

Retrospective Genomics Suggests the Disappearance of a Tiger Shark (*Galeocerdo Cuvier*) Population Off South-Eastern Australia

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Research Article

Keywords: bait capture, micro-evolutionary processes, population structure, temporal trend, historical DNA, elasmobranchs

Posted Date: March 22nd, 2021

DOI: <https://doi.org/10.21203/rs.3.rs-334053/v1>

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Abstract

Over the last century, many populations of sharks have been reduced in numbers by overexploitation or attempts to mitigate human-shark interactions. Still, there is a general perception that populations of large ocean predators cover wide areas and therefore their diversity is less susceptible to local anthropogenic disturbance. Here we report retrospective genomic analyses of DNA using archived and contemporary samples of tiger shark (*Galeocerdo cuvier*) from eastern Australia. Using SNP loci, we documented a significant overall change in genetic composition of tiger sharks born over the last century. The change was most likely due to a shift over time in the relative contribution of two well differentiated, but hitherto cryptic populations. Our data strongly indicate a dramatic shift in relative contribution of the two populations to the overall tiger shark abundance of the east coast of Australia, possibly associated with differences in direct or indirect exploitation rates.

Introduction

Intraspecific genetic diversity is essential for long-term population persistence to avoid the negative effects of inbreeding¹, to buffer against environmental variation in time and space², and to ensure adaptability to a changing environment³. High genetic diversity may also have positive ecosystem effects by promoting productivity, abundance, and stability of community structure^{4–6}. Intraspecific diversity protection is also a specified objective of the Convention on Biological Diversity (CBD; www.cbd.int), but is rarely an integral part of monitoring activities, in particular for species that are not critically endangered^{7,8}. Indeed, the lack of dedicated sampling programs and the many species requiring monitoring makes it logically impossible to oversee intraspecific diversity for most species. In addition, the difficulties connected to the use of high-resolution genetic methods when sample sizes are low or when tissue samples vary in age, composition and provenance⁹ also hinder our ability to assess intraspecific diversity.

The use of DNA extracted from museum specimens, in combination with modern molecular analytical tools, has revolutionized the ability to assess genetic changes at contemporary time scales^{10,11}. Studies on processes such as intra-population loss of diversity, adaptive change caused by evolutionary drivers in the environment, as well as the movement, decline, or extirpation of populations through time and space can now be undertaken^{5,12}. In the marine realm, there are numerous examples of local population reductions and extinctions, in particular for large sharks and rays^{13,14}, but closely coupling these incidences with genetic and genomic data is challenging due to the lack of temporal genetic data for elasmobranch species. Temporal genetic data can be used to test the stability of patterns of population structure, levels of genetic variability, and potential adaptive genetic changes in the timeframe of recent environmental change or exploitation. This is of particular interest in respect to populations of large sharks, where there is a strong need for identification of genetic populations for both conservation and management^{9,15}, as they represent both the relevant unit for evolution and management in order to ensure sustainable exploitation and long-term protection of biodiversity.

The tiger shark (*Galeocerdo cuvier*) is one of the world's largest sharks, with a circumglobal distribution in tropical and sub-tropical waters¹⁶. Satellite-tag tracking studies have revealed extensive movements of several thousand kilometres both within the Atlantic¹⁷ and Indo-Pacific¹⁸ and, possibly, across the Indian Ocean¹⁹. This apparent connectivity within basins implies that genetic structuring over these distances may be limited. This has been corroborated by microsatellite genetic studies that show a general lack of intra-basin structuring, but a clear inter-basin genetic split between the Atlantic and Indo-Pacific Oceans^{20–22}. Accordingly, tiger sharks in Australia are thought to be part of a large Indo-Pacific population. However, population structure of tiger sharks at a local scale remains largely unknown. On the Australian east coast they are found from tropical Queensland to temperate Victoria^{23,24} with some satellite-tagged individuals seen to move as far as New Caledonia and Papua New Guinea^{24,25}. Throughout this distribution, the species is exploited through various fishing operations, from target and bycatch in commercial, artisanal, and recreational fisheries^{26,27}, to shark control operations to improve bather safety^{28,29}, with clear indications of population decline and reduced average size of individuals^{29–31}. Tiger sharks are currently listed as Near Threatened on the International Union for the Conservation of Nature's Red List of Threatened Species IUCN (IUCN) due to a suspected decline by ~30% over the past ~60 years from exploitation in commercial, recreational, and unregulated fisheries, as well as shark control programs³². However, further regional depletions have been suggested. For example, tiger sharks in the Arabian Seas region have been assessed as Vulnerable based on a suspected decline of at least 30% over the past three generations³³. Off Queensland (Australia), tiger shark catch-per-unit-effort has dropped by 74% over the past 25 years, while the average size declined by 21%³¹. Due to its threatened status, potential for over-exploitation and risk to human lives, the tiger shark, like other large sharks, has a high profile in public awareness, and is subject to much debate over its management and conservation actions³⁴.

Here, we investigated a possible link between this apparent decline in tiger shark numbers and its genetic diversity off the east coast of Australia. In order to collect genetic data over the widest spatial and temporal range we extracted DNA from tissue samples taken from shark jaws archived in museums and private collections, or retained as trophies from fishing competitions, and performed retrospective genomic analyses³⁵. We expected to find a single panmictic population in eastern Australia as previously reported²⁰, but we were alert to the possibility of significant changes to the population over the last century as a consequence of documented population reductions.

Results

Bioinformatics pipeline and data filtering

Sequencing yielded an average of 2,833,138 reads per individual, with slightly lower numbers for historical jaw samples (median 1,973,493) than contemporary tissue samples (median 2,301,001). After running the bioinformatics pipeline, 78% of the original sequences went into transcriptome mapping. All samples showed a very low percentage of contaminants, confirming the validity of the capture strategy

for sequencing enrichment with shark DNA. An average of 0.12% of the cleaned reads was of mitochondrial origin and excluded from further analysis, except for the few cases when it was used for species confirmation. Out of the 20,000 catshark derived baits, 4,544 (22.72%) were post-hoc mapped back to the tiger shark transcriptome³⁶ covering 4,137 scaffolds. Of these, 4,143 had captured reads with a depth of coverage higher than non-target regions. Scaffolds with a bait had an average coverage of 68.5X, while scaffolds from off-target regions had an overall average depth of 40.7X (35.8X for historical and 43.4X for contemporary samples). Coverage was higher and less variable in contemporary than in historical samples. Single nucleotide polymorphisms (SNPs) were called from all transcriptomic sequences and we identified 35,061 raw SNP variants for 122 samples in 2,978 reference scaffolds. After filtering, 4,580 SNPs remained. SNPs with significant departures from Hardy-Weinberg Equilibrium (HWE) were removed to produce a final dataset consisting of 1,840 SNP loci genotyped in 115 samples from *G. cuvier* specimens caught between 1939 and 2015. In addition, four samples were removed due to high levels of missing data (below 80% call rate) and four other samples were excluded due to either mislabelling of the samples (identified as a different species), or to a possible contamination with reads from other shark species. The final dataset thus contained 107 samples (Fig. 1, Table S1). Very low levels of DNA damage was observed, confirming DNA was well preserved in jaws³⁵, at least for the relatively short time period (~80 years) compared to true ancient DNA (aDNA) studies. Thus, it is highly unlikely that the final SNPs represent artefacts due to deamination or other DNA damage in the historical jaw samples.

Data analysis for spatial and temporal genomic variability

To examine the stability of patterns over time, we used a temporal genetic analysis based on the back-calculated decade of birth of tiger shark individuals which showed clear evidence of temporal genetic differentiation. This was apparent for the temporal dataset as a whole (Table 1), where pairwise F_{ST} estimates increased with time and with the largest significant break occurring between the oldest samples in comparison to the 1990 and 2000 groups. Temporal differentiation was also apparent for the Tasman Sea samples alone, where the majority of the oldest historical samples originated (Table S2). In contrast, contemporary tiger shark samples (2000 and 2010) from the Gulf of Carpentaria (GCA), Coral Sea (CRS) and Tasman Sea (TAS), showed little evidence of genetic structuring, with estimated non-significant pairwise F_{ST} values of 0.0000 between GCA and CRS, and 0.0012 between CRS and TAS (Table S3). A Principal Coordinates Analysis (PCoA) of spatiotemporal F_{ST} values (Fig. 2) showed a clear temporal separation of samples along axis 1, explaining more than 65% of the variation. The 1910–1960 TAS samples were the most distinct, with the 1970–1980 TAS samples intermediate between the TAS oldest samples and a cluster of the remaining non-differentiated samples. The non-differentiated group included all contemporary samples and historical samples from the GCA (1970–1980). The Principal Component Analysis (PCA) of all spatiotemporally collected individuals (Fig. 3a) supported the genetic differentiation of historical samples as they clustered differently and a proportion of the individuals formed a relatively distinct cluster from the contemporary samples. In contrast to Fig. 3a, an individual based PCA of spatially-collected contemporary samples only (Fig. 3b) did not display any apparent

clustering of individuals according to location. A distinct group of individuals in the historical samples was also supported by the hypothesized scenario of two populations (K=2) evaluated using a Discriminant Analysis of Principal Components (DAPC) (Fig. 4). The analysis showed 60% (12 of 20) of 'cluster 2' individuals in the 1910 – 1960 TAS samples, 38% (5 of 13) in the 1970–1980 samples and only one (1 of 25) in TAS samples from 1990 and 2000. For the GCA samples, only a single 'cluster 2' individual (1 of 23) was found (1990). Likewise, only one 'cluster 2' individual (1 of 16) was found among the CRS 2000 samples. When pooling all putative 'cluster 1' and 'cluster 2' individuals across samples, the resulting mean F_{ST} between the two population groups was 0.015, thus substantially higher than between any pair of spatiotemporal samples, supporting a mixed populations hypothesis. The distribution of F_{ST} values across loci showed that a high number of loci (Fig. S1) contributed to the differentiation, suggesting that genetic differences between the two groups were not caused by technical artefacts or contemporary evolution at one or a few loci. However, while bayescan did not detect any outliers, pcadapt identified 39 possible outlier loci between the two clusters. Most of these loci (34 of 39) showed an overall higher allele frequency for 'cluster 2'. The mean level of heterozygosity in the two groups was noticeably different (Fig. 5), with 0.23 (± 0.0097) for 'cluster 1' and 0.26 (± 0.0104) for 'cluster 2'. Average missing data was lower for 'cluster 1' than 'cluster 2' (0.04% and 2.3%, respectively), likely reflecting the average age of samples. However, there was no correlation between mean individual heterozygosity and proportion of missing data using a linear model ($p = 0.28$; $R^2 = 0.0021$).

Discussion

This is the first study to demonstrate that archived samples in combination with modern genomic tools can reveal temporal changes in biodiversity, which otherwise would have remained unnoticed. We identified genomic differentiation at a relatively local scale for a large migratory shark, the tiger shark. However, this pattern was not apparent in contemporary samples. The data analysis of our spatiotemporal samples suggests that there are two distinct population groups of tiger sharks. One of the population groups was most abundant in the oldest and most southern samples, while it was almost completely absent from contemporary (including southern samples) and most northern samples. We propose that the most parsimonious explanation for this absence is either a shift in relative population abundance or population distribution of two previously cryptic populations of tiger sharks, or both. It is possible that the shift was associated with human activities mirroring the findings from Brown and Roff³⁷ that reported a major decline in abundance of tiger sharks over three generations off the coast of Queensland with greater declines detected at the southern sites. However, we recognise a number of factors could have affected our observations and there may be other alternative causal explanations for this apparent shift in abundance, which we discuss below.

Our results showed evidence of a temporal change in the genetic composition of tiger sharks on the east coast of Australia over the last century, which are unlikely to be the result of technical issues associated with the use of historical DNA (hDNA). In general, capture sequencing of hDNA has a lower sequencing depth and coverage that leads to fewer mapped reads and thus more missing data with increasing

sample age³⁸ which could affect downstream population genomic inferences^{39–41}. Although our historical samples presented fewer reads than the contemporary samples, the amount of missing data was generally very low, even for the oldest samples. For example, in ‘cluster 2’ all except two individuals had less than 5% of missing data (i.e. missing information for 5% of the overall SNP loci). ‘Cluster 2’ individuals generally showed a higher level of heterozygosity than individuals from ‘cluster 1’. This is the opposite of the expected pattern of reduced individual heterozygosity with low coverage caused by allelic drop-out⁴⁰. In addition, the genetic differentiation that we observed between clusters over time was not caused by a few spurious high-differentiated loci (which could be the result of a technical issue in the genomic pipeline), but was found to be spread across the transcriptome. The observed pattern of differentiation was consistent with a scenario of random genetic drift accumulated at an evolutionary time scale in two semi-independent populations of tiger sharks. Thus, the relatively large differentiation between the two putative population clusters, the differences in their levels of heterozygosity, and the finding that not all historical Tasman Sea samples clustered together, renders an alternative hypothesis of a very strong short-term genetic drift in a single panmictic population distributed across the central Indo-Pacific less plausible. In summary, the temporal genetic differentiation was unlikely to be caused by artefacts in sequencing and in the bioinformatics pipeline, by historical genetic drift in a single panmictic population, or by a strong temporal genetic signatures of intra-population environmental selection.

The use of material from historical collections together with genomic-scale analyses provided a relatively large sample size and sufficient statistical power for individual-based cluster analyses, presenting a unique window to explore population composition of tiger sharks in the past. The collection of historical and contemporary samples from the east coast of Australia comprised a huge logistic effort and, at least for the oldest specimens, represents the majority of high quality samples available and practically possible to sample in the community to date. However, our samples are unlikely to provide a full picture of the genetic variation across space and time, including population mixing. As sampling was opportunistic, variation in composition of samples with respect to age and sex, as well as sampling time and method, may have influenced our results. As the oldest samples primarily consisted of large “trophy” fish selectively collected by game-fishermen, they are likely to include a higher proportion of mature females than contemporary samples originating from shark control programmes (drumlines or nets) or research projects. Unfortunately, there is little information on sex for the historical samples. However, a large female bias in sex-ratio is apparent in drumline catches for sharks above approximately 280 cm³⁰ total length. Tagging studies have revealed a variety of behaviours ranging from large-scale movements to considerable site fidelity⁴²; and if tiger sharks show natal site fidelity to specific parturition sites¹⁵, then samples of large, mature females could also explain a higher occurrence of the local (‘cluster 2’) population in the Tasman Sea. However, little is still known about the use of space by tiger sharks, and most of the movements and residency studied so far seem to be connected to feeding grounds or other extrinsic factors¹⁵.

Despite possible sampling-associated uncertainties, the observed patterns of temporal genetic divergence are remarkably clear. Firstly, our results based on contemporary samples are consistent with two recent

studies that detected a single genetic population on the Australian east coast^{20,21}, and are also compatible with the present understanding of population structure in the Indo-Pacific^{21,22}. Secondly, the presence of the two identified cryptic groups was not random in space and time. Individuals from 'cluster 2' were mostly detected in the southern part of the species' distribution, most abundant in the oldest samples, and mostly absent from contemporary samples from the Tasman Sea. Consequently we hypothesize tiger sharks in east Australian waters consisted of at least two populations in the past, but likely comprises a single population now. This may sound counterintuitive in the light of satellite tag-tracking studies that have shown evidence of individuals migrating over 1,000 km^{25,30,43}. However, large migrations and local populations at finer geographical scales are not mutually exclusive, but could be caused by basic "triangle migrations"^{15,44} of fish between parturition sites and juvenile and adult habitats⁴⁵. Despite large migrations, tiger sharks have shown evidence of both site fidelity and residency^{42,46}, i.e. they either stay in or return to specific areas after extensive migrations. This is similar to the white shark, *Carcharodon carcharias*, which displays large-scale migrations, e.g. Southern Ocean⁴⁷; Pacific Ocean⁴⁸; Atlantic Ocean⁴⁹, while also showing fine-scale population structure, e.g. within the Southern Ocean⁵⁰. Little is known as to whether tiger sharks show natal or regional philopatry, as specific pupping grounds or regions have not been identified⁵¹. For some areas, e.g. Hawaiian Islands, tiger sharks seem to have a different pattern of migration depending on their sex, where females show partial migrations possibly related to reproductive purposes while males do not display such behaviour⁴⁶. Further, the species also displays life history traits that are remarkably different to other elasmobranchs, such as only producing single-sired litters while other species employ multiple paternity as a mating strategy⁵². Females producing litters consisting of pups sired by different males is widely documented to increase the genetic quality of offspring, maintain genetic variation in a population, or increase effective population size^{53,54}. The lack of multiple paternity reported for this species may be indicative that tiger shark populations are more vulnerable to the loss of genetic diversity than polyandrous sharks, particularly where overexploitation has resulted in long-term population reductions.

The pattern of population mixing suggests a southern Australian distribution of one of the populations ('cluster 2'), while the apparently more abundant population ('cluster 1') is currently found throughout the entire distribution, with a previous limited intrusion to southern New South Wales (based on the small number of individuals in 'cluster 1' from the historical TAS samples). It is possible that the southerly population ('cluster 2') was a coastal and more resident ecotype and the other population is currently more widespread, more offshore and more migratory, as found in bony fishes such as Atlantic cod (*Gadus morhua*)⁵⁵ and European anchovy (*Engraulis encrasiculus*)⁵⁶ and in marine mammals like the bottlenose dolphin (*Tursiops truncatus*)⁵⁷. Intraspecific differences in movement and residency patterns is also increasingly being reported in sharks⁴⁷, including in large predatory species (e.g.⁴⁷). Importantly, our findings suggest that the current abundance of the putative southerly population has declined compared to pre-1990s levels. The apparent local depletion of tiger sharks at the south eastern distribution of the species in Australia support previous studies showing a reduction in the abundance and mean size of tiger sharks caught^{30,31,37}. Although the species is not commonly commercially

targeted off eastern Australia, the species is caught as bycatch and in Queensland and New South Wales' shark control programs. Off Queensland, tiger shark catch-per-unit-effort has dropped by 74% over the past 25 years, while the average size declined by 21%³¹, with most of the decline occurring in the southern part of the state³⁰. Catch-per-unit-effort also declined in the NSW beach meshing program²⁹. The reduction in a tiger shark population, which coincides with the increase in lethal shark mitigation measures, is of concern and may imply ongoing lethal mitigation measures as possible drivers of population declines. Recent estimates of tiger shark catches obtained from commercial logbook records indicate between 5-10 t routinely caught in Queensland, and approximately 3 t per year in New South Wales. However New South Wales also accounts for approximately 10 t of tiger shark recreational catch^{58,59}. Further, since 1936 the game fishing fishery in New South Wales has targeted larger tiger sharks for capture point scores, and continues to do so over several competitions annually⁶⁰. Illegal foreign fishing vessels, predominantly Taiwanese vessels that target large sharks for their fins, have been apprehended in the Tasman Sea region and north into the Coral Sea, with tiger shark found to comprise about 20% of the total biomass of shark on the Taiwanese vessels⁶¹: the largest tiger shark reported to be 442.1 cm total length⁶¹. Overall, these activities may have selectively removed the southern population ('cluster 2'), most likely through asserting a higher level of exploitation than on the more widespread northern ecotype. Ongoing climate change could also contribute to shifting distributions and abundance of marine fish species and populations⁶², particularly in Australia^{63,64}. In particular, since south-eastern Australia has been identified as one of the global hotspot for ocean warming⁶⁵. Thus, it is possible that increasing sea temperatures could have negatively affected the putative southern population or enhanced the northern population. Genetic analysis of more contemporary samples from along the east coast of Australia and the Pacific Ocean²⁰ using e.g. a targeted genotyping approach focusing on the most informative SNPs⁶⁶, could help elucidating the apparent change of abundance of the two populations of tiger sharks in eastern Australian waters.

The apparent occurrence of localized cryptic populations of tiger sharks, at a finer geographical level than hitherto believed, raises a number of concerns regarding identification and monitoring of intraspecific biodiversity in large sharks. Our study suggests that localized populations may be more common than anticipated from recent genetic studies using markers with lower resolution^{20,67}. This highlights the importance of development of high-resolution genomic resources for elasmobranchs and other high gene flow marine organisms^{68,69} which can provide information on high number of variable sites in the genome (e.g. SNPs). By genotyping many individuals for a high number of SNPs, it may be possible to identify putative populations in species with general low levels of genetic differentiation such as sharks. More importantly, our work also points to significant challenges regarding the scale of current management and biodiversity protection schemes for large sharks. Sustainable management of local populations, through matching the scale of governance with population structure, is important for the protection of the evolutionary legacy of the species and the potential for adapting to future environmental changes⁷⁰. It is also important for the maintenance of healthy marine ecosystems that could provide services to human society⁷¹. Accordingly, management focus will need to include localized protection

measures, such as local seasonal closures or marine reserves to properly match the geographic scale of the population⁷². Specifically, for the east coast of Australia it should be a priority to further confirm our findings, elucidate the current abundance and distribution of the two populations and establish measures to protect the putative southern population component, which appears to have faced a significant historical decline, driven by either direct and indirect exploitation or environmental change.

Method

Sample collection

Tiger shark specimens were caught over a time-span of close to 80 years (1939–2015). Samples originated from north-eastern and eastern Australia, extending from the Gulf of Carpentaria (GCA), through the Coral Sea (CRS) to the Tasman Sea (TAS) (Fig. 1a). Contemporary tissue samples (2000–2015) were obtained as fin-clips from sharks caught in the Queensland Shark Control Program, the New South Wales Shark Meshing Program, commercial and recreational landings, and sharks caught for tagging and tracking research purposes⁷³. Historical samples (1939–1999) comprised dried tiger shark jaws and vertebrae obtained from museum collections, fishers, and other private or public collections. The initial dataset consisted of 115 unique sharks. As tiger sharks are long-lived and the sampled individuals were highly variable in age, we estimated the year of birth for each sample to allow for a more accurate temporal genetic comparison (Fig. 1b). For example, a large shark sampled in 2010 could have been born the same year as a smaller shark sampled in 1990. Individual year of birth was estimated using a locally derived relationship between total length (L_T) and age. Growth rate estimation (t) was based on vertebral aging for both females and males using the Von Bertalanffy growth function (VBGF) (1) as;

$$(1) t = \ln ((-L_T + L_\infty) / (L_\infty - L_0)) / -k^{74},$$

where L_0 and L_∞ represent the length-at-birth and theoretical asymptotic length, respectively, and k represents the growth coefficient. We assumed different parameters for males and females (males: $L_\infty = 441.1$ cm, $k = 0.08$, and $L_0 = 123.4$ cm; females: $L_\infty = 379.9$ cm, $k = 0.06$, and $L_0 = 116.8$ cm), and a combined set where information on sex was not available ($L_\infty = 433.7$ cm, $k = 0.06$, and $L_0 = 121.5$ cm). For individuals with only fork length (L_F) available, total length was calculated using the relationship $L_T = 22.607 + 1.096 L_F^{74}$. For the 20 sharks without length information, total weight (W_T) was used to first obtain L_F using a regression equation parameterized for tiger sharks in the north-western Atlantic⁷⁵, the closest population to our target species from which there is available data ($L_F = ((W_T / 2.5281) \times 10^6) / (1/3.2603)$).

DNA extraction and target capture

Historical tissue material was collected following the protocol described in Nielsen et al.³⁵ and involved the collection of “bio-swarf” produced when drilling a 3.5-mm hole in the calcified cartilage of jaws or vertebrae. Extraction of DNA from the bio-swarf and contemporary fin tissue was performed with the Bioline ISOLATE II Genomic DNA kit according to the manufacturer’s protocol, using 18 – 37 mg (average 27 mg) of tissue per extraction. For genomic library preparation, DNA from contemporary samples was sheared to an average fragment size of 200 bp with a M220 focused ultrasonicator (Covaris, USA). DNA from historical material was fragmented due to degradation over time and therefore used directly. Genomic-capture libraries were prepared using the KAPA Hyper Prep Kit (Kapa Biosystems, USA) according to manufacturer’s instructions. A total of 50 ng input DNA per sample was used with a one in five dilution of the TruSeq DNA HT dual-index adaptors (Illumina, USA). Ten PCR cycles for library amplification were used for the contemporary samples and twelve for the historical. Selected regions of genomic DNA were captured with a MyBaits (MYcroarray) target enrichment kit, consisting of 20.000 biotinylated RNA baits (120 bp each) developed from pancreas, liver and brain derived transcriptome sequences⁷⁶ of the small-spotted catshark (*Scyliorhinus canicula*). At the time of bait development this was the most taxonomically similar species from which a large genomic resource was available for bait design that would likely capture tiger shark transcribed regions. For more details about DNA extraction, library preparation, and bait design see Nielsen et al.³⁵. All DNA samples were captured individually, using 135 ng of DNA library as input, which had previously been treated with a 1x AMPure XP beads clean-up. Hybridization capture of tiger shark DNA was conducted for 24 h at 60 °C in solution for subsequent paired-end (2x125 bp) sequencing on an Illumina HiSeq2000 v4. Prior to sequencing, the captured libraries were amplified using thirteen PCR cycles and purified using 0.8x AMPure XP beads. Quality Control (QC) steps were performed using a Bioanalyzer (Agilent Technologies, CA, USA), thus the final sequencing libraries could be pooled in equal nM concentrations. Samples were sequenced in two lanes. One “historical” lane consisting of 38 jaw samples and 5 fin samples, and a “contemporary” lane with 75 fin samples and 2 jaw samples. The difference in sample number per lane accounted for more variable template numbers among historical samples and thus secured a higher minimum number of sequences per individual. The lanes reciprocally included the same jaw samples that were sequenced in both lanes, similar to one of the contemporary tissue samples in the historical lane, allowing for estimation of “lane effects” and to evaluate reproducibility of multilocus genotypes through the molecular, bioinformatics and population genomics pipelines.

Bioinformatics pipeline and data filtering

We customized a bioinformatics pipeline to ensure removal of potential contaminants, artefacts and low quality reads⁷⁷ before proceeding with the downstream analysis. Briefly, the de-multiplexed reads were controlled for quality using FastQC⁷⁸. All adaptors were removed using AdapterRemoval⁷⁹ and reads were filtered by length and quality, with a minimum length of 30 bp and base quality of 28. Filtered reads were merged using FLASH⁸⁰ with default parameters, and checked for contaminants using Kraken2⁸¹. Both unpaired and concordantly merged reads were mapped against possible sources of contaminants (bacteria, fungi) using the Bowtie2⁸² “sensitive” option. The cleaned reads were also mapped against the

mitochondrial genome of tiger shark (NCBI Reference Sequence: NC_022193.1⁸³). Previous studies have shown that “off-target capture” is common, i.e. capture of genomic regions of both nuclear and mitochondrial origin not matching the baits. For example, in highly degraded samples target template DNA may not bind and amplify as well as in good quality samples and templates, resulting in more amplification of non-targeted regions of the genome^{10,11}. Moreover, mtDNA sequences are commonly captured, or directly sequenced, due to the high copy number of mitochondrial DNA compared to nuclear DNA^{69,84}. This phenomenon may even be desirable, as it allows assessment of mtDNA diversity. After removal of mitochondrial sequences, the reads were mapped against the transcriptome of tiger shark³⁶, using the BWA-mem algorithm. This transcriptome includes 179,867 unique contigs greater than 200 bp length (average 822 bp, maximum 15,892 bp). After mapping, PCR duplicates were removed using Picard-tools (<http://broadinstitute.github.io/picard/>). We checked the patterns of DNA damage of the remaining reads, using MapDamage2.0⁸⁵. Coverage and depth of the target regions were estimated using Samtools⁸⁶, and finally we called SNPs using Freebayes⁸⁷ with default parameters. The raw SNPs obtained were further filtered to keep only biallelic SNPs with quality above 30 and minimum allele count of three. Only SNPs with a maximum level of missing data of 20% were maintained. Additionally, we filtered for excess depth to reduce the possible presence of paralogs and multicopy loci. Linkage disequilibrium (LD) between SNPs within bins of 800 bp (maximum length of the merged reads plus 150 bp each side) was estimated using the prune function in bcftools (SAMtools package), by calculating the square correlation between alleles of each pair of loci, r^2 ⁸⁸ and keeping only SNPs with an $r^2 < 0.25$. To test the reliability of our final SNPs, we compared genotypes for duplicate control samples and only maintained SNPs that matched more than 80% of the pairwise comparisons (to allow for missing data). Finally, we filtered for significant departure from HWE ($p < 0.05$) to remove systematic genotyping errors. All filtering steps but the LD pruning were done using VCFtools⁸⁹.

Data analysis for spatial and temporal genomic variability

Back-calculated year of birth ranged between 1917 (the oldest) and 2012 (the youngest). For all downstream analysis, we grouped samples into four time periods based on their estimated decade of birth: 1910 – 1960, 1970 – 1980, 1990 and 2000, the latter comprising all contemporary samples (2000–2015). These date ranges were used as named time periods throughout the manuscript. The four periods were also associated with different catch rates in the study area, with the highest catch rate between 1960-1980, which significantly decreased after 2000³², especially in the southern part of the Queensland^{9,90}. For the temporal analysis we both estimated temporal genetic differentiation using all samples and a subset of samples from the Tasman Sea, where the majority of temporal samples originated. All estimations of pairwise F_{ST} ⁹¹ between spatial and temporal samples were performed using the R package *StAMPP*⁹² and their significance was assessed with 1,000 permutations over loci. A Principal Coordinates Analysis (PCoA) was applied to the pairwise F_{ST} matrix to summarize and plot the differences reported in the table using the pcoa function in the *ape* 5.0 package⁹³ in R. We performed a Principal Component Analysis (PCA) to explore the spatial and temporal structure of individual genotypes

using the R package *adegenet*⁹⁴. An initial PCA of all samples revealed two identical genotypes (two types of archived tissue from the same individual) and one of them subsequently removed. A PCA of the contemporary samples revealed two extreme outliers of which one of them was identified as another species (spinner shark; *Carcharhinus brevipinna*) based on mtDNA sequences. Both samples were removed from further analysis. In order to test for possible differences in individual heterozygosity among population groups, we used an ad-hoc R script, to calculate the proportion of heterozygous loci out of all genotyped loci for an individual, thereby accounting for differences in total number of genotyped loci (i.e. missing data) across individuals. The boxplot was realized using the *ggplot2* package in R⁹⁵ to highlight the median of each cluster. The distribution of F_{ST} across loci was plotted as a function of heterozygosity using *ggplot2*. Estimates of Weir and Cockerham⁹¹ F_{ST} and heterozygosity were obtained using VCFtools. The F_{ST} was calculated between individuals belonging to different spatiotemporal sample groups. A test of selection was performed using *pcadapt*⁹⁶, with a qvalue of 0.1 as cut-off to identify putative selective outliers and *bayescan*⁹⁷ with parameters -n 5000 -thin 10 -nbp 20 -pilot 5000 -burn 50000 -pr_odds 100. To detect the presence of possible genetic clusters identified by the pairwise F_{ST} , a Discriminant Analysis of Principal Components (DAPC) was applied as it better detects variability among populations⁹⁸ compared to a PCA. For the DAPC we used the *adegenet* package in R, with an a priori assumption of K=2, to further explore the results from the PCA.

Declarations

Acknowledgments

The “Genojaws” project was part funded by the Danish Council for Independent Research Grant DFF – 6108-00583 and Australian Research Grant DP170102043.

We thank the representative of the following institutes and museums that assisted with the sample collections: John West (Taronga Zoo), Sophia Smiley (Waverley Library), Mark McGrouther (Australian Museum), Jeff Johnson (Queensland Museum) and Dennis Reid (formerly New South Wales Department of Primary Industries). We also wish to thank the recreational anglers that permitted us to sample their jaws, or helped us locate other samples: Cameron Baber, Mike Baber, Ivan Bennett, Mark Curtis, Brett Deeney, Neil Grieves, Mark Henrickson, Ken Hoff, Gary Hoff, Leanne Hoff, Gus Johnson, Ed Vander Kruk, David Litchfield, Stuart Lyall, John McIntyre, Mark Mikkelsen, Greg Miles, Brad Page, Nigel Rushworth, Peter Silcock, Mark Stevenson, Terry Titchener, Dave Tubman, Steve Viney, Jamie Ward, Grahame Williams, Karen Wright and David Yiend. Finally, we wish to thank Danielle Davenport and Camilla Christensen for valuable discussions and insights throughout the course of this study.

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E.E.N. and J.R.O. conceived of the study. E.E.N., J.P. and M.B. conducted the field work. A.M., B.J.M., R.H. and E.E.N. wrote the manuscript with input from all other authors. E.E.N., J.R.O. and J.E. conducted the lab work. A.M. conducted all bioinformatics and population genomics analyses with help from R.H. and B.J.M. Input regarding the biology of the species, fisheries data and estimates of length/age were provided by B.H., M.B. J.P. and C.H.

Additional information

Competing Interests statement

The authors have no competing interests.

References

1. Hoffman, J. I. *et al.* High-throughput sequencing reveals inbreeding depression in a natural population. *Proc. Natl. Acad. Sci. U. S. A.* **111**, 3775–3780 (2014).
2. Schindler, D. E., Armstrong, J. B. & Reed, T. E. The portfolio concept in ecology and evolution. *Front. Ecol. Environ.* **13**, 257–263 (2015).
3. Merilä, J. & Hendry, A. P. Climate change, adaptation, and phenotypic plasticity: the problem and the evidence. *Evol. Appl.* **7**, 1–14 (2014).
4. Schindler, D. E. *et al.* Population diversity and the portfolio effect in an exploited species. *Nature* **465**, 609–612 (2010).
5. Mimura, M. *et al.* Understanding and monitoring the consequences of human impacts on intraspecific variation. *Evolutionary Applications* **10**, 121–139 (2017).
6. Hughes, A. R., Inouye, B. D., Johnson, M. T. J., Underwood, N. & Vellend, M. Ecological consequences of genetic diversity. *Ecol. Lett.* **11**, 609–623 (2008).
7. Laikre, L. *et al.* Neglect of genetic diversity in implementation of the convention on biological diversity: Conservation in practice and policy. *Conserv. Biol.* **24**, 86–88 (2010).
8. Hoban, S. M. *et al.* Bringing genetic diversity to the forefront of conservation policy and management. *Conserv. Genet. Resour.* **5**, 593–598 (2013).
9. Dudgeon, C. L. *et al.* A review of the application of molecular genetics for fisheries management and conservation of sharks and rays. *Journal of Fish Biology* **80**, 1789–1843 (2012).
10. Bi, K. *et al.* Unlocking the vault: Next-generation museum population genomics. *Mol. Ecol.* **22**, 6018–6032 (2013).
11. Burrell, A. S., Disotell, T. R. & Bergey, C. M. The use of museum specimens with high-throughput DNA sequencers. *J. Hum. Evol.* **79**, 35–44 (2015).
12. van der Valk, T., Díez-del-Molino, D., Marques-Bonet, T., Guschanski, K. & Dalén, L. Historical Genomes Reveal the Genomic Consequences of Recent Population Decline in Eastern Gorillas. *Curr. Biol.* **29**, 165–170.e6 (2019).
13. Dulvy, N. K. *et al.* Extinction risk and conservation of the world's sharks and rays. *Elife* **2014**, (2014).
14. Dulvy, N. K., Sadovy, Y. & Reynolds, J. D. Extinction vulnerability in marine populations. *Fish Fish.* **4**, 25–64 (2003).
15. Chapman, D. D., Feldheim, K. A., Papastamatiou, Y. P. & Hueter, R. E. There and back again: A review of residency and return migrations in sharks, with implications for population structure and management. *Ann. Rev. Mar. Sci.* **7**, 547–570 (2015).

16. Compagno, L. J. V. FAO species catalogue. Vol. 4. Sharks of the world. An annotated and illustrated catalogue of shark species known to date. Part 2. *Carcharhiniformes* FAO Fish. Synopsis 251–655 (1984).
17. Lea, J. S. E. *et al.* Repeated, long-distance migrations by a philopatric predator targeting highly contrasting ecosystems. *Sci. Rep.* **5**, 1–11 (2015).
18. Ferreira, L. C. *et al.* Crossing Latitudes—Long-Distance Tracking of an Apex Predator. *PLoS One* **10**, e0116916 (2015).
19. Heithaus, M. R., Wirsing, A. J., Dill, L. M. & Heithaus, L. I. Long-term movements of tiger sharks satellite-tagged in Shark Bay, Western Australia. *Mar. Biol.* **151**, 1455–1461 (2007).
20. Holmes, B. J. *et al.* Population structure and connectivity of tiger sharks (*Galeocerdo cuvier*) across the indo-pacific ocean basin. *R. Soc. Open Sci.* **4**, (2017).
21. Pirog, A. *et al.* Genetic population structure and demography of an apex predator, the tiger shark *Galeocerdo cuvier*. *Ecol. Evol.* **9**, 5551–5571 (2019).
22. Sort Matilde, Manuzzi Alice, Jiménez-Mena Belen, Ovenden R Jennifer, Holmes J Bonnie, Bernard M Andrea, Shivji S Mahmood, Meldrup Dorte, Bennett B Mike, Nielsen E Einar1, 5. Come together: Calibration of Tiger Shark (*Galeocerdo cuvier*) microsatellite databases for investigating global population structure and assignment of historical specimens. *Conserv. Genet.*
23. Stevens, J. D. & McLoughlin, K. J. Distribution, size and sex composition, reproductive biology and diet of sharks from Northern Australia. *Mar. Freshw. Res.* **42**, 151–199 (1991).
24. Holmes, B. J. *et al.* Tiger shark (*Galeocerdo cuvier*) movement patterns and habitat use determined by satellite tagging in eastern Australian waters. *Mar. Biol.* **161**, 2645–2658 (2014).
25. Werry, J. M. *et al.* Reef-Fidelity and Migration of Tiger Sharks, *Galeocerdo cuvier*, across the Coral Sea. *PLoS One* **9**, e83249 (2014).
26. Williams, L. E. & Industries., Q. D. of P. *Queensland's fisheries resources: current condition and recent trends 1988-2000 / edited by L. E. Williams.* (Dept. of Primary Industries, 2002).
27. Macbeth, W. G., Geraghty, P. T., Peddemors, V. M. & Gray, C. A. Observer-based study of targeted commercial fishing for large shark species in waters off northern New South Wales. (2009).
28. Paterson, R. A. Effects of long-term anti-shark measures on target and non-target species in Queensland, Australia. *Biol. Conserv.* **52**, 147–159 (1990).
29. Reid, D. D., Robbins, W. D. & Peddemors, V. M. Decadal trends in shark catches and effort from the New South Wales, Australia, Shark Meshing Program 1950 - 2010. *Mar. Freshw. Res.* **62**, 676 (2011).
30. Holmes, B. J. *et al.* Declining trends in annual catch rates of the tiger shark (*Galeocerdo cuvier*) in Queensland, Australia. *Fish. Res.* **129–130**, 38–45 (2012).
31. Roff, G., Brown, C. J., Priest, M. A. & Mumby, P. J. Decline of coastal apex shark populations over the past half century. *Commun. Biol.* **1**, 1–11 (2018).
32. Ferreira, L.C. & Simpfendorfer, C., C. Tiger Shark: *Galeocerdo cuvier*. *IUCN Red List Threat. Species* **8235**, (2019).

33. Jabado, R. W. *et al.* Troubled waters: Threats and extinction risk of the sharks, rays and chimaeras of the Arabian Sea and adjacent waters. *Fish Fish.***19**, 1043–1062 (2018).
34. Muter, B. A., Gore, M. L., Gledhill, K. S., Lamont, C. & Huveneers, C. Australian and U.S. News Media Portrayal of Sharks and Their Conservation. *Conserv. Biol.***27**, 187–196 (2013).
35. Nielsen, E. E. *et al.* Extracting DNA from ‘jaws’: high yield and quality from archived tiger shark (*Galeocerdo cuvier*) skeletal material. *Mol. Ecol. Resour.***17**, 431–442 (2017).
36. Swift, D. G. *et al.* Evidence of positive selection associated with placental loss in tiger sharks. *BMC Evol. Biol.***16**, 1–10 (2016).
37. Brown, C. J. & Roff, G. Life-history traits inform population trends when assessing the conservation status of a declining tiger shark population. *Biol. Conserv.***239**, 108230 (2019).
38. McGaughran, A. Effects of sample age on data quality from targeted sequencing of museum specimens: What are we capturing in time? *BMC Genomics***21**, 188 (2020).
39. Parks, M. & Lambert, D. Impacts of low coverage depths and post-mortem DNA damage on variant calling: A simulation study. *BMC Genomics***16**, 19 (2015).
40. Nielsen, R., Paul, J. S., Albrechtsen, A. & Song, Y. S. Genotype and SNP calling from next-generation sequencing data. *Nature Reviews Genetics***12**, 443–451 (2011).
41. Wang, J. Pedigree reconstruction from poor quality genotype data. *Heredity (Edinb.)***122**, 719–728 (2019).
42. Acuña-Marrero, D. *et al.* Residency and movement patterns of an apex predatory shark (*Galeocerdo cuvier*) at the Galapagos Marine Reserve. *PLoS One***12**, e0183669 (2017).
43. Fitzpatrick, R. *et al.* A Comparison of the Seasonal Movements of Tiger Sharks and Green Turtles Provides Insight into Their Predator-Prey Relationship. *PLoS One***7**, e51927 (2012).
44. Jones, F. R. H. *Fish migration*. (Edward Arnold London, 1968).
45. Krueck, N. C., Treml, E. A., Innes, D. J. & Ovenden, J. R. Ocean currents and the population genetic signature of fish migrations. *Ecology***101**, e02967 (2020).
46. Papastamatiou, Y. P. *et al.* Telemetry and random-walk models reveal complex patterns of partial migration in a large marine predator. *Ecology***94**, 2595–2606 (2013).
47. Bruce, B. D., Stevens, J. D. & Malcolm, H. Movements and swimming behaviour of white sharks (*Carcharodon carcharias*) in Australian waters. *Mar. Biol.***150**, 161–172 (2006).
48. Domeier, M. & Nasby-Lucas, N. Migration patterns of white sharks *Carcharodon carcharias* tagged at Guadalupe Island, Mexico, and identification of an eastern Pacific shared offshore foraging area. *Mar. Ecol. Prog. Ser.***370**, 221–237 (2008).
49. Skomal, G. B., Braun, C. D., Chisholm, J. H. & Thorrold, S. R. Movements of the white shark *Carcharodon carcharias* in the North Atlantic Ocean. *Mar. Ecol. Prog. Ser.***580**, 1–16 (2017).
50. Blower, D. C., Pandolfi, J. M., Bruce, B. D., Gomez-Cabrera, M. D. C. & Ovenden, J. R. Population genetics of Australian white sharks reveals fine-scale spatial structure, transoceanic dispersal events and low effective population sizes. *Mar. Ecol. Prog. Ser.***455**, 229–244 (2012).

51. Holland, K. N. *et al.* A Perspective on Future Tiger Shark Research. *Front. Mar. Sci.***6**, 37 (2019).
52. Holmes, B. J. *et al.* Lack of multiple paternity in the oceanodromous tiger shark (*Galeocerdo cuvier*). *R. Soc. Open Sci.***5**, (2018).
53. Sugg, D. W. & Chesser, R. K. Effective population sizes with multiple paternity. *Genetics***137**, 1147–1155 (1994).
54. Neff, B. D. & Pitcher, T. E. Genetic quality and sexual selection: An integrated framework for good genes and compatible genes. *Molecular Ecology***14**, 19–38 (2005).
55. Hemmer-Hansen, J. *et al.* A genomic island linked to ecotype divergence in Atlantic cod. *Mol. Ecol.***22**, 2653–2667 (2013).
56. Le Moan, A., Gagnaire, P.-A. & Bonhomme, F. Parallel genetic divergence among coastal-marine ecotype pairs of European anchovy explained by differential introgression after secondary contact. *Mol. Ecol.***25**, 3187–3202 (2016).
57. Lowther-Thieleking, J. L., Archer, F. I., Lang, A. R. & Weller, D. W. Genetic differentiation among coastal and offshore common bottlenose dolphins, *Tursiops truncatus*, in the eastern North Pacific Ocean. *Mar. Mammal Sci.***31**, 1–20 (2015).
58. QFish online database, Department of Agriculture & Fisheries Queensland. Accessed 26 May 2020.
59. Rowling, K., Hegarty, A.-M. & Ives, M. Status of fisheries resources in NSW 2008/09. *Status Fish. Resour. NSW 2008/09.* (2010).
60. Pepperell, J. G. Trends in the distribution, species composition and size of sharks caught by gamefish anglers off south-eastern Australia, 1961–90. *Mar. Freshw. Res.***43**, 213–225 (1992).
61. Marshall, L. J. The fin blue line: quantifying fishing mortality using shark fin morphology. (2011).
62. Wisz, M. S. *et al.* Arctic warming will promote Atlantic-Pacific fish interchange. *Nat. Clim. Chang.***5**, 261–265 (2015).
63. Poloczanska, E. *et al.* *Marine Climate Change in Australia: Impacts and Adaptation Responses. 2012 Report Card.* (CSIRO, 2012).
64. Poloczanska, E. S. *et al.* Climate change and Australian marine life. *Oceanogr. Mar. Biol.***45**, 407 (2007).
65. Hobday, A. J. & Pecl, G. T. Identification of global marine hotspots: sentinels for change and vanguards for adaptation action. *Rev. Fish Biol. Fish.***24**, 415–425 (2014).
66. Nielsen, E. E. *et al.* Gene-associated markers provide tools for tackling illegal fishing and false eco-certification. *Nat. Commun.***3**, 1–7 (2012).
67. Bernard, A. M. *et al.* Global population genetic dynamics of a highly migratory, apex predator shark. *Mol. Ecol.***25**, 5312–5329 (2016).
68. Ovenden, J. R., Dudgeon, C., Feutry, P., Feldheim, K. & Maes, G. E. Genetics and Genomics for Fundamental and Applied Research on Elasmobranchs. (Carrier, J. C., Heithaus, M. R., Simpfendorfer, C. A., eds). in *Shark Research: Emerging Technologies and Applications for the Field and Laboratory* (eds. Carrier, J. C., Heithaus, M. R. & Simpfendorfer, C. A.) (CRC Press, 2019).

69. Johri, S., Doane, M., Allen, L. & Dinsdale, E. Taking Advantage of the Genomics Revolution for Monitoring and Conservation of Chondrichthyan Populations. *Diversity***11**, 49 (2019).
70. Hoffmann, A. *et al.* A framework for incorporating evolutionary genomics into biodiversity conservation and management. *Clim. Chang. Responses***2**, 1 (2015).
71. Alexander, S. *et al.* Ecosystem Services: Benefits Supplied to Human Societies by Natural Ecosystems, Vol. 2. *Washington, DC Ecol. Soc. Am.* (1997).
72. Ward-Paige, C. A., Keith, D. M., Worm, B. & Lotze, H. K. Recovery potential and conservation options for elasmobranchs. *J. Fish Biol.***80**, 1844–1869 (2012).
73. Approved by the University of Queensland Animal Ethics Committee (CMS/300/08/DPI/SEAWORLD and CMS/326/11/DPI), the Department of Primary Industries and Fisheries (permit numbers 100541, 165491 and 56095) and the Department of Environment and Resource Management (permit numbers QS2009/GS001, QS2010/MAN26 and QS2010/GS059).
74. Holmes, B. J. *et al.* Age and growth of the tiger shark *Galeocerdo cuvier* off the east coast of Australia. *J. Fish Biol.***87**, 422–448 (2015).
75. Kneebone, J., Natanson, L. J., Andrews, A. H. & Howell, W. H. Using bomb radiocarbon analyses to validate age and growth estimates for the tiger shark, *Galeocerdo cuvier*, in the western North Atlantic. *Mar. Biol.***154**, 423–434 (2008).
76. Mulley, J. F., Hargreaves, A. D., Hegarty, M. J., Heller, R. S. & Swain, M. T. Transcriptomic analysis of the lesser spotted catshark (*Scyliorhinus canicula*) pancreas, liver and brain reveals molecular level conservation of vertebrate pancreas function. *BMC Genomics***15**, 1–18 (2014).
77. Therkildsen, N. O. & Palumbi, S. R. Practical low-coverage genomewide sequencing of hundreds of individually barcoded samples for population and evolutionary genomics in nonmodel species. *Mol. Ecol. Resour.***17**, 194–208 (2017).
78. Andrews, S. A quality control tool for high throughput sequencing data. Available online <http://www.bioinformatics.babraham.ac.uk/projects/fastqc> (2010).
79. Schubert, M., Lindgreen, S. & Orlando, L. AdapterRemoval v2: Rapid adapter trimming, identification, and read merging. *BMC Res. Notes***9**, 88 (2016).
80. Magoč, T. & Salzberg, S. L. FLASH: Fast length adjustment of short reads to improve genome assemblies. *Bioinformatics***27**, 2957–2963 (2011).
81. Wood, D. E., Lu, J. & Langmead, B. Improved metagenomic analysis with Kraken 2. *Genome Biol.***20**, 257 (2019).
82. Langmead, B. & Salzberg, S. L. Fast gapped-read alignment with Bowtie 2. *Nat. Methods***9**, 357–359 (2012).
83. Chen, X., Yu, J., Zhang, S., Ding, W. & Xiang, D. Complete mitochondrial genome of the tiger shark *Galeocerdo cuvier* (Carcharhiniformes: Carcharhinidae). *Mitochondrial DNA***25**, 441–442 (2014).
84. Picardi, E. & Pesole, G. Mitochondrial genomes gleaned from human whole-exome sequencing. *Nature Methods***9**, 523–524 (2012).

85. Jónsson, H., Ginolhac, A., Schubert, M., Johnson, P. L. F. & Orlando, L. MapDamage2.0: Fast approximate Bayesian estimates of ancient DNA damage parameters. in *Bioinformatics***29**, 1682–1684 (Bioinformatics, 2013).
86. Li, H. *et al.* The Sequence Alignment/Map format and SAMtools. *Bioinformatics***25**, 2078–2079 (2009).
87. Garrison, E. & Marth, G. Haplotype-based variant detection from short-read sequencing. (2012).
88. Hill, W. G. & Robertson, A. Linkage disequilibrium in finite populations. *Theor. Appl. Genet.***38**, 226–231 (1968).
89. Danecek, P. *et al.* The variant call format and VCFtools. *Bioinformatics***27**, 2156–2158 (2011).
90. Simpfendorfer, C.A., de Jong, S.K. and Sumpton, W. Long-term trends in large shark populations from inshore areas of the Great Barrier Reef World Heritage Area: results from the Queensland SharkControl Program. *Mar. Trop. Sci. Res. Facil. Transit. Progr. Rep.* (2010).
91. Weir, B. S. & Cockerham, C. C. Estimating F-Statistics for the Analysis of Population Structure. *Evolution (N. Y.)***38**, 1358 (1984).
92. Pembleton, L. W., Cogan, N. O. I. & Forster, J. W. StAMPP: an R package for calculation of genetic differentiation and structure of mixed-ploidy level populations. *Mol. Ecol. Resour.***13**, 946–952 (2013).
93. Paradis, E. & Schliep, K. ape 5.0: an environment for modern phylogenetics and evolutionary analyses in R. *Bioinformatics***35**, 526–528 (2019).
94. Jombart, T. adegenet: a R package for the multivariate analysis of genetic markers. *Bioinformatics***24**, 1403–1405 (2008).
95. Wickham, H. *et al.* Springer-Verlag. New York (2016).
96. Luu, K., Bazin, E. & Blum, M. G. B. pcadapt: an R package to perform genome scans for selection based on principal component analysis. in *Molecular Ecology Resources***17**, 67–77 (Blackwell Publishing Ltd, 2017).
97. Foll, M. & Gaggiotti, O. A genome-scan method to identify selected loci appropriate for both dominant and codominant markers: A Bayesian perspective. *Genetics***180**, 977–993 (2008).
98. Jombart, T., Devillard, S. & Balloux, F. Discriminant analysis of principal components: A new method for the analysis of genetically structured populations. *BMC Genet.***11**, 94 (2010).

Tables

Table 1. Pairwise F_{ST} values between temporal collections. Pairwise F_{ST} values (lower diagonal) and p-values (upper diagonal) to estimate overall genetic differentiation between temporal samples of tiger sharks from east coast Australia. Sample sizes per time period are: 1910–1960 N=21, 1970–1980 N=21, 1990 N=40, 2000 N=25. The star symbol (*) identifies significant values.

	1910–1960	1970–1980	1990	2000
1910–1960	-	0.0900	0.0000*	0.0000*
1970–1980	0.0012	-	0.1780	0.0900
1990	0.0044*	0.0005	-	0.0640
2000	0.0061*	0.0009	0.0009	-

Figures

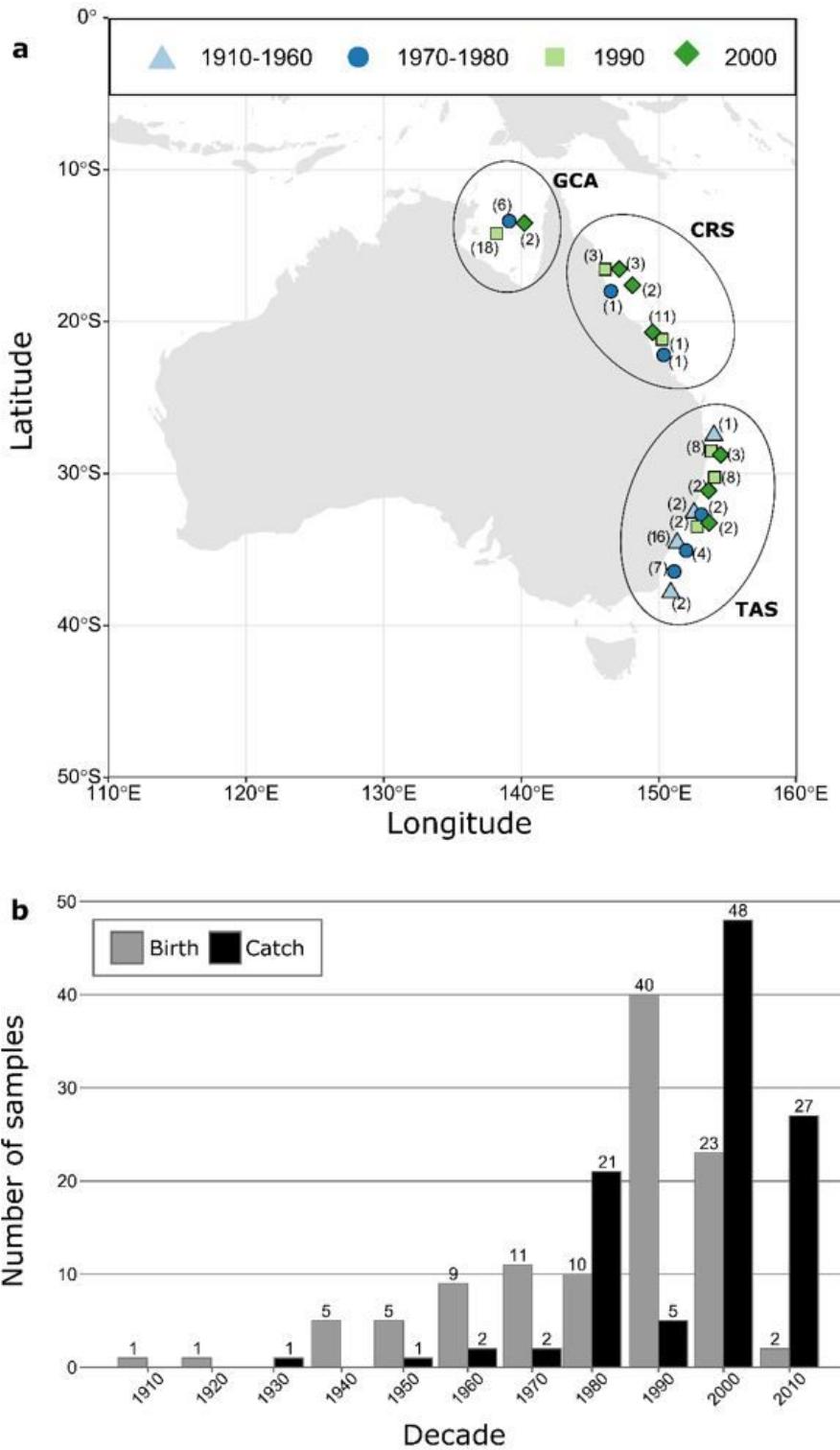


Figure 1

Sampling locations and distribution through time and space. a. Sample distribution along the east-coast of Australia. Samples are grouped by decade of catch (1910–1960, 1970–1980, 1990 and 2000) as explained in the text. The colours identify the four time-periods. The group's names refer to three major spatial collections: Gulf of Carpentaria (GCA), Coral Sea (CRS) and Tasman Sea (TAS). b. The histogram shows the difference between decade of catch and calculated decade of birth and associated sample

numbers, as reported above each bar. Grey bars identify the calculated years of birth, while the black ones refer to the years of catch.

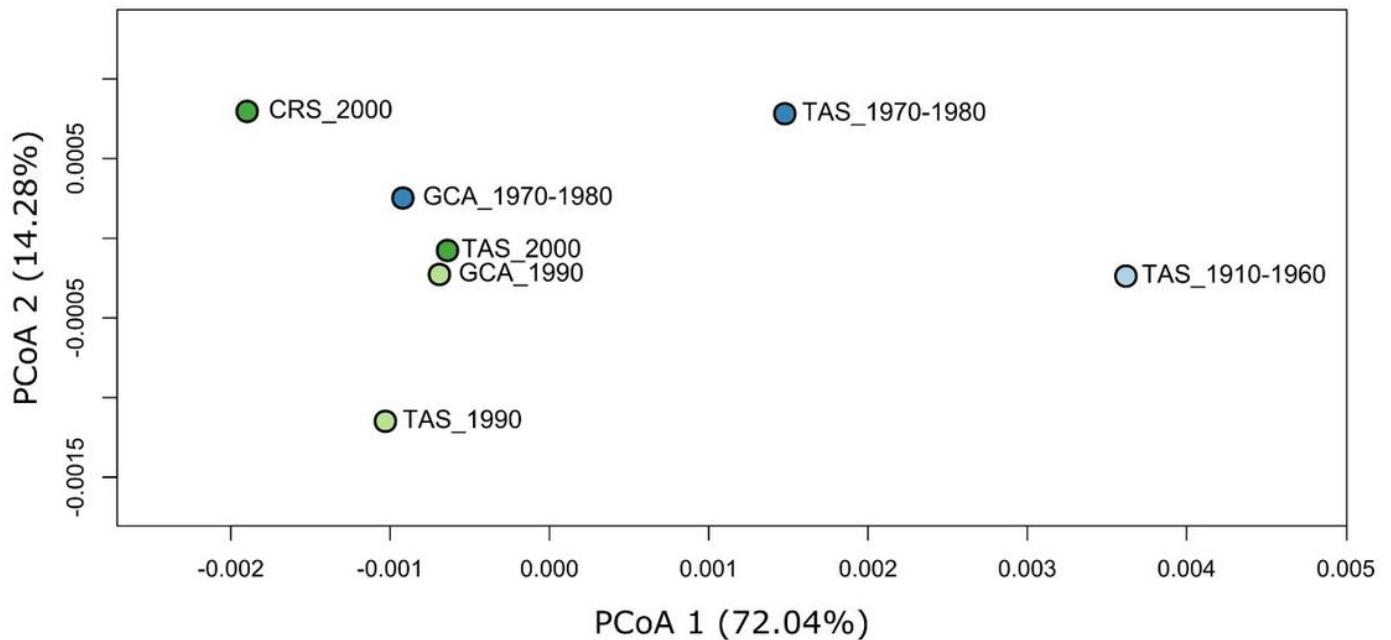
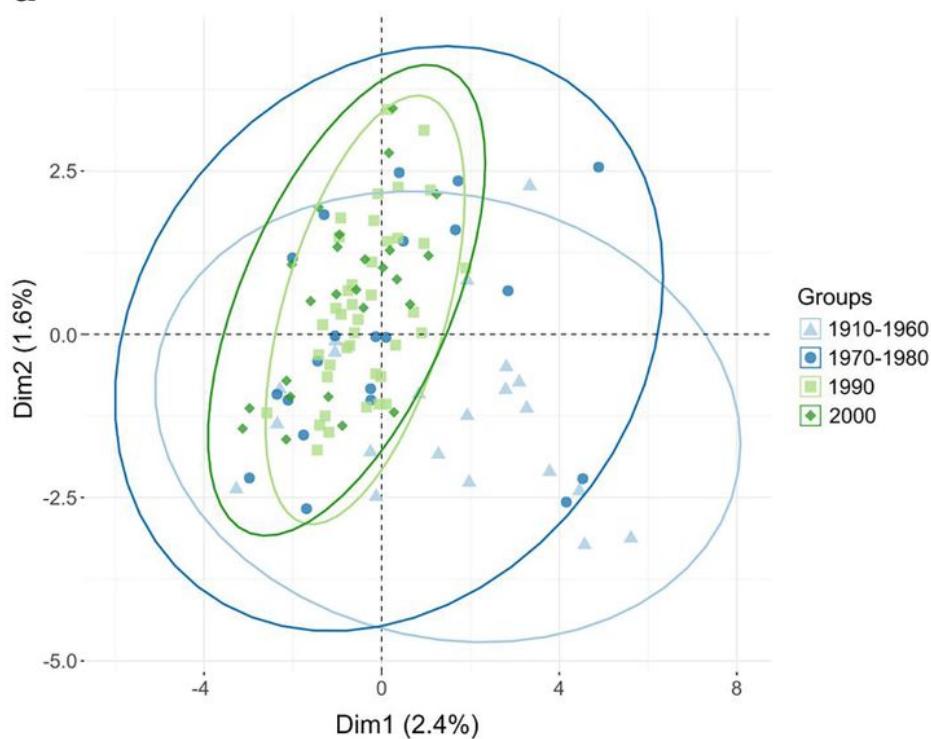
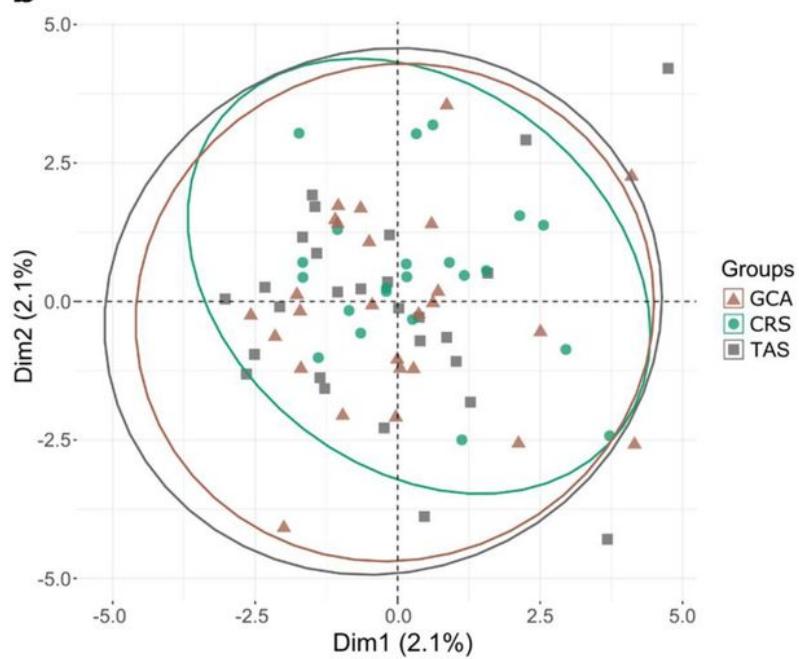


Figure 2

Principal Coordinates Analysis (PCoA) of mean pairwise FST's between spatiotemporal tiger shark samples from eastern Australia. The groups used here are based on back-calculated ages and refer to: Gulf of Carpentaria (GCA), Coral Sea (CRS) and Tasman Sea (TAS). The axis report the percentage of variance explained.

a**b****Figure 3**

Principal Component Analysis (PCA) by time periods and locations. a. PCA of all individual genotypes for the spatiotemporal samples with back-calculated age of birth and b. PCA of contemporary samples based on decade of catch (2000-2010) covering the Gulf of Carpentaria (GCA), Coral Sea (CRS) and Tasman Sea (TAS).

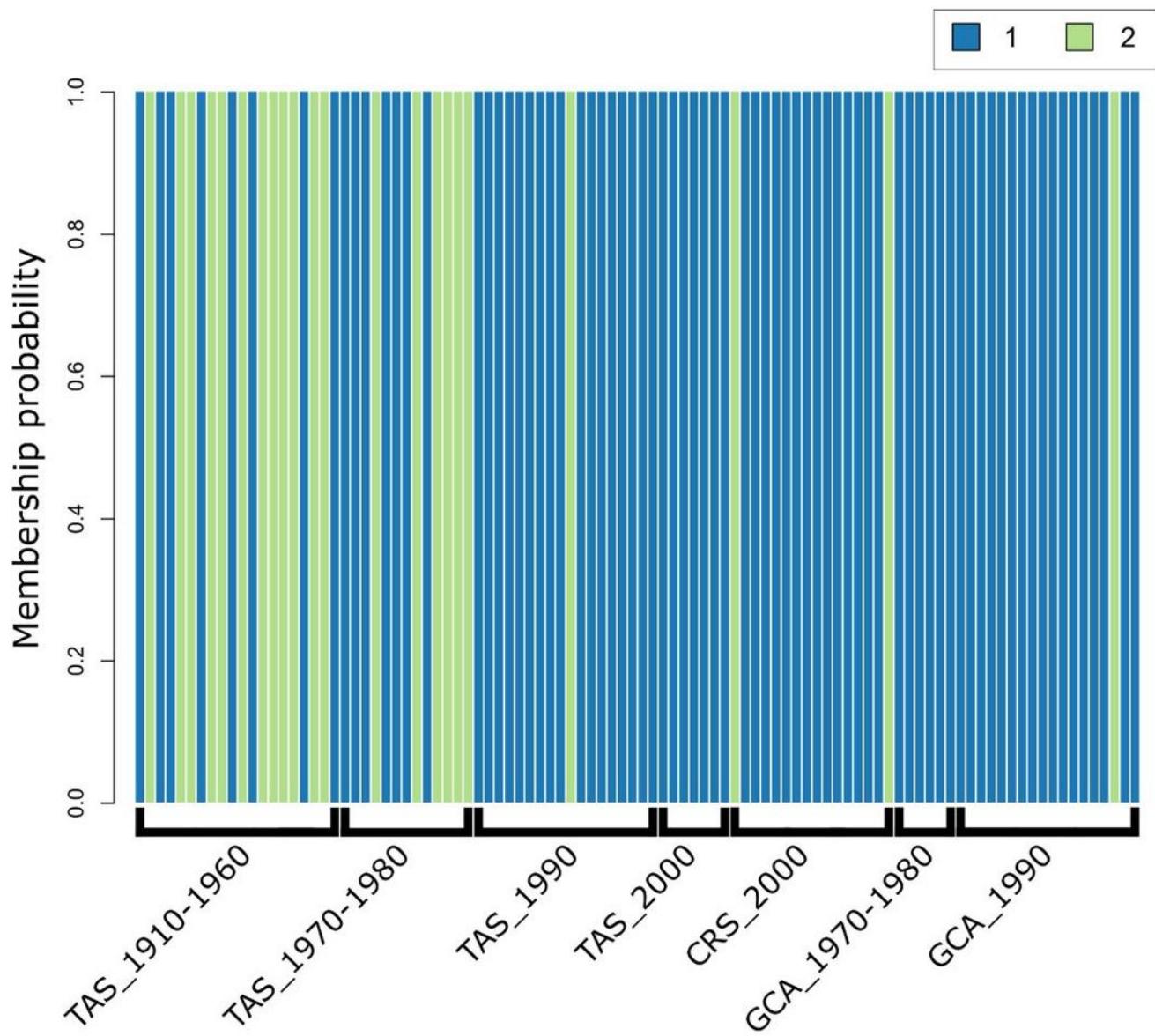


Figure 4

Discriminant Analysis of Principal Components (DAPC) for $K = 2$. The plot illustrates the spatiotemporal occurrence of individuals from the two hypothesized clusters in time and space. Samples grouped by time and space are labelled along the x-axis, only collections encompassing more than six samples were included. The y-axis reports the membership probability of each sample to belong of either clusters ('1' in blue and '2' in green).

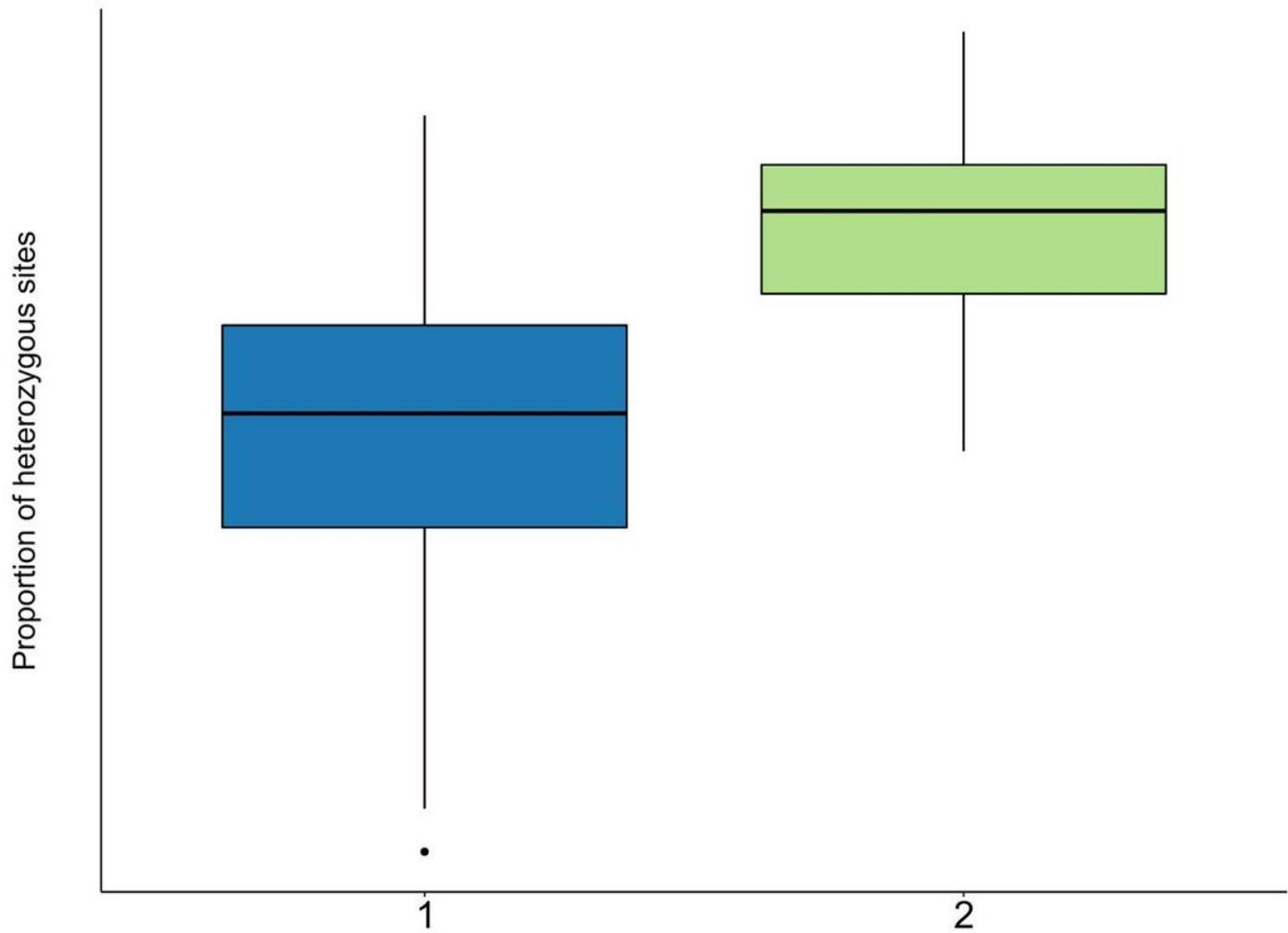


Figure 5

Boxplot of the average proportion of heterozygous SNPs loci for the two clusters. Average proportion of heterozygous SNP loci over total loci genotyped for the two clusters of tiger sharks. Cluster 1 is composed of mainly contemporary and northern samples, while cluster 2 individuals are almost exclusively found in southern historical samples (see Fig. 4 for explanation).

Supplementary Files

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