

## Short Note

### Antarctic Minke Whale (*Balaenoptera bonaerensis*, Burmeister, 1867) in the Tapajós River, Amazon Basin, Brazil

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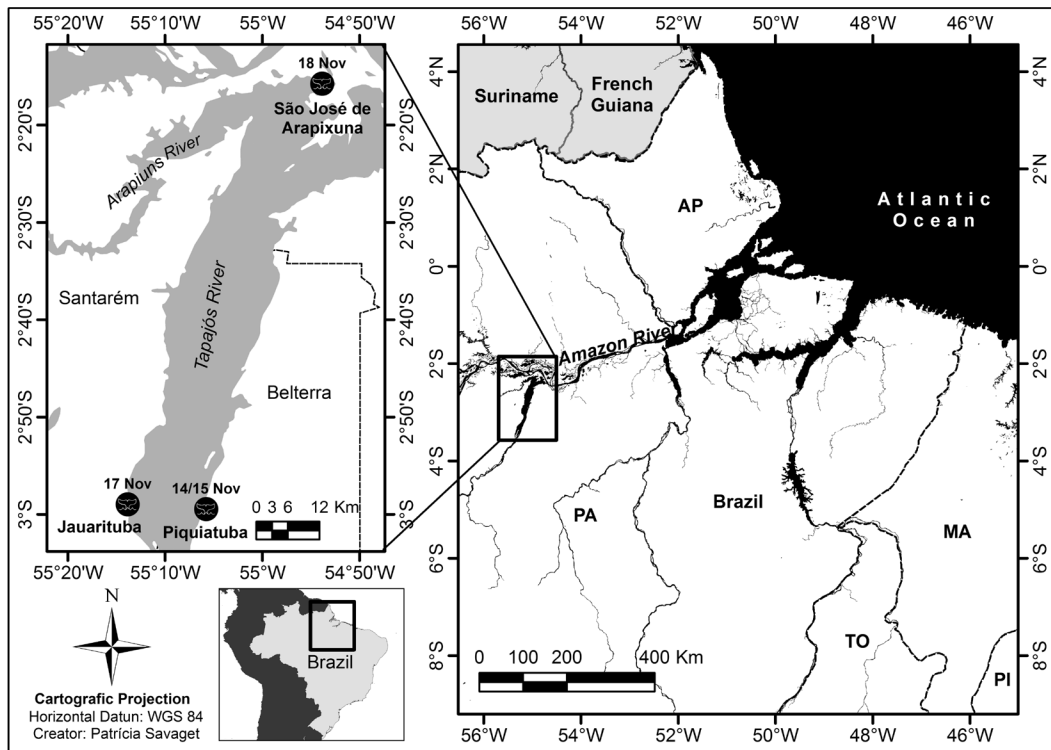
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The Antarctic minke whale (*Balaenoptera bonaerensis*) is distributed throughout the Southern Hemisphere (Rice, 1998; Mead & Brownell, 2005), although there is a single record from Suriname (Rice, 1998). In Brazil, the northeast region has been considered a breeding area for the species (Horwood, 1990; Lucena, 2006), and individuals are often sighted in oceanic waters between 200 and 1,000 m depth and in greater numbers between August and October (Andriolo et al., 2010). This species is listed as data deficient by the International Union for Conservation of Nature (IUCN) (Reilly et al., 2008), and it is currently the most abundant mysticete in the Southern Hemisphere (Beekmans et al., 2010). The first record of *B. bonaerensis* on the north coast of Brazil occurred in the Muriá River, state of Pará, in 2007 (Siciliano et al., 2008). In this report, we recorded the stranding of an individual minke whale in the Tapajós River about 1,000 km from the Atlantic coast.

The minke whale was first sighted by some local inhabitants (traditionally known as *ribeirinhos*) in the shallow waters of the eastern border of the Tapajós River in the community of Piquiatuba, a municipality of Belterra, Pará, Brazil, on 14 November 2007 (Figure 1), nearly a 1,000 km from the sea. The minke whale remained in this location until 15 November when biologists of environmental agencies examined it externally. Although the minke whale seemed to show no external indication of disease, an evaluation of its health had not been possible since no internal examination was done. Many people and boats approached the minke whale, after which the animal left the area. The next day, a rescue team, with the help of speedboats and a helicopter,

went to the sighting area, but the minke whale was not found. It was spotted again by local people on 17 November, in the community of Jauarituba on the western border of the Tapajós River. In this area, the river is nearly 17 km in width. The rescue team flew to this community, but the minke whale left the area before the team's arrival. According to villagers, the animal was hurt by a man with a stick and left the area bleeding.

On 18 November, the minke whale became stranded in a sand bank along the Arapiuns River, near the community of São José de Arapixuna, about 83 km from Jauarituba. The rescue team arrived and found the minke whale in shallow water (1 m deep) with a mud bottom. A visual examination was made of the exposed area of the body (the head and dorsal surfaces), and the underwater area was assessed by palpation since the river water was very dark. The minke whale had a small burn with some skin loss on its dorsal fin; an abrasion posterior to the blow hole; and a shallow wound about 10 cm in diameter on the right flank, close to the abdominal region, probably caused by the stick the previous day. There were no signs of weight loss. To prevent any further sunburn, the minke whale was covered with moistened white sheets. A treatment with a broad-spectrum antibiotic (20 mg/kg of Oxytetracycline) was started by intramuscular injection in the peduncle using a 9 cm needle. The respiration rate was monitored during all observations. The animal stayed apathetic, including when the drug was injected. Plans were made to move the animal to a small river and use nets to establish an enclosure of the area. This would allow collection of blood and other samples to assess the minke



**Figure 1.** Sites where the minke whale (*Balaenoptera bonaerensis*) was sighted in the Tapajós and Arapiuns Rivers, Pará, Brazil, nearly a 1,000 km from the Atlantic Ocean

whale's health status and to care for it until a ship could be prepared to transport it to the sea (a 3-d trip). However, the minke whale suddenly began to make vigorous movements with its tail and body until its head faced the river and then it started swimming. After the minke whale had moved a few hundred meters, it started swimming in circles. This behavior occurred just when the helicopter flew over the area, so it could have been a reaction to the aircraft. The minke whale continued to be monitored, and at least two Amazonian dolphins (*Inia geoffrensis*) were observed swimming close to the animal for a few minutes. The plan to move the minke whale to a small river was aborted at sunset because of bad weather conditions. The minke whale remained under surveillance and was sighted for the last time around 2200 h with the aid of flashlights. On the next day (19 November), the minke whale was not seen, but on 20 November, it was found dead, about 200 m from where it had been sighted for the last time. The animal probably died during the night of 19 November. The necropsy was performed on 20 November, approximately 28 to 36 h after its death.

The minke whale was in moderate decomposition (code three; Geraci & Lounsbury, 2005) with protrusion of the penis and tongue swelling. Loss of skin and the formation of blisters with an accumulation of gas beneath the epidermis were found in localized spots, representing less than 5% of the animal's body. The eyes were absent. The blubber layer was measured below the dorsal fin on the mid-lateral line and had 3 cm thickness. The main areas of injury were the following: hemorrhage on the rectus abdominis muscle and a hematoma forming a cavity of approximately 50 × 30 cm. The stomach was empty, and there were hemorrhagic areas in the stomach walls with loose mucosa. The lungs were congested with blisters on the external surface. Given the decomposition of the carcass, it was not possible to establish the cause of death. Morphometric measurements were collected with the animal alive (except the blubber thickness), following Geraci & Lounsbury (2005). This Antarctic minke whale measured 5.5 m in length (Table 1). Craniometric measurements were collected after recovery and cleaning of the skeleton, along with measures following Zerbini & Simões-Lopes (2000) (Table 2).

**Table 1.** Morphometric measurements of the Antarctic minke whale (*Balaenoptera bonaerensis*) stranded in the Tapajós River

Morphometric parameter	Measurement (m)
Total length	5.5
Tip of maxillary to dorsal fin base	3.7
Tip of maxillary to pectoral fin base	1.74
Mouth length	1.42
Tip of maxillary to blowhole	0.96
Caudal fin maximum width	1.54
Left pectoral fin length of anterior insertion to end	0.9
Left pectoral fin length of axilla to end	0.72
Left pectoral fin maximum width	0.15
Dorsal fin base	0.33
Dorsal fin height	0.22
Blubber layer*	0.03

\*From dead animal on 20 November 2007

One skin sample was collected and preserved in ethanol 70% to confirm genetic identification of the species. Genomic DNA was extracted using a DNeasy tissue kit (Qiagen), and approximately 660 bp of the most variable portion of the mtDNA control region (D-loop) was amplified (primers Dlp-1.5 and Dlp-5; Baker et al., 1993). PCR conditions and sequencing procedures followed the same guidelines described in Engel et al. (2008). Since a large portion of the initial sequence (~360 bp) was not clean due to DNA degradation, the sequence was trimmed to a length of 295 bp and after to 175 bp. A comparison to the GenBank sequences (Benson et al., 2010) from eight minke whale exemplars (four *B. bonaerensis* [EF113728, EF113822, and EF113823: Pastene et al., 2007; and DQ145041: C. S. Baker, direct submission to GenBank in 2005]; two *B. acutorostrata acutorostrata* [EF113835 and EF113895: Pastene et al., 2007]; one *B. a. scammoni* [AY878077: Baker et al., 2000], and one dwarf minke whale [*B. a. subsp.* (EU285375: C. S. Baker & C. Olavarria, direct submission to GenBank in 2007)]) was conducted. The sequences were aligned using Muscle algorithm implemented in *MEGA 6* (Tamura et al., 2013). Phylogenetic analyses using maximum likelihood (ML) method implemented in the program *MEGA 6* were performed to determine the relationship among the query and reference sequences (varying from 333 to 463 bp of length). Furthermore, the minke whale species was identified by submitting the control region sequence to the Web-based program *DNA Surveillance* (Ross et al., 2003), using Version 4.3 of the reference database, and by *Basic Local Alignment Search Tool* (BLAST) searches of GenBank.

The shorter DNA fragment (175 bp) aligned between the positions 162 and 336 of the sequences previously published (336 bp; Pastene et al., 2007). Phylogenetic analyses among the

reference sequences with the sequence of this minke whale from the Tapajós River showed high bootstrap support (> 99%) for the relationship of this minke whale with the four Antarctic minke whale exemplars, which were grouped together. In addition, the analyses based on DNA surveillance showed high bootstrap support (99%), grouping the test sequence with the reference sequences of Antarctic minke whales. *BLAST* also identified Antarctic minke whale for the sequence trimmed in 175 bp. This sequence collapsed in two *B. bonaerensis* haplotypes—Bb083 found in one sample collected in the Antarctic and Bb034 found in one sample collected in Brazil—and two samples collected in the Antarctic. Importantly, this short sequence has six (positions 129, 289, 300, 302, 303, and 304; Pastene et al., 2007) of the eight nucleotide positions that differentiate the two species of minke whales (*B. bonaerensis* and *B. acutorostrata*). These results confirm the utility of molecular genetic methods for the identification of cetacean species.

Since two species of minke whale occur in Brazilian waters—the Antarctic minke whale and the dwarf minke whale (*B. acutorostrata*) (Lodi & Borobia, 2013)—morphometric characters of the skull were also used to identify the minke whale in this stranding. Zerbini & Simões-Lopes (2000) relied on morphological features and morphometric characters of the skull to distinguish these species. Cranial maturity of individuals according to reproductive maturity was tested, and three classes were suggested. The skull measurements of the minke whale in this study corresponded to Class II of *B. bonaerensis* according to Zerbini & Simões-Lopes (Table 2), which correspond to mid-sized animals (possibly older calves or subadults) but are still below the length at sexual maturity. Meirelles & Furtado-Neto (2004) suggested that the distribution of *B. bonaerensis*

**Table 2.** Skull measurements of the Antarctic minke whale stranded in the Tapajós River and range of skull measurements of cranial maturity Class II individuals according to Zerbin & Simões-Lopes (2000)

Morphometric parameter	<i>B. bonaerensis</i> (Tapajós River) (mm)	<i>B. bonaerensis</i> (Zerbin & Simões-Lopes, 2000) (mm)
Condyle premaxillary length	1,280	1,033-1,595
Length of premaxillary	830	655-1,024
Length of maxillary	770	665-1,087
Tip of premaxillary to vertex	850	671-1,042
Tip of premaxillary to nasals	830	566-970
Length of nasals (median)	100	80-156
Breadth of nasals (anterior)	80	58-96
Length of rostrum (base/pterygoid ventral)	780	566-1,014
Breadth of rostrum at middle*	330	187-316
Breadth of rostrum at base	440	313-556
Breadth across maxillaries at vertex	130	88-166
Breadth of frontals across nasals	150	105-198
Breadth between maxillaries at nares	160	112-194
Breadth of skull (squamosal)	690	507-886
Breadth of skull (frontal)	580	446-779
Breadth of skull (maxillaries)	610	442-781
Length of orbit – R	140	102-207
Length of orbit – L	140	104-204
Breadth of exoccipital bone	460	375-659
Breadth across occipital condyles	150	109-238
Height of occipital condyle – R	105	66-152
Height of occipital condyle – L	105	71-147
Breadth of foramen magnum	70	48-110
Height of foramen magnum	60	42-101
Length from foramen magnum to vertex	410	282-486
Tip of premaxillary to anterior end of palatine (median)	820	632-1,005
Tip of premaxillary to posterior end of palatine (median)	1,050	819-1,333
Length of hemimandible (straight) – R	1,210	980-1,595
Length of hemimandible (straight) – L	1,205	981-1,588
Height of hemimandible at coronoid – R	180	137-234
Height of hemimandible at coronoid – L	180	134-233
Height of hemimandible at condyle – R	120	94-169
Height of hemimandible at condyle – L	120	96-166

\*Only measure that does not match with previous study (Zerbin & Simões-Lopes, 2000)

in the South Atlantic Ocean could be extended. Recent records of this species along the northern coast of South America presented herein and in previous studies (Meirelles & Furtado-Neto, 2004; Siciliano et al., 2008; Lodi & Borobia, 2013) could support this assumption and should encourage new studies to investigate the occurrence and distribution of cetaceans in this region.

The occurrence of baleen whales in freshwater systems is not common, although there are occasional records. In Brazil, a pregnant female *B. edeni* was stranded in the Paraguassu River, state of Bahia, in 1981 (Lima et al., 2006). An immature male of the same species remained 100 d in Manning River,

New South Wales, Australia, in 1994 (Priddel & Wheeler, 1997); and despite the time it remained in fresh water, it showed no clinical evidence of physiological damage (Priddel & Wheeler, 1998). In 2007, a mother and calf *Megaptera novaeangliae* appeared in the Sacramento River in California, reaching the ocean 20 d after their first sighting in the river (Gulland et al., 2008). As highlighted by Gulland et al. (2008), the underlying reasons for why whales enter in these atypical locations are unknown, and the health status of the individuals involved has been poorly documented. The minke whale in the Tapajós River was observed for 5 d in fresh water; however, this minke whale must have

taken at least 12 d to move from the Atlantic Ocean to Piquiatuba (almost 1,000 km from the sea), where it was sighted for the first time. A daily average of 78 and 79 km was recorded for two *B. acutorostrata* tagged along the northern coast of Norway (Heide-Jørgensen et al., 2001), and the minke whale of the Tapajós River moved from Jauarituba to São José de Arapixuna (ca. 83 km) in 1 d. Decomposition of the carcass occurred swiftly. Some possible explanations for this fast decomposition include the high temperature of the waters in Tapajós River, around 30° C (Miranda et al., 2009), or a possible septicemia. The cause of death is unclear as is the cause for why this animal entered so far into the Amazonian basin. As far as we are aware, this is the farthest record of a baleen whale out of the sea.

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