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Species identity and human consumption of beaked whales in the Gilbert Islands, Republic of Kiribati

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Abstract

We investigated the species identity and local use of cetaceans on the Gilbert Islands, Republic of Kiribati. Working with the Kiribati Ministry of Environment, Lands and Agricultural Development and Fisheries Division, we visited the islands of Tarawa, Tabiteuea (North), Butaritari and Onotoa from June to July 2009, and collected 24 bones, bone fragments or teeth attributed to recent strandings. The mitochondrial DNA control region or cytochrome *b* was successfully amplified from 12 bones or bone fragments and used to identify four species: *Mesoplodon* sp. representing a new species or subspecies of beaked whale, the dense-beaked whale *Mesoplodon densirostris*, Cuvier's beaked whale *Ziphius cavirostris* and the pygmy sperm whale *Kogia breviceps*. This is the first confirmed identification of the dense-beaked, Cuvier's and pygmy sperm whales from the Gilbert Islands. All specimens were reportedly used for human consumption.

Introduction

The Gilbert Islands are a north to south chain of 16 atolls and coral islands distributed across the equator in the central Pacific Ocean. Along with the largely uninhabited Line Islands and Phoenix Islands, the Gilbert Islands and Banaba (i.e. Ocean Island) form the Republic of Kiribati, with its capital on Tarawa (in the Gilberts). Although the land area of Kiribati is only 726 km^2, its exclusive economic zone (EEZ) is over 3.4 million km^2, an area slightly larger than the total land area of India (Lanteigne, 2012). Despite this vast EEZ, little is known about the diversity or exploitation of cetaceans in the waters of Kiribati. The region around the Gilbert Islands (formerly known as the Kingsmill Group) was recognized by British and American whalers for its abundance of sperm whales *Physeter macrocephalus* (Beale, 1839; Smith *et al*., 2012) and 'whales' were hunted by locals during the first half of the 19th century (Wilkes, 1845). The hunting of 'porpoise' (actually dolphins or other small cetaceans) for human consumption was reported during the 20th century colonial history of the Gilbert Islands (Turbott, 1949) and the traditional shamanistic practice of 'porpoise calling' (again, dolphins or other small cetaceans), suggests a long history of such exploitation (Grimble, 1952). Drive hunting of small cetaceans for human consumption in Kiribati was reported until at least the early 1990s, including melon-head whales *Peponocephala electra* (Brownell, Nowacek & Ralls, 2008) and other unidentified delphinids (Robards & Reeves, 2011), but there are no historical or contemporary records of the exploitation of beaked whales.

Consequently, it came as a surprise when molecular identification of dried whale meat served at a local feast in July 2003 on Tabiteuea Island (southern Gilbert Islands, 1°12′ 14" S, 174°44′ 51" E), provided evidence of a new species (or subspecies) of beaked whale, *Mesoplodon* sp. (Dalebout *et al*., 2007). At the time, members of the local community reported that the meat came from seven 'long whales' driven ashore in the shallow water of the atoll in October 2002 (R. Grace, pers. comm.). Here we report on efforts to investigate exploitation of this unknown species of beaked whales and to better document cetacean diversity in the Gilbert Islands by collecting cetacean artifacts, including dried meat, teeth and bones, during visits to the islands in 2009. Our findings, based on molecular identification of discarded bones and bone fragments, contribute to what is otherwise a dearth of information on cetacean diversity and local use around the Gilbert Islands.

Methods

Collection of bones from the Gilbert Islands

Two coauthors (A. H. and G. S.) visited Bairiki, on the Island of Tarawa, in April 2006 and again in June 2008, to arrange permits from the Kiribati Ministry of Environment, Lands and Agricultural Development (MELAD) and logistical support for visits to the outer islands. From 18 June to 14 July 2009, one of us (A. H.), accompanied by an interpreter and a local Biodiversity Officer, visited three of the outer Gilbert Islands: Onotoa Island, Tabiteuea North and Butaritari Island (Fig. 1). On each island, village elders were shown a copy of *Whales and Dolphins of New Zealand and Australia, an Identification Guide* (Baker, 1999) and asked about local use of whales and dolphins and the location of any meat or skeletal remains.

DNA extraction and species identification

Bones and teeth collected in the Gilbert Islands were exported to New Zealand (with appropriate permits) and stored at the School of Biological Sciences, University of Auckland. The bones were isolated from potential contamination with DNA from other samples of whales and dolphins. The primary laboratory analysis was conducted in a laboratory isolated from cetacean DNA in the School of Biological Sciences, University of Auckland. Each bone was drilled in several areas with a 1.5-mm drill bit as described in Pichler, Dalebout & Baker (2001) and approximately 50 mg of material was collected on sterile foil. Total genomic DNA was isolated from the powdered bone using a Qiagen Blood and Tissue Extraction Kit (Qiagen, Inc., Valencia, CA, USA), following the manufacturer's protocols. Amplification via the polymerase chain reaction (PCR) and sequencing of the mitochondrial (mt) DNA control region (D-loop) and cytochrome *b* gene generally followed protocols described in detail by Dalebout *et al*. (2004). For the control region, a 500-bp fragment was first amplified using the primers M13Dlp1.5 and Dlp5R (Dalebout *et al*., 2004). For most samples (except KI024), this was followed by a nested amplification of a 300-bp fragment using primers M13Dlp1.5 and Dlp4R (Dalebout *et al*., 2004). For amplification of the cytochrome *b* gene, DNA was first concentrated using an Amicon Ultra-0.5 mL centrifugal filter (Millipore, Billerica, MA, USA). Non-nested amplification was then attempted for a 260-bp fragment using the primer pairs, Cyb140-160F, 5′-GATACCTRCACGCAAAYGGG GC-3′ and Cyb305-328R (5′-CACCTCAGAATGATAT TTGTCCTC-3′ (M. L. Dalebout, unpubl. data), or a 190-bp fragment using M13CybMLDF (Dalebout *et al*., 2002) and Cyb196-217R, 5′- AGCCGTAATATAGTCCA CGTCC-3′ (M. L. Dalebout, unpubl. data). PCR products

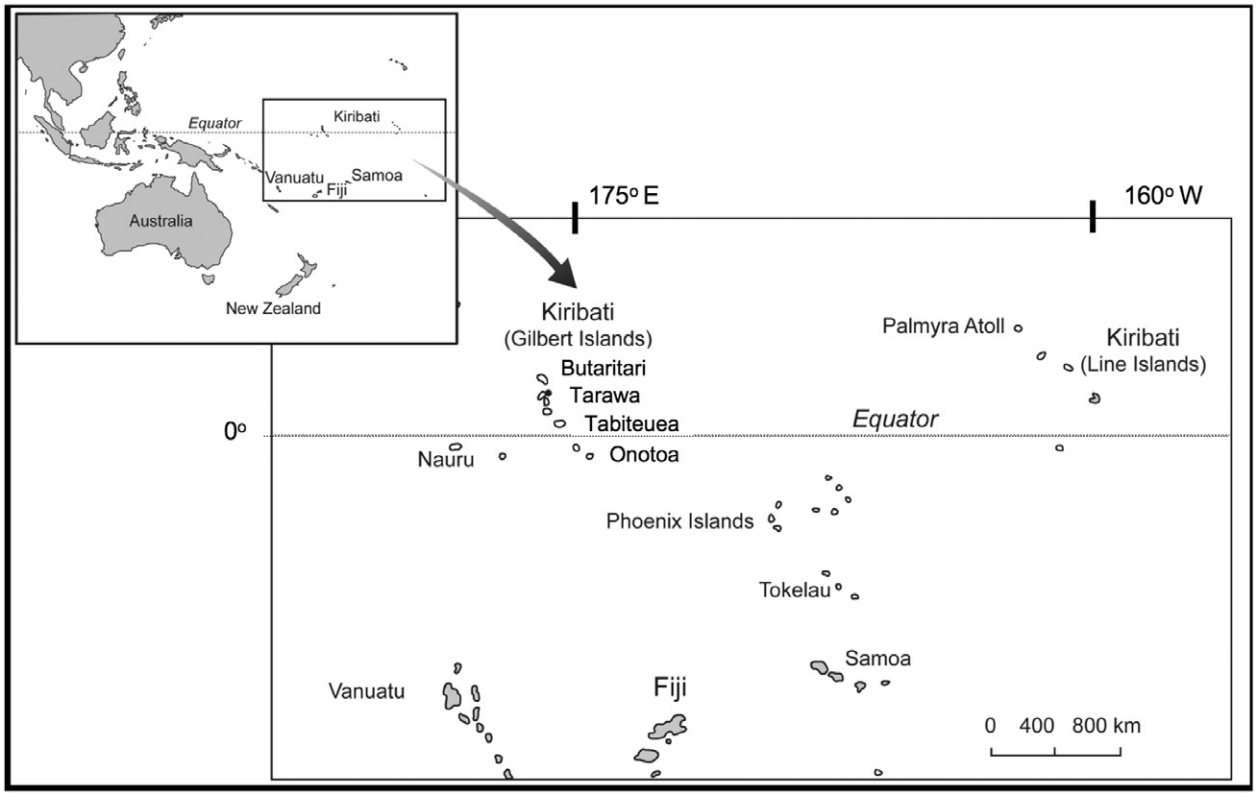

Figure 1 The location of the Gilbert Islands, Republic of Kiribati, relative to other features of the central Pacific Ocean. Shown are the Island of Tarawa (with the capital city of Bairiki) and the three outer islands (Butaritari, Tabiteuea and Onotoa) surveyed in this study.

were purified using Agencourt AMPure XP and sequenced by standard BigDye 3.1 protocol (Applied Biosystems Inc., Foster City, CA, USA) on an ABI 3130 Genetic Analyzer (Applied Biosystems Inc.). Sequences were edited manually using the program GENEIOUS (Drummond *et al*., 2009).

Based on initial success with sequencing and identification, a subset of six bones was transferred to an ancient DNA facility in the Department of Anthropology, University of Auckland for independent replication of DNA extraction and amplification (by coauthor J. R.). The methods for DNA extraction used for these independent bone extractions were modified from that of Matisoo-Smith *et al*. (1997). The methods for amplification and sequencing follow those described earlier.

The species were identified by submitting control region or cytochrome *b* sequences to the web-based program *DNA-surveillance* (Ross *et al*., 2003), as well as by Basic Local Alignment Search Tool (BLAST) searches of GenBank, and by comparison with the sequences from other specimens of *Mesoplodon* sp., as presented in Dalebout *et al*. (2007) and now available as GenBank JX470545. Species identification was based on the grouping of sequences from the bones with the reference sequences, using a neighbor-joining reconstruction as implemented in *DNA-surveillance* (Ross *et al*., 2003), and on matching with GenBank sequences by BLAST search.

Results

Local use and skeletal remains

Tarawa Island

Attempts to collect information on cetaceans from around Tarawa proved unrewarding – there seemed to be little local or institutional knowledge of whales or dolphins. Of some interest was the whaling try pot in use as a water cistern, presumably an artifact of 19th century whaling contact (see Supporting Information Fig. S1). No cetacean artifacts or skeletal material were collected from Tarawa Island.

Tabiteuea North Island

Tabiteuea Island has an extensive coastline and discussions with the locals suggested that strandings were common. Unfortunately, the teeth and bones are considered by the islanders to have no value and are often used as fuel for fires. There was no evidence of remains from the seven whales reportedly killed in 2002 and which provided the dried meat served at the feast in July 2003 (Dalebout *et al*., 2007). The islanders described a recent stranding of two beaked whales, about 3–4 m in length, with two teeth about the size of an index finger. When shown a copy of the identification guide (Baker, 1999), the elders could not make a positive species identification, but described the whale as looking like a cross between Andrew's beaked whale *Mesoplodon bowdoini* and Longman's beaked whale *Indopacetus pacificus*. The islanders reported that the teeth were sold to fisherman from a foreign vessel. No bones could be found and no samples were collected from Tabiteuea North Island.

Butaritari Island

Villagers reported incidental strandings of what were described as 'minke whales' and sperm whales, based on illustrations in the identification guide (Baker, 1999). The teeth and ribs from a sperm whale were displayed by the islanders (see Supporting Information Fig. S1). Islanders also reported the stranding of an unusual whale of approximately 4 m in length, a pointed jaw and a full set of teeth in its lower jaw. Although the stranding had occurred in 2008, the jaw and teeth were still available and were contributed to the collection, along with a sperm whale tooth.

Onotoa Island

The islanders reported that a whale stranded sometime in the previous two months (*c.* April, 2009) at the northern tip of island. The whale was flensed and the meat was dried for eating. The bones were left on site as they are not considered of practical use by the islanders. The whale was described by an elder who participated in the flensing, as about 4–5 m in length, dark in color, lacking baleen or teeth and with a head like a swordfish. When shown a copy of the identification guide (Baker, 1999) the elder described the whale as looking like a cross between Andrew's beaked whale and Longman's beaked whale, similar to the description by elders on Tabiteuea North Island. A number of vertebrae were collected from the site of the stranding and beachcombing revealed older material, including one partial skull and other vertebrae. Some strips of dried meat and 21 skeletal samples were collected from Onotoa Island: 11 vertebrae, 9 bone fragments, and one definite skull fragment.

Species identification of bones

In total, the June to July 2009 expedition to the outer islands was able to collect 24 samples, including one partial skull and a partial lower jaw with teeth and some dried skin attached, and one sperm whale tooth (Table 1). The strips of dried meat prepared for human consumption were also collected on Onotoa, but were destroyed by rats during storage in Tarawa and are not included in Table 1. The majority of samples ($n = 21$) were vertebrae and likely skull fragments collected from Onotoa Island.

Of the 24 samples of bone or teeth, 12 yielded DNA of sufficient quality for PCR amplification and sequencing of mtDNA fragments from the control region and/or cytochrome *b* (Table 1). DNA from six of these 12 samples were independently extracted, amplified and sequenced by the collaborating laboratory. Sequences from seven of the 12 samples were of sufficient length to confirm identity of four species with 100% bootstrap support using *DNA-surveillance* (see Supporting Information Fig. S2). These sequences (six control region and two cytochrome *b*) have

Table 1 Cetacean material collected from the outer islands of the Gilbert group, in the Republic of Kiribati, with species identification based on mtDNA control region (DLP) or CYB sequences

Sample code	Description	Species identification	Island	DLP	CYB
KI001	Fragment	Failed	Onotoa		
KI002*	Fragment	*Mesoplodon densirostris*	Onotoa	**310 bp***	**219 bp***
KI003	Fragment	Failed	Onotoa		
KI004	Vertebra	*Mesoplodon* sp.	Onotoa		255 bp
KI005	Vertebra	Failed	Onotoa		
KI006	Fragment	Failed	Onotoa		
KI007	Fragment	Failed	Onotoa		
KI008	Vertebra	*Mesoplodon* sp.	Onotoa	293 bp	
KI009*	Vertebra	*Ziphius cavirostris*	Onotoa		**236 bp***
KI010	Vertebra	Failed	Onotoa		
KI011*	Vertebra	*Mesoplodon* sp.	Onotoa	**293 bp***	141 bp
KI012*	Vertebra	*Mesoplodon* sp.	Onotoa	**293 bp***	178 bp
KI013	Vertebra	*Mesoplodon* sp.	Onotoa	239 bp	
KI014	Fragment – rib?	Failed	Onotoa		
KI015	Vertebra	*Mesoplodon* sp.	Onotoa	239 bp	
KI016	Vertebra	Failed	Onotoa		
KI017	Vertebra	*Mesoplodon* sp.	Onotoa		171 bp
KI018*	Fragment – skull?	*Mesoplodon densirostris*	Onotoa	**309 bp***	109 bp
KI019*	Skull fragment	*Mesoplodon densirostris*	Onotoa	**311 bp***	136 bp
KI020	Fragment – skull?	Failed	Onotoa		
KI021	Fragment – scapula?	Failed	Onotoa		
KI022[a]	Tooth – sperm whale	Failed	Butaritari		
KI023	jaw – teeth	*Kogia breviceps*	Butaritari		
KI024	jaw – dried skin	*Kogia breviceps*	Butaritari	**487 bp***	
UKIRI[b]	dried meat	*Mesoplodon* sp.	Tabiteuea	**300 bp**	

[a]The sperm whale tooth was identified from its appearance (see Supporting Information).

[b]The sample UKIRI is represented by a partial fragment of the sequence described in Dalebout *et al.* (2007) and now available as GenBank JX470545.

Mesoplodon sp. refers to an unrecognized species or subspecies of beaked whale first identified by Dalebout *et al.* (2007). For samples where DNA extraction or subsequent polymerase chain reaction amplification of mtDNA fragments were not successful, species identification is listed as 'failed'. Sequence lengths in bp are shown for successful DNA identification. An asterisks (*) indicates the six samples for which DNA extraction and amplification were replicated by an independent laboratory (see text). Sequences in bold are included as Supplementary Material and submitted to Dryad.

bp, base pair; DLP, D-loop or control region; CYB, cytochrome *b*; mtDNA, mitochondrial DNA.

been submitted to GenBank as KC540691-KC540698. Sequences from the other five samples were sufficient in length for likely species identification, based on internal matching to the specimens with confident identification. Given the low levels of intra-specific diversity in many beaked whales (Dalebout *et al*., 2004) and the quality of the sequence fragments, there was not sufficient evidence to determine if the bones represented more than one individual of each species.

Sequences from seven of the vertebrae showed an exact (or near exact) match for the control region and/or cytochrome *b* sequences, with the dried whale meat collected on Tabiteuea Island in 2003 (sample code: UKIRI, Dalebout *et al*., 2007). Of these seven vertebrae, two yielded control region sequences of sufficient quality for confident species identification using *DNA-surveillance* (see Supporting Information Fig. S2a). Although recent investigation has matched the mtDNA sequences of the samples from Tabiteuea Island with the holotype of *Mesplodon hotaula* (see Discussion), we refer to this taxon here as *Mesoplodon* sp., pending a comprehensive review and proposal for revised taxonomic ranking.

Three of the bone fragments (including a very dense fragment thought to be part of a skull) were identified as a dense-beaked whale *Mesoplodon densirostris.* All three of these provided confident identification based on control region or cytochrome *b* sequences. A single vertebra was identified as a Cuvier's beaked whale *Ziphius cavirostris*, based on a cytochrome *b* sequence (see Supporting Information Fig. S2b). The partial lower jaw from Butaritari was confirmed to be from a pygmy sperm whale *Kogia breviceps* based on control region sequences from both teeth and dried skin. We were unsuccessful in extracting DNA from the large tooth from Butaritari, but, based on size and shape, it is clear that it represents a sperm whale.

Discussion

Molecular taxonomy of beaked whales

The beaked whales (Family: Ziphiidae) are among the least known of all vertebrate groups. Found in deep oceanic waters, typically far from shore, several species were initially described only from partial skeletal remains found stranded on remote islands and shorelines. Previously thought to include 20 extant species, a recent survey of the molecular systematics of this family resulted in the discovery of a new species, *Mesoplodon perrini*, first identified by phylogenetic analysis of mtDNA sequences (Dalebout *et al*., 2002). Development of a comprehensive molecular taxonomy of the family Ziphiidae resulted in two additional molecular discoveries (Dalebout *et al*., 2004). First, previously misidentified specimens from South Africa were linked to the holotype of Longman's beaked whale from tropical north Queensland, Australia, providing the first evidence of the physical appearance of this species (Dalebout *et al*., 2003). Second, a single tooth and partial skull held in New Zealand institutions were linked to the holotype of Bahamonde's beaked whale *Mesoplodon bahamondi* Reyes, van Waerebeek, Cárdenas and Yáñez, 1995 held in the Museo Nacional de Historia Natural in Santiago, Chile. This finding confirmed the genetic distinctiveness of the species and uncovered a prior description of this species as *Mesoplodon traversii*, dating back to the recovery of a tooth from the Chatham Islands of New Zealand in the 1870s (van Helden *et al*., 2002). Recently, a female and juvenile beaked whale found stranded in New Zealand were identified from mtDNA as *M. traversii*, providing the first evidence of the physical appearance of this species (Thompson *et al*., 2012a). Together, these discoveries and taxonomic revisions demonstrate the power of integrating information from traditional museum specimens and morphological descriptions of stranded specimens with molecular taxonomy for identifying rare and cryptic species of cetaceans, particularly beaked whales (Baker *et al*., 2003; Dalebout *et al*., 2004; Thompson *et al*., 2012b).

Molecular taxonomy also provides a powerful tool for monitoring species exploitation (Baker, 2008), and through market surveys, of documenting species diversity (Baker *et al*., 2006). This can be particularly important where traditional use of cetaceans, including consumption, is increasing (Robards & Reeves, 2011), or where cetacean species, once taken only as 'bycatch', have become targets of artisanal fisheries (Clapham & Van Waerebeek, 2007). However, because of their pelagic habitat and relatively low abundance, beaked whales have seldom been the target of commercial or traditional exploitation (Mead, 2009).

Here we used molecular taxonomy, as implemented in the web-based program *DNA-surveillance* (Ross *et al*., 2003), to confirm the genetic distinctiveness and document local use of a new species or subspecies of beaked whales, first described from dried strips of meat served at a village feast on Tabiteuea Island in 2003 (Dalebout *et al*., 2007). At the time, analyses of mtDNA control region and cytochrome *b* sequences showed the meat came from an unknown member of the beaked whale genus *Mesoplodon*. Subsequent collaborative comparisons showed an exact match of the Tabiteuea sequences with two specimens collected in 2005 on Palmyra Atoll Wildlife Refuge, 2600 km to the northeast (Dalebout *et al*., 2007). Although grouping most closely with *Mesoplodon ginkodens* in phylogenetic reconstructions, the sequences, now including those reported here from Onotoa Island (see Supporting Information Fig. S2a), show a degree of genetic divergence consistent with species-level classifications among mesoplodons (Dalebout *et al*., 2007). Further investigation (Dalebout *et al*., 2012) has matched the mtDNA of these specimens with sequences from the holotype specimen of *M. hotaula*, initially described from a single specimen held at the National Museum, Colombo, Sri Lanka (Deraniyagala, 1963). Shortly after this description, however, *M. hotaula* was synonymized with *M. ginkgodens* (Moore & Gilmore, 1965), which had been described just a few years earlier (Nishiwaki & Kamiya, 1958). With the addition of these new specimens, the ranking of *M. hotaula* as a species or subspecies requires reconsideration, but remains undecided, pending further morphological and genetic comparisons with the small number of existing *M. ginkgodens* specimens (Dalebout *et al*., 2012).

Local use of cetaceans by Gilbertese

We encountered no obvious evidence of the shamanistic practice of 'porpoise calling' documented by Sir Author Grimble during his experiences in the Gilbert Islands prior to World War II (Grimble, 1952). However, on each of the three islands of the Gilbert group, discussions with the elders (through translators with MELAD) confirmed the ongoing local use of cetaceans, including as food for human consumption. Descriptions offered by islanders suggested that cetaceans taken for human consumption are found stranded or are driven ashore if they enter the shallows near the atolls. At the time the meat of the *Mesoplodon* sp. was offered to the visitors of Tabiteuea Island in July 2003, villagers reported that they regularly (several times a year) chased and killed 'long whales' when they come into the shallow waters of the nearby lagoon (R. Grace, pers. comm.). To our knowledge, the drive hunting of beaked whales has never before been reported anywhere in the world. Further investigation is required to document the circumstances under which the beaked whales enter the shallow water of the atolls, and the methods used by the Gilbertese for herding the whales.

Historically, the hunting of small cetaceans was important to island and atoll societies throughout the Pacific Islands, representing the cultural divisions of Micronesia, Polynesia and Melanesia. Some islands with a documented history of hunting cetaceans include: the Mariana Islands (Costenoble, 1905), the Gilbert Islands (Grimble, 1952), Woleai Atoll in Federated States of Micronesia (Alkire, 1968), the Hawaiian Islands (Wilkes, 1845), the Marquesas Islands in French Polynesia (Reeves *et al*., 1999) and the Solomon Islands (Dawbin, 1966). Aside from food, the

teeth and bone from sperm whales and small cetaceans were an important component of traditional jewelry, often in the form of necklaces. These adornments also function to show social status such as rank and leadership (Neich & Pereira, 2004). Unfortunately, there appeared to be little or no cultural use of either the bones or teeth of cetaceans in the islands we visited and so few artifacts were available for confirming the full extent of species involved in subsistence use. In some villages, the bones are burned for fuel and, if teeth are recovered, these are sometimes sold to fishermen from passing foreign vessels.

Species diversity and threats

Our surveys and species identifications provide new, validated records of cetaceans around the remote Gilbert Islands in the Republic of Kiribati. Other than the recent report of *Mesoplodon* sp. (Dalebout *et al*., 2007) and the historical accounts of sperm whales (Townsend, 1935), there are few confirmed records of cetacean species in the Gilbert Islands (Reeves *et al*., 1999). None of the species that we identified were listed in a recent summary of cetaceans used for human consumption in Kiribati (Robards & Reeves, 2011). Our finding of further material from *Mesoplodon* sp. on Onotoa suggests that, like Palmyra Atoll in the Line Islands (Dalebout *et al*., 2007, 2012), the Gilbert Islands are likely to be a hotspot for this species of beaked whale, and raises concerns about its frequency of stranding or potential exploitation for local consumption. The reported large size of the group killed on Tabiteuea and the apparently regular occurrence near small islands (including Palmyra Atoll, Baumann-Pickering *et al*., 2010) suggest an unusual social organization and habitat preference for this previously undescribed species or subspecies.

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Supporting information

Additional Supporting Information may be found in the online version of this article at the publisher's web-site:

Figure S1 Whaling artifacts and cetacean bones collected during surveys of the Gilbert Islands from 18 June to 14 July 2009: (a) whaling 'try-pot', a remnant of 19th-century whaling contact; (b) vertebra (KI004, identified as *Mesoplodon* sp., an unrecognized species or subspecies) and rib bone (KI014, no DNA identification) collected on Onotoa Island; and (c) mandible, rib and teeth of a sperm whale on display in Butaritari Island. Photographs courtesy of Al Hutt.
Figure S2 Representative identification of mtDNA sequences from cetacean material collected from the outer islands of the Gilbert group, in the Republic of Kiribati, using reference sequences available through the web-based application, *DNA-surveillance* (Ziphiidae Vs4.3): (a) control region (DLP) of KI0o2, KI011, KI012, KI018 and KI019; (b) cytochrome *b* (CYB) of KI002 and KI009. The neighbor-joining tree reflects species-specific grouping with bootstrap values based on 500 replications. The program is available at http://www.cebl.auckland.ac.nz:9000/page/whales/title.
Table S1 Sequences of the mtDNA control region (DLP) used in species identification of bones from Gilbert Islands. Shown in FASTA format with sample code followed by primer information
Table S2 Sequences of the mtDNA cytochrome *b* used in species identification of bones from Gilbert Islands. Shown in FASTA format with sample code followed by primer information