

Project Update: December 2018

Background

The southwestern Atlantic Ocean (SWA) is an important habitat for loggerhead sea turtles (*Caretta caretta*), with several important nesting and feeding grounds (Marcovaldi and Marcovaldi 1999, Vélez-Rubio et al. 2013, Carman et al. 2016, Monteiro et al. 2016). The Brazilian nesting population is one of the largest in the world (ca. 7000 to 8000 nests/year; Marcovaldi et al. 2018), with rookeries ranging over a wide latitudinal area from Sergipe (northeast) to Rio de Janeiro (southeast) (Marcovaldi and Chaloupka 2007). These rookeries hold endemic mitochondrial DNA (mtDNA) haplotypes, which provide a unique genetic profile (Reis et al. 2010), and were recognised as three independent management units (MUs, Moritz 1994) within the SWA: (1) the northeastern coast (Sergipe and Bahia), (2) Espírito Santo, and (3) Rio de Janeiro (Shamblin et al. 2014). The genetic differentiation of nesting populations worldwide enables the estimation of the origin of turtles sampled in foraging grounds, which are composed of individuals from multiple rookeries (i.e. mixed stocks), and offer great insights about the migratory behaviour and connectivity of sea turtle populations (Reis et al. 2010, Rees et al. 2017, but see Prosdocimi et al. 2015). Northern and southern Brazil are important foraging grounds for adults and juveniles of loggerheads in both neritic and oceanic habitats (Barros 2010, Marcovaldi et al. 2010, Monteiro et al. 2016). Fishery activities extensively overlap with loggerhead feeding aggregations in these areas, which increases the probability of turtle bycatch and the high fishing-related mortality (Sales et al. 2008, Wallace et al. 2013). However, the effects of this mortality on the demographic and genetic structure of loggerhead populations remain unclear. Populations that decline abruptly in abundance may undergo population bottleneck events, which increase inbreeding and genetic drift, leading to loss of genetic diversity and evolutionary potential (Frankham et al. 1999). Populations that have recently undergone population bottleneck events are rare allele deficiencies, and it is possible to detect this event analysing allelic frequency distribution and the excess heterozygosity in relation to the total number of alleles (Cornuet & Luikart 1996). The current study aims to characterise the genetic composition of loggerhead populations from Brazilian foraging grounds, verify the occurrence of bottleneck events, and assess whether the genetic characteristics of these animals influence their foraging ecology.

Project Progress

In the latter half of 2018, we finished laboratory activities related to molecular analysis. We also finalised the edition and classification of mtDNA haplotypes of loggerheads caught in fisheries activities. To identify the stocks that compound the feeding aggregations in Brazilian waters, we analysed the mtDNA of loggerheads caught in fishing weirs (n=80), pair trawl (n=67) and longline fisheries (n=250). Eleven distinct haplotypes were found, showing different population composition among loggerheads foraging grounds (Table 1). However, it is important to note that all foraging grounds that are overlapping with fisheries in Brazil are composed in greater proportion by individuals from Brazilian rookeries.

Table 1. Haplotype frequencies in percent (%) of loggerhead sea turtles (*Caretta caretta*) bycatch in fisheries activities in Brazilian waters. *** not found.

Haplotype	Fish weir	Pair trawl	Longline fishery
CC-A1.1	1.9	***	***
CC-A2.1	5.8	***	3.3
CC-A3.1	3.9	***	***
CC-A4.1	23.0	31.5	33.8
CC-A4.2	52.0	63.0	42.7
CC-A4.3	***	***	0.8
CC-10.1	1.9	***	***
CC-A11.6	***	1.8	2.4
CC-P1.1	***	1.8	6.5
Cc × Lo	11.5	***	***
New	***	1.8	10.5

The CC-A1.1 is a haplotype frequent in rookeries from eastern coast of United States (USA, Shamblin et al. 2014). The CC-A2.1 is the most geographically widespread haplotype and is present in all of the western Atlantic, eastern Atlantic, and Mediterranean rookeries, except for those in Brazil (Garofalo et al 2009, Reis et al. 2010, Yilmaz et al. 2011). CC-A3.1 has been identified in NW Atlantic rookeries and in Mediterranean nesting sites of Turkey, Lebanon, and Libya (LaCasella et al. 2013). The haplotypes CC-A4.1 and CC-A4.2 are variants of CC-A4, which is the ancestral and exclusive haplotype of the Brazilian populations (Reis et al. 2010, Shamblin et al. 2014). The haplotype CC-A10.1 was reported in rookeries from Florida, USA, and Mexico (Shamblin et al. 2014). The haplotype CC-A11.6 was recognised as an Atlantic lineage that colonised the Indian Ocean, mainly Oman nesting sites (Shamblin et al. 2014). The haplotype Cc-P1.1 was previously assumed to characterise Pacific loggerhead populations nesting in Western Australia and southern Japan (Bowen et al. 1995, Matsuzawa et al. 2016). Comparing polymorphic sites of the new haplotype and the haplotypes previous reported for the Atlantic and the Pacific Oceans and the new haplotype, this new haplotype is a variation of the Cc-P1.1. Two haplotypes typical of olive ridley (*Lepidochelys olivacea*) sea turtles, haplotype E and F (referred to as Cc × Lo haplotype in Table 1, Bowen et al. 1998). Hybrids showed the morphology of pure loggerhead turtles and were identified as such, but their mtDNA haplotype was characteristic of olive ridley rookeries from Brazil and Guinea Bissau (haplotype F), and Suriname (haplotype E and F), showing hybridization between these two species.

Furthermore, throughout this last semester, we finalised the manuscript about origin and foraging ecology of male loggerhead sea turtles entitled "*Origin and foraging ecology of male loggerhead sea turtles from southern Brazil revealed by genetic and stable isotope analysis*" and submitted to *Marine Biology*. In this study, we present the first insights about origin and foraging ecology of male loggerheads from the SWA, by integrating molecular and stable isotope analysis (SIA). Most males exhibit endemic haplotypes from Brazilian rookeries, followed by a low frequency of a haplotype from the North Atlantic and the Mediterranean Sea, as well as Cc × Lo hybrids. SIA showed size-related differences in feeding and habitat use by male loggerheads, with benthic invertebrates dominating the diet of adults, while pelagic prey items dominated the diet of juveniles. Our findings demonstrate the importance of southern Brazil neritic and oceanic habitats for male loggerheads and highlight the value of this area for the maintenance of SWA reproductive management units, which are the main

contributors to these feeding aggregations. The article is currently under review by the journal.

The next steps of our work will be to carry out the Bayesian Mixed Stock Analysis to estimate the contribution of each rookery to the foraging grounds that are overlapping with fisheries in SWA and perform the bottleneck analysis with microsatellites data to assess population bottleneck events in Brazilian loggerhead populations. To verify the occurrence of population bottleneck events, we amplified ten nuclear markers (microsatellites) previously described for sea turtles (Carreras et al. 2007, Monzón-Argüello et al. 2008). For this analysis, we processed 94 tissues samples of loggerhead nesting females from the main rookeries in Brazil: Sergipe (n=13), Bahia (n= 24), Espírito Santo (n=26), and Rio de Janeiro (n=30).

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