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Initial genotype matching of humpback whales from the 2010 Australia/New Zealand Antarctic Whale Expedition (Area V) to Australia and the South Pacific.

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ABSTRACT

Here we present new records of humpback whale migratory connections between Antarctic Area V and migratory corridors of Australia and the South Pacific based on genotype matching (up to 10 microsatellite loci, with sex and mitochondrial DNA). A total of $n = 64$ skin biopsy samples were collected by the Australia/New Zealand Antarctic Whale Expedition during the 2010 austral summer within Antarctic Area V, concentrated between 162°E and 179°W. Comparison of microsatellite genotypes resolved 57 individuals representing 28 females and 29 males. These individuals were then compared to databases of genotypes generated in three independent laboratories, using samples collected in 3 regions: the west coast of Australia (2007, $n = 204$), the east coast of Australia (1996-2010, 5 locations, $n = 865$), and New Zealand/Oceania (1991-2009, 7 locations, $n = 1,203$). Following standardisation of allele bin sizes this comparison revealed 7 likely matches to known migratory corridors; 3 to Hervey Bay, Queensland Australia (south bound migration), 3 to Byron Bay, NSW Australia (north bound migration) and one to Cook Strait, New Zealand (north bound migration). These results are concordant with previously described connections from *Discovery* marking and with results of photo-identification matching from the same expedition, showing a strong connection of Area V to the presumed eastern Australia breeding stock with little or no detectable connection to Oceania.

INTRODUCTION

Prior to protection from commercial whaling, knowledge of movements of individual humpback whales between breeding grounds and migratory corridors to Antarctic feeding Areas IV, V and VI relied on the *Discovery* marking and recovery program (Dawbin 1959, Dawbin 1964, Chittleborough 1965, Dawbin 1966). More recently photo-identification comparisons (e.g. Robbins, et al. 2011, Franklin, et al. In Press), genotype matching (e.g. Steel, et al. 2008, Anderson, et al. 2010) and satellite tagging (e.g. Gales, et al. 2009, Garrigue, et al. 2010, Hauser, et al. 2010) have been used to detect migratory movements of individual whales.

During the 2010 austral summer the Australia/New Zealand Antarctic Whale Expedition (AWE) spent 30 days surveying approximately 5800nm of IWC Antarctic Area V south of 60°S between 150°E and 150°W (Gales 2010). Here we present initial results of a collaborative effort to match genotypes from samples collected during AWE to previously typed individuals from migratory corridors and breeding grounds of Australia and the South Pacific. This study is notable, not only for its large-scale collaborative framework, but also for the success in the matching of microsatellite genotypes through standardisation of allele size binning among three independent laboratories. A parallel study, SC63/SH/16 (Constantine, et al. 2011), which reports on connections made from matching fluke photographs collected during the AWE survey will also be presented. Combined, these reports improve our understanding of humpback whale connections.

METHODS

Collection and genotyping of AWE samples

A total of $n = 64$ humpback whale skin biopsy samples were collected during the Antarctic Whale Expedition (AWE) primarily around the Balleny Islands between 162°E and 179°W. Total cellular DNA was isolated from skin tissue using an automated Promega Maxwell® 16 System. Ten microsatellite loci were amplified for each sample using previously published primers (GT211, GT575 (Bérubé, et al. 2000), GATA417 (Palsbøll, et al. 1997), EV1, EV14, EV37, EV94, EV96 (Valsecchi and Amos 1996) and rw4-10 (Waldick, et al. 1999)) To allow simultaneous amplification of several loci in one PCR, we used a Qiagen Multiplex Kit to multiplex three sets of loci : set 1 (EV37 and GT23); set 2 (EV14, EV96 and GATA417; and set 3 (EV1, EV94 and GT575). GT211 and rw4-10 were amplified individually (Table 1). For each locus, one of the primers within each pair was labelled fluorescently at the 5' end to allow for visualisation of alleles on an automated ABI 3130 sequencer (Applied Biosystems). Each PCR had a final volume of 12.5 µl and included: 1x Qiagen Multiplex PCR Master Mix (containing HotStarTaq®DNA Polymerase, Multiplex PCR Buffer and dNTP mix), 6 mM Mg²⁺, 2 µM of each primer (labelled and non-labelled) and 1 to 8 ng of template DNA. The thermocycling profile consisted of an initial denaturing step of 95°C for 15 minutes, 30 cycles (30 s at 94°C, 90 s at 53°C annealing for rw4-10 and GT211, 58°C annealing for all other loci), 60 s at 72°C) followed by a final extension step of 30 minutes at 60°C. Sex was determined using a fluorescent 5' exonuclease assay producing PCR product from the ZFX and ZFY orthologous gene sequences (Morin, et al. 2005). Sequencing of the mitochondrial (mt) DNA control region (700bp) followed methods described in detail by Olavarría, et al. (2007) with some modifications (contact N.S. for details).

Breeding ground and migratory corridor datasets

Replicates were identified and removed from within the AWE dataset using GenAlex (Peakall and Smouse 2005). Genotypes of the remaining individuals were then compared to a total of $n = 2,068$ previously typed individuals from datasets generated by 3 separate research groups (Table 2 and Figure 1). Dataset 1 (AMMC), comprised $n = 335$ individuals from the west coast of Australia (Exmouth) and two locations on the east coast of Australia (Eden, NSW and Tasmania) typed at a total of 10 loci and generated with the AWE dataset at the Australian Marine Mammal Centre. Dataset 2 (SCU), comprised $n = 734$ individuals from 3 locations on the east coast of Australia (Byron Bay and Ballina, NSW and Hervey Bay, Queensland) typed at up to a total of 13 loci and generated at Southern Cross University (Anderson, et al. 2010). Dataset 3 (SPWRC), comprised $n = 1,203$ individuals from New Zealand and 6 locations throughout Oceania (New Caledonia, Tonga, Niue, Samoa/American Samoa, Cook Islands

and French Polynesia) typed at up to a total of 17 loci and generated by the South Pacific Whale Research Consortium (updated from Steel, et al. 2008) . All 3 datasets also included mtDNA control region sequence and sex information for the majority of samples. The 10 loci typed for AMMC were the same as the 10 loci typed for AWE, 10 of the 17 loci typed for SPWRC were the same as the AWE loci and 8 of the 13 loci typed for SCU overlapped with the AWE loci.

Standardisation of datasets

Due to differences in microsatellite run conditions between the 3 locations, standardisation of allele size was necessary before direct comparisons could be made. The SCU and SPWRC datasets had previously been standardised to each other through an exchange of reference samples as described in Anderson, et al. (2003) with results reported in Anderson, et al. (2010). The AWE/AMMC dataset was then standardised to the SCU and SPWRC dataset using allele frequency calibration (i.e. aligning allele frequency histograms). Reference samples have recently been exchanged between SPWRC and AMMC to check this standardisation.

Initial genotype matching between all 4 datasets was conducted using Cervus (Kalinowski, et al. 2007) using 'relaxed' conditions to avoid false exclusion of true matches due to genotyping or standardisation errors. The relaxed conditions required a minimum of 6 matching loci between any pair of samples. These 'likely matches' were then reviewed for errors at any mismatching loci to confirm identity (Waits and Leberg 2000, Waits, et al. 2001, Hoffman and Amos 2005, Morin, et al. 2010). Where available, variation in mtDNA control region sequences (i.e. haplotypes) and sex were used to confirm matches.

RESULTS

Matching within AWE

Comparison of genotypes within the AWE dataset resolved $n = 57$ unique individuals with a 1:1 sex ratio ($n = 28$ females: $n = 29$ males). Analysis of mtDNA control region sequence resolved 26 haplotypes previously described by Olavarria, et al. (2007) and two new haplotypes.

Matching to breeding grounds and migratory corridors

Comparison of the $n = 57$ AWE individuals to the $n = 2,272$ individuals from West Australia, East Australia, New Zealand and Oceania revealed 7 likely matches representing connections between an Antarctic feeding area and migratory corridors; 3 to Hervey Bay (southbound migration, SCU), 3 to Byron Bay (northbound migration, SCU) and 1 to New Zealand (northbound migration, SPWRC) (Figure 1 and Table 3). Given variation in the number of loci used in each laboratory, the number of overlapping loci within a match ranged from 6 to 10. However, even with only 6 matching microsatellite loci the probability of identity was sufficiently low enough to establish the match with reasonable confidence ($pID, 2.3 \times 10^{-7}$, Table 2) given the relatively small number of pair wise matches ($57 \times 2,272 = 1.3 \times 10^5$). One of the 7 pairs of likely matches (2010AWE215 and Mno04NZ014) showed disagreements at 2 loci (Table 3): a 'soft match' at EV14 (i.e. one sample was homozygous for one allele of the other sample) and 'hard' mismatch at rw4-10 (i.e. no shared alleles between samples). The New Zealand sample (Mno04NZ014) was subsequently re-genotyped for both loci and the soft match at EV14 was found to be the result of allelic dropout. We have not yet had time to resolve the disagreement at rw4-10 but consider that it is likely due to a genotype error.

DISCUSSION

The genotype matches reported here are concordant with results of photo-id matching from this expedition (Constantine, et al. 2011) and with migratory connections between East Australia and Antarctic Area V previously described through *Discovery* marking and recovery, photo-id and genotype matching (e.g. Chittleborough 1965, Dawbin 1966, Anderson, et al. 2010, Franklin, et al. In Press). The six matches to locations along the east coast of Australia add further evidence to the idea that the Balleny Islands are an important feeding area for humpback whales that migrate along this corridor (Rock, et al. 2006, Franklin, et al. In Press)

It is interesting to note that despite the large number of individuals within the Oceania dataset no direct connections were made to any of the Oceania breeding grounds. However, as New Zealand is known to be a migratory corridor for animals that breed in New Caledonia (Garrigue, et al. 2002, Olavarria, et al. 2006) the one match to New Zealand is consistent with the photo-id match to New Caledonia from the AWE (Constantine, et al. 2011) and an earlier genetic match (Steel, et al. 2008) between New Caledonia and Antarctic Area V. The lack of matches to other regions of Oceania suggests that the Balleny Islands area of Antarctic Area V is not a primary feeding ground for these animals. This, along with previously described connections from Tonga and Samoa (Steel, et al. 2008, Robbins, et al. 2011) suggest that animals that breed in island groups east of New Caledonia primarily feed in Antarctic Areas VI and I.

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REFERENCES

- Anderson, M., P. Baverstock, D. Steel, C. S. Baker and C. Olavarria (2003). Standardisation of genetic analysis protocols used in humpback whale research. Final Report to the Department of the Environment and Heritage and the Natural Heritage Trust, Canberra, ACT, pp.
- Anderson, M., D. Steel, W. Franklin, T. Franklin, D. Paton, D. Burns, P. Harrison, P. R. Baverstock, C. Garrigue, C. Olavarria, M. Poole, N. Hauser, R. Constantine, D. Thiele, P. Clapham, M. Donoghue and C. S. Baker (2010). Microsatellite genotype matches of eastern Australian humpback whales to Area V feeding and breeding grounds. Report (SC/62/SH7) to the Scientific Committee of the International Whaling Commission, Agadir, Morocco.
- Bérubé, M., H. Jørgensen, R. McEwing and P. J. Palsbøll (2000). Polymorphic di-nucleotide microsatellite loci isolated from the humpback whale, *Megaptera novaeangliae*. *Molecular Ecology*, 9 (12); 2181-2183.
- Chittleborough, R. G. (1965). Dynamics of two populations of humpback whales, *Megaptera novaeangliae* (Borowski). *Australian Journal of Marine and Freshwater Research*, 16; 33-128.
- Constantine, R., J. Allen, P. Beeman, D. Burns, S. Childerhouse, M. C. Double, P. Ensor, T. Franklin, W. Franklin, N. Gales, C. Garrigue, E. Gates, N. Gibbs, A. Hutsel, C. Jenner, M. Jenner, G. Kaufman, A. Macie, D. K. Mattila, A. Oosterman, D. Paton, J. Robbins, P. Stevick, N. Schmitt, A. Tagarino and K. Thompson (2011). Comprehensive photo-identification matching of Antarctic Area V humpback whales. Report (SC/63/SH16) to the Scientific Committee of the International Whaling Commission, Tromso, Norway.
- Dawbin, W. H. (1959). New Zealand and South Pacific whale marking and recoveries to the end of 1958. *Norsk Hvalfangsttiid*, 48; 213-238.
- Dawbin, W. H. (1964). Movements of humpback whales marked in the southwest Pacific Ocean 1952-1962. *Norsk Hvalfangsttiid*, 53; 68-78.
- Dawbin, W. H. (1966). The seasonal migratory cycle of humpback whales. In *Whales, Dolphins and Porpoises*. K. S. Norris, ed. pp. 145-171. Berkeley: University of California Press.
- Franklin, T., F. Smith, N. Gibbs, S. Childerhouse, D. Burns, D. Paton, W. Franklin, C. S. Baker and P. Clapham (In Press). Migratory movements of humpback whales (*Megaptera novaeangliae*) between eastern Australia and the Balleny Islands, Antarctica, confirmed by photo-identification. *Journal of Cetacean Research Management*.
- Gales, N., M. Double, S. Robinson, M. Jenner, C. Jenner, E. King, J. Gedamke, D. Paton and B. Raymond (2009). Satellite tracking of southbound East Australian humpback whales (*Megaptera novaeangliae*): challenging the feast or famine model of migrating whales. Report (SC61/SH/17) to the Scientific Committee of the International Whaling Commission, Madeira, Portugal.
- Gales, N. (2010). Antarctic Whale Expedition Preliminary science field report and summary. Unpublished report, Hobart, Australia, pp. 21.

- Garrigue, C., A. Aguayo, V. L. U. Amante-Helweg, C. S. Baker, S. Caballero, P. Clapham, R. Constantine, J. Denking, M. Donoghue, L. Flórez-González, J. Greaves, N. Hauser, C. Olavarría, C. Pairoa, H. Peckham and M. Poole (2002). Movements of humpback whales in Oceania, South Pacific. *Journal of Cetacean Research Management*, 4 (3); 255-260.
- Garrigue, C., A. N. Zerbini, Y. Geyer, M.-P. Heide-Jørgensen, W. Hanaoka and P. Clapham (2010). Movements of satellite-monitored humpback whales from New Caledonia. *Journal of Mammalogy*, 91 (1); 109-115.
- Hauser, N., A. N. Zerbini, Y. Geyer, M.-P. Heide-Jørgensen and P. Clapham (2010). Movements of satellite-monitored humpback whales, *Megaptera novaeangliae*, from the Cook Islands. *Marine Mammal Science*, 26 (3); 679-685.
- Hoffman, J. I. and W. Amos (2005). Microsatellite genotyping errors: detection approaches, common sources and consequences for paternal exclusion. *Molecular Ecology*, 14; 599-612.
- Kalinowski, S. T., M. L. Taper and T. C. Marshall (2007). Revising how the computer program cervus accommodates genotyping error increases success in paternity assignment. *Molecular Ecology*, 16 (5); 1099-1106.
- Morin, P. A., A. Nestler, N. T. Rubio-Cisneros, K. M. Robertson and S. L. Mesnick (2005). Interfamilial characterization of a region of the ZFX and ZFY genes facilitates sex determination in cetaceans and other mammals. *Molecular Ecology*, 14 (10); 3275-3286.
- Morin, P. A., K. K. Martien, F. I. Archer, F. Cipriano, D. Steel, J. Jackson and B. L. Taylor (2010). Applied conservation genetics and the need for quality control and reproting of genetic data used in fisheries and wildlife management. *Journal of Heredity*, 101 (1); 1-10.
- Olavarría, C., S. Childerhouse, N. Gibbs and C. S. Baker (2006). Contemporary genetic diversity of New Zealand humpback whales and their genetic relationship with Breeding Stocks D, E, F and G. Report (SC/A06/HW31) to the Intersessional Workshop for the Comprehensive Assessment of Southern Hemisphere Humpback Whales, Scientific Committee of the International Whaling Commission, Hobart, Australia.
- Olavarría, C., C. S. Baker, C. Garrigue, M. Poole, N. Hauser, S. Caballero, L. Flórez-González, M. Bresseur, J. L. Bannister, J. Capella, P. J. Clapham, R. Dodemont, M. Donoghue, C. Jenner, M.-N. Jenner, D. Moro, M. Oremus, D. Paton, H. Rosenbaum and K. Russell (2007). Population structure of South Pacific humpback whales and the origin of the Eastern Polynesian breeding grounds. *Marine Ecology Progress Series*, 330; 257-268.
- Palsbøll, P. J., M. Bérubé, A. H. Larsen and H. Jørgensen (1997). Primers for the amplification of tri- and tetramer microsatellite loci in baleen whales. *Molecular Ecology*, 6 (9); 893-895.
- Peakall, R. and P. E. Smouse (2005). GENALEX 6: genetic analysis in Excel. Population genetic software for teaching and research. *Molecular Ecology Notes*; 1-8.
- Robbins, J., L. Dalla Rosa, J. Allen, D. Mattila, E. Secchi, A. Friedlaender, P. Stevick, D. Nowacek and D. Steel (2011). Return movement of a humpback whale between the Antarctic Peninsula and American Samoa: a seasonal migration record. *Endangered Species Research*, 13 (2); 117-121.
- Rock, J., L. A. Pastene, G. Kaufman, P. Forestell, K. Matsuoka and J. Allen (2006). A note on East Australia Group V Stock humpback whale movement between feeding and breeding areas based on photo-identification. *Journal of Cetacean Research Management*, 8 (3); 301-305.
- Steel, D., C. Garrigue, M. Poole, N. Hauser, C. Olavarría, L. Flórez-González, R. Constantine, S. Caballero, D. Thiele, D. Paton, P. Clapham, M. Donoghue and C. S. Baker (2008). Migratory connections between humpback whales from South Pacific breeding grounds and Antarctic feeding areas based on genotype matching. Report (SC/60/SH13) to the Scientific Committee of the International Whaling Commission, Santiago, Chile.
- Valsecchi, E. and W. Amos (1996). Microsatellite markers for the study of cetacean populations. *Molecular Ecology*, 5; 151-156.
- Waits, J. L. and P. L. Leberg (2000). Biases associated with population estimation using molecular tagging. *Animal Conservation*, 3; 191-199.
- Waits, L. P., G. Luikart and P. Taberlet (2001). Estimating the probability of identity among genotypes in natural populations: cautions and guidelines. *Molecular Ecology*, 10 (1); 249-256.
- Waldick, R. C., M. W. Brown and B. N. White (1999). Characterization and isolation of microsatellite loci from the endangered North Atlantic right whale. *Molecular Ecology*, 8 (10); 1763-1765.

Table 1: The 10 microsatellite loci used to individually identify humpback whales from the AWE.

Loci	Label	Multiplex group	Size range	N alleles
Ev1	VIC	Set 3	123 – 129	4
Ev14	VIC	Set 2	125 – 145	9
Ev37	NED	Set 1	192 – 230	21
Ev94	FAM	Set 3	202 – 220	9
Ev96	FAM	Set 2	147 – 173	13
GATA 417	FAM	Set 2	183 – 282	17
GT 211	FAM	Individually	98 – 120	10
GT 23	VIC	Set 1	101 – 123	9
GT 575	VIC	Set 3	137 – 177	15
rw4-10	VIC	Individually	192 – 216	13

Table 2: The total number of unique genotypes (assumed to represent individual whales) sampled in each region of Oceania and the north and southbound migrations. The number in parenthesis in the Total row is the number of unique genotypes across the entire region (i.e. with between region matches removed)

Region	# loci	# loci shared with AWE	Dataset	Years	# unique genotypes
Oceania breeding grounds					
New Caledonia	17	10	SPWRC	1995-2005	388
Tonga	17	10	SPWRC	1991-2009	371
Samoa/ American Samoa	17	10	SPWRC	2001-2009	88
Cook Islands	17	10	SPWRC	1996-2005	108
French Polynesia	17	10	SPWRC	1997-2007	230
Niue	17	10	SPWRC	2008	3
Total Oceania					1,188 (1,140)
Northbound migration					
Byron Bay, EA	13	8	SCU	1996-2004	337
Eden, EA	10	10	AMMC	2008	43
Tasmania, EA	10	10	AMMC	2006-2008	1
Cook Strait, NZ	17	10	SPWRC	2003-2009	65
Total northbound					446
Southbound migration					
Hervey Bay, EA	13	8	SCU	1997-2003	365
Ballina, EA	13	8	SCU	2003-2004	63
Eden, EA	10	10	AMMC	2008	18
Tasmania, EA	10	10	AMMC	2006-2008	69
Exmouth, WA	10	10	AMMC	2007	204
Total southbound					719
Total breeding and migration					2,353 (2,272)

Table 3: Summary of matching microsatellite genotypes for humpback whales sampled in Area V (n = 57) and the migratory corridors of Oceania and Australia (n = 2,068). -/- indicate an amplification failure at that locus for that sample. Blank cells indicate that a sample was not run for that locus for one of the 3 datasets. Mismatches are shown in bold.

Sample Name	Location	date	dlp	sex	Ev1	Ev14	Ev37	Ev94	Ev96	GATA 417	GT 211	GT 23	GT 575	rw 4-10	# loci	PI
2010AWE014	Area V	1 Mar 2010	SP11	F	123/125	131/137	198/214	212/212	161/163	199/222	106/118	109/115	151/151	204.206	8	4.63 X10 ⁻¹¹
Mn463	Hervey Bay	11 Oct 2000	-	F	123/125	131/137	198/214	212/212	161/163	199/222	106/118		151/151			
2010AWE204	Area V	21 Feb 2010	SP3	M	123/123	131/139	212/214	212/214	171/173	214/222	106/106	109/115	149/151	196/200	7	3.25 x10 ⁻⁹
Mn658	Byron Bay	2004	-	M	123/123	131/139	212/214	212/214	-/-	214/222	106/106		149/151			
2010AWE206	Area V	21 Feb 2010	SP15	M	123/123	131/133	196/214	208/214	157/159	218/226	108/116	113/115	151/151	198/206	6	2.3x10 ⁻⁷
Mn211	Byron Bay	30 June 2004	-	M	123/123	131/133	196/214	208/214	-/-	-/-	108/116		151/151			
2010AWE207	Area V	21 Feb 2010	SP94	M	123/127	131/133	192/198	214/214	153/163	203/210	106/108	109/115	153/157	198/206	7	1.34x10 ⁻⁹
Mn319	Hervey Bay	17 Sept 2003	SP94	M	123/127	131/133	192/198	214/214	-/-	203/210	106/108		153/157			
2010AWE210	Area V	22 Feb 2010	SP94	F	123/123	133/139	198/208	208/220	165/165	199/214	106/112	115/115	143/153	200/204	7	7.7x10 ⁻¹¹
Mn650	Hervey Bay	9 Sept 2002	SP94	F	123/123	133/139	198/208	208/220	-/-	199/214	106/112		143/153			
2010AWE213	Area V	27 Feb 2010	SP102	F	123/127	131/131	194/196	206/212	155/163	199/230	104/108	111/115	137/151	200/204	8	7.4x10 ⁻¹³
Mn519	Byron Bay	2003	SP102	F	123/127	131/131	194/196	206/212	155/163	199/230	104/108		137/151			
2010AWE215	Area V	28 Feb 2010	SP1	M	123/123	133/143	196/216	212/214	161/163	218/238	106/106	115/115	151/153	200/202	9(1)	*7.72x10 ⁻¹³
Mno04NZ014	New Zealand	24 June 2004	SP1	M	123/123	133/143	196/216	212/214	161/163	218/238	106/106	115/115	151/153	196/198		

*PI excluding rw4-10

Figure 1: Map showing the location of microsatellite genotype matches between humpback whales from Antarctic feeding Area V and the migratory corridors of Oceania and Australia. Total number of unique genotypes for each location is shown in parentheses.

