**Supplemental Methods**

*DdRAD Library Preparation*

Extracted genomic DNA was digested with the enzymes *EcoRI* and *SphI*. Reactions were purified using AMPure XP (Agencourt) following complete digestion and adapter oligonucleotides (P1 and P2) were ligated onto fragments. The P1 adapter contains one of 48 possible barcode sequences and the P2 adapter contains one of twelve possible index sequences, allowing individuals to be pooled and sequenced simultaneously. A PCR test was conducted to ensure successful ligation and individuals were pooled and loaded into a Pippin Prep (Sage Science) gel cassette for selection of fragments between 313-437 base pairs in length. Illumina flow cell annealing sequences were added with 14 cycles of PCR. A total of five libraries were prepared and sequenced as paired-end runs on an Illumina HiSeq 4000 DNA sequencer.

*Reference Construction*

 Twenty individuals (17 scalloped hammerheads, 3 Carolina hammerheads, 2 great hammerheads) previously identified based on mtCR haplotypes were sequenced as a paired-end run on an Illumina MiSeq sequencer. The library was prepared following the same method as previously described. The dDocent pipeline [1] was used to assemble a *de novo* reference with the clustering similarity value (c) set to 0.9 (default). Reads were required to have at least 3X coverage (K1 parameter) in at least 2 individuals (K2 parameter) to be included in the reference. To determine the optimal values for K1 and K2, the script *RefMapOpt.sh* (https://github.com/jpuritz/dDocent/blob/master/scripts/RefMapOpt.sh) was used. Given a value of c, the script creates references based on a range of K1 and K2 values and maps sequences back to each reference. The optimal values were determined as those that maximized the total number of mapped reads while minimizing the number of read pairs mapped to different contigs. The final reference contained 39,955 contigs.

*Diagnostic SNPs and Species Identification*

The twenty individuals used to construct the *de novo* reference were screened for single nucleotide polymorphisms (SNPs) that could be used to distinguish scalloped, Carolina and great hammerheads. DDocent was used to map reads and call SNPs, and raw variants were filtered using VCFTools [2] for a minimum quality score of 20, minimum mean depth of 10, 0% missing data, and indels were removed. SNPs were thinned to retain only one SNP per contig. Two sets of diagnostics SNPs were identified, one set to distinguish great hammerheads from scalloped and Carolina hammerheads, and a second set to distinguish scalloped hammerheads from Carolina hammerheads. To identify the first set, allele frequencies were calculated and SNPs that were fixed between great hammerheads and scalloped and Carolina hammerheads (grouped together) were selected. To identify the second set, great hammerheads were removed from the dataset and allele frequencies were recalculated to identify SNPs fixed between scalloped and Carolina hammerheads. The first panel contained 2,695 SNPs, and the second panel contained 1,491.

To identify unknown individuals, dDocent was used to map reads and call SNPs. Raw variants were filtered to only retain diagnostic SNPs using VCFTools. First, individuals were identified as either a scalloped/Carolina hammerhead or great hammerhead using a custom python script to compare genotypes to the first diagnostic panel. Individuals identified as great hammerheads were then removed from the dataset, and the remaining individuals were identified as scalloped or Carolina hammerheads with another python script and the second diagnostic panel. A match of 95% to one species was required for positive species identification.

*Data Filtering*

DDocent was used to map reads and call SNPs for 600 individuals sequenced on an Illumina HiSeq platform. Raw variants were filtered using VCFTools [2]. Individuals with greater than 25% missing data were removed from the dataset. Sites with a sequence quality score less than 20 and genotypes with a quality score less than 30 were removed. Loci were filtered for a genotype call rate of 0.90, minimum allele count of 3, a minimum depth of 5, a mean minimum depth of 15, and maximum depth of 250. Indels were removed from the dataset. Sites were also filtered for mapping quality ratio, quality to depth ratio, allele balance, strand bias, and properly paired status. Finally, loci with more than 15% missing data within any particular sequencing library were removed from analysis, as well as loci with more than 10% missing data within species groups (scalloped hammerhead, Carolina hammerhead, undetermined/hybrid) as determined by the panel of diagnostic SNPs used to identify species. As a quality control measure to identify library effects across separate sequencing runs, 14 individuals were sequenced twice, and replicate individuals were removed prior to further analysis if they were not already filtered in a previous step. After filtering, 7,199 SNPs on 2,512 contigs remained in the dataset. SNPs were thinned to retain one SNP per contig resulting in a final dataset of 2,512 loci and 554 individuals. Mean read depth was 97.00 per individual and 97.24 per locus. Mean missing data was 2.15% per individual and 1.28% per locus.

*Mitochondrial DNA Analysis*

To determine the maternal lineage of hybrids, a 683-base pair region of the mitochondrial control region (mtCR) was sequenced for seven F1 hybrids, eleven scalloped hammerhead backcrosses and one Carolina hammerhead backcross, based on assignments from NewHybrids, with the primers Pro-Shark (5- GCC CTT GGC TCC CAA AGC -3’) and Phe-Shark (5’- TCA TCT TAG CAT CTT CAG TGC CA -3’). Only a subset of hybrids was sequenced because all DNA for some individuals had been used for ddRAD sequencing and no tissue remained to re-extract DNA. PCR amplification was performed in 25 μl reactions containing approximately 50-100 ng template DNA, 1x GoTaq Flexi buffer, 0.2 mM of each dNTP, 1.5 mM MgCl2, 0.25 μM of each primer, and 0.5 units of Taq Flexi DNA polymerase. Amplification was performed using a touchdown PCR protocol that consisted of an initial denaturation for 2 min at 95°C, followed by 40 cycles of 95°C for 1 min, 60°C for 1 min, and 72°C for 1.5 min, with the annealing temperature reduced by 0.5°C each cycle until reaching 55°C. This was followed by a final extension for 10 min at 72°C. The PCR product was visualized on a gel to ensure successful amplification and cleaned with AmpureXP (Agencourt) prior to bidirectional sequencing at Texas A&M University- Corpus Christi Genomics Core Laboratory (http://genomics.tamucc.edu). Sequences were trimmed and edited using Geneious version 7.1.9 [3] and compared to haplotypes available on GenBank.

**References**

1. 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. (doi:10.7717/peerj.431/table-1)

2. Danecek, P. et al. 2011 The variant call format and VCFtools. *Bioinformatics* **27**, 2156–2158. (doi:10.1093/bioinformatics/btr330)

3. Kearse, M. et al. 2012 Geneious Basic: An integrated and extendable desktop software platform for the organization and analysis of sequence data. *Bioinformatics* **28**, 1647–1649. (doi:10.1093/bioinformatics/bts199)