Photo-Identification: A Reliable and Noninvasive Tool for Studying Pink River Dolphins (*Inia geoffrensis*)

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Abstract

Photo-identification is an important tool for studying cetacean residence patterns, population size, movements, and social structure. This knowledge directs conservation and management. We examined the reliability of photo-identification studies of pink river dolphins (Inia geoffrensis) with the hope of encouraging long-term population monitoring programs. From February 2007 to August 2009, 12 surveys were conducted in two locations of the Colombian Amazon and Orinoco river basins. We obtained 795 suitable digital photographs of Inia dolphins. We evaluated the reliability and duration of photo-identification by describing and evaluating the permanence and consistency of eight mark-types. Marks were categorized as reliable (pigmentation patterns on the dorsal ridge, nicks, bends, and wounds) or supplementary based on their prevalence in the population, and gain and loss rates. We created a catalog of well-marked animals, defined as individuals with at least two reliable marks (55% of the images analyzed for this purpose). It contained photographs of the right side of 57 individuals and the left side of 40 individuals. There were 16 individuals with resightings over a 23-mo period. Future field surveys should use digital cameras with long lenses and fast shutter speeds in areas where dolphins are conspicuous when surfacing.

Key Words: pink river dolphins, *Inia geoffrensis*, Amazon, Orinoco, marks, photo-identification

Introduction

The process of recognizing individual cetaceans and tracking them through time allows researchers to answer a wide range of questions related to population size (Wilson et al., 1999), migration (Rock et al., 2006), distribution (Williams et al., 1993), critical habitat (Ingram & Rogan, 2002), and social structure (Whitehead, 2008). Photo-identification is a noninvasive and relatively inexpensive method used to identify individuals (e.g., Hammond et al., 1990). Individuals are recognized from photographs of natural marks such as stripes to identify zebras (Equus burchelli) (Petersen, 1972); nose scars to identify sea otters (Enhydra lutris) (Gilkinson et al., 2007); pigmented spot patterns in leatherback sea turtles (Dermochelys coriacea) (McDonald & Dutton, 1996); and scars, skin patches, color patterns, callosities, and nicks and notches along fin edges to identify whales and dolphins (Hammond et al., 1990). The efficiency of the photo-identification of natural marks has significantly improved during the last decade due to the availability of high-resolution digital cameras, rigorous and standardized protocols for storing and analyzing images, and the use of computer-aided software to assist in the matching of individuals (Hillman et al., 2003; Markowitz et al., 2003). Photo-identification of natural marks became a reliable and widely used tool to help understand the ecology of wildlife populations, and thus, to help in the process of recommending conservation and management actions (e.g., Rock et al., 2006).

Photo-identification of natural marks in freshwater cetacean populations is a challenge. It is not easy to photograph species that spend most of their lives in dark and turbid waters, and, when at the surface, tend to be inconspicuous, shy, and unpredictable. Moreover, given that all cetacean populations that inhabit freshwater ecosystems live in the watersheds of developing countries, the funding and technology available to conduct research are limited. Consequently, photo-identification efforts are conducted mostly by using analog cameras and short-distance lenses, restricting the quantity and quality of information obtained and stored. For all these reasons, studying and identifying freshwater cetaceans using photography has proved challenging.

Some studies, however, have successfully photo-identified cetaceans in freshwater ecosystems using marks (Table 1). For instance, photo-identification studies of cetaceans in Asia use natural marks such as notches, distinctive shapes in the dorsal fin, and white marks to identify Irrawaddy dolphins (Orcaella brevirostris) in riverine and coastal areas (Smith et al., 1997; Parra & Corkeron, 2001; Kreb & Rahadi, 2004). Similarly, studies of river dolphins in Asia use nicks, scars, and white spots on the dorsal ridge and behind the blowhole to identify Baiji (Lipotes vexillifer) of the Yangtze River in China and Ganges susus (Platanista gangetica) in the Karnali River in Nepal (Hua et al., 1990; Zhou et al., 1998; Smith & Reeves, 2000). Photo-identification studies of river dolphins in South America use pigmentation patterns, nicks, wounds, and scratches, to identify pink river dolphins (Inia geoffrensis) in some areas of the Amazon and Orinoco river basins of Colombia, Venezuela, Bolivia, and Peru (Trujillo, 1994; McGuire & Winemiller, 1998; Aliaga-Rossel, 2002; McGuire & Henningsen, 2007). As a result, numerous studies have demonstrated that, in spite of the challenges, cetacean species in freshwater ecosystems have individualspecific natural marks that can be photographed and used for identification.

Little is known about the conservation status of pink river dolphins, *Inia* spp., which are distributed in many rivers and tributaries of the Amazon and Orinoco basins. As a consequence, the *Inia* spp. is listed as *Data Deficient* by the International Union for Conservation of Nature (IUCN) (Reeves et al., 2008). Photo-identification has the potential to be used as a tool to answer questions associated with the criteria (e.g., population size) used by the IUCN, and other organizations, to list species into appropriate threat categories and, thus, to draw attention to their conservation status. For instance,

Table 1. Photo-identification studies of river dolphin populations

Country	Species	Location	Date	Animals identified	# photographs suitable(# photographs collected)	Animals resighted	Maximum time (and distance) between resightings	Source
China	Lipotes vexillifer	Yangtze River	March 86 Dec 87	0	0 (1,000)			Hua et al., 1990
China	L. vexillifer	Yangtze River between Zhenjiang and Hukou	May 89 May 90	7	84 (1,178)	3	373 days (200 km)	Zhou et al., 1998
Nepal	Platanista gangetica	Karnali River			? (1,200)			Smith & Reeves, 2000
Venezuela	Inia	Cinaruco River	Nov 93 June 94	6	32 (2,184)	6	186 days	McGuire & Winemiller, 1998
Bolivia	Inia	Tijamuchi River	Jan 98 Sept 99	2	27	2	239 days	Aliaga- Rossel, 2002
Peru	Inia and Sotalia	Pacaya- Samiria Reserve	1991 2000	72	~270 (9,000)	25	91 months (220 km)	McGuire & Henningsen, 2007
Colombia	Inia	Amazon River and adjacent areas	1991 1993	20	400 (3,600)	Yes		Trujillo, 1994
Colombia	Inia	Amazon and Orinoco River and adjacent areas	Feb 07 Aug 09	57 R* 40 L	795 (6,855)	16	23 months	This study

^{*}R = Inia dolphin's right side identified; L = Inia dolphin's left side identified

while there are robust statistical means to estimate river dolphin population sizes and densities using boat-based surveys, photo-identification effort has been recommended in high-density areas (e.g., confluences and lakes) where boat-based surveys might provide biased estimates (Gomez-Salazar et al., 2011b).

Previous photo-identification studies of river dolphins did not have the capabilities to assess the full potential of research using natural marks. For instance, analog cameras provided a significantly lower quantity and quality of pictures when compared with current digital images (see Table 1). In some cases, photographs were black and white, thus discrimination of colors and details of any natural marks were poor (e.g., Trujillo, 1994; McGuire & Winemiller, 1998). Also, little information was available about the permanence of natural marks as well as the reliability of these marks for identifying individuals and following them over the long term. Hence, using such marks could lead to seriously biased population estimates (e.g., Stevick et al., 2001). And finally, some studies have limited their number of analog photographs taken due to economic constraints (e.g., Aliaga-Rossel, 2002). In summary, more information about the prevalence and reliability of natural marks in Inia populations is needed.

Most of the information available regarding Inia dolphins' natural marks come from studies where individuals have been captured and released (e.g., Martin & da Silva, 2006). Inia dolphins are grey when born and immature, and grey, pink, or blotched pink when adults. Also, when Inia adults are physically active, their coloration can become lighter (Best & da Silva, 1989a, 1989b; da Silva & Martin, 2000). Inia dolphins' natural marks vary with gender and age. For instance, at least in Brazil, due to intense inter-male aggression, males are larger, pinker, more heavily scarred, and have more life-threatening injuries (e.g., broken bones) than females (da Silva, 1994; Martin & da Silva, 2006). Juveniles are less scarred than adults, and toothrake scars are not present in animals during their first 2 y of life (Parra & Corkeron, 2001; Martin & da Silva, 2006). Some of the most permanent marks are found around the blowhole as a result of depigmentation of the skin due to biting from conspecifics and/or contact with rough surfaces such as flooded vegetation (Martin & da Silva, 2006). Other areas of the body, such as pectoral fins and flukes, also have permanent wounds (Martin & da Silva, 2006), but these areas are not often photographed in the wild. In summary, Inia dolphins have conspicuous natural marks that, if photo-identified, could be used to develop noninvasive and longterm studies in some areas of their distribution. Invasive methods for identifying individuals, such as tagging, may have negative impacts on survival or reproduction (e.g., Saraux et al., 2011), thus noninvasive methods, such as photo-identification, are preferable, particularly for those species for which we have conservation concerns.

This study was undertaken to evaluate photo-identification methods of Inia dolphins by (1) describing natural marks suitable for the recognition of individuals, (2) evaluating the permanence of these natural marks, and (3) estimating the proportion of individuals identifiable within the population. As also suggested by McGuire & Henningsen (2007), we predicted that, through the use of digital cameras, the quality and efficiency of the photo-identification of Inia dolphins will improve compared to previous analogue studies (e.g., Trujillo, 1994). Our goal is to provide the basis of a standard operating procedure for photo-identification studies of Inia dolphins in the Amazon and Orinoco Rivers, to suggest how this methodology might be improved, and to encourage the creation of long-term population monitoring programs using this tool.

Materials and Methods

Field Methods

Surveys were carried out from February 2007 to August 2009 in two locations of the Colombian Amazon and Orinoco river basins within study areas comprising approximately 160 and 240 km², respectively (Figure 1). Groups of river dolphins were located visually from a 6-m boat, with a 25-hp outboard engine, at approximately 2-m observation height. A group of river dolphins was defined as animals that were seen together within 250 m of the boat, likely engaged in the same activities (Gomez-Salazar et al., 2011a). We approached groups at a distance of approximately 100 m, and photographic effort started only if individuals remained within 100 m of the boat. The effort ended after 30 min or when individuals could no longer be followed. Digital color photographs were taken using a Nikon D200 (70 to 300 mm lens) and a Nikon D80 (80 to 135 mm lens) at a resolution of $3,872 \times 2,592$ pixels, which were saved in JPEG format. Photographs were taken of all individuals regardless of presence or conspicuousness of marks.

Two types of surveys were conducted. The first (December 2007 through January 2008 and December 2008 through January 2009) was specifically designed for photo-identification. These surveys consisted of encounters during which each encounter was with one group of river dolphins. For each encounter, the location (using a Geographic Positioning System [GPS]), group size, group composition (recorded as the number of adults/juveniles and calves which are dark grey

and less than 1 m long), and habitat type (e.g., main river, tributary, confluence, lake) were recorded. The second type included opportunistic surveys during which encounters were not recorded because observers did not note the identity of the group being photographed.



Figure 1. Study areas in the Colombian Amazon and Orinoco river basins

Photographic Analysis

Picture quality-Each image of Inia dolphins received a Quality rating (Q) between 1 (very poor photograph - not useful) to 5 (very good quality - ideal) (Ottensmeyer & Whitehead, 2003; Auger-Méthé & Whitehead, 2007; Auger-Méthé et al., 2009). The O rating did not depend on the markings of each individual. To do this, first, we assigned a rank from 1 to 5 on each of the following photographic criteria: focus, exposure (contrast of the body against the background), orientation of the dorsal ridge (the angle that the dorsal ridge formed in relation to a plane perpendicular to the axis of the lens), parts of the body visible above water, and size (proportion of the frame occupied by the body). Second, we assigned as the overall Q value the lowest score given to any of the criteria evaluated. For example, an image in perfect focus (5), with a good exposure (5), in which the flank and dorsal ridge of the animal are visible (5), and in which the body of the individual occupies approximately 40% of the frame (5), but in which the dolphin is swimming towards the camera (angle $= 90^{\circ}$ (1), would have an overall Q rating of Q1.

Photographic Matching-We identified and described major mark types of Inia dolphins by using all $Q \ge 3$ photographs. Each mark on each photograph was assigned a mark type (e.g., Trujillo, 1994; McGuire & Henningsen, 2007; Table 3). To facilitate the matching of individuals across surveys, we created a sketch of each distinctive individual per survey and assigned it a temporary catalog number. Best pictures and sketches of each distinctive individual were used to match individuals between surveys by eye. We selected the best picture of each distinctive individual by cropping the picture to contain only the dolphin's body and by enhancing the contrast of the picture using Adobe Photoshop. To assist the matching of individuals by eye, we recorded the mark types and the coloration of each distinctive individual on each survey on a spreadsheet and used these characteristics to find similar individuals in other surveys. We then visually compared the best image of the individuals from each of the surveys to one another, starting with the individuals that appeared to be most similar. We repeated this process separately for photographs taken from both the left and right sides of the Inia dolphins. Each distinct individual was assigned a final left-side and rightside catalog number. All left-side individuals were compared to all right sides to cross-verify identity when possible. For each individual, we recorded the coloration on the flank (grey, pink, or blotched pink) as well as the shape, color, and location on the body of each visible mark. To reduce the probability of errors, at least two people with experience in photo-identification matched each pair of individuals, and the matching was limited to 2 h per person per day.

Permanence and Prevalence of Marks

The permanence of each mark type was evaluated by using distinctive individuals with conspicuous marks such as large wounds, pigmentation patterns, and nicks that were resighted more than once. These animals were followed across resightings within and between surveys to evaluate which mark types changed over time. For each mark type, we calculated the gain and loss rate following the methods of Auger-Méthé & Whitehead (2007). Best photographs ($Q \ge 3$) of all sightings of an individual were displayed on the screen simultaneously to record the marks that were gained, lost, and those that remained the same in subsequent sightings. For each mark type, we calculated the rate of gain (total number of marks gained/sum of the number of months between the first and last sighting of each individual) and the rate of loss (total number of marks lost/sum of the number of months between the first presence of a mark and its last presence or first absence noted). Subsequently, we calculated the prevalence of each mark type across all individuals, defined as the proportion of animals that had a specific mark (Auger-Méthé et al., 2010). Prevalence of marks was estimated for both sides of each dolphin and by using high-quality photographs ($Q \ge 4$).

Proportion of Individuals Identifiable

Based on the gain and loss rates, prevalence of mark types, and their location on the body, we classified these mark types into two categories: (1) reliable and (2) supplementary (see "Results"). The proportion of well-marked animals, defined as individuals with the presence of at least two reliable marks, was calculated. We examined all high-quality photographs ($Q \ge 4$) to estimate the proportion of well-marked individuals in the population.

Results

A total of 6,855 photographs were taken during nine surveys conducted in the Amazon and four surveys conducted in the Orinoco. Of these photographs, *Inia* dolphins were present in 3,734 of the frames, of which 795 were suitable for analysis ($Q \ge 3$) (Table 2), and most of these were focused on the dorsal ridge and the flank. Based on the definitions from previous studies, we found and described eight mark types for *Inia* dolphins: (1) pigmentation patterns, (2) nicks, (3) wounds, (4) scratches, (5) scrapes (e.g., McGuire & Henningsen, 2007), (6) bends in the dorsal ridge, (7) white marks, and (8) black marks (Table 3; Figure 2).

Marks were classified as either reliable or supplementary. Reliable marks lasted for a period of at least 22 mo and were located on *Inia*'s most photographed body parts (dorsal ridge and flank). Reliable marks have low gain (≤ 0.05 per individual per month) and loss (≤ 0.07 per mark per month) rates and are generally prevalent in the population, being found in many $Q \geq 3$ images (Table 4). Reliable marks included pigmentation patterns on the dorsal ridge, nicks, bends, and wounds. Pigmentation patterns on the dorsal ridge are spots of unique and irregular shapes. These have the second highest prevalence values and very low gain and loss rates. Nicks are small indentations in the dorsal ridge that allowed us to match individuals using their left and right sides. Although not as prevalent as pigmentation patterns, they have very low gain and loss rates. Even though wounds and bends were not prevalent (< 0.24), these mark types were very conspicuous, did not change during the study, and, thus, are very promising, reliable mark types. Overall, reliable marks are promising for the long-term identification of *Inia* dolphins in further studies given that they are prevalent and easy to quantify.

		# of photographs							
		Am	azon	Ori	1000				
Survey dates	# days	Total	$Q \ge 3$	Total	$Q \ge 3$				
Feb 07	3	301	47						
July 07	1			142	46				
Dec-Jan 08	9	865	125						
Feb 08	2	149	23						
May 08	1			281	96				
June 08	3	154	37	145	17				
July 08	1		4	186	36				
Oct 08	1	213	69						
Nov 08	1	58	31						
Dec-Jan 09	8	1,206	256						
April 09	1	31	8						
Aug 09	1	3							
Total	32	2,980	600	754	195				

Table 2. Number of days surveyed and photographs taken for each survey

Table 3. Mark types used	to photo-identif	y Inia dolphins;	bold fonts indicate	reliable mark types.
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Mark type	Description	Body location	Color
Pigmentation pattern	Irregular shaped patch of color or discoloration	Head, neck, flank, back, dorsal ridge	Pink or grey
Nick	Indentations	Dorsal ridge	n/a
Wound	Significant losses of tissue and/or mutilations; wounds usually penetrate the skin, blubber, and underlying muscle.	Dorsal ridge, snout, peduncle, or flank	Red, brown (when fresh); later skin color
White mark	Circular or irregular patches and/or white lines; significantly thicker than scratches.	Flank	White
Black mark	Dark coloration of uneven patches	Rear part of the ridge and flank	Black or dark grey
Scratch	One or more straight, circular, semicircular, or "x"-shaped thin lines; these scars are mostly from tooth rakes.	Head, neck, flank, back, dorsal ridge	White
Scrape	Singular, parallel, circular, or semicircular lines or bands	Dorsal ridge and flank	Red or brown
Bend	Bends on sections of the dorsal ridge edge	Posterior part of the dorsal ridge	Skin color

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Supplementary marks can drastically change or disappear within a few days or months and can be located anywhere on the dolphin's body. Supplementary mark types have high gain and loss rates (> 0.10 per individual per month; see Table 4), are not prevalent (Table 4), and/or are very inconspicuous. Thus, these marks were used within surveys to assist in the matching of photographs to individuals but should not be used in long-term photo-identification studies. Supplementary marks in Inia dolphins are scratches; scrapes; black marks; white marks; and pigmentation patterns on the head, neck, and flanks. Scrapes and scratches, although very conspicuous, have the highest loss and gain rates. Scrapes disappeared from some individuals within 1 wk, and scratches disappeared from some individuals within 1 mo. White marks are circular or irregular patches and/or white lines that are significantly thicker than scratches, located

mostly on the dolphins' flanks. Black marks were located on the lower part of the dorsal ridge, and unlike white marks and pigmentation patterns on the dorsal ridge, they did not have a defined shape. Instead, they mostly look like very dark shadows. Pigmentation patterns on the Inia dolphins' heads, although conspicuous, prevalent, and easy to quantify, were not often photographed and, therefore, were only used as supplementary marks when the dorsal ridge was also photographed. Pigmentation patterns on the dolphins' flanks were prevalent: however, the shape of the pigmentation patterns on the flanks was not as well-defined by nearby "anchor" points as the ones located on the dorsal ridge or the head. Thus, pigmentation patterns on the flanks are considered supplementary. Despite this, they were one of the main features used to describe body coloration (see below). In conclusion, supplementary mark types cannot be reliably



Figure 2. Mark types used to photo-identify Inia dolphins

Table 4. Gain and loss rates calculated for five dolphins resigned by right side photographs and 11 dolphins resigned using left side photographs; the prevalence of each mark type was calculated separately for left and right side photographs with qualities $Q \ge 4$ (120 pictures).

			Prevalence			
	Rate of gain [*]	Rate of loss [†]	(proportion of individuals with mark)			
Mark type	(per individual per month)	(per mark per month)	Left side	Right side		
Bend	0.04	0.00	0.17	0.24		
Black mark	0.05	0.03	0.51	0.42		
Scrape	0.36	0.14	0.41	0.46		
Nick	0.05	0.01	0.48	0.46		
Pigmentation pattern (dorsal)	0.05	0.00	0.90	0.92		
Pigmentation pattern (flank)	0.05	0.02	0.48	0.60		
Pigmentation pattern (head)	0.00	0.00	0.58	0.62		
Scratch	0.55	0.11	0.99	0.98		
White mark	0.00	0.00	0.30	0.28		
Wound	0.05	0.07	0.01	0.10		

* (total number of marks gained)/(sum of the number of months between the first and last sighting of each individual)

[†] (total number of marks lost)/(sum of the number of months between the first presence of a mark and its last presence or first absence noted)

used for the long-term identification of *Inia* dolphins and should solely be used to match individuals within surveys.

In this study, coloration on the Inia dolphins' bodies was considered a supplementary mark that assisted in the identification of individuals in the Amazon, mostly within surveys. All dolphins identified in the Orinoco were grey. In contrast, some individuals identified in the Amazon were grey (43% left side, 49% right side), some were pink (14% left side, 17% right side), and some were blotched pink (43% left side, 32% right side). We investigated the color variation of identified Inia dolphins within and between surveys in the Amazon. We found that the tones in the coloration of the pink and blotched pink animals changed (Figure 3). These tones could change within minutes of when a photograph was taken as a result of fluctuations in the Inia dolphins' physical activity, or alternatively due to differences in light levels when photographs were taken. Although these tones changed, becoming lighter or darker, the coloration pattern did not alter, as also suggested in other studies (e.g., Trujillo, 1994; McGuire & Henningsen, 2007). Moreover, individuals seemed to retain some symmetry in the coloration patterns for left and right sides, which, in turn, acted as a supplementary feature when matching left and right sides of some individuals-at least within the same survey. Therefore, given that the color variation did not affect the visualization and features of reliable marks, the pattern of coloration on the Inia dolphins' bodies helped in the matching of individuals in the Amazon.

Well-marked animals, with the presence of at least two reliable marks, were recorded in 66

(55%) of the 120 images ($Q \ge 4$) analyzed for this purpose: 38% had two reliable marks, and 7% had three. Fifty-two percent of the images had just one reliable mark type, and 3% completely lacked reliable marks.

The photo-identification catalog consisted of information, separately for right and left side photographs, about each distinct individual: mark types present; coloration of the animal; best picture taken; Q values of the best picture taken; and, when possible, pictures of the individual from the opposite side. The majority of photographs included in the catalog were Q = 3 (57), followed by Q = 4 (32) and Q = 5 (8). In the Amazon, we identified a total of 41 individuals from photographs of their right side and 28 individuals from the left. In the Orinoco, we identified a total of 16 individuals from photographs of their right side and 12 individuals from the left. We then compared the right and left sides of these individuals. As a result, a total of seven dolphins in the Amazon and two dolphins in the Orinoco were matched with corresponding left and right side photographs. In the Amazon, 15 individuals were resighted over a period of 23 mo. In the Orinoco, one individual was resighted over a period of 1 mo (Table 5).

Discussion

This study, for the first time, evaluated the reliability of marks for the photo-identification of pink river dolphins in the Colombian Amazon and Orinoco river basins. Although the brief surfacing time of *Inia* limits the possibility of visually identifying individuals in the field (da Silva & Martin, 2000), *Inia* dolphins have marks that



Figure 3. Individuals identified and resignted after 12 mo using (a) pigmentation patterns on the dorsal ridge and (b) nicks

can nevertheless be photo-identified. In some locations, dolphins approach boats, and, although unpredictable when surfacing, they can show a large proportion of their bodies. Given that *Inia* dolphins surface conspicuously in some areas of their distribution, we conducted surveys to obtain high-quality digital images of individuals with the goal of describing useful marks for photo-identification.

Due to the higher quality of digital images and faster shooting capacities of new digital cameras relative to analog cameras, we obtained sufficient high-quality photographs (comparable only with the previous study conducted in Colombia; see Table 1) to examine the marks of *Inia* dolphins and to categorize these marks as reliable or supplementary, according to their permanence and consistency.

Reliable mark types in *Inia* dolphins (pigmentation patterns on the dorsal ridge, nicks, bends, and wounds) were defined as marks lasting for a minimum period of 22 mo. These were located on *Inia*'s most photographed body parts (dorsal ridge and flank), had low gain and loss rates, and were either prevalent or very conspicuous. Relying on a single identification mark type could lead to misidentification (Karczmarski & Cockcroft, 1998); therefore, we used at least two reliable marks to identify individuals.

Pigmentation patterns on the dorsal ridge of *Inia* are the most prevalent mark type and are often used as one of the principal marks for photoidentification of the species (e.g., Trujillo, 1994; McGuire & Henningsen, 2007). Pigmentation patterns are likely the result of discoloration of the skin, parasites, or abrasions caused by rubbing against objects or by injuries from their

Date	70223	80102	80105	80513	80612	80622	81004	81102	81203	81207	81211	90106
ID	200	200	200	200	200	200	200	200	200	200	200	200
AMZ-IR-003	CC		CC									
AMZ-IR-004	CC		CC									
AMZ-IR-030									CC	CC		
AMZ-IR-041							CC					CC
ORI-IL-009				MR	MR							
AMZ-IR-020 AMZ-IL-002	CC		CC			AR	CC	AR				
AMZ-IL-003	CC		CC				CC		CC			
AMZ-IL-005	CC									CC		
AMZ-IL-015							CC		CC			
AMZ-IL-019							CC	AR				
AMZ-IL-020								AR	CC			
AMZ-IL-023	CC	CC					CC		CC		CC	
AMZ-IL-024				-			CC		CC			
AMZ-IR-027 AMZ-IL-025								AR	CC		CC	
AMZ-IR-035 AMZ-IL-026										CC	CC	
AMZ-IL-009 AMZ-IR-033			CC						CC			

Table 5. Resighting records of identified Inia dolphins (ID)

CC = Caballo Cocha Lake, MR = Meta River, AR = Amazon River

conspecifics. Pigmentation patterns of many species remain unchanged across multiple years such as in bottlenose whales (Hyperoodon ampullatus) (Gowans & Whitehead, 2001) and blue whales (Balaenoptera musculus) (Sears et al., 1990). In contrast, pigmentation patterns in narwhals (Monodon monoceros) change over time and are only useful for matching individuals within a short period of time (Silverman, 1979). Pigmentation patterns were first described for Inia in the same Amazon location in Trujillo (1994), but it was not known whether these would be reliable marks across multiple years. Although the level of coloration of the pigmentation patterns can change over a short time scale, the pigmentation patterns on the dorsal ridge did not change over the 22 mo of the study. Long-term studies to monitor pigmentation patterns of Inia are required to investigate whether pigmentation patterns remain stable across multiple years. Nicks are indentations on the dorsal fin of many cetacean species that are often used to identify individuals, from both their left and right sides, over the long term (Würsing & Jefferson,

1990). The combination of nicks and pigmentation patterns is useful for the photo-identification of many species such as short-finned pilot whales (*Globicephala macrorhynchus*) and Atlantic white-sided dolphins (*Lagenorhynchus acutus*) (Würsig & Jefferson, 1990). Similarly, nicks and pigmentation patterns were the most promising features for photo-identifying *Inia* dolphins over the long term.

A reliable mark type, described for the first time in this study, is the presence of a bend in the dorsal ridge. Bends were located in the thinnest areas of the dorsal ridge (low, posterior region), possibly triggered by forcible contact with inanimate objects or injuries from conspecifics. The maximum time between resightings of a bend was 2 mo, during which time no changes to the bend were observed. It is unlikely that the original shape of the ridge could be restored after bending, thus bends are a promising feature for identifying *Inia* dolphins in the long term.

Wounds are likely to last for multiple years and are the most conspicuous mark type. Given that river dolphins do not have known natural predators, wounds are presumably caused by interactions with fishing nets or cuts produced by large knives (machete) (e.g., Trujillo, 1994). Hence, these are the least prevalent mark type in our study population. Previous studies found that the majority of resightings of photo-identified Inia dolphins were of the most conspicuous individuals, identified by using wounds and large injuries (McGuire & Winemiller, 2007). The majority of resightings in this study, on the contrary, were of individuals identified by using the most prevalent mark type (pigmentation patterns on the dorsal ridge). In photo-identification studies, it is not the conspicuousness of the marks but their prevalence in the population, as well as their permanence, that will have the largest role in the identification of animals (Hammond et al., 1990). Therefore, wounds, although conspicuous and long lasting, have limited use because of their rarity.

Supplementary mark types (e.g., scratches; scrapes; black marks; white marks; pigmentation patterns on the flank, neck, and head) are, overall, not reliable over the long term but are useful for identifying individuals within the same survey. Scrapes and scratches can be formed from tooth rakes of conspecifics or can be single or parallel lines that may be produced by inanimate objects such as flooded vegetation. These marks cannot be used to identify Inia dolphins for a period longer than 1 wk and 1 mo, respectively. Scratches are highly prevalent on Inia dolphins; however, as has also been observed in other species, they have high gain and loss rates and, thus, limited persistence. For instance, scratches are similar to the linear marks and tooth rakes described in longfinned pilot whales (Globicephala melas), which are also not persistent in the population and therefore not useful for photo-identification studies (Ottensmeyer & Whitehead, 2003; Auger-Méthé & Whitehead, 2007). Black marks appear as shadows, and white marks are irregular features. It is unclear how these are formed, and although their rate of gain and loss is not as high as for scratches and scrapes, these are not prevalent in the population and are very inconspicuous. Overall, supplementary marks, such as scrapes, scratches, white marks, and black marks, should not be used to individually identify Inia dolphins except over the shortest term given their high loss and gain rates and/or lack of prevalence in the population.

Pigmentation patterns in the area around the blowhole are produced as a result of depigmentation of the skin caused by abrasion and biting from conspecifics (Martin & da Silva, 2004). These are common and distinct among *Inia* dolphins (Martin & da Silva, 2004; McGuire & Winemiller, 2007); however, they are considered as supplementary mark types in this study given that few pictures showing Inia's head are available. Pigmentation patterns on the head could become a reliable mark type only if more photographic effort is employed to photograph dolphin heads with their respective flank and dorsal ridge. For instance, bottlenose whales also have unique pigmentation patterns on their heads (Gowans & Whitehead, 2001). Therefore, an independent catalog of the heads was created, and individuals that were matched based on head photographs were compared to individuals matched by their fins to confirm identity and to test the reliability of other marks (Gowans & Whitehead, 2001). Similarly, pigmentation patterns around the blowhole were seen on the Yangtze River dolphin (Hua et al., 1990). While the Yangtze River dolphins surfaced in a manner that allowed easy photo-identification of the head, the surfacing pattern of Inia made the simultaneous photographing of both head and flank a very difficult task, at least in our study. In conclusion, pigmentation patterns in the area of the blowhole could be classified as reliable marks if future studies are able to frequently photograph the head as well as each animal's dorsal ridge and flank.

Pigmentation patterns on the flank were used to describe *Inia*'s body coloration (grey, pink, and blotched pink). Because the level of coloration on the flank changes significantly, and in many instances is very inconspicuous and difficult to quantify, it is not used as a reliable mark type over the long term. However, it was useful to assist in the identification of individuals in the Amazon within surveys and, thus, to match left and right sides of some individuals given the apparent color symmetry. However, more photographs and further analysis are necessary to validate this.

In this study, it was not possible to match the left and right sides for the majority of individuals. Although increasing the photographic effort will help obtain the high-quality photographs required to obtain matches of individuals across sides, we suspect that some individuals may remain impossible to match. For instance, the most prevalent and reliable mark type used—pigmentation patterns in the dorsal ridge—might occur on one of the *Inia* dolphin's sides but not necessarily on its opposite side. This contrasts with nicks since the silhouette of the indentations appears the same from both sides.

The proportion of well-marked *Inia* dolphins (55%) was comparable to that of other species. For instance, 84.0% of narwhals, 33.6% of long-finned pilot whales, and 66.0% of bottlenose whales are identifiable when good quality pictures are examined (Gowans & Whitehead, 2001; Auger-Méthé & Whitehead, 2007; Auger-Méthé et al., 2010).

The success of photo-identification studies of Inia dolphins is greatly influenced by the researchers' ability to obtain high-quality pictures in order to identify these reliable marks. Previous studies have suggested that it is the lack of experience and inadequate equipment that have restricted photoidentification of river dolphins (Hua et al., 1990; McGuire & Winemiller, 2007). Consequently, photo-identification can be improved using digital cameras with fast shutter speeds and long lenses by researchers with considerable experience in photographing river dolphins and/or other very fast-moving subjects. Efficiency will be higher in study areas where dolphins do not avoid boats and surface conspicuously showing a significant portion of their bodies such as at Caballo Cocha Lake on the border between Peru and Colombia, the confluence of the Meta and Orinoco Rivers on the border between Colombia and Venezuela, and Tipishca del Samiria Lake in Peru.

In addition to photo-identification, natural marks have been used in other species to assess an individual's age. For instance, Risso's dolphin (Grampus griseus) adults become lighter with age due to loss of pigment; thus, in photo-identification studies, individuals with a moderate to very high level of scarring are considered adults (Hartman et al., 2008). Marks of Inia could also be used to assess the age and sex of individuals. For example, it was suggested that color, especially the pinkness of some adult males, could potentially be a proxy for Inia's maturity (Martin & da Silva, 2006). This could be investigated by conducting photo-identification studies of marks on individuals that are already sexed, aged, and artificially marked in studies conducted in Brazil. However, these features would not appear to be useful for Inia dolphins in the Orinoco, where all individuals in our study were grey and without pink patches.

To summarize, this study demonstrates that pigmentation patterns on the dorsal ridge, nicks, bends, and wounds are reliable mark types for photo-identification studies of Inia dolphins. In addition, this study provides some direction about how to use and improve upon this methodology, and how to better evaluate other potential reliable marks such as pigmentation patterns in the area of the blowhole. Photo-identification of Inia dolphins in the Amazon and Orinoco using categorical mark types is a promising methodology when using digital cameras with long lenses and fast shutter speeds, as well as in areas where dolphins are more conspicuous when surfacing and, thus, more easily photographed. Further studies should take digital images in RAW format, which require more storage but will reveal greater detail. Despite the broad distribution of Inia dolphins in the

Amazon and Orinoco river basin, little is known about their residence patterns, population size, and social structure. Photo-identification could be an important tool in looking at these ecological data requirements and, thereby, may help to direct conservation and management actions.

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