

Electronic Supplementary Materials

SUPPORTING INFORMATION

Ganges River dolphin: an overview of biology, ecology and conservation status in India

Additional Introductory Material:

Researchers at Patna University (Patna) conducted several surveys in discrete segments in an ~600-km stretch of the Ganges River from Chausa near Buxar (Bihar) to Farakka Barrage (low, gated diversion dam) (West Bengal) near the India-Bangladesh border, using oar-driven country boats during the period of 1991 to 1994; a continuous survey in the Ganges from Patna to Farakka Barrage in November 1994; and surveys from Allahabad to Patna (600 km) in October 1995 and downstream Farakka Barrage in the Bhagirathi-Hooghly Rivers in April 1995, under the Dolphin Conservation Project sponsored by the Ganga Project Directorate, Ministry of Environment and Forests, Government of India (Sinha 1996, 1997, Sinha et al. 2000). During the same period, a study was conducted in discrete segments of the upper reaches of the Ganges River from Rishikesh to Kanpur (approx. 600 km) by Jiwaji University, Gwalior (Behera 1995). Continuous surveys were conducted in the Karnali-Girwa Rivers from Kachali (15 km upstream Chisapani Bridge) on the Karnali gorge in Nepal to Girijapuri Barrage 20 km inside India-Nepal border in 1993 (Smith et al. 1994) and in the Sarda River from Lower Sarda Barrage at Sardanagar to Palya, up to which the river was navigable during the low-water season in March

1994 (Sinha and Sharma 2003a). Continuous surveys were conducted in the Ganges River in a stretch of over 1800 km from Haridwar, at the foothill of the Himalayas, to Farakka near the India-Bangladesh border in 1996-98 (Sinha 1999, Sinha et al. 2000). Further, a dolphin survey was conducted in the Chambal River in the early 1980s (Singh and Sharma 1985).

Thus, a majority of the information on river dolphins in India arises from several surveys and ecological studies that were conducted by Patna University in the mid- and lower reaches of the Ganges and its tributaries and by the World Wildlife Fund (WWF)-India in the upper reaches of the Ganges and some small tributaries (Sinha et al. 2000, Sinha and Sharma 2003a, 2003b, Behera and Rao 1999). With support from the National River Conservation Directorate (earlier Ganga Project Directorate), Ministry of Environment and Forests, Government of India, Patna University undertook intensive studies from 2001 to 2007 in a 500-km stretch of the Ganges in Bihar, where over 50% of the total population of the Ganges River dolphin (currently over 3,000) survive (Sinha et al. 2010). During the same period, local researchers conducted continuous surveys in the Brahmaputra River, a large river of the Ganges river system in India, in the state of Assam (Biswas and Boruah 2000, Wakid, 2005, 2009). Recently, continuous surveys were conducted in a 525-km stretch of the Ganges in Bihar from Sahibganj to Chausa in October-November 2012 and in Gandak River in December 2012. A map of the Ganges basin and major locations mentioned in this article is presented in **Fig. 1**.

We also documented various threats to which the Ganges dolphins were exposed, including directed and incidental killings by the fishermen to extract oil from their blubber for use as bait in the oil fishery in the dolphins' distribution range. In addition, we collected dolphin carcasses from the Ganges and its tributaries in the 1980s and 1990s, tissues of which were analyzed for toxic pollutants, including heavy metals; organochlorines, including polychlorinated

biphenyls (PCBs) and pesticides (e.g., DDT and HCH); organotin compounds; and perfluorinated chemicals (PFCs) (Kannan et al. 1993, 1994, 1997, Senthilkumar et al. 1999, Yeung et al. 2009). One of the projects (Project No. 23), recommended by the IUCN/SSC Cetacean Specialist Group Action Plan, was to develop an alternative to dolphin oil for use as a fish lure (Perrin 1988). As a part of this investigation, we discovered an effective oil from fish scraps as an alternative to dolphin oil (Sinha 2002). The effects of dams and barrages on Ganges dolphins and their prey in the entire distribution range in India were studied by Sinha (2000) and Smith et al. (2000).

Patna University collaborated with the Centre for Cellular and Molecular Biology, Hyderabad, India, to determine the evolutionary relationship of the Ganges dolphin with extinct and extant cetaceans, based on comprehensive analyses of the mitochondrial cytochrome b and nuclear interphotoreceptor retinoid-binding protein gene sequences (Verma et al. 2004). In this regard, several researchers conducted phylogenetic analyses of nucleotide sequences from mitochondrial and nuclear genes of the Ganges dolphins (Cassens et al. 2000, Nikaido et al. 2001, Yan et al. 2005).

Six types of surfacing patterns of the susu, which were dependent on the age and offshore distance of the susu, were reported at the confluence of the Ganges and Gandak near Patna (Sinha et al. 2010a). Recent studies by WWF-India and Japanese researchers on the acoustics of the Ganges dolphin in the upper reaches of the Ganges at Narora are expected to yield information on underwater activities of the susu (Sugimatsu et al. 2011, 2012). Studies on the status, habitat use, and threats that the Ganges dolphins face were conducted in Nepal and Bangladesh (Kasuya 1972, Kasuya and Haque 1972, Smith 1993, Smith et al. 1998, 2001, 2006, 2010).

Additional Text from Taxonomic status and genetics

Gray (1863) first challenged the monophyly of the classical river dolphins. Muizon (1988, 1991) and Heyning (1989) did not consider the platanistoids (i.e., classical river dolphins) to be monophyletic and separated *Platanista* from the remaining river dolphins, based on the study of fossils and facial anatomy; this categorization has been widely accepted. Many morphological analyses emphasized the substantial divergence among the four river dolphin species (Zhou et al. 1979), and this eventually led to the classification of the four genera in four monotypic families (Pilleri and Gahr 1980, Zhou 1982). Rice (1998) considered each of the four families to contain only a single living species, each endemic to a well-defined and restricted geographic setting.

TAXONOMIC STATUS AND GENETICS

There are 88 recognized extant species of cetaceans, of which 41 species that belong to 21 genera are dolphins (iucn-csg.org 2013 accessed on May 7, 2013). Most of the extant dolphins are marine, but several species inhabit, sporadically or exclusively, freshwater. The three obligate riverine dolphins are (i) the blind river dolphin (*Platanista gangetica*), which inhabits the Ganges, Brahmaputra, and Meghna river systems in India, Nepal, and Bangladesh and the Indus river system in Pakistan and India; (ii) the Yangtze river dolphin, or “baiji” (*Lipotes vexillifer*), which inhabited the lower and middle reaches of the Yangtze River in China, but is now extinct, and (iii) the Amazon river dolphin, or “boto” (*Inia geoffrensis*), which is largely distributed in northern South America in the Orinoco and Amazon river systems and the upper Rio Madeira drainage. The fourth species classified as a river dolphin is the La Plata dolphin, or

“franciscana” (*Pontoporia blainvillei*), which is found in estuaries and coastal waters of eastern South America from 19°S (Brazil) to 42°S (Argentina); however, this species is not an obligatory freshwater species.

All river dolphins have a peculiar morphology, with a characteristic long and narrow rostrum (beak) with numerous prehensile teeth, a low triangular dorsal fin supported by connective tissue, broad and visibly fingered flippers, and un-fused cervical vertebrae (which allow considerable neck movement), making the neck flexible (Cassens et al. 2000). The eyes of river dolphins also have been reduced to various degrees (the *Platanista gangetica* even lacks eye lenses and is virtually blind), and their echolocation abilities are more refined than in other cetaceans (Cassens et al. 2000). Considering the early divergence of the “true” river dolphins within the order Cetacea, these shared features offer an excellent example of convergent evolution, despite little genetic connection. In addition, shared skull characteristics led most authors to classify them into a monophyletic group, either in the family Platanistidae or in the superfamily Platanistoidea (Cassens et al. 2000).

For Platanistidae and Iniidae, some subspecies have been designated. The Platanistidae consists of the subspecies of *Platanista gangetica gangetica* (Ganges River dolphin or susu) and *Platanista g. minor* (Indus River dolphin or bhulan). Yang and Kaiya (1999) reported that the difference in the cytochrome b sequence of Ganges and Indus River dolphins was small and supported the belief that the Ganges and Indus River dolphins were probably two subspecies of a single species. They also suggested that, among the four river dolphin families, Platanistidae was the earliest divergent clade. Thus, Platanistidae consists of the “blind” dolphins of the South Asian subcontinent: species *Platanista gangetica*, subspecies *P. g. gangetica*, and *P. g. minor*; and Iniidae consists of the species *Inia geoffrensis* of South America, with the subspecies *I. g.*

geoffrensis (Amazon dolphin or boto), *I. g. humboldtiana* (Orinoco dolphin), and *I. g. boliviensis* (Bolivian dolphin of the Madeira River drainage). There is ongoing debate, in regard to both families, on whether multiple species should be recognized (Cassens et al. 2000; Smith and Reeves 2012; Best and Da Silva 1989; Da Silva 1994). The taxonomic relationships of the river dolphins have been subject to debate for more than a century (Cassens et al. 2000).

Verma et al. (2004) established the evolutionary relationship of the Ganges dolphin with extinct and extant cetaceans based on comprehensive analyses of the mitochondrial cytochrome b and nuclear interphotoreceptor retinoid-binding protein gene sequences. The study suggested that *P. g. gangetica*, a toothed cetacean, is more significantly related to Mysticeti (toothless/baleen whales) than to any other toothed whales. Yan et al. (2005) reported that the *Platanista* lineage is always within the odontocete clade instead of having a closer affinity to Mysticeti, with moderate to high bootstrap values. Nevertheless, they believe that the position of the *Platanista* is more basal, which suggests separate divergence of this lineage well before the other river dolphins and that the exact phylogenetic position of *Platanista* could not be resolved. Nikaido et al. (2001) corroborated the *Platanista* lineage as ancient and stated that this genus was the only living member of a once-diverse clade. The prevention of the looming extinction of this unique clade should be a major conservation priority (Nikaido et al. 2001).

EVOLUTIONARY ADAPTATIONS AND ANATOMICAL PECULIARITIES

Anatomically, *Platanista* is a peculiar mammal (Reeves and Brownell 1989). The skull is highly disparate from that of other members of Cetacea, especially in its having highly pneumatized maxillary crests that arch over the face, which probably act as acoustic reflectors for echolocation generated in the underlying soft nasofacial tissues (Nikaido et al.

2001) (Fig. 2). Herald et al. (1969) assumed that the maxillary flanges act as acoustic baffles to direct sonic pulses into a narrow beam. Scientists who have studied the clicking and echolocation in Ganges dolphins in Sundarbans mangrove forests observed that the centroid frequency of the clicks produced by the dolphins is almost an octave lower than that expected from their size (Jensen et al. 2013). They concluded that the unusual, air-filled, bony maxillary crests found in this species may compensate, in part, for this lower frequency by providing a larger effective baffle and, hence, a more directional sound beam than the biosonar frequency and head size would predict. **The beam width of Ganges dolphins is still wider than that of most other toothed whales, and it is possible that this may facilitate capture of highly maneuverable prey items in shallow and often turbid waters (Jensen et al. 2013).**

The pneumatic sinuses within the maxillary crests arise ventrally in the pterygoid sinus complex of the skull base (Fig. 2). Nikaido et al. (2001) concluded that the complex multi-lobed sinuses have evolved significantly in *Platanista*. Fossils show that complex multi-lobed sinuses evolved in the Squalodelphinidae and that sinus-bearing maxillary crests appeared in the Platanistidae (Nikaido et al. 2001). The degenerate nature of the eye, constant pulse emission, and obstacle-avoidance behavior, both in the natural murky habitat and in captivity, suggest that *Platanista* is an effective echolocator (Herald et al. 1969).

PHYSICAL DESCRIPTION

The Ganges dolphin is gray or brown in color, although sometimes a lighter coloration is exhibited on the ventral side. Their beaks are distinctively swollen at the tip and are highly elongated, attaining 15-27 percent of the length of their bodies depending on age and sex, with large, visible teeth (Fig. 2). On the basis of observations of 36 dolphin carcasses of different age groups at Patna University, we found that beaks' attainment of 15 to 27 percent of the body

length depends on age and sex. We also counted 140 prehensile teeth in both jaws of adult female dolphins, with the upper jaw's tending to have more teeth than did the lower jaw. Although narrow and sharp in youth, the teeth become worn and flattened with age (Anderson 1879). All the rear teeth were found to be worn and flattened in a 2.5 long pregnant dolphin. A newly borne baby was observed to be without teeth, but on the margins of both jaws, there were teeth-like rows of elevations. We observed longer and closely placed teeth at the tip of jaws of a dolphin calf about one month old. In the same calf teeth in the rear part of the jaws were very small with a wide gap between two teeth. This feature is probably an adaptation to hold the nipple between two teeth to suckle milk.

PRIMITIVE CHARACTERISTICS

Platanista g. gangetica bears some of the very primitive characteristics not seen in other cetaceans, viz. presence of caecum at the junction of the small and large intestine (Anderson 1879), and the position of the testes is much more dorsal than in other marine cetaceans (testes are extra-peritoneal in terrestrial mammals). Many of the male oceanic dolphins have their testes situated more dorsally and are much less descended than is seen in *Platanista*. Subcutaneous muscle between two layers of blubber was seen in *Platanista* (pers. comm. Tadasu Yamada). Further, the simple air sacs and accessory air sacs around the nasal passage might indicate that the *Platanista* are more primitive than are other oceanic dolphins (pers. comm. Tadasu Yamada).

BIOLOGY

Ganges dolphins have highly developed sonar systems. They use pulse sounds, but not whistles, to navigate. This allows them to perceive objects, specifically prey, in murky waters. Over a 24-hr period, there is almost always a constant emission of sound; 87% of these sounds are clicks

for echolocation, and the remaining vocalization is used in communication. There have not been enough studies, however, to determine the significance of these communicative sounds (Moreno 2003; Nowak 2003; Reeves and Brownell 1989). Recent studies by WWF-India and Japanese researchers on the acoustics of the Ganges dolphin in the upper reaches of the Ganges River at Narora are expected to yield information on underwater activities of the susu (Sugimatsu et al. 2011; 2012)

BEHAVIOUR

The Ganges dolphin is a solitary animal but occasionally congregates in groups of three to ten individuals. Mothers and calves stay together until the infants are weaned (Sinha 1999). They are found in loose congregations, especially at the confluences where prey congregate. The dolphins swim almost constantly, often on their sides, a method of progression previously unknown among cetaceans (Herald et al. 1969). Shortly after initiating a dive, the dolphin spins 90° on its lateral axis and 180° on its longitudinal axis to swim on its side in the direction opposite from the surfacing direction (Smith 1993). While the dolphins swim sideward, the pectoral flipper either touches the bottom or trails about 2 to 3 cm above it. The tail is higher in the water than is the head, so that the body is at an oblique angle with respect to the bottom of approximately 10°. If the susu were vertical in the water, the head would be bobbing up and down; but because the dolphin is on its side, the head motion becomes a lateral sweep over the bottom (Herald et al. 1969). The flippers are thought to have an important tactile function (Pilleri 1970; Pilleri et al. 1976). This probably explains why the flippers almost feel the bottom as a means to identify the habitat by the nature of its bottom.

It is not possible to identify individuals using photographs because they do not appear to have any unique features, and obtaining pictures of them is prohibitively difficult. It is only when dolphins are kept in clear water in captivity that anything of their underwater behavior can be discerned. Three female Indus dolphins captured by Herald et al. (1969) were kept in holding pens in Karachi en route to the United States, and this was the first time that their unique side-swimming behavior was observed. One pectoral flipper either touched the bottom or trailed just above it. The lower flipper repeatedly touched the bottom during sideward swimming (Pilleri and Pilleri 1987). Pilleri (1970) suggested that side swimming may occur only in certain situations and is an adaptation that allows dolphins to move through shallow water.

SURFACING BEHAVIOR AND DIVE TIMES

Indus and Ganges dolphins surface alone; only mothers and young calves surface in near synchrony. Where the species showed great diversity in surfacing patterns, depending on several environmental factors and age-class, our team studied the surfacing and diving behavior vis-à-vis several covariates in the free-ranging Ganges dolphin. On the Gandak-Ganges confluence at Patna, Ganges dolphins displayed different surfacing patterns depending upon their position from offshore distance as well as on day-hours. Between 25 and 50 m from shore, exposure of head and dorsal fin dominates in adult and sub-adult dolphins (Sinha et al. 2010b). Within the same distance, juveniles expose rostrum, head, and dorsal fin. Feeding activities and adaptive strategies to minimize foraging costs resulted in aggregation and vigorous surfacing of dolphins in the impact zone of confluence.

The adults and sub-adults were found to have different types of surfacing in different day-hours. Exposure of the head and dorsal fin was dominant among adults throughout the day,

while exposure of the rostrum, head, and dorsal fin among the sub-adults was common in the morning and afternoon. Surfacing of adult dolphins was usually vigorous (Leaping / rostrum, head, dorsal fin and fluke rise). The juveniles were observed only during the morning and afternoon and dive-time was highest during morning hours (range 23 – 267 second, mean 142, SD \pm 70). Head and dorsal fin in the morning, and rostrum, head, dorsal fin, and fluke exposures in the afternoon were dominant. *Ad libitum* observation of the dolphin groups showed that they were active, as demonstrated by vigorous and frequent surfacing during morning and afternoon hours. The morning and afternoon were observed to be feeding hours of the Ganges dolphin (Sinha et al. 2010b). Dolphins show mainly the top of the head and back, or the rostrum, head, and back, when surfacing. Breaches are very rare, and the tail flukes are almost never visible. The dolphins were seen following or aggregating in and around the drift fishing nets in shallow zones, most probably to take or catch entangled/escaped fish from the fishing nets (Sinha et al. 2010b).

Dive-time of the Ganges dolphins ranged from 10 to 465 seconds. Dive-times of adults (mean: 129s; range: 10-465s; SD: 87) and sub-adults (mean: 120s; range: 10-385s; SD: 66) were almost similar, whereas juveniles (mean: 92s; range: 20-267s; SD: 58) remained underwater more than did neonates (mean: 59s; range: 25-115s; SD: 26) (Sinha et al. 2010b). In many cetaceans, to cope with the stress of diving, myoglobin concentrations are generally high, but due to their relatively shallow river habitats and short dive periods, myoglobin concentrations in heart and muscle tissues are considerably lower in the Ganges dolphin (Encyclopedia of Life 2011).

MATING AND BIRTH

There is limited information available on the mating systems of Ganges dolphins. Kasuya (1972) estimated the age of sexual maturity to be 10 years or less for both sexes. Kasuya (1972) suggested that males may become sexually mature at about 170 cm in length or less, when they are 10 years old or slightly younger. Our team has often observed mating-like activities in low-water season between March and May, in which four to five males were found chasing one adult female dolphin; this is characterized by vigorous movement with a lot of water splashing, and ultimately one male succeeded. The female and the successful male were found surfacing together, vertically rubbing their ventral portions a couple of times. This appeared to be an indication of successful mating, which, in discussion with local fishermen, was confirmed. Further studies are needed to provide more information on the mating behaviour of dolphins.

Anderson (1879) reported the gestational period to be eight to nine months. Ganges dolphins bear a single offspring of 70-90 cm long. Weaning can begin as early as two months to as late as 12 months, with a typical duration of nine months (Kasuya 1972, Reeves and Brownell 1989). Once offspring have been weaned, they disperse and become independent (Moreno 2003; Nowak 2003; Jefferson et al. 2008).

In *Platanista* there might be two peaks of parturition, one in early summer and the other in early winter. Our team sighted two neonates of the Ganges dolphins in the River Ganges in the last week of October 2012, and 4-5 neonates in the second week of November 2012. We also observed newly born calves in the third week of February 2010 and 2012. Neonates have been sighted in April and October as well. It appears that the parturition occurs during October and May. Kasuya (1972) reported that the juveniles were born from October to March, with a peak in December and January in rivers in Bangladesh. He concluded that, although the parturition of this species can occur during any season of the year, it usually takes place in the low-water

season and not during the flood season, most likely to avoid drowning of neonates in the fast-flowing flooded rivers. Consequently, as the Ganges River is flooded from July through September, parturition does not occur during these months.

The smallest free-swimming dolphin calf was a female, 67.4 cm in length, and weighed 7.3 kg (Kasuya 1972). Four fetuses were between 42.5 and 54.7 cm in length (Harrison 1972). Our team found a 70-cm long dead male fetus, with the placenta attached, floating in the Ganges River in April 1987; it weighed 4 kg. Apparently, it died just after birth or was stillborn. In February 1993, while performing a necropsy of an adult pregnant dead dolphin (2.5 m long and weighing 114 kg), the lead author found a 57-cm fetus in her womb that, apparently, was not mature enough for delivery. During the first week of September 2005, we found a dead pregnant dolphin that had collided with a mechanized vessel; the viscera of the dolphin and a 24-cm long, 116-g fetus, attached to mother's womb with the placenta, were floating in the Ganges River. We also collected the carcass of a 95-cm long male dolphin whose stomach contained milk only; another carcass, of a 105-cm long male, had both milk and remains of immature insects and shrimp in its stomach. It is likely that the latter one was being prepared for weaning.

LIFE SPAN/AGE

Not much is known about the longevity of the Ganges dolphin. Sexual and physical maturity may be attained at the ages of approximately 10 years or less, and more than 20 years, respectively (Kasuya 1972). Physical maturity is attained when all the inter-vertebral discs become completely fused with their respective vertebrae. Females attain sexual maturity when they are between 170 and 200 cm in body length (Kasuya 1972). A 28 year old male Ganges dolphin of 199 cm body length was found to be physically immature (Kasuya 1972). Anderson

(1879) reported a physically mature male dolphin of 211 cm. Similarly, physical maturity was not complete in a 233cm female, but a 252 cm long female was found to be physically matured. Almost all the teeth of the 252-cm female were worn down, and her posterior teeth were flat. The Encyclopedia of Life (2011) reported that the “maximum longevity of a Ganges dolphin may be close to 30 years.” This may be an underestimate, as Kasuya (1972) found a physically immature male who was 28 years old. By counting the number of the opaque layers in dentine, we estimated the age of a 233-cm long female to be 30 years; the female was not completely physically mature. Based on these observations, we expect the life span of the Ganges dolphin to be between 40 and 50 years.

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Fig. 1. Skull of an immature Ganges river dolphin showing (A) maxillary crest and (B) pneumatic sinuses (photos by Samir Kumar Sinha).