

# The foraging ecology of coastal bottlenose dolphins based on stable isotope mixing models and behavioural sampling

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**Abstract** Understanding trophic interactions is critical for elucidating ecological roles of marine predators. We used behavioural observations and stable isotope mixing models to investigate the feeding ecology of Indo-Pacific bottlenose dolphins (*Tursiops aduncus*) in the lagoon of Mayotte (East Africa). We identified prey during 77 % of 54 observed feeding events, observed in both rainy (61 % of events) and dry (39 %) seasons. *Caranx melampygus* and *Gnathanodon speciosus* were involved in 67 % of these events, with *Tylosurus crocodilus* (20 %) and *Mugil cephalus* (13 %) also consumed. Mixing models, based on  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values of skin and blubber ( $n = 30$  samples for both tissues), suggest that behavioural observations are representative of general feeding patterns. Indeed, *C. melampygus* and *T. crocodilus* (*G. speciosus* could not be included in models) were estimated to contribute most to dolphin diets, with mean estimated contributions of 44.6 % ( $\pm 18.9$ ) and 48.1 % ( $\pm 19.1$ ) for skin and 73.7 % ( $\pm 14.9$ ) and 16.9 % ( $\pm 12.4$ ) for blubber, respectively. Our results highlight the value of two independent methods (stable isotopes and behavioural observations) to assess prey preferences of free-ranging dolphins.

## Introduction

A first step in understanding ecological roles of marine predators is characterizing their trophic relationships. However, collecting qualitative and quantitative information on diets of large and mobile marine animals is challenging. This is especially true for cetaceans that are wide-ranging, spend most of their time underwater or are simply difficult to sample due to their elusive nature. Investigating the diet of cetaceans, such as dolphins, is generally limited to the examination of stomach content from stranded and/or incidentally captured individuals. Although stomach content analyses tend to over- or underestimate prey contributions due to differences in their digestion rates (Pierce and Boyle 1991), it allows identification of prey at a fine taxonomic level (Pierce and Boyle 1991; Gannon et al. 1997; Spitz et al. 2011). Nevertheless, specimens required for performing such analyses are often unavailable, especially around tropical islands where cetacean carcasses are quickly scavenged, and therefore opportunities to access biological material from stranded animals might be rare (Kiszka et al. 2010a). The collection of faecal samples from free-ranging cetaceans can be another means of investigating diet (e.g. sperm whales *Physeter macrocephalus*; Smith and Whitehead 2000), but samples are very difficult to collect with unpredictably and fast moving small-sized dolphins.

In the recent years, biopsy samples have been used to investigate the trophic ecology of free-ranging cetaceans, especially using fatty acid signatures and stable isotope analyses (Hooker et al. 2001; Quérouil et al. 2013). The use of naturally occurring nitrogen and carbon stable isotopes provides information on top predator feeding ecology, including cetaceans and delphinids in particular (Di Benedetto et al. 2011; Praca et al. 2011; Méndez-Fernandez et al. 2012). Carbon and nitrogen stable isotope ratios

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depict the primary producers supporting a species' diet and the relative trophic position of the consumer, respectively (Hobson 1999). This approach is generally considered as complementary to other approaches such as behavioural sampling, stomach and faeces content analyses. In marine predators, stable isotope analyses are particularly useful to investigate trophic relationships within a community of predators (Lesage et al. 2001; Zhao et al. 2004; Heithaus et al. 2013), but also ontogenetic, geographic and sex variation in diets and habitat use (Kiszka et al. 2010a; Meissner et al. 2011). Moreover, stable isotopes integrate diet over varying temporal scales depending on the turnover rate of the tissue considered (Hobson and Sease 1998; Outridge and Stewart 1999). A drawback of stable isotopes is that they do not necessarily provide detailed information on the prey types predators consumed. Mass-balance models (i.e. mixing models), however, address this issue by estimating the probability distributions of contributions of different prey sources based on stable isotope data from predator and a range of potential prey (Phillips and Gregg 2003; Urton and Hobson 2005; Parnell et al. 2010). Importantly, these models incorporate uncertainty for each parameter and specific diet-tissue discrimination factors, also called trophic enrichment factors (TEFs). Such methods have begun to advance regional knowledge on the diet and trophic ecology of cetaceans (Jansen et al. 2012; Pomerleau et al. 2012; Witteveen et al. 2012).

Bottlenose dolphins (*Tursiops* spp) are upper trophic level predators that can exploit diverse resources and habitats from inshore to oceanic ecosystems. They also display considerable variation in foraging tactics, and prey selection within and among populations is related to adaptation to local ecological conditions and can be maintained through cultural transmission (Mann and Sargeant 2003; Krützen et al. 2005; Sargeant et al. 2007). Around the island of Mayotte (East Africa, Comoros archipelago), a diverse community of tropical delphinids occurs from coastal to oceanic habitats (Kiszka et al. 2011). The Indo-Pacific bottlenose dolphin (*Tursiops aduncus*), hereafter bottlenose dolphin, is by far the most common cetacean living inside the lagoon and is likely to be the largest top predator. Previous studies highlight its preference for coastal areas (<30 m deep) inside the lagoon and shallow waters of adjacent reef banks (Kiszka et al. 2012). From 2004 to 2008, estimated annual abundances of bottlenose dolphins ranged from  $47 \pm 18$  to  $98 \pm 50$  individuals, suggesting low population size across its range around the island (estimated population home range is  $978 \text{ km}^2$ , Pusineri et al. 2014). Other studies suggest that bottlenose dolphins around Mayotte are at risk from habitat degradation, prey depletion and disturbance (Kiszka et al. 2009, 2012). Based on IUCN guidelines for local classification, bottlenose dolphins around Mayotte have been classified as Endangered (Pusineri et al.

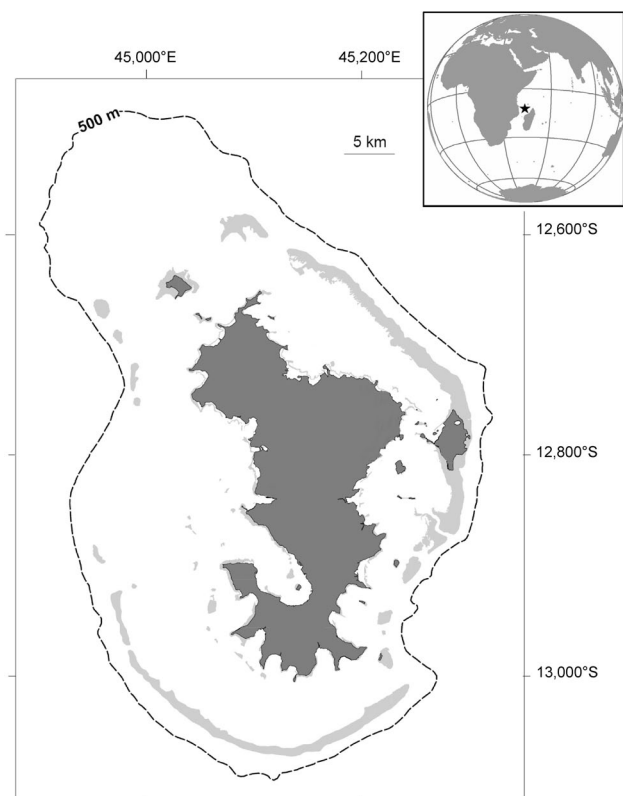
2014). Therefore, information on the ecology of this population is necessary for management and conservation. Dietary information, for example, is needed to infer biological interactions between dolphins and local fisheries.

In this study, we used behavioural observations (feeding events) and stable isotope mixing models from tissues with different turnover rates (2–3 months for skin and significantly longer for blubber, i.e. several months to a year; Hicks et al. 1985; Abend and Smith 1995) to investigate the feeding ecology of bottlenose dolphins around Mayotte. We further investigated whether these techniques provided similar insights into dolphin trophic interactions or whether dolphins foraged on a wider array of taxa than might be predicted based on surface observations.

## Materials and methods

### Data collection

Mayotte is part of the Comoros archipelago located in the northeast of the Mozambique Channel ( $12^{\circ}50'S$ ,  $45^{\circ}10'E$ , Fig. 1). From July 2004 to April 2009, small-boat-based surveys were undertaken around the island during daylight



**Fig. 1** Location of the island of Mayotte and its lagoon (dark grey barrier reef; light grey fringing and double barrier reef)

hours to collect cetacean sighting and behavioural data, as well as blubber and skin biopsies for stable isotope (see Kiszka et al. 2011, 2012 for details on data, sample collection and locations).

Behavioural states were recorded according to Kiszka et al. (2011, 2012). For the present study, we only considered foraging groups in which feeding events were observed. Here, a feeding event refers to the capture of prey by one or several individual dolphins. Prey was frequently seen at the surface during foraging, and bottlenose dolphins were frequently observed with prey at the surface. Prey items were visually identified from the research boat to the lowest possible taxon.

In order to perform stable isotope analysis in dolphins, biopsies (skin and blubber) were collected and samples were treated according to Kiszka et al. (2010b, 2011, 2012). Muscle samples from several fish species were also collected for stable isotope analyses, especially to investigate trophic interactions between delphinids and potential dolphin prey as well as fish species that are found in distinct habitats that should exhibit characteristic  $\delta^{13}\text{C}$  values (seagrass, lagoon and forereef-associated; Kiszka et al. 2011). White dorsal fish muscle samples were collected from a local fish market in April 2009. Tissues were freeze-dried before shipping and subsequent analysis.

#### Stable isotope analyses

In dolphins, stable isotope analyses were performed using skin (epidermis) and blubber samples from biopsies. Tissues were separated before analysis. Ethanol storage may have variable and organism-dependent effects on stable isotope signatures, which are generally greater for  $\delta^{13}\text{C}$  values than for  $\delta^{15}\text{N}$  values (Kaehler and Pakhomov 2001). Ethanol preservation is known to deplete  $\delta^{13}\text{C}$  values of dolphin skin (average depletion of  $-0.48\%$ ; Kiszka et al. 2013). Therefore, we applied this correction factor for  $\delta^{13}\text{C}$  values in skin to run the mixing model. Because lipids are highly depleted in  $\delta^{13}\text{C}$  relative to other tissue components and are a large proportion of the tissues we collected (DeNiro and Epstein 1981; Tieszen et al. 1983), we performed lipid extractions on all samples. For lipid extractions, an aliquot of approximately 100 mg of fine powder was agitated with 4 ml of cyclohexane for 1 h at room temperature; this operation was repeated three times. Next, the sample was centrifuged for 5 min at 4000g, and the supernatant containing lipids was discarded. The sample was dried in an oven at 45 °C for 48 h, and  $0.35 \pm 0.05$  mg subsamples of lipid-free powder were then weighed in tin cups for stable isotope analyses. Stable isotope measurements were performed with a continuous-flow isotope-ratio mass spectrometer (Delta V Advantage, Thermo Scientific, Germany)

coupled to an elemental analyser (Flash EA1112 Thermo Scientific, Italy). Results are expressed in  $\delta$  notation relative to PeeDee Belemnite and atmospheric  $\text{N}_2$  for  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ , respectively, according to the equation:

$$\delta X = (R_{\text{sample}}/R_{\text{standard}} - 1)10^3$$

where X is  $^{13}\text{C}$  or  $^{15}\text{N}$  and R is the isotope ratio  $^{13}\text{C}/^{12}\text{C}$  or  $^{15}\text{N}/^{14}\text{N}$  (Peterson and Fry 1987). Replicate measurements of internal laboratory standards (acetanilide) indicated that measurement errors were  $\pm 0.15$  and  $\pm 0.2\%$  for  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ , respectively. Per cent carbon and nitrogen elemental composition of tissues were obtained using the elemental analyser and used to calculate the sample C/N ratio, indicating good lipid removal efficiency when  $<4$  (Lesage et al. 2010).

#### Data analysis

Variation in  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values among species (i.e. prey and predator) was tested using non-parametric Kruskal–Wallis tests. Mann–Whitney U tests were also used to compare  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values between rainy (November–April) and dry (May–October) seasons, for both skin and blubber tissues. Mixing models were applied with the package SIAR (Stable Isotope Analysis in R; Parnell et al. 2010) to estimate the proportional contribution of potential prey taxa to bottlenose dolphin diets. The mixing model included all but one prey species identified during feeding events, as well as other potential fish prey from representative foraging microhabitats (inner reefs, forereef, seagrass). The fish species selected were pelagic, demersal and benthic species from reef-associated habitats and different trophic levels: *Tylosurus crocodilus* is epipelagic and inhabits waters near reef systems and feeds on pelagic zooplankton; *Mugil cephalus* is demersal and inhabits mud and sand bottoms in the inshore waters and feeds on zooplankton, macroalgae and other benthic organisms; *Mulloidichthys vanicolensis* is demersal on seaward reefs and feeds on small worms and crustaceans; *Siganus argenteus* is demersal and inhabits coastal and inner reef slopes and feeds on algae; *Scarus russelii* is demersal and is found on shallow coastal reefs where it feeds on algae by grazing on coral rubble; *Caranx melampygus* is a demersal and pelagic predator that feeds on small schooling fishes (Randall et al. 1990). The golden trevally *G. speciosus* is not regularly caught by fishermen and could not be included in analyses. It is unlikely that this exclusion dramatically changed mixing model results because this species has similar feeding habits and habitats as *C. melampygus* (Paxton et al. 1989; Smith-Vaniz 1995) and likely is isotopically similar. Therefore, estimate contributions of *C. melampygus* likely represent to combination of this species and *G. speciosus*. trophic enrichment factors (TEFs) or the amount of change in isotope ratios between

diet and consumer tissue (Phillips and Gregg 2003) must be included in mixing models to obtain robust results. Determining TEFs accurately in vertebrates, however, can be particularly challenging because individuals must be held on an isotopically fixed diet in controlled conditions that allow for regular sampling over lengthy periods of time. Furthermore, TEFs are specific to the consumer, tissue and diet (DeNiro and Epstein 1981; Vanderklift and Ponsard 2003), and the use of surrogate or proxy discrimination factors may not be appropriate for species or tissues for which the specific TEF values are unknown (Bond and Diamond 2011). For this reason, we performed three mixing models for skin tissue and one for blubber tissue using different and specific TEFs from the literature (Hobson et al. 1996; Caut et al. 2011; Borrell et al. 2012). Details on TEFs used to run models are summarized in Table 2. SIAR allows incorporating specific TEFs but also the standard deviation for each prey source. This ability to account for variability in the prey mixture is a significant advance in the application of such models to ecological field data (Bond and Diamond 2011). Standard deviations were not available as in Caut et al. (2011), because only one killer whale (*Orcinus orca*) was tested. Therefore, we used the maximum errors measured from the internal laboratory standards (0.2 ‰ for  $\delta^{13}\text{C}$  and 0.15 ‰ for  $\delta^{15}\text{N}$ ).

## Results

### Behavioural observations

From July 2004 to April 2009, a total of 95 groups of bottlenose dolphins were encountered around Mayotte during 224 days of boat surveys. Focal group follows ( $n = 28$ , 25.5 h of active follow) and independent group sightings ( $n = 95$ ) resulted in 54 documented feeding events (Fig. 2 for illustration). Bottlenose dolphin prey was identified, at



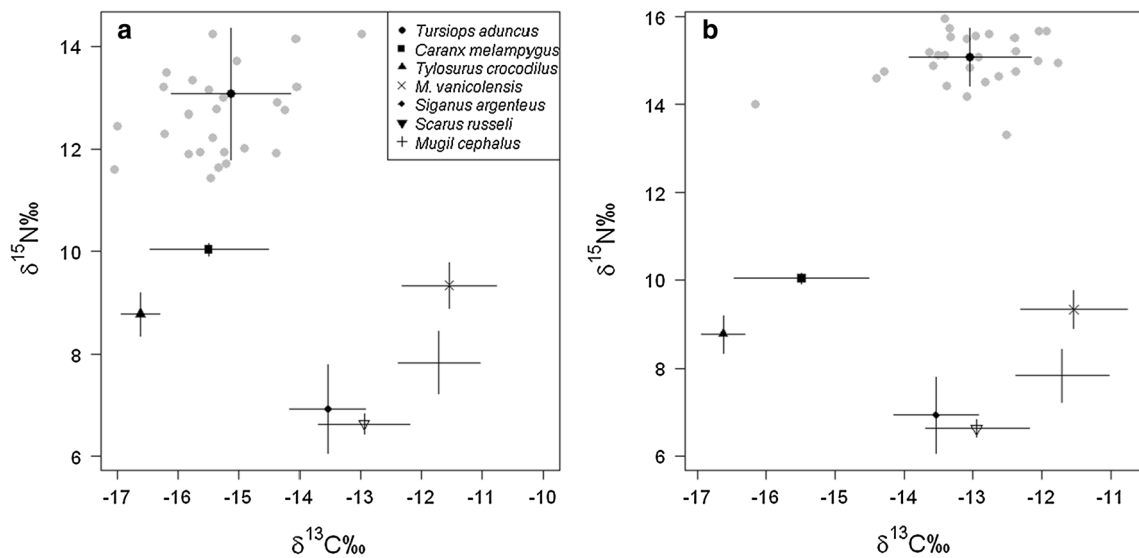
**Fig. 2** Pair of female Indo-Pacific bottlenose dolphins (*Tursiops aduncus*) in the lagoon of Mayotte, handling two large *C. melampygius* (photograph credit: Mathilde Cadier)

least to the family level, for 77 % of feeding events (61 % of events with prey identified observed during the rainy vs. 39 % during the dry season). Two species of Carangidae were identified during 67 % of feeding events where prey were identified with bluefin trevally (*Caranx melampygius*, 82 % of events involving Carangidae) occurring more frequently than golden trevallies (*Gnathanodon speciosus*, 18 %). Other prey species identified included hound needletfish (*Tylosurus crocodilus*, 20 % of feeding events) and mullets (Mugilidae, *Mugil cephalus*, 13 % of feeding events). No seasonal (dry vs. rainy seasons) differences in the relative frequency of prey occurrence could be found. Prey size could not be measured. During one observation of feeding bottlenose dolphins, grey reef sharks (*Carcharhinus amblyrhynchos*) scavenged the remains of several trevallies (*C. melampygius*) caught by a group of 8 dolphins.

### Stable isotope analyses and mixing models

Both skin and blubber of bottlenose dolphins ( $n = 30$ ) had significantly higher  $\delta^{15}\text{N}$  values (Kruskal–Wallis tests; skin:  $H = 46.86$ ,  $df = 6$ ,  $P < 0.001$ , blubber:  $H = 46.87$ ,  $df = 6$ ,  $P < 0.001$ ) than muscle tissue of potential prey species (Fig. 3; Table 1). Blubber isotopic values were higher than skin, for both  $\delta^{15}\text{N}$  (Wilcoxon test:  $W = 93$ ,  $P < 0.001$ ) and  $\delta^{13}\text{C}$  values ( $W = 55.5$ ,  $P < 0.001$ ; Table 1). For  $\delta^{13}\text{C}$ , *M. vanicolensis* had the highest values, while *T. crocodilus* the lowest ( $-11.5$  and  $-16.6$  ‰, respectively). Bottlenose dolphin showed intermediate  $\delta^{13}\text{C}$  values that were significantly different from those of prey species, except for *C. melampygius* and dolphin skin and *S. argenteus* and *S. russeli* compared to dolphin blubber (Post hoc test,  $P > 0.05$ ). Biopsies were collected in both dry ( $n = 12$ ) and ( $n = 18$ ) rainy seasons. No significant isotopic differences were found for either  $\delta^{15}\text{N}$  (skin:  $U = 64$ ,  $P > 0.05$ ; blubber:  $U = 120$ ,  $P > 0.05$ ) or  $\delta^{13}\text{C}$  values (skin:  $U = 99$ ,  $P > 0.05$ ; blubber:  $U = 101$ ,  $P > 0.05$ ).

Mixing model estimates of dietary contributions based on both skin and blubber samples identified *C. melampygius* and *T. crocodilus* as having the highest mean dietary contribution to the diets of bottlenose dolphins (Figs. 4, 5), but their relative estimated contributions varied with model and tissue type. For skin tissue, two of three models (S1, S3) that used different TEFs (Table 2, Fig. 4a, c) suggested that *T. crocodilus* was the most common prey, with a mean estimated contribution of 49.9 % ( $\pm 19.1$ ) to 57.4 % ( $\pm 18.8$ ). *C. melampygius* had the second highest estimated dietary contribution, with 41.9 % ( $\pm 18.1$ ) and 36.3 % ( $\pm 18.7$ ). In contrast, model S2 (Fig. 4a) was more similar to the blubber model with *C. melampygius* contributing a mean estimate of 63.8 % ( $\pm 19.2$ ) of the diet and *T. crocodilus* in second position. For blubber, *C. melampygius* had a mean contribution of 73.7 % ( $\pm 14.9$ ) followed by *T. crocodilus*



**Fig. 3** Carbon ( $\delta^{13}\text{C}$ ) and nitrogen ( $\delta^{15}\text{N}$ ) isotope ratios (mean  $\pm$  SD, ‰) in skin (a) and blubber (b) of Indo-Pacific bottlenose dolphin (*Tursiops aduncus*) (black circle) and in muscle of prey species from Mayotte. All individual points for bottlenose dolphin are shown (grey circles)

**Table 1** Carbon ( $\delta^{13}\text{C}$ ) and nitrogen ( $\delta^{15}\text{N}$ ) isotope ratios (mean  $\pm$  SD, ‰), code of species, number of individuals analysed ( $n$ ) and C/N ratios (mean  $\pm$  SD, ‰) of Indo-Pacific bottlenose dolphin (*Tursiops aduncus*) from Mayotte and its potential prey species

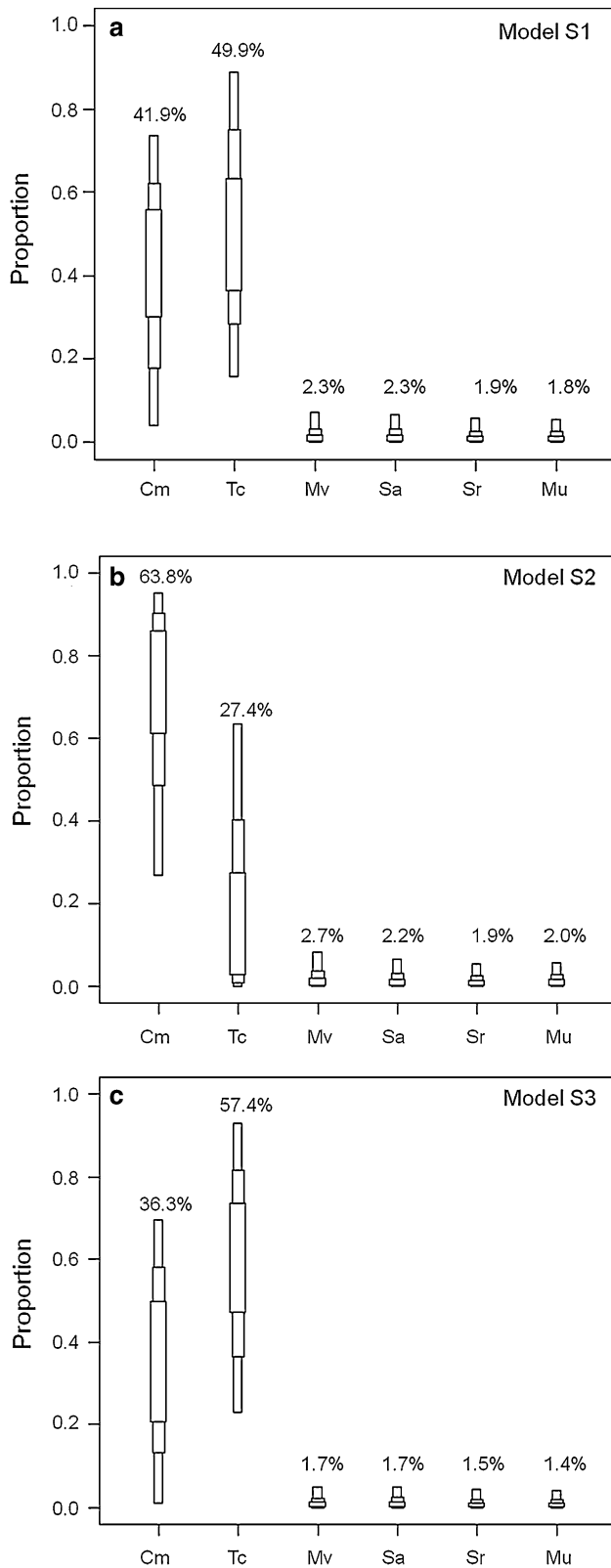
Species	Code	$n$	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$	C:N
Prey					
Bluefin trevally <i>Caranx melampygus</i>	Cm	6	$-15.5 \pm 1.0$	$10.0 \pm 0.1$	$3.09 \pm 0.06$
Hound needlefish <i>Tylosurus crocodilus</i>	Tc	5	$-16.6 \pm 0.3$	$8.8 \pm 0.4$	$3.12 \pm 0.02$
Yellowfin goatfish <i>Mulloidichthys vanicolensis</i>	Mv	5	$-11.5 \pm 0.8$	$9.3 \pm 0.4$	$3.11 \pm 0.02$
Streamlined spinefoot <i>Siganus argenteus</i>	Sa	4	$-13.5 \pm 0.6$	$6.9 \pm 0.9$	$3.14 \pm 0.04$
Russell’s parrotfish <i>Scarus russeli</i>	Sr	5	$-12.9 \pm 0.7$	$6.6 \pm 0.2$	$3.1 \pm 0.01$
Mullet <i>Mugil cephalus</i>	Mc	5	$-11.7 \pm 0.7$	$7.8 \pm 0.6$	$3.2 \pm 0.01$
Predator					
Indo-Pacific bottlenose dolphin <i>Tursiops aduncus</i>					
Skin	TaS	30	$-15.1 \pm 1.0$	$13.1 \pm 1.3$	$3.31 \pm 0.33$
Blubber	TaB	30	$-13.05 \pm 0.9$	$15.1 \pm 0.7$	$2.9 \pm 0.09$

with 16.9 % ( $\pm 12.4$ ; Table 2, Fig. 5). Finally, all mixing models for both tissues suggested that other prey species were less important in the diet of bottlenose dolphins with potential contributions  $<10\%$ , including *M. cephalus*.

**Discussion**

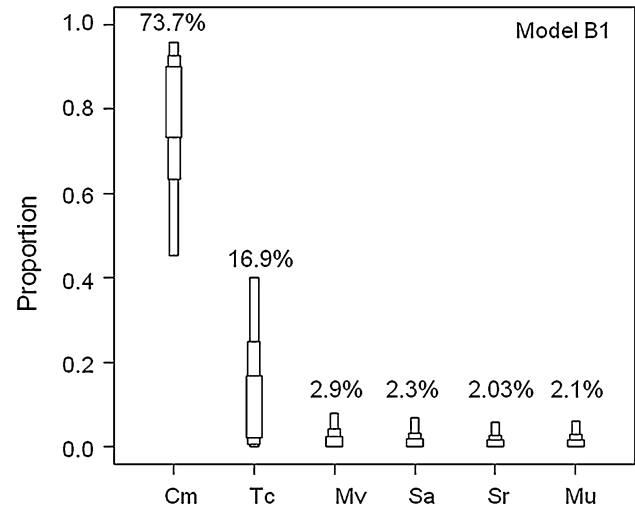
Although mixing models can be useful in elucidating diets, using stable carbon and nitrogen isotopes may be challenging in generalized and opportunistic predators due to

the diversity of possible food sources and consequently stable isotope ratios (Di Benedetto et al. 2011). Thus, the use of complementary methods generally is important for elucidating the feeding ecology of free-ranging cetaceans (Jansen et al. 2012). This study showed the value of using two independent methods in combination to assess the prey preferences of coastal bottlenose dolphins in the lagoon of Mayotte. With few exceptions, there was good agreement between behavioural observations and diets inferred from stable isotope mixing models, suggesting consistent feeding preferences across multiple temporal scales. However,



the relative contribution of prey sources was slightly variable across tissues and the number of prey sources identified varied between methods. Overall, the combination of

**Fig. 4** Results of SIAR mixing models (50, 75 and 95 % credibility intervals) showing the probable sources proportion (%) in diet of Indo-Pacific bottlenose dolphin (*Tursiops aduncus*) from Mayotte, using skin stable isotope ratios (i.e.  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ ). The trophic enrichment factors (TEFs) used to run models and species codes are summarizing in Table 2 (Fig. 4a: Model S1; Fig. 4b: Model S2; Fig. 4c: Model S3)



**Fig. 5** Results of SIAR mixing models (50, 75 and 95 % credibility intervals) showing the probable sources proportion (%) in diet of Indo-Pacific bottlenose dolphin (*Tursiops aduncus*) from Mayotte, using blubber stable isotope ratios (i.e.  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ ) (details as in Fig. 4)

behavioural sampling and isotopic models suggests that bottlenose dolphins primarily feed on high trophic level mesopredators, especially *C. melampygu* and *T. crocodilus*. Behavioural observations suggest that *G. speciosus* and *M. cephalus* are also prey of bottlenose dolphins. *G. speciosus* could not be obtained for this study because they are rarely caught in local fisheries and difficult to target in the field for sampling. Given their similar ecological characteristics and feeding habits, however, it is likely that isotopic values of *G. speciosus* are similar to *C. melampygu*. Therefore, estimated contributions of *C. melampygu* in the mixing model output likely represent a combination of both of these taxa (Paxton et al. 1989; Smith-Vaniz 1995). In contrast, mixing models suggest a very low contribution of *M. cephalus* to dolphin diets, which likely does represent low contributions of this species to assimilated energy. Because no model predicted large contributions of *M. cephalus* to dolphin diets, it is unlikely that inappropriate selection of TEFs (see Table 2) drove this result.

Overall, bottlenose dolphins around Mayotte are essentially epipelagic and demersal predators. *T. crocodilus* is an epipelagic species, inhabiting waters near reef systems that primarily preys upon smaller fish (Randall et al. 1990). *C. melampygu* and *G. speciosus* are pelagic and occasionally

**Table 2** Summary of estimated contributions (mean values) of potential species prey in the diet of Indo-Pacific bottlenose dolphin (*Tursiops aduncus*) from the four different mixing models applied with different trophic enrichment factors (TEFs:  $\Delta^{13}\text{C}$  and  $\Delta^{15}\text{N}$ ) taken in the literature

	Skin			Mean $\pm$ SD	Blubber
	Model S1	Model S2	Model S3		
TEFs	Caut et al. (2011)	Hobson et al. (1996)	Borrell et al. (2012)		Caut et al. (2011)
$\Delta^{13}\text{C}$	2.4 $\pm$ 0.2	2.8 $\pm$ 0.1	1.3 $\pm$ 0.4		2.54 $\pm$ 0.2
$\Delta^{15}\text{N}$	3.2 $\pm$ 0.15	2.3 $\pm$ 0.3	2.8 $\pm$ 0.3		3.15 $\pm$ 0.15
Potential contributions					
Bluefin trevally <i>Caranx melampygus</i>	41.9 $\pm$ 18.1	63.8 $\pm$ 19.2	36.3 $\pm$ 18.7	<b>47.3 <math>\pm</math> 18.7</b>	<b>73.7 <math>\pm</math> 14.9</b>
Hound needlefish <i>Tylosurus crocodilus</i>	49.9 $\pm$ 19.1	27.4 $\pm$ 19.2	57.4 $\pm$ 18.8	<b>44.9 <math>\pm</math> 19.02</b>	<b>16.9 <math>\pm</math> 12.4</b>
Goatfish <i>Mulloidichthys vanicolensis</i>	2.3 $\pm$ 2.3	2.7 $\pm$ 2.7	1.7 $\pm$ 1.6	2.2 $\pm$ 2.2	2.9 $\pm$ 2.6
Streamlined spinefoot <i>Siganus argenteus</i>	2.3 $\pm$ 2.1	2.2 $\pm$ 2.2	1.7 $\pm$ 1.6	2.1 $\pm$ 1.98	2.3 $\pm$ 2.3
Russell's parrotfish <i>Scarus russeli</i>	1.9 $\pm$ 1.8	1.9 $\pm$ 1.8	1.5 $\pm$ 1.4	1.8 $\pm$ 1.7	2.03 $\pm$ 1.9
Mullet <i>Mugil cephalus</i>	1.8 $\pm$ 1.8	2.0 $\pm$ 1.9	1.4 $\pm$ 1.3	1.7 $\pm$ 1.7	2.1 $\pm$ 2.0

The mean values of prey species contributing on average to more than 10 % in the diet of bottlenose dolphin from Mayotte are in bold

benthic predators feeding on small fish, both in the coastal waters and over reef banks outside the inner lagoon (Paxton et al. 1989; Smith-Vaniz 1995). *M. cephalus* is typically an inshore species occurring over soft muddy bottoms off mangroves and in embayments (Harrison 1995). We observed more prey types during behavioural observations than might be expected based upon isotopic mixing models. Moreover, a direct comparison between the isotopic data of bottlenose dolphins and their potential food sources (after the values have been corrected, using any TEFs) showed that the isotopic values of the predator do not get inside the prey polygon, between the isotopic values of their prey. This could be due to not including enough potential prey in mixing models, but also because we observed predation events on species that make up small components of overall diets (e.g. *M. cephalus*). Indeed, we found that dolphins forage heavily on large epipelagic prey that are easily detected from the surface by the observer and can make up a large portion of assimilated biomass. The utility of behavioural observations for determining diets in Mayotte may not be applicable to other locations. Indeed, in many regions, dolphins forage in a number of habitats, or at times, that are difficult to observe. In Mayotte, the water visibility and sighting conditions are excellent. Therefore, behavioural sampling appears to be a valuable method to investigate the foraging ecology of coastal dolphins that inhabit similarly clear waters (Barros and Wells 1998; Herzog and Elliser 2013).

Despite overall agreement, there were slightly different predictions about dietary contributions of prey taxa based

on analyses of skin and blubber. Regardless of the model used, blubber suggested greater contributions of *C. melampygus* than skin did. This could reflect long-term variation in trophic interactions. The turnover rate of skin in the common bottlenose dolphin (*T. truncatus*) is 2–3 months, from the basal lamina to the outermost surface (Hicks et al. 1985). Blubber turnover rate is not well known, but is significantly longer, integrating diet over several months (Abend and Smith 1995). Thus, over longer time scales, *C. melampygus* may make up a greater portion of dolphin diets than they had during the months preceding sampling that are reflected in skin samples. Alternatively, differences among tissues could be due to inaccuracies in the TEFs used in models. Indeed, the use of accurate, species- and diet-specific TEFs is of great importance for using stable isotope mixing models to predict the dietary sources of a consumer (Newsome et al. 2010; Caut et al. 2011; Borrell et al. 2012). Unfortunately, marine mammal TEFs, particularly cetaceans, remain poorly investigated. We attempted to overcome this difficulty by applying multiple assumptions about TEFs, all of which showed general agreement.

Bottlenose dolphins are well known for their dietary flexibility, and populations around the world can range from highly generalized to relatively specialized (Barros and Wells 1998; Mann and Sargeant 2003; Krützen et al. 2005; Sargeant et al. 2007). For example, off the coast of Zanzibar (Tanzania, East Africa), *T. aduncus* prey on a diversity of taxa (Amir et al. 2005) with stomach content analyses documenting 50 species of bony fish and 3 species of squids. Small- and medium-sized neritic fish and cephalopods

contributed substantially to the diet (Amir et al. 2005). Foraging appears to occur over reefs or soft bottoms close to shore. In contrast around Mayotte, most bottlenose dolphins specialize to a greater degree and forage on prey at higher trophic levels. In still other populations, there is specialization within individuals but considerable diversity in prey at the population level. For example, Indian Ocean bottlenose dolphins (*Tursiops cf. aduncus*) in Shark Bay (Western Australia) show high level of flexibility in feeding tactics and resource use, some individuals consuming large predatory fish and others consuming smaller taxa (Mann and Sargeant 2003). Further comparative studies would provide interesting insights into the drivers of specialization (e.g. prey availability, habitat structure, predation risk, population density) at population and individual levels.

Our study also highlighted the utility of biopsies as a minimally invasive approach (Kiszka et al. 2010b) for assessing cetacean feeding ecology when carcasses are unavailable for stomach content analyses. Therefore, in the absence of bottlenose dolphin stomach contents, which provide the most fine scale resolution of prey taxa (Di Benedetto et al. 2011; Dunshea et al. 2013), combining observational and isotopic evidence appears to constitute a powerful approach to investigate the feeding ecology of coastal bottlenose dolphins.

Finally, our results also have implications for management of bottlenose dolphins and their habitats in the lagoon of Mayotte. For example, conflict with existing fisheries is likely to be minimal. Carangids, the primary prey of dolphins, only constitute 7 % of total fishing landings, and hound needlefish are rarely caught (Herfaut 2006). There is, however, increasing development of recreational fisheries targeting trevallies and other large pelagic fishes around Mayotte which could compete with dolphins. Therefore, monitoring recreational fisheries will be important in the future to ensure the sustainability of this economically important activity as well as to ensure that it will not constitute an additional pressure on endangered Mayotte bottlenose dolphins.

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