Identification of young-of-the-year great hammerhead shark Sphyrna mokarran in northern Florida and South Carolina

A. M. Barker, B. S. Frazier, D. M. Bethea, J. R. Gold

and D. S. Portnoy

SEDAR77-RD13

Received: 6/18/2021

Journal of FISH BIOLOGY

fsbi An International Society for Fish Biology

Journal of Fish Biology (2017)
doi:10.1111/jfb.13356, available online at wileyonlinelibrary.com

BRIEF COMMUNICATION

Identification of young-of-the-year great hammerhead shark *Sphyrna mokarran* in northern Florida and South Carolina

A. M. Barker*†, B. S. Frazier‡, D. M. Bethea§, J. R. Gold*
and
D. S. Portnoy*

**Marine Genomics Laboratory, Department of Life Sciences, Texas A&M University–Corpus Christi, 6300 Ocean Dr, Corpus Christi, TX 78412, U.S.A., ‡South Carolina Department of Natural Resources, Marine Resources Research Institute, 217 Ft Johnson Rd, Charleston, SC, 29412, U.S.A. and §NOAA NMFS, Southeast Regional Office, 263 13th Avenue South, St. Petersburg, FL 33701, U.S.A.*

(Received 3 December 2016, Accepted 18 May 2017)

Two sharks, visually identified in the field as young-of-the-year (YOY) scalloped hammerhead *Sphyrna lewini*, were identified as great hammerhead *Sphyrna mokarran* based on nuclear-encoded single nucleotide polymorphisms (SNP) and sequences of mtDNA. Individuals were captured and released in Bulls Bay, SC, and Saint Joseph Bay, FL, in 2013 and 2014, respectively. These findings indicate *S. mokarran* may be pupping in or around these areas and highlight new regions that may be a productive focus for future research on early life history of *S. mokarran*.

Key words: essential fish habitat; molecular identification; morphologically conserved species; shark nursery; *Sphyrna*.

Very little is known about the early life history of the great hammerhead, *Sphyrna mokarran* (Rüppell 1837). Locations of nursery grounds are not well defined and identification of these areas is of importance for management of the resource and conservation of the species (Miller *et al.*, 2014). Large coastal sharks that give birth to live neonates of small size (<70 cm) are expected to utilize discrete nurseries (Branstetter, 1990). The size at birth of *S. mokarran* (50–70 cm (Compagno, 1984) suggests that nursery use would be beneficial to neonates; pupping of *S. mokarran*, however, is thought to occur primarily in offshore waters (Hueter & Tyminski, 2007; Harry *et al.*, 2011). Young-of-the-year (YOY) *S. mokarran* have been observed using nearshore nurseries off the Gulf of Mexico coast of Florida as far north as Yankeetown (29·004467° N; 82·815062° W; Hueter & Tyminski, 2007). Young-of-the-year and juvenile *S. mokarran* < 200 cm total length (L_T) are not known to occur in coastal waters on the east coast of the U.S.A. (Castro, 2011).

†Author to whom correspondence should be addressed. Tel.: +1 760 532 2237; email: abarker@islander.tamucc.edu

To investigate nursery habitat usage of two sympatrically distributed sphyrnids, the scalloped hammerhead *Sphyrna lewini* (Griffith & Smith 1834) and the recently discovered Carolina hammerhead *Sphyrna gilberti* Quattro, Driggers III, Grady, Ulrich & Roberts 2013, double-digest restriction associated DNA sequencing (ddRAD) was used to identify a panel of single nucleotide polymorphisms (SNP) that can be used to differentiate between the species. Because *S. lewini* and *S. gilberti* are conserved morphologically and differ only in the number of precaudal vertebrae (Quattro *et al.*, 2013), the panel allows for conclusive, non–lethal species identification.

Fin clips were collected from eighteen putative YOY *S. lewini* spread across four sites: Corpus Christi, TX (27·689378° N; 97·055843° W), Panama City, FL (29·7326667° N; 85·3691° W), Cape Canaveral, FL (28·389406° N; 80·586626° W), and Bulls Bay, SC (33·009500° N; 79·485346° W). Genomic DNA was extracted using a Mag–Bind Blood & Tissue DNA Kit (Omega Bio-Tek; www.omeganiotek.com). Double-digest restriction associated DNA sequencing (ddRAD) library preparation was conducted following a modified version of Peterson *et al.* (2012; Table S1, Supporting information). The library was sequenced as a paired–end run on one lane of a MiSeq DNA sequencer (Illumina; www.illumina.com). The dDocent pipeline (www.ddocent.com; Puritz *et al.*, 2014) was used for reference construction, mapping reads and SNP calling. A total of 39 011 SNPs were recovered from 4584 contigs (Table S2, Supporting information).

As an initial means of grouping individuals, PCA was run in Adegenet (Jombart, 2008) and three distinct genetic clusters were recovered (Fig. 1 and Table S3, Supporting information). The clusters were highly divergent across all loci ($F_{ST} = 0{\cdot}9–0{\cdot}98$). A total of 846 bp from the mitochondrial control region (mtCR) were sequenced from two to three individuals from each cluster to determine species identity (Table S4, Supporting information). Sequences were compared with haplotypes available on GenBank and three individuals were identified with 99% sequence identity as *S. lewini*, three with 99–100% sequence identity as *S. gilberti and* two with 100% sequence identity as *S. mokarran* (GenBank accession nos. KY315826–KY315830). The first individual identified as *S. mokarran* was captured in Bulls Bay, SC, on 9 July 2013 and the second in St. Joseph Bay, FL, on 5 August 2014 (Fig. 1). Total length was measured at 63·8 cm for the individual captured in SC and 67·0 cm for the individual captured in FL; both fell within the observed size range for neonate *S. mokarran* (Compagno, 1984). A neighbour-joining tree was created from mtCR data with MEGA7 (Kumar *et al.*, 2016) using a Jukes–Cantor substitution model with 500 bootstrap replicates (Table S5, Supporting information). Three groups were recovered with 100% support and were consistent with clusters identified using SNPs in the PCA (Fig. 2). Mean nucleotide divergence between the group identified as *S. mokarran* and other groups was *c.* 16% and mean divergence between *S. lewini* and *S. gilberti* was *c.* 5%. Within-group distances were negligible (0–0·1%; Table I).

Sphyrna mokarran is primarily a tropical species hypothesized to give birth offshore (Harry *et al.*, 2011; Hueter & Tyminski, 2007). The observation of two S. *mokarran* neonates in nearshore habitat of South Carolina and the northern Gulf of Mexico coast of Florida indicates that *S. mokarran* may use nursery habitat further north and further inshore than known previously. Little is known about the early life history of the species and, like other hammerhead sharks, *S. mokarran* is susceptible to over-exploitation (Denham *et al.*, 2007), making identification of essential fish habitat,

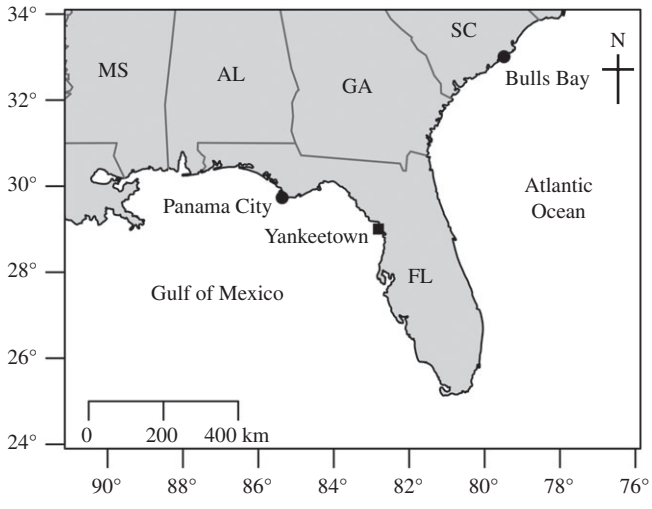

FIG. 1. Map indicating locations of *Spyrna mokarran* neonates identified in the present study (●), and location of previously known northernmost occurrence of *S. mokarran* neonates in the Gulf of Mexico (■).

such as nursery areas, a critical research topic. Given present data, it is not possible to characterize how important these two northern, inshore sites are to *S. mokarran.* Three scenarios may account for the presence of *S. mokarran* in these nurseries. First, it is possible that individuals were pupped elsewhere and subsequently moved into Bulls Bay and St. Joseph Bay after parturition. Given the size of the individuals, however, it is unlikely that they migrated a substantial distance. The capture date of the neonate in Bulls Bay occurred during the time of proposed parturition (Piercy *et al.*, 2010), meaning that the individual probably was born in close proximity to

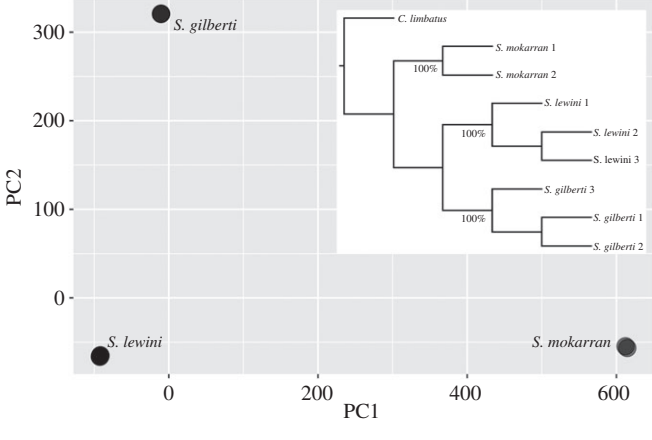

FIG. 2. Results of principal components (PC) analysis, using *Sphyrna* spp. single nucleotide polymorphism data, and a neighbour-joining tree constructed from mitochondrial control region data (mtCR). Both analyses identified three clusters that coincide with identification of three *Sphyrna* species using mtCR basic local-alignment search tool (BLAST) results. *Carcharhinus limbatus* was used as the out group.

TABLE I. Mean between and within-group nucleotide divergence among *Sphyrna* spp., based on mtCR sequences

	S. gilberti	*S. lewini*	*S. mokarran*
S. gilberti	0·001	0·049	0·165
S. lewini	0·049	0·000	0·159
S. mokarran	0·165	0·159	0·000

Bulls Bay. Second, these findings may indicate relatively new nursery habitat usage by *S. mokarran* due to a northward, coastal expansion in nursery usage. Third, several diagnostic features of *S. mokarran* (falcate pelvic fins and nearly straight anterior margin of the cephalofoil) are not as apparent in neonates, causing them to appear relatively similar to neonate *S. lewini* (Castro, 2011). It is possible that neonate *S. mokarran* have been caught in these areas previously but misidentified as *S. lewini*. Such misidentifications are common between morphologically conserved species, especially when one species is expected in a given region or habitat while the other is not (Branstetter, 1982; Tillett *et al.*, 2012). Other potential nursery sites for *S. mokarran* may not yet have been described, in part because of misidentification. Future work is needed to document how frequently neonate *S. mokarran* is encountered in these areas and to estimate the number of breeding females utilizing each site.

We thank M. Ajemian, G. Stunz and D. Adams for sample collection, A. Shaw and E. Vinyard, K. L. Smith, G. A. Casselberry and the interns of the 2014 NOAA Fisheries Panama City Laboratory Shark Population Group Assessment Group for assistance in the field. We also thank C. Hollenbeck, S. O'Leary, S. Willis and J. Puritz for assistance with laboratory work and data analysis and J. Carlson for helpful comments that improved the manuscript. Animals from the GULFSPAN Survey were collected under protocols outlined in Florida Fish and Wildlife Conservation Commission Special Activities License number SAL-14-1292-SRP. Collection of specimens collected in South Carolina was performed under SCDNR Scientific Permit #2212 and was granted an IACUC exemption. Work was funded by U.S. Fish and Wildlife Service Competitive State Wildlife Grant number SC-U2-F15AP00050 and NOAA/NMFS Cooperative Research Program Grant number NA14NMF4540063. This article is publication number 17 of the Marine Genomics Laboratory at Texas A&M University–Corpus Christi, number 111 in the series Genetic Studies in Marine Fishes and contribution 763 of the South Carolina Marine Resources Center.

Supporting Information

Supporting Information may be found in the online version of this paper:

Table S1. Method for double–digest restriction associated DNA sequencing (ddRAD) library preparation and data filtering.

Table S2. Single nucleotide polymorphisms recovered from *Sphyrna* spp.

Table S3. Genepop file used to identify genetic clusters.

Table S4. Mitochondrial control region (mtCR) sequences from two to three individuals from each cluster to determine species identity.

Table S5. Mitochondrial control region (mtCR) sequence alignments used to create neighbour-joining tree.

References

Branstetter, S. (1982). Problems associated with the identification and separation of the spinner shark, *Carcharhinus brevipinna* and the blacktip shark, *Carcharhinus limbatus*. *Copeia* **1982,** 461–465.

Castro, J. I. (2011). *The sharks of North America*. New York, NY: Oxford University Press.

Compagno, L. (1984). FAO Species Catalogue. Vol. 4. Sharks of the world. An annotated and illustrated catalogue of shark species known to date. Part 2 – Carcharhiniformes. *FAO Fisheries Synopsis* **125**, 251–655. Rome: FAO.

Harry, A. V., Macbeth, W. G., Gutteridge, A. N. & Simpfendorfer, C. A. (2011). The life histories of endangered hammerhead sharks (Carcharhiniformes, Sphyrnidae) from the east coast of Australia. *Journal of Fish Biology* **78,** 2026–2051.

Hueter, R. E. & Tyminski, J. P. (2007). Species-specific distribution and habitat characteristics of shark nurseries in Gulf of Mexico eaters off Peninsular Florida and Texas. *American Fisheries Society Symposium* **50**, 193–223. Bethesda, MD: American Fisheries Society.

Jombart, T. (2008). Adegenet: a R package for the multivariate analysis of genetic markers. *Bioinformatics* **24,** 1403–1405.

Kumar, S., Stecher, G. & Tamura, K. (2016). MEGA7: Molecular Evolutionary Genetics Analysis Version 7·0 for bigger datasets. *Molecular Biology and Evolution* **33,** 1870–1874.

Peterson, B. K., Weber, J. N., Kay, E. H., Fisher, H. S. & Hoekstra, H. E. (2012). Double digest RADseq: an inexpensive method for *de novo* SNP discovery and genotyping in model and non-model species. *PLoS ONE* **7,** e37135.

Piercy, A. N., Carlson, J. K. & Passerotti, M. S. (2010). Age and growth of the great hammerhead shark, *Sphyrna mokarran*, in the north-western Atlantic Ocean and Gulf of Mexico. *Marine and Freshwater Research* **61,** 992–998.

Puritz, J. B., Hollenbeck, C. M. & Gold, J. R. (2014). dDocent: a RADseq, variant-calling pipeline designed for population genomics of non-model organisms. *PeerJ* **2,** e431.

Quattro, J. M., Driggers, W. B. I., Grady, J. M., Ulrich, G. F. & Roberts, M. A. (2013). *Sphyrna gilberti* sp. nov., a new hammerhead shark (Carcharhiniformes, Sphyrnidae) from the western Atlantic Ocean. *Zootaxa* **3702,** 159–178.

Tillett, B. J., Field, I. C., Bradshaw, C. & Johnson, G. (2012). Accuracy of species identification by fisheries observers in a north Australian shark fishery. *Fisheries* **127–128**, 109–115.

Electronic References

Branstetter, S. (1990). Early life-history implications of selected carcharhinoid and lamnoid sharks of the northwest Atlantic. *NOAA Technical Report NMFS* **90**, 17–28. Washington, D.C.: U.S. department of Commerce. Available at http://spo.nmfs.noaa.gov/tr90opt.pdf

Denham, J., Stevens, J., Simpfendorfer, C. A., Heupel, M. R., Cliff, G., Morgan, A., Graham, R., Ducrocq, M., Dulvy, N. D., Seisay, M., Asber, M., Valenti, S. V., Litvinov, F., Martins, P., Lemine Ould Sidi, M., Tous, P. & Bucal, D. (2007). *Sphyrna mokarran*. IUCN Red List of Threatened Species. Available at https://doi.org/10·2305/IUCN.UK.2007.RLTS.T39386A10191938.en (last accessed 13 September 2016).

Miller, M. H., Carlson, J., Hogan, L. & Kobayashi, D. (2014). *Status review report: great hammerhead shark (*Sphyrna mokarran*)*. Washington, D.C.: NOAA, U.S. department of Commerce. Available at http://www.cio.noaa.gov/services_programs/prplans/pdfs/ID266_Final_Great_Hammerhead_Shark_Status_Review_Report.pdf

