

Chemical Contaminants, Pathogen Exposure and General Health Status of Live and Beach-Cast Washington Sea Otters (*Enhydra lutris kenyoni*)



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## Chemical Contaminants, Pathogen Exposure and General Health Status of Live and Beach-Cast Washington Sea Otters (Enhydra lutris kenyoni)

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#### I. ABSTRACT

Analyses of blood and liver samples from live captured sea otters and liver samples from beachcast sea otter carcasses off the remote Washington coast indicate relatively low exposure to contaminants, but suggest that even at the low levels measured, exposure may be indicated by biomarker response. Evidence of pathogen exposure is noteworthy - infectious disease presents a potential risk to Washington sea otters, particularly due to their small population size and limited distribution. During 2001 and 2002, 32 sea otters were captured, of which 28 were implanted with transmitters to track their movements and liver and blood samples were collected to evaluate contaminant and pathogen exposure. In addition, liver samples from fifteen beachcast animals that washed ashore between 1991 and 2002 were analyzed to provide historical information and a basis of reference for values obtained from live otters. The results indicate low levels of metals, butyltins, and organochlorine compounds in the blood samples, with many of the organochlorines not detected except polychlorinated biphenyls (PCBs), and a few aromatic hydrocarbons detected in the liver of the live captured animals. Aliphatic hydrocarbons were measurable in the liver from the live captured animals; however, some of these are likely from biogenic sources. A significant reduction of vitamin A storage in the liver was observed in relation to PCB, dibutyltin and octacosane concentration. A significant and strong positive correlation in vitamin A storage in the liver was observed for cadmium and several of the aliphatic hydrocarbons. Peripheral blood mononuclear cell (PBMC) cytochrome P450 induction was elevated in two of 16 animals and may be potentially related to aliphatic and aromatic hydrocarbon exposure. Mean concentration of total butyltin in the liver of the Washington beach-cast otters was more than 15 times lower than the mean concentration reported by Kannan et al. (1998) for Southern sea otters in California. Organochlorine compounds were evident in the liver of beach-cast animals, despite the lack of large human population centers and development along the Washington coast. Concentrations of PCBs and chlordanes (e.g., transchlordane, *cis*-chlordane, *trans*-nonachlor, *cis*-nonachlor and oxychlordane) in liver of Washington beach-cast sea otters were similar to those measured in Aleutian and California sea otters, excluding those from Monterey Bay, which were higher. Mean concentrations of 1,1,1,trichloro-2,2-bis(p-chlorophyenyl)ethanes (DDTs) were lower, and mean concentrations of cyclohexanes (HCH, e.g., alpha BHC, beta BHC, delta BHC and gamma BHC) were slightly higher in Washington beach-cast otters versus those from California and the Aleutians.

Epidemiologically, blood tests revealed that 80 percent of the otters tested positive for morbillivirus and 60 percent for *Toxoplasma*, the latter of which has been a significant cause of mortality in Southern sea otters in California. This is the first finding of positive morbillivirus titers in sea otters from the Northeast Pacific. Individual deaths may occur from these diseases, perhaps more so when animals are otherwise immuno-compromised or infected with multiple diseases, but a population-threatening die-off from these diseases singly is unlikely while population immunity remains high. The high frequency of detection of morbillivirus and *Toxoplasma* in the live otters corresponds well with the cause of death of stranded Washington sea otters reported herein, which has generally been attributable to infectious disease. Washington's sea otter population continues to grow, with over 1100 animals currently

inhabiting Washington waters; however, the rate of growth has slowed over recent years. The population has a limited distribution and has not yet reached its carrying capacity and as such, is still considered at high risk to catastrophic events.

Keywords: sea otter, metals, butyltins, organochlorines, aromatic hydrocarbons, aliphatic hydrocarbons, PCBs, infectious disease, *Toxoplasma*, morbillivirus, vitamin A, cytochrome P450.

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## **II. INTRODUCTION**

The northern sea otter (*Enhydra lutris kenyoni*) population is federally protected under the Marine Mammal Protection Act, and has been listed as endangered since 1981 by Washington State, as it is vulnerable due to small population size and restricted distribution. (Lance et al. 2004) (Figure 1). Sea otters were extirpated from Washington waters in the early twentieth century due to the fur trade (Wilson et al. 1991). In 1969 and 1970, sea otters were translocated from Amchitka, Alaska to the Washington coast to re-establish the population (Jameson et al. 1982). A total of 59 were released in Washington; 30 in 1969 and 29 in 1970. Post release mortality was high the first year with over half the population ending up as beach-cast carcasses. Only 19 animals were counted in 1979, but since then the population has grown at an average annual rate of about 18 percent through 1989. From 1989 to 2004, the average annual rate of increase was about 8.2 percent (Laidre et al. 2002, Jameson and Jeffries 2000, 2004, 2005).





Photo courtesy of C.E. Bowlby

The potential carrying capacity of approximately 1,372 to 2,734 sea otters (Laidre et al. 2002) has not yet been reached for the Washington population. The 2007census report on the Washington sea otter population indicates that the population is continuing to increase in size, with the current estimate at over 1100 animals (Jameson and Jeffries 2007). Results from the 2000 census, however, had the population numbers leveling or decreasing slightly as compared to the 1999 census (Jameson and Jeffries 2000).

With range expansion possible to the south along the Washington coast and east into the Strait of Juan de Fuca, the Washington sea otter population is facing new or additional risks due to increased anthropogenic influences (Figure 2). The human population increases in the eastern portion of the Strait, with several sizeable towns and marinas, and the Strait is a major shipping channel to Puget Sound, Washington and Vancouver, British Columbia. With range expansion to the south along the Washington coast, the sea otters will be entering not only a more developed and populated area, but also a different ecosystem that is associated with different risks, such as a the potential increased exposure to biotoxins like domoic acid in bivalve prey, as well as potential increased sources of contaminants.

The Washington sea otter population is not the only sea otter population potentially at risk. Otter populations along the California coast recently declined during the late 1990s and the Alaskan Aleutian Island population has undergone an alarming decline, and the Prince William Sound population in Alaska lost thousands of otters after the 1989 T/V *Exxon Valdez* spill. Initially, it was postulated that declines in the Alaska Aleutian population have occurred as killer whales (*Orcinus orca*) switch from one prey type to another resulting from declines in the abundance of their primary prey (pinnipeds) (Estes et al. 1998). However, continued declines (up to 90 percent in some locations) lead scientists to speculate that more than predation may be responsible for the decrease in otter numbers (Brownell 1999).

Infectious disease (Hanni et al. 2003, USFWS 2003) and cardiac disease (Kreuder et al. 2003) have been significant mortality factors in California populations. Some have speculated that the decline of the California population may be anthropogenic in nature (Jarman et al. 1996, Kannan et al. 1998). Several reports have recently suggested an increased disease susceptibility resulting from contaminant-induced immunosuppression may be contributing to worldwide declines in marine mammals (Dietz et al. 1989, Osterhaus et al. 1990, Ross et al. 1996, Kannan et al. 1998 and Tanabe et al. 1998). Immunotoxic chemicals include polychlorinated biphenyls (PCBs<sup>1</sup>), polychlorinated dibenzo-*p*-dioxins (PCDDs), polychlorinated dibenzofurans (PCDFs), lead, cadmium, methyl mercury, nickel, butyltins, hexachlorobenzene (HCB), polycyclic aromatic hydrocarbons (PAH) and several pesticides (Ross et al. 1996). A more focused study has revealed that declining populations of sea otters from California and the Aleutian archipelago have higher concentrations of PCBs compared to populations inhabiting southeastern Alaska (Bacon et al. 1999), which could increase their susceptibility to pathogens. The design of our Washington study purposely paralleled some of the analytical parameters in both Alaska and California studies to allow for comparisons between the populations.

Unlike other marine mammals that migrate extensively, sea otters provide an unusual opportunity for study because both the sea otters and their principle prey are relatively sedentary; thus, their contaminant burdens should reflect localized contamination. To address this, in addition to sea otter contaminant residues, data are presented on the chemical residues in their prey. Sea otters also provide an unusual opportunity to study a mid to high-trophic level marine consumer inhabiting highly industrial to extremely remote habitats throughout its occurrence in the Northeast Pacific.

<sup>&</sup>lt;sup>1</sup> A list of acronyms is provided in Appendix 8.

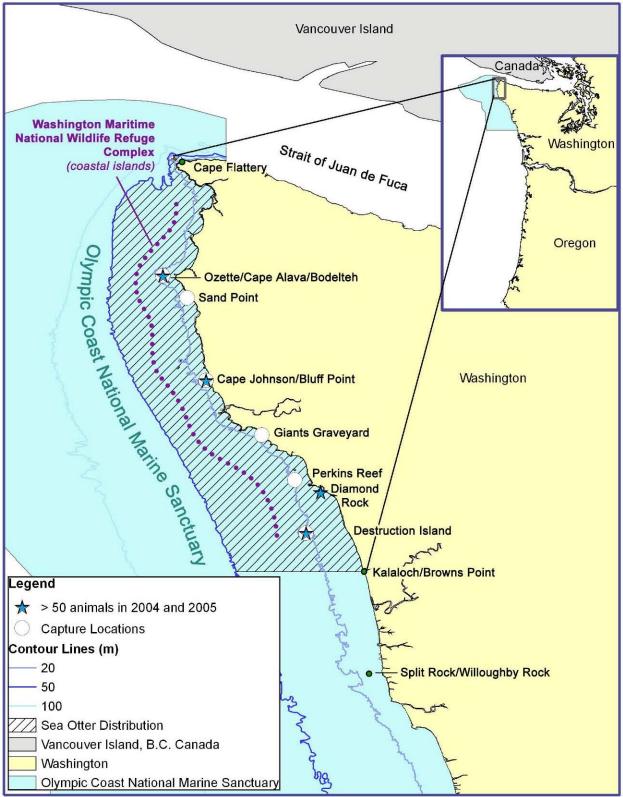


Figure 2. Washington Sea Otter Distribution, High Areas of Use (> 50 animals) and Capture Locations

One of this study's objectives was to develop baseline information on sea otter contaminant and pathogen exposure to compare it to data reported elsewhere. Environmental exposure to contaminants has been associated with a variety of physiological impacts on fish and wildlife. Many fat-soluble environmental contaminants, including PCBs, persist in the environment and present unequivocal risks to the health of wildlife and humans. Their lipophilic nature, environmental persistence, and resistance to metabolic breakdown within organisms leads to biomagnification up the food chain (Cockcroft et al. 1989). As a consequence, fish and invertebrate-eating birds and marine mammals are often exposed to elevated concentrations of these contaminants, and studies suggest that certain populations are suffering from their various toxic effects (Ross et al. 1996). In addition to analyzing organochlorines, analytical scans were completed for other aliphatic and aromatic hydrocarbons and semi-volatile compounds, organotins and metals to complete the baseline assessment. Biomarkers of toxicity, including vitamin A and thyroid hormone concentration, and cytochrome P450 induction were also evaluated, as well as evidence of biotoxins.

Infectious disease exposure (and its potential correlation with immunotoxic chemical concentrations) was also a concern for Washington sea otters. In California, 195 sea otter carcasses were evaluated and 38.5% of the mortalities were caused by infectious diseases (Thomas and Cole 1996). Mortalities reported in California included those from protozoal encephalitis caused by the parasites *Toxoplasma gondii* and *Sarcocystis neurona*.

In summary, this report presents a baseline for the Washington sea otter population as of 2001-2002, in terms of contaminant and pathogen exposure, biomarker responses, and general blood and serum chemistry. It also presents data on their prey to illustrate potential dietary exposure to contaminants and pathogens. Whenever data are available, the Washington sea otter results are compared to those for the California, British Columbia, Alaska-Aleutian Island and Alaska-Prince William Sound populations of otters.

## **III. METHODS**

## **Collection of Tissue Samples from Live Captured Otters**

Sea otters were captured off the coast of Washington in areas of the Washington Maritime National Wildlife Refuge Complex in August of 2001 and 2002. The current distribution of the Washington sea otter population distribution extends from Kalaloch to Pillar Point (Laidre and Jameson 2006), a distance of about 130 kilometers. Specific locations within this range were selected where sea otters are known to form resting rafts, including Destruction Island, Perkins Reef, Bodelteh and Ozette Islands, and Cape Alava (Figure 2).

Sea otters were captured by divers equipped with close circuit (rebreather) SCUBA (to avoid surface disturbance from SCUBA air bubbles), employing a Wilson Trap for an underwater capture (Figure 3a,b). Each animal was transferred and restrained within a net bag, which is closed by a purse line. Captured animals were then transported to the research vessel for surgery. Immobilization and surgical implantation was conducted under the direction of Dr. Carolyn McCormick, a wildlife veterinarian experienced with sea otters. All animals captured except small pups, were immobilized using fentanyl, which is given intramuscularly in combination with valium. This combination produces a safe, short-acting, and easily reversible immobilizing agent suitable for use under field conditions (Monson et al. 2001). Naltrexone was used as an antagonist to insure that animals are recovered prior to release. The wildlife veterinarian administered all drugs. Resuscitation equipment was available in the field and an emergency plan to transport animals needing additional medical attention was in place, but neither was needed.



Figure 3a. Wilson Trap with a Captured Sea Otter (Photo courtesy of C.E. Bowlby)



**Figure 3b. Wilson Traps Stacked on Deck of the Capture Vessel** (Photo courtesy of J.W. Davis)

After an individual was anesthetized, morphometric measurements were taken (Figure 4). Otters were measured, weighed, and sexed, and fecal and urine samples were collected when possible. Each sea otter was marked with a passive integrated transponder (PIT) tag (Thomas et al. 1987) and fitted with unique combinations of colored plastic tags placed in the inter-digital webbing of the hind feet. Any webbing tissue expelled from punching the flipper for the tag was preserved for genetic analyses (currently being conducted by the Seattle Aquarium). A premolar was extracted for age determination. Blood samples were drawn. As part of a concurrent on-going population study, radio transmitters were implanted within the intraperitoneal cavity when size, age, and reproductive condition criteria, as stated in the marine mammal research permit (Appendix 4), were met. Because radio implantation required surgery, we used the opportunity to take fat and liver biopsies for hormone and contaminant chemistry, as per the permit amendment. The surgically implanted radios and transponder chips are permanent, while the flipper tags may eventually dislodge over time. Sea otters were generally released within 2 hours of capture and marking. Mother/pup pairs were released together.

## Figure 4. Ron Jameson Weighing a Captured Sea Otter (Photo courtesy of M.S. Brancato)



The risk to sea otters associated with the capture and surgery activities is very low. During past sea otter research in Washington State, only 1 of 101 captured animals died as a result of the capture procedures (less than 1 percent). No animals succumbed to either capture or surgery during this study effort. In fact, all of the animals implanted with transmitters tracked during this study were alive six months post-capture, well beyond the point when infection or complications associated from the procedures would be expected. Results of the tracking surveys are presented in Appendix 1.

The two primary tissue types collected were blood and liver biopsies. Additional samples were collected as the opportunity arose, such as subcutaneous fat, plug of tissue from punch tagging the animal, feces, urine, hair, and fecal and oral swabs for parasite analysis. Table 1 summarizes the samples collected and type of analyses performed each year.

#### **Collection of Samples from Beach-Cast Otters**

Beach-cast otters used in this study were collected opportunistically between 1991 and 2002 along the Washington coast. Fresh carcasses were collected and shipped on ice to the National Wildlife Health Center in Madison, Wisconsin for necropsy. Tissue samples were archived at the National Wildlife Health Center wrapped in aluminum foil inside a Whirl Pac<sup>®</sup> bag and held frozen at -20°C prior to shipment for contaminant chemistry.

#### **Chemical Analyses**

Whole blood from live-captured otters was used to determine concentrations of metals, butyltins, organochlorines, congener-specific polychlorinated biphenyls (PCBs) and semi-volatile compounds. Liver biopsies from the live-captured otters were analyzed for aliphatic and aromatic hydrocarbons. Liver samples from beach-cast otters collected from Washington beaches were analyzed for residues of organochlorines, metals, and butyltins to provide information regarding historic and current contaminant levels in sea otters of this region. Contaminant analyses were performed by Geochemical & Environmental Research Group (GERG) and Trace Element Research Laboratory (TERL) at Texas A&M University, both contract laboratories for the U.S. Fish and Wildlife Service's (FWS) Analytical Control Facility (ACF). A list of the parameters for each analytical scan is provided in Appendix 2. The analytical methods followed those outlined in NOAA Technical Memorandum NOS ORCA 130 (Lauenstein and Cantillo 1998) unless otherwise specified.

Year	Sample	N	Analyses Performed
Beach-cast	Liver tissue (from dead sea otters	15	Metals, Butyltins, Organochlorines, Congener-Specific
(Stranded)	previously collected)		PCBs and Semi-volatiles. One animal from 2002 also
Animals 1991-			analyzed for Vitamin A levels
2002			
Live Captures	1		
2001	Whole Blood	15	Metals, Butyltins, Organochlorines, Congener-Specific
2001	Whole Blood	15	PCBs and Semi-volatiles
	Serum	15	Serum Chemistry, Cell Blood Counts (CBC) and
			Disease/Pathogen Titers
	Plasma	14	Vitamin A and Thyroid Hormone Levels
	Liver Biopsy	7	Aliphatic and Aromatic Hydrocarbons
	Liver Biopsy	15	Histopathology, RT-PCR, p-450 CYP1A, and Vitamin A
			Levels
	Teeth	14	Age Determination
	Feces swab	15	Parasites
	Feces (scat)	3	Parasites
	Urine	2	Domoic Acid
	Subcutaneous fat	15	Fatty Acid profiles
2002	Whole Blood	15	Metals, Butyltins, Organochlorines, Congener-Specific
			PCBs and Semi-volatiles
	Serum	15	Serum Chemistry, Cell Blood Counts (CBC) and
			Disease/Pathogen Titers
	Plasma	15	Vitamin A Levels
	Plasma	14	Thyroid Hormone Levels
	Liver Biopsy	9	Aliphatic and Aromatic Hydrocarbons
	Liver Biopsy (total includes one	11	Histopathology, RT-PCR, p-450 CYP1A and Vitamin A
	2002 beach-cast animal)		Levels
	Teeth	13	Age Determination
	Feces swab	14	Parasites
	Feces (scat)	4	Parasites
	Urine	1	Domoic Acid
	Subcutaneous fat	15	Fatty Acid profiles

## Table 1. Sea Otter Samples Collected and Analyses Performed

## Organics

Tissue samples were extracted following the method described in MacLeod et al. (1985) with minor revisions from Brooks et al. (1989) and Wade et al. (1988). Briefly, internal standards were added to tissue samples at environmentally relevant concentrations prior to extraction. Tissue samples were then homogenized in methylene chloride (CH<sub>2</sub>Cl<sub>2</sub>) and anhydrous sodium sulfate (Na<sub>2</sub>SO<sub>4</sub>) with a Tissumizer for 3 minutes. The sample mixture was centrifuged and the extract decanted. The extraction process was repeated two more times, with extracts combined and concentrated to 2 ml. Cleanup was done by alumina /silica gel chromatography. Silica gel (20 g, 170°C for 12h, deactivated 5%) was slurry packed over alumina (10 g, 400°C for 4 h, deactivated 1%) using  $CH_2Cl_2$ . The  $CH_2Cl_2$  was replaced with hexane and the tissue extract transferred to the column using 2 ml of hexane. The column was then eluted with 50 ml of pentane to bring off the aliphatics (fraction 1), 200 ml of 1:1 CH<sub>2</sub>Cl<sub>2</sub>:pentane to elute the PAHs, PCBs and organochlorine pesticides (fraction 2). Fraction 2 is concentrated and further cleanup done on the HPLC to remove interfering lipids using size exclusion chromatography. The final extract elution (containing PAHs, PCBs, organochlorine pesticides) was quantitatively analyzed on DB-5 capillary column gas chromatograph (CGC) equipped with a flame ionization detector for aliphatic hydrocarbons, electron capture detector for PCBs and organochlorine pesticides, and CGC/mass spectrometer (MS) in the selected ion monitoring (SIM) to analyze the molecular ions of the compounds of interest.

There are specific cases where analytes requested for pesticide and PCB analyses are known to co-elute with other analytes in the normal CGC with electron capture. These include the pesticide Endosulfan I and the PCB congeners 114 and 157. In these cases, the samples were analyzed by CGC using a mass spectrometer detector in the SIM mode.

To obtain percent moisture and percent lipid, approximately 1 gram of wet sample was weighed into a clean, labeled, preweighed 10 ml beaker. The beaker was placed in a forced air oven at approximately 75 °C for 24 hours. The beaker with the dry sample was then weighed and the percent dry weight is calculated by the formula:

To obtain sample percent lipid, a portion of the extracted lipid containing solution is placed in a pre-weighed 10 ml beaker. The solvent is evaporated from the beaker and the beaker placed in an oven at 105°C for one hour. The evaporated extract is cooled for one hour in a desiccator and reweighed. Percent lipid is calculated by the formula:

Equation % lipid =  $\begin{array}{cc} W_L & V_T \\ ----- & x & ----- \\ W_T & V_{EG} \end{array} X 100$ 

where:  $W_L$  = weight of lipid  $W_T$  = total weight of sample  $V_T$  = total volume of extract  $V_E$  = volume of extract used for lipid determination

The sample percent lipid and percent moisture determinations are presented in Appendix 3 along with the sample weights and volumes.

The concentration of butyltins including tetrabutyltin, tributyltin, dibutyltin, and monobutyltin were determined as described by Wade et al. (1988) for tissues. Tissue samples were homogenized, an aliquot (2-10g, fresh wet) was weighed into a centrifuge tube and surrogate tripropyltin (TPT), approximately 300 ng as Sn added. Samples were extracted for 3 minutes using a Tissumizer after addition of 100 ml of 0.2% tropolone in methylene chloride and sodium sulfate as a drying agent. The samples were then centrifuged and the solvent portion containing the butyltins decanted. Samples were extracted two more times after adding 100 ml of 0.2% tropolone in methylene chloride. Extracts were combined and concentrated on a rotary evaporator to approximately 20 ml. Samples were transferred to 50 ml centrifuge tubes and the methylene chloride was replaced with hexane.

Tissue samples were hexylated in the centrifuge tubes by adding 2 ml of hexylmagnesium bromide under a nitrogen atmosphere and heating at 60° C in a water bath for six hours. Excess hexylmagnesium bromide was neutralized by adding 5 ml of 6 M hydrochloric acid. The organic fraction was removed and saved and the aqueous phase extracted three more times with 10 ml of pentane each time. Sodium sulfate was added to the combined extracts to remove water. The samples were then concentrated to 2 ml, which was transferred to a silica (13.5 g)/alumina (17.0g) column and eluted with 50 ml of pentane. Samples were concentrated to 0.5 ml and tetrapropyltin (4PT), approximately 300 ng as Sn, was added as a recovery standard. Samples were quantitatively analyzed by gas chromatography using a flame photometric detector equipped with a 610 nm filter.

#### **Inorganic Compounds 2002**

#### Digestion of biological tissue.

Liquid or solid biological tissue samples were wet digested with nitric acid and converted into acidic digest solutions for analysis by various atomic spectroscopy methods. When possible, tissue was freeze dried in order to minimize loss of analytes and to facilitate subsequent sample preparation steps, and then homogenized to a fine powder by ball-milling in plastic containers. Approximately 0.20 to 0.25 g of powdered tissue was weighed into a Teflon reaction vessel and 3 ml of HNO<sub>3</sub> were added. The closed reaction vessel was heated in a 130 °C oven until digestion was complete. Samples were then diluted to a final volume of 20 ml with quartz distilled water and stored in 1 oz. polyethylene bottles for later analysis by instrumental techniques.

Analysis of trace metals by inductively coupled plasma optical emission spectroscopy (ICP-OES). (Boron, Barium, Beryllium, Iron, Magnesium, Molybdenum, Strontium, Zinc) Liquid samples were nebulized and the resulting aerosol was transported to the plasma torch. Element-specific atomic-line emission spectra were produced by an inductively coupled argon plasma. The spectra were dispersed by a grating spectrometer, and the intensities of the lines were monitored by photomultiplier tubes or solid state detectors. Samples were quantitated by comparison with external standards. One or more internal standards may be incorporated to compensate for physical effects resulting from viscosity and varying levels of total dissolved solids in the samples. Background correction was required and was measured adjacent to analyte lines on samples during analysis.

# Analysis of trace metals in blood samples by inductively coupled plasma-mass spectroscopy (ICP-MS). (Silver, Aluminum, Arsenic, Cadmium, Chromium, Copper, Manganese, Nickel, Lead, Selenium, Vanadium)

Concentrations of trace elements in blood samples were determined with an atomic spectroscopy method that relies on ionization of sample constituents in a high temperature argon plasma and separation of positively-charged ions on the basis of their mass:charge ratios (m:z) by a quadrupole mass spectrometer. The method offers extremely low detection limits but is subject to interferences from atomic and molecular ions having values within 1 AMU of the target ions. Sample preconcentration and matrix elimination can sometimes eliminate these problems, along with those resulting from high total dissolved solids.

## Digestion of blood and biological tissue for mercury analysis.

Before samples are analyzed by the CVAAS method, mercury is converted to the Hg2+ form. Mercury is digested by a modified version of EPA method 245.5 and 245.6. Sediment and tissue samples can be analyzed either freeze dried or on a wet basis. Tissue samples were homogenized in the original sample containers either after freeze drying or with a Tekmar Tissumizer and subsampled. Samples were digested with nitric acid, sulfuric acid, potassium permanganate, and potassium persulfate in polypropylene tubes in a water bath at 90-95 °C. Before analysis, hydroxylamine hydrochloride was added to reduce excess permanganate and samples were brought to volume with distilled-deionized water.

## Analysis of mercury by cold-vapor atomic absorption spectroscopy (CVAAS).

In this procedure, divalent mercury (Hg++) in aqueous samples (digests of tissue samples) is reduced to the elemental state (Hgo) by a strong reducing agent (stannous chloride). Gaseous Hgo enters the sweep gas and is introduced into an atomic absorption cell, where light produced by a mercury vapor lamp is absorbed by the free Hg atoms. Mercury in the sample was determined by comparing light absorption of the sample with that of external calibration standards.

## Moisture content of tissue samples.

Moisture content was determined by weight loss upon freeze-drying, and is expressed as weight percent of the original wet sample. Depending upon sample size, either the whole sample or a representative aliquot was frozen and then dried under vacuum until a constant weight was attained. Samples were prepared and dried using plastic materials, whenever possible, in order to minimize potential contamination artifacts that might impact subsequent trace element analysis.

## **Inorganic Compounds 2001**

## Trace Metal Total Tissue BOMB Digest

Tissues were digested in heavy-walled, screw-cap Teflon Bombs with concentrated high purity nitric acid. Bombs were heated (for 2-8 hours) and opened three times to release  $CO_2$  build-up. Oven temperature was 129 °C. This procedure results in a total digestion with all trace elements present in the tissue samples being solubilized. Most metals in the digestate were determined by graphite furnace AAS, in which electrical heating is used to produce an atomic cloud. Some elements were typically in high enough concentration (e.g. Zn) to be determined by flame AAS. Mercury was determined by cold vapor atomic absorption spectrometry (AAS), in which Sn2+ is used to reduce Hgo.

## Quality Assurance/Quality Control (QA/QC)

Laboratory QA/QC was under the general supervision of FWS' ACF and their contract laboratories such as Texas A&M University's GERG. The data obtained from the laboratories were reviewed to determine if any should be flagged for quality control issues and excluded from statistical analyses. In addition to review by the principal investigators, the laboratories running the analyses implemented standard QA/QC procedures. Procedural blanks, duplicate samples (with relative percent difference calculated), spike samples and analysis of standard reference materials were all performed by the Texas A&M laboratories to evaluate potential contamination during sampling and to review the accuracy and precision obtained by the laboratories.

The only quality control issue of note is that the variability of Total Petroleum Hydrocarbons (TPH) in the tissue was high (see Table 13). TPH results should be considered estimates. Also, a few of the PCB congeners had data qualifiers for interference and were not included in the total PCB calculation (see Appendix 5 and 6). Typically the analyst will look for an Aroclor pattern and if some PCB congener is out of proportion and/or the peak is not well defined, it will not be included in the total PCB value.

#### **Biomarker Analyses**

## Vitamin A and Thyroid Hormone Analyses

#### **Blood Samples**

Plasma was separated from whole blood and a sample of approximately 1 mL was analyzed for circulatory vitamin A and thyroid hormone concentrations by the Central Laboratory for Veterinarians (Langley, BC, Canada) using their standard laboratory procedures. Vitamin A concentrations in plasma were reported as the total of both free and bound *all-trans* retinol, but did not include retinol esters (that are present in circulation in most carnivores). The thyroid hormones that were measured are tetraiodothyronine (TT4) and triiodothyronine (TT3).

## Liver Samples

Vitamin A concentrations in liver were determined at the Institute of Ocean Sciences (Sidney, BC, Canada) using the method of Mos and Ross (2002). In this method, tissue samples are ground with anhydrous sodium sulphate until a pink powder is obtained. Vitamin A (retinol and retinol-esters) was extracted from this powder in two-step extraction with *n*-hexane. Hexane extracts were subsequently evaporated, and dissolved in a small volume of methanol:dichloromethane (9:1). Vitamin A was quantified by reversed phase HPLC equipped with UV detection (325 nm; Beckman System Gold Solvent Module 126 and Detector Module, respectively) using a Vydac C18 column ( $0.46 \times 25 \text{ cm}$ , 5 µm) and a gradient mobile phase of methanol and dichloromethane at 1.0-1.5 mL min<sup>-1</sup>. Retinol and one retinol ester (retinyl palmitate) were quantified using commercially obtained standards (Sigma-Aldrich). Retinyl-acetate was used an internal standard.

Total retinol content for liver was calculated by addition of retinol and retinyl-palmitate concentrations, in which the retinyl-palmitate concentrations were converted into retinol equivalents by ignoring the mass of the fatty acyl units. A molar ratio of retinol to retinyl palmitate in liver was calculated as a measure of vitamin A mobilization from storage.

## P450 CYP1A Assays

Liver and PBMC total RNA was isolated and quantitative RT-PCR for CYP 1A was performed on the isolated samples following methods described Ben-David et al. (2001). The assays were conducted by a diagnostic laboratory at Purdue University.

Peripheral blood mononuclear cells (PBMC) were isolated from heparinized blood samples taken from live captured sea otters and cryopreserved for analysis of cytochrome P450 (CYP1A) expression using a quantitative reverse transcriptase polymerase chain reaction (RT-PCR). Primers specifically designed for sea otter CYP1A were used.

Liver biopsies from live captured animals were also preserved for analysis of cytochrome P450 (CYP1A1) expression using a quantitative RT-PCR. Liver biopsies were snap frozen in liquid nitrogen for CYP1A expression.

## **Pathogen Analyses**

Serum samples were tested by various diagnostic or research laboratories for antibody to morbilliviruses, *Leptospira* sp., *Brucella* sp., *Toxoplasma gondii, Neospora caninum, Sarcocystis* sp., and Caliciviruses. In some cases tests were run in sequence on the same samples so that serum samples were subsampled by one laboratory before transfer to another laboratory. Samples were tested by the following methods:

## Morbilliviruses:

Serum (virus) neutralization tests were carried out by the Oklahoma State Veterinary Diagnostic Laboratory, Stillwater, Oklahoma. The laboratory method is described in Garner et al. (2000).

#### *Leptospira* sp.:

*Leptospira* microscopic agglutination tests were performed at USDA-National Veterinary Services Laboratory, Ames, Iowa. The method is described in USAHA, 1987.

## Brucella sp.:

Four antibody detection tests for *Brucella* were performed: *Brucella abortus* card, buffered acidified plate agglutination test (BAPA), rivanol, and complement fixation tests. These were performed by the Colorado Department of Agriculture, Lakewood, Colorado. All tests were done according to standard protocols established by the USDA's National Veterinary Service Laboratory.

## Toxoplasma gondii and Neospora caninum:

The  $\overline{T}$ . gondii modified agglutination test and *Neospora* agglutination test were performed by J.P. Dubey, USDA-Agricultural Research Service, Beltsville, Maryland. Methods and results have been previously published in Dubey et al. (2003).

## Sarcocystis sp.:

A direct agglutination test modified for *S. neurona* was performed by David S. Lindsay, Department of Biomedical Sciences and Pathobiology, Virginia-Maryland Regional College of Veterinary Medicine, Virginia Tech, Blacksburg, Virginia. The method is described in Lindsay and Dubey (2001).

## Caliciviruses:

Samples from 2001 were tested using an ELISA test with a recombinant antigen that detects antibody to nearly all known serotypes. The ELISA test was developed and performed in the laboratory of Alvin Smith, College of Veterinary Medicine, Oregon State University, Corvallis, Oregon, cited in Hanni et al. (2003). Samples from 2002 were tested using a serum (virus) neutralization test against serotypes 1, 2, 4, 5, 6, 7, 8, 13, 14; tests were performed at the USDA-National Veterinary Services Laboratory, Ames, Iowa.

In addition to serology, oral and fecal swabs were obtained, when appropriate, from the otters and samples were evaluated for parasites by diagnostic laboratories (Phoenix Central Laboratory, Everett, WA and Swedish Medical Center, Seattle, WA). Opportunistic urine samples were collected from two males in 2001 and one male in 2002. Urine samples were submitted to NMFS' Northwest Fisheries Science Center, Seattle, WA and analyzed for domoic acid. Samples were evaluated using the receptor binding assay and confirmed by a more precise method using High Performance Liquid Chromatography (HPLC). Lastly, 26 of the liver biopsies were fixed in neutral buffered formalin for histopathologic evaluations by a diagnostic laboratory at Purdue University, West Lafayette, IN. Samples were embedded in paraffin, sectioned and stained with hematoxylin and eosin for light microscopy.

#### **Blood Health Screens**

In addition to analysis for specific pathogens, sea otter health was assessed using routine blood chemistry screens. Whole blood was used to determine cell blood counts (CBC) and differentials at a veterinary diagnostic facility (Phoenix Central Laboratory, Everett, WA). Whole blood was centrifuged and the serum submitted for analysis of blood chemistry parameters at the same veterinary diagnostic laboratory.

## **Age Determination**

Cementum age analysis of sea otter teeth was conducted using standardized procedures for wildlife aging by Matson's Laboratory in Milltown, MT. Method is described in Matson (1980).

## **Fatty Acid Profiles**

Profiles of fatty acid concentrations were determined using fat samples collected during the live capture program (VanBlaricom et al. 2007). Samples from 26 of the captured animals were of sufficient quantity to allow for the analyses. Lipids were extracted following the procedures outlined in Folch et al. 1957, as modified by Iverson et al. 2001.

## **Prey Analyses**

As part of a concurrent project, NOAA's Olympic Coast National Marine Sanctuary (OCNMS) and a multi-agency and private sector team collected several species of marine invertebrates along the outer coast of Washington within sea otter range. Species collected included the shore crab *Hemigrapsus nudus*, mussels *Mytilus californianus* and *Mytilus edulis/trossulus*, the whelk Nucella lamellosa, and the limpet Tectura scutum. Chemical scans for organochlorines, aromatic hydrocarbons and metals were performed by NOAA's Environmental Assessment Program, Northwest Fisheries Science Center in Seattle, WA on composite tissue samples of each species from four different locations within the Washington sea otter range. In addition, NOAA's Status and Trends, Mussel Watch program surveyed its Cape Flattery, Washington site in 2000 and 2002, which is also within the Washington sea otter range. Besides the chemical scans indicated above, this project also measured butyltins. The invertebrate data presented are species of organisms that sea otters could forage on (Bowlby et al. 1988; Estes et al. 2003; VanBlaricom et al. 2007), including crab species observed in the scat samples from the live captured otters in this study and organisms mentioned as prey items in the ground-based tracking studies mentioned in Appendix 1 (Laidre and Jameson 2006). Analytical methods similar to those described for sea otter tissues were used for the analyses of the invertebrate samples.

#### **Data Analyses**

Summary statistics including mean, standard deviation, and maximum concentrations were determined for all detected analytes. Means were calculated using one-half the detection limit for values less than the detection limit and also without non-detects. Data are presented by gender and age for ease in viewing trends.

Metals				Organo-		Aliphatics 2001	Aromatics	
Live	Metals Beach-	Butyltins Live	Butyltins	chlorines	PCB Live	& 2002 Live	2001& 2002	Semi-volatiles
Capture	cast	Capture	Beach-cast	Beach-cast	Capture	Capture	Live Capture	Beach-cast
n = 30	<i>n</i> = 15	<i>n</i> = 30	<i>n</i> = <i>15</i>	<i>n</i> = 15	<i>n</i> = 30	n = 16	<i>n</i> = 16	<i>n</i> = 15
Whole								
Blood	Liver	Whole Blood	Liver	Liver	Whole Blood	Liver	Liver	Liver
Aluminum				PCB-				
(Al)	Aluminum (Al)	Dibutyltin	Dibutyltin	TOTAL	PCB1	n-docosane	Unresolved	1,2,3,4-
Arsenic								
(As)	Arsenic (As)	Monobutyltin	Monobutyltin		PCB8/5	n-eicosane	Complex	Tetrachlorobenzene
Boron (B)	Cadmium (Cd)		Tributyltin		PCB28	n-heptacosane	Mixture	1,2,4,5-
Barium			-			_		
(Ba)	Chromium (Cr)				PCB42/59/37	n-heptadecane		Tetrachlorobenzene
Cadmium								
(Cd)	Copper (Cu)				PCB44	n-hexacosane		
Copper								
(Cu)	Iron (Fe)				PCB49	n-hexadecane		
Iron (Fe)	Mercury (Hg)				PCB101/90	n-nonacosane		
Mercury								
(Hg)	Magnesium (Mg)				PCB105	n-nonadecane		
Magnesium								
(Mg)	Manganese (Mn)				PCB135	n-octacosane		
Lead (Pb)	Nickel (Ni)				PCB138/160	n-octadecane		
Selenium								
(Se)	Selenium (Se)				PCB153/132	n-pentacosane		
Strontium								
(Sr)	Strontium (Sr)				PCB175	n-pentadecane		
Vanadium								
(V)	Zinc (Zn)				PCB180	n-tetracosane		
Zinc (Zn)					PCB187	n-triacontane		

Table 2. Analytes Where Greater than 50 Percent of Samples Were Above Detection Limits

Analyses performed were chemical scans, thus a wide range of parameters were analyzed. Appendix 2 provides a full list of the parameters analyzed. Table 2 presents a list of the analytes for which greater than 50 percent of the results were above the detection limit to minimize the reporting of false positives.

Only those chemicals detected are presented in the data tables in the text. All results of detected concentrations are reported in the data tables regardless of their toxicity; however, only those considered to be of potential consequence are discussed. In general, the results are reported in ppm wet weight; however for metals, dry weight was also presented for ease in comparison to literature values. In some cases, analytes were summed to provide exposure to a class of compounds. Those summed include: butyltins, e.g., the sum of tetra-, tri-, di- and monobutyltins; total PAH; and several organochlorine pesticides were summed, e.g., total DDT (1,1,1,-trichloro-2,2-bis(p-chlorophyenyl))ethanes including p,p'-DDE, p,p'-DDD and p,p'-DDT); hexachlorocyclohexanes (HCH isomers); chlordanes (*trans*-chlordane, *cis*-chlordane, *trans*-nonachlor, *cis*-nonachlor and oxychlordane), and cyclohexanes (HCHs, e.g., alpha BHC, beta BHC, delta BHC and gamma BHC).

Statistical analyses beyond summary statistics were not performed on analytes unless 50 percent of the samples were above the detection limit. Prior to statistical analyses, data normality was tested using Shapiro-Wilk normality test and normal probability plots. Data that was found to be not normal were natural log, square root, or rankit transformed. Appendix 10 provides the normality test results and which contaminants were transformed. Statistical analyses were performed using SPSS® Base 10.0 statistical software (SPSS Inc.).

Stepwise multiple linear regression analysis in SPSS Software was used to determine if age, gender and body weight of sea otters were significant predictors of chemical concentrations, biomarker assays or positive pathogen titers. Gender was a categorical variable in the regressions. Regressions were run excluding non-detects. Significance of the overall regression was evaluated with the standard F-test with the probability of F to include a predictor at 0.50 and the probability of F to remove a predictor at 0.1. When more than one predictor is significant, the p-value for the partial t-test coefficient is also reported. If the only significant predictor was gender, the categorical variable, then the means for each gender are reported rather than the R<sup>2</sup> value. Tests were considered significant with an alpha level  $\leq 0.05$ . Estimated R<sup>2</sup> values, when significant, were considered strong only if the value was at least 0.70, meaning 70 percent of the relationship could be explained by the regression.

Correlations between chemical concentrations and positive pathogen titer or biomarker responses were evaluated using the nonparametric Spearman's rho correlation as were biomarkers responses in blood versus liver. Significance was evaluated at an alpha level of 0.05.

Analysis of Variance (ANOVA) was conducted to compare vitamin A levels in lactating versus non-lactating female otters. Significance was evaluated at an alpha level of 0.05.

Statistics performed using the fatty acid analyses focused on two fatty acids, linoleic and linolenic acid, because they cannot be endogenously synthesized by mammals, and therefore can only stem from dietary sources. Statistical analyses were performed by VanBlaricom et al. 2007

and are summarized here. Univariate analyses of variance were performed to compare mean values among classes within categories of age and weight classes, sex and capture location.

For the cell blood count and serum chemistry data, comparisons were made among Washington, Alaska and California sea otters using Tukey multiple comparisons, or t-tests when only two sets of data were available. Only summary statistics were available for Alaska and California studies, so approximate normality was assumed.

#### **IV. RESULTS**

#### **Live Captured Otters**

A total of thirty-two sea otters were captured during the 2001 and 2002 field seasons, of which 27 were adults or sub-adults and five were pups (Table 3). Samples were not taken from pups that weighed less than 10 kilograms (kg) and transmitters were not implanted in pups less than 15 kg in weight, as per permit requirements for this study (Appendix 4). One pup weighed more than 15 kg and was implanted, thus, 28 animals were implanted with radio-transmitters in an effort to track movements of individuals. Biological samples were collected from 30 animals as two were less than 10 kilograms. Of the animals captured, 21 were female and 11 were male, with samples collected from 20 females and 10 males (Table 3). Sea otter adults and sub-adults ranged from 2 to 16 years of age based on cementum layer analysis of pre-molars that were extracted during the capture procedures. Adult females ranged between 20 and 31 kg, while adult males ranged from 27 to 50.5 kg. The 50.5 kg male captured off Destruction Island, which is part of the Washington Maritime National Wildlife Refuge Complex, is the heaviest sea otter ever captured, to the best of our knowledge. Capture locations for each animal and general statistics are presented in Table 3. Body or nutritional condition was calculated using a ratio of weight(kg)/length(cm), revealing that these animals did not vary considerably in condition. The condition ratio for female adults ranged from 0.18 to 0.24, and with the single otter (WA106) excluded, the range was even tighter 0.18 to 0.22. Subadult females were similar 0.17 and 0.19. Adult males ranged 0.25 to 0.33 and subadult males from 0.21 to 0.23. The oldest male otter captured had the highest ratio, a 10 year old male, WA102.

#### **Beach-Cast Otters**

Liver samples were collected from 15 beach-cast (dead, stranded) sea otters between 1991 and 2002, with samples archived at the USGS National Wildlife Health Center. The information available on these animals is presented in Table 3. This sample set may be biased in that emphasis is placed on otters collected in 2000 during a mortality event and thus, may not represent general trends in mortality. Of these otters, ten are male and five are female. The nine individuals that were aged range from 2 to 13 years. Body or nutritional condition was calculated using a ratio of weight(kg)/length(cm), revealing that these animals were in poor condition as compared to the live captured animals, although the male with the highest ratio of weight to length, otter number 14717 (Table 3) was a beach cast animal. Cause of death of the beach cast animals reported herein is generally attributed to infectious disease (Table 3).

## **Analytical Results**

#### Overview

Results of the analyses are presented in tabular form with summary statistics including the mean calculated without non-detects, the mean calculated with one half the detection limit for non detects, standard deviation, and maximum concentrations. Only those analytes detected are presented in the data tables. Appendix 2 provides a full list of the parameters analyzed. A list of the contaminants detected in over 50 percent of the otter samples analyzed is presented in Table 2. Summary statistics are presented for each gender and for the sample population overall. Animal age is also provided and the data are presented from youngest to oldest otter within a gender, for ease in viewing gender and age differences in analyte concentrations. For the most part, no significant age nor gender biases were found, perhaps due to the small sample size and low contaminant residues measured. Because the condition or nutrition ratio (weight/length) did not vary considerably statistics were not performed based on condition ratio.

Whole blood samples from live captured animals had detectable concentrations of PCBs, heavy metals and butyltins. Liver samples from live captured animals had detectable concentrations of several of the aliphatic hydrocarbons, but only a few aromatic hydrocarbons were detected. Liver from beach-cast animals contained measurable concentrations of several metals, such as cadmium, copper and zinc, and several organochlorines including PCBs, chlordane and DDT metabolites. Below is a summary of the findings for each group of compounds.

## Metals

Whole blood from the live otter captures was analyzed for metals in both 2001 and 2002. In 2001, detection limits were much higher and only six metals were detected (Table 4); chromium, copper, iron, magnesium, selenium and zinc. Of these, iron and magnesium are of little toxicological concern.

With lower detection limits in 2002, 15 metals were detected in the whole blood of the live sea otters captured (Table 4), but mean concentrations of only six metals were greater than 1 ppm, and four of these (iron, magnesium, aluminum and boron) are of little toxicological concern.

Concentrations of heavy metals of potential toxicological concern – arsenic, cadmium, chromium, copper, lead, manganese, mercury, nickel, selenium, silver and zinc – were low for all samples, with only mean concentrations of selenium and zinc above 1 ppm. Three of the otters had whole blood concentrations of selenium of over 3 ppm. The oldest animal captured, a 16 year old female (WA110), had the highest whole blood concentration of zinc (4.43 ppm). Although a literature search was performed, no results for these compounds were found for comparison.

	a	~					-	Conditio	
Otter #	Study Year	Capture Location	Adult/ Pup	Sex	Age	Weight (kg)	Length (cm)	n Ratio (kg/cm)	Notes
Live Otte		2000000	- «P			(8)	(011)	(118, 0111)	
									Lactating, mother of
WA084	2001	Cape Alava	Adult	F	4	23.5	129.5	0.18	WA085
WA085	2001	Cape Alava	Pup	F	<1	3	NM		Pup of WA084
WA086	2001	Ozette River	Adult	F	5	21	120	0.18	Lactating, mother of WA087
WA087	2001	Ozette River	Pup	М	<1	5	NM		Pup of WA086
WA088	2001	Ozette River	Adult	F	13	22	124	0.18	
WA089	2001	Cape Alava	Adult	F	6	23	124	0.19	
WA090	2001	Cape Alava	Adult	F	9	24	125	0.19	
WA091	2001	Cape Alava	Adult	F	7	22	123	0.18	
WA092	2001	Cape Alava	Pup	М	<1	16	105	0.15	Pup of WA093
WA 002	2001	Come Alexa	A .d. 14	Б	10	27	124	0.20	Lactating, mother of
WA093	2001	Cape Alava	Adult	F F	10		134		WA092
WA094	2001	Cape Alava	Adult	F F	5 4	23 23	127	0.18	
WA095	2001 2001	Sand Point	Adult	г F		23	127	0.18	
WA096	2001	Sand Point Destruction	Adult	F	10	21	126	0.21	
WA097	2001	Island	Adult	М	8	41	134	0.31	
WA098	2001	Destruction Island	Subadult	М	3	30	130	0.23	
WA098 WA099	2001	Perkins Reef	Subadult	F	3	24	124	0.23	
WA099 WA100	2001	Perkins Reef	Adult	г F	4	24	124	0.19	
WA054	2001	Cape Alava	Adult	F	15	27	128	0.21	
WA034	2002	Destruction	Adult	Г	15	21	127	0.21	
WA102	2002	Island	Adult	М	10	50.5	151	0.33	
WA103	2002	Sand Point	Adult	F	9	23	126	0.18	
WA104	2002	Sand Point	Pup	М	<1	11	95	0.12	
WA105	2002	Destruction Island	Adult	М	5	38	142	0.27	
WA106	2002	Perkins Reef		F	6	31	131	0.24	
WA107	2002	Destruction Island	Adult	М	6	36.5	138	0.26	
WA 100	2002	Destruction	Call 1 1	м	2	21	122	0.22	
WA108	2002	Island	Subadult	М	3	31	132	0.23	
WA109	2002	Perkins Reef	Subadult	F	3	20	119	0.17	
WA110	2002	Perkins Reef Giants	Adult	F	16	25	125	0.20	
WA111	2002	Graveyard	Adult	F	4	26	128	0.20	
WA112	2002	Cape Johnson	Adult	М	6	37	146	0.25	
WA113	2002	Cape Johnson	Subadult	М	2	27	130	0.21	
WA114	2002	Destruction Island	Adult	F	6	30	135	0.22	
		Destruction							
WA115	2002	Island	Pup	F	<1	15	106	0.14	l

Table 3. General Information on Live Captured and Beach-Cast Sea Otters

Otter #	Year Collected	Stranding Location	Adult/Pup	Sex	Age	Weight (kg)	Length (cm)	Condition Ratio (kg/cm)	Cause of Death <sup>b</sup>
Beach-Cast Otters									
10385	1991	Cedar Creek	Adult	М	U	28	138.5	0.20	Infectious Disease
13827	1995	Cape Alava	Adult	F	U	24	NM		Other
13555	1995	Cape Alava	Adult	М	U	31.2	147	0.21	Infectious Disease
14325	1996	Yellow Banks	Adult	F	U	20	132	0.15	Infectious Disease
14717	1997	Bowman Creek	Adult	М	U	59	147	0.40	Other
15713-01	1998	Shipwreck Point	Adult	М	8	28.1	143	0.20	Infectious Disease
16904-01	2000	Roosevelt Beach	Adult	М	12	41.5	144	0.29	Infectious Disease
16961-01	2000	Ocean Park	Subadult	F	3	19.5	125	0.16	Infectious Disease
16961-02	2000	Pacific Beach	Pup	М	<1	15.1	114	0.13	Infectious Disease
16961-03	2000	Ocean City	Subadult	F	2	17	117	0.15	Infectious Disease
16961-04	2000	Roosevelt Beach	Subadult	М	2	23.4	143	0.16	Infectious
17058-01 <sup>a</sup>	2000	Ocean City	Adult	М	U	37.8	124	0.30	Infectious Disease
17315-01	2001	Beach 1	Adult	F	13	19.6	142	0.14	Emaciation
18124-01	2002	Pacific Beach	Subadult	М	3	19.6	141	0.14	Infectious Disease
18316-01	2002	Kalaloch	Subadult	М	1	22.2	137.5	0.16	Infectious Disease

Table 3. General Information on Live Captured and Beach-Cast Sea Otters; continued

U = unknown

NM= not measured

a. Approximate length and weight because carcass had no rear foot or tail. Also coded WA019.

b. Data from USGS-National Wildlife Center. One of five categories: emaciation, infectious disease, other causes (e.g., twisted gut, heart failure), trauma, undetermined. Infectious diseases include: protozoal meningoencephalitis (from both *Toxoplasma* and *Sarcocystis*), morbillivirus infection, leptospirosis and bacterial infections. Otters may have more than one disease.

weight)										
Otter	Gender	Age	Aluminum	Arsenic	Boron	Barium	Cadmium	Chromium	Copper	Iron
WA115	F	<1	1.6	0.135	2.04	1.23	0.00632	< 0.0571	0.871	641
WA099	F	3	< 20	< 0.4	< 1.6	< 4	< 0.08	< 0.4	0.712	620
WA109	F	3	1.25	0.116	1.62	0.943	0.00834	< 0.0402	0.95	712
WA084	F	4	< 20	< 0.4	< 1.6	< 4	< 0.08	< 0.4	0.695	545
WA095	F	4	< 20	< 0.4	< 1.6	< 4	< 0.08	< 0.4	0.838	520
WA100	F	4	< 20	< 0.4	< 1.6	< 4	< 0.08	0.421	0.686	559
WA111	F	4	0.924	0.124	1.19	0.708	0.00588	< 0.0328	0.845	625
WA086	F	5	< 20	< 0.4	< 1.6	< 4	< 0.08	< 0.4	0.725	548
WA094	F	5	< 20	< 0.4	< 1.6	< 4	< 0.08	< 0.4	1.18	583
WA089	F	6	< 20	< 0.4	< 1.6	< 4	< 0.08	< 0.4	0.823	533
WA106	F	6	0.874	0.134	1.11	0.603	0.007	< 0.0388	0.946	676
WA114	F	6	1.55	0.169	2.15	1.19	0.0102	< 0.0478	1.11	904
WA091	F	7	< 20	< 0.4	< 1.6	< 4	< 0.08	< 0.4	0.8	576
WA090	F	9	< 20	< 0.4	< 1.6	< 4	< 0.08	< 0.4	0.686	548
WA103	F	9	0.842	0.191	1.24	0.584	0.0205	< 0.0318	0.922	634
WA093	F	10	< 20	< 0.4	< 1.6	< 4	< 0.08	< 0.4	0.752	492
WA096	F	10	< 20	< 0.4	< 1.6	< 4	< 0.08	0.414	0.762	494
WA088	F	13	< 20	< 0.4	< 1.6	< 4	< 0.08	< 0.4	0.745	464
WA054	F	15	0.988	0.275	1.23	0.727	0.00565	< 0.0317	0.893	479
WA110	F	16	1.81	0.134	2.41	1.44	0.00929	< 0.0584	0.823	690
WA092	М	<1	< 20	< 0.4	< 1.6	< 4	< 0.08	< 0.4	0.724	533
WA104	М	<1	2.1	0.183	2.46	1.45	0.0144	< 0.0644	0.848	621
WA113	М	2	0.973	0.117	1.52	0.703	< 0.00190	< 0.038	0.967	660
WA098	М	3	< 20	< 0.4	< 1.6	< 4	< 0.08	0.465	0.875	555
WA108	М	3	0.788	0.12	1.23	0.594	< 0.00163	< 0.0326	0.844	657
WA105	М	5	0.798	0.114	1.27	0.578	< 0.00162	< 0.0325	0.805	646
WA107	M	6	0.931	0.109	1.19	0.725	< 0.00154	< 0.0308	0.854	631
WA112	M	6	0.895	0.112	1.52	0.658	< 0.00184	< 0.0369	0.951	631
WA097	М	8	< 20	< 0.4	< 1.6	< 4	< 0.08	0.59	0.903	482
WA102	М	10	1.1	0.138	1.37	0.76	< 0.00161	< 0.0322	0.859	630
Females	Mean <sup>a</sup>		6.492	0.184	1.13	1.571	0.0277	0.150	0.838	592
	Mean <sup>b</sup>		1.230	0.160	1.62	0.928	0.0091	0.418	0.838	592
n = 20	Std dev		4.414	0.038	0.52	0.574	0.0158	0.126	0.135	102
	Max		1.81	0.275	2.41	1.44	0.0205	0.421	1.18	904
Males	Mean <sup>a</sup>		3.759	0.149	1.30	1.147	0.0139	0.139	0.863	605
	Mean <sup>b</sup>		1.084	0.128	1.51	0.781	0.0144	0.528	0.863	605
n = 10	Std dev		4.440	0.041	0.27	0.656	n/a	0.252	0.046	63
	Max		2.1	0.183	2.46	1.45	0.0144	0.59	0.967	660
All	Mean <sup>a</sup>		5.581	0.172	1.19	1.430	0.0231	0.146	0.846	596
	Mean <sup>b</sup>		1.162	0.145	1.57	0.860	0.0097	0.473	0.846	596
n = 30	Std dev		4.504	0.042	0.51	0.619	0.0177	0.157	0.116	90
	Max		2.1	0.275	2.46	1.45	0.0205	0.59	1.18	904
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 Table 4. Detectable Metals in Whole Blood of Live Captured Sea Otters 2001-2002 (ppm, wet weight)

a. mean calculated using 1/2 detection limit for non-detects; b. mean calculated without non-detects; Metals below detection limits: Ag, Be, Mo, Ni

 Table 4. Detectable Metals in Whole Blood of Live Captured Sea Otters 2001-2002 (ppm, wet weight); continued

Otter	Gender	Age	Mercury	Magnesium	Manganese	Lead	Selenium	Strontium	Vanadium	Zinc
WA115	F	<1	0.0257	57.6	< 0.114	0.0212	0.927	0.114	0.0782	2.65
WA099	F	3	< 0.16	47.8	< 0.8	< 0.4	3.5	< 0.4	< 0.4	2.91
WA109	F	3	0.0041	53.7	< 0.0800	0.00927	2.06	0.0668	0.0539	3.66
WA084	F	4	< 0.16	48.6	< 0.8	< 0.4	1.78	< 0.4	< 0.4	2.69
WA095	F	4	< 0.16	48	< 0.8	< 0.4	1.36	< 0.4	< 0.4	2.71
WA100	F	4	< 0.16	43.9	< 0.8	< 0.4	2.39	< 0.4	< 0.4	2.47
WA111	F	4	0.0414	53.4	< 0.0660	0.0071	2.05	0.0737	0.0435	3.22
WA086	F	5	< 0.16	49	< 0.8	< 0.4	1.48	< 0.4	< 0.4	2.58
WA094	F	5	< 0.16	45.8	< 0.8	< 0.4	1.43	< 0.4	< 0.4	2.77
WA089	F	6	< 0.16	46.8	< 0.8	< 0.4	1.4	< 0.4	< 0.4	2.43
WA106	F	6	0.0628	54.4	0.22	0.0146	2.15	0.0536	0.0542	3.57
WA114	F	6	0.0988	63.2	< 0.0960	0.0173	2.62	0.0727	0.0638	4.03
WA091	F	7	< 0.16	50.9	< 0.8	< 0.4	1.63	< 0.4	< 0.4	3.25
WA090	F	9	< 0.16	47.9	< 0.8	< 0.4	1.4	< 0.4	< 0.4	2.74
WA103	F	9	0.125	52.3	< 0.0640	0.00583	0.671	0.102	0.0458	2.75
WA093	F	10	< 0.16	46.7	< 0.8	< 0.4	1.89	< 0.4	< 0.4	2.46
WA096	F	10	< 0.16	44.7	< 0.8	< 0.4	2.57	< 0.4	< 0.4	2.65
WA088	F F	13 15	< 0.16	48.2	< 0.8	< 0.4	2.34	< 0.4	< 0.4	2.26
WA054 WA110	F F	15	0.0719 0.0074	48.6 55.7	< 0.0630 < 0.117	0.00432 0.00901	1.68 2.71	0.167	0.0453 0.0775	2.43 4.43
WA110 WA092	г М	<1	< 0.16	47.7	< 0.117	< 0.4	0.897	0.0678	< 0.4	2.49
WA092 WA104	M	<1	0.0287	54.8	< 0.129	0.0109	0.435	0.17	0.0923	2.49
WA104 WA113	M	2	0.0237	51	< 0.0760	0.00762	1.81	0.0701	0.0499	3.83
WA098	M	3	< 0.16	46.6	< 0.8	< 0.4	4.04	< 0.4	< 0.4	2.95
WA108	M	3	0.0591	50.5	< 0.0650	0.00667	2.85	0.0713	0.0437	3.4
WA105	M	5	0.0594	48.3	< 0.0650	0.00647	4.4	0.0449	0.0596	3.38
WA107	M	6	0.0776	50	< 0.0620	0.00574	1.96	0.0518	0.0421	3.18
WA112	М	6	0.0603	47.6	< 0.0740	0.00716	1.58	0.0392	0.0507	3.47
WA097	М	8	< 0.16	47.5	< 0.8	< 0.4	3.66	< 0.4	< 0.4	2.78
WA102	М	10	0.154	53.4	< 0.0640	0.00731	2.16	0.0496	0.0412	3.01
Females	Mean <sup>a</sup>		0.0699	50.4	0.27	0.1244	1.90	0.156	0.1431	2.93
	Mean <sup>b</sup>		0.0546	50.4	0.22	0.0111	1.90	0.090	0.0578	2.93
n = 20	Std dev		0.0292	4.8	n/a	0.0950	0.67	0.060	0.0720	0.58
	Max		0.125	63.2	0.22	0.0212	3.5	0.167	0.0782	4.43
Males	Mean <sup>a</sup>		0.0761	49.7	n/a	0.0652	2.38	0.110	0.0980	3.14
	Mean <sup>b</sup>		0.0744	49.7	n/a	0.0074	2.38	0.071	0.0542	3.14
n = 10	Std dev		0.0335	2.3	n/a	0.0943	1.10	0.073	0.0747	0.26
	Max		0.154	54.8	0	0.0109	4.4	0.17	0.0923	3.83
All	Mean <sup>a</sup>		0.0719	50.2	0.23	0.1047	2.06	0.140	0.1281	3.00
	Mean <sup>b</sup>		0.0639	50.2	0.22	0.0094	2.06	0.081	0.0561	3.00
n = 30	Std dev		0.0297	4.2	n/a	0.0970	0.94	0.067	0.0740	0.53
	Max	1 (2, 1	0.154	63.2	0.22	0.0212	4.4	0.17	0.0923	4.43

a. mean calculated using 1/2 detection limit for non-detects; b. mean calculated without non-detects; Metals below detection limits: Ag, Be, Mo, Ni

In general, metals concentrations in whole blood of live captured otters did not show a significant relationship to age or gender. There were several statistically significant relationships between animal body weight and metal concentrations: mercury (p = 0.013,  $R^2 = 0.391$ ), selenium ( $p = 0.016 R^2 = 0.189$ ), strontium (p = 0.001,  $R^2 = 0.617$ ), and vanadium (p = 0.017,  $R^2 = 0.364$ ). Again, in all cases except strontium, although statistically significant the relationship was not strong, as indicated by the low adjusted r-squared values.

Liver samples from the 15 beach-cast otters were analyzed for metals and are presented as both wet weight (Table 5) and dry weight (Table 6) for comparison with literature values. There was considerable variability in the results as illustrated by the large standard deviations for several of the metals. In some cases this variability could be explained by one or two individuals. Of note is the wide range in aluminum (<5.0 to 198 ppm dw), copper (15.3 to 191 ppm, dw) and zinc concentrations (100 to 258 ppm, dw). The 2 year old male (16961-04) had the highest concentrations of copper (191 ppm, dw), mercury (41.2 ppm, dw) and selenium 18.3 ppm, dw), the 3 year old male (18124-01) had elevated manganese and zinc in the liver, and male sea otter 13555 had more than double the liver concentration of aluminum than any of the other animals. Copper concentrations in the beach-cast animals cannot be compared to those in the live captured animals because of the difference in sample matrix, liver versus whole blood, respectively. Several metals of toxicological significance exceeded a mean of 10 ppm in the liver of the beach-cast animals, including aluminum, cadmium, copper, mercury, manganese and zinc.

Regression analysis to evaluate relationships between age, gender (body weights were not known) and metal concentrations in the liver of beach-cast animals revealed one significant relationship. Cadmium concentrations were significantly higher in female animals (p = 0.047, mean for females of 6.03 ppm ww, mean for males of 2.03 ppm ww; Table 5).

## Organotins

Whole blood samples from the 2001 and 2002 live captured animals were analyzed for butyltins (i.e., tetra-, tri-, di- and monobutyltin) (Table 7). No tetrabutyltin was detected and tributyltin (TBT), the more toxic moiety, was detected in only one 10-year old male animal (WA102). Regression analysis revealed no significant relationships between age, gender or body weight and total butyltin concentration in whole blood. Total butyltin concentrations ranged from non-detect to 0.427 ppm (wet weight), with a mean of 0.105 ppm (Table 7).

Butyltins were also measured in liver from fifteen beach-cast otters from the Washington coast (Table 8). Total butyltin liver concentrations in beach-cast Washington sea otters was similar to that in whole blood of live captured animals, ranging from 0.0553 to 0.447 ppm wet weight; however, unlike whole blood results for live captured animals, the concentration of tributyltin in liver of beach-cast animals was above the detection limit for all but two animals. Regression analysis revealed no significant relationship between age or gender and total butyltin concentration in these samples.

								tterb (p	<u>pm,</u>	8										
Otter	Age	Gender	Silver	Aluminum	Arsenic	Boron	Cadmium	Chromium	Copper	Iron	Mercury	Magnesium	Manganese	Molybdenum	Nickel	Lead	Selenium	Strontium	Vanadium	Zinc
16961-03	2	F	NA	7.64	< 0.170	< 0.681	4.73	0.188	25.8	257	1.9	202	3.5	< 0.681	< 0.170	< 0.170	1.88	0.714	< 0.170	34.1
16961-01	3	F	NA	30.9	0.209	< 0.691	3.13	< 0.173	22.3	199	1.22	279	2.53	< 0.691	< 0.173	< 0.173	1.68	1.34	< 0.173	38
17315-01	13	F	NA	1.49	0.405	< 0.563	4.93	0.228	50.3	540	4.64	173	2.78	< 0.563	0.335	< 0.141	3.31	< 0.141	< 0.141	38.5
14325	U	F	NA	13.6	0.158	< 0.564	16.6	0.344	28	293	5.44	573	5.8	< 0.564	0.337	< 0.141	2.2	1.09	< 0.141	43.6
13827	U	F	NA	17	0.336	< 0.661	5.25	0.226	26.7	342	0.505	188	2.73	< 0.661	0.355	< 0.165	1.22	0.382	< 0.165	36.4
16961-02	<1	Μ	NA	5.67	0.275	< 0.527	0.717	< 0.132	32.9	461	3.48	323	2.99	< 0.527	< 0.132	< 0.132	1.77	0.711	< 0.132	51.2
16961-04	2	Μ	NA	14	0.269	< 0.644	2.07	< 0.161	61.6	438	13.2	273	4.8	< 0.644	< 0.161	< 0.161	5.87	0.482	< 0.161	71.3
18124-01	3	Μ	NA	< 1.43	0.472	< 0.570	2.51	< 0.143	20.7	682	2.45	166	6.18	< 0.570	< 0.143	< 0.143	3.24	< 0.143	< 0.143	73.4
15713-01	8	Μ	NA	5.08	0.376	< 0.669	1.36	0.337	9.49	545	0.968	212	2.87	< 0.669	0.411	< 0.167	0.672	< 0.167	0.663	48.7
14717	U	Μ	NA	11.1	0.349	< 0.531	3.89	0.338	4.06	154	0.444	157	3.68	< 0.531	0.436	< 0.133	0.446	< 0.133	< 0.133	48.4
16904-01	12	Μ	NA	20	< 0.131	< 0.525	0.624	0.159	16.8	288	4.89	108	1.38	< 0.525	< 0.131	< 0.131	3.58	0.163	< 0.131	28.1
17058-01	U	Μ	NA	5.65	< 0.161	< 0.643	1.57	< 0.161	35.7	643	5.63	256	2.33	< 0.643	< 0.161	< 0.161	3.76	< 0.161	< 0.161	56.8
13555	U	Μ	NA	58.3	0.382	< 0.589	1.13	0.319	37.6	499	5.35	217	3	< 0.589	0.285	< 0.147	4.02	0.264	< 0.147	44.2
10385	U	Μ	NA	< 1.2	0.131	< 0.481	5.39	0.167	9.78	285	0.338	163	3.02	< 0.481	0.483	< 0.120	0.456	< 0.120	< 0.120	40.7
18316-01	1	Μ	0.0519	0.853	0.469	1.14	1	< 0.106	6.53	662	1.19	183	5.45	0.231	< 0.106	0.0437	3.98	0.296	0.166	38.4
Females	Mean <sup>a</sup>		-	14.1	0.24		6.9	0.21	30.6	326	2.7	283	3.47		0.240		2.06	0.719		38.1
n = 5	Mean <sup>b</sup>			14.1	0.28		6.93	0.25	30.6	326	2.7	283	3.47		0.342		2.06	0.882		38.1
	Std dev			11.1	0.11		5.5	0.07	11.2	130	2.2	167	1.35		0.011		0.78	0.421		3.5
	Max			30.9	0.41		16.6	0.34	50.3	540	5.4	573	5.80		0.355		3.31	1.340		43.6
Males	Mean <sup>a</sup>			12.2	0.29		2.03	0.17	23.5	466	3.8	206	3.57		0.203		2.78	0.228		50.1
n = 10	Mean <sup>b</sup>			15.1	0.34		2.0	0.26	23.5	466	3.8	206	3.57		0.404		2.78	0.383		50.1
	Std dev			18.5	0.11		1.54	0.09	18.2	178	3.9	64	1.48		0.085		1.85	0.217		14.1
	Max			58.3			5.4	0.34	61.6	682	13.2	323	6.18		0.483		5.87	0.711	0.663	73.4
All	Mean <sup>a</sup>			12.8	0.27		3.7	0.18	25.9	419	3.4	232	3.54		0.215		2.54	0.392		46.1
n = 15	Mean <sup>b</sup>			14.7	0.32		3.7	0.26	25.9	419	3.4	232	3.54		0.377		2.54	0.605		46.1
	Std dev			15.5	0.11		4.0	0.08	16.2	173	3.4	110	1.39		0.069		1.58	0.399		12.9
	Max		0.0519	58.3		1.14	16.6	0.34	61.6	682	13.2	573	6.18	0.231	0.483	0.0437		1.340	0.663	73.4
L							-					-	-	-		_		-		

## Table 5. Detectable Metals in Liver of Beach-Cast Sea Otters (ppm, wet weight)

a. mean calculated using 1/2 detection limit for non-detects; b. mean calculated without non-detects; Metals analyzed that were below detection limit: Ba, Be; units: ppm; Sample matrix: liver; n=15

Otter	Gender	Age	Silver	Aluminum	Arsenic	Boron	Cadmium	Chromium	Copper	Iron	Mercury	Magnesium	Manganese	Molybdenum	Nickel	Lead	Selenium	Strontium	Vanadium	Zinc
16961-03	F	2	NA	22.4	< 0.500	< 2.00	13.9	0.552	75.9	755	5.57	592	10.3	< 2.00	< 0.500	< 0.500	5.53	2.1	< 0.500	100
16961-01	F	3	NA	89.6	0.605	< 2.00	9.06	< 0.500	64.5	575	3.54	807	7.34	< 2.00	< 0.500	< 0.500	4.87	3.89	< 0.500	110
17315-01	F	13	NA	5.31	1.44	< 2.00	17.5	0.811	179	1920	16.5	613	9.89	< 2.00	1.19	< 0.500	11.8	< 0.5	< 0.500	137
14325	F	U	NA	48.3	0.559	< 2.00	58.9	1.22	99.3	1040	19.3	2030	20.6	< 2.00	1.19	< 0.500	7.81	3.88	< 0.500	154
13827	F	U	NA	51.3	1.02	< 2.00	15.9	0.685	80.7	1040	1.53	568	8.25	< 2.00	1.07	< 0.500	3.7	1.16	< 0.500	110
16961-02	М	<1	NA	21.5	1.05	< 2.00	2.72	< 0.500	125	1750	13.2	1220	11.3	< 2.00	< 0.500		6.72	2.7	< 0.500	194
16961-04	М	2	NA	43.6	0.835	< 2.00	6.42	< 0.500	191	1360	41.2	847	14.9	< 2.00	< 0.500		18.3	1.5	< 0.500	221
18124-01	М	3	NA	< 5.00	1.66	< 2.00	8.81	< 0.500	72.4	2390	8.58	581	21.7	< 2.00	< 0.500	< 0.500	11.4	< 0.5	< 0.500	258
15713-01	М	8	NA	15.2	1.12	< 2.00	4.07	1.01	28.4	1630	2.89	633	8.58	< 2.00	1.23	< 0.500	2.01	< 0.5	1.98	
14717	М	U	NA	41.9	1.31	< 2.00	14.6	1.27	15.3	579	1.67	592	13.9	< 2.00	1.64	< 0.500	1.68	< 0.5	< 0.500	182
16904-01	М	12	NA	76.1	< 0.500	< 2.00	2.38	0.607	63.9	1100	18.6	412	5.25	< 2.00	< 0.500	< 0.500	13.6	0.622	< 0.500	107
17058-01	М	U	NA	17.6	< 0.500	< 2.00	4.87	< 0.500	111	2000	17.5	795	7.26	< 2.00	< 0.500	< 0.500	11.7	< 0.5	< 0.500	177
13555	М	U	NA	198	1.3	< 2.00	3.85	1.08	128	1700	18.2	736	10.2	< 2.00	0.97	< 0.500	13.7	0.897	< 0.500	150
10385	М	U	NA	< 5.00	0.545	< 2.00	22.4	0.694	40.7	1190	1.41	680	12.6	< 2.00	2.01	< 0.500	1.9	< 0.5	< 0.500	170
18316-01	M	1	0.177	2.91	1.6	3.9	3.42	< 0.361	22.3	2260	4.05	624	18.6	0.79	< 0.361	0.149	13.6	1.01	0.565	131
	Mean <sup>a</sup>			43.4	0.77		23.1	0.70	99.9	1066	9.3	922	11.3				6.7	2.3		122
n = 5	Mean <sup>b</sup>			50.6	0.83		6.90	0.58	104.1	1108	16.4	776	9.8				9.8	2.2		
	Std dev			32.1	0.46		20.3	0.36	46.0	516.8	8.1	627	5.3				3.2	1.6		
	Max			89.6	1.44		58.9	1.22	179	1920	19.3	2030	20.6				11.8	3.9		
Males	Mean <sup>a</sup>			42.2	0.99		7.35	0.58	79.8	1596	12.7	712	12.4		0.73		9.5	0.8		
n = 10	Mean <sup>b</sup>			47.6	1.17		15.4	0.97	77.7	1575	9.2	785	13.2		1.33		7.9	1.7		101
	Std dev			59.5	0.51		6.44	0.41	57.4	554.9	12.2	216	5.1		0.69		6.0	0.8		44
	Max			198	1.66		22.4	1.27	191	2390	41.2	1220	21.7		2.01		18.3	2.7	1.98	
All	Mean <sup>a</sup>			42.6	0.92		12.6	0.62	86.5	1419	11.6	782	12.0		0.75		8.6	1.3		
n = 15	Mean <sup>b</sup>			48.7	1.09		12.6	0.88	86.5	1419	11.6	782	12.0		1.33		8.6	2.0		100
	Std dev			50.7	0.49		14.2	0.38	53.1	584	10.8	391	5.0		0.61		5.2	1.3		45
	Max		0.177	<b>198</b>	1.66	3.9	58.9	1.27	191	2390	41.2	2030	21.7	0.79	2.01	0.149	18.3	3.9	1.98	258

## Table 6. Detectable Metals in Liver of Beach-Cast Sea Otters (ppm, dry weight)

a. mean calculated using 1/2 detection limit for non-detects; b. mean calculated without non-detects; Metals analyzed that were below detection limit: Ba, Be; U = unknown

Otters 20	01-2002	(hhu	, wet weight)			
Otter	Gender	Age	Monobutyltin	Dibutyltin	Tributyltin	<b>Total BTs</b> <sup>c</sup>
WA115	F	<1	0.141	0.0302	< 0.010	0.1762
WA099	F	3	0.0162	0.0194	< 0.010	0.0406
WA109	F	3	0.0231	< 0.010	< 0.010	0.0331
WA084	F	4	0.0260	0.0373	< 0.010	0.0683
WA095	F	4	0.0950	0.0325	< 0.010	0.132
WA100	F	4	0.0321	0.0153	< 0.010	0.0524
WA111	F	4	0.172	0.120	< 0.010	0.297
WA086	F	5	< 0.010	< 0.010	< 0.010	0.0150
WA094	F	5	0.0366	< 0.010	< 0.010	0.0466
WA089	F	6	0.0295	0.0146	< 0.010	0.0491
WA106	F	6	0.0180	0.0106	< 0.010	0.0336
WA114	F	6	0.0796	0.0196	< 0.010	0.104
WA091	F	7	0.0155	0.0150	< 0.010	0.0355
WA090	F	9	< 0.010	< 0.010	< 0.010	0.0150
WA103	F	9	0.0146	< 0.010	< 0.010	0.0246
WA093	F	10	0.0153	< 0.010	< 0.010	0.0253
WA096	F	10	0.0261	0.0218	< 0.010	0.0529
WA088	F	13	0.0163	< 0.010	< 0.010	0.0263
WA054	F	15	0.103	0.135	< 0.010	0.243
WA110	F	16	0.292	0.130	< 0.010	0.427
WA092	М	<1	0.0127	0.0119	< 0.010	0.0296
WA104	М	<1	0.0506	0.0255	< 0.010	0.0811
WA113	М	2	0.0645	0.0488	< 0.010	0.118
WA098	М	3	0.0616	< 0.010	< 0.010	0.0716
WA108	М	3	0.0785	0.0575	< 0.010	0.141
WA105	М	5	0.0129	< 0.010	< 0.010	0.0229
WA107	М	6	0.0208	0.0520	< 0.010	0.0778
WA112	М	6	0.179	0.0744	< 0.010	0.258
WA097	М	8	0.0217	< 0.010	< 0.010	0.0317
WA102	М	10	0.246	0.154	0.0140	0.414
Females	Mean <sup>a</sup>		0.0581	0.0318		0.0949
n = 20	Mean <sup>b</sup>		0.0640	0.0463		0.0949
_	Std dev		0.0725	0.0428		0.1103
	Max		0.292	0.135		0.427
Males	Mean <sup>a</sup>		0.0748	0.0439		0.1246
n = 10	Mean <sup>b</sup>		0.0748	0.0606		0.1246
	Std dev		0.0778	0.0462		0.1233
	Max		0.246	0.154	0.014	0.414
All	Mean <sup>a</sup>		0.0637	0.0358		0.1048
n = 30	Mean <sup>b</sup>		0.0679	0.0513		0.1048
	Std dev		0.0734	0.0436		0.1136
	Max		0.292	0.154	0.014	0.427
·	aulated usi	n = 1/2	latastion limit for			

 Table 7. Detectable Butyltins (BT) in Whole Blood of Live Captured Sea

 Otters 2001-2002 (ppm, wet weight)

a. mean calculated using 1/2 detection limit for non-detects; b. mean calculated without non-detects; c. Total BT calculated using 1/2 detection limit for non detects; No tetrabutyltin was detected in any samples analyzed; Detection limit for each butyltin = 0.010 ppm.

Otter	Gender	Age	Monobutyltin	Dibutyltin	Tributyltin	Total BT <sup>c</sup>
16961-03	F	2	0.0183	0.0228	0.052	0.0931
16961-01	F	3	0.0188	0.0472	0.0884	0.154
17315-01	F	13	0.141	0.208	0.0348	0.384
14325	F	U	0.0672	0.0516	0.0763	0.195
13827	F	U	0.117	0.0708	0.0224	0.21
16961-02	М	<1	0.017	0.0207	0.0574	0.0951
16961-04	М	2	0.0463	0.07	0.0795	0.196
18124-01	М	3	0.183	0.162	0.102	0.447
15713-01	М	8	0.0451	0.0527	< 0.0180	0.107
14717	М	U	0.0338	0.0145	< 0.0140	0.0553
16904-01	М	12	0.0494	0.0613	0.0605	0.171
17058-01	М	U	0.0577	0.107	0.0481	0.213
13555	М	U	0.104	0.0541	0.0973	0.255
10385	М	U	0.0851	0.0553	0.0251	0.166
18316-01	М	1	0.103	0.137	0.0268	0.267
Females	mean <sup>a</sup>		0.0725	0.0801	0.0548	0.207
n = 5	mean <sup>b</sup>		0.0725	0.0801	0.0548	0.2072
	Std dev		0.056	0.0735	0.0276	0.109
	max		0.141	0.208	0.0884	0.384
Males	mean <sup>a</sup>		0.0724	0.0735	0.0513	0.197
n = 10	mean <sup>b</sup>		0.0724	0.0735	0.0621	0.1972
	Std dev		0.0484	0.0478	0.0344	0.111
	max		0.183	0.162	0.102	0.447
all	mean <sup>a</sup>		0.0724	0.0757	0.0524	0.201
n = 15	mean <sup>b</sup>		0.0724	0.0757	0.0593	0.2006
	Std dev		0.049	0.055	0.0313	0.106
	max		0.183	0.208	0.102	0.447

 Table 8. Detectable Butyltins (BT) in Liver of Beach-Cast Sea Otters (ppm, wet weight)

a. mean calculated using 1/2 detection limit for non-detects; b. mean calculated without nondetects; c. sum calculated using 1/2 detection limit for non-detects; No tetrabutyltin was detected in any samples analyzed; U = unknown

### **Organic Compounds**

### **Aliphatic and Aromatic Hydrocarbons**

Liver of 16 of the 2001 (n=7) and 2002 (n=9) live captured animals was evaluated for aliphatic and polycyclic aromatic hydrocarbons (PAHs) (Tables 9 and 10). The detection limit varies by sea otter due to the amount of tissue available for analysis, ranging from 0.658 to 3.57 ppm, wet weight for aliphatics and from 0.068 to 1.360 ppm wet weight for PAHs. Several of the aliphatics were detected in more than 50 percent of the live captured otters. Aliphatics with the highest concentrations - [nhexadecane (maximum concentration of 436 ppm, standard deviation of 134), nnonacosane (maximum concentration of 83.6 ppm, standard deviation of 24.7), noctacosane (maximum concentration of 99.3 ppm, standard deviation of 31.3)] - also showed considerable variability with large standard deviations (Table 9). Elevated concentrations were particularly apparent for a 3 year old female otter (WA099), which not only had the highest number of maximum concentrations of aliphatic compounds but also had detectable concentrations of more of the aliphatic compounds than any other otter. A 5 year old female (WA086) also had maximum concentrations of numerous of the aliphatics. Regardless, regression analysis revealed only one significant gender relationship to aliphatic concentrations. Nonacosane was significantly higher in females versus males (p = 0.048, mean concentration for females is 54.1 ppm ww, mean for males is 29.9 ppm ww).

As expected, very few PAHs were detected in liver of live animals, and only unresolved complex mixture were detected in at least 50 percent of the animals evaluated (Table 10). Maximum concentrations of the individual aromatic hydrocarbons ranged from non-detect to 2.28 ppm. As with the aliphatics, female sea otter WA099 had some of the highest PAH concentrations observed, primarily naphthalenes (naphthalene, C1-naphthalene and 2-methylnaphthalene) (Table 10). Regression analysis revealed no significant gender, age or body weight relationship to PAH concentrations.

#### **Semi-volatile Compounds**

Semi-volatile compounds were detected in only one whole blood sample from 30 live captured animals: 1,2,3,4-tetrachlorobenzene, which measured 0.00151ppm in sea otter WA094.

Two semi-volatile compounds were also determined in liver samples from eight of the 15 beach-cast otters (Table 11): 1,2,3,4-tetrachlorobenzene (mean of 0.00907 ppm) and 1,2,3,5-tetrachlorobenzene (mean of 0.0255 ppm). Regression analysis revealed no significant gender, body weight or age relationship to concentrations of these semi-volatiles.

Otter	Gender	Age	Detection Limits	n-decane (C10)	n-docosane (C22)	n-dodecane (C12)	n- dotriacontane (C32)	n-eicosane (c20)	n-heneicosane (C21)	n- hentricontane (C31)	n- heptacosane (C27)	n- heptadecane (C17)	n-hexacosane (C26)	n-hexadecane (C16)	n-nonacosane (C29)	n-nonadecane (C19)	n-octacosane (C28)	n-octadecane (C18)
WA099	F	3	2.38	8.72	5.6	9.54	4.01	9.69	5.56	<2.38	18.7	16.6	17.5	436	80.5	15.4	19.6	15.5
WA109	F	3	3.45	< 3.45	< 3.45	< 3.45	< 3.45	< 3.45	< 3.45	4.03	7.17	< 3.45	8.02	< 3.45	34.8	< 3.45	6.8	< 3.45
WA095	F	4	1.16	<1.16	2.01	2.21	1.26	12.3	5.03	<1.16 <3.03	6.61	16.1	5.18	167	47.4	13.3	7.97	15 26.7
WA086	F	5	3.03	< 3.03 < 0.676	4.03 <0.676	5.07 <0.676	<3.03 <0.676	20.7 <0.676	7.32 <0.676	< 3.03	12 <0.676	27.3 0.933	9.94 0.747	203 0.796	8.28 68.3	21.5 <0.676	12.5 78.5	0.742
WA114 WA091	F F	6 7	0.68 0.66	< 0.678	< 0.070	< 0.070	< 0.678	< 0.070	< 0.070	< 0.658	< 0.070	4.94	1.65	64.8	39.5	< 0.070 4.57	2.08	4.67
WA091 WA090	г F	9	1.04	< 1.04	1.17	2.03	<1.04	5.35	2.95	< 1.04	3.76	4.94 9.54	3.49	136	67.7	8.08	5.35	8.9
WA050 WA054	F	15	2.78	<2.78	4.07	<2.78	<2.78	3.3	<2.78	3.5	5.05	4.13	5.97	2.86	56.7	5.43	70.1	5.35
WA110	F	16	0.71	< 0.714	0.947	< 0.714	1.06	< 0.714	< 0.714	1.06	1.73	< 0.714	1.92	< 0.714	83.6	< 0.714	99.3	< 0.714
WA113	М	2	0.81	< 0.806	< 0.806	< 0.806	0.982	< 0.806	< 0.806	1.2	1.68	< 0.806	1.89	< 0.806	22.6	< 0.806	27	< 0.806
WA098	М	3	2.17	<2.17	3.17	4.94	2.8	3.23	<2.17	<2.17	12.4	9.46	10.4	301	12.1	4.22	12.5	5.72
WA108	М	3	1.72	<1.72	<1.72	<1.72	<1.72	2.24	<1.72	<1.72	<1.72	3.45	1.93	2.67	43.8	3.56	53.6	3.6
WA105	М	5	1.61	< 1.61	<1.61	< 1.61	< 1.61	< 1.61	<1.61	<1.61	<1.61	2.36	<1.61	1.79	28.7	2.45	34.7	2.48
WA107	М	6	3.57	<3.57	< 3.57	<3.57	<3.57	<3.57	<3.57	<3.57	<3.57	6.2	4.07	4.63	64.1	4.74	76	5.7
WA112	М	6	1.35	<1.35	<1.35	<1.35	<1.35	<1.35	<1.35	<1.35	<1.35	<1.35	<1.35	<1.35	29.4	<1.35	33.1	<1.35
WA097	М	8	1.79	6.05	4.78	3.74	2.54	11	5.55	<1.79	9.22	11.6	7.33	212	8.63	13.6	9.87	12.6
Females	Mean	a			2.38	2.7	1.35	6.68	3.02	1.45	6.35	9.07	6.05	113	54.1	7.86	33.6	8.77
n = 9	Mean	b			2.77	4.09	2.11	9.61	3.99		7.1	11.36	6.05	144	54.1	11.4	33.6	11
	Std de	ev			1.76	2.92	1.12	6.66	2.45	1.38	5.83	9.19	5.27	145	24.2	7.41	37.9	8.8
	Max			8.72	5.6	9.54	4.01	20.7	7.32	4.03	18.7	27.3	17.5	436	83.6	21.5	99.3	26.7
Males	Mean				1.78	1.89	1.49	2.88			3.92	4.88	3.87	74.7	29.9	4.24	35.3	4.45
n = 7	Mean	b			3.98	4.34	2.11	5.49			7.77	6.61	5.12	104	29.9	5.71	35.3	6.02
	Std de	ev			1.63	1.76	0.88	3.72			4.82	4.36	3.7	127	19.1	4.45	23.2	4.18
	Max			6.05	4.78	4.94	2.8	11	5.55	1.79	12.4	11.6	10.4	301	64.1	13.6	76	12.6
All	Mean	a		1.7	2.12	2.34	1.41	5.01	2.4	1.27	5.28	7.24	5.09	96	43.5	6.27	34.31	6.88
n = 16	Mean			7.39	3.04	4.16	2.11	8.24	3.87		7.28	9.38	5.72	128	43.5	8.8	34.31	8.91
	Std de	ev		2.33	1.68	2.44	1	5.75	2.24	1.05	5.38	7.57	4.64	134	24.7	6.38	31.32	7.29
	Max			8.72	5.6	9.54	4.01	20.7	7.32	4.03	18.7	27.3	17.5	436	83.6	21.5	99.3	26.7

Table 9. Detectable Aliphatic Compounds in Liver of Live Captured Sea Otters (ppm, wet weight)

a. mean calculated using 1/2 detection limit for non-detects; b. mean calculated without non-detects; Aliphatics analyzed that were below detection limit: n-hentriacontane & n-tritriacontane

$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	Table 7.	2000	ceas.	e i inp		ompou				1				,		continu
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	Otter	Gender	Age	Detection Limits		n- pentadecane (C15)	n-tetracosane (C24)	n-tetradecane (C14)	tetr:	n-tr		n-tridecane (C13)	n- tritriacontane (C33)	n-undecane (C11)		Pristane
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	WA099	F	3	2.38	13.2		8.64	19.6	< 2.38	12.7	5		< 2.38	18		10.3
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	WA109	F	3	3.45	7.89	< 3.45	8.04	< 3.45	< 3.45	4.31	5.48	< 3.45	< 3.45	< 3.45	< 3.45	< 3.45
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	WA095	F	4	1.16												
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	WA086	F	5	3.03												
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	WA114	F	6	0.676	< 0.676	0.723	0.78	< 0.676	< 0.676	< 0.676	< 0.676	< 0.676	0.77	< 0.676	< 0.676	< 0.676
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	WA091	F	7	0.658	1.53		1.34	3.13	< 0.658	1.45	0.77	1.36	< 0.658	2.69	1.65	2.19
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	WA090	F	9	1.04	2.91	5.38	1.79	4.17	1.68	2.79	1.18	2.13	< 1.04	5.96	2.97	3.33
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	WA054	F	15	2.78	6.75	<2.78	7.62	< 2.78	< 2.78	3.64	5.63	<2.78	< 2.78	< 2.78	<2.78	< 2.78
$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	WA110	F	16	0.714	1.95	< 0.714	2.07	< 0.714	< 0.714	1.37	1.39	< 0.714	1.46	< 0.714	< 0.714	< 0.714
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	WA113	М	2	0.806	1.77	1.76	1.65	< 0.806	< 0.806	1.58	1.19	< 0.806	< 0.806	< 0.806	< 0.806	< 0.806
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	WA098	Μ	3	2.17	7.96	8.76	5.24	13.3	<2.17	9.72	3.45	6.02	1.02	14.5	4.81	5.67
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	WA108	М	3	1.72	2.55	2.14	2.38	< 1.72	<1.72	1.78	1.8	<1.72	< 1.72	< 1.72	< 1.72	< 1.72
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	WA105	Μ	5	1.61	< 1.61	< 1.61	< 1.61	< 1.61	< 1.61	< 1.61	< 1.61	<1.61	<1.61	<1.61	< 1.61	< 1.61
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	WA107	Μ	6	3.57	4.61	< 3.57	4.28	< 3.57	< 3.57	4.44	< 3.57	< 3.57	3.93	< 3.57	< 3.57	< 3.57
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	WA112	М	6	1.35	<1.35	< 1.35	< 1.35	< 1.35	< 1.35	1.61	<1.35	<1.35	<1.35	<1.35	<1.35	<1.35
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	WA097			1.79	6.8	8.26	4.74	13.5	< 1.79	8.16	2.76	4.34	<1.79	8.99	5.87	6.43
$ \begin{array}{ c c c c c c c c c c c c c c c c c c c$	Females	Mean	a		5.48	5.7	4.37	5.44	-	4.5	2.89	1.91	1.05	4.05	2.86	3.72
$ \begin{array}{ c c c c c c c c c c c c c c c c c c c$	n = 9	Mean	b		6.12	7.97	4.37	6.81		4.73	2.63	2.96	1.05	7.79	4.38	5.93
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $		Std de	ev		4.25	5.69	3.12	6.48		3.99	2.13	1.68	0.51	5.55	2.85	3.82
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		Max			13.2	15.5	8.64	19.6	1.68	12.7	5.63	6	1.73	18	8.04	10.3
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	Males	Mean	a		3.6	3.46	2.82	4.48		4.01	1.78	2.13	1.46	4	2.17	2.38
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	n = 7	Mean	b		4.74	5.23	3.66	6.04		5.71	3.11	5.18	1.23	11.7	5.34	6.05
All $n = 16$ Mean <sup>a</sup> 4.654.723.695.020.984.292.421.234.032.563.13 $N = 16$ Mean <sup>b</sup> 5.596.884.126.482.865.082.773.71.139.114.665.96Std dev3.744.842.76.130.523.71.781.850.845.362.543.3					2.92	3.5	1.91	6.11		3.58	1.02	2.18	1.14	5.54	2.23	2.56
n = 16 Mean <sup>b</sup> 5.59 6.88 4.12 6.48 2.86 5.08 2.77 3.7 1.13 9.11 4.66 5.96 Std dev 3.74 4.84 2.7 6.13 0.52 3.7 1.78 1.85 0.84 5.36 2.54 3.3		Max			7.96	8.76	5.24	13.5		9.72	3.45	6.02	3.93	14.5	5.87	6.43
Std dev         3.74         4.84         2.7         6.13         0.52         3.7         1.78         1.85         0.84         5.36         2.54         3.3	All	Mean	a		4.65	4.72	3.69	5.02	0.98	4.29	2.4	2	1.23	4.03	2.56	3.13
Std dev         3.74         4.84         2.7         6.13         0.52         3.7         1.78         1.85         0.84         5.36         2.54         3.3	n = 16	Mean	b		5.59	6.88	4.12	6.48	2.86	5.08	2.77	3.7	1.13	9.11	4.66	5.96
Max         13.2         15.5         8.64         19.6         1.79         12.7         5.63         6.02         3.93         18         8.04         10.3		Std de	ev		3.74			6.13			1.78		0.84	5.36	2.54	3.3
		Max			13.2	15.5	8.64	19.6	1.79	12.7	5.63	6.02	3.93	18	8.04	10.3

Table 9. Detectable Aliphatic Compounds in Liver of Live Captured Sea Otters (ppm, wet weight); continued

a. mean calculated using 1/2 detection limit for non-detects; b. mean calculated without non-detects; Aliphatics analyzed that were below detection limit: n-hentriacontane & n-tritriacontane

 Table 10. Detectable Polycyclic Aromatic Hydrocarbon Compounds in Liver of Live

 Captured Sea Otters (ppm, wet weight)

Capturet			<u>(ppm,</u>	wet we	igni)							
Otter	Gender	Age	Detection Limits	2- methylnaphthalene	C1-naphthalenes	C2-Phenanthrenes & Anthracenes	C3-fluorenes	Biphenyl	Fluorene	Naphthalene	Unresolved Complex Mixture	Total Petroleum Hydrocarbons
WA099	F	3	1.1	1.07	1.69	< 1.07	< 1.07	1.74	< 1.07	1.44	< 1.07	< 1.07
WA109	F	3	0.3	< 0.345	< 0.345	< 0.345	0.378	< 0.345	< 0.345	< 0.345	291	50.5
WA095	F	4	0.5	< 0.523	< 0.523	< 0.523	< 0.523	0.881	< 0.523	0.53	2090	< 0.523
WA086	F	5	1.4	< 1.36	< 1.36	< 1.36	< 1.36	2.28	< 1.36	1.43	< 1.36	< 1.36
WA114	F	6	0.1	< 0.068	< 0.068	< 0.068	0.0686	< 0.068	< 0.068	< 0.068	382	< 0.135
WA091	F	7	0.3	< 0.296	0.414	< 0.296	< 0.296	0.719	< 0.296	0.47	< 0.296	< 0.296
WA090	F	9	0.5	< 0.469	0.58	< 0.469	< 0.469	0.763	0.556	0.625	< 0.469	< 0.469
WA054	F	15	0.3	< 0.278	< 0.278	< 0.278	0.278	< 0.278	< 0.278	< 0.278	380	< 0.556
WA110	F	16	0.1	< 0.071	< 0.071	< 0.071	< 0.071	< 0.071	< 0.071	< 0.071	459	131
WA113	М	2	0.1	< 0.081	< 0.081	< 0.081	< 0.081	< 0.081	< 0.081	< 0.081	177	139
WA098	М	3	1	< 0.978	1.05	< 0.978	< 0.978	1.77	< 0.978	1.28	< 0.978	< 0.978
WA108	М	3	0.2	< 0.172	< 0.172	< 0.172	0.211	< 0.172	< 0.172	< 0.172	306	9.2
WA105	М	5	0.2	< 0.161	< 0.161	< 0.161	0.195	< 0.161	< 0.161	< 0.161	195	296
WA107	М	6	0.4	< 0.357	< 0.357	< 0.357	0.359	< 0.357	< 0.357	< 0.357	424	102
WA112	М	6	0.1	< 0.135	< 0.135	0.138	1.16	< 0.135	< 0.135	< 0.135	229	< 0.27
WA097	М	8	0.8	< 0.804	0.996	< 0.804	< 0.804	1.44	< 0.804	1.04	< 0.804	< 0.804
Females	Mean <sup>a</sup>			0.445		0.291	0.752		0.542	796	45.5	
n = 9	Mean <sup>b</sup>			1.28			0.242		0.899	720	90.8	
	Std dev			0.52		0.212	0.796		0.55	648	61.8	
	Max		1.07	1.69		0.682	2.28	0.556	1.44	2090	131	
Males	Mean <sup>a</sup>			0.357		0.408	0.523		0.396	473	109	
n = 7	Mean <sup>b</sup>			1.61		0.138	0.481		1.16	266	137	
	Std dev			0.457		0.364	0.746		0.528	368	120	
	Max			1.05	0.138	1.16	1.77		1.28	1090	296	
All	Mean <sup>a</sup>			0.407		0.342	0.652		0.478	655	80.9	
n = 16	Mean <sup>b</sup>			1.37		0.14	0.379		0.974	493	121	
	Std dev			0.479		0.284	0.758		0.528	553	98.9	
	Max		1.07	1.69	0.682	1.16	2.28	0.556	1.44	2090	296	
a mean cal	culated usi	nσ 1/2	) detect	ion limit t	for non_de	etects h	mean calo	ulated w	ithout nor	-detects.	PΔHc	

a. mean calculated using 1/2 detection limit for non-detects; b. mean calculated without non-detects; PAHs analyzed that were below detection limit: naphthalene, 1,6,7-Trimethyl-naphthalene, 1-methylnaphthalene, 1-methylnaphthalene, 2,6-dimethylnaphthalene, acenaphthalene, acenaphthalene, anthracene, benzo(a)pyrene, benzo(b)fluoranthene, benzo(e)pyrene, benzo(g,h,i)perylene, benzo(k)fluoranthene, C1-chrysenes, C1-dibenzothiophenes, C1-Fluoranthenes & Pyrenes, C1-fluorenes, C1-Phenanthrenes & Anthracenes, C2-chrysenes, C2-dibenzothiophenes, C2-naphthalenes, C2-fluorenes, C3-chrysenes, C3-dibenzothiophenes, C3-naphthalenes, C4-naphthalenes, C4-Phenanthrenes & Anthracenes, chrysene, dibenzothiophene, fluoranthene, indeno(1,2,3-cd)pyrene, perylene, phenanthrene, pyrene, Benzo(a)anthracene, Dibenz(a,h)anthracene

Otter	Gender	Age	Detection Limits	1,2,3,4- Tetrachlorobenzene	1,2,4,5- Tetrachlorobenzene
17315-01	F	13	0.00431	0.00776	0.021
14325	F	U	0.00410	0.00521	0.0199
13827	F	U	0.00472	0.0406	0.0502
15713-01	М	8	0.00446	0.00585	0.0193
14717	М	U	0.00463	< 0.00463	0.0218
13555	М	U	0.00568	< 0.00568	0.0189
10385	М	U	0.00735	< 0.00735	0.0486
18316-01	М	1	0.00862	< 0.00862	< 0.00862
Females	Mean <sup>a</sup>			0.0179	0.0304
n = 3	Mean <sup>b</sup>			0.0179	0.0304
	Std dev			0.0197	0.0172
	Max			0.0406	0.0502
Males	Mean <sup>a</sup>			0.0038	0.0226
n = 5	Mean <sup>b</sup>			0.0059	0.0272
	Std dev			0.0014	0.0161
	Max			0.0059	0.0486
all	Mean <sup>a</sup>			0.0091	0.0255
n = 8	Mean <sup>b</sup>			0.0149	0.0285
	Std dev			0.0129	0.0158
	Max			0.0406	0.0502

 Table 11. Detectable Semi-volatile Compounds in Liver of Beach-Cast Sea Otters

 (ppm, wet weight)

a. mean calculated using 1/2 detection limit for non-detects; b. mean calculated without nondetects; U = Unknown; The only detectable semi-volatile found in live sea otters was 1,2,3,4-Tetrachlorobenzene (0.00151ppm in WA094).

### Organochlorines

Both organochlorine scans and congener-specific PCB analyses were performed on whole blood samples from live captured Washington sea otters and on liver from the beach-cast animals. Only one otter had detectable organochlorines other than PCBs in whole blood and that was otter WA098, which had a measured level of 0.18 ppm p.p'-DDE. Because concentrations of individual congeners were quite low, the sum of the PCBs is presented in Table 12, with the detection limit and concentration for each congener provided in Appendix 5. Total PCB concentration ranged from 2.15 to 15 ppb wet weight when one-half the detection limit was used in the calculations and from 0.944 to 12.4 ppb wet weight when zero was used for congeners below the detection limit. Total PCB concentrations were significantly higher in males (mean = 7.34 ppb) than females (mean = 4.69; p = .035) when the sum was calculated without non detects. Several of the congener concentrations revealed a significant relationship to a predictor. Gender differences were significant for (1) PCB 101/90, (p = 0.005, mean for males is 0.000247 ppb ww, mean for females is 0.000124 ppb ww; (2) PCB 138/160 (p = 0.034. mean for males is 0.001151 ppb ww, mean for females is 0.000705 ppb ww); and (3) PCB 153/132 (p = 0.048, mean for males is 0.727 ppb ww, mean for females is 0.055 ppb ww). In all three instances, the PCB congeners were higher in males than females, as would be expected since females can rid themselves of contaminants through lactation and giving birth. Age differences were significant for several congeners as well: PCB 105 (p = 0.045;  $R^2 = .228$ ), PCB 180 (p = 0.002;  $R^2 = .303$ ), and PCB 187 (p = 0.004;  $R^2 = .353$ ). PCB congener concentrations with a relationship to age were higher in younger versus older animals.

Liver samples from fifteen beach-cast sea otters was analyzed for organochlorines (Table 13). Only PCBs were detected in more than 50 percent of the animals and these were less than 1 ppm. The organochlorines were also summed by group and are presented along with total butyltin for easier comparison to literature values (Table 14); however, a direct comparison cannot be made with live Washington animals since liver biopsies from live animals were not analyzed for organochlorines. None of the beach-cast Washington otters had liver concentrations exceeding 0.85 ppm for DDT (1,1,1,-trichloro-2,2-bis(p-chlorophyenyl)ethanes), cyclohexanes (HCHs), chlordanes (CHL), PCBs or total butyltins (BT). Congener-specific PCB detection limits and results are presented in Appendix 6. Regression analysis revealed no significant relationships between gender or age and total PCB concentrations.

Otter	Gender	Age	Sum of All PCB Congeners <sup>a</sup>	Sum of All PCB Congeners <sup>b</sup>
WA115	F	<1	11.6	9.06
WA099	F	3	5.27	3.69
WA109	F	3	14.7	12.0
WA084	F	4	5.17	3.83
WA095	F	4	4.10	2.55
WA100	F	4	3.18	1.41
WA111	F	4	9.93	7.74
WA086	F	5	4.07	2.59
WA094	F	5	2.39	1.01
WA089	F	6	2.72	1.15
WA106	F	6	8.03	6.18
WA114	F	6	9.05	7.15
WA091	F	7	5.62	3.45
WA090	F	9	3.58	1.91
WA103	F	9	12.7	9.98
WA093	F	10	2.15	0.940
WA096	F	10	2.94	1.12
WA088	F	13	3.05	1.25
WA054	F	15	10.2	8.18
WA110	F	16	10.8	8.52
WA092	М	<1	4.68	2.84
WA104	М	<1	12.1	10.2
WA113	М	2	12.3	9.96
WA098	М	3	4.76	3.81
WA108	М	3	15.0	12.4
WA105	М	5	9.19	6.95
WA107	М	6	12.1	10.1
WA112	М	6	11.5	9.50
WA097	М	8	3.46	1.66
WA102	М	10	7.93	5.98
Females	Mean		6.57	4.69
N = 20	Std dev		3.94	3.56
	Max		14.7	12.0
	Min		2.15	0.944
Males	Mean		9.30	7.34
N = 10	Std dev		3.94	3.65
	Max		15.0	12.4
	Min		3.46	1.66
All live	Mean		7.48	5.57
N = 30	Std dev		4.09	3.75
	Max		15.0	12.4
	Min		2.15	0.944

 Table 12. Total PCB Concentration in Whole Blood of Live Captured Sea Otters

 (ppb, wet weight)

a. total PCBs calculated using 1/2 detection limit for non-detects; b. total PCBs calculated without nondetects; Congeners with interference noted by the laboratory are not included in this sum; List of summed PCB congeners: 1, 7/9, 8/5, 16/32, 18/17, 22/51, 24/27, 25, 26, 28, 29, 30, 31, 33/20, 39, 40, 41/64, 42/59/37, 44, 45, 46, 47/75, 48, 49, 52, 53, 60/56, 63, 66, 67, 69, 72, 74/61, 81, 82, 84, 85, 87/115, 92, 95/80, 97, 99, 101/90, 105, 107, 110, 118, 119, 128, 130, 135, 136, 138/160, 141/179, 146, 149/123, 151, 153/132, 156, 158, 166, 167, 170/190, 171/202, 172, 174, 175, 176/137, 177, 178, 180, 183, 187, 191, 193, 194, 195/208, 196, 197, 199, 200, 205, 206, 207, 209.

Otter	Gender	Age	Aldrin	alpha BHC	alpha chlordane		cis- nonachlor	delta BHC	Dieldrin	gamma BHC	heptachlor epoxide	Mirex	o,p'-DDT
16961-03	F	0	NA	<.0100		<.0100	<.0100	NA	<.0100		-		<.00410
16961-01	F	3	NA	<.0100	<.0100	<.0100	<.0100	NA	<.0100				<.0100
17315-01	F	13	<.00431	<.00431	<.00431	0.0372	<.00431	<.00431	0.0485	<.00431	0.00973	<.00431	<.0100
14325	F	U	<.00410	<.00410	<.00410	0.0065	<.00410	<.00410	0.00882	0.00965	0.00559	<.00410	<.0100
13827	F	U	0.0342	<.00472	0.0309	0.0659	<.00472	0.0085	0.0231	0.0566	<.00472	<.00472	<.0100
16961-02	М	<1	NA	<.0100	<.0100	<.0100	<.0100	NA	<.0100	<.0100	<.0100	<.0100	<.00431
16961-04	М	2	NA	<.0100	<.0100	<.0100	<.0100	NA	<.0100	<.0100	<.0100	<.0100	0.0136
18124-01	М	3	NA	<.0100	<.0100	<.0100	<.0100	NA	<.0100	<.0100	<.0100	<.0100	<.0100
15713-01	М	8	<.00446	0.00469	<.00446	0.0622	<.00446	<.00446	0.016	0.00616	<.00446	<.00446	<.00446
14717	М	U	<.00463	0.00464	<.00463	0.0138	<.00463	<.00463	0.00729	0.00585	<.00463	<.00463	<.00463
16904-01	М	12	NA	<.0100	<.0100	<.0100	<.0100	NA	<.0100	<.0100	<.0100	<.0100	<.0100
17058-01	М	U	NA	<.0100	<.0100	<.0100	<.0100	NA	<.0100	<.0100	<.0100	<.0100	<.0100
13555	М	U	<.00568	<.00568	<.00568	0.0138	0.00764	<.00568	0.015	<.00568	<.00568	0.00828	<.00568
10385	М	U	0.0167	0.00777	0.0183	0.0514	<.00735	<.00735	<.00735	0.0399	<.00735	<.00735	<.00735
18316-01	М	1	<.00862	<.00862	<.00862	<.00862	<.00862	<.00862	0.00908	<.00862	<.00862	<.00862	<.00862
Females	Mean <sup>a</sup>					0.0239			0.0181	0.0157			
n = 5	Mean <sup>b</sup>					0.0365			0.0268	0.0331			
	Std dev					0.0297			0.0201	0.0332			
	Max		0.0342		0.0309	0.0659	0.005	0.0085	0.0485	0.0566	0.00973	0.005	0.0136
Males	Mean <sup>a</sup>			0.0049		0.0171			0.0076	0.0084			
n = 10	Mean <sup>b</sup>			0.0057		0.0353			0.0118	0.0173			
	Std dev			0.0018		0.0252			0.0043	0.0196			
	Max		0.0167	0.00777	0.0183	0.0622	0.00764	0.00431	0.016	0.0399	0.005	0.00828	0.005
all	Mean <sup>a</sup>			0.0044		0.0193			0.0111	0.0108			
n = 15	Mean <sup>b</sup>			0.0057		0.0358			0.0183	0.0236			
	Std dev			0.0018		0.0247			0.0144	0.0233			
	Max	1/2 1 1	0.0342	0.00777	0.0309	0.0659	0.00764	0.0085	0.0485	0.0566	0.00973	0.00828	0.0136

Table 13. Organochlorines in Liver of Beach-Cast Sea Otters (ppm, wet weight)

a. mean calculated using 1/2 detection limit for non-detects; b. mean calculated without non-detects; U = unknown; NA = Not analyzed; The only detectable organochlorine other than PCBs measured in live otters was in the whole blood of otter WA098 - p,p'-DDE at 0.18 ppm.

Otter	Gender	Age	Oxy- chlordane	p,p'- DDT	PCB- TOTAL	trans- nonachlor	Aroclor- 1242	Aroclor- 1248	Aroclor- 1254	Aroclor- 1260
16961-03	F	2	0.0073	<.00410	0.503	0.0137	<.0410	<.0410	<.0410	0.503
16961-01	F	3	<.0100	<.0100	0.407	<.0100	NA	NA	NA	NA
17315-01	F	13	<.0100	<.0100	0.258	0.0147	NA	NA	NA	NA
14325	F	U	<.0100	<.0100	0.176	<.0100	NA	NA	NA	NA
13827	F	U	<.0100	<.0100	0.0888	<.0100	NA	NA	NA	NA
16961-02	М	<1	0.00917	<.00431	0.446	0.0124	<.0431	<.0431	0.178	0.268
16961-04	М	2	<.00472	0.0064	0.486	0.00775	0.17	0.194	0.122	<.0472
18124-01	М	3	0.0188	<.0100	0.718	0.0244	NA	NA	NA	NA
15713-01	М	8	0.00965	<.00446	0.82	0.00954	<.0446	<.0446	0.246	0.533
14717	М	U	<.00463	<.00463	0.26	<.00463	<.0463	0.104	<.0463	0.156
16904-01	М	12	<.0100	<.0100	0.4	<.0100	NA	NA	NA	NA
17058-01	М	U	<.0100	<.0100	0.255	<.0100	NA	NA	NA	NA
13555	М	U	0.00818	<.00568	0.821	0.0312	<.0568	<.0568	0.411	0.411
10385	М	U	<.00735	<.00735	0.407	<.00735	0.102	0.122	0.102	0.0815
18316-01	М	1	<.00862	<.00862	0.0936	<.00862	NA	NA	NA	NA
Females	Mean <sup>a</sup>				0.4484	0.0088				
n = 5	Mean <sup>b</sup>				0.2870	0.0142				
	Std dev				0.0460	0.0041				
	Max				0.503	0.0147				0.503
Males	Mean <sup>a</sup>		0.0067		0.3897	0.0105		0.0692	0.1955	0.2954
n = 10	Mean <sup>b</sup>		0.0115		0.4707	0.0171		0.1400	0.2118	0.2899
	Std dev		0.0049		0.2470	0.0102		0.0476	0.1246	0.1841
	Max		0.0188	0.0064	0.821	0.0312	0.17	0.194	0.411	0.533
all	Mean <sup>a</sup>		0.0064		0.4093	0.0099		0.0733	0.1575	0.2911
n = 15	Mean <sup>b</sup>		0.0106		0.4093	0.0162		0.1400	0.2118	0.3254
	Std dev		0.0047		0.2353	0.0085		0.0476	0.1246	0.1862
	Max		0.0188	0.0064	0.821	0.0312	0.17	0.194	0.411	0.533

Table 13. Organochlorines in Liver of Beach-Cast Sea Otters (ppm, wet weight); continued

a. mean calculated using 1/2 detection limit for non-detects; b. mean calculated without non-detects; U = unknown; NA = Not analyzed; The only detectable organochlorine other than PCBs measured in live otters was in the whole blood of otter WA098 - p,p'-DDE at 0.18 ppm.

Otter	Gender	Age	DDTs	HCHs	CHLs	PCBs	BTs
16961-03	F	2	0.0359	0.015	0.02	0.503	0.0931
16961-01	F	3	0.0867	0.015	0.02	0.407	0.154
17315-01	F	13	0.0912	0.0437	0.0259	0.258	0.384
14325	F	U	0.0775	0.0203	0.0251	0.176	0.195
13827	F	U	0.0228	0.133	0.0434	0.0888	0.21
16961-02	М	<1	0.094	0.015	0.02	0.446	0.0951
16961-04	М	2	0.439	0.015	0.0532	0.486	0.196
18124-01	М	3	0.239	0.015	0.0297	0.718	0.447
15713-01	М	M 8 0.08		0.0753	0.0237	0.82	0.107
14717	М	U	0.0304	0.0266	0.0093	0.26	0.0553
16904-01	М	12	0.0852	0.015	0.02	0.4	0.171
17058-01	М	U	0.0307	0.015	0.02	0.255	0.213
13555	М	U	0.527	0.0223	0.0499	0.821	0.255
10385	М	U	0.0237	0.103	0.0293	0.407	0.166
18316-01	М	1	0.0192	0.017	0.0172	0.0936	0.267
Females	Mean		0.0628	0.0454	0.0269	0.448	0.207
N = 5	Std dev	7	0.0313	0.0504	0.0096	0.0460	0.109
	Max		0.0912	0.133	0.0434	0.503	0.384
	Min		0.0228	0.015	0.02	0.4	0.0931
Males	Mean		0.158	0.032	0.0272	0.390	0.197
n = 10	Std dev	7	0.184	0.0311	0.0141	0.290	0.111
	Max		0.527	0.103	0.0532	0.821	0.447
	Min		0.0192	0.015	0.0093	0.089	0.0553
All	Mean		0.126	0.0364	0.0271	0.409	0.201
n = 15	Std dev	7	0.156	0.0373	0.0124	0.235	0.106
	Max		0.527	0.133	0.0532	0.821	0.447
	Min		0.0192	0.015	0.0093	0.089	0.0553

 Table 14. Organochlorines by Group in Liver of Beach-Cast Sea Otters

 (ppm, wet weight)

Totals and means calculated using 1/2 detection limit for non detects; U = Unknown

DDTs = p,p'-DDD + p,p'-DDE + p,p'-DDT

HCHs = alpha BHC + beta BHC + delta BHC + gamma BHC

CHLs = alpha chlordane + cis-nonachlor + oxychlordane + trans-nonachlor

BTs = monobutyltin + dibutyltin + tributyltin + tetrabutyltin

### **Biomarkers**

### Vitamin A and Thyroid Hormones

Plasma and liver tissue were used to determine vitamin A levels (Table 15). Plasma was also analyzed for thyroid hormones. The mean value of vitamin A in the plasma of all blood samples was 170 ug/L, while the liver value was 9.35 ug/L, (Table 15). Vitamin A concentrations in the liver and plasma were not significantly correlated with one another (p=0.315 n=24). Concentrations of the two retinoids, retinol (fat soluble) and retinyl-palmitate (storage ester form), were above detection limits in all samples analyzed. Three additional unquantified peaks detected in sea otter liver samples may represent other retinol esters.

Regression analyses to determine if vitamin A or thyroid hormone level related to the age, gender or body weight of the animals studied found three significant gender relationships and one significant weight relationship. Females had a greater concentration of all-trans retinyl palmitate (p=0.005,  $r^2=0.292$ , n=25) in the liver (mean for males is 34.76 ug/g, mean for females is 397.18 ug/g); as well as a greater concentration of total retinol (p = 0.010,  $r^2 = 0.255$ , n = 25) in the liver (mean for males is 38.22 ug/g, mean for females is 306.31 ug/g). The ratio of all-trans retinol to all-trans retinyl palmitate was higher in males (p = 0.49,  $r^2 = 0.159$ , mean for males is 61.46 and the mean for females is 54.61). All-trans retinyl palmitate also had a significant relationship with gender and age (p = 0.003, partial t-test coefficient significance = 0.001 for gender and 0.046 for age,  $R^2 = 0.411$ ).

The thyroid hormone tetraiodothyronine/thyroxine (T4) had a significant relationship with animal body weight, (p < 0.001,  $R^2 = 0.436$ ) with larger animals having less of the hormone. ANOVA was also used to evaluate if lactating females differed in vitamin A circulatory or liver levels from levels in non-lactating reproductive aged females (females become reproductive at 4-5 years; Kenyon 1969, Jameson and Johnson 1993). Of the 17 females of reproductive age captured, three were lactating. There was no significant difference between lactating and non lactating females in the vitamin A levels in plasma (p = 0.154) or liver samples (p = 0.726).

When comparing vitamin A concentrations in liver to contaminant concentrations in liver and whole blood samples, a negative correlation was observed between concentration of PCBs in whole blood and all-trans retinyl palmitate (RP) in liver (p < 0.001), total retinol (TR) in liver, (p < 0.001), and the ratio of all trans retinol (R) to RP in liver (p < 0.001). There was no significant relationship between whole blood PCB concentrations and all-trans retinol (R) in the liver or plasma, nor was there a significant relationship between PCB concentration in the liver and the thyroid hormones tetraiodothyronine (T4) and triiodothyronine (T3) in the plasma.

In addition to PCBs, a significant reduction of vitamin A storage in the liver was observed in relation to dibutyltin and octacosane concentration (Table 18). A significant and strong positive correlation in vitamin A storage in the liver was observed for cadmium and several of the aliphatic hydrocarbons. Several additional contaminants were significantly correlated with Vitamin A indices but no others were particularly strong correlations.

WA115FWA099FWA109FWA084FWA095FWA095FWA100FWA111FWA086FWA086FWA094FWA094FWA095FWA094FWA094FWA095FWA091FWA091FWA093FWA093FWA094FWA095FWA095FWA096FWA096FWA097MWA104MWA105MWA105MWA107MWA108MWA107MWA102MWA102MWA103MWA104MWA105MWA105MWA102MWA102MWA102MWA102MWA102MManesMan	<1 3 3 4 4 4 4	[ <i>R</i> ] ug/L 150 190 200	<b>[R] in ug/g</b> NA	[RP] in ug/g	TR in ug/g	R:RP	nmol/L	nmol/L
WA099FWA109FWA084FWA095FWA100FWA104FWA086FWA087FWA094FWA094FWA094FWA106FWA107FWA091FWA091FWA093FWA093FWA094FWA095FWA096FWA096FWA097MWA103MWA104MWA105MWA107MWA107MWA102MWA102MWA103MWA104MWA105MWA107MWA102MWA102MWA103MeanMalesMean	3 3 4 4 4	190	NA	27.4				nn0t/L
WA109FWA084FWA095FWA100FWA104FWA086FWA087FWA089FWA106FWA107FWA091FWA091FWA091FWA093FWA093FWA094FWA093FWA094FWA095FWA096FWA096FWA097MWA103MWA104MWA105MWA107MWA107MWA102MWA102MWA103MWA104MWA105MWA107MWA102MWA102MWA103MeanManesMeanMalesMean	3 4 4 4 4			NA	NA	NA	29	0.4
WA084FWA095FWA100FWA101FWA086FWA087FWA089FWA089FWA106FWA107FWA091FWA091FWA093FWA093FWA094FWA095FWA096FWA097MWA098MWA104MWA105MWA107MWA107MWA107MWA102MWA103MWA104MWA105MWA105MWA107MWA102MWA103MeanManesMeanMalesMean	4 4 4	200	8.34	191.58	113.71	22.97	25	0.8
WA095         F           WA100         F           WA111         F           WA086         F           WA084         F           WA089         F           WA089         F           WA106         F           WA107         F           WA114         F           WA091         F           WA092         F           WA093         F           WA093         F           WA093         F           WA094         F           WA095         F           WA096         F           WA097         M           WA113         M           WA104         M           WA105         M           WA107         M           WA107         M           WA102         M           WA103         M           WA104         M           WA105         M           WA107         M           WA102         M           WA103         M           WA104         M           WA105         M           WA107	4 4	200	11.55	31.66	115.15	12.08	58	0.5
WA100         F           WA111         F           WA086         F           WA094         F           WA089         F           WA089         F           WA106         F           WA107         F           WA108         F           WA091         F           WA091         F           WA091         F           WA093         F           WA093         F           WA093         F           WA094         F           WA095         F           WA096         F           WA097         M           WA103         M           WA104         M           WA103         M           WA104         M           WA105         M           WA107         M           WA107         M           WA102         M           WA103         M           WA104         M           WA105         M           WA107         M           WA102         M           WA103         M           WA104	4	120	6.41	283.34	162.24	44.22	23	NSQ
WA111         F           WA086         F           WA094         F           WA089         F           WA106         F           WA107         F           WA114         F           WA090         F           WA091         F           WA090         F           WA091         F           WA092         F           WA093         F           WA094         F           WA095         F           WA096         F           WA097         M           WA103         M           WA104         M           WA105         M           WA108         M           WA107         M           WA102         M           WA103         M           WA104         M           WA105         M           WA105         M           WA102         M           WA103         M           WA104         M           WA105         M           WA105         M           WA102         M           WA103		NA	5.99	660.47	369.25	110.28	NS	NSQ
WA086         F           WA094         F           WA089         F           WA089         F           WA106         F           WA114         F           WA091         F           WA092         F           WA093         F           WA093         F           WA093         F           WA093         F           WA094         F           WA095         F           WA096         F           WA097         M           WA103         M           WA104         M           WA105         M           WA106         M           WA107         M           WA107         M           WA102         M           WA102         M           WA103         M           WA104         M           WA105         M           WA102         M           WA103         M           WA104         M           WA105         M           WA102         M           WA103         Mean	4	170	2.05	236.98	132.39	115.49	21	0.9
WA094         F           WA089         F           WA106         F           WA106         F           WA114         F           WA091         F           WA090         F           WA090         F           WA090         F           WA091         F           WA092         F           WA093         F           WA094         F           WA0954         F           WA054         F           WA092         M           WA110         F           WA093         M           WA104         M           WA105         M           WA107         M           WA107         M           WA107         M           WA102         M           WA102         M           WA103         M           WA104         M           WA105         M           WA107         M           WA102         M           WA103         M      //wwwwwwwwwwwwwwwwwwwwwwwwwwwwwwwwwww	4	190	NA	NA	NA	NA	15	0.6
WA089         F           WA106         F           WA114         F           WA091         F           WA090         F           WA090         F           WA090         F           WA093         F           WA094         F           WA095         F           WA096         F           WA097         M           WA092         M           WA104         M           WA105         M           WA108         M           WA103         M           WA104         M           WA105         M           WA106         M           WA107         M           WA102         M           WA103         M           WA104         M           WA105         M           WA107         M           WA102         M           WA097         M           WA102         M           WA103         Mean           Maks         Mas	5	140	16.04	1731.14	968.16	107.95	22	0.5
WA106         F           WA114         F           WA091         F           WA090         F           WA090         F           WA093         F           WA093         F           WA096         F           WA097         F           WA098         F           WA092         M           WA104         M           WA105         M           WA108         M           WA107         M           WA105         M           WA107         M           WA102         M           WA103         M           WA104         M           WA105         M           WA105         M           WA102         M           WA103         M           WA104         M           WA105         M           WA102         M           WA103         M           WA104         M           WA105         M           WA102         M           WA103         M           WA104         M	5	120	11.54	418.36	241.64	36.26	20	NSQ
WA114         F           WA091         F           WA090         F           WA093         F           WA093         F           WA094         F           WA0954         F           WA054         F           WA054         F           WA054         F           WA054         F           WA054         M           WA110         F           WA092         M           WA103         M           WA104         M           WA105         M           WA107         M           WA107         M           WA102         M           Makes         Mean	6	230	12.37	1207.33	676.40	97.61	28	1.1
WA091         F           WA090         F           WA103         F           WA093         F           WA096         F           WA096         F           WA097         F           WA098         F           WA092         M           WA110         F           WA092         M           WA104         M           WA103         M           WA104         M           WA105         M           WA107         M           WA107         M           WA102         M           WA103         M           WA104         M           WA105         M           WA105         M           WA102         M           WA097         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	6	220	NA	NA	NA	NA	14	0.2
WA090         F           WA103         F           WA093         F           WA096         F           WA096         F           WA088         F           WA054         F           WA054         F           WA092         M           WA100         F           WA092         M           WA104         M           WA105         M           WA108         M           WA107         M           WA107         M           WA102         M           WA103         M           WA104         M           WA105         M           WA107         M           WA102         M           WA097         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	6	120	14.87	38.91	8.97	11.46	16	0.3
WA103         F           WA093         F           WA096         F           WA088         F           WA054         F           WA054         F           WA054         F           WA054         F           WA054         F           WA054         F           WA10         F           WA092         M           WA104         M           WA105         M           WA105         M           WA107         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	7	190	1.54	81.90	46.59	53.04	22	NSQ
WA093         F           WA096         F           WA088         F           WA054         F           WA054         F           WA054         F           WA054         F           WA054         F           WA110         F           WA092         M           WA104         M           WA103         M           WA108         M           WA105         M           WA107         M           WA107         M           WA097         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	9	270	4.00	219.27	124.59	54.88	25	0.8
WA096         F           WA088         F           WA054         F           WA054         F           WA054         F           WA054         F           WA054         F           WA092         M           WA104         M           WA103         M           WA098         M           WA108         M           WA105         M           WA107         M           WA107         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	9	130	34.23	242.22	1419.65	30.75	23	0.3
WA088         F           WA054         F           WA100         F           WA092         M           WA104         M           WA104         M           WA103         M           WA098         M           WA108         M           WA105         M           WA107         M           WA097         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	10	160	4.18	75.44	45.67	18.05	20	0.4
WA054         F           WA110         F           WA092         M           WA104         M           WA104         M           WA104         M           WA104         M           WA104         M           WA105         M           WA105         M           WA107         M           WA107         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	10	200	2.32	327.72	182.56	141.42	15	0.5
WA110         F           WA092         M           WA104         M           WA103         M           WA098         M           WA108         M           WA105         M           WA107         M           WA097         M           WA097         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	13	210	18.91	991.27	564.11	52.43	24	NSQ
WA092         M           WA104         M           WA103         M           WA098         M           WA108         M           WA105         M           WA105         M           WA107         M           WA107         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	15	140	1.74	3.51	24.44	10.19	NA	NSQ
WA104         M           WA113         M           WA098         M           WA108         M           WA105         M           WA107         M           WA107         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	16	170	5.26	10.87	11.77	9.31	12	0.6
WA113         M           WA098         M           WA108         M           WA105         M           WA107         M           WA107         M           WA107         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	<1	180	7.82	66.45	44.36	8.50	28	NSQ
WA098         M           WA108         M           WA105         M           WA107         M           WA107         M           WA107         M           WA107         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	<1	210	NA	NA	NA	NA	51	1.6
WA108         M           WA105         M           WA107         M           WA107         M           WA107         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	2	110	35.44	39.16	56.91	4.76	24	0.4
WA105         M           WA107         M           WA112         M           WA097         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	3	150	0.16	69.43	38.35	423.88	26	0.7
WA107         M           WA112         M           WA097         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	3	130	1.08	2.43	0.41	13.30	27	0.6
WA112         M           WA097         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	5	170	10.27	17.04	5.85	7.27	13	0.9
WA097         M           WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	6	200	NA	NA	NA	NA	17	0.7
WA102         M           Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	6	130	9.16	11.08	62.85	5.26	9	0.4
Females         Mean           n = 20         Std dev           Min         Max           Males         Mean	8	110	6.86	66.35	43.35	9.67	5	0.6
n = 20 Std dev Min Max Males Mean	10	230	1.73	6.17	53.65	19.02	16	0.6
Min Max Males Mean		175	9.49	397.18	306.31	54.61	23	0.6
Max Males Mean		42	8.36	485.35	391.48	43.37	10	0.3
Males Mean		120	1.54	3.51	8.97	9.31	12	0.2
Males Mean		270	34.23	1731.14	1419.65	141.42	58	1.1
		162	9.06	34.76	38.22	61.46	22	0.7
n = 10 <b>Std dev</b>		43	11.3	29.18	23.10	146.52	13	0.4
Min		110	0.16	2.43	0.41	4.76	5	0.4
Max		230	35.44	69.43	62.85	423.88	51	1.6
All Mean		170	9.35	281.20	220.52	56.80	22	0.6
n = 30 Std dev		42	9.17	432.50	344.41	86.75	11	0.3
Min	<u> </u>	110	0.16	2.43	0.41	4.76	5	0.2
Max		270	35.4	1731.14	1419.65	423.88	58	1.6

Table 15. Vitamin A and Thyroid Hormone Levels for Live Captured Sea Otters (Plasma and Liver)

NSQ = Not sufficient quantity; NS = No sample available; NA = Not analyzed; [R] = all-trans retinol; [RP] = all-trans retinyl palmitate; [TR] = total retinol = [R]+0.55\*[RP]; T4 = tetraiodothyronine/thyroxine; T3 = triiodothyronine

The T4 thyroid hormone level was significantly negatively correlated with eiocosane in the seven animals with detectable levels of eiocosane. T4 was significantly negatively correlated with selenium and positively correlated with strontium but neither correlation was strong. T3 was significantly negatively correlated with copper, but the correlation was not strong (Table 18).

# P450 CYP1A Assays

The cytochrome P450 blood (PBMC) and liver results for the 2001 live captured otters are presented in Table 16. The results are not yet available for the live captured animals from 2002 The mean PBMC P450 value for the 16 live captured otters was  $7.57 \times 10^6$  molecules p450/100ng total RNA, while the mean liver value was  $1.85 \times 10^6$  molecules p450/100ng total RNA. Although the mean is presented, variability in the data, particularly the blood data, is considerable as can be seen by the large standard deviations; therefore, evaluating mean cytochrome P450 values does not reveal as much as reviewing the individual results or medians. Six of the animals for which P450 was analyzed in the PBMC had less than 1 x 10<sup>6</sup> molecules p450/100ng total RNA, which is indicative of animals living in a clean environment (B. Ballachey pers. comm. 2006). Three of the animals had PBMC levels less than  $4 \times 10^{6}$  but greater than 1 x 10<sup>6</sup> molecules p450/100ng total RNA. Cytochrome P450 induction was considerably higher in the remaining two animals, at  $17.90 \times 10^6$  for otter WA099 and 54.10 x 10<sup>6</sup> molecules p450/100ng total RNA for otter WA093. There was not a significant correlation between blood and liver P450 values (p=0.505) – in fact liver induction of P450 in otter WA099 was less than one molecule p450/100ng total RNA and otter WA093 was slightly above the mean. Liver levels were less variable than the blood values, ranging from  $0.2 \times 10^6$  to  $4.8 \times 10^6$ molecules p450/100ng total RNA. There were no significant correlations between contaminant concentrations and cytochrome P450 results for PBMC. Cytochrome p450 in the liver was negatively correlated with total PCB (p = 0.01). Regression analysis revealed no significant gender, age or body weight relationship to the P450 assay results for PBMC or liver.

## Pathogens

Individual results for the serological tests are provided in Table 17. In summary, 24 of 30 animals or 80 percent, were positive for antibodies to morbilliviruses; 1 of 30 (3 %) was positive for antibodies to *Leptospira* sp.; no animals were positive for antibodies to *Brucella* or caliciviruses; 18 of 30 (60 %) were positive for antibodies to *T. gondii*; 15 of 30 (50 %) were positive for *N. caninum*, and 4 of 14 (29 %) were positive (7 samples are pending) for *Sarcocystis* sp. antibodies. The otters generally had higher titers to the two distemper strains of morbillivirus than the cetacean-derived strains indicating that the population's exposure is probably to Canine Distemper Virus (CDV) or Phocine Distemper Virus (PDV).

Oral swabs, fecal swabs and scat samples were obtained, when possible, from live captured otters. All bacterial flora and parasites found in such samples were normal and as expected for sea otter residing in the wild due to the prey items they consume. The only obviously identifiable prey item in the scat was the shore crab, *Hemigrapsus nudus*. No domoic acid was detected using the receptor binding assay or high performance liquid chromatography (HPLC) in urine samples collected opportunistically from three animals. Liver biopsies evaluated for histopathology did not reveal any major lesions.

Otter	Gender	Age	PBMC Molecules p450/100ng total RNA	Liver Molecules p450/100ng total RNA
WA099	F	3	17.90E+06	0.27E+06
WA084	F	4	0.23E+06	NA
WA095	F	4	NA	4.75E+06
WA100	F	4	ND	0.30E+06
WA086	F	5	3.67E+06	2.34E+06
WA094	F	5	ND	3.24E+06
WA089	F	6	0.51E+06	2.37E+06
WA091	F	7	2.94E+06	1.66E+06
WA090	F	9	ND	2.40E+06
WA093	F	10	54.10E+06	2.44E+06
WA096	F	10	3.10E+06	2.16E+06
WA088	F	13	0.19E+06	1.14E+06
WA087	М	<1	0.25E+06	NA
WA092	М	<1	0.32E+06	0.20E+06
WA098	М	3	0.07E+06	1.16E+06
WA097	М	8	NA	1.42E+06
Females	Mean		10.3E+06	2.1E+06
n = 12	Std dev		18.6E+06	1.3E+06
	Min		0.19E+06	0.27E+06
	Max		54.1E+06	4.8E+06
Males	Mean		0.21E+06	0.93E+06
n = 4	Std dev		0.13E+06	0.64E+06
	Min		0.07E+06	0.20E+06
	Max		0.32E+06	1.4E+06
All	Mean		7.57E+06	1.9E+06
n = 16	Std dev		16.3E+06	1.3E+06
	Min		0.07E+06	0.20E+06
	Max		54.1E+06	<b>4.8E+06</b>
	Median		0.51E+06	1.91E+06

Table 16. Cytochrome P450 (CYP1A1) Blood (PBMC) and Liver AssayResults for 2001 Live Captured Sea Otters

ND = no data, sample not viable for analysis; NA = not analyzed. Mean and median for PBMC calculated using n=11; mean and median for liver calculated using n=14.

Tuble 1	1.00		Incloudy I		ave Cup	ureu Sea Ollo			
Otter	Gender	Age	Canine Distemper Virus (CDV)	Dolphin Morbillivirus (DMV)	Phocine Distemper Virus (PDV)	Porpoise Morbillivirus (PMV)	Toxoplasma gopendingi modified agglutination test (MAT)	Neospora Agglutination Test (NAT)	Sarcocystis sp.
WA115	F	<1	< 8	< 8	< 8	< 8	200	< 40	NA
WA099	F	3	≥256	96	128	48	< 25	25	negative
WA109	F	3	< 8	< 8	< 8	< 8	< 25	< 40	NA
WA084	F	4	≥256	96	192	32	> 200	< 25	negative
WA095	F	4	64	24	48	8	< 25	< 25	negative
WA100	F	4	48	48	96	32	> 200	< 25	pending
WA111	F	4	48	12	48	24	< 25	< 40	NA
WA086	F	5	≥ 256	32	128	16	> 200	< 25	500
WA094	F	5	≥ 256	64	≥256	32	> 200	< 25	500
WA089	F	6	192	16	≥256	24	< 25	25	negative
WA106	F	6	64	16	64	32	> 200	40	NA
WA114	F	6	≥ 256	128	128	64	200	160	NA
WA091	F	7	≥256	96	192	32	< 25	< 25	negative
WA090	F	9	64	24	48	8	> 200	50	negative
WA103	F	9	48	8	24	12	> 200	40	NA
WA093	F	10	< 8	< 8	< 8	< 8	< 25	< 25	negative
WA096	F	10	32	25	32	16	> 200	< 25	negative
WA088	F	13	48	8	32	8	< 25	< 25	negative
WA054	F	15	16	< 8	8	< 8	< 25	160	pending
WA110	F	16	96	24	64	12	> 200	< 40	NA
WA092	Μ	<1	< 8	< 8	< 8	< 8	> 200	< 25	100
WA104	Μ	<1	< 8	< 8	< 8	< 8	< 25	< 40	NA
WA113	Μ	2	32	<8	16	< 8	< 25	40	NA
WA098	Μ	3	96	32	64	24	> 200	25	negative
WA108	Μ	3	< 8	< 8	< 8	< 8	> 200	40	pending
WA105	Μ	5	≥256	64	96	48	< 25	40	pending
WA107	Μ	6	64	24	48	24	> 200	40	pending
WA112	Μ	6	16	< 8	12	< 8	100	80	pending
WA097	Μ	8	192	16	128	16	> 200	25	500
WA102	Μ	10	192	16	96	16	> 200	160	pending

## Table 17. Serum Antibody Results for Live Captured Sea Otters

NA = not analyzed; Bold indicates positive antibody titers; No animals were positive for antibodies to *Brucella*, *Leptospira hardjo*, *Leptospira icterohaemorrhagiae*, *Leptospira pomona*, *Leptospira bratislava*, or *caliciviruses*; Sea otter WA095 was the only animal that tested positive for antibodies to *Leptosira grippotyphosa*.

### **Relationship Between Biomarkers Contaminant Concentrations**

Although sea otters are a high trophic level consumer, concentrations of chemicals that tend to bioaccumulate, such as heavy metals, PCBs, chlorinated hydrocarbons insecticides (DDT and derivatives), and butyltins, were low in the animals captured off Washington in 2001 and 2002. However, there were many significant correlations between contaminant concentrations and the Vitamin A and thyroid biomarker responses. Vitamin A responses were significantly correlated with several metals, butyltins, PCBs and several aliphatics, while thyroid responses were significantly correlated with a few of the metals. The significant correlations are listed in Table 18.

Cytochrome P450 induction was elevated in PMBC samples from two of the otters, one of which also had the highest aliphatic and aromatic hydrocarbon concentrations, the other of which the liver was not analyzed for hydrocarbons; however the latter did exhibit elevated serum enzyme alkaline phosphatase. Cytochrome p450 in the liver was negatively correlated with total PCB (p = 0.01; Table 18).

### **Blood Health Screens**

Hematology values (cell blood counts or CBCs) and serum chemistry results are presented in Tables 19 and 20, respectively. Cell blood counts varied little among otters while serum chemistry parameters were more variable. Much of the variability in blood and serum values is explained by one or two otters. Sea otter WA102, the oldest male otter captured (10 years old) had the lowest platelet count at 64 x 1,000/UL – the range excluding this animal was from 160 to 405 x 1,000/UL, and for all but three animals, the platelet count was above 200 x 1,000/UL. WA102 also had one of the two lowest white blood cell counts at 3.8 10<sup>3</sup> cells/mm<sup>3</sup>. One animal, WA054, a 15 year old female, drove the high variability observed in percent neutrophils and lymphocytes, with the highest percent neutrophils (84%) and the lowest percent lymphocytes (10%) – and likewise, the lowest white blood cell count at 2.9 10<sup>3</sup> cells/mm<sup>3</sup>. Percent eosinophils was also highly variable, due to two otters with high percentages, WA089 at 30 percent and WA088 at 21 percent – the other animals ranged from 0 to 16 percent. WA089 also had the lowest glucose level at 75 mg/dL. WA106 had the highest glucose level, 192 mg/dL. Blood urea nitrogen (BUN) ranged from 39 (WA098) to 81 (WA103) mg/dL, with much of the variability explained by animals at either end of the range.

<u>Vitamin A</u> All-Trans Retir	yl Palmitate	(RP)		Total Retinol (TR =	[R]+0.55*[RP])			 R:RP ratio	i.		
Contaminant	Correlation Coefficient	Signific ance	N	Contaminant	Correlation Coefficient	Signific ance	N	Contaminant	Correlation Coefficient	Signific ance	N
Cadmium	0.9429	0.0048	6	Iron	-0.4166	0.0383	25	Iron	-0.4520	0.0233	25
Iron	-0.5289	0.0066	25	Magnesium	-0.3997	0.0478	25	Magnesium	-0.5690	0.0030	25
Magnesium	-0.5490	0.0045	25	Selenium	-0.6070	0.0013	25	Zinc	-0.5548	0.0040	25
Selenium	-0.4778	0.0157	25	Zinc	-0.6048	0.0014	25	Dibutyltin	-0.5536	0.0323	15
Zinc	-0.6533	0.0004	25	Total Butyltin	-0.4651	0.0191	25	Sum of PCBs <sup>a</sup>	-0.6209	0.0009	25
Dibutyltin	-0.7643	0.0009	15	Sum of PCBs <sup>a</sup>	-0.5674	0.0031	25	Sum of PCBs <sup>b</sup>	-0.6246	0.0008	25
Total Butyltin	-0.5266	0.0068	25	Sum of PCBs <sup>b</sup>	-0.5662	0.0032	25	Octacosane	-0.6095	0.0159	15
Sum of PCBs <sup>a</sup>	-0.7432	0.0000	25	Eiocosane	0.6667	0.0499	9				
Sum of PCBs <sup>b</sup>	-0.7415	0.0000	25	Heptadecane	0.8112	0.0014	12				
Eiocosane	0.7333	0.0246	9	Hexadecane	0.6014	0.0386	12				
Heptadecane	0.7622	0.0040	12	Nonadecane	0.7091	0.0146	11				
Hexadecane	0.6294	0.0283	12	Octacosane	-0.6416	0.0099	15				
Nonadecane	0.6273	0.0388	11	Octadecane	0.7902	0.0022	12				
Octacosane	-0.7006	0.0036	15								
Octadecane	0.7203	0.0082	12								
Pentadecane	0.7818	0.0075	10								

# Table 18. Significant Correlations Between Biomarkers, Positive Pathogen Titers and Contaminants Vitamin A

<u>Thyroid</u>							
Tetraiodothyro	onine/thyroxin	e (T4)		Triiodothyronine (1	[3]		
Contaminant	Correlation Coefficient	Signific ance	N	Contaminant	Correlation Coefficient	Signific ance	N
Selenium	-0.4195	0.0263	28	Copper	-0.5873	0.0032	23
Strontium	0.5897	0.0265	14				
Eicosane	-0.8547	0.0143	7				

<u>p450</u> p450 Liver			
Contaminant	Correlation Coefficient	Signific ance	N
Sum of PCBs <sup>a</sup>	-0.6484	0.0121	14
Sum of PCBs <sup>b</sup>	-0.6308	0.0156	14

	per Virus (CDV)		
Contaminant	Correlation Coefficient	Signific ance	N
Mercury	-0.5223	0.0458	15

a. total PCBs calculated using 1/2 detection limit for non-detects; b. total PCBs calculated without non-detects

Regression analysis revealed several significant relationships, though the relevance is not known and  $R^2$  values were all low, meaning that little of the relationship could be explained by the significant predictors. Regression analysis results include the following significant relationships: (1) between gender and age and white blood cell count (p = 0.002, partial t-test coefficient = 0.001 for gender and 0.016 for age,  $R^2 = 0.611$ ), with a significant difference in white cell counts between males (5.69) and females (7.41) (n=30, p=0.009); (2) between age and red blood cell count (p = 0.028,  $R^2 = 0.16$ ); (3) between body weight and mean corpuscular volume (p = 0.034,  $R^2 = 0.151$ ); (4) between gender and mean corpuscular hemoglobin concentration (p = 0.007, mean for males is 33.9 g/dL, mean for females is 35.0 g/dL; and (5) between age and percent neutrophils (p = 0.045,  $R^2 = 0.154$ ). Significant relationships were also determined for some of the serum chemistry parameters: (1) between BUN/creatinine ratio and age (p = 0.014,  $R^2 = 0.22$ ); (2) triglycerides and age (p = 0.022,  $R^2 = 0.365$ ); (3) cholesterol and gender (p = 0.008,  $R^2 = 0.253$ ); (4) GGT and weight (p = 0.023,  $R^2 = 0.189$ ); and (5) alanine aminotransferase and age (p = 0.015,  $R^2 = 0.215$ ).

## **Fatty Acid Profiles**

Fatty acids were used to investigate the diet of the live captured otters by comparing the fatty acids among age and weight classes, sex and capture location (VanBlaricom et al. 2007). Of the eight univariate analyses of variance, results of seven were not significant and the authors believe that the eighth, a comparison among weight classes, was a false positive result. The authors found meaningful variation in concentrations of both linoleic and linolenic acids in individual otters, with values of linoleic acid varying up to 110x and linolenic acid up to 70x among individual otters.

## **Analysis of Prey Items**

Marine invertebrates were collected from the intertidal area within the Washington sea otter range for a program being conducted concurrently with the sea otter study. In addition, NOAA Status and Trends, Mussel Watch Program has a sampling station within the Washington sea otter range at Cape Flattery that was sampled in 2000 and 2002. On both these programs invertebrates that are potential prey for sea otters were analyzed for a suite of chemicals similar to those for sea otters in this study, providing information on potential dietary exposure of sea otters to contaminants (Tables 21, 22 and 23).

As might be expected, chemical concentration varied by species. In general, the predator (whelk), predatory/scavenger (crab), and grazer (limpet) contained higher concentrations of metals than did the filter feeders (mussels) (Table 21). Concentrations of metals in bivalves on the Washington coast generally have higher cadmium and arsenic concentration than those in Puget Sound possibly due to the metal-rich upwelling of sea water along the coast. Butyltin residues in mussels were determined on the Mussel Watch Program (Table 22). TBT was quantified in only one sea otter, yet it is the primary congener detected in the mussels from 2000 and 2002. Similar to sea otters, organochlorines, except for PCBs, were generally not detected or at very low concentrations in invertebrates samples. PCB concentrations followed the same pattern as metals with higher concentrations in the predators, scavengers and grazer than in filter feeders. Additional invertebrate data from the Mussel Watch Program are provided in Appendix 7, which illustrates PAH and individual organochlorine compound data.

# Table 19. Cell Blood Counts (CBC) for Live Captured Sea Otters

Table 19				ounts (	(CDC)	, 101 1		apiure		/11015						1	
Otter	Gender	Age	White Blood Cells (WBC)	Red Blood Cells (RBC)	Hemoglobin (HGB)	Hematocrit (HCT)	Mean Corpuscular Volume (MCV)	Mean Corpuscular Hemoglobin (MCH)	Mean Corpuscular Hemoglobin Concentration (MCHC)	Red Cell Distribution width (RDW)	Platelets	Mean Platelet Volume	Percent Neutrophils	Percent Lymphocytes	Percent Monocytes	Percent Eosinophils (EOS)	Percent Basophils
WA115	F	<1	8.2	5.54	18.3	53.7	97	33	34.1	NA	266	NA	32	66	0	2	0
WA099	F	3	7.8	5.32	20.6	59.9	113	38.7	34.4	14.9	204	10.1	44	35	5	16	0
WA109	F	3	7.8	5.69	20.5	60.4	106	36	33.9	NA	268	NA	48	38	4	9	1
WA084	F	4	8.4	5.45	19.1	56.8	104	35.1	33.6	14	209	10.9	42	47	2	9	0
WA095	F	4	6.9	4.77	17.6	50.2	105	37	35.2	13.1	292	11.4	42	41	6	11	0
WA100	F	4	9.6	4.67	17.2	45.3	97	36.9	38	19.8	402	12.9	44	44	2	10	0
WA111	F	4	5.4	5.14	18.8	54.5	106	36.6	34.6	NA	305	NA	35	64	1	0	0
WA086	F	5	7.8	5.05	19.6	56.5	112	38.8	34.7	15.1	217	11	62	28	5	5	0
WA094	F	5	10.7	4.94	18.0	51.2	104	36.4	35.2	12.4	253	10.2	74	18	6	2	0
WA089	F	6	9.3	5.27	19.7	55.1	105	37.5	35.8	13.3	343	9.4	44	22	4	30	0
WA106	F	6	7.5	5.34	19.4	55.4	104	36.4	35.1	NA	310	NA	NA	NA	NA	NA	NA
WA114	F	6	6.9	5.25	20.0	57.8	110	38.2	34.7	NA	291	NA	60	23	5	12	0
WA091	F	7	7.3	5.24	20.0	56.1	107	38.2	35.7	12.7	206	11.4	54	28	10	6	2
WA090	F	9	6.9	5.33	20.0	56.6	106	37.6	35.3	14	252	11.9	53	35	5	7	0
WA103	F	9	8.1	5.05	19.0	54.8	109	37.6	34.7	NA	405	NA	71	23	0	5	1
WA093	F	10	7.4	5.21	19.6	55.9	107	37.7	35.1	13.3	160	11.7	67	23	6	3	1
WA096	F	10	5.4	4.44	16.5	46.6	105	37.1	35.4	12.6	294	10.4	46	43	5	5	1
WA088	F	13	7.8	5.15	18.5	51.6	100	36	36	14.7	299	9.7	33	42	3	21	1
WA054	F	15	2.9	4.27	14.2	43.5	102	33.2	32.6	NA	NA	NA	84	10	6	0	0
WA110	F	16	6.1	4.91	18.2	52.3	106	37.2	34.9	NA	317	NA	55	42	1	2	0
WA092	М	<1	6.0	5.06	18.0	51.4	102	35.6	35.1	14.5	295	8.9	50	36	8	6	0
WA104	М	<1	5.5	4.85	17.2	51.5	106	35.6	33.5	NA	226	NA	NA	NA	NA	NA	NA
WA113	М	2	4.0	5.37	19.1	55.4	103	35.6	34.5	NA	261	NA	NA	NA	NA	NA	NA
WA098	М	3	8.1	5.22	20.1	59.6	114	38.4	33.6	13.7	191	12.3	38	46	6	10	0
WA108	М	3	5.4	5.75	19.9	58.1	101	34.6	34.2	NA	263	NA	NA	NA	NA	NA	NA
WA105	М	5	6.4	5.53	20.0	60.8	110	36.1	32.8	NA	250	NA	61	31	3	4	1
WA107	М	6	6.1	5.48	18.8	56.7	104	34.2	33.1	NA	264	NA	51	36	7	5	1
WA112	Μ	6	4.3	4.38	17.7	51.5	118	40.4	34.3	NA	269	NA	NA	NA	NA	NA	NA
WA097	М	8	7.3	4.54	18.0	52.6	116	39.6	34.2	13.6	235	9.9	37	53	5	4	1
WA102	М	10	3.8	5.24	18.6	56.1	107	35.6	33.2	NA	64	NA	57	31	6	6	0
Females	Mea	n	7.4	5.10	18.7	53.7	105	36.8	35.0	14.2	279	10.9	52	35	4	8	0.4
n = 20	Std o	dev	1.7	0.355	1.53	4.55	4.17	1.56	1.08	2.00	64.4	1.0	14	15	3	8	0.6
	Min		2.9	4.27	14.2	43.5	97.0	33	32.6	12.4	160	9.4	32	10	0	0	0
	Max		10.7	5.69	20.6	60.4	113	38.8	38.0	19.8	405	12.9	84	66	10	30	2
Males	Mea	n	5.7	5.14	18.7	55.4	108	36.6	33.9	13.9	232	10.4	49	39	6	6	0.5
n = 10	Std o	lev	1.4	0.439	1.03	3.50	6.10	2.13	0.723	0.493	65.4	1.7	10	9	2	2	0.5
	Min		3.8	4.38	17.2	51.4	101	34.2	32.8	13.6	64.0	8.9	37	31	3	4	0
	Max		8.1	5.75	20.1	60.8	118	40.4	35.1	14.5	295	12.3	61	53	8	10	1
All	Mea		6.8	5.12	18.7	54.3	106	36.7	34.6	14.1	262	10.8	51	36	4	8	0.4
n = 30	Std o	dev	1.8	0.378	1.37	4.24	4.98	1.73	1.09	1.78	67.5	1.1	13	13	2	7	0.6
	Min		2.9	4.27	14.2	43.5	97.0	33	32.6	12.4	64.0	8.9	32	10	0	0	0
	Max		10.7	5.75	20.6	60.8	118	40.4	38.0	19.8	405	12.9	84	66	10	30	2
									ont: MCV								

WBC,  $10^3$  cells/mm<sup>3</sup>; RBC,  $10^6$  cells/mm<sup>3</sup>; HGB, g/dL; HCT, percent; MCV, fL; MCH, pg; MCHC, g/dL; RDW, %, Platelets, x1000/uL. Mean platelet volume,  $10^3/\mu$ L; NA = not analyzed

	Gender	Age	Glucose (mg/dL)	Blood Urea Nitrogen (BUN), (mg/dL)	Creatinine (mg/dL)	<b>BUN/Creatinine Ratio</b>	Uric Acid (mg/dL)	Sodium (mequiv/L)	Potassium (mequiv/L)	Na/K Ratio	Chloride (mequiv/dL)	Carbon Dioxide (meq/L)	Calcium (mg/dL)	Phosphorus mg/dL)	Total Protein (g/dL)	Albumin (A), (g/dL)	Globulin (calculated) (G), (g/dL)	A/G Ratio
WA115	F	<1	146	51	1	51	NA	151	4.4	34	108	24	10	7.3	5.9	3.1	2.8	1.1
WA099	F	3	115	40	0.5	80	2	155	4	NA	111	21	9.9	2.7	7.4	3.1	4.3	0.7
WA109	F	3	120	47	0.5	94	NA	152	4.7	32	111	19	10.1	4.3	6.4	3.3	3.1	1.1
WA084	F	4	132	72	0.6	120	1.6	154	4.2	NA	113	19	9.4	4.3	7.2	2.9	4.3	0.7
WA095	F	4	101	62	0.6	103.3	2.1	153	4.9	NA	112	21	9.4	6.5	6.5	2.9	3.6	0.8
WA100	F	4	93	62	0.5	124	2.1	153	4.5	NA	116	19	10.6	3.8	6.8	3.0	3.8	0.8
WA111	F	4	106	46	0.6	76.67	NA	152	4.7	32	109	24	9.5	5.3	6.3	3.1	3.2	1.0
WA086	F	5	125	74	0.5	148	2	154	4.3	NA	115	18	9.2	3.2	6.6	2.8	3.8	0.7
WA094	F	5	152	44	0.5	88	1.7	154	3.7	NA	113	22	9.2	3.2	7.6	2.9	4.7	0.6
WA089	F	6	75	51	0.5	102	2.1	155	4.5	NA	114	25	9	4.3	6.5	2.6	3.9	0.7
WA106	F	6	192	43	0.6	71.67	NA	155	4.6	34	112	22	9	3.3	6.3	2.9	3.4	0.9
WA114	F	6	125	62	0.4	155	NA	154	4.6	33	113	22	8.6	3.0	7.5	2.9	4.6	0.6
WA091	F F	7 9	120	42	0.5	84	1.7	158	4.3	NA	115	24	9.1	5.3	6.5	3.0	3.5	0.9
WA090	г F	9	96 121	69 81	0.7	98.6 162	3 NA	155 160	4.8	NA 31	112 119	20	9.3 8.8	5.4	7.1 7.4	3.1 2.5	4.0	0.8
WA103 WA093	г F	9	131	44	0.3	146.7	NA	155	4.5	NA	119	20 22	0.0 9.8	4.8			4.9	0.5
WA095 WA096	г F	10	153 98	44	0.5	80	1.1 1.5	155	4.5	NA	113	22	9.8 9.3	6.7 4.1	5.5 6.8	3.1 2.6	2.4 4.2	1.3 0.6
WA090 WA088	F	10	101	52	0.5	104	1.5	151	4.3	NA	112	20	9.3 8.9	3.9	6.9	2.0	4.2	0.6
WA033 WA054	F	15	139	71	0.5	142	NA	162	4.2	39	112	23	9.8	5.8	7.3	3.2	4.1	0.8
WA110	F	16	95	64	0.4	160	NA	153	5	31	110	22	9.6	4.9	6.9	3.1	3.8	0.8
WA092	M	<1	122	50	0.5	100	1.4	157	4.4	NA	117	25	8.7	3.5	6.6	2.8	3.8	0.7
WA104	M	<1	161	42	0.3	140	NA	156	5.2	30	115	20	10.3	7.2	5.1	3.1	2.0	1.6
WA113	M	2	107	43	0.6	71.67	NA	155	4.6	34	115	24	9.3	4.5	6.9	3.1	3.8	0.8
WA098	M	3	120	39	0.5	78	2	150	4.8	NA	112	20	10.1	5.0	6.7	3.0	3.7	0.8
WA108	М	3	99	47	0.5	94	NA	153	4.7	33	111	22	9.5	3.1	6.7	3.3	3.4	1.0
WA105	М	5	131	60	0.8	75	NA	159	4.6	35	116	24	9	3.0	6.3	2.9	3.4	0.9
WA107	М	6	103	66	0.6	110	NA	152	4.6	33	111	21	9.1	4.4	6.8	3.0	3.8	0.8
WA112	М	6	87	73	0.6	121.7	NA	155	4.7	33	115	20	8.5	3.5	6.4	2.8	3.6	0.8
WA097	М	8	86	44	0.8	55	3.2	152	4.2	NA	114	23	9.3	3.1	7.7	2.7	5.0	0.5
WA102	М	10	151	77	0.6	128.3	NA	155	4.5	34	116	20	9.1	4.7	6.4	2.9	3.5	0.8
Females	Me	an	121	56	0.5	110	1.89	154	4.5	33	113	22	9.4	4.6	6.8	2.9	3.8	0.8
n = 20	Std	dev	27	13	0.1	33.2	0.46	2.78	0.34	2.6	2.84	2.0	0.5	1.3	0.55	0.21	0.6	0.2
	Mi	n	75	40	0.3	51	1.1	151	3.7	31	108	18	8.6	2.7	5.5	2.5	2.4	0.5
	Ma	x	192	81	1	162	3	162	5.1	39	119	25	10.6	7.3	7.6	3.3	4.9	1.3
Males	Me	an	117	54	0.6	97.4	2.2	154	4.6	33	114	22	9.3	4.2	6.6	3.0	3.6	0.87
n = 10	Std	dev	25	14	0.1	27.6	0.92	2.67	0.26	1.6	2.15	2.0	0.56	1.3	0.65	0.18	0.7	0.29
	Mi	n	86	39	0.3	55	1.4	150	4.2	30	111	20	8.5	3.0	5.1	2.7	2.0	0.5
	Ma	x	161	77	0.8	140	3.2	159	5.2	35	117	25	10.3	7.2	7.7	3.3	5.0	1.6
All	Me	an	119	55	0.6	105	1.95	154	4.5	33	113	22	9.4	4.5	6.7	2.9	3.8	0.82
n = 30	Std	dev	26	13	0.1	31.5	0.55	2.7	0.32	2.1	2.66	2.0	0.51	1.3	0.58	0.2	0.7	0.23
	Mi	n	75	39	0.3	51	1.1	150	3.7	30	108	18	8.5	2.7	5.1	2.5	2.0	0.5
	Ma	X	192	81	1	162	3.2	162	5.2	39	119	25	10.6	7.3	7.7	3.3	5.0	1.6

Table 20.	Serum	Chemistry	<b>Results for</b>	Live	Captured Sea Otters
		Chempery			

NA = not analyzed

	<u>e 20.</u>	Ser	um C	nemi	stry F	tesuit	s for L	ive Ca	apture	a sea	Otters;	cont	nuea				
Otter	Gender	Age	Triglycerides (mg/dL)	Cholesterol (mg/dL)	Total Bilirubin (mg/dL)	Direct Bilirubin (mg/dL)	Gamma Glutamyltransfera se (GGT)	Alkaline Phosphatase (AP)	Lactic Dehydrogenase (LD)	Aspartate Aminotransferase (AST)	Alanine Aminotransferase (ALT)	Iron (µg/dL)	Hemolytic Index	Lipemic Index	Icteric Index	Osmolality	Anion Gap
WA115	F	<1	NA	147	0.2	NA	14	332	NA	182	163	NA	38	10	0	315	19
WA099	F	3	49	162	0.2	0	11	110	253	98	83	237	NA	NA	NA	NA	NA
WA109	F	3	NA	129	0.2	NA	13	171	NA	201	176	NA	52	9	0	315	22
WA084	F	4	80	303	0.3	0	13	136	306	215	234	338	NA	NA	NA	NA	NA
WA095	F	4	57	211	0.1	0	20	115	323	161	217	191	NA	NA	NA	NA	NA
WA100	F	4	49	150	0.2	0.1	5	82	336	126	106	195	NA	NA	NA	NA	NA
WA111	F	4	NA	170	0.1	NA	14	116	NA	145	122	NA	66	10	0	314	19
WA086	F	5	87	187	0.2	0	13	91	212	192	196	326	NA	NA	NA	NA	NA
WA094	F	5	69	195	0.2	0	16	91	308	157	214	265	NA	NA	NA	NA	NA
WA089	F	6	36	183	0.2	0	10	75	396	172	146	188	NA	NA	NA	NA	NA
WA106	F	6	NA	144	0.1	NA	12	131	NA	120	127	NA	60	9	0	323	21
WA114	F	6	NA	149	0.1	NA	17	102	NA	150	178	NA	58	24	0	324	19
WA091	F	7	54	161	0.3	0.1	17	96	522	300	210	263	NA	NA	NA	NA	NA
WA090	F	9	60	220	0.3	0	31	148	494	355	373	260					
WA103	F	9	NA	199	0.1	NA	17	82	NA	283	305	NA	53	8	0	343	21
WA093	F	10	38	152	0.2	0.1	14	309	338	129	176	190	NA	NA	NA	NA	NA
WA096	F	10	44	138	0.2	0	13	78	245	117	108	116	NA	NA	NA	NA	NA
WA088	F	13	34	136	0.2	0	16	85	295	160	184	218	NA	NA	NA	NA	NA
WA054	F	15	NA	235	0.1	NA	51	60	NA	293	403	NA	37	7	0	342	20
WA110	F	16	NA	123	0.1	NA	10	116	NA	231	180	NA	65	6	0	322	21
WA092	Μ	<1	88	166	0.3	0	20	134	437	264	221	185	NA	NA	NA	NA	NA
WA104	М	<1	NA	196	0.1	NA	9	242	NA	116	139	NA	28	10	0	324	21
WA113	М	2	NA	128	0.2	NA	16	102	NA	206	154	NA	80	5	0	318	16
WA098	М	3	103	157	0.2	0	11	122	330	130	130	242	NA	NA	NA	NA	NA
WA108	М	3	NA	152	0.1	NA	14	164	NA	225	185	NA	51	6	0	316	20
WA105	М	5	NA	132	0.1	NA	17	122	NA	149	126	NA	69	6	0	333	19
WA107	М	6	NA	123	0.1	NA	17	102	NA	136	136	NA	66	8	0	321	20
WA112	Μ	6	NA	124	0.1	NA	22	103	NA	192	153	NA	67	10	0	328	20
WA097	М	8	44	137	0.2	0.1	21	95	264	347	280	148	NA	NA	NA	NA	NA
WA102	М	10	NA	122	0.2	NA	44	140	NA	474	438	NA	55	4	0	333	19
Females	Mea	n	55	175	0.2	0.0	16	126	336	189	195	232	54	10	0	325	20
n = 20	Std	dev	17	43.6	0.1	0.0	9.6	71.8	94	70	83	63	11	6	0	11.6	1.2
	Min		34	123	0.1	0	5	60	212	98	83	116	37	6	0	314	19
	Max	[	87	303	0.3	0.1	51	332	522	355	403	338	66	24	0	343	22
Males	Mea	n	78	144	0.2	0.0	19	133	344	224	196	192	59	7	0	325	19
n = 10	Std	dev	31	24	0.1	0.1	9.7	44	87	113	98	47	17	2	0	6.87	1.6
	Min		44	122	0.1	0	9	95	264	116	126	148	28	4	0	316	16
	Max	[	103	196	0.3	0.1	44	242	437	474	438	242	80	10	0	333	21
All	Mea	n	59	164	0.2	0.0	17	128	337	201	195	224	56	9	0	325	20
n = 30	Std	dev	21	40.6	0.1	0.0	9.5	63	90	86	87	61	14	5	0	9.38	1.4
	Min		34	122	0.1	0	5	60	212	98	83	116	28	4	0	314	16
	Max	[	103	303	0.3	0.1	51	332	522	474	438	338	80	24	0	343	22

Table 20. Serum Chemistry Results for Live Captured Sea Otters; continued

Max1033030.30.1513325224744383388024034322NA=not analyzed; GGT, gamma glutamyltransferase (1 U/L = 16.67 nkat/L); AP, alkaline phosphatase (1 U/L = 16.67 nkat/L);LD, lactic dehydrogenase (1 U/L = 16.67 nkat/L); AST, aspartate aminotransferase (1 U/L = 16.67 nkat/L); ALT, alanine aminotransferase (1 U/L = 16.67 nkat/L).

Species	Study	N		Aluminum	Arsenic	Cadmium	Chromium	Copper	Iron	Mercury	Magnesium	Manganese	Nickel	Lead	Selenium	Silver	Strontium	Tin	Vanadium	Zinc
Hemigrapsus nudus	OCNMS	18	Mean	266		0.53		16.4	286		7721	21.8				3.14	2131		1.60	45.3
Shore crab	Biomarker		std dev	150		0.13		7.2	227		800	7.2				0.38	210		0.36	9.06
			Min	93		0.50		7.2	70		6210	12.4				2.57	1700		1.18	29.6
			Max	603		1.04		30.5	816		9000	32.4				4.12	2520		2.36	62.1
Mytilus californianus	OCNMS	22	Mean	316		8.28		6.1	421		5594	12.4				0.50	54		1.59	122
Mussel	Biomarker		std dev	224		4.57		1.5	224		1379	4.6				0.00	19		0.96	30.6
			Min	43.8		3.56		4.45	110		3280	6.64				0.5	27.4		0.5	72.1
			Max	824		17.00		10.9	890		7900	22.8				0.50	92		3.63	183
Mytilus edulis/trossulus	OCNMS	1	Mean	701		5.66		6.0	531		5700	22.9				0.50	68		3.34	119
Mussel	Biomarker																			
Nucella lamellosa	OCNMS	7	Mean	110		37.73		12.5	159		6487	11.1				3.14	610		1.17	150
Whelk	Biomarker		std dev	41		25.06		12.4	121		2789	4.0				1.27	248		0.11	85.5
			Min	55		14.90		0.5	79		2710	7.6				1.69	313		1.01	62.1
			Max	185		84.10		30.6	429		10500	18.2				5.01	974		1.30	301
Tectura scutum	OCNMS	1	Mean	2050		28.20		11.5	3360		3890	50.4				0.50	70		7.81	59.3
Limpet	Biomarker																			
<i>Mytilus edulis</i> mussel	NOAA Mussel Watch	U	Mean	369	11.7	12.8	1.65	7.92	766	0.195		12.4	2.13	0.756	2.81	0.99		0		129

Table 21. Metals in Invertebrates Collected in the Intertidal Area of the Washington Sea Otter Range (ppm, dry weight)

N = number of composite samples, with composites made up of at least 10 individuals from the same location. U = unknown. Blank values indicate not analyzed.

# Table 22. Butyltins in Composite Mussel Samples from NOAA Status and Trends,Mussel Watch Program Cape Flattery Site (ppb, dry weight)

Year and Species	Monobutyltin	Dibutyltin	Tributyltin	Total Butyltins
2000 Mytilus californianus	0	0	9.61	9.61
2002 Mytilus edulis	0.645	1.50	8.17	10.32

				Total	Total	Total
Species	Ν		Total PCB	HCH	Chlordane	DDTs
Hemigrapsus nudus	18	mean	4.42	0.29	0.02	0.64
Shore crab		std dev	11.4	0.31	0.05	1.13
		min	ND	ND	ND	ND
		max	49	1.1	0.17	3.3
Mytilus californianus	22	mean	2.49	0.54	0.18	0.52
Mussel		std dev	0.56	0.28	0.42	0.65
		min	1.50	0.18	ND	ND
		max	3.6	1.4	1.4	2.1
Mytilus edulis/trossulus	1	mean	3.2	0.71	0.24	0.13
Mussel						
Nucella emarginata	15	mean	8.0	0.21	0.75	0.49
Whelk		std dev	5.0	0.25	2.12	0.36
		min	2.4	ND	ND	ND
		max	24	0.7	7.8	1
Nucella lamellosa	9	mean	4.9	0.40	0.17	0.89
Whelk		std dev	2.8	0.36	0.17	0.70
		min	2.0	ND	ND	ND
		max	11.5	0.975	0.405	2.4
Tectura scutum	14	mean	4.4	0.46	0.06	0.12
Limpet		std dev	3.0	0.25	0.24	0.14
		min	2.0	ND	ND	ND
		max	13	0.95	0.88	0.4

# Table 23. Organochlorines by Compound Group: Invertebrates Collected in the Intertidal Area of the Washington Sea Otter Range (ppb, wet weight)

N = number of composite samples, with composites made up of at least 10 individuals from the same location.

ND=Not detected

## **V. DISCUSSION**

The Washington sea otter population has not faced the extent of declines that the California and two of the three Alaska populations have faced in recent years, yet growth of the Washington population has slowed before reaching its estimated carrying capacity. A greater number of mortalities and a lower census number than expected in 2000, and potential range expansion leading to increased exposure to biotoxins and anthropogenic effects led to concerns about the Washington population and development of this study. This study was conducted to obtain baseline data for chemical contaminants and pathogens, compare across populations, and evaluate if health stressors in the Washington population could be identified. Data for archived stranded (beach-cast) animals are also presented, but comparison of the beach-cast animals to the live captured animals was not possible because of the different sample matrices. Livers of beach-cast animals were analyzed, while most of the chemical analyses performed on tissues from the live captured animals were on whole blood.

Contaminant levels are low in Washington sea otters, with very few exceptions. A discussion of each group of analytes is presented below, comparing Washington otters to those in Alaska and California, or to confamilial species or other marine mammals where literature was available. Whole blood chemical contaminant data were not readily available so much of the discussion of contaminants pertains to the beach-cast animals. In general, Washington sea otters appear to reflect chemical concentrations and blood and serum chemistry most similar to California sea otters versus stock in Alaska. Of the Alaska stocks, Washington sea otters appear to reflect conditions most similar to those of the Aleutian Islands, which makes sense as the otters came from Amchitka Island.

### Metals

Metals concentrations in the liver of Washington beach-cast animals were similar to those reported in the literature for other marine mammals in the Northeast Pacific but within the range of those reported for confamilial species (Tables 5, 6, 24 and 25). Chromium, copper, mercury, manganese, strontium and zinc were all within the range reported by Kannan et al. (2008) in California, Washington, and Alaska otters. Cadmium levels were similar to those in Alaska sea otters, but considerably less than those reported for California otters. Aluminum, cadmium, copper, selenium and zinc concentrations in the liver of Washington beach-cast otters were generally similar to those reported in gray whales from the west coast of North America (Varanasi et al. 1993) and higher than those reported for confamilial species. Mercury concentrations in Washington sea otters were higher than those reported for gray whales, but similar to those reported for confamilial species, Alaska beluga whales and dolphins from other areas. The detection limit for lead was not low enough to allow for comparison as concentrations in liver of the beach-cast animals were below the detection limit of 0.5 ppm. As noted above, a 2 year old male (16961-04) had the highest concentrations of copper, mercury and selenium – concentrations that were considerably higher than those recorded in confamilial species, only two of which were from the Pacific Northwest.

			Silver	Aluminum	Barium	Cadmium	Chromium	Copper	Iron	Mercury	Magnesium	Manganese	Molybdenum	Lead	Selenium	Strontium	Vanadium	Zinc	
Species	Region	Ν				Ű	-						Σ		_		Ň		Source/Study
Enhydra lutris	Washington	15		42.6		12.6	0.62	86.5	1419	11.6	782	12			8.6	1.3		156	Present Study
Enhydra lutris	California	6	1.5			53	4.2	110		13		12	0.69	0.35		1.5	0.36	210	Kannan et al. 2008
Enhydra lutris	Washington*	3	0.78			35	0.52	84		8.6		13	0.59	0.06		3.3	0.19	150	Kannan et al. 2008
Enhydra lutris	PWS, Alaska	2	1.6			7.6	0.49	210		11		35	1.6	1.2		0.4	0.22	310	Kannan et al. 2008
Enhydra lutris	Adak Island	2	0.9			4.9	0.61	56		1.3		22	0.56	0.08		2.1	0.23	140	Kannan et al. 2008
Enhydra lutris	Kamchatka	5	0.84			15	0.49	47		1.8		8.6	0.68	0.23		1.1	0.05	110	Kannan et al. 2008
Enhydra lutris	California	80	1.59		0.02	91.9	0.53	133		17.8		16.9	0.52	0.22		1.46	0.18	230	Kannan et al. 2006
Lutra lutra	Great Britain	51					0.27	28.27				7.637						111.67	Mason and Stephenson, 2001
Lutra lutra	Ireland	39					0.024	23.77				4.91						83.8	Mason and Stephenson, 2001
Lutra lutra	Denmark	65					0.081	21.1				3.52						92.13	Mason and Stephenson, 2001
Lutra lutra	Europe	12 (juvenile)				0.1*		51.3*		1.2*								132.3*	Hyvarinen et al. 2003
Lutra lutra	Europe	10 (female)				0.58*		33*		3.8*								101*	Hyvarinen et al. 2003
Lutra lutra	Europe	9 (male)				0.6*		35.8*		5.5*								10.94*	Hyvarinen et al. 2003
Lutra lutra	Austria	15				0.36		38.7		1.01*				0.37				92.6	Gutleb et al. 1998
Lutra lutra		7				0.31		17.1		0.65*				0.83				96.2	Gutleb et al. 1998
Lutra lutra	Czech Republic	5				1.51		23.1		14.43*				0.39				162.4	Gutleb et al. 1998
Mustela vison		20		3.56	0.07	0.16	1.65	23.6	1009	4.28	602	8.71	1.19	0.31	2.28	0.37		95	Harding et al. 1999
Lutra canadensis	PNW			2.77	0.11	0.42	1.24	24.9	1121	2.68		10.8		0.77	1.92	0.21		80	Harding et al. 1999

Table 24. Literature Summary of Mean Concentrations of Metals in Liver of Sea Otter and Confamilial Species (ppm, dry weight)

All values are dry weight except those marked with asterisk (\*), which are wet weight and can be compared to Table 5.

Washington beach cast sea otters used in the Kannan et al. 2008 study likely include some of the animals used in the present study

Species	Belu	·	Gray v		Gray	vhale	Gray whale	Striped and bottlenose dolphins	Bottlenose dolphin, common dolphin, melon-headed whale
			Northern Paci						East Coast of
Location	Alas		Coa		WA, CA,		Bering Sea	Mediterranean	Australia
Weight basis	wet we	eight	dry we	eight	wet w	eight	wet weight	wet weight	wet weight
Ν	24		5		10	)	5	23	6
	mean $\pm$ std dev	range	mean $\pm$ std dev	range	mean $\pm$ SE	mean $\pm$ SE range 1		range	range
Silver	$18.56\pm12.58$	0.637-107.4			$0.017\pm0.003$	0.01-0.02	$0.31\pm0.064$		
Aluminum					32 ± 15	6.4-150	$4.2 \pm 2.7$		
Arsenic	$0.18\pm0.13$	0.065-0.815			$0.34\pm0.062$	0.047-0.7	$0.32\pm0.028$		0.2-0.76
Barium					$0.061\pm0.016$	0.01-0.12			
Cadmium	$1.266 \pm 0.309$	0.455-3.645	$1.77 \pm 1.11$	0.81-3.62	$4.3 \pm 1.2$	0.06-6.2	$0.21\pm0.04$	0.07-9.0	0.02-21
Chromium					$0.16\pm0.031$	nd-0.25	$0.29\pm0.019$		0.1-1.2
Copper	$25.57 \pm 17.52$	3.97-123.8	$57.3 \pm 87.7$	3.37-228	$9.2 \pm 2.2$	0.63-25	$16 \pm 3.4$	4.3-24	2.1-18
Iron	$444.5 \pm 127.1$	100-780.5	$580 \pm 334$	165-1239	$2300\pm950$	120-4200	$340 \pm 67$	163-842	
Mercury	$19.86\pm9.48$	0.704-72.9			$0.056\pm0.012$	0.009-0.12	$0.16\pm0.061$	1.3-550	0.72-141
Magnesium	$137.2 \pm 29.0$	58.5-219							
Manganese	$2.295 \pm 0.346$	1.617-3.357	$7.2 \pm 6.57$	0.11-16.94	$3 \pm 0$		$3.1 \pm 0.18$	0.03-6.5	
Nickel			$1.84 \pm 0.22$	1.53-2.16	$0.23 \pm 0.13$	nd-0.35	$0.039 \pm 0.014$		0.05-0.56
Lead			$2.06 \pm 1.06$	0.78-3.62	$0.12 \pm 0.022$	0.02-0.27	$0.06 \pm 0.013$		<0.04-0.17
Selenium					$2 \pm 0.3$	0.35-3.4	$1.1 \pm 0.15$		1.5-58
Strontium	$12.92 \pm 8.00$	1.078-75.51			$1.1 \pm 0.56$	0.2-1.8			
Vanadium	$0.094 \pm 0.042$	0.015-0.279					$0.52 \pm 0.055$		
Zinc	$25.95 \pm 2.50$	20.9-38.53	141 ± 32	96-173	99 ± 16	1.6-160	$28 \pm 1.9$	15-115	22-144
Source/Study	Becker et al. 2000		Mendez et al. 200		Varanasi et al. 1	993	Tilbury et al. 2002	Roditi-Elsar et al. 2003	Law et al. 2003

# Table 25. Literature Summary of Metals in Liver of Marine Mammals (ppm)

Concentrations of heavy metals of potential toxicological concern in the whole blood samples from live captured animals were low (or not detected) in all samples, with only mean concentrations of selenium and zinc above 1 ppm. Concentrations in blood samples of live captured animals were not as variable as those in the liver of beach-cast animals, possibly due to the difference in the number of years over which the samples were collected. The liver is a storage organ while contaminants detected in blood reflects current transport which may also explain the difference in variability. Although a literature search was performed, no results were found for comparison of metals in whole blood.

### **Organotins – Butyltins**

Total butyltin liver concentrations in Washington beach-cast otters were within the range reported by Kannan et al. (1998) for California sea otters; however, the mean total butyltin concentration in Washington beach-cast otters was more than 15 times lower than the mean concentration reported by Kannan et al. (1998) for California sea otters (0.0724 versus 1.3 ppm) (Tables 8 and 26). Also, the contribution of tributyltin (TBT) to the sum of total butyltins was considerably less in Washington sea otters versus those reported by Kannan et al. (1998) (Tables 8 and 26). Total butyltin liver concentrations in Washington otters were greater than those reported by Murata et al. (2008) for Alaska and Russian sea otters, but less than reported for California sea otters. The California sea otter ranges includes populated areas including Monterey, Moss Landing and Santa Cruz with higher use of TBT than the less populated coastline of Washington. Butyltins degrade over time and this difference could also be due to the difference in the time frame in which the studies were conducted. These results could reflect different exposure pathways between the populations, or that exposure levels have decreased over time with the limit on usage and then the eventual ban of TBT-containing antifouling paints.

Tanabe et al. (1998) has shown that pinnipeds and otters have lower butyltin residues than cetaceans, and postulates this may be due to molting of the fur. However, from the total butyltin values reported in the literature it appears most Steller sea lion and cetacean liver concentrations are in the same range as those for Washington sea otters (Tables 8 and 26).

No tetrabutyltin was detected and TBT, the more toxic moiety, was detected in the whole blood of only one 10-year old male animal. Butyltins do not pose a risk to Washington sea otters from dietary exposure (MacLellan et al. 1998), and concentrations currently in the water column and sea otter prey should continue to decline following the ban on TBT-based paints.

Species	Location	Ν	Statistic	MBT	DBT	ТВТ	Total BTs	Source
Sea Otter	Washington	15	Mean	0.0724	0.0757	0.0524	0.201	Present
			Range	0.017 - 0.183	0.0145 - 0.208	0.0224 - 0.102	0.0553 - 0.447	Study
Sea Otter	California	28	Mean	0.0503	0.2267	0.1904	0.4711	Murata
			Range	0.0019 - 0.610	0.004 - 2.4	0.0010 - 1.3	0.021 - 4.1	et al. 2008
Sea Otter	Russia	5	Mean	0.0048	0.0023	0.0071	0.035	Murata
			Range	0.0027-0.0099	0.0076-0.059	0.002 - 0.017	0.018 - 0.079	et al. 2008
Sea Otter	Alaska	4	Mean	0.012	0.07	0.051	0.12	Murata
			Range	0.0021 - 0.028	0.0024 - 0.170	<0.0003-0.076	0.0065 - 0.27	et al. 2008
Sea Otter	Washington	3	Mean	0.015	0.12	0.071	0.21	Murata
			Range	0.0068 - 0.027	0.084 - 0.180	0.046 - 0.110	0.160 - 0.250	et al. 2008
Sea Otter	California	35	Mean	0.07	0.67	0.56	1.3	Kannan
			Range	<0.007 - 0.36	0.021 - 5.82	0.019 - 3.02	0.04 - 9.2	et al. 1998
Steller Sea	Japan	6	Mean	0.093	0.11	0.021	0.22	Kim
Lion			Range	0.052 - 0.13	0.051 - 0.14	0.016 - 0.031	0.18 - 0.3	et al. 1996
Dall's Porpoise	Japan	13	Mean	0.12	0.44	0.21	0.79	Nakata
			Range	0.047 - 0.22	0.027 - 1.1	0.08 - 0.45	0.26 - 1.8	et al. 2002
Pelagic	England &	16	Mean	0.0043	0.077	0.028	0.11	Law
Cetaceans <sup>a</sup>	Wales		Range	<.003 - 0.082	0.019 - 0.23	<.003 - 0.035	0.019 - 0.312	et al. 1999

 Table 26. Literature Summary of Butyltins (BT) in Liver of Marine Mammals Including

 Sea Otters (ppm, wet weight)

a. including white-sided dolphin, white-beaked dolphin, striped dolphin, common dolphin, Risso's dolphin, long-finned pilot whale, fin whale, minke whale, pygmy sperm whale, Sowerby's beaked whale, Blainville's beaked whale, Northern bottlenose whale

#### **Organic Compounds**

#### Aliphatic and Polycyclic Aromatic Hydrocarbons and Semi-volatile Compounds

Aliphatic and aromatic hydrocarbons were measured in liver samples from live captured animals but not in the beach-cast animals. Semi-volatiles were analyzed in liver of some of the beachcast animals and in blood of the all of the live captured animals. Concentrations of PAHs and semi-volatiles were low or not detected in live captured Washington sea otter liver and blood; however, several aliphatic hydrocarbons were detected in the liver of the live captured otters. The low levels of PAHs was expected as vertebrates, such as sea otters, rapidly metabolize PAHs to more polar compounds that are excreted to the bile and eliminated from the animal. Semivolatiles were also low or not detected in the beach-cast otter liver samples. Although a literature search was conducted, only data from the T/V Exxon Valdez studies were found for comparison to the Washington sea otter data. Ballachey and Kloecker (1997) analyzed hydrocarbon residues in tissues of 12 carcasses from Southeast Alaska with no known history of exposure to oil as a baseline for those collected that were exposed to the Exxon Valdez spill. Control animals included 11 from a native subsistence hunter and one drowned in a gill net. Muscle, kidney and liver were analyzed. Similar to Washington otters, concentrations of the aromatic hydrocarbons were low, with several not detected or below the method detection limit. However, aliphatic hydrocarbon concentrations in the liver from southeast Alaska sea otters were considerably less than that of Washington otters. For instance, the three aliphatics in highest concentration in liver from the live captured Washington otters – n-hexadecane, n-nonacosane, and n-octacosane - were below the method detection limit in many cases for the southeast Alaska otters. Concentrations of the aliphatics above the method detection limit were an order of magnitude lower in the Southeast Alaska sea otter livers versus those from Washington (Table 27). This is surprising because the low molecular weight compounds would be expected to be lost from the environment quickly, yet several are measurable in the Washington sea otters, such as n-decane, n-undecane, n-dodecane, n-tridecane and n-tetradecane. Washington sea otters live in a habitat rich in macroalgae, which is a biogenic source of alkanes, particularly C15, C17 and C19 (Clark and Blumer 1967, Lytle et al. 1979, Youngblood et al. 1971). These biogenic sources may explain some of the higher alkane levels in the liver of Washington versus Alaska otters. Additional research into biogenic sources and the potential contribution from natural hydrocarbons seeps or diapiric muds on the Washington coast may explain the results obtained. Fingerprinting of natural oils on the Olympic Peninsula has been conducted (Kvenolden et al. 1991) and 18  $\alpha$  (H)- and 18  $\beta$  (H) oleanane (C30) could be used as biomarkers when chemical typing. In addition, three oil spills have occurred on the Washington coast within the sea otter range since the reintroduction of sea otters to the area - Meigs, Nestucca and Tenyo Maru - that may contribute to the residue levels observed. Evidence of exposure to petroleum hydrocarbons may be indicated by the amount of unresolved complex for the aromatic hydrocarbons -acomponent that is considered a degraded fossil fuel petroleum residue (Lytle et al. 1979). The unresolved complex was one of only two of the parameters detected in over 50 percent of the animals (the other was total petroleum hydrocarbons). The mean concentration of the unresolved complex was 655 ppm wet weight with a standard deviation of 553 ppm driven by the single animal,WA095, with an unresolved complex concentration of 2090 ppm wet weight, more than four times higher than the next highest concentration (459 ppm wet weight).

			n-decane (C10)	n-undecane (C11)	n-dodecane (C12)	n-tridecane (C13)	n-tetradecane (C14)	n-pentadecane (C15)	n-hexadecane (C16)	n-heptadecane (C17)	n-octadecane (C18)	n-nonadecane (C19)	n-eicosane (c20)	n-heneicosane (C21)	n-docosane (C22)	n-tricosane (C23)	n-tetracosane (C24)	n-pentacosane (C25)	n-hexacosane (C26)	n-heptacosane (C27)	n-octacosane (C28)	n-nonacosane (C29)	n-triacontane (C30)	n-hentricontane (C31)	n-dotriacontane (C32)	n-tetratriacontane (C34)	n-tritriacontane	Phytane	Pristane
	Mean <sup>a</sup>	n=16	7.39	9.11	4.16	3.7	10.3	6.88	128	9.38	8.91	8.8	8.24	4.82	3.04	2.91	4.12	5.59	5.72	7.28	34.31	43.5	4.82	2.45	2.11	1.68	1.79	4.66	5.96
	Std dev		2.33	5.36	2.44	1.85	6.13	4.84	134	7.57	7.29	6.38	5.75	2.24	1.68	1.78	2.7	3.74	4.64	5.38	31.32	24.7	3.7	1.05	1	0.52	0.84	2.54	3.3
	Min <sup>b</sup>		6.05	2.69	1.6	1.36	3.13	0.723	0.796	0.933	0.742	2.45	2.24	2.5	0.947	0.77	0.78	1.53	0.747	1.68	2.08	8.28	1.37	1.06	0.982	1.68	0.77	1.65	2.19
This study	Max		8.72	18	9.54	6.02	19.6	15.5	436	27.3	26.7	21.5	20.7	7.32	5.6	5.63	8.64	13.2	17.5	18.7	99.3	83.6	12.7	4.03	4.01	1.79	3.93	8.04	10.3
	Mean <sup>a</sup>	n=12	<dl< td=""><td>0.059</td><td><dl< td=""><td>0.068</td><td><dl< td=""><td>0.132</td><td><dl< td=""><td>0.039</td><td><dl< td=""><td>0.025</td><td><dl< td=""><td>0.032</td><td>0.073</td><td>0.04</td><td>0.045</td><td>0.124</td><td>0.043</td><td>0.035</td><td>0.032</td><td>0.036</td><td>0.036</td><td>0.046</td><td>0.051</td><td><dl< td=""><td>0.045</td><td><dl< td=""><td>0.075</td></dl<></td></dl<></td></dl<></td></dl<></td></dl<></td></dl<></td></dl<></td></dl<>	0.059	<dl< td=""><td>0.068</td><td><dl< td=""><td>0.132</td><td><dl< td=""><td>0.039</td><td><dl< td=""><td>0.025</td><td><dl< td=""><td>0.032</td><td>0.073</td><td>0.04</td><td>0.045</td><td>0.124</td><td>0.043</td><td>0.035</td><td>0.032</td><td>0.036</td><td>0.036</td><td>0.046</td><td>0.051</td><td><dl< td=""><td>0.045</td><td><dl< td=""><td>0.075</td></dl<></td></dl<></td></dl<></td></dl<></td></dl<></td></dl<></td></dl<>	0.068	<dl< td=""><td>0.132</td><td><dl< td=""><td>0.039</td><td><dl< td=""><td>0.025</td><td><dl< td=""><td>0.032</td><td>0.073</td><td>0.04</td><td>0.045</td><td>0.124</td><td>0.043</td><td>0.035</td><td>0.032</td><td>0.036</td><td>0.036</td><td>0.046</td><td>0.051</td><td><dl< td=""><td>0.045</td><td><dl< td=""><td>0.075</td></dl<></td></dl<></td></dl<></td></dl<></td></dl<></td></dl<>	0.132	<dl< td=""><td>0.039</td><td><dl< td=""><td>0.025</td><td><dl< td=""><td>0.032</td><td>0.073</td><td>0.04</td><td>0.045</td><td>0.124</td><td>0.043</td><td>0.035</td><td>0.032</td><td>0.036</td><td>0.036</td><td>0.046</td><td>0.051</td><td><dl< td=""><td>0.045</td><td><dl< td=""><td>0.075</td></dl<></td></dl<></td></dl<></td></dl<></td></dl<>	0.039	<dl< td=""><td>0.025</td><td><dl< td=""><td>0.032</td><td>0.073</td><td>0.04</td><td>0.045</td><td>0.124</td><td>0.043</td><td>0.035</td><td>0.032</td><td>0.036</td><td>0.036</td><td>0.046</td><td>0.051</td><td><dl< td=""><td>0.045</td><td><dl< td=""><td>0.075</td></dl<></td></dl<></td></dl<></td></dl<>	0.025	<dl< td=""><td>0.032</td><td>0.073</td><td>0.04</td><td>0.045</td><td>0.124</td><td>0.043</td><td>0.035</td><td>0.032</td><td>0.036</td><td>0.036</td><td>0.046</td><td>0.051</td><td><dl< td=""><td>0.045</td><td><dl< td=""><td>0.075</td></dl<></td></dl<></td></dl<>	0.032	0.073	0.04	0.045	0.124	0.043	0.035	0.032	0.036	0.036	0.046	0.051	<dl< td=""><td>0.045</td><td><dl< td=""><td>0.075</td></dl<></td></dl<>	0.045	<dl< td=""><td>0.075</td></dl<>	0.075
Ballachey	Std dev			0.007		0.015		0.001		0.004		0.012			0.017	0.015	0.016	0.188	0.01	0.013	0.009	0.007	0.007						0.049
and Kloecker	Min <sup>b</sup>			0.055		0.047		0.131		0.036		0.014		0.032	0.058	0.019	0.03	0.033	0.029	0.022	0.022	0.03	0.03	0.046	0.051		0.045		0.01
	Max			0.064		0.1		0.133		0.044		0.045		0.032	0.112	0.07	0.08	0.506	0.057	0.056	0.044	0.044	0.044	0.046	0.051		0.045		0.138

Table 27. Comr	arison of Aliphatics in S	ea Otter Liver Sa	nples Collected in Southeast	t Alaska and Washington (ppm, wet weight)

a. mean calculated without non-detects; b. minimum detected value; Aliphatics analyzed in this study that were below detection limit: n-hentriacontane & n-tritriacontane; <DL

= below method detection limit

#### **Organochlorines** (OCs)

Organochlorines have been implicated in the decline of several other marine mammal species (Tanabe et al. 1994) and river otter (Mason 1989). Kannan et al. (2007) found that elevated PCB concentrations in sea otter liver were strongly associated with infections diseases. In this study, only one otter had a detectable blood level of organochlorines, other than PCBs (WA098) which was p,p'-DDE at 0.18 ppm. In general, Washington beach-cast sea otters had similar liver concentrations of those organochlorine compounds measured by Bacon et al. (1999) in California and Aleutian sea otters and Kannan et al. (2008) in Alaskan sea otters, and less than those measured by Nakata et al. (1998) and Kannan et al. (2004, 2008) in California sea otters (Tables 10, 11 and 28).

A review of the literature yields a wide range of PCB concentrations (0.00081 to 222.3 ppm, wet weight) in liver of sea otters, river otters, and pinnipeds, (Bacon et al. 1999, Elliott et al. 1999, Kajiwara 2001, Kannan et al. 2004, Leonards 1997, Nakata et al. 1998 and Troisi 1997), some of which may be explained by summation of different congeners, lipid normalization, species and location of animals (Tables 28 and 29). A comparison of the liver PCB concentrations in the Washington beach-cast sea otters with literature values reveals that Washington concentrations are on the lower to mid-range of means reported for sea otters, and are lower than most river otter and pinnipeds reported. Mean total PCB concentration in liver of Washington sea otters is similar to those reported in Prince William Sound and Adak Island sea otters in Alaska (Kannan et al. 2008). Mean total PCB concentration of 0.40 ppm is higher than that found in liver of beach-cast Aleutian sea otters (0.31 ppm), California sea otters (0.19 ppm), and Southeast Alaska otters (0.008 ppm), (Bacon et al. 1999); however, it is one fourth the mean concentration reported by Nakata et al. (1998) for California sea otters (1.6 ppm), and less than that reported by Kannan et al. (2004), (1.93 ppm), Kannan et al. (2006), (0.624 ppm) and Kannan et al, (2008), (1.1 ppm) for California otters. PCB concentrations in livers of beach-cast Washington otters were similar to those of the other California otters reported by Kannan et al. (2004) when the high outliers from Monterey Bay are excluded.

Although not exhaustive, a review of the literature for PCB concentrations in the blood of marine mammals yielded results for seals (DeSwart et al.1994; Young 1998; Beckman et al. 1999), all of which were higher than mean concentrations of the Washington sea otters by at least an order of magnitude. However, even at the low levels of PCB measured in the whole blood samples from live captured animals, we still observed a significant reduction of vitamin A storage in the liver, a biomarker of exposure (see below); therefore, continued monitoring of PCB in the Washington population is suggested.

Location	Ν	Statistic	PCBs	DDTs	HCHs	CHLs	НСВ	Source
								Present
Washington	15	Mean	0.409	0.126	0.0364	0.0271	ND	Study
								Kannan et al
California	6	Mean	1.100	2.300	0.013	0.110	ND	2008
								Kannan et al
Washington*	6	Mean	0.210	0.160	0.016	0.022	ND	2008
Prince								
William								Kannan et al
Sound, AK	3	Mean	0.420	0.210	0.0053	0.077	ND	2008
								Kannan et al
Adak Island	2	Mean	0.390	0.065	0.0072	0.0092	ND	2008
		Mean	.624					Kannan et al
California	80	Range	.003-5.96	ND	ND	ND	ND	2007
		Mean	1.93	2.02	0.0328	0.125		Kannan et al
California	11	Range	0.28-8.7	0.29-5.9	0.009-0.0076	0.01-0.05	ND	2004
		Mean	1.6	1.8	0.032	0.093	0.0023	Nakata et al.
California	20	Range	0.06-8.7	0.28-5.9	0.005-0.13	0.0041-0.5	0.0007-0.008	1998
								Bacon et al.
Aleutian	7	Mean	0.31	0.036	ND	0.015	0.002	1999
								Bacon et al.
California	9	Mean	0.19	0.85	ND	0.031	0.002	1999
Southeast								Bacon et al.
Alaska	7	Mean	0.008	0.001	ND	0.001	0.001	1999

 Table 28. Literature Review of Organochlorines in Liver of Beach-Cast Sea Otters (ppm, wet weight)

ND=Not Determined

\* Washington beach cast sea otters used in the Kannan et al. 2008 study likely include some of the animals used in the present study.

# Table 29. Literature Summary of PCBs in Liver of Confamilial Species and Marine Mammals (ppm, wet weight)

Species	Location	Statistic	РСВ	Source
Pinnipeds <sup>a</sup>	California	Mean	0.11	Kajiwara et al. 2001
L		Range	0.005-0.41	5
Harbor seal	England	Mean	0.00293	Troisi et al. 1997
		Range	0.00001-0.01674	
Eurasian		Mean		
otter	Netherlands		70.994	Leonards et al. 1997
		Range	4.441-222.3	
River otter	Washington	Mean	.170	
		Range	.0161-10.087	Grove and Henry 2007

a. elephant seal and harbor seal

#### DDT (1,1,1,-trichloro-2,2-bis(p-chlorophyenyl)ethanes

Mean DDT concentrations in liver of Washington beach-cast animals did not exceed 1 ppm, which is considerably lower than that observed in California sea otters (Bacon et al. 1999; Kannan et al. 2004, 2008; and Nakata et al. 1998) but higher than that observed in Alaskan sea otters (Bacon et al. 1999) (Tables 11 and 28).

#### Cyclohexanes (HCHs)

Mean cyclohexane concentration in the liver of Washington beach-cast sea otters was similar to those observed by Kannan et al. (2004) and Nakata et al. (1998) in California beach-cast otters and two and three times greater than those observed by Kannan et al. (2008) in California and Alaska (Table 28).

#### Chlordanes (CHLs)

Chlordanes in liver of Washington beach-cast otters were considerably lower than those reported by Kannan et al. (2004, 2008) and Nakata et al. (1998) but similar to those reported by Kannan et al. (2008) for Alaska others and those reported by Bacon et al. (1999) for both California and Aleutian sea otters (Table 28).

Thus, from our analyses it seems the levels of OCs found in the tissues of Washington sea otters is not markedly different from other investigation findings for various marine mammal species with presumably similar exposure pathways for these chemicals.

#### **Biomarkers**

#### Vitamin A

Vitamin A is a collective name for a group of fat-soluble molecules (retinoids) essential to growth, development, reproduction and immunocompetence. Because vitamin A has been shown to be sensitive to disruption by organochlorine contaminants (PCBs and dioxin-like compounds especially), levels of this dietary hormone have been widely used as biomarkers of toxicity in both adult (Tabuchi et al. 2006; Mos et al. 2007; De Swart et al. 1994; Brouwer et al. 1989) and young harbor seals (*Phoca vitulina*) (Simms et al. 2000, Jenssen et al. 1994), sea lions (Debier et al. 2005), river otters (Murk et al. 1996) and a variety of bird species (Harris et al. 2007; Murk et al. 1994; Spear et al. 1986). Decreased vitamin A levels can indicate that the health of a species is adversely affected by contaminant exposure that may be playing a part in impaired reproduction and an increased incidence of disease (Spear et al. 1989).

Kannan et al. (2000) established organochlorine tissue residue threshold values for seals, otters and minks from published field and laboratory studies. Toxicological endpoints elicited by the aquatic mammals in response to this dietary dose of chemicals included altered vitamin A levels, reduced thyroid hormone concentrations, and immunosuppression (Kannan et al. 2000). These physiological effects were observed at liver concentrations ranging from 6.6 to 11 ug PCB/g depending on the species of marine mammal. In this study we did not measure PCBs or other organochlorines in liver of the live captured sea otters. PCB concentrations in the liver of the beach-cast animals were above the range determined by Kannan et al. (2000) to elicit a physiological response. In our study, concentrations of contaminants in the liver of beach-cast sea otters were relatively low and similar to those in other sea otter populations in the United States. Chemical concentrations in the whole blood and liver of live captured otters were also low. However, even the low level of PCB exposures we observed have been associated with the sensitivity of vitamin A storage enzymes in laboratory animals (Zile 1992), and we observed a significant reduction of vitamin A storage in the liver. Significant negative correlations were found between PCBs and all-trans retinyl palmitate, total retinol, and the R:RP ratio, though not the correlation with the R:RP ratio was not particularly strong. The disruption of these compounds is associated with a displacement of hormone by PCB metabolites at the transport protein complex (TTR-RBP). In summary, since measurable levels of PCBs were found in beach-cast livers and at levels above those known to cause physiological stress, it is not surprising to see the decreased levels of retinols and thyroid hormones in the blood of live Washington sea otters sampled.

Vitamin A in the liver was also reduced in relation to dibutyltin and octasosane concentrations, but significantly and positively correlated to several of the other aliphatic hydrocarbons, and positively and particularly strongly correlated to cadmium concentrations (Table 18). Harris et al. (2007) also found a strong positive correlation between hepatic vitamin A concentration and kidney cadmium concentration but not so to liver or plasma concentrations of cadmium. In all cases our sample size is relatively small (6 to 15), making interpretation of the results problematic.

Vitamin A increases in the liver with age, decreases during disease, and can decrease over the course of lactation. Thyroid hormones change over the course of development and may also be affected by the animal's condition because they function in thermoregulation. No significant or strong relationship ( $\mathbb{R}^2$  values were less than 0.40) was observed between age, gender or body weight of the sea otters and vitamin A or thyroid hormone levels, which may be due to our small sample size.

#### Cytochrome P450 CYP1A Assays

The oxidative metabolizing enzyme cytochrome P450 (CYP1A) plays a role in the detoxification of some contaminants, such as polycyclic aromatic hydrocarbons and chlorinated hydrocarbons. Cytochrome P450 (CYP1A) assays were conducted on the peripheral blood mononuclear cells (PBMC) isolated from heparinized blood and on liver biopsies in order to determine if blood and liver P450 levels correlated and if the P450 enzyme induction found in liver tissue correlated with any chemical concentrations. Molecular biomarker procedures may allow us to evaluate potential chemical-specific stressors to the sea otters, and help predict overall sea otter health.

Cytochrome P450 CYP1A1 is an isozyme in the cytochrome P450 superfamily. Its role in metabolizing chemicals has been well studied, though not in sea otters. Cytochrome P450 molecular biomarker assays of PBMC (peripheral blood mononuclear cell) and liver have been used to evaluate potential chemical-specific stressors to Alaska and California sea otters, thus we undertook these assays for the Washington otters for comparison. The development of the technique using blood (PBMC) allows for a less invasive approach to determine if the animals are potentially stressed due to specific toxins. For instance, the mean level expression in PBMC from sea otters captured in non-oiled areas in Prince William Sound, Alaska is reported as 0.12 x

 $10^{6}$  as compared to 1.96 x  $10^{6}$  in oiled areas in 1994-1996 (Snyder et al. 2002). The mean level expressed in PBMC from Washington otters was considerably higher, at 7.57 x  $10^6$  molecules p450/100ng total RNA in blood samples. Unpublished data provided by USGS (B. Ballachey 2006) using the same methods employed on the Washington project, revealed that California  $(43.97 \times 10^6 \text{ molecules p} 450/100 \text{ ng total RNA})$  and Washington  $(7.57 \times 10^6 \text{ molecules})$ p450/100ng total RNA) mean PBMC cytochrome P450 expression was closer to animals experiencing the *Exxon Valdez* spill (22.11 x  $10^6$  molecules p450/100ng total RNA) than to southeast Alaska animals  $(1.02 \times 10^6 \text{ molecules p450/100ng total RNA})$  or those from a control site for the *Exxon Valdez* study (1.66 x  $10^6$  molecules p450/100ng total RNA). The mean liver value in Washington otters was  $1.85 \times 10^6$  molecules p450/100ng total RNA. The mean Washington sea otter liver value was more in line with the only other liver values available for comparison – those from otters exposed to the *Exxon Valdez* spill (5.90 x  $10^6$  molecules p450/100ng total RNA) and the control site  $(1.03 \times 10^6 \text{ molecules p450}/100 \text{ ng total RNA})$ (unpublished data provided by USGS, B. Ballachey 2006). As indicated above, the mean may not be an adequate statistic since variability in the P450 results from Washington sea otters is considerable, with the PBMC mean driven by two otters. This variability is also apparent in the data provided by USGS. When medians are compared, the Washington otters PBMC expression  $(0.51 \times 10^6 \text{ molecules p450/100ng total RNA})$  is comparable to the Alaska sea otters from unoiled areas in Prince William Sound (1.05 x  $10^6$  molecules p450/100ng total RNA) and Southeast Alaska ( $1.02 \times 10^6$  molecules p450/100ng total RNA); however, the median for the California otters is still comparatively high (4.09 x  $10^6$  molecules p450/100ng total RNA) and closer to that of oiled animals from Alaska ( $4.89 \times 10^6$  molecules p450/100ng total RNA).

Chemical concentrations in Washington sea otters were low, perhaps explaining why there were few significant relationship between the cytochrome P450 results and the chemical concentrations. Only total PCBs and P450 in the liver were found to be significantly correlated. Despite these low concentrations, liver from one of the two otters with increased PBMC P450 induction also had the highest aliphatic and aromatic hydrocarbon concentrations (WA099). Liver from the other otter with increased PBMC P450 induction was not analyzed for hydrocarbons; however it did exhibit elevated serum enzyme alkaline phosphatase, one of the serum enzymes that has been shown to increase in response to hydrocarbon exposure (Ballachey et al. 2003).

#### Pathogens

The following text is quoted almost verbatim from the results obtained from the USGS National Wildlife Health Center.

#### Serology

The high prevalence (80 percent) of antibody titers to morbilliviruses in the live captured animals suggests that the Washington sea otter population is fairly well-protected against a widespread morbillivirus outbreak. Individual deaths may occur but a population-threatening die-off from this disease is unlikely while population immunity remains high. This is the first finding of positive morbillivirus titers in sea otters. No positive titers were found in a survey of Alaska and

California sea otters (Hanni et al. 2003), although British Columbia has detected *Morbillivirus* in river otters using the marine environment (Nichol et al. 2003).

Only one animal had a low positive antibody titer to one serovar of *Leptospira* sp. From leptospirosis deaths in Washington sea otters during the second year of this study (including the animal identified as "18316-01" sampled in this study) we know that sea otters are susceptible to this disease. These results suggest that exposure to this agent was not widespread at the time of the study. Hanni et al. (2003) reported one instance of this antibody titer in California otters as well, and none in the Alaska population.

*Brucella* sp. has been found in a harbor seal, and antibodies have been detected in both harbor seals and sea lions in Puget Sound, Washington, and in sea otters from California and Alaska. Negative *Brucella* sp. results in sea otters suggests that they are not exposed to this potential zoonotic disease agent on the outer coast of Washington or that our sample population is small. Hanni et al. (2003) detected *Brucella* sp. antibodies in both Alaska and California animals.

Toxoplasmosis has been diagnosed in sea otters dying in Washington and California (Dubey et al. 2003) and has been cited as major cause of mortalities and the slowed recovery rate for California sea otters (Kreuder et al. 2003). The high prevalence of antibody titers (60 percent) found in this study is similar to previous studies of California otters (Dubey et al. 2003; Hanni et al. 2003). The explanation for the seroprevalence remains unexplained and the cause postulated for California, that exposure is likely from oocysts in feces of felids entering the marine system through runoff, is not as likely for Washington sea otters since their exposure to such sources would not be great. The Washington coast is relatively pristine with only small villages located along the shore adjacent to the sea otter range, and the domestic cat population is small. Wild cats may be a source, but it is not known how significant this source would be. As a post pilot study to this capture operation we collected sea otter prey to evaluate oocyst content as providing a potential dietary exposure route for sea otters. The study was small and we (Dubey et al. unpublished data) did not detect any oocysts, however, Miller et al. (2008) reported Type X *T. gondii* in one of the marine bivalves analyzed.

The presence of antibody against *Neospora caninum* in sea otters is a new finding from this study (first reported in Dubey et al. 2003). The significance of this finding is unclear; no other evidence of infection and no disease from this agent has been reported in sea otters.

*Sarcocystis neurona* has caused fatal encephalitis in Washington sea otters in the past. Other *Sarcocystis* sp. have been shown to be present in Washington sea otters, but not associated with significant disease. The *Sarcocystis* serologic test used in this study has not been tested to determine its specificity for *S. neurona* in sea otters. Therefore until the test is proven specific, positive antibody in sea otters should be considered evidence of exposure to *S. neurona* or other similar *Sarcocystis* sp.

No calicivirus-associated disease has been described in sea otters. A past serologic survey for this agent found few low positive titers in California, but none in Alaskan sea otters (Hanni et al. 2003).

Based on these findings for Washington live and beach-cast otters and those of other sea otter populations, continued serological testing is recommended to monitor the health of the Washington sea otter population.

#### Biotoxin

In addition to anthropogenic contaminants, concern has been raised by scientists studying biotoxin levels along the outer coast. There may be a connection between anthropogenic activities (nutrient loading) and the increased incidence of harmful algal blooms (NOAA 1999). Biotoxin monitoring in shellfish is ongoing by the Quinault Nation, Quileute and Makah tribes and the Washington Department of Health (WDOH), and during this study, it was part of the Olympic Region Harmful Algal Bloom partnership. This monitoring information provides a useful database for potential exposure of sea otters to biotoxins, such as domoic acid produced by a diatom, and Paralytic Shellfish Poisoning (PSP) toxins produced by dinoflagellates, both of which can accumulate in invertebrate prey of sea otters. In 1998 in California, there was a California sea lion (Zalophus californianus) mortality event that, through weight-of-evidence studies, indicated that domoic acid exposure, from ingestion of its prey, was the cause of seizures, loss of neurological function, and ultimately death of these animals (Lefebvre et al. 1999; Scholin et al. 2000). Domoic acid has also recently been linked to reproductive failure in pinnipeds such as the California sea lion (Brodie et al. 2006). Domoic acid has been associated with seabird die offs in California (Work et al. 2003), and one domoic acid poisoning has been presumed for a California sea otter (Kreuder et al. 2003). A die off of sea otters in the Aleutians in 1987 was also partly attributed to PSP toxins (DeGange and Vacca 1989); however, Kvitek et al. (1991) indicates that sea otters may actually be able to avoid prey containing elevated PSP.

In this study, urine was opportunistically collected from three animals for domoic acid determination and all were negative. This is certainly not conclusive and if warranted by observations of increased harmful algal blooms, increased occurrence of domoic acid in shellfish or increased sea otter mortalities, particularly any reported during harmful algal blooms, additional study may be undertaken. This would be of increased importance if the Washington sea otter population continued their range expansion southwards into areas known to have more regular elevated levels of domoic acid.

#### **Blood Health Screens**

Washington sea otter cell blood counts (CBC) and serum chemistry were generally similar to those reported for California sea otters (Hanni et al. 2003). Alaska otter CBC and serum chemistries often statistically differed from Washington and California sea otters (Table 30). Table 30 presents a comparison of mean values for Alaska, California and Washington otters for those parameters that overlapped between studies and the statistical comparison of the means is also presented. Gender differences could be evaluated for Prince William Sound, Alaska and Washington otters. Gender differences in some of the parameters were apparent in both the Alaska and Washington populations – for white blood cell counts and glucose, counts or concentrations were higher in female otters, and gamma-glutamyltransferase 1 was higher in male otters.

Other parameters that differed notably between the sea otter populations included phosphorus, which was highest in the Aleutian Island animals; lactic dehydrogenase, which was statistically lower in Washington otters; and aspartate aminotransferase, which was statistically higher in Prince William Sound animals (Table 30). Diet could potentially explain the difference in

Table 30. Comparison of Hematology and Serum Chemistry Variables (Mean ± Standard Deviation) for Adult Sea Otter Populations in the United States: Prince William Sound (1992<sup>1</sup>), Aleutian Island (1995-2000<sup>2</sup>), California (1995-2000<sup>2</sup>), and Washington (2001-2002) Sea Otters

Variable	Location	Mean Sig	Std dev	N
White Blood Cells	Alaska, PWS	8.84 <sup>a</sup>	2.02	49
	California	7.2 <sup>b</sup>	1.9	48
	Washington	6.8 <sup>b</sup>	1.8	30
Red Blood Cells	Alaska, PWS	5.07 <sup>a</sup>	0.30	49
	California	5.1 <sup>a</sup>	0.5	48
	Washington	5.12 <sup>a</sup>	0.378	30
Hemoglobin	Alaska, PWS	20.60 <sup>a</sup>	1.10	49
6	California	18.7 <sup>b</sup>	2.2	48
	Washington	18.7 <sup>b</sup>	1.37	30
Hematocrit	Alaska, PWS	59.28 <sup>a</sup>	3.47	49
	California	55.5 <sup>b</sup>	5.4	48
	Washington	54.3 <sup>b</sup>	4.24	30
Mean Corpuscular Volume	Alaska, PWS	117.34 <sup>a</sup>	4.43	49
Ĩ	California	108.8 <sup>b</sup>	5.3	48
	Washington	106 <sup>c</sup>	4.98	30
Mean Corpuscular	Alaska, PWS	40.73 <sup>a</sup>	1.60	49
Hemoglobin	California	37.4 <sup>b</sup>	2.4	48
C C	Washington	36.7 <sup>b</sup>	1.73	30
Mean Corpuscular	Alaska, PWS	34.70 <sup>a</sup>	0.93	49
Hemoglobin Concentration	California	35.4 <sup>b</sup>	1.1	48
-	Washington	34.6 <sup>a</sup>	1.09	30
Red Cell Distribution	Alaska, PWS	12.83 <sup>a</sup>	0.57	49
Width	Washington	14.1 <sup>a</sup>	1.78	15
Neutrophils	California	52 <sup>a</sup>	10	48
*	Washington	51 <sup>a</sup>	13	25
Lymphocytes	California	30 <sup>a</sup>	10	48
	Washington	36 <sup>a</sup>	13	25
Monocytes	California	6 <sup>a</sup>	4	48
2	Washington	4 <sup>a</sup>	2	25
Eosinophils	Alaska, PWS	0.41 <sup>a</sup>	0.39	49
I	California	11 <sup>b</sup>	7	48
	Washington	8 <sup>b</sup>	7	25
Platelets	California	242 <sup>a</sup>	95	48
	Washington	262 <sup>a</sup>	67.5	30
Glucose	Alaska, PWS	146.99 <sup>a</sup>	32.19	53
	Alaska, AI	148 <sup>a</sup>	46	54
	California	112 <sup>b</sup>	26	49
	Washington	119 <sup>b</sup>	26	30
Blood Urea Nitrogen	Alaska, PWS	50.18 <sup>a</sup>	10.36	53
C C	Alaska, AI	51 <sup>a</sup>	11	54
	California	57 <sup>b</sup>	12	49
	Washington	55 <sup>a,b</sup>	13	30

	Alaska, PWS	0.01	0.1.1	
Creatinine	Alaska, AI	0.81 <sup>a</sup>	0.14	53
	California	0.4 <sup>b</sup>	0.2	53
	Washington	$0.6$ $^{\circ}$ 0.6 $^{\circ}$	0.07 0.1	49
	-		0.1	30
Uric Acid	Alaska, PWS	2.54 <sup>a</sup>	0.70	53
	Washington	1.95 <sup>a</sup>	0.55	15
Sodium	Alaska, PWS	154.25 <sup>a</sup>	1.69	53
	Alaska, AI	157 <sup>a,b</sup>	10	54
	California	148 <sup>b</sup>	4	49
	Washington	154 <sup>a</sup>	2.7	30
Potassium	Alaska, PWS	4.17 <sup>a</sup>	0.33	53
	Alaska, AI	4.3 <sup>a,b</sup>	0.5	54
	California	4.4 <sup>b</sup>	0.4	49
	Washington	4.5 <sup>b</sup>	0.32	30
Chloride	Alaska, PWS	118.61 <sup>a</sup>	2.48	53
Chionde	Alaska, AI	115 <sup>b</sup>	2.48	53 54
	California	113 112 <sup>b</sup>	9	54 49
	Washington			
	Alaska, PWS	113 <sup>b</sup>	2.66	30
Calcium	Alaska, AI	8.73 <sup>a</sup>	0.46	53
	California	9.2 <sup>b</sup>	0.7	54
	Washington	8 <sup>c</sup>	0.6	49
		9.4 <sup>b</sup>	0.51	30
Phosphorus	Alaska, PWS	4.28 <sup>a</sup>	1.38	53
	Alaska, AI	5.9 <sup>b</sup>	1.3	54
	California	3.8 <sup>a</sup>	0.9	49
	Washington	4.5 <sup>a</sup>	1.3	30
Albumin	Alaska, PWS	2.92 <sup>a</sup>	0.18	53
	Alaska, AI	2.6 <sup>b</sup>	0.3	54
	California	2.7 <sup>b</sup>	0.2	49
	Washington	2.9 <sup>a</sup>	0.2	30
Globulin	Alaska, PWS	3.88 <sup>a</sup>	0.48	53
	Alaska, AI	4.3 <sup>b</sup>	0.5	54
	California	4.3 <sup>b</sup>	0.5	49
	Washington	3.8 <sup>a</sup>	0.7	30
Total Protein	Alaska, AI	6.9 <sup>a</sup>	0.7	54
	California	6.9 <sup>a</sup>	0.8	49
	Washington	6.7 <sup>a</sup>	0.58	30
Cholesterol	Alaska, AI	174 <sup>a</sup>	57	54
	California	174 164 <sup>a</sup>	54	49
	Washington	164 <sup>a</sup>	40.6	30
Total Dilimbin	Alaska, PWS	0.40 <sup>a</sup>	0.14	
Total Bilirubin	Alaska, AI	0.40 <sup>b</sup>		53 54
	California		0.1	54 40
	Washington	$0.1^{\circ}$	0.04	49
	Alaska, PWS	0.2 <sup>b</sup>	0.1	30
Gamma-		15.70 <sup>a</sup>	3.95	53
glutamyltransferase 1	Washington	17 <sup>a</sup>	9.5	30

Alkaline	Alaska, PWS	83.24 <sup>a</sup>	25.88	53
phosphatase	Alaska, AI	112 <sup>b,c</sup>	35	54
	California	98 <sup>a,c</sup>	27	49
	Washington	128 <sup>b</sup>	63	30
Lactic dehydrogenase	Alaska, AI	573.8 <sup>a</sup>	73.7	54
	California	532.3 <sup>a</sup>	62.8	49
	Washington	337 <sup>b</sup>	90	15
Aspartate	Alaska, PWS	282.42 <sup>a</sup>	56.91	53
aminotransferase	Alaska, AI	173 <sup>b</sup>	75	54
	California	208 <sup>b</sup>	70	49
	Washington	201 <sup>b</sup>	86	30
Alanine	Alaska, PWS	235.98 <sup>a</sup>	68.40	53
aminotransferase	Alaska, AI	205 <sup>a</sup>	71	54
	California	230 <sup>a</sup>	65	49
	Washington	195 <sup>a</sup>	87	30

1). <sup>1</sup>Results for Alaska sea otters from Ballachey et al. 2003 are reported only from unoiled otters – Western Prince William Sound (PWS).

2.) <sup>2</sup>California and Aleutian Island (AI)/Elfin Cove results from Hanni et al. 2003, genders combined.

Sig = statistically significant; Same letter superscript indicates there is not a significant difference in the results for those locations. Different letter superscript indicates a significant difference (p<0.05) between locations.

White blood cells (103 cells/mm3); Red blood cells (106 cells/mm3); Hemoglobin (g/dL); Hematocrit (%); Mean corpuscular volume (fL); Mean corpuscular hemoglobin (pg); Mean corpuscular hemoglobin concentration (g/dL); Red cell distribution width (%); glucose (mg/dL); Blood urea nitrogen (mg/dL); creatinine (mg/dL); uric acid (mg/dL); sodium (mequiv/L); potassium (mequiv/L); chloride (mequiv/dL); calcium (mg/dL); phosphorus (mg/dL); albumin (g/dL); globulin (g/dL); total protein (g/dL); cholesterol (mg/dL); total bilirubin (mg/dL); gamma glutamyltransferase (1 U/L = 16.67 nkat/L); alkaline phosphatase (1 U/L = 16.67 nkat/L); lactic dehydrogenase (1 U/L = 16.67 nkat/L); aspartate aminotransferase (1 U/L = 16.67 nkat/L);

glucose, phosphorus, sodium and creatinine levels between Alaska populations and those in Washington and California. The Aleutian Island animals feed on fish more than do the other populations. The relevance of these differences in CBC and serum chemistry, if any, is unknown, but may be explained by a difference in laboratory methods, physiological differences, contaminant exposure, parasite loads, or as previously mentioned, sea otter diet.

#### **Fatty Acid Profiles**

The authors found meaningful variation in concentrations of both linoleic and linolenic acids in individual otters, suggesting substantial differences in foraging habits of the individual otters within the same age class, weight, location or gender (VanBlaricom et al. 2007). This result is consistent with foraging variation documented in California sea otters (Estes et al. 2003). The authors recommend that Quantitative Fatty Acid Signature Analysis be performed to estimate the diet consumed. This requires knowledge of fatty acid profiles in prey items that are collected from the same location the sea otter are captured and then modeling the combination of fatty acids from the prey that would most likely lead to the distribution of fatty acids found in the sea otters.

#### **Analysis of Prey**

Chemical concentrations in sampled prey were low, as reflected also in sea otter tissue concentrations.

#### **Management Actions/Recommendations**

Based on the results of this investigation, the following direct management actions are recommended:

- 1) Data from this study should be used as a baseline from which to compare the future health of the Washington sea otter population over time. The data can also be used in an injury assessment, such as Natural Resource Damage Assessments (NRDA) that might result from or as a part of contaminant releases. As such, this information is useful to resource agencies in addition to the USFWS, such as Olympic Coast National Marine Sanctuary, Washington Department of Fish and Wildlife, Olympic National Park and the primary spill response agencies (NOAA, the US Coast Guard and the Washington Department of Ecology) and should be provided to them.
- 2) Results should continue to be used by resource managers as done so within, to evaluate the health of Northeast Pacific sea otter populations, comparing a relatively stable population of otters (Washington) with populations experiencing declines (Alaska and California) to better understand the reason for the declines.
- 3) The information should be provided to the team responsible for the Washington State Recovery Plan for the Sea Otter, for incorporation into future revisions to the recovery plan.
- 4) The information should be provided to agencies to help meet their mandate for resource protection, in particular sea otter population protection (i.e., management actions under the Marine Mammal Protection Act such as stock assessments as well

as responding to stranded sea otters and carcass recovery from Washington beaches). Agencies with this mandate include the USFWS, Washington Department of Fish and Wildlife and the Olympic Coast National Marine Sanctuary.

- 5) An overview or summary of the report should be produced for public outreach/awareness purposes and web posting.
- 6) The results of this study should contribute to the design of futures studies of the Washington sea otter population. For instance, serology and blood health screens should continue for specific parameters that have been shown to respond to exposure to hydrocarbons (e.g., increased alanine aminotransferase and alkaline phosphatase) or parasite loads (e.g., eosinophils).

From this study a baseline has been developed for future assessment of the Washington sea otter population. Based on the findings of low PCB concentrations yet a significant relationship between these and reduced Vitamin A liver production, and the potential relationship between aliphatic and aromatic hydrocarbon concentrations and P450 induction measured in two of the otters, we believe periodic monitoring using a more limited set of parameters is warranted. Analysis of PCBs, aliphatic and aromatic hydrocarbons, and potentially a subset of metals is also warranted. Biomarkers, such as Vitamin A analysis and cytochrome P450, should also be measured to generate additional data from which broader conclusions might be drawn. Additional serology is also needed since pathogen exposure is quite evident in the Washington population with both morbillivirus (80%) and *Toxoplasma* (60%), which has been determined to be a significant cause for California sea otter mortalities, evident in the Washington sea otter population tested, and infectious disease was determined the primary cause of death for the majority of the subsample of the beach-cast animals reported herein.

Analyses of blood and liver samples from live captured sea otters and liver samples from beach-cast sea otter carcasses off the remote Washington coast indicate relatively low exposure to contaminants, but suggest that even at the low levels measured, exposure may be indicated by biomarker response. The results indicate low levels of metals, butyltins, and organochlorine compounds in the blood samples, with many of the organochlorines not detected except polychlorinated biphenyls (PCBs), and a few aromatic hydrocarbons detected in the live captured animals. Evidence of pathogen exposure presents a potential risk to Washington sea otters, particularly due to their small population size and limited distribution. Washington waters (Jameson and Jeffries 2007); however, the population remains well under estimated carrying capacity, has a limited distribution and has not yet reached its carrying capacity and as such, is still considered at high risk to catastrophic events.

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#### APPENDICES

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# Appendix 1. Tracking and Ground Studies Conducted in Support of the Environmental Contaminant and General Health Investigation

Appendix 1a. Report on Tracking Radio-tagged Sea Otters Along the Washington Coast from 2001 to 2003

Report on Tracking Radio-tagged Sea Otters along the Washington Coast from 2001 to 2003

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For: Cooperative Agreement No. 134101J014

15 June 2004

#### Appendix 1a Report on Tracking Radio-tagged Sea Otters along the Washington Coast from 2001 to 2003

#### Introduction

This report summarizes radio-tag tracking activities conducted under Cooperative Agreement No. 134101J014 between Washington Department of Fish and Wildlife and U.S. Fish and Wildlife Service. This effort was part of a cooperative interagency research project to monitor the distribution, numbers, movements, health and contaminant/biotoxin levels in Washington's sea otter population.

Sea otters were captured and radio-tagged off the Olympic Peninsula in 2001 (15 otters) and 2002 (13 otters) (Tables 1 and 2). The purpose of the project was to detect the presence of radio-tagged otters both within and outside the current Washington range. Aerial surveys were determined to be the most efficient technique to detect radio-tagged otters and were conducted within the current sea otter range in Washington (Destruction Island to Pillar Point) as well as potential range of sea otters from the Columbia River to Destruction Island; Pillar Point to Elwha River; and along Vancouver Island, British Columbia from Race Rocks to Esteven Point.

#### Methods

WDFW personnel conducted aerial surveys in a Cessna 185 to locate radio-tagged sea otters. Surveys were flown at 800 feet and 100 knots airspeed. Surveys covered all known and potential range of sea otters in Washington and along Vancouver Island. Telemetry equipment included right and left mounted H-style yagi antennas, Advanced Telemetry System (ATS) scanning receiver and switch box for determining specific locations of radio-tagged otters. For each survey, all radio-tagged otters known to be alive were programmed into the scanning receiver. Resight information including date, time, location, latitude/longitude and activity were recorded in a telemetry log (Appendix 1).

During surveys, the receiver scan interval was set at 2-4 seconds per frequency to allow detection of each radio-tag. At known sea otter concentration sites along the Olympic Peninsula (Destruction Island, Diamond Point, Perkins Reef/Rocks 443, Cape Johnson, Cedar Creek, Yellowbanks, Sand Point, Cape Alava and Duk Point), the aircraft circled and scanned 4-6 times through all programmed radiotag frequencies in order to detect resting and active otters. If a radiotag was detected, the switch box and receiver gain were manipulated to determine specific location of the radio-tagged otter.

Once the specific location of the radio-tagged otter was determined, latitude and longitude was recorded using an onboard GPS unit. Activity patterns (active, resting or dead) were determined based on signal type and recorded. For an active otter, the signal was heard while on the surface and lost when diving; for a resting animal, the signal was continuous; and for dead otter a continuous mortality signal was received. Following determination of a radio-tag location, that frequency was deleted from the scan cycle to reduce the time required scanning remaining undetected radio-tags. A mortality signal from a radio-tagged otter was investigated to determine status and location. In some cases a false mortality signal was detected from a radio-tagged live otter by determining the animal was actively diving. If a radio-tagged otter was determined to be dead, its location was recorded and frequency deleted from subsequent survey scans. Capture and resight locations were plotted by latitude

and longitude (in hddd<sup>0</sup> mm.mmm' format) on maps generated up MapSource BlueChart Version 5.0 from Garmin. Distances between capture and resight locations were measured along the coast by following the 40-foot isobath contour line using the MapSource distance/bearing feature.

#### Results

In 2001, from August 3-11, a total of 15 sea otters were captured and implanted with radiotags at Ozette River (2 otters), Cape Alava (7 otters), Sand Point (2 otters), Perkins Reef/ Rocks 443 (2 otters) and Destruction Island (2 otters) (Table 1). In 2002, from August 13-22, a total of 13 sea otters were captured and implanted with radio-tags at Cape Alava (1 otter), Sand Point (1 otter), Cape Johnson (2 otters), Giant's Graveyard (1 otter), Perkins Reef/Rocks 443 (3 otters) and Destruction Island (5 otters) (Table 2).

Between 2 October 2001 and 9 December 2003, telemetry surveys were conducted through potential and current range of sea otters in Washington from the Columbia River to the Elwha River (23 surveys) as well as along Vancouver Island, British Columbia from Race Rocks to Esteven Point (2 surveys) (Table 3). Surveys averaged 5.9 hours (range 3.3-7.7 hours) for a total of 146.5 hours of flight time.

For the 15 otters radio-tagged in 2001, one (Otter 95) was never heard; two (Otters 88 and 94) were heard only once; and one (Otter 97) was heard only twice. Of the remaining 11 otters radio-tagged in 2001, movement distances from capture location to resight location (Table 4) ranged up to 2 miles (Otter 98); up to 5 miles (Otter 86); up to 10 miles (Otter 89, 93, 99, and 100); up to 15 miles (Otter 90 and 96); up to 20 miles (Otter 91); and up to 37 miles (Otter 84 and 92). Extensive movement and interchange throughout their known range was recorded for the majority of otters radio-tagged in 2001 with the greatest movements between Cape Alava and Destruction Island (Otter 84 and 92). Otter 91 moved from Cape Alava to Slant Rock in the Strait of Juan de Fuca. The farthest distance from capture to resight location was for Otter 97 that was recovered dead on Shi-Shi Beach, a distance of 47 miles from its capture location at Destruction Island. Three of the otters implanted in 2001 died (Otter 88, 91 and 97).

For the 13 otters radio-tagged in 2002, one (Otter 54) was never heard; one (Otter 108) was heard only once, and one (Otter 113) only twice. Of the remaining 10 otters radio-tagged in 2002, movement distances from capture location to resight location (Table 5) ranged up to1 mile (Otter 107); up to 4 miles (Otter 103 and 109); up to 13 miles (Otter 102, 106 and 110); and up to 30 miles (Otter 105, 111, 112 and 114). Otters radio-tagged in 2002 showed extensive interchange throughout their range similar to the otters radio-tagged in 2001, with exchange recorded between Destruction Island and Cedar Creek (Otter 105); Cape Johnson and Destruction Island (Otter 112); Destruction Island to Cape Johnson (Otter 114); and Giant's Graveyard to Ozette River (Otter 111). Two of the otters implanted in 2002 died (Otter 54 and 103).

Although there was extensive movements and interchange between areas along the Washington coast, none of the radio-tagged otters were located outside of their currently defined range.

Five of the twenty-five sea otters implanted with radio-tags (20 percent) had died by 9 December 2003.

<u>NO.</u>	DATE	<b>LOCATION</b>	SEX	AGE	Wt (kg)	<u>TX FREQ.</u>	<b>LATITUDE</b>	<b>LONGITUDE</b>
84	8/3/01	CAPE ALAVA	F	А	23.5	164.334	48 10.21	124 44.53
85	8/3/01	CAPE ALAVA	F	Р	3	NONE	48 10.21	124 44.53
86	8/4/01	<b>OZETTE RIVER</b>	F	А	21	164.576	48 10.81	124 43.32
87	8/4/01	<b>OZETTE RIVER</b>	Μ	Р	5	NONE	48 10.81	124 43.32
88	8/6/01	<b>OZETTE RIVER</b>	F	А	22	164.697	48 10.86	124 43.28
89	8/6/01	CAPE ALAVA	F	А	23	164.495	48 10.08	124 44.72
90	8/6/01	CAPE ALAVA	F	А	24	164.054	48 10.08	124 44.72
91	8/6/01	CAPE ALAVA	F	А	22	164.451	48 10.08	124 44.72
92	8/6/01	CAPE ALAVA	Μ	Р	16	164.294	48 10.09	124 44.72
93	8/6/01	CAPE ALAVA	F	А	27	164.516	48 10.09	124 44.72
94	8/7/01	CAPE ALAVA	F	А	23	164.135	48 10.10	124 44.69
95	8/7/01	SAND POINT	F	А	23	164.674	48 07.25	124 43.06
96	8/7/01	SAND POINT	F	А	27	164.396	48 07.25	124 43.06
97	8/9/01	DESTRUCTION I.	Μ	А	41	164.014	47 40.89	124 28.44
98	8/9/01	DESTRUCTION I.	Μ	А	30	164.173	47 40.89	124 28.44
99	8/9/01	PERKINS/443	F	А	24	164.596	47 46.77	124 29.64
100	8/11/01	PERKINS/443	F	А	27	164.216	47 46.77	124 28.44

Table 1. Summary of 2001 Washington sea otter captures.

Table 2. Summary of 2002 Washington sea otter captures.

<u>NO.</u>	DATE	<b>LOCATION</b>	SEX	AGE	Wt (kg)	TX FREQ.	LATITUDE	<b>LONGITUDE</b>
102	8/13/02	DESTRUCTION I.	Μ	А	50.5	164.353	47 40.93	124 28.49
103	8/17/02	SAND POINT	F	А	23	164.373	48 07.30	124 42.91
104	8/17/02	SAND POINT	Μ	Р	11	NONE	48 07.30	124 42.91
105	8/18/02	DESTRUCTION I.	Μ	А	38	164.931	47 40.78	124 28.61
106	8/18/02	PERKINS/RK 443	F	А	31	164.882	47 46.84	124 29.63
54	8/16/02	CAPE ALAVA	F	А	27	164.754	48 10.26	124 44.49
107	8/19/02	DESTRUCTION I.	Μ	А	36.5	164.433	47 40.84	124 28.48
108	8/19/02	DESTRUCTION I.	Μ	А	31	164.801	47 40.84	124 28.48
109	8/19/02	PERKINS/RK 443	F	SA	20	164.973	47 46.81	124 29.66
110	8/19/02	PERKINS/RK 443	F	А	25	164.922	47 46.48	124 29.66
111	8/20/02	GIANTS GRVYD	F	А	26	164.694	47 51.02	124 33.98
112	8/21/02	CAPE JOHNSON	Μ	А	37	164.862	47 59.02	124 41.02
113	8/21/02	CAPE JOHNSON	Μ	А	27	164.553	47 59.02	124 41.02
114	8/22/02	DESTRUCTION I.	F	А	30	164.773	47 40.39	124 29.34
115	8/22/02	DESTRUCTION I.	F	Р	15	NONE	47 40.39	124 29.34

DATE	AREA OF RADIOTELEMETRY SURVEY	HRS
10/2/01	Columbia River to Neah Bay	5.7
10/4/01	Elwha River to Split/Willoughby Rocks	4.0
10/23/01	Cancelled due to bad weather	
10/25/01	Cancelled due to bad weather	
10/30/01	Cancelled due to bad weather	
11/6/01	Columbia River to Lyre River	7.7
11/8/01	Grays Harbor to Lyre River (with broken coaxial cable)	7.2
11/13/01	Cancelled due to bad weather	
11/15/01	Cancelled due to bad weather	
11/20/01	Cancelled due to bad weather	
11/27/01	Pacific Beach to Elwha River	6.8
2/4/02	Columbia River to Seal/Sail Rocks	5.0
2/26/02	Columbia River to Elwha River	4.0
4/2/02	Elwha River to Columbia River	7.2
5/20/02	Columbia River to Elwha River	4.2
5/22/02	Pt. Grenville to Seal/Sail Rocks	5.7
6/19/02	Willapa Bay to Neah Bay	3.5
10/16/02	Pacific Beach to Pillar Point	6.5
10/17/02	Race Rocks to Esteven Point BC	7.5
11/26/02	Pt. Grenville to Tatoosh Island	3.3
1/16/03	Cancelled due to bad weather	
2/10/03	Columbia River to Lyre River	6.5
4/9/03	Elwha River to Quinault River	7.7
4/10/03	Race Rocks to Esteven Point BC	7.0
4/12/03	Cancelled due to bad weather	
6/22/03	Cancelled due to fog/weather	
7/7/03	Columbia River to Elwha River	3.9
7/8/03	Destruction Island to Neah Bay	5.8
7/9/03	Pacific Beach to Elwha River	6.5
7/10/03	Destruction Island to Sekiu River	5.9
8/18/03	Grays Harbor to Giants Graveyard (end with fog to north)	5.1
9/11/03	Cancelled due to bad weather	
10/21/03	Cancelled due to bad weather	
11/3/03	Pt. Grenville to Pillar Point	6.0
11/12/03	Pt. Grenville to Sekiu River	7.0
12/9/03	Columbia River to Elwha River	6.8

### Appendix 1a Table 3. Summary of Sea Otter Telemetry Surveys from 2001 to 2003.

TABLE 4.	DISTA	NCE (N	/ILES)	FROM	CAPTU	ire loo		NS OF S	SEA OT	TERS F	RADIO	TAGGED	) IN 20	01.	
								SEA O	TTER N	IUMBEF	२				
DATE	84	86	88	89	90	91	92		94	95	96	97	98	99	100
10/2/01	32.7	1.3		1	4.4			0.3			3.3	3.3	0.2		
10/4/01	33		1.7	0.1	0.3						3.1	0.1	0.1	6	7
11/6/01	31					19.3		1.4	0.2				1.7	5.5	
11/27/01	37.3	1.1		0.4	10.2	19.4	37.4	10			1.5		0.5	6.3	6.5
2/4/02			<11.3>	•	0.7	20	37.7	10.4			9.9		0.1		
4/2/02	31.1	5.4		0.9	10.2		39.4	9.3			11.8		0.1	0.1	8.7
5/20/02	37.5	0.5		0.3	6.1	<23.6	37.2	3.4				<47.3	0.6	6.6	6.6
5/22/02	30.8	1.7		3.1	6		37.9	0.4			6.7		0.1	7.2	7.2
6/19/02					3.1						2.3		0.8		7.9
10/16/02	24.6	1.9		6	0.1		13.8							7.6	
11/26/02		1.7			11.1		14.8	10.8						7.2	
2/10/02					0.3			10.3						0.1	
4/9/02							13.4								
7/7/03		1.9		3.1			38.5	3.2			0.3				7.1
7/8/03	31.7														
11/3/03				1.2										0.6	
11/12/03	24.8			3.4											
12/9/03	25														7.7

TABLE 5.	DISTAN	ICE (M	ILES) F	ROM	CAPTU	RE LOO	CATION	S OF S	EA OT	TERS R	ADIOT	AGGED	) IN 20	02.
							SEA C	TTER N	NUMBE	R				
DATE	102	103	105	106	54	107	108	109	110	111	112	113	114	
10/16/02	0.1	0.1	30.1	5.6		0.5	0.4	4.2	5.5	0.9	24.6	1.8		
11/26/02			0.5	5.9		1.1		0.1	3	13.8	24.6		0.1	
2/10/03		3.5	0.1	2.7	<24.3>	•		0.3	6.5				1.1	
4/9/03	0.8	1.2	0.6	7.8					2.9		0.3		25.8	
7/7/03		1.1	4.9	7.3		0.3		3	3				0.6	
7/8/03	13.2									7	0.2	0.2		
8/18/03	9.8		0.1					0.5	6.4				0.1	
11/3/03		<2.7>	1.5							9.9	25.1			
11/12/03			0.6			0.9			21	26				
12/9/03									12.7	1.6				

#### Sea Otter Aerial Survey Telemetry Resight Logs for 2001-2003.

### 2001 Sea Otter Radiotelemetry Resight Form

DATE: 2 October 2001 (Grays Harbor to Neah Bay) SURVEY TYPE: Aerial OBSERVERS: Steve Jeffries and Dyanna Lambourn

Otter No.	Age/ Sex	Radiotag Frequency	Capture Location	Resight Time	Resight Location	Latitude	Longitude	Activity	Miles from Capture Site	Comments
84	AD/F	164.334	Cape Alava	11:50	Diamond Pt.	47 45.20	124 27.33	Resting	32.7	
85	P/F		Cape Alava					8		
86	AD/F	164.576	Ozette R.	12:15	Cape Alava	48 09.90	124 44.30	Resting	1.3	
87	P/M		Ozette R.		<b>^</b>					
88	AD/F	164.697	Ozette R.							
89	AD/F	164.495	Cape Alava	13:05	Ozette Island	48 09.21	124 44.65	Resting	1.0	
90	AD/F	164.054	Cape Alava	12:54	Father & Son	48 13.60	124 42.02	Active	4.4	Rapid pulse rate
91	AD/F	164.451	Cape Alava							Interference
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava	12:15	Cape Alava	48 09.76	124 44.35	Resting	0.3	
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point	12:15	Cape Alava	48 09.88	124 44.30	Resting	3.3	
97	AD/M	164.014	Destruction	11:49	Abbey Island	47 42.90	124 25.50	Active	3.3	
98	AD/M	164.173	Destruction	13:13	Destruction Is	47 40.72	124 28.43	Resting	0.2	
99	AD/F	164.596	Rks/#443							
100	AD/F	164.216	Rks/#443							

# 2001 Sea Otter Radiotelemetry Resight Form

DATE: 4 October 2001 (Elwha River to Split/Willoughby Rks) SURVEY TYPE: **Aerial** OBSERVERS: Steve Jeffries and Dyanna Lambourn

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava	14:17	Diamond Pt	47 45.23	124 27.30	Resting	33.0	
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	13:52	Duk Pt	48 12.05	124 42.12	Active	1.7	
89	AD/F	164.495	Cape Alava	13:57	Cape Alava	48 09.90	124 44.23	Resting	0.1	
90	AD/F	164.054	Cape Alava	13:57	Cape Alava	48 09.85	124 44.28	Resting	0.3	Rapid pulse
91	AD/F	164.451	Cape Alava							Interference
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point	13:57	Cape Alava	48 09.80	124 44.25	Resting	3.1	
97	AD/M	164.014	Destruction	14:22	Destruction	47 40.70	124 28.52	Resting	0.1	
98	AD/M	164.173	Destruction	14:22	Destruction	47 40.65	124 28.50	Resting	0.1	
99	AD/F	164.596	Rks #443	14:12	GiantsGrvyd	47 51.00	124 33.80	Active	6.0	
100	AD/F	164.216	Rks #443	14:22	Destruction	47 40.66	124 28.00	Resting	7.0	

# 2001 Sea Otter Radiotelemetry Resight Form

DATE: 6 November 2001 (Columbia River to Lyre River) SURVEY TYPE: **Aerial** OBSERVERS: Steven Jeffries and Dyanna Lambourn

Otter No.	Age/ Sex	Radiotag Frequency	Capture Location	Resight Time	Resight Location	Latitude	Longitude	Activity	Miles from Capture Site	Comments
84	AD/F	164.334	Cape Alava	13:06	Hoh Head	47 46.56	124 29.95	Active	31.0	
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.							
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava	14:03	E. Slant Rk	48 23.51	124 38.26	Resting	19.3	Interference
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava	13:54	Ozette R.	48 11.16	124 23.02	Active	1.4	
94	AD/F	164.135	Cape Alava	13:52	Cape Alava	48 10.12	124 43.88	Active	0.2	
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction							
98	AD/M	164.173	Destruction	11:45	Midway DI	47 41.27	124 26.46	Active	1.7	
99	AD/F	164.596	Rks #443	13:22	Strwbrry Pt	47 50.40	124 33.66	Active	5.5	
100	AD/F	164.216	Rks #443							

### 2001 Sea Otter Radiotelemetry Resight Form

DATE: 8 November 2001 (Grays Harbor to Lyre River w/ broken coaxial cable) SURVEY TYPE: **Aerial** OBSERVERS: Steve Jeffries and Dyanna Lambourn, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.							
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava							Interference
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction							
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks #443							
100	AD/F	164.216	Rks #443							

Telemetry malfunction-coaxial cable to receiver broken

## 2001 Sea Otter Radiotelemetry Resight Form

DATE: 27 November 2001 (Pacific Beach to Elwha River) SURVEY TYPE: **Aerial** OBSERVER: Steven Jeffries, WDFW

Otter No.	Age/ Sex	Radiotag Frequency	Capture Location	Resight Time	Resight Location	Latitude	Longitude	Activity	Miles from Capture Site	Comments
84	AD/F	164.334	Cape Alava	11:14	W DI	47 40.63	124 29.27	Active	37.3	
85	P/F	101.551	Cape Alava	11.11	W DI	17 10.05	12129.27	Tietre	57.5	
86	AD/F	164.576	Ozette R.	12:08	Cape Alava	48 10.35	124 44.50	Resting	1.1	
87	P/M	101.570	Ozette R.	12.00	Cupernava	10 10:55	12111.50	resting	1.1	
88	AD/F	164.697	Ozette R.							
89	AD/F	164.495	Cape Alava	12:03	Cape Alava	48 10.35	124 44.81	Resting	0.4	
90	AD/F	164.054	Cape Alava	13:34	Anderson Pt	48 18.18	124 40.07	Resting	10.2	Rapid pulse
91	AD/F	164.451	Cape Alava	12:30	E. of Slant	48 23.58	124 40.30	Resting	19.4	Interference
92	P/M	164.294	Cape Alava	13:09	NE DI	47 40.90	124 28.60	Active	37.4	
93	AD/F	164.516	Cape Alava	12:18	Anderson Pt	48 18.43	124 40.99	Active	10.0	
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point	11:51	N Yellwbnk	48 06.23	124 42.15	Resting	1.5	
97	AD/M	164.014	Destruction							
98	AD/M	164.173	Destruction	11:08	NE DI	47 40.99	124 29.18	Active	0.5	
99	AD/F	164.596	Rks #443	11:29	GiantsGrvyd	47 51.13	124 33.74	Resting	6.3	
100	AD/F	164.216	Rks #443	12:57	GiantsGrvyd	47 51.20	124 34.21	Resting	6.5	

## 2002 Sea Otter Radiotelemetry Resight Form

DATE: 4 February 2002 (Columbia River to Seal/Sail Rocks) SURVEY TYPE: **Aerial** OBSERVERS: Steve Jeffries, WDFW

Otter No.	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	13:58	Waatch Pt	48 20.00	124 40.7	Dead	11.3	Mortality signal
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava	13:43	Cape Alava	48 10.40	124 45.80	Active	0.7	Rapid but active
91	AD/F	164.451	Cape Alava	14:06	Slant Rock	48 23.60	124 39.70	Resting	20.0	interference
92	P/M	164.294	Cape Alava	13:16	DI	48 40.80	124 28.60	Active	37.7	
93	AD/F	164.516	Cape Alava	13:56	Andrsn Pt	48 18.4	124 40.9	Active	10.4	
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point	13:51	Pt of Arches	48 15.1	124 42.3	Resting	9.9	
97	AD/M	164.014	Destruction							
98	AD/M	164.173	Destruction	13:12	DI	47 40.8	124 28.6	Resting	0.1	
99	AD/F	164.596	Rks #443							
100	AD/F	164.216	Rks #443							
Ref		164.754	OLM	11:37	OLM					

### 2002 Sea Otter Radiotelemetry Resight Form

DATE: 26 February 2002 (Columbia River to Destruction Island) Note: end at DI due to bad interference SURVEY TYPE: **Aerial** OBSERVERS: Steve Jeffries, WDFW and Peter Olesiuk, DFO

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead						
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava							
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction							
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks #443							
100	AD/F	164.216	Rks #443							

## 2002 Sea Otter Radiotelemetry Resight Form

DATE: 2 April 2002 (Elwha River to Columbia River) SURVEY TYPE: **Aerial** OBSERVERS: Steve Jeffries, WDFW and Peter Olesiuk, DFO

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava	11:55	Rks 443	47 46.89	124 29.76	Resting	31.1	
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.	11:28	Sand Pt	48 06.49	124 42.94	Active	5.4	
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava	11:20	S. Ozette Is	48 09.28	124 44.54	Resting	0.9	w/ raft of 30
90	AD/F	164.054	Cape Alava	11:02	N Andrsn Pt	48 18.04	124 41.51	Resting	10.2	
91	AD/F	164.451	Cape Alava							Bad interference
92	P/M	164.294	Cape Alava	12:10	DI east	47 40.86	124 28.78	Active	39.4	w/ 180
93	AD/F	164.516	Cape Alava	11:07	Portage Head	48 17.40	124 41.70	Active	9.3	Swimming north
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point	12:35	S. Portge Hd	48 16.57	124 41.96	Active	11.8	
97	AD/M	164.014	Destruction							
98	AD/M	164.173	Destruction	12:09	DI east	47 40.86	124 28.78	Active	0.1	w/ 180
99	AD/F	164.596	Rks #443	11:56	Rks 443	47 46.89	124 29.50	Active	0.1	
100	AD/F	164.216	Rks #443	13:55	DI west	47 40.22	124 29.35	Resting	8.7	

### 2002 Sea Otter Radiotelemetry Resight Form

DATE: 20 May 2002 (Columbia River to Elwha River) SURVEY TYPE: **Aerial** OBSERVER(S): Steve Jeffries and Dyanna Lambourn, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location		_	-	<b>Capture Site</b>	
84	AD/F	164.334	Cape Alava	14:43	DI	47 41.45	124 28.40	Resting	37.5	
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.	15:32	S. of Ozette R	48 10.69	124 44.33	Resting	0.5	w/10
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava	15:27	Cape Alava	48 09.86	124 44.33	Resting	0.3	w/6
90	AD/F	164.054	Cape Alava	15:20	Yellowbanks	48 05.22	124 41.77	Resting	6.1	w/24 rapid pulse
91	AD/F	164.451	Cape Alava	15:57	Waadah N side	48 23.13	124 36.00	Dead	23.6	Mortality on beach
92	P/M	164.294	Cape Alava	14:43	DI	47 41.45	124 28.40	Active	37.2	
93	AD/F	164.516	Cape Alava	15:39	Duk	48 12.08	124 42.34	Resting	3.4	w/30
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	15:44	ShiShi N. end	48 16.65	124 40.80	Dead	47.3	Dead at north end
98	AD/M	164.173	Destruction	14:43	DI	47 41.45	124 28.40	Active	0.6	
99	AD/F	164.596	Perkins/#443	14:43	DI	47 41.45	124 28.40	Active	6.6	
100	AD/F	164.216	Perkins/#443	14:43	DI	47 41.45	124 28.40	Resting	6.6	

### 2002 Sea Otter Radiotelemetry Resight Form

DATE: 22 May 2002 (Pt Grenville to Seal/Sail Rks) SURVEY TYPE: **Aerial** OBSERVER: Steve Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava	12:31	Rks 443	47 46.89	124 29.76	Active	30.8	w/50
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.	13:14	Duk	48 12.08	124 42.34	Resting	1.7	w/40
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava	13:15	Duk	48 12.08	124 42.34	Resting	3.1	w/40
90	AD/F	164.054	Cape Alava	12:57	Yellowbanks	48 05.22	124 41.77	Active	6.0	rapid pulse/alive
91	AD/F	164.451	Cape Alava	Dead				Dead		
92	P/M	164.294	Cape Alava	12:12	DI	47 40.86	124 28.78	Active	37.9	w/100
93	AD/F	164.516	Cape Alava	13:11	Cape Alava	48 09.86	124 44.33	Resting	0.4	w/8
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point	13:14	Duk	48 12.08	124 42.34	Resting	6.7	w/40
97	AD/M	164.014	Destruction	Dead				Dead		
98	AD/M	164.173	Destruction	12:12	DI	47 40.86	124 28.78	Resting	0.1	w/100
99	AD/F	164.596	Perkins/#443	12:13	DI	47 40.86	124 28.78	Resting	7.2	w/100
100	AD/F	164.216	Perkins/#443	12:15	DI	47 40.86	124 28.78	Active	7.2	w/100

### 2002 Sea Otter Radiotelemetry Resight Form

DATE: 19 June 2002 (Willapa Bay to Neah Bay) SURVEY TYPE: **Aerial** OBSERVER: Steve Jeffries, WDFW

Otter No.	Age/ Sex	Radiotag Frequency	Capture Location	Resight Time	Resight Location	Latitude	Longitude	Activity	Miles from Capture Site	Comments
84	AD/F	164.334	Cape Alava	Time	Location				Capture Site	
-		104.334	1							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava	16:50	Duk	48 12.11	124 42.20	Resting	3.1	w/60
91	AD/F	164.451	Cape Alava	Dead				Dead		
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point	16:38	Yellowbanks	48 05.44	124 42.34	Resting	2.3	w/15
97	AD/M	164.014	Destruction	Dead				Dead		
98	AD/M	164.173	Destruction	16:06	DI east	47 40.18	124 29.00	Active	0.8	w/150
99	AD/F	164.596	Rks #443							
100	AD/F	164.216	Rks #443	15:58	DI west	47 40.15	124 29.35	Resting	7.9	w/15

### 2002 Sea Otter Radiotelemetry Resight Form

DATE: 16 October 2002 (Pacific Beach to Pillar Point)

SURVEY TYPE: Aerial

OBSERVERS: Steve Jeffries, WDFW and Kristen Walker, Portland State University

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location		C	•	<b>Capture Site</b>	
84	AD/F	164.334	Cape Alava	11:05	GiantsGravyd	47.51.06	124 33.68	Resting	24.6	
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.	13:21	Cape Alava	48 09.77	124 44.67	Active	1.9	Weak signal
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava	11:59	Pt of Arches	48 14.56	124 42.95	Active	6.0	
90	AD/F	164.054	Cape Alava	11:38	Cape Alava	48 09.90	124 44.50	Active	0.1	Rapid pulse rate
91	AD/F	164.451	Cape Alava	Dead				Dead		
92	P/M	164.294	Cape Alava	11:18	Bluff Pt	47 58.58	124 40.81	Resting	13.8	
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	Dead				Dead		
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks #443	10:27	DI	47 40.51	124 29.76	Active	7.6	
100	AD/F	164.216	Rks #443							

## 2002 Sea Otter Radiotelemetry Resight Form

DATE: 16 October 2002 (Columbia River to Pillar Point) SURVEY TYPE: Aerial OBSERVERS: Steve Jeffries, WDFW and Kristen Walker, Portland State University

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
102	AD/M	164.353	DI	10:18	DI	47 41.54	124 28.70	Resting	0.1	
103	AD/F	164.373	Sand Point	11:33	Sand Pt	48 07.15	124 42.72	Active	0.1	
104	P/		Sand Point							
105	AD/M	164.931	DI	13:13	Norwgn Mem	48 02.92	124 42.12	Active	30.1	
106	AD/F	164.882	Rks #443	11:01	Toleak Pt	47 50.50	124 32.48	Resting	5.6	
54	AD/F	164.754	Cape Alava							
107	AD/M	164.433	DI	10:16	DI	47 40.91	124 29.12	Resting	0.5	
108	AD/M	164.801	DI	12:36	DI	47 40.95	124 29.00	Active	0.4	
109	SA/F	164.973	Rks #443	10:48	Toleak Pt	47 49.80	124 32.20	Active	4.2	Rapid pulse rate
110	AD/F	164.922	Rks #443	10:47	Strawberry Pt	47 50.72	124 32.94	Active	5.5	
111	AD/F	164.694	GiantsGravyrd	12:55	Strawberry Pt	47 50.70	124 32.80	Resting	0.9	
112	AD/M	164.862	Cape Johnson	12:31	DI	47 40.93	124 27.86	Active	24.6	
113	AD/M	164.553	Cape Johnson	11:13	Cape Johnson	47 57.0	124 39.80	Active	1.8	
114	AD/F	164.773	DI							
115	P/F		DI							

## 2002 Sea Otter Radiotelemetry Resight Form

DATE: 17 October 2002 (Race Rks to Esteven Pt, BC) SURVEY TYPE: **Aerial** OBSERVERS: Steve Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead						
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava	Dead						
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	Dead						
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks 443							
100	AD/F	164.216	Rks #443							

## 2002 Sea Otter Radiotelemetry Resight Form

DATE: 26 November 2002 (Pt Grenville to Tatoosh Island) SURVEY TYPE: Aerial OBSERVERS: Steve Jeffries

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.	15:04	Cape Alava	48 09.70	124 44.56	Active	1.7	
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead						
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava	15:14	Anderson Pt	48 18.50	124 39.20	Active	11.1	Rapid/mortality
91	AD/F	164.451	Cape Alava	Dead						
92	P/M	164.294	Cape Alava	15:32	CapeJohnsn	47 5760	124 39.80	Active	14.8	
93	AD/F	164.516	Cape Alava	15:13	Anderson Pt	48 18.65	124 39.91	Active	10.8	
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	Dead						
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks #443	14:28	DI	47 40.90	124 28.91	Resting	7.2	
100	AD/F	164.216	Rks #443							

## 2002 Sea Otter Radiotelemetry Resight Form

DATE: 26 November 2002 (Pt Grenville to Tatoosh Island) SURVEY TYPE: Aerial OBSERVERS: Steve Jeffries

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
102	AD/M	164.353	DI							
103	AD/F	164.373	Sand Point							
104	P/		Sand Point							
105	AD/M	164.931	DI	14:24	DI	47 41.20	124 28.70	Active	0.5	
106	AD/F	164.882	Rks #443	14:42	GiantsGravyrd	47 50.88	124 33.70	Active	5.9	
54	AD/F	164.754	Cape Alava							
107	AD/M	164.433	DI	14:20	DI	47 40.45	124 29.77	Resting	1.1	
108	AD/M	164.801	DI							
109	SA/F	164.973	Rks #443	15:48	Rk 443	47 46.79	124 29.49	Active	0.1	rapid/mortality
110	AD/F	164.922	Rks #443	15:42	Goodmn Crk	47 49.24	124 30.50	Resting	3.0	
111	AD/F	164.694	GiantsGravyrd	14:21	DI	47 40.45	124 29.77	Active	13.8	
112	AD/M	164.862	Cape Johnson	14:25	DI	47 41.20	124 28.70	Active	24.6	
113	AD/M	164.553	Cape Johnson							
114	AD/F	164.773	DI	14:27	DI	47 40.49	124 29.50	Active	0.1	
115	P/F		DI							

## 2003 Sea Otter Radiotelemetry Resight Form

DATE: 10 Feb 2003 (Columbia River to Lyre River) SURVEY TYPE: **Aerial** OBSERVER: Steve Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava	13:45	Cape Alava	48 09.92	124 44.23	Active	0.3	Rapid pulse rate
91	AD/F	164.451	Cape Alava	Dead	_			Dead		
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava	14:08	Anderson Pt	48 17.83	124 40.90	Resting	10.3	
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	Dead				Dead		
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks #443	13:05	Rks 443	47 46.81	124 29.70	Active	0.1	
100	AD/F	164.216	Rks #443							

## 2003 Sea Otter Radiotelemetry Resight Form

DATE: 10 Feb 2003 (Columbia River to Lyre River) SURVEY TYPE: **Aerial** OBSERVER: Steven Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
102	AD/M	164.353	DI							
103	AD/F	164.373	Sand Point	13:54	Cape Alava	48 10.12	124 44.19	Active	3.5	
104	P/		Sand Point							
105	AD/M	164.931	DI	12:52	DI	47 40.80	124 28.53	Active	0.1	
106	AD/F	164.882	Rks #443	13:01	Diamond Rk	47 45.23	124 27.24	Active	2.7	
54	AD/F	164.754	Cape Alava	14:20	Waadah	48 22.68	124 35.84	Dead	24.3	Rapid, tag on beach
107	AD/M	164.433	DI							
108	AD/M	164.801	DI							
109	SA/F	164.973	Rks #443	13:10	Rks 443	47 46.55	124 29.84	Active	0.3	Rapid pulse, alive
110	AD/F	164.922	Rks #443	13:18	GiantsGrvyrd	47 51.08	124 33.76	Resting	6.5	
111	AD/F	164.694	Giants Gravyrd							
112	AD/M	164.862	Cape Johnson							
113	AD/M	164.553	Cape Johnson							
114	AD/F	164.773	DI	12:49	DI	47 40.74	124 28.42	Resting	1.1	
115	P/F		DI							

## 2003 Sea Otter Radiotelemetry Resight Form

DATE: 9 April 2003 (Elwha River to Quinault River) SURVEY TYPE: **Aerial** OBSERVER: Steven Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location		_		<b>Capture Site</b>	
84	AD/F	164.334	Cape Alava							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava	Dead				Dead		
92	P/M	164.294	Cape Alava	12:23	Bluff Pt	47 58.75	124 41.31	Resting	13.4	
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	Dead				Dead		
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks #443							
100	AD/F	164.216	Rks #443							

## 2003 Sea Otter Radiotelemetry Resight Form

DATE: 9 April 2003 (Elwha River to Quinault River) SURVEY TYPE: **Aerial** OBSERVER: Steven Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location		_		Capture Site	
102	AD/M	164.353	DI	12:46	DI	47 40.27	124 28.61	Active	0.8	
103	AD/F	164.373	Sand Point	12:09	Sand Pt	48 08.47	124 43.92	Active	1.2	
104	P/		Sand Point							
105	AD/M	164.931	DI	12:46	DI	47 40.27	124 28.61	Active	0.6	
106	AD/F	164.882	Rks #443	12:46	DI	47 40.27	124 28.61	Active	7.8	
54	AD/F	164.754	Cape Alava	Dead	Waadah			Dead	24.3	Tag on beach
107	AD/M	164.433	DI							
108	AD/M	164.801	DI							
109	SA/F	164.973	Rks #443							
110	AD/F	164.922	Rks #443	12:36	GoodmanCrk	47 49.05	124 30.20	Resting	2.9	
111	AD/F	164.694	GiantsGravyrd							
112	AD/M	164.862	Cape Johnson	12:21	Bluff Pt	47 58.75	124 41.31	Resting	0.3	
113	AD/M	164.553	Cape Johnson							
114	AD/F	164.773	DI	12:21	Bluff Pt	47 58.75	124 41.31	Resting	25.8	
115	P/F		DI							

## 2003 Sea Otter Radiotelemetry Resight Form

DATE: 10 April 2003 (Race Rks to Estevan Pt, BC) SURVEY TYPE: **Aerial** OBSERVER: Steven Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava	Dead				Dead		
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	Dead				Dead		
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks #443							
100	AD/F	164.216	Rks #443							

## 2003 Sea Otter Radiotelemetry Resight Form

DATE: 10 April 2003 (Race Rks to Estevan Pt, BC) SURVEY TYPE: **Aerial** OBSERVER: Steven Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
102	AD/M	164.353	DI							
103	AD/F	164.373	Sand Point							
104	P/		Sand Point							
105	AD/M	164.931	DI							
106	AD/F	164.882	Rks #443							
54	AD/F	164.754	Cape Alava	Dead				Dead		
107	AD/M	164.433	DI							
108	AD/M	164.801	DI							
109	SA/F	164.973	Rks #443							
110	AD/F	164.922	Rks #443							
111	AD/F	164.694	GiantsGravyrd							
112	AD/M	164.862	Cape Johnson							
113	AD/M	164.553	Cape Johnson							
114	AD/F	164.773	DI							
115	P/F		DI							

## 2003 Sea Otter Radiotelemetry Resight Form

DATE: July 7, 2003 (Columbia River to Elwha River) SURVEY TYPE: **Aerial** OBSERVER: Steven Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.	1758	Duk	48 12.20	124 42.10	Active	1.9	
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava	1757	Duk	48 12.18	124 42.16	Active	3.1	
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava	Dead				Dead		
92	P/M	164.294	Cape Alava	1709	DI	47 40.80	124 28.68	Active	38.5	
93	AD/F	164.516	Cape Alava	1756	Duk	48 12.15	124 42.14	Active	3.2	
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point	1748	Sandpoint	48 07.55	124 42.80	Active	0.3	
97	AD/M	164.014	Destruction	Dead				Dead		
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks #443							
100	AD/F	164.216	Rks #443	1710	DI	47 40.74	124 28.69	Resting	7.1	Rapid mort. signal

### 2003 Sea Otter Radiotelemetry Resight Form

DATE: July 7, 2003 (Columbia River to Elwha River) SURVEY TYPE: **Aerial** OBSERVER: Steven Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
102	AD/M	164.353	DI							
103	AD/F	164.373	Sand Point	1749	Weddng Rks	48 08.22	124 43.12	Active	1.1	
104	P/		Sand Point							
105	AD/M	164.931	DI	1722	HohR mouth	47 44.90	124 27.19	Active	4.9	
106	AD/F	164.882	Rks #443	1710	DI	47 40.72	124 28.64	Active	7.3	
54	AD/F	164.754	Cape Alava	Dead	Waadah			Dead		Tag buried in sand
107	AD/M	164.433	DI	1705	DI	47 40.68	124 28.72	Active	0.3	
108	AD/M	164.801	DI							
109	SA/F	164.973	Rks #443	1720	HohR mouth	47 44.95	124 27.24	Resting	3.0	
110	AD/F	164.922	Rks #443	1719	HohR mouth	47 44.85	124 27.20	Active	3.0	
111	AD/F	164.694	GiantsGravyrd							
112	AD/M	164.862	Cape Johnson							
113	AD/M	164.553	Cape Johnson							
114	AD/F	164.773	DI	1707	DI	47 40.65	124 28.60	Active	0.6	
115	P/F		DI							

#### 2003 Sea Otter Radiotelemetry Resight Form

DATE: July 8, 2003 (Destruction Island to Neah Bay) Note: scan frequencies not heard on 7/7/03 flight SURVEY TYPE: Aerial OBSERVERS: Steven Jeffries, WDFW and Lisa Triggs, Pt. Defiance Zoo and Aquarium

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location			-	<b>Capture Site</b>	
84	AD/F	164.334	Cape Alava	1204	Rk 443	47 46.29	124 29.70	Active	31.7	
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead						
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava	Dead						
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	Dead						
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Perkins/#443							
100	AD/F	164.216	Perkins/#443							

#### 2003 Sea Otter Radiotelemetry Resight Form

DATE: July 8, 2003 (Destruction Island to Neah Bay) Note: scan frequencies not heard on 7/7/03 flight SURVEY TYPE: Aerial OBSERVERS: Steven Jeffries, WDFW and Lisa Triggs, Pt. Defiance Zoo and Aquarium

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
102	AD/M	164.353	DI	1214	GiantsGravyrd	47 50.92	124 33.72	Active	13.2	Signal weak
103	AD/F	164.373	Sand Point							
104	P/		Sand Point							
105	AD/M	164.931	DI							
106	AD/F	164.882	Rks #443							
54	AD/F	164.754	Cape Alava	Dead				Dead		
107	AD/M	164.433	DI							
108	AD/M	164.801	DI							
109	SA/F	164.973	Rks #443							
110	AD/F	164.922	Rks #443							
111	AD/F	164.694	GiantsGravyrd	1204	Rks 443	47 46.20	124 29.70	Active	7.0	
112	AD/M	164.862	Cape Johnson	1224	Bluff Pt	47 59.20	124 41.00	Resting	0.2	
113	AD/M	164.553	Cape Johnson	1224	Bluff Pt	47 59.20	124 41.00	Active	0.2	
114	AD/F	164.773	DI							
115	P/F		DI							

#### 2003 Sea Otter Radiotelemetry Resight Form

DATE: July 9, 2003 (Pacific Beach to Elwha River) Note: only scan frequencies not heard on 7/7-8/03 SURVEY TYPE: **Aerial** OBSERVERS: Steven Jeffries, WDFW and Lisa Triggs, Pt. Defiance Zoo and Aquarium

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava	Dead				Dead		
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	Dead				Dead		
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks #443							
100	AD/F	164.216	Rks #443							

#### 2003 Sea Otter Radiotelemetry Resight Form

DATE: July 9, 2003 (Pacific Beach to Elwha River) Note: only scan frequencies not heard on 7/7-8/03 SURVEY TYPE: Aerial OBSERVERS: Steve Jeffries, WDFW and Lisa Triggs, Pt. Defiance Zoo and Aquarium

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				<b>Capture Site</b>	
102	AD/M	164.353	DI							
103	AD/F	164.373	Sand Point							
104	P/		Sand Point							
105	AD/M	164.931	DI							
106	AD/F	164.882	Rks #443							
54	AD/F	164.754	Cape Alava	Dead				Dead		
107	AD/M	164.433	DI							
108	AD/M	164.801	DI							
109	SA/F	164.973	Rks #443							
110	AD/F	164.922	Rks #443							
111	AD/F	164.694	GiantsGravyrd							
112	AD/M	164.862	Cape Johnson							
113	AD/M	164.553	Cape Johnson							
114	AD/F	164.773	DI							
115	P/F		DI							

#### 2003 Sea Otter Radiotelemetry Resight Form

DATE: July 10, 2003 (Destruction Island to Sekiu River Note: only scan frequencies not heard on 7/7-9/03) SURVEY TYPE: Aerial OBSERVERS: Steven Jeffries, WDFW and Lisa Triggs, Pt. Defiance Zoo and Aquarium

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava	Dead				Dead		
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	Dead				Dead		
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks #443							
100	AD/F	164.216	Rks #443							

### 2003 Sea Otter Radiotelemetry Resight Form

DATE: July 10, 2003 (Destruction Island to Sekiu River Note: only scan frequencies not heard on 7/7-9/03) SURVEY TYPE: **Aerial** OBSERVER: Steven Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
102	AD/M	164.353	DI							
103	AD/F	164.373	Sand Point							
104	P/		Sand Point							
105	AD/M	164.931	DI							
106	AD/F	164.882	Rks #443							
54	AD/F	164.754	Cape Alava	Dead				Dead		
107	AD/M	164.433	DI							
108	AD/M	164.801	DI							
109	SA/F	164.973	Rks #443							
110	AD/F	164.922	Rks #443							
111	AD/F	164.694	GiantsGravyrd							
112	AD/M	164.862	Cape Johnson							
113	AD/M	164.553	Cape Johnson							
114	AD/F	164.773	DI							
115	P/F		DI							

### 2003 Sea Otter Radiotelemetry Resight Form

DATE: 18 August 2003 (Grays Harbor to Giants Graveyard) Note: end at Giants Graveyard due to fog to north SURVEY TYPE: Aerial OBSERVER: Steve Jeffries

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				<b>Capture Site</b>	
84	AD/F	164.334	Cape Alava							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava	Dead				Dead		
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	Dead				Dead		
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks #443							
100	AD/F	164.216	Rks #443							

### 2003 Sea Otter Radiotelemetry Resight Form

DATE: 18 August 2003 (Columbia River to Giants Graveyard Note: end at Giants Graveyard due to fog to north) SURVEY TYPE: Aerial OBSERVER: Steve Jeffries

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				<b>Capture Site</b>	
102	AD/M	164.353	DI	12:25	GoodmnCrk	47 49.25	124 30.55	Active	9.8	weak
103	AD/F	164.373	Sand Point							
104	P/		Sand Point							
105	AD/M	164.931	DI	11:50	DI	47 40.70	124 28.61	Active	0.1	
106	AD/F	164.882	Rks #443							
54	AD/F	164.754	Cape Alava	Dead				Dead		
107	AD/M	164.433	DI							
108	AD/M	164.801	DI							
109	SA/F	164.973	Rks #443	12:03	Rks 443	47 47.22	124 29.66	Resting	0.5	mortality signal
110	AD/F	164.922	Rks #443	12:10	GiantsGrvyrd	47 51.15	124 33.55	Resting	6.4	
111	AD/F	164.694	GiantsGravyrd	12:12	GiantsGrvyrd	47 51.09	124 33.49			
112	AD/M	164.862	Cape Johnson							
113	AD/M	164.553	Cape Johnson							
114	AD/F	164.773	DI	12:09	GiantsGrvyrd	47 51.10	124 33.72	Active	0.1	weak
115	P/F		DI							

## 2003 Sea Otter Radiotelemetry Resight Form

DATE: 21 October 2003 (Cancelled bad weather) SURVEY TYPE: **Aerial** OBSERVER: Steven Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava	Dead				Dead		
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	Dead				Dead		
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks #443							
100	AD/F	164.216	Rks #443							

### 2003 Sea Otter Radiotelemetry Resight Form

DATE: 3 November 2003 (Point Grenville to Pillar Point) SURVEY TYPE: Aerial OBSERVER: Steve Jeffries

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				<b>Capture Site</b>	
84	AD/F	164.334	Cape Alava							
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava	14:06	S. Ozette Is	48 09.10	124 44.31	Resting	1.2	
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava	Dead				Dead		
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	Dead				Dead		
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks #443	13:28	Rks 443	47 47.26	124 29.58	Active	0.6	
100	AD/F	164.216	Rks #443							

## 2003 Sea Otter Radiotelemetry Resight Form

DATE: 3 November 2003 (Point Grenville to Pillar Point) SURVEY TYPE: **Aerial** OBSERVER: Steve Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				<b>Capture Site</b>	
102	AD/M	164.353	DI							
103	AD/F	164.373	Sand Point	13:59	Yellowbanks	48 05.13	124 41.35	Dead	2.7	mortality
104	P/		Sand Point							
105	AD/M	164.931	DI	13:14	DI	47 40.52	124 30.32	Active	1.5	active but mort.signal
106	AD/F	164.882	Rks #443							
54	AD/F	164.754	Cape Alava	Dead				Dead		
107	AD/M	164.433	DI							
108	AD/M	164.801	DI							
109	SA/F	164.973	Rks #443							
110	AD/F	164.922	Rks #443							
111	AD/F	164.694	GiantsGravyrd	13:41	Chilean Mem.	47 57.11	124 40.06	Resting	9.9	
112	AD/M	164.862	Cape Johnson	13:12	DI	47 40.64	124 30.71	Active	25.1	
113	AD/M	164.553	Cape Johnson							
114	AD/F	164.773	DI							
115	P/F		DI							

## 2003 Sea Otter Radiotelemetry Resight Form

DATE: 12 November 2003 (Point Grenville to Sekiu River) SURVEY TYPE: Aerial OBSERVER: Steve Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava	15:18	GiantsGrvyrd	47 51.06	124 33.22	Resting	24.8	In raft w/ 10 El's
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava	13:15	N. Sand Pt	48 07.29	124 43.15	Active	3.4	
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava	Dead				Dead		
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	Dead				Dead		
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Perkins/#443							
100	AD/F	164.216	Perkins/#443							

## 2003 Sea Otter Radiotelemetry Resight Form

DATE: 12 November 2003 (Point Grenville to Sekiu River) SURVEY TYPE: Aerial

OBSERVER: Steve Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location		_		<b>Capture Site</b>	
102	AD/M	164.353	DI							
103	AD/F	164.373	Sand Point	Dead				Dead		
104	P/		Sand Point							
105	AD/M	164.931	DI	15:03	DI NE	47 40.41	124 29.12	Active	0.6	active but mort. sig
106	AD/F	164.882	Rks #443							
54	AD/F	164.754	Cape Alava	Dead				Dead		
107	AD/M	164.433	DI	12:57	DI W	47 40.30	124 29.41	Active	0.9	
108	AD/M	164.801	DI							
109	SA/F	164.973	Rks #443							
110	AD/F	164.922	Rks #443	13:37	N Cedar Crk	48 01.24	124 41.31	Active	21.0	
111	AD/F	164.694	GiantsGravyrd	13:41	Ozette R W	48 10.49	124 43.41	Active	26.0	Swimming to Canball
112	AD/M	164.862	Cape Johnson							
113	AD/M	164.553	Cape Johnson							
114	AD/F	164.773	DI							
115	P/F		DI							

### 2003 Sea Otter Radiotelemetry Resight Form

DATE: 9 December 2003 (Columbia River to Elwha River) SURVEY TYPE: **Aerial** OBSERVER: Steven Jeffries, WDFW

Otter	Age/	Radiotag	Capture	Resight	Resight	Latitude	Longitude	Activity	Miles from	Comments
No.	Sex	Frequency	Location	Time	Location				Capture Site	
84	AD/F	164.334	Cape Alava	11:59	GiantsGrvyrd	47 51.01	124 33.83	Resting	25.0	w/12 in raft
85	P/F		Cape Alava							
86	AD/F	164.576	Ozette R.							
87	P/M		Ozette R.							
88	AD/F	164.697	Ozette R.	Dead				Dead		
89	AD/F	164.495	Cape Alava							
90	AD/F	164.054	Cape Alava							
91	AD/F	164.451	Cape Alava	Dead				Dead		
92	P/M	164.294	Cape Alava							
93	AD/F	164.516	Cape Alava							
94	AD/F	164.135	Cape Alava							
95	AD/F	164.674	Sand Point							
96	AD/F	164.396	Sand Point							
97	AD/M	164.014	Destruction	Dead				Dead		
98	AD/M	164.173	Destruction							
99	AD/F	164.596	Rks #443							
100	AD/F	164.216	Rks #443	11:30	DI	47 40.46	124 29.25	Resting	7.7	Rapid mortality

## 2003 Sea Otter Radiotelemetry Resight Form

DATE: 9 December 2003 (Columbia River to Elwha River) SURVEY TYPE: Aerial OBSERVER: Steve Jeffries, WDFW

Otter No.	Age/ Sex	Radiotag Frequency	Capture Location	Resight Time	Resight Location	Latitude	Longitude	Activity	Miles from Capture Site	Comments
102	AD/M	164.353	DI	TIME	Location				Capture Site	
102	AD/M AD/F	164.373	Sand Point	Dead				Dead		
104	P/		Sand Point							
105	AD/M	164.931	DI							
106	AD/F	164.882	Rks #443							
54	AD/F	164.754	Cape Alava	Dead				Dead		
107	AD/M	164.433	DI							
108	AD/M	164.801	DI							
109	SA/F	164.973	Rks #443							
110	AD/F	164.922	Rks #443	12:05	James Island	47 54.69	124 38.98	Active	12.7	
111	AD/F	164.694	GiantsGravyrd	11:50	N. Toleak Pt	47 49.75	124 33.16	Active	1.6	
112	AD/M	164.862	Cape Johnson							
113	AD/M	164.553	Cape Johnson							
114	AD/F	164.773	DI							
115	P/F		DI							

# Appendix 1b. Activity Budgets and Prey Consumption for the Washington Sea Otter, *Enhydra lutris kenyoni*: 2003 and 2004

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> Activity Budgets and Prey Consumption for the Washington Sea Otter, Enhydra lutris kenyoni: 2003 and 2004

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Walker, K.A., Davis, J.W., and D.A. Duffield. Activity Budgets and Prey Consumption for the Washington Sea Otter, *Enhydra lutris kenyoni*: 2003 and 2004.

#### ABSTRACT

Northern sea otters, *Enhydra lutris kenyoni*, were extirpated from the Washington coast in the early 1900's. Reintroductions of sea otters from Amchitka Island, Alaska occurred in 1969 and 1970. By 2005, 814 sea otters were occupying a range from Destruction Island north to Anderson Point, with the majority of individuals located south of La Push, Washington. The focus of this study was to describe local abundance, diurnal activity budgets, and variability in prey consumption at four locations in the current Washington sea otter range. Local abundance varied among the locations. Overall, sea otters primarily spent their daylight hours resting (62.0%), followed by grooming (19.7%), and then feeding (7.6%). Activities varied by location, age, group size, tide, and time of day. Diet consisted predominantly (82%) of crabs, clams, and sea stars. In each study area, one main prey item comprised at least 34% of the total prey consumed; however, the main prey item differed among locations. Study locations were utilized differently, some as resting/pup-rearing sites and others as feeding sites. Overall diurnal feeding activity in this study (7.6%) suggested high food availability in the current Washington sea otter range, signifying that the Washington population is still below equilibrium density.

#### **INTRODUCTION**

Sea otters (*Enhydra lutris kenyoni*) were common along the Washington coast until they were extirpated during the fur trade (Scheffer 1940, Riedman and Estes 1990). The current Washington State sea otter population was subsequently founded from sea otters translocated from Amchitka Island, Alaska. Two groups of translocated sea otters were introduced in 1969 and 1970, resulting in a total of 59 otters (41 females and 18 males) being released into Washington waters over these two years (Bowlby *et al.* 1988). It is believed that the current population was re-established from no more than 43 otters, and possibly as few as 10 (Jameson *et al.* 1982).

Between 1977 and 1989, records indicate that the Washington population increased from 19 to 208 otters (Jameson 1998, Lance *et al.* 2004). Since 1989, the Washington Department of Fish and Wildlife (WDFW) had observed continued growth to a total of 604 animals by 1999 (Lance *et al.* 2004). In the latest census conducted in July 2005, 814 sea otters were counted, a 10% increase from 2004 (Jameson and Jeffries 2005). Overall, there has been an average annual rate of increase of 8.2% since 1989 and it would appear that the Washington sea otter population is still in a positive growth phase (Jameson and Jeffries 2005).

The Washington State sea otter population is currently protected under the Marine Mammal Protection Act of 1972 and under endangered species legislation in the State of Washington. As sea otter numbers continue to increase in Washington there are many management issues that are of concern: oil spill and anthropogenic contaminant risks, shellfish fisheries conflicts, and the effects of sea otters on coastal marine ecosystem structure and dynamics (Lance *et al.* 2004). Ecological factors such as distribution, abundance and prey consumption, as well as time and activity budgets need to be documented not only to aid in these management decisions, but also to support the WDFW Sea Otter Recovery Plan (Lance *et al.* 2004) which is aimed at ensuring that a self-sustaining population of sea otters will eventually exist in Washington.

Bowlby et al. (1988) presented data on Washington sea otter abundance, distribution, and activity budgets collected in 1986 & 1987. However, little dietary composition data for sea otters in Washington has been published, unlike other regions in which sea otters inhabit, most likely due to the remote nature of the area and presence of complex weather patterns which make land-based observations more difficult. Dietary consumption information is especially critical because in recent years there has been an increased presence of immuno-suppressing contaminants in sea otters and there may be a relationship between sea otters ingesting contaminated prey items and mortality events and incidence of pathogens observed in the Washington population (Lance et al. 2004, Dubey et al. 2003). Accordingly, to provide these data and to address changes in the previously measured population parameters, this research study was designed to investigate prey consumption, as well as, local abundance, group size and diurnal activity patterns for sea otters in four known high abundance locations along the Washington State Olympic Peninsula. Additional analyses were conducted to assess the influence of environmental variables such as tide, water and air temperature, time of day, age, and date/year, on these ecological measures.

#### **MATERIALS AND METHODS**

Research was conducted in the Olympic Coast National Marine Sanctuary on the Washington State Olympic Peninsula during the spring and summer months of 2003 and 2004. This was a land-based observation study using coastal viewing areas which were close to large rafts of sea otters and which were accessible by backcountry trails and 4-wheel drive vehicles. Four locations were selected for the concentrated survey effort (Duk Point, Cape Alava, Sand Point, and Norwegian Memorial; Fig.1). These sites were chosen based on accessibility and viewing conditions, as well as to complement toxicological and epidemiological studies being conducted in the area by the U.S. Fish and Wildlife Service.

#### Activity Budgets and Prey Consumption

Based on methods used by Estes *et al.* 1982, an effort was made to evenly distribute observations across time periods for all locations to avoid spatial segregation bias due to point-in-time sampling. However, the actual total amount of time spent at each study location was dependent upon clear viewing conditions and upon accessibility to the study area based on tidal cycles.

When sea otters were in unobstructed viewing areas at the four study locations, sea otter activity was observed from the coast by scanning the water using Cabela Alaskan Guide 10 X 40 binoculars (Cabela, Sidney, Nebraska). An AT-80 high definition Swarovski spotting scope (Swarovski Optik, Cranston, Rhode Island) was used to facilitate closer observations of prey consumption and behavioral activities. During each observation period, the largest number of sea otters in the area was noted every half hour. Date, time, location, observable tidal cycle (high, mid, low), and weather conditions were also recorded.

An ethogram was constructed based on a study by Packard and Ribic (1982), and activity budget data were collected. Activity budget categories included resting, grooming, foraging, traveling, playing, nursing, and other (i.e. copulating, startled). A scan sample method, based on a study by Estes *et al.* (1982), was used to record activity. Scans were conducted with a spotting scope to determine the total number of sea otters concentrated in one location and their individual activity. Age (adult or pup) and the number of sea otters per group at each location were recorded. One minute was spent in each field of view visible through the spotting scope, so as not to exclude otters underwater. This was based on an average reported dive time for sea otters of one minute (Estes *et al.* 1982). During every scan, each sea otter observed was classified as performing one of the seven behaviors. The scan was repeated every thirty minutes, with the total time at each area being as equally distributed as possible among the locations. Activity budgets were calculated from these observations.

Prey consumption data were collected by direct observation of individual sea otters feeding. Foraging observations (eating or engaged in foraging dives) were made between activity budget scans. Once a foraging otter was located, data collection would begin. If more than one individual was observed feeding, the otter closest to shore was observed to facilitate identification of prey species. Once an individual was no longer actively foraging,

moved out of sight, or when the activity budget scan was to resume, the feeding observation was terminated. Location, time of day, and tidal cycle were recorded for all feeding observations, along with sex and age (pup or adult), if identifiable. Sex of the adults was determined by the presence of either a penile or testicular bulge on the male lower abdomen, or the presence of abdominal teats on the female (Riedman and Estes 1990). Pups were not sexed due to difficulties in distinguishing reproductive anatomy. Age was categorized as adult or pup (approximately 0-24 weeks old). Additional data collected during a feeding observation included: the total number of successful and unsuccessful dives, the duration of each observed foraging dive, the number of prey brought to the surface after a foraging dive, and the identification of the prey item. A successful dive was defined by the capture of a prey item. If prey items were not brought back to the surface, the dive was classified as unsuccessful.

Taxonomic classification of prey species was determined to the lowest possible taxon. If there was difficulty in classifying an individual prey species, then the major taxonomic group was indicated (i.e. crab, clam, sea star, mussel, etc.). Prey were classified as unknown when items were either hidden in the sea otter's forepaws, or the otter was too far away for complete confidence in classification.

Tide height data and water and air temperature data for 2003 and 2004 were obtained from the CO-OPS database (<u>http://co-ops.nos.noaa.gov/data\_res.html</u>).

### **Statistical Methods**

All statistical tests were conducted using SPSS (SPSS, Inc. Version 11.5 Chicago, Illinois). A one-way analysis of variance (ANOVA) was conducted to test for significant differences in overall mean abundance (the mean number of sea otters present at a given location). Tukey Honestly Significant Difference (HSD) post hoc tests were used to test for significant differences among locations.

To be able to compare differences in daily activity patterns among the four study locations, the number of sea otters performing a specific behavior was divided by the total number of sea otters observed during the half hour time interval, giving a percentage of time spent in that activity. Chi square contingency tables were then used to test whether each activity was uniform among the four locations, as well as between all paired locations, and, similarly, to test uniformity in prey consumption among locations and by gender.

Stepwise logistic regression was used to investigate the relationship between behavioral activities (adults and pups together) and 10 predictor variables -location, date, year, time (second order polynomial; time and time squared), tide (actual height in meters and daily tidal cycle coded – high, mid, low), water temperature, age, and group size.

Differences in dive durations among locations were tested using one-way ANOVA, followed by a Tukey HSD post hoc comparison. The effects of gender and females with pups on dive duration were also tested using a one-way ANOVA.

The total numbers of successful *vs*. unsuccessful dives were analyzed by logistic regression analysis to look for relationships between unsuccessful dives and location, gender of the sea otter, and the presence of a dependent pup with a foraging mother.

#### **RESULTS**

#### Local Abundance

Significant differences in overall local abundance were found among the four locations (ANOVA:  $F_{(3,315)} = 28.75$ , p < 0.001; Fig. 2). Follow-up pair-wise comparisons found that mean sea otter abundance at Cape Alava and Sand Point did not differ significantly (p = 0.304), however all other pair-wise comparisons were significant at  $p \le 0.01$ . Duk Point had the highest average number of otters observed per  $\frac{1}{2}$  hr activity scan, followed by Cape Alava, Sand Point, and then Norwegian Memorial. The highest number of sea otters observed overall during a single  $\frac{1}{2}$  hr activity scan was 104 at Duk Point (2004), with Cape Alava having the second highest count of 86 sea otters (2003).

#### **Activity Budget Data**

The combined time and activity budget data for 2003-2004 showed that sea otters spent 62.3% of their daylight hours resting, 19.7% grooming, 7.6% feeding, with playing, traveling, nursing, and "other" activities comprising the remaining 10.4% of the activity budget (Table 1). While, overall, sea otters spent the majority of the daylight hours resting, the proportion of the time spent in resting, feeding, traveling, and "other" activities varied significantly among locations ( $\chi^2 = 83.09$ , p < 0.001;  $\chi^2 = 91.96$ , p < 0.001;  $\chi^2 = 44.50$ , p < 0.001;  $\chi^2 = 33.72$ , p < 0.001, respectively). On the other hand, diurnal grooming, playing, and nursing activities were not significantly different among the four study sites ( $\chi^2 = 7.74$ , p = 0.05;  $\chi^2 = 6.70$ , p = 0.08;  $\chi^2 = 4.91$ , p = 0.18, respectively).

The different predictor variables - location, date, year, time (second order polynomial), tide (actual height and coded), water temperature, age, and group size, had effects on the various activities.

Resting Activity: Seven of the predictor variables were significantly related to resting activity and accounted for 14% of the variation in feeding (Wald's  $\chi^2$  statistic = 923.1, p < 0.001, Cox-Snell R<sup>2</sup> = 0.14). The greatest contribution to the log-odds of resting was group size; resting activity was higher in larger groups (p < 0.001). Age (adult *vs.* pup) was the second most influential variable with pups resting more than adults (p < 0.001). There was greater resting activity in 2003 (p < 0.001), and resting was less frequent in the morning hours (time; p = 0.026) with a peak in the afternoon (time sq.; p = 0.002). Resting activity increased as tide height decreased (p = 0.021) and as water temperature decreased (p = 0.04).

Grooming Activity: Seven of the predictor variables were significantly related to grooming activity (Wald's  $\chi^2$  statistic = 231.0, p < 0.001), however, the regression model accounted for very little of the observed variability (Cox-Snell R<sup>2</sup> = 0.04). Age was the greatest contributor to the log-odds of grooming, with adults grooming more than pups (p < 0.001). Grooming activity was observed in greater frequency in 2004 (p < 0.001), however this varied among the observation days (p = 0.002). The probability of sea otters grooming increased as tide height decreased (p < 0.001). Grooming had a greater probability of occurring in smaller groups (p < 0.001), and was more common in the middle of the day (time: p = 0.013 and time sq.: p = 0.006).

*Feeding Activity*: Seven of the predictor variables were significantly related to feeding activity, accounting for 21% of the variation in feeding (Wald's  $\chi^2$  statistic =

1469.2, p < 0.001, Cox-Snell R<sup>2</sup> = 0.21). The greatest contribution to the log-odds of feeding was group size: sea otters rarely fed in large groups (p < 0.001). Tide height was the second most influential variable, with the probability of sea otters feeding increasing as tide height increased (p < 0.001). The probability of sea otters feeding was different among the locations (p < 0.001). Adults fed more than pups (p < 0.001), and there was a peak in foraging activity in the late morning and a decrease in the late afternoon (time sq; p = 0.057). Feeding activity was greater in 2004 (p = 0.001), and decreases in water temperature were related to an increase in the log-odds of feeding (p = 0.014).

*Playing Activity*: Four of the predictor variables were significantly related to play activity, but accounted for only 1% of the variability (Wald's  $\chi^2$  statistic = 74.4, p < 0.001, Cox-Snell R<sup>2</sup> = 0.01). The greatest contribution to the log-odds of playing was group size, with the probability of playing increasing as group size decreased (p < 0.001). The probability of sea otters playing decreased in the morning (p < 0.001) and increased in the afternoon hours (p < 0.001), and there was variability in play activity among the observation dates (p = 0.003).

*Traveling Activity*: Six of the predictor variables were significantly related to traveling activity, but accounted for very little of the variability in travel activity (Wald's  $\chi^2$  statistic = 222.97, p < 0.001, Cox-Snell R<sup>2</sup> = 0.04). The greatest contribution to the logodds of traveling was group size (p < 0.001); sea otters traveled in small groups (one to twenty-one individuals), with the exception of occasions where, for example, the majority of a group were resting, became startled and all rapidly swam together in one direction. Traveling significantly decreased in the afternoon hours (p < 0.001) and was observed more often in 2003 (p < 0.001). The probability of sea otters traveling increased as both water temperature (p = 0.002) and tide height (p = 0.028) increased. Adults traveled significantly more than pups (p < 0.001).

*Nursing Activity*: Only pups were classified as nursing with one predictor variable, group size, significantly related to nursing activity (Wald's  $\chi^2$  statistic = 16.48, p < 0.001, Cox-Snell R<sup>2</sup> = 0.03), again accounting for very little variability. As group size decreased, the probability of sea otter pups nursing increased (p = 0.007). In fact, nursing behavior was only witnessed in groups smaller than 30 sea otters, and was most often seen when there was only the mother-pup pair present.

*Other Activity*: The last criterion variable was whether or not sea otters were engaged in "other" activities, 83% of which were acts of copulation. Three of the predictor variables were significantly related to other activity (Wald's  $\chi^2$  statistic = 222.97, p < 0.001, Cox-Snell R<sup>2</sup> = 0.01). The greatest contribution to the log-odds of other activity was the tide height, with other activity increasing during higher tides (p < 0.001). The probability of other activities increased as group size decreased to between two to nine individuals (p < 0.001) and these activities decreased in the morning hours (p < 0.001).

#### **Diet Composition**

Sea otters were observed consuming crabs (Order Decapoda), kelp crabs (*Pugettia producta*), clams (Order Bivalvia), Pacific littleneck clams (*Protothaca staminea*), octopus (*Octopus dofleini*), sea stars (*Pisaster sp.*), Dawson's sea stars (*Solaster dawsoni*), sea

cucumbers (*Cucumaria miniata*), mussels (*Mytilus sp.*), snails (Class Gastropoda), red sea urchins (*Strongylocentrotus franciscanus*), and unknown bivalves (Order Bivalvia).

Overall, sea otters consumed primarily crabs (45.0%), clams (23.1%), and sea stars (13.9%) (Table 2). All other food items together comprised approximately 18.0% of the total dietary composition. In each study area, one main prey type comprised at least 34% of the total consumption of all identifiable prey, but the main prey types differed among locations. The proportion of clams, crabs, sea stars, and mussels all differed significantly among locations ( $\chi^2 = 171.83$ , p < 0.001;  $\chi^2 = 18.09$ , p < 0.001;  $\chi^2 = 118.80$ , p < 0.001;  $\chi^2 = 16.71$ , p = 0.001, respectively).

Overall, females consumed significantly more clams than males ( $\chi^2 = 106.71$ , p < 0.001). Males consumed significantly greater amounts of crabs ( $\chi^2 = 4.05$ , p = 0.044), mussels ( $\chi^2 = 14.24$ , p < 0.001), and unknown prey items ( $\chi^2 = 20.22$ , p < 0.001) than females. All other prey items were consumed uniformly by both sexes.

#### **Dive Durations**

The mean duration for all observed foraging dives was 35.6 sec. The shortest dive recorded (5.3 sec) was by a male at Sand Point who consumed an unknown prey item, and the longest (122 sec) was an unsuccessful dive recorded for an adult female at Norwegian Memorial. The longest dive in which a prey item was captured, was 93.9 sec by a female at Sand Point.

Longer dives occurred when sea otters captured clams ( $X = 45.8 \pm 17.7 \text{ sec}$ , n = 49) and sea stars ( $X = 42.5 \pm 17.9 \text{ sec}$ , n = 17). The shortest dives were for small unknown prey items ( $X = 22.4 \pm 15.7 \text{ sec}$ , n = 25). On 80.1% of observed foraging dives, one prey item was captured, two prey items were observed 18% of the time, and three prey items were captured on 1.9% of the foraging dives.

Dive durations differed significantly among the four locations (ANOVA:  $F_{(3,198)} = 16.65$ , p < 0.001), with location alone explaining 19% of the variation in dive length. The greatest mean dive duration was seen at Norwegian Memorial and was significantly different from both Sand Point (p < 0.001) and Duk Point (p < 0.001). Sea otters at Duk Point dove longer than the otters at Sand Point (p = 0.032). Mean dive durations were not significantly different for males, females, or females with pups (ANOVA:  $F_{(2,143)} = 1.72$ , p = 0.183).

#### Successful vs. Unsuccessful dives

A significant relationship existed between the number of unsuccessful dives and location (Wald's  $\chi^2$  statistic = 10.79, p = 0.001). Duk Point and Cape Alava had the highest proportion of unsuccessful dives with a mean percent of 24% and 21.7%, respectively. The lowest number of observed unsuccessful dives occurred at Sand Point and Norwegian Memorial (15% and 12.3%, respectively).

No relationship was found between the number of unsuccessful dives and the gender of the otters (Wald's  $\chi^2$  statistic = 0.493, p = 0.483), nor between the number of unsuccessful dives and independent females and females with pups (Wald's  $\chi^2$  statistic = 0.590, p = 0.442).

#### DISCUSSION

Lower percentages of foraging activities are believed to occur in areas where sea otter populations are below equilibrium density, as a consequence of still abundant sessile prey (Estes et al. 1982, Garshelis 1983, Estes et al. 1986). Garshelis et al. (1986) proposed a relationship between the amount of time sea otters spent foraging and food availability and that one could use time budget data to assess the availability vs. limitation of prey in different parts of a population's range. This has been supported by studies on diurnal patterns of activity for sea otters at Amchitka Island, Alaska, where the population is assumed to be at or near equilibrium density vs. a population at Attu Island, Alaska which is considered to be below equilibrium density (Estes et al. 1982, Estes 1990). The Amchitka Island population spent 51-58% of the day feeding, while in the Attu Island population sea otters spent only 16-18% of their day feeding. Furthermore, given limited prey availability at Amchitka Island, Estes et al. (1982) concluded that the otters adjusted their foraging activities to exploit fish (60% of their diet), which also resulted in changes in the their diurnal feeding activities. Similarly, a small translocated population of sea otters at Blanco Reef in Oregon (below equilibrium) spent 17% of the day feeding (Estes et al. 1982), compared to the California population (assumed to be approaching equilibrium density) where Estes et al. (1986) found that sea otters spent 21-28% of the day feeding.

The translocated sea otter population in Washington was studied in 1986 and 1987 (Bowlby *et al.* 1988) and at that time exhibited feeding frequencies of 9.5-11.2%, respectively. The overall diurnal feeding activity reported in this study for 2003 and 2004 (7.6%) is similarly low and suggests that there is high food abundance in the Washington sea otter population range. This would indicate that the Washington population is still below equilibrium density, as predicted by an estimated carrying capacity of 2,734 sea otters for this range (Laidre *et al.* 2002).

This study concentrated on four coastal locations in Washington where sea otters had been previously reported. Local abundance fluctuated within and among locations throughout the day and from one observation day to the next. The average local sea otter abundance was greatest at Duk Point and lowest at Norwegian Memorial. Activity budgets were also different among the locations and were influenced by a number of confounding variables: location, tide height, water temperature, time of day, group size, and age. Overall, the highest frequency of resting behavior was observed at Cape Alava, which also had the lowest frequency of feeding activities of all locations. Duk Point had the second highest frequency of resting activity and the second lowest frequency of feeding activity. Conversely, sea otters at Sand Point and Norwegian Memorial exhibited the highest frequencies of feeding activity and the lowest resting activity. This suggests that Duk Point and Cape Alava may be used more as resting and/or pup-rearing sites, while Sand Point and Norwegian Memorial are used more as feeding sites.

In this study, for all Washington locations combined, there was an increase in foraging activities beginning around 0930, a peak occurring in the late morning, and then a significant decrease in foraging activities occurring after 1300. Traveling activities also decreased during the afternoon hours. In California, sea otters tended to have crepuscular peaks in activity, with feeding behavior peaking in the early morning hours (0600) and again in the afternoon (1600-1800) (Estes *et al.* 1986, Ralls and Siniff 1990). At Amchitka

Island, Alaska, similar foraging patterns were observed, with peaks in foraging activity occurring at 0800 and again at 1800-1900 (Estes *et al.* 1982). In contrast, at Attu Island, Alaska, and Blanco Reef, Oregon, sea otters foraged throughout the day (Estes *et al.* 1982). Patterns of behavior appear to be specific to location, which could be influenced by the availability of resources, topography, weather and seasonal effects, and social structure (Garshelis 1983).

In the current study there was a close relationship between tide height and feeding with the greatest feeding activity occurring at the highest tides of the day. The exception to this trend was at Norwegian Memorial where at low tide the greatest feeding occurred. Bowlby *et al.* (1988) also found an increase in foraging behavior at Cape Alava and Sand Point in 1986-87 and at Duk Point in 1987 during higher tide levels, although at Cape Johnson (similar to Norwegian Memorial) the opposite was found to be true. The observations from both studies suggest that activity may be influenced by a combination of variables (e.g. time, location, tide height, group size, age, water temperature) and that these patterns cannot be generalized.

Sea otters have been shown to select prey items that maximize their energy intake, presumably to meet their high metabolic requirements (Ostfeld 1982, Garshelis 1983). As preferred prey items become less abundant, sea otters spend more time foraging in search of the prey items, and switch to other prey items (Ostfeld 1982, Garshelis 1983). For example, red abalones have a very high caloric content (994 kcal) compared with kelp crabs (43 kcal) (Ostfeld 1982). Although more time is spent harvesting the abalone (Ostfeld 1982), the overall net energy gain makes it beneficial for sea otters to consume such prey items when available. Prey items thought to be of high preference to sea otters include: sea urchins, abalone, cancer crabs, and large clams (Kenyon 1969, Garshelis 1983). Less preferred prey (also of lower caloric content) include: snails, kelp crabs, sea stars, mussels, and chitons (Ostfeld 1982, Garshelis 1983).

Studies at Amchitka Island, Alaska, showed that the majority of prey items consumed were sea urchins and fish, while at Attu Island, Alaska, the main prey was sea urchins (Estes *et al.* 1982). In contrast, in Prince William Sound, Alaska, sea otters consumed primarily clams (> 75%) and crabs (Garshelis 1983, Garshelis *et al.* 1986). In California, sea otters consumed primarily crabs, clams, sea urchins, abalone, and mussels (Ostfeld 1982, Ralls *et al.* 1995). From previous studies, Washington sea otters have been shown to feed on crabs, sea urchins, sea cucumbers, sea stars, clams, mussels, snails, octopus, abalones, and chitons (Bowlby *et al.* 1988, Riedman and Estes 1990, Lance *et al.* 2004).

In this study, sea otters in Washington consumed primarily crabs (45.0%), clams (23.1%), and sea stars (13.9%). The remaining prey items consisted of unidentified bivalves, mussels, octopus, snails, sea cucumbers, and a sea urchin. At each location the main prey type was different (Norwegian Memorial - clams; Cape Alava and Sand Point - crabs; Duk Point - sea stars). Almost half of the prey consumed at Duk Point in 2003 and 2004 were sea stars and mussels, which based on the caloric classification by Ostfeld (1982) and Garshelis (1983), are of lower caloric content. This suggests that prey of higher preference, in terms of caloric content, are less abundant at Duk Point.

Over the years, there has been a notable shift in diet at Sand Point and Duk Point suggesting a change in the availability of prey items. Prey consumption at Duk Point in

1987 consisted of primarily clams, then octopus (Bowlby *et al.* 1988). In the 2003 and 2004 study, the main prey items included sea stars and crabs, followed by clams, mussels, unknown bivalves, octopus, and sea cucumbers. Prey consumed at Cape Alava and Sand Point in 2003 and 2004 consisted of primarily crabs, then clams. In 1986 and 1987, Bowlby *et al.* (1988) found that while sea otters at Cape Alava consumed crabs and clams, as well as sea cucumbers, at Sand Point the prey items consumed were primarily octopus, then sea urchins.

Change in prey composition over time has been shown to be correlated with the overall length of time in which sea otters have occupied an area (Estes *et al.* 1978, Ostfeld 1982, Garshelis 1983). In Washington, sea otter abundance has been shown by Kvitek *et al.* (1989) to have a negative correlation with the distribution and abundance of invertebrate prey. For example, they found that in Washington, prey items were larger in size and more abundant outside of the primary range that sea otters inhabited. They also found that increases in kelp cover at Cape Alava have occurred since the reintroduction of sea otters back into Washington waters and that by 1988, sea urchins at Cape Alava were almost nonexistent (Kvitek *et al.* 1989). Only one sea urchin was noted during all foraging observations in all the locations of the current study. These observations support the findings by Kvitek *et al.* (1998) that increases in sea otter abundance were positively correlated with the abundance of algal species and the reduced presence of sea urchins (*Strongylocentrotus sp.*). Therefore it would be predicted that areas which have been occupied longer in Washington by sea otters, such as Sand Point and Cape Alava, or that have higher abundance counts such as Duk Point, would have fewer sea urchins.

Although dive durations are correlated with the depths to which sea otters are diving, variation among locations and between sexes have also been recorded (Garshelis 1983). Foraging dives have been found to have a mean duration ranging from 85 sec in Southeast Alaska (Bodkin *et al.* 2004), to 61 sec in Prince William Sound (Garshelis 1983), and 56.4 sec in California (Loughlin 1977). The longest dives ever recorded were 265 sec and 246 sec in California (Loughlin 1977, Riedman and Estes 1990), and 205 sec in Prince William Sound (Garshelis 1983).

In this study, dive durations ranged from 5.3 sec to 122.0 sec, with mean dive duration of 35.6 sec. There were differences between locations and these local differences probably relate to differences in prey availability. For example, Sand Point had a high dive success rate, the highest feeding activity, and the least amount of time spent foraging. Of the four locations, prey availability would be predicted to be highest at this location.

The current Washington sea otter population is still growing, but does not appear to be currently expanding its range, with the exception of a few sightings of single otters outside of the population range, south near Cape Arago and Cape Disappointment, OR, and north in Makah Bay, WA (Jameson and Jeffries 2004). Based on feeding activity for the four locations, food availability does not appear to be limited and it would seem that the Washington sea otter population has the potential to continue to grow.

There is concern that, in the event of a single oil spill or catastrophic event, the entire Washington population could be severely affected due to their restricted distribution along the coast. The *Exxon Valdez* oil spill of 1989 was a disastrous situation, with sea otters suffering the greatest mortality of all the marine mammals affected (Garshelis 1997).

Furthermore, the constant change in sea otter abundance observed during this study from site visit to site visit indicated a high degree of movement between locations. With this movement, there is a strong possibility of conflict with vessels transporting petroleum and other hazardous materials. Continuing to monitor distribution, prey consumption, foraging behaviors and movement will be of extreme importance for determining areas of suitable habitat and predicting future movements along the Washington coast.

Prey consumption data is of particular and timely significance because of the concerns surrounding current mortalities and contaminant findings. Contaminants such as butyltins, PCBs, and organochlorine pesticides, have been detected in sea otters in California (Kannan et al. 2004). Contaminant levels in Washington sea otter population are currently being investigated by the U.S. Fish and Wildlife Service and its partners. Recent necropsy findings in Washington have revealed the presence of acanthocephalan peritonitis, protozoal encephalitis (caused by the protozoal parasite T. gondii or S. neurona), and leptospirosis (Lance et al. 2004, Dubey et al. 2003). Sea otters captured in Washington in 2000 and 2001 have also shown positive titers to morbilliviruses (Lance et al. 2004). Industrial and agricultural compounds are making their way into sea otter tissues probably via trophic transfer and recent investigations into the possibility of protozoal parasites also being transferred via trophic levels are also being investigated (Lance et al. 2004), hence the importance of continuing to collect prey consumption data. The data collected in this study regarding current prey composition per location, and the changes over time, are being incorporated into current U.S. Fish and Wildlife Service toxicological and epidemiological studies which are attempting to determine direct correlations between prey consumption and the recent contaminant and mortality findings in the Washington sea otter population.

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Table 1. Activity percentages for 2003 and 2004 by age and location.

	No. of Animal				Activity %			
Location	Observations	Resting	Grooming	Feeding	Playing	Traveling	Nurse	Other
Cape Alava								
adults	1026	72.1	20.1	3.1	0.8	3.4	0	0.5
pups	119	73.1	6.7	4.2	3.4	5.0	7.6	0
Total	1145	72.2	18.7	3.2	1.1	3.6	0.8	0.4
Duk Point								
adults	3579	61.8	19.7	6.7	1.8	9.6	0	0.4
pups	240	75.4	5.4	4.2	2.5	4.2	7.9	0.4
Total	3819	62.7	18.8	6.5	1.9	9.2	0.5	0.4
Sand Point								
adults	1484	53.4	23.5	12.6	2.4	8.0	0	0.1
pups	168	74.4	5.9	6.6	0.6	4.2	8.3	0
Total	1652	55.6	21.7	12.0	2.3	7.6	0.8	0.01
Norwegian Memorial								
adults	429	55.5	24.0	11.7	0.9	5.6	0	2.3
pups	71	77.5	5.6	8.5	2.8	2.8	2.8	0
Total	500	58.6	21.4	11.2	1.2	5.2	0.4	2.0
All locations								
adults	6518	61.1	20.9	7.8	1.7	8.0	0	0.5
pups	598	74.9	5.8	5.4	2.1	4.2	7.4	0.2
Grand Total	7116	62.3	19.7	7.6	1.8	7.6	0.6	0.4

	Clam	Pacific Little- neck clam	Crab	Kelp crab	Octopus	Sea Star	Dawsons Sun Star	Sea Cucumber	Mussel	Snail	Unk. Bivalve	Red Sea Urchin
Duk Point	26 (14.7)	0	48 (27.1)	1 (0.6)	3 (1.7)	60 (33.9)	1 (0.6)	1 (0.6)	19 (10.7)	0	18 (10.2)	0
Cape Alava	14 (23.7)	0	38 (64.4)	0	1 (1.7)	4 (6.8)	0	0	0	0	1 (1.7)	1 (1.7)
Sand Point	21 (12.8)	0	103 (62.8)	1 (0.6)	4 (2.4)	1 (0.6)	0	0	10 (6.1)	2 (1.2)	22 (13.4)	0
Norwegian Memorial	16 (21.1)	33 (43.4)	18 (23.7)	5 (6.6)	0	0	0	0	0	1 (1.3)	3 (4.0)	0
Total	77 (16.2)	33 (6.9)	207 (43.5)	7 (1.5)	8 (1.7)	65 (13.7)	1 (0.2)	1 (0.2)	29 (6.1)	3 (0.6)	44 (9.2)	1 (0.2)

Table 2. Number of prey items consumed by location (total percentage of prey consumed), unknown items excluded.

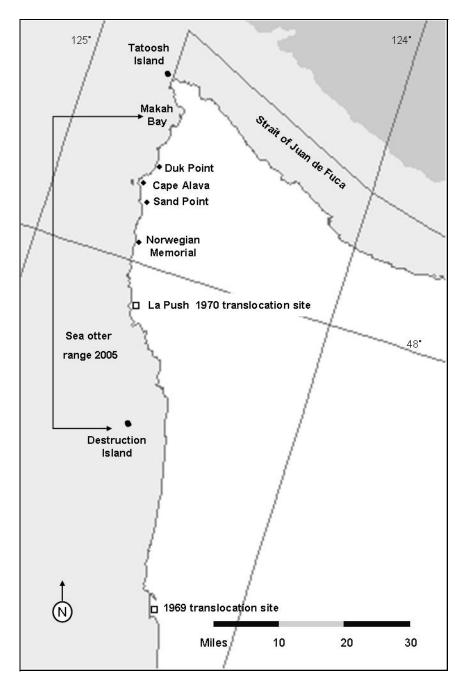


Figure 1. Study locations, current range, and release sites of the translocated Washington sea otter population. Map created using Map Maker: National Atlas of the United States, March 24, 2006, <u>http://nationalatlas.gov</u>.

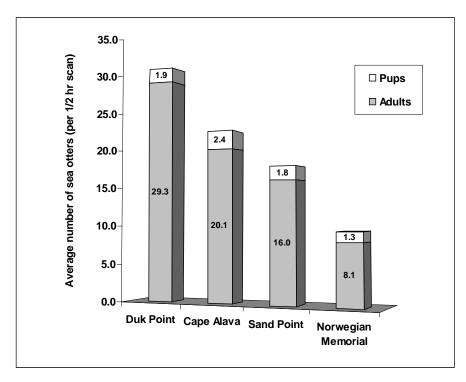


Figure 2. Average abundance of sea otters per ½ hour activity scan totaled over 2003 and 2004. All pairwise location comparisons were significantly difference from one another ( $p \le 0.01$ ) except for Cape Alava and Sand Point.

## **Appendix 2.** List of the Analytes Scanned in Whole Blood or Liver Samples of Washington Sea Otters

Metals Silver Aluminum Arsenic Boron Barium Barium Cadmium Chromium Copper Iron Mercury	Aliphatics n-decane n-docosane n-dodecane n-dotriacontane n-eicosane n-heneicosane n-hentriacontane n-heptacosane n-heptadecane n-hexacosane n-hexadecane	Organochlorines Aldrin alpha BHC alpha chlordane beta BHC cis-nonachlor delta BHC Dieldrin endosulfan II endrin gamma BHC gamma chlordane HCB
Magnesium Manganese Molybdenum Nickel Lead Selenium Strontium Vanadium	n-nonacosane n-nonadecane n-octacosane n-octadecane n-pentacosane n-pentadecane n-tetracosane n-tetradecane n-tetradecane n-tetratriacontane	Heptachlor heptachlor epoxide mirex o,p'-DDD o,p'-DDE o,p'-DDT oxychlordane p,p'-DDD p,p'-DDE
ButyltinsDibutyltinMonobutyltinTetrabutyltinTributyltinSemi-volatiles1,2,3,4-Tetrachlorobenzene1,2,4,5-Tetrachlorobenzene	n-triacontane n-tricosane n-tridecane n-tritriacontane n-undecane phytane pristane	p,p'-DDT PCB-TOTAL toxaphene trans-nonachlor pentachloro-anisole chlorpyrifos AROCLOR-1242 AROCLOR-1248 AROCLOR-1254 AROCLOR-1260 AROCLOR-1268

#### Polycyclic Aromatic Hydrocarbons

1,6,7-Trimethylnaphthalene 1-methylnaphthalene 1-methylphenanthrene 2,6-dimethylnaphthalene 2-methylnaphthalene acenaphthalene acenaphthene anthracene benzo(a)pyrene benzo(b)fluoranthene benzo(e)pyrene benzo(g,h,i)perylene benzo(k)fluoranthene biphenyl C1-chrysenes C1-dibenzothiophenes C1-Fluoranthenes & Pyrenes C1-fluorenes C1-naphthalenes C1-Phenanthrenes & Anthracenes C2-chrysenes C2-dibenzothiophenes C2-fluorenes C2-naphthalenes C2-Phenanthrenes & Anthracenes C3-chrysenes C3-dibenzothiophenes C3-fluorenes C3-naphthalenes C3-Phenanthrenes & Anthracenes C4-chrysenes C4-naphthalenes C4-Phenanthrenes & Anthracenes chrysene dibenzothiophene fluoranthene

fluorene indeno(1,2,3-cd)pyrene naphthalene perylene phenanthrene pyrene unresolved complex mixture Benzo(a)anthracene Dibenz(a,h)anthracene

Appendix 2
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	P
<u>PCBs</u>	P
PCB1	P
PCB7/9	P
PCB8/5	P
PCB15	P
PCB16/32	P
PCB18/17	P
PCB22/51	P
PCB24/27	P
PCB25	P
PCB26	P
PCB28	P
PCB29	P
PCB30	P
PCB31	P
PCB33/20	P
PCB39	P
PCB40	P
PCB41/64	P
PCB42/59/37	P
PCB44	P
PCB45	P
PCB46	P
PCB47/75	P
PCB48	P
PCB49	P
PCB52	P
PCB53	P
PCB60/56	P
PCB63	P
PCB66	P
PCB67	P
PCB69	P
PCB70	P
PCB72	P
PCB74/61	P
PCB77	P
PCB81	P
PCB82	P
PCB83	P
PCB84	P
PCB85	P
PCB87/115	P
PCB/55	P
PCB92	P

PCB95/80
PCB97
PCB99
PCB101/90
PCB105
PCB107
PCB110
PCB114
PCB118
PCB119
PCB126
PCB128
PCB129
PCB130
PCB135
PCB136
PCB138/160
PCB141/179
PCB146
PCB149/123
PCB151
PCB153/132
PCB156
PCB158
PCB166
PCB167
PCB169
PCB170/190
PCB171/202
PCB172
PCB174
PCB175
PCB176/137
PCB177
PCB178
PCB180
PCB183
PCB185
PCB187
PCB189
PCB191
PCB193
PCB194
PCB195/208
PCB196
PCB197

PCB199 PCB200 PCB201 PCB1/157/173 PCB203/196 PCB205 PCB206 PCB207 PCB209

## Appendix 3. Percent Lipid and Percent Moisture Used in Analyses Performed by GERG at Texas A&M University

Otter	Sample Volume (ml)	Percent Lipid
WA054	15	0.19
WA084	15	0.23
WA086	15	0.18
WA088	15	0.11
WA088	15	0.11
WA089	15	0.11
WA090	15	0.21
WA091	15	0.12
WA092	15	0.15
WA093	15	0.33
WA094	15	0.27
WA095	15	0.10
WA096	15	0.16
WA097	15	0.10
WA098	15	0.15
WA099	15	0.08
WA100	15	0.13
WA101	14	0.16
WA102	15	0.10
WA103	15	0.11
WA104	10	0.10
WA105	15	0.15
WA106	15	0.11
WA107	15	0.14
WA108	15	0.03
WA109	15	0.03
WA110	15	0.08
WA111	15	0.01
WA112	15	0.09
WA113	15	0.16
WA114	15	0.14
WA115	12	0.09

## Live Otters (sample matrix: blood)

Otter	Sample Weight (g)	Sample Volume (ml)	Percent Lipid	Percent Moisture
16904-01			2.0	73.7
16961-01			5.53	65.5
16961-02			2.96	73.7
16961-03			4.94	65.9
16961-04			3.7	67.8
17058-01			3.63	67.9
18124-01			1.92	71.5
17315-01	15		1.0	68.5
15713-01	15		2.4	64.1
14717	15		3.6	72.1
14325	15		2.9	69.7
13827	15		2.7	67.5
13555	15		4.8	69.7
10385	15		1.8	69.4
18316-01 (liver)	25		6.18	61.88
18316-01 (blood)		15	0.19	

Appendix 3 continued; Beach-Cast Otters (sample matrix: liver)

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## Appendix 4. Marine Mammal Scientific Research Permit

Appendix 5. Congener-Specific PCB Detection Limits and Concentrations Measured in	
Whole Blood Live-Captured Sea Otters (ppb, wet weight)	

Whole Di		c Cur	/ui cu	Sca Otters	( <b>PP</b> 2, "	ce weigi	10)				
Otter	Gender	Age	PCB1	PCB101/90	PCB105	PCB107	PCB110	PCB118	PCB128	PCB130	PCB135
WA115	F	<1	0.979		0.089					0.124	
WA099	F	3		0.163	0.050	0.053	0.051	0.157			0.070
WA109	F	3	1.625		0.017					0.157	
WA084	F	4		0.176	0.050		0.060	0.133	0.054		0.067
WA095	F	4		0.095		0.095		0.100			0.075
WA100	F	4		0.130							0.069
WA111	F	4	0.285		0.056					0.144	
WA094	F	5	0.047			0.053					0.066
WA086	F	5	0.049	0.142		0.052		0.078			0.073
WA089	F	6		0.081							0.070
WA106	F	6	1.045		0.007						0.011
WA114	F	6	0.815		0.008					0.069	
WA091	F	7		0.158				0.204			0.086
WA103	F	9	2.006		0.004						
WA090	F	9	0.079	0.136		0.066		0.076			0.073
WA096	F	10		0.078		0.069					0.076
WA093	F	10									0.035
WA088	F	13		0.081		0.050					0.066
WA054	F	15	0.700		0.028			0.088			
WA110	F	16	1.288		0.007					0.068	
WA104	М	<1	0.901	0.405	0.024	0.105		0.065			0.049
WA092	М	<1		0.164		0.090		0.091			0.093
WA113	М	2	1.216	0.209	0.077					0.083	
WA098	М	3		0.226	0.043	0.080	0.114	0.125			0.100
WA108	М	3	0.945		0.028					0.575	
WA105	М	5	0.721		0.010						
WA112	М	6	0.910		0.090	0.025				0.331	
WA107	М	6	0.774	0.356	0.030					0.106	
WA097	М	8		0.121				0.137			0.061
WA102	М	10	0.320		0.022			0.048			
Females	Mean		0.811	0.124	0.032	0.063	0.056	0.119	0.054	0.112	0.064
N = 20	Std dev		0.663	0.037	0.029	0.016	0.006	0.048		0.042	0.020
	Max		2.006	0.176	0.089	0.095	0.060	0.204	0.054	0.157	0.086
Males	Mean		0.827	0.247	0.040	0.075	0.114	0.093		0.274	0.076
N = 10	Std dev		0.273	0.111	0.029	0.035		0.038		0.230	0.024
	Max		1.216	0.405	0.090	0.105	0.114	0.137		0.575	0.100
All live	Mean		0.817	0.170	0.036	0.067	0.075	0.109	0.054	0.184	0.067
N = 30	Std dev		0.534	0.093	0.028	0.024	0.034	0.044		0.167	0.021
	Max		2.006	0.405	0.090	0.105	0.114	0.204	0.054	0.575	0.100
NA = Not or		lantra		ate values be	low dataat					•	

NA = Not analyzed; blank cells indicate values below detection limit

DIUUU L	ave-Cap	uiuiu	i Sea Otters	<u>, (hhnè u</u>	et weight),	continu	cu			
Otter	Gender	Age	PCB138/160	PCB146	PCB149/123	PCB151	PCB153/132	PCB156	PCB16/32	PCB170/190
WA115	F	<1	1.353				0.915			
WA099	F	3	0.718	0.093			0.611		0.065	0.148
WA109	F	3	1.623				1.062			
WA084	F	4	0.597	0.085	0.047		0.594		0.163	0.151
WA095	F	4	0.510	0.054			0.339	0.053	0.076	0.124
WA100	F	4	0.367				0.163			
WA111	F	4	1.509				0.911			
WA094	F	5	0.198				0.074		0.058	
WA086	F	5	0.406				0.382		0.120	0.109
WA089	F	6	0.260				0.100		0.055	0.072
WA106	F	6	0.868				0.426			
WA114	F	6	1.062				0.503			
WA091	F	7	0.592	0.068			0.663		0.086	0.165
WA103	F	9	1.041				0.622			
WA090	F	9	0.296				0.225		0.075	0.121
WA096	F	10	0.233				0.115		0.083	
WA093	F	10	0.186				0.075		0.081	0.057
WA088	F	13	0.227				0.099		0.141	0.084
WA054	F	15	0.920				0.627			
WA110	F	16	1.140				0.607			
WA104	М	<1	1.265				1.016			
WA092	М	<1	0.541		0.068		0.382		0.072	0.143
WA113	М	2	1.643				1.088			
WA098	М	3	0.561	0.080	0.199	0.051	0.511		0.049	0.152
WA108	М	3	1.945				1.475			
WA105	М	5	0.821				0.530			
WA112	М	6	2.145				0.658			
WA107	М	6	1.274				0.742			
WA097	М	8	0.426				0.205		0.072	0.063
WA102	М	10	0.893				0.664			
Females	Mean		0.705	0.075	0.047		0.456	0.053	0.091	0.115
N = 20	Std dev		0.460	0.017			0.306		0.035	0.037
	Max		1.623	0.093	0.047		1.062	0.053	0.163	0.165
Males	Mean		1.151	0.080	0.134	0.051	0.727		0.064	0.119
N = 10	Std dev		0.606		0.093		0.374		0.013	0.049
	Max		2.145	0.080	0.199	0.051	1.475		0.072	0.152
All live	Mean		0.854	0.076	0.105	0.051	0.546	0.053	0.085	0.116
N = 30	Std dev		0.546	0.015	0.082		0.349		0.033	0.038
	Max		2.145	0.093	0.199	0.051	1.475	0.053	0.163	0.165
NA - Not		blank	cells indicate				1		0.100	0.100

Appendix 5. Congener-Specific PCB Detection Limits and Concentrations Measured in Whole Blood Live-Captured Sea Otters (ppb, wet weight); continued

NA = Not analyzed; blank cells indicate values below detection limit

WHOIC L		VU-Ua	aptureu Sea	Ollers	ppn, wei	weight)	, continue	u			
Otter	Gender	Age	PCB171/202	PCB174	PCB175	PCB177	PCB18/17	PCB180	PCB183	PCB187	PCB194
WA115	F	<1			0.718			0.527		0.135	
WA099	F	3	0.065					0.457	0.071	0.129	
WA109	F	3			0.817			0.680		0.073	
WA084	F	4	0.050					0.453	0.088	0.171	
WA095	F	4			0.057			0.362	0.069	0.117	0.050
WA100	F	4			0.052			0.203		0.050	
WA111	F	4			0.618			0.457		0.139	
WA094	F	5						0.122			
WA086	F	5			0.049			0.336	0.065	0.088	0.048
WA089	F	6						0.201			
WA106	F	6			0.510			0.137			
WA114	F	6			0.555			0.247		0.020	
WA091	F	7			0.068			0.509	0.087	0.105	
WA103	F	9			0.803			0.276		0.043	
WA090	F	9						0.291		0.071	
WA096	F	10			0.050			0.146			
WA093	F	10						0.093	0.049		
WA088	F	13						0.133			
WA054	F	15			0.582			0.291			
WA110	F	16			0.650			0.277		0.046	
WA104	М	<1			0.585			0.591		0.178	
WA092	М	<1		0.064		0.061		0.360	0.071	0.114	
WA113	М	2			0.693			0.634		0.121	
WA098	М	3	0.072	0.046	0.037	0.066		0.322	0.077	0.178	
WA108	М	3			0.769			0.613		0.209	
WA105	М	5			0.632			0.237			
WA112	М	6			0.636			0.449		0.044	
WA107	М	6			0.629		0.473	0.532		0.121	
WA097	М	8			0.055			0.228			
WA102	М	10			0.555			0.297		0.068	
Females	Mean		0.058		0.425			0.310	0.072	0.091	0.049
N = 20	Std dev		0.011		0.317			0.161	0.015	0.045	0.001
	Max		0.065		0.817			0.680	0.088	0.171	0.050
Males	Mean		0.072	0.055	0.510	0.064	0.473	0.426	0.074	0.129	
N = 10	Std dev			0.013	0.270	0.004		0.158	0.004	0.057	
	Max		0.072	0.064	0.769	0.066	0.473	0.634	0.077	0.209	
All live	Mean		0.062	0.055	0.460	0.064	0.473	0.349	0.077	0.106	0.049
N = 30	Std dev		0.002	0.033	0.295	0.004	0.175	0.167	0.012	0.052	0.04)
1, 50	Max		0.072	0.013	0.275	0.066	0.473	0.680	0.013	0.002	0.001
	IVIAX		0.072	0.004	0.01/	0.000	0.473	0.000	0.000	0.209	0.030

## Appendix 5. Congener-Specific PCB Detection Limits and Concentrations Measured in Whole Blood Live-Captured Sea Otters (ppb, wet weight) ; continued

NA = Not analyzed; blank cells indicate values below detection limit

	ive Cup	uicu		crs (ppo		ni), conti	nucu				
Otter	Gender	Age	PCB196	PCB199	PCB22/51	PCB24/27	PCB28	PCB30	PCB31	PCB33/20	PCB41/64
WA115	F	<1					0.267				
WA099	F	3	0.066	0.092			0.151		0.065		
WA109	F	3			0.258		0.302			0.129	
WA084	F	4	0.076	0.059		0.072	0.054				
WA095	F	4	0.072	0.078			0.095				
WA100	F	4					0.095		0.053		
WA111	F	4			0.133		0.206				
WA094	F	5					0.070				
WA086	F	5	0.067	0.090			0.065				
WA089	F	6					0.070				
WA106	F	6					0.190	0.241			
WA114	F	6					0.149				
WA091	F	7	0.076	0.085			0.090				
WA103	F	9			0.265		0.334				
WA090	F	9		0.059			0.063				
WA096	F	10					0.103				
WA093	F	10				0.054	0.048				0.042
WA088	F	13					0.066				
WA054	F	15					0.115				
WA110	F	16			0.297		0.286				
WA104	М	<1			0.163		0.174			0.172	
WA092	М	<1	0.064	0.094			0.117				
WA113	М	2					0.170				
WA098	М	3	0.046	0.061			0.079				0.040
WA108	М	3			0.244		0.352			0.298	
WA105	М	5					0.294				
WA112	М	6			0.119		0.156				
WA107	М	6			0.169		0.216	0.876		0.500	
WA097	М	8					0.134				
WA102	М	10					0.125				
Females	Mean		0.071	0.077	0.238	0.063	0.141	0.241	0.059	0.129	0.042
N = 20	Std dev		0.005	0.015	0.072	0.013	0.092		0.008		
	Max		0.076	0.092	0.297	0.072	0.334	0.241	0.065	0.129	0.042
Males	Mean		0.055	0.078	0.174		0.182	0.876		0.323	0.040
N = 10	Std dev		0.013	0.023	0.052		0.084			0.166	
	Max		0.064	0.094	0.244		0.352	0.876		0.500	0.040
All live	Mean		0.067	0.077	0.206	0.063	0.155	0.558	0.059	0.275	0.041
N = 30	Std dev		0.007	0.017	0.068	0.003	0.090	0.330	0.008	0.167	0.041
	Max		0.010	0.015	0.000	0.072	0.352	0.876	0.065	0.500	0.001
NIA – Not		hlant			elow detecti		0.354	0.070	0.005	0.500	0.072

Appendix 5. Congener-Specific PCB Detection Limits and Concentrations Measured in Whole Blood Live-Captured Sea Otters (ppb, wet weight) ; continued

NA = Not analyzed; blank cells indicate values below detection limit

TT HOLE I	noou Li		ipiul cu Dea	Otters (	<u>. pp.,</u>		, com	innaca			
Otter	Gender	Age	PCB42/59/37	PCB44	PCB45	PCB49	PCB52	PCB60/56	PCB69	PCB7/9	PCB8/5
WA115	F	<1		3.289		0.330					0.339
WA099	F	3	0.223					0.091			
WA109	F	3		3.535		0.595				0.781	0.355
WA084	F	4	0.086				0.084	0.107		0.066	
WA095	F	4	0.074								
WA100	F	4	0.144					0.080			
WA111	F	4		2.691		0.328					0.257
WA094	F	5	0.043					0.075		0.155	
WA086	F	5	0.048				0.050	0.092		0.049	
WA089	F	6	0.051					0.079			
WA106	F	6		2.277		0.233					0.240
WA114	F	6		2.471		0.294			0.551		0.406
WA091	F	7	0.109					0.137			
WA103	F	9		3.480		0.678					0.429
WA090	F	9	0.071					0.096			0.052
WA096	F	10	0.082					0.089			
WA093	F	10	0.050					0.068		0.049	
WA088	F	13	0.142					0.089			
WA054	F	15		2.553		0.260				1.806	0.209
WA110	F	16		2.835		0.345				0.243	0.427
WA104	М	<1		2.546		0.318				1.085	0.592
WA092	М	<1	0.077					0.103			
WA113	М	2	0.096	3.005		0.293				0.208	0.425
WA098	М	3	0.039				0.081	0.054		0.059	
WA108	М	3		3.344		0.318	0.994				0.254
WA105	М	5	0.140	2.787	0.236	0.291					0.252
WA112	М	6	0.117	2.643		0.300				0.462	0.412
WA107	М	6		2.665		0.329					0.322
WA097	М	8	0.054					0.100			
WA102	М	10		2.500		0.269					0.214
Females	Mean		0.094	2.892		0.383	0.067	0.091	0.551	0.450	0.302
N = 20	Std dev		0.053	0.483		0.162	0.024	0.019		0.652	0.125
	Max		0.223	3.535		0.678	0.084	0.137	0.551	1.806	0.429
Males	Mean		0.087	2.784	0.236	0.303	0.538	0.086		0.453	0.353
N = 10	Std dev		0.038	0.298		0.021	0.646	0.027		0.453	0.133
	Max		0.140	3.344	0.236	0.329	0.994	0.103		1.085	0.592
All live	Mean		0.091	2.841	0.236	0.345	0.302	0.090	0.551	0.451	0.324
N = 30	Std dev		0.048	0.397		0.123	0.462	0.020		0.562	0.127
	Max		0.223	3.535	0.236	0.678	0.994	0.137	0.551	1.806	0.592
			0,220	0.000	0.200	0.070	0.774	0.107	0.001	1.000	

## Appendix 5. Congener-Specific PCB Detection Limits and Concentrations Measured in Whole Blood Live-Captured Sea Otters (ppb, wet weight) ; continued

NA = Not analyzed; blank cells indicate values below detection limit

	UIC DI	Ju Live-	Capiarca		hhn, act	weight),
Gender	Age	PCB84	PCB87/115	PCB92	PCB95/80	PCB99
F	<1					
F	3					0.103
F	3					
F	4		0.048		0.150	0.084
F	4					0.050
F	4					
F	4					
F	5				0.052	
F	5				0.075	0.053
F	6	0.053			0.057	
F	6					
F	6					
F	7				0.068	0.090
F	9					
F	9				0.064	
F	10					
F	10				0.057	
F	13				0.068	
F	15					
F	16					
М	<1					
М	<1				0.069	
М	2					
М	3		0.037	0.047	0.083	0.093
М	3					
М	5					
М	6					
М	6					
М	8					
М	10					
Mean		0.053	0.048		0.074	0.076
Std dev					0.032	0.023
Max		0.053	0.048		0.150	0.103
			0.037	0.047	0.076	0.093
Std dev						
			0.037	0.047		0.093
		0.053	0.043	0.047	0.074	0.079
Ivrean				5.5.1		
Mean Std dev			0.008		0.028	0.022
	Gender  F G  F  F  F  F  F  F  F  F  F  F  F	Gender     Age       F     <1	GenderAgePCB84F<1	GenderAgePCB84PCB87/115F<1	GenderAgePCB84PCB87/115PCB92F<1	F $<1$ $<1$ $<1$ $<1$ F       3 $<1$ $<1$ $<1$ F       4 $<0.048$ $<0.150$ F       4 $<0.048$ $<0.150$ F       4 $<0.048$ $<0.150$ F       4 $<0.048$ $<0.150$ F       4 $<0.048$ $<0.050$ F       5 $<0.053$ $<0.052$ F       6 $0.053$ $<0.057$ F       6 $0.053$ $<0.057$ F       6 $0.053$ $<0.057$ F       6 $0.053$ $<0.058$ F       9 $<0.053$ $<0.068$ F       9 $<0.051$ $<0.068$ F       9 $<0.051$ $<0.057$ F       10 $<0.051$ $<0.057$ F       10 $<0.051$ $<0.053$ F       10 $<0.051$ $<0.057$ F       16 $<0.0101$ $<0.068$ F       16 $<0.037$ $<0.047$

Appendix 5. Congener-Specific PCB Detection Limits and Concentrations Measured in Whole Blood Live-Captured Sea Otters (ppb, wet weight); continued

NA = Not analyzed; blank cells indicate values below detection limit

Otter	Gender	Age	PCB1	PCB101/90	PCB105	PCB107	PCB110	PCB118	PCB119	PCB128	PCB130	PCB135	PCB136	PCB138/160	PCB141/179	PCB146
13827	F	U	506.0	47.40	3.32	2.71	0.94	3.88		0.91	0.40			10.00	0.56	3.30
14325	F	U	77.3	8.94			0.27	6.42	2.33	3.49	1.76			37.20	3.35	12.20
17315-01	F	13	109.0	14.30	25.00	1.52	4.14	11.90		5.37	1.74			40.70	1.44	11.50
18316-01	М	1	0.5	4.29	0.67			2.70		1.42	1.12		0.21	10.98		3.32
10385	М	U	105.0	27.20	3.06	3.12	1.16	3.44		1.02				13.40	0.47	2.54
13555	М	U	125.0	29.60	17.00		15.40	35.20	1.51	12.40	4.26	13.70		76.40	5.20	18.40
14717	М	U	75.2	5.02	5.09	1.13		8.08		2.60	0.54			17.30	0.52	5.23
15713-01	М	8	75.8	28.40	40.70	1.92	5.24	27.10		16.90	6.50			104.00	4.64	20.90
Females	Mean		230.8	23.55	14.16	2.12	1.78	7.40	2.33	3.26	1.30			29.30	1.78	9.00
N = 3	Std dev		238.9	20.83	15.33	0.84	2.07	4.10		2.24	0.78			16.81	1.43	4.95
	Max		506.0	47.40	25.00	2.71	4.14	11.90	2.33	5.37	1.76			40.70	3.35	12.20
	Min		77.3	8.94	3.32	1.52	0.27	3.88	2.33	0.91	0.40			10.00	0.56	3.30
Males	Mean		76.3	18.90	13.30	2.06	7.27	15.30	1.51	6.87	3.11	13.70	0.21	44.42	2.71	10.08
N = 5	Std dev		47.3	13.04	16.56	1.00	7.33	14.89		7.30	2.79			42.98	2.57	8.84
	Max		125.0	29.60	40.70	3.12	15.40	35.20	1.51	16.90	6.50	13.70	0.21	104.00	5.20	20.90
	Min		0.5	4.29	0.67	1.13	1.16	2.70	1.51	1.02	0.54	13.70	0.21	10.98	0.47	2.54
All live	Mean		134.2	20.64	13.55	2.08	4.53	12.34	1.92	5.51	2.33	13.70	0.21	38.75	2.31	9.67
N = 8	Std dev		154.8	15.06	14.90	0.82	5.68	12.17	0.58	5.95	2.24			34.60	2.05	7.21
	Max		506.0	47.40	40.70	3.12	15.40	35.20	2.33	16.90	6.50	13.70	0.21	104.00	5.20	20.90
	Min		0.5	4.29	0.67	1.13	0.27	2.70	1.51	0.91	0.40	13.70	0.21	10.00	0.47	2.54

# Appendix 6. Congener-Specific PCB Detection Limits and Concentrations Measured in Liver of Beach-Cast Sea Otters (ppb, wet weight)

Otter	Gender	Age	PCB149/123	PCB151	PCB153/132	PCB156	PCB158	PCB16/32	PCB166	PCB167	PCB170/190	PCB171/202	PCB172	PCB174	PCB175
13827	F	U	1.92		9.98			2.20		0.37	5.35	0.70		0.25	7.24
14325	F	U	0.44	3.15	50.50		3.18	1.32		1.08	16.20	9.53	3.57	4.29	2.95
17315-01	F	13	5.74	3.14	25.90		2.91			0.68	10.90	7.07	2.06	0.84	3.45
18316-01	М	1	0.59	0.40	9.94	1.05	1.20	1.38		0.56	3.35	1.45		0.16	2.74
10385	М	U	2.62		15.50			1.98			6.10	1.66	0.72	0.65	5.26
13555	М	U	34.10	8.32	70.30		4.42	0.29		2.38	14.00	18.00	2.46	9.50	5.36
14717	М	U	0.32	0.41	5.43		1.85	0.98			4.03	2.34	0.80	0.28	1.94
15713-01	М	8	8.71	3.44	45.20		7.28		7.51	4.35	23.50	18.90	4.59	2.84	
Females	Mean		2.70	3.15	28.79		3.05	1.76		0.71	10.82	5.77	2.82	1.79	4.55
N = 3	Std dev		2.73	0.01	20.41		0.19	0.62		0.36	5.43	4.56	1.07	2.18	2.35
	Max		5.74	3.15	50.50		3.18	2.20		1.08	16.20	9.53	3.57	4.29	7.24
	Min		0.44	3.14	9.98		2.91	1.32		0.37	5.35	0.70	2.06	0.25	2.95
Males	Mean		9.27	3.14	29.27	1.05	3.69	1.16	7.51	2.43	10.20	8.47	2.14	2.69	3.82
N = 5	Std dev		14.29	3.74	27.70		2.77	0.71		1.90	8.56	9.12	1.82	3.96	1.75
	Max		34.10	8.32	70.30	1.05	7.28	1.98	7.51	4.35	23.50	18.90	4.59	9.50	5.36
	Min		0.32	0.40	5.43	1.05	1.20	0.29	7.51	0.56	3.35	1.45	0.72	0.16	1.94
All live	Mean		6.80	3.14	29.09	1.05	3.47	1.36	7.51	1.57	10.43	7.46	2.37	2.35	4.13
N = 8	Std dev		11.42	2.90	23.61		2.17	0.69		1.54	7.10	7.45	1.53	3.25	1.87
	Max		34.10	8.32	70.30	1.05	7.28	2.20	7.51	4.35	23.50	18.90	4.59	9.50	7.24
	Min		0.32	0.40	5.43	1.05	1.20	0.29	7.51	0.37	3.35	0.70	0.72	0.16	1.94

Appendix 6. Congener-Specific PCB Detection Limits and Concentrations Measured in Liver of Beach-Cast Sea Otters (ppb, wet weight); continued

Appendix 6. Congener-Specific PCB Detection Limits and Concentrations Measured in Liver of Beach-Cast Sea Otters (ppb, wet
weight); continued

Otter	Gender	Age	PCB176/137	PCB177	PCB178	PCB18/17	PCB180	PCB183	PCB187	PCB191	PCB193	PCB194	PCB195/208	PCB196	PCB197	PCB199
13827	F	U		1.10		30.40	8.72	23.50	11.90	0.51	0.49	1.46	0.82	2.81	0.48	3.66
14325	F	U		8.13	4.41	0.44	46.90	36.60	26.80	2.56	2.73	9.71	4.72	12.80	0.40	13.50
17315-01	F	13		4.79	3.21		31.30	17.20	22.70	0.69	1.53	2.96	1.27	4.52	0.57	5.41
18316-01	М	1	1.70	0.80	1.35	0.49	5.61	2.82	5.55		0.32	0.94	0.33		0.64	2.47
10385	М	U		1.39	1.68	21.10	13.20	12.60	15.30			2.36	0.76	3.99		5.13
13555	М	U	6.74	11.80	6.54	0.61	45.40	15.60	37.40	0.59	1.13	6.39	1.97	10.50	0.68	8.76
14717	М	U		0.78	0.20	1.04	12.80	21.80	3.16	0.30	0.19	0.69	0.45	1.78	0.61	1.75
15713-01	М	8		5.67	4.98		83.00	36.00	34.80	1.17	1.89	8.43	5.80	17.50	1.83	13.80
Females	Mean			4.67	3.81	15.42	28.97	25.77	20.47	1.25	1.58	4.71	2.27	6.71	0.48	7.52
N = 3	Std dev			3.52	0.85	21.18	19.20	9.90	7.70	1.13	1.12	4.39	2.13	5.34	0.09	5.25
	Max			8.13	4.41	30.40	46.90	36.60	26.80	2.56	2.73	9.71	4.72	12.80	0.57	13.50
	Min			1.10	3.21	0.44	8.72	17.20	11.90	0.51	0.49	1.46	0.82	2.81	0.40	3.66
Males	Mean		4.22	4.09	2.95	5.81	32.00	17.76	19.24	0.69	0.88	3.76	1.86	8.44	0.94	6.38
N = 5	Std dev		3.56	4.77	2.68	10.20	32.40	12.28	16.07	0.44	0.79	3.47	2.29	7.08	0.59	4.97
	Max		6.74	11.80	6.54	21.10	83.00	36.00	37.40	1.17	1.89	8.43	5.80	17.50	1.83	13.80
	Min		1.70	0.78	0.20	0.49	5.61	2.82	3.16	0.30	0.19	0.69	0.33	1.78	0.61	1.75
All live	Mean		4.22	4.31	3.20	9.01	30.87	20.76	19.70	0.97	1.18	4.12	2.02	7.70	0.75	6.81
N = 8	Std dev		3.56	4.08	2.25	13.30	26.60	11.46	12.84	0.83	0.93	3.55	2.09	5.95	0.49	4.73
	Max		6.74	11.80	6.54	30.40	83.00	36.60	37.40	2.56	2.73	9.71	5.80	17.50	1.83	13.80
	Min		1.70	0.78	0.20	0.44	5.61	2.82	3.16	0.30	0.19	0.69	0.33	1.78	0.40	1.75

Otter	Gender	Age	PCB200	PCB201	PCB203/196	PCB205	PCB206	PCB207	PCB209	PCB22/51	PCB24/27	PCB25	PCB26	PCB28	PCB29	PCB33/20
13827	F	U		0.45			0.55		0.47		5.59		3.96	24.30		21.40
14325	F	U	0.54	0.70		1.29	6.96		3.72				0.19	0.90		0.20
17315-01	F	13					3.58		1.92							
18316-01	М	1			1.66					1.15	0.24	0.79		0.35	0.48	
10385	М	U					1.14							7.87		0.83
13555	М	U		2.27		0.70	4.99		1.13					0.62		
14717	М	U					1.30	0.24	1.30				0.72	1.18		
15713-01	М	8					8.11	0.89	8.26							
Females	Mean		0.54	0.57		1.29	3.70		2.04		5.59		2.08	12.60		10.80
N = 3	Std dev			0.17			3.21		1.63				2.66	16.54		14.99
	Max		0.54	0.70		1.29	6.96		3.72		5.59		3.96	24.30		21.40
	Min		0.54	0.45		1.29	0.55		0.47		5.59		0.19	0.90		0.20
Males	Mean			2.27	1.66	0.70	3.89	0.56	3.56	1.15	0.24	0.79	0.72	2.51	0.48	0.83
N = 5	Std dev						3.33	0.46	4.07					3.59		
	Max			2.27	1.66	0.70	8.11	0.89	8.26	1.15	0.24	0.79	0.72	7.87	0.48	0.83
	Min			2.27	1.66	0.70	1.14	0.24	1.13	1.15	0.24	0.79	0.72	0.35	0.48	0.83
All live	Mean		0.54	1.14	1.66	1.00	3.80	0.56	2.80	1.15	2.92	0.79	1.63	5.87	0.48	7.48
N = 8	Std dev			0.99		0.42	3.00	0.46	2.89		3.78		2.04	9.47		12.06
	Max		0.54	2.27	1.66	1.29	8.11	0.89	8.26	1.15	5.59	0.79	3.96	24.30	0.48	21.40
	Min		0.54	0.45	1.66	0.70	0.55	0.24	0.47	1.15	0.24	0.79	0.19	0.35	0.48	0.20

Appendix 6. Congener-Specific PCB Detection Limits and Concentrations Measured in Liver of Beach-Cast Sea Otters (ppb, wet weight); continued

Appendix	x 6. Con	igene	r-Specif	ic PCB	Detection	Limits and	Concent	trations	Measu	red in Liv	er of Be	ach-Ca	st Sea Of	tters (pp	ob, wet
weight);	continue	ed													

Otter	Gender	Age	PCB39	PCB40	PCB41/64	PCB42/59/37	PCB44	PCB45	PCB46	PCB47/75	PCB48	PCB49	PCB52	PCB53	PCB60/56	PCB63
13827	F	U		6.76	3.88	31.20			0.37			1.69	8.32		4.23	12.10
14325	F	U			1.94							1.05	6.48		2.94	
17315-01	F	13		0.53	2.54			0.99			1.06	2.69	7.04		2.91	
18316-01	М	1	1.23			0.35			0.23			1.65	2.97	1.72		
10385	М	U		3.82	5.00	15.90			0.55				5.64		5.66	2.14
13555	М	U		1.44	2.90	0.96	3.25			3.81		2.53	11.00		4.81	0.38
14717	М	U		4.74	2.12						0.20	0.51	5.47		3.90	
15713-01	М	8		1.14	3.66						8.34	9.08	13.80		2.84	
Females	Mean			3.64	2.79	31.20		0.99	0.37		1.06	1.81	7.28		3.36	12.10
N = 3	Std dev			4.41	0.99							0.83	0.94		0.75	
	Max			6.76	3.88	31.20		0.99	0.37		1.06	2.69	8.32		4.23	12.10
	Min			0.53	1.94	31.20		0.99	0.37		1.06	1.05	6.48		2.91	12.10
Males	Mean		1.23	2.79	3.42	5.74	3.25		0.39	3.81	4.27	3.44	7.78	1.72	4.30	1.26
N = 5	Std dev			1.77	1.23	8.81			0.23		5.75	3.85	4.46		1.21	1.25
	Max		1.23	4.74	5.00	15.90	3.25		0.55	3.81	8.34	9.08	13.80	1.72	5.66	2.14
	Min		1.23	1.14	2.12	0.35	3.25		0.23	3.81	0.20	0.51	2.97	1.72	2.84	0.38
All live	Mean		1.23	3.07	3.15	12.10	3.25	0.99	0.38	3.81	3.20	2.74	7.59	1.72	3.90	4.87
N = 8	Std dev			2.44	1.09	14.62			0.16		4.47	2.90	3.42		1.08	6.32
	Max		1.23	6.76	5.00	31.20	3.25	0.99	0.55	3.81	8.34	9.08	13.80	1.72	5.66	12.10
	Min		1.23	0.53	1.94	0.35	3.25	0.99	0.23	3.81	0.20	0.51	2.97	1.72	2.84	0.38

Otter	Gender	Age	PCB66	PCB67	PCB69	PCB72	PCB74/61	PCB8/5	PCB82	PCB84	PCB85	PCB87/115	PCB92	PCB95/80	PCB97	PCB99
13827	F	U	45.60	9.02		3.25	8.85	66.60	5.54			0.74	5.78		5.71	8.32
14325	F	U	6.86	9.19	0.85		1.79	21.90				1.12	5.59			9.41
17315-01	F	13	2.93				1.28	8.90				2.51	8.96	2.87	1.39	8.48
18316-01	М	1	1.47			334.23	0.66	1.07	0.29		0.81	0.92	0.81			3.67
10385	М	U	21.90	3.85		3.86	3.14	27.70	4.38			1.37	5.58		4.53	6.02
13555	М	U	4.08	16.00			3.02	0.53	7.36	1.40		7.64	21.10	9.67	7.51	21.60
14717	М	U	14.00	11.60			0.79	18.40				1.31	2.36			5.51
15713-01	М	8	15.30	3.74		0.33	1.86	17.10				6.99	15.00	5.54	2.55	21.40
Females	Mean		18.46	9.11	0.85	3.25	3.97	32.47	5.54			1.46	6.78	2.87	3.55	8.74
N = 3	Std dev		23.58	0.12			4.23	30.27				0.93	1.89		3.05	0.59
	Max		45.60	9.19	0.85	3.25	8.85	66.60	5.54			2.51	8.96	2.87	5.71	9.41
	Min		2.93	9.02	0.85	3.25	1.28	8.90	5.54			0.74	5.59	2.87	1.39	8.32
Males	Mean		11.35	8.80		112.81	1.89	12.96	4.01	1.40	0.81	3.65	8.97	7.61	4.86	11.64
N = 5	Std dev		8.43	6.05		191.77	1.18	11.83	3.55			3.36	8.74	2.92	2.50	9.04
	Max		21.90	16.00		334.23	3.14	27.70	7.36	1.40	0.81	7.64	21.10	9.67	7.51	21.60
	Min		1.47	3.74		0.33	0.66	0.53	0.29	1.40	0.81	0.92	0.81	5.54	2.55	3.67
All live	Mean		14.02	8.90	0.85	85.42	2.67	20.27	4.39	1.40	0.81	2.83	8.15	6.03	4.34	10.55
N = 8	Std dev		14.60	4.69		165.88	2.66	21.06	3.00			2.83	6.78	3.43	2.44	7.01
	Max		45.60	16.00	0.85	334.23	8.85	66.60	7.36	1.40	0.81	7.64	21.10	9.67	7.51	21.60
	Min		1.47	3.74	0.85	0.33	0.66	0.53	0.29	1.40	0.81	0.74	0.81	2.87	1.39	3.67

Appendix 6. Congener-Specific PCB Detection Limits and Concentrations Measured in Liver of Beach-Cast Sea Otters (ppb, wet weight); continued

Appendix 7a

### Appendix 7. Polycyclic Aromatic Hydrocarbons and Organochlorines in Invertebrate Prey Species

## 7a. Detectable Polycyclic Aromatic Hydrocarbons Measured in Mussels from the NOAA Status and Trends, Mussel Watch Program Cape Flattery Site in 2002 (ppb, dry weight)

Analyte	Concentration in <i>Mytilus edulis</i> 2002				
1,6,7-Trimethylnaphthalene	1.2				
1-Methylnaphthalene	3.9				
1-Methylphenanthrene	0.8				
2,6-Dimethylnaphthalene	2.8				
2-Methylnaphthalene	12.2				
Acenaphthene	0.6				
Anthracene	0.2				
Biphenyl	5.9				
C1-Naphthalenes	10.5				
C1-Phenanthrenes/Anthracenes	3.8				
C2-Fluoranthenes/Pyrenes	0.7				
C2-Naphthalenes	8.5				
C2-Phenanthrenes/Anthracenes	4.2				
C3-Naphthalenes	4.5				
C4-Naphthalenes	2.6				
Chrysene	1.7				
Dibenzothiophene	0.5				
Fluoranthene	1.5				
Fluorene	1.5				
Naphthalene	9.9				
Phenanthrene	7.1				
Pyrene	0.7				

PAHs that were not detected: Acenaphthylene, Benzo[a]pyrene, Benzo[b]fluoranthene, Benzo[e]pyrene, Benzo[k]fluoranthene, C1-Chrysenes, C1-Fluoranthenes/Pyrenes, C1-Fluorenes, C2-Chrysenes, C3-Chrysenes, C3-Fluorenes, C3-Phenanthrenes/Anthracenes, C4-Chrysenes, C4-Phenanthrenes/Anthracenes, Dibenzo[a,h]anthracene, Perylene

(ppp, ary v	weign																										
Year and Species	alpha chlordane	alpha BHC	beta BHC	cis-nonachlor	Dieldrin	Endosulfan II	Endrin	gamma chlordane	heptachlor-epoxide	Hexachlor	Mirex	0,p'-DDD	o,p'-DDE	o,p'-DDT	oxychlordane	Pentachloro-anisole	p,p'-DDD	p,p'-DDE	p,p'-DDT	Trans-Nonachlor	Total chlordanes	Total DDT	Total dieldrin	Total HCH	Total PCB	1,2,3,4- Tetrachlorobenzene	1,2,4,5- Tetrachlorobenzene
2000 Mytilus californianus		2.471	1.483		2.842			0.618	0.865	0.371		0.124		0.124	0.247	1.606	0.988	4.695	0.247	0.618	8.155	6.178	2.842	4.571	10.872	0.124	3.212
2002 Mytilus edulis	6.298	2.203	1.807	0.111	3.44	0.186	1.039	0.78	0.569	0.297	0.099	0.393	0.495	0.334		2.71	0.978	5.123	0.161	0.445	7.312	7.484	3.44	4.863	8.566	0.136	0

## 7b. Organochlorines in Mussels from NOAA Status and Trends Mussel Watch Program, Cape Flattery Site in 2000 and 2002 (ppb, dry weight)

## Appendix 8. List of Acronyms/Abbreviations

4PT – tetraprylin AAS – atomic absorption spectrometry ACF – Analytical Control Facility Ag – silver Al – aluminum AMU – atomic mass unit ANOVA - analysis of variance As – arsenic B – boron Ba – barium Be – beryllium BHC – benzene hexachloride BT – butyltins BUN - blood urea nitrogen C – Celsius CBC - cell blood counts Cd – cadmium CDV – Canine Distemper Virus CGC – capillary gas chromatography CHL – chlordanes  $CO_2$  – carbon dioxide Cr – chromium Cu – copper CVAAS – cold vapor atomic absorption spectroscopy DDD - dichlorodiphenyldichloroethane DDE - dichlorodiphenylethylene DDTs -dichloro-diphenyl-trichloroethane (1,1,1,-trichloro-2,2-bis(p-chlorophyenyl)ethanes DMV – Dolphin Morbillivirus EPA – Environmental Protection Agency Fe – iron FWS – Fish and Wildlife Service (United States) g – gram GERG - Geochemical and Environmental Research Group HCB – hexachlorobenzene HCH - cyclohexanes Hg – mercury HNO<sub>3</sub> – nitric acid HPLC – high performance liquid chromatography ICP – inductively coupled plasma optical emission spectroscopy ICP-MS – coupled plasma-mass spectroscopy Kg – kilograms M - molar m:z – mass charge ratio MAT – modified agglutination test Mg – magnesium

ml - milliliter Mn – manganese Mo-molybdenum NA - no sample available Na<sub>2</sub>SO<sub>4</sub> - sodium sulfate Narcan - Nalazone hydrochloride NAT - Neospora Agglutination Test ng - nanograms Ni – nickel nm - nanometer NMFS - Northwest Fisheries Science Center NOAA – National Oceanographic and Atmospheric Administration NSQ – Not sufficient quantity OCNMS - Olympic Coast national Marine Sanctuary OCs - Organochlorines oz. – ounce PAH – polycyclic aromatic hydrocarbons PBMC – Peripheral Blood Mononuclear Cell PIT – passive integrated transponder tag Pb-lead PCB – polychlorinated biphenyls PCDD – polychlorinated dibenzo-p-dioxins PCDF - polychlorinated dibenzofurans PDV – Phocine Distemper Virus PMV - Porpoise Morbillivirus ppb – parts per billion ppm – parts per million QA/QC – quality assurance quality control R- all-trans retinol RP – all-trans retinyl palmitate Se – selenium SIM - p3Sn – Tin Sr – strontium T3 – triiodothyronine T4 – tetraiodothyronine/thyroxine TBT - tributyltin TERL – Trace Element Research Laboratory TPH – total petroleum hydrocarbons TPT – tripropyltin TR - total retinol ([R]+0.55\*[RP])USFWS – United States Fish and Wildlife Service USGS – United States Geological Survey V – vanadium WBC – white blood cells WDFW - Washington Department of Fish and Wildlife wt-weight

## Appendix 9. Analyses and Results Pending and Samples Archived

## Genetics

Tissue plugs leftover from the tagging procedures were preserved and transferred to the Seattle Aquarium so that genetic analysis can be conducted on the captured otters. Results are pending.

### Fur and Whiskers

Other samples, like fur and whiskers were taken and archived in the event that they would be useful to other researchers or were needed for further analysis under this proposal.

### Serology

Sarcocystis results are pending for seven of the live captured sea otters.

### **Peripheral Blood Mononuclear Cells (PBMC)**

PBMC results are pending for the 2002 live captured sea otters.

Contaminant Group	Contaminant	Transformation	Shapiro-Wilk Significance
Gloup	Aluminum (Al)	Rankit	0.99
	Arsenic (As)	Rankit	0.99
	Boron (B)	Rankit	0.99
	Barium (Ba)	Rankit	0.99
Ie	Cadmium (Cd)	Square root	0.157
iptu	Copper (Cu)	None	0.05
Metals live capture	Iron (Fe)	ln	0.236
s liv	Mercury (Hg)	none	0.595
etals	Magnesium (Mg)	Square root	0.445
We	Lead (Pb)	ln	0.313
	Selenium (Se)	None	0.261
	Strontium (Sr)	ln	0.243
	Vanadium (V)	ln	0.087
	Zinc (Zn)	Square root	0.99
	Aluminum (Al)	ln	0.903
	Arsenic (As)	None	0.573
ght	Cadmium (Cd)	Square root	0.054
Metals beach cast wet weight	Chromium (Cr)	Square root	0.07
vet	Copper (Cu)	None	0.184
ıst v	Iron (Fe)	None	0.628
h ca	Mercury (Hg)	Square root	0.364
eac	Magnesium (Mg)	ln	0.312
b b	Manganese (Mn)	None	0.109
[eta]	Nickel (Ni)	None	0.847
Z	Selenium (Se)	None	0.47
	Strontium (Sr)	None	0.062
	Zinc (Zn)	None	0.406
ltins ve			
Butyltins Live	Total Butyltins	ln	0.196
	Monobutyltin	None	0.263
Butyltins Beach cast	Dibutyltin	ln	0.548
ach	Tributyltin	None	0.465
$\mathbf{B}_{\mathbf{F}}$ B	Total BT	None	0.223

## Appendix 10. Normality Tests

Contaminant			Shapiro-Wilk
Group	Contaminant	Transformation	Significance
	PCB1	None	0.422
	PCB8/5	None	0.591
	PCB28	ln	0.337
	PCB42/59/37	None	0.3
	PCB44	None	0.161
	PCB49	Rankit	0.99
	PCB101/90	ln	0.404
PCBs	PCB105	ln	0.353
РС	PCB135	Rankit	0.99
	PCB138/160	ln	0.263
	PCB153/132	None	0.162
	PCB 175	Rankit	0.99
	PCB180	None	0.257
	PCB187	None	0.581
	total PCBs A	Rankit	0.99
	total PCBs B	Rankit	0.99
Organochlorines Beach cast			
Org	total PCB	None	0.3
	n-docosane	None	0.482
	n-eicosane	None	0.259
	n-heptacosane	None	0.311
	n-heptadecane	None	0.169
	n-hexacosane	None	0.06
ŝ	n-hexadecane	None	0.05
Aliphatics	n-nonacosane	None	0.487
liph	n-nonadecane	None	0.081
A	n-octacosane	ln	0.558
	n-octadecane	None	0.07
	n-pentacosane	None	0.367
	n-pentadecane	None	0.57
	n-tetracosane	None	0.294
	n-triacontane	None	0.269
AHs	Unresolved Complex		
$\mathbf{P}_{I}$	Mixture	Rankit	0.913
Semivolatiles PAHs	1,2,3,4- Tetrachlorobenzene	N/A	N/A
Sem	1,2,4,5- Tetrachlorobenzene	None	0.077

Contaminant			Shapiro-Wilk
Group	Contaminant	Transformation	Significance
oid	Vit A-plasma [R] ug/L	None	0.328
Ъуг	Vit A-liver [R] in ug/g	ln	0.161
T br	[RP] in ug/g	ln	0.542
Vitamin A and Thyroid	TR in ug/g	ln	0.281
in /	R:RP	ln	0.361
tam	T4 nmol/L	ln	0.101
Vi	T3 nmol/L	ln	0.092
p450	PBMC Molecules p450/100ng total RNA	ln	0.422
đ	Liver Molecules p450/100ng total RNA	None	0.365
	White Blood Cells (WBC)	None	0.778
	Red Blood Cells (RBC)	None	0.270
	Hemoglobin (HGB)	Rankit	0.229
	Hematocrit (HCT)	None	0.136
	Mean Corpuscular Volume (MCV)	None	0.383
nts	Mean Corpuscular Hemoglobin (MCH)	None	0.957
Cell Blood Counts	Mean Corpuscular Hemoglobin Concentration (MCHC)	None	0.319
Cell	Red Cell Distribution width (RDW)	Rankit	0.721
	Platelets	None	0.275
	Mean Platelet Volume	None	0.982
	Percent Neutrophils	None	0.451
	Percent Lymphocytes	None	0.674
	Percent Monocytes	None	0.315
	Percent Eosinophils (EOS)	ln	0.785
	Percent Basophils	N/A	

Contaminant			Shapiro-Wilk
Group	Contaminant	Transformation	Significance
	Glucose (mg/dL)	None	0.400
	Blood Urea Nitrogen		
	(BUN), (mg/dL)	Rankit	0.255
	Creatinine (mg/dL)	N/A	
	<b>BUN/Creatinine Ratio</b>	None	0.274
	Uric Acid (mg/dL)	None	0.072
	Sodium (mequiv/L)	ln	0.057
	Potassium (mequiv/L)	None	0.762
	Na/K Ratio	None	0.061
	Chloride (mequiv/dL)	None	0.512
	Carbon Dioxide (meq/L)	None	0.125
	Calcium (mg/dL)	None	0.517
	Phosphorus mg/dL)	ln	0.357
	Total Protein (g/dL)	None	0.297
	Albumin (A), (g/dL)	None	0.284
	Globulin (calculated)		
	(G), (g/dL)	None	0.447
<u>y</u> r	A/G Ratio	ln	0.212
nist	Triglycerides (mg/dL)	None	0.236
her	Cholesterol (mg/dL)	ln	0.047
m C	Total Bilirubin (mg/dL)	N/A	
Serum Chemistry	Direct Bilirubin (mg/dL)	N/A	
	Gamma Glutamyltransferase (GGT)	Rankit	0.197
	Alkaline Phosphatase (AP)	Rankit	0.275
	Lactic Dehydrogenase (LD)	None	0.265
	Aspartate Aminotransferase (AST)	ln	0.448
	Alanine	ln.	0.469
	Aminotransferase (ALT)	ln None	0.468 0.669
	Iron (µg/dL) Hemolytic Index		0.545
	•	None	
	Lipemic Index	ln N/A	0.099
	Icteric Index	N/A None	0.006
	Osmolality	None	0.096
	Anion Gap	Rankit	0.990

Shapiro-Wilk Significance is for transformed data unless data unless data were not transformed

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