

2006 SYMPOSIUM ON SOUTHERN RESIDENT KILLER WHALES



APRIL 3-5, 2006

NOAA WESTERN REGIONAL CENTER AUDITORIUM
7600 SAND POINT WAY NE, BUILDING 9
SEATTLE, WA 98115



Fisheries and Oceans
Canada



SOUTHERN RESIDENT KILLER WHALE SYMPOSIUM
APRIL 3-5, 2006
NOAA WESTERN REGIONAL CENTER AUDITORIUM
7600 SANDPOINT WAY NE
SEATTLE, WASHINGTON

PROPOSED AGENDA

SYMPOSIUM OBJECTIVES

1. Present new research results;
2. Examine how this information moves us forward in informing conservation actions; and
3. Discuss what additional research, if any, is needed.

MONDAY, APRIL 3

- 1:00 – 1:20 Welcome, Introductions, and Agenda Review
- Welcome and opening comments – *Linda Jones, Northwest Fisheries Science Center (NWFSC)*
 - Opening comments – *Usha Varanasi, NWFSC Science Director*
 - Introductions, agenda review, and ground rules – *Paul De Morgan, RESOLVE*
- 1:20 – 2:35 Management Needs
- NOAA Fisheries, NW Regional Office – Regulatory status of SRKWs and science needs to conserve them – *Garth Griffin*
 - Washington Department of Fish and Wildlife – *Rocky Beach*
 - Fisheries and Oceans Canada – Recovery planning and research needs – *Marilyn Joyce*
- 2:35 – 2:55 Review of High Priority Research Questions from Previous Workshops
- Overview – *Mike Ford, NWFSC*
 - Question and answer

GENETICS SESSION

High Priority Research Questions:

- *Are the North Pacific resident killer whales a distinct species or subspecies?*
- *What are the patterns of mating within the Southern Residents, and between the Southern Residents and other eastern North Pacific killer whale populations? Are intrinsic demographic problems, such as a lack of appropriate mates or inbreeding depression, limiting the Southern Resident's recovery?*

- *What was the historical size of the Southern Resident killer whale population? Is there genetic evidence that the Southern Resident's have experienced a severe genetic bottleneck?*

2:55 – 3:15 Genetics Presentations (Session Leader: Mike Ford)

- Mike Ford, NWFSC – Killer whale taxonomy and “species” status under the Endangered Species Act

3:15 – 3:30 BREAK

3:30 – 4:20 Genetics Presentations (cont.)

- Phil Morin, Southwest Fisheries Science Center – Genetic analysis of killer whale (*Orcinus orca*) historical bone and tooth samples to identify western U.S. ecotypes
- Phil Morin, Southwest Fisheries Science Center – On the use of AFLP markers for taxonomic study of killer whales (R. LeDuc, N. Hedrick, and R. Pitman)
- Rus Hoelzel, University of Durham – Evolution of population genetic structure of killer whales (*Orcinus orca*) in the North Pacific

4:20 – 4:50 Genetics Discussion

1. What are the significant results in terms of answering the high priority research question(s) or in better understanding the population and its conservation?
2. How can these results be used to inform the conservation of the Southern Resident killer whales?
3. Are there critical information gaps that still need to be addressed for conservation? If so, what are they? What method(s) is most likely to successfully address the gap taking into account biases and cost and logistical limitations?

POPULATION DYNAMICS SESSION

High Priority Research Questions:

- *What is the population growth rate of Southern Residents and what causes variation in the rate from that of Northern Residents?*
- *What factors are affecting the reproductive success of SRKWs?*
- *What are the factors affecting mortality?*
- *Do changes in the social structure affect the population's ability to recover?*

4:50 – 5:30 Population Dynamics Presentations (Session Leader: Eli Holmes)

- Eli Holmes, NWFSC – Southern Resident killer whale population dynamics and population viability analysis (P.R. Wade and K.C. Balcomb III)

- Peter Olesiuk, Fisheries and Oceans Canada – Life history and population dynamics of resident killer whales (*Orcinus orca*) in British Columbia and neighbouring waters

5:30 ADJOURN FOR THE DAY

6:30 – 8:30 EVENING RECEPTION: WATERTOWN HOTEL
4242 ROOSEVELT WAY, NE

All Symposium participants are invited to attend an evening reception at the Watertown Hotel from 6:30 p.m. to 8:30 p.m. Dr. Usha Varanasi, Northwest Fisheries Science Center Science Director, will welcome everyone and offer opening comments.

TUESDAY, APRIL 4

8:00 – 8:30 COFFEE AVAILABLE

POPULATION DYNAMICS SESSION (CONT.)

8:30 – 8:45 Population Dynamics Presentations (cont.)

- Ken Balcomb, Center for Whale Research – Long-term social dynamics of fish-eating killer whales (K.M.Parsons; K.C. Balcomb; J.W. Durban; J. K.B.Ford)

8:45 – 9:05 Population Dynamics Discussion

1. What are the significant results in terms of answering the high priority research question(s) or in better understanding the population and its conservation?
2. How can these results be used to inform the conservation of the Southern Resident killer whales?
3. Are there critical information gaps that still need to be addressed for conservation? If so, what are they? What method(s) is most likely to successfully address the gap taking into account biases and cost and logistical limitations?

CONTAMINANTS SESSION

High Priority Research Questions:

- *What are the differences in levels and patterns of contaminants among the Southern Resident pods, as well as differences between Southern Residents and other eastern North Pacific killer whale populations?*
- *Is there a relationship between exposure to contaminants in Southern Resident whales and their survivorship or reproductive success?*

9:05 – 9:55 Contaminants Presentations (Session Leader: Peggy Krahn)

- Peter Ross, Fisheries and Oceans Canada – Persistent organic pollutants in marine mammals inhabiting the transboundary waters of British Columbia-Washington
- Gina Ylitalo, NWFSC and Sandie O’Neill, WDFW – Regional patterns of persistent organic pollutants in five Pacific salmon species (*Oncorhynchus* spp) and their contributions to contaminant levels in northern and Southern Resident killer whales (*Orcinus orca*)

9:55 – 10:15 Contaminants Discussion

1. What are the significant results in terms of answering the high priority research question(s) or in better understanding the population and its conservation?
2. How can these results be used to inform the conservation of the Southern Resident killer whales?

3. Are there critical information gaps that still need to be addressed for conservation? If so, what are they? What method(s) is most likely to successfully address the gap taking into account biases and cost and logistical limitations?

10:15 – 10:30 BREAK

PREY SESSION

High Priority Research Questions:

- *What are the important prey species for SRKWs?*
- *Is prey abundance adequate to support SRKW populations?*
- *Is the quality of the prey adequate to meet the nutritional and energetic needs of killer whales?*

10:30 – 12:05 Prey Presentations (Session Leader: John Ford)

- John Ford, Fisheries and Oceans Canada – Selective foraging in resident killer whales
- Shannon McClusky, University of Washington – Movement patterns and population trends of Southern Resident killer whales in relation to relative abundance and distribution of Pacific Salmon in the Puget Sound – Georgia Basin
- Peggy Krahn, NWFSC – Feeding ecology of eastern North Pacific killer whales *Orcinus orca* from fatty acid, stable isotope and organochlorine analyses of blubber biopsies
- Brad Hanson, NWFSC – Investigations of associations of southern resident killer whales and their prey
- Jeff Haymes, WDFW – Washington Chinook Salmon abundance (B.Sanford)
- John Ford, Fisheries and Oceans Canada – Linking prey and population dynamics: did food limitation cause recent declines of resident killer whales?

12:05 – 12:35 Prey Discussion

1. What are the significant results in terms of answering the high priority research question(s) or in better understanding the population and its conservation?
2. How can these results be used to inform the conservation of the Southern Resident killer whales?
3. Are there critical information gaps that still need to be addressed for conservation? If so, what are they? What method(s) is most likely to successfully address the gap taking into account biases and cost and logistical limitations?

12:35 – 1:50 LUNCH

ENERGETICS SESSION

High Priority Research Question:

- *Is the quality of the prey adequate to meet the nutritional and energetic needs of killer whales?*

1:50 – 2:20 Energetics Presentations (Session Leader: Dawn Noren)

- Robin Dunkin, University of California, Santa Cruz – Seasonal variation in energetic status and body condition in killer whales (T.M. Williams)
- Samuel Wasser, University of Washington – Non-invasive monitoring of physiological health of Southern Resident killer whales

2:20 – 2:35 Energetics Discussion

1. What are the significant results in terms of answering the high priority research question(s) or in better understanding the population and its conservation?
2. How can these results be used to inform the conservation of the Southern Resident killer whales?
3. Are there critical information gaps that still need to be addressed for conservation? If so, what are they? What method(s) is most likely to successfully address the gap taking into account biases and cost and logistical limitations?

DISTRIBUTION AND HABITAT SESSION

High Priority Research Questions:

- *What habitats are important for Southern Resident killer whales, particularly in outer coastal waters?*

2:35 – 4:00 Distribution and Habitat (Session Leader: Brad Hanson)

- Ken Balcomb, Center for Whale Research – Winter distribution of Southern Resident killer whales, 2003-2006
- Jeff Nystuen, Applied Physics Laboratory, University of Washington – Listening for orcas in the underwater racket of Cape Flattery and Haro Strait
- John Hildebrand, Scripps Institution of Oceanography, University of California, San Diego – Killer whale acoustic monitoring in the Coastal Waters of Washington
- Donna Hauser, University of Washington – Effects of environmental factors and temporal scale on pod-specific Southern Resident killer whale summer distribution patterns: implications for designating critical habitat

4:00 – 4:15 BREAK

- 4:15 – 4:45 Distribution and Habitat Discussion
1. What are the significant results in terms of answering the high priority research question(s) or in better understanding the population and its conservation?
 2. How can these results be used to inform the conservation of the Southern Resident killer whales?
 3. Are there critical information gaps that still need to be addressed for conservation? If so, what are they? What method(s) is most likely to successfully address the gap taking into account biases and cost and logistical limitations?

VESSEL INTERACTIONS SESSION

High Priority Research Question:

- *Does vessel presence affect Southern Resident killer whales?*

- 4:45 – 5:45 Vessel Interactions Presentations (Session Leader: Dawn Noren)
- Dave Bain, University of Washington – Land-based studies of the effects of vessel traffic on the behavior of Northern and Southern Resident killer whales (*Orcinus* spp.)
 - Dawn Noren, NWFSC – Behavioral energetics of Southern Resident killer whales
 - Jennifer Marsh, University of Washington – Social behavior of Southern Resident killer whales (*Orcinus orca*)

5:45 ADJOURN FOR THE DAY

WEDNESDAY, APRIL 5

8:00 – 8:30 COFFEE AVAILABLE

VESSEL INTERACTIONS SESSION (CONT.)

8:30 – 9:00 Vessel Interactions Discussion

1. What are the significant results in terms of answering the high priority research question(s) or in better understanding the population and its conservation?
2. How can these results be used to inform the conservation of the Southern Resident killer whales?
3. Are there critical information gaps that still need to be addressed for conservation? If so, what are they? What method(s) is most likely to successfully address the gap taking into account biases and cost and logistical limitations?

NOISE EFFECTS SESSION

High Priority Research Question:

- *Does vessel noise adversely affect Southern Resident killer whales?*

9:00 – 10:35 Noise Effects Presentations (Session Leader: Brandon Southall)

- Brandon Southall, Office of Science and Technology, NMFS – Acoustic creatures in noisy environments: effects of sound on marine wildlife
- John Hildebrand, Scripps Institution of Oceanography, University of California, San Diego – Ambient noise in the Haro Strait from whale watching and commercial vessels
- Val Veirs, Colorado State – Average levels and power spectra of ambient sound in the habitat of Southern Resident orcas
- Mike Wolfson, Applied Physics Laboratory, University of Washington – The acoustic environment of Haro Strait: A data-model comparison of shipping traffic noise (Chris Jones)
- Andrew Foote, University of Durham – Acoustic studies of the Southern Resident killer whale population: implications for remote acoustic monitoring and indications of vocal behavioral change due to vessel noise

10:35 – 10:50 BREAK

10:50 – 11:20 Noise Effects Discussion

1. What are the significant results in terms of answering the high priority research question(s) or in better understanding the population and its conservation?

2. How can these results be used to inform the conservation of the Southern Resident killer whales?
3. Are there critical information gaps that still need to be addressed for conservation? If so, what are they? What method(s) is most likely to successfully address the gap taking into account biases and cost and logistical limitations?

INVITED PANEL ROUNDTABLE

The following individuals will be reflecting on the Symposium presentations and discussions and offering their insights and views on related questions:

- Ken Balcomb, Center for Whale Research
- John Durban, Alaska Fisheries Science Center
- John Ford, Fisheries and Oceans Canada
- Brandon Southall, Office of Science and Technology, NMFS
- Laurie Weitkamp, NWFSC
- Judy Zeh, University of Washington

11:20 – 12:30 Invited Panel Roundtable and Open Discussion – Session I

- After assessing all the information, what are the significant results in terms of answering the high priority research question(s) or in better understanding the population and its conservation?
- Can the risk factors that have been identified be ranked in terms of their impacts to the population?
- What are the potential interactions between risks factors?
- How can these results be used to inform the conservation of the Southern Resident killer whales?

12:30 – 1:45 LUNCH

1:45 – 2:45 Open Discussion – Session I (continued)

2:45 – 3:30 Invited Panel Roundtable and Open Discussion – Session II

- What are the highest priorities for future research?
- Are there critical information gaps that still need to be addressed for conservation? If so, what are they? What method(s) is most likely to successfully address the gap taking into account biases and cost and logistical limitations?

3:30 – 3:45 BREAK

3:45 – 4:30 Open Discussion – Session II (continued)

4:30 – 4:45 Next Steps and Acknowledgments

4:45 ADJOURN

Killer whale taxonomy and “species” under the Endangered Species Act

Michael Ford

Northwest Fisheries Center, NOAA Fisheries, Seattle, WA.

The ESA allows listing of species, subspecies, and (for vertebrates only) “distinct population segments” (DPS). In 1996, the US Fish and Wildlife Service and US National Marine Fisheries Service (NMFS) issued a joint policy defining a DPS as a population that is both discrete (demographically isolated) and biologically significant to the species as a whole. The question of “significance” was a significant factor influencing the listing decision of the Southern Residents. In 2002, the NMFS declined to list the Southern Resident population because it failed to meet the DPS criteria. In 2004, after a court ruling on how the NMFS should consider scientific uncertainty relating to the taxonomy of *Orcinus orca*, a NMFS Biological Review Team (BRT) determined that the Southern Residents met the criteria for being a DPS of an unnamed subspecies of resident killer whales. In considering the Southern Resident’s “significance”, the review team considered four factors: ecological setting, the population’s range compared to other populations, genetic differentiation, and behavioral and cultural diversity. After considering these factors and the recent results of a NMFS-sponsored international workshop that reviewed killer whale taxonomy, the BRT concluded that the Southern Resident population was a DPS of an unnamed North Pacific resident (fish eating) subspecies of killer whale.

Genetic analysis of killer whale (*Orcinus orca*) historical bone and tooth samples to identify western U.S. ecotypes (Manuscript in press, *Marine Mammal Science*)

Phillip A. Morin^{1*}, Richard G. LeDuc¹, Kelly M. Robertson¹, Nicole M. Hedrick¹, William F. Perrin¹, Michael Etnier², Paul Wade², Barbara L. Taylor¹

(1)NMFS, Southwest Fisheries Science Center, 8604 La Jolla Shores Drive, La Jolla, CA 92037, U.S.A.

(2) National Marine Mammal Laboratory, 7600 Sand Point Way N.E., Seattle, WA 98115

Introduction:

Three different types of killer whales have been recognized in the eastern North Pacific. Available evidence indicates the “resident” type eats fish, and is possibly a salmon specialist in some areas (Ford et al. 1998; Saulitis et al. 2000). The “transient” type eats primarily marine mammals (Ford et al. 1998; Saulitis et al. 2000). The “offshore” type has been observed apparently eating fish but its prey preferences are not well described (Ford et al. 2000).

Three discrete populations of resident-type whales have been described. In summer, the “southern resident” population is found primarily in Washington and southern British Columbia. Their home range during the spring, summer, and fall includes the inland waterways of Puget Sound, the Strait of Juan de Fuca, and the Strait of Georgia, where they are known to eat Chinook and chum salmon. Their occurrence in the coastal waters off Washington, Vancouver Island, and more recently off the coast of Oregon and central California has been documented (Krahn et al. 2002).

The southern resident population declined substantially from 1996 to 2001 (Krahn et al. 2002), and has been listed as endangered under the Endangered Species Act (ESA) (Krahn et al. 2002; Reeves et al. 2004). This population may have been significantly larger in the past (Krahn et al. 2002), and may have had a wider distribution. Substantial declines in native salmon populations have been documented in this region in recent decades (Krahn et al. 2002). Salmon-eating killer whales, if formerly present at all of the major salmon runs and dependent on them, may have constricted their range to inland Washington (in summer) subsequent to the salmon population declines. An understanding of the significance of the population requires a better understanding, if possible, of the historical distribution of this population and the other ENP ecotypes.

This study focused on 30 historical samples (Table 1), most of which were from bone and teeth of animals sampled before 1980 south of inland Washington waters (Figure 1). We have used ancient DNA analysis methods to assay diagnostic sites in mitochondrial DNA haplotypes to classify the historical bone and tooth samples, to determine the historical distribution of the ENP haplotypes along the Washington, Oregon, and California coasts.

Results:

The DL5/DH6 portion of the killer whale mitochondrial control region contains diagnostic nucleotide sites that distinguish the killer whale ecotypes found in the eastern

North Pacific (Table 2) . We obtained DNA sequence data from 25 of 30 samples (83%). Of the 25 sequences, eight (8) were of the offshore haplotype, twelve (12) were of the transient haplotype, four (4) were of the southern resident haplotype, and one (1) was of the northern resident haplotype (Table 1). In every case except one, diagnostic haplotypes were from animals sampled in the region of their current known distribution. Specifically, southern resident haplotypes were from samples collected in Washington, and transient and offshore haplotypes were found in all regions. The one exception was a single northern resident haplotype found from a sample collected somewhere off the coast of California (Table 1). This sample represents the southernmost sample to date of that population, if it is indeed from California as museum records indicate.

Six whales that were genetically identified had stomach content data available as well. All of the stomach contents were consistent with what is known of killer whale diets for each ecotype .

Conclusions:

These data from historical samples, combined with museum and NMFS records from the original sample collections, provide additional insight into the historical distributions of killer whale populations, ecotypes and their diets. These data provide no evidence to support the hypothesis that the southern resident killer whale population was formerly distributed south of Washington state along the Oregon and California coasts. However, only six samples from northern California, in the hypothesized historical range of southern residents, were available, so given the small sample size and the presence of other killer whale ecotypes known to live at least part of the year off of the western US coast, these data do not exclude the possibility that southern residents also once included these areas in their normal range. Further sampling of museum samples of whales from this region might further define the historical ranges of each of the ENP killer whale ecotypes.

References:

- Ford, J. K. B., G. M. Ellis and K. C. Balcomb. 2000. Killer whales: The natural history and genealogy of *Orcinus orca* in British Columbia and Washington State. Vancouver, BC, Canada, UBC Press.
- Ford, J. K. B., G. M. Ellis, L. G. Barrett-Lennard, A. B. Morton, R. S. Palm and K. C. Balcomb. 1998. Dietary specialization in two sympatric populations of killer whales (*Orcinus orca*) in coastal British Columbia and adjacent waters. *Canadian Journal of Zoology* 76(8): 1456-1471.
- Krahn, M. M., P. R. Wade, S. T. Kalinowski, M. E. Dahlheim, B. L. Taylor, M. B. Hanson, G. M. Ylitalo, R. P. Angliss, J. E. Stein and R. S. Waples. 2002. Status review of Southern Resident killer whales (*Orcinus orca*) under the Endangered Species Act. NOAA Technical Memorandum NMFS-NWFSC 54: 1-133.
- Reeves, R. R., W. F. Perrin, B. L. Taylor, C. S. Baker and S. L. Mesnick. 2004. Report of the Workshop on the Shortcomings of Cetacean Taxonomy in Relation to Needs of Conservation and Management. Cetacean Systematics: Approaches in genetics, morphology, and behavior, April 30 - May 2, La Jolla, CA, USA.

Saulitis, E., C. Matkin, L. G. Barrett-Lennard, K. Heise and G. M. Ellis. 2000. Foraging strategies of sympatric killer whale (*Orcinus orca*) populations in Prince William Sound, Alaska. *Marine Mammal Science* 16(1): 94-109.

Table 1:

Killer whale sample information. Under “Haplotype ID”, O=offshore, T=transient, SR=southern resident, NR=northern resident, as determined from DNA sequences.
*samples that were re-extracted and re-sequenced.

LAB ID	FIELDID	TISSUE TYPE	AMOUNT USED (g)	DNA concentration (copies/ μ l)	Haplotype ID	SOURCE	YR	LOCALITY
26641	SMI602	tooth	0.2	69		ARCHEOLOGICAL-NMML		CA, SAN MIGUEL IS.
34079	USNMA0013018	muscle	0.15	107	T	USNM	mid-1800's	CA
34080	USNMA0016487	skull bone	0.19	94	O	USNM	mid 1800's	CA
34081	USNMA0016488	tooth	0.2	16*	NR	USNM	mid 1800's	CA
34082	USNMA0016625	tooth	0.19	1548	SR	USNM	<1883	WA, CLALLAM CO., CAPE FLATTERY
34087	USNMA0049909	periosteal bone	0.15	--		USNM		CA, SANTA CATALINA IS., AVALON
34100	LACM54444	tooth	0.19	802	T	STRAND-CA-LACM	1973	CA, SANTA CRUZ IS., WILLOWS ON SE. SIDE
34515	LACM22791	tooth	0.12	1870	O	STRAND-MEXICO-LACM	1951	MEXICO, BAJA CALIFORNIA, BAHIA DE SEBASTIAN VISCAINO
34516	LACM30461	skull bone	0.14	1		STRAND-CA-LACM	1927	CA, LOS ANGELES CO., HERMOSA BEACH
34517	LACM52455	skull bone	0.21	57	O	STRAND-CA-LACM	1961	CA, ORANGE CO., NEWPORT BEACH
34521	LACM72550	skull bone	0.17	4695	O	STRAND-CA-LACM	1985	CA, HUNTINGTON BEACH, BOLSA CHICA STATE BEACH
37270	SBMNH979	tooth	0.12	2108	O	STRAND-CA-SBMNH	1981	CA, SANTA CRUZ IS., CHINA HARBOR
37271	SBMNH1546	tooth	0.22	258	T	STRAND-CA-SBMNH	1977	CA, MORRO BAY SAND SPIT, 1/4 MI. N. OF HAZ AND CANYON
37272	SBMNH4074	tooth	0.15	0.3	T	STRAND-CA-SBMNH	1990	CA, SANTA BARBARA CO., SANTA CRUZ IS.
37273	SBMNH-NA-CA-125-13C-2	tooth	0.13	11		Archeological-CA-SBMNH		CA
39060	NMML0078	tooth	0.18	2674	T	NMML	1961	CA, SAN FRANCISCO
39061	NMML0079	tooth	0.13	105	T	NMML	1963	CA
39062	NMML0080	tooth	0.18	2377	O	NMML	1964	CA
39063	NMML0081	tooth	0.21	224*	T	NMML	1965	CA, PT. CONCEPTION
39064	NMML0082	tooth	0.19	3037	T	NMML	1966	CA, OFF SAN FRANCISCO
39065	NMML0083	tooth	0.16	37060	SR	NMML	1967	WA, YUKON HARBOR, PUGET SOUND
39066	NMML0084	tooth	0.14	3495	T	NMML	1967	CA
39067	NMML0085	tooth	0.12	28240	T	NMML	1967	CA
39068	NMML0086	tooth	0.19	11		NMML	1970	WA, WHIDBEY IS., PUGET SOUND
39069	NMML0087	tooth	0.14	--*	O	NMML	1966	CA, MORRO BAY

39070 NMML0088	tooth	0.23	--*	SR	NMML	1967	WA, YUKON HARBOR, PUGET SOUND
39071 NMML0089	tooth	0.18	--*	SR	NMML	1967	WA, YUKON HARBOR, PUGET SOUND
39075 MVZ-129686	a=tooth, b=muscle	.19 .18	107800 201	T	MUSEUM- BERKELEY-MVZ	1962	CA, SAN FRANCISCO CO., 70 MI. W SAN FRANCISCO
39076 MVZ-134462	muscle (dry)	0.08	391600	T	MUSEUM- BERKELEY-MVZ	1966	CA, SONOMA CO., GOAT RK., MOUTH OF RUSSIAN RIVER
39077 MVZ-184169	vertebrae	0.24	38*	O	MUSEUM- BERKELEY-MVZ	1964	CA, 15 MI. W SAN MIGUEL IS.

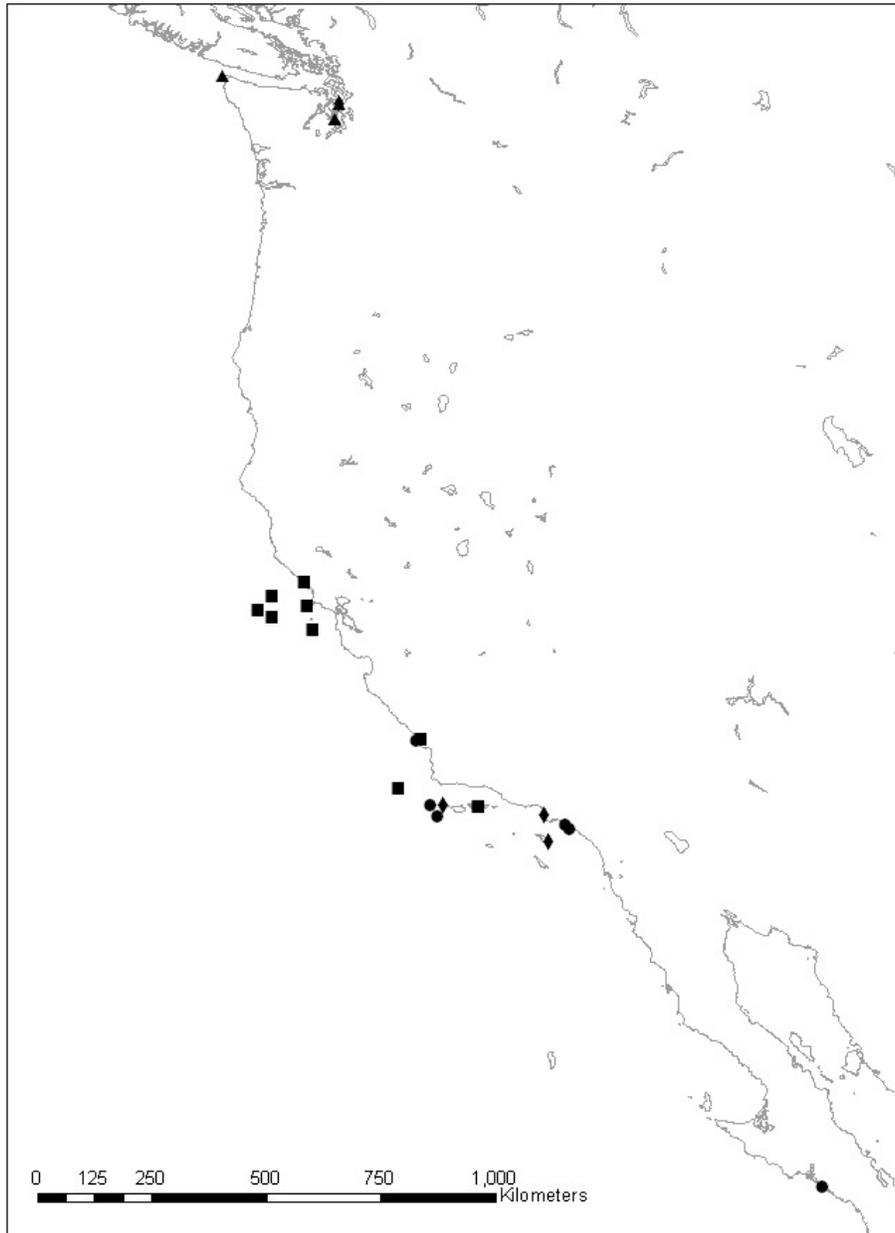
Table 2:

Diagnostic sites for eastern North Pacific killer whale sequences. The numbers refer to the variable sites in the DNA sequence.

Variable Site	Offshore	Transient	Southern Resident	Northern Resident
1	C	T	C	C
2	C	C	C	T
3	T	C	T	T
4	C	T	T	T

Figure 1:

Original sampling locations (when known) for killer whale samples used in this study. Samples with imprecise collection locations (e.g., 'California') are not shown, or are shown in general location (e.g., Cape Flattery, WA). Haplotypes are: Square = transient, circle = offshore, triangle = southern resident, diamond = unknown.



On the use of AFLP markers for taxonomic study of killer whales

R. LeDuc, N. Hedrick, and R. Pitman
Southwest Fisheries Science Center, NMFS

INTRODUCTION:

The taxonomic uncertainty surrounding killer whales has direct consequences for their management and conservation. Specifically, recognition of a population as a distinct population segment (DPS) requires that the taxon to which the population belongs be defined. However, the number of extant species of killer whales is still an open question. Reproductive isolation between North Pacific killer whale ecotypes is indicated by their ecological distinctness and behavioral segregation and has been supported by data from the maternally inherited mitochondrial genome. However, the nuclear genetic (i.e., microsatellite) analyses to date (Hoelzel 2002) have not corroborated these results. This may be due in part to the nature of microsatellite data; these highly variable markers are useful for differentiating populations, but are rarely used for examining species-level differences. In other words, unlike sequence data which can reveal fixed differences or phylogenetic monophyly, microsatellite data do not lend themselves to using simple criteria for discerning species differences. It has also been difficult to put the variation seen in the North Pacific into a context of global variation in killer whales, primarily due to the paucity of samples and data from outside the Pacific basin.

Recent work (Kingston and Rosel 2004) used amplified fragment length polymorphisms (AFLPs) to successfully discriminate between closely related species of small delphinids, species that do not always clearly segregate in analyses of mitochondrial sequence data. Unlike microsatellites, which depend on variation in the number of repeats within relatively few markers, AFLP variation represents base substitutions recorded from a broader sampling of the overall genome. Here we test the feasibility of using AFLP markers to examine differentiation within Antarctic killer whales using samples already on hand at the SWFSC.

There are three distinct morphotypes inhabiting the Antarctic (Pitman and Ensor 2003): Type A, a typical killer whale that inhabits open water and is thought to feed primarily on minke whales; Type B, a smaller form that favors the pack ice and specializes on pinnipeds; and Type C, another small, ice-inhabiting form that specializes on fish. In addition to the variation in body size, each of these forms is also distinct in its color pattern of dorsal cape and eye patch shape. An initial study using mitochondrial sequence data (LeDuc and Pitman 2004) found several fixed differences between Type A and the two other types, and a single fixed difference between Types B and C. It is hoped that the AFLP data will shed some light on any differentiation of the nuclear genome that may exist.

MATERIALS AND METHODS:

Samples

Six samples from Antarctic waters were used in this study, two each of the three morphotypes discussed above (samples A1, A2, B1, B2, etc.). All were collected as biopsies from free-swimming killer whales. DNA was extracted using standard methodologies.

AFLPs

The AFLP assay was run according to a modified protocol of Kingston and Rosel (2004). The resulting data are an amplified view of the fingerprint created by performing initial restriction enzyme cuts and subsequent sequence specific amplifications in each sample's genome. This method is composed of five main steps; digestion, adaptor ligation, pre-selective PCR, selective PCR using fluorescent primers, and electrophoresis and analysis.

Total DNA was digested with two restriction enzymes simultaneously, Eco RI and TaqI (Kingston and Rosel, 2004). Restriction enzymes cut the DNA into fragments wherever specific short sequences of bases occur. Double stranded synthetic adapters were then ligated to the fragments, providing DNA of known sequence attached to the ends of the fragments. These adapters served as templates for the annealing of primers in the subsequent PCR. Two rounds of PCR amplified the labeled fragments. The first, pre-selective PCR, amplified the fragments using primers complementary to the adapters. This step served to increase the total number of copies of the fragments. The second, selective fluorescent PCR, utilized primers with three additional bases that extended into the native DNA fragment beyond the adapter sequence. This allowed selection of different fragment sets depending on the bases chosen for the primers and present in the fragment. These fluorescent PCR products were run on an Applied Biosystems 3100 sequencer per Applied Biosystems AFLP protocols and settings. The resulting data files were analyzed and scored using Genemapper version 4.0 (Applied Biosystems, Foster City, CA).

RESULTS AND DISCUSSION:

The different combinations of pre-selective and selective PCR constituted 48 assays applied to the samples. Seventeen of these assays have been examined to date. The data from each assay consisted of fluorescent bands representing alleles from a broad range of sizes. Each allele was scored as present (1) or absent (0) in each individual. The genetic source of variation can arise in two different ways. First, a mutation in the short sequences recognized by the restriction enzymes will result in the loss of a particular fragment, which in turn will not be amplified in the subsequent PCR steps. Second, mutations in the sequence near the end of the fragment will mean that the primers in the second, selective, PCR will not anneal and the fragment will fail to amplify. In either case, it is thought that the presence and absence of alleles reflects point mutations within the sequence of the DNA fragments.

Only alleles that were between 75 and 300 base pairs long were used, as this size range proved to be the most reliable in replication. The number of alleles present in the

killer whales, as well as the proportion of these that were variable, differed greatly among assays, with some yielding very few total and/or none that were variable. From the seventeen assays examined, there were a total of 249 variable alleles within killer whales.

Various types of analyses of these binary data can be performed, such as the principal coordinates analyses conducted by Kingston and Rosel (2004), or even tree-building methods such as parsimony. However, here we only attempt to characterize the amount and general nature of the variation. Of the 249 variable alleles, the proportion displayed by individual samples averaged 0.482, with a low of 0.426 (A2) and a high of 0.562 (A1). Table 1 shows the percent of allelic states that were different between samples.

Table 1. The proportion of AFLP alleles that differed between samples (i.e., present in one and absent in another), and averages between types.

Sample	A2	B1	B2	C1	C2
A1	0.474	0.554	0.582	0.562	0.486
A2		0.474	0.494	0.418	0.446
B1			0.301	0.313	0.341
B2				0.325	0.417
C1					0.309
Averages					
A vs. B	0.526				
A vs. C	0.478				
B vs. C	0.348				

In spite of the small sample size, the present results are an encouraging sign that the amount and type of variation in AFLP data make this a useful tool for examining the amount of genetic differentiation among killer whale types. In some respects, the pattern of variation is similar to that of the mitochondrial data, in that the differences between B and C (0.348, ranging from 0.313 – 0.417) are on a similar scale as the differences within each form (0.301 and 0.309 for B and C, respectively), although they do not overlap. The primary overlap in values is between the within-A comparison (0.474) and between A and the other two forms: equal to one of the A-B comparisons and exceeding two of the A-C comparisons. This result is not surprising in light of our current understanding. If the Type A killer whales are seasonal visitors in pursuit of migratory minke whales, they likely represent multiple populations from lower latitudes, perhaps even from different ocean basins. These would therefore be expected to exhibit a greater degree of genetic diversity within the morphotype. The sampling may reflect this; while each sample of Type B and C killer whales was sampled fairly close to its partner (both Type Bs came from near the Antarctic Peninsula and both Cs from near the Ross Sea), the two Type A samples were taken from fairly disparate locations (60-12 ° S 45-30 ° W and 62-56 ° S 128-57 ° E).

CONCLUSIONS

The present results, though encouraging, are certainly preliminary. Besides the larger sample set available from LeDuc and Pitman (2004), additional samples of all three

forms from new areas have been collected and are awaiting importation. In addition, the remainder of the 48 assays conducted has yet to be examined. Nevertheless, the results indicate that AFLP data can serve as a valuable tool for examining variation within and between killer whale types and populations, not only in the Antarctic but in the North Pacific and other ocean basins.

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Evolution of population genetic structure of killer whales (*Orcinus orca*) in the North Pacific

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Killer whales have revealed very little mtDNA variation worldwide (Figure 1; Hoelzel et al. 2002), however there are fixed differences between some populations (e.g. Hoelzel et al. 1998). Hoelzel et al. (2002) suggested that they had been through a population bottleneck (based especially on tests for neutrality, coalescent based evidence for expansion, and the pattern of diversity), and noted that if so the magnitude of mtDNA differences among populations may sometimes reflect differences among remnant lineages that survived the bottleneck, rather than differentiation related to time in isolation. This together with the matrilineal structure of local populations means that mtDNA is relatively uninformative about migration among populations of this species (except to show that female migration has not occurred between those populations that show fixed mtDNA differences). However, male-mediated gene flow remains a possibility. Here we present data based on 16 nuclear (microsatellite DNA) markers. These loci are biparentally inherited, and therefore reflect the dispersion of both males and females. We show evidence for historical gene flow among all of the North Pacific populations included in our sample (southern residents, Southeast Alaskan residents, Bering Sea residents, Russian (Kamchatka) residents, Southeast Alaskan transients, Californian transients, and offshores), but significant levels of differentiation both for paired comparisons and for all populations combined. Populations of the same ecotype in allopatry showed a similar level of differentiation as that seen between different ecotypes (transients and residents) in sympatry, and there was a linear relationship between genetic differentiation (measured using F_{ST}) and geographic distance between putative populations within an ecotype.

This study also further developed a new method of analysis of population structure, proposed by Nielsen and Wakeley (2001), and extended by Hey and Nielsen (2004), with a data set of 15 microsatellite loci (omitting one locus to avoid problems associated with possible deviation from the stepwise mutation model). The goal was to assess the size and age of regional populations, and to determine whether or not they have been exchanging genes. Pairs of populations were analyzed under the Isolation with Migration model (Figure 2). This is a general model of population divergence in which an ancestral population splits into two descendant populations. Importantly the two descendant populations may exchange genes (Hey and Nielsen 2004; Nielsen and Wakeley 2001). The method simultaneously estimates the time point at which populations began to diverge, the average rate of gene flow since that time point, and the modern and ancestral effective population size.

The assessment of population structure based on Bayesian likelihood estimates (using STRUCTURE) is given in Figure 3. The highest likelihood was found for $K = 7$ populations, and the likelihood value was flat among the four replicate runs. All putative populations were supported with the exception of the 'Bering Sea' population, which appeared to be composed of a mixture of animals from Kamchatka and Southeast Alaska, perhaps indicating a population

boundary in the Bering Sea region. Note that all of these animals have the northern resident mtDNA haplotype, as do the Southeast Alaskan residents, while the Kamchatka animals have the southern resident mtDNA haplotype. After assigning 7 populations as indicated by the initial runs, we could identify several putative migrants. These are indicated in Figure 3 with arrows. Only those with a significance of $p < 0.001$ are shown. One of these suggests a migration event between the Offshore and Transient communities (between the two main mtDNA lineages in the North Pacific).

Further assessment of gene flow was undertaken using the private allele method, the coalescent method implemented in MIGRATE, and the MCMC simulation method (the isolation with migration model) adapted to this analysis as part of this study (Figure 4). The pattern and level of gene flow indicated was similar using all three methods. In each case low levels of gene flow were indicated between all pairs of putative populations, and the magnitude did not differ for comparisons within compared to between ecotypes (transients vs residents).

The estimated effective size of regional populations were fairly small, generally under 1000 and possibly less than 50 (depending on the mutation rate assumed). Further, the marine-mammal eating ecotype showed consistently larger effective population sizes than the fish-eating ecotype (e.g. using a mutation rate of 7.5×10^{-8} , the average for Alaskan residents was 67 while the average for Alaskan transients was 205). A second pattern is that the regional populations appear to have persisted for thousands of years and possibly, depending on the mutation rate, for tens of thousands of years (but less than 20,000 years). The effective population size estimates of the sampled populations were invariably a small fraction of the estimate for the ancestral population. It is possible that the sampled regional populations all evolved as founders from one or a few larger ancestral populations. For example, the coastal habitat occupied by these populations only became available after the ice retreated some 18,000 years ago, and coastal populations may have been founded after that. However if the ancestors of these populations were themselves exchanging genes, then the large estimated sizes for ancestral populations could be a byproduct of the method, which imposes a strict two population/one ancestor model on a species that may well have had ongoing gene flow among multiple regional populations.

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Figure 1: Linearized neighbour joining tree. Kimura two-parameter genetic distances are shown. Bootstrap values based on 1,000 replications (after Hoelzel et al. 2002). First three letters of haplotype code indicate oceanic location (e.g. ENP= eastern North Pacific).

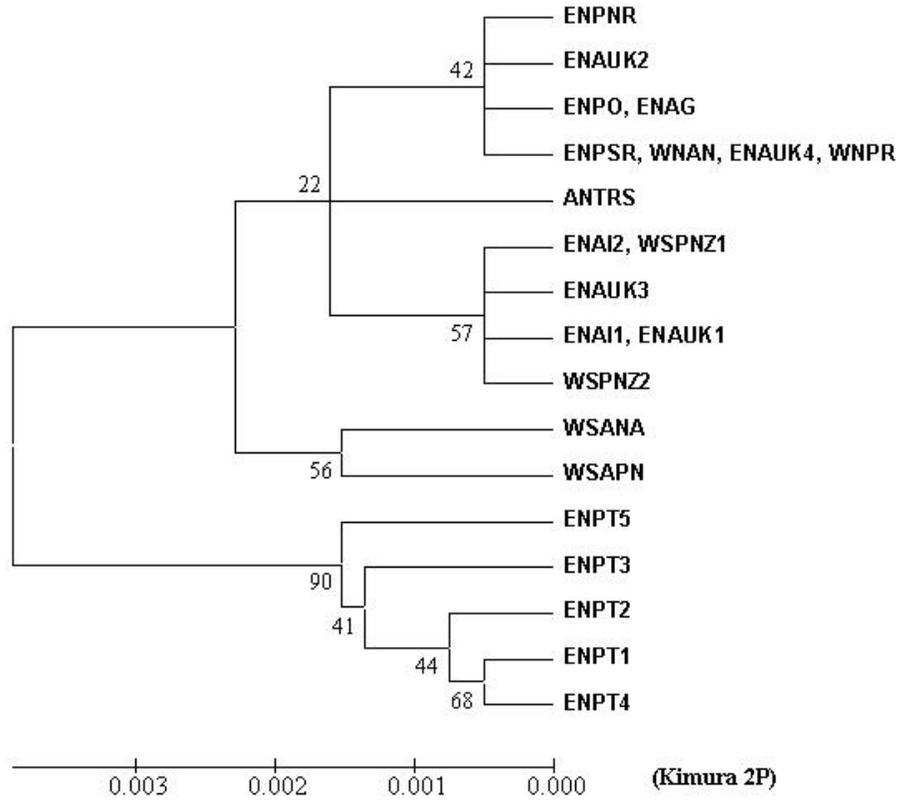


Figure 2: The Isolation with Migration model, showing model parameters (Hey and Nielsen 2004)

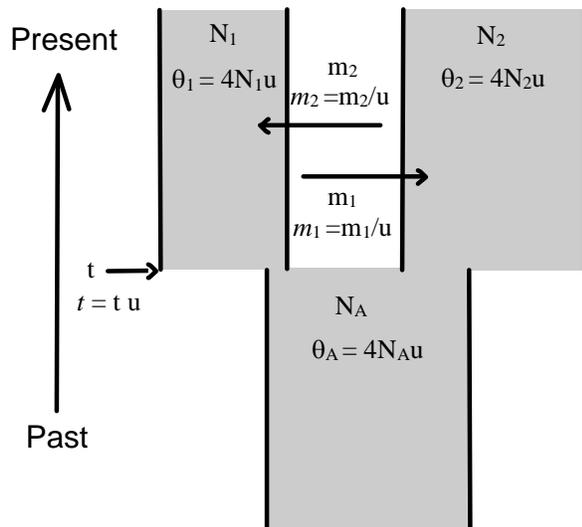


Figure 3: Proportional assignment to one of seven putative populations (in seven different colors) for each of the 211 individual whales in the study. Population of origin is indicated below the histogram, and putative migrants are indicated above with arrows.

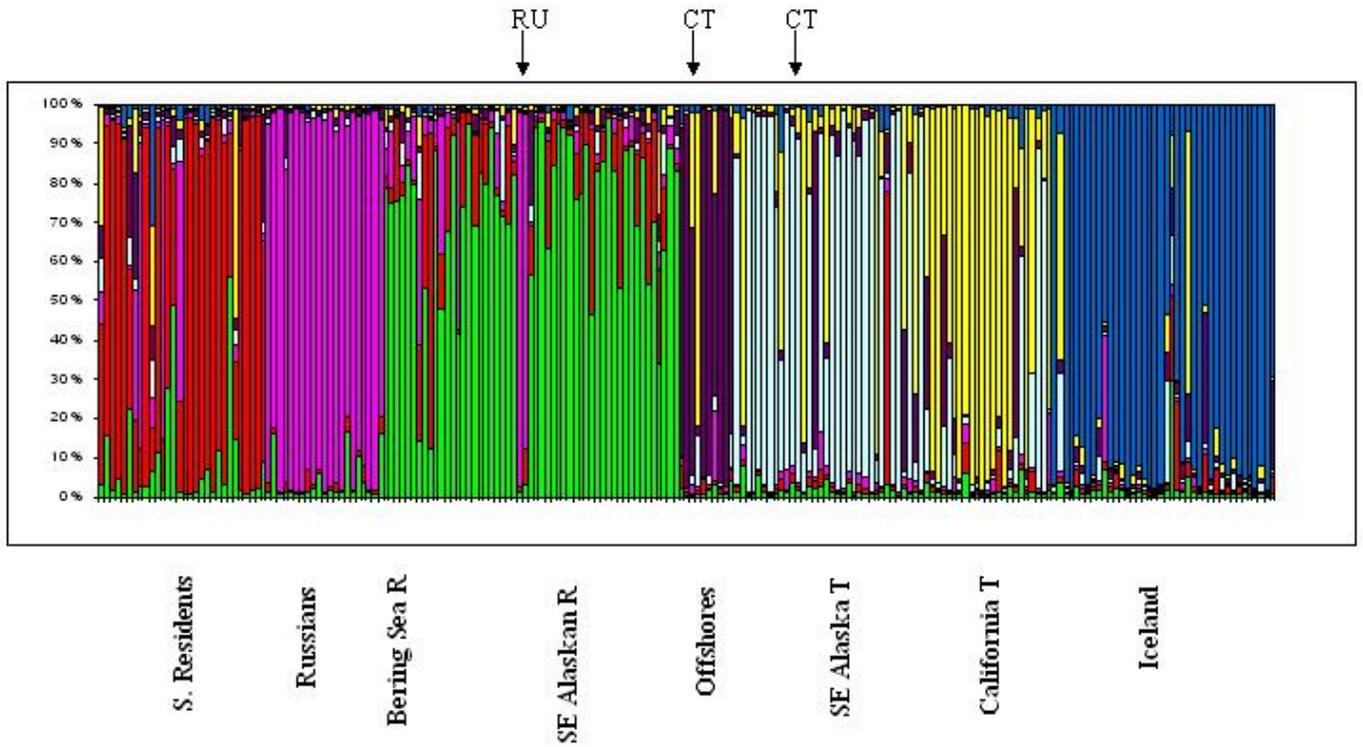
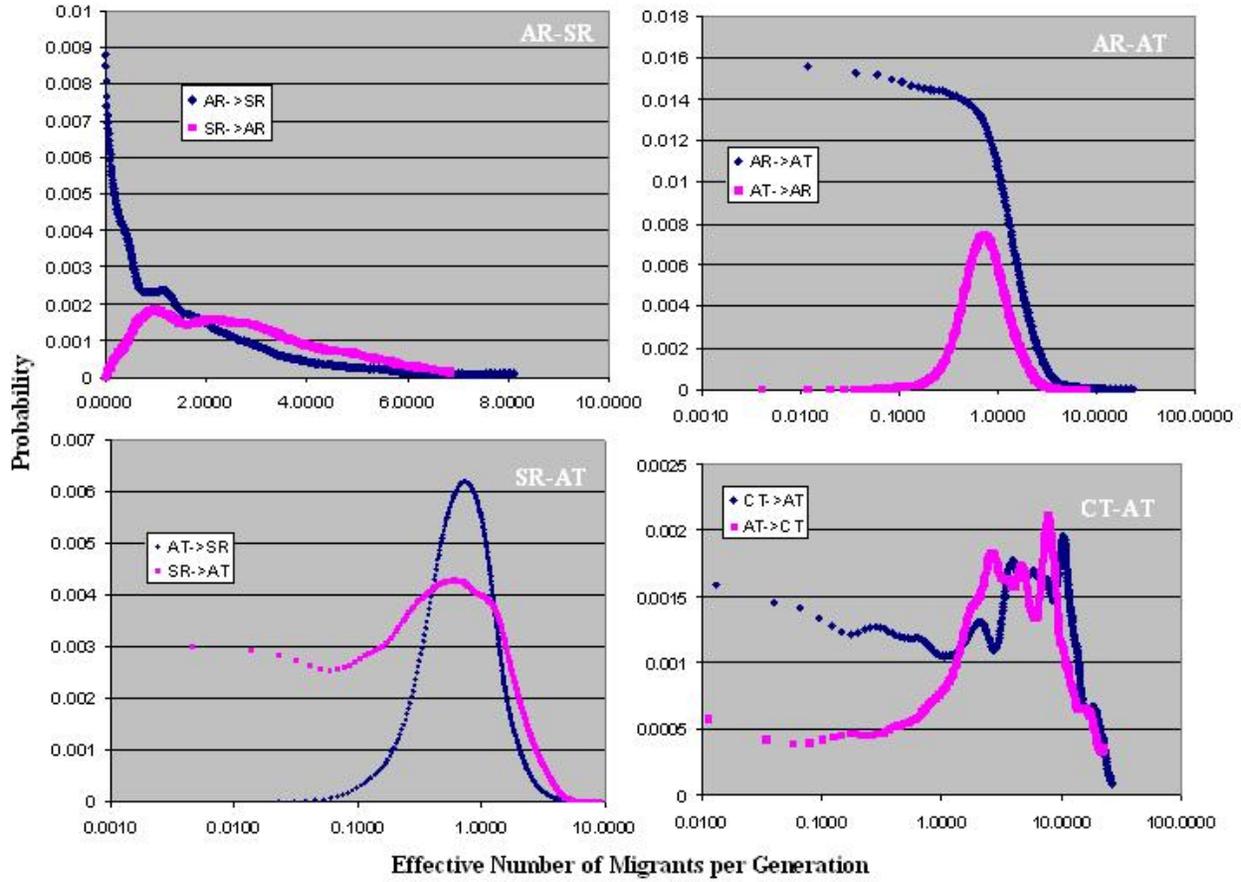


Figure 4: Migration rates estimates based on MCMC simulations. AR= Alaskan residents, SR= southern residents, AT= Alaskan transients, CT= Californian transients. Migration estimates are directional (see keys).



Southern Resident Killer Whale Population Dynamics and Population Viability Analysis

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In May and June of each year since 1973–74, the Center for Whale Research, Friday Harbor, Washington, has taken photographs that identify every Southern Resident individual. As a result, the annual survey amounts to a census of the entire population. This census allows a detailed examination of the population dynamics of the Southern Resident community, including the survival and fecundity of individual animals of known age. In 1974 the population comprised 71 whales, whereas the census in the summer of 2003 counted 83 whales, representing an overall annual increase of 0.4% per year. However, the population has fluctuated considerably over the 29 years of the study (Figure 1). Using the maximum recorded population size of 97 animals in 1996, the Southern Resident killer whale population declined by 2.2% per year between 1996 and 2003.

Large differences exist in survival rates of Southern Residents among different age and sex categories. Reproductive-age females had the highest survival rate, followed by juveniles, post-reproductive-age females and young males. Calves and old males had the lowest survival rates. In addition, there have also been large changes in survival rates through time for all age and sex categories. Survival has shifted from relatively high levels in the 1970s, to low levels in the early 1980s, to high levels again in the late 1980s and early 1990s, then returned to low levels through 2001. Survival rates improved in 2001 and 2002, but they were not as high as in previous periods of relatively high survival.

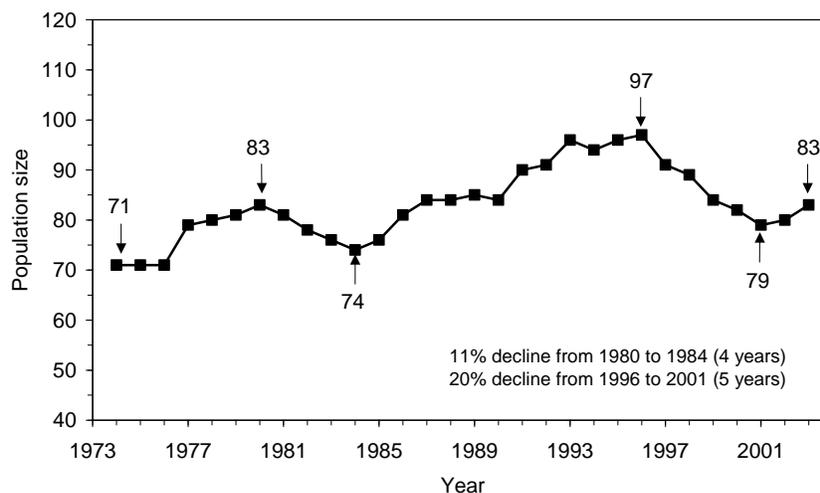


Figure 1. Southern Resident killer whale population size in June through time (includes L98).

A Population Viability Analysis (PVA) was conducted to synthesize the potential biological consequences of a small population size, a slowly increasing or a declining population trend and potential risk factors. The probability of the Southern Resident population going extinct was estimated using demographic information from the yearly census. Both the probability of extinction (defined as <1 male or female) as well as the probability of “quasi-extinction,” (defined as ≤ 10 males or females) were determined, because the BRT believed that a population at the quasi-extinction level would be “doomed” to extinction, even though literal extinction might still take decades for long-lived killer whales. Under the assumption that growth rates in the future will accurately be predicted by the full 29-year time series of available data (the most optimistic scenario considered), the model predicted the probability that Southern Resident would become extinct was <0.1 –3% in 100 years and 2–42% in 300 years. If a quasi-extinction threshold was used instead of actual extinction, the predicted probability of meeting the threshold ranged 1–15% in 100 years and 4–68% in 300 years. For both scenarios, the higher percentages in each range were associated with higher probability and magnitude of catastrophic mortality events (e.g., oil spills), as well as with a smaller carrying capacity (i.e., $K = 100$) (Figure 2). When it was assumed that the population survival for the last 10 years would best predict the future (most pessimistic model), the analysis predicted a probability of extinction of 6–19% in 100 years and 68–94% in 300 years. If a quasi-extinction threshold was used in lieu of actual demographic extinction, the predicted probability of meeting the threshold ranged from 39–67% in 100 years to 76–98% in 300 years (Figure 2).

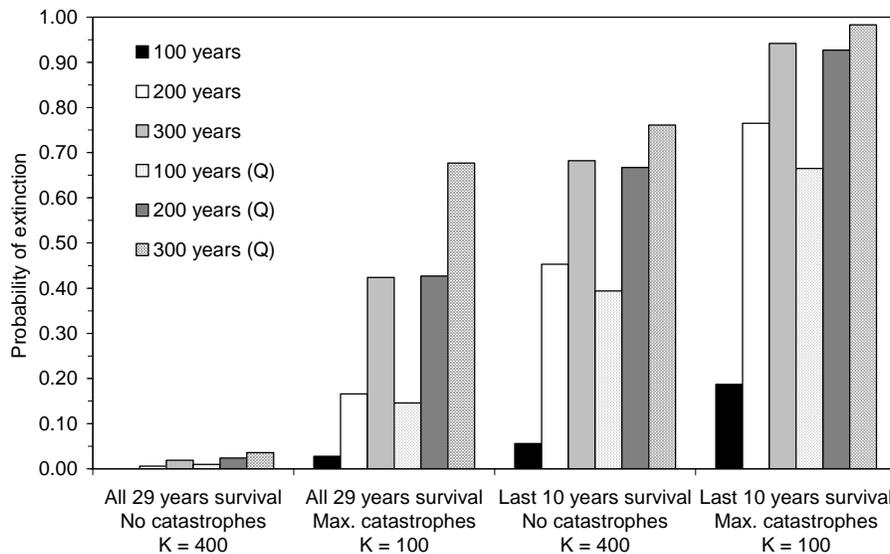


Figure 2. Estimated risk of extinction (<1 male or female) or quasi-extinction (Q; ≤ 10 males or females) at 100, 200 and 300 years for the Southern Resident population, assuming 1974–2003 (29-year) survival rates continue, with no catastrophes and with the maximum probability of catastrophe (two catastrophes every 100 years, with a 20% decline in the population following the catastrophes); or assuming survival rates for the last 10 years continue, with no catastrophes and with the maximum probability of catastrophe.

Life History and Population Dynamics of Resident Killer Whales (*Orcinus orca*) in British Columbia and Neighbouring Waters

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Annual photo-identification surveys conducted between 1973-75 and 2004 were used to estimate life history parameters and develop a population model for the northern resident population of killer whales that inhabits coastal waters of British Columbia. During the 1970's, 80's and early 90's, the population grew exponentially ($r^2=0.986$; $F_{1,22}=1,568.5$; $P<0.001$) at an annual rate of 2.6% (95% CI 2.48-2.76%) (Figure 1). Although the population almost doubled in size from about 125 to 217 animals, there was no evidence of a slowing of the growth rate ($F_{1,21}=0.25$; $P=0.622$), suggesting the population was unrestrained and increasing at its maximum intrinsic rate. The population peaked abruptly in the mid-1990s, declined by 7-9%, and then exhibited a small increase, resulting in no discernible trend over the last decade ($F_{1,10}=1.36$; $P=0.271$), indicating that something was restraining its growth. Life history and population parameters were thus estimated separately for 1973-96, a period of unrestrained growth; and 1996-2004, a period of no net change. During the period of unrestrained growth, females had a mean life expectancy of 46 years and maximum longevity was on the order of 80 years. Females typically gave birth to their first viable calf at 14.1 years of age ($SE=0.050$; range 10-21 years) and those that survived produced a total of 4.7 calves at mean intervals of 4.9 years ($SE=0.18$; range 2-11 years) over a reproductive lifespan typically lasting about 24 years. Older females exhibited reproductive senescence, with about 50% being post-reproductive by 38 years of age, and none reproducing after 46 years of age. Based on development of the dorsal fin – a secondary sexual characteristic – males typically attained sexual maturity at 13.0 years of age ($SE=0.046$; range 9-18 years) and the fin continued to develop for an average of 5.5 years ($SE=0.113$; range 3-7 years), such that males had typically attained physical maturity by 18.5 years of age. Males had a mean life expectancy of 31 years and maximum longevity was probably on the order of 60-70 years. Mortality curves were U-shaped for both sexes, indicating most mortality occurred early and late in life, but the right limb was steeper for males, resulting in a sex ratio that was progressively skewed toward females with increasing age (1:1 at age 15, 2:1 by age 34, and 3:1 by age 41 years). A sex- and age-structured model incorporating these parameters predicted that a population would increase at a rate of 2.4% per annum and be comprised of 46% juveniles, 22% reproductive females, 10% post-reproductive females, and 22% adult males. During 1973-96, the study population actually increased at 2.6% and was comprised, on average, of 46% juveniles, 21% reproductive females, 11% post-reproductive females and 22% adult males, indicating a good fit with the model predictions. Surprisingly, there were no major changes in reproductive parameters as the population stabilized during 1996-2004. Mean age at first birth increased slightly but significantly from 14.1 to 15.4 years ($t_{49}=3.23$; $P=0.002$), mean age of onset of post-reproductive senescence increased from 38.4 to 40.6 years ($t_{61}=2.84$; $P=0.006$), and

calving intervals were marginally longer (5.5 versus 4.9 years; $t_{97}=2.92$; $P=0.091$) (Figure 2). The overall effect was a slight drop in the estimated reproductive potential of females from 4.7 to 4.5 calves. The recent decline in productivity was due almost entirely to increases in mortality, which were evident and statistically significant ($2.61 < \chi^2 < 7.75$; $P < 0.05$) across all sex- and age-categories (Figure 3). Survival of viable calves to age 15 (about the age they are recruited to the adult population) dropped from 80% to 61%, and mean life expectancy declined from 46 to 30 years for females and from 31 to 19 years for males. Because the increase in mortality was broadly distributed across all sex- and age-classes, the predicted sex and age structure of the stable population remained almost unchanged at 47% juveniles, 24% reproductive females, 11% post-reproductive females, and 18% adult males.

The life history parameters for neighbouring resident killer whale populations in Washington and Alaska appear to fall within the range of our unrestrained and stable models for northern BC residents, suggesting the models represent the general population biology of the resident ecotype of killer whale. We believe such models provide a useful construct for exploring and developing a better understanding of the factors that may regulate or impact killer whale populations, and this will be illustrated for the southern resident population.

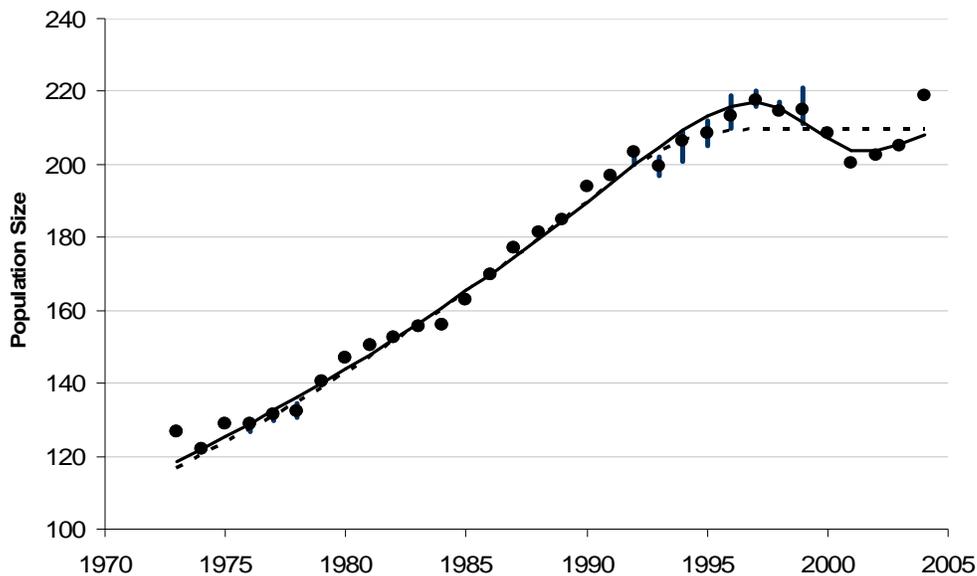


Figure 1. Trends in size of the northern resident killer whale population during the study period. The symbols represent the annual abundance estimates, and vertical bars the potential range in population size due to uncertainty in the exact year of births and deaths. The solid line represents a generalized logistic with a two-year lag, and the dashed line a generalized logistic with no lag.

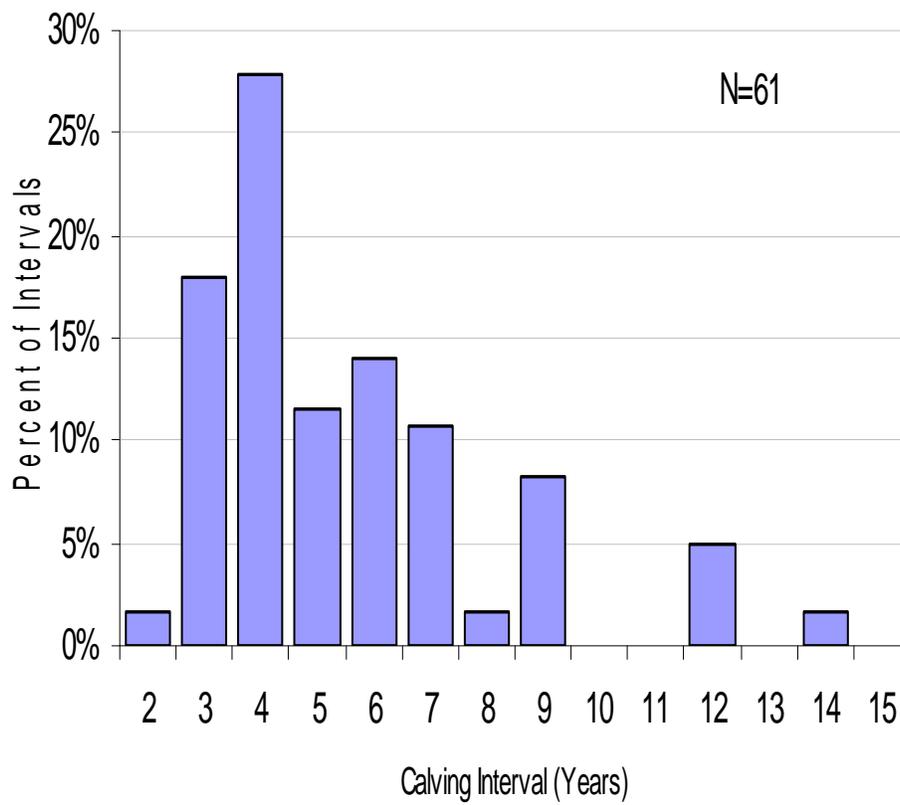
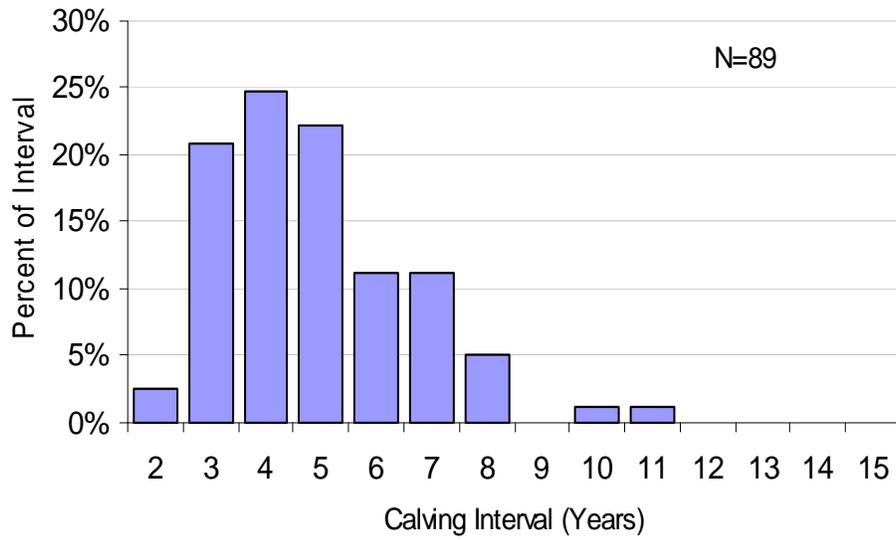


Figure 2. Observed calving intervals completed during 1983-95 (top panel) and 1996-2004.

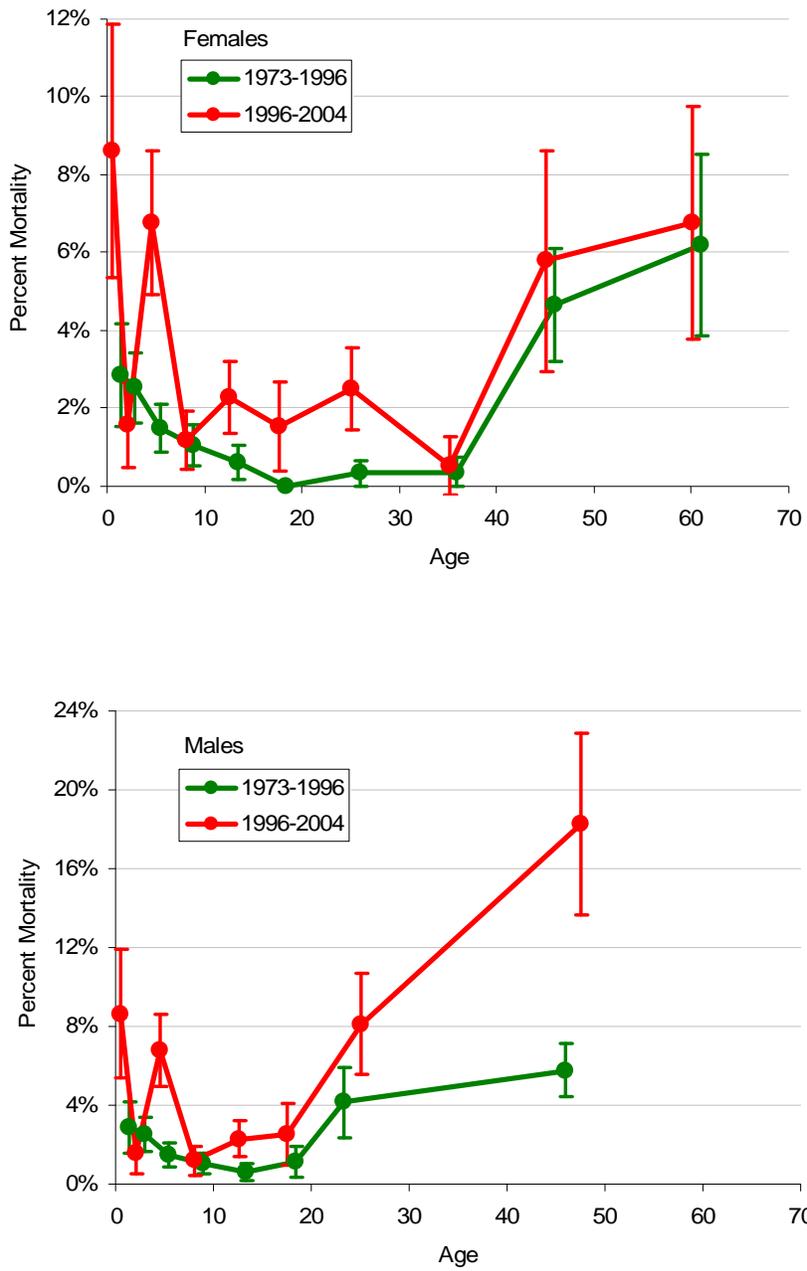


Figure 3. Age-specific annual mortality rates by age-category for females (top panel) and males (lower panel). The vertical bars represent standard errors for each estimate. Note that male mortality rates are plotted on a scale twice that of females.

Long-term Social Dynamics of Fish-eating Killer Whales

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Among marine mammals, odontocete cetaceans provide some extreme examples of highly stable matrilineal social structuring. The natal philopatry of both males and females exhibited by fish-eating killer whales in the eastern North Pacific is often cited as a unique example of such sociality in mammals. These whales have been characterized as comprising closed maternal lineages from which effective dispersal has never been documented. However, even such apparently stable social systems may experience changes over time in response to critical changes their ecosystem. The so-called southern resident killer whale (SRKW) population, consisting of less than 100 whales, has become the focus of much conservation and management concern in recent years. This population, typically described as consisting of three matrilineally-based social units ('pods'), has been photographically censused on an annual basis for nearly three decades, providing a unique opportunity to examine temporal changes in the social dynamics of this long-lived highly social cetacean. Here we use this longitudinal dataset to assess changes in the social structure of the SRKW population at the level of both the individual and the matriline. Using both coefficients of association and novel Bayesian clustering methods, we quantify the stability of social affiliations within this killer whale population based on data from more than 1360 encounters, and 118000 high quality individual killer whale identifications. Inter-annual comparisons of social patterns from these photographic data demonstrate dramatic changes in the social affiliations of these killer whales within the last decade, coinciding with the most recent period of population decline. Moreover, pod-specific changes suggest differences in the social stability within pods. Understanding these social dynamics may provide insight into the influence of social structure on population demographics, and help identify critical changes in key ecological forces driving such social changes.

Persistent organic pollutants in marine mammals inhabiting the transboundary waters of British Columbia - Washington

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The long-lived and high trophic level nature of many marine mammals renders them vulnerable to accumulating often very high concentrations of persistent chemicals, including pesticides, industrial by-products and flame retardants. In the case of killer whales (*Orcinus orca*), some of the older individuals currently frequenting the coastal waters of British Columbia (BC) and Washington State were born during the First World War, well before the advent of widespread chemical manufacture and use. Offspring of killer whales born today are exposed to thousands of chemicals that have been inadvertently or deliberately introduced into the environment over the past 60 years. The use of polychlorinated biphenyls (PCBs) as heat- and fire-resistant fluids for transformers and industrial machinery between 1929 and 1977 in North America resulted in widespread contamination of biota. Using biopsy samples, we have since found that the resident and transient killer whales in the NE Pacific Ocean are among the most PCB-contaminated marine mammals in the world. We have attributed this PCB contamination to a combination of 'global' (salmonid) and 'local' (non-salmonid) sources, whereby Puget Sound represents a regional hotspot. The more recent use of polybrominated diphenyl ethers (PBDEs) as flame retardants in consumer textiles, electronics and plastics represents a current concern for toxicologists and regulators. We have found moderate levels of PBDEs, as well as other classes of new generation fire retardants, in BC's killer whales. However, our temporal studies of the non-migratory harbour seal (*Phoca vitulina*) profile the rapid emergence of PBDEs as a major chemical concern in BC's coastal food web. Like resident killer whales, coastal grizzly bears feed heavily on returning salmon. While we have previously established that killer whales are highly contaminated with POPs, we have now found that salmon-eating 'maritime' bears are exposed to higher levels of many POPs than their 'interior' counterparts. We estimate that salmon provide up to 70% of the organochlorine pesticides, 90% of the polychlorinated biphenyls (PCBs), and 85% of the lighter polybrominated diphenyl ethers (PBDEs) found in maritime grizzly bears. Although living in fundamentally different environments, the killer whales and maritime grizzly bears of British Columbia are inextricably linked to the open Pacific Ocean by virtue of their shared 'culture of salmon'. The physical and chemical properties of PCBs and PBDEs that confer benefits upon commercial applications represent the very same properties that have caused widespread environmental problems: the chemicals are resistant to breaking down in their

intended use, as well as in the environment and in biota. In addition, many flame retardants resemble natural hormones and can lead to a disruption of endocrine processes in exposed individuals. We have found that several biological endpoints in free-ranging harbour seals inhabiting the transboundary waters of BC and Washington have been adversely affected by exposure to environmental contaminants. These effects include a contaminant-related alteration of i) circulatory vitamin A and thyroid hormone levels, ii) expression of thyroid hormone and Aryl hydrocarbon receptors, and iii) immune function. More research is needed to better characterize the nature of health risks to killer whales, although inter-species extrapolation provides a basis for concern. Given the global nature of contaminant dispersion, both national and international level efforts are required to regulate persistent organic pollutants, mitigate contaminated sites, and protect local killer whale habitat. The risk of contaminant-related adverse health effects presents an additional conservation concern to killer whales and other high trophic level species that may also be threatened by depleted salmon stocks, habitat degradation and climate change. The degree to which killer whales and other high trophic level marine mammals are exposed to persistent flame retardants, coupled with their vulnerability to adverse health effects, highlights the need for a 'weight of evidence' approach in research, conservation planning and regulatory processes. In this context, novel, minimally-invasive, alternative and surrogate approaches to conducting research on endangered southern resident killer whales must form a central research strategy in support of conservation and management efforts.

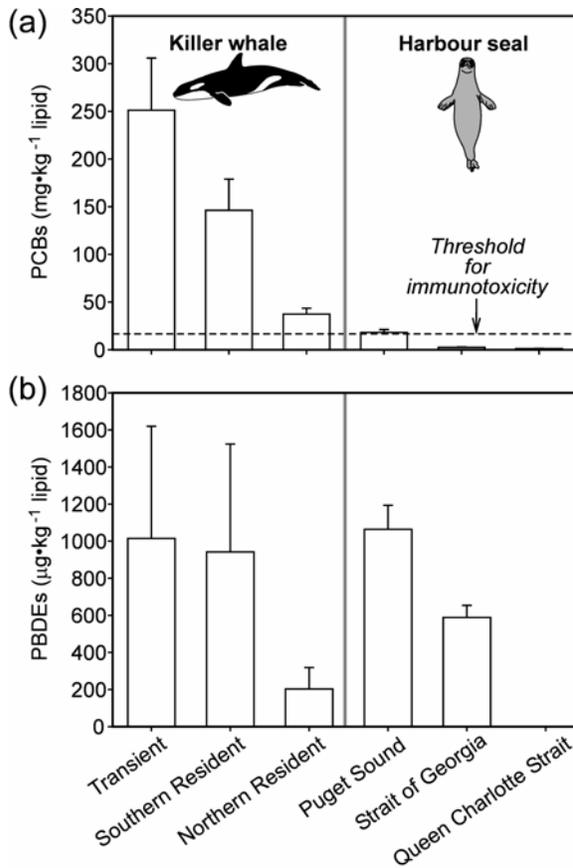


Figure 1: The southern resident killer whales that frequent the transboundary waters of British Columbia and Washington State are among the most PCB-contaminated marine mammals in the world, and are at risk for adverse health effects including reproductive impairment, developmental abnormalities and immunotoxicity (from Ross, 2006).

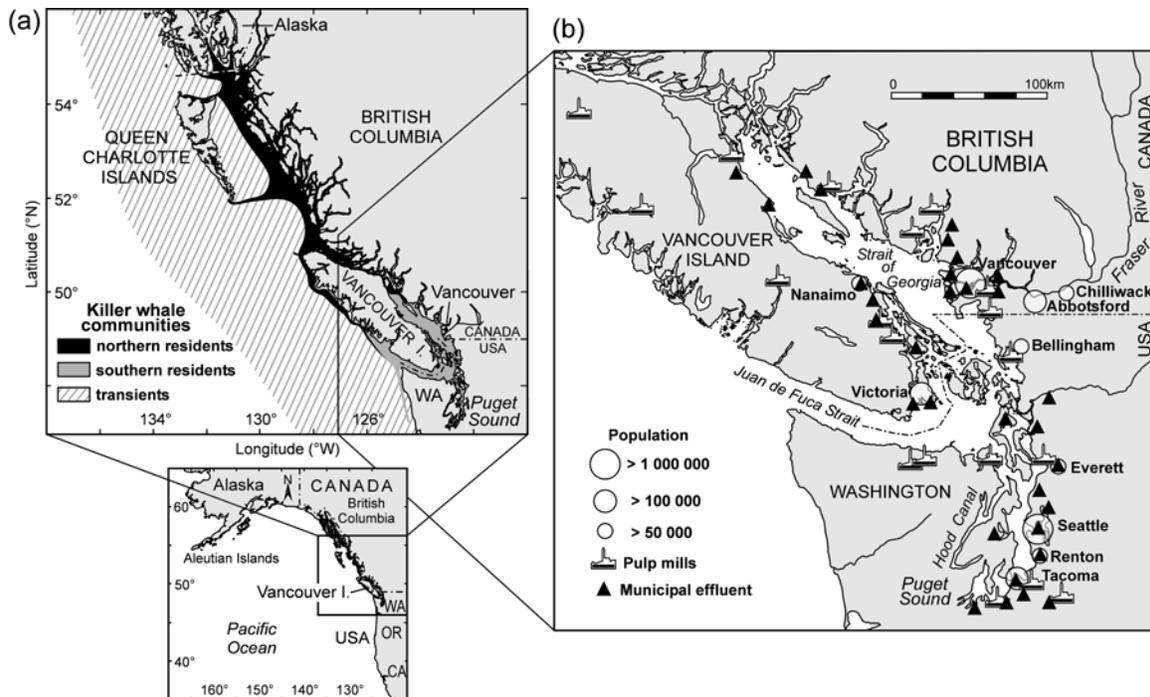


Figure 2: The transboundary coastal waters of southern British Columbia and northern Washington State represent critical feeding areas for the southern resident killer whales. This watershed is also home to over seven million human inhabitants. The combined effects of noise and disturbance, toxic chemicals, and reduced prey availability, represent important conservation concerns to these whales (from Ross, 2006).

Ross, P.S. 2006. Fireproof killer whales: Flame retardant chemicals and the conservation imperative in the charismatic icon of British Columbia. *Can. J. Fish. Aquat. Sci.* **63**: 224-234.

Regional patterns of persistent organic pollutants in five Pacific salmon species (*Oncorhynchus spp*) and their contributions to contaminant levels in northern and southern resident killer whales (*Orcinus orca*)

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Previous studies on killer whales (*Orcinus orca*) have shown that southern residents contain higher concentrations of persistent organic pollutants (POPs) than northern residents (Ross *et al.*, 2000; Rayne *et al.*, 2004) and other North Pacific resident killer whale populations (Ylitalo *et al.*, 2001; Herman *et al.*, 2005). Elevated contaminant exposure in southern residents may be attributed to dietary differences between the two whale populations or to regional differences in concentrations of POPs in their prey. Based on observational data and stomach contents analyses, Ford *et al.* (1998) identified Pacific salmon (*Oncorhynchus spp*), especially Chinook salmon (*O. tshawytscha*), as the primary prey of southern and northern resident killer whales in their summer feeding ranges. The main objective of this study was to determine if Pacific salmon had species-specific regional body burdens of contaminants that could differentially affect contaminant levels in northern and southern residents. An additional objective was to measure proximate composition (amount of protein, lipid and carbohydrate) and estimate caloric content of Pacific salmon as an indicator of species- and regionally-specific variation in nutritional quality of prey to killer whales.

Free-ranging populations of anadromous Pacific salmon are generally assumed to have low levels of POPs because the majority of their growth (and therefore contaminant uptake) occurs in open water of the Pacific Ocean. However, the oceanic distributions of the five Pacific salmon species (*i.e.*, Chinook, coho, chum, pink, sockeye) differ and, as a result, can influence their exposure to contaminants. For example, when pink, chum and sockeye salmon enter the marine environment, they rapidly migrate northward and westward through coastal waters of North America and are found in the open waters of the North Pacific, Gulf of Alaska and Bering Sea by the end of their first year at sea (Quinn, 2005). In contrast, Chinook and coho, have a more coastal marine distribution along the continental shelf compared to other salmon populations, although, populations within these two species can also differ in their marine distribution (Quinn, 2005).

To determine if Pacific salmon populations present in the summer feeding areas of northern and southern resident whales had species- and regionally-specific body burdens of contaminants, we analyzed POPs in whole body samples of five species of Pacific salmon collected from northern British Columbia to the central California coast. All five species were each sampled in two regions, the non-urbanized north-central coast of British Columbia that is frequented by northern resident killer whales and in the urbanized Puget Sound - Strait of Juan de Fuca region that is frequented more by

southern resident killer whales (Table 1). Additionally, Chinook salmon were sampled in the coastal waters on the inside of Vancouver (near Robson Bight), the Lower Columbia River (spring and fall runs) and central coast of California (Table 1). We sampled terminal marine and in-river fisheries, at times aligned with the return time for specific salmon stocks, to increase the likelihood of sampling particular stocks of fish returning to a specific river. The presumed river-stock associated with each sampling location is noted in Table 1.

Table 1. Number of individual fish of each species collected at each sampling location for chemical, proximate and stable isotope analyses. Contaminants samples were based on whole body samples of individual Chinook salmon and composite samples (5-6 individuals) for other salmon species. Proximate analyses were based on composite samples of males or females, each with 2-3 fish per composite. Note: Only 13 of the Fraser and 20 of the Columbia River Fall Chinook were analyzed for contaminants but proximate analyses were based on all fish collected.

Region	Sampling Location	River Stock Origin	Species						
			Chinook			Sockeye	Pink	Chum	Coho
			Fall	Spring	Resident				
North/ central coast BC	Chatam Sound	Skeena	30			29	30		
	Dean Channel	Kimsquit					30	30	
East coast Vancouver Island	near Robson Bight	Fraser/Nimkish	30						
Strait of Juan de Fuca/ Puget Sound	Lopez Sound	Fraser				30			
	Skagit River	Skagit					30		
	near Apple Cove Point	Puget Sound mixed			34		30	11	
	Nooksack/ Duwamish/ Nisqually rivers	Puget Sound mixed	36					29	
Columbia River	Lower Columbia River	Columbia	30	20					
Central coast of California	near Point Arena	Sacramento/ Joaquin	29						

Overall, concentrations of POPs were higher in coho and Chinook populations that have more coastal distributions than those measured in salmon species (e.g., chum, pink, sockeye) with more oceanic distributions. For pink, chum, and sockeye salmon, PCB levels were higher for fish caught in the more urbanized region of Puget Sound/Strait of Juan de Fuca region than in the less urbanized north/central coast of British Columbia

area, whereas concentrations of DDTs were similar between regions. PBDEs were below the limit of quantitation in all pink, chum and sockeye sampled in this study. Concentrations of PCBs, PBDEs and DDTs were higher in coho salmon from Puget Sound compared to coho from the central BC coast. Regional variation in POP exposure was also evident in Chinook salmon (Figure 1) and appears to be associated with differences in marine distribution of these species. For example, Chinook salmon returning to Puget Sound had significantly higher concentrations of PCBs and PBDEs

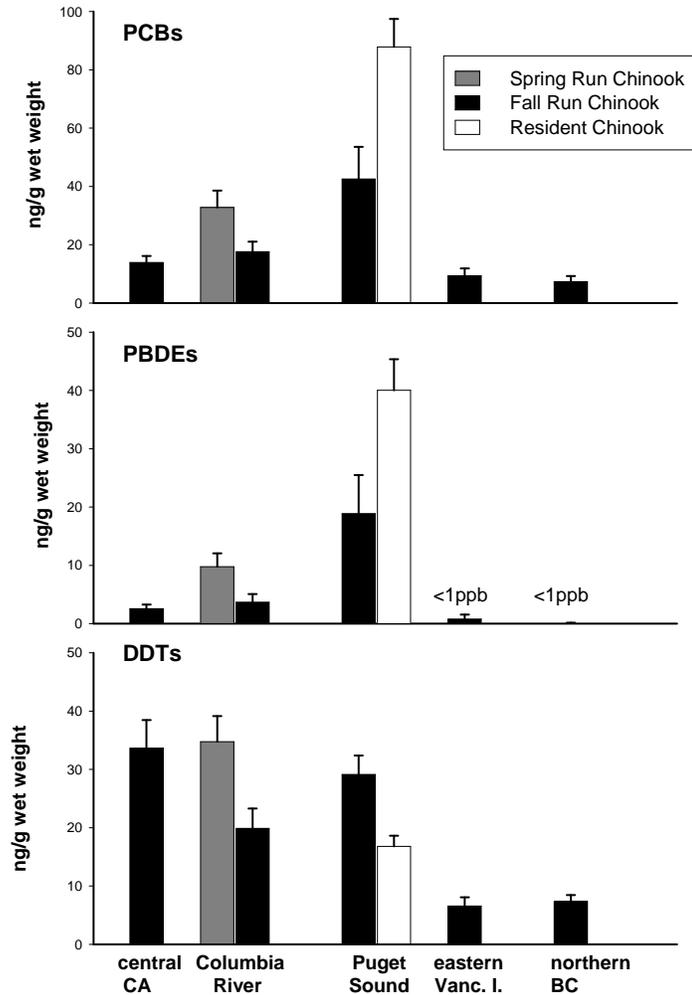


Figure 1. Concentrations of persistent organic pollutants (\pm 95% CI) in whole body samples of individual Chinook salmon caught in terminal fishing areas. River populations represented include fish returning to the Sacramento/San Joaquin (central CA; n = 29), the Columbia River (spring run n = 20; fall run n = 17), the Nooksack, Duwamish and Nisqually (Puget Sound; n = 36), the Fraser and Nimpkish (eastern coast Vancouver Island; n = 13; Georgia Basin) and the Skeena rivers (northern BC; n = 30). Additionally data are shown for sub-adult chinook salmon that were resident in Puget Sound in the winter months (termed “resident chinook” n = 44).

compared to other Pacific coast salmon populations we sampled. Furthermore, Chinook salmon that resided in Puget Sound in the winter rather than migrate to the Pacific Ocean

(“residents”) had the highest concentrations of POPs, followed by Puget Sound fish populations believed to be more ocean-reared. Fall Chinook from Puget Sound have a more localized marine distribution in Puget Sound and the Georgia Basin than other populations of Chinook from the west coast of North America and are more contaminated with PCBs (2 to 6 times) and PBDEs (5 to 17 times).

Overall, the total caloric content per kg of fish was higher for Chinook and sockeye salmon than the other salmon species we sampled, due to their higher fat content. Moreover, because of their greater size, the total Kcal per fish was highest for Chinook salmon. Regional differences in caloric content among Chinook populations were also observed. Puget Sound Chinook had lower caloric content (total Kcals per fish) than summer/fall run of Chinook salmon from all regions we sampled (Figure 2). Assuming that the fish we sampled were representative of the sizes of fish available for killer whale consumption, whales feeding on non-resident Chinook salmon in Puget Sound would need to eat 1.5 to 1.8 times as many Chinook salmon as animals feeding outside Puget Sound to obtain the same caloric content.

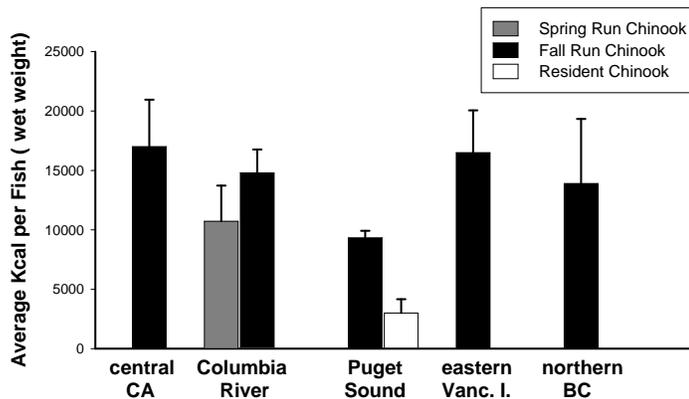


Figure 2. Average estimated caloric content (\pm 95% CI) of whole body samples of individual Chinook salmon caught in terminal fishing areas. River populations represented include fish returning to the Sacramento/San Joaquin (central CA), the Columbia River (spring and fall runs), the Nooksack, Duwamish and Nisqually (Puget Sound), the Fraser and Nimpkish (eastern coast Vancouver Island) and the Skeena rivers (northern BC).

In summary, regional body burdens of contaminants in Pacific salmon, and Chinook salmon in particular, could contribute to the higher levels of contaminants in southern resident killer whales. In addition to contamination, regional differences in caloric content of Chinook salmon from Puget Sound further reduce their quality as prey to southern resident killer whales.

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Selective foraging in resident killer whales

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A previous study of the diet of the resident and transient ecotypes using opportunistic collection of prey remains from kill sites as a primary measure of prey selection found that resident killer whales feed predominantly on salmonids, particularly on chinook salmon (*Oncorhynchus tshawytscha*). To address uncertainties concerning potential biases in the prey fragment sampling technique and questions regarding seasonal and geographic variability in diet, we conducted dedicated field studies of foraging behaviour in resident killer whales during 1997-2005. These studies used focal-animal and focal-group observations as well as underwater video monitoring to document details of prey capture, handling and consumption. Foraging by resident killer whales often involves cooperation among kin-related group members, and prey items are frequently brought to the surface and shared by two or more whales. Adult males share prey less often than do females and juveniles. Prey sharing does not appear to be related to prey size. All common prey species and all age (i.e., size) categories of these species were shared. Prey fragments left at kill sites result mostly from prey handling and sharing, and appear to be reliable indicators of selection for different salmonid species by resident killer whales. Chinook is the predominant prey species taken by both northern and southern resident communities during May-August, but chum salmon (*O. keta*) is more prevalent in September-October, at least in northern residents (Figure 1). Coho salmon (*O. kisutch*) are taken in low numbers in June-October, but sockeye (*O. nerka*) and pink (*O. gorbuscha*) salmon are not significant prey species despite their high seasonal abundance. No significant differences were observed in the diets of different pods or clans. Non-salmonid fishes do not appear to represent an important component of resident whale diet during May-October. Their strong preference for chinook salmon may influence the year-round distribution patterns of resident killer whales in coastal British Columbia and adjacent waters.

This report is available on the Canadian Science Advisory Secretariat website at:
http://www.dfo-mpo.gc.ca/csas/Csas/Publications/ResDocs-DocRech/2005/2005_041_e.htm

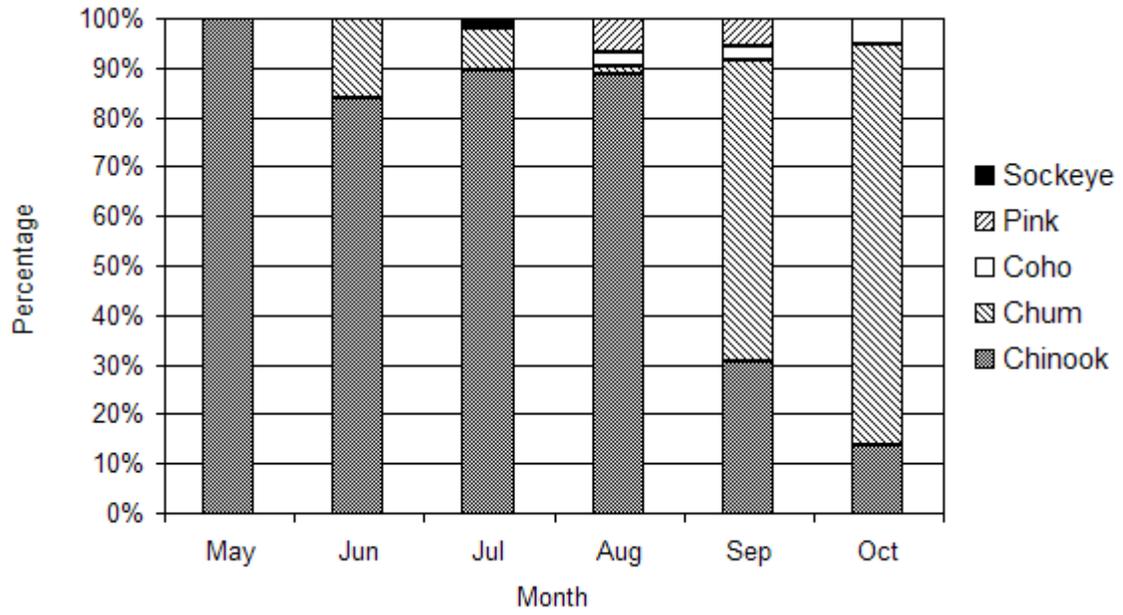


Figure 1. Frequency distribution of salmonid species in resident killer whale feeding events during May-October ($n = 396$ kills).

Movement Patterns and Population Trends of Southern Resident Killer Whales in Relation to Relative Abundance and Distribution of Pacific Salmon in the Puget Sound-Georgia Basin.

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Introduction

This research project was divided into two major components: spatial analysis of whale movement patterns across time and space, and population trend analysis of whales and Pacific salmon at various spatial scales and lag periods. Investigations into the spatial distribution and characterization of the spatial structure of an animal population has become increasingly important for ecologists to understand how a population interacts with the environment (Monestiez et al. 2006). The use of space is vital to the study of populations and individuals at many levels and for many disciplines, including population biology, conservation biology, and behavioral ecology (Whitehead 2001). For questions concerning the relationship between predators and their prey, it is crucial to take into consideration the spatial and temporal patterns of both predator and prey. The spatial dynamics and social structure of most medium to large-sized carnivores are most significantly influenced by the distribution and abundance of preferred prey (Patterson and Messier 2001).

This project links the spatially explicit movement behavior of the southern resident killer whale community (SRC) with spatially distributed estimates of salmon abundance, with the intention of investigating relationships between the SRC and the various salmon species. Research questions include: 1) Does whale space use and movement shape complexity differ between periods of population increase and decrease? 2) Do any changes in space use patterns coincide with changes in salmon catch? 3) Do whales occur in fishery management areas more often than by chance? 4) Are there significant correlations between the whales and salmon and if so, are they pod, species, or region specific?

Methods and Results

The project addresses issues of representation, characterization, and modeling of individual pod movement (J, K, and L) by statistical week (week 1 begins on the first Monday of the year and proceeds through December 31st, resulting in 53 statistical weeks per year) from 1991 through 2001, and reports the results of a series of spatial pattern analysis approaches used to investigate variations in shape and size of pod movement behavior between and within salmon management areas. All whale sightings recorded during 1991-2001 in the Puget Sound-Georgia Basin were used in this analysis. The movement behavior of each pod of the SRs is modeled as “space use polygons” by statistical week. The shape of the “space used” is then spatially intersected with salmon management areas which contain attributes of weighted fish catch for each statistical week (Figure 1). In this way, quantitative pattern metrics of use areas as a function of an external parameter, in this case, fish density, are derived. Two pattern metrics were calculated to address questions of whale movement. 1) Total area (TA), which quantified space used by a pod in a given week (measured in hectares) and 2) Landscape Shape

Index (LSI), which is a measure of shape complexity. Increasing LSI values imply more convoluted shape patterns.

It was hypothesized that whales would cover more area and their shape patterns would be more convoluted during periods of lower prey availability. Increased space use and movement complexity would indicate searching behavior. In contrast, whales would remain in a smaller area and their shape patterns would be less convoluted during periods of higher prey availability. Mean annual values of TA and LSI from the early 1990's, when the SRC was generally increasing in population, were compared to mean annual TA and LSI values from the late 1990's, when the SRC was generally decreasing in population. The differences in mean values of both TA and LSI were significantly different between the early 1990's (1991-1995) and the latter 1990's (1996-2001) for the entire SR population as well as each individual pod (Figures 2 and 3). Salmon catch per unit effort (CPUE) data for each species will be compared between these time periods. Generally, catch was significantly lower in the latter 1990's than in the early 1990's. This appears to reflect both a decrease in salmon abundance (reflected by decreased CPUE) and an increase in regulations (reflected by a total lack of fishing effort in some management areas beginning in 1998).

Intra-annual differences in TA and LSI values were also compared. It was hypothesized that during the late spring and early summer whales would exhibit higher space use and more complex movement patterns than in the late summer and fall due to the presence of more abundant and concentrated salmon runs in the Georgia Basin- Puget Sound region in the second half of the year. However, the mean TA used by J pod from week 1 up to week 30 was statistically not different from the mean TA used from week 30 to the end of the year, with the exception of the years 1996 and 2001. K pod showed significant intra-annual differences in TA in years 2000 and 2001. Similar to J pod, L pod showed significant differences in TA values between the first and second part of the year in 1996 and 2001. Only two years showed significant differences between the mean LSI up to week 30 and the mean LSI from week 30 to the end of the year for J pod: 1995 and 1996. Results showed that the mean LSI values for K pod were statistically different between the spring- early summer and late summer- early fall in years 2000 and 2001. For L pod, the years 1996 and 2001 had statistically different LSI values in the weeks prior to week 30 compared to the period from week 30 to the end of the year. It should be noted that sightings of the whales are extremely limited in the winter and spring months. Results would be significantly more robust if more sightings were available.

A chi-square test was used to investigate whale presence in salmon management areas reporting the highest density of salmon catch (regardless of species) on a weekly basis. Whale presence in these areas was not statistically different than would be expected based on random distribution, with the exception of years 1994 and 2000. This test may be performed on combined salmon catch data (all species combined) or individual species treated separately. An example of the latter type of test might be identification of the area of highest catch for Chinook salmon for each week. Analysis of salmon catch and time spent by each pod in management areas will also be completed.

The second part of this study tested the relationship between the SRs at the population and pod levels to different species of salmon at various spatial scales and temporal lag periods. Lag periods of 0-5 years were used to test correlations between fluctuating salmon availability and demographic trends of SR whales. Lag periods of up

to five years were chosen in consideration of the approximately 17 month gestation period, 1.5 year lactation period (Baird 2001), and the necessary relative fitness of an adult female prior to conception.

Salmon escapement data from all regions (see figure 4) were totaled and tested against total SR population and for each pod using Spearman's rank correlation test. Individual pod data were also tested against each species of salmon for each region where data were available. When totaled salmon by species was tested against total SR population, few significant correlations were found. Correlations that did exist at the total population scale were between the SRs and normal chum, early chum, and sockeye. Contrary to expectation based on observational and scale sampling evidence of chinook predation, no significant correlations between total chinook and total SRs were found.

When the scale of analysis was decreased to pod, species, and area specificity, different correlation patterns emerged. Pods were tested against totaled salmon species and then tested against the different salmon species in each of the regions. When salmon was totaled across all regions, J pod showed significant relationships with chinook, early and normal chum, coho, and sockeye. K pod was correlated to the same species, as well as pink salmon. L pod did not show highly significant correlations with any of the salmon species. When salmon were divided by natal region, pod specific correlation patterns emerged. Overall, J and K pod showed similar correlation patterns to salmon of various regions. The biggest difference among the pod specific correlations was the absence of significant correlations between L pod and chum salmon in central and southern Puget Sound while both J and K pods showed significant correlation across all time lags tested with Puget Sound chum. Total run size data will also be tested against whale population trends and the results will be compared.

Discussion

This study was the first to combine disparate observational datasets from single point and continuous time observations into a single dataset of space utilization. It represents a pioneering attempt to spatially analyze whale movement patterns in relation to varying salmon abundance. There are extremely limited data regarding prey preferences of the SRs and an immediate need to protect resources most crucial to the survival of the population. Any relationships that can be shown, even on a coarse scale, to be significant could have far reaching implications in the management of both the SRs and the salmon runs on which they appear to depend. Preliminary results from this study indicate that the whales cover more area and move in a more erratic pattern during years of lower salmon catch and during years when the whale population was declining. It is likely that the whales were food limited during the latter 1990's and were expending more energy in search of less available prey. To further investigate this pattern, and to compare years of more consistent sighting effort, modeling observational whale sighting data from 2002 to the present, when the whales have once again been increasing in numbers, would be an important next step in understanding whale movement behavior.

Given predator responses in other systems to variations in prey abundance or availability, and the SRCs perceived preference for salmonid prey, testing correlations between salmon and whale population trends seems a reasonable and necessary step towards understanding killer whale foraging ecology and population dynamics. Correlations among specific pods, prey species, or regions would be particularly

beneficial in aiding management plans aimed at bolstering the SRs or particular salmon run populations. Preliminary results indicate that chinook spawning in regions 10, 40 and 50, early chum spawning in region 50, normal chum spawning in regions 40, 50 and 60, coho spawning in region 20, and Fraser river sockeye should be considered conservation priorities as important prey for the SRC. Because L pod exhibited fewer correlations with salmon than J and K pods did, and because L pod has fluctuated in numbers more dramatically than the other two pods, more effort should be focused on the prey preferences of L pod whales.



Figure 1. A visual example of modeling one statistical week of observational whale data taken from point observations to intersected movement shape polygons.

Total Area Used

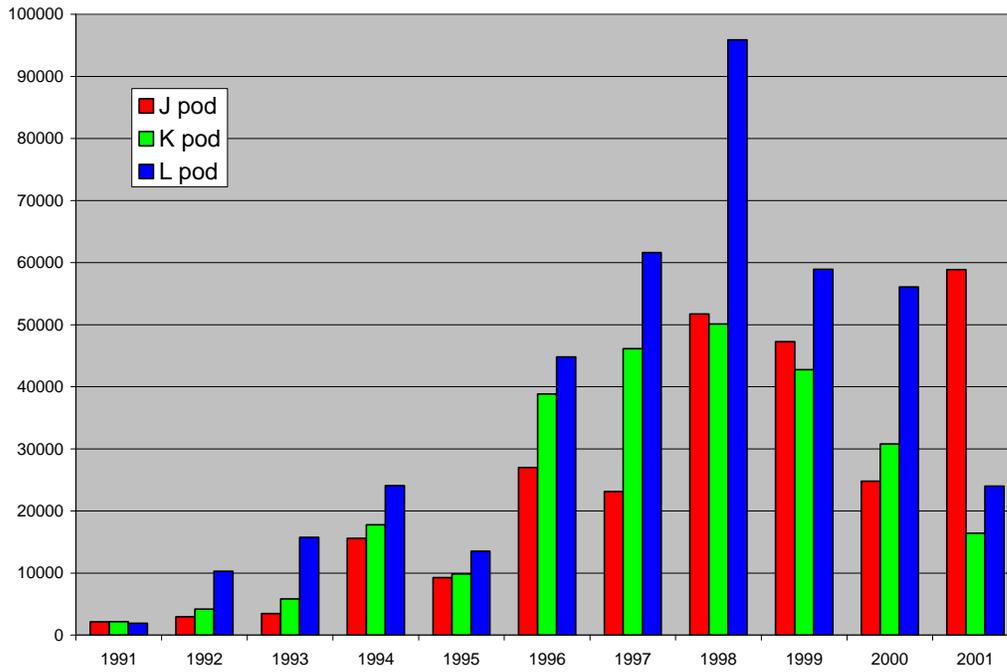


Figure 2. Total average annual space use by pod from 1991-2001.

Landscape Shape Index

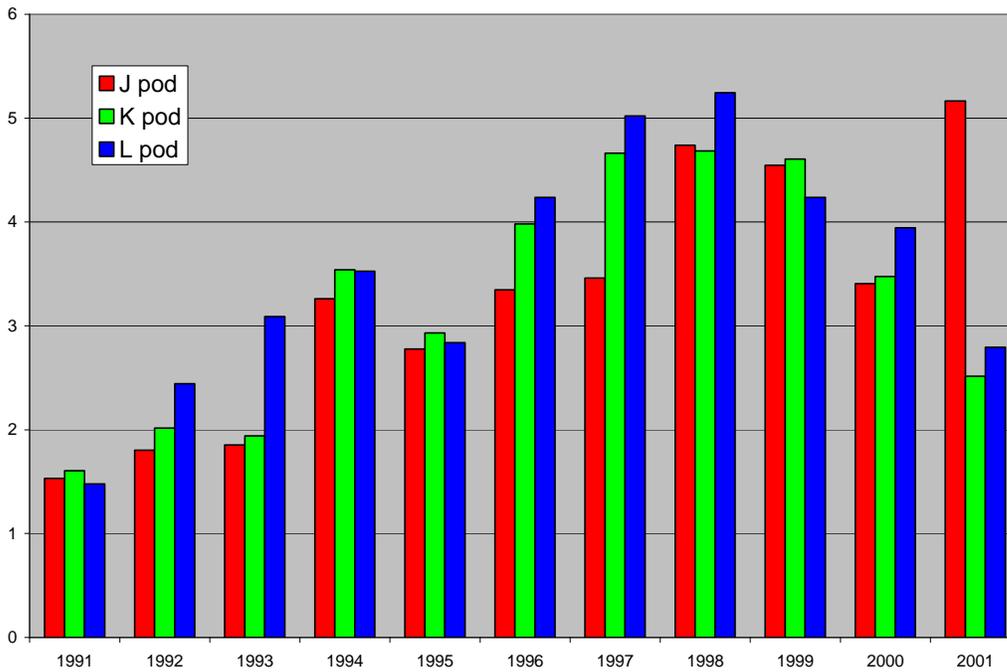


Figure 3. Annual average landscape shape metric by pod from 1991-2001.

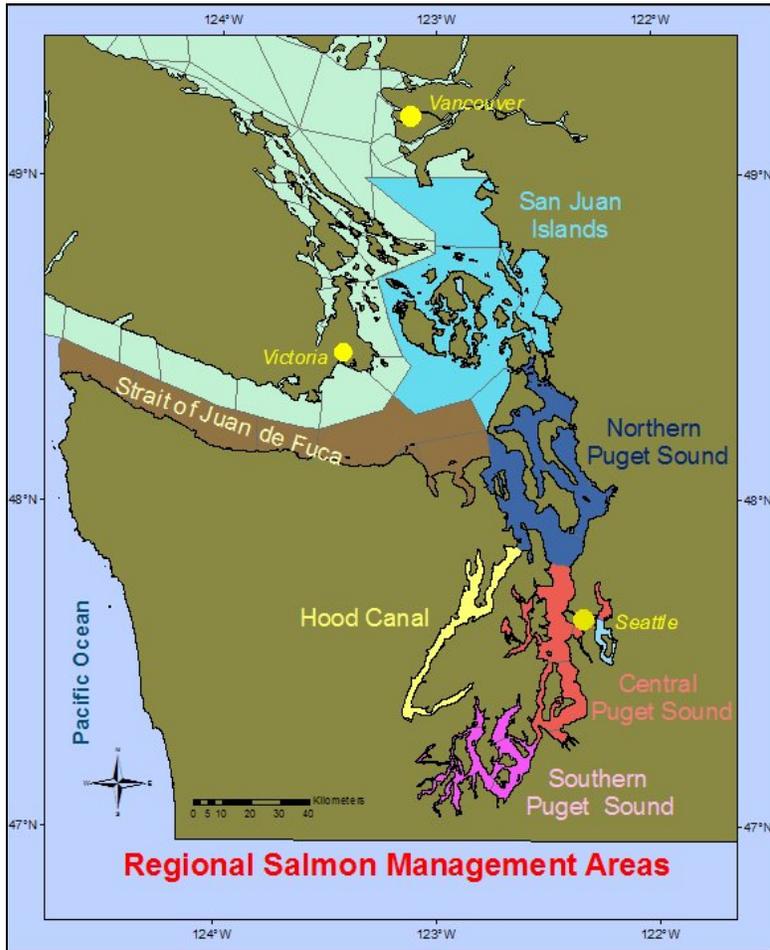


Figure 4. Regions of escapement grouping. Strait of Juan de Fuca (10), San Juan Islands (20), Northern Puget Sound (30), Central Puget Sound (40), Southern Puget Sound (50), and Hood Canal (60). Note: Sockeye escapement to the Fraser River is grouped in region 20.

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Feeding ecology of eastern North Pacific killer whales *Orcinus orca* from fatty acid, stable isotope, and organochlorine analyses of blubber biopsies

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ABSTRACT:

Blubber biopsy samples from eastern North Pacific killer whales *Orcinus orca* were analyzed for fatty acids, carbon and nitrogen stable isotopes and organochlorine contaminants. Fatty acid profiles were sufficiently distinct among the 3 reported ecotypes ('resident,' 'transient' or 'offshore') to enable individual animals to be correctly classified by ecotype and also by mitochondrial DNA (mtDNA) haplotype. Profiles of PCBs also enabled unambiguous classification of all 3 killer whale ecotypes, but stable isotope values lacked sufficient resolution. Fatty acid, stable isotope and PCB profiles of the resident and transient ecotypes were consistent with those expected for these whales based on their reported dietary preferences (fish for resident whales, marine mammals for transients). In addition, these ecotype profiles exhibited broad similarity across geographical regions, suggesting that the dietary specialization reported for resident and transient whales in the well-studied eastern North Pacific populations also extends to the less-studied killer whales in the western Gulf of Alaska and Aleutian Islands. Killer whales of the same ecotype were also grouped by region of sample collection. The mean stable isotope ratios of various regional groups differed considerably, suggesting that the prey preferences of these North Pacific killer whales may be both region and ecotype specific. Furthermore, 3 specific ecotypes of killer whales were found to have measured stable isotope values that were consistent with dietary preference reported in the literature. Finally, although the offshore population had blubber fatty acid profiles implicating fish as its primary prey, contaminant and stable isotope results were equally congruent with predation on marine mammals.

Herman, D.P., D.G. Burrows, P.R. Wade, J.W. Durban, C.O. Matkin, R.G. LeDuc, L.G. Barrett-Lennard, and M.M. Krahn. 2005. Feeding ecology of eastern North Pacific killer whales *Orcinus orca* from fatty acid, stable isotope, and organochlorine analyses of blubber biopsies. *Marine Ecology Progress Series*, 302: 275-291.

DIET STUDIES OF “SOUTHERN RESIDENT” KILLER WHALES IN THEIR SUMMER AND FALL RANGE: PREY SAMPLING AND BEHAVIORAL CUES OF PREDATION

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“Southern resident” killer whales are found primarily in the inshore waters around southern Vancouver Island and in northwestern Washington in the summer and fall months. The diet of individuals in this population is incompletely understood. Unlike the sympatric population of so-called “transient” killer whales (see Baird and Dill 1995), documented observations of predatory interactions with other species of marine mammals have been extremely rare (Ford et al. 1998). Based on the lack of such observations, combined with occasional anecdotal observations of predation on fish and the spatial and temporal co-occurrence of whales and pre-spawning runs of salmon, it is widely accepted that this population feeds primarily, if not entirely, on fish, at least in summer and fall. However, relatively little is known about which species of fish are taken and how this varies seasonally or between pods. The only published studies of diet, involving analyses of available stomach contents and collection of fish scales from behind foraging whales, concluded that salmon, and primarily chinook salmon, form the vast majority of the diet (Ford et al. 1998, 2005). Sample sizes for those studies were relatively small (samples from 68 predation events and 5 stomach contents from “southern residents”), given that these were collected over a 30 year time span, and information on the details of predation on fish are limited. Hoelzel (1993) noted that “a series of fast turns and rolls at the surface [were] always seen when an interaction with fish prey was apparent”. In their scale sampling study, Ford et al. (1998) used cues “such as rapid acceleration, sudden direction changes, or circling” as indicators of potential predation.

The primary purpose of this study was to gain additional information on the diet of this population through the collection of prey remains from foraging whales, as well as examine the behavioral cues associated with predation to assess potential biases associated with the prey sampling methodology.

Field activities were based out of Friday Harbor, WA, and were undertaken during short periods each month from June to September 2005 and during August/September 2004, using a 6-m rigid-hulled inflatable boat. In October sampling was conducted opportunistically when the whales entered Puget Sound. Killer whales were found primarily based on sightings reported by other researchers, commercial whale watching operators, or The Whale Museum Soundwatch Program. Information recorded for each encounter included location, pod(s) present, the number of boats with whales, and the approximate area covered by the group of whales (an indicator of how spread out the group was). If whales were not approached closely (see below), information on pod identity was provided by other vessels on site (e.g., from the Soundwatch Program or other researchers); for groups that were approached closely we used the catalog of van Ginneken

et al. (2004) to identify individuals, and digital photographs (using a Canon SLR with 100-300 mm zoom lens) were taken to confirm identity of difficult-to-identify whales.

To minimize public relations conflicts, whales were not approached closely if they were nearshore (< 500 m) in areas of high human habitation, or if more than a couple whale watching boats were with the whales and the whales were in a tight group (e.g., if the whales were in an area of 0.25 km² or less). If whales were not close to shore or in a tight group in close proximity to whale watching vessels, collection of samples was undertaken in two different circumstances, during “follows” of one or more focal whales, or opportunistically, if the number of whales present within several hundred meters of the vessel was too great to keep track of individual whales. Given our attempts to minimize public relations conflicts, we did not attempt to distribute effort randomly or evenly between different pods or individuals, but chose whales to follow based primarily on distance from commercial or recreational whale watching vessels. During focal follows, information on speed (slow, medium, fast), directionality (directional/non-directional), spacing between individuals (tight, loose, spread), and orientation (linear, flank, non-linear) were recorded every ten minutes. Identity of individual whales in the focal group were recorded when possible, using the catalog of van Ginneken et al. (2004). The size of the focal group was recorded as it changed, with an operational definition of the “group” being those whales within an approximately 300 m radius around the research boat that could be continuously observed and potentially approached if cues of predation were observed. The radial distance for including individuals in the “group” decreased with increasing glare or deteriorating sea conditions. Distance between the boat and whales during observations varied depending on the number of whales being observed and their configuration relative to each other, and ranged from approximately 10 to 300 m.

Several cues were used to trigger close approaches to look for fish parts in the “fluke prints” (glassy areas of water caused by upwelling from the whales’ tail as it dives). Cues included both high energy and low energy state activities. High energy activities included fast directional surfacings (FDS) that were out of context (i.e., not part of a series of fast directional surfacings characteristic of fast travel), fast and moderate non-directional surfacings, and (for pairs of whales) a sudden decrease in one whale’s speed associated with a long-dive by a second whale in a pair. Non-directional surfacings were defined as surfacings where the whale’s trajectory changed part-way through the surfacing. Fast non-directional surfacings (FNDS) involved generation of white water when the whale surfaced. Chases involved a prolonged series of FNDSs and prolonged high-speed surface swimming (with part of the body visible).

Low energy cues included moderate non-directional surfacings (MNDS, which did not involve the generation of white water when the whale surfaced, yet were at higher swim speeds than a typical surfacing), direction change of a whale in the group (toward the focal whale), and surfacing after a long dive. In addition, other subtle cues included a fish observed in the whale’s mouth or a pause at the surface by an associated whale.

Upon observing one or more of these cues we would note the time, approach the fluke print of the target whale, and record the time of our arrival to the fluke print (to the second). We would also note the presence of other whales nearby and record information on the context (e.g., social interactions with the target whale). Approaches were made in a way to avoid or minimize disturbance to the whale(s) present, by slowing the vessel speed either to a stop in the fluke print (if the whale(s) was still actively milling), or matching the speed of the whale(s) upon arrival at the fluke print. Reactions of whales to follows and close approaches were recorded. Once at the fluke print, we recorded whether fish, fish scales, or fish parts were observed, and if seen, estimated the number of prey parts visible in the water column. When no prey parts were

observed in the first fluke print we would proceed to subsequent fluke prints to search for prey parts. During all surfacings when in close proximity to whales (i.e., less than 20 m) one observer on the boat would attempt to watch the mouth-line of the whales as they surfaced to try to assess whether the whales were carrying prey. A long-handled fine-mesh net was used for collecting parts observed in the water, which were stored in ziplock bags in a cooler on ice packs while in the field. Prey samples (comprised of one or more prey parts) were later frozen for analyses. To confirm the identity of difficult to identify individuals, we attempted to obtain photographs of the dorsal fin of whales from which prey remains were collected.

We spent time on the water in presence of whales on 25 days and logged 170.25 hours of effort during 5 sampling periods in 2004 and 2005 (Table 1). Follows and approaches were typically undertaken in the late afternoon and evening hours due to the large number of boats with the whales in the morning through mid-day. Cues prompting close approaches occurred on 136 different occasions, including 37 associated with high energy behaviors and 99 with lower energy behaviors. Prey or prey parts were collected in 18 (49 %) and 31 (31%) of these cases. Of the samples we collected, 49 individual fish were identified. The majority (75%) were chinook, 18% were chum (mostly from October), and 6% were coho.

Of significance was the collection of a substantial number of samples during more subtle low energy activities. There were numerous cases where from a distance the behaviors could have been misinterpreted as social activity. In particular, the observation of a change in direction by a whale in the group and converging with the focal whale as well as the occurrence of fish parts in several consecutive fluke prints suggests that prey manipulation and sharing might be occurring on a regular basis. Prey sharing would help explain the preferential selection of the relatively large chinook and chum and why they are brought to the surface. The large size of these prey would be more likely to allow sharing, and by bringing them to the surface they can be manipulated and shared while breathing following a long dive. Another reason some fish are likely handled near the surface is that in some cases we observed fish being chased just below the surface of the water. The surface of the water likely acts as a highly effective barrier in open water to “corner” a fish.

Systematic focal animal behavioral observations provide a variety of information that will be useful for management needs. Obtaining a representative sample across seasons is an important first step in establishing current baseline prey selection against which to evaluate future trends. Determining which cues represent foraging behavior will likely allow foraging rate determination – potentially a reflection of prey availability. Locations of predation events will also contribute to critical habitat delineation. Identification of prey to species, and ideally to stock, will provide important information on how to better manage prey resources.

Focal behavior follows also lends itself to the collection of fecal material from known animals. Fecal samples collected as part of this study will potentially provide additional information on prey selection (through genetics) as well as contribute tissue for whale genetics studies and material suitable for assessment of health parameters.

Table 1. Southern resident killer whale predation event sampling effort, predations cues, and prey samples collected.

Effort by month				
Dates	# days of effort	# hours on effort	# of cues	# of prey samples collected
Jun 2005	6	43.1	17	2
Jul 2005	5	34.6	22	12
Aug 2005	5	37.6	52	18
Aug/Sep 2004	6	47.6	23	10
Oct 2005	2	7.4	22	7

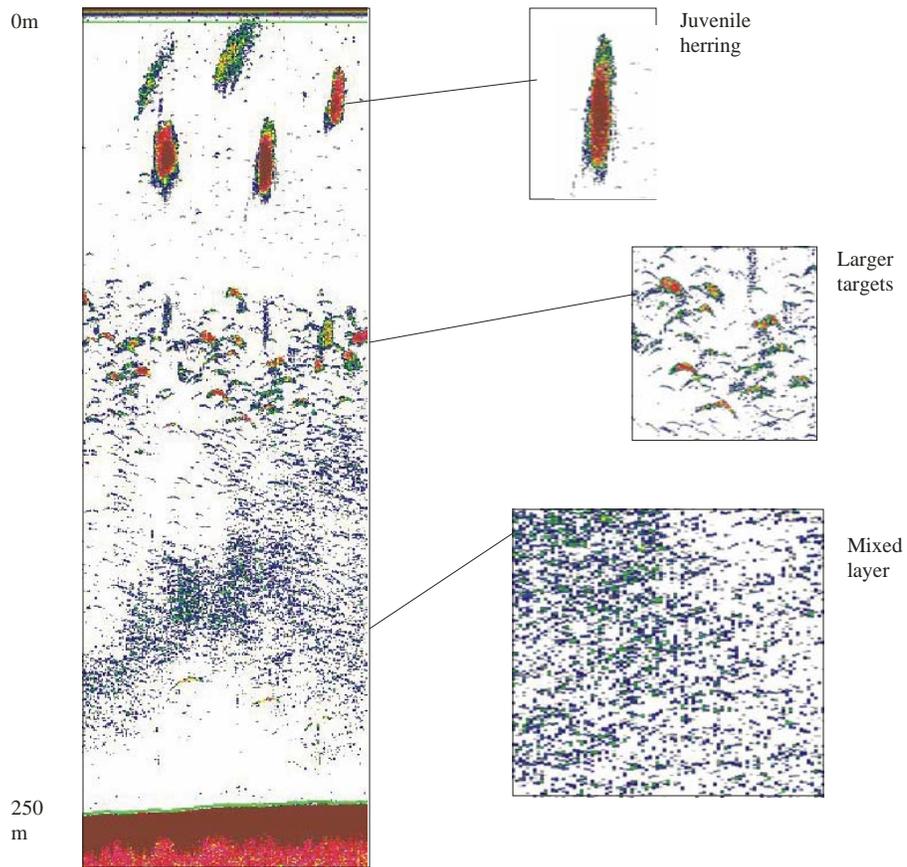


Figure 1. Example of an echogram showing the three backscatter categories used in echo integration.

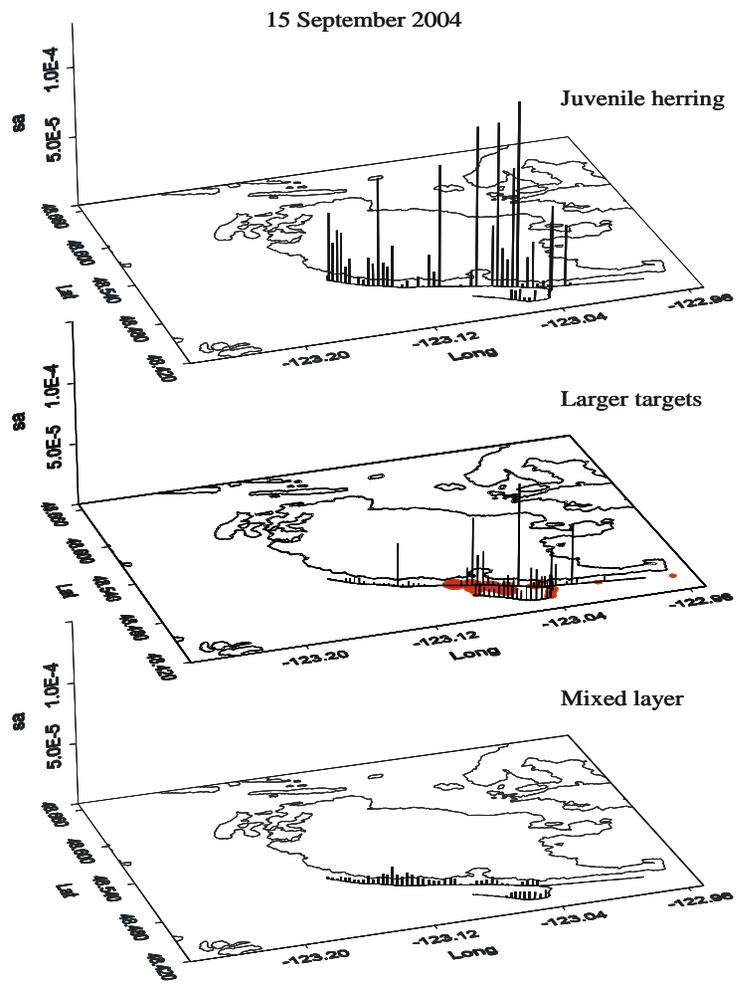


Figure 2a. Relative acoustic densities (s_a , units $m^2 \cdot m^{-2}$) of the three backscattering layers in the Haro Strait region on 15 September 2004. Red dots indicate killer whale locations.

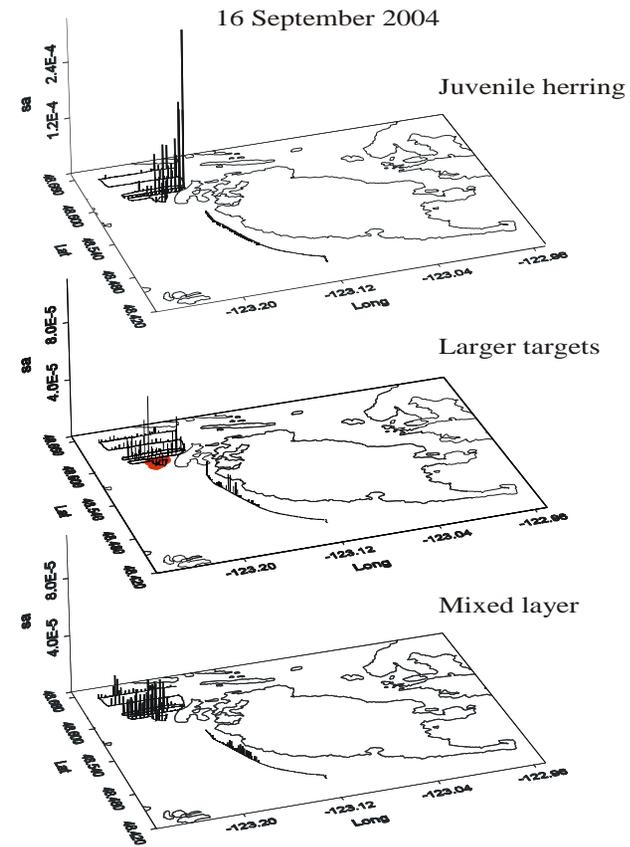


Figure 2b. Relative acoustic densities (s_a , units $m^2 \cdot m^{-2}$) of the three backscattering layers in the Haro Strait region on 16 September 2004. Red dots indicate killer whale locations.

Washington Chinook Abundance Abstract

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Over the last 30 years the total adult return of Puget Sound origin Chinook returning to the Strait of Juan de Fuca and Puget Sound has varied from a high of 292,000 in 1990 to a low of 118,000 in 1992. However this was followed by a general increase over the next ten years with an estimated 280,000 returning in 2001. Average abundance during the 1980s was 235,000; during the 1990s -172,000; and the 2000-2004 average is 222,000.

The majority of Chinook returning to Puget Sound are hatchery fish, comprising about 76% of the total return, with the natural-origin recruits (wild fish) now comprising about 24% of the total return. This level is slightly down from an average of 28% during the late 1970s and early 1980s.

Although Chinook are listed under the ESA, this is not the result of recent downward trends in total abundance. The Chinook ESU (Evolutionary Significant Unit) listing was directed at continued low returns of naturally spawning fish representing 22 distinct populations within Puget Sound.

Coastal Chinook populations consist of the north coastal rivers, Grays Harbor and Willapa Bay. Since the mid-1980 the total run sizes have ranged from a high of about 182,000 in 1988 to a low of 42,000 in 2002. Since 2002 adult returns have increased, with 2004 return of 110,000.

Total Chinook returns entering the Columbia River have varied widely over the years with a low return of 382,000 in 1995 and a high of 1,345,000 in 2003. The returns seem almost cyclic in nature, but are better explained due to variations in freshwater and ocean conditions.

In summary, within all three major areas of Chinook production, there has been no definable trend in terms of abundance since the mid-1970s. All regions do exhibit significant variations in run sizes from year to year, and this can largely be attributed to changes in freshwater and ocean conditions. With regard to the decrease in the whale population from 1995 through 2001, Puget Sound Chinook exhibited a 60% increase during this period.

There has been a historical decrease in the adult size of Chinook salmon returning to Puget Sound. The average body weight of Chinook in recent years is 71% of the average weight in the 1970s, and 56% the average weight observed in the 1950s. This is likely due to the higher composition of hatchery fish, which tend to return in higher percentages as 3 year-old fish rather than four and five-year old fish. However, the reduction of the total biomass of Chinook per unit abundance does not appear to coincide with the reduction in the whale population.

Regarding other salmon species, there is also no apparent trend in total abundance that would be identified as a contributor to the decline in the whale population.

Linking prey and population dynamics: did food limitation cause recent declines of resident killer whales?

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Northern and southern resident killer whales in British Columbia and Washington State experienced declines in abundance of up to 20% during 1996-2001. As a result, these populations were listed under Canada's Species-at-Risk Act as Threatened and Endangered, respectively, and southern residents have recently been listed as Endangered under the U.S. Endangered Species Act. Potential factors contributing to these declines include environmental contaminants, physical and acoustic disturbance, and changes in the availability or quality of food. Our recent research has shown that chinook salmon and, to a lesser extent, chum salmon, are important prey for resident killer whales, but other smaller salmonid species are not. The whales' strong preference for chinook salmon is likely due to this species' large size, high lipid content and, unlike other salmonids, its year-round presence in the whales' range. In this study, we assess whether food limitation was potentially a significant factor in recent declines of these whale populations. We examined the relationship between trends in killer whale population dynamics based on long-term photo-identification data, and abundance levels of chinook and chum salmon off the B.C. and Washington coasts over the past 25 years. Resident killer whale population productivity is regulated primarily by changes in survival rather than reproduction. Periods of decline resulted from unusually high mortality rates that were experienced by all age- and sex-classes of whales and were synchronous in the two socially-isolated resident communities. Fluctuations in observed versus expected age- and sex-specific mortality rates showed a very strong relationship to changes in coast-wide chinook salmon abundance, but no relationship to chum salmon abundance. A sharp drop in coast-wide chinook abundance during the late 1990s was strongly correlated with a significant decline in resident whale survival (Figure 1). The whales' preference for chinook salmon is likely due to this species' relatively large size, high lipid content and, unlike other salmonids, its year-round presence in the whales' range. Resident killer whales may be especially dependent on chinook during winter, when this species is the primary salmonid available in coastal waters, and the whales may be subject to nutritional stress leading to increased mortality if the quantity and/or quality of this prey resource declines. Although chinook salmon is clearly of great importance, determining whether this species is the principal factor limiting resident whale productivity will require on-going monitoring of both salmon and whale population trends.

A report describing this work is available on the Canadian Science Advisory Secretariat website at:

http://www.dfo-mpo.gc.ca/csas/Csas/Publications/ResDocs-DocRech/2005/2005_042_e.htm

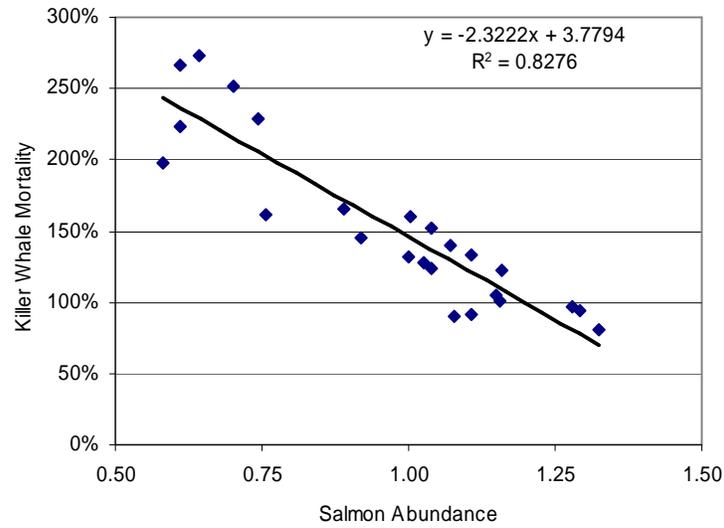


Figure 1. Percentage of expected mortality rates observed in northern and southern resident killer whale communities combined as a function of total chinook salmon abundance (all Pacific Salmon Commission Chinook Technical Committee index regions combined), 1979-2004. Mortality deviations are lagged by 1 year after chinook abundance, 3-yr running average mortality.

Killer Whale Energetics: Do they really have “killer appetites”?

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The debate concerning marked declines of marine mammal populations in the North Pacific has focused in part on predation by transient killer whales. Central to the argument is the energetic demands of the whales and how these demands are translated into food consumption rates. Data on the energetic demands of cetaceans are logistically difficult and often energetic demand is inferred from changes in body condition or caloric intake. However, these results may be complicated by seasonal changes in energy demand and the deposition and utilization of stored energy reserves in blubber. The goals of this study were to, 1) assess whether odontocetes in general and killer whales in particular have unusually high energetic requirements, 2) quantify seasonal variation in energetic demand and caloric intake in odontocetes, and 3) determine the relationship between energetic demand, energetic intake, and two often used proxies for energetic status (total body mass and blubber thickness).

We simultaneously measured metabolic rate, caloric intake, total body mass and blubber thickness across seasons in trained, adult male Atlantic bottlenose dolphins (*Tursiops truncatus*, mean body mass = 189 kg) and one adult female killer whale (*Orcinus orca*, body mass = 1800kg). Resting metabolic rate was determined from open-flow respirometry and was correlated to monthly averages of caloric intake and blubber thickness. External (air and water temperatures) and internal (plasma steroid hormone concentrations) cues that may influence energetic requirements were also measured.

We found that the mean resting metabolism of dolphins, 5.02 ± 0.85 SE $\text{mlO}_2 \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$, was approximately 2.1 times that predicted for terrestrial mammals of similar body size. Killer whales followed similar trends for sea otters, pinnipeds and other cetaceans with a resting metabolism that was 1.5 - 2.0 times predicted values. Field metabolic rates (FMR) estimated from these values also showed similar trends for odontocetes and other marine mammals as described by,

$$\text{FMR}_{\text{marine mammal}} = 1443.8 \text{mass}^{0.75} \quad (n = 10 \text{ species}, r^2 = 0.948, p < 0.001)$$

where FMR is in $\text{kJ} \cdot \text{day}^{-1}$ and mass is in kg.

Across seasons, we found that mean resting metabolic rate was not significantly different between summer (4.93 ± 0.24 SE $\text{mlO}_2 \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$) and winter (5.11 ± 0.15 SE $\text{mlO}_2 \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$) ($p = 0.525$; $F = 0.4107$) in bottlenose dolphins. Conversely, caloric intake decreased 41 - 46% during the summer period and body mass increased by 7.9 - 8.6% during the same period. Mean blubber thickness was 10.2% greater in winter compared to summer. Preliminary data from the killer whale indicates similar seasonal trends in energetic demand and caloric intake. While metabolic demand appeared to remain stable (2.49 ± 0.09 SE $\text{mlO}_2 \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$), caloric intake varied over two fold. Body mass was also significantly greater in the winter (1853.4 ± 12.5 kg) compared to summer (1783.33 ± 15.6 kg) ($p = 0.0022$, $F = 12.22$). These changes in caloric intake, body mass,

and blubber thickness were independent of air and water temperature (air: $p = 0.21$, $F = 1.59$; water: $p = 0.28$, $F = 1.16$). In the bottlenose dolphins, testosterone concentration was positively correlated with body mass and negatively correlated with blubber thickness and caloric intake.

The results indicate that killer whales do not have elevated metabolic demands compared to other marine mammals including smaller odontocetes. Rather, the high potential impact of killer whales on prey populations is attributed to the exceptional size of the individual predator (where adult whales are 10 to 20 times the mass of the biggest land carnivore) and to the large number of whales comprising social hunting groups.

Our results also indicate that there can be significant seasonal differences in the caloric requirements of cetaceans and that point estimates for a single species could underestimate the energetic requirements for a population by as much as 50%. Furthermore, there are significant time lags (1-2 months) between when a change in energetic demand occurs, and when caloric intake actually responds. This temporal disparity could have implications for management especially in a species such as the killer whale, which relies in part on salmon, a seasonally abundant prey source. Interactions between the availability of high quality prey, seasonal fluctuations in energetic demand, and potential time lags between energetic status and body fat stores could be important parameters to consider in proper management steps to protect marine mammal populations.

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Non-invasive Monitoring of Physiological Health of Southern Resident Killer Whales.

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Understanding the relative influences of the various pressures hypothesized to be responsible for the southern resident killer whale (SRKW) population decline could greatly enhance mitigation efforts aimed at their recovery. Our research aims to use a suite of fecal-based measures of physiological health to determine the relative importance of five hypotheses for the SRKW decline. These hypotheses include:

1. Disturbance stress from
 - (a) boat traffic and/or
 - (b) noise from US naval sonar testing.
2. Declining prey base
3. Physiological impacts from toxins
4. Declining reproductive health
5. Chronic pathogen exposure

We describe the validation of several non-invasive physiological measures from KW feces that are ideally suited to discriminate between the above pressures. These measures include: glucocorticoid (GC) metabolites resulting from adrenal activation as a generalized stress index; thyroid hormone (T3) measures of nutritional stress; estradiol (E2), progesterone (P4), and testosterone (T) as measures of reproductive function; and immunoglobulin (IgG and IgA) measures of infection and immunosuppression. Some of the above measures also show characteristic changes in response to toxin exposure. The relative importance of the 5 hypothesized pressures responsible for the SRKW decline should be ascertainable by the combined profiles of the above physiological measures in comparisons between the southern residents and less troubled populations.

For example, any one of the 5 hypothesized pressures could be associated with elevated fecal glucocorticoids in southern residents relative to less impacted subpopulations. However, the reduced prey and toxin hypotheses should be distinguished from the disturbance hypothesis by relatively low fecal thyroid hormone (T3) in feces; T3 is lowered in response to nutritional stress as well as by a number of endocrine disrupting toxins such as PCBs. The combination of glucocorticoids and T3 could also help tease apart the reduced prey hypothesis from the toxin hypothesis, as well as indicate a possible synergy between them. High PCB exposure under nutritional stress tends to lower glucocorticoids and T3, whereas glucocorticoids are elevated and T3 decreased in response to PCB exposure without accompanying nutritional stress. Endocrine disruption (hypothesis 3) might be further indicated by a marked change in the relative amounts of estradiol and testosterone metabolites in feces. Reduced reproductive function (hypothesis 4) would be associated with reduced T in males, and reduced P in females at each stage of gestation (as determined by backdating from the estimated time of parturition). Finally, the pathogen hypothesis should be indicated by

high variance in IgG and IgA. SRKWs are expected to be immunosuppressed, on average, from any of the pressures in hypotheses 1-3. However, chronic pathogen pressure under such conditions should cause a relatively larger portion of the SRKW to experience infection, followed by a strong immune response. The resultant high and low immune responses across individual southern residents would produce relatively high between-individual variation in their immunoglobulin levels compared to less stressed populations.

When investigating fecal-based physiological measures in a new species, one must show that the molecule is present in sufficient quantities in feces and can be reliably measured across its range of concentration. We describe validation studies for each of the endocrine measures described above. (Validation of the immunoglobulins are still in process and will not be described further.)

We ultimately plan to use detection dogs, specially trained to locate scat (Wasser et al 2004) from free-ranging KW, to enhance sampling efficiency. To illustrate this method, we describe the application of detection dogs to collect scat from North American right whales in the Bay of Fundy, conducted in collaboration with R. Rolland and S. Kraus of the NE Aquarium (Rolland et al (in press)).

METHODS

Fecal samples from captive killer whales were acquired from SeaWorld. Wild killer whale fecal samples were collected by NMFS scientists from Springer, just prior to her translocation back to her natal pod, and from an adult male from J pod in the Puget Sound. Parallelism tests were conducted on these fecal samples to assess assay reliability for detection of glucocorticoid, T3, estradiol, progesterone, and testosterone metabolites across their ranges of concentration. Hormones were extracted using methods described by Wasser et al. (2000). Extracts were serially diluted and each dilution assayed to confirm that the slope of the resultant dilution curve was parallel to that of the standard curve used to estimate their respective hormone concentration. We also compared fecal T3 concentrations of a captive killer whale of unknown sex to a wild adult male SRKW to provide preliminary comparisons of captive versus wild differences in nutritional status.

RESULTS

All hormone serial dilutions from killer whale fecal extracts were parallel to their respective standard curves, demonstrating that we are accurately measuring them across their ranges of concentration (Figs 1-5).

The T3 concentration of the captive killer whale was four-fold higher than the T3 concentration of the wild male, providing preliminary evidence of a nutritional deficit in the wild KW examined. However, additional comparisons will be required to further confirm this assertion.

Scat detection dogs provided a greater than five fold increase in sampling of North Atlantic right whale scat compared to sampling by researchers skilled in right whale scat collection (Rolland et al. in press). This sampling method holds considerable promise for acquiring the sample size needed to compare the physiological status of SRKW to other comparison populations using feces. Gonadal hormones extracted from the right whale

samples were also able to distinguish immature from sexually mature males and females, as well as reliably distinguish between resting, lactating and pregnant adult females (Rolland et al. 2005). Glucocorticoids from these samples were able to distinguish stress associated with fishing net entanglement, poor health, and mating competition in males (Hunt et al. in press).

IMPLICATIONS FOR SRKW CONSERVATION

We have validated a suite of fecal-based physiological measures on captive and southern resident killer whales for discriminating between the various hypotheses for the decline of the SRKW. These methods, coupled with use of scat detection dogs to acquire sufficient sample sizes from free ranging cetaceans, hold considerable promise for discriminating between pressures impacting SRKW recovery. We are now in the process of training dogs to detect SRKW feces and hope to pilot this method this spring. Other information, such as a host of genetic indices (Wasser et al. 2004), prey preferences and pathogen identification can also be acquired from such samples.

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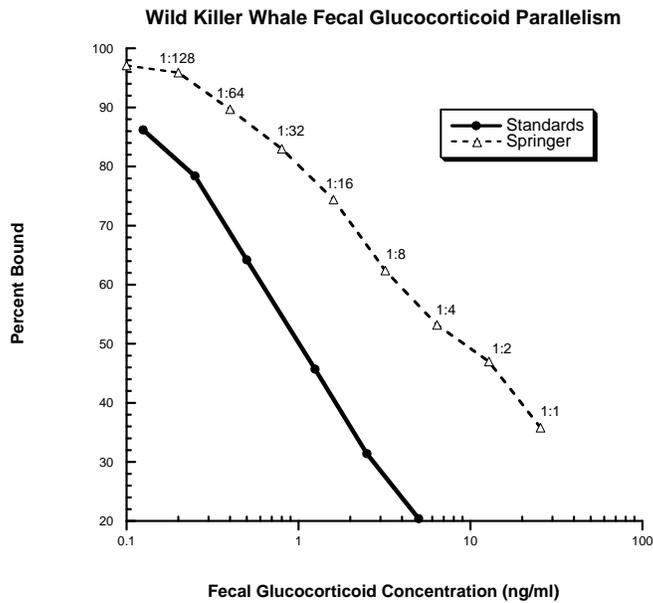


Fig. 1. Glucocorticoid parallelism of a serially-diluted fecal sample from Springer compared to known standard concentrations of the hormone. Parallel lines indicate the reliable measurement of the hormone in feces across its range of concentration.

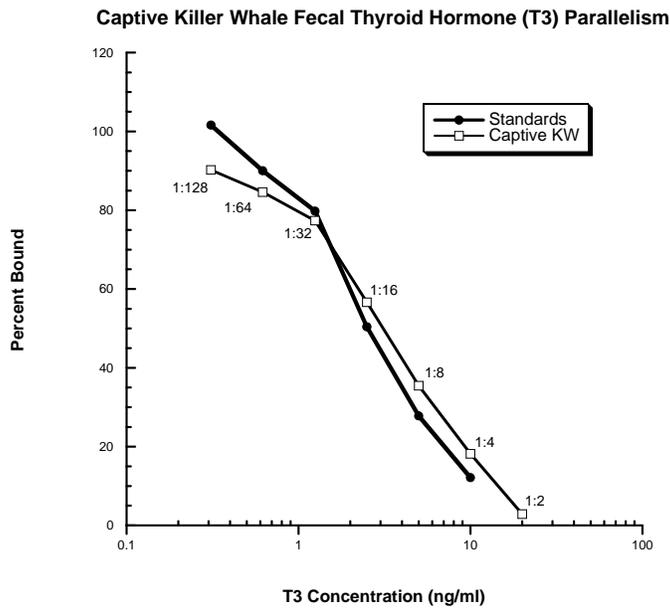


Fig. 2. Thyroid hormone parallelism of a serially-diluted fecal sample from a captive killer whale compared to known standard concentrations of the hormone.

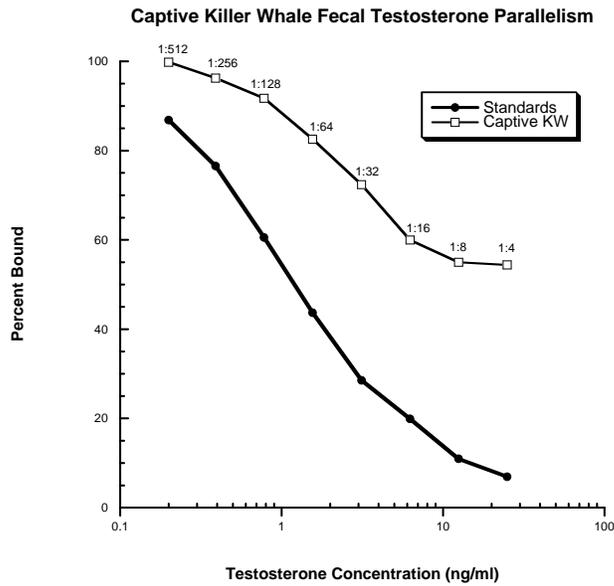


Fig. 3. Testosterone parallelism of a serially-diluted fecal sample from a captive killer whale compared to known standard concentrations of the hormone.

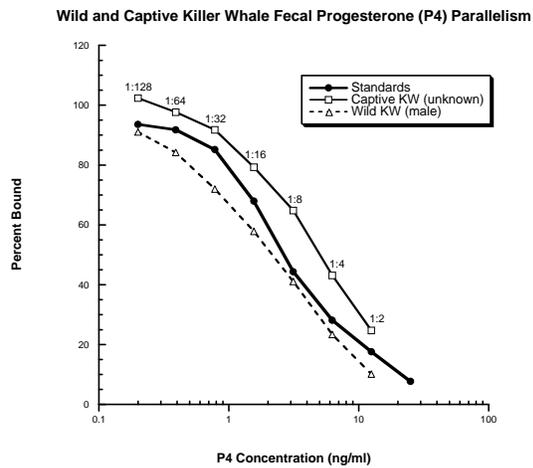


Fig. 4. Progesterone parallelism of a serially-diluted fecal sample from a captive killer whale and a free ranging SRKW (J6) compared to known standard concentrations of the hormone.

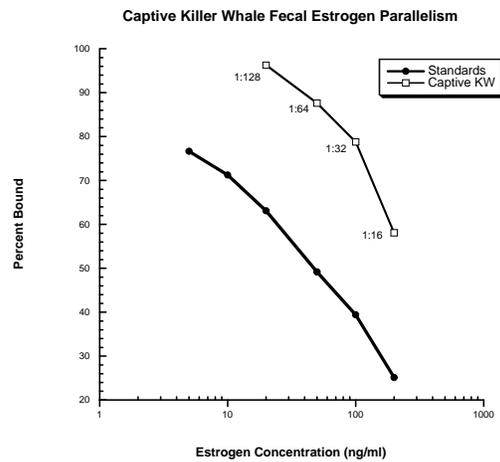


Fig. 5. Estrogen parallelism of a serially-diluted fecal sample from a captive killer whale compared to known standard concentrations of the hormone.

Winter Distribution of Southern Resident Killer Whales, 2003-2006

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The presence of southern resident killer whales (SRKW's) in marine waters from central Vancouver Island to central California was assessed during winter months from 1 October 2003 to 1 April 2006 using sightings reports from the public, confirmation of SRKW reports using photographs and acoustic techniques, and follow-up interviews with selected individuals. An existing Puget Sound sighting report system (Orcanetwork.org) was enhanced for this purpose, and thirty road trips were conducted during three winters to outer coastal ports and harbors within the study area to distribute fifteen hundred laminated posters in order to establish a coastal sighting network for reporting killer whale sightings. Six hundred of these laminated posters were posted in high visibility locations around the ports and harbors and along the routes between them. A toll-free number operated by Orcanetwork.org was available for respondents to call in or email at any time to report killer whale sightings.

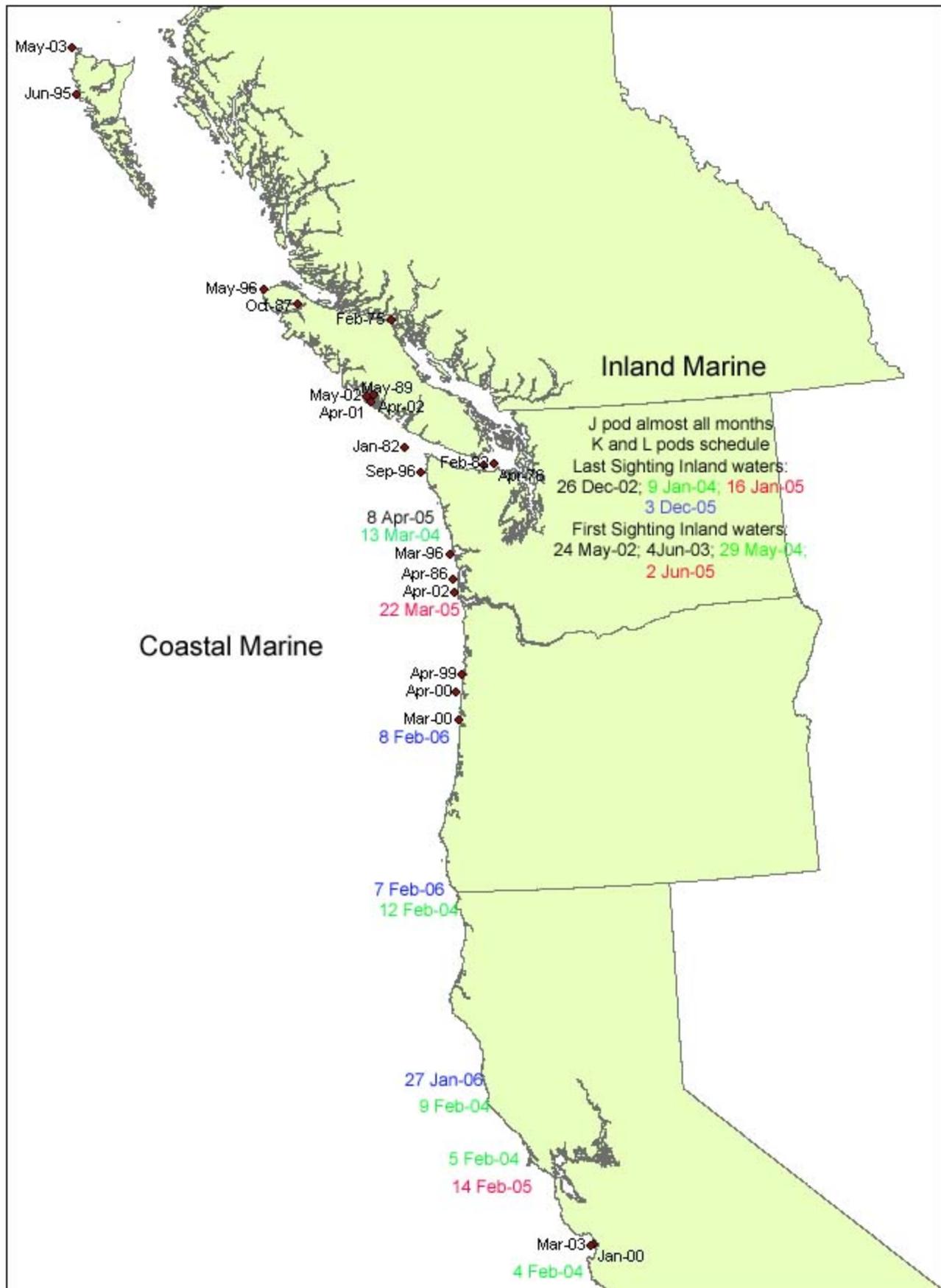
More than one thousand public sightings reports were received during the winter study periods (2003/04, 2004/05, 2005/06), often with multiple reports of the same occurrence, suggesting that observer coverage was good in the interior marine waters of Washington state. In all three years, during autumn and early winter months all three pods foraged in or near greater Puget Sound and lower Georgia Strait. Sixty-four coastal killer whale sightings reports were received during the study period that suggest the late winter coastal distribution for the majority of SRKW's can be characterized as travels southward along the coast from Washington toward central California, commencing in January; and then, travels northward from central California to British Columbia commencing mid-February to early March. The whales' travels appear to be exclusively in continental shelf waters, and often near coastal (within 12nm of shore), although that may be an artifact of reporting (observers are typically land-based or aboard near-coastal vessels). It is tempting to seek correlation of the whales' distribution with the distribution and abundance of prey stocks of salmon.

One pod, J pod (currently numbering 24 individuals) appears to travel less extensively, and is more frequently reported in inland marine waters of Washington state and British Columbia during all winter months. The largest pod, L pod (currently numbering 43 individuals) appears to dissociate into matrilineal groups, one of which ("L12's", currently numbering 10 individuals) is often sighted alone, or (eg., during March 2006 sightings in Puget Sound) in loose association with K pod (currently numbering 20 individuals). The size of a pod reported in a public sighting provides a clue regarding whether the whales being reported *might* be SRKW's, but these sightings reports alone reveal only an apparent pattern of distribution that requires confirmation. Photographs or acoustic recordings are essential for confirming any report. For many reports, the whales were often quite spread out over many miles of ocean during winter months, making observers' estimates of numbers problematic. The locations of all sightings reports confirmed to be or reasonably certain to be SRKW's are plotted (Figure 1).

Many winter killer whale reports throughout the study area can be attributed to the “transient” ecotype of killer whale, particularly when predations on other marine mammals are reported with the sightings, or if the reports are with photographs or from experienced observers. There was an obvious increase of sightings reports of “transient” killer whales in inland marine waters of the Pacific Northwest during autumn and winter months of all three winters of this study period, and the reports were typically of small groups of 1-7 individuals. The most recent winter was unusual in this respect, with our encountering a group of 31 “transient” killer whales in Haro Strait on 11 February 2006. Many of these “transient” killer whales subsequently traveled deep into Puget Sound from 12-15 February before some of them went north in Georgia Strait on 16 February, and others went west in the Strait of Juan de Fuca. We photo-identified all of the “transient” ecotype whales in this Haro Strait encounter, and matched many of them to ID photographs taken prior from California to Alaska. One sub-group of six whales from this encounter also spent six months in Hood Canal last winter/spring/early summer (Jan-July 2005).

Some of the sightings reports with photographs taken in February and March 2006 suggest that a large number of L pod whales (25-30 of 43) were foraging between Point Reyes, California and the central Oregon coast, during the same approximate time period that the remainder of L pod (L12's) and K pod were foraging further north and entering Puget Sound on 1 March 2006. On 10 March, a sighting of 12 whales in Swanson Channel suggests that these whales may have traveled further north into the Canadian Gulf Islands and Georgia Strait. The location of J pod was unknown between 10 February and 17 March 2006.

One six year old male SRKW (L98) was accidentally killed by the propeller of a slow moving ocean tug in Nootka Sound, British Columbia on 10 March, 2006, thus ending a dilemma concerning what to do about “lost” juvenile whales that become separated from their natal pod. We can only surmise from Luna's long solitude that the pods of SRKW's did not regularly come into Nootka Sound in the recent five years.



Listening for Orcas in the Underwater Racket of Cape Flattery and Haro Strait

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1. Introduction

The underwater ambient sound field contains a lot of information about the marine environment. By passively monitoring the underwater sound we can make quantitative measurements of physical processes such as wind speed and rainfall rate, detect and monitor sound-producing marine animals, especially whales, and monitor anthropogenic activities, including shipping, sonars and other sound-producing human activities. Identifying these sources of sound, their duration and relative loudness as a function of frequency and location provides a fundamental basis of information needed to address the question of the impact of anthropogenic noise on the marine environment.

2. Long-term Monitoring

Measurements over many months or years are needed to establish baseline sound budgets that can be used to access climatic changes or trends in biological or human activities. As part of research to establish rainfall climatologies over the ocean, an acoustic rain gauge was developed (Ma and Nystuen, 2005). This instrument is a small, relatively inexpensive, low-power, low duty-cycle acoustic recorder that can be easily attached to surface or sub-surface ocean moorings (Fig.1). It is autonomous and can be deployed for up to one year without servicing. It records acoustic spectra, rather than actual time series, greatly reducing the amount of data storage needed to describe the sound field. For the purpose of monitoring the entire sound field, these instruments have been renamed Passive Aquatic Listeners (PALs).



Fig. 1. Passive Aquatic Listener (PAL)

Spectral and temporal characteristics of different sound sources allow acoustic classification of each source. Fig. 2 shows a typical week-long record of underwater sound in a marine environment. Sound levels vary widely and at different time scales. The slowly varying background, at the time scale of hours, is closely associated with wind speed, and can be used to quantitatively measure wind speed when no other sound sources are present (Vagle et al. 1990). Shorter duration loud events, at a time scale of minutes, are associated with rainfall and shipping. Typical spectral characteristics for these sound sources

are shown in Figure 3 and provide a basis for objective classification. Rainfall is relatively loud at higher frequencies, and shipping is relatively loud at lower frequencies. The objective classification of these sound sources is superimposed on the sound record shown in Fig. 2.

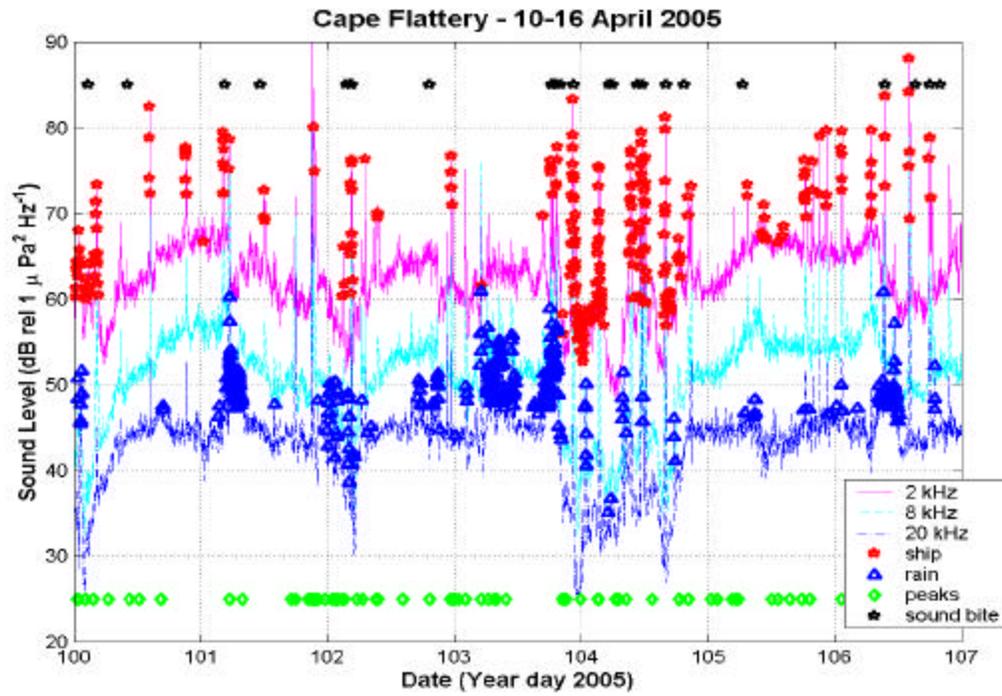


Figure 2. A week-long underwater marine sound record. The sound level density is shown for 2, 8 and 20 kHz along with the acoustic classification of the source (ship, rain, peaks). Times for "sound bites" are also shown.

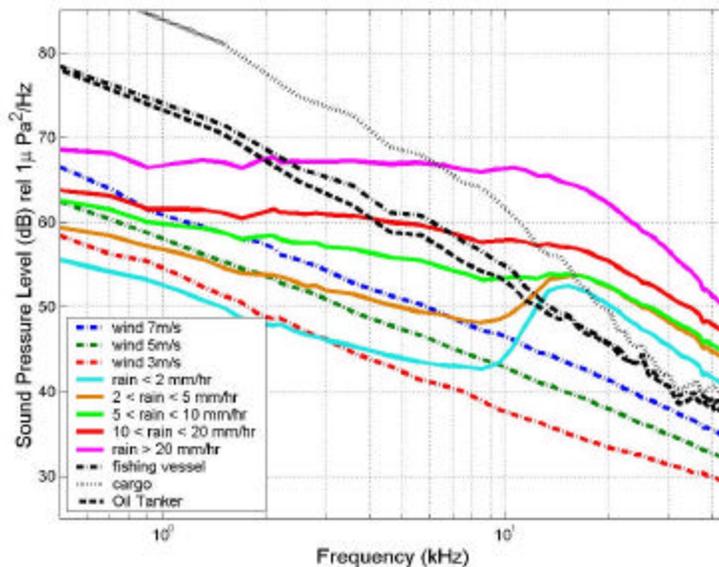


Figure 3. Typical spectral sound signatures for wind, rain and ships. Wind is relatively quiet, with a distinctive spectral slope. Rain is relatively louder at high frequency. Ships are relatively louder at low frequencies.

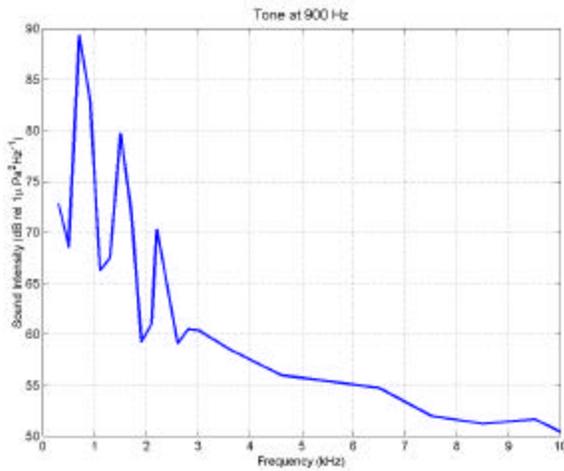


Fig. 4. Spectral signature showing a "peak" in the spectrum. This is consistent with a whale "whistle" and potentially indicates detection. This record is from the Bering Sea (May 2004).

At an even shorter time scale, calls associated with marine mammals, or sonar pings, are present. These can be detected by sampling the sound field several times within a few seconds. If one of

samples is much louder than the others, then a "transient" sound is detected, and can be classified. These sounds include whistles and clicks that have unique features that can be used to identify the species, or even sub-species (or pod) of the animals producing the sound. An example of a whistle is shown in Fig. 4. Times when such "peaks" in the acoustic record are present are also indicated in Fig. 2. These are potentially "whale detections".

3. Validation

Validation of the acoustic classification of whale detection either requires visual observation (very difficult) or the actual time series to which experienced researchers can listen. The processing software of the PAL has been modified to allow the storage of selected "time bites" based on the suspected sound source. During 2005, modified PALs were deployed at Cape Flattery and in Haro Strait (Fig. 5). At each location, multiple "time bites" verified acoustic detection of whales, including orcas (Fig. 6), and, at the Haro Strait site, were also corroborated by visual observations.

Fig. 5. Locations where PALs have been deployed in the Puget Sound Region. PALs were deployed at Cape Flattery and Haro Strait in 2005. PALs are currently deployed at Cape Flattery and Westport (Jan – July 2006).



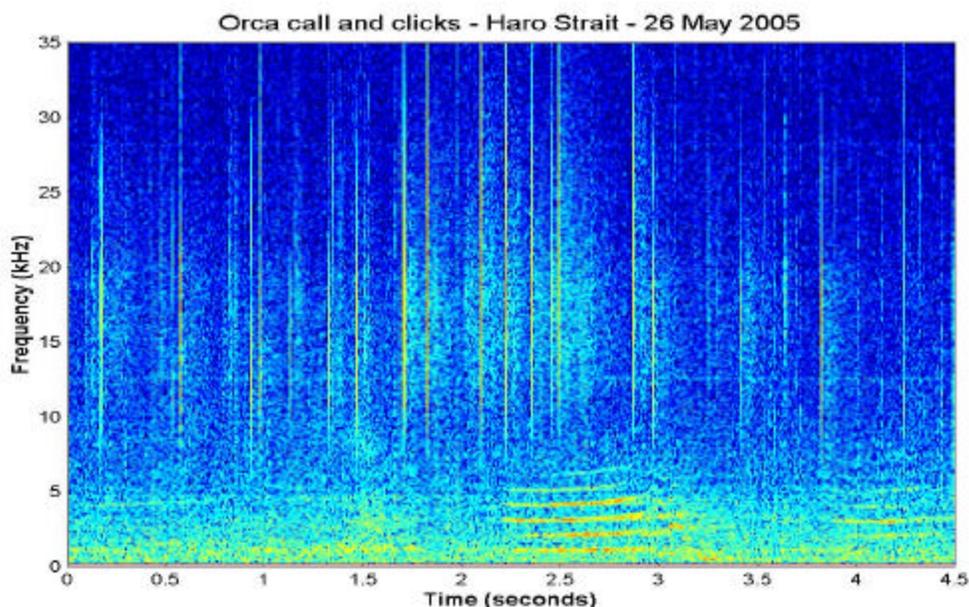


Fig. 6. An example of a "sound bite" of an orca call and clicks in Haro Strait displayed as a sonogram (time versus frequency with level in color). These "sound bites" are used to verify the identity of the sound source.

4. Sound Budgets

Once the sound sources are identified, components of the sound budget can be quantified. Table 1 shows the percentage of time that different sound sources are detected for 3 different locations: Cape Flattery, Haro Strait and the Bering Sea. Haro Strait is a location with heavy shipping traffic and the Bering Sea location is relatively "pristine" with minimal shipping. Table 1 also reports the relatively "loudness" of these different sound sources for two different frequency bands: 2 kHz and 20 kHz. Shipping is the loudest sound source at 2 kHz, whereas rainfall is relatively loud at 20 kHz. This points out that sound budgets depend on frequency as well as location.

5. Conclusions

Low-duty cycle acoustic recorders have been deployed in the Puget Sound region to monitor the ambient sound field. These instruments can quantify the sound budget by objectively identifying the sound source. The signal from wind, rain, ships and whales are reported. Validation of acoustic classification is achieved by recording short "sound bites." Multiple sound bites verified the detection of orcas at the measurement locations. These instruments are capable of long-term deployments in harsh weather conditions on sub-surface moorings that are cheaper, safer and easier to maintain than surface moorings.

Table 1. Sound Budget Statistics

Dominate Sound Source (% of time present)					
	Ships	Wind	Rain	Peaks	
Cape Flattery (Apr 2005)	21%	70%	6%	3%	
Haro Strait (May 2005)	23%	53%	8%	12%	
Bering Sea (May 2004)	1%	85%	6%	7%*	
Sound Levels at 2 kHz (dB relative to 1 $\mu\text{Pa}^2 \text{Hz}^{-1}$)					
	Ships	Wind	Rain	Peaks	All
Cape Flattery (Apr 2005)	62 ± 7	54 ± 5	57 ± 5	57 ± 7	55 ± 7
Haro Strait (May 2005)	63 ± 7	55 ± 6	58 ± 8	59 ± 8	58 ± 8
Bering Sea (May 2004)	72 ± 5	60 ± 7	64 ± 6	62 ± 7	60 ± 8
Sound Levels at 20 kHz (dB relative to 1 $\mu\text{Pa}^2 \text{Hz}^{-1}$)					
	Ships	Wind	Rain	Peaks	All
Cape Flattery (Apr 2005)	33 ± 8	35 ± 6	49 ± 6	34 ± 5	34 ± 8
Haro Strait (May 2005)	40 ± 6	40 ± 4	52 ± 5	41 ± 6	42 ± 6
Bering Sea (May 2004)	44 ± 6	42 ± 5	48 ± 2	41 ± 6	42 ± 5

* sampling was biased against storage of "peaks"

References:

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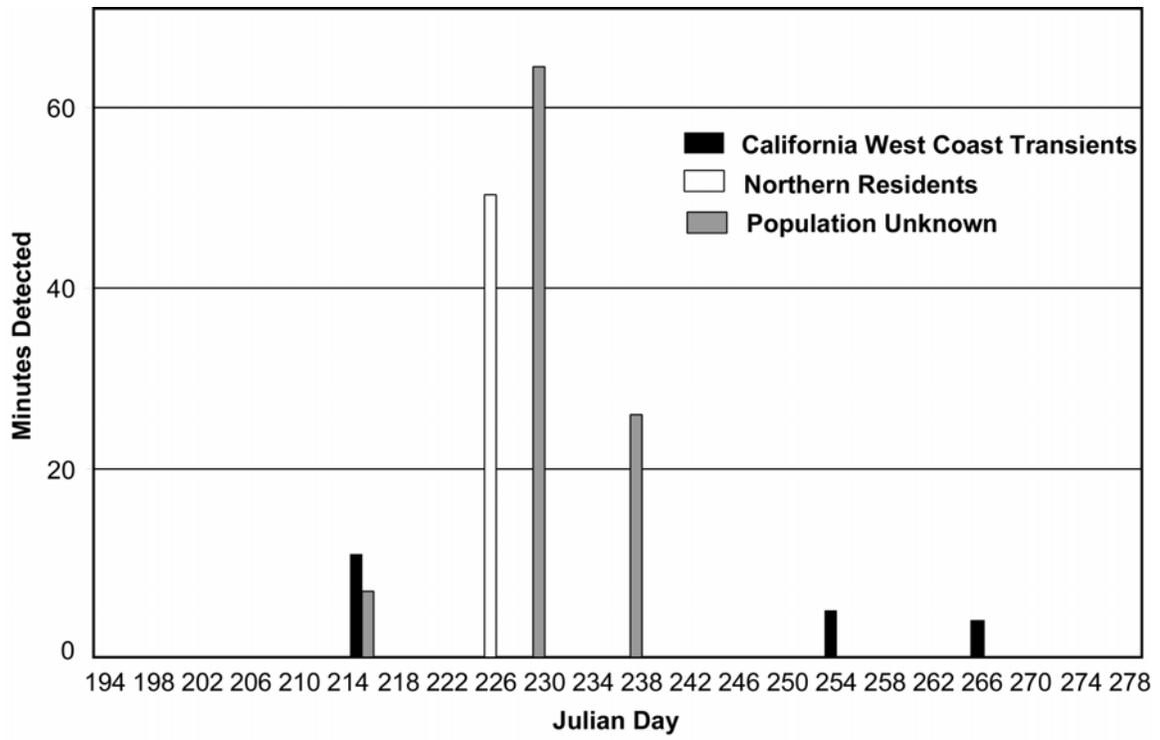
S Vagle, WG Large and DM Farmer, 1990: An evaluation of the WOTAN technique for inferring oceanic wind from underwater sound. *J. Atmos. and Ocean. Tech.* 7, 576-595.

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Killer Whale Acoustic Monitoring in the Coastal Waters of Washington

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A High-frequency Acoustic Recording Package (HARP) was deployed approximately 15 miles off the Washington coast within the southern portion of the Olympic Coast National Marine Sanctuary off Westport, WA in July, 2004 to conduct an acoustic survey for cetaceans. The HARP sampled at 80 kHz sample rate continuously for three months. Several odontocete species have been identified in the acoustic record, including killer whales. From mid-July to early October, killer whale vocalizations were detected on seven occasions, with calling heard for as few as five minutes up to an hour. Based on the occurrence of discrete calls, two populations of killer whales have been identified from these recordings. Northern Residents were detected in mid-August and West Coast transient killer whales, representing the California dialect, were heard in early August and on two days in mid-September. Three additional recordings of killer whales from August have not been identified to a specific population. On-going monthly visual surveys conducted from Westport, WA (John Calambokidis, Cascadia Research) have observed killer whales on three occasions since July, 2004. Photos of the killer whales indicate two of these sightings consisted of groups of transients. None of these sightings were associated with acoustic detections of killer whales. Acoustic recording has continued since October, 2004 at a site further offshore, and at sites off Westport and Cape Elizabeth during March – May 2005, although these data have not been analyzed to date. Acoustic detection of killer whale calls is a promising method for monitoring the presence of killer whales along the outer Washington coast. Killer whale recordings from this dataset were identified to population by Volker Deecke at UBC.



Effects of environmental factors and temporal scale on pod-specific SRKW summer distribution patterns: implications for designating critical habitat

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Background

Recent population declines of Southern Resident killer whales (SRKW) have led to several US and Canadian conservation listings, and recovery planning necessitates designating critical habitat. Although this killer whale population is arguably the most extensively studied world-wide, there is still considerable uncertainty over something as vital as how the population is distributed in time and space. Thus, quantifying patterns of pod-specific SRKW distribution and identifying key areas used by SRKW is a necessary first step towards designating critical habitat and studies assessing risk of potential factors on habitat use. This information can then be used to inform critical habitat designation, a condition of listing under the US Endangered Species Act.

The overarching goal of our research is to determine SRKW summer distribution patterns within Washington and British Columbia inshore waters. Our approach was to consider, first, pod-specific summer distribution and, second, overall SRKW space use. At the pod level, our goals were to: (1) determine and compare pod-specific distribution and high use regions; (2) evaluate degree of spatial overlap and specialization by pods; and (3) analyze how distribution relates to static environmental factors (i.e. depth, slope, and distance from nearest shore). Lastly, we describe the effects of temporal scale on overall SRKW distribution patterns and how temporal scale may affect management and conservation planning.

Methods

We modeled SRKW summer space use patterns via a platform of opportunity; sightings were reported by a centralized spotting network of commercial whale watchers (the 'Pager Network') from May to September 1996-2001. We attempted to understand and quantify potential biases associated with these opportunistic data using a field study (Hauser et al. in review). Distribution was first modeled using kernel density functions in ArcGIS (ESRI v. 9.1) to describe relative frequency of sightings by area. A relatively novel spatial modeling approach using principles of landscape ecology was used to compare among pods and between temporal scales where absolute pod sighting densities were normalized into five relative classes for comparison of 'low' to 'high' use regions. The class representing the highest use area was considered 'core'. We also considered associations of pod high use areas with depth, slope, and distance to nearest shore. Normalized kernel densities and core areas were also modeled for each week, month, and annual season of overall SRKW sightings to evaluate the effects of temporal scale on distribution.

Pod-specific distribution and core regions

Summer distribution maps of relative use showed intriguing similarities and potentially important differences among J, K, and L pods (Fig. 1). The highest density of all pod sightings occurs in Haro Strait along the west side of San Juan Island into Boundary Pass. Regions in the Strait of Juan de Fuca along Vancouver Island also contribute a higher sighting density to L pod distribution. All three pods display similar space use in areas that are commonly classified as low to high density regions, with common core areas occurring in Haro Strait (Fig. 2, panel a). However, there are also areas that are very frequently used by one pod and very rarely by other pods, suggesting specialization. L pod exhibits such 'specialized' areas in the Strait of Juan de Fuca while J pod specializes in small regions of Rosario Strait and Active Pass. K pod does not have such specialized areas. All three pods tend to overlap in the overall extent of distribution, but L pod ranges furthest south, and J pod north (Fig. 2, panel c). Lastly, pod core areas are shown in figure 2, panel d. The west side of San Juan Island in Haro Strait is classified as core for all three pods, but regions along Vancouver Island in the Strait of Juan de Fuca are part of L pod's core region and J pod has core regions leading into Swanson Channel. These analyses highlight areas of high conservation interest, based on relative occurrence as well as key pod differences. Habitat management could protect areas of high occurrence by pod, particularly for L pod which has recently experienced greater population declines than J and K pod. L pod, in particular, appears to have very different space use tendencies.

Environmental effects on pod-specific distribution

Spatially explicit correlations of pod density with static environmental factors suggest that pods respond similarly to their environment (Fig. 3), despite exhibiting some distinct distribution patterns described previously. Core regions of SRKW pods appear to occur in deep, near-shore, moderately sloping regions of Washington and British Columbia inshore waters. Core regions for each pod occur in areas that are deeper and steeper than what is available in the study area and are very near-shore. However, the study area is entirely composed of inshore waters and SRKW pods do not appear to select core areas necessarily closer than what is available. Continued research into the mechanisms for these environmental associations is warranted, including spatially explicit analyses of prey and environment.

Effect of temporal scale on overall SRKW distribution

The effect of temporal scale on overall SRKW distribution was examined using several metrics frequently employed in landscape ecology that address the composition and configuration of distribution patterns. Specifically, we analyzed variations in core regions at weekly, monthly, and seasonal temporal scales. SRKW clearly exhibit spatially complex, dynamic summer distribution patterns, with greater variation at finer temporal scales. Figure 5 illustrates spatially explicit analyses of core classification frequency at each temporal scale, identifying areas that were rarely to always considered core among iterations of each temporal scale. Some regions, like those in Haro Strait, were always or near always considered core while others were considered core ephemerally, regardless of temporal scale. Likewise, some areas were never considered core. These results

suggest that areas that are often considered core, regardless of temporal scale, may be good management target regions. Additionally, variation in distribution patterns is minimized at coarser temporal scales (annual seasons), but coarser temporal scales also encompass the greatest total amount of core space (Fig. 5) that may lead to greater mitigation with potential population risk factors than finer temporal scales (weekly) that include a much smaller total amount of space. A dynamic management plan designating habitat protection of core areas on a fine scale at appropriate times may provide the most specific protection of the population as well as limit mitigating factors.

Summary

SRKW exhibit non-random, non-homogeneous summer distribution patterns. Furthermore, SRKW display complex, dynamic summer space use patterns that vary among pods and temporal scale as well as in relation to the available habitat features. Our key findings are that:

- (a) regions of Haro Strait are considered core for all pods, at each temporal scale;
- (b) J and L pods show specialization in certain areas;
- (c) although there are appears to be dynamic and variable distribution patterns among the pods, each pod responds similarly to environmental factors;
- (d) core regions are associated with deeper and steeper inshore waters than what is available in the study region; and,
- (d) scale matters to how managers designate critical habitat.

Ultimately, we conclude that critical summer habitat designation can be accordingly specific to pod and temporal scale, as well as in relation to environmental factors within Washington and British Columbia inshore waters.

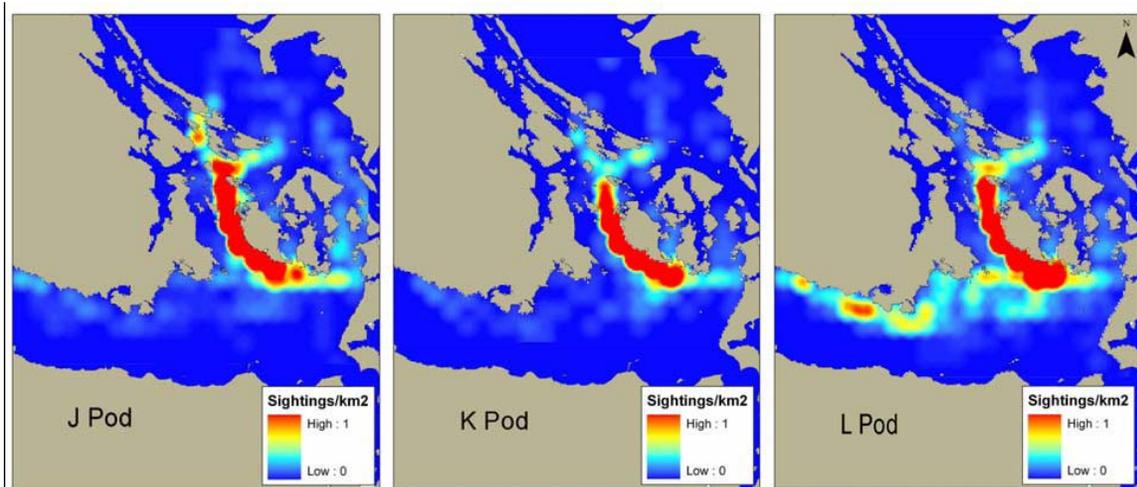


Figure 1. Distribution maps for J, K, and L pod groups showing the density of sightings/km², normalized as the proportion of the maximum density for each pod. Red coloration represents higher sighting density and blue coloration represents low sighting density.

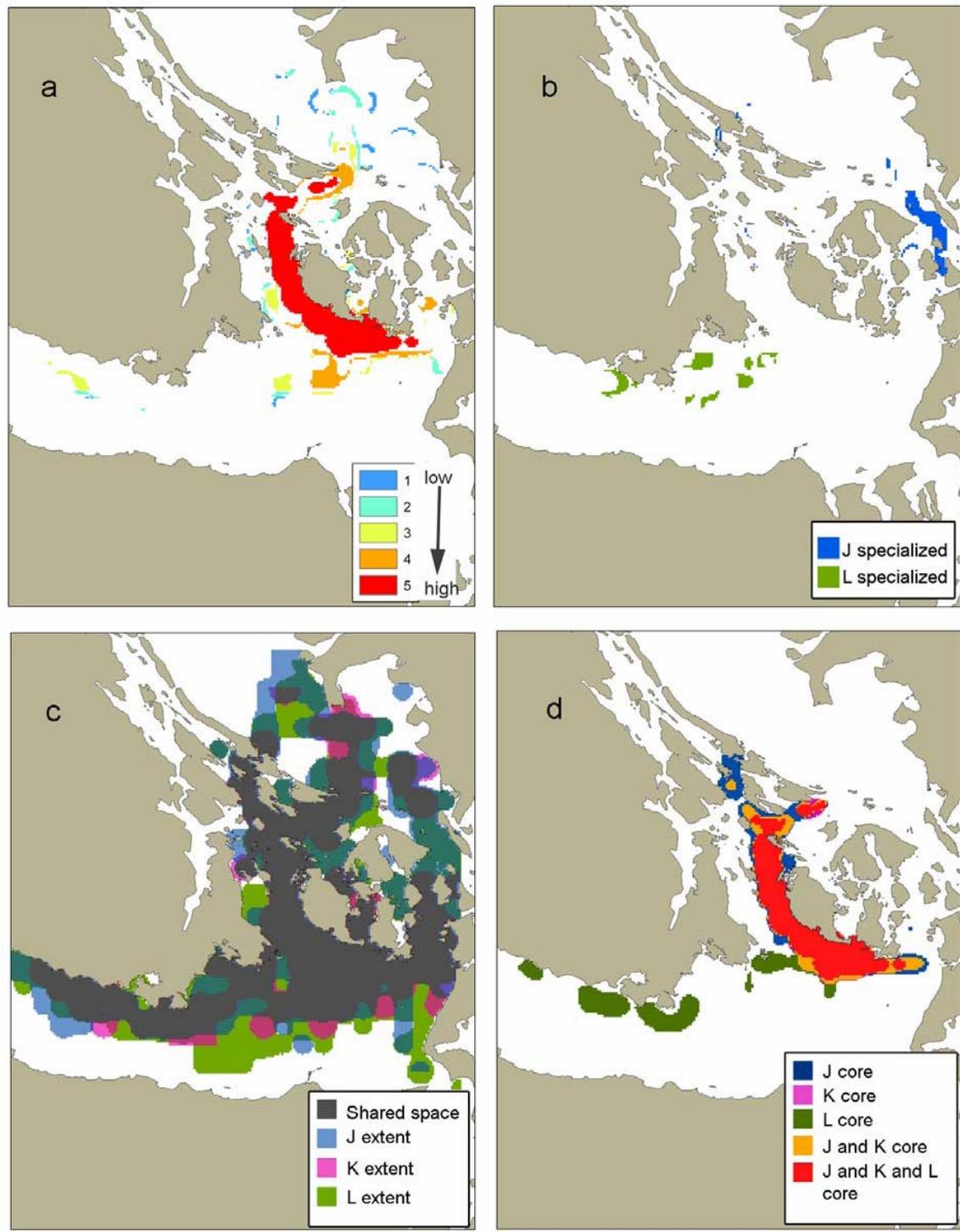


Figure 2. Pod-specific space use illustrating common classification of space by all pods (a), areas where a single pod is found at high densities while the other two pods are classified as low density (b), overall extent of space used by each pod (c, with 30% transparency for each pod), and regions classified as core for one or more pods (d).

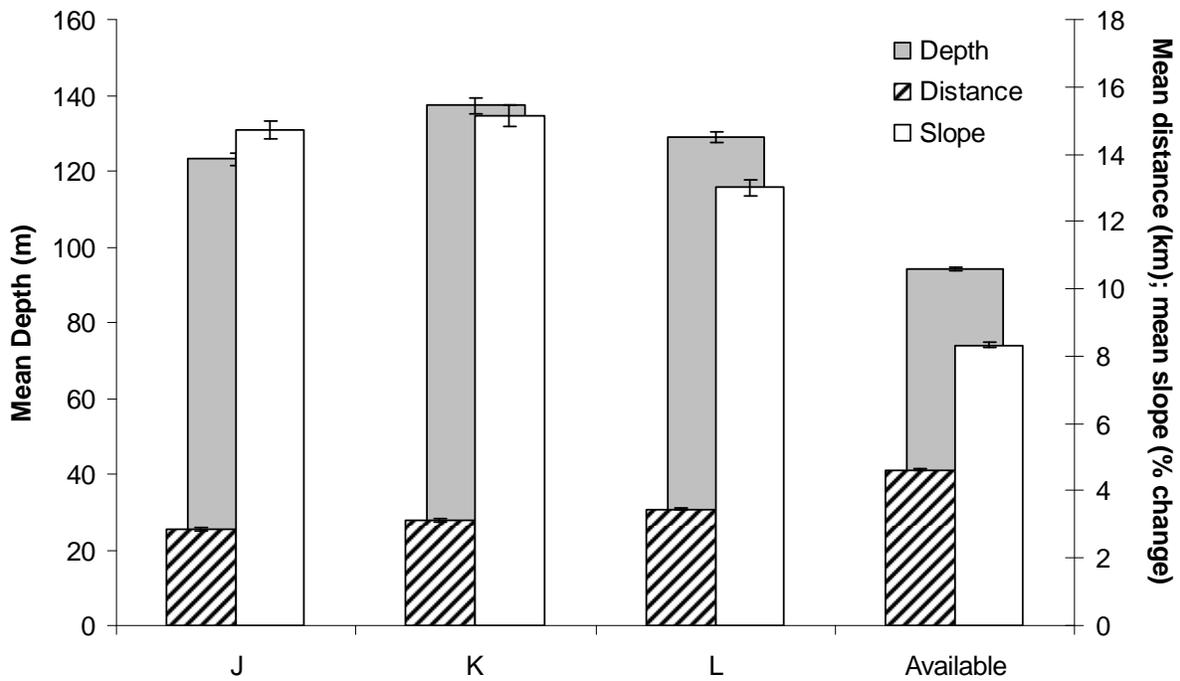


Figure 3. Mean depth (m), distance from nearest shore (km), and slope (+/-1 se) for each pod's core region as well as those available throughout the study region.

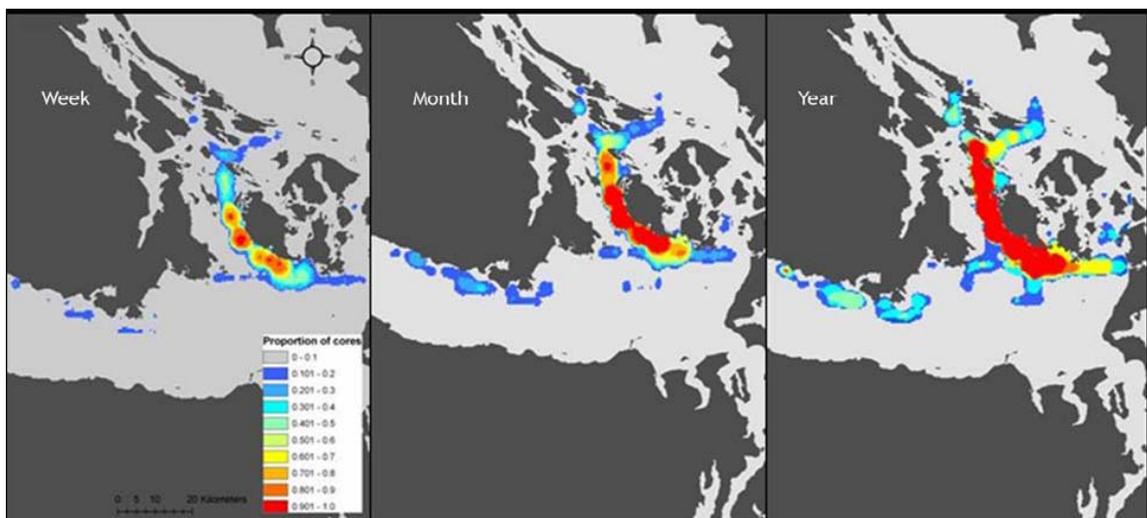


Figure 4. Maps representing the proportion of areas classified as core at each temporal scale (week, month, and annual summer season), where warm colors (reds) are areas that are more frequently considered core regions and cool colors (blues) were considered core but only rarely.

LAND-BASED STUDIES OF THE EFFECTS OF VESSEL TRAFFIC ON THE BEHAVIOR OF NORTHERN AND SOUTHERN RESIDENT KILLER WHALES (*ORCINUS SPP.*)

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Vessel traffic is being evaluated as a factor that may have contributed to Southern Resident Killer Whales becoming endangered. To determine whether vessels affect the behavior of resident killer whales, we conducted an observational study of Southern Residents in Haro Strait, Washington, from 2003-5 at two different sites along San Juan Island and an experimental study of Northern Residents in Johnstone Strait, British Columbia, in 2004 along West Cracroft Island. The studies focused on whale behaviors that may correlate with energy expenditure (directness of travel, changes in speed and direction, surface active behavior, and respiration rate) or energy acquisition (time spent foraging). Vessel number and proximity were treated as independent variables that may influence how vessels affect whale behavior.

Materials and Methods

Data collected include: theodolite tracks of focal individuals, along with observations of their behavior; and scan sampling of activity states of subgroups, along with counts of vessels at various distances from each subgroup. Theodolite tracks were summarized in terms of directness (distance traveled / distance progressed) and deviation (mean changes in direction) indices, and travel speed. Rates of respiration and display behaviors were also determined for each focal sample. Vessel number and distance were used as candidate explanatory variables for differences in track indices and other behavior, along with natural factors such as sex, age, pod membership, time of day, time of year, geographic location, tide and current. The natural factors were considered as variables which might confound the results. Vessel traffic was not controlled in work with Southern Residents. For Northern Residents, whale watching vessels took direction from the research team. This allowed obtaining 20 minutes of “control” data with no vessels present within 1 km, followed by 20 minutes of “treatment” data while whale watching vessels followed their normal operating practices. These experimental tracks were supplemented with observational data as were collected with Southern Residents.

Data on Southern Residents were collected from 28 July to 30 September 2003, 1 May to 31 August 2004, and 15 May to 31 July 2005 from two land-based observation sites on the west side of San Juan Island. Data on Northern Residents were collected from 1 July through 11 September 2004 from the south side of West Cracroft Island.

Theodolite data were collected using electronic theodolites connected to portable computers running Theoprog. Theodolite heights above mean lower low water were 99 and 72 m at the San Juan Island North and South sites, respectively, and 50 m above the mean water level on West Cracroft

Island. Tidal corrections to theodolite heights were based on tables for the Southern Resident study and measured for the Northern Resident study.

Focal individuals were drawn from all age and sex classes in the Southern Resident study (roughly half of the individuals in the population were tracked at least once). Experimental tracks of Northern Residents were limited almost exclusively to adult males, but opportunistic tracks were obtained of both males and females and adults and juveniles to assess the robustness of the experimental results.

At each surfacing, the time of taking a breath and additional behavioral events such as breaches, pectoral fin slaps, and fluke slaps were noted. The theodolite was used to determine positions of surfacings, as well as the positions of vessels surrounding the focal animal. Positions were used to calculate swimming speeds, directness of the path taken, magnitudes of changes in direction, and respiratory intervals. Monte Carlo simulations were used to estimate the statistical significance of the differences in the distributions of behavioral indices between the vessels within and no-vessels within 1000 m treatments. Generalized additive models (GAMs) were used to assess the importance of vessel and non-vessel factors in determining behavioral parameters and to allow treatment of vessel numbers and proximity as continuous rather than binary variables.

Activity states of subgroups and number and proximity of vessels were recorded using scan samples every 15 minutes. Individuals were assigned to subgroups based on a 100m chain rule. Probabilities for transitions among activity states were calculated and placed in a matrix. Log-linear analysis and AIC were used to assess the importance of vessel and non-vessel factors in determining transition probabilities.

Results: Southern Residents

Southern Residents spent a high percentage of time in proximity to vessels (Table 1). The maximum and mean numbers of vessels recorded are shown in Tables 2 and 3. Monte Carlo simulations suggested that travel became less direct, turns became sharper, dives became longer, and surface active behavior became more frequent in the presence of vessels in 2003-4 at both sites combined (Table 4). However, these results should be interpreted with caution as they may be confounded by factors other than vessel traffic. GAM analysis confirmed vessel effects contributed to significant results, although natural factors were important as well. E.g., paths became significantly more direct as the distance to the nearest vessel increased, K pod tended to travel along straighter paths than the other two pods, and directness increased with age (Table 5). Whales resting while waiting for other whales who took a less direct path would expend approximately half the energy of the other whales while waiting.

Whales were more likely to forage and less likely to travel at the South Site than the North Site (Figure 1). Whales spent significantly more time traveling and less time foraging when boats were within 400 m than when the closest vessel was farther away at the North Site (Figures 2-3). These figures show there are still effects to distances greater than the 100m in the current guidelines, but our scan sample data lack the resolution and sample size to determine where the effects become negligible. Pods did not differ significantly in the way vessel presence influenced activity state in 2003-4. Figure 5 shows the results of tests for effects of **Location**, **Year**, and the presence of **Boats** within 100 meters on the probability of transitions between **Preceding** and **Succeeding** activity states. The comparison between the top center condition and the top left condition shows the presence of **Boats** within 100 m significantly affected transition probabilities. The comparison between the top center and second from the top center show **Location** had a significant effect. The comparison between the top left and second from the top left conditions show the effects of **Boats** and **Location** did not confound each other. Other comparisons were statistically insignificant (year, interaction terms).

Results: Northern Residents

Responses of the 16 adult male killer whales tracked differed significantly between treatment levels (Wilcoxon's test $P=0.0148$). Swimming path became less direct when approached by few boats, and whales increased directness when approached by many boats (Figure 4). Pooling both treatments, reducing the comparison to presence vs. absence, would have masked these significant responses with strong statistical confidence (Wilcoxon's test $P>0.999$). Consistent with previous experiments, inter-breath interval, swimming speed, angle between successive dives, and rate of surface active behavior did not differ significantly. This apparent distinction between "few" and "many" boats, though, was supported by 140 opportunistic observations on 26 whales from the population of 216. Generalized Additive Models were used to control for effects of potentially confounding variables, and confirmed a non-linear relationship between the number of boats approaching within 1000m and a whale's swimming path directness, with an inflection point around 3 boats. One objective measure of this inflection point (refitting the GAMs, but placing knots manually from 1-16 boats, and minimizing AIC score) showed that whale behavior was best described as a non-linear function of boat number, with a knot placed at three boats, which suggests that the experimental track treatment level of few (1-3) versus many (>3) boats was appropriate. The avoidance response by northern resident killer whales differed when there were few versus many boats, and this result has important implications for the design and interpretation of other vessel impact assessments. Pooling all traffic conditions into categories of absence versus presence of boats could allow researchers to falsely conclude that there was no impact of boat traffic on whale behavior when, in fact, there was more than one response. We recommend that presence/absence analyses be supplemented by more detailed analyses when possible.

Conclusions:

Both vessel proximity and number appear to be factors affecting killer whale behavior in ways that may have energetic consequences, and such consequences would be important to the recovery of a food-limited population. Though small at any given moment, the cumulative effect of many vessels operating for many hours may be equivalent to removals in excess of Potential Biological Removal (PBR). Still, the effects reported here are too small for their elimination to be expected to allow downlisting the population. Our data indicate vessels did influence behavior at distances greater than 100m. This indicates there is management value to maintaining at least a 100m no-boat zone around whales. Additional data are required to assess whether other characteristics of vessels like type, size and speed influence whale behavior and to refine the relationship among vessel effects and number and distance. Other types of data (e.g., monitoring noise and prey capture events, and range-wide behavior budgets) are required to determine whether the behavioral effects documented here in fact have biologically significant energetic consequences.

Tables and Figures

Table 1. Percentage of scan samples with at least one vessel within the distances shown of the subgroup in Southern Residents in 2003-5. WOT = Whale Oriented Traffic, NOT = Not whale Oriented Traffic, and ANY refers to the presence of at least one of either type of vessel.

Year	Within 100 m			Within 400 m			Within 1000 m			Field of View
	ANY	WOT	NOT	ANY	WOT	NOT	ANY	WOT	NOT	ANY
2003	23.3	22.5	0.8	81.0	75.6	9.7	90.1	84.2	19.0	99.5
2004	28.8	26.7	4.9	56.7	51.6	11.2	75.7	69.4	21.5	98.5
2005	28.5	26.9	2.6	58.7	55.3	8.9	79.5	74.1	21.4	97.5

Table 2. Maximum number of vessels in scan samples within the distances shown of the subgroup in Southern Residents in 2003-5. WOT = Whale Oriented Traffic, NOT = Not whale Oriented Traffic, and ANY refers to the presence of at least one of either type of vessel.

Year	Within 100 m			Within 400 m			Within 1000 m			Field of View
	ANY	WOT	NOT	ANY	WOT	NOT	ANY	WOT	NOT	ANY
2003	7	7	1	19	19	5	36	23	15	109
2004	9	8	3	16	14	6	42	40	13	78
2005	17	17	5	31	31	14	35	35	14	57

Table 2. Mean number of vessels in scan samples within the distances shown of the subgroup in Southern Residents in 2003-5. WOT = Whale Oriented Traffic, NOT = Not whale Oriented Traffic, and ANY refers to the presence of at least one of either type of vessel.

Year	Within 100 m			Within 400 m			Within 1000 m			Field of View
	ANY	WOT	NOT	ANY	WOT	NOT	ANY	WOT	NOT	ANY
2003	0.35	0.34	0.01	3.08	2.91	0.17	7.09	6.54	0.55	42.26
2004	0.58	0.52	0.06	2.88	2.58	0.31	10.18	9.18	1.00	25.05
2005	0.57	0.52	0.04	4.14	3.79	0.35	10.48	9.55	0.93	19.38

Table 4. Behavioral indices for Southern Residents in the absence (distance to nearest vessel > 1000m) and presence of vessels in 2003-4.

Behavior	Value		Significance
	Without Boats N = 21	With Boats N = 100	
Directness Index	83.28	74.82	P < .001
Deviation Index	25.63	28.58	P < .01
Respiratory Interval (sec)	42.58	49.15	P < .001
Surface Active (/hr)	0.62	2.60	P < .001
Surface Speed (m/hr)	6638	6404	P > .1

Table 5. Effects of natural (year, pod membership and age) and vessel related variables on directness index.

Formula:

$$DI \sim \text{YEAR} + \text{POD} + s(\text{AGE}) + \text{PCA} + s(\text{BOATS}) - 1$$

Parametric coefficients:

	Estimate	std. err.	t ratio	Pr(> t)
YEAR	-0.00014202	1.553e-05	-9.145	2.9417e-16
PODJ	-0.040724	0.03777	-1.078	0.28262
PODK	0.090947	0.04594	1.98	0.049479
PODL	-0.050198	0.0372	-1.349	0.17916
PCA	3.4896e-05	1.596e-05	2.186	0.030307

Approximate significance of smooth terms:

	edf	chi.sq	p-value
s(AGE)	1.967	4.6553	0.098005
s(BOATS)	1	5.4682	0.020626

R-sq.(adj) = 0.0544 Deviance explained = 53.3%

GCV score = 0.060885 Scale est. = 0.058298 n = 164

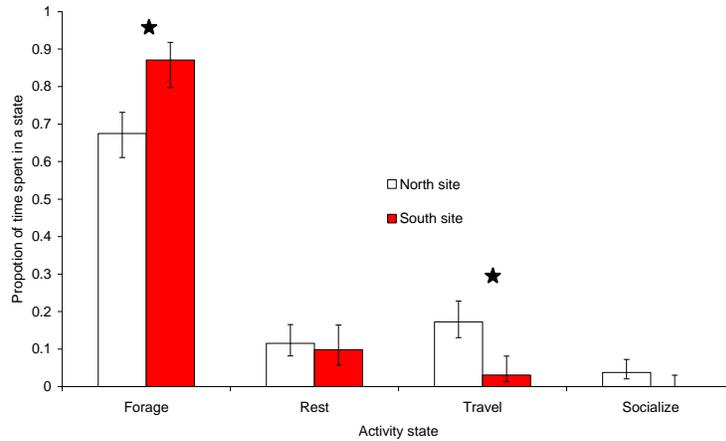


Figure 1. The proportion of time focal killer whales spent in each activity state (their activity budget) depending on the site sampled. Error bars are 95% confidence intervals. Black stars indicate differences that are significant at the 0.05 level in Southern Residents in 2003-4 for all vessel conditions combined.

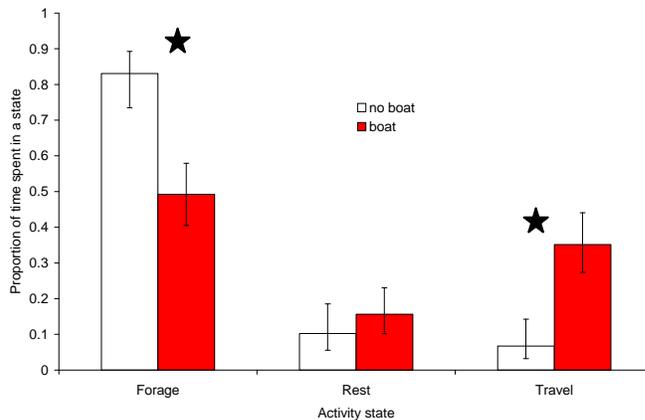


Figure 2. The proportion of time focal Southern Resident killer whales spent in each activity state (activity budget) depending on the presence of boat within 400m of them at the North site in 2003-4. Error bars are 95% confidence intervals. Black stars indicate differences that are significant at the 0.05 level.

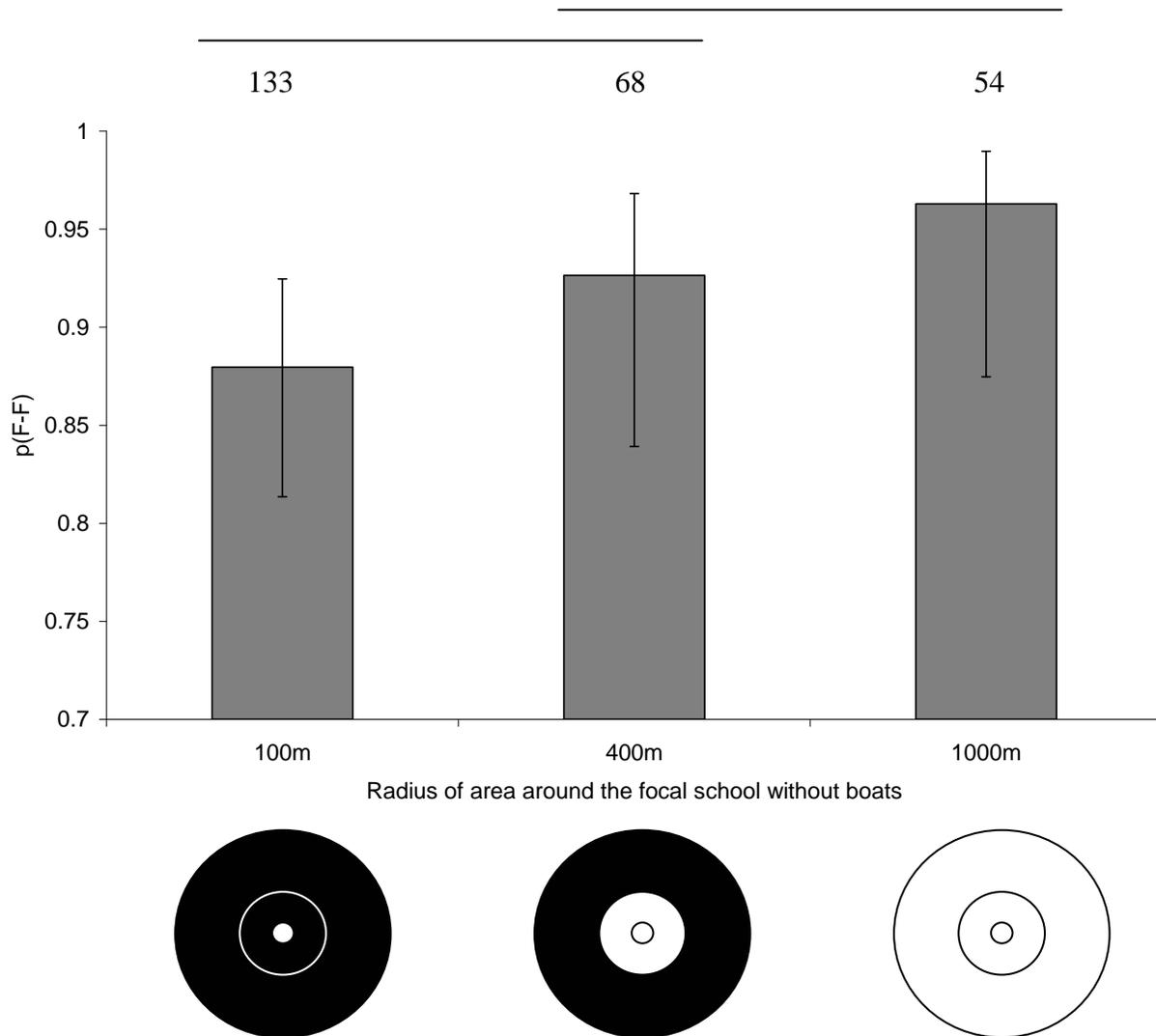


Figure 3. Likelihood that Southern Resident killer whales stay foraging when foraging, $p(F-F)$, when there is no boat present within 100m, 400m, or 1000m of the focal school at the North site in 2003-4. The diagram describes these three treatments in which the focal group is at the centre of the concentric doughnuts (100m, 400m, and 1000m radii) and the boat exposure is represented in black. Error bars are 95% confidence intervals. Whales are significantly more likely to stay foraging when foraging when there are no boats within 1000m as opposed to when there are no boats within 100m of them (as shown by the horizontal lines above the figure which link situations that could not be significantly distinguished from one another). The number of transitions observed is given above each bar.

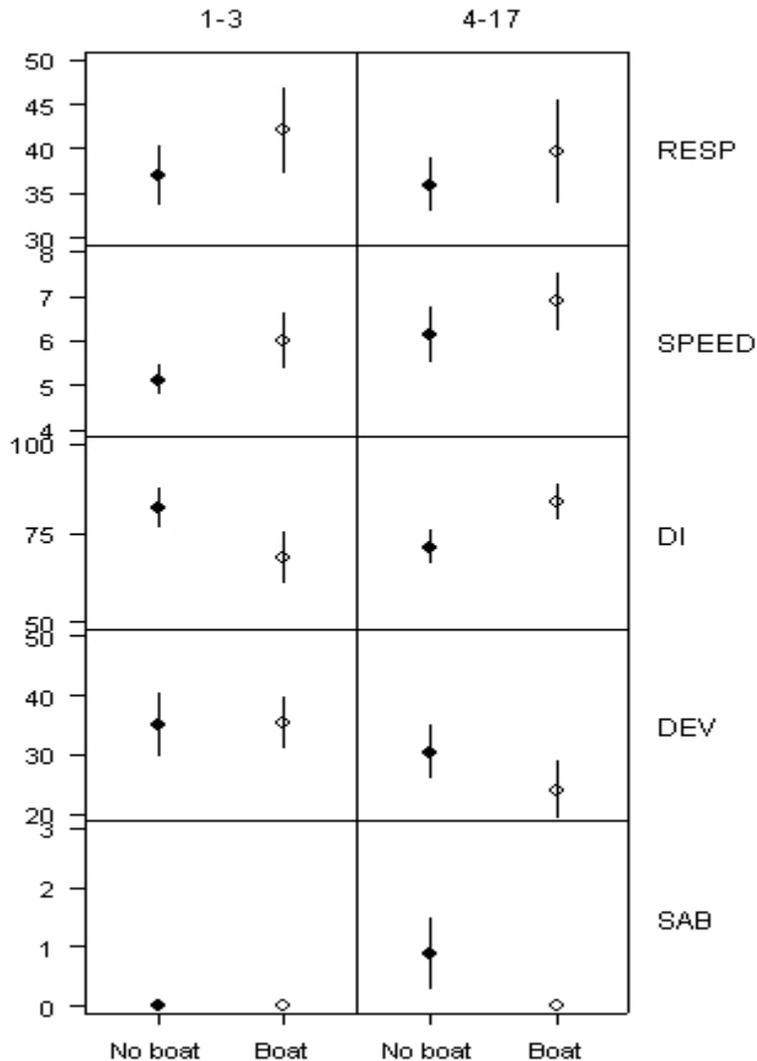
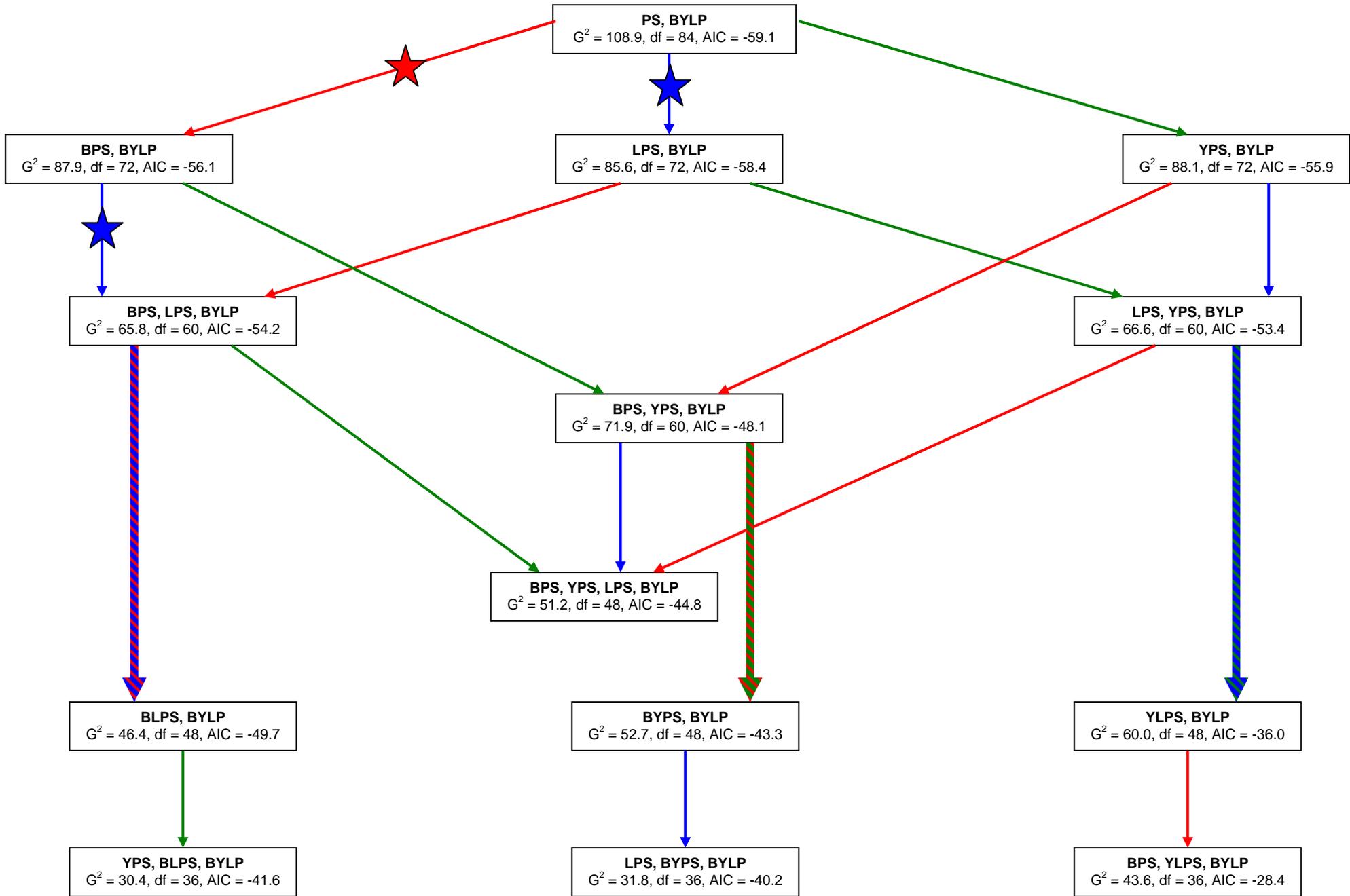


Figure 4. Behavioral responses (mean \pm se of the difference in whale behavior during control and treatment phases) of whales to experimental approach under two treatment conditions: approach to within 1000m by 1-3 boats, or by 4-17 boats. (a) RESP: Average time between respirations, or inter-breath interval(s); (b) SPEED: swimming speed (km/h); (c) DI: directness index (unitless); (d) DEV: deviation index (degrees); (e) SAB: surface active behavior (bouts/h). Note that all sample sizes are the same: 8 no-boat observations followed immediately by 8 treatment observations, for each of the two treatment levels, for a total of 32 observations.

Figure 5. (next page). Tests of boat presence within 100m (*B*), site (*L* for location to avoid confusion in abbreviations), and year of sampling (*Y*) effects on behavior state transitions (*PS*) using log-linear analyses for Southern Residents in 2003-4. Models and their respective goodness-of-fit G^2 statistics, degrees of freedom, and AIC values are shown in the boxes (adapted from Caswell 2001). Terms added are color-coded. Blue arrows represent the addition of a site effect (*LS*, *LPS* terms added to the previous model), red arrows represent the addition of a boat effect (*BS*, *BPS*), and green arrows represent the addition of a year effect (*YS*, *YPS*). To those terms corresponds an increment in G^2 and degrees of freedom, which are used to test for the significance of the term addition. Arrows are marked with a star when the term addition is significant ($p < 0.05$).



Behavioral Energetics of Southern Resident Killer Whales in the Presence of Vessels

Dawn P. Noren, NOAA Northwest Fisheries Science Center
Jennifer A. Marsh, University of Washington
Jim C. Ha, University of Washington

Introduction

Vessel disturbance has been identified as one of the potential risk factors to Southern Resident killer whales (Krahn *et al.* 2002). Since the late 1980s, these whales have experienced a significant increase in vessel-based whale watching (private and commercial) in the San Juan Islands (Baird 1999, Otis and Osborne 2001). In addition, other vessel activities, such as sports fishing, ferry and freighter traffic, have also increased in this region (Baird 1999).

Vessels may impact killer whales in several ways. Past studies have quantified impacts of vessels on Southern Resident killer whales by modeling the potential acoustic effects (Erbe 2002) or measuring short-term responses of individual whales to vessels (Kriete 2002). Northern Resident killer whales as well as other cetacean species can demonstrate horizontal and/or vertical avoidance behavior in response to boats (Nowacek *et al.* 2001, Williams *et al.* 2002, Lusseau 2003, Ng and Leung 2003). Agonistic behaviors, such as tail and pectoral fin slaps on the surface of the water, may also be displayed (Williams *et al.* 2002).

Although cetaceans tend to respond to boat traffic with stereotyped, short-term avoidance tactics, determining a link between short-term responses and a long-term effect is difficult. By collecting behavioral data that can be quantified in terms of energetic costs to individuals, we can begin to understand how short-term avoidance tactics may have long lasting effects. The purpose of this study is to determine if adult Southern Resident killer whales demonstrate avoidance behaviors in response to vessels off San Juan Island. Furthermore, if behavioral changes are demonstrated in the presence of vessels, energetic costs associated with these changes will be calculated to assess whether these changes are energetically significant.

Methods

Research was conducted in inshore waters near San Juan Islands, USA and off the east coast of Vancouver Island and the southern Gulf Islands, CA from late August through mid-September in 2003, and from early June through mid-September in 2004 and 2005. Data was collected from a 5.18-meter Bayliner power boat with a 90 hp two-stroke outboard motor in 2003 and 2004 and from a 7.92-meter Pacific aluminum skiff with a 225 hp four-stroke outboard motor in 2005. The research vessel departed from the west side of San Juan Island each morning between 0600 and 0800, weather permitting. Data were collected in Beaufort sea state ≤ 3 and while visibility conditions were adequate for locating and following killer whales. Each day, the Southern Resident killer whales were located by searching areas they frequent and by monitoring the VHF radio and pager system used by commercial whale-watchers. When killer whales were sighted, the boat approached to within approximately 100 m to allow for positive identification of individuals, and then retreated to a working distance of >100 m for the collection of data.

Data were collected continuously from individual adult male and female Southern Resident killer whales using a focal follow approach. Data were recorded using Event 3.0 Software created by J. Ha on a Palm III x e while the boat was motionless or traveled at a slow speed in parallel with the focal whale at a distance >100 m. During focal follows, one identified whale was followed for the collection of continuous behavioral and energetic data. For the duration of each focal follow,

the occurrence of every surface active behavior (e.g., spy hop, breach, tail slap, pec slap, porpoise, etc.), initiation of each dive, termination of each dive, and each breath taken by the individual was recorded. Swim speed through the water was estimated by paralleling the animal at a distance of >100 m and matching the speed of the boat with that of the whale. Prior to and during ten-minute intervals throughout the focal follow, GPS position, pod identification, focal group size, spatial (contact, tight, loose, spread) and formation (flank, linear, non-linear) arrangement of the focal group, and number of boats within approximately 1000 m of the whale were recorded. Boats were categorized as private, commercial whale watch, or kayak. Distances between the whale and specific vessels (closest, second closest, and the research vessel) were measured using a laser range finder at least every 10 minutes, or more frequently if vessel placement changed dramatically. Focal follows were conducted on several adult males and females each day. Individuals were only followed a second time in one day if several hours had elapsed since the first focal follow and other variables, such as focal group size and composition, vessel traffic, and geographic location had changed.

Due to the large sexual dimorphism in Southern Resident killer whales and the documented difference in responses of male and female Northern Resident killer whales to vessels, data from males and females were analyzed separately. Focal follows included in the analyses consisted of 15 – 80 continuous minutes during which the time of the initiation and end of every dive, every breath taken during each surface interval, and the occurrence of each surface active behavior (spy hop, porpoise, breach, tail slap, and pec slap) were recorded with certainty. Because this is an on-going study and additional data will be collected, only preliminary, less sophisticated analyses of the data have been conducted. General linear model analyses were used to assess the significance of relationships between whale behaviors (dive duration, post-dive surface duration, the ratio between post-dive surface duration and previous dive duration, number of surface active behaviors, swim speed, and respiration rates), pod size, and vessel parameters (number of vessels present and the closest distance measured between the focal individual and a vessel). Although interactions between several variables are potentially significant, these interactions were not addressed in these preliminary analyses. Additional more sophisticated analyses will be conducted in the future.

Results

In 2003, a pilot study was conducted for 8 days, resulting in 7 days of data collection. In 2004, 65 days in the field resulted in 27 days of data collection. With the use of the larger research vessel in 2005, the number of days in the field increased to 72 and resulted in 39 days of data collection. However, data from 2005 are still being prepared for analysis and are not presented here. Analyses of continuous focal follows ≥ 15 minutes from 2003 and 2004 (2003: $n=10$ male follows; 2004: $n=31$ male follows and $n=10$ female follows) are presented.

The preliminary analyses suggest that only a few of the recorded behaviors change in response to the number of vessels present. There were no significant relationships between the number of vessels within 1000 m and swim speed, respiration rate, or the total number and rate of surface active behaviors (all $P>0.05$). Furthermore, there were no relationships between the number of individuals in the focal group of killer whales and these behaviors (all $P>0.05$). However, the relationships between these behaviors and other characteristics of the focal group (e.g., pod identification and spatial and formation arrangement) still need to be assessed.

Relationships between the number of vessels present and diving parameters are more complex. The preliminary results suggest that these relationships may differ between males and females (Table 1; Figures 1, 2, and 3) In addition, the relationships may depend on whether there are few (0-15) or many (>15) vessels present (Table 1, Figures 1 and 2). Specifically, in males, surface duration and the ratio between surface duration and the previous dive duration significantly

decrease with increasing number of vessels present when the number of vessels within 1000 m are low (0-15), but when there are many (>15) vessels present, there is no significant relationship between these variables.

Table 1. Summary of statistics for relationships between average surface and dive durations and average number of vessels present during focal follows ≥ 15 minutes in duration.

Focal Follows	Surface Duration	Dive Duration	Surface : Dive Dur.
2003 Adult Males			
All (n=10)	Decrease (p=0.02)	No Change (p=0.47)	Decrease (p<0.001)
With 0-15 vessels (n=9)	Decrease (p=0.02)	No Change (p=0.99)	Decrease (p=0.03)
With >15 vessels (n=1)	-----	-----	-----
2004 Adult Males			
All (n=31)	No Change (p=0.31)	No Change (p=0.51)	No Change (p=0.51)
With 0-15 vessels (n=23)	Decrease (p=0.02)	Decrease (p=0.021)	Decrease (p=0.01)
With >15 vessels (n=8)	No Change (p=0.10)	No Change (p=0.14)	No Change (p=0.08)
2004 Adult Females			
All (n=10)	No Change (p=0.45)	No Change (p=0.83)	No Change (p=0.29)
<i>All had 0-15 vessels</i>			

In addition there was no relationship between the closest vessel distance and the total number or rate of surface active behaviors (Fig. 4, all $P>0.05$). Yet, on two occasions females repeatedly performed tail slaps when approached within <100 m by a small motorized boat.

Discussion

Focal follows can provide detailed information to assess some behavioral responses to vessel disturbance in Southern Resident killer whales. These preliminary results suggest that the relationships between the number of vessels present within 1000 m and diving parameters are not simple and may differ between males and females. For the limited sample of data collected from females, there appear to be no relationships between the number of vessels present and surface and dive durations. In contrast, the number of vessels present within 1000 m has a slight but statistically significant effect on surface duration and the ratio of surface duration to previous dive duration in males. However, these relationships are complex and appear to change when there are few (≤ 15) versus several (>15) vessels present. Additional data and more sophisticated analyses are needed to assess the effect of the number of vessels present on surface duration, dive duration, and the ratio of surface duration to previous dive duration when few and many vessels are present. Furthermore, additional data from females are needed to better assess these relationships in female killer whales.

Furthermore, diving parameters and the occurrence of surface active behaviors do not appear to be related to the closest distance between the focal whale and a vessel. However, observations during the study indicate that when vessels approach whales closely while under power, surface active behaviors such as tail slaps may occur. Thus, a more focused study on the mode of operation of vessels in close proximity (≤ 100 m) of Southern Resident killer whales is warranted to better understand what variables may elicit more energetically expensive responses.

Finally, once the behavioral responses of the killer whales are better understood, metabolic rates for adult female and male Southern Resident killer whales will be estimated using physiological and behavioral data collected during this study and metabolic and respiration rate data collected by Kriete (1995) and Noren (unpublished data, 2005-2006). Estimates will be used to determine whether there are increased metabolic costs to killer whales when vessels are present.

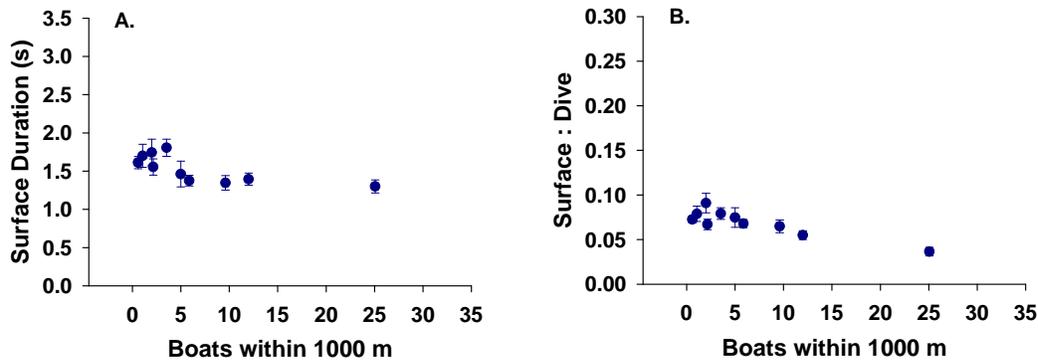


Figure 1. Relationship between the number of vessels within 1000 m of the focal whale and the A) surface duration and B) ratio of surface duration to previous dive duration during follows of individual adult male SRKWs in 2003. Mean values for each focal follow ($n=10$) are presented with ± 1 SE bars. Statistics for the relationships are reported in Table 1.

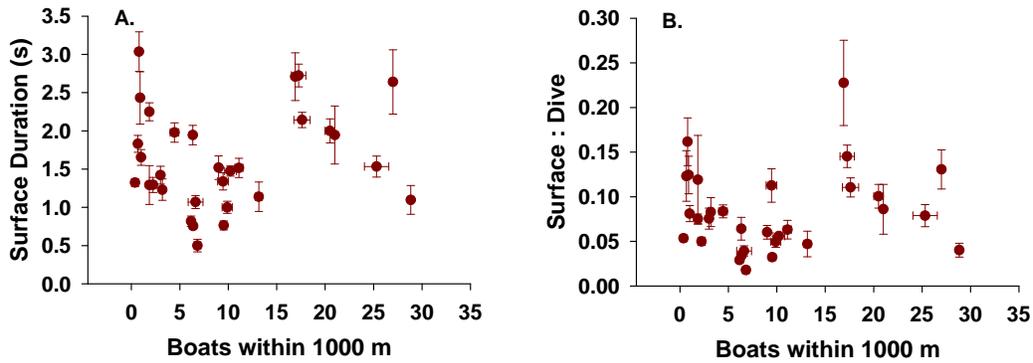


Figure 2. Relationship between the number of vessels within 1000 m of the focal whale and the A) surface duration and B) ratio of surface duration to previous dive duration during follows of individual adult male SRKWs in 2004. Mean values for each focal follow ($n=31$) are presented with ± 1 SE bar. Statistics for the relationships are reported in Table 1.

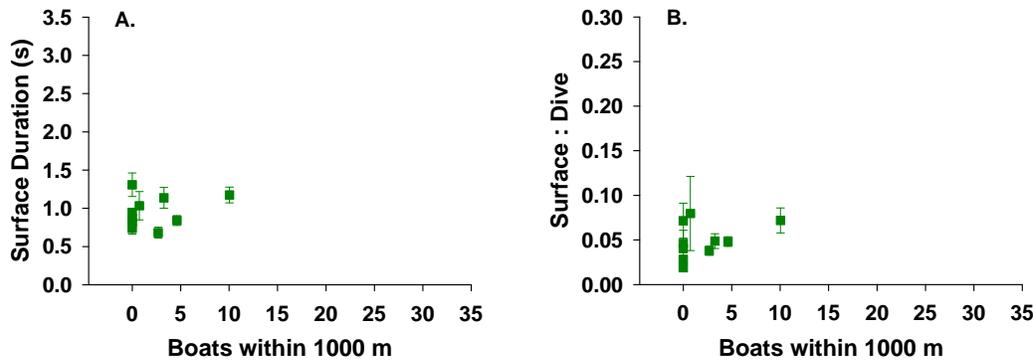


Figure 3. Relationship between the number of vessels within 1000 m of the focal whale and the A) surface duration and B) ratio of surface duration to previous dive duration during follows of individual adult female SRKWs in 2004. Mean values for each focal follow ($n=10$) are presented with ± 1 SE bar. Statistics for the relationships are reported in Table 1.

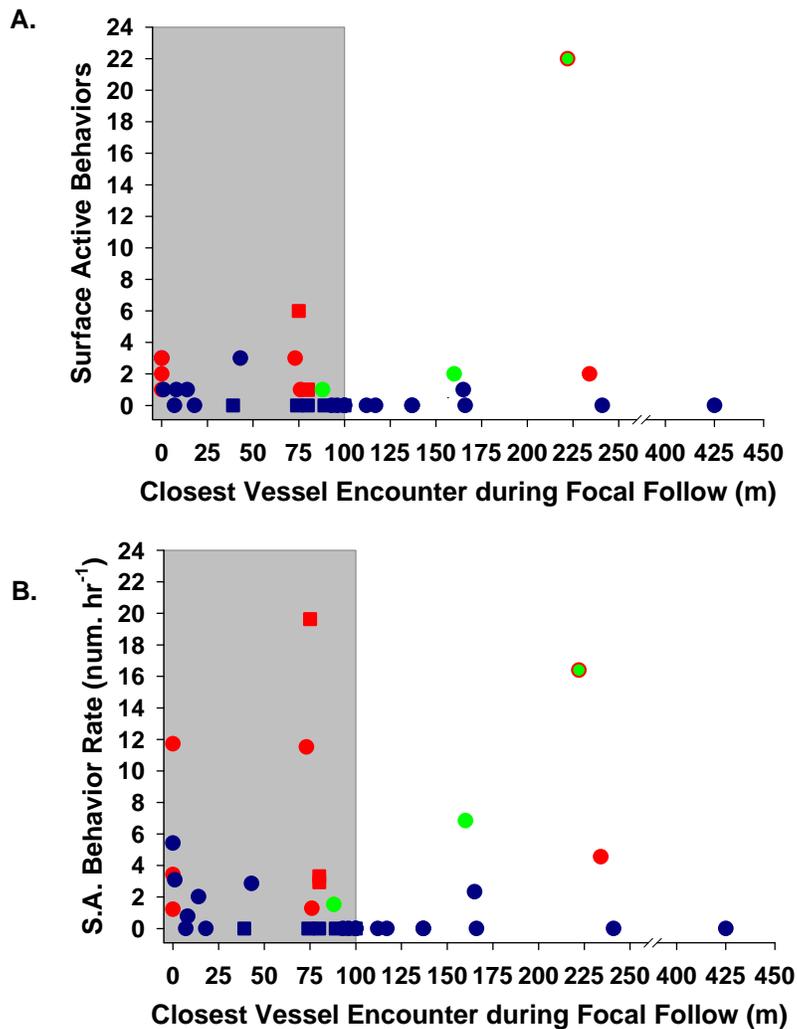


Figure 4. Relationship between the closest vessel encounter and the A) total number of surface active behaviors that occurred and B) calculated rate of surface active behaviors per hour for each individual focal follow from adult SRKW in 2004. The closest vessel encounter is defined as the closest distance that was measured between a vessel (moving and/or stationary) and the focal animal during the entire focal follow. The gray shaded region (0-100 m) represents the “NO-GO ZONE” for vessel whale watching guidelines. Data from males (●) and females (■) are presented. Blue Symbols designate that the closest measured vessel encounter and the surface active behavior(s) did not occur simultaneously. Red Symbols designate that the closest measured vessel encounter and the surface active behavior(s) occurred simultaneously. Green Symbols designate that the surface active behavior(s) occurred when the only vessel present was the research vessel and that the distance between the focal whale and the research vessel was >215 m. The Green Symbol with Red Outline designates that the surface active behaviors occurred when the research vessel was the closest and only vessel near the focal whale and was at a distance >215 m from the whale.

Social Behavior of Southern Resident Killer Whales (*Orcinus orca*)

Jennifer A. Marsh and Jim C. Ha
University of Washington

Introduction

Inshore, or resident, killer whales are one of three distinct populations of killer whales in the Pacific Northwest. Resident killer whales are characterized by large group sizes and a piscivorous diet. They are highly social mammals that live in stable subgroups of several related females and their young. Offspring of both sexes remain in their natal group for life, and groups are organized along matrilineal lines. Resident society members can be assigned to a series of social units according to maternal genealogy. The bonds within social units get progressively weaker, as relatedness between individuals decreases incrementally for subpods, pods, clans, and communities.

The focus of the research presented here is the social behavior of southern resident killer whales. Since these whales are highly cognitive and display an extremely cohesive social structure, fluctuations in social behavior among killer whales have important implications for the benefits of pod cohesion. If the relative amount of social behavior wanes over time due to ecological variables, killer whales may lose the group-living benefits such as alloparental care, group foraging, sensory integration, and cultural trait transmission.

As a measure of social behavior, percussive behaviors were examined in this study since they are considered communicative and may convey information to conspecifics. Synchronous surfacing was also examined in this study, as surfacing together represents a social bond between two animals and has previously been used to define the strength of affiliation among conspecifics in bottlenose dolphins. Our specific hypothesis was that the number of commercial and or private boats would affect the rates of percussive and synchronous surfacing behavior in southern resident killer whales. Additionally, the effect of other variables on social behavior were also examined, such as killer whale group size, pod, time of day, and year.

Methods

Research was conducted in the inshore waters of Washington State. Data were collected between June and September each year over the three years, 2003-2005. A general authorization for scientific research has been granted for this project by the National Marine Fisheries Service (File No. 965-1632).

Data collection procedure

For 2003 and 2004, a 17-foot Bayliner power boat with a 90 hp two-stroke outboard motor was used to collect data in the study area. For 2005, data were collected from the R/V Noctiluca, a 7.92 meter Pacific power boat with a 225 hp four-stroke outboard motor provided by NOAA NMFS NWFSC. The research vessel departed from Wescott Bay or Snug Harbor on San Juan Island each morning at approximately 0600, weather permitting, and data were collected in Haro Strait, Boundary Pass, Rosario Strait, and waters adjacent to San Juan Islands. All data collection was conducted in Beaufort sea state ≤ 3 and under visibility conditions adequate for locating and following killer whales. The general research method was to locate killer whales by boat each morning by searching frequent foraging locales. When whales were located, the boat approached to within approximately 100m to allow for positive identification of individuals by sight, and then retreated to > 100 m for subsequent behavioral observations. Trained undergraduates from the University of Washington assisted in behavioral data collection during 10 minute periods.

Data were collected using Event 3.0 software created by J. Ha on a Palm III μ . Prior to each 10 minute period, pod identification, GPS position, spatial arrangement, group size, and number of boats present was recorded. Group size was defined as the total number of killer whales in the visual range of the observer. Boats were categorized as private vessel, commercial whale watch vessel, or kayak within visual range of the researcher, and within approximately 0.5 miles of the

whales. Spatial arrangement categories for whales included contact, tight, loose, and spread. All occurrence sampling was performed for specific behaviors including breach, half breach, tail slap, inverted tail slap, pectoral fin slap, spy hop, physical contact, cartwheel, and synchronous surfacing.

Statistical analysis

All hypotheses were tested using appropriate statistical methods. When pertinent, assumptions of statistical tests were checked, including normality (skewness and kurtosis) as well as homoscedasticity (Bartlett's Test for Homogeneity of Variance). When necessary, appropriate transformations were performed and assumptions rechecked before performing parametric statistics. Nonparametric statistical methods were used when necessary. All statistical analyses were performed using Systat 7, and a probability of 0.05 was used as the criterion for rejection of the null hypotheses. Percussive and synchronous surfacing behaviors were divided by killer whale group size before plotting in order to address group size effects on results. While interactions were not specifically addressed in this preliminary analysis, there is clearly potential for significant interactions among the variables examined. Final analyses will address this issue in depth.

Results

Sampling

Data collection began June 2, 2003 and continued through September 17, 2003; June 1 – September 17, 2004; June 1 – August 31, 2005. For 2003, data were successfully collected on 33/69 field days, resulting in 333 10 minute samples, and a mean sampling rate of 10.09 ± 5.49 samples/day. In 2004, data were collected on 33/72 field days, resulting in 307 samples, and a mean sampling rate of 9.30 ± 6.62 samples/day. For 2005, data were collected on 28/53 field days, resulting in 362 samples, and a mean sampling rate of 12.93 ± 6.16 samples/day.

Synchronous Surfacing

Group size was found to have a significant inverse relationship with synchronous surfacing ($F_{1,581} = 37.579$, $P < 0.001$). Pod was also found to have a significant relationship with synchronous surfacing ($F_{2,581} = 39.413$, $P < 0.001$; Figure 1), as did year ($F_{2,581} = 93.183$, $P < 0.001$; Figure 2). Additionally, commercial boat number was found to have a significant inverse relationship with synchronous surfacing ($F_{1,581} = 3.982$, $P = 0.046$; Figure 3). Conversely, time of day was found to have a significant direct relationship with synchronous surfacing ($F_{2,581} = 14.416$, $P < 0.001$; Figure 4). No clear pattern of significance was found between synchronous surfacing and private boat number.

Percussive Behavior

Group size and percussive behavior showed a significant inverse relationship ($F_{1,581} = 3.958$, $P = 0.047$). Pod and percussive behavior also showed a significant relationship ($F_{2,581} = 3.308$, $P = 0.037$; Figure 5). A significant relationship was also found between percussive behavior and year ($F_{2,581} = 32.587$, $P < 0.001$; Figure 6). No significant relationship was found between percussive behavior and time of day, commercial boat, or private boat number.

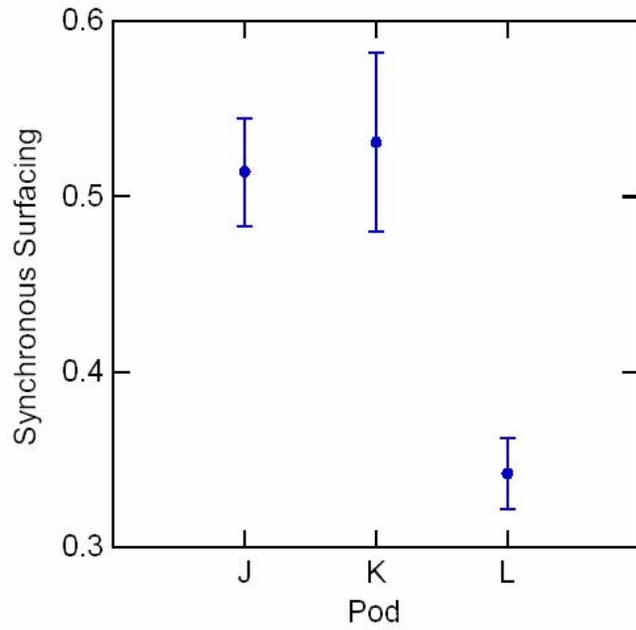
Discussion

Synchronous surfacing appeared to be affected by several variables, including group size, commercial boat number, time of day, year, and pod. These results will be examined further in the near future as more sophisticated statistical methods are employed to detail interactions and possible confounds.

Percussive behavior was seen to vary significantly with group size, pod and year. As percussive behaviors are considered to be communicative, it is apparent that these behaviors may be a tool these animals use to inform conspecifics about their environment, including who is present, as well as how many other whales are found nearby.

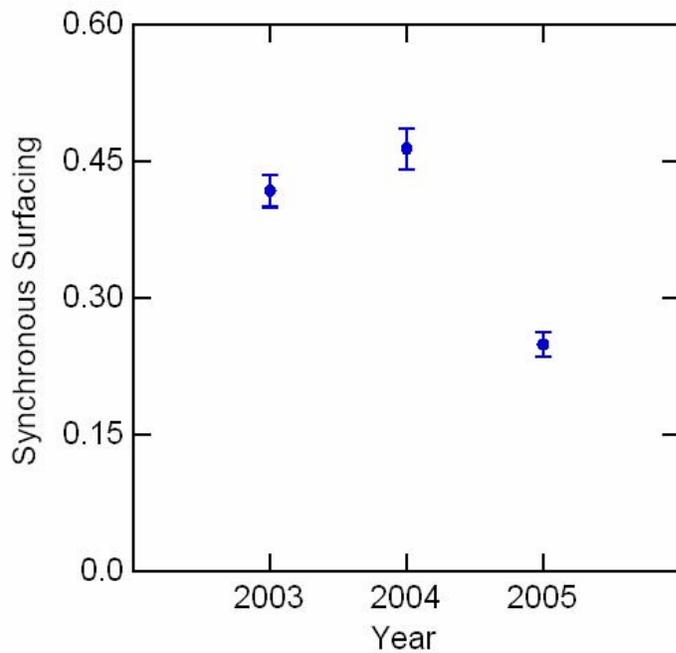
Further data will be analyzed using a combination of principal components analysis and general linear modeling to examine complex interactions among predictor variables. Power

analysis will also be conducted. We will ultimately propose a model of behavior which will include



possible influences on social behavior, only a portion of which are discussed here.

Figure 1. Synchronous surfacing per whale by pod. Bars represent standard error.



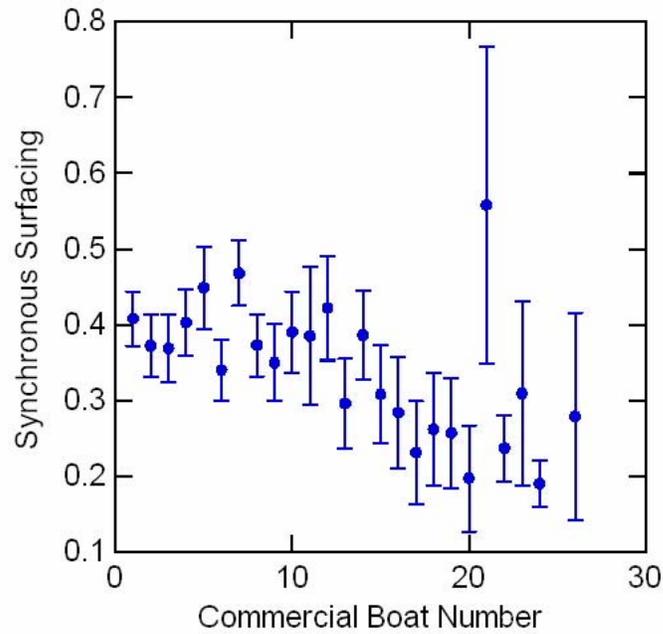


Figure 2. Synchronous surfacing per whale by year. Bars represent standard error.

Figure 3. Synchronous surfacing per whale by commercial boat number. Bars represent standard error.

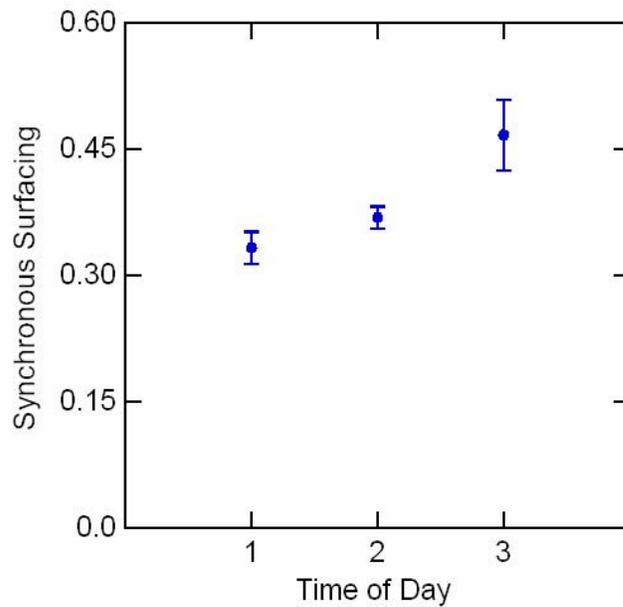


Figure 4. Synchronous surfacing per whale by time of day. Time of day categories include 1: 0600-1000; 2:1000-1400; 3:1400-1800. Bars represent standard error.

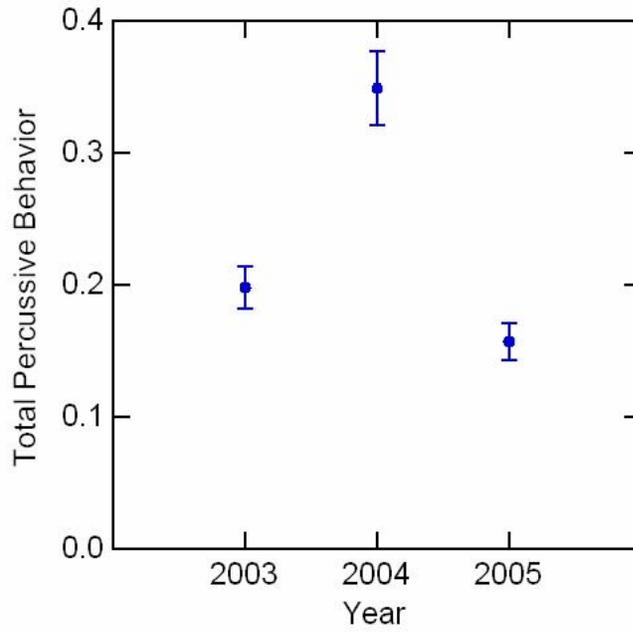
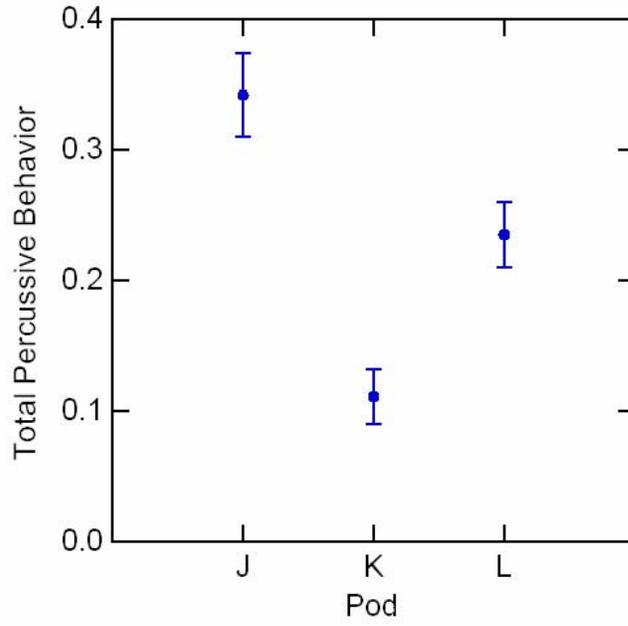


Figure 5. Total percussive behavior per whale by pod. Bars represent standard error.
Figure 6. Total percussive behavior per whale by year. Bars represent standard error.

Acoustic Creatures in Noisy Environments: Effects of Sound on Marine Wildlife

Brandon L. Southall, NOAA Ocean Acoustics Program, National Marine Fisheries Service, Office of Science and Technology

Sound is a dominant feature of the marine environment and the primary modality for conveying information over any appreciable space under water. Most if not all marine animals rely to some extent on sound for a wide range of biological functions, including: communication, navigation, foraging, and predator detection. Natural physical processes produce sounds that can travel great distances underwater and dominate local ambient noise in certain frequency bands. Human activities can also introduce sound into the marine environment, either incidental to industrial activities or intentionally in the context of navigation or remote sensing.

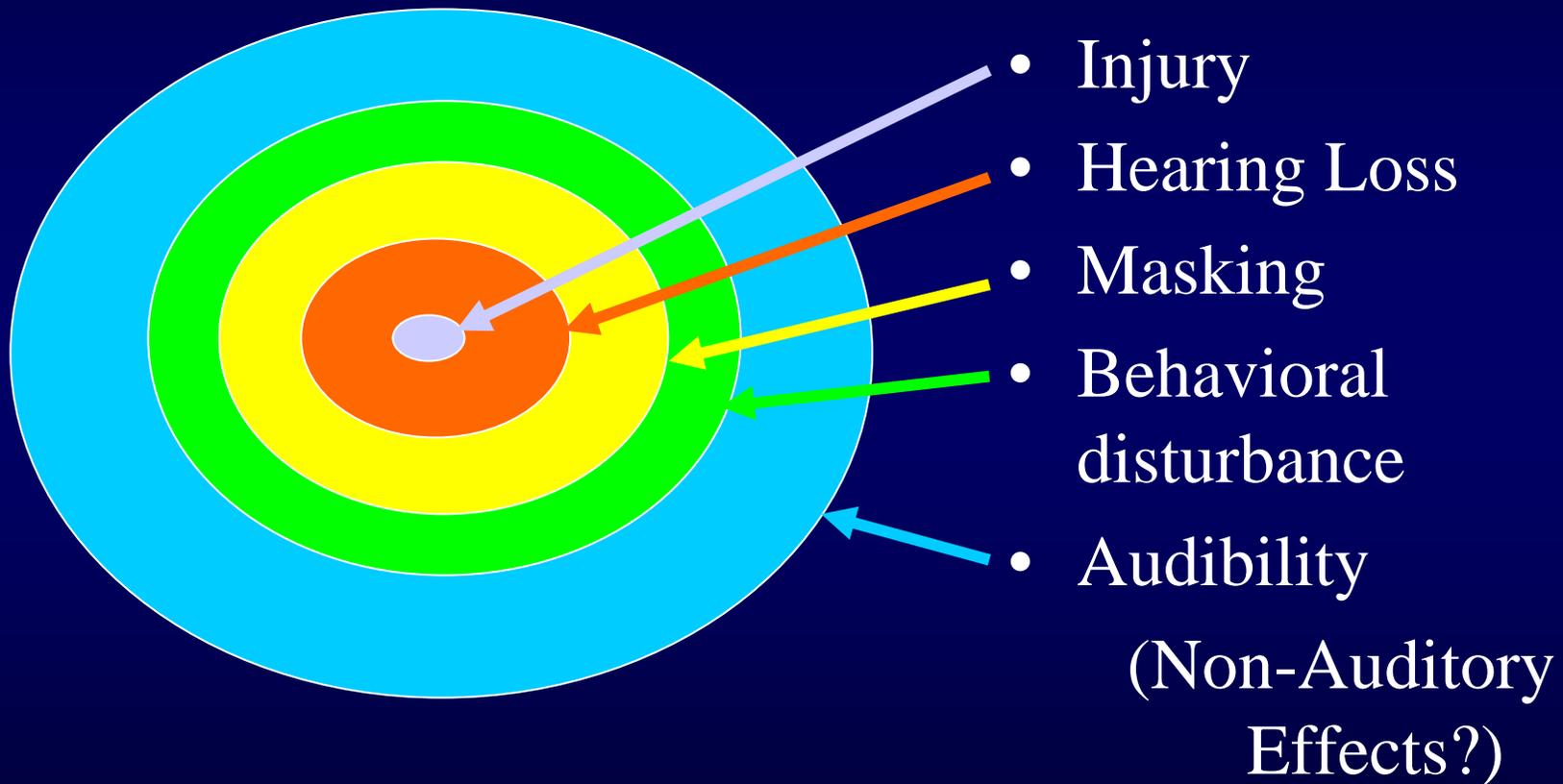
The ocean is both a relatively noisy place, as a result of these varied sources, and a highly complex acoustic environment. The acoustic characteristics received from a particular source at some distance can be strongly affected by a range of factors other than simply sound level and frequency, including: the presence of reflective surfaces; complex bathymetry; variations in temperature, pressure, and salinity; and other factors. Sound sources, environmental factors, and characteristics of receiving individuals exposed to a specific sound are all critical aspects in assessing how the receiver may react.

Sounds may have a wide range of possible effects on animals, including: simple detection; behavioral responses of variable magnitude; auditory masking (noise interference); temporary or permanent reductions in hearing sensitivity; and, in extreme cases, death (Fig. 1). There are numerous critical features in determining which among these effects an animal exposed to a particular sound may experience. In addition to the received sound characteristics, these include physiological, behavioral, motivational, and other features of the individual in question. One key feature is the relationship between frequencies of interest and those of the species in question (Fig. 2). For a number of reasons, among them recent concerns regarding the potential for anthropogenic sound to affect marine life, there has been a considerable amount of research conducted to understand the relevant considerations more fully.

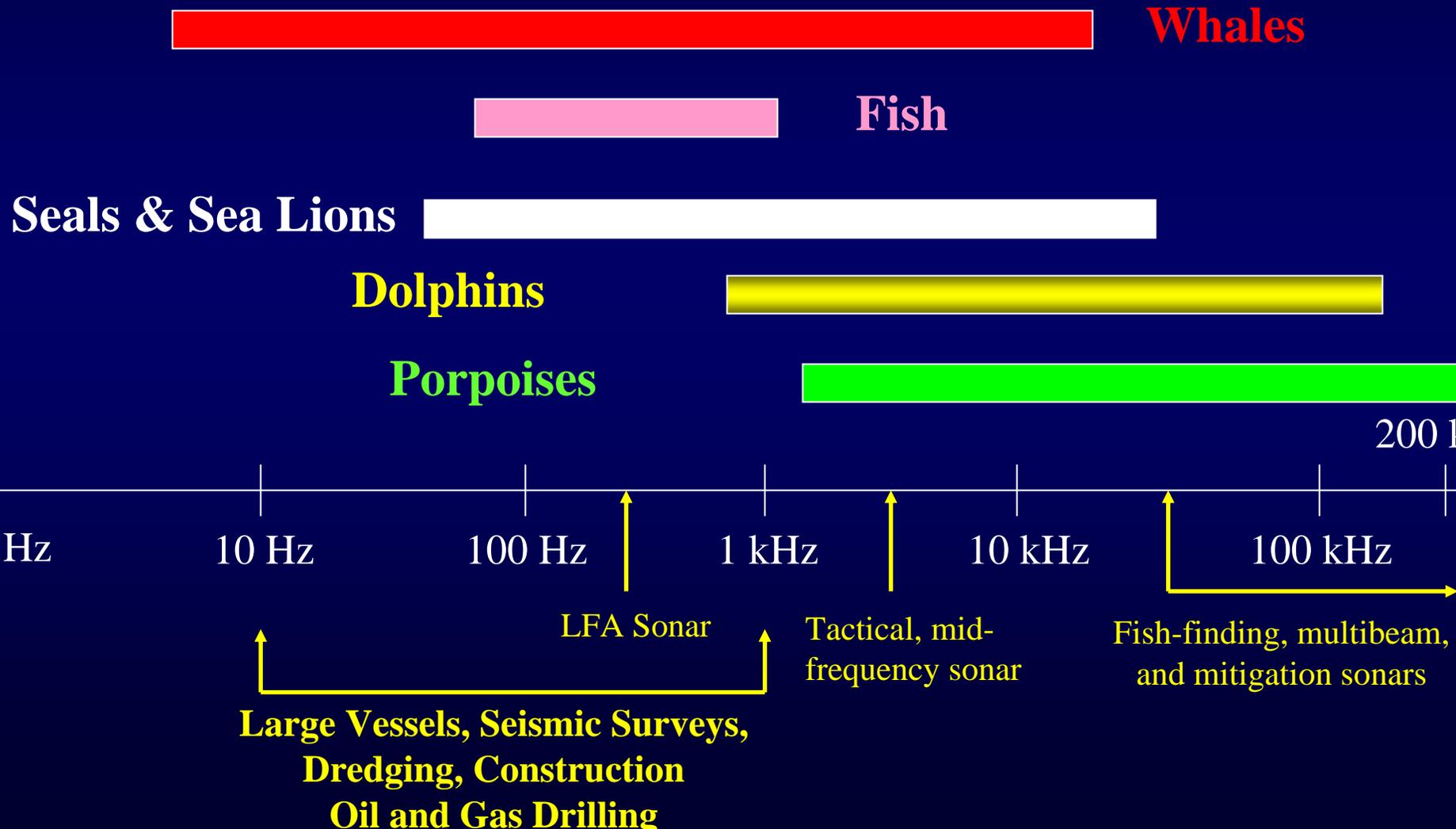
Field measurements of sounds produced by animals in their natural environment and behavioral responses of individuals provide critical information on natural communication systems. Laboratory investigations elucidate basic sensory capabilities and the extent to which they may be affected by noise in various ways. Passive acoustic sensing consequently can provide critical, detailed information on: presence, activity, and in some cases abundance of marine species; natural forces related to the assessment of climatological and geological processes; and the extent to which human sound sources are affecting the acoustic habitats of certain marine environments. It can be used further as a means of assessing ambient noise budgets over space and time, which are the baseline against which specific effects of explicit sound sources must be considered. Active acoustic deployments can provide a powerful means of investigating physical features of marine environments and detecting specified marine species.

This presentation will consider a number of basic acoustic principles and relevant considerations, including advances in research that bear on our ability to gauge the relative importance of human sound introduction among other known anthropogenic stressors. Additionally, data regarding sound production, use, and detection in killer whales will be discussed briefly, in order to set the stage for presentations on important and fascinating targeted research on issues related to killer whale acoustics.

Spatial Zones of Noise Influence



Frequency Relationships Between Marine Animal Sounds and Human Noise Sources



Ambient noise in the Haro Strait from whale watching and commercial vessels

John Hildebrand

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This study provides baseline data to characterize the noise exposure that southern resident killer whales may experience within the Haro Strait. Ambient noise data were collected, providing source levels from a representative sample of major vessel classes under typical operating conditions. Data for this project were collected during shipboard operations on October 11-12, 2003 and May 28-31, 2004 in the Haro Strait. The locations for data collection were along the west side of San Juan Island, near Limekiln Lighthouse. A vertical hydrophone array of 3-5 elements was used during this project to collect ambient noise data. The hydrophone array was calibrated at the US Navy's TRANSDEC facility.

Data on vessel noise were collected both opportunistically (without control of the vessel operation) and under a controlled data collection protocol (with the cooperation of the vessel). All large commercial vessel data were collected opportunistically. Small vessel data were collected both opportunistically and under the controlled protocol.

Data were collected opportunistically from the following classes of vessels. The numbers of vessels sampled for each class are given in parentheses.

- **Commercial** – tug (4), dry bulk (5), container (5), and ore carrier (1)
- **Private** – sport fish (6), yacht (12), cabin cruiser (9), whaler (2), RHIB (3), Puget trawler (2), sailboat (2)
- **Whale Watch** – catamaran (1), yacht (2), jet RHIB (1), sport fish (3), RHIB (3), Puget trawler (1), Al hull (1), TBD (7)

The protocol for controlled noise data collection from individual whale watching vessels was as follows:

1. cruising at 10 knots pass the hydrophones at an approximate range of 200 m
2. cruising at 4 knots pass the hydrophones at an approximate range of 200 m
3. Idle for 1 minute at approximately 350 m range, then proceed to 200 m range
4. Idle for 1 minute at 200 m range
5. Accelerate to cruising speed
6. Terminate test at range of approximately 500 m

Data were collected from the following fifteen vessels using the above protocol: *Orca Spirit*, *Explorathor*, *Annie Mae*, *Olympus*, *Starlet*, *ORCA*, *Bon Accord*, *Mercury*, *K-Ko*, *Ziphid*, *NOVA*, *Countess*, *Supercat*, *SoundWatch*, *Kitti Wake*.



Figure 1. Commercial container ship, Hanjin Marseilles (51,000 Gross Tons) recorded in the Haro Strait on May 30, 2004.



Figure 2. Whale watch vessel, Annie Mae (sport fishing type), recorded under a controlled protocol May 30, 2004.



Figure 3. Whale watch vessel, Explorathor (jet RHIB type), recorded under a controlled protocol May 31, 2004.



Figure 4. Whale watch vessel, Olympus (yacht type), recorded under a controlled protocol May 31, 2004.

Average levels and power spectra of ambient sound in the habitat of southern resident orcas

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Abstract

We used a pair of hydrophones to monitor sound pressure levels (SPL, dB re 1 μ Pa) and frequency spectra of ambient sounds in Haro Strait during 18 months (April 2004 - November 2005). Half-hour average SPL in the broad frequency band 0.1-15 kHz ranges from ~95-130 dB. The overall average SPL is ~115 dB; the SPL is ~2 dB during summer days. The broadband ambient sound field in Haro Strait is dominated by noise from large vessels (commercial ships) that increase SPL ~20-25 dB for 10-30 minute periods (Figure 1) and cause the long-term 20 dB difference between minimum and average SPL. Smaller vessels (motor boats) increase SPL about as much as ships (~15-20 dB) but do so for short periods of time (Figure 2). Nevertheless, boats add ~2 dB to the average SPL during mid-afternoon hours in the summer and increase spectrum levels in the 10-20 kHz band. In this frequency band, spectrum levels at mid-day during a summer day are more than 10 dB re 1 μ Pa/Hz^{1/2} higher than during a summer night or a day or night in the winter. (Figures 3 & 4)

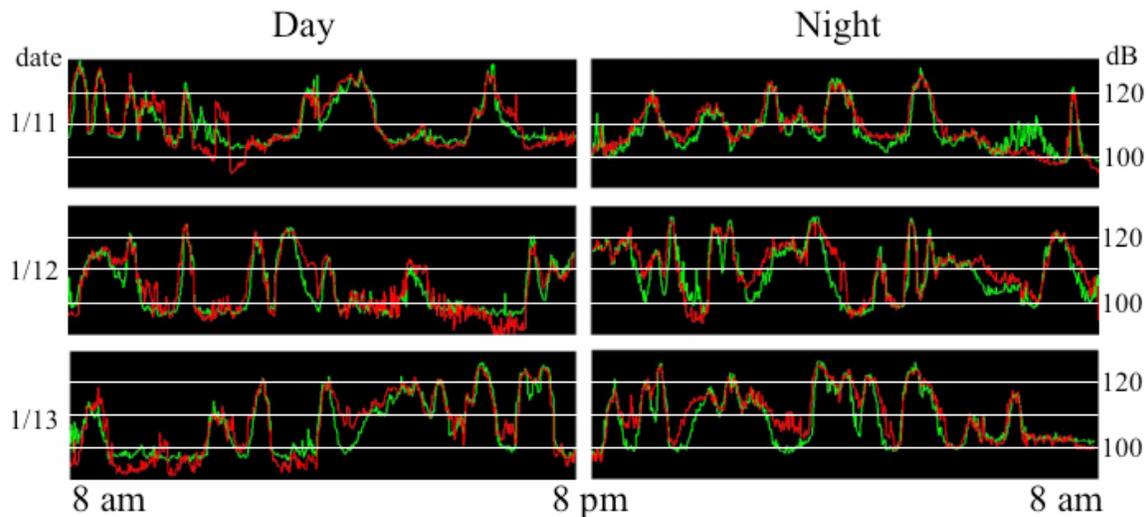


Figure 1: Three consecutive winter days (Jan 11-14, 2005) of ambient sound amplitude received at two widely spaced hydrophones in the Orcasound array. One hydrophone signal is in red; the other is in green. The two plots in each row include data from 8:00-24:00 of the date that labels the row (at left), as well as the first eight hours (0:00-8:00) of the subsequent day.

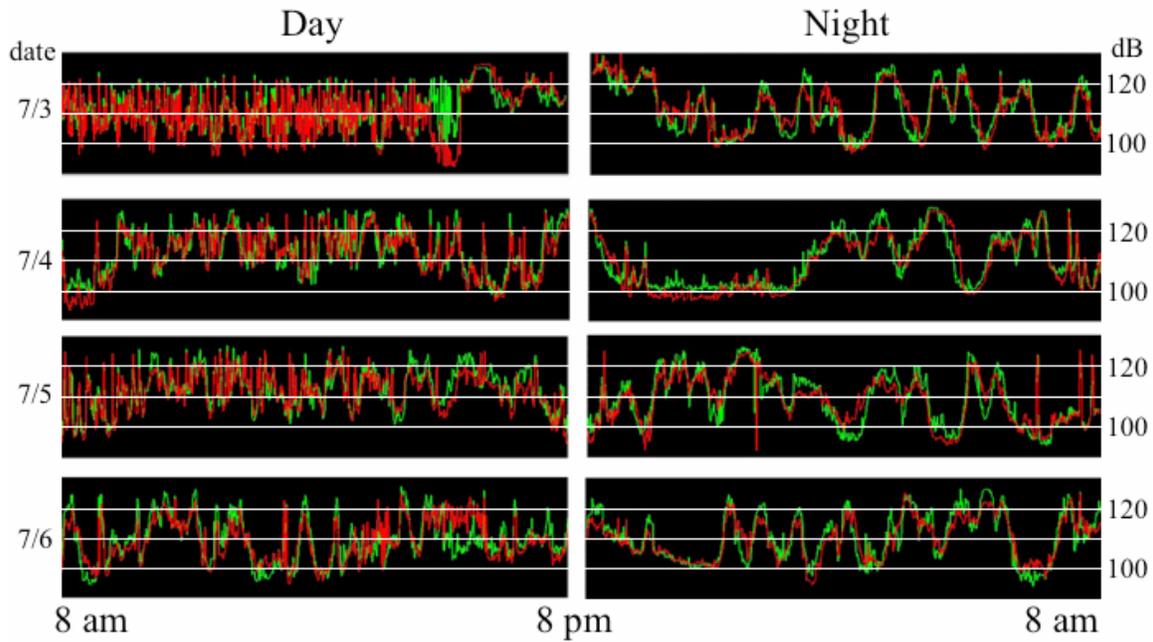


Figure 2: Four consecutive summer days of ambient sound amplitude (dB) received at the Orcasound array. This period includes the Fourth of July weekend, 2004). Colors and time axis are the same as in Figure 1.

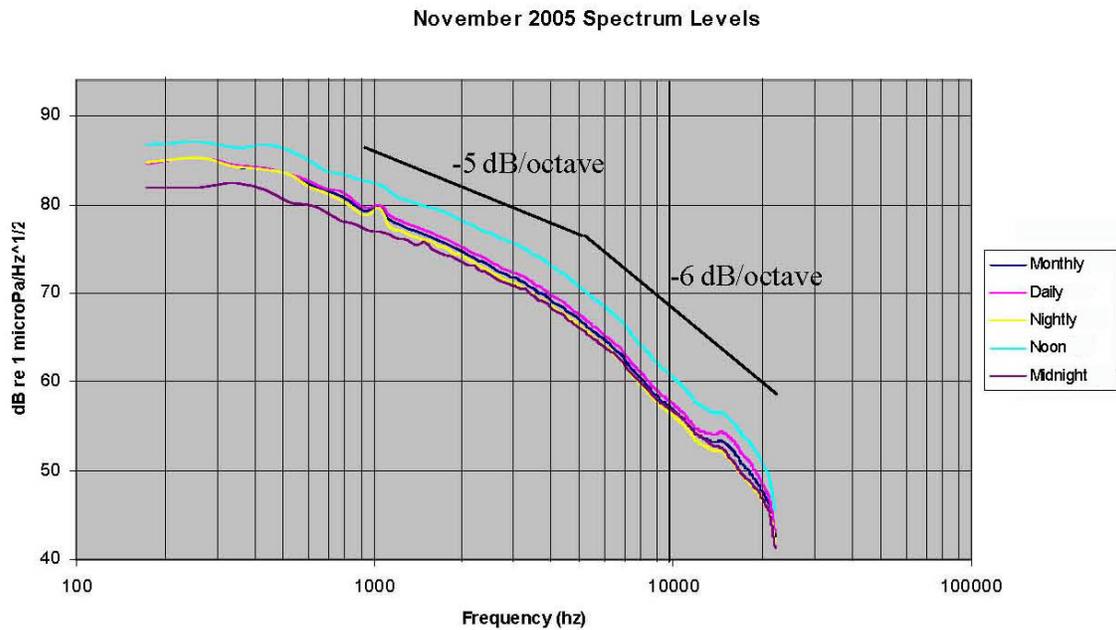


Figure 3 Spectrum levels for November, 2005.

July 2005 Spectrum Levels

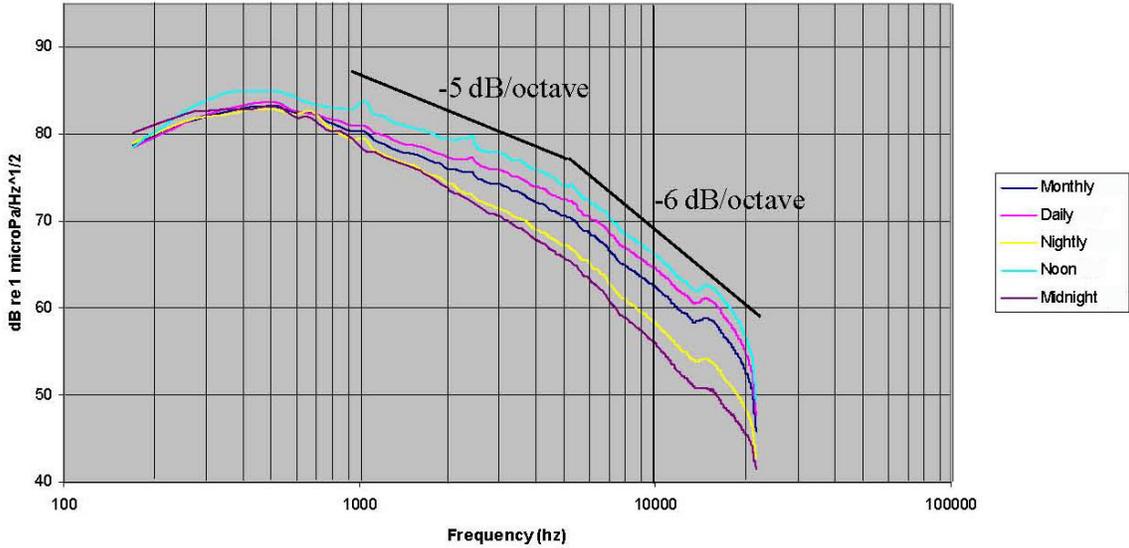


Figure 4: Spectrum levels for July, 2005

The Acoustic Environment of Haro Strait: A data-model comparison of shipping traffic noise.

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Understanding the acoustic environment of the Southern Resident killer whales is a prerequisite to predicting the impact of anthropogenic sounds on an individual orca's ability to forage and communicate in the presence of background noise sources. Several factors lend to make the acoustic environment of the Haro Strait sufficiently complicated. Of primary interest is the highly variable bathymetry in the Strait, characterized by a relatively deep canyon with a very steep wall at the western coast of San Juan Island, as illustrated in Figure 1. The sediment properties have large variations as well. The steep walls about the west side of San Juan Island are exposed rock with silt and sand material lie on the bottom of the channel. Because of the strong and variable currents, the thickness of this sediment layer will vary temporally and spatially. The sea surface varies with space and time in accordance with wind stress. All of these factors contribute to the unique and complex nature of this acoustic environment. Indeed, the only benign factor is the sound speed velocity in the water column; to first order it is constant. It should also be noted that these factors defining the acoustic environment become more important as the acoustic frequency of propagation increases.

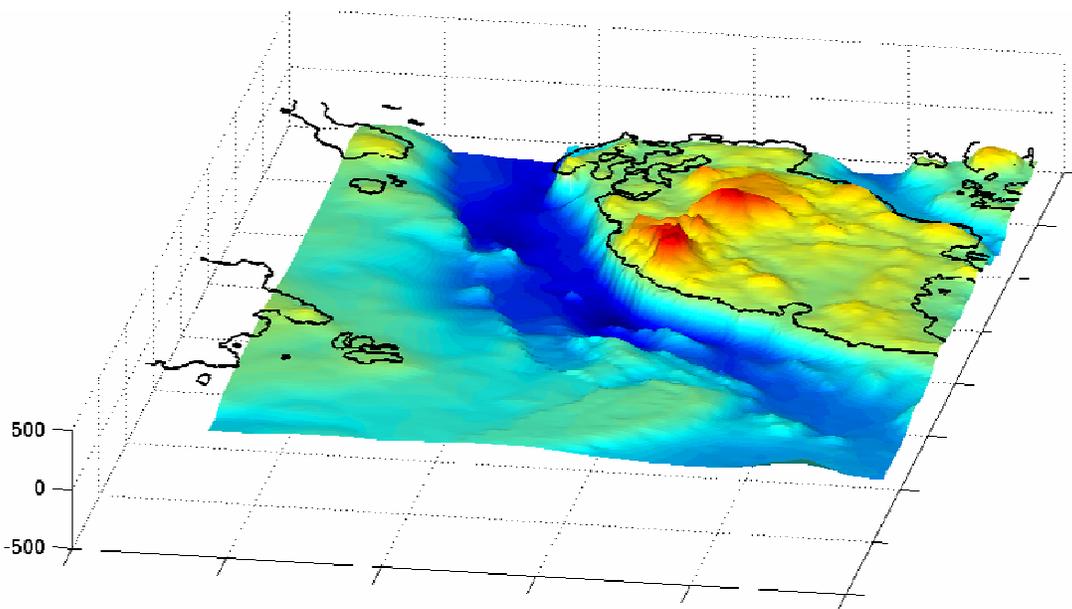


Figure 1: Bathymetry of the Haro Strait illustrating the contrast between the relatively deep channel and the steep wall at the western coast of San Juan Island. The horizontal scale is about 25 km square. The darkest blue designates a water depth of about 300 m.

VTS Traffic Day 151, 2004

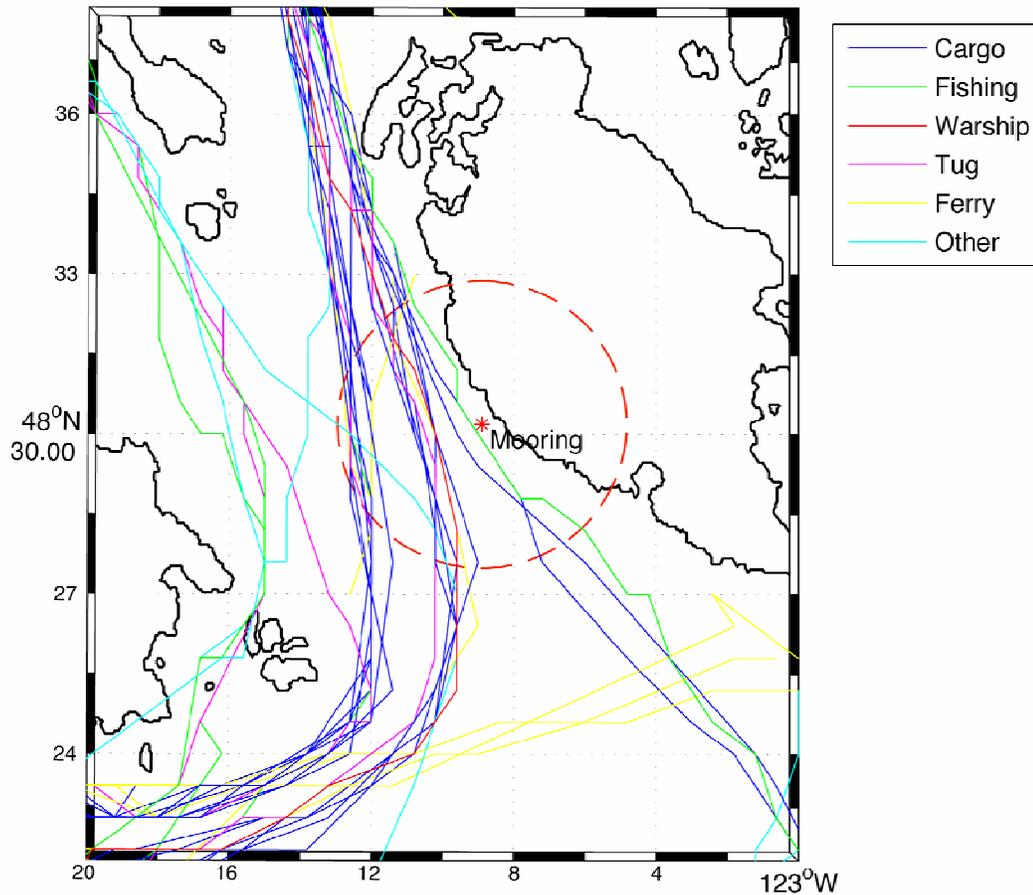


Figure 2: VTS tracks of shipping traffic in the Haro Strait for one day. A 5 km range ring (dashed red) is shown around the mooring location.

Killer whales communicate and forage by echo-location in the frequency range of 1 to 40 kHz (wavelengths of 4 cm to 1.5 m). The details of the surface and bottom at spatial scales of several meters and smaller will affect how sound propagates throughout this range of frequencies. This talk will focus on the lower end of this frequency range, illustrating propagation and variability at 3.6 kHz. However, the modeling methodology can be extended to higher and lower frequencies, as applicability requires.

Due to the complexity of this environment, one cannot expect measurements of acoustic signals recorded at one location to correlate with recordings at a different location. Thus understanding this acoustic environment through measurements alone would involve numerous hydrophones moored throughout Haro Strait, and would be prohibitively expensive. For this reason, numerical modeling is an attractive approach. However, in order to gain confidence in the modeling approach, it is useful to test the model results against measurements if they are available, as will be discussed.

During May and June of 2004, acoustic data was recorded on a mooring off the western coast of San Juan Island. Additionally, the Vessel Tracking System (VTS) database of ship traffic in the vicinity of Haro Strait was analyzed to isolate vessels

passing within 5 kilometers of the mooring. This provided a way to identify the location of a sound source (a passing ship) in the acoustic measurements. Figure 2 illustrates the variety and density of ship traffic for a given day.

Using ships whose VTS signatures reported a constant speed provides approximately constant source levels, and the measurements of the ship signatures would vary only due to differences in the propagation path from the ship location to fixed hydrophone position. This is nearly optimal for testing an acoustic propagation model in this environment. The optimal test would involve detailed knowledge of the source level of a particular ship. Figure 3 illustrates such a ship track and the associated acoustic data from the APL PAL instrument.

The acoustic model requires various environmental inputs, such as bathymetry, geo-acoustic properties of the sediments, water sound velocity, and bottom and surface roughness. The best available deterministic environmental data for the Haro Strait was used, and several reasonable scenarios were used for the roughness of the sea surface and bottom, as well as the choice of sediment thickness and properties. Due to the lack of knowledge of the geo-acoustic properties of the bottom as well as surface wave heights, a range of realistic values for these inputs to the acoustic model were used. As mentioned above, this uncertainty in the environmental inputs of the model becomes more relevant as the acoustic frequency of propagation increases.

Because the acoustic frequencies of propagation are sufficiently high, a statistical approach is applied, whereby ‘realizations’ of the surface and bottom roughness are created, and the acoustic model is run through each realization, finally obtaining a distribution of received acoustic signals. This ‘Monte-Carlo’ approach is a standard approach used in the ocean acoustics modeling and reveals that the model used in this study compares well with the data. In addition, it provides insight on the variability in the acoustic levels an orca would hear due to the time varying sea surface roughness. Figure 4 illustrates an example of the model/data comparison results for the ship illustrated in the previous figure.

This talk will discuss some details of this preliminary analysis, and will conclude by discussing potential future directions for this research, with the ultimate goal of providing a cost efficient method to ascertain the impact of shipping noise in the Haro Strait on the Southern Resident Orcas.

We gratefully acknowledge support from Brad Hanson and Linda Jones at the Northwest Fisheries Science Center (NWFSC).

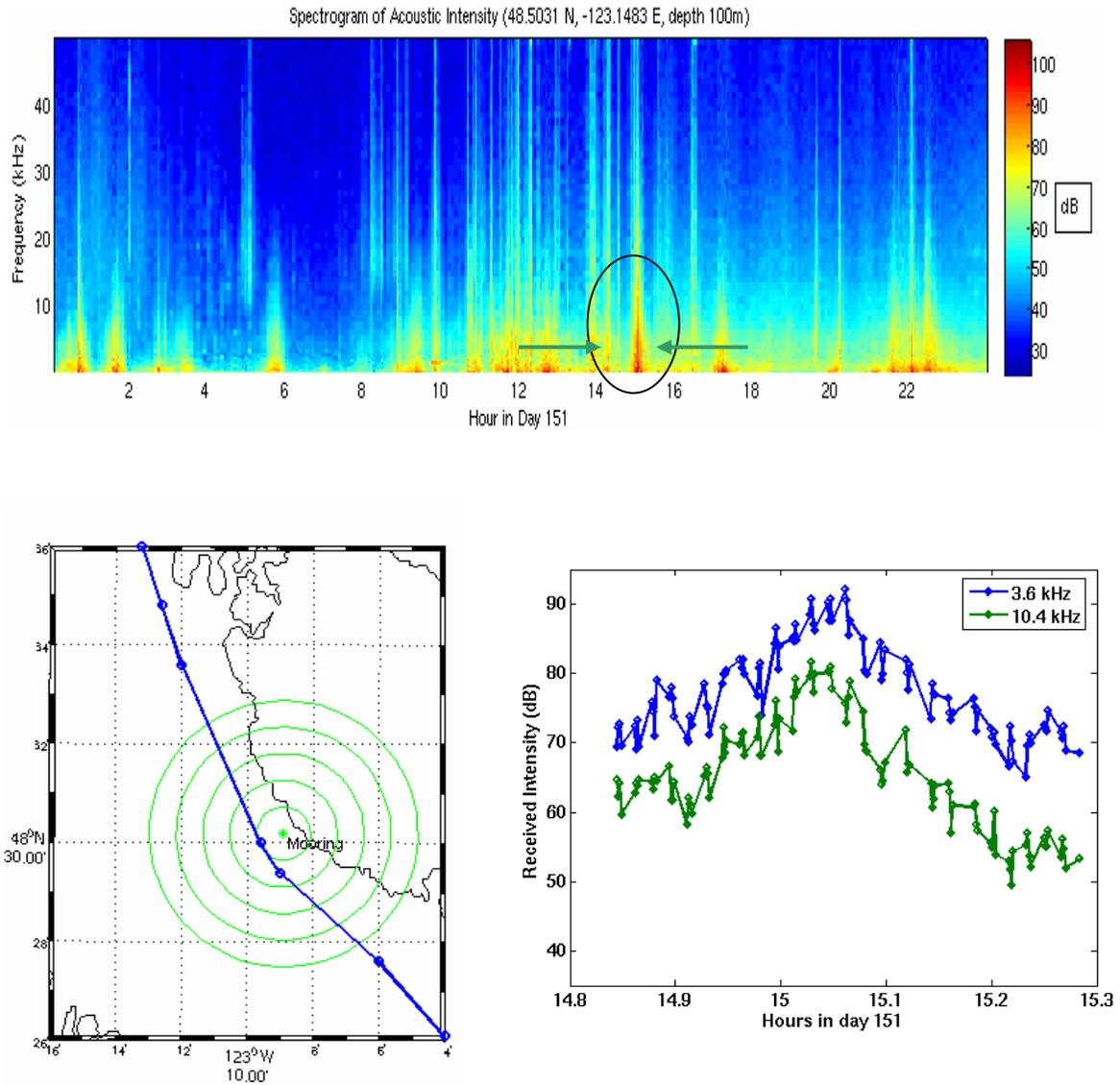


Figure 3: The spectral signature of an individual ship is shown (circled) in the PAL spectrogram (upper panel). The associated VTS ship track (lower left panel) shows that the ship passed within 1 kilometer of the mooring; concentric range circles are successively shown in green every 1 km. The spectral signature as a function of time as the ship passes the mooring is shown in the lower right panel for two frequencies of interest.

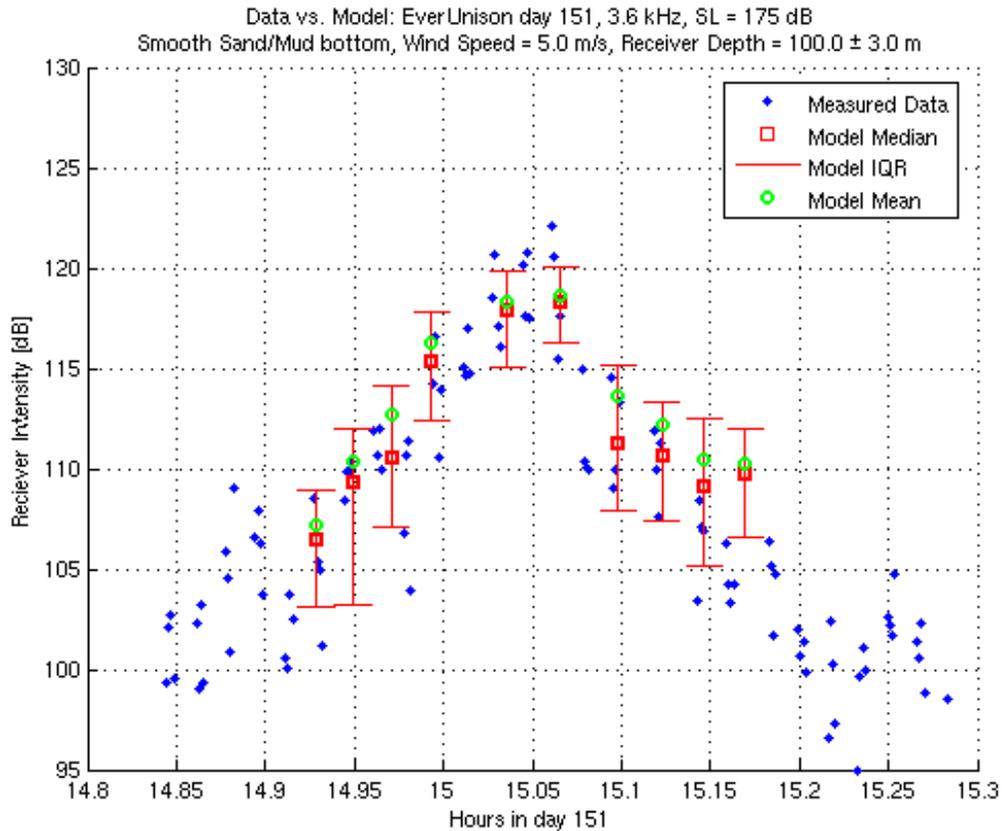


Figure 4. Comparison of model results and PAL data for the track of a single ship, as shown in Figure 3. The model results are derived by Monte-Carlo simulation using 20 realization of the environment with a wind speed of 5 m/s (~10 kts). Model results are illustrated with the mean, median, and data inter-quartile range (IQR) to illustrate data dispersion. The IRQ is the difference between the 75th and 25th percentiles in the random acoustic intensity. Data dispersion of >5 dB is observed in the model. Each point in time in the plot corresponds to a position of the ship. Each model data point corresponds to a position of the ship at 1 km range increments from the mooring as the ship passes traveling north, as illustrated in the previous figure by the intersections of the ship track and the concentric range circles. The sediment is modeled as a smooth sand/mud bottom.

Acoustic studies of the Southern Resident Killer whale population: implications for remote acoustic monitoring and indications of vocal behavioral change due to vessel noise.

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Each of the Southern Resident pods produces a distinctive repertoire of call types and has a dominant call type that can account for over 50% of the relative call usage (Hoelzel and Osborne 1986; Ford 1991). Suggested functions of stereotyped calls include group cohesion and coordination of foraging (Hoelzel and Osborne 1986; Ford 1989, 1991; Miller 2002, 2006). Call types can remain in the repertoire over a period of at least 28 years (Ford 1991).

It is important to know the full year-round range and habitat usage of the three pods that make up this population so that the sources of chemical and noise pollution, possible seasonal changes in diet and other possible threats be identified and the appropriate management steps taken. Acoustic monitoring has the benefit of suitability for remote deployment, low cost compared to ship-borne visual surveys, 24 hour detection capabilities and could have a range of several kilometres greater than visual sighting range in most sea states. Group specific call type repertoires could be important in facilitating remote acoustic monitoring.

We compared each pod's proportional call-type usage for two time periods (1977-1981 and 2001-2003) using a Pearson's product moment correlation coefficient. We analysed over 100 hours from 278 recording sessions of the Southern Residents, recorded between 1977 and 2003 and classified 16,153 calls into 28 call types. A further 2,017 calls were not classified either because they were too faint to reliably distinguish or the calls were characterized as non-stereotyped. Consistent with Ford (1991) we detected no substantial change over time to call-type structure or pod-specific repertoires, and only rarely used call types were recorded in one time period, but not the other. Although each pod shared a number of call types with the other two, the proportional usage was highly distinctive among pods in both time periods (Figure 1). The proportional usage of call-types was strongly correlated between time periods for each pod (Figure 2). This suggests that there is overall stability in the proportional usage of call types within each pod's repertoire, though the correlation results may be inflated by one or two outlying data points for J and L pods, and there are some proportional usage differences (Figure 2). However, the predominance of the two most common call types, (S1 & S4 for J pod; S16 & S17 for K pod; S19 & S2iii for L pod), in each of the Southern Resident pod's repertoires are pod distinctive and stable enough over decadal timescales to facilitate remote monitoring of this population using acoustic recording devices without accompanying visual tests.

A subset of two-component call types (S2i, S13, S36 & S37ii, S42 & S44) that were not commonly used by any of the pods when recorded on their own were more frequently recorded in multi-pod recordings. However, the primary and secondary call-types of each pod present in the various multi-pod aggregations remained prominent and should allow identification of the constituent pods within multi-pod aggregations.

The whale-watching fleet contains 72 commercial vessels and an average of 22 boats follow observed groups during daylight hours. The number of vessels increased progressively over the past decade (Foote et al. 2004). Impacts to killer whales, ranging from masking to a permanent threshold shift, have been predicted from vessel noise depending on the running speed, distance and vessel type of the boat (Bain and Dahlheim 1994; Erbe 2002). We address the problem of boat noise by testing hypotheses based on the theory that increasing redundancy through increased signal rate or signal duration should effectively increase the signal to noise ratio and lower the detection threshold. If the recent increase in noise associated with increased vessel numbers in the presence of the Southern Resident killer whales were masking vocal communication, we predict that one or possibly more of such anti-masking strategies would be adopted. We analysed the total duration of primary call types for each of the three Southern Resident pods J, K and L. Vessel noise presence was detected aurally, and by spectral energy and waveform amplitude. No attempt was made to quantify the number of boats present from the recordings. Call durations were compared in only two categories: motor boats present or absent for the duration of the recording. We found no significant difference in the duration of primary calls of each pod J, K and L in the presence vs. absence of boats for the first two periods, (1977-1981, 1990-1992) but a significant increase in call duration for all three pods in the presence of boats during the 2001-2003 period (Figure 3).

An increase in call duration could reduce masking by increasing redundancy, temporal summation or increased intra-narial pressure. The average number of vessels attending the whales increased approximately 5-fold from 1990 to 2000 (Foote et al. 2004). The concurrent change in call duration by all three pods suggests a threshold level of disturbance beyond which anti-masking behaviour began.

Bain, D. E. & Dahlheim, M. E. 1994. Effects of masking noise on detection thresholds of killer whales. In: *Marine mammals and the Exxon Valdez* (Ed. by T. R. Loughlin), pp. 243-256. San Diego: Academic Press.

Erbe, C. 2002. Underwater noise of whale-watching boats and potential effects on killer whales (*Orcinus orca*), based on an acoustic impact model. *Marine Mammal Science*, 18, 394-418.

Foote, A. D., Osborne, R. W. & Hoelzel, A. R. 2004. Environment: Whale-call response to masking boat noise. *Nature*, 428, 910.

Ford, J. K. B. 1989. Acoustic behaviour of resident killer whales (*Orcinus orca*) off Vancouver Island, British Columbia. *Canadian Journal of Zoology*, 67, 727-745.

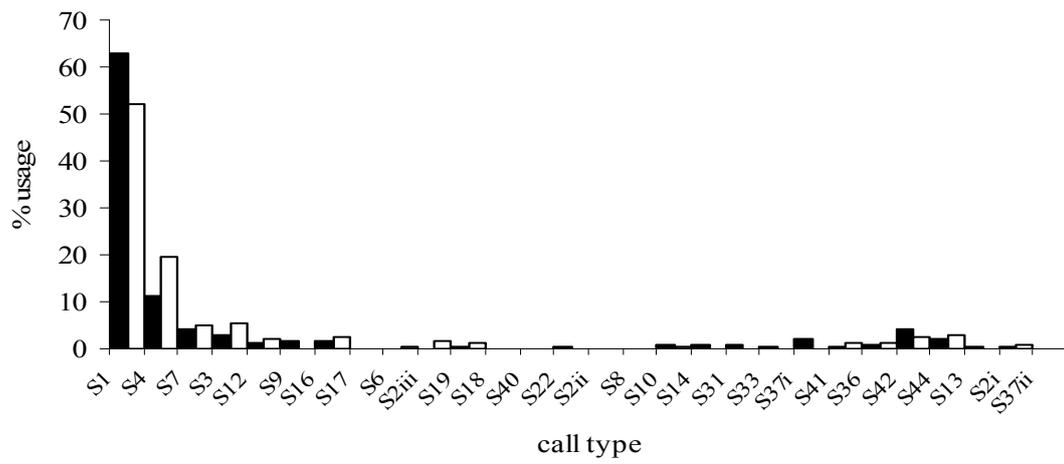
Ford, J. K. B. 1991. Vocal traditions among resident killer whales (*Orcinus orca*) in coastal waters of British Columbia. *Canadian Journal of Zoology*, 69, 1454-1483.

Hoelzel, A. R. & Osborne, R. W. 1986 Killer whale call characteristics: Implications for cooperative foraging strategies. In: *Behavioral biology of killer whales* (Ed. by B. Kirkevold & J. S. Lockard), pp. 373-403. New York: Alan R. Liss.

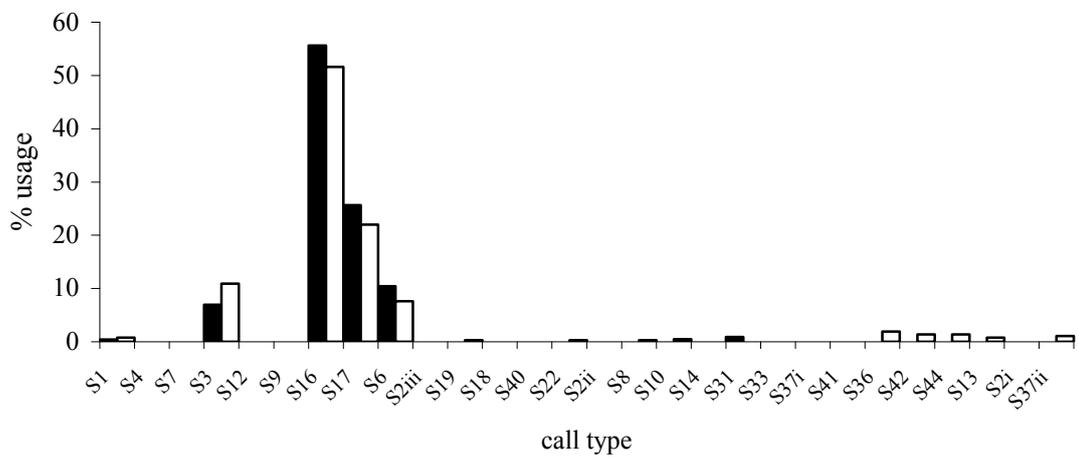
Miller, P. J. O. 2002. Mixed-directionality of killer whale stereotyped calls: a direction of movement cue? *Behavioral Ecology & Sociobiology*, 52, 262-270.

Miller, P. J. O. 2006. Diversity in sound pressure levels and estimated active space of resident killer whale vocalizations. *Journal of Comparative Physiology A*. doi 10.1007/s00359-005-0085-2

(a)



(b)



(c)

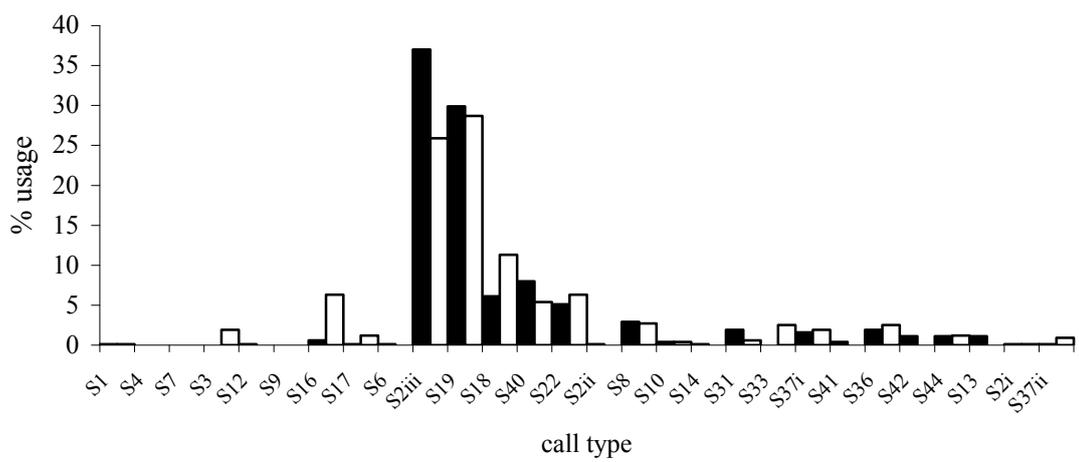
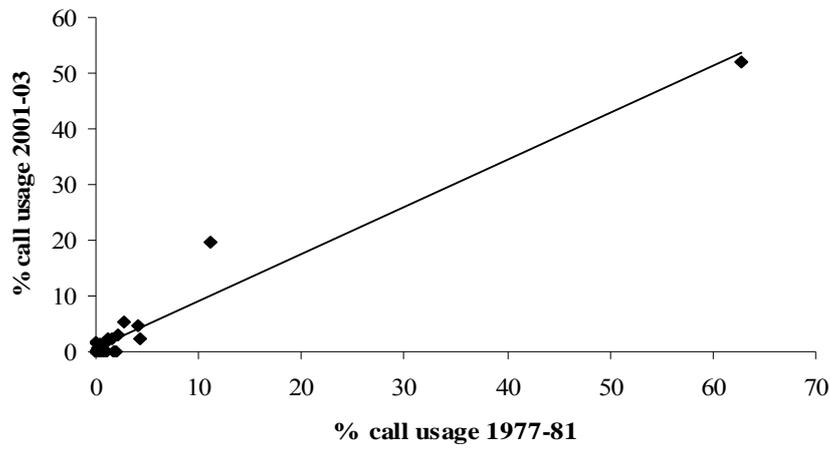
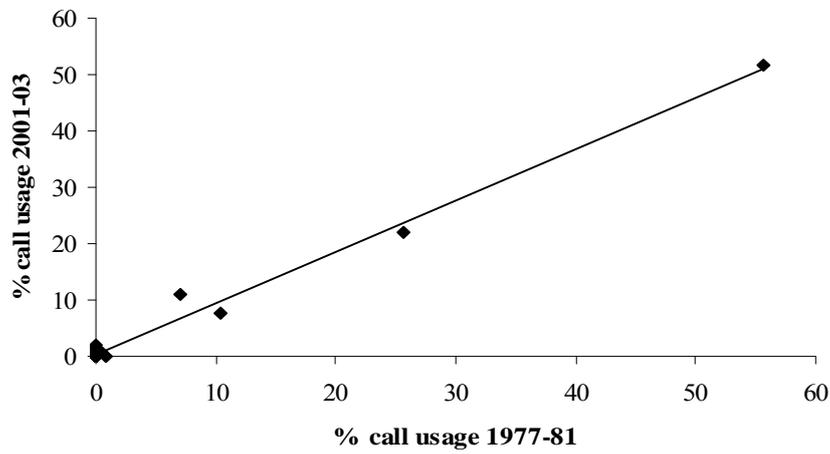


Figure 1. A comparison of call type usage between two time periods ■: 1977-1981 and □: 2001-2003 by: (a) J pod; (b) K pod; (c) L pod.

(a)



(b)



(c)

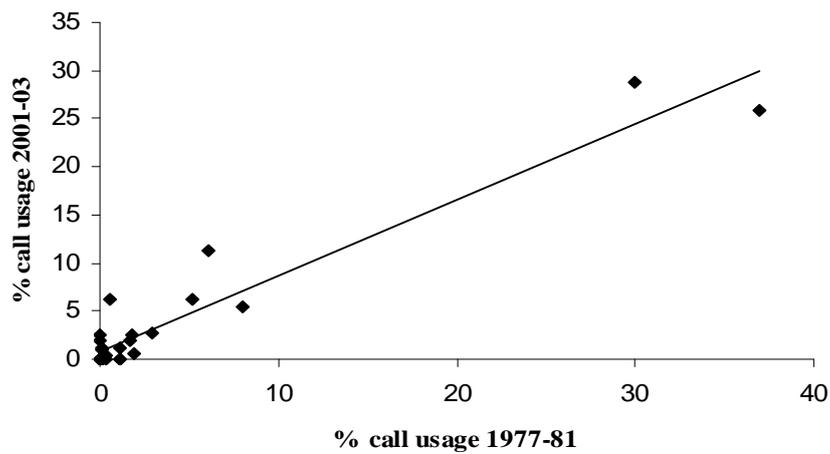


Figure 2. A plot of relative frequency of use of each call type in time period 1977-1981 compared against time period 2001-2003 by: (a) J Pod; (b) K Pod; (c) L pod.

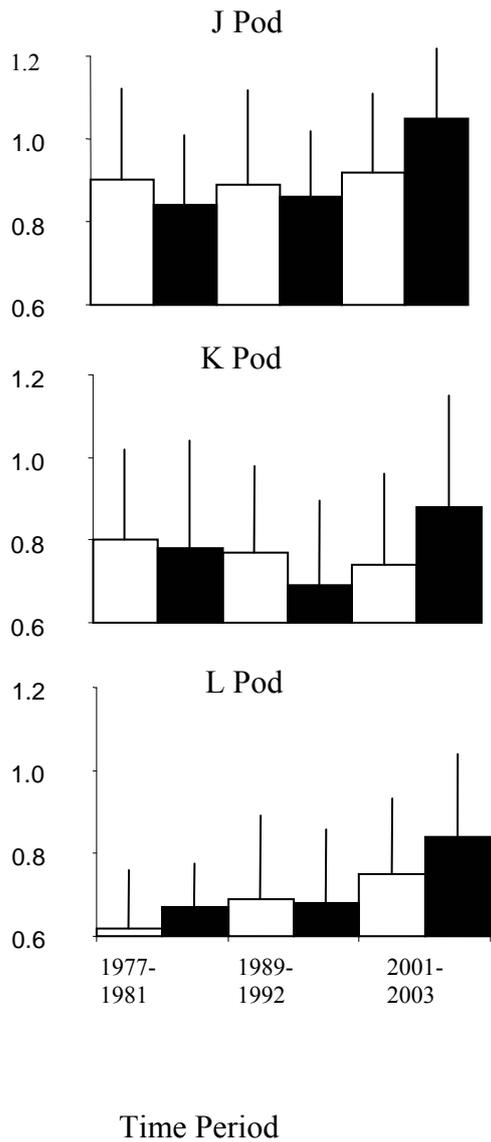


Figure 3. Call duration (seconds) for each pod comparing recordings with (black) and without (white) boats for each time period 1.(1977-81), 2.(1990-92) and 3.(2001-03) (error bars show 1 s.d.).