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Marine mammals from northeast atlantic: relationship between their trophic status as determined by $\delta^{13}C$ and $\delta^{15}N$ measurements and their trace metal concentrations

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Abstract

The relationship between trophic position through $\delta^{13}C$ and $\delta^{15}N$ and trace metal concentrations (Zn, Cd, Cu and Hg) was investigated in the tissues of six marine mammal species from the Northeast Atlantic: striped dolphin *Stenella coeruleoalba,* common dolphin, *Delphinus delphis*, Atlantic white-sided dolphin *Lagenorhynchus acutus*, harbour porpoise *Phocoena phocoena*, white beaked-dolphin *Lagenorhynchus albirostris,* grey seal *Halichoerus grypus* stranded on French Channel and Irish coasts. White-beaked dolphins, harbour porpoises, white-sided dolphins, common and striped dolphins display the same relative and decreasing trophic position, as measured by $\delta^{15}N$ values, along both the Irish and French channel coasts, reflecting conservative trophic habits between these two places. Hepatic and renal Cd concentrations were significantly correlated to muscle $\delta^{13}C$ and $\delta^{15}N$ values while Hg, Zn and Cu did not. These results suggest that Cd accumulation is partly linked to the diet while other factors such as age or body condition might explain Hg, Zn or Cu variability in marine mammals. Combined stable isotope and trace metal analyses appear to be useful tools for the study of marine mammal ecology.

Keywords: Marine mammals; Stable isotopes; Heavy metals; Trophic transfer; Northeast Atlantic; *Delphinus delphis*; *Stenella coeruleoalba*; *Phocoena phocoena*; *Lagenorhynchus albirostris*; *Lagenorhynchus acutus*; *Halichoerus grypus*

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1. Introduction

Over 40 species of cetaceans and pinnipeds occur throughout the Northeast Atlantic as defined by the OSPAR Convention (OSPAR, 2000). Among these species, the common dolphin, *Delphinus delphis*, the striped dolphin, *Stenella coeruleoalba*, the harbour porpoise, *Phocoena phocoena*, the white-beaked dolphin, *Lagenorhynchus albirostris*, the white-sided dolphin, *Lagenorhynchus acutus* and the grey seal, *Halichoerus grypus* are regularly observed within the Channel and Celtic Sea (Hammond et al., 1995; OSPAR, 2000; Rogan & Berrow, 1996).

Marine mammals are top predators and their key role in the structure of the marine ecosystem has often been suggested (Bouquegneau, Debacker, & Gobert, 1997; Bowen, 1997; Pauly, Trites, Capuli, & Christensen, 1998). Published information on diet composition and trophic status of these species in Irish waters and the French Channel is sparse (Rogan & Berrow, 1996) and is required to understand the role of marine mammals in ecosystem dynamics. Moreover, in marine mammals, diet is the main pollutant contamination pathway and might influence their contaminant load (reviewed by Aguilar, Borrel, & Pastor, 1999; Das, Debacker, Pillet, & Bouquegneau, in press).

Dietary studies are often performed by stomach content analysis or field observations. In marine mammals, the use of naturally occurring stable isotopes of carbon (^{13}C) and nitrogen (^{15}N) has recently provided new insights into their feeding ecology (e.g. Abend & Smith, 1995; Burns, Trumble, Castellini, & Testa, 1998; Hobson, Sease, Merrick, & Piatt, 1997; Hobson & Welch, 1992; Kelly, 2000; Lesage, Hammil, & Kovaks, 2001; Smith, Hobson, Koopman, & Lavigne, 1996). The method is based on the demonstration that stable isotope ratios of a consumer are related to those of their prey (De Niro & Epstein, 1978, 1981; Peterson & Fry, 1987). nitrogen-15 typically shows a stepwise increase with trophic level within a food chain (Cabana & Rasmussen, 1994; Hobson & Welch, 1992; Thompson, Furness, & Lewis, 1995). The carbon-13 value is close to that of the diet and is preferentially used to indicate relative contributions to the diet of different potential primary sources in a trophic network, indicating for example the aquatic vs. terrestrial, inshore vs. offshore, or pelagic vs. benthic contribution to food intake (Dauby, Khomsi, & Bouquegneau, 1998; Hobson, Ambrose, & Renaud, 1995; Smith et al., 1996). When using stable isotopes to assess diets of animals feeding at or near the top of the trophic web on several prey items, many of which may have similar isotopic signatures, clear distinctions about the diet are more difficult to determine. However, it may be possible to infer the general trophic level at which animals are feeding, by applying and comparing ^{15}N and, to a limited extent ^{13}C) step-wise enrichment values (Kurle & Worthy, 2001). Furthermore, stable isotope analysis is often used to provide a continuous variable with which to assess both trophic level (Hobson et al., 1995; Michener & Schell, 1994) and trophic transfer of contaminants (Das, Lepoint, Loizeau, Debacker, Dauby, & Bouquegneau, 2000; Kidd, Hesslein, Fudge, & Hallard, 1995). In previous studies, stable isotope ratios and trace metal concentrations were determined in common and striped dolphin tissues from the Northeast Atlantic (Das et al., 2000) and high renal Cd concentrations encountered were assumed to be related to the diet.

In this study, we examine closer and compare the relationship among six marine mammal species from the Irish and French channel coasts, the grey seal, the harbour porpoise, the striped dolphin, the common dolphin, the white-sided dolphin and the white-beaked dolphin, using a multidisciplinary approach based on stable isotope ($\delta^{13}C$ and $\delta^{15}N$) and trace metal analyses. We also examined mercury (Hg), cadmium (Cd), zinc (Zn) and copper (Cu) concentrations for evidence of diet transfer, specific bioaccumulation or biomagnification processes or differences between the two regions.

2. Materials and methods

2.1. Collection and storage

Liver, kidney and muscle samples were collected from seven striped dolphins, 24 common dolphins, 13 harbour porpoises, four white-beaked dolphins, five white-sided dolphins and two grey seals found stranded or incidentally caught in fish nets. between 1989 and 1993 (counties of Cork, Galway, Kerry, Meath, Clare and Waterford) and the northern French Atlantic coast (region of Cotentin) between 1998 and 2001.

2.2. Analytical methods

2.2.1. Zn, Cd and Cu analyses

After being weighed and dried for 48 h at 110 °C, samples were digested with a solution of nitric acid (Merck 456) and slowly heated to 100 °C until complete digestion. Atomic absorption spectrophotometry (ARL 3510) was used to determine Cu, Zn and Cd concentrations. Concentrations are expressed as $\mu g\ g^{-1}$ dry weight (dw).

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

2.2.2. Hg analyses

Hg was analysed by flameless atomic absorption spectrophotometry (Perkin-Elmer MAS-50A) after sulphuric acid digestion, as described by Joiris, Holsbeek, Bouquegneau, and Bossicart (1991). Quality control measurements for total mercury included replicate analysis resulting in coefficients of variation $<10\%$ and analysis of certified material (DORM-1, NRC, Canada).

2.2.3. Stable isotope measurements

Organisms may vary in their concentrations of lipids. As lipids have been shown to be depleted in ^{13}C relatively to the diet (Tieszen, Boutton, Tesdahl, & Slade, 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 ground with a mortar and pestle into powder. Stable isotope measurements were performed on a V.G. Optima (Micromass) IR–MS coupled to a N-C-S elemental analyser (Carlo Erba) for automated analyses. Routine measurements were precise to within 0.3 $^{0}/_{00}$ for 13-carbon and 15-nitrogen. Stable isotope ratios were expressed in δ notation according to the following equation:

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

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

Carbon and nitrogen ratios are expressed relative to the v-PDB (Vienna Peedee Belemnite) standard and to atmospheric nitrogen, respectively. Reference materials were IAEA-N1 ($\delta^{15}N = +0.4 \pm 0.2‰$) and IAEA CH-6 (sucrose) ($\delta^{13}C = -10.4 \pm 0.2‰$).

2.3. Data treatment

Kolmogorov–Smirnov test was used to test for data departure to normality. When not normally distributed the variables were log-transformed. Effect of species and sampling location on δ^{13}C and δ^{15}N values or trace metal concentrations were tested simultaneously using multivariate analysis of variance (two-way MANOVA) followed by post-hoc multiple comparison tests (LSD test). Parametric Spearman–coefficient has been used to test correlations between the values. Results were judged significant when $P < 0.01$ unless otherwise stated.

3. Results

3.1. Stable isotope analysis

δ^{15}N and δ^{13}C analyses were performed on liver and muscle of six marine mammal species (Table 1). The grey seal and the white-sided dolphin were excluded from statistical treatment due to the small sample size and availability for the two regions.

δ^{13}C measurements varied significantly with both species and sampling location while mean δ^{15}N value remained similar for French Channel and Irish coasts (two-way MANOVA, univariate results see Table 2). In both locations, the lower δ^{15}N values occurred in the striped dolphin, significantly depleted compared with harbour porpoise (post-hoc LDS test, $P < 0.0001$) and white-beaked dolphin (post-hoc LSD test, $P < 0001$). Mean δ^{15}N data did not differ significantly between striped and common dolphins (post-hoc LSD test, $P = 0.06$). For both ecosystems, striped dolphins were significantly depleted in carbon-13 compared with white-beaked dolphin (post-hoc LSD test, $P < 0.0005$) and harbour porpoises (post-hoc LSD test, $P < 0.005$) but were similar to common dolphins (post-hoc LSD test, $P = 0.1$). Mean δ^{13}C values did not differ significantly between harbour porpoise and white-beaked dolphin ($P > 0.1$).

Table 1
δ ^{15}N and δ ^{13}C values in muscles of marine mammals from the French Channel and Irish coasts

	Channel coast		Irish coast	
	δ^{13}C	δ^{15}N	δ^{13}C	δ^{15}N
Striped dolphin *Stenella coeruleoalba*	−16.7 (−16.7) + 0.4 (−17.1/−16.4) $n = 3$	11 (10.1) + 1.8 (9.8–13.1) $n = 3$	−17.5 (−17.5) + 0.1 (−17.7/−17.4) $n = 3$	10.8 (11) + 0.6 (10.2–11.3) $n = 3$
Common dolphin *Delphinus delphis*	−16.5 (−16.6) + 0.5 (−17.1/−15.6) $n = 8$	12.1 (12.2) + 0.4 (11.4–12.6) $n = 8$	−17.1 (−17.1) + 0.4 (−18/−16.5) $n = 14$	12.2 (12.4) + 1 (10–14.3) $n = 14$
Atlantic white-sided dolphin *Lagenorhynchus acutus*	na		−17.0 (−17.2) + 0.5 (−17.4/−16.4) $n = 4$	12.7 (12.6) + 0.5 (12.1–13.4) $n = 4$
Harbour porpoise *Phocoena phocoena*	−16.1 (−15.8) + 0.6 (−17.1/−15.8) $n = 4$	16.5 (17.1) + 2.4 (13.2–18.8) $n = 4$	−16.5 (−16.4) + 0.7 (−17.2/−15.1) $n = 7$	14.1 (13.9) + 1.6 (12–17.2) $n = 7$
White-beaked dolphin *Lagenorhynchus albirostris*	−15.4 $n = 1$	16.5 $n = 1$	−16.3 (−16.4) + 0.3 (−16.6/−16) $n = 3$	15.8 (16.4) + 2.3 (13.3–17.8) $n = 3$
Grey seal *Halichoerus gryphus*	−15.4 $n = 1$	18.3 $n = 1$	na	

Data is given as average (median) + standard deviation, (minimum – maximum); *n*, number of samples, na, not available.

Table 2
Influence of species and sampling locations (two-way MANOVA results, univariate specific effects) on metal concentrations and stable isotope ratios in the tissues

Factors	Species	Geographic location
Liver		
Zn (log)	$F_{3,23} = 1.4$, $P > 0.2$	$F_{1,23} = 0.001$, $P > 0.9$
Cu (log)	$\mathbf{F_{3,23} = 4.1}$, $\mathbf{P < 0.02}$	$F_{1,23} = 0.9$, $P > 0.3$
Cd (log)	$\mathbf{F_{3,26} = 20.2}$, $\mathbf{P < 0.0001}$	$F_{1,26} = 0.6$, $P > 0.4$
Hg (log)	$F_{3,14} = 1.2$, $P > 0.3$	$F_{1,14} = 0.7$, $P > 0.4$
Kidney		
Zn (log)	$\mathbf{F_{3,23} = 5.9}$, $\mathbf{P < 0.005}$	$F_{1,23} = 0.16$, $P > 0.6$
Cu (log)	$\mathbf{F_{3,23} = 4.7}$, $\mathbf{P < 0.015}$	$F_{1,23} = 3.9$, $P > 0.06$
Cd (log)	$\mathbf{F_{3,26} = 20.3}$, $\mathbf{P < 0.0001}$	$F_{1,26} = 1.15$, $P > 0.2$
Hg	$F_{3,14} = 0.8$, $P > 0.5$	$F_{1,14} = 0.05$, $P > 0.8$
Muscle		
Zn (log)	$\mathbf{F_{3,23} = 12.2}$, $\mathbf{P < 0.0001}$	$F_{1,23} = 2.98$, $P > 0.09$
Cu (log)	$F_{3,23} = 0.6$, $P > 0.6$	$F_{1,23} = 3.3$, $P > 0.08$
Hg (log)	$F_{3,14} = 0.06$, $P > 0.9$	$F_{1,14} = 0.001$, $P > 0.9$
δ^{13}C	$\mathbf{F_{3,35} = 6.4}$, $\mathbf{P < 0.002}$	$\mathbf{F_{1,35} = 11.7}$, $\mathbf{P < 0.002}$
δ^{15}N	$\mathbf{F_{3,35} = 22.01}$, $\mathbf{P < 0.0001}$	$F_{1,35} = 2$, $P > 0.1$

ns, not significant, $P > 0.1$; log: indicate the data were log-transformed before statistical treatment to ensure a normal distribution

The mean muscle and liver $\delta\ ^{13}C$ values were significantly more negative for the animals from the Irish coasts compared with animals from the Northern French coast (post-hoc LSD test, $P < 0.0002$) (Fig. 1).

3.2. Metal level in the tissues

Geographic location did not affect trace metal concentrations while Zn, Cu and Cd displayed strong-interspecific differences (Table 3). Hg remained similar between species and locations. Striped dolphin displayed higher renal Zn concentrations than harbour porpoise (post-hoc LSD test, $P < 0.01$) and common dolphins (post-hoc LSD test, $P < 0.005$), which in turn were higher than those of white-beaked dolphin (post-hoc LSD test, $P < 0.05$). Striped dolphin displayed the highest hepatic and renal Cd concentrations significantly higher than those of common dolphin (post-hoc LSD test, $P < 0.0006$), which in turn were higher than those of harbour porpoise (post-hoc LSD test, $P < 0.0001$) and white beaked dolphin (post-hoc LSD test, $P < 0.01$).

The highest hepatic Cu was measured in the liver of the harbour porpoise and the lowest in the common dolphin (post-hoc LSD test, $P < 0.005$). Other species displayed similar Cu concentrations.

3.3. Relationship between stable isotopes and heavy metals

No clear relationship was observed between hepatic or muscular $\delta^{15}N$ and $\delta^{13}C$ and hepatic, renal or muscular Hg concentrations, neither within a species nor when

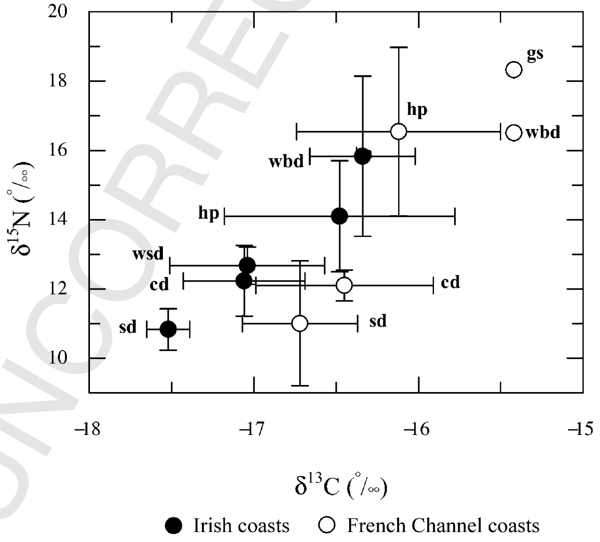

Fig. 1. Muscle $\delta^{13}C$ and $\delta^{15}N$ in marine mammals from Irish, French Channel coasts (Sd: striped dolphin; cd: common dolphin; wsd: white-sided dolphin; hp: harbour porpoise; wbd: white-beaked dolphin, gs: grey seal).

Table 3
Zn, Cd, Cu and Hg concentrations (μg g^{-1} dry weight) in the liver, muscle and kidney of marine mammals from the French Channel and Irish coasts

		French Channel coasts				Irish coasts			
		Zn	Cd	Cu	Hg	Zn	Cd	Cu	Hg
Striped dolphin ***Stenella coeruleoalba***	Liver	**140** (155) + 41 (94–171) *n* = 3	**35** (4.6) + 55 (3–99) *n* = 3	**26** (25) + 9.2 (18–36) *n* = 3	**37** (40) + 25 (11–60) *n* = 3	**185** (172) + 60 (133–250) *n* = 3	**38** (37) + 13 (25–51) *n* = 3	**39** (37) + 9 (30–49) *n* = 3	**41** (45) + 31 (19–63) *n* = 2
	Muscle	**33** (33) + 0.9 (33–34) *n* = 3	< **0.1** (< 0.1) + 0.2 (< 0.1 / 0.3) *n* = 3	**7.8** (7.5) + 1.2 (6.9–9.1) *n* = 3	**3.7** (4.1) + 1.5 (2–4.8) *n* = 3	**47** (42) + 9 (42–58) *n* = 3	**0.2** (0.3) + 0.2 (< 0.1–0.4) *n* = 3	**6** (5) + 1.7 (4.7–7.8) *n* = 3	**4** (4) + 0.9 (3.4–4.6) *n* = 2
	Kidney	**112** (124) + 35 (73–140) *n* = 3	**71** (22) + 104 (< 0.1 / 190) *n* = 3	**16** (15) + 2.6 (14–19) *n* = 3	**8** (7.4) + 4.3 (3.9–13) *n* = 3	**150** (150) + 15 (136–163) *n* = 4	**150** (141) + 35 (118–199) *n* = 4	**18** (18) + 4.6 (13–24) *n* = 4	**15** (15) + 5 (11–18) *n* = 2
Common dolphin ***Delphinus delphis***	Liver	**141** (128) + 31 (106–177) *n* = 5	**1.5** (1.3) + 1.3 (0.3–3.7) *n* = 5	**17** (15) + 3 (14–22) *n* = 5	**124** (29) + 156 (2.2–320) *n* = 5	**150** (152) + 35 (81–220) *n* = 14	**6.8** (4.6) + 6.9 (1.2–27) *n* = 14	**20** (20) + 4.6 (13–28) *n* = 14	**46** (19) + 55 (4-1 63) *n* = 8
	Muscle	**32** (33) + 6 (20–39) *n* = 8	< **0.1** (< 0.1) (< 0.1) *n* = 8	**5.1** (4.8) + 1.4 (3.8–7) *n* = 8	**2.7** (1.4) + 2.9 (0.3–9.1) *n* = 8	**48** (43) + 13 (35–80) *n* = 14	**0.3** (0.2) + 0.3 (< 0.1-1) *n* = 14	**6** (5.8) + 1.7 (3.6–9.8) *n* = 14	**2.7** (2.7) + 1.2 (1.5–4.9) *n* = 3
	Kidney	**84** (92) + 23 *n* = 5	**8.7** (7.8) + 8.1 (1-22) *n* = 5	**13** (14) + 5.6 (7.5–21) *n* = 5	**13** (4.1) + 5.1 (6.8–20) *n* = 5	**97** (96) + 23 (53–130) *n* = 12	**33** (23) + 25 (6–72) *n* = 12	**13** (12) + 3.2 (8.2–19) *n* = 12	**8** (6) + 5 (2-14) *n* = 7
White-sided dolphin ***Lagenorhynchus acutus***	Liver	na				**136** (136) + 140 (37–235) *n* = 2	**2.8** (2.8) + 4 (< 0.1 / 5.7) *n* = 2	**18** (18) + 22 (2.7–33) *n* = 2	**150** *n* = 1
	Muscle					**41** (34) + 24 (22–75) *n* = 4	**3.6** (0.3) + 6.7 (0.1–14) *n* = 4	**4.8** (4.7) + 1.7 (3.2–6.4) *n* = 4	**2.7** (2.7) + 1.2 (1.5–3.9) *n* = 3
	Kidney					**97** (97) + 10 (90–104) *n* = 2	**9.6** (9.6) + 14 (< 0.1 / 19) *n* = 2	**7.7** (7.7) + 6.2 (3.4–12) *n* = 2	**7.8** *n* = 1

(*continued on next page*)

Table 3 (*continued*)

		French Channel coasts				Irish coasts			
		Zn	Cd	Cu	Hg	Zn	Cd	Cu	Hg
Harbour porpoise ***Phocoena phocoena***	Liver	**258** (140)+246 (126–628) *n*=4	**0.3** (0.3)+0.2 (0.1–0.5) *n*=4	**111** (103)+69 (36–203) *n*=4	**8.9** (3.5)+11 (3–26) *n*=4	**173** (135)+98 (91–380) *n*=8	**0.6** (0.6)+0.4 (<0.1/1.1) *n*=8	**22** (23)+11 (3–39) *n*=8	**24** (5.3)+42 (4.1–99) *n*=5
	Muscle	**126** (102)+49 (101–199) *n*=4	<**0.1** (<0.1) *n*=4	**12** (13)+4.6 (5.2–15) *n*=4	**3.5** (3.5)+2.4 (1.8–5.2) *n*=2	**45** (45)+8 (32–56) *n*=7	<**0.1** (0.1)+0.1 (<0.1/0.2) *n*=7	**4.6** (5)+1.6 (1.2–6) *n*=7	**3.6** (3.1)+1.4 (2.5–5.6) *n*=4
	Kidney	**99** (96)+8 (93–111) *n*=4	**1.5** (0.3)+2.7 (<0.1 / 5.6) *n*=4	**32** (27)+16 (18–55) *n*=4	**4** (3.1)+2.5 (2.3–7.6) *n*=4	**90** (90)+16 (70–112) *n*=7	**3.9**(3.7)+3.4 (3.7)+3.4 (0.4–10) *n*=7	**13** (13)+1.6 (11–15) *n*=7	**2.2** (2.1)+0.6 (1.6–2.9) *n*=4
White-beaked dolphin ***Lagenorhynchus albirostris***	Liver	**96** *n*=1	**0.4** *n*=1	**27** *n*=1	**229** *n*=1	**101** (101)+28 (81–121) *n*=2	**0.3** (0.3)+0.1 (0.2–0.3) *n*=2	**24** (24)+14 (14–34) *n*=2	na
	Muscle	**67** *n*=1	**0.9** *n*=1	**5.5** *n*=1	**4.2** *n*=1	**97** (110)+39 (53–127) *n*=3	<**0.1** (<0.1) *n*=3	**5** (4.3)+2 (3.5–7.2) *n*=3	na
	Kidney	**37** *n*=1	**0.9** *n*=1	**8.2** *n*=1	**5.6** *n*=1	**73** (73)+41 (44–102) *n*=2	**0.3** (0.3)+0.03 (0.2–0.3) *n*=2	**7.6** (7.6)+6.1 (3.3–12) *n*=2	na
Grey seal ***Halichoerus grypus***	Liver	**204** (204)+59 (163–246) *n*=2	**1.5** (1.5)+1.0 (0.8–2.2) *n*=2	**51** (51)+7 (46–56) *n*=2	**368** (368)+59 (326–409) *n*=2	na			
	Muscle	**92** *n*=1	**0.3** *n*=1	**3.7** *n*=1	**7.6** *n*=1				
	Kidney	**150** *n*=1	**5.7** *n*=1	**17** *n*=1	**49** *n*=1				

Data is given as a mean (median)±standard deviation, range of concentrations (minimum–maximum); *n*, number of samples; na, not available.

all species were considered ($P > 0.01$). A decreasing relationship was observed between muscle δ^{15}N and cadmium concentrations (log-transformed) in the liver (Pearson product–moment correlation, $r = -0.68$, $P < 0.0001$, $n = 35$) and in the kidney (Pearson product–moment correlation, $r = -0.76$, $P < 0.0001$, $n = 31$, Fig. 2). A similar correlation was found between δ^{13}C values and cadmium concentrations (log-transformed) in the liver (Pearson product–moment correlation, $r = -0.47$, $P < 0.005$, $n = 35$) and in the kidney (Pearson product–moment correlation, $r = -0.56$, $P < 0.001$, $n = 31$, Fig. 3). Zn and Cu in liver, kidney or muscle were not correlated with either muscle δ^{15}N or δ^{13}C values.

4. Discussion

The usefulness of δ^{13}C and δ^{15}N signatures as a measure of trophic status in studies of mercury accumulation has been reported recently for a variety of species, including marine mammals (Atwell, 1998; Bearhop, Waldron, Thompson, & Furness, 2000; Braune, Donaldson, & Hobson, 2002; Kidd, 1995; Thompson, Bearhop, Speakman, & Furness, 1998a,1998b), However the relationship with other metals has received less attention (Camuso, Martinotti, Balestrinni, & Guzzi, 1998; Das et al., 2000). Marine mammals generally have high mercury and cadmium concentrations in their tissues compared to other marine groups, which is thought to be a consequence of their high position in the food web (Atwell, Hobson, & Welch, 1998; Jarman, Hobson, Sydeman, Bacon, & McLaren, 1996; Thompson, 1990). In the present study, although differences in mean mercury levels among species were not statistically significant, high mercury concentrations have been measured in the livers of the white-beaked dolphin and the grey seal from the channel coast (Table 1).

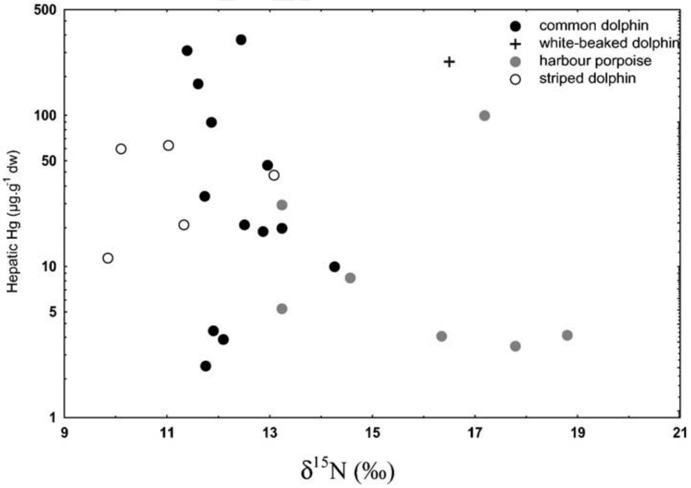

Fig. 2. Relationship between muscle δ^{15}N and hepatic Hg concentration using a log-scale in marine mammals from Irish and French Channel coasts (Pearson product–moment correlation, $P > 0.2$, $n = 26$).

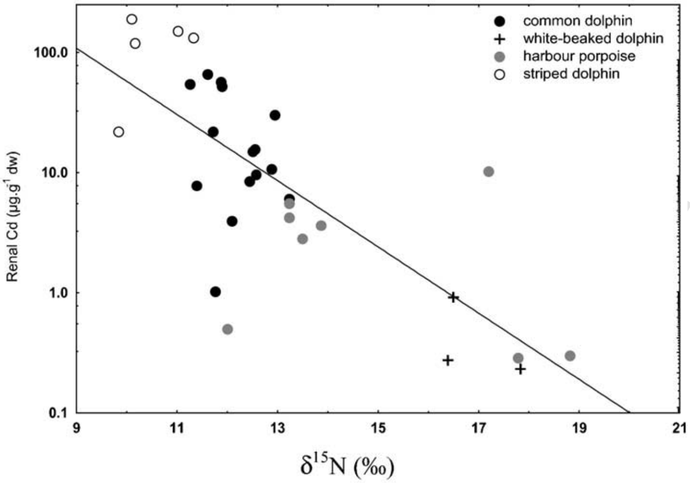

Fig. 3. Relationship between muscle δ^{15}N and renal Cd concentration using a log-scale in marine mammals from Irish and French Channel coasts (Pearson product–moment correlation $r = -0.76$, $P < 0.0001$, $n = 31$).

Elevated Hg values have been described previously in one white-beaked dolphin from the Irish Sea (around 108 µg g^{-1} dw assuming a mean water content of 75% for the tissues) and in 12 grey seals from the Liverpool Bay (mean: 590 µg g^{-1} dw) (Law, Jones, Baker, Kennedy, Milnes, & Morris, 1992). Positive correlations between δ^{15}N values and Hg concentrations have previously been described for freshwater fish (Kidd et al., 1995), in the blood of the great skua chicks (Bearhop et al., 2000) and within a trophic web (Atwell et al., 1998), suggesting that part of the Hg variation may be linked to a bioamplification process. In the present study we did not find any positive relationship between δ^{15}N (or δ^{13}C) values and mercury concentrations either for all species together (Fig. 2), or between species. The wide range of δ^{15}N and δ^{13}C values observed among the six marine mammal species supports the hypothesis of a different trophic status (Fig. 1). Indeed, when considering all the individuals together, δ^{15}N values range from 9‰ to more than 19‰. The δ^{15}N and δ^{13}C of marine predator tissues is determined initially by the isotopic composition of the baseline phyto- and zooplankton sources, technically measured in the particulate organic matter (POM). No POM data are available for the Celtic Sea or the French Channel. A mean δ^{15}N value of 5‰ is generally used for offshore POM (Tucker, Sheats, Giblin, Hopkinson, & Montoya, 1999) but data may vary with their sampling origin, from 4.5‰ in the Gulf of St-Lawrence (Northwest Atlantic) to 5.0 ± 1.2‰ in some salt marshes within the Northeast French Atlantic (Lesage et al., 2001; Riera, Stal, Nieuwenhuize, Richard, Blanchard, & Gentil, 1999), reaching even higher values (up to 9‰) in the coastal part of the North Sea (Mariotti, Lancelot, & Billen, 1994; Middelburg & Nieuwenhuize, 1998). As a result, part of the δ^{15}N interspecific variation in marine mammals might be related to coastal vs. offshore δ^{15}N signature of the primary producers. Indeed, some species such as the striped dolphin are typically oceanic while others, such as the harbour porpoise are more coastal (Evans et al., 1987). However, these geographic differences observed for POM do not explain the large $\delta^{15}N$ range displayed by the six marine mammal species of this study and these $\delta^{15}N$ values may reflect specific trophic status. In both areas, high muscle and hepatic $\delta^{15}N$ values suggest a higher trophic position for white-beaked dolphins and harbour porpoises compared to common or striped dolphins. A high $\delta^{15}N$ value has also been measured in the grey seals but must be confirmed on a larger sampling (Fig. 1). Moreover, white-beaked dolphins, harbour porpoises, white sided dolphins, common and striped dolphins display the same relative and decreasing trophic position, as measured by $\delta^{15}N$ values, along both the Irish and French Channel coasts, reflecting conservative trophic habits between these two locations.

While previous studies suggest that mercury levels tend to be greater in tissues of higher trophic level organisms, it is unclear to what extent this is the result of biomagnification through the food web or bioaccumulation within organisms over time (Atwell et al., 1998). This is difficult to determine as top-predators, such as marine mammals are long-living species and results are weakened by the absence of age data. Relationships between age and Hg have reported for various porpoise (Siebert et al., 1999), dolphin (Honda & Tatsukawa, 1981; Honda, Tatsukawa, Itano, Miyazaki, & Fujiyama, 1983) and seal species (Anan et al., 2002). In this study, total concentrations of mercury were measured, but it is well known that organic and inorganic species of mercury have very different dynamics in marine mammals. Methyl-mercury is the main form present in the prey (fish and invertebrates) and then is stored indefinitely as tiemmanite (HgSe) in the liver of marine mammals (Nigro & Leonzio, 1996).

In contrast to Hg, a significant decreasing relationship between muscular $\delta^{15}N$ and $\delta^{13}C$ values and renal Cd suggest that some of the variation can be linked to dietary specialisation (Figs. 3 and 4). The tendency observed is, as muscle $\delta^{15}N$ and $\delta^{13}C$ increase, cadmium concentrations decrease, with values ranging between those of striped dolphins (maximum renal cadmium, minimum muscle $\delta^{15}N$ and $\delta^{13}C$) and grey seals and white-beaked dolphins (minimum renal cadmium, maximum muscle $\delta^{15}N$ and $\delta^{13}C$).

$\delta^{13}C$ is more useful to indicate the origin of carbon sources than as an indicator of the trophic level. The general pattern of inshore, benthos linked food webs being more enriched in ^{13}C compared with offshore, pelagic food webs presents a potentially useful tool. For example, $\delta^{13}C$ values are typically higher in coastal or benthic food webs than in offshore food webs (Hobson, 1999). A clear relation is observed between increasing muscle $\delta^{13}C$ of the six marine mammal species and oceanic vs. coastal habitat preference (Fig. 1). The $\delta^{13}C$ depletion observed for striped, common and white-sided dolphins would therefore presumably reflect a greater reliance on offshore food while the higher mean values observed for harbour porpoise, white-beaked dolphin and grey seal correspond to their preference for a more coastal habitat (Fig. 1). The white-beaked dolphin has less oceanic preferences than the Atlantic white-sided dolphin (Cipriano, 2002; Kinze, 2002) which is in agreement with its higher $\delta^{13}C$ values. High Cd concentrations have been reported previously

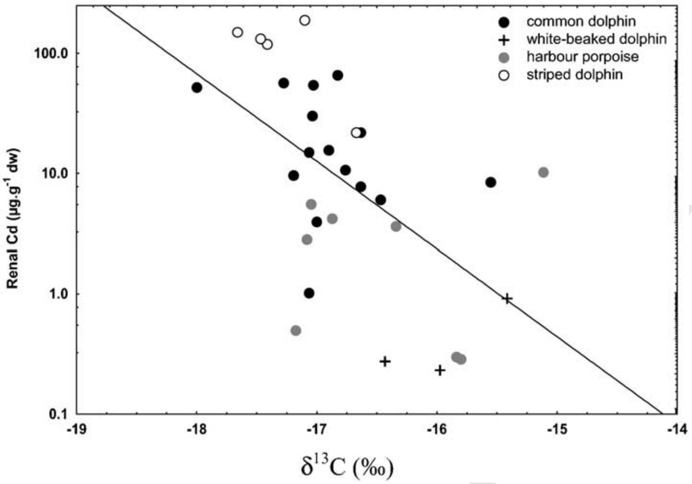

Fig. 4. Relationship between muscle δ^{13}C and renal Cd concentration using a log-scale in marine mammals from Irish and French Channel coasts (Pearson product–moment correlation $r = -0.56$, $P < 0.001$, $n = 31$).

in the livers and kidneys of by-caught striped and common dolphins from the Northeast Atlantic (Das et al., 2000) and oceanic cephalopods constituted a significant part of their diet (Hassani, Antoine, & Ridoux, 1997). Previous studies have highlighted the ability of cephalopods to concentrate cadmium in the digestive gland, even in unpolluted areas like the Kerguelen Islands (Bustamante, Cherel, Caurant, & Miramand, 1998). Oceanic cephalopods are, indeed, considered an essential link for cadmium transfer in marine trophic food chains (Bustamante, Caurant, Fowler & Miramand, 1998; Law, Morris, Allchin & Jones, 1997). Our results suggest that the high Cd values encountered in striped and common dolphins from the Irish and the French Channel coasts are partly diet related as a result of ingestion of prey with low δ^{15}N and δ^{13}C values and high Cd levels. Such high Cd levels can be found in oceanic cephalopods (Bustamante, Caurant et al., 1998) which furthermore display typical low δ^{15}N values (Hooker et al., 2001; Ostrom et al., 1993) compared to individuals collected within the Southern North Sea bight (Das et al., 2002). In contrast the diet of the harbour porpoise, white-beaked dolphins and grey seals are likely to rely more on fish species (Evans, 1987; Rogan & Berrow, 1996).

No relation was found between isotopic composition and either Zn or Cu in the tissues suggesting that the large value range observed for these species is not diet related. Other factors such as body condition influenced by nutritional stress are likely to be involved. Zn, Cu and Cd values measured in these stranded common and striped dolphins are similar to that reported for the same species individuals by-caught in the Bay of Biscay in 1993 (Das et al., 2000). Harbour porpoises that died from infectious diseases displayed significantly higher Zn and Hg concentrations than healthy porpoises that died from physical trauma while Cu and Cd did not differ between the two groups (Bennet et al., 2001). Previous studies have also documented an increase of $\delta^{15}N$ values in starving animals as they might use their proteins for survival (Gannes, Martinez del Rio, & Koch, 1998). In birds, nutritional stress caused substantial increases in diet-fractionation values due either to mobilization and redeposition of proteins elsewhere in the body or amino acid composition changes in the tissues (Gannes et al., 1998; Hobson & Clark, 1992). In contrast, Arctic ground squirrels (*Spermophilus parryii plesius*) in poor and excellent body condition had similar $\delta^{15}N$ values (Ben-David, McColl, Boonstra, & Karels, 1999). Similarly, muscle $\delta^{15}N$ and $\delta^{13}C$ values do not differ between porpoises from the North Sea displaying a poor, moderate of good body condition allowing the use of muscle tissue for stable isotope studies (Das, unpublished data).

To conclude, white-beaked dolphins, harbour porpoises, white sided dolphins, common and striped dolphins display the same relative and decreasing trophic position, as measured by $\delta^{15}N$ values, both the Irish and French channel coasts, reflecting conservative trophic habits between these two places.

Hepatic and renal Cd concentrations were significantly correlated to muscle $\delta^{13}C$ and $\delta^{15}N$ values while Hg, Zn and Cu did not. These results suggest that Cd accumulation is partly linked to the diet while other factors such as age or body condition might explain Hg, Zn or Cu variability in marine mammals. Combined stable isotope and trace metal analyses appear as promising and powerful tools for the study of marine mammal ecology. Further work should concentrate on using the stable isotope method to further explore the behaviour and transfer of trace metals in the marine environment. However, further data on other trophic components from the Northeast Atlantic should allow a better understanding of trophic contaminant transfer.

5. Uncited references

Caurant et al., 1994; Couperus, 1997; Gowans and Whitehead, 1995; Titllemier et al., 2002

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