



## Mating sanctuary for whitespotted eagle rays (*Aetobatus narinari*) in Brazilian southeastern waters

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### ABSTRACT

This article provides the first records of courtship behavior and a mating event of free-ranging whitespotted eagle rays (*Aetobatus narinari*) in Brazilian waters. These observations shed light on utilizing the marine no-take zone surrounding the Anchieta Island State Park as a mating site in the South Atlantic region. Sightings of the species in shallower areas notably increased throughout summer months (from January to March, 2023 and from January to February, 2024), which aligns with its known reproductive period. Specifically, the mating event occurred in March 2023, followed by observations of courtship behavior in January 2024. While the population dynamics and impact of fisheries remain poorly understood, the species is frequently captured by traditional fishing fleets and traded locally. This reiterates the importance of the local Marine Protected Areas (MPAs) and underlines the urgency for their expansion, alongside the need for further studies that support specific management actions.

**Keywords:** Autecology, Citizen science, Elasmobranchs, Marine protected areas, Endangered species

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The Elasmobranchii (sharks, rays and skates) is one of the most diverse group of vertebrates, not only in terms of species richness but mainly by the variety of strategies displayed by their representants (Compagno, 1990). These organisms

can be classified according to where they spend most of their life cycle, living at oceanic or coastal zones and sporadically interchanging the use of such habitats due to feeding and reproductive purposes (Compagno, 2008; Camhi et al., 2009; Ajemian et al., 2012). Regarding reproduction, seasonal migrations to inshore areas have been suggested (e.g., bays and estuaries), either for mating or parturition (Lessa et al., 1986; Araújo et al., 2016; Rangel et al., 2018). However, although the use of more sheltered regions reduces natural mortality by predation and/or competition in earlier life stages, this behavior enhances the threats over the species since those habitats are highly impacted by human activities (i.e., fishery target, bycatch, habitat destruction and pollution) (Camhi et al., 2009; Ferretti et al., 2010; Dulvy et al., 2017, 2021).

Today, more than 30% of the marine elasmobranchs are classified in threatened categories (Dulvy et al., 2021) and a fewer than a third of them are coastal related species (Camhi et al., 2009). Notably, the Aetobatidae family stands out within this group, with all its constituent species classified as either vulnerable or endangered by the International Union for Conservation of Nature (IUCN). Particularly, the circumstances of the white spotted eagle ray, *Aetobatus narinari* (Euphrasen, 1790) is troubling. Encompassing an expressive part of its territory (Sales et al., 2019), populations living at Brazilian waters lack information (i.e., population biology and fishing effects) (ICMBio, 2016) to enable a proper evaluation about its conservation status, and is considered data deficient (SiBBr, 2023). Moreover, its benthopelagic habit associated with movements to nursery grounds on shallower regions (Yokota and Lessa, 2006; Bassos-Hull et al., 2014) expose them to several fisheries performed across a range of depths (e.g., beach seines, gillnets, corral fishing and longlines) (Tagliafico et al., 2012; ICMBio, 2016; Rangel et al., 2021).

These factors likely hinder the population reestablishment of *A. narinari*, as this species exhibits delayed maturation and requires a minimum of one year to bear up to five embryos (Schluessel et al., 2010; Araújo et al., 2022). Given this scenario, since July 2022 the species

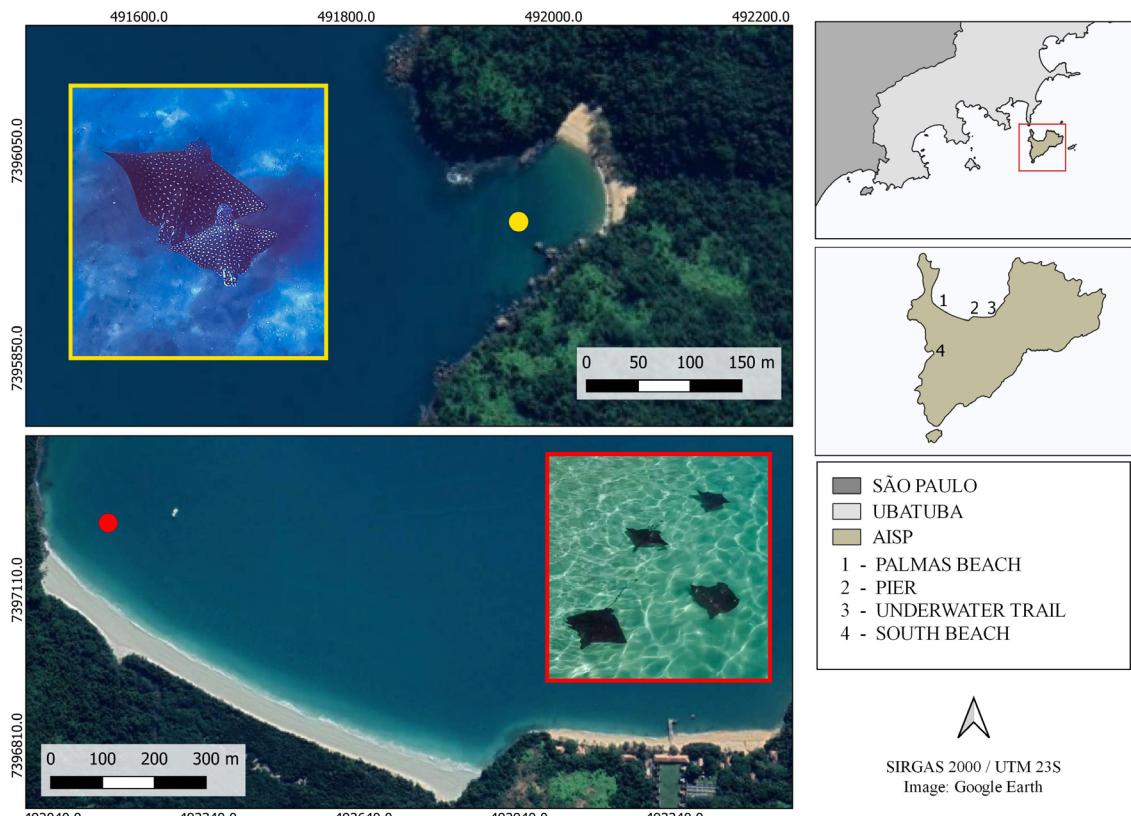
occurrence in the surrounding waters of the Parque Estadual Ilha Anchieta (Anchieta Island State Park – AIS) has been systematically monitored. This monitoring initiative is part of an educational program led by the Diving into Conservation project which engages AIS visitors as citizen scientists, providing opportunistic records of the species. Additionally, volunteers affiliated with the project conduct sampling campaigns during both winter and summer months using drones, Baited Remote Underwater Videos (BRUVs) and Diver Operated Videos (DOVs).

AIS is a protection area (IUCN Category II) (Dudley, 2008) located 8 km off the southeastern Brazilian coast (23° 32' 22" S - 45° 3' 54" W). Encompassing a 17 km perimeter, the park seeks the full protection of natural resources, covering a significant remnant of the Atlantic Forest (Guillaumon et al., 1989). Despite its relevance for the associated terrestrial fauna, AIS's boundaries have been safeguarded by a marine no-take zone since 1983 (SUDEPE, 1983). Both protection areas collectively form an integral component of the Marine Protected Areas (MPAs) network (São Paulo, 2008), contributing to the broader framework of Geobiodiversity Protection Zonation within the larger Marine Environment Protection Area of the North Coast of São Paulo state (APAMLN). Activities are restricted inside them, permitting only low-impact tourism with the development of sustainable and environmental education activities such as hiking, wildlife watching, swimming and diving as well as scientific research (Forestry Foundation, 2020). Importantly, these areas serve as critical refuges for numerous commercially valuable species (e.g., *Micropogonias furnieri*, *Epinephelus marginatus*, *Caranx latus*) which are currently threatened according to IUCN (IUCN, 2024).

Four different locations were monitored daily during winter (July/2022 and August/2022) and summer (from January/2023 to March/2023 and from January/2014 to February/2024) campaigns. Baited Remote Underwater Videos (BRUVs) were deployed near the pier and South Beach of AIS, whereas Diver Operated Videos (DOVs) were utilized at South Beach, Palmas Beach, and along the underwater trail. Concurrently, citizen science activities were conducted at both South

and Palmas Beaches (Figure 1). As the South and Palmas Beaches are part of major bays in the island, two transects were pre-established at each bay side, being sampled twice: first the diver swam right above the rocky reefs towards the ocean and then returned above the bottom sand. Due to the extension of the underwater trail and its morphology, a single transect was set parallel to the rocky reef. Species were recorded by GoPro

Hero 9® or GoPro 360® in both methodologies, taking from three to ten minutes per DOV and an hour per BRUV. In January/2024, pilot samplings were conducted using the DJI mini 2 drone to enhance record acquisition efficiency. This aimed to expand the coverage area more efficiently, directing the DOV teams to the beaches where specimens of *A. narinari* had been previously identified by the drone.



**Figure 1.** Map of the Anchieta Island State Park pointing out the surveilled locations (numbers) and highlighting the Palmas and South Beaches (red and yellow spots), places where the courtship behavior and the mating event of the *Aetobatus narinari* were recorded.

On the morning of March 26, 2023, the project's volunteers were performing a DOV sampling at the bay of South Beach when an adult couple of *A. narinari* was recorded. They were seen fifteen minutes apart next to the rocky reefs, exhibiting evasive swimming behavior upon detection of the diver's presence. The male and female individuals were around one meter and 1.60 meter wide, respectively, and both lacked their tails. Approximately fifteen minutes later (at 7:30 AM) the female was sighted again, swimming randomly

over the sand bottom in the middle of the bay. After nine minutes displaying this behavior, the male appeared and initiated pursuit. First, the male kept a certain distance above the female, joined the tips of its pectoral fins and dipped quickly towards the female, gripping its dorsum (Figure 2 a-c). Subsequent to this approach, the male nibbled the female's dorsal skin and positioned itself behind the female (Figure 2 d), albeit no scars were discerned. After this courtship behavior, the male gripped the posterior portion of the female's right

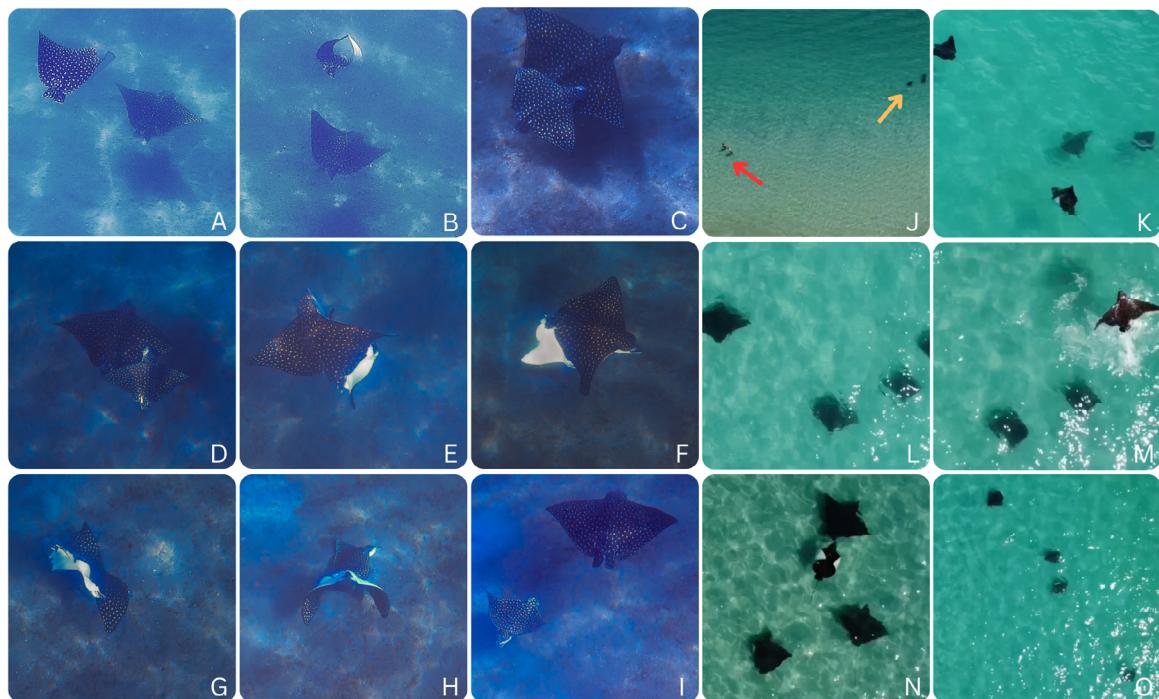
pectoral fin and assumed a ventral position, inserting only one clasper into its cloaca (Figure 2 e-f). While coupling, the specimens rotated, sinking down with the female keeping motionless, laying on the seafloor, and ending below the male (Figure 2 g). The female then reoriented itself upwards, hindering further clasper insertion, and swam away from the male who attempted pursuit, leading both out of sight (Figure 2 h-i). The entire mating event took two minutes and no cloud of semen was observed, probably due to the suspension of sand particles by the rotation of the specimens above the seafloor.

Later, at 11:00 AM of January 15, 2024, the project's volunteers were drone surveilling the area when four adult specimens of *A. narinari*, one female and three males, were recorded at Palmas Beach. They were initially on the surf zone close to bathers (Figure 2 j), with males chasing the female, which started to evade their attempts and swam to offshore. The female was larger (around 1.70 meter wide) than the males (between 1.40 – 1.60 meter wide), one of which lacked its tail. Throughout the observed behavior, the female maintained a leading position ahead of the males (Figure 2 k), evading their outflanking attempts towards the surf zone (Figure 2 l). In one of these attempts, the largest male leapt out of the water in an effort to intercept the female (Figure 2 m); however, it swerved, keeping the front position. A few seconds later, one of the smaller males attempt to grip the posterior portion of the female's right pectoral fin, which responded by accelerating and eluding the male advance (Figure 2 n-o). The group swam from the central region of Palmas Beach to its left extremity, ultimately reaching the surf zone. But none of the males' attempts at mating were successful. The entire event lasted six minutes, after which all individuals swam offshore, disappearing. The complete video of both records are available on [Zenodo](#).

Similar courtship and copulating behaviors were noticed in North Atlantic and even Old World specimens (Sales et al., 2019), at the time identified as *A. narinari*. Observing unsuccessful mating attempts, Tricas (1980) verified the male approximation from above, trying to mount

and nibbling the female's dorsum, as well as the female containment, by biting its pectoral fins. Nibbling and holding behaviors were also observed in free-ranging (McCallister et al., 2020) and captive specimens (Uchida et al., 1990). During copulation, males assumed an abdomen-to-abdomen position with insertion of one clasper and rotation of both specimens in a clockwise direction, ending with one of them laying on its back above the tank floor. As previously described by McCallister et al. (2020), *A. narinari* exhibits a mating behavior consisting of five stages (Yano et al., 1999), although several strategies might be employed in the courtship stage conditionally to female receptiveness. Such strategies may include males swimming around females, attempts to corral and more abrupt approaches after failed mounting attempts, sometimes directly progressing to holding the female's pectoral fins or inflicting dorsal gouges that may leave scars (Tricas, 1980).

Use of coastal and insular regions by elasmobranchs for mating and parturition is well recorded (Lessa et al., 1986; Wetherbee et al., 2007; Hazin et al., 2008; Karlovic et al., 2021), being essential to identify and monitor such places for conservation of threatened species (Anderson et al., 2021). To our knowledge, this study presents the first documented instances of courtship and mating behavior of *A. narinari* in Brazilian waters, underscoring AIS as a significant mating site for the species. Although no newborns or young of the year were found in the monitored areas, we believe that the surroundings of the AIS serve as a nursery habitat. Sightings of the species in the AIS's shallow waters (< 5 meters deep) increased from January to late March 2023, when ten records were communicated to us by citizen scientists and other two were made by the project's team. Moreover, between January and February 2024, there was an 8 percent increase in species records despite the lower sampling period. As suggested by Tagliafico et al. (2012), the mating period of the *A. narinari* starts on February right after parturition, aligning with our findings and the increasing records of mature specimens in other Brazilian regions (e.g., Araújo et al., 2022).



**Figure 2.** Captures of the mating and courtship events of the *Aetobatus narinari*, showing: the DOV record of (A) the chasing stage, with the male above the female, keeping a certain distance; (B) the mount attempt, with the male joining its pectoral fins and dipping into the female's direction; (C-D) the holding behavior, where the male positioned itself behind the female's pectoral fin after nibbling its dorsum; (E) copulation, where the male assumed a ventral position, (F) inserting one clasper into the female's cloaca and (G) both specimens rotated, sinking down to the seafloor; (H-I) separation, with the female assuming a position that hindered clasper insertion, separating from the male and swimming away. The drone record of (J) the beginning of courtship behavior, with specimens on the surf zone (yellow arrow, the reddish one indicate bathers); (K) the chasing stage, with three males behind the female; the intercept attempts were (L) the males tried to outflank the female to the surf zone; (M) they jumped out of water and (N) tried to hold the female by gripping its pectoral fins; ending with (O) the female escape by speeding up its velocity.

All together, these factors reiterate the important role played by the MPAs network and underline the necessity to expand their boundaries. Due to its conservation status in São Paulo waters (*i.e.*, near threatened – São Paulo, 2018), populations of *A. narinari* have been caught by traditional fleets and traded on local markets (Rodrigues et al., 2021), although its population trends remain unrecognized (ICMBio, 2016). Additionally, the observation of specimens lacking tails suggests incidental capture in local fisheries, as tail docking is a common practice when individuals are caught and subsequently released.

While such aspects were not assessed, clarifying its truly conservation status and supporting specific management programs,

the establishment of newer MPAs seems to be the most urgent action for species conservation. For example, the Parque Estadual Marinho Tartaruga-de-Pente (Hawksbill Turtle's Marine State Park – HTMSP), which is still in the proposal stage (Forestry Foundation, 2021). Ensuring AISPs and other local MPAs effectiveness consist in one of the greatest challenges for marine conservation in Brazil (Motta et al., 2021), especially due to intense anthropogenic activities in their surroundings (Imoto et al., 2016; Angelini et al., 2018; Pincinato and Gasalla, 2019). In this regard, the creation of the HTMSP will strengthen the MPAs network, enabling the use of financial funds for fully protected areas (*e.g.*, environmental compensation; Brazil, 2002) and supporting a

focused management for the area with its own definitions of use and control actions. Additionally, further studies that elucidate the unknown aspects about the species' biology and ecology, such as expanding surveillance to identify its key habitat on the AISP's surroundings, are also demanded.

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## AUTHOR CONTRIBUTIONS

T.C.K.: Conceptualization; Formal Analysis; Methodology; Visualization; Project Administration; Funding Acquisition; Writing – original draft; Writing – review & editing.  
 G.R.L.: Investigation; Formal Analysis; Methodology; Visualization; Writing – original draft; Writing – review & editing.  
 A.R.L.; I.L.A.; Y.A.Y.; A.A.V.A.: Investigation; Formal Analysis; Writing – review & editing.  
 L.S.A.; F.G.B.; M.T.A.: Investigation; Formal Analysis; Methodology; Writing – review & editing.  
 F.S.M.C.: Investigation; Formal Analysis; Methodology; Writing – original draft; Writing – review & editing.  
 C.Y.K.: Investigation; Methodology; Writing – review & editing.  
 L.C.C.: Conceptualization; Formal Analysis; Methodology; Visualization; Project Administration; Funding Acquisition; Writing – review & editing.  
 J.M.S.: Investigation; Formal Analysis; Methodology; Writing – review & editing.  
 P.S.M.: Formal Analysis; Resources; Writing – review & editing.  
 J.F.D.: Formal Analysis; Funding Acquisition; Supervision; Writing – review & editing.

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