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Monitoring of four rehabilitated Amazonian manatees

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The Amazonian manatee *Trichechus inunguis* (Natterer, 1883) is endemic to the seasonally flooded Amazonian basin rivers and wetlands. Manatees inhabit lacustrine systems and floodplains (flooded forests), in white and tannic slow-moving black waters (Best, 1984; for description of water types, see Rios-Villamizar *et al.*, 2013). During the low water season, when most of the floodplains are dry, manatees stay in the largest lakes (locally called *cochas* in Peru) and in slow current parts of deep rivers (Timm *et al.*, 1985). The IUCN classifies the Amazonian manatee as Vulnerable (criterion A3cd) as a result of poaching, increasing accidental mortality in fishing nets, impacts of climate change, loss and degradation of habitat due to deforestation, pollution from oil spills and mercury from the gold mining industry (Marmontel *et al.*, 2016). During the past several decades, this species has suffered a significant population decline throughout its range. This decline has been the result of over-hunting (Domning, 1982) for local consumption and small-scale trade (Marmontel *et al.*, 2016), which continues to this day.

The Association for the Conservation of Amazonian Biodiversity (ACOBIA-DWAZOO) is a nonprofit conservation partnership based in Iquitos, Peru, that manages the Amazonian Rescue Center (Centro de Rescate Amazónico CREA). CREA rehabilitates and releases back into the wild manatee calves that have been entangled in fishing nets, or captured after the mother has been hunted for meat and oil. In order to monitor and document the outcome of the manatee's rehabilitation program, researchers use radio telemetry tags to follow them after their release. The goals of this part of the program are: a) to monitor and document the adaptation of the animals to their new environment, b) to evaluate the released manatees' behavior towards humans, c) to describe the use of habitat during the different seasons/hydrological cycles (high and low water levels), and d) to evaluate the effectiveness of outreach efforts and assess the local citizens' attitude towards manatees. The rehabilitation process of calves normally takes around three years. During this period, the calves are kept in individual pools of approximately 1.5 m x 3 m x 1.2 m. Initially they are hand-fed with a special formula consisting of 50% powdered milk and 50% lactose-free infant formula. This diet is gradually replaced with water lettuce (*Pistia stratiotes*). At the age of two the calves are weaned from bottle-feeding and moved to a communal pool (approximately 15 m x 7 m x 1 m) with other manatees, where they are fed water lettuce only. This contrasts with their diet in the wild, based on two species of the Gramineae family, *Paspalum repens* and *Echinochloa polystachya* (up to 96% of the samples), with *Eichhornia crassipes* in third place (Colares and Colares, 2002). However, the nutritional value of *P. stratiotes* and its accessibility and easy collection made it an ideal food, containing as much as 8.6% of crude protein, compared to a maximum of 4% in *Eichhornia crassipes* (Vázquez *et al.* 1998). The experience in CREA shows that a diet limited to water lettuce is enough for the adequate growth of Amazonian manatees, which is periodically checked by weighing and measuring the captive animals. By the time calves are two years old, they are tame and approach visitors, who are allowed to touch them, for environmental education purposes. However, this may result in an unnatural behavior, with animals approaching boats when released. Therefore,

when the animals are apt to be returned to the wild, it is necessary to assess whether that situation remains. After the period in the rescue center, the animals are transfered to a pre-release area with no direct or visual contact with people, where they are maintained in semi-captive conditions for one year and are expected to lose their bond with humans. During this time they are supplied solely with water lettuce. The criteria used to select which animals will be set free are body size and time passed since their arrival at the CREA, though no standardized guideline is used.

On 22 April 2014, four Amazonian manatees named Sol, Liberty, Yuri and Yanayacu were released back into their natural habitat, in the Cocha Zapote Lake (04º18'47" S, 73º18'49" W) San Juan de Yanayacu, Tamshiyacu-Tahuayo Communal Reserve, Loreto, Peru. Yuri and Yanayacu were adults (based on Rodrigues *et al.*, 2008), whereas Sol and Liberty were juveniles at that time (Table 1). After being transported by boat from Iquitos to the release site in the vicinities of San Juan de Yanayacu village, the manatees were kept for a day in a small plastic pool, where local villagers were allowed to visit but not to touch them. Besides that, once in the release area the animals did not pass any period of time in enclosures, but they were directly freed in the wild (hard-released). An Advanced Telemetry Systems 160 MHz band VHF radio transmitter was adapted to each manatee with a leather belt adjusted with steel nuts over a PVC plate. The transmitter was wrapped in heat-shrink tubing covered with self-amalgamating tape to enhance its durability. The belt was made of materials which prevented unnecessary discomfort to the manatee and which would quickly deteriorate, allowing the tag to be released and fall off the manatee after a short period of time. Once in place, the belt was fastened around the animal's peduncle. It was assumed that the transmitters would likely not be retrieved from the animals.

The location where the manatees were released met the following requirements: a) the species was already present in the area, and b) the area was effectively protected by law, and local people were aware of the manatees' endangered status and the importance of their protection. The release area consists in a mosaic of flooded forests, rivers, lakes and lagoons, which potentially can sustain a healthy manatee population, given the large amount of suitable food and habitat. Manatees are known to be present in the area, but there is no available information about the present or past population size. However, local people agree that the species is scarcer than it was some decades ago, even if poaching is no longer reported in the area. In contrast, poaching still occurs in other areas of Peru, such as Pacaya-Samiria National Reserve and its core area[1].

The movements, behavior and adaptations of the four studied animals may be representative of ecological aspects of wild manatees. The conclusions of this study may be helpful in order to improve techniques and to correct mistakes in future releases. Monitoring the manatees was carried out during two seasons (high and low water). Field work was conducted during three continuous weeks every month, from approximately 08:00h to midday on most of the days. The researchers would follow the manatees using the strength and direction of the radio signal to locate them, using the "homing" technique (see Ryan, 2011). The following parameters were recorded in the field: habitat, water depth, social interactions between the studied animals, daily and seasonal movement patterns, and behavior towards humans; in short, those patterns which could be studied with telemetry and without visual

Table 1. Information on four rehabilitated and released Amazonian manatees. mo = month; y = year. Source: CREA internal data.

Individual	Sex	Origin	Estimated age		Weight (kg)	
			Rescue	Release	Rescue	Release
Sol	F	Puerto Sol, Ucayali River basin. Requena, Loreto 04°36'51" S; 73°33'25" W	3 mos	2 ys, 7 mos	16	82.5
Liberty	M	Libertad, Ucayali River basin. Requena, Loreto 04°31'45" S; 73°27'11" W	3-4 mos	2 ys	17	74
Yanayacu	M	San Juan de Yanayacu, Amazonas River basin. Maynas, Loreto. 04°18'32" S; 73°17'25" W	1 mo	6 ys, 5 mos	18	223
Yuri	F	Yurimaguas, Huallaga River basin Alto Amazonas, Loreto 05°54'00" S; 76°05'00"W	2 mos	6 ys, 11 mos	20	241.5

[1]Soto, A. and The Nature Conservancy. (2007) *Caza del manatí amazónico en la Reserva Nacional Pacaya Samiria*. Lima, Perú: Centro de Datos para la Conservación. Facultad de Ciencias Forestales, Universidad Agraria La Molina, Lima, Peru.

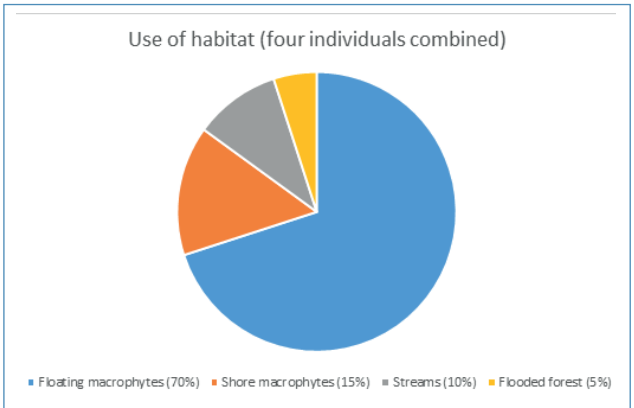

Figure 1. Use of habitat by the four individuals combined, period 22 April to 27 August 2014. *Hab. 1*: Floating macrophytes (70%); *Hab. 2*: Shore macrophytes (15%); *Hab. 3*: Streams (10%); *Hab. 4*: Flooded forest (5%).

contact with the animals. For this reason, other objectives like feeding and body condition (to assess adaptation to their new environment) could not be studied. Field monitoring took place from 22 April to 27 August 2014. On 16 July the transmitters of Yuri and Yanayacu failed to emit a signal. This may have been caused by the failure of the battery or because of loss of the belts and attached transmitters. The total number of days that the animals were radio-tracked was 97. The total number of records was 220, 118 of them corresponding to Sol and Liberty, and 102 to Yuri and Yanayacu.

The following habitats were used by the manatees: floating macrophytes, shore macrophytes, flooded forests, and streams (see Fig. 1). Floating macrophytes consist on floating aquatic vegetation, which are not rooted or associated with the shore, with the most important plant species being *P. stratiotes, Eichhornia azurea*, *E. crassipes, Montrichardia linifera, Pontederia rotundifolia, Polygonum hydropiperoides, Nymphaea* sp., *Salvinia auriculata,* and *Utricularia foliosa*. Shore macropytes refers to plants associated with shores of lakes, occupied by emergent grasses and flowering plants of *Paspalum* sp., *Polygonum* sp., with associated floating plants. Flooded forests provide little shelter and food, but were eventually used to move between more productive areas, especially during the drop of water levels, when the animals were forced to increase their movements. Streams (including Yanayacu river) have scarce vegetation cover, but connect different masses of water with better conditions, and thus were used during active movement behavior. Floating macrophytes was, by far, the most used habitat, followed by shore macrophytes. During the entire study period the juvenile individuals (Sol and Liberty) remained together in Tipashiro Lake and its surrounding areas. The adults (Yuri and Yanayacu) also remained together during the whole period, except for 27 May when both animals were at a considerable distance (1 km approx.) from one another. This contrasts with Kendall and Orozco (2014), who assure that Amazonian manatees do not seem to have strong social bonds with other individuals, apart from the mother-calf relationship. The close relationships shown by the manatees in this study could be an unnatural behavior, caused for their previous captivity.

For the subadults we estimated a minimum home range of 0.53 km^2, constructing the minimum convex polygon around the data: using a basic GIS program (*Google Earth*), the perimeter of the area bounded by the locations of the individuals was calculated. Because of intrinsic error in the methods used to obtain the locations, these data should be considered with caution.

For the adults, different areas of occupancy were defined. From 22 April to 30 June, their home range (calculated by the minimum convex polygon technique) was estimated at 2.82 km^2. On 30 June, the decrease in water levels forced the animals to leave the area, using the only natural exit, through Moena

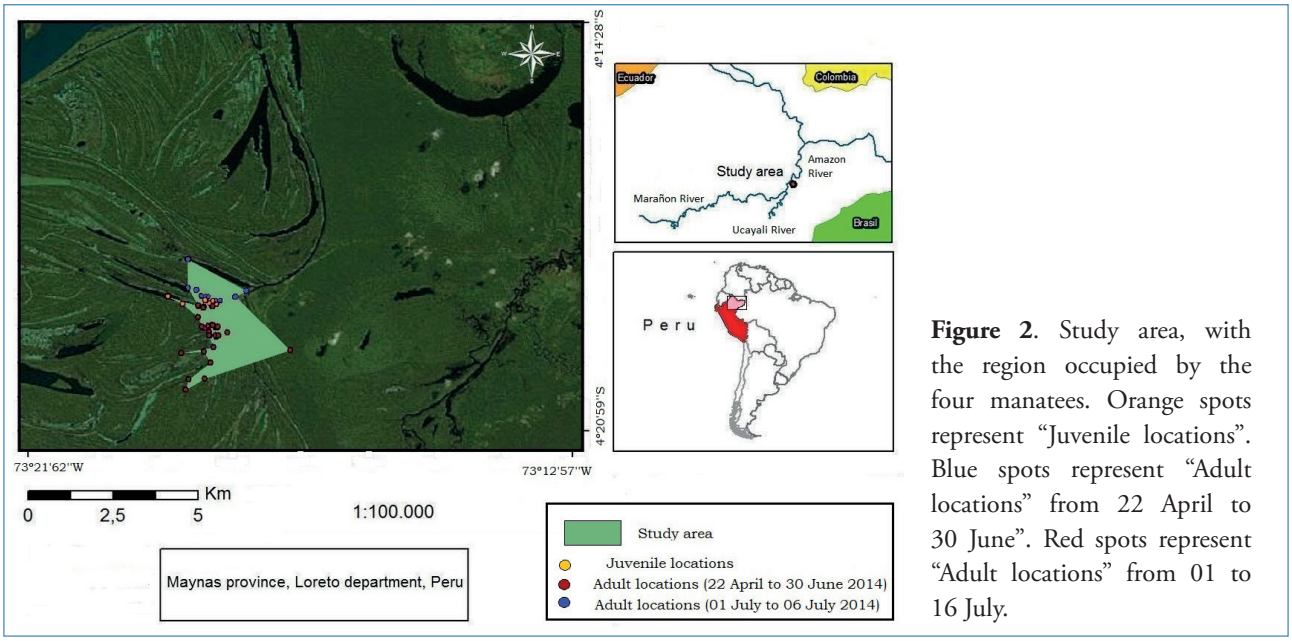

Figure 2. Study area, with the region occupied by the four manatees. Orange spots represent "Juvenile locations". Blue spots represent "Adult locations" from 22 April to 30 June". Red spots represent "Adult locations" from 01 to 16 July.

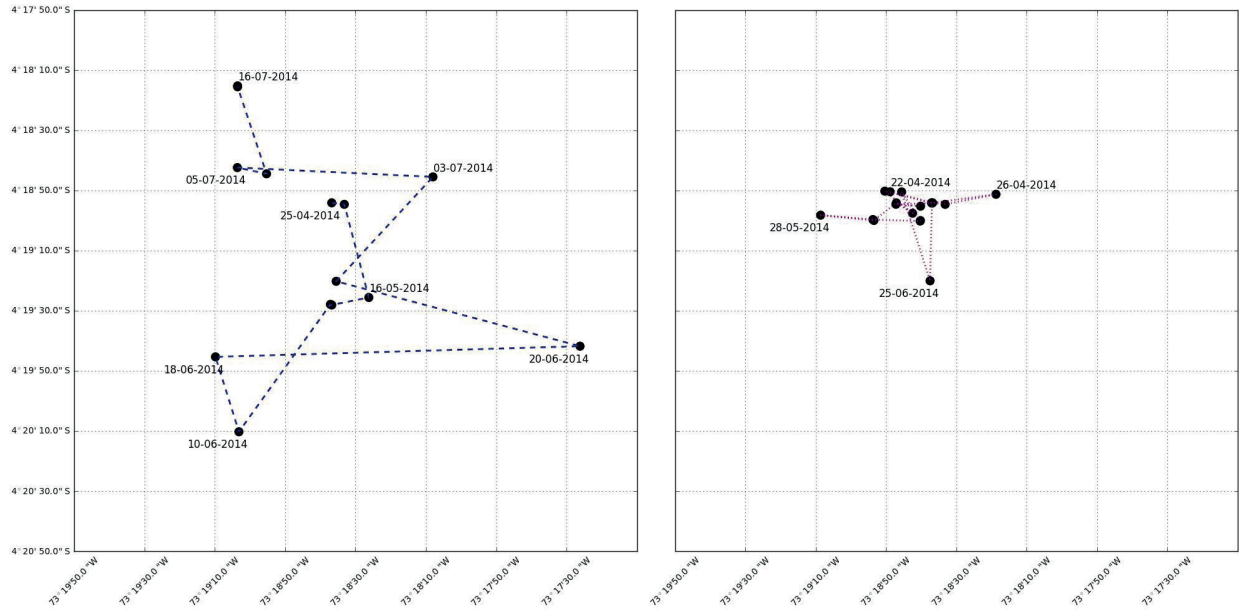

Figure 3. Movement patterns. Left image represents adults, right image represents juveniles. Different locations in the images are connected by traced or dotted lines, respectively, for representation purposes. Lines do not necessarily replicate the route actually taken by the animals from one point to another

Lake, near the area where they had been released. From 30 June to 10 July, Yuri and Yanayacu settled around the confluences of the lakes Moena and Zapote and the Yanayacu River, which was used to move from one lake to another. From 10 to 16 July (when the last radio signal of both animals was recorded), the animals moved to deeper waters in the nearby Cocha Zapote Lake. An area of occupancy of 1.45 km^2 was estimated for the period 1-16 July 2014. During the entire study period, the adult manatees, Yuri and Yanayacu, occupied an area of about 4.27 km^2. Figure 2 shows the areas of occupancy of the four individuals. The movement patterns are represented in Figure 3.

Arraut *et al.* (2009) found that Amazonian manatees selected the habitat with aquatic macrophytes and floating grasses during high water, but they were not accessible during the low water season. In contrast, in our study area these habitats were favored in both seasons. This seems to indicate that as long as depth allows it, these habitats are preferred by the manatees, and they do not need to migrate to deeper bodies of water to feed. In fact, the two juveniles (Sol, Liberty) did not leave their area of occupancy during the low water level season, since their territory was deep enough and food was plenty. Furthermore, the two adult individuals, who were forced to abandon their first area of occupancy, settled in a nearby area, although it is possible they may have migrated to Yanayacu or Amazon River after 16 July, when contact was lost. These results indicate that migration during low water level season, from seasonally flooded habitat to deep rivers and lakes where food is scarce, may be a prevalent behavior only when local conditions impose such behavior.

The total number of movements larger than 0.5 km and made over a 24-hour period was eight (8.33% of the total records). Six of them were made by the adult manatees and two by the juveniles. The longest movement recorded for an adult individual was 2.53 km on 22 June, and 1.4 km for the juveniles, on 25 April.

Water depths used by the four manatees depended on flood stage. This item was recorded using a Secchi disk. The average depth of the four individuals combined was 3.7 m. Sol and Liberty occupied the same lagoon during the entire study, and depth records decreased over the study period, from 6.85 m on 26 April, to 1.6 m on 13 August. The adults occupied different areas throughout the study period, with the minimum depth of 1.3 m recorded on 30 June. In July, when they moved to a deeper lake, the depth of that area increased to 5.5 m. So we can suggest that the drop of the water levels was the reason that motivated the adults to migrate.

There were no differences in behavior, movements, or habitat selection between the male and female manatees. This contrasts with Landeo-Yauri *et al.* (2017), in whose study "Females showed higher level of residence than males, and stayed within the release area, while males left the lake during the season of lowering-water levels". Moreover, the two adults showed a definite higher rate of mobility.

During the field study the animals were never seen directly; therefore we were unable to assess body condition or witness feeding behavior. When we approached the manatees in an unmotorized canoe they did not flee or leave the area (as was concluded by the radio signals); however, we were unable to

observe them. We concluded from this that they were showing a passive defensive behavior by submerging themselves and waiting for the threat to pass, which normally took around 15-30 min. One of the criteria for determining the effectiveness of the rehabilitation process was to check that the animals avoid human presence, what they seem to have done.

Our results show that captive rehabilited manatees can survive in the wild, if prior to their release they become unaccustomed to human presence. The evidences for this statement are: a) the four animals remained in areas with plenty of food and shelter for the whole period, b) the adults migrated to deeper lakes, with enough aquatic vegetation, during low-water levels, and c) no approaching to boats was observed for any of the individuals. When we were near they did not flee, but showed a passive defensive behavior, staying plunged. Nonetheless, these conclusions must be considered with caution, given the small sample of time and individuals considered. There is no evidence to suggest that, after the tracking period, the animals were successful in their adaptation, and less so to confirm that they have bred, thus contributing with the population recovery to their natural levels prior to big scale hunting.

A special effort was made in community outreach. Prior to the release, CREA's educational team spent four days in San Juan de Yanayacu, devoting two sessions to visiting the neighboring villages of Nuevo Junín and Ayacucho de Tipishca. In those three localities meetings were held with the authorities, who have at all times been supportive of the work done by CREA. Those meetings gave the project official recognition and prestige, and more importantly, allowed us to spread our message to the whole community. A series of activities was carried out to introduce the problems regarding manatee conservation, and explaining the importance of the species in the ecosystem. All dwellers in the three communities were invited to those activities, although the focus was on children and teenagers. The real effectiveness of the community engagement was not quantified by a balanced methodology, such as interviews. Nonetheless, the author was living in San Juan de Yanayacu from 22 April to 27 August 2014 and was able to check how the project was accepted by the whole community, their awareness of the species' situation, and a complete end to poaching of the species, at least by the residents of San Juan de Yanayacu. This acceptance was helped by the fact that two families (of the ten who live in the village year-round) received payment, for providing services to the researcher during the period he spent in the village. Moreover, every family in the community benefited from the rise in the number of tourists and others visitors. At the end of the research period, the whole population of San Juan de Yanayacu expressed their desire to foster future manatee releases. We therefore suggest that the Amazonian manatee rescue program is successful, but can be improved. The main hindrance for the future success of the program is the economic limitations of the organizations involved. If this was solved, it would be possible to obtain radio telemetry materials of higher quality. Equally, future researchers should use more standardized protocols and come into contact with specialists, to increase our knowledge in Amazonian manatee ecology, with positive impacts on conservation.

The following recommendations are made for future releases:

a) The use of transmitters with longer duration and emission power is recommended, as well as belts with more durable stitching - all these details are very important if we are to locate the animals for longer periods and over longer distances;

b) Considering that the local people are committed to the conservation of the species, we would highly recommend the future release of manatees in this area; it exhibits perfect conditions for a recovery of the population of the species, which would be accelerated by future releases;

c) The educational program returned very positive results, so it is necessary to continue with this work; however, we should also include a standardized methodology that allows the researchers to evaluate the extent of the change in perception that the community has for the species;

d) Given the warm response of the local people throughout the project, we recommend that some kind of reward should be given for this attitude; for example, educational materials could be given to children, and we should look for ways of hiring some people all year round, not just during the presence of the researchers. One possibility is to introduce the figure of "manatee guardian", for instance;

e) For future releases, use of the same area is highly recommended, given the quality and quantity of optimal habitat and the commitment of local authorities and residents.

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