



REPORTS

Tuna and Dolphin Associations in the North-east Atlantic: Evidence of Different Ecological Niches from Stable Isotope and Heavy Metal Measurements

K. DAS†*, G. LEPOINT†, V. LOIZEAU‡, V. DEBACKER†, P. DAUBY† and J. M. BOUQUEGNEAU†

†Department of Oceanology, University of Liège, B6 Sart-Tilman, B-4000 Liège, Belgium

‡DEL/CCM, Centre IFREMER de Brest, BP 70, 29280 Plouzane, France

Associations of tunas and dolphins in the wild are quite frequent events and the question arises how predators requiring similar diet in the same habitat share their environmental resources. As isotopic composition of an animal is related to that of its preys, stable isotope ($^{13}\text{C}/^{12}\text{C}$ and $^{15}\text{N}/^{14}\text{N}$) analyses were performed in three predator species from the North-east Atlantic: the striped dolphin, *Stenella coeruleoalba*, the common dolphin *Delphinus delphis* and the albacore tuna, *Thunnus alalunga*, and compared to their previously described stomach content. Heavy metals (Cd, Zn, Cu and Fe) are mainly transferred through the diet and so, have been determined in the tissues of the animals. Tuna muscles display higher $\delta^{15}\text{N}$ than in common and striped dolphins (mean: 11.4 vs. 10.3‰ and 10.4‰, respectively) which reflects their higher trophic level nutrition. Higher $\delta^{13}\text{C}$ are found in common (−18.4‰) and striped dolphin (−18.1‰) muscles than in albacore tuna (−19.3‰) probably in relation with its migratory pattern. The most striking feature is the presence of two levels of cadmium concentrations in the livers of the tunas (32 mg kg^{−1} dry weight (DW) vs. 5 mg kg^{−1} DW). These two groups also differ by their iron concentrations and their $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ liver values. These results suggest that in the Biscay Bay, tunas occupy two different ecological niches probably based on different squid inputs in their diet. © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: marine mammals; tunas; stable isotopes; heavy metals; food webs; North-east Atlantic.

Introduction

Small pelagic dolphins are commonly associated with tunas in mixed-species aggregations. These polyspecific associations have been extensively studied in the tropical Pacific (Perrin *et al.*, 1973; Au and Pitman, 1986; Au, 1991; Scott and Cattanach, 1998) and comprise several species, like tunas, marine mammals, seabirds or sharks. In general, these polyspecific aggregations seem to form when social species of similar foraging join to form larger groups to increase feeding success and to better avoid predators (Scott and Cattanach, 1998). These animals are known to feed, interact and travel together for various periods of time (Au, 1991). In the feeding aggregations observed in tropical waters, prey is driven to the surface by yellowfin tunas and is abundant and diverse enough to allow dolphins and other species like seabirds and sharks to feed at the same time (Au and Pitman, 1986). In the North-east Atlantic three predator species are often caught together in the same nets during commercial seining operations: the albacore tuna (*Thunnus alalunga*), the striped dolphin (*Stenella coeruleoalba*) and the common dolphin (*Delphinus delphis*). As they are known to feed together, the question arises about the possible competition or commensal relationship that might occur between them. These kinds of alimentary studies are often performed by stomach content analysis (Perrin *et al.*, 1973; Hassani *et al.*, 1997) or field observations (Au and Pitman, 1986). However, these useful data reflect the most recent meal and may not be representative of the overall diet. In marine mammals, the use of naturally occurring stable isotopes of carbon and nitrogen has provided complementary data to their feeding ecology (Hobson and Welch, 1992; Abend and Smith, 1995; Smith *et al.*, 1996; Hobson *et al.*, 1997; Burns *et al.*, 1998). Stable isotope analysis has emerged as a powerful tool to trace diet as isotope

*Corresponding author.

E-mail address: krishna.das@student.ulg.ac.be

ratios of a consumer are related to those of their preys (DeNiro and Epstein, 1978, 1981; Peterson and Fry, 1987). Indeed, the carbon and nitrogen isotope values ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) differ between organisms and their diets because of a slight selective retention of the heavier isotope and excretion of the lighter one. As a result, organisms have a higher δ value than their diet. Nitrogen-15 typically shows a stepwise increase with trophic level within a food chain (Hobson and Welch, 1992; Cabana and Rasmussen, 1994; Thompson *et al.*, 1995) with a trophic enrichment value of about 3‰ (Hobson and Welch, 1992; Michener and Schell, 1994). On the contrary, an animal has $\delta^{13}\text{C}$ values close to that of its diet. (DeNiro and Epstein, 1978; Hobson and Clark, 1992; Thompson *et al.*, 1995; Smith *et al.*, 1996). Thus, carbon-13, rather than being a reliable indicator of the trophic level, is generally used to indicate relative contributions to the diet of different potential primary sources in a trophic network (Rau *et al.*, 1992; Havelange *et al.*, 1997; Dauby *et al.*, 1998), indicating for example the aquatic vs. terrestrial, inshore vs. offshore or pelagic vs. benthic contribution to food intake (Hobson *et al.*, 1995; Smith *et al.*, 1996).

Since stable isotopes ratios of consumers' tissues are derived from assimilated food, the tissue reflects dietary input integrated over time, not just the last food intake. The metabolic rates and the subsequent turn-over of elements differ between liver and muscle, allowing the evaluation of diet integrated between different periods of time (e.g., days, weeks or months) (Tieszen *et al.*, 1983; Hobson *et al.*, 1996, 1997).

We had the opportunity to use samples from three species from the North-east Atlantic: the striped dolphin, the common dolphin and the albacore tuna, whose stomach contents have been previously determined by Hassani *et al.* (1997). In order to contribute to the knowledge of ecological aspects of polyspecific associations, we have studied their isotopic composition ($\delta^{15}\text{N}$ and $\delta^{13}\text{C}$). Combined with the isotopic ratio determinations, heavy metal analyses have been performed in the tissues of the three predators. Heavy metals are merely transferred through the diet. Indeed, heavy metal levels found in marine organisms depend not only on the environment contamination but also on several other ecological or physiological factors (Bouquegneau and Joiris, 1988; André *et al.*, 1990a,b; Caurant *et al.*, 1994), among which the diet and the position in the trophic web are determining elements (Bouquegneau and Joiris, 1992; Cabana and Rasmussen, 1994; Kidd *et al.*, 1995; Stewart *et al.*, 1997).

By using a combination of stable isotope and heavy metal analyses, our aim is to compare the diet and the position in the trophic web of three predators from the North-east Atlantic, the striped dolphin, the common dolphin and the albacore tuna and to contrast our results with stomach content analysis previously described by Hassani *et al.* (1997).

Materials and Methods

Collection and storage

The 23 striped dolphins (*S. coeruleoalba*) (10 females and 13 males) and 10 common dolphins (*D. delphis*) (5 females and 5 males) used in this study have originated from the Bay of Biscay in the North-east Atlantic. The locations of the captures covered an area between 46° and 50° North and 9°–20° West. They were accidentally captured during commercial tuna seining operations from May to September 1993, from which 20 albacore tuna fishes (*T. alalunga*) (9 females and 11 males) were collected. This collection was conducted by the *Institut Français de Recherche pour l'Exploitation de la Mer (IFREMER)* from Brest (France) to assess ecological risk linked to the North-east Atlantic French drift-net activity (Goujon *et al.*, 1993; Goujon, 1996).

Tissue samplings were performed at the fishing site and frozen immediately in liquid nitrogen and then kept in a freezer (−20°C) until storage in Oceanology Laboratory (Liège, Belgium).

Dolphin sex and age determinations were realized by Collet (1993a,b), who showed an exponential length-age relationship (see also Di-Méglio *et al.*, 1996). The ages of dolphins were estimated by counting tooth growth layer groups (GLGs). The total lengths from the snout to fluke notch were determined for each individual by Goujon (1996). All tuna specimens are juveniles (4–5 years old) with length ranging from 75 to 81 cm while the ages of the dolphins range from 0.1 to 20 years old (Collet, 1993a,b).

Analytical methods

Zn, Cd, Fe and Cu analyses. After being weighed and dried for 48 h at 110°C, samples were digested with a mixed solution of hydrochloric (Merck 317) and nitric (Merck 456) acids (1:3, v:v) and slowly heated to 100°C until complete digestion. Atomic absorption spectrophotometry (ARL 3510) was used to determine heavy metal concentrations (Cu, Zn, Cd, Fe). Pb, Ni, Cr contents were also determined but the results most often were below the detection limits (unpublished data). Concentrations are expressed as mg kg^{−1} dry weight (DW).

Parallel to the samples, a set of certified material samples (CRM 278 Community Bureau of Reference, Commission of the European Communities) was also analysed to ensure the method's sensitivity. Recoveries ranged from 92% to 102% for Cu, Zn, and Fe and 88% for Cd. Limits of detection were 0.01 µg g^{−1} DW for Cu, 0.33 for Zn, and 0.22 for Cd.

Stable isotope measurements. Organisms may vary in their concentrations of lipids. As lipids have been shown to be depleted in ¹³C relatively to the diet (Tieszen *et al.*, 1983), they were extracted from samples using repeated rinses with 2:1 chloroform: methanol prior to analysis. After drying at 50°C (48 h), samples were grounded into

a homogeneous powder. Carbon dioxide and nitrogen gas were analysed on an V.G. Optima (Micromass) IR-MS coupled to an N-C-S elemental analyser (Carlo Erba). Routine measurements are precise to within 0.5‰ for carbon-13 and 0.6‰ for nitrogen-15. Stable isotope ratios were expressed in δ notation according to the following

$$\delta X = [(R_{\text{sample}}/R_{\text{standard}}) - 1] \times 1000,$$

where X is ^{13}C or ^{15}N and R is the corresponding ratio $^{13}\text{C}/^{12}\text{C}$ or $^{15}\text{N}/^{14}\text{N}$.

Carbon and nitrogen ratios are expressed relative to the Vienna Pee Dee Belemnite (VPDB) standard and to atmospheric nitrogen, respectively.

Data treatment

Parametric and non-parametric tests were used to compare different groups: Kolmogorov–Smirnov test was used to assume the normality of the data. ANOVA followed by post-hoc multiple comparison tests have been used to compare the data between the different species. When the necessary assumptions to realize ANOVA were not gathered (normality of the variables and homogeneity of variances), Kruskal–Wallis was used followed by multiple comparisons based on the Kruskal–Wallis rank sums to test for pairwise differences among species. Mann–Whitney U test was performed to compare differences among sexes or groups when variances were not homogenous. Spearman coefficient was used to test correlations between the values. Results were judged significant when $p < 0.01$ (Scherrer, 1984).

Results

Metal level in the tissues

In dolphin and tuna tissues, Zn, Cd and Cu and Fe mean concentrations are higher in liver than in muscle (Table 1). In dolphins, Cd displays the highest mean concentration in kidney. Fe is always higher in the blood due to the hemoglobin respiratory pigment. In both mammal gland and testis, iron and zinc were more concentrated than copper and cadmium. Zn concentrations in the liver are similar for the three species (ANOVA, $F = 1.6$, $p > 0.05$). Hepatic copper levels are not significantly different for the two dolphin species but tunas display lower copper concentrations than common dolphins (ANOVA: $F = 3.07$, $p < 0.01$). Hepatic iron concentrations in striped dolphins are consistently greater than those in common dolphins, which in turn are greater than those in albacore tunas (ANOVA: $F = 0.94$, $p = 0.04$). Cadmium concentrations in the livers are not significantly different for the three species (ANOVA: $F = 0.3$, $p > 0.05$). The standard deviations of cadmium mean concentrations both in kidney and livers of dolphins (Table 1) are very large. Metal levels are similar for both genders in the three species (data not shown), but a significant increase of this metal level with

length of individuals appears in the livers of both striped and common dolphins (Fig. 1). Such a relationship was not found between the size of the tunas and their hepatic cadmium concentrations (Fig. 2). The most striking feature of Fig. 2 is that the tuna group is heterogeneous. One group of tunas is heavily contaminated by cadmium (mean $32 \text{ mg kg}^{-1} \text{ DW}$) while the other one displays lower cadmium levels (mean: $5 \text{ mg kg}^{-1} \text{ DW}$; Mann–Whitney U test: $p < 0.0005$). Zinc and copper are similar for both groups while iron, $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ are significantly different (Table 2). In the muscles, there are no significant differences between the two groups of tunas where cadmium concentrations were always under the detection limit.

When the three species are compared together (Table 1), tunas display lower zinc and copper concentrations in muscles than the two dolphin species (Kruskal–Wallis: $p < 0.001$ and ANOVA: $F = 1.4$, $p < 0.001$, respectively).

Stable Isotope Analyses

Isotopic compositions are similar for both sexes in the three species and no effect of age has been observed ($p > 0.05$, data not shown).

Tunas display significantly higher $\delta^{15}\text{N}$ in muscles than striped and common dolphins: 11.4‰ vs. 10.4‰ and 10.3‰ , respectively (ANOVA: $F = 3.3$, $p = 0.00002$ and $p = 0.0001$, respectively) (Table 1). The mean $\delta^{15}\text{N}$ values in muscles do not vary significantly between the dolphin species: $\delta^{15}\text{N}$ of $+10.3\text{‰}$ in the muscles of common dolphins and $+10.4\text{‰}$ in the muscles of striped dolphins (ANOVA: $p > 0.05$).

Tunas display a muscle $\delta^{13}\text{C}$ significantly lower than dolphins (-19.3‰ vs. -18.1‰ and -18.4‰ for striped and common dolphins respectively (ANOVA: $F = 0.6$, $p = 0$ and $p = 0.001$, respectively).

Individual results of $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values in muscles and livers are presented in Figs. 3 and 4. In the muscles, specific regrouping appears: tunas are completely isolated from dolphins with a higher $\delta^{15}\text{N}$ and a lower $\delta^{13}\text{C}$ (Fig. 3). The common dolphin data are also concentrated in a small area of the figure (medium $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values) while the striped dolphin displays a wide range of $\delta^{15}\text{N}$ values overlapping the two other species.

In livers, no regrouping between individuals from the same species occurs (Fig. 4). However, tunas with low cadmium contamination display lower $\delta^{15}\text{N}$ values than contaminated tunas (ANOVA: $p = 0.000016$), striped and common dolphins (ANOVA: $p = 0.02$ and $p < 0.001$, respectively; Table 2, Figs. 5 and 6). Common dolphins display medium $\delta^{15}\text{N}$ mean values between striped dolphins and tunas with high cadmium levels, the difference between these latest being significant (ANOVA, $F = 1.17$, $p = 0.002$). $\delta^{13}\text{C}$ mean values in the livers are significantly lower for tunas with low cadmium levels compared to other tunas (ANOVA: $p = 0.02$), striped dolphins ($p < 0.0005$) and common dolphins ($p < 0.002$). The $\delta^{13}\text{C}$ mean values are similar for tunas

TABLE 1

Trace elements concentrations (mg kg⁻¹DW) and isotopic composition of three main predators (*Stenella coeruleoalba*, *Delphinus delphis*, *Thunnus alalunga*) of the North-east Atlantic expressed as a mean $\pm$ standard deviation, range of concentrations (minimum–maximum); *N*: number of samples; nd: non-determined.

		<i>N</i>	Zn	Cd	Cu	Fe	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$
<i>Stenella coeruleoalba</i>	Liver	22	167 $\pm$ 7.8 (33–385)	17 $\pm$ 15 (0.2–51)	43 $\pm$ 54 (7–272)	974 $\pm$ 363 (393–1792)	–17.8 $\pm$ 0.5 (–18.9/–17.08)	10.8 $\pm$ 0.6 (9.4–11.9)
	Muscle	23	41 $\pm$ 16 (27–101)	0.37 $\pm$ 1 (<0.05–5)	6 $\pm$ 1.4 (1–9)	577 $\pm$ 221 (96–1131)	–18.1 $\pm$ 0.5 (–19.2/–17.2)	10.4 $\pm$ 0.75 (8.8–12.1)
	Kidney	23	113 $\pm$ 24 (76–168)	91 $\pm$ 57 (0.1–199)	19 $\pm$ 6 (9–30)	716 $\pm$ 226 (413–1413)	nd	nd
	Mammal gland	4	71 $\pm$ 30 (47–115)	1.3 $\pm$ 0.9 (0.4–2)	4.5 $\pm$ 1 (3–6)	273 $\pm$ 69 (187–344)	nd	nd
	Testis	2	45–157	0.6–3.2	3–3.2	120–215	nd	nd
	Blood	12	23 $\pm$ 12 (12–52)	0.65 $\pm$ 0.7 (<0.05–2.5)	2.8 $\pm$ 1.3 (1.5–6)	2510 $\pm$ 264 (2034–2972)	nd	nd
<i>Delphinus delphis</i>	Liver	10	153 $\pm$ 27 (117–197)	15 $\pm$ 17 (0.4–58)	32 $\pm$ 12 (19–52)	706 $\pm$ 227 (326–1096)	–17.8 $\pm$ 0.5 (–18.9/–17.3)	11.1 $\pm$ 0.4 (10.4–11.8)
	Muscle	10	50 $\pm$ 46 (27–181)	0.1 $\pm$ 0.2 (<0.05–0.5)	5 $\pm$ 1 (3–7)	470 $\pm$ 95 (278–560)	–18.4 $\pm$ 0.5 (–19.1/–17.7)	10.3 $\pm$ 0.3 (9.8–10.8)
	Kidney	10	109 $\pm$ 21 (86–146)	71 $\pm$ 61 (3–169)	20 $\pm$ 8 (27–181)	707 $\pm$ 224 (396–1032)	nd	nd
	Mammal gland	4	74 $\pm$ 17 (56–97)	0.6 $\pm$ 0.9 (<0.09–1.9)	3.5 $\pm$ 1.4 (2–5)	188 $\pm$ 90 (110–305)	nd	nd
	Testis	1	99	3.1	4.2	278	nd	nd
	Blood	8	21 $\pm$ 9 (13–40)	1 $\pm$ 1 (<0.07–4)	4 $\pm$ 2 (2–9)	2289 $\pm$ 504 (1153–2729)	nd	nd
<i>Thunnus alalunga</i>	Liver	20	152 $\pm$ 46 (107–265)	17 $\pm$ 14 (3–41)	12 $\pm$ 6 (5–29)	429 $\pm$ 343 (101–1391)	–18.4 $\pm$ 0.8 (–19.8/–16.9)	10.8 $\pm$ 0.8 (9.4–12.7)
	Muscle	12	16 $\pm$ 3 (12–21)	<0.07	1 $\pm$ 0.6 (0.5–3)	49 $\pm$ 10 (28–65)	–19.3 $\pm$ 0.6 (–20.4/–18.5)	11.4 $\pm$ 0.4 (10.9–12.2)

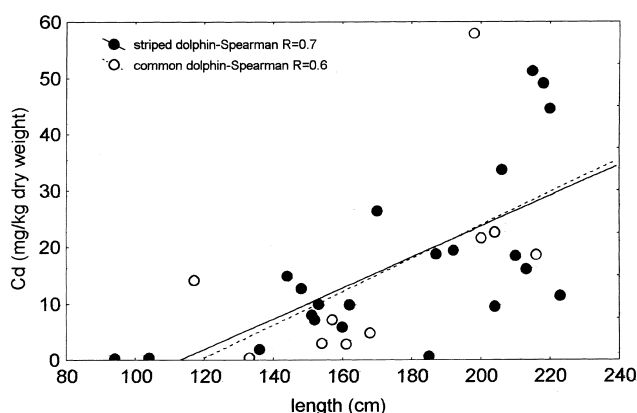


Fig. 1 Relationship between the size of the dolphins and cadmium concentrations in livers.

contaminated by Cd, striped and common dolphins ($p > 0.05$).

Discussion

When species require similar resources in the same habitat, as striped and common dolphins and albacore do in the Biscay Bay, they tend to reduce the competition by occupying different physical locations or feeding on different preys, as it has already been observed for other small odontocetes (Gowans and Whitehead, 1995). A comparative dietary study of albacore and both dolphin species from North-east Atlantic has been re-

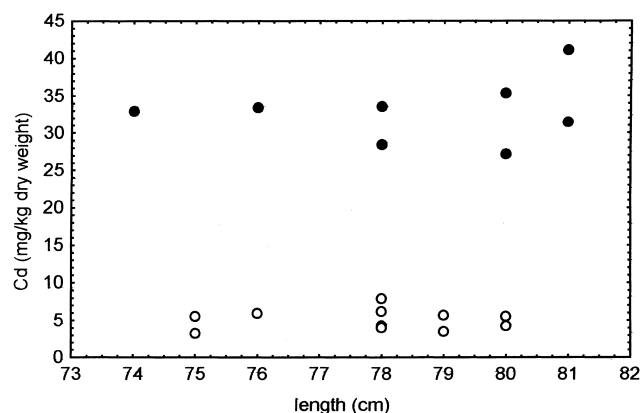


Fig. 2 Relationship between the cadmium concentrations in the liver and the length of the tunas. The black dots are the cadmium contaminated group.

alized and published by Hassani *et al.* (1997). These authors have shown that common and striped dolphins have a more preferential cephalopod diet. Fish ranked second in the food of the common dolphin and only third in the striped dolphin, in which crustaceans were more important in relative abundance. Fish present in the dolphin stomach were mostly small mesopelagic fish as Myctophidae or Sternoptychidae. The food of albacore was mainly fish (mostly the small Sternoptychidae *Maurolicus muelleri*) and to a lesser extent, crustaceans (Hassani *et al.*, 1997). However, the common dolphin samples included all the fish species found in the diet of

TABLE 2

The Zn, Cu, Cd, Fe, $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values in the livers of the two groups of tunas.

	Cadmium contaminated group $N = 11$	Low cadmium contaminated group $N = 9$
Zn	159 ± 4	146 ± 41
Cu	13 ± 5	10 ± 7
Cd*	32 ± 4	5 ± 1
Fe*	722 ± 312	189 ± 72
$\delta^{15}\text{N}^*$	11.5 ± 0.6	10.2 ± 0.5
$\delta^{13}\text{C}^{**}$	-18.1 ± 0.9	-18.7 ± 0.5

*Significantly different with $p < 0.001$ (Mann-Whitney U test).

** $p = 0.05$.

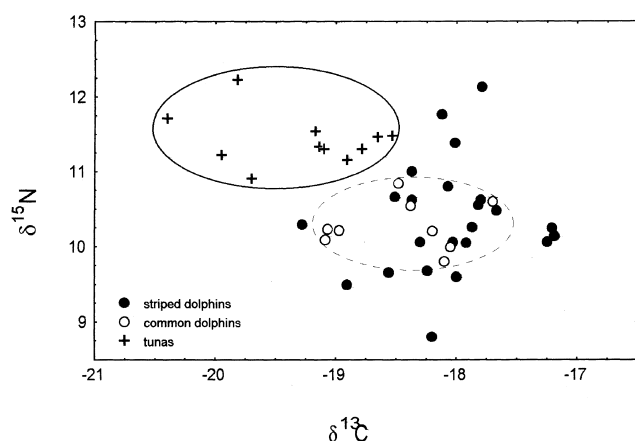


Fig. 3 The $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ stable isotope values in the muscles of tunas, striped and common dolphins.

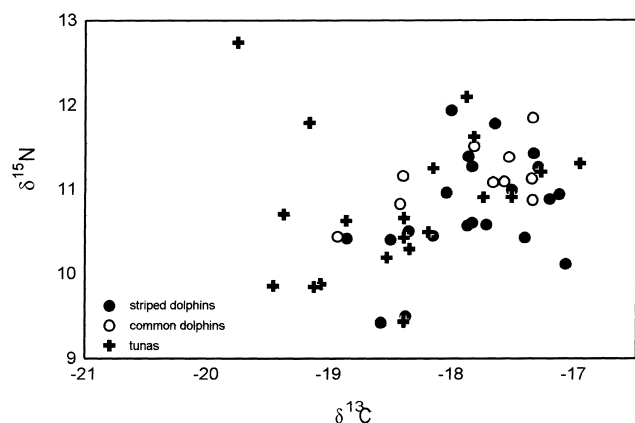


Fig. 4 The $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ stable isotope values in the livers of tunas, striped and common dolphins.

the albacore whereas only one species of prey was found in both striped dolphin and tuna stomachs. Each species seems to have preferential preys but it has been demonstrated that striped and common dolphins are quite opportunistic feeders and can take advantage of seasonally or locally abundant preys (Würtz and Marrale, 1993; Blanco *et al.*, 1995; Young and Cockcroft, 1995). For example, in the Northwest Atlantic, the diet of common dolphins consists mainly of longer sized fish as

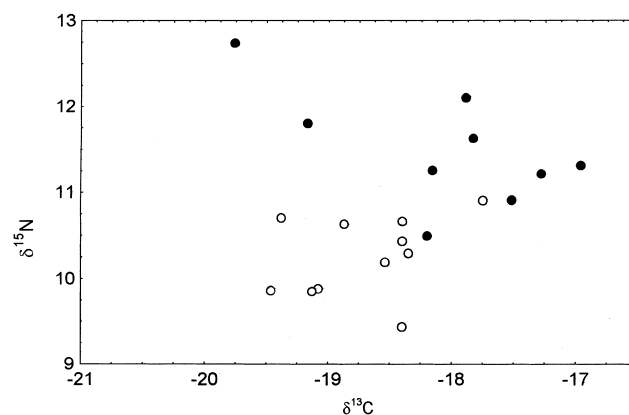


Fig. 5 Comparison of the stable isotope values in livers between highly (●) and low-contaminated groups of tunas (○).

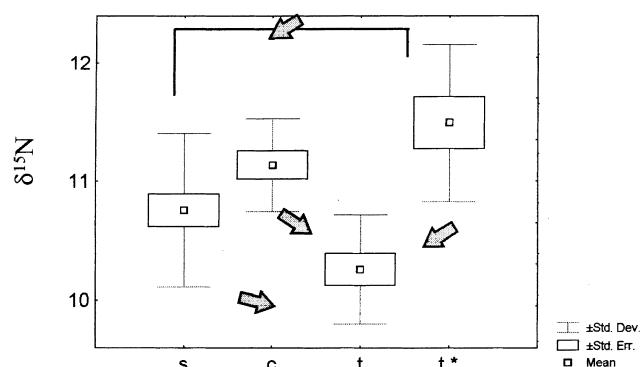


Fig. 6 Comparison of $\delta^{15}\text{N}$ in livers of striped dolphins (s), common dolphins (c), tunas contaminated by cadmium (t^*) and other tunas (t). Arrows indicate significant decreasing difference between groups (ANOVA).

capelins (*Mallotus villosus*) and mackerels (*Scomber scombrus*) (Overholtz and Waring, 1991; Ostrom *et al.*, 1993). Isotopic data on delphinids are scattered (Hobson and Welch, 1992; Ostrom *et al.*, 1993). Ostrom *et al.* (1993) have found in the muscles of one common dolphin from the Western North Atlantic a $\delta^{15}\text{N}$ and a $\delta^{13}\text{C}$ of $+14.8\text{‰}$ and -17.8‰ , respectively). Our isotope values in muscles of the common dolphin are low compared to these data. These high $\delta^{15}\text{N}$ values measured by these authors are consistent with a higher trophic level and the piscivorous diet of common dolphin in the studied geographical area (Ostrom *et al.*, 1993).

Our isotopic data on muscles which integrate a long-term nutrition, also suggest a specific diet: tunas are completely isolated from the other two species with a higher $\delta^{15}\text{N}$ which reflects their higher trophic level nutrition compared with dolphins (Fig. 3). The common dolphin data are also concentrated in a small area of the figure while the striped dolphin displays a wide range of $\delta^{15}\text{N}$. This large dispersion of the individual data may reflect more opportunistic trophic habits of striped dolphins compared to the common dolphin.

The trophic position of the tuna is rather uncommon because generally, in stable isotopes studies, marine

mammals display the highest $\delta^{15}\text{N}$ (Hobson and Welch, 1992; Ostrom *et al.*, 1993; Hobson *et al.*, 1995). Moreover, tunas exhibit lower $\delta^{13}\text{C}$ values than both dolphin species. Both $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values in muscles indicate that different sources of food take part in the diet of tunas compared to striped and especially common dolphins despite a substantial overlap in diet. Tunas present a strong migratory cycle: during the spring, they are found offshore from the Azores and then migrate within the Bay of Biscay during May to August (quoted by Goujon, 1996). Measured lower $\delta^{13}\text{C}$ data presumably reflect a greater reliance on offshore food in relation with this migration. Stable carbon isotopes have proven most useful in identifying where particular organisms feed, and $\delta^{13}\text{C}$ values are typically higher in coastal or benthic food webs than in pelagic food webs (Hobson *et al.*, 1995, 1997).

The considerable overlap in values among the three species and the absence of specific regrouping in the livers (Fig. 4) indicate isotopic similarity among their respective recent diets.

From an ecotoxicological point of view, animals feeding on cephalopods tend to concentrate higher cadmium content (Bouquegneau and Joiris, 1992; Caurant *et al.*, 1994; Bouquegneau *et al.*, 1997). Indeed, cephalopods have been shown to concentrate high level of this metal (Law *et al.*, 1997). Therefore, it is quite striking to find similar cadmium levels in the livers of the three species as very few squids are found in the tuna diet (Hassani *et al.*, 1997). However when we go through individual tuna results (Fig. 2 and Table 2), it appears that there are two levels of iron and cadmium in the livers of tunas: one group is highly contaminated by cadmium as the other one displays low levels. There is no relationship between the size of the tunas and the hepatic cadmium concentrations. On the contrary, there is an increase of hepatic cadmium concentrations of both dolphin species with length (Fig. 1), which reflects a constant intake of cadmium contaminated preys associated with a long half-time of elimination of the pollutant (10–30 yr as quoted by Wagemann *et al.*, 1990). The absence of such a correlation in the tunas suggests that the high hepatic cadmium content might not reflect a chronic contamination but be the result of a punctual ingestion of cadmium contaminated preys. These results are enhanced by $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ differences in the livers of tunas (Fig. 5 and Table 2). It clearly appears that isotopic composition differs strongly between the cadmium contaminated group of tunas and the second one, indicating that two different food habits were co-existing sympatrically and temporally. Differences in iron concentrations between the two groups of tunas are still to be elucidated. Cadmium levels and isotope ratios in muscles are similar in the two groups of tunas suggesting that these feeding habits might be relatively recent.

Despite an apparent large overlap in stable isotope values in livers of the three species (Fig. 4), different

groups can be pointed out (Fig. 6). Tunas with low cadmium contamination display lower $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ mean values than other tunas and both dolphins, suggesting a greater contribution of lower trophic level preys in their diet. On the contrary, common dolphins and tunas contaminated by Cd display similar $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values in their liver confirming that tunas share more prey similarities with common dolphins than striped dolphins (Hassani *et al.*, 1997). In fact, common dolphins seem to occupy a medium trophic position between striped dolphins and tunas with high Cd levels (Fig. 6). These observed differences combined with cadmium concentrations in livers probably result from differential reliance on squids. Our observations are enhanced by stomach analysis studies carried out by Hassani *et al.* (1997) who have suggested that the diet of the tuna may differ: when tuna was captured alone or in the same nets as common dolphins, fish was predominant in the stomach as described earlier. However, these authors have noticed that, when tunas were captured in the same nets as striped dolphins, their stomach contents became similar to dolphins with a high squid content suggesting that feeding behaviour of albacore may differ when it co-occurs with striped dolphin. On the contrary, the presence of common dolphins would have no influence on the feeding habits of tunas (Hassani *et al.*, 1997).

$\delta^{15}\text{N}$, $\delta^{13}\text{C}$, iron and cadmium also suggest different feeding habits between these three predators. While striped and common dolphins seem to differ little in their diet, tunas seem to occupy two potential different ecological niches in the Bay of Biscay. These different niches are based on at least two types of preys, influencing both their isotope values and their cadmium and iron content in livers. These two striking behaviours were relatively recent from the collection of the samples, in agreement with the rapid turn-over of livers compared to muscles. An experimental study carried on gerbils (*Meriones unguiculatus*) has determined a faster turn-over rate of carbon in livers (half-life: 6.4 days) than in muscles (half-life: 27.6 days) (Tieszen *et al.*, 1983). This behaviour is likely to be in relation with the migration pattern of the tunas. When tunas get through the Biscay Gulf, some individuals become associated with dolphins sharing similar prey preferences as squids, suggesting a potential competition. For unclear reasons, some tunas display different feeding particularities mainly based on small Sternoptychidae. As tuna feeding behaviour may differ in relation with the dolphin species it is associated to, two hypotheses can be formulated considering these tunas which recently fed on Sternoptychidae:

- (1) Tunas and common dolphins were hunting together preys that were abundant and varied enough to allow specific preferences within a common dolphin/tuna school or, as suggested by Hassani *et al.* (1997),
- (2) these tunas were not associated with dolphins.

To conclude, combined stable isotopes and heavy metal analyses appear as powerful tools for delineating

the trophic relationship within these mixed-species aggregations. Further researches on other organisms from this area will help to get a better understanding on heavy metal transfer and different alimentary relations within high trophic levels.

Thanks to Mr Alain Abarnou (IFREMER, Brest) and his comments: without him this work would not have been possible. We are grateful to Mr Biondo (Oceanology Laboratory, Liège, Belgium) for his valuable technical assistance and to Mr. Michel Goujon, Loïc Antoine (IFREMER, Brest, France), Vincent Ridoux and Sami Hassani (Oceanopolis, Brest, France) who collaborated to perform all the sampling and collect the biological informations. The personal comments of Sami Hassani on dolphin and tuna diets helped us to improve this manuscript. We also wish to thank Mr Holsbeek and Pr Joiris from VUB (Belgium) for their valuable advices. Thanks also to Miss N. Poisseroux for her logistical assistance.

This study was supported by grants from *Modèle – Environnement, Fondation Lefranc* and from the Belgian Office for Scientific, Technical and Cultural Affairs (contract MN/DD/50).

- Abend, A. G. and Smith, T. D. (1995) Differences in ratios of stable isotopes of nitrogen in long-finned pilot whales (*Globicephala melas*) in the western and eastern North Atlantic. *ICES Journal of Marine Science* **52**, 837–841.
- André, J. M., Amiard-Triquet, C., Boudou, A. and Ribeyre, F. (1990a) Cadmium contamination of tissues and organs of Delphinids species (*Stenella attenuata*). Influence of biological and ecological factors. *Ecotoxicological and Environmental Safety* **20**, 290–306.
- André, J. M., Ribeyre, F. and Boudou, A. (1990b) Mercury contamination level and distribution in tissues and organs of Delphinids (*Stenella attenuata*) from the Eastern Tropical Pacific, in relation to biological and ecological factors. *Marine Environmental Research* **30**, 43–72.
- Au, D. W. (1991) Polyspecific nature of tuna schools: shark, dolphin and seabird associates. *Fishery Bulletin* **89**, 343–354.
- Au, D. W. and Pitman, R. L. (1986) Seabird interactions with dolphins and tuna in the Eastern Tropical Pacific. *The Condor* **88**, 304–317.
- Blanco, C., Aznar, J. and Raga, J. A. (1995) Cephalopods in the diet of the striped dolphin *Stenella coeruleoalba* from the western Mediterranean during an epizootic in 1990. *Journal of Zoology* **237**, 151–158.
- Bouquegneau, J. M. and Joiris, C. (1988) The fate of stable pollutants-heavy metals and organochlorines. In *Marine Organisms. Advances in Comparative and Environmental Physiology*, vol. 2, pp. 219–247. Springer, Berlin.
- Bouquegneau, J. M. and Joiris, C. (1992) Ecotoxicology of stable pollutants in cetaceans: organochlorines and heavy metals. In *Whales: Biology – Threats – Conservation. Symposium Proceedings (Brussels, 5–7 June 1991)*, ed. J. J. Symoens, pp. 247–250. Royal Academy of Overseas Sciences (Brussels).
- Bouquegneau, J. M., Debacker, V., Gobert, S. and Nellissen, J.P. (1997) Toxicological investigations on four sperm whales stranded on the Belgian coast: inorganic contaminants. In *Sperm whale deaths in the North Sea: Science and Management. Bulletin de l'Institut Royal des Sciences Naturelles de Belgique* eds. T. G. Jacques and R. H. Lambertsen, pp. 75–78. 67-supplement.
- Burns, J. M., Trumble, S. J., Castelline, M. A. and Testa, J. W. (1998) The diet of Weddells seals in MuMurdo Sound, Antarctica as determined from scat collections and stable isotopes analysis. *Polar Biology* **19**, 272–282.
- Cabana, G. and Rasmussen, J. B. (1994) Modelling food chain structure and contaminant bioaccumulation using stable nitrogen isotopes. *Nature* **372**, 255–257.
- Caurant, F., Amiard, J. C., Amiard-Triquet, C. and Sauriau, P. G. (1994) Ecological and biological factors controlling the concentrations of trace elements (As, Cd, Cu, Hg, Se, Zn) in Delphinids *Globicephala melas* from the North Atlantic Ocean. *Marine Ecology Progress Series* **103**, 207–219.
- Collet, A. (1993a) Age et statut reproducteur des cétacés capturés. Resultats préliminaires et analyse partielle des données. *Rapport Gerdeau* 93. 19p.
- Collet, A. (1993b) Age et statut reproducteur des cétacés capturés. Resultats préliminaires et analyse partielle des données. *Rapport Gerdeau* 93. 9p.
- Dauby, P., Khomsi, A., Bouquegneau, J. M. (1998) Trophic relationships within intertidal communities of the Brittany coasts: a stable carbon isotope analysis. *Journal of Coastal Research* **14**, 1202–1212.
- DeNiro M. J. and Epstein S. (1978) Influence of the diet on the distribution of carbon isotopes in animals. *Geochimica Cosmochimica Acta* **42**, 495–506.
- DeNiro M. J. and Epstein S. (1981) Influence of the diet on the distribution of nitrogen isotopes in animals. *Geochimica Cosmochimica Acta* **45**, 341–351.
- Di-Méglio N., Romero-Alvarez R. and Collet A. (1996) Growth comparison in striped dolphins, *Stenella coeruleoalba*, from the Atlantic and Mediterranean coasts of France. *Aquatic Mammals* **22**, 11–21.
- Goujon, M., Antoine, L. and Collet A. (1993) Incidental catches of cetaceans by the French albacore tuna driftnet fishery-preliminary results. ICES Statutory meeting 1993, N13, Ref. H: 17.
- Goujon, M. (1996) Captures accidentelles du filet maillant dérivant et dynamique des populations de dauphins au large du Golfe de Gascogne. *Les Publications du Laboratoire Halieutique* **15**, 239. Laboratoire Halieutique de l'ENSAR.
- Gowans, S. and Whitehead, H. (1995) Distribution and habitat partitioning by small odontocetes in the Gully, a submarine canyon on the Scotian shelf. *Canadian Journal of Zoology* **73**, 1599–1608.
- Hassani, S., Antoine, L., Ridoux, V., (1997) Diet of Abacore, *Thunnus alalunga*, and dolphins, *Delphinus delphis* and *Stenella coeruleoalba*, caught in the North-east Atlantic albacore drift-net fishery: A progress report. *Journal of Northwest Atlantic Fishery Science* **22**, 119–123.
- Havelange, S., Lepoint, G., Dauby, P. and Bouquegneau J. M. (1997) Feeding of the spard fish *Sarpa salpa* in a seagrass ecosystem: diet and carbon flux. *P.S.Z.N.I. Marine Ecology* **18**, 289–297.
- Hobson K. A. and Clarck R. G. (1992) Assessing avian diets using stable isotopes. II: Factors influencing diet-tissue fractionation. *Condor* **94**, 189–197.
- Hobson K. A. and Welch H. E. (1992) Determination of trophic relationships within a high Arctic food web using $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ analysis. *Marine Ecology Progress Series* **84**, 9–18.
- Hobson, K. A. K., Ambrose, W. G. and Renaud, P. E. (1995) Sources of primary production, benthic-pelagic coupling, and trophic relationships within the North-east water polynia: insights from $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ analysis. *Marine Ecology Progress Series* **128**, 1–10.
- Hobson, K. A., Schell, D., Renouf, D. and Noseworthy E. (1996) Stable-carbon and nitrogen isotopic fractionation between diet and tissues of captive seals: implications for dietary reconstructions involving marine mammals. *Canadian Journal of Fisheries and Aquatic Sciences* **53**, 528–5333.
- Hobson, K. A., Sease, J. L., Merrick, R. L. and Piatt, J. F. (1997) Investigating trophic relationships of pinnipeds in Alaska and Washington using stable isotopes ratios of nitrogen and carbon. *Marine Mammal Science* **13**, 114–132.
- Kidd, K. A., Hesslein, R. H., Fudge, R. J. P. and Hallard, K. A. (1995) The influence of trophic level as measured by $\delta^{15}\text{N}$ on mercury concentrations in freshwater organisms. *Water, Air and Soil Pollution* **80**, 1011–1015.
- Law, R. J., Morris, R. J., Allchin, C. R. and Jones, B. R. (1997) Metals and chlorobiphenyls in tissues of sperm whales (*Physeter macrocephalus*) and other cetaceans species exploiting similar diets. In *Sperm Whale Deaths in the North Sea: Science and Management*, eds. T. G. Jacques and R. H. Lambertsen, pp. 79–89. Bulletin de l'Institut Royal des Sciences Naturelles de Belgique 67-supplement.
- Michener, R. H. and Schell, D. M., (1994) Stable isotope ratios as tracers in marine aquatic foodwebs. In *Stable Isotopes in Ecology and Environmental Science*, eds. K. Lajtha and R. H. Michener, pp. 138–186. Blackwell Scientific Publications, Oxford, UK.
- Ostrom, P. H., Lien, J. and Macko, S. A. (1993) Evaluation of the diet of Sowerby's beaked whale, *Mesoplodon bidens*, based on isotopic comparisons among north-western Atlantic cetaceans. *Canadian Journal of Zoology* **71**, 858–861.
- Overholtz, W. J. and Waring, G. T. (1991) Diet composition of pilot whales *Globicephala* sp. and common dolphins *Delphinus delphis* in the mid-Atlantic Bight during spring 1989. *Fishery Bulletin, US*, **89**, 723–728.
- Perrin, W. F., Warner, R. R., Fiscus, C. H. and Holts, D.B. (1973) Stomach contents of porpoise, *Stenella* sp., and yellowfin tuna, *Thunnus albacares* in mixed species aggregations. *Fishery Bulletin* **71**, 1077–1091.

- Peterson, B. J. and Fry, B. (1987) Stable isotopes in ecosystem studies. *Annual Review of Ecology and Systematics* **18**, 293–320.
- Rau, G. H., Ainley, D. G., Bengtson, J. L., Torres, J. J. and Hopkins, T. L. (1992) $^{15}\text{N}/^{14}\text{N}$ and $^{13}\text{C}/^{12}\text{C}$ in Weddell seabirds, seals, and fish: implications for diet and trophic structure. *Marine Ecology Progress Series* **84**, 1–8.
- Scherrer, B. (1984) Biostatistiques. 850 pp. Gaëtan Morin Québec, Canada.
- Scott, M. D. and Cattanach, K. L. (1998) Diel patterns in aggregations of pelagic dolphins and tunas in the eastern Pacific. *Marine Mammal Science* **14**, 401–428.
- Smith, R. J., Hobson, K. A., Koopman, H. N. and Lavigne, D. M. (1996) Distinguishing between populations of fresh-and salt-water harbour seals (*Phoca vitulina*) using stable-isotope ratios and fatty acid profiles. *Canadian Journal of Fisheries and Aquatic Sciences* **53**, 272–279.
- Stewart, F. M., Philips, R.A., Catry, P. and Furness, R.W. (1997) Influence of species, age and diet on mercury concentrations in Shetland seabirds. *Marine Ecology Progress Series* **151**, 237–244.
- Thompson, D. R., Furness, R. W. and Lewis, S. A. (1995) Diets and long-term changes in $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values in northern fulmars *Fulmarus glacialis* from two North-east Atlantic colonies. *Marine Ecology Progress Series* **123**, 3–11.
- Tieszen, L. L., Boutton, T. W., Tesdahl, K. G. and Slade, N. A. (1983) Fractionation and turn-over of stable carbon isotopes in animal tissues: implication for $\delta^{13}\text{C}$ analysis of diet. *Oecologia (Berlin)* **57**, 32–37.
- Wagemann, R., Stewart, R. E., Béland, P. and Desjardins, C. (1990) Heavy metals and selenium in tissues of beluga whales, *Delphinapterus leucas*, from the Canadian Arctic and the St. Lawrence estuary. *Canadian Journal of Fisheries and Aquatic Sciences* **224**, 191–206.
- Würtz, M. and Marrale, D. (1993) Food of striped dolphin, *Stenella coeruleoalba*, in the Ligurian Sea. *Journal of the Marine Biological Association of UK* **73**, 571–578.
- Young, D. D. and Cockcroft, V. G. (1995) Stomach contents of stranded common dolphins *Delphinus delphis* from the south-east of Southern Africa. *Zeitschrift für Säugetierkunde* **60**, 343–351.
-