Journal of Heredity Advance Access published May 14, 2014

Journal of Heredity
doi:10.1093/jhered/esu029

Nuclear and Mitochondrial Patterns of Population Structure in North Pacific False Killer Whales (*Pseudorca crassidens*)

Karen K. Martien, Susan J. Chivers, Robin W. Baird, Frederick I. Archer, Antoinette M. Gorgone, Brittany L. Hancock-Hanser, David Mattila, Daniel J. McSweeney, Erin M. Oleson, Carol Palmer, Victoria L. Pease, Kelly M. Robertson, Gregory S. Schorr, Mark B. Schultz, Daniel L. Webster, and Barbara L. Taylor

From the NOAA Southwest Fisheries Science Center, 8901 La Jolla Shores Drive, La Jolla, CA 92037 (Martien, Chivers, Archer, Hancock-Hanser, Pease, Robertson, and Taylor); Cascadia Research Collective 218 1/2 W, 4th Avenue, Olympia, WA 98501 (Baird, Schorr, and Webster); NOAA Southeast Fisheries Science Center, 101 Pivers Island Road, Beaufort, NC 28516 (Gorgone); Hawaiian Islands Humpback Whale National Marine Sanctuary, 726 S. Kihei Road, Kihei, HI 96753 (Mattila); Wild Whale Research Foundation, Box 139, Holualoa, HI 96725 (McSweeney); NOAA Pacific Islands Fisheries Science Center, 2470 Dole St., Honolulu, HI 96822 (Oleson); Department of Land Resource Management, PO Box 496, Palmerston, NT 0831, Australia (Palmer); Research Institute for Environment and Livelihoods, Charles Darwin University, Casuarina NT 0811, Australia (Palmer); and the University of Melbourne, Faculty of Medical and Dental Health Sciences, Department of Biochemistry and Molecular Biology, Bio21 Molecular Science and Biotechnology Institute, 30 Flemington Road, Parkville, Victoria 3010, Australia (Schultz)

Address correspondence to Karen K. Martien at the address above, or e-mail: Karen.Martien@noaa.gov

Data deposited at Dryad: http://dx.doi.org/doi:10.5061/dryad.2pq32 Available after 1 year embargo.

Abstract

False killer whales (*Pseudorca crassidens*) are large delphinids typically found in deep water far offshore. However, in the Hawaiian Archipelago, there are 2 resident island-associated populations of false killer whales, one in the waters around the main Hawaiian Islands (MHI) and one in the waters around the Northwestern Hawaiian Islands (NWHI). We use mitochondrial DNA (mtDNA) control region sequences and genotypes from 16 nuclear DNA (nucDNA) microsatellite loci from 206 individuals to examine levels of differentiation among the 2 island-associated populations and offshore animals from the central and eastern North Pacific. Both mtDNA and nucDNA exhibit highly significant differentiation between populations, confirming limited gene flow in both sexes. The mtDNA haplotypes exhibit a strong pattern of phylogeographic concordance, with island-associated populations sharing 3 closely related haplotypes not found elsewhere in the Pacific. However, nucDNA data suggest that NWHI animals are at least as differentiated from MHI animals as they are from offshore animals. The patterns of differentiation revealed by the 2 marker types suggest that the island-associated false killer whale populations likely share a common colonization history, but have limited contemporary gene flow.

Subject areas: *Population structure and phylogeography*

Key words: *cetacean, island-associated, male-mediated gene flow, population structure, social structure*

The impact of evolutionary and demographic forces on a genetic locus will depend on the transmission mode, ploidy, mutation rate and mechanism, and functional constraints of that locus. Differences in the patterns of genetic diversity and differentiation from multiple loci can therefore provide insight into the evolutionary history, dispersal patterns, social

structure, and mating patterns of populations. Differentiation in mitochondrial DNA (mtDNA) is expected to be higher than that in nuclear markers due to differences in effective population size between the mitochondrial and nuclear genomes. However, when the difference between the 2 marker types is greater than expected, it can indicate either sex-biased dispersal or male-mediated gene flow without dispersal (Peters et al. 2012). Sometimes mtDNA and nuclear DNA (nucDNA) show contrasting patterns of genetic differentiation, with the hierarchical relationships between populations differing between the 2 marker types (e.g., Gomez et al. 2002). Such complex patterns of genetic structure can result from stochastic variation, selective sweeps, historical events such as introgression or may reflect differences between the colonization history of populations (reflected in mtDNA genealogies) and current patterns of nuclear gene flow (reflected in nucDNA).

We examine the colonization history, social structure, and contemporary differentiation among populations of false killer whales (*Pseudorca crassidens*; Owen 1846) in the North Pacific by comparing phylogeographic patterns and genetic differentiation from mtDNA and nucDNA. False killer whales are a large delphinid found in tropical and subtropical waters throughout the world. Like many marine species, their offshore distribution makes them difficult to study. Most of what is known about false killer whales comes from studies around the Hawaiian Archipelago. Three populations have been described within Hawaiian waters based on photo-identification, satellite telemetry, and genetic data (Chivers et al. 2007; Baird et al. 2008, 2010, 2013). The main Hawaiian Islands (MHI) and Northwestern Hawaiian Islands (NWHI) populations are small, with the MHI population estimated to number only about 151 (coefficient of variation [CV] = 0.20) individuals (Oleson et al. 2010). The only abundance estimate available for the NWHI population is 552 (CV = 1.09) individuals, but there is a large uncertainty associated with this estimate because it is based on only a small number of encounters during a single survey within their range (Bradford et al. 2014). The offshore population is larger—abundance within the 200-nmi Exclusive Economic Zone (EEZ) around Hawai'i is estimated at 1552 (CV = 0.66) individuals (Bradford et al. 2014). However, that may be an underestimate, as the range of the offshore population is unknown and likely extends beyond the EEZ. The MHI, NWHI, and offshore populations are managed as separate population stocks under the US Marine Mammal Protection Act (Carretta et al. 2013).

The MHI population is the subject of an ongoing photo-identification study that dates back to 1986 (Baird et al. 2008, 2012). High resighting rates, sighting histories that span decades for many individuals, and telemetry data that show animals spending the majority of their time in water less than 1000-m deep (Baird et al. 2010, 2012) demonstrate that this is a resident, island-associated population (Baird et al. 2008, 2010, 2012). Though the ranges of the MHI and offshore populations do overlap slightly (Baird et al. 2010), there are no documented interactions between the 2 populations.

The MHI population exhibits strong social structure. Social network analysis revealed the existence of 3 large social clusters that interact regularly but differ in their habitat usage patterns (Baird et al. 2012). There is evidence that the MHI population has undergone a dramatic decline over the past 2 decades (Reeves et al. 2009; Oleson et al. 2010). If the MHI population were extirpated, the habitat might not be recolonized quickly, just as the waters of the Mediterranean surrounding Italy have remained devoid of false killer whales for over 50 years following extirpation due to hunting (Stanzani and Piermarocchi 1992; Reeves and Notarbartolo di Sciara 2006). Following a status review (Oleson et al. 2010), the MHI population was listed as endangered under the US Endangered Species Act (Federal Register 2012).

Far less is known about the NWHI population, which was only discovered in 2010 (Baird et al. 2013). Photo-identification matches between animals encountered near Nīhoa in 2010 and animals encountered near Kaua'i in 2008 suggest long-term site fidelity. Telemetry data indicate that the population ranges from Kaua'i to at least Gardner Pinnacles and maintains a close association with islands, atolls, and seamounts (Baird et al. 2013; Baird RW, unpublished data). Though the range of the NWHI and MHI populations overlap at Kaua'i, they have never been sighted together.

A previous genetic study comparing the MHI and offshore populations revealed strong phylogeographic structuring of haplotypes within North Pacific false killer whales, with animals from the MHI population possessing closely related haplotypes not found elsewhere in the Pacific (Chivers et al. 2007). In this study, we use a substantially expanded sample set, including samples from the previously unsampled NWHI, and data from both mitochondrial control region sequences and 16 microsatellite loci to further investigate population structure in North Pacific false killer whales. We compare the patterns of genetic differentiation among the MHI, NWHI, and offshore populations as revealed by the 2 marker types to gain insight into the colonization history of the Hawaiian Archipelago and the social structure and patterns of gene flow among these populations. We then compare the habitat characteristics of the MHI versus the NWHI to examine the factors that may have driven the evolution of island-associated populations in this otherwise offshore species.

Materials and Methods

The Samples

The sample set consisted of 276 tissue samples collected from false killer whales biopsied at sea (n = 271 including 11 sampled by observers during long-line fishing operations), in captivity (n = 3), or stranded on the beach (n = 2) between 1983 and 2011 (Figures 1 and 2). All tissue samples (i.e., skin or muscle) were preserved frozen or in a 20% dimethylsulfoxide solution saturated with NaCl (Amos and Hoelzel 1991; Amos 1997) and archived in the Southwest Fisheries Science Center's (SWFSC) Marine Mammal and Turtle Molecular Research Sample Collection.

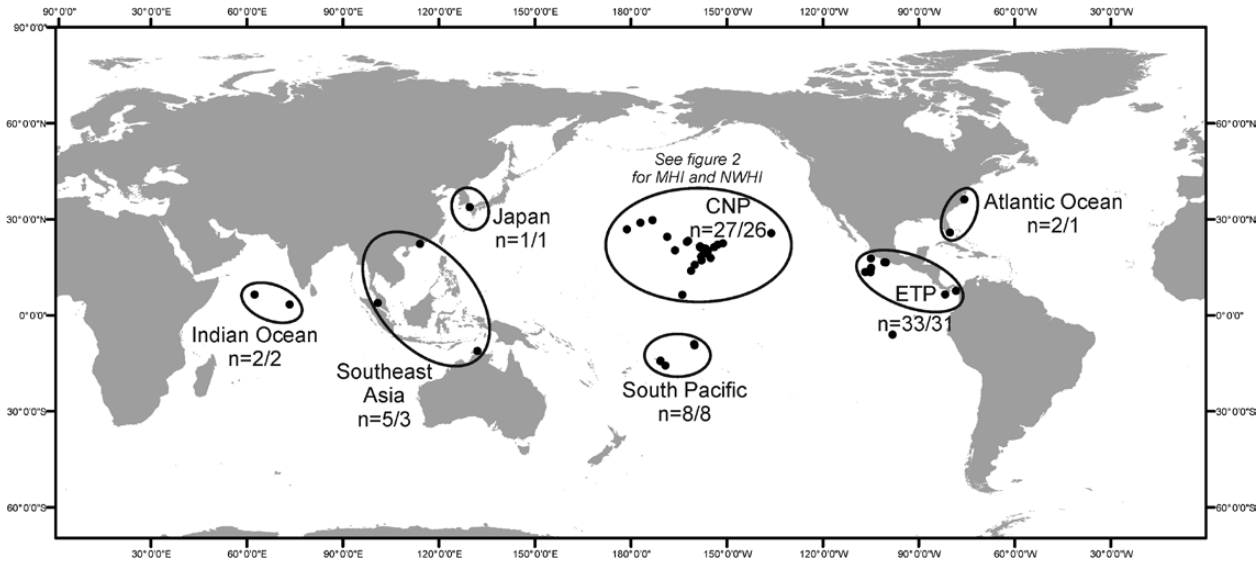

Figure 1. Collection locations of false killer whale samples. The groupings referred to in Table 2 and Figure 3 are indicated, with the exception that "East Pacific" in Figure 3 refers to the ETP stratum plus one encounter (2 samples) south of the ETP. The CNP stratum consists of all samples within the ellipse except for those included in the MHI and NWHI strata (see Figure 2). Sample sizes are listed for the mtDNA data set followed by the nucDNA data set. Only the MHI, NWHI, CNP, and ETP strata are included in most analyses.

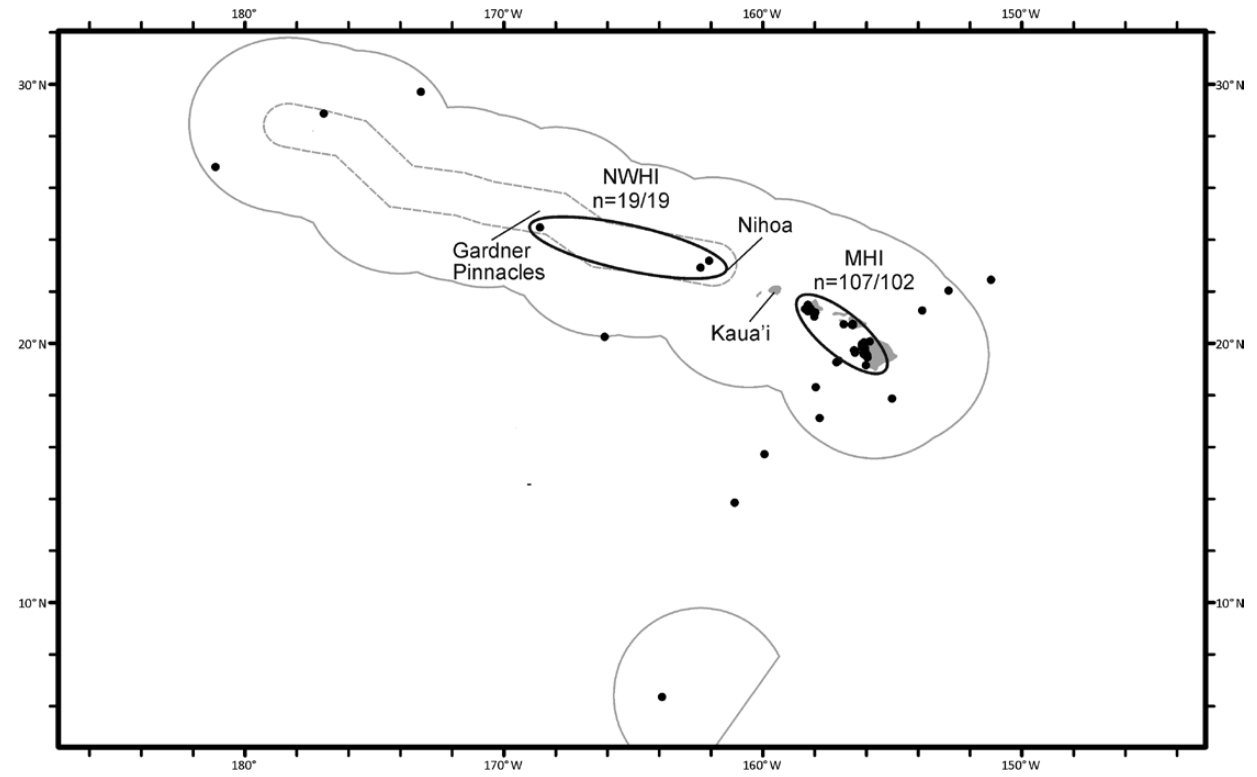

Figure 2. Collection locations of false killer whale samples around the Hawaiian Archipelago. Ellipses indicate the samples included in the NWHI and MHI strata. All other samples on this map are part of the CNP stratum. Sample sizes for the mtDNA/nucDNA data set are indicated. Solid gray lines show the boundaries of the Hawai'i and Palmyra (bottom center) EEZs. The dashed gray line shows the boundary of the Papahānaumokuākea Marine National Monument.

We focused our analyses on samples from the eastern North Pacific, as that is the area from which the majority of our samples were collected. We divided the eastern North Pacific into 4 strata—the MHI, NWHI, Central North Pacific (CNP), and Eastern Tropical Pacific (ETP; Figure 1). The CNP and ETP were delineated based on oceanographic and biogeographic features (Reilly 1990; Longhurst 1998), as well as on sample distribution, whereas the MHI and NWHI strata were delineated based primarily on photo-identification and satellite tag data. The MHI stratum was defined as all samples collected from groups containing any individuals that were part of the Hawai'i insular social network, as determined by analyses of the photo-identification catalog and association pattern data (Baird et al. 2008, 2012). The NWHI stratum contains individuals from 4 encounters (Table 1), all of which occurred very close to islands or atolls. Three of these encounters involved a satellite-tagged individual that remained close to the NWHI during the entire 52-day tag duration (Baird et al. 2013). There were also multiple photo-identification matches among these encounters and between them and individuals encountered off of Kaua'i in 2008, suggesting multiyear fidelity to the Hawaiian Archipelago (Baird et al. 2013). None of the individuals from these encounters nor from the encounter off of Kaua'i in 2008, link to the MHI social network. The fourth NWHI encounter was not linked photographically or through satellite tag data to the

Table 1 Characteristics of the false killer whale groups with multiple samples collected

Encounter number	Samples collected	Samples sequenced	Unique individuals	Final sample size	Collection date	Haplotype
CNP						
1	3	3	3	3	21 April 2008	9, 25
2	4	4	3	3	10 November 2010	32
3	6	4	4	4	18 August 2005	7, 9
4	4	4	3	3	1 September 2010	9
5	5	5	4	4	10 September 2010	30
ETP						
6	5	5	4	4	24 August 1998	9
7	15	15	12	12	14 November 2000	9, 10
8	7	6	3	2	21 August 2003	11
9	13	13	13	8	14 November 1999	10
10	3	3	3	2	7 October 2000	12
NWHI						
11	9	9	8	8	26 September 2010	1, 31
12	5	5	4	4	7 October 2010	1
13	11	11	8	6	21 October 2010	1, 31
MHI						
14	7	7	7	7	30 September 2002	1, 2
15	3	3	2	2	13 September 2004	1
16	12	12	10	10	7 August 2005	1, 2, 5
17	7	7	6	6	16 July 2008	1, 2
18	4	4	4	4	26 July 2008	1, 2
19	4	4	2	2	18 December 2009	1, 2
20	7	7	6	6	19 December 2009	1
21	3	3	1	1	28 July 2010	1, 2
22	7	7	6	6	11 August 2010	1, 2
23	6	6	4	4	14 August 2010	1, 2
24	12	12	10	10	20 August 2011	1, 2
25	5	5	2	2	25 August 2011	1, 2
26	4	4	2	2	9 December 2000	1, 2
27	3	3	2	2	21 December 2000	1
28	6	6	6	6	28 February 2001	1, 2
29	4	4	2	2	1 May 2002	1
30	22	22	18	18	26 May 2003	1
31	3	3	2	2	15 October 2010	1
32	5	5	5	5	10 December 2009	1
American Samoa	4	4	4	4	7 October 2006	17
Australia	3	3	3	3	23 November 2009	5
Southwest Pacific	3	3	2	2	15 October 2006	28

The columns show the total number of biopsy samples collected from the group ("Samples collected"), the number successfully sequenced ('Samples sequenced'), the number of "Unique individuals" once replicates (including those sampled in previous encounters) were removed, the "Final sample size" after closely related individuals were removed, the date of the encounter, and the haplotypes detected in the group. The name of the stratum to which each group belongs is identified under each regional heading.

other NWHI encounters. However, because this encounter occurred within the known range of the NWHI population (based on satellite tag data; Baird RW, unpublished data) and all 4 individuals from this encounter possessed haplotypes only found among the MHI and NWHI samples (see Results), they were included in the NWHI stratum.

Baird et al. (2012) used a social network derived from photo-identification data and an analysis of network modularity to identify social clusters within the MHI population. They identified 3 large social clusters (clusters 1, 2, and 3), as well as 4 small, peripheral clusters primarily composed of animals only sighted once. For some analyses, we stratified the MHI population according to the social clusters identified by Baird et al. (2012). For these analyses, we only included samples from Baird et al.'s clusters 1, 2, and 3 due to the uncertainty associated with their other clusters and the small number of samples available from them. We also excluded samples from individuals that were not photographically identified, as they were not included in Baird et al.'s social cluster analysis.

Laboratory Processing

The 5' end of the hypervariable mtDNA control region was amplified from extracted genomic DNA (lithium chloride protocol: Gemmell and Akiyama 1996; sodium chloride protocol: Miller et al. 1988; Qiagen DNeasy Blood and Tissue Kit #69506) using the polymerase chain reaction (PCR) and then sequenced using standard techniques (Saiki et al. 1988; Palumbi et al. 1991). The PCR cycling profile consisted of 90 °C for 2.5 min, followed by 35 cycles of 94 °C for 50 s, an annealing temperature of 60 °C for 50 s, and 72 °C for 1.5 min, then a final extension of 72 °C for 5 min. The sequence was generated in 2 parts, each of which was sequenced in both directions using Applied Biosystems Inc. (ABI, Foster City, CA) model 377, 3100 and 3730 sequencers. For the 5' segment, we used primers H16498 (5'-CCTGAAGTAAGAACCAGATG-3') (Rosel et al. 1994) and L15829 (5'-CCTCCCTAAGACTCAAGG-3') (developed at the SWFSC), and for the 3' segment, we used primers H497 (5'-AAGGCTAGGACCAAACCT-3') and L16218 (5'-TGGCCGCTCCATTAGATCACGAGC-3') (both developed at the SWFSC). The 3' segment of approximately 573 base pairs included an approximately 20–base-pair section of overlap with the first 395 base pairs of the 5' segment to ensure all sequences were complete. The final sequences were 947 base pairs long and were aligned using SEQED, version 1.0.3 (ABI) and Sequencher software (versions 4.1 and 4.8; Gene Codes, Ann Arbor, MI).

Samples were genotyped using microsatellite DNA primers for 16 dinucleotide loci: D12t derived from beluga whales (*Delphinapterus leucas*) (Buchanan et al. 1996), EV1t and EV14t derived from sperm whales (*Physeter macrocephalus*), EV94t derived from humpback whales (*Megaptera novaenglia*) (Valsecchi and Amos 1996), KWM2at, KWM2b, and KWM12at derived from killer whales (*Orcinus orca*) (Hoelzel et al. 1998), SW19t derived from sperm whales (Richard et al. 1996), SL125t and SL1026t derived from spinner dolphins (*Stenella longirostris*) (Galver 2002), and TexVet5t (Rooney et al. 1999), Ttr11, Ttr34, Ttr48, Ttr58, and TtrRC11 (Rosel et al. 2005) derived from common bottlenose dolphins (*Tursiops truncatus*). Extracted DNA was amplified using published protocols (Martien et al. 2012). The PCR thermal cycling profile for these primers was 90 °C for 2.5 min, followed by 35 cycles of 94 °C for 45 s, 1 min at annealing temperature, and 72 °C for 1.5 min, then a final extension of 72 °C for 5 min. The optimal annealing temperatures were 48 °C for KWM2at and KWM2b, 50 °C for KWM12at, 54 °C for EV14t, 55 °C for D12t, EV1t, EV94t, SL125t, SL1026t, SW19t, TexVet5t, Ttr11, and TtrRC11, 57 °C for Ttr34 and Ttr48, and 60 °C for Ttr58.

Size and purity of the amplicon were assessed electrophoretically. Genotype data were generated on ABI genetic analyzers (models 3100 and 3730) using a commercial internal lane standard (ROX500®; PE Applied Biosystems Inc.). ABI's GENEMAPPER (version 4.0) software was used to make preliminary allele fragment size "calls." GENEMAPPER's calls were reviewed and, if necessary, manually edited to finalize calls. Data generated on the ABI 3100 were normalized from runs of a set of samples on the ABI 3730 using the program Allelogram (Morin et al. 2009b). The size of each allelic pair for each locus constituted the raw data for analyses.

Samples were genetically sexed using the zinc finger (ZFX and ZFY) genes. Prior to 2005, sex determinations were completed according to Fain and LeMay (1995). After 2005, a Real-Time PCR (Stratagene, La Jolla, CA) assay was used as described in Morin et al. (2005).

Data Review

Ten percentage of the sample set, chosen at random, was replicated for each marker during data generation, and these records were reviewed for consistency in allele size scoring. Discrepancies between replicate genotypes were used to calculate the per-allele error rate. After data generation had been completed, all allele size calls were reviewed by a second, independent genotyper. Inconsistencies between the original data set and that generated by the independent genotyper were jointly reviewed by both genotypers and treated as missing data if unresolvable.

Prior to analyses, the final nucDNA data set was reviewed for quality (Morin et al. 2010). Samples that could not be consistently replicated, were missing data for >25% of the markers, or were homozygous at 9 or more loci were deemed to be of poor quality and removed from the data set. The program MICROCHECKER (version 2.2.3; Van Oosterhout et al. 2004) was used to examine the markers for allelic dropout and null alleles. Deviation from Hardy–Weinberg equilibrium (HWE) was assessed for each microsatellite locus using exact tests of HWE (Guo and Thompson 1992) and tests for heterozygote deficiency (Raymond and Rousset 1995), as implemented in GENEPOP version 4 (Rousset 2008). The same software was used to evaluate linkage disequilibrium for each pair of loci using Fisher's method and the Markov chain method. All HWE and linkage disequilibrium

tests were conducted using 1000 dememorization steps, 100 batches, 1000 iterations per batch. The tests were conducted separately for each geographic stratum and combined across strata to calculate a global P value for each locus (Fisher 1935). The jackknife procedure described in Morin et al. (2009a) was used to identify samples that were highly influential (i.e., log-odds greater than 2) in deviations from HWE. The genotypes identified by the jackknife procedure were removed from the data set.

We used the program GenAlEx version 6.4 (Peakall and Smouse 2006) to calculate both the probability that 2 randomly chosen individuals would possess the same multilocus genotype (PI) and the probability that full siblings would share the same genotype (PISibs; Taberlet and Luikart 1999). Pairs of samples that matched in sex, mtDNA haplotype, and microsatellite genotype were considered duplicate samples. When available, photo-identification data were also used to identify duplicate samples from the same individual. The program DROPOUT (McKelvey and Schwartz 2005) was used to identify additional pairs of samples whose genotypes differed at 4 or fewer loci. These pairs could represent duplicate samples with genotyping errors. One sample from each duplicate pair was removed prior to analysis.

Two hundred twenty-four of the samples came from 36 groups (sensu Baird et al. 2008) from which 3 or more samples were collected (Table 1). Due to the strong social structure in false killer whales (Baird et al. 2008, 2012), taking multiple samples from the same group could result in sampling closely related individuals, which could bias our sample. Such a bias is not a concern in the MHI, where nearly 70% of the population has been sampled. However, such a bias is possible in the other strata and could lead some of our analyses to overestimate the amount of population structure within our data (Rodríguez-Ramilo and Wang 2012). We therefore compared relatedness within and among groups from all of our sampling strata other than MHI in order to determine whether the groups are more related than expected. Using the program GenAlEx v. 6.41 (Peakall and Smouse 2006), we calculated Queller and Goodnight's (1989) relatedness within each group and compared it to the distribution of values expected if the individuals in the group had been drawn at random from the allele frequencies for their sampling stratum. For groups that were significantly more related than expected, we identified the individual that had the highest average relatedness to the other members of the group and removed that individual from the data set. We then iteratively reran the relatedness analyses to determine whether the remaining samples were still more closely related than expected, removing samples one at a time as required, until the mean within-group relatedness was not significantly higher than expected.

We reviewed the haplotype data published by Chivers et al. (2007) to ensure data quality. All samples with unique haplotypes (i.e., not present in any other sample) were resequenced 2 or more times to confirm the sequence. We submitted all sequence data to GenBank and all microsatellite genotypes to Dryad for archival purposes.

Analyses of Genetic Data

Genetic Diversity

We identified haplotypes and quantified genetic variability in terms of haplotypic diversity (h) and nucleotide diversity (π) using ARLEQUIN, version 3.11 (Excoffier et al. 2005). For the MHI stratum, we also used ARLEQUIN to calculate Tajima's D (Tajima 1989) and Fu's F_s (Fu 1997) to look for evidence of population expansion or a bottleneck. For the nucDNA data set, we used custom code (available upon request) written in R (R Development Core Team 2011) to calculate the number of alleles per locus, as well as observed and expected heterozygosity, and FSTAT (Goudet 2001) to calculate allelic richness.

To select the most appropriate nucleotide substitution model, we used Akaike information criterion (Akaike 1974) and Bayesian information criterion (Schwarz 1978) with jModelTest version 2.1.1 (Guindon and Gascuel 2003; Darriba et al. 2012). We looked for phylogeographic patterns in the mtDNA data by generating a median joining network using the algorithm of Bandelt et al. (1999), implemented in the software package Network 4.5.1.0 (available from http://www.fluxus-engineering.com/sharenet.htm). We used the Maximum Parsimony option (Polzin and Daneschmand 2003) to identify and eliminate unnecessary median vectors and links and the Star Contraction option (Forster et al. 2001) to simplify the final network for easier visualization. The final network was compared to one without star contraction (data not shown) to ensure that star contraction did not alter or obscure any phylogenetic patterns.

Genetic Differentiation

We conducted both global and pairwise tests of the null hypothesis of no population structure among strata by conducting a global Fisher's Exact test of differentiation (Raymond and Rousset 1995), as implemented in ARLEQUIN (Excoffier et al. 2005), for the mtDNA sequence data set and using a χ^2 test (Roff and Bentzen 1989) for the nucDNA data set. The χ^2 test was implemented using custom code written in R (available upon request). Statistical significance was determined from 10 000 random permutations of each data set, with $\alpha = 0.05$. A correction for multiple tests was not applied to interpret results because each test was testing an independent hypothesis. Corrections for multiple tests effectively reduce the critical value (α), or Type I error rate, at the expense of the Type II error rate (Perneger 1998). Consequently, inappropriate application of correction factors can have serious conservation management implications.

Pairwise estimates of genetic differentiation between strata were also calculated using Φ_{ST} for the mtDNA sequence data, and both F_{ST} (Wright 1931; Weir and Cockerham 1984) and F'_{ST} (Meirmans 2006) for the nucDNA microsatellite data. Φ_{ST} was calculated using the program Arlequin (Excoffier et al. 2005), with the genetic distances between haplotypes calculated using the nucleotide substitution model selected by the jModelTest analysis. F_{ST} and F'_{ST} were calculated using custom code written in R (available upon request).

We tested for evidence of sex-biased dispersal in the microsatellite data set using the biased dispersal test of Goudet et al. (2002), as implemented in FSTAT (Goudet 2001). This test looks for evidence of first-generation immigrants within a sample. We examined differences between males and females with respect to mean and variance of assignment indices, F_{IS}, F_{ST}, relatedness, and within-group gene diversity (H_S) and assessed significance through 1000 permutations. To test for evidence of sex-biased dispersal within the mtDNA data set, we used custom R code (available upon request) to create an mtDNA implementation of the same sex-biased dispersal test used for the microsatellite data set. For the mtDNA data set, we only compared F_{ST} values between males and females, and again assessed significance through 1000 permutations.

We used the Bayesian clustering program STRUCTURE 2.3.3 (Pritchard et al. 2000; Falush et al. 2003; Hubisz et al. 2009) to cluster all samples collected from the central and eastern Pacific Ocean (i.e., samples from Japan and Southeast Asia were excluded) on the basis of their microsatellite genotypes. We had STRUCTURE cluster the samples into k = 1–6 groups and ran STRUCTURE 5 times for each k. To confirm convergence, we compared estimates of likelihood and ancestry across runs. We evaluated support for different values of k by comparing the mean log-likelihood for each model and by calculating ΔK (Evanno et al. 2005). All analyses were run for 1 000 000 steps following a burn-in of 50 000 and assumed an admixture model with correlated frequencies. All other parameters were left at program defaults. We used CLUMPP (Jakobsson and Rosenberg 2007) to average the ancestry coefficients of individuals across replicate runs and graphed the results with Distruct (Rosenberg 2004).

We stratified the samples from the central and eastern Pacific according to the groups identified by STRUCTURE and used the program GeneClass2 (Piry et al. 2004) to conduct an assignment test. Though STRUCTURE can also be used for assignment tests, GeneClass2 has the advantage that, in addition to identifying the most likely source of a sample, it can identify source populations that can be excluded as the source of a sample. We used the assignment criterion of Paetkau et al. (1995) to calculate the likelihood of each sample having originated in each stratum and used Paetkau et al.'s (2004) resampling method to determine whether any of the strata could be excluded as the potential source of each sample. We used L_home as the likelihood value, as it is possible that some individuals could have originated in unsampled source populations. For all analyses, we set the default frequency for missing alleles at 0.01, performed 1000 resampling events, and set the α to 0.01 as recommended by Piry et al. (2004).

To test for evidence of isolation-by-distance we used GenAlEx6 (Peakall and Smouse 2006) to calculate the genotypic (Peakall 1995; Smouse and Peakall 1999) and geographic distance between all pairs of individuals and conduct a Mantel test. We conducted the test within each of our 4 North Pacific strata (Figures 1 and 2), within the groups identified by STRUCTURE, and within the entire eastern and central Pacific combined. We assessed the significance of the Mantel Tests through 999 permutations.

We used the nucDNA data set and the program Migrate 3.6.4 (Beerli 2006) to estimate the mutation-scaled effective population sizes ($\Theta = 4N_e\mu$) and asymmetric mutation-scaled dispersal parameters ($M = m/\mu$) between our 4 North Pacific strata. We used a Bayesian search strategy with 8 replicate chains, each consisting of a 100 000 step burn-in followed by a 2 500 000 step chain that was sampled every 100 steps. For Θ we used a uniform prior from 0 to 100 with a window of 25, whereas for M we used a windowed exponential prior with minimum of 0, mean of 500, maximum of 1000, and sampling window of 250. The priors were chosen based on preliminary runs and sampled via Slice Sampling. Chain convergence was confirmed through estimates of effective sample size and by comparing results across 3 independent runs.

Results

Data Review

The probability of 2 individuals possessing the same multilocus genotype was 1.23×10^{-18} for unrelated individuals and 2.70×10^{-7} for full siblings, indicating that the microsatellite loci were adequate for identifying unique individuals. Using available photographs and genotypes from the 16 nucDNA markers, 51 duplicate samples from 44 individuals were identified and removed prior to analyses. In all cases, duplicate samples of a given individual were sampled within the same stratum. In addition to the removal of duplicate samples, 11 samples were eliminated from the mtDNA analysis and 22 samples from the nucDNA analysis due to poor sample quality. Encounters 9, 10, and 13 (Table 1) exhibited significantly higher within-encounter relatedness than the other encounters. We eliminated 5, 1, and 2 samples from these encounters, respectively, to reduce the within-group relatedness to the point where it was no longer significantly higher than expected. After these exclusions, the data sets used for all summary statistics and analyses included 206 samples in the mtDNA data set and 195 samples in the nucDNA data set.

Two samples were identified in the HWE jackknife analysis as having likely genotyping errors. Both were homozygous for rare alleles, one at locus D12t and one at locus Ttr11. The genotypes of these samples at these loci were set to null for all analyses.

Locus D12t was out of HWE in the MHI stratum and EV94t was out of HWE in the CNP stratum (Supplementary Table 1 online). No other deviations were detected for any other loci in the MHI or CNP strata, and no deviations were detected for any loci in the other strata. The Fisher's Exact test for linkage disequilibrium across populations was significant for 8 pairs of loci: SW19t/Ttr48, EV94t/Ttr58, SL1026t/TtrRC11, EV94t/KWM2at, KWM2at/SL1026, KWM2at/Ttr58, KWM2b/SL125t, and EV14t/TexVet5t. Most of the linkage disequilibrium was found in the MHI stratum, with only 3 locus pairs deviating significantly from linkage disequilibrium in 2 strata. None were significant for more than 2 strata. Deviations from HWE and linkage equilibrium are not unexpected in a small population from which more than half of the individuals have been sampled, like the MHI insular false killer whales. Thus, all markers were retained.

Twenty-four samples were randomly chosen for replication. These samples were genotyped an average of 2.1 times at each nuclear locus, for a total of 1380 allele calls. Eighteen errors were detected, resulting in a per-allele error rate of 0.013. Resequencing of samples revealed errors in some of the Chivers et al. (2007) sequences and resulted in the elimination of 7 haplotypes: 3, 4, 8, 13, 14, 15, and 22 (numbers correspond to the haplotype numbers in Chivers et al. Figure 4b). Thus, there were 17 rather than 24 haplotypes among the samples included in that study. The numbers assigned to those haplotypes were not reused. Furthermore, we identified 2 errors that had resulted in Chivers et al. concluding that one animal sampled around Palmyra possessed haplotype 2 and one animal sampled around the MHI had haplotype 9 (see Chivers et al. 2007; Figure 3). In fact, the haplotypes of those individuals were reversed.

Genetic Diversity

We identified 25 haplotypes among the 206 mtDNA sequences representing individual animals in the data set (Table 2), including 8 additional haplotypes (haplotypes 25–32) identified among the samples added to the Chivers et al. (2007) data set. There were 32 polymorphic sites including 30 substitutions (29 transitions and 1 transversion) and 2 insertions/deletions in the 947 base-pair sequences. The best-fit substitution model was the Tamura and Nei (1993) model with invariant sites (TrN+I). The observed nucleotide diversity was low (i.e., π = 0.32%; Table 3) compared with most other delphinids (e.g., spotted dolphins, Escorza-Trevino et al. 2005; bottlenose dolphins, Martien et al. 2012), but comparable to estimates from other closely related species such as killer whales and pilot whales (Hoelzel et al. 1998; Oremus et al. 2009). Overall haplotypic diversity was 0.740 (± 0.028; Table 3). For the MHI stratum, neither Tajima's D (D = −0.579, P = 0.324) nor Fu's F_s (F_s = 2.212, P = 0.872) provided evidence of population expansion or bottleneck, as neither was significantly different from zero.

We detected more than one haplotype in 17 of the 35 groups where 3 or more individuals were sampled (Table 1). In all but 3 of these cases (encounters 1, 14, and 19), both haplotypes were detected in females, confirming that groups encountered in the field are not strictly matrilineal (Chivers et al. 2007).

Observed heterozygosity and allelic richness for the 16 nucDNA markers were similar across all strata (Table 3). The number of alleles detected at the different loci ranged from 5 (Ttr34) to 16 (TtrRC11; Supplementary Table 1 online).

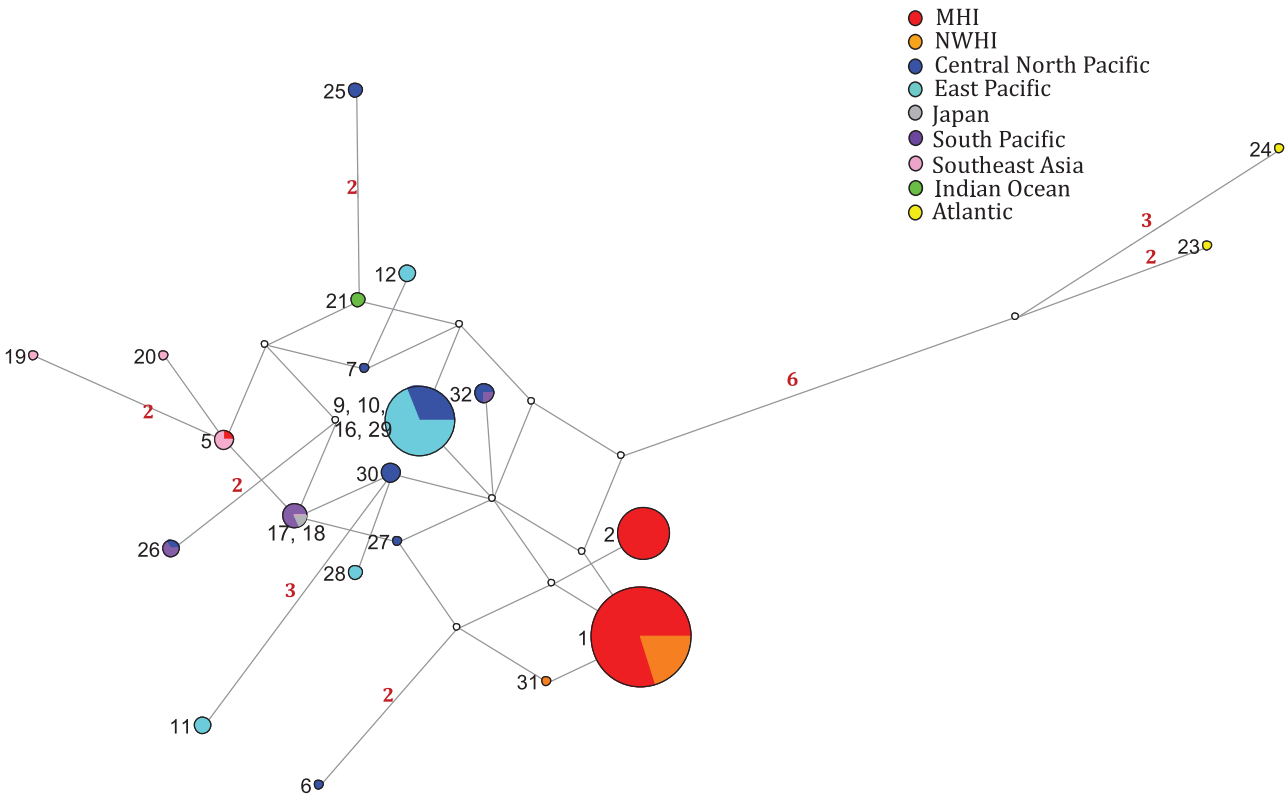

Figure 3. Median joining network of all false killer whale haplotypes. Nodes are color-coded according to the geographic region from which they were sampled (see Figure 1). The size of nodes is proportional to the frequency of the haplotype. Small white circles represent haplotypes inferred by the analysis but not present in our data set. Lengths of lines connecting nodes are proportional to the inferred number of mutations separating haplotypes. Small red numbers above the lines indicated the number of mutational events, if greater than one.

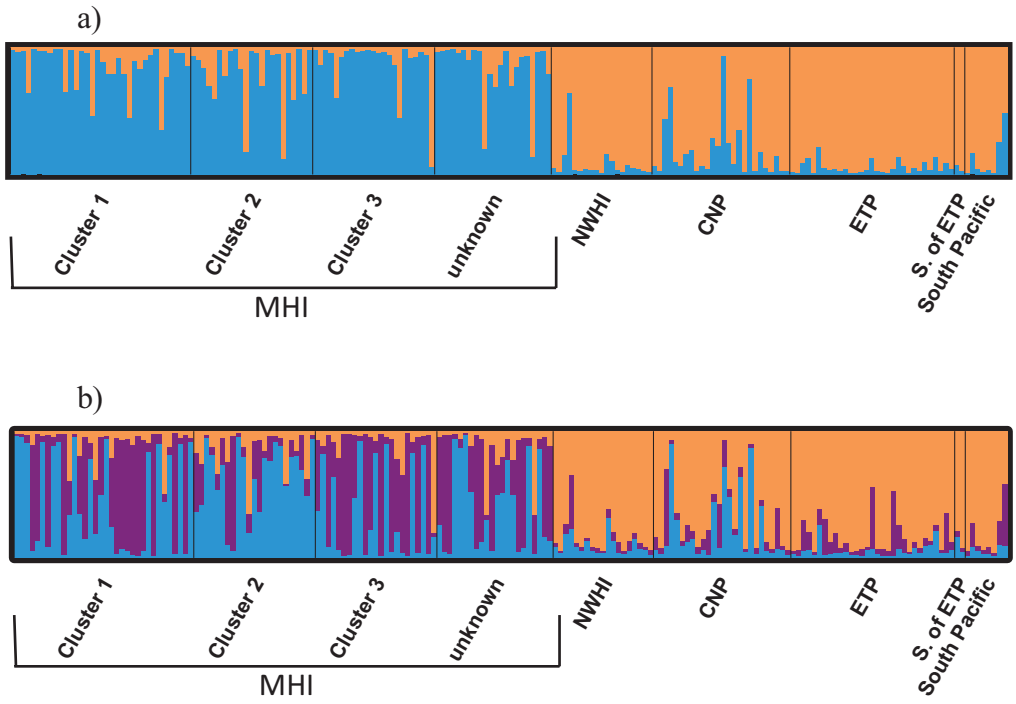

Figure 4. Graphical representation of STRUCTURE results for models containing (**a**) 2 and (**b**) 3 groups. Each vertical bar represents an individual and is shaded as to the proportion of the individual's ancestry that is attributable to groups 1 (orange), 2 (blue), and 3 (purple), as defined by STRUCTURE. MHI samples are divided into the 3 social clusters identified by Baird et al. (2012) as well as those samples for which social cluster affiliation is unknown. The remaining ndividuals are arranged into the strata indicated on Figures 1 and 2. 'S. of ETP' refers to 2 samples collected south of our ETP stratum.

Phylogeographic Structure

Phylogeographic concordance is evident in the distribution of haplotypes (Figure 3). The 2 Atlantic Ocean samples cluster together in the median joining network and are 10 nucleotide substitutions from the nearest Pacific haplotypes. The 3 haplotypes from the MHI and NWHI (1, 2, and 31) also cluster together. Haplotypes from the South Pacific, Southeast Asia, and Japan also appear near each other on the median joining network, but are more similar to the east Pacific and central North Pacific haplotypes than are the Hawaiian haplotypes.

One group (encounter 16, Table 1) sampled off the island of Hawai'i included a male with haplotype 5, a haplotype also identified from animals sampled off Northern Australia. This was the only animal associated with either of the island-associated populations that possessed a haplotype found elsewhere in the Pacific. This animal was photographed and judged "distinctive" (meaning it would be readily identifiable in subsequent photographs), but has never again been sighted (Baird RW, unpublished data). Of the 10 individuals photographically identified from this group, the individual with haplotype 5 is the only one that has not been resighted.

Genetic Differentiation

We found evidence of statistically significant genetic differentiation among strata. Global tests of differentiation revealed significant ($P < 0.0001$) genetic divergence between strata overall in both the mtDNA and nucDNA data sets, rejecting the global null hypothesis of no population structure. All pairwise comparisons among geographic strata were statistically significant in both data sets (Table 4). In the mtDNA data set, genetic divergence (Φ_{ST}) between the MHI and NWHI ($\Phi_{ST} = 0.131$) was similar to that between the CNP and ENP ($\Phi_{ST} = 0.091$), whereas divergence values between the island-associated strata and the offshore strata were much higher, ranging from 0.618 to 0.709 (Table 4). In the nucDNA data set, in contrast, divergence values were much smaller and similar across all comparisons, with F_{ST} estimates ranging from 0.012 to 0.031 (Table 4). The lowest divergence values calculated from the nucDNA data set were from the 3 comparisons involving the CNP. No evidence for sex-biased dispersal was detected in either data set using the bootstrap analysis.

We also found significant genetic differentiation among the social clusters within the MHI population (Table 4). In the mtDNA data set, clusters 1 and 2 were not significantly differentiated from each other, though both were significantly differentiated from cluster 3. This result was driven by the fact that haplotype 2, while at frequencies of 0.441 and 0.417 in clusters 1 and 2, respectively, was completely absent from cluster 3. In the nucDNA data set, estimates of F_{ST} ranged from 0.008 to 0.012 (Table 4).

Table 2 Haplotype frequencies for all false killer whales after replicates were removed

Haplotype	MHI	NWHI	CNP	ETP	South Pacific	West Pacific	Indo-Pacific	Indian Ocean	Atlantic Ocean
1 (EF601197)	79	18							
2 (EF601198)	27								
5 (EF601201)	1						3		
6 (EF601204)			1						
7 (EF601202))			1						
9 (EF601207)			14	14					
10 (EF601208)				13					
11 (EF601209)				2					
12 (EF601210)				2					
16 (EF601205)			1						
17 (EF601206)					5				
18 (EF601216)						1			
19 (EF601217)						1			
20 (EF601218)							1		
21 (EF601214)								2	
23 (EF601219)									1
24 (EF601220)									1
25 (HQ438483)			2						
26 (HQ438484)			1		2				
27 (HQ438485)				1					
28 (HQ438486)					2				
29 (HQ438487)				1					
30 (KJ567087)			4						
31 (KJ567088)		1							
32 (KJ567089)			3		1				

Column names correspond to the strata indicated in Figures 1 and 2. GenBank accession numbers are given in parentheses.

Table 3 Estimates of genetic diversity of false killer whales for the mtDNA and nucDNA data sets for the 4 CNP and eastern North Pacific strata, MHI social clusters, and for all samples

Stratum	mtDNA				nucDNA				
	n	N_h	H	π	n	N_A	H_o	H_e	A_R
MHI	107	3	0.395 ± 0.043	0.0009 ± 0.0007	102	7.69 ± 2.24	0.744 ± 0.153	0.746 ± 0.144	6.34 ± 1.56
NWHI	19	2	0.105 ± 0.092	0.0001 ± 0.0002	19	6.75 ± 2.49	0.789 ± 0.177	0.741 ± 0.165	6.68 ± 2.84
CNP	27	8	0.712 ± 0.085	0.0030 ± 0.0018	26	7.88 ± 2.90	0.733 ± 0.152	0.764 ± 0.132	7.32 ± 2.52
ETP	33	6	0.676 ± 0.052	0.0022 ± 0.0014	31	8.50 ± 2.78	0.752 ± 0.144	0.756 ± 0.134	7.43 ± 2.18
Cluster1	34	2	0.508 ± 0.029	0.0011 ± 0.0008	34	6.50 ± 1.90	0.741 ± 0.143	0.731 ± 0.146	5.59 ± 1.44
Cluster2	24	3	0.554 ± 0.053	0.0014 ± 0.0010	23	6.50 ± 1.75	0.753 ± 0.169	0.751 ± 0.157	5.83 ± 1.57
Cluster3	23	1	0.000 ± 0.000	0.0000 ± 0.0000	23	6.38 ± 1.54	0.740 ± 0.198	0.745 ± 0.134	5.70 ± 1.18
All samples	206	25	0.740 ± 0.028	0.0032 ± 0.0019	195	10.56 ± 3.50	0.744 ± 0.136	0.761 ± 0.142	10.47 ± 3.44

Allelic richness was calculated using a minimum sample size of 18 for the geographic strata, 12 for the social clusters, and 168 for all samples.

The STRUCTURE analyses also detected genetic structure within the North Pacific. The highest mean log-likelihood and highest ΔK occurred when the samples were divided into 3 groups (Supplementary Table 2 online). However, the mean log-likelihood for the model with 2 groups was only slighter lower. In the model with 2 groups, the MHI samples derived an average of 15% of their ancestry from group 1 and 85% from group 2, whereas the remaining samples derived an average of 89% of their ancestry from group 1 and only 11% from group 2 (Figure 4). Increasing the number of groups to 3 had little effect on the assignment of non-MHI samples, but resulted in the MHI samples being split between groups 2 and 3 (Figure 4). The assignment of MHI samples to groups 2 and 3 did not vary significantly between the social clusters (Anova of logit-transformed assignment probabilities, P value = 0.101). When we used STRUCTURE to analyze the MHI and non-MHI samples separately, the model with only one group was strongly supported in both cases, indicating that there is no population structure detectable by STRUCTURE within either of the 2 groups. We therefore conclude that the model with 2 groups provides the best fit to our data. We henceforth refer to the 2 groups identified by STRUCTURE as the MHI (which is identical to the MHI stratum used in other analyses; Figure 4), and the Eastern Pacific, which includes all samples used in the STRUCTURE analyses except those from the MHI.

Table 4 Estimates of genetic differentiation between eastern and CNP strata and between MHI social clusters

Comparison	mtDNA			nucDNA	
	Φ_{ST}	χ^2 *P* value	F_{ST}	F'_{st}	χ^2 *P* value
MHI versus NWHI	0.131	0.005	0.030	0.119	<0.0001
MHI versus CNP	0.658	<0.0001	0.012	0.047	<0.0001
MHI versus ETP	0.709	<0.0001	0.023	0.090	<0.0001
NWHI versus CNP	0.618	<0.0001	0.018	0.074	<0.0001
NWHI versus ETP	0.695	<0.0001	0.031	0.121	<0.0001
ETP versus CNP	0.091	<0.0001	0.012	0.050	0.0002
Among social clusters					
1 versus 2	−0.029	0.5710	0.008	0.029	0.015
1 versus 3	0.376	0.0001	0.012	0.048	0.001
2 versus 3	0.342	0.0006	0.012	0.051	0.004

Based on the assignment test conducted with GeneClass2, the proportion of samples assigning most strongly to the source population from which they were collected was 0.931 (95/102) for the MHI samples and 0.867 (78/90) for the Eastern Pacific samples. There was one sample from each population for which the population where it was collected could be excluded as a possible source. One of these was a sample collected in the Eastern Pacific for which both putative source populations could be excluded. The Eastern Pacific could be excluded as a possible source for 5% (5/102) of the MHI samples, whereas the MHI could be excluded as a possible source for 37% (33/90) of the Eastern Pacific samples. One of the MHI samples that assigned more strongly to the Eastern Pacific population was the individual that possessed haplotype 5, which was also found in Australia. That individual had an exclusion probability from the MHI of 0.027, slightly higher than the critical value of 0.01 used to identify migrants.

We did not find evidence of isolation-by-distance within the MHI, NWHI, CNP, or ETP strata, nor did we find isolation-by-distance when we combined all of these strata. However, when we excluded the MHI samples and analyzed all other samples from the Eastern Pacific, we did find statistically significant ($P = 0.002$) evidence of isolation-by-distance, with geographic distance accounting for 6.9% of the variation in genetic distance between individuals.

The migration parameters estimated by MIGRATE had broadly overlapping 95% confidence intervals, all of which included zero (Supplementary Figure 1 online). Similarly, the estimates of Θ produced by MIGRATE had overlapping confidence intervals that included zero (Supplementary Figure 2 online). The estimates therefore did not provide any insight into the relative sizes of or rates of dispersal between our 4 North Pacific strata.

Discussion

Patterns of Differentiation

Our analyses of both mitochondrial and nuclear genetic variation revealed limited gene flow between populations within the North Pacific, including between the 2 island-associated populations in the Hawaiian Archipelago. mtDNA suggests strong maternally based fidelity to populations, and even to social clusters within populations. The phylogeographic pattern suggests that the MHI and NWHI populations have a common colonization history and that they have been closed to immigration from the offshore realm for long enough to evolve new haplotypes *in situ*. The fact that the populations share their most common haplotype but that each population has its own unique haplotype, each apparently derived from the common haplotype, suggests that the 2 island-associated populations may have been isolated from each other for a long time as well. Haplotype frequencies differ significantly between the MHI and NWHI populations. However, this result is due to the absence of haplotype 2 from the NWHI population. Nothing is known regarding the social structure of the NWHI population. If it is comprised of distinct social clusters, like the MHI population (Baird et al. 2012), it is likely that most of our NWHI samples come from a single social cluster since they were collected from encounters that are linked by association (Baird et al. 2013). Given that the mtDNA differentiation between the NWHI and MHI populations is comparable to that seen between social clusters in the MHI population, more samples are needed from the NWHI population to confirm the absence of haplotype 2 from that population and allow more robust estimation of the level of mtDNA differentiation between it and the MHI population.

The nuclear data set also indicates limited gene flow between the MHI and NWHI populations, with the NWHI animals representing a separate population. The STRUCTURE analyses strongly supported the separation of the MHI and NWHI, but fail to identify the social clusters within the MHI, indicating that differentiation between the MHI and NWHI populations is considerably greater than between MHI social clusters. The difference in allele frequencies between these 2 populations was highly significant. The F_{ST} and F'_{ST} estimates between them were among the highest of any comparison we made and were approximately 3 times larger than the estimates between MHI social clusters (Table 4).

In contrast to the phylogeographic structure revealed by the mtDNA data set, analyses of the nuclear data set that did not require *a priori* stratification of the samples suggest that the NWHI is as or more differentiated from the MHI as it is from the remainder of the North Pacific. STRUCTURE groups

the NWHI samples with all offshore samples, to the exclusion of the MHI. Similarly, the Mantel test indicates that all North Pacific samples except for the MHI conform to an isolation-by-distance model, again suggesting that the NWHI population has greater genetic connectivity to offshore animals than to the MHI population.

The different patterns of differentiation revealed by the mtDNA and nucDNA data sets may reflect a difference between the colonization history and contemporary gene flow patterns within the Hawaiian Archipelago. The fact that the MHI and NWHI share a mitochondrial lineage suggests that they have a shared colonization history, either of one population founding the other, or both being founded simultaneously by related individuals. Strong site fidelity and an absence of immigration, either due to social exclusion of immigrants or reduced fitness of immigrants in the novel insular environment, could have prevented the mixing of haplotypes and allowed the evolution of new haplotypes *in situ*. However, the nuclear analyses that grouped the NWHI with offshore samples rather than MHI suggests that, in the nuclear genome, the NWHI population is more genetically similar to the offshore populations than it is to the MHI population. Discordant patterns between maternally and biparentally inherited markers have been attributed to colonization histories that differ from patterns of ongoing, male-mediated gene flow in other studies as well (Comes and Abbot 1998; Gomez et al. 2002). Similarly, strong social structure and small population size can result in the shared colonization history reflected in the mitogenome of 2 populations being quickly obscured in their nuclear genomes (e.g., Northeast Atlantic bottlenose dolphins; Mirimin et al. 2011).

The apparent similarity in the nuclear genome between the NWHI population and the rest of the North Pacific should be viewed with caution, however, due to the small sample sizes from these strata. The MHI population is much better sampled than any other region, comprising half of our sample set. The fact that we have sampled nearly two-thirds of that population may enable STRUCTURE to identify that very well-sampled gene pool as a unique group, while small sample sizes from the remaining strata may result in insufficient power to detect structure in the remainder of the data set. Nonetheless, the fact that the NWHI animals clustered with the offshore samples instead of with MHI animals suggests that they are at least as different from the MHI animals as they are from the offshore animals. Additional samples from the NWHI population are needed in order to better evaluate its differentiation from the other strata.

There was one sample in our data set that deviated from the strong pattern of geographic concordance in the mitochondrial data set—a male sampled off the coast of Hawai'i Island that possessed a haplotype also detected off the coast of Australia and not closely related to the other insular haplotypes. This animal assigned more strongly to the eastern Pacific than to the MHI population. Though its *P* value from the exclusion test was not quite statistically significant, it is the only animal from that encounter that has never again been sighted or rebiopsied. These results suggest that it may not be a member of the MHI population, but rather an offshore animal that was interacting with the MHI population. If that is the case, it is the first documented interaction between the populations.

Our estimates of F_{ST} and F'_{ST} were not entirely consistent with the patterns suggested by STRUCTURE and Mantel tests, instead showing that differentiation was highest in comparisons involving the NWHI and lowest in comparisons involving the CNP. However, the magnitude of differentiation between populations, as measured by *F*-statistics, depends not only on the rate of gene flow between them but also on their effective population sizes. Consequently, the relative magnitude of F_{ST} and F'_{ST} for different population pairs tells us little about the relative rates of gene flow when those pairs differ substantially in abundance.

Evidence of Sex-Biased Dispersal

For populations with ongoing gene flow that is not sex-biased, we would expect F_{ST} (or Φ_{ST}) for mtDNA markers to be 4 times larger than for nucDNA markers due to the differences in inheritance modes (Larsson et al. 2009). The ratio of mtDNA Φ_{ST} to nucDNA F_{ST} between MHI and NWHI roughly conformed to this expectation. However, for all other comparisons the ratio was substantially higher, ranging from 8 to 55. For Φ_{ST} versus F'_{ST}, the ratio was 1.1 for MHI versus NWHI and ranged from 1.8 to 14 for all other comparisons. Ratios differing from 4 are not unexpected, given the high variance in estimates of F_{ST}. Nonetheless, the discrepancy between estimates of differentiation for the 2 data sets indicates some degree of male-mediated gene flow between these populations. The strong phylogeographic pattern in the mtDNA data set and the absence of haplotype 2 from one of the MHI social clusters indicates that both sexes exhibit strong fidelity to their natal social cluster. If mating occurs between populations and social clusters, the mtDNA pattern would be maintained while allowing for gene flow in the nuclear genome. This type of social organization has been observed in fish-eating killer whales in the nearshore waters of the temperate North Pacific (Ford et al. 2011; Parsons et al. 2013). These killer whales are highly social, with both sexes exhibiting strong philopatry to natal social groups, known as "pods" (Baird and Whitehead 2000; Ford et al. 2011). This social structure has resulted in very low levels of genetic variation, with each pod possessing a single mitochondrial haplotype (Hoelzel et al. 1998; LeDuc et al. 2008; Parsons et al. 2013). Pods from the same population share identical or similar haplotypes, whereas different populations typically do not share haplotypes (Parsons et al. 2013). Mating occurs both within and between pods and, at lower rates, between populations (Ford et al. 2011), resulting in lower levels of nuclear than mitochondrial differentiation between populations (Parsons et al. 2013).

Unlike killer whales, in which the pattern of pod-specific haplotypes is consistent across most of the North Pacific, our results indicate that phylogeographic structuring of false killer whale haplotypes occurs primarily within the Hawaiian Archipelago (Figure 3). In fact, the most common haplotype

in the North Pacific (haplotype 9) was detected in animals ranging from the coast of Mexico to the northwestern edge of the Hawaiian EEZ and is more similar to the haplotype from the Indian Ocean than it is to the Hawaiian insular haplotypes. This pattern suggests that there is greater site fidelity within the Hawaiian Archipelago than there is in the remainder of the Pacific.

Though the difference in estimates of differentiation for the mtDNA versus nucDNA data set is greater than expected in the absence of sex-biased gene flow, F_{ST} estimates based on microsatellites are known to be downwardly biased due to their high levels of diversity and their mutation model, which results in high homoplasy (see Meirmans and Hedrick 2011). F'_{ST} attempts to correct for diversity, but in no way corrects for homoplasy (Rousset 1996; Kronholm et al. 2010). The degree of bias associated with estimates of differentiation derived from a microsatellite data set can be assessed by comparison with other nuclear markers, such as single nucleotide polymorphisms (SNPs) or nuclear sequence (Smith et al. 2007; Morin et al. 2012). The lower mutation rates and low level of homoplasy in these markers render them less vulnerable to bias, though many more markers are necessary to achieve comparable statistical power (Morin et al. 2009c). Consequently, it would be useful to further investigate the extent of male-mediated gene flow among North Pacific false killer whale populations using SNP or nuclear sequence loci.

Oceanographic Influences on Differentiation

The evolution of 2 separate island-associated populations in this otherwise offshore species may be driven by the unique habitats associated with the Hawaiian Archipelago. The Hawaiian Archipelago lies in the center of the North Pacific central gyre, an area of low productivity once thought to be largely homogenous (Schmelzer 2000). However, in the last 2 decades oceanographic research has revealed considerable variability around the island chain. The large, steep-sided islands in the MHI disrupt the flow of the prevailing wind and ocean currents, producing nearshore upwelling and persistent eddies on the leeward sides of the islands that may increase productivity (Doty and Oguri 1956; Seki et al. 2002). Productivity is likely further enhanced by runoff from the islands, which brings nutrients into the nearshore environment. The MHIs are surrounded by large areas of shallow water habitat, which support diverse reef and benthic communities not present in the offshore waters. All of these factors combine to result in an abrupt habitat discontinuity between the insular environment and the immediately adjacent offshore realm. The resource specialization and behavioral adaptations necessary to efficiently exploit such different habitats have been proposed as important mechanisms driving population differentiation in other cetaceans (Hoelzel et al. 1998; Natoli et al. 2005; Möller et al. 2007; Martien et al. 2012) and may explain the evolution of insular populations of false killer whales and other marine mammal species around the MHI (Andrews et al. 2010; Martien et al. 2012).

Like the MHI, the NWHI encompass large areas of shallow water habitat. However, the dramatic difference in landmass between the MHI and NWHI and the more northerly location of the NWHI result in considerable oceanographic differences between the 2 biomes. The mountainous islands of the MHI have a combined land area of approximately 16 000 km^2, whereas the NWHI, which consist of submerged and nearly submerged atolls and islands, have a total land area of only 8 km^2 (Baker et al. 2011). Consequently, many of the factors contributing to increased productivity in the nearshore waters immediately surrounding the MHI are absent in the NWHI. Furthermore, the NWHI are located in an area where cool, vertically mixed, high surface chlorophyll water from the subarctic mixes seasonally with warmer, vertically stratified, low surface chlorophyll water of the North Pacific central gyre (Polovina et al. 1994, 2001). This mixing zone, known as the Transition Zone Chlorophyll Front (TZCF), results in much higher average productivity in offshore waters surrounding the NWHI (Schmelzer 2000; Polovina et al. 2001, 2008), reducing the habitat differences between nearshore and offshore waters in that part of the archipelago. The southern extent of the TZCF varies over seasonal and decadal scales (Polovina et al. 1994, 2001, 2008), resulting in high interannual variability in oceanographic conditions that has been linked to variability in the abundance and survival rates of primary producers, reef fishes, spiny lobsters, seabirds, and Hawaiian monk seals in the NWHI (Polovina et al. 1994; Schmelzer 2000; Baker et al. 2007).

Differences between the MHI and NWHI have been observed in the population structure and social organization of several species of marine mammals (Andrews et al. 2010; Baker et al. 2011; Baird et al. 2013). Andrews et al. (2010) observed less genetic structuring and higher group stability in spinner dolphins in the NWHI than the MHI, both of which they attributed to differences in the availability of resting habitat and prey resources. The endangered Hawaiian monk seal also exhibits genetic differentiation and dramatically different population trends between the NWHI and MHI (Baker et al. 2007, 2011). Similarly, restricted gene flow between MHI and NWHI false killer whale populations may reflect differences in foraging strategies and hunting methods needed in order to exploit resources in the different environments. The cooperative hunting methods employed by MHI false killer whales may result in reduced fitness or social exclusion of would-be immigrants, as has been suggested for other species (Baird and Whitehead 2000).

Summary

Our results corroborate the findings of previous photo-identification (Baird et al. 2008), satellite tagging (Baird et al. 2010, 2012), and genetic (Chivers et al. 2007) studies that demonstrated the ecological and genetic uniqueness of MHI false killer whales. The phylogeographic signal revealed by mtDNA indicates strong fidelity of both sexes over a long period of time. The apparent *in situ* evolution of haplotypes not found in the offshore population combined with the unique habitat in which this population resides suggest that this population

may have evolved local adaptations to their particular insular habitat, a possibility that should be further investigated. nucDNA suggest that the MHI population is as different from the NWHI population as it is from offshore animals, while the failure of STRUCTURE to delineate the social clusters within the MHI suggests that the social affiliations captured by the photo-identification data are more fluid than population-level distinctions. The recent severe decline in the abundance of the MHI population and the unique anthropogenic threats faced by marine species in the MHI (Baker et al. 2011) raise concern regarding the persistence of this population.

Supplementary Material

Supplementary material can be found at http://www.jhered.oxfordjournals.org/.

Funding

National Marine Fisheries Service (Office of Protected Resources, Pacific Island Regional Office, Southwest Fisheries Science Center's Marine Mammal and Turtle Division, Pacific Islands Science Center Protected Species Division); the Wild Whale Research Foundation; and the U.S. Navy (grant numbers N000141010686 from the Office of Naval Research and N00244-10-1-0048 from the Naval Postgraduate School).

Acknowledgements

We thank Drs C. Anderson, Ministry of Fisheries and Agriculture, Republic of the Maldives; S. Jaaman, University of Malaysia; S. Hung, Agriculture, Fisheries and Conservation Department, Hong Kong; J. Robbins, Provincetown Center for Coastal Studies; T. Jefferson and W. F. Perrin, SWFSC; and the NMFS, Pacific Islands Regional Office for providing samples to us. We are also grateful for the dedication and talent of many people who participated in the collection of biopsy samples during the research cruises that made this study possible, including the chief scientists, officers, and crews of the NOAA ships *David Starr Jordan*, *McArthur*, *McArthur II*, *Malcolm Baldridge*, *Oscar Elton Sette*, and the UNOLS ship *Endeavor*. We thank the Island Marine Institute, Maui, Hawai'i, J. Mobley of the University of Hawai'i, and the Wild Whale Research Foundation, Hawai'i, for providing research vessels for sampling in nearshore areas around Hawai'i, as well as J. Aschettino for assistance in the field. Samples collected in the United States were taken under NMFS permits 1026/689424, 774–1714, and 14097. Procedures for ensuring animal welfare during biopsy sampling were approved as part of the Scientific Research permits approval process. Samples collected outside US jurisdiction were imported under CITES Import permits. All data were generated in the SWFSC Genetics Laboratory with assistance from A. Konopacki, K. Coultrup, A. Frey, A. Lang, C. LeDuc, P. Morin, A. Rosenberg, and B. Sutton. P. Olson assisted with the review of photographs collected from encounters with false killer whales in the ETP. Thanks to D. Prosperi, who assisted with data management and archiving, and B. Cozzi for bringing to our attention the extirpation of false killer whales from Italian waters. M. Ford, M. Jenson, B. Perrin, P. Rosel, J. Jackson, A. Foote, and an anonymous reviewer provided helpful comments for improving earlier versions of this manuscript.

References

Akaike H. 1974. A new look at the statistical model identification. IEEE Trans Automat Contr. 19:716–723.

Amos W. 1997. Marine mammal tissue sample collection and preservation for genetic analyses. In: Dizon AE, Chivers SJ, Perrin WF, editors. Molecular genetics of marine mammals. Lawrence (KS): Society of Marine Mammalogy. p. 107–113.

Amos W, Hoelzel AR. 1991. Long-term preservation of whale skin for DNA analysis. Rep Int Whal Comm Sp Is. 13:99–103.

Andrews KR, Karczmarski L, Au WW, Rickards SH, Vanderlip CA, Bowen BW, Gordon Grau E, Toonen RJ. 2010. Rolling stones and stable homes: social structure, habitat diversity and population genetics of the Hawaiian spinner dolphin (*Stenella longirostris*). Mol Ecol. 19:732–748.

Baird RW, Gorgone AM, McSweeney DJ, Webster DL, Salden DR, Deakos MH, Ligon AD, Schorr GS, Barlow J, Mahaffy SD. 2008. False killer whales (*Pseudorca crassidens*) around the main Hawaiian Islands: long-term site fidelity, inter-island movements, and association patterns. Mar Mammal Sci. 24:591–612.

Baird RW, Hanson MB, Schorr GS, Webster DL, McSweeney DJ, Gorgone AM, Mahaffy SD, Holzer DM, Oleson EM, Andrews RD. 2012. Range and primary habitats of Hawaiian insular false killer whales: informing determination of critical habitat. Endanger Species Res. 18:47–61.

Baird RW, Oleson EM, Barlow J, Ligon AD, Gorgone AM, Mahaffy SD. 2013. Evidence of an island-associated population of false killer whales (*Pseudorca crassidens*) in the Northwestern Hawaiian Islands. Pacific Sci. 67:513–521.

Baird RW, Schorr GS, Webster DL, McSweeney DJ, Hanson MB, Andrews RD. 2010. Movements and habitat use of satellite-tagged false killer whales around the main Hawaiian Islands. Endanger Species Res. 10:107–121.

Baird RW, Whitehead H. 2000. Social organization of mammal-eating killer whales: group stability and dispersal patterns. Can J Zool. 78:2096–2105.

Baker JD, Harting AL, Wurth TA, Johanos TC. 2011. Dramatic shifts in Hawaiian monk seal distribution predicted from divergent regional trends. Mar Mammal Sci. 27:78–93.

Baker J, Polovina J, Howell E. 2007. Effect of variable oceanic productivity on the survival of an upper trophic predator, the Hawaiian monk seal *Monachus schauinslandi*. Mar Ecol Prog Ser. 346:277–283.

Bandelt HJ, Forster P, Röhl A. 1999. Median-joining networks for inferring intraspecific phylogenies. Mol Biol Evol. 16:37–48.

Beerli P. 2006. Comparison of Bayesian and maximum-likelihood inference of population genetic parameters. Bioinformatics. 22:341–345.

Bradford AL, Forney KA, Oleson EM, Barlow J. 2014. Accounting for subgroup structure in line-transect abundance estimates of false killer whales (*Pseudorca crassidens*) in Hawaiian waters. PLoS One. 9:e90464.

Buchanan FC, Friesen MK, Littlejohn RP, Clayton JW. 1996. Microsatellites from the beluga whale Delphinapterus leucas. Mol Ecol. 5:571–575.

Carretta J V, Oleson E, Weller DW, Lang AR, Forney KA, Baker J, Hanson B, Martien KK, Muto MM, Lowry MS, et al. 2013. U.S. Pacific marine mammal stock assessments: 2012. La Jolla (CA): NOAA Technical Memorandum NMFS-SWFSC-504. p. 385.

Chivers SJ, Baird RW, McSweeney DJ, Webster DL, Hedrick NM, Salinas JC. 2007. Genetic variation and evidence for population structure in eastern North Pacific false killer whales (*Pseudorca crassidens*). Can J Zool. 85:783–794.

Comes HP, Abbot RJ. 1998. The relative importance of historical events and gene flow on the population structure of a Mediterranean ragwort, *Senecio galicus* (Asteraceae). Evolution. 52:355–367.

Darriba D, Taboada GL, Doallo R, Posada D. 2012. jModelTest 2: more models, new heuristics and parallel computing. Nat Methods. 9:772.

Doty MS, Oguri M. 1956. The island mass effect. J du Cons Int pour l'Exploration del a Mer. 22:33–37.

Escorza-Trevino S, Archer FI, Rosales M, Lang AM, Dizon AE. 2005. Genetic differentiation and intraspecific structure of Eastern Tropical Pacific spotted dolphins, *Stenella attenuata*, revealed by DNA analyses. Conserv Genet. 6:587–600.

Evanno G, Regnaut S, Goudet J. 2005. Detecting the number of clusters of individuals using the software STRUCTURE: a simulation study. Mol Ecol. 14:2611–2620.

Excoffier L, Laval G, Schneider S. 2005. Arlequin (version 3.0): an integrated software package for population genetics data analysis. Evol Bioinform Online. 1:47–50.

Fain SR, LeMay JP. 1995. Gender identification of humans and mammalian wildlife species from PCR amplified sex-linked genes. Proc Am Acad Forensic Sci.:34.

Falush D, Stephens M, Pritchard JK. 2003. Inference of population structure using multilocus genotype data: linked loci and correlated allele frequencies. Genetics. 164:1567–1587.

Federal Register. 2012. Endangered and threatened wildlife and plants: endangered status for the main Hawaiian Islands insular false killer whale distinct population segment. Fed Regist. 77:70915–70939.

Fisher RA. 1935. The logic of inductive inference (with discussion). J R Stat Soc. 98:39–82.

Ford MJ, Hanson MB, Hempelmann JA, Ayres KL, Emmons CK, Schorr GS, Baird RW, Balcomb KC, Wasser SK, Parsons KM, et al. 2011. Inferred paternity and male reproductive success in a killer whale (*Orcinus orca*) population. J Hered. 102:537–553.

Forster P, Torroni A, Renfrew C, Röhl A. 2001. Phylogenetic star contraction applied to Asian and Papuan mtDNA evolution. Mol Biol Evol. 18:1864–1881.

Fu YX. 1997. Statistical tests of neutrality of mutations against population growth, hitchhiking and background selection. Genetics. 147:915–925.

Galver LM. 2002. The molecular ecology of spinner dolphins, *Stenella longirostris*: genetic diversity and population structure [Masters thesis]. San Diego (CA):University of California, San Diego. p. 192.

Gemmell NJ, Akiyama S. 1996. An efficient method for the extraction of DNA from vertebrate tissues. Trends Genet. 12:338–339.

Gomez A, Adcock GJ, Lunt DH, Carvalho GR. 2002. The interplay between colonization history and gene flow in passively dispersing zooplankton: microsatellite analysis of rotifer resting egg banks. J Evol Biol. 15:158–171.

Goudet J. 2001. FSTAT, a program to estimate and test gene diversities and fixation indices (version 2.9.3). Available from: http://www2.unil.ch/popgen/softwares/fstat.htm.

Goudet J, Perrin N, Waser P. 2002. Tests for sex-biased dispersal using bi-parentally inherited genetic markers. Mol Ecol. 11:1103–1114.

Guindon S, Gascuel O. 2003. A simple, fast, and accurate algorithm to estimate large phylogenies by maximum likelihood. Syst Biol. 52:696–704.

Guo SW, Thompson EA. 1992. Performing the exact test of Hardy-Weinberg proportion for multiple alleles. Biometrics. 48:361–372.

Hoelzel AR, Dahlheim M, Stern SJ. 1998. Low genetic variation among killer whales (*Orcinus orca*) in the eastern north Pacific and genetic differentiation between foraging specialists. J Hered. 89:121–128.

Hubisz MJ, Falush D, Stephens M, Pritchard JK. 2009. Inferring weak population structure with the assistance of sample group information. Mol Ecol Resour. 9:1322–1332.

Jakobsson M, Rosenberg NA. 2007. CLUMPP: a cluster matching and permutation program for dealing with label switching and multimodality in analysis of population structure. Bioinformatics. 23:1801–1806.

Kronholm I, Loudet O, de Meaux J. 2010. Influence of mutation rate on estimators of genetic differentiation–lessons from Arabidopsis thaliana. BMC Genet. 11:33.

Larsson LC, Charlier J, Laikre L, Ryman N. 2009. Statistical power for detecting genetic divergence - organelle versus nuclear markers. Conserv Genet. 10:1255–1264.

LeDuc RG, Robertson KM, Pitman RL. 2008. Mitochondrial sequence divergence among Antarctic killer whale ecotypes is consistent with multiple species. Biol Lett. 4:426–429.

Longhurst A. 1998. Ecological geography of the sea. San Diego (CA): Academic Press.

Martien KK, Baird RW, Hedrick NM, Gorgone AM, Thieleking JL, McSweeney DJ, Robertson KM, Webster DL. 2012. Population structure of island-associated dolphins: evidence from mitochondrial and microsatellite markers for common bottlenose dolphins (*Tursiops truncatus*) around the main Hawaiian Islands. Mar Mammal Sci. 28:E208–E232.

McKelvey KS, Schwartz MK. 2005. DROPOUT: a program to identify problem loci and samples for noninvasive genetic samples in a capture-mark-recapture framework. Mol Ecol Notes 5:716–718.

Meirmans PG. 2006. Using the AMOVA framework to estimate a standardized genetic differentiation measure. Evolution. 60:2399–2402.

Meirmans PG, Hedrick PW. 2011. Measuring genetic differentiation: F_{ST} and related measures. Mol Ecol Resour. 11:5–18.

Miller SA, Dykes DD, Polesky HF. 1988. A simple salting out procedure for extracting DNA from human nucleated cells. Nucleic Acids Res. 16:1215.

Mirimin L, Miller R, Dillane E, Berrow SD, Ingram S, Cross TF, Rogan E. 2011. Fine-scale population genetic structuring of bottlenose dolphins in Irish coastal waters. Anim Conserv. 13:1–12.

Möller LM, Wiszniewski J, Allen SJ, Beheregaray LB. 2007. Habitat type promotes rapid and extremely localised genetic differentiation in dolphins. Mar Freshw Res. 58:640–648.

Morin PA, Archer FI, Pease VL, Hancock-Hanser BL, Robertson KM, Huebinger RM, Martien KK, Bickham JW, George JC, Postma LD, et al. 2012. Empirical comparison of single nucleotide polymorphisms and microsatellites for population and demographic analyses of bowhad whales. Endanger Species Res. 19:129–147.

Morin PA, Leduc RG, Archer FI, Martien KK, Huebinger R, Bickham JW, Taylor BL. 2009a. Significant deviations from Hardy-Weinberg equilibrium caused by low levels of microsatellite genotyping errors. Mol Ecol Resour. 9:498–504.

Morin PA, Manaster C, Mesnick SL, Holland R. 2009b. Normalization and binning of historical and multi-source microsatellite data: overcoming the problems of allele size shift with allelogram. Mol Ecol Resour. 9:1451–1455.

Morin PA, Martien KK, Archer FI, Cipriano F, Steel D, Jackson J, Taylor BL. 2010. Applied conservation genetics and the need for quality control and reporting of genetic data used in fisheries and wildlife management. J Hered. 101:1–10.

Morin PA, Martien KK, Taylor BL. 2009c. Assessing statistical power of SNPs for population structure and conservation studies. Mol Ecol Resour. 9:66–73.

Morin PA, Nestler A, Rubio-Cisneros NT, Robertson KM, Mesnick SL. 2005. Interfamilial characterization of a region of the ZFX and ZFY genes facilitates sex determination in cetaceans and other mammals. Mol Ecol. 14:3275–3286.

Natoli A, Birkun A, Aguilar A, Lopez A, Hoelzel AR. 2005. Habitat structure and the dispersal of male and female bottlenose dolphins (*Tursiops truncatus*). Proc Biol Sci. 272:1217–1226.

Oleson EM, Boggs CH, Forney KA, Hanson MB, Kobayashi DR, Taylor BL, Wade PR, Ylitalo GM. 2010. Status review of Hawaiian Insular false killer whales (*Pseudorca crassidens*) under the Endangered Species Act. Honolulu (HI): NOAA Technical Memorandum NMFS-PIFSC-22. p. 139.

Oremus M, Gales R, Dalebout ML, Funahashi N, Endo T, Kage T, Steel D, Baker CS. 2009. Worldwide mitochondrial DNA diversity and phylogeography of pilot whales (*Globicephala* spp.). Biol J Linn Soc. 98:729–744.

Paetkau D, Calvert W, Stirling I, Strobeck C. 1995. Microsatellite analysis of population structure in Canadian polar bears. Mol Ecol. 4:347–354.

Paetkau D, Slade R, Burden M, Estoup A. 2004. Direct, real-time estimation of migration rate using assignment methods: a simulation-based exploration of accuracy and power. Mol Ecol Notes. 6:288–295.

Palumbi SR, Martin AP, Romano S, McMillan WO, Stice L, Grawbowski G. 1991. A simple fool's guide to PCR. Honolulu (HI): University of Hawai'i.

Parsons KM, Durban JW, Burdin AM, Burkanov VN, Pitman RL, Barlow J, Barrett-Lennard LG, LeDuc RG, Robertson KM, Matkin CO, et al. 2013. Geographic patterns of genetic differentiation among killer whales in the northern North Pacific. J Hered. 104:737–754.

Peakall R. 1995. Evolutionary implications of allozym and RAPD variation in diploid populations of dioecious buffalograss *Buchloe dactyloides*. Mol Ecol. 4:135–147.

Peakall R, Smouse PE. 2006. GENALEX 6: genetic analysis in Excel. Population genetic software for teaching and research. Mol Ecol Notes. 6:288–295.

Perneger TV. 1998. What's wrong with Bonferroni adjustments. BMJ. 316:1236–1238.

Peters JL, Bolender KA, Pearce JM. 2012. Behavioural vs. molecular sources of conflict between nuclear and mitochondrial DNA: the role of male-biased dispersal in a Holarctic sea duck. Mol Ecol. 21:3562–3575.

Piry S, Alapetite A, Cornuet JM, Paetkau D, Baudouin L, Estoup A. 2004. GENECLASS2: a software for genetic assignment and first-generation migrant detection. J Hered. 95:536–539.

Polovina JJ, Chai F, Howell E a., Kobayashi DR, Shi L, Chao Y. 2008. Ecosystem dynamics at a productivity gradient: A study of the lower trophic dynamics around the northern atolls in the Hawaiian Archipelago. Prog Oceanogr. 77:217–224.

Polovina JJ, Howell E, Kobayashi DR, Seki MP. 2001. The transition zone chlorophyll front, a dynamic global feature defining migration and forage habitat for marine resources. Prog Oceanogr. 49:469–483.

Polovina JJ, Mitchum GT, Graham NE, Craig MP, Demartini EE, Flint EN. 1994. Physical and biological consequences of a climate event in the central North Pacific. Fish Oceanogr. 3:15–21.

Polzin, T, Daneschmand SV. 2003. On Steiner trees and minimum spanning trees in hypergraphs. Oper Res Lett 31:12–20.

Pritchard JK, Stephens M, Donnelly P. 2000. Inference of population structure using multilocus genotype data. Genetics. 155:945–959.

Queller DC, Goodnight KF. 1989. Estimating relatedness using genetic-markers. Evolution (N. Y). 43:258–275.

R Development Core Team. 2011. R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing.

Raymond M, Rousset F. 1995. Genepop (Version-1.2) - Population-Genetics Software for Exact Tests and Ecumenicism. J Hered. 86:248–249.

Reeves RR, Leatherwood S, Baird RW. 2009. Evidence of a possible decline since 1989 in false killer whales (*Pseudorca crassidens*) around the main Hawaiian Islands. Pacific Sci. 63:253–261.

Reeves RR, Notarbartolo di Sciara G (compilers and editors). 2006. The status and distribution of cetaceans in the Black Sea and Mediterranean Sea. 5–7 March 2006, Monaco. Malaga, Spain: IUCN Centre for Mediterranean Cooperation. p. 142.

Reilly SB. 1990. Seasonal changes in distribution and habitat differences among dolphins in the eastern tropical Pacific. Mar Ecol Progress Series. 66:1–11.

Richard KR, Whitehead H, Wright JM. 1996. Polymorphic microsatellites from sperm whales and their use in the genetic identification of individuals from naturally sloughed pieces of skin. Mol Ecol. 5:313–315.

Rodríguez-Ramilo ST, Wang J. 2012. The effect of close relatives on unsupervised Bayesian clustering algorithms in population genetic structure analysis. Mol Ecol Resour. 12:873–884.

Roff DA, Bentzen P. 1989. The statistical analysis of mitochondrial DNA polymorphisms: chi 2 and the problem of small samples. Mol Biol Evol. 6:539–545.

Rooney AP, Merritt DB, Derr JN. 1999. Microsatellite diversity in captive bottlenose dolphins (*Tursiops truncatus*). J Hered. 90:228–231.

Rosel PE, Dizon AE, Heyning JE. 1994. Genetic analysis of sympatric morphotypes of common dolphins (genus *Delphinus*). Mar Biol. 119:159–167.

Rosel PE, Forgetta V, Dewar K. 2005. Isolation and characterization of twelve polymorphic microsatellite markers in bottlenose dolphins (*Tursiops truncatus*). Mol Ecol Notes 5:830–833.

Rosenberg NA. 2004. Distruct: a Program for the Graphical Display of Population Structure. Mol Ecol Notes 4:137–138.

Rousset F. 1996. Equilibrium values of measures of population subdivision for stepwise mutation processes. Genetics. 142:1357–1362.

Rousset F. 2008. genepop'007: a complete re-implementation of the genepop software for Windows and Linux. Mol Ecol Resour. 8:103–106.

Saiki RK, Gelfand DH, Stoffel S, Scharf SJ, Higuchi R, Horn GT, Mullis KB, Erlich HA. 1988. Primer-directed enzymatic amplification of DNA with a thermostable DNA polymerase. Science. 239:487–491.

Schmelzer I. 2000. Seals and seascapes: covariation in Hawaiian monk seal subpopulations and the oceanic landscape of the Hawaiian Archipelago. J Biogeogr. 27:901–914.

Schwarz G. 1978. Estimating the dimension of a model. Ann Stat. 6:461–464.

Seki MP, Lumpkin R, Flament P. 2002. Hawai'i cyclonic eddies and blue marlin catches: the case study of the 1995 Hawaiian International Billfish Tournament. J Oceanogr. 58:739–745.

Smith CT, Antonovich A, Templin WD, Elfstrom CM, Narum SR, Seeb LW. 2007. Impacts of marker class bias relative to locus-specific variability on population inferences in Chinook Salmon: a comparison of single-nucleotide polymorphisms with short tandem repeats and allozymes. Trans Am Fish. Soc. 136:1674–1687.

Smouse PE, Peakall R. 1999. Spatial autocorrelation analysis of individual multiallele and multilocus genetic structure. Heredity (Edinb). 82(Pt 5):561–573.

Stanzani AL, Piermarocchi C. 1992. Cattura di alcuni individui di Pseudorca crassidens (Owen, 1846) in Adriatico. Atti della Soc Ital di Sci Nat e del Mus Civ di Stor Nat di Milano. 133:89–95.

Taberlet P, Luikart G. 1999. Non-invasive genetic sampling and individual identification. Biol J Linn Soc. 68:41–55.

Tajima F. 1989. Statistical method for testing the neutral mutation hypothesis by DNA polymorphism. Genetics. 123:585–595.

Tamura K, Nei M. 1993. Estimation of the number of nucleotide substitutions in the control region of mitochondrial DNA in humans and chimpanzees. Mol Biol Evol. 10:512–526.

Valsecchi E, Amos W. 1996. Microsatellite markers for the study of cetacean populations. Mol Ecol. 5:151–156.

Van Oosterhout C, Hutchinson WF, Willis DPM, Shipley P. 2004. MICRO-CHECKER: software for identifying and correcting genotyping errors in microsatellite data. Mol Ecol Notes. 4:535–538.

Weir BS, Cockerham CC. 1984. Estimating F-Statistics for the analysis of population-structure. Evolution. 38:1358–1370.

Wright S. 1931. Evolution in Mendelian populations. Genetics. 16:97–159.

Received August 22, 2013; First decision October 7, 2013, Accepted March 28, 2014

Corresponding editor: Jennifer Jackson