

Research Article

Vertical movements of the southern stingray, *Dasyatis americana* (Hildebrand & Schroeder, 1928) in the Biological Reserve of the Rocas Atoll, Brazil

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ABSTRACT. The southern stingray, *Dasyatis americana*, one of the most common species of rays of the family Dasyatidae, is distributed in tropical and subtropical waters of the western Atlantic Ocean. In Brazil, it occurs especially in the northeast coast, being usually sighted in Fernando de Noronha Archipelago and in the Rocas Atoll. The present research sought to elucidate the pattern of vertical movements of the *D. americana* inside the Rocas Atoll and in its surroundings, through the use of satellite electronic transmitters PSAT (Pop-up Satellite Archival Tags), as well as to understand the role of Rocas Atoll in the conservation of this species. The vertical distribution of depth and temperature were obtained from two adult female stingrays, identified as Ray I and Ray II, measuring approximately 95 and 108 cm disc wide respectively. The results indicated a strong preference for both specimens for the shallow and warm waters of the Rocas Atoll. Ray I and Ray II remained 91.4% and 86.3% of the monitoring period, respectively, in warm waters, at temperatures >28°C and up to 5 m in depth. The results show that areas with shallow waters constitute one of the essential habitats for the species in Rocas Atoll, with a likely occurrence of site fidelity.

Keywords: Dasyatidae, essential habitats, animal behavior, site fidelity, northeastern Brazil.

Desplazamientos verticales de la raya-látigo americana, *Dasyatis americana* (Hildebrand & Schroeder, 1928) en la Reserva Biológica del Atolón de las Rocas, Brasil

RESUMEN. La raya-látigo americana *Dasyatis americana*, una de las especies más comunes de la familia Dasyatidae, se distribuye en aguas tropicales y subtropicales del Océano Atlántico occidental. En Brasil, ocurre sobre todo en la costa noreste, siendo comúnmente avistada en Fernando de Noronha y en el Atolón Las Rocas. En la presente investigación se analizó el patrón de desplazamiento vertical de la *D. americana* al interior del Atolón Las Rocas y en sus alrededores, mediante satélites transmisores electrónicos PSAT (Pop-up Satellite Archival Tags), así como la comprensión del rol de este atolón en la conservación de esta especie. La distribución en profundidad y temperatura vertical se obtuvieron de dos hembras adultas de rayas, identificadas como Raya I y Raya II, que medían aproximadamente 95 y 108 cm de ancho de disco, respectivamente. Los resultados indicaron una fuerte preferencia de los dos especímenes por aguas poco profundas y cálidas del Atolón Las Rocas. Raya I y Raya II permanecieron 91,4% y 86,3% del período de seguimiento, respectivamente, en aguas cálidas con temperaturas >28°C y hasta 5 m de profundidad. Los resultados muestran que las zonas de aguas someras constituyen uno de los hábitats esenciales para esta especie en el Atolón Las Rocas, e indican una probable fidelidad con este lugar.

Palabras clave: Dasyatidae, hábitats esenciales, comportamiento animal, fidelidad al lugar, noreste de Brasil.

INTRODUCTION

The southern stingray, *Dasyatis americana* (Hildebrand & Schroeder, 1928) is one of the most common members of Dasyatidae family (Last & Stevens, 1994). The species can reach up to 150 cm of disk width (Figueiredo, 1977) and has a late sexual development:

females are mature from 75 cm of disc width and males from 51 cm disc width (Henningesen, 2000; Ramírez-Mosqueda *et al.*, 2012). *D. americana* is often found in tropical and subtropical waters of the western Atlantic Ocean, including the Gulf of Mexico and the Caribbean Sea and is particularly abundant along the shores of Florida and Bahamas (Bigelow & Schroeder,

1953). In Brazil, it occurs especially in the northeast coast, usually being sighted in Fernando de Noronha Archipelago and in Rocas Atoll (Aguiar *et al.*, 2009). Overall the species is found in coastal and insular shallow waters, with maximum record depth reported for the species in 53 m (IUCN, 2015). Featuring demersal habits, the species is an opportunistic identified the mesopredator, feeding mainly of small benthic invertebrates (Gilliam & Sullivan, 1993; Ebert & Cowley, 2003).

The importance of the identification of essential habitats for fish communities and their diversity is recognized and discussed by several authors (Anderson *et al.*, 1989; Aguilar *et al.*, 2009). The selection of specific habitats within the same ecosystem for different purposes and life stages is a crucial element in the strategy of survival and reproductive success of any specie, in particular in the marine environment, where environmental variables such as salinity, dissolved oxygen (Heithaus & Delius, 2009; Hasler *et al.*, 2009) and temperature (Farrugia *et al.*, 2011), among others, often reach levels that can endanger the survival of several species that inhabit it. Thus, identifying and protecting of habitats essenciais from the species, in this context including the vertical distribution of individuals in the water column, is crucial in order to understand fish communities and for species conservation and management activities (Steimle & Zetlin, 2000).

Similarly to other species of rays, there is lack of information on the life history and essential habitats of the members of the genus *Dasyatis*. Studies that address aspects related to its horizontal and vertical movement are particularly rare, especially for *D. americana* (Corcoran *et al.*, 2013). Tilley *et al.* (2013) investigated the horizontal movement pattern of *D. americana* in Glovers Reef Atoll, Belize, as of acoustic telemetry techniques. The authors observed that, in general, mean stingray activity space was relatively small (<0.5 km²) with daytime activity space significantly larger than nighttime activity space. Aspects related to the vertical displacement pattern of the species, including its distribution in different extracts of depth and temperature have not yet been reported in the literature.

An important tool used to study the pattern of movement of marine fish, which has been widely used around the world, is the satellite telemetry. The transmitters via satellite PSAT type (Pop-up Satellite Archival Tags) have been developed to supply the need for long-term data collection. This kind of technology has already been used in research on several different species, including tunas (Block *et al.*, 1998; Lutcavage *et al.*, 1999; Sibert *et al.*, 2003; Teo *et al.*, 2007; Whitlock *et al.*, 2012), billfish (Graves *et al.*, 2002;

Kerstetter *et al.*, 2003; Horodysky & Graves, 2005), turtles (Swimmer *et al.*, 2002), squids (Bazzino *et al.*, 2010), and other teleost (Horodysky & Graves, 2005; Sims *et al.*, 2009), in addition to sharks (Weng & Block, 2004; Bonfil *et al.*, 2005; Stokesbury *et al.*, 2005; Hammerschlag *et al.*, 2010) and rays (Le Port *et al.*, 2008; Wearmouth & Sims, 2009).

The first PSAT data published for a stingray was conducted in the northeast coast of New Zealand. The investigation of horizontal and vertical movement pattern, including the observation of temperature and depth variables from the stingray *Dasyatis brevicaudata* showed that the analyzes of geolocation, depth and temperature indicated that species moved largest distances (≤ 25 km) (Le Port *et al.*, 2008). However for a adequate comprehension of the essential habitats utilized by the group, greater efforts need to be employed in order to understand the movement pattern of species of rays, particularly of *Dasyatis* genre.

This technological innovation, together with the use of existing tools, have allowed the identification of migratory movements associated with feeding and reproductive behavior, as well as environmental factors, in several of the studied species (Arnold & Dewar, 2001; Boustany *et al.*, 2002). More recently, this technology has been used in the study of vertical and horizontal movements of elasmobranchs species in Environmental Protection Areas (EPA) in Brazil, following the example of research that has been conducted inside the EPA of Fernando de Noronha - Rocas - São Pedro and São Paulo Archipelago (Macena, 2010; Bezerra, 2013).

Several researches that have tried to assess the benefits of MPAs for the conservation of elasmobranchs suggest that, currently, these areas offer variable levels of protection for the populations usually exploited (Bonfil, 1997; Chapman *et al.*, 2005; Carraro & Gladstone, 2006; Robbins *et al.*, 2006; Heupel *et al.*, 2010; Knip *et al.*, 2012). However, if the spatial and temporal dynamics of the life history of the species are considered during the planning and development of these areas, there is a much higher probability they will serve as an effective management tool in the conservation of elasmobranchs (Le Port *et al.*, 2012).

In northeast Brazil, the Rocas Atoll is officially categorized as a Biological Reserve, an Integral Protection Conservation Unit, which is the most restrictive under Brazilian law. Within the area of the Atoll, the exploitation of natural resources, recreational activities and visitation, except those with scientific and educational purposes, are strictly prohibited. That might be one of the reasons why Rocas Atoll has one of the highest rate of endemism among Brazilian oceanic islands, according to their geographical isolation (Serafini *et al.*, 2010).

The Rocas Atoll is still used as an area for feeding, reproduction and nursery for some species of sharks and rays, being the region of great importance for elasmobranch fauna (Castro & Rosa, 2005; Freitas *et al.*, 2006; Garla *et al.*, 2006; Wetherbee *et al.*, 2007). In this context, the present study investigated the pattern of vertical movements of the *D. americana* inside and around Rocas Atoll, to better understand its behavior, as well as the role that isolated Atoll might have in the conservation of the species.

MATERIALS AND METHODS

Study area

The Rocas Atoll (03°51'30"S, 033°49'29"W), the only atoll in the South Atlantic, was the first biological reserve to be established in Brazil, in 1978. Constituted by a reef ring extending over an area of approximately 7.5 km², it is located about 260 km east of the city of Natal, in northeast Brazil, and 145 km west of Fernando de Noronha Archipelago. The region is located over a volcanic mountain that integrates the chain of underwater mountains of Fernando de Noronha, which is composed of a string of seamounts with east-west direction (Gorini & Bryan, 1976).

The location is considered an ecological sanctuary, for harboring a large number of sea birds, migratory and resident turtles that use the site for nesting and feeding, as well as several species of fish and shellfish, some of which are endemic to this insular ecosystem. The region is embedded in the South Equatorial Current, which flows from east to west (Pereira *et al.*, 2010), and has a system of semidiurnal and mesotides, with a maximum height of 3.8 m (Gherardi & Bosence, 1999). Several natural pools are formed in the reef flat, with communication or not with the adjacent deep ocean.

Programming of electronic transmitters (MiniPATs)

The satellite transmitters PSAT type (Pop-up Satellite Archival Tags) MiniPAT model was programmed to collect data at 60-second intervals for all monitoring period, producing histograms every 6 h from depth and temperature variations. Electronic transmitters were preprogrammed to collect data for 60 days. Due to the *D. americana* ecology, the transmitters have been programmed to be automatically released from the specimens after 96 h of constant depth and temperatures. Data transmitted to the satellite system ARGOS were downloaded and decoded by WC-DAP software (Data Analysis Programs).

For pattern analysis of vertical movement of the *D. americana* were considered two variables: temperature and depth frequented by the species during the moni-

toring period. These variables were also analyzed for factor period (day and night). In this sense we consider the time of day between 05:00-17: 59 h and time of night among 18:00 to 4:59. In order to investigate possible differences in the preferences of daily patterns of vertical movement, the percentage of time spent for the rays in different extracts of depth and temperature for the day and night were tested separately, for the animals, using the non-parametric Kolmogorov-Smirnov test. Statistical analyzes were performed in BioEstat 5 software.

Transmitter attachment

Two rays of the species *D. americana* were tagged with electronic transmitters (MiniPATs) during a scientific expedition to the Rocas Atoll, which occurred in March 2013, during a period of 25 days. To avoid the handling of animals, the tagging procedure was performed through free diving (Fig. 1). The MiniPAT was firstly fixed to a stainless steel tip and was then applied with the help of a spear, by a diver, in the posterior region of the pectoral fin (adapted from Speed *et al.*, 2013).

RESULTS

Transmitter performance

The distribution of depth and temperature was obtained for two female stingrays, both presumably adults, with an estimated size of 108 and 95 cm of disc width. Both rays were marked in Zulu pool, inside the Rocas Atoll. The Ray I was tagged on 10/03/2013 (0352.375N 3347.888W) and Ray II on 10/03/2013 (0352.366N 3347.898W). Both specimens showed good vital conditions during the procedure. However, the MiniPat Ray II was released prematurely, remaining on the animal for only 8 days, despite it had been programmed to collect data for 60 consecutive days. Ray I, however, remained with the transmitter during all pre-programmed period (Table 1). This specimen was observed days after it was tagged with the electronic satellite transmitter, in its natural habitat. The total percentage of data decoded and successfully transmitted through the ARGOS satellite system was 67% for Ray I and 64% for Ray II. Due to technical problems, it was not possible to obtain the geographical location of the tagged animals during the monitoring period.

Habitat use and temperature preferences

The results indicate a clear vertical pattern of habitat use for the *D. americana*, with both specimens showing a remarkable preference for the shallow and warm waters of Rocas Atoll. Ray I and Ray II remained 91.4% and 86.3% of the monitored period, respectively,

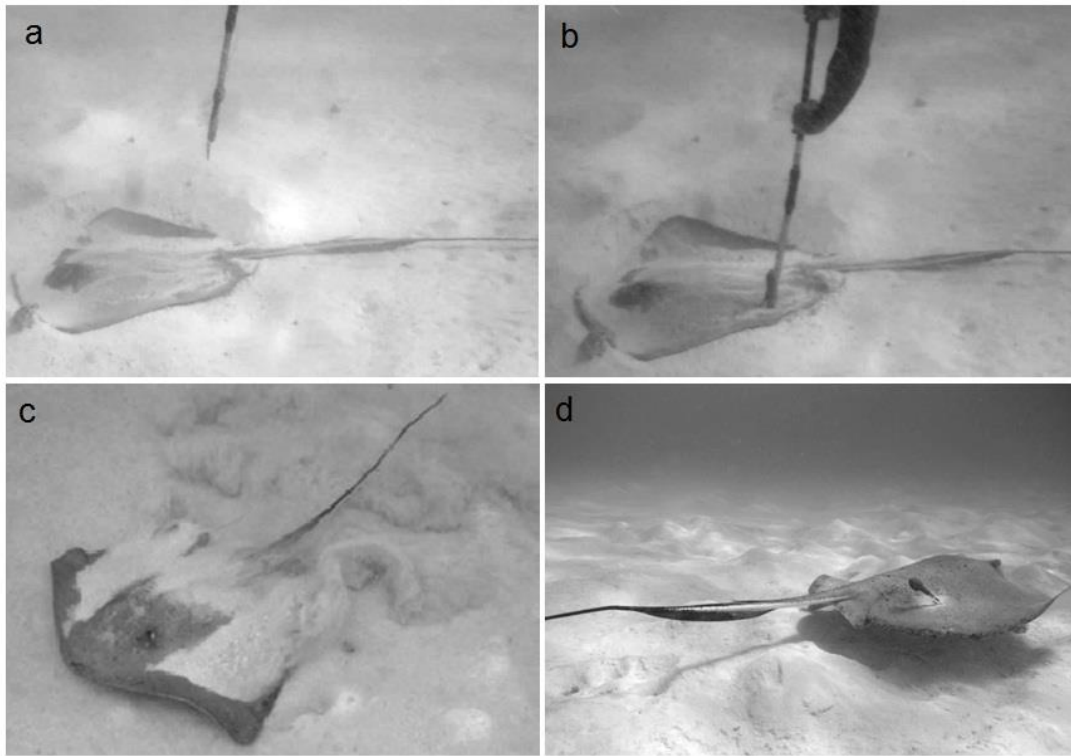


Figure 1. Technique used for tagging the southern stingray *D. americana*, in natural environment with satellite transmitter (MiniPat) in the Biological Reserve of Rocas Atoll. a) Approximation of a diver with a spear carrying the MiniPat, behind and lateral to the animal, b) transmitter application in the lateral region of the pectoral fin, c) fast movement after the tagging procedure, d) individual swimming naturally with the electronic transmitter fixed properly and with the least amount of handling.

Table 1. Summary of information related to monitored rays with electronic satellite transmitters (MiniPat) inside the Rocas Atoll.

Ray	DW (cm)	Sex	Data tagging	Tagging location		Monitoring period (days)	Retention period (days)
				Latitude	Longitude		
Ray I	108,0	F	10/03/2013	0352.375N	3347.888W	60	60
Ray II	95,0	F	11/03/2013	0352.366N	3347.898W	60	8

in warm waters with temperatures above 28°C, at depths of up to 5 m (Fig. 2).

Throughout the monitoring period, Ray I remained 93% of daytime and 94% of nighttime, at depths from surface to 5 m. The depth range most frequented by the specimen was between 1 and 2 m, staying in this layer 52.1% and 54.8% of the day and night, respectively (Fig. 3). The specimen spent only 7% of the day and 6% of the night in depths greater than 10 m. The temperature range frequented by Ray I was very similar between daytime and nighttime, spending 89.2% of the day and 91.4% of the night at temperatures between 27 and 31°C. Ray I stayed in water with lower temperatures (21 to 27°C) in only 8.9% of the day and 8.2% of the night. The similarity of time spent by Ray I

between daytime and nighttime, in different layers of depth and temperature, was confirmed statistically (Depth: $D = 0.286$, $P = 0.938$; Temperature: $D = 0.143$, $P = 0.985$).

In a manner similar to Ray I, Ray II spent most of its time at depths from surface to 5 m, staying within this layer 70.7% and 72.4% of the time, during day and night, respectively. Differently from Ray I, however, Ray II preferred different depth ranges between daytime and nighttime, remaining 27.9% of the day in water depths between 10 and 20 m; and 34.3% of the night very close to the surface (0-1 m). Although the general temperature distribution also differed, it remained close to the one shown by Ray I, spending 88.7% of the day and 91.4% of the night at temperatures

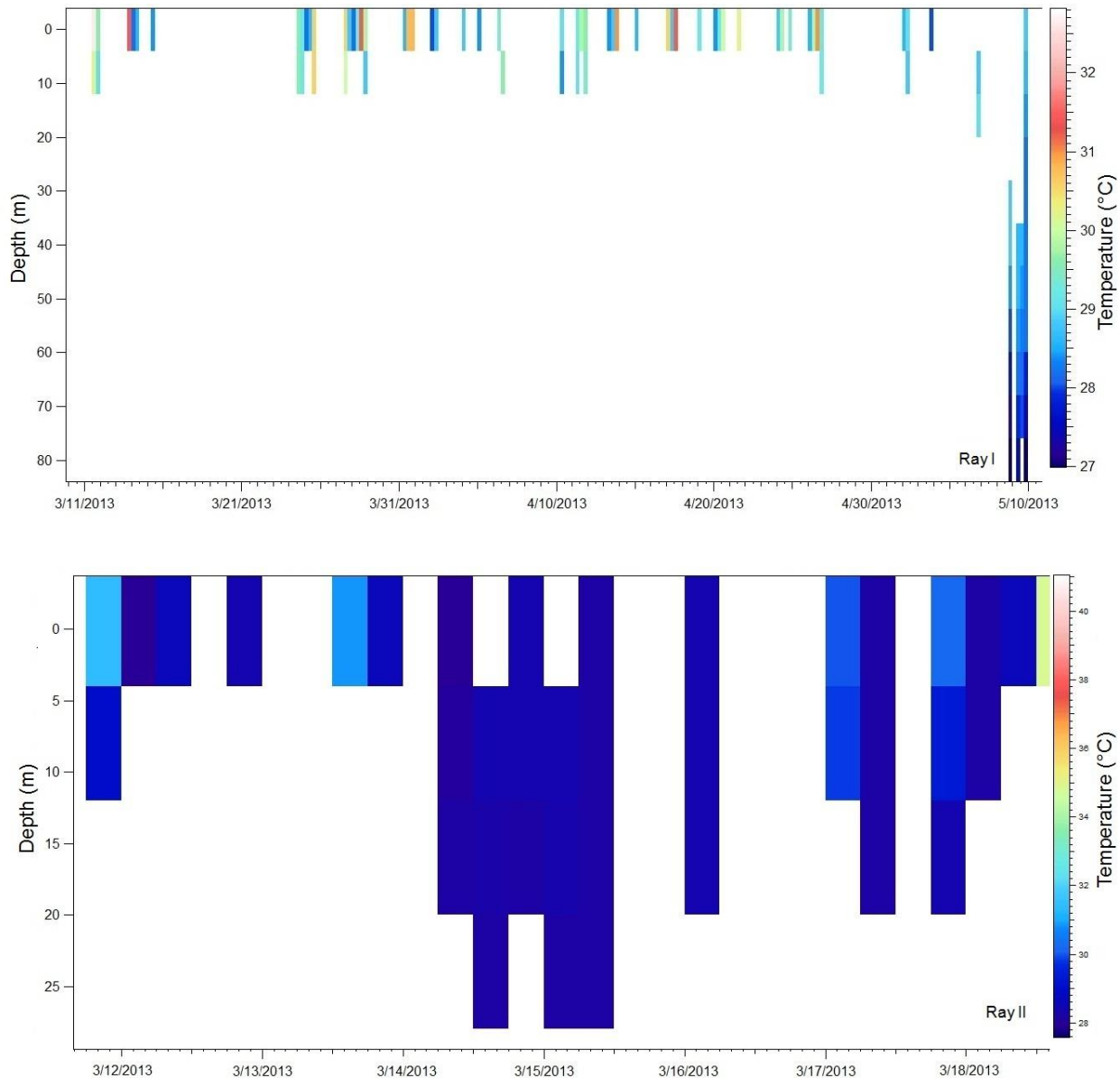


Figure 2. Depth and water temperature profiles (from summary data every 6 h), experienced by the Ray I and Ray II, monitored with electronic transmitters (MiniPat) in the Biological Reserve of Rocas Atoll. The colors represent the temperature gradient of seawater.

between 27 and 31°C, and 78.7% and 85.6% of the day and night, respectively, in temperatures between 27 and 29°C. Lower temperatures (24 to 27°C) were recorded in only 6.6% of the day and 6.0% of the night. The differences between the proportions of time used by Ray II in different depths and temperatures throughout the day and night, were not statistically significant (Depth: $D = 0.375$, $P = 0.627$; Temperature: $D = 0.25$, $P = 0.964$).

Despite the clear preference of both animals for the warm waters close to the surface, both rays performed

vertical movements venturing in deeper waters, with lower temperatures. Ray I performed a greater number of dives than Ray II, particularly in the last days of the monitoring period. The deepest dive conducted by Ray I happened on 09/05/2013, when it reached 77.5 m in depth, at a temperature of 25.3°C. Ray II, which prematurely lost the transmitter, naturally exhibited a smaller number of dives, reaching on 15/03/2013, 24.0 m depth in water with 28.1°C of temperature.

The data were obtained every 5 min through the function time series of the transmitters; and indicate

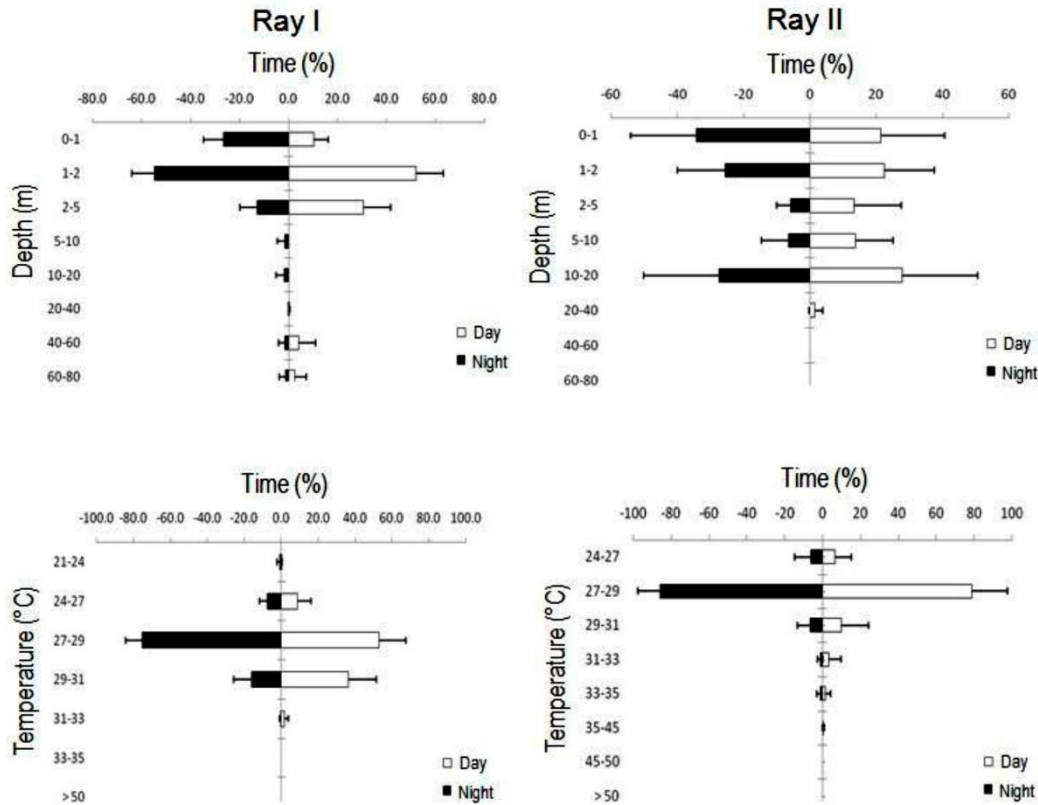


Figure 3. Frequency relative (%) in relation to the depths and temperatures experienced during the vertical movement, the Ray I (right) and the Ray II (left) throughout the day and night, monitored with electronic transmitters (MiniPat) in the Biological Reserve of Rocas Atoll.

Table 2. Summary statistics (average, minimum, maximum and standard deviation) of values reading temperature and depth experienced by rays monitored with electronic satellite transmitters (MiniPat) in the Biological Reserve of Rocas Atoll.

Period	Ray (n = 2)	Average depth (m)	Minimum depth (m)	Maximum depth (m)	Standard deviation
Day	Ray I	2.40	0	59.00	6.17
Night	Ray I	3.82	0	77.50	11.14
Day	Ray II	4.91	0	24.00	5.61
Night	Ray II	4.60	0	17.00	6.04

depth and temperature cumulative frequency (Table 2) similar for both individuals. The Ray I experienced temperatures that varied between 23.8 and 33.3°C (mean \pm SD) (28.1 ± 1.0) and depths from 0 to 77.5 m (3.0 ± 8.5), while Ray II stayed at temperatures ranging between 25.7 and 34.4°C (28.3 ± 1.0) and depths from 0 to 24 m (4.8 ± 5.8).

The vertical daily movements performed by Ray I clearly show that the specimen preferred shallow waters with depths above 5 m, between 01:00 and 11:00 pm, with higher temperature also occurring in the same period, while between 12:00 am and 12:00 pm that animal dived into deeper waters, below 10 m. The

vertical habitat utilization for Ray II, however, did not show any clear pattern, with regard to a 24-h period (Figs. 4a-4b, 5a-5b).

DISCUSSION

Transmitter performance

In the last decades, several methods to attach electronic transmitters to stingrays, to monitor their movements, have been used without negative effects to animal welfare (Blaylock, 1990; Collins *et al.*, 2007; Le Port *et al.*, 2008). However, in most cases, these attachment techniques require the capture and restraint of animals

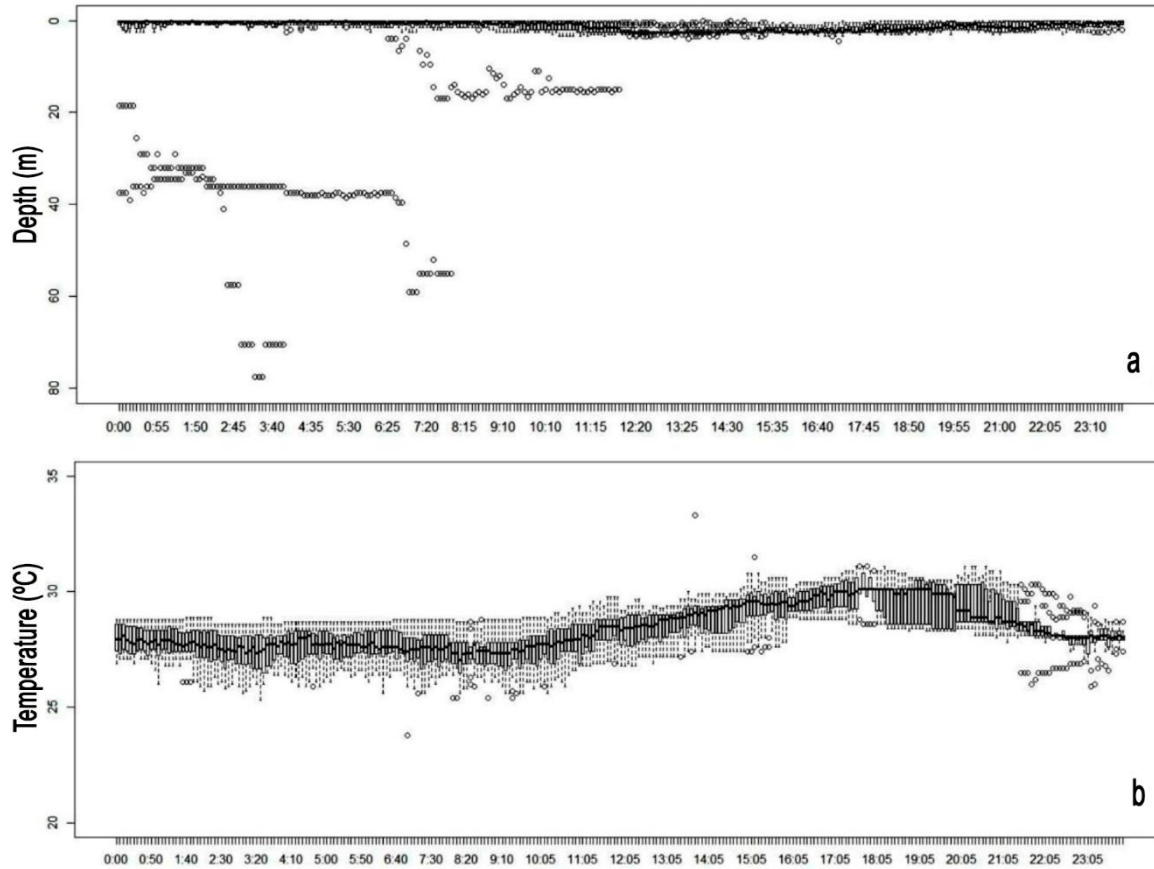


Figure 4. Box plots showing the distribution of maximum and minimum depth a) and temperature b) relative to the 24-hour cycle for Ray I monitored with electronic transmitters (MiniPat) in the Biological Reserve of the Rocas Atoll.

on board, with risks for the researcher in handling potentially dangerous species as well as a higher likelihood of trauma to the handled animals (Speed *et al.*, 2013).

The use of tagging techniques that do not require the capture of the animal, allowing it to stay all the time in its natural environment, reduces significantly the risk of harming both the handler and the tagged specimens (Sundstrom *et al.*, 2001). The successful use of diving weapons and spear guns for external tagging of large elasmobranchs, for instance, has been reported in various studies with nurse sharks (*Ginglymostoma cirratum*), manta rays (*Manta birostris*) and whale sharks (*Rhincodon typus*) (Pratt & Carrier, 2001; Wilson *et al.*, 2006; Dewar *et al.*, 2008).

Despite the technical difficulties associated with tagging and tracking elasmobranchs in tropical reef environments are well recognized (Nelson, 1977; Bres, 1993; Simpfendorfer & Heupel, 2004), the choice for a spear gun to carry out the external tagging of *D. americana*, with Mini-Pat transmitters, by free-diving, proved to be very safe and efficient in this research. The

direct observation of Ray I, days after it was tagged with the electronic satellite transmitter, in its habitat, allowed to suggest that the species was behaving naturally, undertaking its usual activities inside the Rocas Atoll, apparently not bothered by the transmitter it was carrying.

Habitat use and temperature preferences

Both tagged rays exhibited comparable vertical movement profiles, staying throughout the monitoring time in rather shallow depth (less than 5 m). Both specimens remained less than 10% of their time in water depths greater than 10 m. A similar trend was also observed in other researches on other *Dasyatidae* species, *Pastinachus atrus* and *Urogymnus asperrimus*, in Ningaloo Reef, Western Australia (Speed *et al.*, 2013). According to those authors, the rays tagged with acoustic transmitters showed a highly sedentary behavior, remaining in shallow waters (1-10 m deep) most of the time, with some specimens staying in the same place for periods of about 8 h with normal behaviors of feeding and resting. The sedentary behavior in shallow

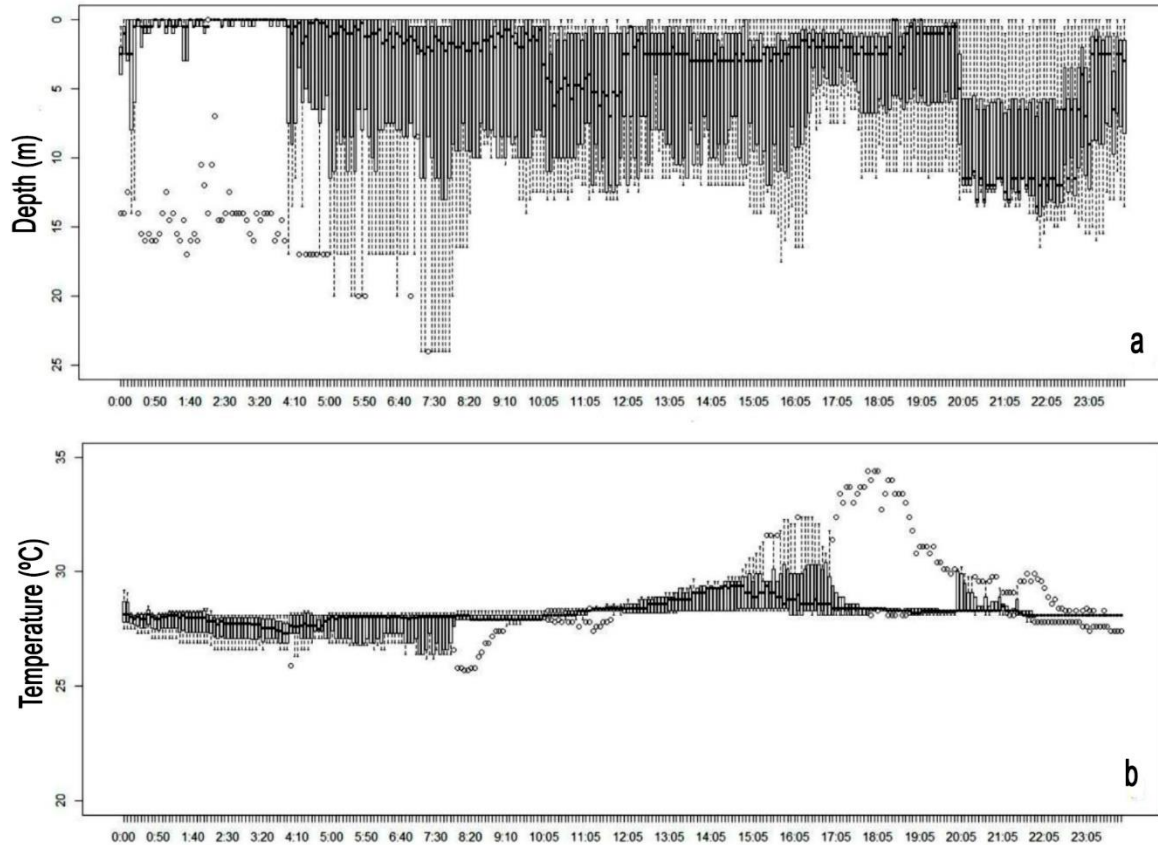


Figure 6. Box plots showing the distribution of maximum and minimum depth a) and temperature b) relative to the 24-hour cycle for Ray II monitored with electronic transmitters (MiniPat) in the Biological Reserve of the Rocas Atoll.

and warm water observed for some Dasytidae family species, irrespective of day or night, may be associated with feeding process, since the species feed on prey that are also commonly found in quite shallow depths.

The behavior of the tagged specimens to remain most of the time in shallow waters of Rocas Atoll, observed in this study for both monitored specimens, is similar to that described in studies which evaluated the movement pattern of *D. americana* in the Cayman Islands and the Glovers Reef Atoll, Belize (Corcoran *et al.*, 2013; Tilley *et al.*, 2013). These authors also cited that beyond the high site fidelity, in general, the average activity space (<0.5 km²) used by the species is considerably small.

The preference for shallow waters found in the present study for *D. americana* seems to be closely related to the biological characteristics of the species, as well as to the study area. According to Bigelow & Schroeder (1953), the species that make up the *Dasyatis* genus are characterized by having strictly benthic feeding habits, eating especially epibenthic species,

mostly, crustaceans and small teleosts, among other invertebrates (Gilliam & Sullivan, 1993; Carqueja *et al.*, 1995; Silva *et al.*, 2001; Ebert & Cowley, 2003). Therefore, the fact that *D. americana* occupy primarily coastal and shallow waters and have a feeding behavior strongly associated to the sandy bottom (Bigelow & Schroeder, 1953; Figueiredo, 1977; Menni & Stehmann, 2000) certainly accounts to the high percentage of time that the two tagged specimens remained in shallow water inside the reserve.

The vertical daily pattern of movement performed by Ray I, which remained with the transmitter during all the programming period, shows that the specimen preferred shallower water in most of the day, but would rather be in deeper water most of the night. Other researches on the movements of *D. americana*, recorded a higher degree of activity and space use at night, than during the day (Corcoran *et al.*, 2013). This pattern of behavior confirms that *D. americana* is a species with naturally nocturnal behavior, foraging especially during this period (Gilliam & Sullivan, 1993).

This kind of behavior has also been reported for several other coastal elasmobranch species that occur in shallow water (McKibben & Nelson, 1986; Cartamil *et al.*, 2003; Vaudo & Lowe, 2006; Collins *et al.*, 2007; Chapman *et al.*, 2009). For the Ray II the absence of a clear distinction between the activities carried out during day and night, in this study, may be related to possible changes in animal behavior after the tagging procedure. Some behavioural changes observed for fish species may be related to the physiological and biochemical effects of exhaustive exercise associated with capture and handling from the animals (Wells *et al.*, 1986; Hoolihan *et al.*, 2011).

The most preferred temperature range (27 to 29°C) experienced by the *D. americana* in Rocas Atoll was coherent with the temperature variation observed in the region, which has an average of 27°C, although it may go beyond 40°C in closed pools (Gherardi & Bosence, 1999; Pereira *et al.*, 2010). The permanence of the species in higher temperatures during the monitoring period is probably associated to their high frequency of occurrence in shallow locations of the Rocas Atoll (at depths of up to 5 m), observed in the present research.

Although infrequent, the record of dives deeper than 70 m, in this study, seems to indicate that the species occasionally makes incursions outside of the reef ring, staying for unknown periods in the region around the Rocas Atoll. Despite it was not possible to obtain the geographical locations of the monitored specimens with satellite transmitters, the profile of the vertical habitat use seems to indicate that *D. americana* does not make large migrations beyond the Rocas Atoll. It also indicate the low depth areas inside the Atoll as an essential habitat for the species.

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