Antarctic Minke Whales (Balaenoptera bonaerensis) near the Equator

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Introduction

The Antarctic minke whale (*Balaenoptera bonaerensis*) undertake seasonal migrations between feeding grounds in the Antarctic waters in the summer (south of 60°S), and breeding grounds in the tropical or temperate regions in the winter. Sighting data from the period 1976–1987showed that *B. bonaerensis moved* southward from the breeding areas by October-November, and that most of them had migrated into Antarctic waters by January (Kasamatsu *et al.*, 1995).

B. bonaerensis has been the main target of the whaling industry off Brazil where at least 14,000 specimens were caught (Zerbini *et al.*, 1997, Lucena, 2006). Only recently there was an effort of evaluating the relative abundance of *B. bonaerensis* and other baleen whales on their former breeding ground off Northeast Brazil (Andriolo *et al.*, 2010).

Here we report on two Antarctic minke whales stranded on the northern coast of Brazil in January and November 2007, respectively, and a third specimen that ventured the Amazon river in April 2009 (herein called whale#1, whale#2 and whale#3). Together, they represent the first concrete evidence of Antarctic minke whales on the North region of Brazil. In addition, they could point to a northern range extension of *B. bonaerensis* during their annual migration to the tropics, actually crossing the Equator. On the other hand, whether these specimens represent strays or an evidence of a truly migratory pathway is a matter of speculation.

Methods

The Northern Brazilian coast remained neglected by marine mammal researchers until very recently. Only in November 2005 a study group was established and dedicated themselves to aquatic mammal studies. The so called "Grupo de Estudos de Mamíferos Aquáticos da Amazônia (GEMAM)" is affiliated to Museu Paraense Emílio Goeldi (MPEG). This Museum has the third largest mammal collection in Brazil. In the past 5 years, GEMAM staff has recovered over 300 specimens of aquatic mammals, mainly from the coast of Pará state. A review of aquatic mammals 'records is available for the northern coast (Siciliano *et al.*, 2008) and many other studies are under way along the coast. Strandings along the Pará coast or inland are now reported on a regular basis, whenever the stranding site is accessible.

DNA sequencing: whale #1

Genomic DNA was extracted from one sample of a supposed Antarctic minke whale carcass (MPEG 38487) following a modified standard phenol-chloroform protocol described by Sholl *et al.* (2008).

The complete cytochrome *b* gene (1140 bp) was amplified using primers CBout1 (5'AATGAYATGAAAARYCATCGTTG-3') and CB-out2 (5'TCTTCCTTGAGTCTTAGGGAG-3'; (Cassens *et al.*, 2000). Amplifications were carried out in 50 ml reactions containing 250 ng to 1.0 mg of DNA, dNTPs (0.5 mM/ml), primers (0.3 pmol/ml), Taq DNA polymerase (Invitrogen, 0.04 U/ml) and amplification buffer under the following conditions: $94^{\circ}C$ (1 min), and 35 cycles at $94^{\circ}C$ (1 min), $55^{\circ}C$ (1 min) and $72^{\circ}C$ (90s). Four primers were used for sequencing: CB-out1 and CB-out2, which were used as external primers, and two internal primers: CB-in1 (5'-TTRTTRGATCCTGTTTCRTG-3') and CB-in2 (5'- TGAGGACAAATATCATTYTGAG-3'; (Cassens *et al.*, 2000). The PCR products were purified with 'GFXTM PCR DNA and Gel Band Purification' kit (Amersham Pharmacia), and both strands were sequenced in ABI Prism 3730 automatic sequencer. Sequences were edited with BioEdit Sequence Alignment Editor version 7.0 (Hall, 1999). A blast search using the nucleotide-nucleotide basic local alignment (blastn) search tool was done using the sequences generated.

Results

Specimens account

Details of the three stranding events of Antarctic minke whales focused in this study are given below.

Strandings events

Whale #1: On 9 January 2007 a whale stranded alive on the mouth of Rio Muriá (00°43'28.2"S, 047°54'46.8"W), Curuçá, Pará (Figure 1, Table 1). It was pushed back to sea by local fishermen and villagers. But on the 15th January it was found dead on a sand bar called Lombo da Ganância (00°33'24.87"S, 047°41'9.13"W) at Vila de Camará, Marapanim. The carcass was in advanced decomposition. It was an adult male, TL 10,37m (total length). Samples were collected for genetic and contaminant load studies. The stomach was empty. Skull and vertebrae were collected and deposited at Coleção de Mastozoologia, Museu Paraense Emílio Goeldi (MPEG 38487). DNA sequencing of muscle samples of this specimen revealed a 99% of similarity to the Antarctic minke whale (*Balaenoptera bonaerensis*) (AP006466) when compared to other baleen whales (Figures 2). This is the first report of an Antarctic minke whale for the northern Brazilian coast (Siciliano *et al.*, 2008).

Whale #2: On 14 November 2007 a live stranded whale was found at the locality of Piquiatuba, National Forest of Tapajós, Belterra, Pará (02°51'57,57"S, 055°3'55,97"W). It was a young male, TL 5,51m. After this first noticeable stranding of a whale and consequent public attraction, somehow the whale returned to the river. On the 17th the specimen was resighted at Jaguarituba, Resex Tapajós-Arapiuns, Santarém, on the right margin of Tapajós river. On the 18th November the whale got stranded again on a river bank at Comunidade São José do Arapixuna, near the mouth of Arapiuns river. On the 19th several attempts were made to push back the whale to the river, but on the 20th the carcass was found stranded on a river bank near the mouth of Arapiuns river. Necropsy was conducted by specialized veterinarians and results will be published elsewhere. The complete skeleton in mounted at "Faculdades Integradas do Tapajós (FIT)" in Santarém, Pará.

Whale #3: On April 2009 a live stranded Antarctic minke whale was found on a mudflat of the Amazon river, inside the Biological Reserve of Parazinho, Amapá (00°53'N, 049°59'W) (Lima *et al.*, 2010). It was a young female, TL 5,38m, that was pushed back to the sea during high tides and apparently was not seen again (Lima *et al.*, 2010).



Figure 1.Stranding locations of Antarctic minke whale (*Balaenoptera bonaerensis*) on the northern Brazilian coast. Note inland stranding site of whale#2, Tapajós river, Pará State.



Figure1.Neighbor-joining analysis has grouped the stranded baleen whale on the coast of Marapanim, Pará, Brazil (MPEG 38487) with *B. bonaerensis*.

Discussion

The present report is intended to first document a series of recent strandings of Antarctic minke whales near the Equator. In fact, the northern Brazilian coast has large areas still uncovered by marine mammal researchers. The large mangrove areas and mudflat beds are naturally difficult to survey by any conventional method. For this reason, there is a lack of background information on stranded cetaceans for that particular area of Brazil. Here we present two new stranding records of Antarctic minke whales in Northern Brazil, both stranded alive. A third specimen has indeed crossed the Equator and apparently has survived after beaching on a mudflat in the Amazon river mouth (Lima *et al.*, 2010). Using standard molecular sequencing we confirmed the identification of one of the stranded whale (MPEG 38487) as *B. bonaerensis*.

More recently, Glover *et al.* (2010) based upon analysis of mitochondrial and microsatellite DNA profiles, has presented the observation on a single *B. bonaerensis* in 1996, and a hybrid with maternal contribution from *B. bonaerensis* in 2007, in the Arctic Northeast Atlantic. These observations present evidence of a transequatorial migration of Antarctic minke whales. Further evidence of such destination to the Equator is given by Meirelles and Furtado-Neto (2004) and Meirelles *et al.* (2009) based on a report on a stranded Antarctic minke whale on the central coast of Ceará, Northeast region of Brazil (03°32'34"S, 038°50'55"W). In addition, Husson, (1978) mentioned the occurrence of a stranded minke whale in Suriname (04°N, 56°W). Whether these specimens represent strays or an evidence of a truly migratory pathway is a matter of speculation. It is clear the need of future surveys for evaluating cetacean occurrence and distribution patterns on the northern Brazilian coast in order to clarify these questions.

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Table 1. Antarctic minke whales on the North region of Brazil (2007-2010).

Strandings		Coordinates	Date of stranding	Field/Museum number	TL (m)	Gende r	Specimen data	Source
Balaenoptera bonaerensis								
Whale #1	Rio Muriá, Curuçá, PA	00°43'S, 047°54'W	9 Jan.2007	MPEG 38487	10.37	М	Stranded alive	Siciliano <i>et al</i> ., 2008
Whale #2	Piquiatuba, Rio Tapajós, Belterra, PA	02°51'S, 055°3'W	14 Nov. 2007	FIT Santarém	5.51	Μ	Stranded alive	This study
Whale #3	Rio Amazonas, Biological Reserve of Parazinho, AP	00°53'N, 049°59'W	15 Apr. 2009	-	5.38	F	Stranded alive	Lima <i>et al.</i> , 2010

PA = Pará State, AP = Amapá State