



STRANDED TURTLES IN THE SOUTHEAST COAST OF **BRAZIL: WHICH ARE THE RELEVANT FACTORS?**

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Abstract

Sea turtles are one of the many marine species in constant threat by the human population. To help the species conservation, studies based on stranded turtles found in coastal areas are crucial, since they can provide important data about the cause of death. Therefore, we evaluated the stranding patterns that occur in a South American region, on the southeast coast of Brazil, from 2011 to 2016. Our findings show that dead or alive stranded turtles exhibit different stranding patterns, which might be explained by oceanographic processes (like coastal/oceanic currents and upwelling), nutrient enrichment in coastal areas, and biological factors (like parasitosis and plastic debris ingestion). We expect the results presented here, shall shed light on this issue and contribute to the development of intervention plans to diminish life-threatening problems to sea turtles.

Keywords: Chelonia mydas; fishing; parasites; health; immunity

INTRODUCTION

Among marine fauna, sea turtles belong to the group with long-distance migratory behavior, occupying niches at different geographical regions, according to their developmental stage (BOLTEN, A., 2003). Therefore, they are highly susceptible to suffer from anthropic threats, like accidental capture and boat collision, ingestion of plastic waste, debris, among others (Hamann et al. 2010; Santos et al. 2015; Reis et al. 2017). Five sea turtle species are found in Brazilian coast: loggerhead (*Caretta caretta* Linnaeus, 1758), green turtle (*Chelonia mydas* Linnaeus, 1758), leatherback (*Dermochelys coriacea* Vandelli, 1761), olive ridley (*Lepidochelys olivacea* Eschscholtz, 1829) and hawksbill (*Eretmochelys imbricata* Linnaeus, 1766) (MARCOVALDI; DEI-MARCOVALDI, 1999).

Sea turtles in Brazil are threatened by incidental capture in fishing artifacts (BUGONI et al., 2008; MARCOVALDI; DEI-MARCOVALDI, 1999) and marine pollution (BJORNDAL, K.

A.; BOLTEN, A. B.; LAGUEUX, 1994; IVAR-DO-SUL; COSTA, 2007; SANTOS, Robson Guimarães et al., 2015; TOURINHO; IVAR DO SUL; FILLMANN, 2010). Besides these anthropic threats, turtles can also suffer from debilitating diseases, like fibropapillomatosis (HERBST, 1994; JONES et al., 2016), which can impair animal ability to feed or swim, eventually causing death (GEORGE, 1997; RENAN DE DEUS SANTOS et al., 2017; SANTOS, M. et al., 2015). Dead or sick turtles end up stranded at coastal areas, becoming part of an important database that is fundamental for ecological studies to help species conservation (POLI et al., 2014). Moreover, the records of stranded turtles are useful for a better understanding of the environmental conditions of coastal areas, serving as a baseline for environmental management and conservation plans.

Previous studies about stranded sea turtles in South America have described their occurrence in the Northeast region of Brazil (MASCARENHAS, Rita; SANTOS, Robson; ZEPPELINI, 2004), showing that *C. mydas* is the major species found, with fishing and trash consumption being the major human threats (POLI et al., 2014). Here we expanded this edge of knowledge, studying stranded turtles in a Southeast region of Brazilian coast, inferring what could be the possible causes for their stranding pattern.

MATERIAL AND METHODS

1. Study area and monitoring strategy

This study evaluated data of stranded turtles found between 2011 to 2016, on the Southeast coast of Brazil. This coastal shore corresponds to 763 km between the north of Espírito Santo state (18°20'09" S and 39°39'49" W) and the central region of Rio de Janeiro state (22° 56' 11" S and 42° 38' 02" W). Two monitoring strategies were adopted: by daily regular beach monitoring (walking or aboard vehicles) or by rescue calls (by phone calls to rehabilitation centers located between these coordinates). In this last strategy, stranding occurrences were reported to the researchers through partnerships with environmental agencies and the local community. The area studied was divided into 11 evaluation points (P1 to P11), based on their proximity to monitoring centers. The database used in this study is part of a continuous monitoring program established by the Brazilian government as a precondition for oil and gas exploitation by Petrobras Inc. Therefore, the results of these studies were compiled in the 6th annual report called Programa de Monitoramento de Praias, Área da Bacia de Campos e Espírito Santo.

2. Turtles and classification of health status

All species found in the Brazilian coast (*C. mydas*, *C. caretta*, *E. imbricata*, *L. alvacea*, and *D. coriacea*) were identified and considered in our study. However, we excluded all reproductive activity registries from our data analysis since these are not considered stranding events. Each stranded turtle was initially classified as dead or alive. When alive, an overall health score was determined for the animal. The health scores is based upon three qualitative values:: Good (when animals had no clinical signs of disease, the amount of muscle and fat were within a predetermined average based upon age, and the turtle's reflexes were normal), bad (when turtles were debilitated, with clear signs of advanced disease stage and exhibiting almost no reflexes), and intermediate (when the animal overall clinical health was somewhere between the two extremes).

3. Classification of anthropic or natural causes interfering with turtle health

For each turtle found, any signs of interaction with products from human origin/activities was determined. When a turtle died during treatment, a necropsy analysis was performed to determine whether death was caused by anthropic or natural causes. Animals with anthropic signs of death were divided into three categories: by fishing (the presence of injuries incurred

after interaction with fishing lines from nets or fishing rods), by boats (corresponding to cases when turtles present clear signs of injuries caused by collision with boats or their propellers), or by trash (corresponding to cases where death was caused by the appearance of plastics or other human-produced materials attached to their bodies or inside their gastrointestinal tract). Turtles suffering from natural causes were also divided into three groups: from tumors (corresponding to cases exhibiting signs of abnormal tissue growth, often fibropapillomas), from parasites (corresponding to ecto- or endo-parasites, the latter being the most common most common), or from predation (corresponding to clear signs of predator attack).

4. Statistical analysis

The normal distribution of our data was confirmed by a Kolmogorov–Smirnov test. Comparisons between monitoring points (P1 to P11) were performed by ANOVA with Tukey correction. The analyses were performed by Graphpad Prism version 5.00 for Mac (Graphpad Software, San Diego, California, USA). For the statistical analyses, results were considered significant for p < 0.05.

Multivariate analyses were performed to simultaneously compare and identify the list of anthropic or natural factors that were the most relevant for each health status group. The relevance of a characteristic is determined by the magnitude and direction of its corresponding vector, more important vectors point to the associated group. This analysis was carried out with the FITOPAC software (Unicamp, SP, Brazil).

RESULTS AND DISCUSSION

1. The occurrence of stranded turtles in the southeast coast of Brazil

From the total of 22,585 stranded turtles, 1,266 were found alive (5.6%). Evaluating all points where this total amount of stranded turtles was found, P8 was the point with the highest number of cases (Figure 1). After it, the numbers of turtles found in P6 and P7 were also higher than all other points (Figure 1). The highest occurrence of strandings may be related to the large-scale currents pattern. At the Brazilian coast, the rise of South Atlantic Central Water (SACW) dictates the upwelling events at South-Southeast region (COELHO-SOUZA et al., 2012), which is highly dependent of wind direction. These events are more evident at Cabo Frio region (23° S, 42° W) and they are more frequent and intense during

austral springs and summers (CALADO; GANGOPADHYAY; SILVEIRA, DA, 2008; CASTELAO; BARTH, 2006; REIS, E. C.; GOLDBERG; LOPEZ, G. G., 2017). Moreover, the geometrical line of the Brazilian coast changes after Cabo de São Tomé (also located just below P8), from North-South to East-West, as well as the bottom ocean topography, both having a high impact in this upwelling (RODRIGUES; LORENZZETTI, 2001). Since the Cabo Frio upwelling is right below P8, it is possible that it contributes for the high numbers of strandings of dead turtles at this point. During the summer, the intensification of the upwelling process associated with the northeast wind may make the region attractive to live turtles due to the greater supply of food. Already in the winter, a period of weak upwelling, the current pattern is inverted, with intensification of the southwest wind that pushes the surface water towards the continent, causing stacking of water on the coast and increasing the probability of the dead-turtle strandings (Reis et al. 2017).

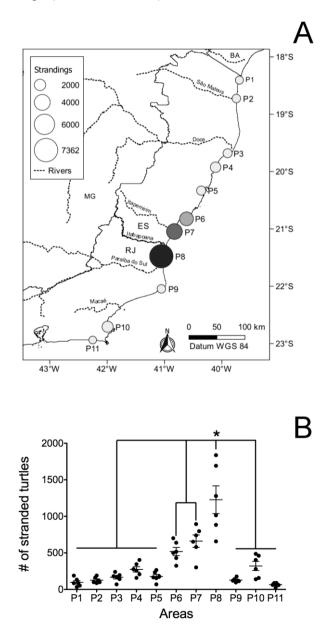


Figure 1 – The number of stranded turtles at the southeast Brazilian coast. From 2011 to 2016 (6 yrs.), the amounts of stranded turtles were counted on the southeast coast of Brazil. (A) Each area where turtles were found is represented by a circle, where the higher radius and darker colors are directly related to higher amounts of stranded turtles. (B) Each point represents the number of stranded turtles found in each year. The mean <u>+</u> standard deviation (SD) of each area is represented. *p<0.05.</p>

It is interesting to notice that, every year where the P6 and P7 numbers of strand turtles were high, P8 followed the same tendency (Table 1). However, for P10, an inverse correlation was observed, when we consider thresholds of 1,200 and 400 turtles for P8 and P10, respectively; in these cases, when P8 numbers were higher than 1,200 stranded turtles, P10 numbers were lower than 400 stranded turtles, and viceversa (Table 1). Besides the considerations about the dynamics of coastal/oceanic streams already discussed, it is possible that other factors may contribute to these stranding differences cited above. For example, after a long period of migration between Africa and South America, through the South Atlantic Subtropical Gyre, it is possible that many debilitated turtles arrive to each coast and start to look for food in high primary productivity regions (LUSCHI et al., 1998; PAPI et al., 2000). Therefore, variations of food ingestion at arrival may contribute to have turtles with variable health status at each coast, as we described here.

	Areas										
Years	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11
2011	134	87	196	403	235	701	797	1837	139	139	89
2012	189	99	240	345	265	500	894	1692	177	157	59
2013	88	163	181	276	166	639	740	1279	113	280	88
2014	86	99	172	257	177	517	653	883	121	461	72
2015	53	190	142	207	155	430	580	1014	120	487	24
2016	34	124	69	158	69	324	302	657	96	403	62
Total	584	762	1000	1646	1067	3111	3966	7362	766	1927	394

Table 01 – Numbers of stranded turtles found in different areas of Brazilian southeast coast*.

*During 2011 to 2016 (6 yrs.), the amounts of stranded turtles were counted in different areas at the southeast coast of Brazil.

When we considered live-stranded turtles only, their higher numbers at P8 than other evaluated places (Figure 2, A and B) also corroborate to the hypothesis of the Brazilian Current influence in turtle stranding. However, there was one difference in this trend, since the numbers of live stranded turtles at P7 seem to follow in the opposite direction. While the total amount of stranded turtles at P7 is high, the numbers of live-stranded turtles are low (Figures 1 vs. 2). Despite being influenced by passive transport of marine currents, turtles can actively change their route in search of areas with higher primary productivity and food, favoring foraging activity. Therefore, P7 possibly is an area without any coastal attractive trait (e.g., food source) for live turtles (since P7 area is a long sandy coastline), keeping them far from the coastal line. This nutritional need hypothesis is supported by the amount of river mouths at P8, known as upwelling area (COELHO-SOUZA et al., 2012) and high primary productivity (REIS, E. C. et al., 2010). Among these river mouths we found the Paraíba do Sul River, the largest river on the Brazilian Southeast, with a river mouth of 50 km (PACHECO et al., 2017). Paraíba do Sul River basin includes one of the most populated area of Brazil, which may influences positively the primary productivity because of wastewater discharges (urban and industrial) and nutrients runoff from agriculture (ARBUCKLE; DOWNING, 2001).

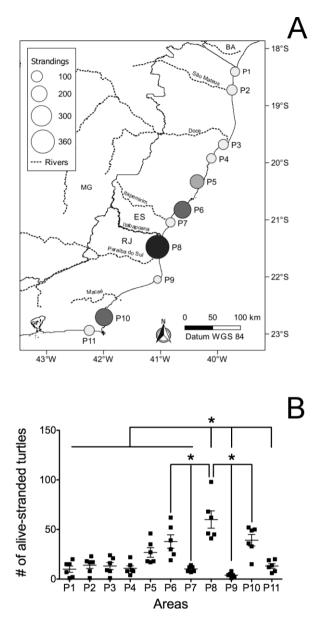


Figure 2 – The number of alive-stranded turtles at the southeast Brazilian coast. From 2011 to 2016 (6 yrs.), the amounts of alive-stranded turtles were counted on the southeast coast of Brazil. (A) Each area where turtles were found is represented by a circle, where the higher radius and darker colors are directly related to higher amounts of stranded turtles. (B) Each point represents the number of stranded turtles found in each year. The mean <u>+</u> standard deviation (SD) of each area is represented. *p<0,05.</p>

Nutrient enrichment of water bodies is a growing global problem and excessive nutrients is the primary cause of eutrophication in coastal waters (HOWARTH; MARINO, 2006; SINHA; MICHALAK; BALAJI, 2017). Associated impacts include the increase in primary productivity and occurrence of algal blooms. Urbanized watersheds such as the Paraíba do Sul River usually provide higher inputs of nutrients and organic matter to coastal areas. This pattern could be intensified in rainy periods, being result from the impact of high stormwater runoff on the watershed. Ovalle et al (2013) conducted a long-term study to

evaluate hydro-chemical variation in the basin outlet through estimations of annual fluxes and identification of their controlling factors in the Paraíba do Sul River. The authors found that concentrations of organic matter and nutrients like dissolved organic carbon (DOC), ammonium, nitrite and phosphate had increased values during the high discharge period. Seasonal patterns of DOC, phosphate and the reduced forms of dissolved inorganic nitrogen has a strong linkage to transient hydrological flow paths and the main source of these compounds is the mineralization of organic matter in natural and agricultural soils of the watershed (OVALLE et al., 2013). The results indicated that point and diffuse anthropogenic inputs delivery large amounts of nutrients into the Paraíba do Sul River channel (OVALLE et al., 2013).

The nutrient enrichment on coastal water, resulting from both the continental fluvial supply from the Paraíba do Sul River and the Cabo Frio upwelling, supports the hypothesis of a relative increase in primary productivity in P8, providing higher availability of food and serving as attractive for turtles that come from their long journey across the Atlantic Ocean. In this sense, we have two important events for the increase of primary productivity, one of a natural order, the Cabo Frio upwelling, and another strongly intensified by human action, the fluvial contribution of the Paraíba do Sul river which due to great extension and complexity of the watershed is an immense challenge for the water management of water quality and coastal ecological dynamics.

Still corroborating that the high primary productivity attracts live turtles to the shore, at Cabo Frio region, tropical algae species adapted to cold water have a reproductivity peak due to upwelling (GUIMARAENS, DE; COUTINHO, 1996). For example, green algae *Ulva sp.* reproductivity peak occurs during upwelling season (GUIMARAENS; PAIVA; COUTINHO, 2005) and this is one of the food sources of adult green turtles as shown by other authors (SANTOS, Robson G. et al., 2011; VÉLEZ-RUBIO et al., 2016).

As mentioned above, the health status of the turtles can be one important factor to influence these stranding patterns. Possibly, that animals with a good health status could decide to stay in the Brazilian Current, while animals with intermediate or bad health status would not. Alternatively, animals with good health status may not want to leave the Brazilian Current while turtles with intermediate or bad health status would leave, chasing for food and recovery. Then, to evaluate these possibilities, we divided these numbers of alive turtles in three different groups: turtles with good, intermediate or bad health status (Figure 3).

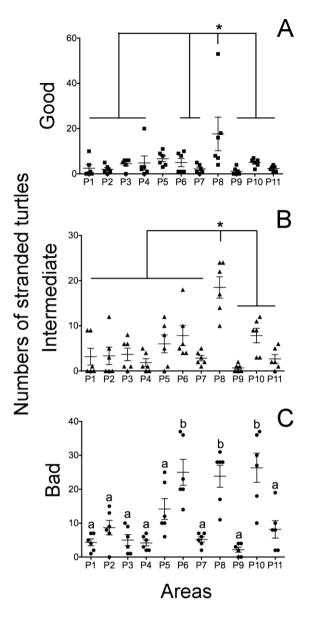


Figure 3 – The number of stranded turtles found at different health status. From 2011 to 2016, the number of stranded turtles found in Good (A), Intermediate (B), or Bad (C) health status were counted on the southeast coast of Brazil. Each point represents the number of stranded turtles found by year. The mean <u>+</u> standard deviation (SD) of each area is represented. *p<0,05. In C, different letters represent statistically different data.

According to our data (Figure 3), P8 was also the area with the highest amounts of animals found (from all health status here determined), which corroborates with the hypothesis that oceanic currents are the major contributors to this stranding. However, one interesting occurrence was observed. When we performed this division of animals with different health status, turtles with a bad health status had also a broad preference to strand, being found at P6, P8, and P10. In these cases, it seems that turtles with a bad health status have decided to leave the oceanic current and have moved near to the shore. It is possible this behavior is motivated by the amount of food sources presented at these areas, since

these animals would need to spend energy to recover from their debilitate health status. This hypothesis is supported by two facts: i) *Chelonia mydas* is the most common species found in all areas (Figure 4) and ii) a predominant presence of *Ulva fasciata* algae in these areas, one of the components of *C. mydas* diet (SANTOS, Robson G. et al., 2011). Since P6, P8, and P10 areas are close to urban areas, this causes an impact in the coastal distribution of algae, with a predominant distribution of *U. fasciata* because their intrinsic characteristic. Their spores can use bacterial extracellular substances for substrate adherence (SHIN, 2008; SINGH et al., 2013) and they are resistant to environmental contaminants, making them useful in macroalgal bioassays (YOSHIOKA et al., 2016). This algae consumption may not help these debilitated turtles, since it was already shown that *U. fasciata* could also accumulate higher amounts of heavy metals in these conditions (LEE; WANG, 2001) and, therefore, accumulate these metals inside *C. mydas* bodies. This Ulva sp. restricted diet could lead to a micronutrient debt (SANTOS, Robson G. et al., 2011), increasing immunological and hormonal deficits, which in turn could be associated to fibropalillomatosis development (SANTOS, M. et al., 2015; SANTOS, RG et al., 2010).

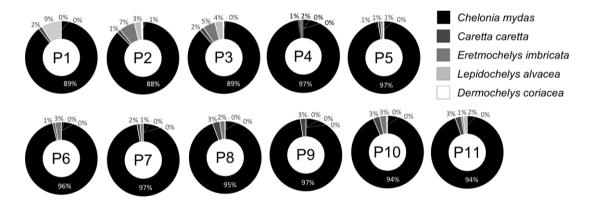


Figure 4 – Percentage of turtle species found in each area at the southeast Brazilian coast. From 2011 to 2016 (6 yrs.), the amounts of each species of stranded turtles were counted on the southeast coast of Brazil. Each pie chart is numbered according to the data of a monitored area (from P1 to P11). The percentages of each species found are represented.

2. Types of human interactions and health issues found in live-stranded turtles

We already observed that there were patterns in stranding of turtles along the studied area. However, these patterns have not indicated if interactions between turtles and human activities had influenced. Thus, we decided to evaluate what were the types of human interactions or problems from natural causes, that could be observed in these live-stranded turtles. In Figure 5, in all places but P1, fishing was the most important interaction with good health score turtles. At P1, interaction with boats and predation were the most observed interactions. It is interesting to note that P8 was the only point with higher strands of turtles with good health score than other points (Figure 4A). It seems these juvenile turtles are captured in gillnets, a common fishing equipment used in Brazilian coast, mainly in small scale fisheries (LÓPEZ-BARRERA; LONGO; MONTEIRO-FILHO, 2012). Other authors have already identified anthropic interactions with turtles at the South of the region here described, during 2008 to 2010, where fishing, boat collision, and trash consumption were described (REIS, E. C.; GOLDBERG; LOPEZ, G. G., 2017). Therefore, our study is confirming the historic sequence of events that may contributed to stranding of turtles.

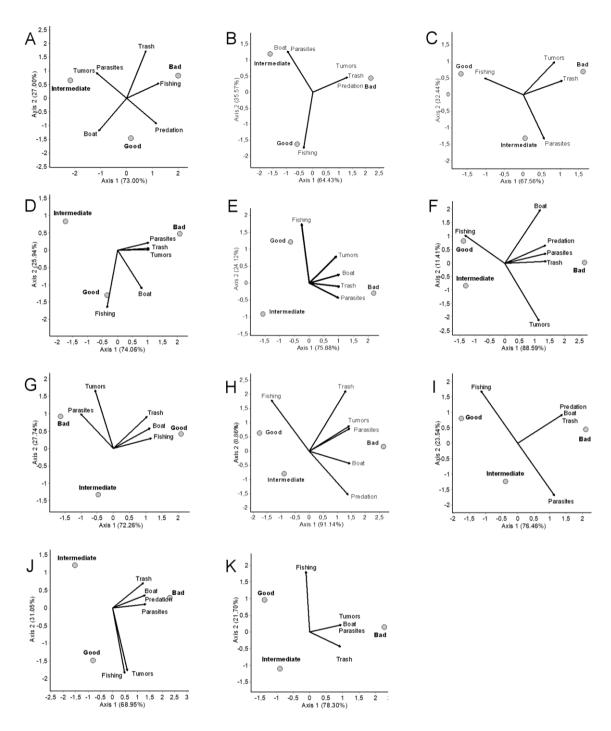


Figure 5 – Anthropic interactions or problems from natural causes found in alive stranded turtles. During 2011 to 2016 (6 yrs.), interactions with anthropic products (fishing, boat, or trash) or problems from natural causes (tumors, parasites, or predation) were evaluated in all alive stranded turtles found from P1 to P11 points (A to K, respectively). The proximity and length of each vector to a turtle health status (Good, Intermediate, or Bad) indicates its relevance. When a factor is missing, its relevance is null.

Animals with intermediate health score were also found at higher numbers at P8 than other points (Figure 4B). Again, fishing was the most frequently human interaction found in juvenile turtles (Figure 5H). We highlight that P8 is a region with high primary productivity, leading to high diversity of fishes and, consequently, more fishing practices. Second to fishing, parasites were the most common natural interference found in animals with intermediate health score in most of these points (Figure 5). Possibly, some immunosuppression is already in place, compromising their fight against parasites.

Corroborating with this immunosuppression hypothesis, more animals with a bad health score were found at P6, P8, and P10 points (Figure 4C) and the most common find in these animals were parasites (Figure 5F, 5H, and 5J). Therefore, likely this bad heath score is interfering with their immune system and making them more susceptible to infections. Trash consumption was also found in these turtles with a bad health score from 2 of these 3 points (Figure 5F and 5J), and it is known that trash ingestion is an important cause of juvenile green turtle deaths (SANTOS, Robson Guimarães et al., 2015). Therefore, it is also possible that plastic debris ingestion would cause harm to their immune responses. Predation was also well represented in these animals from P6 and P10 (Figure 5F and 5J), which can be caused by their debilitated state and diminished capacity scape from predators.

After all these analyses, specific turtle-stranding regions are found at this part of the Brazilian coast, and factors like coastal/ocean currents, turtle health status, and feeding habits would explain why this stranding is happening. However, our results only indicate that human interactions like fishing and littering could be the most important factors to contribute for turtle deaths. Future studies will be necessary to understand the dynamics of these type of human interactions and their contribution to the death-stranded turtles found in this part of the Brazilian coast.

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