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ORIGINAL PAPER

Genetic fingerprinting reveals natal origins of male leatherback turtles encountered in the Atlantic Ocean and Mediterranean Sea

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Abstract Understanding population dynamics in broadly distributed marine species with cryptic life history stages is challenging. Information on the population dynamics of sea turtles tends to be biased toward females, due to their accessibility for study on nesting beaches. Males are encountered only at sea; there is little information about their migratory routes, residence areas, foraging zones, and population boundaries. In particular, male leatherbacks (*Dermochelys coriacea*) are quite elusive; little is known about adult and juvenile male distribution or behavior. The at-sea distribution of male turtles from different breeding populations is not known. Here, 122 captured or stranded male leatherback turtles from the USA, Turkey, France, and Canada (collected 1997–2012) were assigned to one of nine Atlantic basin populations using genetic analysis with microsatellite DNA markers. We found that all turtles originated from western Atlantic nesting beaches (Trinidad 55%, French Guiana 31%, and Costa Rica 14%). Although genetic data for other Atlantic nesting populations were represented in the assignment analysis (St. Croix, Brazil, Florida, and Africa (west and south), none of the male leatherbacks included in this study were shown to originate from these populations. This was an unexpected result based on estimated source population sizes. One stranded turtle from Turkey was assigned to French Guiana, while others that were stranded in France were from Trinidad or French Guiana breeding populations. For 12 male leatherbacks in our dataset, natal origins determined from the genetic assignment tests were compared to published satellite and flipper tag information to provide evidence of natal homing for male leatherbacks, which corroborated our genetic findings. Our focused study on male leatherback natal origins provides information not previously known for this cryptic, but essential component of the breeding population. This method should provide a guideline for future studies, with the ultimate goal of improving management and conservation strategies for threatened and endangered species by taking the male component of the breeding population into account.

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Introduction

Marine animals exhibit varying patterns of dispersal and distribution in relation to their breeding sites. Individuals capable of long-range movements may still breed seasonally in discrete colonies (Hoffman and Forcada 2012). Some

marine and terrestrial species exhibit a distinct preference for particular sites; this behavior results in site fidelity (Baker et al. 1995), with some species exhibiting sex-biased variability in their site fidelity. In northern fur seals, Baker et al. (1995) demonstrated that fidelity to a natal rookery was evident for both males and females, and fidelity increased with age, resulting in animals nearing breeding age remaining closer to their natal rookeries. While females may exhibit very strict site fidelity (Antarctic fur seal females return to less than a body length (~2 m) of their natal site; Hoffman and Forcada 2012), less is known about male philopatry or dispersal in migratory species. For example, due to selective forces such as foraging competition and a harsh physical environment, the wandering albatross exhibits differential migration resulting in sex-segregated foraging zones (Åkesson and Weimerskirch 2014).

Marine turtles occupy broad geographic ranges including separate breeding and foraging areas; however, the boundaries of these ranges are difficult to resolve (Wallace et al. 2010). Like the seals and birds described above, they are highly migratory, traveling great distances between reproductive sites and foraging grounds (Hamann et al. 2010). While breeding areas are relatively discrete geographically and genetically, foraging areas for marine turtles may comprise individuals from multiple populations and geographic locations (Bowen et al. 2005; Murphy et al. 2006; Bolker et al. 2007; Doyle et al. 2008; López-Mendilaharsu et al. 2009; Wallace et al. 2010; Tucker et al. 2014). Their broad distribution makes them vulnerable to a variety of anthropogenic threats including interactions with fishing gear (Stewart et al. 2016), entanglement, and marine debris ingestion at varying spatial scales, which are important to understand for management purposes because certain populations (especially small ones) may be differentially affected by these threats (Stewart et al. 2016).

Understanding migration routes, population boundaries, high-use areas, and connections that exist among rookeries and foraging grounds are key to sea turtle conservation (Hamann et al. 2010; Casale et al. 2012) because protection may require cooperation among multiple jurisdictions and countries and multiple methods of assessing threats to individual populations. Traditionally, long-range turtle movements have been tracked using conventional tagging and satellite telemetry methods (e.g., Block et al. 2011; Bailey et al. 2012; Fossette et al. 2014; Hays et al. 2016). However, advances in genetics now contribute to our knowledge of population structure, movements, distribution, and behavior of sea turtles (Hamann et al. 2010; Jensen et al. 2013; Komoroske et al. 2017). One powerful genetic tool, assignment testing, may be used to determine the source nesting populations of individual animals sampled in foraging or developmental habitats (Stewart et al. 2013). Female sea turtles have previously been shown to have high levels of site fidelity to a beach or region (Bowen and Karl 2007; Wallace et al. 2010; Stewart and Dutton 2014; Bannister et al. 2016), and distinct genetic signatures of populations (or management units, MUs) reaffirm these findings (e.g., Dutton et al. 2013; Jensen et al. 2013; Roden et al. 2013; Shamblin et al. 2014). Foraging site fidelity is also observed in many sea turtle species, and most foraging areas are composed of mixed stocks (i.e., they are a mixture of individuals from various nesting populations) (James et al. 2005; Velez-Zuazo et al. 2008 Schofield et al. 2010).

Information on male marine turtle life history is limited compared to the more accessible females, and their spatial and temporal distributions are not well understood. Unlike females, they rarely come ashore; thus, most studies require the difficult task of locating and then humanely capturing them in water at breeding or foraging grounds (Fitzsimmons et al. 1997; James et al. 2005; Innis et al. 2010). Generally, it has been assumed that male-mediated gene flow plays a significant role in genetic diversity for turtles, even if the rate of gene flow is low. Although female marine turtles are known to return to natal regions when ready to breed, less is known about the fidelity of male turtles. Satellite-tracking studies show that mature male turtles travel to breeding grounds with some level of fidelity (James et al. 2005), and it is thought that these areas do represent natal rookeries (James et al. 2005; Stewart et al. 2013). In a tagging and telemetry study of green turtles at French Frigate Shoals, Dizon and Balazs (1982) concluded that adult males and females were faithful to a particular breeding area. In a subsequent tagging study of green turtles in the southern Great Barrier Reef, Limpus (1993) found males to exhibit fidelity to a particular courtship area, returning in successive breeding years. To further test the extent of male marine turtle philopatry, a genetic study of male mitochondrial DNA was performed on breeding male green turtles in Australia (FitzSimmons et al. 1997). The data indicated male green turtles, like females, are philopatric to courtship areas within their natal regions. Other studies have used satellite telemetry and capture–mark–recapture with flipper tags to study the broad movement patterns of foraging male and female leatherbacks encountered in waters off eastern Canada (James et al. 2005, 2007). Results suggested that males found in these temperate foraging grounds migrate with fidelity to waters adjacent to nesting colonies throughout the Caribbean, and Central and South America (James et al. 2005).

Male leatherback turtles (*Dermochelys coriacea*) are particularly elusive as this species has cryptic early life stages and males are rarely seen. There are not many data on male leatherback movements, mating behavior, long-term fidelity to courtship areas, or philopatry to breeding areas in the vicinity of natal beaches (but see James et al. 2005). The purpose of this study was to investigate the natal origins of male leatherbacks found in foraging areas or stranded

along beaches using genetic assignment testing. This type of genetic test assigns unknown individuals to their population of origin based on their multilocus genotype, and the expected probabilities of that genotype occurring in each of the potential source populations (Manel et al. 2005; Kalinowski et al. 2007). We were interested in assessing the sources of male leatherbacks found far afield from their breeding areas, which gives an indication of how far they have dispersed from their presumed natal regions. This work is a first step in refining population dynamics assumptions and parameters, to examine whether males in foraging areas come disproportionately from one (or a few) breeding population exclusively or whether multiple breeding populations are represented in the adult male at-sea distribution.

Materials and methods

Samples We searched the National Marine Mammal and Sea Turtle Research Collection at the Southwest Fisheries Science Center for skin and DNA samples that had been collected from male leatherbacks in the Atlantic from 1997 to 2012. Samples were collected by collaborators from stranding events, in-water captures, entanglement studies, and were from four countries: Canada ($n = 79$), USA ($n = 34$), France ($n = 8$), and Turkey ($n = 1$) (Fig. 1). All turtles were classified as Entangled (live), Stranded (usually dead), or Foraging (live capture) (Online Resource Table 1). Stranded animals were assumed to have been foraging nearby when discovered and sampled based on findings by Hart et al. (2006).

Genetic analysis We used standard manufacturer protocols and laboratory procedures to extract genomic DNA from 139 leatherback tissue samples using one of several extraction methods as discussed in Dutton et al. (2013). The extracted DNA, in addition to positive and negative controls, was amplified using polymerase chain reaction (PCR) with 15 polymorphic marine turtle microsatellite loci [N32 (Dutton 1995), 14-5, LB110, LB128, LB141, LB142, LB145, LB143, LB133, LB123, LB125, LB157, LB158 (Roden and Dutton 2011), C102, and D1 (Dutton and Frey 2009)]. Detailed primer information and reaction conditions are described in Roden and Dutton (2011), Dutton and Frey (2009), Dutton (1995), and Dutton et al. (2013). Positive controls of leatherback samples with known genotypes were included in each PCR and genotyped as well to detect any shifts in allele size throughout the study; however, no changes were detected. In addition, PCR-negative controls were included in all PCR reactions and included in the genotyping to detect potential contamination. The products of each PCR reaction, including positive and negative controls, were checked for amplification using 2% agarose gels stained with ethidium bromide (Maniatis et al. 1982). The reaction products were then separated by electrophoresis with an ABI Genetic Analyzer (ABI 3100, ABI 3130, or ABI Prism 3730) using ROX 500 fluorescent size standard (Applied Biosystems Inc, Foster City, CA, USA). No PCR multiplexing was performed; however, markers with microsatellite size ranges that do not overlap were multiplexed on the genetic analyzer. The PCR products of ten microsatellite loci were combined according to the following loci pairings: N32/D1, 14-5/LB123, LB128/LB143, LB125/LB157, LB158/C102. PCR products for microsatellite loci LB133, LB145, LB142, LB141, LB110 were not multiplexed for analysis on the ABI Genetic Analyzer. We used GeneMapper 4.0 (Applied Biosystems Inc, Foster City, CA, USA) to score alleles; each allele call was verified manually.

Statistical analysis: assignment tests To assign turtles to probable source populations, we used the microsatellite allele frequencies generated from nine demographically independent populations (DIPs) of Atlantic leatherbacks (collected between 1992 and 2008) described by Dutton et al. (2013) as our genetic baseline dataset. The nine populations included Brazil, Costa Rica (Atlantic), Florida, St. Croix (USVI), Trinidad and Tobago, French Guiana (including Suriname), Gabon, Ghana, and South Africa (Fig. 2). All individuals in this dataset ($n = 1417$) were nesting females and had genotypes for at least 11 of 15 microsatellite loci (genotype success rate >70% across 15 loci). These 15 loci were chosen based on genotyping

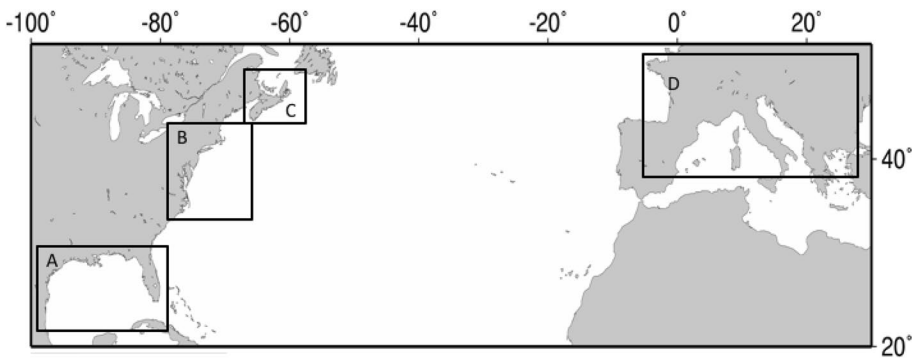

Fig. 1 Sample location areas for 122 leatherback turtles, resulting from strandings, or live-capture/entanglement studies in *A*. Southeast USA ($n = 4$). *B* Northeast and mid-Atlantic USA ($n = 30$). *C* Nova Scotia, Canada ($n = 79$). *D* Europe (France and Turkey, $n = 9$)

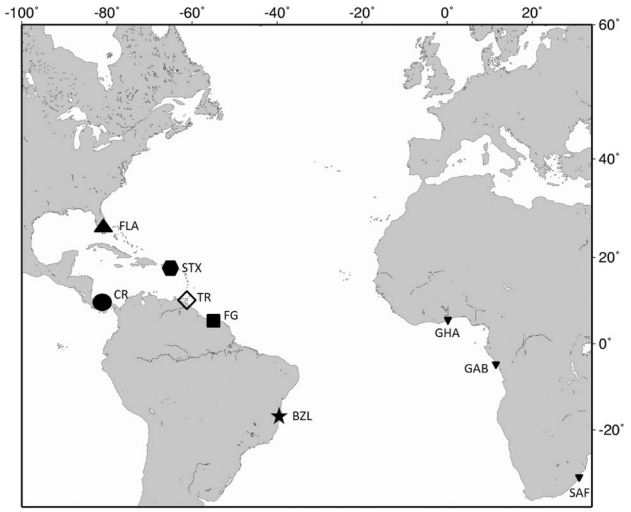

Fig. 2 Source populations for leatherback turtles in our genetic assignment reference dataset. These populations were identified in Dutton et al. (2013) and include Brazil—BZL (*star*), Florida—FLA (*triangle*), Costa Rica—CR (*circle*), French Guiana—FG (*square*), Trinidad—TR (*diamond*), St. Croix—STX (*hexagon*) and populations in Africa (*inverted triangles*) (Ghana—GHA, Gabon—GAB, and South Africa—SAF)

reliability, consistency, and breadth of data for these samples. There were no null alleles and no linkage disequilibrium at any of the loci for the baseline dataset, and all populations individually met the conditions for Hardy–Weinberg equilibrium at $p < 0.05$ (for details see Roden and Dutton 2011 and Dutton et al. 2013). For our male leatherback dataset, 139 individuals were genotyped with 15 marine turtle microsatellite markers. Of these, 122 individuals genotyped at a minimum success rate of 70% across 15 microsatellite loci to be considered for subsequent assignment analysis. The largest subset of males in this study were from Nova Scotia ($n = 76$) and were previously analyzed in Stewart et al. (2013). Three additional foraging live-caught male leatherbacks not included in Stewart et al. (2013) were included in this assignment study. In addition, 43 males were added from strandings, entanglements, and live captures throughout the Atlantic and Mediterranean to complete this dataset ($n = 122$).

The assignment testing (AT) program ONCOR (Kalinowski et al. 2007) was used to assign male turtles to source nesting beaches. We previously demonstrated the accuracy of this method for assigning female turtles to source nesting beaches based on genotypic data (Stewart et al. 2013), using data from tag returns and satellite tracks to confirm source populations. Within ONCOR, we used the genetic reference dataset as described above and designated the sampled male leatherbacks as the mixture population. We used the Individual Assignment option to assign each male turtle to one of nine baseline populations.

Results

Samples and genetic analysis We had 122 samples in our dataset. The average size of captured turtles (mean ± SD) was 150.7 ± 8.1 cm CCL ($n = 89$) and 110.2 ± 6.6 cm CCW ($n = 66$). The range of CCL measurements was 130.4–171.0 cm, while the range of CCW was 97.1–133.0 cm. The majority of these turtles would be considered adults, especially considering that all were classified in the field as males, and males do not exhibit sexual characteristics (long tail) until they are mature (Stewart et al. 2007).

Using ONCOR, we found that the majority of the 122 analyzed turtles—55% (67) were assigned to Trinidad, while 31% (38) were assigned to French Guiana and 14% (17) were assigned to Costa Rica (Online Resource Table 1). Of the 122 turtles, 22 had an assignment probability >80% to Trinidad, 15 to French Guiana, and 8 to Costa Rica. Turtles that came from both Trinidad and French Guiana had primary assignments that were >80% at a rate of 33 and 39%, respectively. In contrast, the rate of precision for Costa Rica was higher. For turtles assigned to Costa Rica, 47% were (8/17) assigned with >80% accuracy. For probabilities <80% (turtles assigned primarily to Trinidad or French Guiana), we often see the secondary assignment to be the opposite rookery (e.g., turtle ID 76346 assigned to Trinidad at 78.7% and secondarily to French Guiana at 21.2%; Online Resource Table 1). No turtles were assigned to the baseline populations of Brazil, Florida, St. Croix, South Africa, Gabon, or Ghana.

Twelve out of 122 male leatherbacks had satellite-tracking data to reference (James et al. 2005; Dodge at el. 2014; Table 1) and to compare and verify assignment probabilities. Of these, we considered ten assignments to be validated and considered correct. Seven male leatherback turtles were tracked from offshore Nova Scotia, Canada. According to the ONCOR assignment analysis, five were assigned with high probabilities (>70%) to Trinidad and French Guiana. The other two males foraging off Nova Scotia had primary assignments to Costa Rica at probabilities of 81.2 and 53.6%. The remaining five turtles were tracked from off the coast of Massachusetts, USA. They all were assigned primarily to Trinidad at probabilities ranging from 49.9 to 87.3% as assigned to that region. Of all assignments, two (Turtle ID 37400 and 91164; Table 1) did not match the tracking data.

Discussion

Our findings provide insights into the putative population origins of male leatherbacks, and we found that all turtles were assigned to three of the nine possible genetically characterized breeding populations of Trinidad, French

Table 1 Assignment study sample subset of 12 male leatherbacks with known satellite-tracking end point data paired with calculated primary and secondary genetic source population assignments and associated probabilities

Turtle ID	Capture type	Country	Place	Assign #1	Probability (%)	Assign #2	Probability (%)	Tracking location (PTT)	Source	References
37400	Foraging	Canada	Offshore Nova Scotia	Costa Rica	81.2	Trinidad	17.5	Bocas del Toro Peninsula, Panama	Mike James	Stewart et al. (2013)
37403	Foraging	Canada	Offshore Nova Scotia	Trinidad	71.7	Fr. Guiana	28.3	Trinidad	Mike James	Stewart et al. (2013)
77396	Foraging	Canada	Offshore Nova Scotia	Fr. Guiana	85.0	Trinidad	14.9	French Guiana and Suriname	Mike James	Stewart et al. (2013)
77403	Foraging	Canada	Offshore Nova Scotia	Fr. Guiana	82.4	Trinidad	17.3	Grenada, St. Vincent/St. Lucia	Mike James	Stewart et al. (2013)
77406	Foraging	Canada	Offshore Nova Scotia	Trinidad	88.2	Fr. Guiana	8.0	Trinidad/Grenada/St. Vincent	Mike James	Stewart et al. (2013)
91168	Foraging	Canada	Offshore Nova Scotia	Fr. Guiana	89.9	Trinidad	10.1	Grenada/Carriacou	Mike James	Stewart et al. (2013)
91164	Foraging	Canada	Offshore Nova Scotia	Costa Rica	53.6	Trinidad	43.9	Grenada/Carriacou	Mike James	Stewart et al. (2013)
79656	Foraging	USA	Offshore, MA	Trinidad	69.8	Fr. Guiana	22.1	Tobago Basin	Kara Dodge	Dodge et al. (2014)
79659	Foraging	USA	Offshore, MA	Trinidad	85.0	Fr. Guiana	15.0	Tobago Basin	Kara Dodge	Dodge et al. (2014)
79662	Foraging	USA	Offshore, MA	Trinidad	58.4	Fr. Guiana	41.5	Offshore Matura Beach, Trinidad	Kara Dodge	Dodge et al. (2014)
88972	Entanglement	USA	Nantucket Sound, MA	Trinidad	87.3	Fr. Guiana	6.8	Offshore Trinidad	Kara Dodge	Dodge et al. (2014)
156961	Foraging	USA	Offshore Nantucket, MA	Trinidad	49.9	Costa Rica	44.6	Gulf Stream/South Atlantic Bight	Kara Dodge	Unpublished data

Guiana, and Costa Rica. The probability precision for turtles belonging to the Trinidad and French Guiana nesting populations may reflect the relatively weak differentiation that exists between them (Dutton et al. 2013). Populations linked by high rates of dispersal or overlap will be genetically similar, and assignments to one or the other population may be equally likely. Although the two populations were found to be weakly, but significantly differentiated using microsatellites in a stock structure analysis (Dutton et al. 2013), the mtDNA markers used in that study were not sufficient to detect any differences between the two nesting sources. This is the most likely reason for the lower probability in the assignment tests for turtles assigned to those rookeries.

For turtles satellite tracked from the Northwest Atlantic, these foraging animals were tracked to French Guiana/Suriname, Panama, and Trinidad/Grenada. One male turtle was assigned to French Guiana at a certainty of 85% (turtle ID: 77396, Table 1 and Online Resource Table 1), which matched its satellite track to that region. Two turtles were assigned incorrectly to their natal regions according to the assignment testing vs. satellite tracking (turtle ID: 91164 and 37400, Table 1 and Online Resource Table 1). One individual (turtle ID 91164) was tracked to Grenada, but was assigned to Costa Rica with a 53.6% probability and had a secondary assignment to Trinidad. This finding raises the alternative hypothesis that adult males may not always return to their natal rookery. However, Grenada was not represented in the genetic baseline dataset, and more likely the reason for the conflict and low degree of assignment certainty. This turtle was adult sized (CCL = 150.6, CCW = 105.6), so if male turtles do return to natal beaches for breeding, we may assume this turtle was from Grenada but the analysis was forced to choose a source from the populations in our baseline dataset. The second turtle (turtle ID 37400) was tracked to Panama, but assigned to Costa Rica at a confidence probability of 81.2%. This higher degree of assignment certainty tells us that this turtle may in fact be from Panama, but due to proximity of the nesting beaches in Panama and Costa Rica, the two populations may be genetically similar. Like Grenada, our baseline dataset does not have nesting female samples from Panama, but a contiguous breeding population may exist there and be the reason why the turtle was assigned to this region. Dodge et al. (2014) found evidence of a shared breeding population through tag–recapture of an adult female nesting in both Panama and Costa Rica during a single breeding season. Additional evidence for nesting site interchange between Costa Rica and Panama was documented by Chacón-Chaverri and Eckert (2007) and Ordoñez et al. (2007). This finding emphasizes the need to strengthen our rookery genetic baseline dataset and sample missing populations for a more complete analysis. The other nine turtles with satellite tagging data were

tracked to the Trinidad/Tobago/Grenada/French Guiana region and also assigned to those populations.

Leatherbacks are occasionally found in the Mediterranean (Rees et al. 2004; Levy et al. 2005; Sönmez et al. 2008). Geldiay et al. (1995) had reported that leatherbacks were present in Turkish waters but were not often seen, while Groombridge (1990) suggested that leatherbacks regularly enter the Mediterranean Sea and are seen mainly in the westernmost portion. We had a sample from a male leatherback that was stranded in Turkey (turtle ID: 9796, Online Resource Table 1; Taskavak et al. 1998), which was confirmed as a subadult male during necropsy (CCL = 136 cm, CCW = 100 cm; weight = 200 kg). This turtle was assigned to French Guiana at a probability of 82%, with Trinidad as a secondary assignment. A female turtle that was stranded in Turkey in 2005 carried flipper tags that had been applied at Matura Beach, Trinidad (Sönmez et al. 2008). This fortuitous tag return along with our genetic assignment of the male leatherback that was stranded earlier provide evidence that leatherbacks from western Atlantic nesting populations do cross the Atlantic and enter the Mediterranean Sea.

All of the male turtles were assigned to one of three nesting populations (Trinidad, French Guiana, and Costa Rica), and there was a notable absence of turtles from the African populations (Gabon, Ghana and South Africa). It is somewhat surprising that we did not find any turtles from other nesting areas in the western Atlantic including Florida, St. Croix, or Brazil. If one assumes that turtles are randomly mixing in foraging areas in relative proportions to source nesting populations, we should have seen some contributions from the smaller rookeries. Based on the following estimated female nesting population sizes for the western Atlantic (Brazil: 150, Costa Rica: 2000, Florida: 750, French Guiana: 3200, Trinidad: 9500, and St. Croix: 750) and assuming that there are at least as many males in each population as there are females (estimates from Stewart and Dutton 2014), then we should have seen male turtles assigned to nesting populations in the following numbers: Brazil: 1, Costa Rica: 15, Florida: 5, French Guiana: 24, Trinidad: 71, and St. Croix: 6. Instead, we found a greater number than expected from Costa Rica (17 vs. 15), almost double the number that would be expected from French Guiana (38 vs. 24) and less from Trinidad than expected (67 vs. 71). Therefore, the population distribution of the assigned turtles was significantly different than what we would have expected had the turtles been distributed in proportion to population size ($\chi^2 = 20.659$, $p < 0.05$). There are several plausible reasons for this result: (1) leatherbacks may not randomly distribute themselves in foraging areas in proportion to their nesting population sizes, a conclusion reached by Stewart et al. (2016) based on leatherback bycatch samples collected in the pelagic longline fisheries of the USA, (2) adult sex ratios within nesting populations may not be 1:1 and perhaps some populations have more breeding males than other populations, (3) there may be some mechanism of sex-biased dispersal to preferred foraging areas, and (4) sampling bias in our collection; because there are large populations that contribute animals to the foraging population, we may have by chance sampled those populations, missing the fewer turtles that belonged to the smaller populations. Even though James et al. (2007) found an in-water female to male ratio of 1.86:1 while Dodge et al. (2011, 2014) and Innis et al. (2014) found more males in foraging populations off New England and Florida, at a ratio of 1.3 males:1 female (12 males, 9 females, 8 unknown sex), we would expect to find males from all populations, even just one or two from the smaller populations. Out of 288 foraging leatherbacks from Canada, there were 177 females and 83 males (28 unknown sex; Stewart et al. 2013), and for both males and females, small populations were less represented than large populations.

Stewart and Dutton (2014) determined an approximate 50:50 sex ratio in the St. Croix population that mated in 2010. This apparent lack of males assigned to Florida and St. Croix warrants further investigation into adult male numbers (determined through breeding sex ratio evaluation) to determine whether males are under-represented in these breeding populations. Sex-biased dispersal to different foraging grounds might explain the absence of males from the smaller nesting populations in our results. In a study looking at longline bycatch from the Atlantic, Stewart et al. (2016) found that there were differences in the distributions of turtles from nesting grounds in three main foraging areas in the Atlantic; more turtles from the Costa Rica population were found in the Gulf of Mexico than in other areas and although the turtles were not parsed out by sex for that study (sex information was not available), this may be an indication that there is some differential distribution of turtles to foraging grounds from different nesting aggregations. We found that no male turtles were assigned to the African rookeries and this follows the pattern of what we know about the dispersal patterns of those turtles: they forage in the southwest Atlantic, off Uruguay and Brazil (Billes et al. 2006; Witt et al. 2011; Dutton et al. 2013; Fossette et al. 2014; Prosdoscimi et al. 2014). Additionally, the discovery of leatherbacks in the Mediterranean from western Atlantic rookeries (Trinidad and French Guiana) fits the model of a north–south split for leatherback foraging; turtles in the northern hemisphere forage in the north, while those in the southern hemisphere forage south (Witt et al. 2011; Prosdoscimi et al. 2014).

Male marine turtles are critical components of populations. Their study is of particular interest due to concern over climate change. Climate change may pose challenges for sea turtles, which exhibit temperature-dependent sex determination (TSD). Incubation temperature during development determines the sex of hatchlings; generally, higher temperatures yield more females while lower temperatures

produce more males with a pivotal temperature of approximately 29 °C (Davenport 1997). This potentially puts sea turtles at risk from global climate change. Increases in nest temperatures, due to warmer beaches, may bias development in favor of the production of females throughout a nesting season (Davenport 1997; Fuentes et al. 2010; Laloe et al. 2014), with some studies predicting complete feminization if global temperatures continue to rise (Mrosovsky et al. 1984). This feminization could ultimately lead to the decline of populations if there are not enough males being produced to contribute to the breeding population. This potential threat reinforces the need for the study and protection of male sea turtles.

This is a first look at determining the source populations for male leatherbacks throughout the Atlantic and we recommend that all stranding and bycatch sampling protocols strive to record both the sex and size of the sampled animals. This additional information adds value for determining natal populations as well as informing population dynamics analyses. Using even more informative markers may give higher resolution and certainty to the probabilities of assignment, particularly for cases where nesting populations are in close proximity or are only mildly differentiated. In addition, analyzing more samples from areas where there are gaps (along the southeastern seaboard and Gulf of Mexico) would be helpful in expanding the geographic coverage to help answer the question of rookery-biased dispersal to foraging areas. The results from our study advance sea turtle conservation and population assessments by allowing the inclusion of information on males in foraging areas by determining which nesting populations they belong to.

Understanding how turtles move between nesting beaches and foraging areas, as well as how they mix on the foraging grounds, is important for assessing how threats may affect populations in different ways. This information would also be helpful in developing responses or mitigation plans to lessen these threats. If males do frequent different foraging grounds and in differing proportions than the females (possibly due to sex-biased dispersal), it will be important to understand the population-specific threats to males. With concerns about climate change potentially reducing the male component of all breeding populations, it is important to understand baseline distributions and dispersal, population size and threats on foraging grounds or migratory pathways, specifically for male turtles.

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Compliance with ethical standards

Ethical standards All applicable international, national, and/or institutional guidelines for the care and use of animals were followed.

Conflict of interest The authors declare that they have no conflict of interest.

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