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Notes

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New information on the diet of True's beaked whale (*Mesoplodon mirus*, Gray 1850), with insights into foraging ecology on mesopelagic prey

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Ziphiidae species contribute up to 30% of the world's cetacean biodiversity. These species are generally difficult to study as they are deep diving animals, and until recently most of the information on this group has been obtained from stranded individuals. However, during the last two decades, our knowledge on the ecology of several species in this family has dramatically increased. Different studies in Ireland have suggested that the deep areas of the Rockall Trough, Porcupine Bight, and slope systems off the northwest of Ireland, where bathymetry might be >1,000 m depth, may provide important habitats for beaked whales (Boisseau *et al*. 2011, Wall *et al.* 2013, Oudejans 2014, Rogan *et al*. 2017). Despite the information compiled in these studies, knowledge on the ecology of True's beaked whales is poorly known and usually gleaned from the small number of strandings. A comprehensive study of beaked whale strandings on the Irish

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coast was published by Rogan and Hernandez-Milian (2011), which reported 132 records of five Ziiphidae species from 1800 to 2009. A total of 13 True's beaked whales (*Mesoplodon mirus*, Gray 1850) have been reported stranded on Irish coasts in the last 200 yr; however, only five were recovered for postmortem examination, including the three animals that stranded in 2013 which are reported here. Dietary information was not obtained from previous strandings.

Mesopelagic fish and squid constitute part of the deep scattering layer (DSL) (Irigoien *et al*. 2014). The DSL is usually located between 200 and 1,000 m depth in the North Atlantic Ocean, with a typical fish community dominated by Myctophidae and Sternoptychidae (Fennel and Rose 2015). Inference from acoustic studies of two species of beaked whales (using acoustic tags) suggests that beaked whales might be foraging from depths of ~400 m (Tyack *et al*. 2006, Madsen *et al*. 2014). True's beaked whale diet has been poorly investigated due to the difficulty in observing animals foraging at sea or obtaining stomach contents from individuals. Only three publications describing diet from stomach contents of this species are available. Sekiguchi *et al*. (1992) identified three squid beaks of *Teuthowenia* sp. and the remains of three unidentified fish from a True's beaked whale, stranded in South Africa; in 2004, a True's beaked whale stranded in São Sebastião (Brazil) presented some anthropogenic debris but no food remains (Souza *et al*. 2005); 7 yr later another True's beaked whale stranded at Waiatoto Spitz (New Zealand), again, unidentified squid beaks and fish remains were found (Constantine *et al*. 2014). To the authors' knowledge, all True's beaked whales stranded and examined in the North Atlantic region did not contain identifiable prey items in their stomachs.

In 2013 three True's beaked whales (two adult females and a female calf) were reported stranded on May 12 (TBW 1/13; 55.35°N, 7.34°W), 13 (TBW 2/13; 55.29°N, 7.29°W) and 27 (TBW 3/13; 53.41°N, 10.14°W) on the west and northwest coast of Ireland, http://www.iwdg.ie) (Lusher *et al*. 2015) and recovered for postmortem examination following standard protocols. Whales stranded on the northwest coast (TBW 1/13 and TBW 2/13) were in a fresh condition allowing a thorough postmortem examination, whereas TBW 3/13 (stranded on the west coast) was in a moderate state of decomposition and a minimum postmortem examination was carried out. Two individuals (TBW 1/13 and TBW 2/13) were closely related and were most likely a mother and her calf, whereas the whale which stranded 2 wk later belonged to a different maternal lineage (Lusher *et al*. 2015).

Full digestive tracts, from mouth to anus, were collected from all individuals to study diet, parasites and ingestion of anthropogenic material (see Lusher *et al*. 2015). Contents were collected following the standard protocol established by Lusher *et al*. 2015. All prey remains were identified to the lowest possible taxon using published guides (*e.g*., Smale *et al*. 1995, Campana 2004, Tuset *et al*. 2008) and the UCC meso- and bathypelagic fish bones reference collection. Standard measurements of cephalopod beaks, otoliths, and bones were taken to reconstruct length and biomass of prey using back-calculation regression equations (Table S1). Prey category importance was evaluated using two widely used quantitative indices: percentage of prey by number (%N) and percentage of reconstructed biomass (%W). Digestive tracts from the two adult whales contained mostly remains of fish prey, while the calf only presented a small amount of a white-yellow substance in the stomach and unknown digested remains in its intestines (Table 1). 104 out of 106 prey items were fish, where 18% of the prey remains were identified using bones (*e.g*., vertebrae, cleithra, premaxillae) instead of otoliths. In addition, a large squid (~250–300 mm mantle length) was

Table 1. Prey species identified from both adults and the calf of True's beaked whales stranded in Ireland in 2013. N is number of prey occurring and W is the reconstructed biomass. Percentage importance by number (%N), and percentage importance by weight (%W) per individual are presented in brackets. Length (L) and weight (W) of fish is in millimeters and grams, respectively (see Table S1 for equations).

	True's beaked whale 1/13						True's beaked whale 3/13						TBW 2/13
	N (N%)	W (%W)	Length range	L average	W range	W average	N (%N)	W (%W)	Length range	L average	W range	W average	N
Anguilliformes													
Nemichthys scolopaceus							8 (25.8)	900.8 (24.4)	65[a]	65	112.6	112.6	
Osmeriformes													
Microstoma microstoma	2 (2.7)	48.1 (0.6)	161.7–168.7	165.2	22.6–24.5	24.1							
Xenodermichthys copei	1 (1.3)	24.8 (0.3)	—	—	—	—							
Stomiiformes													
Argyropelecus aculeatus	1 (1.3)	6.6 (0.1)	63.3	—	6.6	—							
A. hemigymnus	2 (2.7)	8.7 (0.1)	48.9–60.4	54.6	3.0–5.7	4.4							
A. olfersii	1 (1.3)	30.1 (0.4)	103.8	—	30.1	—							
Argyropelecus spp.	12 (16.0)	91.1 (1.1)	43.1–95.1	63.3	2.0–23.0	7.6	3 (9.7)	145.6 (4.0)	105.1–136.2	119.9	31.3–69.2	48.5	
Sternoptyx spp.	1 (1.3)	12.4 (0.2)	—	—	—	—	1 (3.2)	101.7 (2.8)	132.5	—	101.7	—	
Astronethes spp.							1 (3.2)	6.6 (0.2)	11.0	—	6.6	—	
Myctophiformes													
Benthosema glaciale							2 (6.5)	3.6 (0.1)	54.7–56.6	55.6	1.7–1.9	1.8	
Cerastocopelus maderiensis	1 (1.3)	2.1 (<0.0)	—	—	—	—							
Diaphus spp.							1 (3.2)	0.4 (<0.0)	28.7	—	0.4	—	
Lampanyctus ater							1 (3.2)	2.1 (<0.0)	72.4	—	2.1	—	
Lampanyctus macdonaldi							5 (16.1)	11.7 (0.3)	58.6–83.8	73.7	1.0–3.4	2.3	
Lampanyctus spp.	19 (25.3)	118.7 (1.4)	74.7–122.7	97.9	2.3–12.3	6.3							

(Continued)

Table 1. *(Continued)*

	True's beaked whale 1/13						True's beaked whale 3/13						TBW 2/13
	N (N%)	W (%W)	Length range	L average	W range	W average	N (%N)	W (%W)	Length range	L average	W range	W average	N
Myctophum punctatum							1 (3.2)	1.5 (<0.0)	34.6	—	1.5	—	
Notoscopelus kroyeri	2 (2.7)	17.9 (0.2)	97.1–99.2	98.1	8.6–9.2	8.9							
Notoscopelus spp.	1 (1.3)	29.6 (0.4)	—	—	—	—							
Unknown Myctophidae							1 (3.2)	10.7 (0.3)	100.8	—	10.7	—	
Gadiformes													
Coelorhinchus spp.	1 (1.3)	448.5 (5.4)											
Halargyreus johnsoni	3 (4.0)	701.5 (8.5)	193.7–307.8	262.7	85.0–341.1	233.8							
Gadiculus argenteus thori	4 (5.3)	1.1 (<0.0)	62.2–87.0	73.2	0.1–0.5	0.3							
Micromesistius poutassou	1 (1.3)	867.4 (10.5)	—	—	—	—							
Stephanoberyciformes													
Scopelogadus spp.	3 (4.0)	24.4 (0.3)	51.4–78.0	63.8	3.7–14.1	8.1							
Cetomimiformes													
Rondeletia loricata	1 (1.3)	17.0 (0.2)	—	—	—	—							
Scorpaeniformes													
Helicolenus dactylopterus	1 (1.3)	3.6 (<0.0)	—	—	—	—							
Perciformes													
Aphanopus carbo	13 (17.3)	5,838.1 (70.4)	267.8–974.2	697.7	19.7–1,107.9	449.1	2 (6.5)	2,503.1 (67.9)	877.6–1,110.1	993.9	777.6–1,725.5	1,251.6	
Unknown fish	3 (4.0)	NA	—	—	—	—	5 (16.1)	NA	NA	—	—	—	1
Cephalopod	2 (2.7)	NA	—	—	—	—							
Milk													Yes
Total	75	8,291.7		94.6		118.5	31	3,687.7		143.4		141.8	1

[a]Half of maximum length reported in FISHBASE was used.

found in the mouth of TBW 1/13 at the time of the stranding, although it was not recovered.[2] A minimum of 23 deep sea fish species from at least 13 families were identified (Table 1). Myctophidae was the most abundant group (32%N), with *Lampanyctus* (Bonaparte, 1840) species making up to 83%N followed by *Notoscopelus* (Günther, 1864) species (13%N). The second most abundant family was Sternoptychidae (18%N), and *Argyropelecus* species was the only identified fish within the family. The third most abundant family in terms of number was Trichiuridae (14%N), with the black scabbard fish (*Aphanopus carbo*, Lowe 1839) as the only species identified (Table 1, Fig. 1a). Twenty-four out of 90 fish identified (27%) were typically bathypelagic prey, while 16 (18%) were typically mesopelagic prey. The remaining 55% can be considered meso- and bathypelagic fish (Table 1, Fig. 1a). The anguilliform *Nemichthys scolopaceus* (J. Richardson 1848) was found in significant numbers (26%N, 24%W) in the female stranded in Galway (Table 1). In general, over 50% of the prey found in the stomachs of the True's beaked whales in this study comprised myctophids, sternoptychis, and stomiids, and it is reasonable to assume that they were feeding within the DSL. Furthermore, most of the prey eaten by the True's beaked whales in this study were meso- or bathypelagic fish usually found in waters 200–800 m deep, suggesting that this species forage on mid-water pelagic fish that they catch either during deep dives or feeding at night when prey undertake vertical migrations.

Prey length ranged from 11 mm (*Astronesthes* spp., J. Richardson 1845) to 1,100 mm (black scabbardfish). Most of prey items were smaller than 120 mm, with 75% below 100 mm length (Fig. 1b, Table 1). Five larger prey were found in TBW 1/13: they were the gadoids blue whiting (*Micromesistius poutassou*, A. Risso 1827), rattail (*Coelorhinchus* spp., Giorna 1809), and the slender codling (*Halargyreus johnsonii*, Günther 1862), which ranged between 200 and 300 mm in length. In TBW 3/13, the larger prey items were two black scabbardfish (878 and 1,110 mm). Most of the biomass contribution (70%) was made by black scabbardfish. Gadiformes contributed 17% of the biomass with three main species, blue whiting, slender codling, and the rattail contributing the most (Table 1). The occurrence of larger black scabbardfish in both adults is of note. Black scabbardfish is a deep-water species of high commercial interest in the Northeast Atlantic, showing size and maturity segregation, with large individuals found in spawning areas in Madeira and smaller, immature individuals west of the British Isles (Riberio Santos *et al.* 2013*a*). In this area, black scabbardfish are caught at depths between 700 and 1,000 m and may feed in the DSL (Ribeiro Santos *et al.* 2013*b*). The reconstructed size range of black scabbardfish is consistent with sizes reported from the west of Ireland (*e.g.*, Ribeiro Santos *et al.* 2013*a, b*), although the smaller reconstructed size is smaller than previously reported for this area. Reconstructed size and biomass is generally obtained using back-calculation equations constructed from fish otoliths/bones and fish size/weight that have not suffered any digestion process; otoliths and bones of black scabbardfish were slightly eroded and the original size and biomass of prey may be greater. The long, cylinder shape of black scabbardfish may be suitable for True's beaked whale to consume given their elongated beaks and using their assumed feeding mechanism of suction feeding.

Identification of a large number of species follows the same pattern as in other studies which suggest that True's beaked whales have a generalist foraging behavior

[2]Personal observation by Mr. Emmett Johnson, National Parks and Wildlife Service; e-mail: emmett.johnston@ahg.gov.ie; 15 May 2013.

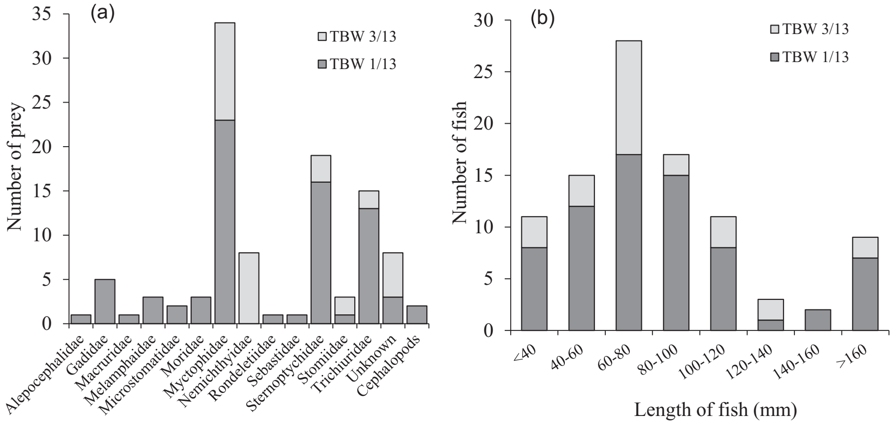

Figure 1. Number of prey items found in digestive tracts of the two adults of True's beaked whales stranded in Ireland in 2013 (a) in numbers separated by family, and (b) by reconstructed length.

(MacLeod *et al.* 2003, MacLeod 2005). This kind of strategy might be related to the amount of time and energy that this whale species spends during foraging dives or to prey behavior. If these predators undertake high energy demanding dives, high energy density prey would be expected within their diet (Spitz *et al.* 2010). In this case, the average energy density of the prey is 3–4 kj/g (Spitz *et al.* 2010). Low energy prey might be useful if they are aggregated in a small area reducing the energy consumption on finding and catching the prey. It is not clear whether mesopelagic fish form schools (*e.g.*, Saunders *et al.* 2013) or gather in high concentrations (Irigoien *et al.* 2014), but in either case, the concentration of these fish will increase the encounter chances, especially for smaller prey.

Debris was found in the stomachs of both adult females (TBW 1/13 and TBW 3/13, see Lusher *et al.* 2015) within the connecting chambers and the main stomach (Fig. S1). Previously, two other True's beaked whales that stranded in Ireland (Gassner *et al.* 2005) and Brazil (Souza *et al.* 2005) contained debris in their digestive tracts. Presence of debris in the digestive tracts of cetaceans has been suggested to cause internal injuries and malnutrition (Baulch and Perry 2014); however, the True's beaked whales that stranded in Ireland did not have any injuries in their digestive tracts and they seemed to be in good nutritional state based on their dorsal blubber thickness (30 mm, TBW 1/13; 23 mm, TBW 2/13; and 39 mm, TBW 3/13).

Many causes of death have been reported for cetaceans from pathological diseases to anthropogenic activities, but intense anthropogenic sound seems to be the most important threat to *Mesoplodon* spp. such as True's beaked whale (Cox *et al.* 2006). It is generally accepted if these whales are hunting when an acoustic impact occurs, they will have prey items in their stomachs, while if they are sick their stomachs may be empty. Both adult whales in this study had many prey remains present in their stomachs, indicating a recent meal. Whereas, the calf, which stranded 2 d after the mother, had a fluid consistent with milk in its stomach, suggesting that its last meal was at least 2 d before it died.

Superficial injures were recorded on the skin of TBW 1/13, with two large healed marks (at the right pectoral fin, 10 cm length, and anterior to the genital slit, 25 cm length), and two scratches on the right lateral side (140 cm length) of the whale associated with the separation of the muscular fascia and the subcutaneous blubber. Hemorrhagic areas were found in muscle and the abdominal cavity as well as in different organs. Lungs presented a multifocal severe and interstitial emphysema, and a multifocal bronchospasm and bronchopneumonia associated with parasite cysts. Several cestode cysts of the genus *Phyllobothrium* spp. (Cestoda: Tetraphyllidea) were found in the retroperitoneal blubber, as well as peritoneal-associated *Monorygma grimaldii* cysts (Cestoda: Tetraphyllidea). Bronchopneumonia was associated with a parasitic infection of *Phyllobothrium* spp. Intestines were inflamed and the uterus was enlarged, suggesting recent pregnancy. Two corpora lutea/albicantia were detected on the left ovary. Mammary glands contained milk, suggesting that this individual was lactating. Histological analysis revealed that this whale suffered a lung fat embolism (grade 1), and presented a severe multifocal interstitial emphysema, with bullous multifocal subpleural emphysema, and alveolar oedema. Presence of bubbling was detected on the surface of the kidneys and histological analysis revealed a multifocal interstitial chronic linfociti nephritis. The liver was too decomposed to carry out histological analysis and no histological analysis of other organs was carried out. This microscopic analysis suggests the whale stranded alive and died as a result of cardiorespiratory failure after beaching. The low grade of lung fat embolism (grade 1) detected in TBW 1/13 can be associated with soft tissue trauma during the stranding event (Fernandez *et al.* 2005). This animal showed chronic multiorganic pathologies, including a parasitic pneumonia, which indicates an unhealthy condition that may have played a role in the cause of standing and death after beaching. This whale did not show systemic gas embolism as has been described in beaked whale mass strandings linked to sonar (Fernandez et al. 2005, Cox *et al.* 2006, D'Amico *et al.* 2009).

The calf (TBW 2/13) presented bruising consistent with a live stranding. Free yellow fluid in the interconnection of the scapula and pectoral bones was noted, with a possible localized infection. The abdominal cavity contained blood and the intestines were inflamed. A small number of nematode parasites and cysts were found in the lungs. Ovaries were undeveloped and presented a lobed appearance. It is reasonable to suggest that the calf stranding occurred after separation from the mother.

Macroscopic examination of the organs of the second adult female (TBW 3/13) did not show any abnormality. Few *M. grimaldii* cysts were detected in the peritoneal cavity. The digestive tract presented some anomalies. The main stomach chamber lining had some small areas with black discoloration. Intestines were found to be inflated at the time of postmortem examination and showed a midgut volvulus or contortion with the outside walls attached. Gross examination of the internal area showed an enlargement and possibly fibrotic intestinal lining. Samples for histopathological analysis were not collected preventing an accurate determination of cause of dead. The time difference in strandings excludes these events being considered a mass stranding. However, the fact that both adults had full or partly digested prey suggests that they were foraging prior to death and that death was likely sudden. Unfortunately, no cause of death could be determined.

Foraging ecology studies allow us to understand variations in organisms' habits, and obtain some insights into their behavior, as they might change their dietary preferences, and therefore change their trophic interactions. Information obtained in this study represents an important contribution to future research on the trophic ecology of True's beaked whale. Further samples need to be collected to confirm the piscivorous diet of this beaked whale species, in combination with other approaches such as stable isotope analysis.

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Supporting Information

The following supporting information is available for this article online at http://onlinelibrary.wiley.com/doi/10.1111/mms.12430/suppinfo.

Figure S1. Diagram of the different compartments of a True's beaked whale stomach (after Mead 2007). The different aspects of the internal stomach tissue is shown in the drawing and pictures. Macroplastic items found in both adult females are shown. (A) 7.1 × 2.2 cm shotgun cartridge found in the main stomach of TBW 3/13 and (B) 4.2 × 3.1 cm fragment found in the connecting chambers of TBW 1/2013 (see Lusher *et al.* 2015).

Table S1. Regression equations used for estimating fish sizes and biomass. SL: standard length, TL: total length, Lpa: pre-anal length, FW: total weight, OL: otolith length, OW: otolith width; all size measurements in mm and weight in grams. Sources are as follows: Ba: Battaglia *et al*. (2010), Do: Dorel (1986), Fo: Froese *et al*. (2014), F&E: Fock and Elrich (2010), Ha: Härkonen (1986), Ja: Jaramillo *et al*. (2014), Mc: McCarthy *et al*. (2011), Ol: Olafsdottir e*t al*. (2016), Petrakis *et al*. (1999), Sa: Santos *et al*. (2007), UCC: UCC reference collection. For those otoliths where regressions were not available, the length estimation was obtained by extrapolation from the UCC mesopelagic fish bones reference collection.