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ORIGINAL ARTICLE

Influence of lunar phases and oceanographic parameters on green turtle nesting in Rocas Atoll

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Abstract
Animals can synchronize their reproductive behavior patterns with biological rhythms and environmental conditions. Understanding these patterns is particularly critical for conserving endangered species like sea turtles. This study analyzed the relationship between the nesting behavior of the green turtle (*Chelonia mydas*) and lunar luminosity, lunar phases, and sea surface temperature (SST) in the Atol das Rocas Biological Reserve, Brazil. Field data were obtained through nightly monitoring during the breeding seasons from 2018 to 2020. Although lunar luminosity and lunar phases did not exhibit a statistically significant effect, the highest number of nesting occurrences was observed during the full moon and new moon, which may be related to the spring tide. The SST was the only variable showing a statistically significant relationship with complete oviposition, incomplete oviposition, and false crawl occurrences, indicating the preference of turtles to come ashore at higher temperatures (around 29.5 °C). Furthermore, while lunar luminosity and lunar phases may have some influence on the nesting pattern of the green turtle, they do not limit it, as turtles come ashore under different observed environmental conditions.

Keywords Marine turtles · *Chelonia mydas* · Nesting · Lunar cycle · Sea surface temperature · Ecology

Introduction

Biological rhythms (*e.g.*, lunar cycle and tides) and sea temperature are essential to understanding animal behavior. Biological rhythms can be classified into ultradian, circadian and infradian, referring to the annual seasonality, the tidal cycle and the lunar phases (Marques and Menna-Barreto 2007). Sea turtles are susceptible to environmental temperature due to ectothermy (Weishampel et al. 2010), evidencing a close relationship with the oceanographic conditions of their occurrence area. The synodic lunar cycle has an average period of 29.53 days, presenting four main lunar phases (or angles): New Moon (0° and 360°), First Quarter (90°), Full Moon (180°) and Third Quarter (270°) (Nakamura et al. 2019). This movement influences the tidal cycle and the levels of environmental light, as well as the behavior and physiology of some plants and animals (Kronfeld-Schor et al. 2013; Marques and Menna-Barreto 2007).

Some organisms present patterns of reproductive behavior's synchronization with the effects of lunar phases, such as some fish families (Serranidae, Labridae and Siganidae) (Takemura et al. 2010; Ikegami et al. 2014), the one-night mass spawning of over 100 coral species on the Great Barrier Reef (Australia), the *Sesarma haematocheir* (De Haan, 1833) crab in Japan, and even more complex animals such as the Eurasian badger mammal (*Meles meles* Linnaeus, 1758) (Kronfeld-Schor et al. 2013). In addition, the lunar cycle influences the women's menstrual cycle, despite the anthropocentric hegemonic view that humans would not be part of nature's processes (Cutler 1980; Law 1986; Zimeck 2006).

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Several authors have observed the correlation of the lunar cycle with the reproductive behavior of sea turtles; *Caretta caretta* (Linnaeus, 1758) (Burney et al. 1990); *Dermochelys coriacea* (Vandelli, 1761) (Law et al. 2010); *Lepidochelys olivacea* (Eschscholtz, 1829) (Hughes and Richard 1974) and *Eretmochelys imbricata* (Linnaeus, 1766) (Nakamura et al. 2019). However, few studies use this approach in *Chelonia mydas* (Linnaeus, 1758), especially in Brazil, where no study was performed despite its broad occurrence along the Brazilian coast.

The green turtle (*Chelonia mydas*) is currently threatened, being classified as Endangered (EN) worldwide (IUCN 2022) and Near Threatened (NT) in Brazil (MMA 2022). The green turtle nesting in Brazilian territory occurs mainly on oceanic islands such as the Rocas Atoll Biological Reserve, the second largest reproductive site of this species, behind only Trindade Island (Bellini et al. 2013). Studies in areas with minimal human activity, such as the Rocas Atoll, can be excellent alternatives to analyzing the mechanisms of the synchronization of turtles with environmental variables (Marques and Menna-Barreto 2007; Pike 2008; Bèzy et al. 2020).

Understanding the influence of environmental variables on the nesting behavior of sea turtles provides important information about their response to global environmental changes. In addition, these findings may assist in the management of more effective local actions for their conservation (Pike 2008). In this sense, this study aimed to study the influence of lunar luminosity, lunar phases and sea surface temperature (SST) on the nesting behavior of the Rocas Atoll population of the green turtle, *C. mydas*.

Material and methods

Study area

The Rocas Atoll Biological Reserve is an oceanic island located in the Northeast of Brazil (Fig. 1), comprising a total area of 360 km^2 delimited by the isobath of 1000 m. The Rocas Atoll is an elliptical ring reef with approximately 6.5 km^2 and two main islands: Farol Island and Cemitério Island (Kikuchi 2002; MMA 2007; Pereira et al. 2010). The site is of great importance for biodiversity conservation, standing out as a key location for the protection of endemic and endangered species and being a feeding, resting and breeding area for *C. mydas* (Kikuchi 2002; MMA 2007). The study area is semidiurnal mesotidal, with a range of 2.8 m during spring tides and 1.7 m during neap tides (Kikuchi 2002; Costa et al. 2017).

The study area is influenced by the South Equatorial Current and has an average sea surface temperature (SST) of 27 °C in its external region (Ferreira et al. 2012; Kikuchi 2002). Two reef passages connect the atoll lagoon to the ocean. During low tide, these are the only communications between its exterior and interior. Therefore, tidal variation, together with the geomorphology of the region, causes large variations in SST and salinity in the lagoons and coral pools, which can reach 39° in the pools, due to the restricted water circulation (Kikuchi 2002; MMA 2007; Costa et al. 2017).

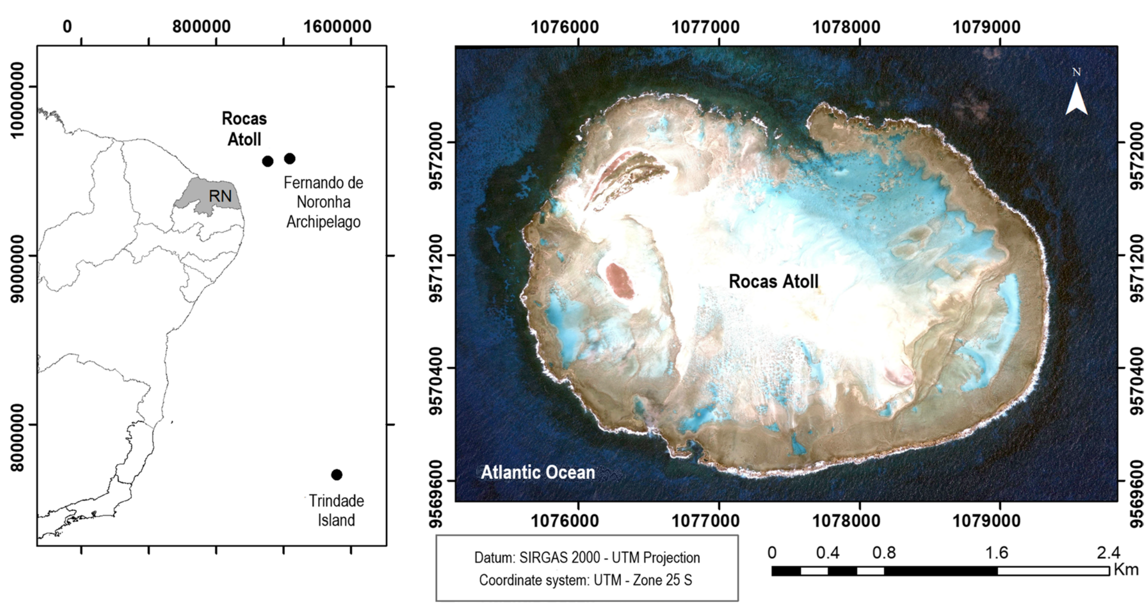

Fig. 1 Main nesting sites of *C. mydas* Atlantic Equatorial, Brazil, with emphasis in the Rocas Atoll

Sampling

The data used in this study were obtained while monitoring the breeding seasons from 2018 to 2020 on Farol Island, which is 674 m length and 327 m width, with 1,800 m of coastal extension (Pereira et al. 2010). Beach monitoring was daily (with the exception of days with bad weather, such as rainy days), at night and the period between three hours before and three hours after the high tides. Night patrolling depends on tides, because in the Rocas Atoll turtles, turtles preferentially nest at night during the flood and high tide, avoiding the nesting site due to the exposure of the reef zone during the low and ebb tide, making it difficult for animals to access the sandy beach (Bellini et al. 2013; Grossman et al. 2009; Silva and Godoy 2016). Three people made up the monitoring team, selected by the ICMBio (Instituto Chico Mendes de Conservação da Biodiversidade). Data collected embraced date and time of occurrence, location, animal identification (e.g., presence of metal tag or microchip), biometric data (curvilinear measurement of carapace length and width) and occurrence type. The data used in this study refer to sea turtles that nest before sunrise.

Occurrences were classified as complete oviposition (CO) – turtle performs all stages of the nesting process; incomplete oviposition (IO) – turtle prepares the nest site and/or digs an egg chamber, but does not deposit eggs within the chamber; and false crawl (FC) – when the turtle emerges and returns to the sea without performs any other step in the nesting process (Nakamura et al. 2019; Nishizawa et al. 2013). This study was carried out with the authorization of the Biodiversity Authorization and Information System (Sisbio) license nº 59809.

Data analysis

To understand the relationship between turtle occurrence and the lunar angle and Sea Surface Temperature (SST), we considered 756 occurrences recorded in the breeding seasons from 2018 to 2020. For the relationship between breeding occurrences and lunar luminosity, we considered 578 records obtained in 2019 and 2020, since we lack data on the time of occurrence for 2018.

Lunar luminosity – the illuminated face of the Moon facing Earth – was determined based on the Double Precision Library of Celestial Mechanics, Astrometry and Astrodynamics (Libnova: <http://libnova.sourceforge.net/index.html>) for the coordinates of the study area. In this sense, the occurrence data of the 2019 and 2020 breeding seasons were crossed with the illuminated percentage of the visible face of the moon (0—100%) at the time of each occurrence.

The lunar calendar of the Astronomy Department of IAG/USP was used together with the total occurrences of the seasons from 2018 to 2020 to investigate the lunar phases and tidal types. The lunar angle—angular position between Sun-Moon-Earth—was used as a continuous variable from 0° to 360°. To facilitate the discussion, four main angles were considered as reference: 0° = new moon; 90° = first quarter; 180° = full moon; 270° = third quarter; 360° = new moon (Nakamura et al. 2019).

The oceanographic and meteorological data of Rocas Atoll are sparse due to the absence of buoys and monitoring stations. Therefore, the TSM data were obtained on the Giovanni platform (NASA 2020) through the Aqua-MODIS satellite with nocturnal data with 4 km spatial resolution, 8-day temporal resolution, and 11-microns spectral resolution.

Statistical analysis

The Generalized Linear Mixed Model (GLMM) was used to analyze the relationship between occurrence type and SST (Bolker et al. 2009). Initially, a Poisson distribution (Family) with a logarithmic link function was considered to generate the model, but the data presented overdispersion. Therefore, we used the negative binomial distribution to develop the definitive model.

We used a simple linear regression model to analyze the relationship between lunar luminosity and reproductive occurrences. The analyzes were performed with the packages *stats* (R Core Team 2020) and *lme4* (Bates et al. 2015) in the R Environment (R Core Team 2020). The significance level was $p < 0.05$ (Banerjee et al. 2009).

We examined the lunar angle as a continuous variable to assess uniformity using the Kuiper test (Kuiper 1960; Jammalamadaka and SenGupta 2001). This analysis was conducted utilizing the circular statistic package (Agostinelli and Lund 2023) within the R Environment.

Results

The reproductive seasons of 2018 to 2020 resulted in 756 occurrence records of *Chelonia mydas* in the Farol Island of the Rocas Atoll, of which 568 culminated in nests (complete oviposition—CO), 97 unsuccessful nest attempts (incomplete oviposition—IO), and 91 were tracks without oviposition (false crawl—FC). The nesting period mainly occurred from January to April, showing the nesting peak in February and March (Fig. 2), which followed the study by Bellini et al. (2013). However, the nests' number was heterogeneous over the three years. In 2020, there were approximately twice as many occurrences (53.34% of total occurrences) as compared to the same period in 2018 (28.87%) and 2019 (17.78%) (Fig. 2).

We found no significant relationship between occurrence type and lunar luminosity (CO, -0.14 ± 0.11, $p = 0.23$; IO, -0.04 ± 0.02, $p = 0.14$; FC, -0.04 ± 0.03,

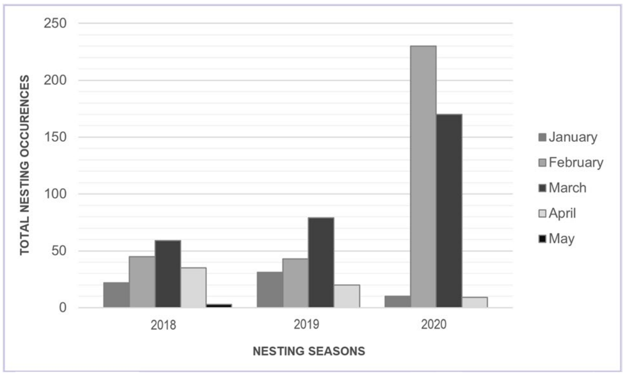

Fig. 2 Monthly distribution of *Chelonia mydas* nesting occurrences in the 2018–2020 breeding seasons in the Rocas Atoll, Equatorial West Atlantic

$p = 0.15$) (estimate ± standard error, p-value) (Fig. 3). The Kuiper test performed no statistically significant relationship between lunar angle and total occurrences ($t = 1.61$, $p > 0.1$, $n = 166$ observations) (test statistic, p-value, n observations) (Fig. 4). The sea surface temperature presented a significant influence on the three occurrence types (CO, $p < 0.01$; IO, $p = 0.05$; FC, $p < 0.01$; Table 1). The results showed the species' preference for reaching the beach at higher temperatures (≥ 29.5 °C) (Fig. 5). This pattern was observed for CO and IO, while FC presented a more heterogeneous distribution.

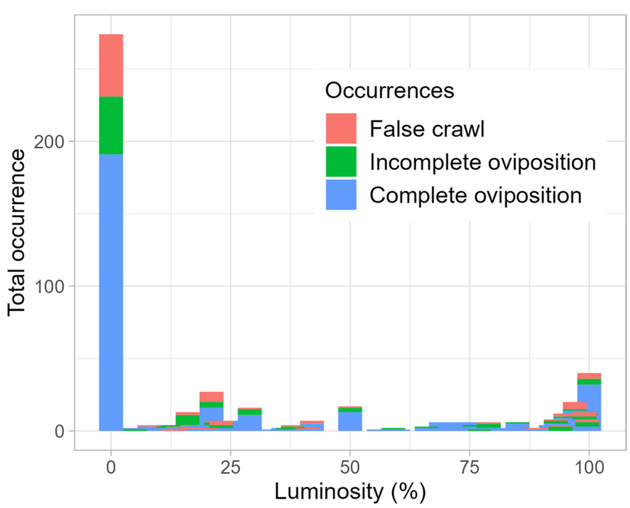

Fig. 3 Relationship between lunar luminosity and oviposition of turtles in the 2019–2020 breeding season in the Rocas Atoll. Distribution of the frequency of occurrences with recorded time ($n = 578$) related to the respective luminous percentage of the moon in the sky (0—100%) at the time of occurrence (false crawl; incomplete oviposition; complete oviposition)

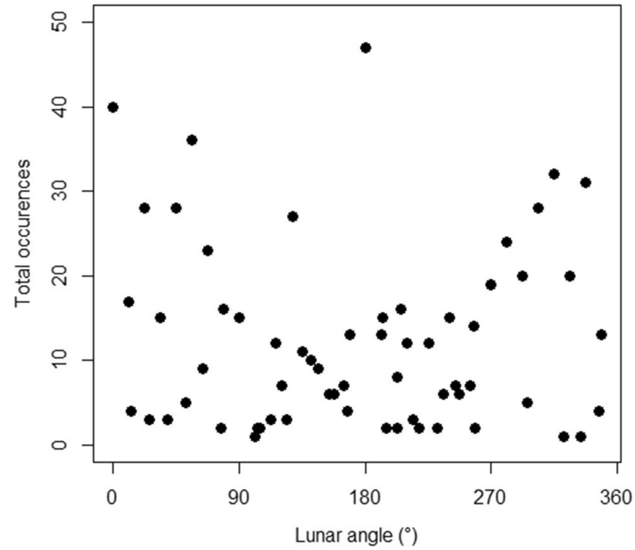

Fig. 4 Effect of lunar phases on *C. mydas* nesting behavior during the 2018–2020 breeding seasons. Distribution of total occurrences ($n = 756$) related to lunar angle, where 0° = new moon; 90° = first quarter; 180° = full moon; 270° = third quarter; 360° = new moon

Discussion

The number of studied nests ($n = 568$) is not representative of the total observed in the Rocas Atoll since it did not consider the occurrences from Cemetery Island, those of sporadic days in which there was no monitoring, or those occurring in other months of the reproductive period, such as December.

The heterogeneous distribution of the number of nests between the analyzed years (Fig. 2) may be related to the possibility that the green turtles nesting in the Rocas Atoll comes from different feeding sites, which influences their return from the feeding area to the nesting area, interfering with their remigration interval (Agostinho et al. 2021).

Turtles do not usually make nests in consecutive years. Specifically, the average turtle's remigration period in the Rocas Atoll is 3.5 years (Bellini et al. 2013). Therefore, it is possible that different individuals were sampled in each year of this study, which could justify the observed distribution. In addition, the high level of interannual variation found in the number of nesting females in populations of *C. mydas* could be related to environmental conditions (Broderick et al. 2003).

Lunar luminosity

The illuminated percentage of the visible face of the moon allows analyzing the influence of lunar brightness during the nesting moment when the configuration of the study area is ideal, i.e., it does not present artificial lighting. This variable

Table 1 Relationship between Sea Surface Temperature (SST) with occurrence types of *Chelonia mydas* from the Rocas Atoll

Effects	Complete oviposition		Incomplete oviposition		False crawl	
Fixed effect	Estimate ± SE	*p*	Estimate ± SE	*p*	Estimate ± SE	*p*
SST	0.22 ± 0.04	< 0.01	0.21 ± 0.11	0.05	-0.44 ± 0.08	< 0.01
Random effect	Variance ± SD		Variance ± SD		Variance ± SD	
Total occurrences	0.32 ± 0.57		1.47 ± 1.21		0.55 ± 0.74	

SE standard error, *SD* standard deviation

was not intended to infer about the lunar phase, given that 50% illuminated represents both the first quarter and third quarter. Also, 0% does not just represent the new moon but also occurrences where the moon had not yet risen in the sky. It is important to note that the Moon can be seen for 12 h in a 24-h period, rising every day 48 min later (Garrison 2014).

The lack of significant influence of the lunar luminosity on occurrence types could be related to the sampling size of three reproductive seasons, which could be insufficient to generalize patterns. In addition, other biotic and abiotic factors could influence the analysis. For example, cloudy nights could affect lunar luminosity despite the lunar cycle. Another important point is that during a lunar cycle, half of the nights will have no moon in the sky, which makes sense of the result found of the peak nesting around zero angle.

Thus, even knowing studies that show that high lunar luminosity can discourage green turtle nesting, as in João Vieira Island (Guinea-Bissau) (Ferreira 2012), and that in the Rocas Atoll, the turtles' eggs and hatchlings are preyed on by the crab *Johngarthia lagostoma* (H. Milne-Edwards, 1837) (Silva and Godoy 2016). Therefore, avoiding illuminated nights for nesting could be a strategy to decrease predation risks, since they become less exposed on darker nights. We could not relate sea turtle nesting patterns to lunar luminosity at the Rocas Atoll (Fig. 3).

Lunar phases

The lunar influence on nesting patterns may be associated with lunar luminosity and tides. We found no statistical relationship between the distribution of nesting occurrences and lunar phases; the recorded lunar angles are uniformly distributed. However, upon observing the sum of occurrences recorded at each lunar angle (Fig. 4), the highest number of nesting occurrences was at angles 180° (full moon, $n = 47$) and 0° (new moon, $n = 40$), as represented in the scatter plot. Such a pattern suggests a potential association with the spring tide, characterized by the highest tidal amplitudes, providing for a longer time for the turtles to emerge and nest on the beach. This relationship could be further explored in future research.

The Poilão Island (Guinea-Bissau) is similar to the Rocas Atoll, in which the reef area is also exposed at low tide. Thus, turtles nest at high tide, with a prevalence of nesting in areas above the highest spring tide (Patricio et al. 2018). Therefore, considering that green turtles tend to select nesting sites with a low risk of flooding (Patrício et al. 2018), the *C. mydas'* departure of the water to nest during the spring high tide requires a shorter distance in the sand to lay eggs away from the maximum limit of the spring tide, which may be a strategic behavior to ensure the success of the nest and less effort from the female.

Similar behavior has been observed in leatherback turtles (*Dermochelys coriacea*) in French Guiana, which adjust their return to nest close to new and full moons, with nesting peaks at spring tide (Girondot and Fretey 1996). In Florida, United States, the loggerhead turtle (*Caretta caretta*) is more likely to nest during the new and full moons (Burney et al. 1990). In contrast, the hawksbill turtle (*Eretmochelys imbricata*) in Rio Grande do Norte, Brazil, showed a higher nesting frequency during the waxing and waning moons (Nakamura et al. 2019).

The behavior may be regulated by some biological clock mechanism that induces nesting in the same tide, since these alternating phases correspond to the same type of tide every 15 days (spring or neap tide). Therefore, testing this hypothesis in the future with females marked by a metal tag or microchip in the study area would be interesting. Additionally, analyzing more reproductive seasons could help investigate whether there is a tendency to nest at specific lunar angles, as shown in other studies.

Sea surface temperature

In the Atlantic Ocean, three buoys located between latitudes 0°S—8°S, where the Rocas Atoll is located, presented the annual peak of TSM in the months of March, April and May (Ferreira et al. 2012), when the nesting season also occurs at the site. In this study, *Chelonia mydas* preferred to ascend to the beach for nesting; at sea surface temperatures ≥ 29.5 °C (Fig. 5a, b). A more heterogeneous distribution was observed in FC (Fig. 5c). Still, it was not considered relevant for the discussion since the water temperature can influence the turtle's exit from the water for the oviposition process, but not because of its nesting behavior (occurrence type). It is noteworthy that nesting during these higher temperatures

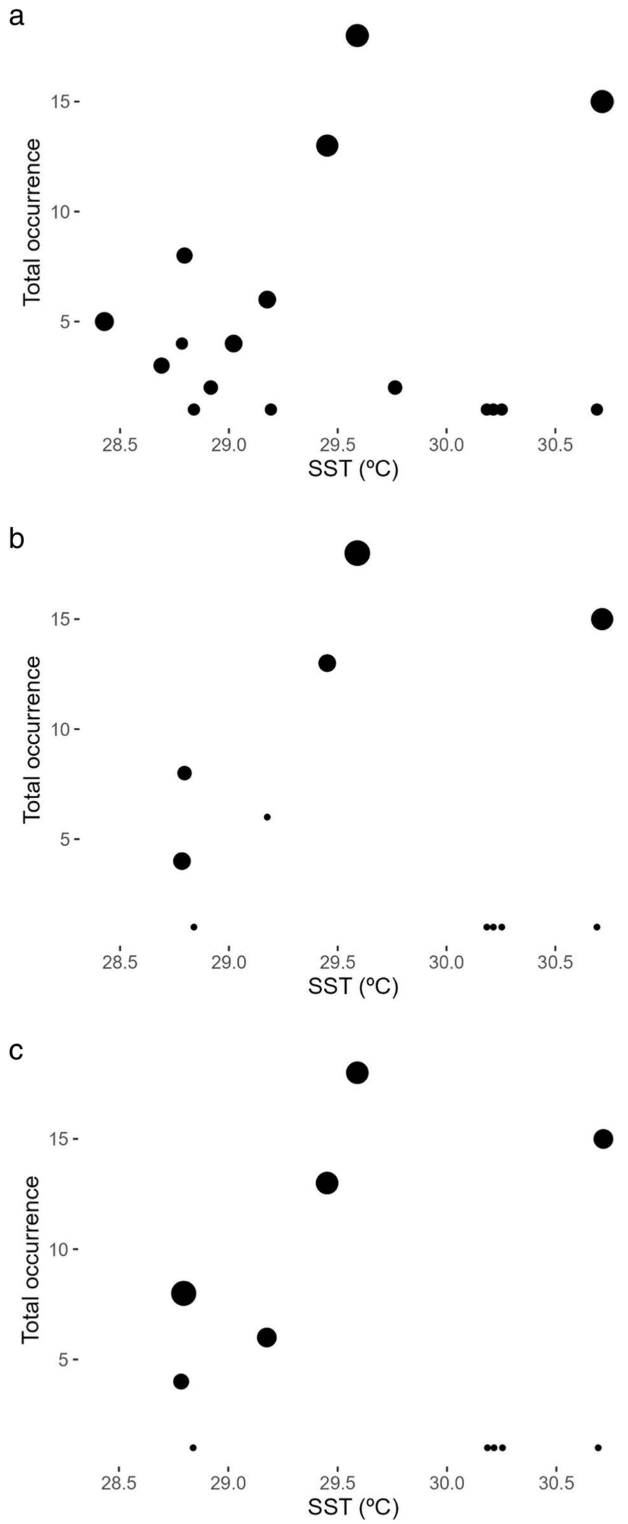

Fig. 5 Effect of Sea Surface Temperature (SST) on *C. mydas* nesting behavior during the 2018–2020 breeding seasons. **a** complete oviposition; **b** incomplete oviposition; **c** false crawl. Distribution of total occurrences ($n = 756$) related to TSM. The number and position of the circles represent the total number of occurrences at each temperature (°C); the size of the circles indicates the relative proportion of the number of complete oviposition—a, incomplete oviposition—b and false crawl—c

is related to normal conditions of annual climate variation. There is no data indicating that turtles could take advantage of climate change (Hawkes et al. 2009).

The nesting preference for SSTs around 29.5 °C observed in this study is similar to the pivotal temperature of ~ 29 °C, which determines the production of 50% females and 50% males in *C. mydas* nests (Broderick et al. 2000; Godley et al. 2002). Although this pivotal temperature is related to the temperature of the sand at first, this relationship may indicate a strategic behavior of turtles' environmental perception, starting from the time they leave the water to nest to obtain greater success in the sexual determination of their hatchlings. Such a relationship was recently observed for the hawksbill turtle (*E. imbricata*), which changed its peak nesting period to months with milder SST to preserve the sex ratio of hatchlings in the Rio Grande do Norte, Brazil, suggesting that this species can be responding to climate change (Oliveira et al. 2020).

The animals most dependent on environmental conditions will likely be the most affected by climate change. Thus, understanding the effect of SST on turtle nesting can help predict the behavior of the green turtle in the face of climate change, as well as SST anomaly phenomena in the East Pacific, such as El Niño, which affected corals in the Rocas Atoll in 2009–2010 (Ferreira et al. 2012) and SST anomalies over the tropical Atlantic (Atlantic Dipole) (Nóbrega et al. 2016).

This work analyzed the synergistic effect of environmental variables on the nesting behavior of *C. mydas* in the Rocas Atoll and concluded that the preference of turtles to emerge and nest on the beach occurs at higher temperatures (TSM), around 29.5 °C. Green turtles in Rocas Atoll seem to respond to the difference of fraction of degree Celsius in relation to the SST, in a current alarming scenario of climate crisis, the impacts of climate change may cause the decrease and loss of the nesting area in oceanic islands (Witt et al. 2010), such as Rocas Atoll, and change the nesting behavior and consequently the distribution of this species. Furthermore, while lunar luminosity and lunar phases may influence the nesting pattern of green turtles, they do not limit it, as turtles come ashore under different observed environmental conditions.

We suggest that other environmental conditions should be reported in future studies, such as precipitation and SST anomalies (e.g., El Niño, La Niña, and Atlantic Dipole). It is also suggested the development of studies in the other two main nesting sites of *C. mydas* in Brazil – Trindade Island and Fernando de Noronha Archipelago – to verify if the behavior related to lunar phases and tides occurs due to the immersion and emergence configuration of the coastline of each specific area or due to a biological rhythm of species synchronicity. In addition, a larger sampling size would be more suitable for the statistical analysis of lunar phases since a study encompassing more seasons could sample the same turtles returning to nest after the remigration period, favoring the observation of possible individual nesting patterns. Understanding the influence of environmental variables on the nesting behavior of the green turtle can improve the monitoring effort for its conservation, in addition to understanding how this endangered species will respond to natural climate changes and, mainly, those caused by humans.

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Data Availability The data sets generated and/or analyzed during this study are available from the corresponding author on reasonable request.

Declarations

All applicable international, national, and/or institutional guidelines for the care and welfare of animals were followed. The sampling of green turtles on Rocas Atoll was authorized by the Brazilian Government by Biodiversity Authorization and Information System (SISBIO) through the sampling license number 59809.

Competing interests All authors certify that they have no affiliations with or involvement in any organization or entity with any financial interest or non-financial interest in the subject matter or materials discussed in this manuscript.

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