Unexpected patterns of global population structure in melon-headed whales *Peponocephala electra*

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ABSTRACT: Foraging specialization, environmental barriers, and social structure have driven the development of strong genetic differentiation within many marine species, including most of the large dolphin species commonly referred to as 'blackfish' (subfamily Globicephalinae). We used mitochondrial sequence data (mtDNA) and genotypes from 14 nuclear microsatellite loci (nDNA) to examine patterns of genetic population structure in melon-headed whales *Peponocephala electra* (MHWs), poorly known members of the blackfish family for which genetic structuring is unknown. MHWs are globally distributed in tropical and subtropical waters, and have formed resident populations around oceanic islands. They frequently mass strand, suggesting strong social cohesion within groups. Based on these characteristics, we hypothesized that MHWs would exhibit strong regional genetic differentiation, similar to that observed in other members of the Globicephalinae subfamily. Instead we found only moderate differentiation (median mtDNA Φ_{ST} = 0.204, median nDNA F_{ST} = 0.012) among populations both within and between ocean basins. Our results suggest that populations of MHWs that are resident to oceanic islands maintain a higher level of genetic connectivity than is seen in most other blackfish. MHWs may be more behaviorally similar to delphinids from the Delphininae subfamily (particularly the spinner dolphin *Stenella longirostris*), which are known to form coastal and island-associated resident populations that maintain genetic connectivity either through occasional long-distance dispersal or gene flow with larger pelagic populations. Our results suggest that differences in social organization may drive different patterns of population structure in social odontocetes.

KEY WORDS: Genetic structure · Social structure · Differentiation · Oceanic islands · Hawai'i · Cetacean

INTRODUCTION

Despite a lack of obvious barriers to movement, many broadly distributed marine species show a high degree of genetic structure, sometimes on surprisingly small spatial scales (e.g. Fontaine et al. 2007, Martien et al. 2014, Viricel & Rosel 2014, Gaos et al. 2016). Many potential drivers of population structure within the marine realm have been identified, including resource and habitat specialization, physiographic and oceanographic barriers, and social organizations that limit gene flow between groups. The patterns of genetic structuring and the scales at which it occurs varies widely among species, and sometimes even between populations of the same species (e.g. Bérubé et al. 1998, Andrews et al. 2010,

Gaos et al. 2016, Viricel et al. 2016). Nonetheless, because closely related species often share similar life history and behavioral traits, broadly similar patterns of genetic structure often emerge among them. For example, the cetacean subfamily Globicephalinae, commonly referred to as 'blackfish,' is comprised of several species of large, dark-colored dolphins, including killer whales *Orcinus orca*, false killer whales *Pseudorca crassidens*, pygmy killer whales *Feresa attenuata*, long-finned pilot whales *Globicephala melas*, and short-finned pilot whales *G. macrorhynchus*. Each of these species exhibits strong social structure and deep divergences within the mitochondrial genome, both within and between ocean basins (Bigg et al. 1987, Amos et al. 1993, Baird et al. 2008, 2012, McSweeney et al. 2009, Oremus et al. 2009, Morin et al. 2010a, Parsons et al. 2013, Martien et al. 2014).

Melon-headed whales *Peponocephala electra* (MHWs) are poorly known members of the blackfish subfamily. However, much of what we do know about MHWs suggests that they might also form strongly differentiated regional populations (Brownell et al. 2009b, Aschettino et al. 2012). They are globally distributed in oceanic habitats of tropical and subtropical waters, particularly in continental and insular slope waters between 20°S and 20°N (Perryman 2002). MHWs also mass strand frequently (Brownell et al. 2009b), a recurring pattern observed in highly social species, such as pilot whales (Olson & Reilly 2002). A near mass stranding event (i.e. animals that would have stranded were it not for human intervention) in Hawai'i (USA) associated with a naval exercise utilizing mid-frequency sonar (Southall et al. 2006, Brownell et al. 2009b), and a mass stranding in Madagascar associated with a multibeam sonar survey (Southall et al. 2013) both raise concern that this species may be particularly vulnerable to anthropogenic sounds.

MHWs are typically seen far offshore in deep water. However, observational, photo-identification, and anecdotal evidence suggests the presence of island-associated populations of MHWs around Palmyra Atoll and the Marquesas Islands in French Polynesia (Brownell et al. 2009b), the Hawaiian Archipelago (Aschettino et al. 2012), and around Mayotte in the Mozambique Channel (Kiszka et al. 2011). The best-studied populations are those around the main Hawaiian Islands. Aschettino et al. (2012) used social network analysis of photo-identification data to identify and describe the ranges of 2 populations. The larger of these, referred to as the Hawaiian Islands (HI) population, has an estimated abundance of 5794

individuals (CV = 0.20; Aschettino 2010). It ranges in deep waters (>1000 m) among all of the main Hawaiian Islands as well as into offshore waters to at least several hundred kilometers offshore (Woodworth et al. 2012, Baird 2016). In contrast, the 'Kohala resident population', estimated to number only 447 individuals (CV = 0.12; Aschettino 2010), is generally restricted to the small shallow-water shelf (median sighting depth of 381 m) off the northwestern coast of Hawai'i Island (Fig. 1; Baird 2016). Although the ranges of these populations partially overlap, individuals from the 2 populations have never been observed together despite photo-identification records dating back more than 2 decades. A Bayesian analysis of the sighting records for these 2 populations showed that the dispersal rate between them is low enough to render them demographically independent (Aschettino et al. 2012).

Although the distribution of MHWs, their apparent strong social structure, and the existence of island-associated populations all suggest that they may have patterns of genetic differentiation similar to those of other blackfish, there are several aspects of MHW behavior that distinguish them from the rest of the subfamily. They are typically seen in large aggregations, with mean group sizes in the hundreds (Brownell et al. 2009b, Hamilton et al. 2009, Kiszka et al. 2011, Baird et al. 2013). In contrast, mean group sizes of false killer whales, short- and long-finned pilot whales, pygmy killer whales, and killer whales are <30 (Amos et al. 1991, Baird & Dill 1996, Ottensmeyer & Whitehead 2003, Baird et al. 2008, 2013, Hamilton et al. 2009). Furthermore, near some oceanic islands MHWs have been documented undertaking regular daily shoreward–offshore movements. During the day they rest in large groups near the shelf/insular slopes or just offshore of barrier reefs, while at night they break off into smaller groups and move to deep offshore water to forage (Brownell et al. 2009b). This daily movement pattern is very similar to that seen in spinner dolphins *Stenella longirostris* around oceanic islands (Lammers 2004, Oremus et al. 2007), where it is thought to reduce predation risk from open-ocean sharks (Kiszka et al. 2015). However, this behavior has never been observed for any other species of blackfish.

Because MHWs have never been studied genetically, it is unknown whether these behavioral differences from other blackfish will result in different patterns of genetic structure. We hypothesized that the strong social structure and fine-scale population structure identified in previous observational and

Fig. 1. Melon-headed whale *Peponocephala electra* samples from the Hawaiian Islands (USA) population (closed circles) and the Kohala resident population (open triangles). Inset: global sampling distribution
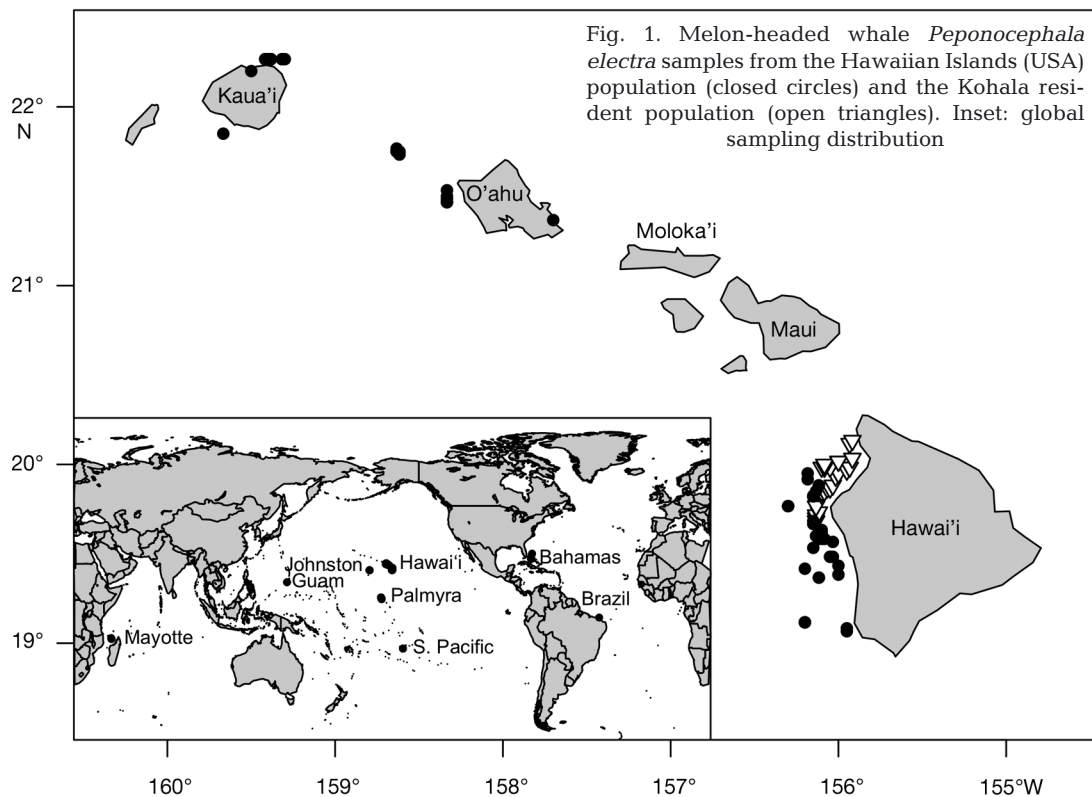
photo-identification studies would translate into strong genetic differentiation at the regional and global scale, and low mitochondrial haplotypic diversity, similar to what is seen in other blackfish. We tested this hypothesis using mitochondrial (mtDNA) sequence data and genotypes from 14 nuclear microsatellite loci (nDNA) from multiple putative populations from the Pacific Ocean. We also compared the Pacific samples to a small set of samples from the Atlantic and Indian Oceans to look for evidence of strong mitochondrial divergence between ocean basins, as has been described in other species of blackfish.

MATERIALS AND METHODS

Sample set

Our sample set consisted of 232 samples collected from animals biopsied at sea (n = 225), stranded dead on shore (n = 6), or from sloughed skin (n = 1). All samples were part of the NMFS Marine Mammal and Sea Turtle Research (MMASTR) Collection, where they were preserved in either ethanol or salt-saturated DMSO and frozen at −20°C, or they were frozen at −80°C without preservative.

We stratified samples geographically and further stratified the Hawaiian samples into the 2 populations identified by Aschettino et al. (2012; Fig. 1). Hawaiian samples were assigned to the Kohala resident population if they were from animals known to be members of that population based on photo-identification data or if they were sampled from a group containing known members of that population. All other Hawaiian samples were assigned to the Hawaiian Islands population. There were no instances of animals having been sighted with members of both populations.

Laboratory processing

Genomic DNA was extracted using a lithium chloride protocol (Gemmell & Akiyama 1996), sodium chloride protocol (Miller et al. 1988), or Qiagen DNeasy Blood and Tissue Kit (no. 69506). The 5′ end of the hypervariable mtDNA control region was amplified and sequenced in 2 parts on an Applied Biosystems 3730 sequencer. The PCR cycling profile and sequencing primers were as described by Martien et al. (2014). Sequences were assembled and aligned using SEQED version 1.0.3 (ABI) and Sequencher software (versions 4.1 and 4.8; Gene Codes), resulting in final sequences 961 bp long.

We genotyped the samples at 14 dinucleotide microsatellite loci: Ttr11, Ttr58, and TtrRC11, derived from bottlenose dolphin *Tursiops truncatus* (Rosel et al. 2005); KWM12at and KWM2at, derived from *Orcinus orca* (Hoelzel et al. 1998); SW19t (Richard et al. 1996), EV1t, and EV14t (Valsecchi & Amos 1996), all derived from sperm whale *Physeter macrocephalus*; SL125t and SL849t, derived from *Stenella longirostris* (Galver 2002); D17t, derived from beluga whale *Delphinapterus leucas* (Buchanan et al. 1996); SAM25t, derived from North Atlantic right whale *Eubalaena glacialis* (Waldick et al. 1999); and GATA53, derived from humpback whale *Megaptera novaeangliae* (Palsbøll et al. 1997). DNA was amplified using the protocols and PCR thermal cycling profiles described by Martien et al. (2014). The annealing temperature and fluorescent tag used for each locus are provided in Table S1 in the Supplement at www.int-res.com/articles/suppl/m577p205_supp.pdf.

Amplicons were visualized on electrophoresis gels and then genotyped on an ABI 3730 genetic analyzer using a commercial internal lane standard (ROX500®; ABI). Allele size calls were made using ABI's GeneMapper (version 4.0) software. Ten percent of samples were randomly chosen for replication in order to estimate error rates. Negative and positive controls were included on every genotyping plate to monitor for contamination and ensure high quality genotypes. We used real-time PCR (Stratagene) of the zinc finger (*ZFX* and *ZFY*) genes to genetically sex the samples (Morin et al. 2005).

Data review

We reviewed all replicate nDNA genotypes for consistency. We calculated the per-allele error rate using only the random replicates. Once data generation was complete, a second, independent genotyper reviewed 20% of allele size calls, chosen at random. Any discrepancies between the original calls and those of the second genotyper were jointly reviewed by both genotypers. Discrepancies that could not be resolved were treated as missing data.

We followed the quality protocols described by Morin et al. (2010b) for the nDNA data set. Samples that were homozygous at ≥75% of the loci, had genotypes at <12 loci, or could not be consistently replicated were excluded from the data set. We used exact tests of Hardy-Weinberg equilibrium (HWE) and tests for heterozygote deficiency to assess each locus for deviation from HWE. We looked for evidence of linkage between pairs of loci using Fisher's

method and the Markov chain method. We used 1000 dememorization steps, 100 batches, and 1000 iterations per batch for both the HWE and linkage disequilibrium tests. We conducted the tests separately for each stratum and then combined p-values across strata to obtain a global p-value for each locus. We identified and excluded from the data set individual genotypes that were highly influential (log-odds >2) in deviations from HWE based on the jackknife procedure described by Morin et al. (2009). All HWE and linkage disequilibrium tests, including the HWE jackknife, were conducted in the R package strataG (R Development Core Team 2014, Archer 2016), which implements the software GENEPOP version 4 (Rousset 2008).

We identified pairs of samples with identical or nearly identical (differing at ≤4 loci) genotypes using the program DROPOUT (McKelvey & Schwartz 2005). For pairs identified by DROPOUT with mismatching loci, we reviewed the raw data and, when necessary, re-genotyped in order to resolve the conflict. To ensure the quality of the mtDNA data, we reviewed all unique haplotypes (i.e. those represented by a single sample) ≥2 times to confirm the accuracy of the sequence.

Data analysis

For mtDNA, we used Arlequin version 3.11 (Excoffier et al. 2005) to estimate haplotypic diversity (H) and nucleotide diversity (π). For the nDNA data set, we used strataG to estimate allelic richness (A_R), observed heterozygosity (H_o), expected heterozygosity (H_e), and the mean number of alleles per locus (N_A). We calculated all diversity estimates both within strata and for the whole data set.

We used the algorithm of Bandelt et al. (1999) as implemented in PopArt (http://popart.otago.ac.nz) to generate a median-joining network of our sequences. To estimate the magnitude of genetic differentiation between pairs of strata, we calculated both F_{ST} and Φ_{ST} (Excoffier et al. 1992) for the mtDNA data set and F_{ST} and F'_{ST} (Meirmans 2006) for the nDNA. F_{ST} is downwardly-biased when diversity within populations is high. F'_{ST} and Φ_{ST} both correct for within-population diversity and therefore do not exhibit this bias (Meirmans & Hedrick 2011). Φ_{ST} was derived from Wright's (1965) formulae and therefore tracks the expected values from those formulae, whereas F'_{ST} does not (Kronholm et al. 2010). Φ_{ST} requires the use of a substitution model to estimate the genetic distance between pairs of haplotypes. We used jMod-

elTest 2.1.1 (Guindon & Gascuel 2003, Darriba et al. 2012) to determine which substitution model best fit our sequence data.

To determine whether the magnitude of mtDNA differentiation that we observed among MHW strata was comparable to that for other blackfish, we compared our Φ_{ST} estimates to published estimates for resident killer whales (Parsons et al. 2013), false killer whales (Martien et al. 2014), long-finned pilot whales (Oremus et al. 2009), and short-finned pilot whales (Oremus et al. 2009, Van Cise et al. 2016) (Table S2 in the Supplement). We grouped the Φ_{ST} values according to whether they were from comparisons of strata within the same ocean basin or between ocean basins and used Mann-Whitney U-tests to determine whether values were significantly lower in MHWs than in other blackfish.

We assessed the statistical significance of genetic differentiation between pairs of strata for both data sets using χ^2 permutation tests (10 000 permutations), which are more powerful for detecting genetic differentiation than the permutation tests associated with F-statistics (Hudson et al. 1992, Goudet et al. 1996). We only included strata with at least 5 samples in the estimates of differentiation. All F_{ST}, F'_{ST}, and χ^2 analyses were conducted in strataG, while Φ_{ST} was calculated in Arlequin v.3.11.

We used the Bayesian clustering algorithm implemented in the program STRUCTURE (Pritchard et al. 2000, Falush et al. 2003) to cluster samples into groups based on their nDNA genotypes. We first included all samples from the Pacific Ocean in the analysis and had STRUCTURE cluster the samples into $K = 1$ to 6 groups. We ran the analysis first with an uninformative prior (LOCPRIOR=F), and a second time with a prior based on sampling location (LOCPRIOR=T), with sampling location set equal to the stratum from which each sample was collected. Because STRUCTURE tends to only identify the highest level of genetic structure within a data set, we re-ran the analyses but only included samples from the Hawaiian Islands and Kohala resident populations and had STRUCTURE cluster the samples into $K = 1$ to 3 groups. We repeated each analysis 5 times for each value of K and compared estimates of ancestry and likelihood across runs to confirm convergence. We compared the mean log-likelihood for each value of K to determine the value best supported by our data. We also calculated ΔK, another statistic used for determining which value of K best fits the data (Evanno et al. 2005). All analyses ran for 1 000 000 steps following a burn-in of 50 000 and an admixture model with correlated frequencies be-

tween groups. We left all other parameters at program defaults. We averaged ancestry coefficients of individuals across replicate runs using CLUMPP (Jakobsson & Rosenberg 2007). All STRUCTURE and CLUMPP analyses were conducted from within the strataG package.

We also used GENELAND (Guillot et al. 2005) to cluster the samples into groups, again using their nDNA genotypes. GENELAND uses a Bayesian clustering algorithm very similar to the one implemented in STRUCTURE. However, the 2 programs differ with respect to the priors that can be used to inform the clustering. Unlike STRUCTURE, which allows the user to specify a prior on the group membership of each sample, GENELAND only assumes that clusters will exhibit some degree of geographic contiguity, as is the case in most natural populations. The geographic coordinates from which samples were collected are incorporated into the analysis, but no *a priori* information regarding group membership is used. The spatial prior used by GENELAND enables it to detect weaker population structure than is detectable with STRUCTURE without biasing it toward identifying the groups the researcher suspects exist (François et al. 2006, Olsen et al. 2014). GENELAND can be instructed to ignore spatial data, in which case the clustering algorithm is the equivalent to that used when STRUCTURE is run with an uninformative prior and no admixture (Guillot et al. 2005).

We ran GENELAND both with and without spatial data. For both analyses we first conducted 10 independent runs in which the number of groups (K) represented by the data was allowed to vary from 1 to 10. These runs were checked for convergence and used to determine the modal value of K. We then ran a final analysis with K fixed at the modal value in order to estimate individual assignments to groups. We post-processed the final run with a burn-in of 2000 iterations in order to determine the posterior assignment probabilities for individuals, using a spatial domain of 200 pixels by 200 pixels. All runs consisted of 500 000 Markov Chain Monte Carlo iterations, with results saved every 100 iterations, and assumed correlated allele frequencies. Because GENELAND uses straight-line distance in its calculations of geographic proximity, it cannot properly account for the movement barriers between ocean basins. Therefore, we limited our GENELAND analyses to only samples collected from the Pacific Ocean, and excluded data from 2 stranded animals due to uncertainty regarding the precise geographic origin of those samples.

RESULTS

Data review

We identified 10 pairs of samples that had identical genotypes. All pairs of duplicates also had matching haplotypes, were of the same sex, and were collected from the same stratum. We eliminated 1 sample from each pair. We excluded 14 samples from the mtDNA data set because we were unable to obtain high-quality sequences from them. There were 28 samples for which we were unable to generate reliable genotypes. These included all samples from Mayotte, from which we were unable to extract sufficient quality DNA for genotyping. We excluded all of these samples from the nDNA data set, as well as 1 sample for which we had insufficient tissue for inclusion in the nDNA laboratory processing. Following all exclusions, our final mtDNA data set included 208 samples, while the nDNA data set included 193 individuals (Tables 1 & 2).

Four samples were identified in the HWE jackknife analysis as outliers, indicating that they likely contained genotyping errors (Morin et al. 2010b). Each of these samples was homozygous at the locus in question (1 at locus EV14t, 3 at GATA53). The genotypes of these samples at these loci were set to null for all analyses. Although 3 loci deviated significantly from HWE in a single stratum, none of these deviations was considered significant when p-values were combined across strata using Fisher's method. Fisher's exact test for linkage disequilibrium was not significant for any pairs of loci.

Genetic diversity

We detected 45 unique haplotypes (Table 1) and 33 variable sites. Haplotypes 1 and 2 dominated the samples, occurring in nearly half (88/208) of the samples. Most other haplotypes (36/45) were unique to a single stratum. Haplotypic diversity (H) was much lower in the Bahamas ($H = 0.473$, $\pi = 0.001$) than in other strata ($H = 0.694–0.933$; $\pi = 0.002–0.005$). The next lowest haplotypic diversity was in the Kohala Resident population. Nucleotide diversity and estimates of diversity in the nDNA data set were similar across all strata. The number of alleles detected at the microsatellite loci ranged from 6 (Ttr58) to 13 (KWM12at and KWM2at; Table S1). The nucleotide substitution model that best fit the data was the model of Tamura & Nei (1993) with gamma = 0.632. All mtDNA sequences were submitted to GenBank

(accession numbers are given in Table 1), and genetic data and strata assignments of all samples are available from the corresponding author.

Genetic structure

The median-joining network did not reveal any phylogeographic structure in the mtDNA data set (Fig. 2). The 2 most common haplotypes are not phylogenetically closely related to each other, and were found in nearly all strata. Although haplotypes 1 and 2 were not found in the Atlantic samples, the 4 haplotypes we identified in the Atlantic (29, 30, 31, and 41) are all more similar to haplotype 2 than haplotype 2 is to haplotype 1.

Nearly all pairs of strata were significantly differentiated in the mtDNA data set (Table 3). The only exceptions were a comparison involving Johnston Atoll and one involving Mayotte, both of which have small sample sizes. Φ_{ST} values were highest between the Bahamas and the other strata, ranging from 0.193 to 0.592. Φ_{ST} values were lowest in comparisons involving the Hawaiian Islands population ($\Phi_{ST} = 0.018–0.363$). Φ_{ST} values were significantly lower than those reported in other studies of blackfish overall and when restricted to only within- or between-ocean basin comparisons (all Mann-Whitney U comparisons, $p < 10^{-10}$; Table S2).

Patterns of differentiation in the nDNA data set were similar to those seen in the mtDNA data set (Table 3). All pairwise comparisons were statistically significant except for those comparing Johnston Atoll to other Pacific strata. Mayotte and the South Pacific did not have enough samples in the nDNA data set to allow pairwise comparisons. F_{ST} values in the nDNA data set were generally low, ranging from 0.026 to 0.050 for comparisons involving the Bahamas and from −0.009 to 0.014 for comparisons among Pacific strata.

When we used an uninformative prior with respect to group membership, STRUCTURE failed to detect any genetic structuring within the nDNA data set, favoring the model with only 1 group ($K = 1$) (Table S3) regardless of whether all Pacific samples (Fig. 3A) or only those from the Hawaiian Islands and Kohala resident populations (Fig. 3D) were included in the analysis.

When we used our *a priori* stratum assignments as a prior and included all Pacific samples, likelihood was maximized when $K = 2$ (Table S3). The strongest individual assignments were for the Palmyra samples, which had a mean assignment of 95.9% to Group 2

Table 1. Melon-headed whale *Peponocephala electra* haplotype frequencies by stratum (sample sizes in parentheses below locations). GenBank accession numbers are given in parentheses in the first column. HI: Hawaiian Islands

Haplotype	Bahamas (11)	Brazil (1)	Guam (2)	Kohala (43)	Johnston (10)	Mayotte (7)	HI (72)	Palmyra (56)	S. Pacific (6)
1 (KT223048)			1	2		1	19	14	
2 (KT223049)				22	1	2	13	13	
3 (KT223050)							4		
4 (KT223051)							6		
5 (KT223052)							1		
6 (KT223053)				1	2		5		
7 (KT223054)					1		2		
8 (KT223055)				9					
9 (KT223056)				2					
10 (KT223057)							4		
11 (KT223058)							1		
12 (KT223059)							4		
13 (KT223060)			1				1		
14 (KT223061)					5		7	2	
15 (KT223062)							1		
16 (KT223063)				1			1		
17 (KT223064)								3	
18 (KT223065)								2	
19 (KT223066)								1	
20 (KT223067)					1				
21 (KT223068)								1	
22 (KT223069)								1	
23 (KT223070)								1	
24 (KT223071)								1	
25 (KT223072)								3	
26 (KT223073)								1	
27 (KT223074)								3	
28 (KT223075)							1		
29 (KT223076)	2								
30 (KT223077)	8								
31 (KT223078)		1							
32 (KT223079)				4					
33 (KT223080)				2					
34 (KT223081)							1		
35 (KT223082)							1		1
36 (KT223083)									2
37 (KT223084)									1
38 (KT223085)									1
39 (KT223086)								1	1
40 (KT223087)								6	
41 (KT223088)	1								
42 (KT223089)						3			
43 (KT223090)						1			
44 (KT223091)								2	
45 (KT223092)								1	

(Fig. 3B, Table 4). All of the other samples were assigned predominantly to Group 2, with the exception of the South Pacific samples, which had approximately equal ancestry from both groups (Fig. 3, Table 4). Likelihood was only slightly lower for the model with 3 groups ($K = 3$), and the individual assignments revealed additional structure, with the Kohala and Hawaiian Islands samples deriving most of their ancestry from different groups (Fig. 3C, Tables 4 & S3).

When only samples from the Hawaiian Islands and Kohala populations were included in the analysis, the highest likelihood was for the model with $K = 1$, but the model with $K = 2$ had only slightly lower likelihood (Table S3). Further, the model with $K = 2$ clearly separated the Kohala samples and the Hawaiian Islands samples into separate groups (Fig. 3E), with the Kohala samples deriving an average of 87.0% of their ancestry from Group 1 and the Hawai-
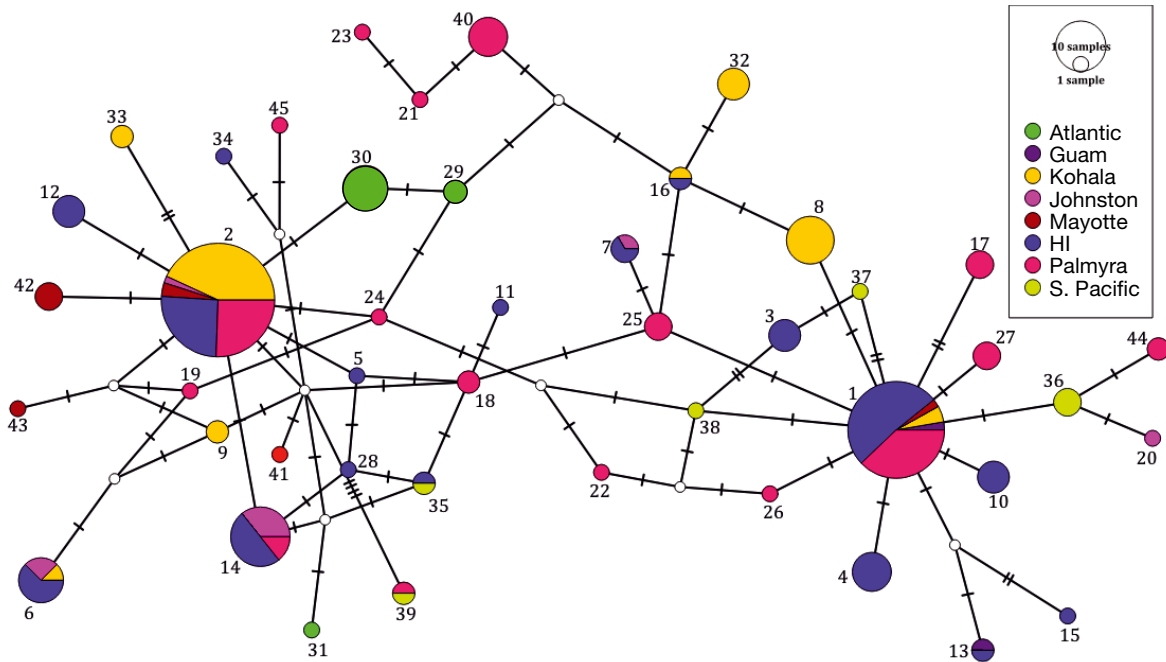
Fig. 2. Median-joining network. Each colored node represents a haplotype of melon-headed whales *Peponocephala electra*, while white nodes represent haplotypes that are hypothesized to exist but were not detected within our data set. Nodes are color-coded according to the stratum in which a given haplotype was detected. Each hash mark on lines connecting nodes represents 1 mutational change. Numbers next to nodes correspond to haplotype numbers in Table 1, and node size is proportional to the frequencies of haplotypes

Table 2. Estimates of genetic diversity in the mtDNA and nDNA data sets of melon-headed whales *Peponocephala electra*, by stratum and overall. Columns show sample size (n), number of haplotypes (N_h), haplotypic diversity (*H*), nucleotide diversity (π), mean number of alleles per locus (N_A), allelic richness (A_R), expected heterozygosity (H_e), observed heterozygosity (H_o), and the overall p-value from an exact test of Hardy-Weinberg equilibrium (HWE p). Allelic richness within strata was based on a minimum sample size of 7, while overall allelic richness was based on a sample size of 186. We did not calculate allelic richness in Guam or the South Pacific due to the low sample size. HI: Hawaiian Islands. na: not analyzed due to insufficient samples

| | mtDNA | | | | nDNA | | | | | |
Stratum	n	N_h	*H*	π	n	N_A	A_R	H_e	H_o	HWE p
Bahamas	11	3	0.473	0.001	10	4.43	0.470	0.652	0.681	0.466
Brazil	1	1	NA	NA	0	na	na	na	na	na
Guam	2	2	1.000	0.002	2	2.50	na	0.619	0.500	0.682
Kohala	43	8	0.694	0.003	41	7.00	0.172	0.713	0.725	0.861
Johnston	10	5	0.756	0.002	10	5.29	0.534	0.708	0.695	0.790
Mayotte	7	4	0.810	0.003	0	na	na	na	na	na
HI	72	17	0.877	0.003	70	8.21	0.120	0.733	0.727	0.591
Palmyra	56	17	0.873	0.003	56	7.71	0.141	0.723	0.726	0.168
S. Pacific	6	5	0.933	0.005	4	4.07	na	0.753	0.661	0.943
Overall	208	45	0.899	0.004	193	10.57	0.055	0.737	0.706	0.891

ian Islands samples deriving 89.0% of their ancestry from Group 2.

GENELAND identified 3 groups within our Pacific samples, regardless of whether we incorporated spatial information into the analysis (Table 5). When spatial data were ignored, most Palmyra samples clustered into their own group, but the 2 Hawaiian strata were not well differentiated. When the spatial data were incorporated, the resulting cluster corresponded closely to previously described populations: Group 1 contained all samples from Palmyra and no other samples, Group 2 contained 39 out of 41 (95.1%) Kohala samples, plus 8 of the 69 (11.6%) Hawaiian Islands samples, and all remaining sam-

Table 3. Pairwise estimates of genetic differentiation in melon-headed whales *Peponocephala electra* between strata in both the mtDNA and nDNA data sets. Φ_{ST} values were calculated using the Tamura & Nei (1993) model of nucleotide substitution with gamma = 0.632. Comparisons involving Mayotte and the South Pacific were only conducted for the mtDNA data set due to the small sample size for these strata in the nDNA data set. **Bold**: significant at p < 0.05. HI: Hawaiian Islands, na: not analyzed due to insufficient samples

Comparison	mtDNA			nDNA		
	Φ_{ST}	F_{ST}	χ^2 p-value	F_{ST}	F'_{ST}	χ^2 p-value
Bahamas vs. Kohala	0.193	0.385	**<0.001**	0.035	0.113	**0.001**
Bahamas vs. Johnston	0.493	0.390	**<0.001**	0.026	0.080	**0.019**
Bahamas vs. Mayotte	0.401	0.381	**<0.001**	na	na	na
Bahamas vs. HI	0.363	0.272	**<0.001**	0.033	0.113	**0.001**
Bahamas vs. Palmyra	0.343	0.278	**<0.001**	0.050	0.163	**0.001**
Bahamas vs. South Pacific	0.592	0.335	**0.001**	na	na	na
Kohala vs. Johnston	0.198	0.239	**<0.001**	−0.009	−0.030	0.937
Kohala vs. Mayotte	0.104	0.126	**0.006**	na	na	na
Kohala vs. HI	0.075	0.117	**<0.001**	0.006	0.021	**0.004**
Kohala vs. Palmyra	0.061	0.097	**<0.001**	0.014	0.050	**0.001**
Kohala vs. South Pacific	0.216	0.221	**<0.001**	na	na	na
Johnston vs. Mayotte	0.233	0.197	**0.008**	na	na	na
Johnston vs. HI	0.177	0.099	0.067	−0.006	−0.022	0.884
Johnston vs. Palmyra	0.219	0.140	**<0.001**	0.007	0.026	0.114
Johnston vs. South Pacific	0.249	0.165	**0.011**	na	na	na
Mayotte vs. HI	0.170	0.067	**0.019**	na	na	na
Mayotte vs. Palmyra	0.204	0.057	0.070	na	na	na
Mayotte vs. South Pacific	0.341	0.131	**0.045**	na	na	na
HI vs. Palmyra	0.018	0.015	**<0.001**	0.010	0.035	**0.001**
HI vs. South Pacific	0.030	0.099	**<0.001**	na	na	na
Palmyra vs. South Pacific	0.069	0.101	**<0.001**	na	na	na

ples (4.9% of Kohala samples, 88.4% Hawaiian Islands samples, and 100% of Guam, Johnston, and South Pacific samples) were assigned to Group 3 (Table 5).

DISCUSSION

Patterns of mitochondrial divergence

Based on available knowledge for MHWs prior to our study, we predicted we would find strong differentiation between putative populations and deep divergences within the MHW mitochondrial sequences, similar to what has been reported for other blackfish species (e.g. Oremus et al. 2009, Parsons et al. 2013, Martien et al. 2014, Van Cise et al. 2016). However, our data did not reveal any such pattern. Rather, we detected a highly reticulated haplotype network in which closely related haplotypes are as likely to be from different ocean basins as they are from the same population. Our sample size from the Atlantic Ocean was inadequate to assess genetic diversity there, or to allow robust conclusions regarding genetic differentiation between ocean basins. Nonetheless, the placement of the 4 Atlantic haplo-

types detected in our sample set suggests that increased sampling from the Atlantic is unlikely to reveal a deep divergence between ocean basins. Haplotypic diversity in most strata (Table 2) was also higher than is typical for other blackfish (e.g. Amos et al. 1993, Oremus et al. 2009, Parsons et al. 2013, Martien et al. 2014). Haplotypic diversity was similar to that observed in smaller delphinids such as spinner dolphins (Oremus et al. 2007, Andrews et al. 2010), pantropical spotted dolphins *Stenella attenuata* (Escorza-Trevino et al. 2005), and common bottlenose dolphins (Tezanos-Pinto et al. 2009, Martien et al. 2012), species that also do not exhibit deep mitochondrial divergences.

The deep mitochondrial divergences observed within previously studied blackfish species, including killer whales, long-finned pilot whales, and false killer whales, are believed to result from strong fidelity to natal populations (Amos et al. 1993, Baird 2000, Barrett-Lennard 2000, Ford et al. 2011, Martien et al. 2014). Although interbreeding among populations can result in nuclear gene flow, a lack of dispersal between populations precludes the exchange of mtDNA, allowing mtDNA lineages to diverge via drift. Furthermore, restricted mtDNA gene flow results in lower mitochondrial effective population
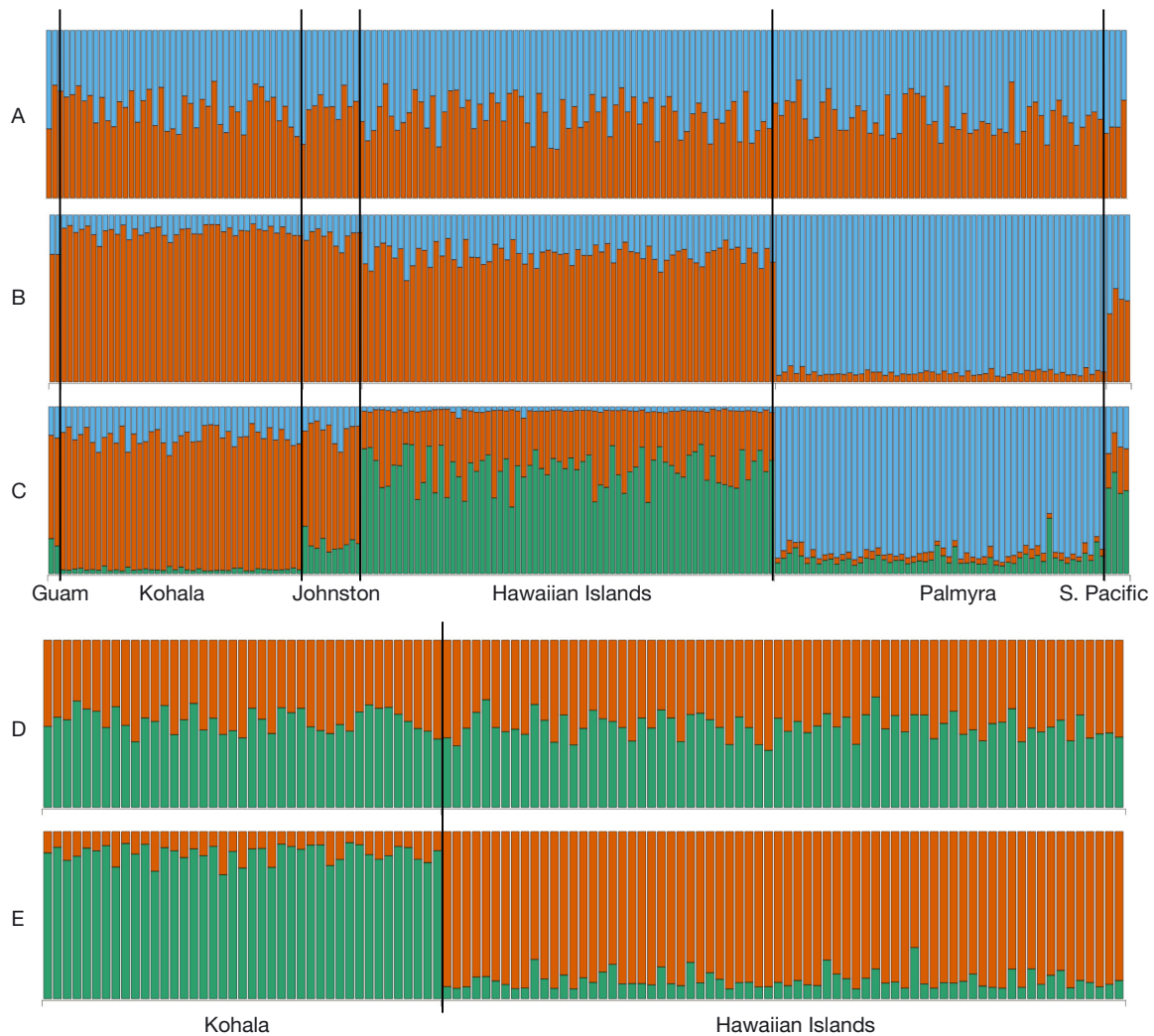
Fig. 3. STRUCTURE results for analyses using melon-headed whale *Peponocephala electra* nDNA from (A–C) all Pacific samples and (D,E) only samples from the Hawaiian Archipelago. For analysis with all Pacific samples, results are shown for models in which (A) LOCPRIOR = F and $K = 2$, (B) LOCPRIOR = T and $K = 2$, and (C) LOCPRIOR = T and $K = 3$. For analyses with only Hawaiian samples, results are shown for models where $K = 2$ with (D) LOCPRIOR = F and (E) LOCPRIOR = T. Each bar represents a single individual, and is shaded to show the proportion of that individual's ancestry that is attributable to each group (A–C: blue = Group 1, orange = Group 2, green = Group 3; D,E: green = Group 1, orange = Group 2)

size, contributing to low haplotypic diversity in these species. Our results suggest that MHWs do not exhibit strong fidelity to natal groups, but rather disperse between populations at a rate high enough to prevent the development of geographically restricted mitochondrial lineages, enabling them to maintain higher haplotypic diversity than other species of blackfish.

Population structure

Our analyses did reveal significant differentiation between most pairs of strata, confirming that these are distinct populations between which gene flow is restricted. However, the magnitude of mtDNA differentiation between populations was significantly lower than what has been reported in other species of blackfish. Estimates of differentiation for both the mtDNA and nDNA data sets were highest between the Bahamas and all Pacific strata, indicating that movement rates between the ocean basins are lower than within the Pacific, as would be expected. However, movement rates appear to be high enough to prevent the development of high levels of differentiation or strong phylogeographic structure.

STRUCTURE was unable to detect any genetic structuring among Pacific samples when we used an

Table 4. Mean ancestry of individual melon-headed whales *Peponocephala electra* from each stratum to the groups identified by STRUCTURE. Results are shown for the model that used stratum membership as a prior

| Stratum | STRUCTURE | | | | |
| | K = 2 | | K = 3 | | |
	Group 1	Group 2	Group 1	Group 2	Group 3
Guam (n = 2)	0.614	0.386	0.633	0.178	0.188
Kohala (n = 43)	0.668	0.332	0.796	0.179	0.025
Johnston (n = 10)	0.689	0.311	0.677	0.143	0.180
HI (n = 72)	0.498	0.502	0.348	0.025	0.627
Palmyra (n = 56)	0.041	0.959	0.041	0.868	0.091
S. Pacific (n = 6)	0.333	0.667	0.243	0.231	0.525

Table 5. Proportion of individual melon-headed whales *Peponocephala electra* from each stratum assigned to groups identified by GENELAND. Results are shown for analyses both with and without geographic coordinates of samples incorporated. wGC and w/oGC: with and without geographic coordinates, respectively

| Stratum | w/oGC | | | wGC | | |
	Group 1	Group 2	Group 3	Group 1	Group 2	Group 3
Guam (n = 2)	0.000	0.000	1.000	0.000	0.000	1.000
Kohala (n = 43)	0.116	0.442	0.395	0.000	0.951	0.049
Johnston (n = 10)	0.000	1.000	0.000	0.000	0.000	1.000
HI (n = 72)	0.194	0.361	0.403	0.000	0.116	0.884
Palmyra (n = 56)	0.929	0.054	0.018	1.000	0.000	0.000
S. Pacific (n = 6)	0.000	0.000	1.000	0.000	0.000	1.000

uninformative prior. This result is consistent with previous performance tests that showed that with uninformative priors, STRUCTURE generally fails to detect genetic structure when genetic differentiation is at the magnitude we detected among Pacific strata (Latch et al. 2006, Waples & Gaggiotti 2006). The strength of assignments increased substantially when we used stratum assignments as a prior in the STRUCTURE analyses, although most samples still exhibited considerable mixed ancestry. The greatest change resulting from the use of a location prior came in the analysis that only included samples from the Hawaiian Archipelago. Mean ancestry estimates increased from ~50% (i.e. no information in the data; Fig. 3D) to >85% when the informative prior was used (Fig. 3E). Although the model that incorporates group membership priors was designed to improve the performance of STRUCTURE in the face of low levels of differentiation, the fact that it uses hypothesized group membership—the very thing it is trying to estimate—as a prior leaves it vulnerable to producing results that are driven more by the prior than by the data. The dramatic change in ancestry esti-

mates when we incorporated an informative prior, combined with the fact that the posterior assignments closely match the prior assignments, suggests that this may be the case in our analysis.

Even in the absence of spatial data, GENELAND was able to detect genetic structure within our data set due to the fact that it uses a no-admixture model, while we selected a model with admixture in our STRUCTURE analyses. The no-admixture model is known to be more powerful for detecting structure, but it has the limitation of being unable to detect admixture (Pritchard et al. 2000). When we incorporated spatial data, GENELAND identified 3 groups, which correspond to our 3 strata with the highest sample sizes: Palmyra, Hawaiian Islands, and Kohala. The 3 remaining Pacific strata (Guam, Johnston, and South Pacific) all assigned strongly to the same group as the Hawaiian Islands samples. However, the assignments of these strata may reflect their small sample sizes rather than a genetic similarity to the Hawaiian Islands.

Unlike STRUCTURE, GENELAND does not incorporate any information regarding stratum assignment for individual samples. The only information available to the analysis was the nDNA genotype and sampling location of each individual. It assumes some degree of spatial coherence of clusters, which is what we would expect if genetic structure were correlated with ecological factors such as habitat or prey distribution, but in no way constrains the geographic size or location of the clusters. Thus, it is striking that the geographic range of the Kohala cluster as identified by GENELAND aligns with the range identified by photo-identification analyses—the small, shallow-water shelf off the northwest coast of Hawai'i Island. The low haplotypic diversity that we detected in the Kohala samples further supports the conclusion that these animals represent a small population that exchanges very few dispersers with the larger Hawaiian Islands population (Aschettino 2010, Aschettino et al. 2012).

The adjacency of the ranges of the Kohala and Hawaiian Islands populations combined with their large differences in both abundance and habitat led

Aschettino et al. (2012) to suggest that the 2 populations are exploiting different foraging niches, as has been suggested for other island-associated cetaceans within the Hawaiian archipelago (Baird et al. 2009, Andrews et al. 2010, Martien et al. 2012, 2014, Courbis et al. 2014). Aschettino et al. (2012) hypothesized that the unique oceanographic characteristics of the 'Alenuihāhā Channel, which separates the islands of Hawai'i and Maui, may result in a sufficient density of prey to sustain the Kohala population in a very small range.

The presence of a behaviorally unique small population of MHWs immediately adjacent to the 'Alenuihāhā Channel is cause for concern, due to frequent naval operations utilizing mid-frequency sonar within the channel. MHWs appear to be particularly sensitive to anthropogenic sound (Southall et al. 2006, Brownell et al. 2009b), a fact that has been exploited in many parts of the world in drive hunts where sound is used to drive MHWs toward the beach where they can be more easily killed or captured (Brownell et al. 2009a). The limited range of the Kohala coast population limits their ability to move away from harmful sound sources, and increases the risk that they will be displaced into unfavorable or unfamiliar habitat if they do (Forney et al. 2017). Thus, the unique characteristics of the Kohala resident population combined with their proximity to a known threat warrant additional conservation and management attention for this small, demographically independent population.

Possible role of behavior

The differences in genetic structure between MHWs and other blackfish may be linked to their foraging strategies and social organization. At some islands, the behavior of MHWs resembles a fission–fusion type structure, where small groups coalesce into larger aggregations during the day and then disperse at night (Brownell et al. 2009b). A similar organization has been described for spinner dolphin populations in the Society Archipelago in French Polynesia (Oremus et al. 2007) and the main Hawaiian Islands (Norris et al. 1994), for example. Spinner dolphins also exhibit patterns of genetic differentiation similar to what we found for MHWs, with moderately differentiated small, resident populations occurring in the nearshore waters around islands (Galver 2002, Andrews et al. 2010). These similarities in the patterns of genetic structure support the hypothesis that the behavior of MHWs is more simi-

lar to that of spinner dolphins than it is to that of other blackfish.

Like spinner dolphins, MHWs are nocturnal mesopredators that forage primarily on predictable and relatively abundant mesopelagic squids and fishes that undertake daily vertical migrations (e.g. Brownell et al. 2009b, Kiszka et al. 2011). In contrast, at least some species of blackfish that exhibit strong genetic structuring feed on prey that are less predictable and more difficult to catch. False killer whales, for example, feed on large pelagic fish (Baird et al. 2008) and Bigg's killer whales feed on marine mammals (Bigg et al. 1987, Baird & Dill 1996). Short-finned pilot whales, which feed in the deep scattering layer, also appear to target large, fast-moving, evasive prey that can be difficult to locate (Aguilar Soto et al. 2008). Accessing these fast-swimming, patchily distributed, and hard-to-catch prey is likely more challenging for large homeothermic predators, and these cetaceans may rely upon strong, stable social bonds within foraging groups (Bigg et al. 1990, Baird & Dill 1996, Baird et al. 2008). Indeed, false killer whales resident to the main Hawaiian Islands have been observed engaging in cooperative hunting and prey sharing (Baird et al. 2008). Differences in foraging strategies have also been evoked to explain differences in group size and social organization between mammal-eating and fish-eating killer whales (Baird & Whitehead 2000).

Overall, MHWs have relatively large group sizes compared to other species of blackfish (e.g. Brownell et al. 2009b, Baird et al. 2013), which might be a strategy to reduce predation risk from oceanic predators such as large pelagic sharks (Gygax 2002; for a review, see Kiszka et al. 2015). Large groups imply some higher levels of intra-species competition. Therefore, MHWs might need to undertake extensive movements and/or rely on relatively more productive areas in tropical and typically oligotrophic ecosystems, which could increase the size of their home range and the extent of their movements. However, the ecological and social drivers of the magnitude of the movements of MHWs need to be further investigated.

The patterns of genetic differentiation in spinner dolphins and MHWs could result from long-distance dispersal of individuals among populations, or via gene flow with widespread oceanic populations (Oremus et al. 2007, Tezanos-Pinto et al. 2009). Large oceanic populations of MHWs are known to exist, at least within the Pacific Ocean (Brownell et al. 2009b, Hamilton et al. 2009), but also most likely in the Indian Ocean (Mannocci et al. 2014). Although most

of our samples came from animals known to show some long-term fidelity to islands, satellite tag data show that animals from the Hawaiian Islands population actually spend most of their time offshore (Baird 2016). This stratum also exhibited the lowest levels of nDNA differentiation and the weakest clustering in both the STRUCTURE and GENELAND analyses. Thus, it is possible that these individuals represent an oceanic population that shows occasional fidelity to the Hawaiian Islands, and serves as a conduit for gene flow throughout the central Pacific. Collecting samples from the open ocean would be valuable for further evaluating this possibility, and elucidating the patterns of connectivity among MHW populations, particularly between island-associated and oceanic populations.

Conclusions

Though our results confirm the existence of demographically independent populations in Hawai'i identified in observational and photo-identification studies, they disprove our original hypothesis that MHW would exhibit the same global patterns of deep mitochondrial divergences and strong genetic structuring revealed in studies of other blackfish species. Comparative studies of groups of closely related species, such as the blackfish, can provide insight into the biological and ecological drivers of different patterns of genetic structure. Future studies linking foraging tactics and social structure of MHWs would provide valuable information for evaluating the importance of these aspects of life history in influencing the patterns of genetic differentiation in blackfish and other delphinids.

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Unexpected patterns of global population structure in melon-headed whales *Peponocephala electra*

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Table S1. Locus-specific estimates of genetic diversity for microsatellite loci. Columns show fluorescent tag (*Tag*) and annealing temperature (T_A) used for the locus, number of alleles detected (# alleles), number of samples with missing genotypes (# missing), allelic richness (A_R), expected heterozygosity (H_e), and observed heterozygosity (H_o). Allelic richness was based on a minimum sample size of 185.

Locus	Tag	T_A (°C)	# alleles	# missing	A_R	H_e	H_o
D17t	FAM	55	12	0	0.064	0.840	0.802
EV14t	HEX	56	12	6	0.067	0.657	0.656
EV1t	HEX	55	7	3	0.038	0.556	0.554
EV94t	FAM	55	10	1	0.054	0.815	0.790
GATA53	HEX	58.4	7	4	0.039	0.655	0.613
KWM12at	FAM	52	13	7	0.072	0.813	0.833
KWM2at	HEX	45	13	2	0.070	0.878	0.870
SAM25t	FAM	55	9	7	0.050	0.815	0.806
SL125t	FAM	55	9	7	0.050	0.688	0.672
SL849t	FAM	55	8	0	0.043	0.576	0.610
SW19t	FAM	55	7	8	0.039	0.683	0.659
Ttr11	FAM	55	12	1	0.064	0.850	0.850
Ttr58	FAM	60	6	2	0.032	0.600	0.600
TtrRC11	FAM	55	10	1	0.054	0.801	0.801

Table S2. Published values of Φ_{ST} for fish-eating killer whales (KW), false killer whales (FKW), short-finned pilot whales (SFPW), and long-finned pilot whales (LFPW) used to generate Figure 3. Strata are defined in source material. Abbreviations: ETP = Eastern Tropical Pacific; HI = Hawaiian Islands; MHI = main Hawaiian Islands; NWHI = Northwest Hawaiian Islands.

Taxon	Strata	Comparison type	Φ_{st}	Source
SFPW	South Pacific vs Atlantic	between	0.589	Oremus et al. 2009
SFPW	N. Japan vs Atlantic	between	0.741	Oremus et al. 2009
SFPW	S. Japan vs Atlantic	between	0.619	Oremus et al. 2009
LFPW	N. Atlantic vs New Zealand	between	0.648	Oremus et al. 2009
LFPW	N. Atlantic vs Tasmania	between	0.265	Oremus et al. 2009
SFPW	Indian Ocean vs California Current	between	0.549	Van Cise et al. in press
SFPW	Indian Ocean vs ETP	between	0.47	Van Cise et al. in press
SFPW	Indian Ocean vs S. Japan	between	0.534	Van Cise et al. in press
SFPW	Indian Ocean vs Hawai'i	between	0.738	Van Cise et al. in press
SFPW	Indian Ocean vs South Pacific	between	0.202	Van Cise et al. in press
SFPW	Indian Ocean vs SE Asia	between	0.157	Van Cise et al. in press

Taxon	Strata	Comparison type	Φ_{st}	Source
SFPW	N. Atlantic vs California Current	between	0.78	Van Cise et al. in press
SFPW	N. Atlantic vs ETP	between	0.712	Van Cise et al. in press
SFPW	N. Atlantic vs S. Japan	between	0.851	Van Cise et al. in press
SFPW	N. Atlantic vs HI	between	0.882	Van Cise et al. in press
SFPW	N. Atlantic vs South Pacific	between	0.58	Van Cise et al. in press
SFPW	N. Atlantic vs SE Asia	between	0.816	Van Cise et al. in press
SFPW	N. Atlantic vs Indian Ocean	between	0.674	Van Cise et al. in press
FKW	MHI vs NWHI	within	0.131	Martien et al. 2014
FKW	MHI vs Central N. Pacific	within	0.658	Martien et al. 2014
FKW	MHI vs ETP	within	0.709	Martien et al. 2014
FKW	NWHI vs Central N. Pacific	within	0.618	Martien et al. 2014
FKW	NWHI vs ETP	within	0.695	Martien et al. 2014
FKW	ETP vs Central N. Pacific	within	0.091	Martien et al. 2014
SFPW	N. Japan vs South Pacific	within	0.545	Oremus et al. 2009
SFPW	S. Japan vs South Pacific	within	0.58	Oremus et al. 2009
SFPW	S. Japan vs N. Japan	within	0.566	Oremus et al. 2009
LFPW	Tasmania vs New Zealand	within	0.423	Oremus et al. 2009
KW	Gulf of Alaska vs E. Aleutians-Trinity	within	0.18	Parsons et al. 2013
KW	Gulf of Alaska vs C. Aleutians	within	0.783	Parsons et al. 2013
KW	E. Aleutians-Trinity Islands vs C. Aleutians	within	0.915	Parsons et al. 2013
KW	Gulf of Alaska vs W. Aleutians-Russia	within	0.962	Parsons et al. 2013
KW	E. Aleutians-Trinity Islands vs WAL-RUS	within	1	Parsons et al. 2013
KW	C. Aleutians vs W. Aleutians-Russia	within	0.131	Parsons et al. 2013
SFPW	S. Japan vs California Current	within	0.809	Van Cise et al. in press
SFPW	Hawai'i vs California Current	within	0.852	Van Cise et al. in press
SFPW	South Pacific vs California Current	within	0.521	Van Cise et al. in press
SFPW	SE Asia vs California Current	within	0.736	Van Cise et al. in press
SFPW	S. Japan vs ETP	within	0.777	Van Cise et al. in press
SFPW	Hawai'i vs ETP	within	0.823	Van Cise et al. in press
SFPW	South Pacific vs ETP	within	0.479	Van Cise et al. in press
SFPW	SE Asia vs ETP	within	0.685	Van Cise et al. in press
SFPW	HI vs S. Japan	within	0.574	Van Cise et al. in press
SFPW	Spac vs S. Japan	within	0.655	Van Cise et al. in press
SFPW	SE Asia vs S. Japan	within	0.105	Van Cise et al. in press
SFPW	HI vs Spac	within	0.744	Van Cise et al. in press
SFPW	HI vs SE Asia	within	0.639	Van Cise et al. in press
SFPW	SE Asia vs Spac	within	0.48	Van Cise et al. in press

Table S3. Mean log-likelihood and ΔK for STRUCTURE models.

Sample Set	Model	Likelihood	
		Mean ln(K)	ΔK
	$K = 1$	**-5226.30**	NA
Hawaiian samples only	$K = 2$	-5406.54	NA
LOCPRIOR = F	$K = 3$	-6289.48	NA
	$K = 1$	**-5226.00**	NA
Hawaiian samples only	$K = 2$	-5249.20	NA
LOCPRIOR = T	$K = 3$	-5516.94	NA
	$K = 1$	**-8716.44**	NA
	$K = 2$	-9138.90	-422.46
All Samples	$K = 3$	-8923.58	215.32
LOCPRIOR = F	$K = 4$	-9017.96	-94.38
	$K = 5$	-8899.84	118.12
	$K = 6$	-9209.82	-309.98
	$K = 1$	-8716.70	NA
	$K = 2$	**-8669.76**	9.014
All Samples	$K = 3$	-8701.46	0.815
LOCPRIOR = T	$K = 4$	-8725.28	12.45
	$K = 5$	-9034.68	0.939
	$K = 6$	-9240.90	NA