable isotope analysis. Mar Coast Fish 7(1):537–548. https://doi.org/10.1080/19425120.2015.1094157

Woods B, Trebilco R, Walters A, Hindell M, Duhamel G, Flores H, Moteki M, Pruvost P, Reiss C, Saunders RA, Sutton C, Gan YM, Van de Putte A (2022) Myctobase, a circumpolar database of mesopelagic fishes for new insights into deep pelagic prey fields. Sci Data 9(1):404. https://doi.org/10.1038/s41597-022-01496-y

Woods BL, Van de Putte AP, Hindell MA, Raymond B, Saunders RA, Walters A, Trebilco R (2023) Species distribution models describe spatial variability in mesopelagic fish abundance in the Southern Ocean. Front Mar Sci 9:981434. https://doi.org/10.3389/fmars.2022.981434

Wootton RJ (2012) Ecology of teleost fishes (Vol.1) Springer Science and Business Media.

Worm B, Tittensor DP (2011) Range contraction in large pelagic predators. Proc Natl Acad Sci 108(29):11942–11947. https://doi.org/10.1073/pnas.1102353108

Worm B, Davis B, Kettemer L, Ward-Paige CA, Chapman D, Heithaus MR, Kessel ST, Gruber SH (2013) Global catches, exploitation rates, and rebuilding options for sharks. Mar Policy 40:194–204. https://doi.org/10.1016/j.marpol.2012.12.034

Young JW, Lansdell MJ, Campbell RA, Cooper SP, Juanes F, Guest MA (2010) Feeding ecology and niche segregation in oceanic top predators off eastern Australia. Mar Biol 157:2347–2368. https://doi.org/10.1007/s00227-010-1500-y

Young JW, Hunt BP, Cook TR, Llopiz JK, Hazen EL, Pethybridge HR, Ceccarelli D, Lorrain A, Olson RJ, Allain V, Menkes C, Patterson T, Nicol S, Lehodey P, Kloser RJ, Arrizabalaga H, Choy CA (2015) The trophodynamics of marine top predators: current knowledge, recent advances and challenges. Deep Sea Res Part II: Top Stud Oceanogr 113:170–187. https://doi.org/10.1016/j.dsr2.2014.05.015

Zhang B, Pethybridge H, Sutton C, Virtue P, Li Y (2024) Total mercury concentrations in Tasman sea mesopelagic fish: exploring biotic and abiotic drivers. Mar Pollut Bull 206:116676. https://doi.org/10.1016/j.marpolbul.2024.116676