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NATURAL RESOURCES DAMAGE ASSESSMENT

DRAFT PRELIMINARY STATUS REPORT

US Fish and Wildlife Service

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Alaska Fish and Wildlife Research Center

Study Title:	Assessment of the Magnitude, Extent, and Duration of Oil Spill Impacts on Sea Otter Populations in Alaska.
Study ID Number:	Marine Mammals Study Number 6
Project Leaders:	B. E. Ballachey, J. L. Bodkin and D. Burn
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Preliminary Damage Assessment Report

Marine Mammal Study 6 20 November 1991

EXECUTIVE SUMMARY

Damages to sea otters resulting from the T/V <u>Exxon Valdez</u> oil spill can be categorized into two temporal components: 1) acute mortality, and 2) chronic damages.

Acute mortality was partially reflected in a total of about 1,000 sea otter carcasses collected during or shortly after the spill. It is probable that some additional number of sea otters became oiled and subsequently died, and that their carcasses were never recovered. Preliminary results of autopsies of intact carcasses indicate less than 5% of mortalities were clearly not spill related.

Three approaches were taken to estimate the total number of sea otter mortalities that resulted from acute exposure to oil. One method estimates the number of unrecovered carcasses based on the probability of carcass recovery. Available information suggests a about 75% of sea otter carcasses are not recoverable. Another method compares estimates of sea otter abundance before and after the spill. These comparisons suggest a large number of otters, relative to the number of carcasses retrieved, suffered acute mortality. A third method consists of an intersection model to estimate mortality based on potential exposure to oil and observed mortality rates dependent on degree of oiling. Simulations of this model indicate that about 52% of the Kenai Peninsula sea otter population was potentially exposed to oil. This model may be applied throughout the spill zone to provide an estimate of total mortality. A synthesis of these methods of loss estimates suggest between 3,500 and 5,500 sea otter may have died as a result of acute exposure to oil following the Exxon Valdez spill.

Chronic damages to sea otters may result from sub-lethal initial exposure and continued exposure to environmental hydrocarbons. Indirect damages, either chronic or acute, may result from affected sea otter prey populations. Preliminary findings of Coastal Habitat and Shellfish studies have identified elevated levels of hydrocarbons in intertidal and subtidal sediment samples collected within the spill zone. Additionally, hydrocarbon analysis of benthic marine invertebrates indicate that high levels of hydrocarbons persist in several species previously identified as sea otter prey in western Prince William Sound.

Preliminary results of several sea otter damage assessment studies indicate that sub-lethal, chronic exposure may be damaging sea otters at the biochemical, physiological and population levels. Comparisons of pre- and post-spill estimates of sea otter abundance, based on boat surveys the shore, found non-oiled areas underwent a 13.5% increase in abundance, while oiled areas underwent a 34.6% decrease. In addition, the post-spill population in the oiled area is significantly lower than the best pre-spill estimate, indicating a real decline on the order of 1600 otters initially, and up to 2200 in subsequent years. No change in abundance was detected between July 1990 and July 1991 surveys.

Results of a 1991 field study evaluating sea otter prey selection and foraging success indicate sea otters have not altered their diet over the past decade and continue to rely primarily on clams and mussels as forage. Given hydrocarbon contamination in bi-valve mollusks, the prey study described a pathway for continued exposure of sea otters to environmental hydrocarbons. In addition, other damage assessment studies have documented decreases in abundance of mussels in oiled areas which may further impede recovery of sea otter population.

Analysis of age class composition of beach cast sea otters in western Prince William Sound from 1974-84, in 1989 and following the spill in 1990-91 detected significant differences between prespill and spill year distributions, and pre-spill and post-spill distributions. Sall year and post-spill distributions were not significantly different. The proportion of prime age animals dying in western Prince William Sound increased in both the spill year (1989) and post-spill years (1990 and 1991). The observed differences represent a shift from a pre-spill composition of principally young and aged animals to an increased proportion of prime-age animals during and following the spill. The observed changes in the age distributions of dying sea otters suggest a prolonged, spill-related affect on at least the western Prince William Sound sea otter population.

Results of clinical, hematologic and serum chemistry analyses of otters which died shortly (within the first 10 days) after entering the rehabilitation centers indicate that the majority of sea otters that died acutely appear to have succumbed to shock. Terminal generally included lethargy and hypothermia, often signs accompanied by anorexia, convulsions, and hemorrhagic diarrhea. The most common hematologic abnormalities included lymphopenia, leukopenia, and anemia. The most prevalent syndromes identified in these otters by clinical chemistry included azotemia, hyperkalemia, hypoglycemia, hepatocellular leakage, and hypoproteinemia/hypoalbuminemia.

Blood samples collected from wild caught otters in 1990 and 1991 identified significant differences in several blood parameters between eastern and western Prince William Sound. Hematologic and serum chemical analyses of adult male sea otters found significantly higher hematocrits and hemoglobins in the west. Western males had significantly higher eosinophil counts, suggesting systemic hypersensitivity reactions. Serum sodium and serum chloride were significantly higher and serum potassium lower in western males. Although there were no significant differences in hematologic parameters between east and west female otters, some chemistry changes were present which were consistent with changes observed in the males. However, the degree of difference was small so that data must be interpreted cautiously. As a group, western sea otter pup hematocrits, hemoglobins, and red cell counts were significantly lower than those of eastern pups, suggesting a mild anemia in the west pups. Hematologic and clinical chemistry differences between eastern pups were of equivocal biologic significance, and trends seen in adults were not present in the pups observed to date.

Over 400 sea otter tissue samples, predominatly liver, fat, and blood, have been analyzed for hydrocarbon contaminants. An exploratory data analysis has been initiated. Results will allow identification of linkages between exposure and effects observed in the population.

Results of a study on survival of sea otter pups demonstrate significantly higher post weaning mortality in western Prince William Sound, compared to controls in the east. In contrast, survival of adult females sea otters was significantly higher in western Prince William Sound compared to controls in the east. However, pupping rates of adult females and survival of those pups through weaning in 1990 and 1991 were similar between eastern and western Prince William Sound and were considered normal.

In conclusion, results of damage assessment studies to date suggest initial damages were extensive, killing between 3,500 and 5,500 sea otters. Additionally, results of several NRDA studies suggest chronic damages to sea otters are occurring which may preclude or delay recovery of affected populations. Although the conclusions presented here should be considered preliminary, evidence of persistent damages is compelling and warrants continued investigation.

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Section 1

BOAT-BASED SURVEYS OF SEA OTTERS IN PRINCE WILLIAM SOUND, ALASKA

Douglas M. Burn

SUMMARY

As part of the Natural Resources Damage Assessment (NRDA) studies conducted during spill year 3, boat-based surveys of sea otters in Prince William Sound were continued. The results of these third year surveys indicate that the otter population may have stabilized, but is still below pre-spill levels.

OBJECTIVES

The purpose of this study was to estimate post-spill sea otter population size in Prince William Sound, in order to determine initial and continuing damages related to the <u>Exxon Valdez</u> oil spill.

INTRODUCTION

Within the first few weeks of the Exxon Valdez oil spill, the sea otter quickly became the most vivid symbol of the damage to wildlife in Prince William Sound, Alaska. The fact that sea otters suffered injury by the spill cannot be refuted. It is only the magnitude of this injury that is in dispute. Various methods have been used to extrapolate the number of carcasses recovered to total mortality, based on carcass recovery rates. This study attempts to make direct comparisons between pre- and post-spill population estimates, in order to quantify the net change in the otter population.

STUDY METHODOLOGY

Study Area and Survey Strata

The study area consists of the waters of Prince William Sound (PWS), Alaska, exclusive of Hawkins Island Cutoff and Orca Inlet (Figure 1). Previously, the study area had been divided into 3 distinct survey strata: shoreline, coastal, and pelagic. The shoreline strata was based on shoreline transects surveyed by Irons, Nysewander and Trapp (1988) during the summers of 1984 and 1985, and is defined by the 200m-wide strip adjacent to the coastline. Areas outside of this 200m-wide strip were not systematically surveyed prior to the spill. In an attempt to quantify the PWS otter population as a whole, the areas greater than 200m from shore were divided into sampling "blocks" based on a 5-minute latitude/longitude grid system. The coastal stratum was comprised of those blocks that are immediately adjacent to 1km or more of shoreline. The pelagic stratum was comprised of those blocks that are adjacent to less than 1km of shoreline. Within each block, a number of 200m-wide strip transects were designated for sampling.

This original designation of coastal and pelagic strata was of little value to analysis of sea otter data (it must be emphasized that these data are collected as part of a joint seabird/marine mammal survey). Therefore, areas outside of the shoreline stratum were restratified, based on their distance to otter foraging habitat as defined by the 20m bathymetric contour. Those blocks whose mean distance is greater than 5km from foraging habitat are considered "offshore" blocks, while those whose average distance is less than 5km are considered "nearshore" blocks. While this new stratification still suffers from the artificial rigidity of the grid system, I feel it is an improvement over the original scheme.

Oiling Classification

The determination of oiled vs. non-oiled transects and blocks is of major importance in the estimation of damage to the sea otter population from these data. The pathways taken by the oil is a fundamental question in virtually all damage assessment studies. Ironically, it is one area of investigation that has been virtually ignored on a comprehensive basis, even though estimation of damages is extremely sensitive to these determinations.

Given the numerous sources of oiling information, with no means of evaluating the relative strengths and weaknesses of each, at this point in time any reasonable stratification of oiling seems as good as any other. The oiling classification presented in this report is based on Alaska Department of Environmental Conservation (ADEC) overflight data from the time of the spill. Aerial observations were used to create a GIS coverage depicting the movement of oil over the surface of the water. While it must be recognized that this interpretation of oiling has certain inconsistencies when compared to shoreline oiling and/or the National Oceanographic and Atmospheric Administration's (NOAA) HAZMAT model, it is the oiling picture most often used to depict the geographic extent of the spill.

Since sea otters are highly mobile animals, it is possible that otters inhabiting areas outside the pathway of the oil could have swam into the area of the slick and become oiled. For this reason, a buffer zone of 5km was added to the ADEC overflight data to represent an area within which otters might have been affected by oil. Shoreline transects, and nearshore and offshore blocks with any area located within 5km of the oil are therefore classified as oiled (Figure 2).

Survey Dates

Pre-spill data of Irons et al. (1988) were collected for shoreline transects during the summers of 1984 and 1985. These data serve as a baseline for comparison with post-spill surveys. For the purposes of this report, the pre-spill effort is referred to as survey 0. To date, 9 post-spill surveys have been completed. The dates of all surveys are listed below:

Survey	Ô	07/07/84	-	08/24/84
		05/22/85		08/31/85
Survey	1	06/15/89	-	07/06/89
Survey	2	07/12/89	-	07/26/89
Survey	3	08/07/89	-	07/26/89
Survey	4	03/12/90	-	03/22/90
Survey	5	06/08/90	-	06/23/90
Survey	6	07/06/90	-	07/30/90
Survey	7	08/06/90	-	08/22/90
Survey	8	03/07/91	-	03/23/91
Survey	9	07/03/91	-	07/22/91

<u>Sample Size</u>

The baseline survey of Irons et al. (1988) consisted of 718 shoreline transects, of which 708 are within the PWS study area (the remaining 10 transects are within the Orca Inlet and Hawkins Island Cutoff areas). The nearshore and offshore strata were not sampled by Irons et al. (1988).

Post-spill surveys were initially conducted during the summer of 1989 as a random sample of approximately 25% of available shoreline transects and the original coastal and pelagic blocks. Due to logistical constraints, only the shoreline stratum was sampled during survey 1. All three strata have been sampled in each survey since July 1989. Once the initial random sample of transects and blocks was chosen, each successive survey replicated the same sampling units to allow for comparison over time.

In order to complete winter surveys when weather conditions are often more constraining, only a subset of the summer transects and blocks (approximately 14% of all shoreline transects and nearshore and offshore blocks) were sampled.

Beginning with survey 5 in June 1990, an additional 25 transects were added to the shoreline sample, increasing the proportion sampled from 25% to 29%. These additional transects were randomly selected from both oiled and non-oiled western Prince William Sound. Sample sizes of the nearshore and offshore strata were not increased.

Analytical Methods

Sea otter density and abundance estimates are calculated using ratio estimator techniques (Cochran, 1977).

Shoreline sea otter density is calculated as a ratio:

$$R = \frac{\sum y}{\sum x}$$

where: R = shoreline sea otter density y = number of sea otters within shoreline transect x = area of shoreline transect in km²

Standard error of this ratio is calculated as:

$$\mathbf{s}(R) = \frac{1}{\bar{x}} \sqrt{\frac{\sum (y - Rx)^2}{n(n-1)} \frac{N - n}{N}}$$

where: s(R) = standard error of R N = total number of shoreline transects n = number of sampled shoreline transects

The ratio estimate of shoreline sea otter population:

$\hat{Y}_{R} = RX$

where: \hat{Y}_R = ratio estimate of shoreline sea otter population X = total area of all shoreline transects in km²

Standard error of this estimate is calculated as:

 $s(\hat{Y}_R) = s(R)X$

where: $s(\hat{Y}_R)$ = standard error of the ratio estimate \hat{Y}_R

Nearshore and offshore sea otter densities are calculated within each block as:

$$r_{B} = \frac{\sum y_{t}}{\sum x_{t}}$$

where:

- - $x_t = area of nearshore/offshore transect(s) sampled$

within survey block in km²

Otter densities are calculated for the nearshore and offshore strata as:

$$R = \frac{\sum x_B r_B}{\sum x_B}$$

where: R = sea otter density in nearshore and offshore strata $x_{R} =$ total area of sampled survey block in km^{2}

Standard error of this ratio is calculated as:

$$s(R) = \frac{1}{\bar{x}_B} \sqrt{\frac{\sum (r_B x_B - R x_B)^2}{n(n-1)}}$$

where: s(R) = standard error of R N = total number of nearshore/offshore blocks n = number of sampled nearshore/offshore blocks

Estimate of nearshore and offshore otter population size:

$\hat{Y}_{R} = RX$

where:

 \hat{Y}_R = ratio estimate of nearshore/offshore otter population X = total area of all nearshore/offshore habitat in km²

Standard error of the estimated population size:

 $s(\hat{Y}_R) = s(R)X$

where: $s(\hat{Y}_R) = standard error of estimate \hat{Y}_R$

STUDY RESULTS

Numbers of shoreline transects sampled and sea otters counted are presented in Table 1. Numbers of nearshore blocks sampled and otters counted are presented in Table 2. With the exception of one animal sighted during the July 1990 survey, no otters have ever been observed in the offshore stratum. Summary tables and estimates for this stratum are therefore omitted.

All estimates presented in this report are uncorrected for

sightability. A pilot study conducted concurrently with the August 1990 survey estimated an overall sightability factor of 0.70. This value is based on a relatively small sample size of ground-truthing segments (n=21). While it is unclear how accurate this sightability value is, it is certain that these boat-based observations do miss some proportion of the otters within a transect. Abundance estimates presented here should be considered minimum values.

Sea otter density and abundance estimates for the shoreline stratum are presented in Table 3. In the non-oiled area, otter densities exhibited a modest increase (13.5%) between the prespill surveys of Irons et al. (1988) and the summer 1989 surveys. This would suggest that the otter population in these areas was increasing. Otter densities in the oiled area underwent a decline of approximately 34.6% during the same time period. Surveys conducted after summer 1989 show further declines in otter density in oiled areas, suggesting continuing injury to the population. However, otter density in non-oiled areas also exhibit a decline during the same time period. With the exception of the July 1990 survey, otter densities in the oiled area are consistently lower than those in the non-oiled area, which is exactly opposite the pre-spill pattern.

Otter density and abundance estimates for the nearshore stratum are presented in Table 4. Due to the relatively low level of sampling effort, abundance estimates for this stratum have low precision. In every survey, otter densities are much lower in the oiled area than in the non-oiled area. However, lacking prespill information on this stratum, it is unclear if this pattern is due to oil or some other factors. Of particular interest is the peak in otter density seen in all 3 July surveys for the nonoiled area. These fluctuations in otter densities have profound effects on the overall population estimates.

Ratios of nearshore to shoreline abundance were calculated for the 6 post-spill summer surveys in order to develop a correction factor to apply to the pre-spill shoreline estimate of Irons et al. (1988). For the non-oiled area, the mean nearshore/shoreline ratio is 1.855 (s.e. 0.363), and for the oiled area 0.852 (s.e. 0.187). The standard error of the mean of the 6 ratios was used to calculate an approximate 95% confidence interval around the adjusted pre-spill estimates. Adjustments for population growth during the period between 1984/85 and the moment of the spill have not been made. An increase in population during that time would logically result in larger estimates of damages. Total population estimates for the Prince William Sound study area are presented in Table 5. These data suggest that the current postspill population size is approximately 6000 otters.

Population trends in non-oiled, oiled, and all areas are presented in Figures 3-5. For comparison, the pre-spill

population estimate is also shown. In the non-oiled area, there is considerable overlap between pre- and post-spill population estimates. There is virtually no overlap in between estimates in the oiled area. These results would suggest that a real decline in sea otter abundance has occurred within the oiled area. The difference between point estimates is on the order of 1600 otters initially in summer 1989, and later 2200 otters in summer 1990 and 1991 (Figure 6). Adjusting these values for otters not seen, these loss estimates would be 2285 and 3143 otters, respectively. Given that approximately 500 oil-related otter carcasses were recovered at the time of the spill, these data suggest a carcass recovery rate of 21%.

STATUS OF INJURY ASSESSMENT

The task of estimating damages to sea otters is an extremely complicated one. Other sections of this report deal with physiological changes on the organismal level. The effect of low levels of hydrocarbon residues in blood and/or tissues of an individual otter may be insignificant on the population level. Conversely, population size may fluctuate over time for a variety of natural causes. Attempts to discern which population changes are directly related to the <u>Exxon Valdez</u> oil spill should only be made with a thorough understanding of the qualifying assumptions involved.

First, the geographic extent of the spill itself is not definitively known. Due to the mobility of sea otters, the actual extent of the oil effects on the sea otter population is also unknown. Results from this study suggest that the otter population in the Prince William Sound study area has undergone a decline relative to the 1984/85 surveys. The proportion of this decline attributable to oil is directly dependent upon classification of sampling units as oiled vs. non-oiled.

Second, pre- and post-spill comparisons are limited by the quality of the pre-spill data set. Results from post-spill surveys indicate that considerable numbers of sea otters frequent the waters greater than 200m from shore, and that proportions of otters within the shoreline and nearshore strata vary seasonally. Lack of pre-spill data from the nearshore stratum is a severe limitation to these comparisons. In addition, the 4-5 year period between pre- and post-spill surveys is problematic. The distribution and abundance of the population at the moment of the spill may or may not have been comparable to that of the 1984/85 survey.

Finally, the survey design for the waters greater than 200m from shore (i.e. the nearshore and offshore strata) is poor. Sampling units should be redesigned to increase precision, while still maintaining comparability with previous surveys. As a population monitoring tool, the current survey could only detect large population changes, primarily due to low precision in the nearshore stratum. The cooperative nature of this survey (seabirds and marine mammals are counted) dictates that the summer survey be conducted during July to minimize the influence of migratory bird species. In addition, we now have a period of 3 consecutive July surveys completed. Unfortunately, July also appears to be the summer month with the greatest number of otters moving into the nearshore stratum. This causes the July results to be the least precise of any summer survey.

Given these numerous assumptions and caveats, there remain several important results from this study. First, in direct comparisons between pre-and post-spill shoreline data, non-oiled areas underwent a 13.5% increase, while oiled areas underwent a 34.6% decrease. And second, the post-spill population in the oiled area is significantly lower than the best pre-spill estimate, indicating a real decline on the order of 1600 otters initially, and up to 2200 in subsequent years (estimates uncorrected for sightability). On the positive side, the population showed little change between July 1990 and July 1991 surveys, suggesting that perhaps the effects of the spill on a population level have subsided. However, this can not be determined conclusively without further monitoring of the population.

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	Non	-Oiled Are	ea	C	iled Area	
Survey Date	Sampled Transects	Area (km²)	Count of Otters	Sampled Transects	Area (km²)	Count of Otters
PWS Study Area	301	387.0		441	453.3	
Summer 1984/85	285	367.6	1666	423	417.5	2191
June 1989	68	85.2	445	115	121.7	400
July 1989	69	86.7	460	118	125.4	414
August 1989	69	86.7	425	118	125.4	464
March 1990	38	49.6	216	61	66.6	173
June 1990	78	96.6	305	134	139.3	219
July 1990	78	96.6	253	134	139.3	384
August 1990	78	96.6	388	134	139.3	411
March 1991	38	49.5	195	61	66.6	123
July 1991	78	96.6	294	134	139.3	406

Table 1.1 Numbers of shoreline transects sampled and sea otters counted in Prince William Sound, Alaska.

		Noi	n-Oiled Are	<u>ea</u>		Oiled Area	
_	Survey Date	Sampled Blocks	Area (km²)	Count of Otters	Sampled Blocks	Area (km²)	Count of Otters
	PWS Study Area	118	2400.1		147	4573.8	
	Summer 1984/85						
	June 1989						
	July 1989	28	571.7	61	36	1140.3	31
	August 1989	28	571.7	23	36	1140.3	19
	March 1990	17	416.4	23	30	975.3	21
	June 1990	27	543.6	44	34	1090.7	25
	July 1990	28	571.7	62	36	1140.3	19
	August 1990	28	571.7	42	36	1140.3	15
	March 1991	17	416.4	21	30	975.3	9
	July 1991	27	543.6	61	36	1140.3	18

Table 1.2 Numbers of nearshore blocks sampled and sea otters counted in Prince William Sound, Alaska.

	<u>N</u> Density	<u>on-Oiled Are</u> Estimated	<u>ea</u>	Density	<u>Oiled Area</u> Estimated	
Survey Date	(otters/km ²)	Abundance	95% c.i	(otters/km ²)	Abundance	95% c.i.
Summer 1984/85	4.53	1754	±143	5.25	2285	±101
June 1989	5.22	2020	±586	3.29	1430	±362
July 1989	5.31	2053	±631	3.30	1438	±360
August 1989	4.90	1897	±635	3.70	1611	±460
March 1990	4.35	1685	±566	2.60	1130	±199
June 1990	3.16	1221	±511	1.58	690	±331
July 1990	2.62	1013	±291	2.76	1200	±397
August 1990	4.02	1554	±439	2.95	1284	±419
March 1991	3.94	1524	±510	1.85	804	±161
July 1991	3.04	1177	±362	2.91	1268	±287

Table 1.3 Summary of shoreline sea otter density and abundance estimates for Prince William Sound, Alaska.

Survey Date	<u>N</u> Density (otters/km ²)	on-Oiled Are Estimated Abundance	<u>ea</u> 95% c.i	Density (otters/km ²)	<u>Oiled Area</u> Estimated Abundance	<u>Area</u> ated ance 95% c.i.		
Summer 1984/85					67 - 14 - 14 - 14 - 14 - 14 - 14 - 14 - 1			
June 1989								
July 1989	1.33	3204	±1865	0.31	1416	±1274		
August 1989	0.49	1180	±735	0.18	838	±754		
March 1990	0.67	1604	±886	0.24	1120	±879		
June 1990	0.99	2370	±1784	0.26	1190	±890		
July 1990	1.30	3126	±3094	0.18	846	±701		
August 1990	0.86	2070	±1182	0.15	704	±473		
March 1991	0.57	1368	±497	0.11	498	±414		
July 1991	1.27	3050	±1861	0.18	801	±796		

Table	1.4	Summary	of	nearshore	sea	otter	density	and	abundance	estimates	for	Prince
		William	Sou	und, Alaska	a.							

Table 1.5 Summary of sea otter abundance estimates for Prince William Sound, Alaska. Summer 1984/85 estimate based on shoreline/nearshore ratios from 6 post-spill summer surveys.

	<u>Non-Oiled Area</u>		<u>Oiled</u> Estimated	Area	<u>All Areas</u> Estimated		
Survey Date	Abundance	95% c.i	Abundance	95% c.i.	Abundance	95% c.i.	
Summer 1984/85	4992	±1248	4249	±846	9241	±1508	
June 1989							
July 1989	5257	±1969	2854	±1324	8111	±2373	
August 1989	3077	±971	2449	±884	5526	±1313	
March 1990	3289	±1051	2250	±901	5539	±1384	
June 1990	3592	±1856	1880	±950	5471	±2084	
July 1990	4140	±3107	2099	±813	6239	±3212	
August 1990	3624	±1261	1988	±632	5612	±1411	
March 1991	2892	±712	1301	±444	4194	±840	
July 1991	4228	±1895	2069	±846	6297	±2076	



Prince William Sound study area, showing survey strata.



Figure 1.2 Oiling classification of sampling units within the Prince William Sound study area.



Figure 1.3. Post-spill sea otter population trend in the non-oiled areas of Prince William Sound, Alaska. Error bars represent 95% confidence intervals.



Figure 1.4. Post-spill sea otter population trend in the oiled areas of Prince William Sound, Alaska. Error bars represent 95% confidence intervals.



Figure 1.5. Post-spill sea otter population trend in the all areas of Prince William Sound, Alaska. Error bars represent 95% confidence intervals.



Figure 1.6 Estimated net change in sea otter population in the oiled areas of Prince William Sound, relative to best pre-spill estimate.

DEVELOPMENT OF SEA OTTER SURVEY TECHNIQUES

James L. Bodkin and Mark S. Udevitz

SUMMARY

Field trials were conduced in 1991 to evaluate the Piper PA-18 Super-Cub as a platform from which to estimate sea otter density. At least 90% of the sea otters observed in survey units by ground based observers were also detected by intensive aircraft searches at each of 3 altitudes using a fixed search pattern. The proportion of sea otters detected was also greater than or equal to .90 for intensive searches using each of 3 search patterns at a fixed altitude. Sea otter detectability increased and its rate of increase decreased with increasing search intensity. Results of these investigations suggest that additional work toward development of a Super-cub based sea otter survey technique is justified.

A boat-based survey was designed and implemented in 1991 to estimate ratios of independent to dependent sea otters and patterns of habitat use by sea otters in portions of Prince William Sound. The ratio of independent to dependent sea otters was similar among study areas along oiled and un-oiled shoreline in the western sound and along un-oiled shorelines in the eastern sound.

OBJECTIVES

1) Evaluate the feasibility of using the Piper PA-18 aircraft as platform for estimating sea otter density.

2) Design and implement a small boat survey to estimate ratios of dependent to independent sea otters and patterns of habitat use by sea otters in oiled and non-oiled portions of Prince William Sound.

INTRODUCTION

Initial damages to the sea otter population resulting from the T/V Exxon Valdez oil spill included lethal and sub-lethal levels of direct exposure. One method used to estimate the total immediate loss to the sea otter population in Prince William Sound was a comparison of estimates of sea otter abundance based on boat surveys conducted before and after the spill. Boat surveys were used to estimate sea otter density in 1989 and 1990 in order to be consistent with the method used before the spill. This consistency was necessary for assessing the immediate loss,

but it has become evident that the boat survey methodology, as conducted, will not provide the accuracy or precision necessary to monitor the smaller annual changes that are likely to occur in the post-spill population (Burn 1990, Udevitz et al. 1990).

Several methods have been used in the past to obtain estimates of sea otter abundance and distribution. Most surveyors of sea otter abundance have acknowledged that some animals were probably missed (Kenyon 1969, Estes 1977) but have generally failed to quantify biases or estimate precision.

Counts of sea otters from the ground have been generally recognized as providing the most accurate estimates of nearshore sea otter abundance (Schneider 1971). Estes and Jameson (1988) estimated that the probability of sighting sea otters was 94.5% for standardized shoreside counts, using two experienced observers, high-resolution 10X binoculars and 50X Questar telescopes (New Hope, PA). This was the first study to rigorously evaluate the effect of activity, group size and distance from observer on sighting probability of sea otters. Their results provide a baseline, against which other methods might be evaluated. However, due to limited access and transportation along most coastlines, ground counts can not be used over the large geographic areas occupied by most sea otter populations.

Sea otter surveys have commonly been conducted from vessels of various sizes (Ebert, 1968, Jameson et al. 1986, Johnson 1987, Pitcher 1989). Pitcher (1989) suggested that applications of boat counts should be limited to assessing trends in abundance and geographic distribution. Schneider (1971) felt boat counts provided higher counts than aerial surveys. Johnson (1987) found that counts from boats were 1.7 times greater than helicopter counts in Prince William Sound. Udevitz et al. (1990) determined that detection of sea otters in boat-based surveys is reduced due to avoidance behavior of the otters as well as sightability problems. In addition to surveying sea otter abundance, boat surveys have been used to provide indices of reproductive rates in sea otter populations (Estes 1990).

Helicopters have occasionally been used to conduct surveys of sea otter populations (Estes 1971, Schneider 1971, Pitcher 1975). Schneider (1971) concluded that helicopters counts were two to four times higher than fixed-wing counts but that shore counts may be three times greater than helicopter counts. Johnson (1987) found helicopter counts inferior to boat counts. Preliminary, quantitative studies reported by Douglas et al. (1990) suggest that rotary-winged aircraft might be suitable as an observation platform. Cost and safety considerations of rotary-wing aircraft over water may preclude their wide spread use in sea otter surveys. Aerial counts of sea otters from fixed-wing aircraft have been used for several decades throughout their range (Kenyon 1969, Wild and Ames 1974 and Simon-Jackson et al. 1986). Fixed-wing surveys have usually been based on single survey lines along a shoreline where sea otters may occur. Kenyon (1969) felt aerial surveys provided higher counts than those obtained from small boats, and estimated he observed 50-75% of the sea otters present from the air. Schneider (1971) felt only 10-25% of the sea otters were counted in a fixed-wing survey. Studies conducted by California Dept. of Fish and Game estimated that 25-40% of the sea otters are missed by fixed-wing counts (Wild and Ames 1974, Geibel and Miller, 1984). Traditionally, fixed-winged surveys have been conducted without standardized protocols. Many different types aircraft (e.g., DC-3, Grumman Goose, Cessna 185, Cessna 206, Piper PA-18) have been used, each with very different flight characteristics (e.g., speed, maneuverability, viewing attributes). Procedures commonly varied between surveys (e.g., airspeed, altitude, number of observers, weather conditions, flight patterns). While many of these survey attributes have been recognized as affecting counts, the magnitudes of these effects have not been previously quantified.

The objective of most sea otter surveys has been to describe changes that may have occurred in the abundance and distribution of the species over time or to provide a baseline against which future counts may be compared. Two factors have generally led to difficulty in interpreting survey data. First, with few exceptions (Estes and Jameson 1988, and Jameson et al. 1986) survey methodologies have not been standardized. Secondly, except for the ground counts described by Estes and Jameson (1988) and Jameson et al. (1986), survey methods have not been rigorously tested to determine the proportion of the animals actually observed or the effects of activity and environmental conditions on sightability.

The long term objective of this study is to develop and implement standardized survey methodologies that will provide improved estimates of sea otter density and reproductive rates and that are applicable throughout the species' range. The first phase of this work consisted of trials conducted in April and July, 1991 to evaluate the Piper PA-18 Super-Cub as a survey platform for estimating sea otter density. The Super-Cub has been selected repeatedly for wildlife survey work based on its slow stall speed and high degree of maneuverability (Erickson and Siniff 1964, LeResche and Rausch 1974, Gasaway et al. 1986). It seats one pilot and one passenger in tandem, an arrangement recommended by Erickson and Siniff (1964) as allowing navigation and observation to occur from the same spatial orientation in the plane. The second phase of this project was the development and implementation of a systematic boat survey in August and September, 1991 to document rates of reproduction in western Prince William Sound. Data analysis has not yet been completed,

but we present preliminary results in this report.

METHODS

Aircraft Evaluation

Two series of trials were conducted to evaluate the Piper PA-18 aircraft as a platform for estimating sea otter density. All trials used ground based observers to quantify the proportion of animals detected from the air. The first series of trials was conducted in April 1991 to assess the effect of altitude (altitude evaluation) on sea otter detectability. The second series of trials was conducted in July 1991 to assess the effect of search pattern (pattern evaluation) on sea otter detectability.

Trials were conducted on areas of ocean (survey units) that did not contain canopy forming kelp, were large enough to contain a full search pattern, allowed unrestricted observation from an adjacent vantage point, and contained 1 or more otters immediately prior to arrival of the aircraft. Survey units were selected by ground crews based on previous reconnaissance and observation of the area immediately before ground crew deployment. All survey units for the altitude evaluation were located in Eastern Prince William Sound. Survey units for the pattern evaluation were scattered throughout Prince William Sound, though most were in the west.

Ground crews approached each selected survey unit after a thorough study of the area from offshore, taking care to minimize disturbance to sea otters. After deploying themselves at the vantage point, the ground crew defined the boundaries of the unit, established an orientation for the aerial search pattern within the unit and determined the position and activity of each otter within the unit. The ground crew then contacted the aircraft by VHF radio to begin the trial. Ground observations followed methods established by Estes and Jameson (1988). Immediately prior to arrival of the aircraft, the ground crew recorded the location, group size, number of dependent pups and activity of each otter or group of otters. Activity categories included swimming (changing location), resting (stationary on water surface) and diving (stationary and temporarily submerging). The ground crew also recorded the location and behavior of all otters observed outside the boundaries of the unit, observations regarding changes in sea otter activity associated with the approach of the aircraft, and the time the aircraft entered and departed the unit. Following the departure of the aircraft, the ground crew was transported by boat to the next survey unit.

Altitude evaluation trials were conducted at 46m, 92m and 137m

observed by both crews (b_i) , the number observed only by the ground crew (g_i) , and the number observed only by the aerial observer (a_i) in the observation circle or strip were determined. The number of otters in the circle or strip before any response to the approaching aircraft was determined based on ground crew observations prior to the arrival of the aircraft.

Sea otter detection probabilities (detectabilies) for the aerial observer were estimated as

$$\hat{P}_{d} = \frac{\sum_{i=1}^{L} b_{i}}{\sum_{i=1}^{L} (b_{i} + g_{i})}$$

where r is the number of trials. Detectabilities were also estimated separately for each trial as $b_i/(b_i+g_i)$. Kruskal-Wallis tests were used to evaluate differences in detection probabilities among altitudes and patterns. Fisher's exact test for contingency tables was used to evaluate the effect of altitude and pattern on the proportion of trials in which all otters were detected and the proportion of trials in which otters exhibited disturbance behavior. All statistical tests were conducted at the .05 significance level.

Reproduction

Estimates of annual reproduction, as indicated by ratios of independent to dependent sea otters and patterns of habitat use were obtained from small (<10m) boat surveys. Surveys were conducted from 13 August through 11 September 1991.

Sample units corresponded to the coastline transects established by Irons et al. (1988) and extended offshore out to the 100m depth contour or 1/2 the distance to the opposing shoreline, whichever was less. A subset of sample units was randomly selected to be surveyed in each of 3 strata. Strata consisted of heavily oiled and non-oiled nearshore habitat in western Prince William Sound and non-oiled habitat in eastern Prince William Sound.

The survey vessel maneuvered about 200 to 300m offshore, and out to the offshore boundry as necessary to observe and classify all otters within each selected sample unit. Boat speed was maintained at less than 15 mph. Surveys were conducted only when viewing conditions are considered good or better (calm to light winds, sea state less than Beaufort 2).

Surveys crews consisted of two observers, including the boat operator. Crews used high resolution binoculars and a Questar telescope. Otters were classified as either dependent or independent. Dependent otters were defined as sea otters smaller

than, and in close association with, an adult. This definition included, but was not limited to, pups in close physical contact, nursing, receiving food from, swimming with or being groomed by an adult sea otter. Independents were defined as all other sea otters. Crews recorded the number of dependent and independent sea otters found in each sample unit.

Each sample unit was classified by coastline physiography and bathymetry into one of six categories. Coastline physiography was categorized as protected bay, open coast, or island. Bathymetry was categorized as either shallow (less than 31m deep for more than 50% of the sample unit's length, 200 to 300m offshore) or deep (greater than 31m deep for more than 50% of the sample unit length). Depth determinations were based on navigational charts and fathometer readings taken during the survey.

Ratios of independent to dependent sea otters were obtained for each stratum and for each habitat type by summing over all sample units within each stratum or habitat type. Proportion of dependent sea otters was calculated for each transect. Kruskal-Wallis tests were used to evaluate differences in proportions among areas. Statistical tests were conducted at the .05 significance level.

RESULTS

Aircraft Evaluation

We conducted 98 trials, observing 329 groups of one or more sea otters for a total of 741 animals in our preliminary evaluation of the Piper Super-cub as a survey platform. Intensive searches resulted in detectability estimates greater than or equal to .90 for all patterns and altitudes investigated (Tables 3.1 and 3.2). All otters were detected in over half of the samples (Tables 3.1 and 3.2). The type of avoidance behavior observed in boat surveys (Udevitz et al. 1990), in which otters leave the search area before the survey platform arrives, was not observed in response to the aircraft. However, on some occasions it was apparent that otters were disturbed and began diving, swimming out of the area, or swimming erratically within the search area in response to the aircraft after it arrived.

For a fixed search pattern, detection probability and proportion of samples in which all otters were detected tended to decrease slightly with altitude, but differences were not significant (Table 3.1). The proportion of samples in which otters appeared to be disturbed by the aircraft was higher at 150 ft than for 300 or 450 ft., but again the difference was not significant (Table 3.1).

Detectability estimates for initial strip counts ranged from .52 to .72 (Figure 1). Detectability increased sharply with the first 2 to 3 circles or ovals after the strip count and continued to increase slightly for the next 3 to 4 circles or ovals. No new otters were ever detected after the 7th circle or oval.

Detection probability and proportion of samples in which all otters were detected tended to increase with decreasing pattern size, but differences were not significant (Table 3.2). There were no significant differences in the amount of disturbance resulting from the search patterns (Table 3.2).

Reproduction

We surveyed 17 transects in non-oiled eastern Prince William Sound, 48 in non-oiled and 54 in oiled habitat in western Prince William Sound, observing 547 independent and 249 independent sea otters. Ratios of independent to dependent sea otters were 72:28, 68:32 and 68:32 in the eastern, western non-oiled and western oiled areas, respectively. Differences in proportions of dependent sea otters among areas were not significant. Dependent to independent ratios for each habitat type represented by 10 or more transects varied from 68:32 to 72:28 (Table 3.3).

DISCUSSION

Aircraft Evaluation

We were not able to detect any effect of altitude on detectability over the narrow range of altitudes we considered. We would expect detectability to decrease substantially at altitudes much greater than those we considered. In general, safety is expected to increase with altitude (for altitudes up to at least 164m). Forty-six meters was considered the minimum altitude safe enough for conducting this type of survey work. We selected an altitude of 92m for conducting the pattern evaluation because it could provide an added margin of safety without appreciably decreasing detectability.

We were also not able to detect any differences in detectability among the intensive search patterns we considered. In the absence of strong differences in detectability, selection of a search pattern could be based on the probability of encountering otters in each search. We hypothesize that this probability decreases with decreasing the size of the search pattern, thus increasing the number of replicated searches necessary to obtain a detection probability estimate with a given level of precision.

Sea otters occasionally exhibited behaviors in response to the aircraft that may have made them more difficult to detect. All of these behaviors have also been observed in response to boats

in boat surveys (Udevitz et al. 1990). These behaviors occurred after the arrival of the survey platform at the search area and included diving and swimming away from the platform. In boat surveys, however, some otters were able to leave the search area before the survey platform arrived and thus avoid any possibility of detection. The aircraft approached search areas more quickly and disturbed otters were not able to leave before the aircraft arrived. This suggests that reductions in detectability due to disturbance by the survey platform may be less of a problem for aircraft than for boats.

Detectabilities were unacceptably low for uncorrected strip counts, but these were substantially improved by subsequent intensive searches using any of the investigated patterns. The data suggest that the most efficient search intensity might consist of 3 circles or ovals after an initial strip count. Almost all of the additional otters are detected during the first 3 circles or ovals. The time and fuel required for more than 3 circles or ovals would probably not justify the slight increase in detectability estimates they would provide. Even with intensive searches, however, not all of the otters were detected. Population size estimates based on correction factors derived from these types of intensive counts can be expected to be negatively biased on the order of 10%. This amount of bias may represent a substantial improvement over previously used methods, but probably cannot be considered negligible for most applications.

Results presented here are based on a preliminary analysis of data from the 1991 field season. We are in the process of refining the analyses reported here and conducting additional analyses, including investigations of

1) the possibility of over-counts by the air crew due to otters entering the area during the intensive search,

2) the effects of otter activity, group size, and viewing conditions on detectability,

3) sightability as a function of distance from the aircraft and otter group size (based on line transect data),

4) the number of systematically (or randomly) located intensive searches required to obtain detectability estimates of a given precision (based on simulations from sea otter distribution data),

5) differences in detectability of independent and dependent sea otters, and

6) the power of these analyses.

Additional work will be required to integrate the results of this study into a survey technique. Such a technique might employ counts of sea otters within strips of sea otter habitat and adjustments to account for missed otters based on intensive searches over portions of the strips. We anticipate designing a trial survey in Prince William Sound and continuing the evaluation of detectability from a Super-Cub in other habitat types in 1992.

Reproduction

Preliminary analysis of data from the reproduction survey suggests that any differences in ratios of independent to dependent sea otters among stratum or habitat types must be small. These ratios may serve as indices of pup production. However, care must be used in interpreting comparisons of these ratios obtained from different areas (e.g., zones or habitat types) because the proportion of sexually mature females may also vary among areas.

Results presented here are based on a preliminary analysis of data from the 1991 field season. We are in the process of completing analyses of

1) statistical differences in independent to dependent ratios among habitat types

2) the patterns of habitat utilization in relation to the availability of habitat types, and

3) the power of these analyses.

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Table 2.1. Estimates of detectability and related parameters for 3 different altitudes (750 m circle search pattern without preceding strip count, circling continued until 5 minutes past last new otter sighting .

		Altitude	
	46m	92m	137m
Trials ^a	13	12	12
Groups ^b	58	43	44
Otters°	133	104	106
Detectability	.92	.91	.90
Detect=1 ^d	.62	.50	.50
Disturbance	.23	.08	.08

^aNumber of trials.

^bTotal number of otter groups observed by ground crews.

'Total number of otters observed by ground crews.

^dProportion of samples with detectability = 1.

^oProportion of samples in which otters appeared to be disturbed by the aircraft activity.

Table 2.2. Estimates of detectability and related parameters for 3 search patterns (strip count followed by circles or ovals until 5 minutes past last new otter sighting, 300 ft altitude).

		Pattern	
-	400 m circle	750 m circle	800 m oval
Trials ^a	20	19	22
Groups ^b	58	40	86
Otters°	113	72	213
Detectability	.96	.93	.90
Detect=1 ^d	.80	.79	.68
Disturbance°	.15	.26	.19 ^f

^aNumber of trials.

^bTotal number of otter groups observed by ground crews.

'Total number of otters observed by ground crews.

^dProportion of samples with detectability = 1.

^oProportion of samples in which otters appeared to be disturbed by the aircraft activity.

^fBased on 21 samples for which the presence or absence of disturbed behavior was recorded.

Table 2.3. Number of sample units (N) and numbers and ratios of dependent and independent sea otters by habitat type (S=shallow open coast; SB=shallow bay; Is= shallow island; D=deep open coast; DB=deep bay; Isd= deep island).

Habitat	N	Independent	Dependent	Ratio
S	27	226	105	68:32
SB	33	226	108	68:32
Is	10	40	16	72:28
D	31	42	16	72:28
DB	9	7	0	100:0
IsD	8	6	4	33:67
Total	119	547	249	69:31



Figure 1. Detection probability as a function of search effort (number of additional circles or ovals after initial strip counts) for 3 search patterns.

Section 3

INTERSECT MODEL OF SEA OTTER MORTALITY

James L. Bodkin and Mark S. Udevitz

SUMMARY

Conceptual development of the intersect model to estimate potential exposure of sea otters to oil along the Kenai Peninsula is Exposure was measured in terms of the amount and complete. duration of oil in an otter's vicinity (gallon*days). Preliminary simulations indicate potential exposure of approximately 1,211 sea otters to some degree of oiling. Potential exposure within this group averged 32,655 gallon*days (SE = 3,902 gallon*days) ranging from 1,100 to 155,100 gallon*days of oil. Refinement of model components is continuing. We anticipate that this model will provide an estimate of sea otter mortality along the Kenai Peninsula resulting from the T/V Exxon Valdez oil spill. The model may may be extended to allow application throughout the spill zone. Extensions will depend on obtaining suitable representations of sea otter abundance and distribution in the additional areas.

OBJECTIVES

Develop a model capable of estimating rates of exposure to oil, degree of oiling and mortality of sea otters associated with oil released from the Exxon Valdez along the Kenai Peninsula, Alaska, 1989.

INTRODUCTION

Following the release and subsequent movement of oil from the T/V Exxon Valdez, live and dead oiled sea otters were observed within Prince William Sound and along the Kenai Peninsula. Oiled sea otter carcasses were retrieved and live oiled otters were captured for transport to rehabilitation centers in Valdez, Seward and Homer. The sum of dead oiled otters plus live oiled otters minus pre-spill dead otters provides a minimum estimate of the number of sea otters coming into contact with spilled oil. It is probable that some additional number of animals became oiled and died and their carcasses remained unrecovered while others may have become oiled and survived.

The purpose of this study is to develop an analytical model, relating exposure of sea otters to oil with subsequent mortality along the Kenai Peninsula. The approach involves estimating the abundance and distribution of sea otters in near-shore and offshore habitat along the Kenai Peninsula at the time of the spill, estimating the level of exposure of each otter, estimating the

degree of oiling received by otters at each exposure level, and estimating the mortality rate associated with each degree of oiling. This information will be integrated by the model to provide an estimate of the total spill induced mortality for this area.

STUDY METHODOLOGY

A computer model developed by NOAA (OSSM) was used to simulate the distribution of oil particles as they traveled through Prince William Sound and along the Kenai Peninsula. The OSSM model traces the movement of 10,000 particles of oil (each representing about 1,100 gallons, and covering about .3 to 1 mi² of sea surface) from their origin at Bligh reef. Under this model, about 1,250 (12%) of the oil particles moved out of Prince William Sound and along the Kenai Peninsula. We used the location of each particle at 3 hour intervals to represent the distribution of oil over time, along the Kenai Peninsula.

The abundance and distribution of sea otters in near-shore and offshore habitat along the Kenai Peninsula at the time the oil passed through was estimated based on the spring 1989 helicopter survey (Douglas et al. 1990). The location of each observed otter was recorded during the survey on large scale maps. These locations were used as an estimate of the distribution of otters at the time of the spill. The total number of otters present was estimated as described by Douglas et al. (1990).

In order to measure exposure, an exposure region was defined for each otter as a circle with radius 1.4 km centered at the otter's location during the survey. This radius represents the average distance sea otters were observed to move between successive radio relocations recorded between 18 and 36 hours apart in California (Ralls et al. 1988). The Ralls et al. (1988) data includes movements of adult and sub-adult male and female sea otters (n=38).

The number of gallons per day times the number of days (gallon*days) that oil was within an exposure region was used as a measure of the exposure of that location to oil. This exposure was estimated for each otter location by counting the number of OSSM oil particles that were within its exposure region during each 24 hour interval, multiplying by 1100, and summing over the time period of the spill. The proportion of the observed otters at each location was used to estimate the proportion of the population with that location's level of exposure.

Data for relating exposure levels to oiling and mortality of otters were collected within two areas of Prince William Sound. The first of these areas was Herring Bay $(60^{\circ} 28'N \ 147^{\circ} \ 45'W)$, on the north end of Knight Island where heavy oiling was observed to persist

over time, most otters were oiled, the degree of oiling was heavy and mortality rates were high. The second site comprised the northeast third of Prince of Wales Passage, including Iktua Bay $(60^{\circ}$ 06'N 148° 00'W), between Evans and Bainbridge Island. This area was lightly oiled along most of the shoreline and oil appeared to pass through in a short time. Most sea otters were either non-oiled or lightly oiled and mortality was relatively low. Similar data are not available for the Kenai Peninsula. We consider these exposure levels and subsequent mortality estimates to possibly be biased low for the following reason. We were not at the capture sites at the time of initial oiling. Animals that suffered immediate mortality were removed from the pool of potential captures prior to our arrival. At the time of initial oiling, many sea otters in Prince of Wales Passage were observed in oil (B. Garrot, pers. comm.) and may have died and drifted out of the area prior to our arrival approximately two days later.

Comparisons of mortality rates between Prince William Sound and the Kenai Peninsula would initially indicate substantial differences between areas. However, sea otter capture efforts did not begin along the Kenai until 29 April, 1989. Bodkin and Weltz (1990) describe a pattern of declining degree of oiling and resultant mortality as the time interval between exposure and capture increased. This pattern led to diminishing sea otter capture efforts in the Sound about 21 April and a shift in the effort to the Kenai Peninsula where initial oiling occurred on or about 1 April. A comparison of the proportions of animals in each oiling category suggests more similar oiling rates within samples captured after more than about 2 weeks of potential exposure (Table 3.1). We suggest the delayed capture response along the Kenai Peninsula contributed to the observed differences in mortality rates between areas.

During the first 3 weeks of April, attempts were made to capture all otters in these areas with dip-nets and tangle-nets (Bodkin and Weltz 1990). Each otter was classified into 1 of 4 categories based on the quantity of oil observed on its pelage at the time of capture. The degree of oiling categories were defined as follows; heavy = complete or nearly complete coverage of the pelage with visible oil, moderate = partial oiling of about 25-50% of the pelage with visible oil, light = oil not easily visible or detectable, or a small proportion (<10%) of the pelage containing visible oil, and none = oil not visually or tactically evident on the pelage. The proportion of animals in these areas exhibiting each of the above degrees of oiling was estimated by the proportion of the of the animals captured or handled in each area that were classified into each category.

A total of 43 sea otters were captured or handled in the Prince William Sound areas, with 11 from Herring Bay and 32 from Prince of Wales Passage. 31 otters were captured with dip-nets and 12 were captured with tangle nets. Tangle nets are usually assumed to obtain an unbiased sample of otters with respect to age and sex (Bodkin and Weltz 1990). Although sample sizes are small, the proportion of oiled otters obtained by each capture method was similar (92% oiled/ 8% non-oiled, dip-nets; 91% oiled/ 9% nonoiled, tangle nets), suggesting that if tangle nets obtained an unbiased sample with respect to oiling, then the dipnet samples were also unbiased.

With the exception of five non-oiled animals that were released after capture, all other captured otters were transported to rehabilitation centers, where they were cleaned and held. Mortality rates were estimated for each of the oiling categories as the proportion of animals that died. We assumed the five non-oiled Sixty-five percent of the observed mortality animals survived. occurred within 5 (65%) days of arrival (mean = 7.1 days; range 0 to 34 days) at a rehabilitation center. Pups born at the rehabilitation facilities, otters with an undetermined oiling status and otters exhibiting obvious non-oil related pathology (eg., paralysis or blindness) were excluded from oiling and mortality rate calculations (Table 3.1). Mortality rates of nonoiled sea otters were not used in the model because selective capture of debilitated animals would have resulted in relatively high mortality among these animals.

The range of exposure levels encountered in the 2 Prince William Sound areas will be estimated by calculating the exposure in gallon*days as described above for 10 randomly distributed points within each area. High exposure will be defined as greater than or equal to the minimum exposure value obtained for Herring Bay. Low exposure will be defined as less than or equal to the maximum exposure value obtained for Prince of Whales Passage. Moderate exposure will be defined as values between these 2 extremes.

The proportion of the estimated total near-shore and off-shore Kenai Peninsula sea otter population in high, moderate and low exposure categories will be determined based on the their estimated exposure values and the scale developed for the Prince William Sound areas. For the portion that have high exposure values, the proportion with each degree of oiling will be estimated using the proportions from Herring Bay. For the portion that have low exposure values, the degree of oiling proportions will be obtained from Prince of Whales Passage. For each of these segments of the population, the total mortality will be estimated by taking each of the products of the total population estimate, the exposure level proportion, a corresponding degree of oiling proportion and its associated mortality rate and then summing over the degree of oiling categories. Overall mortality rates for the high and low exposure categories will be estimated as the total mortality for that portion of the population divided by the size of that portion of the population. The overall mortality rate for the moderate exposure category will be estimated as the mean of the mortality rates for the high and low categories. Total mortality for the moderate exposure category will be estimated as the product of the total population estimate, the moderate exposure level proportion and its associated mortality rate. The total mortality for the Kenai Peninsula otters will be estimated as the sum of the totals for the three exposure categories for the near-shore and offshore habitats.

STUDY RESULTS

Helicopter surveys of the abundance and distribution of sea otters along the Kenai Peninsula during April 1989 counted 1,114 sea otters on transects. 1,083 and 31 sea otters were counted on nearshore and off-shore transects, respectively. These counts provided estimates of 1275 sea otters (SE = 26) in 778 km² of near-shore habitat and 1053 sea otters (SE = 215) in 3,353 km² offshore habitat.

Rates of oiling, degree of oiling and associated mortality rates for Herring and Prince of Wales Passage, with comparisons from sea otters captured throughout Western Prince William Sound and the Kenai Peninsula are presented in Table 3.1.

Results of preliminary intersect simulations indicate 122 of the near-shore sea otter exposure regions (buffers) were intersected with OSSM oil point vectors on one or more days between 24 March and 23 May 1989. These 122 buffers represent about 393 sea otters that encountered potential exposure to oil for one or more days. Potential exposure levels for this group in nearshore habitat averaged 33,583 gallon*days (range 1,100 to 155,100 gallon*days; SE = 4,037 gallon*days). Nine sea otter buffers occurring in offshore habitat were intersected with OSSM oil point vectors. These nine buffers represent about 818 sea otters that encountered potential exposure to oil. Potential exposure for this group averaged 20,044 gallon*days (range 1,100 to 144,100 gallon days; SE = 15,554).

STATUS OF INJURY ASSESSMENT

Progress to date in this study has been the conceptual development of an intersect model to estimate sea otter mortality as a result of exposure to oil along the Kenai Peninsula. Preliminary simulations of the model indicate exposure to about 1211 (52%) of the Kenai Peninsula sea otter population. We are currently developing estimates of exposure levels for the Prince William Sound capture sites.

The near-shore habitats of the Kenai Peninsula represent areas for which have some of the best data on otter populations during the spill. This approach could be extended with a coarser resolution

to other areas of the spill zone for which our information on otter abundance and distribution is not as precise.

We are unable to provide an estimate of our confidence in the value of this model, our its products, at this time. We anticipate this model will produce a reasonable estimate of sea otter exposure to oil and subsequent mortality, given an accurate representation of sea otter abundance and distribution, and relationships of exposure levels to mortality rates.

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Table 3.1 Sea otter mortality rates as related to degree of oiling for Herring Bay, Prince of Wales Pass, all of Western Prince William Sound, for animals captured in Western Prince William Sound after 13 April, and for all animals captured along the Kenai Peninsula (H=heavy, M=moderate) L=light, N=none).

Degree of Oiling		H 	M 	L	N
Herring Bay (N=11) Mortality	N ¥	5 .45 .80	4 .36 .50	2 .18 .50	0 0 0
Prince of Wales Pass (N=32) Mortality	N ¥	0 0 0	3 .09 .33	19 .59 .37	10 .31 .30
Combined Mortality (both sites)		.80	.43	.38	.30
All Western Prince William Sound (N=118) Mortality	N ¥	50 .42 .76	14 .12 .50	44 .37 .31	10 .09 .70
Western Prince William Sound ¹ (N=27) Mortality	N X	3 .11 .33	5 .19 .20	16 .62 .06	3 .08 .67
Kenai Peninsula (N=124) Mortality	N &	3 .02 .00	19 .15 .11	70 .56 .11	32 .26 .12

¹sea otters captured after 13 April, 1989 (13 or more days after initial oil exposure).

Section 4

SEA OTTER PREY SELECTION AND FORAGING SUCCESS IN WESTERN PRINCE WILLIAM SOUND, ALASKA, 1991

Angela Doroff and James Bodkin

SUMMARY

Sea otter prey were determined by visual observation and scat analysis in western Prince William Sound in areas of heavy, moderate and no shoreline oiling. Patterns of prey use did not differ from historic data at Green and Montague Islands. Principle prey species were clam, mussel and crab for all study areas; clams were the most frequently retrieved prey item. There were no significant differences in forage success rates or mean number of prey retrieved per dive among areas. Mussel and clam were the most frequently observed prey in sea otter scat at Green Island. Prey species were collected at each site to assess variability in contamination of individual prey items. Samples currently wait hydrocarbon analysis. Sea otters were not observed to reject prey after retrieval in areas of heavy shoreline oiling. Continued exposure of sea otters to hydrocarbons through their prey remains likely in the spill area.

OBJECTIVES

1. To describe prey species and the relative frequency with each which prey species is consumed by sea otters in 3 areas affected by the <u>Exxon</u> <u>Valdez</u> oil spill.

2. To determine foraging success rates in each of three study areas

3. To compare prey species and foraging success rates from the Green Island area to historic data from the same region.

4. Estimate mean size and determine approximate caloric value per prey item.

5. To collect tissue samples of sea otter prey to assess hydrocarbon contamination in individual prey items.

INTRODUCTION

Sea otters have occupied southwestern Prince William Sound since at least the early 1950's and had expanded their range north to include western Knight Island by approximately 1970 (Lensink 1962; Garshelis 1986). In a study of sea otter forage behavior conducted during 1980-81 at Green Island (Fig. 1) sea otters spent approximately 50% of a 24 hr period foraging, and during the winter months (November-April) foraging activity increased (Garshelis 1986). Garshelis (1986) and Johnson (1987) concluded the sea otter population in western Prince William Sound at Green Island was food resource limited.

Sea otters commonly prey on a variety of benthic marine invertebrates that inhabit coastal waters ranging in depth from the intertidal to approximately 20 fathoms (Kenyon 1969). Principal prey species identified in Prince William Sound in the past include clam, mussel and crab (Calkins 1978; Garshelis 1986; Johnson 1987). Relative to other marine mammals, sea otters have a high metabolic demands to maintain their body temperature in cold North Pacific waters (Costa and Kooyman 1984). Juvenile and adult sea otters consume between 20-30% of their body weight per day (Kenyon 1969).

Damages to the nearshore benthic community resulting from the Exxon Valdez oil spill (EVOS) may influence the recovery of sea otters in affected habitats. Probable mechanisms of influence include 1) availability, and decreased food 2) consumption of prey To contaminated by hydrocarbons. evaluate hydrocarbon contamination in Prince William Sound, shellfish and coastal sediments have been sampled in portions of the sea otter range by the coastal habitat and fish/shellfish damage assessment studies. Preliminary results have shown elevated levels of hydrocarbons in sediments and bivalve mollusks (A. Weiner, unpub rpt 1991). However, information is lacking on the variation in contamination of individual prey items in areas where sea otters are known to forage.

Our objectives were 1) to describe prey species and the relative frequency with which each prey species is consumed by sea otters in 3 areas affected by the <u>Exxon Valdez</u> oil spill, 2) to determine foraging success rates in each of three study areas and compare prey species and foraging success rates from Green and Montague Islands area to historic data from the same region, and 3) to collect tissue samples of sea otter prey in areas where foraging occurs to determine hydrocarbon content of prey.

METHODS

Study Areas

Sea otter prey species were determined at three areas within western Prince William Sound, Alaska. Study areas included portions of Montague, Green and Squirrel Islands (Fig. 1) and were selected by the following criteria: 1) relative degree of shoreline oiling (based on DEC shoreline oiling maps) with Squirrel, Green and Montague Islands representing heavy, heavy to moderate and no shoreline oiling, respectively 2) capture location of sea otters from which blood and subcutaneous fat samples were available from living sea otters and 3) sample sites of other NRDA studies collecting sea otter prey species and sediments for hydrocarbon analysis. All areas contained primarily soft bottom sediments with broad expanses of shallow (< 10m) subtidal areas. Each study area was delineated by the viewing distance which observers could identify sea otter prey species from shore-based vantage points (maximum viewing distance approximately 1km). The approximate area monitored for foraging animals was 4, 5 and 6 km² for Squirrel, Green and Montague Islands, respectively. During the course of the study, survey flights were conducted to estimate the number of sea otters within and adjacent to the study areas. A survey flight was conducted 21 May, 1991 for Green Island in a 185 Cessna aircraft at airspeed of 60 kph and altitude of approximately 153m. The aircraft maintained a distance of 100m from shore. When large groups of sea otters were encountered, the aircraft circled until the observer felt the count was accurate. A second survey was conducted 22 July, 1991 for the Squirrel and Montague Island study Methods were the same as the previous survey with the areas. exception that the aircraft used was a Piper PA-18 Supercub.

Prey Identification

Observations were made by trained individuals with the aid of high resolution telescopes (Questar Corporation, New Hope, PA) and 10X binoculars. Data recorded included sex, age class of focal animal (adult or juvenile), number of prey and relative prey size (A: < 5 cm, B: \geq 5 to < 7 cm , C: \geq 7 cm to < 9 cm, D: \geq 9 to < 12 cm and E: \geq 12 cm), dive interval, surface interval, and prey item to lowest identifiable taxon. Repeated dives were recorded for a focal animal until a maximum of 50 identifiable prey items were observed per individual or until the animal was lost or discontinued foraging. Radio-implanted sea otters from damage assessment studies were used as focal animals when feasible. Focal animal selection was random when more than one otter was feeding at an observation site. Adult animals were categorized as male, independent female or female with a pup. Juveniles were identified as small dark-headed otters estimated to be less than 24 months of age. A minimum sample of 500 identified prey items were recorded for each study area. Data was collected only during daylight hours, during all tidal cycles. Tidal state and estimated water depth from navigational charts were recorded for all observations.

Study areas were sampled consecutively: 17 April - 11 May Green Island, 14 June - 3 July Squirrel Island, and 13 July - 24 July Montague Island. Each study area had several vantage points and observers rotated through them at irregular time intervals (weather and viewing conditions influenced our ability to have fixed schedules).

Data analysis

Kruskal-Wallis tests were used to test differences among the proportion of clam, crab and mussel in the diet of adult sea otters of varying sex classes (independent female, female with pup, male, and adult of unknown sex) and areas for all bouts having a records of \geq 10 dives.

A successful dive was defined as any dive which obtained a prey item. Success rates were determined for all bouts of adult and juvenile sea otters having ≥ 10 dives. Unsuccessful dives were included in the data analysis; dives of unknown result were not included. An arcsin transformation was used to normalize distributions and a 2-way analysis of variance was used to test for differences among sex classes and areas. Differences in success rate by age (adult and juvenile) were tested by t-test for samples of equal variance. Data were normalized by an arcsin transformation.

Number of prey per dive was averaged per bout for all adult animals by sex class (independent female, female with pup, male, and adult of unknown sex) and area. Dives containing mussel were excluded for this analysis due to their clumped growth pattern and the difficultly in obtaining accurate counts on a per dive basis. Dives of unknown result were disregarded in this analysis. A 2-way analysis of variance was used to test differences among sex class and area.

Scat Analysis

During the period 20 April-2 May 1991, 253 sea otter scat samples were examined in the field along 8.5 km of beach on western Green and Barrier Islands (Fig.1). For each scat sample encountered the following information was recorded: location, single or multiple defecation, and the species of prey represented (when determined) and relative percentage each species contributed to the entire scat. Notes were made on unidentified, noncalcareous material within the scat. Data were compiled as frequency of occurrence of each species (Fig.2).

Prey Collection

Within each study area all forage observations collected were plotted on a gridded map with Universal Transverse Mercator coordinates. A polygon was drawn around the outermost foraging observations with a 100 m^2 grid overlay. Ten 100 m^2 plots were chosen at random for each area. Sites for clam collection within each plot were selected by a random boat anchor drop in each plot and searching with the aid of SCUBA (Self Contained Underwater Breathing Apparatus) for desired species from the anchor point in concentric circles. Water depth averaged 8m (range 5 - 12m). Clams were excavated with a venturi dredge (Keene Engineering Northridge, CA) powered with a Briggs and Staton 8 hp motor on the rear deck of a 7.6 m Boston Whaler. Divers attempted to locate 3 butter clams (Saxidomus giganteus) at each plot. This could not be accomplished at all plots and the design was broadened to include all known prey species of clam. Clams were brought to the surface in mesh dive bags, wrapped in chemically cleaned aluminum foil (Acetone and Hexane washed) and frozen in a small propane freezer aboard the 7.6 When more than 3 clams were retrieved from a m Boston Whaler. single plot, clams were randomly selected for hydrocarbon analyses. When more than 1 species was collected, additional samples were randomly selected to compare hydrocarbon levels in species among plots and areas. For hydrocarbon analysis, bivalves were thawed and all tissue was placed in chemically clean jars. Seventy-nine bivalve tissue samples currently await analysis.

RESULTS

Prey Analysis

Portions of 38 forage bouts (29 adult and 9 juvenile) were recorded at Green Island. At Squirrel Island, portions of 69 forage bouts (68 adult and 1 juvenile) were recorded and, at Montague Island, portions of 72 forage bouts were observed (69 adult and 3 juvenile).

Clams were retrieved on 67% (n=405), 34% (n=858) and 46% (n=735) of all successful dives recorded for adults at Green, Squirrel and Montague Islands respectively and were consistently the most frequently retrieved prey item. Figure 3 summarizes adult sea otter prey as the number of dives retrieving clam, crab, mussel, other invertebrates, and unidentified prey for each study area. Juveniles (for all areas pooled) also retrieved clams most frequently (51% n=427). The ratio of dives retrieving mussel/dives retrieving clam for juveniles and adults was 0.61 and 0.25, respectively.

Table 1 summarizes variation of prey use by adults at each study

area. No significant differences were detected in independent females, females with pups, males and adults of unknown sex in percentage of dives retrieving clam, crab, and mussel at Green (P=0.78, P=0.45, P=0.82) or Montague (P=0.21, P=0.88, P=0.74) Islands. Significant differences were detected in the percentage of dives retrieving clams (P=0.01) among sexes for Squirrel Island; no significant differences were detected for crab (P=0.24) or mussel (P=0.38). No significant differences were detected among areas for the percentage of dives retrieving clam (P=0.09), however, significant differences were detected for crab (P=0.001) and mussel (P=0.03).

Greater than 70% of clams identified to species in all areas were butter clams (<u>Saxidomus giganteus</u>). The most frequent prey items identified to species were blue mussel (<u>Mytilis edulis</u>) for Green and Squirrel Islands and Helmet crabs (<u>Telmessus spp.</u>) for Montague Island.

Mean success rates for adult and juvenile sea otters in all study areas were 90% (n=82) and 92% (n=10), respectively. Success rates did not differ significantly (P=0.49) between adult and juvenile sea otters. At Green Island, adult and juvenile success rates were 87% (n=16) and 98% (n=8), respectively. Forage success did not differ significantly among sex classes (P=0.12) or among areas (P=0.29). The interaction between site and sex class was not significant (P=0.09).

Mean number of prey retrieved per dive averaged 1.2, 1.0 and 1.3 for Squirrel, Green and Montague Islands, respectively. No significant differences were detected among areas (P=0.11) or sex classes (P=0.83). The interaction between site and sex class was not significant (P=0.30).

Blue mussel (<u>M. edulis</u>) was observed most frequently (60%) in 253 sea otter scat examined on northeastern Green Island (Table 2). Clam species, primarily <u>Protothaca staminea</u> and <u>S. giganteus</u>, were identified in 46% of all scat examined. Crab and other small invertebrates were found in 19% and 20%, respectively. Of scats containing a single prey type, 76 were mussel, 23 clam and 13 contained one of the following: scallop (<u>Chlymys sp.</u>), snail (<u>Natica sp.</u>), Cockle (<u>Clinocardium sp.</u>), limpet (<u>Notoacmea scutum</u>).

Prey Sampling

A total of 79 prey were sampled from 20 plots from Green (n=7), Squirrel (n=7) and Montague (n=6) Islands. The species composition of the sample is as follows: 23 <u>Macoma spp.</u>, 20 <u>S. giganteus</u>, 24 <u>P.</u> <u>staminea</u>, 6 <u>Humilaria kennerleyi</u>, 5 <u>Gari californica</u> (species yet to be verified), and 1 <u>Serripes groenlandicus</u>. Mean shell length and wet meat weight for sampled species are as follows: <u>Macoma</u> 42 mm and 4.7g, <u>S. giganteus</u> 44 mm and 10.7g, <u>P. staminea</u> 40 mm and 8.9g, <u>H. kennerleyi</u> 46 mm and 8.9g, <u>G. californica</u> 46 mm and 8.6g, and <u>S. groenlandicus</u> 56 mm and 16.2g.

All samples have been submitted for hydrocarbon analysis; results are not yet available.

DISCUSSION

Sea otter diet has remained relatively constant over time at Green Island and Montague Strait. Calkins (1978) reported major prey in Montague Strait were clam (<u>S. giganteus</u>), crab (<u>Telmessus spp</u>.) and sea stars (<u>Evasterias troschelii</u>). Johnson (1987) reported clam and mussel as the most important prey for adult and subadult animals at Green Island. Clams were the most frequently identified prey retrieved and mussel the most frequent prey in scats in our study. Study results are also consistent with the findings of Garshelis (1986) with the exception that clams comprised only 67% of the diet in our study compared to > 75% at Green Island during 1980.

Differences in prey use were not detected among sexes with the exception of percentage of clam at Squirrel Island. Johnson (1987) found highly significant differences among age classes; our sample sizes were insufficient to test age related differences in prey consumption. Juveniles consumed a higher proportion of mussels than did adult animals in our study areas. Mean number of prey per dive at Green Island was 1.0 (n=35, sd=0.38). Johnson (1987) was unable to determine mean prey per dive because clam size was too small to accurately determine the number of prey being consumed. Mean prey per dive were calculated only for those dives which prey number could be determined.

Forage success did not differ among sex classes or among areas in our study. Johnson (1987) observed success to be relatively constant in areas of different demographic composition with slightly lower success for prey items requiring excavation.

Sea otters in Prince William Sound haul out regularly throughout the year but more frequently in the winter months (Johnson 1987). Scat deposits collect on snow berms above mean high tide making it feasible to estimate over-winter diets by scat analysis. Scat analysis provides useful information on prey items often too small to be identified visually. Late April and early May scats were examined at Green Island. Mussels were the most frequent prey item identified in scat samples, followed by clam (Fig. 2). For large clams, shells are removed prior to ingestion but small clams (< 5 cm) are often ingested with shell parts (Johnson 1987). Visual observation data suggest few large clams are retrieved by sea otters (79% < 5cm, 20% 5-7 cm, and 1% 7-9 cm, n=479). Overall mean shell length of clams encountered while collecting prey was 43 mm (range 31-61mm). These observations are consistent with Garshelis (1986) and Johnson (1987) views that western Prince William Sound may be food resource limited.

Visual observations and scat analysis yield different results when comparing prey species frequency in sea otter diet. By visual observation, clams were most frequently retrieved and by scat analysis mussel was most frequent. We believe these differences reflect differences in methodology rather than a seasonal shift in prey preference. Scat analysis is biased toward small prey items where nondigestable parts are ingested and against prey where no hard parts are ingested. Shells of mussel tend to be thinner and consistently < 5cm in length where as shells of clams 5cm and greater will less likely be ingested.

Resampling of unmarked foraging otters likely occurred in our study area. Comparing aerial survey counts of each study area to numbers of bouts sampled provides a general measure of potential resample intensity. Aerial count to sample size ratio for Green, Squirrel and Montague were: 27/38, 23/69 and 65/72, respectively. Within or adjacent to the Green, Squirrel and Montague Island study areas were 9, 20 and 30, respectively, tagged sea otters from capture efforts during 1989-90. Forage observations occurred on only 2 tagged animals (1 at Green Island and 1 at Squirrel Island) throughout the entire observation period and repeat observations did not occur for these tagged individuals.

Sea otters demonstrated the ability to detect and reject prey contaminated by paralytic shellfish poisoning (Kvitek et al in press). We postulated sea otters may also be able to detect prey contaminated by hydrocarbons, however, foraging otters were not observed to reject retrieved prey at the surface in any of the study areas.

The results of this study indicate that sea otters have not changed their food habits in western Prince William Sound over the past decade. Sea otters continue to rely principally on clams and mussels as prey. High levels of hydrocarbons have been observed in both clams and mussels in the spill area (NRDA damage assessment studies-ref?). While sea otters continue to rely on these two prey items, continued exposure to hydrocarbons is likely in areas affected the EVOS. Analysis of prey samples collected during this study will allow us to estimate hydrocarbon exposure rates and to determine variation among individual prey items at each study site.

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Table 4.1 Median proportion of dives retrieving clam, crab, and mussel for adult sea otters (<u>Enhydra lutris</u>) in Prince William Sound, Alaska, 1991.

Green Island					Squirrel Island				Montague Island			
Sex Class	Clam	Crab	Mussel	* N ¹	Clam	Crab'	Mussel	N ¹	Clam	Crab*	Mussel*	NI
Independent female	0.41	0.0	0.18	2	0.29 [@]	0.0	0.72	10	0.36	0.03	0.0	6
Female with pup	0.42	0.0	0.0	7	0.15 [@]	0.63	.053	18	0.53	0.88	0.0	15
Independent male	0.79	0.0	0.0	14	0.58 [@]	0.16	0.16	6	0.5	0.05	0.0	9
Sex unknown	0.41	0.0	0.0	1	0.96@	0.0	0.0	2	0.76	0.10	0.0	3

¹ Number of forage bouts containing ≥ 10 dives for each sex class.

[@] Significant differences among sex classes in the proportion of dives retrieving clam (P = 0.01).

* Significant differences among areas in the proportion of dives retrieving crab (P< 0.01) and Mussel (P = 0.03).



Figure**4**3. Composition of prey by visual observation in western Prince William Sound. AK.

Figure 4.1. Sea otter forage study area locations in western Prince William Sound, Alaska, 1991.



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Figure⁴2. The composition of Mussel, Clam, Crab and other small invertebrates in 253 sea otter scats examined during 20 April to 2 May 1991, in Western Prince William Sound, Alaska.



- 1) <u>Mytilis</u> edulis
- 2) Frequency of <u>Saxidomas giganteus</u>, <u>Protothaca staminea</u>, <u>Humilaria kennerleyi</u>, and unknown shell fragments.
- 3) Species not identified.
- 4) SSCL equivalent to: Scallop (<u>Chlymys sp.</u>), Snail (<u>Natica sp.</u>), Cockle (<u>Clinocardium sp.</u>), Limpet (<u>Notoacmea scutum</u>), and other unidentified snail shell fragments.

Number of Scats

Table 4.2 The composition of Mussel, Clam, Crab and other small invertebrates in 253 sea otter scats examined during 20 April to 2 May 1991 in Wester Prince William Sound, Alaska.

The number of scat containing the following percentages of	Mussel, Clam	, Crab and SSCL
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Total	Percer	ntage of							
	<u>100</u> %	90%	75%	50%	25%	10%	5%	in sample	total-No. of scat
Mussel ¹	76	24	10	13	14	6	10	153	(60%)
Clam ²	23	22	8	15	21	10	17	116	(46%)
Crab ³	0	2	2	5	21	10	7	47	(19%)
SSCL^4	13	4	5	8	4	6	10	58	(20%)

¹ <u>Mytilus</u> edulis

- ² <u>Protothaca</u> <u>staminea</u>, <u>Saxidomas</u> <u>giganteus</u>, <u>Humilaria</u> <u>kennerleyi</u>: includes unidentified shell fragments.
- ³ Species not identified
- ⁴ SSCL equivalent to: Scallop (<u>Chlymys sp.</u>), Snail (<u>Natica sp.</u>), Cockle (<u>Clinocardium sp.</u>), limpet (<u>Notoacmea scutum</u>), and other unidentified snail shell fragments.

Section 5

POST-SPILL SEA OTTER MORTALITY IN PRINCE WILLIAM SOUND

Daniel H. Monson

SUMMARY

We compared the age class distributions of beach cast sea otter carcasses found in Prince William Sound from 1974 through 1986 (pre-spill), in 1989 as a direct result of the Exxon Valdez oil spill (spill year) and in 1990 and 1991 (post-spill). There were significant differences between pre-spill and spill year distributions and pre-spill and post-spill distributions. Spill year and post-spill distributions were not significantly different. The proportion of prime age animals dying in western Prince William Sound increased in both the spill year (1989) and post spill years (1990 and 1991). The observed differences represent a shift from a pre-spill composition of principally young and aged animals to an increased proportion of prime-age animals during and following the spill. The observed changes in the age distributions of dying sea otters suggest a prolonged, spill related affect on at least the western Prince William Sound sea otter population. Carcass recovery rates and the sex ratio of dying animals did not differ pre- to post-spill.

OBJECTIVES

- 1. To test the hypothesis that pre-spill levels of mortality (number of carcasses per linear kilometer of beach surveyed) are not significantly different from post-spill levels of mortality in Prince William Sound.
- 2. To test the hypothesis that the proportion of female carcasses found on beaches in pre-spill surveys is not significantly different from proportions found in post-spill beach surveys in Prince William Sound.
- 3. To test the hypothesis that the proportion of prime-age carcasses found on beaches in pre-spill surveys is not significantly different from proportions found in post-spill beach surveys in Prince William Sound.

INTRODUCTION

Following the EXXON VALDEZ oil spill (EVOS) sea otter populations in the path of the spill experienced severe mortality resulting in the collection of hundreds of carcasses (DeGange and Lensink, 1990). Acute and chronic exposure to hydrocarbons persisting in the environment may result in additional losses. Characteristics of mortality for these populations (i.e. mortality rates, age-class and sex composition of mortality) can be monitored using information gained from beach cast carcasses.

Kenyon (1969), Johnson (1987) and Bodkin and Jameson (1991) document and describe patterns of mortality for sea otter populations. Kenyon (1969) describes the age structure of dying sea otters at Amchitka Island in the western Aleutian chain as consisting primarily of juveniles (70%). Among adults found dead or dying, most showed signs of aging. Data from California suggest a similar mortality pattern with the majority of annual mortality comprised of sub-adult sea otters (Bodkin and Jameson, 1991). Johnson (1987) collected beach cast carcasses in Prince William Sound from 1974 through 1984. At Green Island, 163 carcasses were collected during this period. Aged carcasses were comprised of 45% juveniles (0 to 1 year) and 40% adults greater than 8 years of age (figure 5.1). This area has supported sea otters since at least the 1950's (Lensink, 1962) and has a well established population. In an area at the front of an expanding sea otter population in eastern Prince William Sound, Johnson collected only 25 carcasses during the same period. Juveniles comprised 17% of aged animals and 78% were greater than 8 years of age (Fig. 5.2). Thus, under normal conditions, age composition of beach cast carcasses would likely show similar bimodal patterns with prime-age mortality remaining relatively low and rates for juveniles and old sea otters higher depending upon habitat conditions such as the availability of resources.

The most sensitive indicator of change in mortality patterns may be the proportion of prime-age carcasses recovered. An increase in this proportion indicates altered mortality rates for the reproducing population which has the greatest impact on the stability of the population.

This study examines patterns of sea otter mortality as indicated by beach-cast carcasses in 1989, 1990 and 1991, following the EVOS and compares those data with pre-spill data collected by Johnson (1987).

STUDY METHODOLOGY

A beach survey, following methodology described by Johnson (1987), was conducted in portions of Prince William Sound between May 10 and May 19, 1990, and 12 April and 2 May, 1991. Study sites were the same as Johnson's (1987) and included Green Island, within the spill zone, and the Port Gravina area of northeastern Prince William Sound, outside the spill zone. Agencies and individuals known to be working in these areas were contacted to assure that carcasses would not be removed before our systematic search. above sea level. Trials were conducted in sets of 3, with one trial at each altitude, in random order, within each set. All altitude evaluation trials were conducted using a 750m circle intensive search pattern. In this pattern, the aircraft was piloted along the circumference of a 750m diameter circle while the aerial observer viewed the circumscribed area. The aircraft pilot generally was unable to assist in visual observation due to the technical aspect of the survey procedures. Aircraft speed was maintained as close as possible to 28m/sec (55mph). The pilot used a stopwatch, airspeed and minute of turn to define the 750m diameter circle (128 seconds to complete, 32 seconds through each quadrant). The location and orientation of the circle was indicated by markers positioned at the vantage point by the ground crew. The aerial observer recorded the time, location, group size, number of pups and activity of each new sea otter or group of sea otters observed. Circling was continued until 5 minutes had elapsed without any new otters being observed.

Pattern evaluation trials were conducted using 3 different intensive search patterns in conjunction with a strip count. The same aircraft, but different pilots, were used for the altitude and pattern evaluations. All pattern evaluation trials were conducted at an altitude of 92m above sea level and at a speed of Each trial began with a strip count in which the plane 28m/s. flew along one edge of a 400m strip while the aerial observer recorded the location, group size, number of pups and activity of each sea otter or group of sea otters observed in the strip. Width of the strip was determined by the aerial observer using distance indicators marked on the wing struts. The length of the strip was either 400m, 750m or 800m, depending on the subsequent search pattern. Immediately following the strip count, the plane began one of three search patterns over the strip that had just The aircraft was piloted along the circumference been counted. of either a 400m diameter circle, a 750m diameter circle, or a 400m x 800m oval while the aerial observer viewed the circumscribed area. Selection of the search pattern was made by the ground crew according to the distribution of sea otters and the physiography of the coastline, while attempting to obtain an equal number of trials for each pattern. Ground crews indicated the location and orientation of each strip, circle and oval with markers at the vantage point. The pilot used techniques analogous to those developed for the 750m circle to maintain each of the other 2 search patterns. The aerial observer recorded the circle or oval number, location, group size, number of pups and activity of each new sea otter or group of sea otters observed Intensive search patterns were continued during the search. until 5 minutes had elapsed without any new otters being observed.

At the end of each day, ground and aerial crews compared the mapped locations of all observed otters. For the otters present in trial i, i=1,...,r, when the aircraft arrived, the number

Rates of carcass deposition (number/kilometer of beach surveyed) were determined for systematic beach surveys in the Green Island area. Proportions of female and prime age carcasses were determined for Green Island and the Port Gravina area. Prime-age was defined as between 2 and 8 years of age, based on pre-spill beach-cast carcass collections (Johnson 1987).

The date of collection, carcass location, relative position on the beach, and condition, along with the sex and estimated age were recorded for each carcass. Only carcasses determined to have been deposited over the previous winter/spring were used in the analysis. This determination was based upon the presence of hide and cartilage on skeletal remains and the location of remains with respect to the previous years layer of dead vegetation (above or below). Skulls and baculum (when present) were collected. A premolar was removed for age analysis (Garshelis 1984).

The U. S. Fish and Wildlife Service requested the reporting and collection, when possible, of all sea otter carcasses found during the second and third year's spill monitoring and clean-up efforts. Carcasses were reported or collected throughout the spill area by various federal and state agencies as well as private groups and individuals during spring and summer, 1990 and 1991. When possible, each carcass was examined and a tooth collected by Service biologists. Only data from carcasses actually recovered from beaches are included in this analysis.

All teeth were sectioned and read (aged) by Gary Matson (Matson's lab, Milltown, MT) and many were read independently by James Bodkin (AFWRC, Anchorage, AK). Each assigned a quality to the reading as a measure of confidence in the age. Matson's age was used for all analyses with the second reading serving as verification.

Differences between age distributions and sex ratios were tested using Fisher's Exact Test (2-tailed) or a standard Chi-Square test. Where significant differences were not seen between post-spill years, data were pooled. In addition, if differences were not observed between Green Island data and incidental data collected in other areas of western Prince William Sound, data were pooled as all post-spill mortality data. Pooled data were then tested against pre-spill data and data collected at the time of the spill. Differences were considered significant at p < 0.05.

STUDY RESULTS

Systematic Beach Survey

Eighteen carcasses were recovered from 54 kilometers of coast at Green Island (oiled area) in 1990 for a recovery rate of 0.33/km. Sex was determined for 14 carcasses and 6 (43%) were female. Fifteen carcasses provided teeth for aging. Five (33%) were

juveniles, 3 (20%) prime-age and 7 (47%) older than 8 years of age. In 1991, 9 carcasses were recovered on Green Island beaches for a recovery rate of 0.17/km. Sex was determined for 4 carcasses and 3 (75%) were female. Eight provided a tooth and 4 (50%) were juvenile and 4 (50%) were prime-age. There were no significant differences between carcass recovery rates and sex composition between years or between pre and post-spill data at Green Island. The difference between 1990 vs 1991 age distributions approached significance (p = 0.08) and the pooled post-spill age distribution (Fig. 5.3) was not significantly different from pre-spill data. Six recently deposited carcasses were recovered from 53 kilometers of coast in the Port Gravina (non-oiled) area in 1990 for a recovery rate of 0.11/km. Of these 4 (67%) were female. All 6 were aged and 2 (33%) were juvenile, 3 (50%) were prime-age and 1 (17%) was older than 8 years of age. No carcasses were recovered on these beaches in 1991. Carcass recovery rates could not be determined for pre-spill years. The proportion of females in the post-spill sample was significantly higher than the pre-spill sample (p < 0.001). Sample sizes were insufficient to test for differences in age structure between pre and post-spill.

Incidental Carcass Collection

Multi-agency beach assessment crews examined approximately 1105 km of beach in the spill zone of western Prince William Sound during the spring of 1990 (Dan Edging, pers. com.). In addition, approximately 500 km of beach were walked by the Prince William Sound Conservation Alliance (PWSCA) looking for oil and clean-up debris (David Janka, pers. com.). The over-lap between beaches walked by the PWSCA and the survey crews is not known but assumed to be great. These groups collected 52 carcasses from western Prince William Sound beaches of which 46 came from oil impacted areas (Table 5.2) for a recovery rate of between 0.03 - 0.04/km. The PWSCA collected 21 (40%) with the rest collected by EXXON crews, the U.S. Fish & Wildlife Service (USFWS), the US Forest Service (USFS), Alaska Dept. of Fish & Game (ADF&G), the Alaska Dept. of Environmental Conservation (ADEC) and various private individuals and groups. Of the 52 carcasses collected incidental to other work in the Sound 37 provided teeth for aging. Twelve (32%) were juvenile, 18 (49%) were prime-age and 7 (19%) were greater than 8 years of age. Sex was determined on 15 individuals and 3 (20%) were female.

Multi-agency beach survey crews examined 385 km of beach in the spring of 1991 (1991 Work Program Status, Continuing Programs Aug. 15, 1991). The PWSCA participated in these surveys but did no additional surveys. Eight carcasses were reported in 1991 and 7 were collected for a recovery rate of 0.02/km. Six carcasses have been aged and 1 (17%) was a juvenile, 3 (50%) were prime age and 2 (33%) were greater than 8 years old.

No significant differences were observed in sex ratios or age distributions between years for incidental samples. Combined post-spill age distribution (Fig. 5.4) was significantly different from pre-spill data at Green Island (p < 0.0001). There were no significant differences between age distributions or sex ratios for combined post-spill systematic and incidental data. We pooled incidental and systematic sex ratio and age for western Prince William Sound for 1990 and 1991. Age (Fig. 5.5) and the sex ratios for carcasses collected post-spill for all of western Prince William Sound were significantly different than pre-spill data from Green Island (p = 0.0005 and p = 0.05 respectively). Combined sex ratio data for western Prince William Sound are summarized in Table 5.2.

DISCUSSION

Sea otter mortality patterns vary between areas and over time as a result of factors such as seasonal weather patterns, yearly changes in otter distribution and abundance, sexual segregation of the population within an area and condition of the supporting habitat (ie. resource availability). Kenyon (1969), Johnson (1987), and Bodkin and Jameson (1991) using beach cast carcass data describe a general pattern of mortality for sea otter populations. A major proportion of dying sea otters are sub-adults (45%-70%). Mortality rates of prime age animals are generally low (5%-15%). Mortality rates for young of the year and aged animals increases with length of time otters have occupied an area. The majority of losses are observed in late winter/early spring when old and young animals in food-limited areas appear stressed. A peak in mortality has also been observed during late summer and early fall corresponding to the peak pup weaning periods of populations in California (Bodkin and Jameson, 1991). These studies conclude that data from beach cast carcasses can be a valuable indicator of trends in mortality for sea otter populations.

Systematic Beach Survey

Sea otter densities were considered to be relatively stable at Green Island at the time of Johnson's (1987) study. Rates of carcass deposition on Green Island beaches ranged from 0.13/km to 0.63/km ($X^2 = 0.31/km$, sd = 0.20). The spring 1990 rate of carcass deposition 0.33/km and mean post-spill rate (1990 and 1991 combined) of 0.26/km fall near the pre-spill mean. However, 1989-1991 winter densities of sea otters in the Green Island area may have been significantly lower than pre-spill densities due to loses observed during the EVOS, thus this rate may be high relative compared to sea otter abundance. Burn (Section 1, this document) found consistently lower densities of sea otters in coastal portions of Prince William Sound affected by oil. At this time we do not have information on winter densities of sea otters in this area. An accurate determination of winter densities will be important in interpreting pre and post-spill carcass deposition rates.

The overall 1976-1986 sex ratio of 99 sea otter carcasses identified to sex and collected at Green Island was 59 females (60%). This proportion did not change when pups were excluded from the analysis (47 female of 78 = 60%). This may reflect the areas status as a female area (Johnson, 1987). The post-spill sex ratio of carcasses identified to sex was not significantly different between years. The combined post-spill proportion of female carcasses (10 of 19 = 53%) does not significantly differ from the pre-spill proportion of females (60%).

The number of carcasses recovered in the non-oiled area of Port Gravina totaled only 25 over 10 years (2.5/year) prior to the EVOS (Johnson, 1987). The 6 carcasses recovered in the 1990 spring survey may reflect an increase in mortality expected as the length of time sea otters have occupied the area increases. Pre-spill data were collected during the first years of re-occupation as an advancing front of male sea otters moved through this area (Johnson, 1987). Mortality rates can be expected to have been low at this time as resources were abundant (Garshelis et. al., 1986). However, no carcasses were collected in the Spring of 1991, thus several more years of data collection may be needed before inferences can be made about mortality rates in the area.

One (6%) of 18 carcasses identified to sex in the Port Gravina area by Johnson (1987), was identified as female. This reflects the dominance of males in the area which was, at the time, the expanding front of the Prince William Sound sea otter population (Johnson, 1987). This contrasts with the Green Island pre-spill data of 60% (n = 99) female sea otters (significantly different at p < 0.001) which had a long established population at the time and contained female areas (Johnson, 1987). The spring 1990 beach survey recovered 67% (n = 6) female carcasses in the Port Gravina and is not significantly different from that observed at Green Island prior to the EVOS. This indicates the move of females into the area as the expanding population front of males moved east.

A summary of Johnson's pre-spill mortality data for the Green Island area indicates the proportion of prime-age carcasses found on beaches ranged from 0 to 29% with a 9 year mean of 14% (n=142 carcasses, sd=0.097) (Fig. 5.1) The spring 1990 and 1991 age distributions were not significantly different (but approached significance, p = 0.08). The basic age structure found in the spring 1990 survey (33% juvenile, 20% prime, 47% old, n = 15) was similar to the over-all average pre-spill structure at Green Island (Fig. 5.1). However, the 1991 distribution of 50% juvenile and 50% prime age (n = 8), had higher a proportion of prime age than any single year of pre-spill data. However, sample size was inadequate

for a valid statistical comparison. The 1990 and 1991 age data were pooled (Fig. 5.3) and the combined age distribution of carcasses recovered at Green Island was not significantly different from pre-spill data. Several years of data may be needed before trends can be determined.

The pre-spill proportion of prime-age carcasses was low in the Gravina area of eastern Prince William Sound. Only 5% (n = 18) of aged carcass collected from 1974 to 1984 were prime-age (Fig. 5.2). The post-spill recovery of 50% (n = 6) prime-age carcass in spring 1990 may indicate an unusual year of sea otter mortality in this area, however the small sample size precluded valid statistical testing. No carcasses were collected in this area in 1991 and a number of years data will be needed before trends can be determined.

Age distribution of carcasses collected prior to the EVOS at Green Island and the Port Gravina area were significantly different (p = 0.01) due to an even lower rate of prime-age mortality in the Port Gravina pre-spill data. This finding along with what seem to be lower carcass deposition rates (none could be determined for prespill data at Port Gravina) are consistent with the conclusions drawn above for differences in mortality between long occupied vs newly established sea otter habitat.

Incidental Carcass Collection

Ages have been determined for 351 carcasses collected in Prince William Sound at the time of the EVOS. Of these, 152 (43%) were prime-age animals (Fig. 5.6). Mortality during the spill was expected to have been equal for all age classes and thus provide an estimate of the sea otter age distribution of the population at that time. If oil spill effects did not persist in the Sound during the winter of 1989/1990, we would expect the age distribution of mortality to return to the "normal" bimodal pattern (i.e., mortality dominated by old and very young animals), observed by Johnson prior to the spill in Prince William Sound.

The age distribution of carcasses collected by beach crews since the spill is not significantly different between years. And, the combined incidental post-spill data is not significantly different from the age distribution of carcasses collected at the time of the spill (Figs. 5.4 and 5.6). In addition, age data from incidental and systematic carcass collections were not significantly different. In fact, the combined age distribution of carcasses collected post-spill in western Prince William Sound (incidental and systematic) is very similar to the distribution observed at the time of the spill (Figs. 5.5 and 5.6) and both distributions are significantly different from pre-spill data (p = 0.0005 for postspill data and p < 0.0001 for data collected at the time of the spill). This may be an indication of continuing damages to the sea otter population in oiled areas of Prince William Sound.

These data indicate all age groups may be experiencing equal mortality rates (ie. the probability of death is equal for juvenile, prime age and old sea otters) in portions of western Prince William Sound. This conclusion has serious implications for recovery of sea otter populations within the spill area. If this trend continues, the reproductive portion of the population may suffer increasingly significant losses and impede or prevent sea otter populations from recovering in the near future (next 10 years) or possibly precipitate a further decline in populations within the spill area.

Combined post-spill sex ratios for all of western Prince William Sound (Table 5.2) were significantly different from pre-spill data collected at Green Island and data collected at the time of the spill (38% female for post-spill data vs 60% female for both prespill data and data collected during the spill, p = 0.05 and p =0.02 respectively). If the sex ratio from carcasses collected at the time of the spill is representative of actual sex ratio of the population of western Prince William Sound, these data indicate unequal levels of mortality for male and female otters post-spill. Sea otters do segregate by sex and this finding may reflect some factor influencing mortality in male areas more than female areas. However, male carcasses are more likely to be identified due to the presence of a baculum even when heavily scavenged. Sex has not been determined for the majority of incidentally collected carcasses and only 34 (41%) of 83 post-spill carcasses have been identified to sex. Thus, the post-spill data may be biased towards identification of male carcasses.

The number of carcasses collected in 1991 declined from 1990. However, this is most likely a reflection of the intensity of effort between the two years. Survey crews and the PWSCA searched more beach in 1990 than 1991 (1600 km vs 385 km). In addition reporting of carcasses (particularly heavily scavenged carcasses) was not as consistent in 1991 as the previous year. Thus the change in number of carcasses recovered may reflect reduced effort rather than a change in mortality between 1990 and 1991.

Changes in post-spill sea otter mortality may be reflected by changes in the age and sex ratios or beach cast sea otter carcasses and, to a lesser extent, the rate of carcass deposition on Prince William Sound beaches. The results of this study demonstrate that the age structure of the dying western Prince William Sound sea otter population after the oil spill is comprised of a significantly higher proportion of prime age animals than prior to the spill. The extent of the effects of this pattern of mortality on the recovery of the sea otter population remain unknown.

Reliability of Age Estimates

For a sample of 339 teeth read by two independent readers, the readings were highly correlated (r = 0.94, p < 0.0001). If only the teeth for which both biologists were highly confident of the reading are considered (n = 137), the correlation improves to r = 0.97 (p < 0.0001). In addition, these two biologists along with one other have performed blind readings of ten known-age teen from California and there was a high degree of accuracy and precision among readers. Questions on aging of sea otters using tooth cementum annuli should be addressed to Jim Bodkin at the Alaska Fish and Wildlife Research Center (tel. 907 786-3450).

STATUS OF INJURY ASSESSMENT

The prime-age sea otter mortalities observed through spill year 3 are evidence of an abnormal pattern of mortality continuing in parts of Prince William Sound. A high proportion of the prime-age carcasses were recovered in areas that received heavy oiling in both 1990 and 1991. The abnormal mortality pattern may be associated with oil contamination in these areas and indicate continuing damage to the population. Further monitoring of carcass deposition, with systematic searches of areas heavily impacted by oil, will provide data on the persistence and specific causes of this abnormal mortality pattern.

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1990 and 1991 locations of sea otter carcasses found incidental to beach monitoring and clean-up activities in Prince William Sound. Table 5.1

Area	Spill Zone	Number
Knight Island	Yes	15
Eleanor Island	Yes	5
Naked Island	Yes	11
Perry Island	Yes	7
Long Island	Yes	1
Craft Island area	Yes	4
Evans Island	Yes	5
Latouch Island (SW Prince William Sound)	Yes	2
Green Island ¹	Yes	28
Stockdale Harbor (Montague Island)	No	1
Whittier area (NW Prince William Sound)	No	5
Valdez area	No	1
Unknown location within the Sound	Unk	1
Total collected		86

Total collected

¹Includes 27 carcasses collected during systematic beach walks.

Table 5.2 Sex ratios of beach cast sea otter carcasses collected in western Prince William Sound before, during, and after the EXXON Valdez oil spill.

	Fe	male	P	Total		
Time Period	number	percent	number	percent	<u>collected</u>	
Pre-Spill ¹	59	60%	40	40%	99	
1989 ²	312	60%	208	40%	520	
Post-Spill ²	13	38%	21	62%	34	

¹Includes onln carcasses collected at Green Island. ²Includes carcasses collected all over western Prince William Sound.



Figure 5.1. Age distribution of 142 sea otter carcasses collected on Green Island, Prince William Sound, Alaska, 1974-1984. (note: Ages > 10 combined at 10)



Figure 5.2. Age distribution of 18 sea otter carcasses collected in eastern Prince William Sound, Alaska, 1974-1984.



Figure 5.3. Age distribution of 23 sea otter carcasses collected on Green Island, Prince William Sound, Alaska, 1990-1991.



Figure 5.4. Age distribution of 43 sea otter carcasses collected in western Prince William Sound, Alaska, 1990-1991 (excluding Green Island).



Figure 5.4. Age distribution of 43 sea otter carcasses collected in westerm Prince William Sound, Alaska, 1990-1991 (excluding Green Island).



Figure 5.5. Age distribution of 66 sea otter carcasses collected in western Prince William Sound, Alaska, 1990-1991 (including Green Island) ..



Figure 5.6. Age distribution of 437 sea otter carcasses collected in Prince William Sound, Alaska, 1989.

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Section 6

HYDROCARBON CONTAMINATION OF SEA OTTER TISSUE SAMPLES

D. Mulcahy & B.E. Ballachey

SUMMARY

Sea otter tissue samples have been collected to measure hydrocarbon contamination following exposure of the otters to oil from the Exxon Valdez, and to evaluate associations between hydrocarbon levels and other information gathered on the sea otters, including clinical and histopathologies, and life histories. Samples are from sea otters that entered the rehabilitation centers, carcasses collected in the oil zone post-spill (1989, 1990 and 1991), and otters captured in Prince William Sound post-spill (1989, 1990 and 1991). Sea otters in southeast Alaska were sampled to provide a non-exposed control group. To date, over 400 samples have been analyzed. The hydrocarbon data, which consist of 93 primary analytes, calculated ratios and summations, have been entered into the sea otter data base, and an exploratory analysis of the data initiated. Results on approximately 200 more samples, including the control group, are pending. Full analyses of the data are expected to identify potential linkages between hydrocarbon contamination and deleterious effects associated with oil exposure noted in concurrent NRDA studies on sea otters. In addition, published literature on hydrocarbon levels in mammalian tissues is scant and this study will provide unique and valuable reference data on hydrocarbons in the event of future exposures of marine mammals to petroleum.

OBJECTIVES

The objective of this study is to evaluate levels of petroleum hydrocarbons and hydrocarbon metabolites in sea otter tissue samples as indicators of exposure to oil from the <u>Exxon Valdez</u>, and to relate contaminant levels to other data gathered on the sea otters, including clinical records, gross, clinical and histological pathologies, and rates of reproduction and survival. To address this objective, samples from the following groups are being examined:

- A. Sea otters from the rehabilitation centers: 1) otters which died at the centers, with tissue samples collected at necropsy (see Appendix A), and 2) fat from sea otters captured in oiled areas, treated at the rehabilitation centers, and released into the wild in 1989 (see Appendix C).
- B. Carcasses of sea otters recovered following the spill (1989, 1990 and 1991). For many of these, necropsy and

histopathological information is also available (see Appendix A).

- C. Blood and fat from sea otters captured in eastern and western Prince William Sound for radiotelemetry and bioindicator studies in 1989, 1990 and 1991 (see Appendices B and D).
- D. Sea otters from south-east Alaska, representing a non-oiled control. In 1991, blood samples were collected from live caught otters, and tissue samples were collected from carcasses taken by a subsistence hunter.

The nine hypotheses listed below were initially formulated regarding hydrocarbon levels in tissue samples collected from sea otters at the rehabilitation centers, and from carcasses recovered following the spill.

- Hypothesis 1. A. Levels of hydrocarbon contamination do not vary within tissue types between animals. B. Levels of hydrocarbon contamination do not vary across tissues from a single animal.
- Hypothesis 2. Hydrocarbon levels in sea otter tissues are independent of the degree of oiling.
- Hypothesis 3. Hydrocarbon levels in sea otter tissues do not vary as a result of duration of potential exposure to oil.
- Hypothesis 4. Hydrocarbon levels in sea otter tissue are independent of time between spill and exposure.
- Hypothesis 5. Among lightly oiled sea otters exposed to oil for more than 20 days, hydrocarbon levels in tissues do not vary as a result of age of oil.
- Hypothesis 6. A. Hydrocarbon levels in sea otter carcasses recovered in 1990 from western Prince William Sound do not differ from controls.
 B. Hydrocarbon levels in sea otter carcasses recovered in 1990 from western Prince William Sound do not differ from those in carcasses of lightly oiled sea otters from western Prince William Sound in 1989.
- Hypothesis 7. Hydrocarbon levels in tissue samples from sea otters exhibiting no external sign of oiling do not differ from control animals.
- Hypothesis 8. Fetuses and pups of oiled female sea otters do not exhibit elevated hydrocarbon levels in tissues.

Hypothesis 9. Hydrocarbon levels in tissues of lightly oiled sea otters that died shortly after arrival at rehabilitation centers (within 3-4 days) do not differ from those of lightly oiled animals that survived 3-4 weeks.

INTRODUCTION

Alkane and aromatic hydrocarbons in tissues of sea otters which died during and after the EXXON VALDEZ oil spill, and sea otters captured in the course of concurrent NRDA studies are being analyzed. These data are expected to identify which sea otters collected alive or dead were exposed to oil from the spill and which sea otters have levels of petroleum hydrocarbons in their tissues that exceed normal background levels. Such information is critical in interpreting mortality and morbidity data being collected in several studies. Hydrocarbon data on individual sea otters and on similar groupings of sea otters will eventually be collated with gross and histopathological data obtained through necropsy of dead sea otters, and with demographic, life history and movement patterns of living sea otters.

Extensive laboratory analysis has been done of alkane and aromatic hydrocarbon levels in sea otter tissues submitted during these investigations. Scant information exists in the published literature for the levels of hydrocarbons to be considered normal in marine mammal tissues, the metabolic alteration of ingested and absorbed petroleum hydrocarbons which produced altered hydrocarbon products in tissues, and the level of non-petroleum, biogenically produced hydrocarbons. Interpretation of the detailed data obtained for the many analytes obtained in our studies is complicated by the lack of guidance provided by the published Much of the analysis done during our studies will literature. undoubtedly provide pathways for future investigation of similar events.

STUDY METHODOLOGY

Hydrocarbon Analyses

Samples were submitted through NRDA Technical Services Study Number 1 for hydrocarbon analyses. Two major classes of petroleum hydrocarbons, 1) alkanes or aliphatic compounds, and 2) aromatic compounds, are the focus of the analyses done on the samples. Alkane hydrocarbons contain only single bonds between carbon atoms and may be normal, branched or cyclic. Aromatic hydrocarbons consist of various combinations of the six-carbon benzene ring.

Hydrocarbon analysis of submitted tissue and fluid samples yield concentrations of 28 alkane and 39 aromatic primary analytes. In addition, seven alkane and 19 aromatic ratios or summations of the primary analytes are then calculated, for a total of 93 data points from each sample (Tables 6.1 and 6.2). While the primary analytes carry important information about each sample, the ratios and summations may offer a simpler first approximation of the data.

Alkane and aromatic hydrocarbons are widespread in nature. Some are produced by marine and terrestrial organisms, some are introduced into the environment from oil seeps and weathering of petrogenous rock and some come from sources of human pollution other than the EXXON VALDEZ oil spill. Determination of the source of hydrocarbons present within an animal is complicated, and made more difficult by the animal's metabolism of the hydrocarbons for storage and elimination. As general guidelines, tissues from nonexposed animals contain moderate levels of biogenic hydrocarbons such as C17, pristane, and odd carbon chain normal alkanes (C25 through C33). Oil is indicated in the alkanes by high levels of phytane and an constant level series of odd and even carbon chain normal alkanes. Oil is indicated in the aromatics by alkylated naphthalenes, phenanthrenes and dibenzothiophenes. The presence of the unresolved complex mixture (UCM) and the calculated carbon preference index (CPI) is an excellent indicator of the presence of degraded oil. Table 6.3 provides a brief explanation of some of the hydrocarbon measures considered to be of value in detecting exposure to petroleum.

Data Analyses

The hydrocarbon data is generally not normally distributed, but is characterized by a preponderance of low values with points extending through a $3-4 \log_{10}$ range. A \log_{10} transformation is being evaluated to change the distribution of the data from one dominated by measurements in the lowest intervals to one which is more normally distributed (Snedecor and Cochran, 1967; Humphrey et al. 1987).

Samples

Tissue samples were collected according to protocols established in Technical Services Study Number 1. For the hypotheses listed above, results of analyses on the following animals and tissues are being examined:

Hypothesis 1. Ten sea otter carcasses were selected that had been obtained in Western Prince William Sound, assessed as having the same degree of oiling (heavy) and all of which had died before 11 April 1989. The following tissues were collected and submitted for hydrocarbon analysis: liver, kidney, brain, muscle, intestine, fat (from six animals only) and testes (from two animals only). Levels of the alkane and aromatic hydrocarbons are being compared between tissue types of each animal (Hypothesis 1B) and the same organ compared among sea otters (Hypothesis 1A).

- Hypothesis 2. Animals in this group fit the following criteria: all were collected between 2 April 1989 and 9 April 1989, all were obtained in Western Prince William Sound, and all spent between 0 and 7 days at rehabilitation centers before dying. Four sea otters were heavily oiled and seven otters were lightly oiled. Liver tissue was submitted for analysis.
- Hypothesis 3. Animals selected for this test were controlled for degree of oiling (heavy or moderate), collected during the same period (for each group), and were brought to rehabilitation centers before dying. Six animals were considered to have been exposed to oil for less than 2 days and five animals for 8 to 11 days. Liver tissue was submitted for analysis.
- Hypothesis 4. Moderately and heavily oiled sea otters were selected for this test. The animals were collected over a time gradient following the spill. The time gradient was established by collecting animals from three different geographic locations at different distances from the origin of the spill. Ten animals were collected from western Prince William Sound (these were the same animals used in Hypothesis 1), eight animals from the Kenai Peninsula, and eight from Kodiak Island. Liver tissue was submitted for analysis.
- Hypothesis 5. Lightly oiled sea otters were collected from three different geographic locations (western Prince William Sound, Kenai Peninsula and Kodiak Island). These otters had been exposed to oil for less than 20 days. The oil at the three locations is presumed to have a differing hydrocarbon composition and different physical properties due to aging. Liver, hair and intestinal tissues were submitted for analysis.
- Hypothesis 6. Hydrocarbon levels in tissues of sea otters found dead in the second year of the oil spill will be compared to the levels obtained from analysis of tissues from 11 sea otters collected by a subsistence hunter in southeast Alaska (Hypothesis 6A). Five sea otter carcasses recovered in Prince William Sound in 1990 will be used. The hydrocarbon levels in the 1990 sea otter carcasses will also be compared to the levels in tissues of lightly oiled sea otters that died in rehabilitation centers in 1989, the first year of the spill (Hypothesis 6B). Seven sea otters have been selected from 1989 to be compared with the five sea otters from 1990. Liver tissue was submitted for analysis.
- Hypothesis 7. Ten otters were selected from those collected in 1989 that showed no external signs of oiling. The hydrocarbon

levels found in their liver tissues will be compared to the levels in the liver tissues of the 11 subsistence-collected sea otters from southeast Alaska.

- Hypothesis 8. Hydrocarbon levels in liver samples of nine sea otter neonates or fetuses born to oiled females in rehabilitation centers are to be compared with levels in livers from fetuses and neonates from unoiled female sea otters.
- Hypothesis 9. Liver tissue was collected from 7 sea otters that died within 3 to 4 days following their arrival in rehabilitation centers. Hydrocarbon levels will be compared to levels in liver tissue from animals that survived in rehabilitation centers for 3 to 4 weeks before dying.

The control group consists of 11 sea otters killed by a subsistence hunter in southeast Alaska in the spring of 1991. Liver, kidney, muscle, and fat were collected. These tissues await submission for hydrocarbon analysis.

Female sea otters and sea otter pups were captured in 1989 and 1990 for instrumentation as part of the NRDA radiotelemetry studies, and fat and blood samples were collected at the time of surgery. Details of the capture and sampling methods are presented in Appendices B and D. Blood was also collected from adult male sea otters caught in 1990 for a NRDA study on bioindicators of oil exposure. The sea otters were captured in eastern Prince William Sound (considered the non-oiled control area), and within or immediately adjacent to the oil spill zone (treatment area) in western Prince William Sound. Areas peripheral to or on the edge of the spill zone in western Prince William Sound where sea otters were captured included Port Chalmers and Stockdale Harbor on Montague Island, and Channel and Little Green Islands near Green Island. Hydrocarbon analyses are complete on samples from 55 adult female otters, but are not yet complete on the pups or adult males.

Forty-five sea otters captured and treated at the rehabilitation centers were radio-instrumented prior to release in the summer of 1989. Fat tissues were collected from these animals at the time of surgery, and hydrocarbon analyses are complete. A summary of the survival, reproduction and movements of these otters is presented in Appendix C.

Necropsies have been done on all sea otter carcasses that were in suitable condition. Pathologists certified by Board of the American College of Veterinary Pathology performed the necropsies and completed the histopathology for carcasses recovered in 1989. Tissues from 51 oiled sea otters and six unoiled sea otters that died in the rehabilitation centers were taken for histopathological examination (Appendix A). An additional 214 necropsies were done on frozen sea otter carcasses to determine gross pathological changes. A suite of tissues was sampled from the carcasses at the time of necropsy, but only a subset (predominately liver samples) has currently been analyzed. Remaining samples are maintained in frozen storage pending the need for additional analyses.

STUDY RESULTS

Most of the hydrocarbon analyses completed to date have been on liver, fat and blood tissues. Some hypotheses required the use of other specific tissue types, and the submission and completion of hydrocarbon analyses on these samples have been given priority. Although many additional samples have been collected, it is not anticipated that analysis of them all will be required. A subset of the samples in storage will be submitted in 1992 to complete the data set and strengthen the value of the statistical analysis.

Data on over 400 samples has been entered into the sea otter data base, and an exploratory data analysis is underway to provide guidance in determining the variables of greatest value for identifying hydrocarbon exposure. Due, however, to the complex nature of the data (93 variables; Tables 6.1 & 6.2), the lack of previous studies providing guidelines, and the incomplete hydrocarbon analyses on certain samples, including the control group, preliminary results are not presented in this document.

Blood and fat samples collected from sea otter pups have been submitted for hydrocarbon analyses but results have not yet been returned. Data analyses will be conducted when these data are complete.

Fat samples were obtained from sea otters that were captured in oiled areas, treated at the rehabilitation centers, and instrumented with a radio for telemetry prior to release in 1989. These samples have been analyzed, and data analysis is pending.

For sea otter carcasses on which gross, clinical and histological pathology is available, tissues from several organs have been analyzed, and additional samples are being submitted to coordinate the sample analysis with the histopathological study on these carcasses. Data analyses will be completed at a later date.

The components and concentrations of hydrocarbons present in sea otters in and around the spill area must be evaluated against a control group of sea otters from well outside the spill area. Tissues from a group of 11 sea otters from southeast Alaska were obtained in the spring of 1991 from a subsistence hunter. The use of sea otters from southeast Alaska as controls is predicated on that geographic area being "pristine," at least on the basis of large, known oil spills. The control tissues have not yet been analyzed, but will be submitted in early 1992. Without the data describing the levels of hydrocarbons in these otters, the interpretation of hydrocarbon levels in the tissues from animals

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within the spill zone must be limited to comparisons of levels and the presence or absence of specific hydrocarbon components. Conclusive statements as to the contribution of the oil spill to total body loads of hydrocarbons and the presence of specific components of the hydrocarbon series must be made cautiously until control data is available.

Most studies of oil in the marine environment have dealt with the persistence of oil in sediment and in marine invertebrates (Cretney et al. 1987a; 1987b). A few studies have focused on fish (Al-Saad 1990; Luquet et al. 1983). Very little information is available on uptake and metabolism of petroleum hydrocarbons in mammalian systems, and almost nothing has been published on the "background" levels of alkane and aromatic hydrocarbons found in marine mammals living in non-oiled environments. Mammals do have advanced metabolic systems for handling toxic compounds, and if an animal survives an acute encounter with oil, it has the capacity to metabolize the hydrocarbons that are systemic. For example, the action of inducible mixed function oxidase (MFO) enzymes converts the hydrocarbons to water-soluble forms for excretion (Engelhardt 1982). This means that the form of the oil constituents are altered from the original to allow storage and elimination. Because of the paucity of data concerning the metabolism of petroleum hydrocarbons in mammalian systems, research into how the mammalian system handles oil contamination and how petrogenic hydrocarbon compounds are metabolically altered for storage and elimination would be beneficial.

STATUS OF INJURY ASSESSMENT

An exploratory analysis of hydrocarbon data is underway to test a number of hypotheses concerning the presence, detectability and composition of petroleum residues in organs of sea otters killed during or shortly after the oil spill. More than 400 samples, primarily liver, fat and blood, have been analyzed to date. Samples of other tissues have been collected and will be submitted to augment the results already obtained.

Preliminary results are not presented at this time in part due to the complex nature of the data and the risks inherent in drawing conclusions prior to complete analysis of all available data. To interpret the results, control samples against which samples from oiled areas can be compared are needed; these samples are slated for analysis in early 1992. Further examination of the data will focus on the biological significance of the differences noted by relating the findings to clinical, pathological and life history information on the otters, as available.

Hydrocarbon analyses of several categories of samples are pending:

1)

Control samples (tissues) from south-east Alaska

- 2) Blood and fat from sea otter pups and adult males in Prince William Sound
- 3) Fat from sea otters which survived treatment at the rehabilitation center
- 4) Additional tissues from sea otter carcasses, to coordinate with the histopathological studies.

Although the threat of exposure to oil spills and other sources of petroleum continues for many marine mammals, published information on the metabolism of petroleum hydrocarbons in mammalian systems is Research should be encouraged into how the mammalian minimal. system handles oil contamination and how petrogenic hydrocarbon compounds are metabolically altered for storage and elimination. The lack of any prior data on hydrocarbon levels in mammalian tissues has increased the difficulty of analyzing the data gathered following the 1989 oil spill. Assuming the appropriate data analyses can be completed (dependent on continuing funding in 1992), the hydrocarbon contaminant data collected as part of the NRDA process on sea otters should prove to be of major importance for examining linkages between exposure and recovery of the sea otter populations in oiled areas. Furthermore, the data set will be of exceptional value in provision of reference baselines for sea otters and other marine mammals in the event of future oil spills or other events involving exposure to petroleum compounds.

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Table 6.1. Interpretation of Hydrocarbon Data.

- 1. TOTAL ALKANES. The best comparison will be to the total alkanes found in organs from the unoiled, control animals. Until those analyses are available, use >1 μ g/g (>1000ng/g) as a threshold level for determining the presence of oil.
- ODD: EVEN CARBON NUMBER. This ratio, which should apply to the high molecular weight n-alkanes (C25 through C34), should be 1 in petroleum and >1 if from natural sources. Use <1 as an indicator of oiling.
- 3. C17, PRISTANE AND ODD CARBON CHAIN NORMAL ALKANES (C25 THROUGH C34). Biogenic aliphatic hydrocarbons.
- 4. PHYTANE AND A SERIES OF ODD AND EVEN CARBON NUMBER ALKANES. Petrogenic aliphatic hydrocarbons. Their presence is an indicator of oil contamination but levels should be compared to unoiled controls.
- PRISTANE: PHYTANE. This ratio should be very high (≈20?) for uncontaminated material. Levels of about 1 to 3 are indicative of oil contamination.
- 6. ALKYLATED NAPHTHALENES, PHENANTHRENES AND DIBENZOTHIOPHENES. Petrogenic aromatic hydrocarbons. Their presence is an indicator of oil contamination but levels should be compared to unoiled controls.
- 7. UNRESOLVED COMPLEX MIXTURE (UCM). Petrogenic hydrocarbons. Its presence is an indicator of oil contamination but levels should be compared to unoiled controls.
- C18: PHYTANE. This ratio can be used as an indicator of biodegradation of oil. Use ≥1 as an indicator of recent oiling.
- 9. CARBON PREFERENCE INDEX (CPI). Equal to 1.0 for petroleum and <3.0 can be used to indicate oil contamination.

ALKANES:

1.	C10	11. C20	21.	C30 31. UNRESOLVED COMPLEX MIXTURE
2.	C11	12. C21	22.	C31 (UCM)
3.	C12	13. C22	23.	C32 32. ODD: EVEN
4.	C13	14. C23	24.	C33 33. PRIS:PHY
5.	C14	15. C24	25.	C34 34. C18: PHYTANE
6.	C15	16. C25	26.	PRISTANE 35. CARBON PREFERENCE INDEX
7.	C16	17. C26	27.	PHYTANE (CPI)
8.	C17	18. C27	28.	TOTAL ALKANES
9.	C18	19. C28	29.	TOTAL ODD ALKANES
10.	C19	20. C29	30.	TOTAL EVEN ALKANES

AROMATICS:

1.	NAPHTHALENE	33.	CHRYSENE
2.	C1-NAPHTHALENE	34.	C1-CHRYSENE
з.	C2-NAPHTHALENE	35.	C2-CHRYSENE
4.	C3-NAPHTHALENE	36.	C3-CHRYSENE
5.	C4-NAPHTHALENE	37.	C4-CHRYSENE
6.	TOTAL METH. NAPHTHALENES	38.	TOTAL METH. CHRYSENES
7.	TOTAL NAPHTHALENES	39.	BENZO(b) FLUORANTHENE
8.	BIPHENYL	40.	BENZO(k) FLURANTHENE
9.	ACENAPHTHYLENE	41.	BENZO (e) PYRENE
10.	ACENAPHTHENE	42.	BENZO (a) PYRENE
11.	FLUORENE	43.	PERYLENE
12.	C1-FLUORENE	44.	IDENO([1,2,3-cd)PYRENE
13.	C2-FLUORENE	45.	DIBENZO(a, h) ANTHRACENE
14.	C3-FLUORENE	46.	BENZO(g,h,i)PERYLENE
15.	TOTAL METH. FLUORENES	47.	TOTAL PARENT AROMATICS
16.	PHENANTHRENE	48.	TOTAL METHYLATED AROMATICS
17.	ANTHRACENE	49.	LOW MOL. WT. AROMATICS
18.	C1-PHENANTHRENE	50.	BENZO(a)PYR.+ BENZO(e)PYR.
19.	C2-PHENANTHRENE	51.	HIGH MOL. WT. AROMATICS
20.	C3-PHENANTHRENE	52.	TOTAL AROMATICS
21.	C4-PHENANTHRENE	53.	TOTAL PARENTS: TOTAL METH. ARO.
22.	TOTAL METH. PHENANTHRENES	54.	NAPHTHALENE: TOTAL METH. NAPS.
23.	TOTAL PHENANTHRENES	55.	FLUORENE: METH. FLUORENES
24.	DIBENZOTHIOPHENE	56.	PHENANTHRENE:METH. PHENS.
25.	C1-DIBENZOTHIOPHENE	57.	DIBENZOTHIOPHENE:METH. DIBS.
26.	C2-DIBENZOTHIOPHENE	58.	CHRYSENE: METH. CHRYSENE
27.	C3-DIBENZOTHIOPHENE		
28.	TOTAL METH. DIBENZOTHIOPHI	ENES	
29.	FLUORANTHENE		
30.	PYRENE		
31.	C1-FLUORANTHENE-PYRENE		
32.	BENZ[a]ANTHRACENE		

Table 6.3. Calculated Ratios and Summations.

ALKANES:

- 28. TOTAL ALKANES=(C-EVEN+C-ODD+UCM+PRISTANE+PHYTANE)
- 29. TOTAL ODD=(C11+C13+C15+C17+C19+C21+C23+C25+C27 +C29+C31+C33)
- 30. TOTAL EVEN=(C10+C12+C14+C16+C18+C20+C22+C24+C26+C28 +C30+C32+C34)
- 32. ODD: EVEN= (TOTAL ODD/TOTAL EVEN)
- 33. PRIS: PHY=(PRISTANE/PHYTANE)
- 34. C18: PHYTANE=(C18/PHYTANE)
- 35. CARBON PREFERENCE INDEX (CPI)=(2[C27+C29]/[C26+C28+C30])

AROMATICS:

- 6. TOTAL METH. NAPHTHALENES=(C1-NAPHTHA+C2-NAPHTHA +C3-NAPHTHA+C4-NAPHTHA)
- 7. TOTAL NAPHTHALENES=(NAPHTHA+TOTAL METH. NAPHTHALENES)
- 15. TOTAL METH. FLUORENES=(C1-FLUORENE+C2-FLUORENE +C3-FLUORENE)
- 22. TOTAL METH. PHENANTHRENES=(C1-PHENANTHRENE +C2-PHENANTHRENE+C3-PHENANTHRENE+C4-PHENANTHRENE)
- 23. TOTAL PHENANTHRENES=(PHENANTHRENE +TOTAL METH. PHENANTHRENE)
- 28. TOTAL METH. DIBENZOTHIOPHENES=(C1-DIBENZOTHIOPHENE +C2-BIBENZOTHIOPHENE+C3-DIBENZOTHIOPHENE)
- 38. TOTAL METH. CHRYSENES=(C1-CHRYSENE+C2-CHRYSENE +C3-CHRYSENE+C4-CHRYSENE)
- 47. TOTAL PARENT AROMATICS (SELECTED) = (NAPHTHA+FLUORENE +PHENANTHRENE+DIBENZOTHIOPHENE+CHRYSENE)
- 48. TOTAL METH. AROMATICS (SELECTED)=(TOTAL METH. NAPHTHALENES+TOTAL METH.FLUORENES+TOTAL METH. PHENANTHRENES+TOTAL METH. DIBENZOTHIOPHENE)
- 49. LOW MOL. WT. AROMATICS=(NAPHTHA+TOTAL METH. NAPHTHALENES+BIPHENYL+ACENAPHTHALENE+ACENAPTHENE +FLUORENE+TOTAL METH.FLUORENE+PHENANTHRENE +ANTHRACENE+TOTAL METH. PHENANTHRENE)=TOTAL AROMATICS FROM NAPHTHA TO C4-PHENANTHRENE)

Table 6.3 continued. Calculated Ratios and Summations.

50. BENAPYR + BENEPYR=(BEN[A]PYR+BEN[E]PYR)

- 51. HIGH MOL. WT. AROMATICS=(CHRYSENE+TOTAL METH. CHRYSENES +DIBENZOTHIOPHENE+TOTAL METH. DIBENZOTHIOPHENES +FLUORANTHRENE+PYRENE+C1-FLUORANTHENE-PYRENE +BEN[A]ANTHRACENE+BEN[B]FLUORANE+BEN[K]FLUORANE +PERYLENE+I123CDPYRENE+D.B.AHANTHRACENE +B.GHI PERYLENE+{BENAPYR+BENEPYR})
 - =TOTAL AROMATICS FROM DIBENZOTHIOPHENE TO B.GHI PERYLENE)
- 52. TOTAL AROMATICS=(LOW MOL. WT. AROMATICS+HIGH MOL. WT. AROMATICS)
- 53. TOTAL PARENTS: TOTAL METH. AROMATICS=(TOTAL PARENT AROMATICS/TOTAL METH. AROMATICS)
- 54. NAPHTHALENE: TOTAL METH. NAPS.=(NAPHTHALENES/HIGH MOL. WT. AROMATICS)
- 55. FLUORENE: METH. FLUORENES=FLUORENE/TOTAL METH. FLUORENE)
- 56. PHENANTHRENE: METH. PHENANTHRENES=(PHENANTHRENES/TOTAL METH. PHENANTHRENES)
- 57. DIBENZOTHIOPHENE: METH. DIBENZOTHIOPHENES= DIBENZOTHIOPHENES+TOTAL METH. DIBENZOTHIOPHENES)
- 58. CHRYSENE:METH. CHRYSENE=(CHRYSENE+TOTAL METH. CHRYSENES)

Section 7A

Clinical and Clinical Laboratory Correlates in Sea Otters Dying Acutely in Rehabilitation Centers

A. H. Rebar

INTRODUCTION

Following the oil spill caused by the grounding of the Exxon Valdez oil tanker in Alaska's Prince William Sound, a major effort was made to treat oiled sea otters in rehabilitation centers in an attempt to return them to the wild. Sea otters presented to the centers were classified as heavily oiled, moderately oiled, lightly oiled, or unoiled on the basis of visual examination. This report details the clinical findings and principal laboratory changes in otters which died shortly (within the first 10 days) after entering the rehabilitation centers regardless of degree of oiling or approach to therapy. Another report (Appendix A) describes the gross pathologic and histopathologic alterations in the same as well as other otters dying acutely in the centers. Later reports will address the clinical pathology of otters surviving in the centers for longer period.

METHODS

Twenty three otters have been selected for inclusion in this report. Otters were selected based upon completeness of clinical records and availability of detailed gross and histopathologic necropsy reports. Otters were divided into three groups on the basis of degree of oiling. Otters with greater than 60% of their bodies covered by oil were classified as heavily oiled, otters with 30-60% coverage were considered moderately oiled, and those with evidence of oiling but less than 30% coverage were classified as lightly oiled.

Clinical laboratory data for these twenty three otters are more varied in terms of completeness and reliability than either clinical records or pathology reports. Laboratory data were collected primarily to aid the clinician in clinical assessment and treatment; consequently, there is considerable variation in the amount and type of laboratory data available for each otter. Logistical problems in sample management also impact upon the consistency of the laboratory data. The centers were established under emergency conditions without proper laboratory equipment. Consequently, laboratory samples were sent to four different reference laboratories for evaluation, and comparing data from these laboratories must be approached with extreme caution. Additionally, inclement weather hindered the transport of some samples to laboratories. In some cases, laboratory data were totally lost as a result of these delays. In other instances, results were reported but have not been included in this summary because the problems in sample management were considered so great that laboratory data were regarded as invalid. Every effort has been made to include only that data which resulted from acceptably collected, transported, and processed samples.

The majority of the samples were processed through the PML laboratories in Seattle, Washington. Sea otter hematology and clinical chemistry reference ranges have been established for this laboratory through the analysis of hematology samples collected from 8 and serum samples collected from 26 clinically normal adult sea otters from southeastern Alaska. These reference ranges are listed in Table 7.1. The reference range was considered to be the mean +/-2 standard deviations with one exception. The reference range for lymphocytes was too large to be clinically useful. Consequently, lymphocyte counts of less than 1000 were regarded as lymphopenia and greater than $6000/\mu$ 1 as lymphocytosis (standard for most mammals). All PML generated laboratory data in this report have been compared to these reference ranges. No specific sea otter reference ranges have been developed for any of the other laboratories and the few results generated in those laboratories must be interpreted empirically. Extreme caution has been used in interpreting these results.

RESULTS

Tables 7.2-4 summarize the individual laboratory findings and clinical observations for the 23 heavily, moderately, and lightly oiled otters included in this report. Table 7.5 summarizes the principal syndromes which the laboratory data and clinical comments delineate.

The most common clinical syndrome seen regardless of degree of oiling was shock. Shock was usually characterized by hypothermia and lethargy and often by hemorrhagic diarrhea. Shock was rarely observed at the time of presentation but in heavily and moderately oiled otters generally developed within 48 hours of initial examination at the centers. In the lightly oiled otters included in this report, shock generally occurred during the second week of captivity.

A high proportion of otters in all three groups died with convulsions. Four of seven heavily oiled, four of five moderately oiled, and three of nine lightly oiled otters were seizuring at or near the time of death.

Anorexia was also a common clinical problem for otters in the rehabilitation centers. Anorexia was reported in three of seven heavily oiled otters, two of five moderately oiled otters, and one of nine lightly oiled otters.

From a laboratory data perspective, the most common hematologic abnormalities included lymphopenia and leukopenia, usually accompanied by increased numbers of immature neutrophils. Among heavily oiled animals, six of six had lymphopenia and four of six had leukopenia with increased immature neutrophils. Of the moderately oiled otters, four of five had lymphopenia and two of five had leukopenia with increased immature neutrophils. Of the lightly oiled otters, four of seven had lymphopenia and two of seven had leukopenia with increased immature neutrophils.

Anemia was also a relatively common hematologic abnormality. Altogether, nine of eighteen otters had anemia. This included three of six among the heavily oiled otters, one of five of those which were moderately oiled, and five of seven of the lightly oiled otters. Reticulocyte counts were not done and blood films were not available for retrospective study; as a result, the anemias could not be further classified.

The most prevalent syndromes identified by clinical chemistry included azotemia, hyperkalemia, hypoglycemia, hepatocellular leakage, and hypoproteinemia/hypoalbuminemia.

Azotemia, indicated by elevated serum urea. was the most common finding followed closely by hepatocellular leakage as indicated by elevated serum transaminases. Six of six heavily oiled otters, three of four moderately oiled otters, and six of nine lightly oiled otters were azotemic. Urine specific gravities were not available to help differentiate prerenal from renal azotemia. However, using the conservative assumption that serum urea levels about 200 mg/dl indicated renal azotemia, one of four moderately oiled otters, and three of nine lightly oiled otters were in renal failure when sampled near death. Evidence of hepatocellular leakage was present in five of five heavily oiled otters, three of four moderately oiled otters, and five of nine lightly oiled otters.

Hypoglycemia, hypoproteinemia/hypoalbuminemia, and hyperkalemia were somewhat less frequent alterations. They were found in nearly equal proportions in all of the three groups.

DISCUSSION

The vast majority of the sea otters that died acutely (within the first ten days of confinement) in the Valdez and Seward rehabilitation centers appear to have succumbed to shock. Terminal signs included lethargy, anorexia, and convulsions, often accompanied by hemorrhagic diarrhea.

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Whether or not shock was a direct effect of oiling or primarily an indirect effect secondary to confinement and handling in the rehabilitation centers is difficult to assess. Lightly oiled otters were as likely to die from shock as heavily oiled ones, suggesting that confinement was more important than direct exposure to oil. However, more heavily oiled otters developed signs of shock more rapidly than lightly oiled otters. In addition, heavily oiled otters generally had a higher proportion of laboratory abnormalities than lightly oiled otters. These findings suggest that at the least, exposure to oil was an important predisposing factor.

Laboratory findings correlated for the most part with clinical observations. The most frequent hematologic findings was lymphopenia. The most common cause of lymphopenia in animals is systemic stress which results in increased endogenous production of glucocorticoids and resultant sequestration and possible destruction of circulating lymphocytes. Certainly the otters treated in rehabilitation centers were stressed.

Leukopenia characterized by neutropenia with increased numbers of immature neutrophils (a degenerative left shift) was also extremely common. A degenerative left shift suggests severe inflammation. It is frequently observed in animals suffering from diarrhea with bowel stasis, proliferation of intestinal bacteria, and subsequent systemic endotoxemia. Such a scenario is likely in those otters suffering from diarrhea and hemorrhagic bowel syndrome.

Hyperkalemia and hypoproteinemia/hypoalbuminemia were probably also related to diarrhea and hemorrhagic bowel syndrome. Hyperkalemia was at least partially the result of release of potassium from dying cells in the hemorrhagic bowel. Acidosis, a common accompaniment of diarrhea, can also be a cause of hyperkalemia. Hypoproteinemia and hypoalbuminemia were probably the result of protein loss in the diarrhea fluid.

Azotemia was the most common syndrome identified from the laboratory data. As stated before, since urine specific gravities were not available for these animals, prerenal azotemia could not be absolutely differentiated from renal azotemia. However, necropsies did not reveal significant renal lesions (see Pathology report). It is therefore most likely that shock led to prerenal azotemia as a result of decreased renal perfusion. In the few animals that probably had true renal azotemia (those with serum urea nitrogen values greater than 200 mg/dl), it is likely that long-standing reduced renal perfusion eventually led to primary renal injury near the time of death.

Hypoglycemia probably resulted from anorexia. Sea otters have high metabolic rates and devour large quantities of food. Anorexia would be expected to quickly deplete hepatic glycogen stores, resulting in hypoglycemia. Hypoglycemia was probably the cause of many of the terminal convulsions.

Hepatocellular leakage may have been a reflection of primary hepatotoxicity but was more likely a nonspecific change associated with anorexia. In anorexia, decreased availability of carbohydrates as an energy source leads to mobilization of fat from tissues stores to the liver. Increases in fat within hepatocytes result in increased cell membrane permeability with leakage of transaminases into the blood. In these otters, elevated transaminases correlated most frequently with hepatic lipidosis histopathologically (Appendix A).

The cause(s) of anemia in these otters is difficult to determine, especially since the anemias were not well characterized. Certainly in those animals with hemorrhagic bowel syndrome, blood loss must be considered a contributing factor. Oil exposure has been reported to cause Heinz body hemolysis in birds; the possibility of Heinz body hemolysis in the otters can neither be substantiated nor refuted, but laboratory reports give no indication that Heinz bodies were ever observed.

Section 7B

SUMMARY REPORT: HEMATOLOGY AND CHEMISTRY

A.H. Rebar

Male Adult Sea Otters - 1989 and 1990

Hematologic and serum chemical analyses were performed on samples collected from 43 male adult Prince William Sound sea otters in 1989 and 1990. Twelve of these were from the east (unoiled) side while 31 were from the west. Data from the eastern otters were used to establish reference ranges (mean+/-2 standard deviations) for each of the analytes measured. Population data as well as individual animal data were compared to these reference ranges.

Hematologic differences between eastern and western adult males were minimal. Both hematocrits and hemoglobins were significantly higher in western otters than in eastern otters but the biologic significance of this is equivocal. From a biological perspective, the difference in oxygen carrying capacity of the blood at the values recorded is minimal. Western males had significantly higher absolute eosinophil counts, suggesting the possibility of systemic hypersensitivity reactions in western otters.

The important serum chemical differences between eastern and western males were found in protein and electrolyte levels. Western males had higher serum protein and serum globulin levels than eastern males. These findings suggest greater antigenic stimulation (more inflammatory and/or infectious conditions) in western than in eastern otters. This finding was supported by the trend (although not statistically significant) toward higher circulating neutrophil numbers in western males.

Serum sodium and serum chloride were significantly higher and serum potassium was significantly lower in western males than in their eastern counterparts. This pattern of electrolyte change was unusual; the possibility of stress-induced hyperadrenalism should be ruled out by measurement of serum cortisol levels.

Other differences between western and eastern males included lower lactate dehydrogenase (LDH), phosphorus, and glucose, and higher cholesterol, and carbon dioxide in western otters. LDH differences were disregarded because of the nonspecific nature of the enzyme. Glucose differences were not of biological importance because the relationship of sample collection to eating could not be ascertained and was undoubtedly not constant. Phosphorus differences were not important when considered in light of the large reference range. Cholesterol differences, like LDH, were not specific, difficult to interpret, and probably biologically insignificant. Differences in carbon dioxide levels were biologically insignificant.

Female Adult Sea Otters - 1989 and 1990

Hematologic and serum chemical analyses were performed on samples collected from a total of 103 female Prince William Sound sea otters in the fall of 1989 and the spring of 1990. Forty of these were from the east (unoiled) side while 63 were from the west (oiled) side. Data from the eastern otters were used to establish reference ranges (mean +/-2 standard deviations) for each of the analytes measured. Population data as well as individual animal data were compared to this reference range. There were no differences in hematologic parameters between eastern and western female otters. Some chemistry changes were present, but the degree of difference was small so that data must be interpreted cautiously.

Mean serum protein, albumin, and globulin levels for western female otters were within the eastern reference range. However, total protein and serum globulin levels were slightly higher in western females, a noteworthy finding since it mirrors the same trend seen in western adult males in 1989 and 1990. As mentioned previously, a trend to higher globulin levels suggests possible antigenic stimulation.

Liver enzyme levels (SGPT, alkaline phosphatase) for western females also fell within the eastern reference range but again mean values were somewhat higher for the western females than for the eastern otters. This suggests the possibility of subclinical liver disease and merits careful monitoring in the coming months and years. This is particularly important in light of elevated liver enzymes seen in otters brought to rehabilitation centers shortly after the oil spill. SGOT and LDH levels followed the same patterns as SGPT and alkaline phosphatase; however, these enzymes are nonspecific and are influenced by factors such as hemolysis. Consequently, less interpretive significance was given to these values.

Electrolyte levels in western females also generally fell within eastern reference ranges. Once again, however, a similar pattern to that seen in adult males was present; that is sodium and chloride mean values were somewhat higher and potassium mean values were somewhat lower in western females than in those from the east. Whether this subtle difference was biologically significant is still uncertain. However, as with males, a possible stress-induced hyperadrenalism should be ruled out by measurement of serum cortisol levels.

7 - 7

Sea Otter Pups - 1990

Hematologic and serum chemical analyses were performed on samples collected from a total of 57 Prince William Sound sea otter pups in the fall of 1990. Fifteen of these otters were from the east (unoiled) side of the Sound while 42 were from the western (oiled) side. Data from the eastern side were used to establish reference ranges for each of the analytes measured. Population data as well as individual animal data were compared to this reference range.

As a group, western pup hematocrits, hemoglobins, and red cell counts were significantly lower than those of eastern pups. From a biological perspective, these reductions were minimal but supported by individual animal data. MCHC was also significantly lower in western pups, but the degree of this decrease was not regarded as biologically significant.

When considered collectively, red cell data suggest a mild anemia in western pups. Although reticulocyte counts were not available, the normal red cell indices suggested that this anemia was most likely nonregenerative. The degree of anemia was minimal so that biological significance was equivocal.

Leukocyte group data for easter versus western pups showed minimal differences. The only real difference was a higher mean lymphocyte count in western pups. When the individual animal data was examined, this difference truly reflected a higher absolute lymphocyte count in a number of western pups. Lymphocytosis may be seen in animals with chronic antigenic stimulation; however, this change is usually accompanied by hypergammaglobulinemia and therefore elevated serum globulin levels. Globulin levels in western pups were normal, so it was difficult to suggest chronic antigenic stimulation in the present In some animals, most notably cats, lymphocytosis is seen case. when the animal is excited at the time of sample collection. While this may also be the case in sea otters, it is difficult to rationalize a relatively greater effect in western than in eastern pups. It is noteworthy that the eosinophilia seen in western adult males was not a prominent finding in the pups.

The only other interesting hematologic observation was the presence of a significantly elevated mean platelet count in western pups. On an individual animal basis, this elevation was seen in a relatively high proportion of individuals (10 of 24 measured). While the elevation appeared to be real, the degree of elevation from a biological perspective was not extreme and no particular interpretation could be attached to this finding.

Chemistry data were relatively unremarkable. Statistically significant differences in BUN, LDH, glucose, and triglycerides were not regarded as biologically significant. Both sodium and

chloride levels were statistically lower in western pups than in those from the east. The degree of reduction was so slight as to be of no biologic consequence; however, the observation is noteworthy in that it is in direct contrast to findings described earlier for adults where sodium and chloride were increased while potassium was reduced.

In summary, hematologic and clinical chemical differences between eastern pups were not striking and at best were of equivocal biologic significance. Perhaps the most significant observation was that trends seen in adults were not present in the pups observed to date.

*****	Mean	2SD	Reference Range
WBC	9175.00	4507.00	4668.00-13682.00
HGB	18.88	1.94	16.94-20.82
RBC	5.08	0.67	4.41-5.75
НСТ	62.00	9.43	52.57-71.43
MCV	122.25	11.99	110.26-134.24
MCH	37.25	2.78	34.47-40.03
MCHC	30.63	2.63	28.00-33.26
PLATELETS	295.00	128.75	166.25-423.75
(1000)			
SEGS	4136.00	2918.94	1217.06-7054.94
LYMPHS	3837.50	3750.26	87.24-7587.76
MONOS	208.50	99.57	108.93-308.07
EOS	942.00	719.27	222.73-1661.27
BASOS	51.00	102.40	0.00-153.40
GLUCOSE	156.04	92.48	63.56-248.52
TOTPROTEIN	6.30	1.26	5.04-7.56
CREATANINE	0.60	0.25	0.35-0.85
URIC ACID	2.41	1.20	1.21-3.61
CHOLERSTEROL	130.46	42.03	88.43-172.49
TRIGLYCERIDES	72.08	59.91	12.17-131.99
ALK PHOS	96.15	54.81	41.34-150.96
SGOT	202.81	280.50	0.00-483.31
SGPT	181.04	147.33	33.71-328.37
LDH	373.73	319.81	53.92-693.54
TOTBILI	0.49	0.37	0.12-0-0.86
SODIUM	148.12	21.11	127.01-169.23
POTASSIUM	3.93	0.72	3.21-4.65
CHLORIDE	111.35	15.29	96.06-126.64
CALCIUM	8.62	1.39	7.23-10.01
PHOSPHORUS	4.46	1.75	2.71-6.21
ALBUMIN	2.66	0.49	2.17-3.15
GLOBULIN	3.65	0.98	2.67-4.63
BUN	50.08	22.50	27.58-72.58
СРК	1642.89	4554.90	0.00-6197.79

Table 7.1 Reference Ranges for Sea Otters in Southeast Alaska (Samples analyzed at PML Laboratory)

Reference Range equal to mean +/-2 standard deviations Sample size for CBC is 8 Sample size for Chem values is 26 (19 females and 9 males) Samples size for CPK is 18 All otters at least one year of age

HE	NO T	OT NE	UT	LYMP	Т	OT	UR	EA ALA	AN ALK	,						
OTTER#	CRIT	WBC	PHIL	Bands C	YTE G	LUC P	ROT ALE	BU GLO	B NITR	ATRA	PHOS	ΝΑΚ	CL	Ρ		
VZ111 ¹	± ↓	ŧ	ŧ	Ť	Ť	N	ţ	± 4	Ν	î	Ť	Ν	-	-	-	-
VZ070 ²	-	-	-	-	-	Ν	N	Ν	Ν	î	Ť	Ν	±	1	N	Ť
VZ006 ³	Ν	Ť	Ŷ	Ť	ŧ	11	Ν	ŧ	Ν	Ť	Ť	Ν	Ν	Ν	±	1 1
VZ135⁴	Ļ	Ť	N	Ť	1	N	±↓	±↓	Ν	Ť	† †	ſ	Ν	Ν	Ν	Ť
VZ035 ⁵	Ť	Ν	Ν	N	Ť	-	-	-	-	-	-	-	-	-	-	-
VZ023 ³	¥	ŧ	t	t	Ť	±↓	± ↓	± ↓	Ν	Ť	Ť	Ν	Ν	Ν	Ν	± ↑
VZ013 ⁶	Ν	Ν	Ν	t	¥	11	ŧ	ţ	t	1	-	Ν	Ν	t	Ν	î
Total:	3/6↓	4/6↓	3/6	ł	6/6↓	3/6↓	4/6↓	5/6↓	1/6↓				1/5∔	_		
	1/6† 2/6N	6/6N	3/6	5/6† N 1/6N	3/	6N 2/6	N 1/6N	5/6N		6/6† 5/5	5/5† 5N 4/5N	3/5N 4/5I	V	2/5†	1/5 t	5/5†
Kev:	N =	= Norm	nal		↑ ↑ =	= Mark	ed increa	se								

Table 7.2 Principal laboratory and clinical findings in heavily oiled sea otters dying acutely (within 10 days of presentation) at rehabilitation centers.

 $\downarrow \downarrow$ = Marked decrease

= Moderate decrease \uparrow = Moderate increase

 $\pm \uparrow$ = Marginal increase

 $\pm \downarrow$ = Marginal decrease

³ Not available.

 ¹ Normal at presentation but mild hyperthermia, died in seizures within 12 hours.
 ² Within 2-3 days - became anorectic, lethargic and developed dark tarry stools. Developed seizures.

⁴ Lethargic and anorectic, hypothermic, seizured. Died within 4 days.

⁵ Dark diarrhea, lethargic.

⁶ Anorectic, dark tarry stools, seizured, died.

HEMO TOT NEUT		L۷	LYMP TOT				UREA			N	ALK						
OTTER#	CRIT	WBC	PHIL	Bands	CYTE	GLUC	PROT	ALBU	J GLC	B NITR	ATRA	PHOS	NA K	CL	P		
VZ113 ¹	N	ţ	Ļ	Ť	ţ	t		N	Ļ	N	† †	-	N	N	Ť	N	t
VZ100 ²	Ν	ŧ	± ↓	Ν	Ť	-		ŧ	±↓	ţ	-	Ť	Ť	Ν	Ť	Ν	Ť
VZ085 ³	N	Ν	Ν	N	Ť	t	Ł	= ↓	N	±↓	Ν	t	Ν	Ν	Ν	Ν	Ť
VZ080 ⁴	Ν	ŧ	Ν	Ν	ŧ	N		N	Ť	Ν	Ť	Ν	N	Ν	Ť	Ν	Ť
VZ043 ⁵	-	-	-	-	-	-		-	-	-	-	-	-	-	-	-	-
VZ109 ⁶	Ļ	¥	ţ	Ť	N	ŧ		N	N	Ν	Ť	† †	Ν	Ν	Ť	Ν	Ť
VZ045⁵	-	-	-	-	-	-		-	-	-	-	-	-	-	-	-	-
Total:	1/5↓	4/5↓	3/5	ł	4/5	5 4 2	2/4↓	2/5↓	2/5↓	2/5↓							
	4/5N	1/5N	2/5	2/51 N 3/5N	t N 1/E	5 N 1	/4 † /4N	3/5N	1/5† 2/5N	3/5N	3/4↑ 1/4N	3/4† 1/4N	1/5† 4/5N	5/5N	4/5† 1/5N	5/51 5N	ł

 $\pm \uparrow$ = Marginal increase

Table 7.3: Principal laboratory and clinical findings in heavily oiled sea otters dying acutely (within 10 days of presentation) at rehabilitation centers.

Key:

N = Normal

↑↑ = Marked increase

= Moderate decrease † = Moderate increase

 $\downarrow \downarrow$ = Marked decrease

 $\pm \downarrow$ = Marginal decrease

¹ Emaciated, pregnant at presentation, started to abort, became lethargic and hypothermic, seizured, died 36 hours after admission.

² Fair condition at presentation, died within 24 hours after seizures and anorexia.

³ Died within 24 hours, lethargic and hypothermic.

⁴ Within 24 hours of presentation; lethargic, hypothermic, vomiting. Died at 36 hours in seizures.

⁵ Omit

⁶ Thin on presentation. Within 24 hours - diarrhea, anorexia, decreased appetite. Died within 4 days with tarry diarrhea, lethargy, terminal seizures.
Kev:	N =	= Norn	nal		† †	= Ma	rked increa	se								
	2/7N	5/7N	5/71	5/7 f N 2/7N	1 1 3/7N	/9↑ 4/9N	5/9N	5/9N	6/9† 6/9N	5/9† 3/9N	2/9† 4/9N	6/91 7/9N	9N	6/9 3/9N 9/	}† ′9N 3/!	9N
Total:	5/7↓	2/7↓	2/7	ļ	4/7↓	4/9↓	4/9↓	4/9↓	3/9↓							
SW120 ⁶	-	-	-	-	-	î	ţ	ţ	±↓	Ť	t	Ν	Ν	Ť	Ν	î
VZ136⁵	ŧ	Ν	Ν	Ť	N	ſ	N	ŧ	Ν	Ť	Ť	N	N	Ť	Ν	î
VZ081 ⁴	Ť	Ν	N	† †	N	ţ	N	Ν	N	† †	1	N	N	Ť	Ν	î
VZ011 ¹	Ť	f	Ť	Ť	¥	ţ	¥	± ↓	±↓	Ť	Ν	N	N	Ť	Ν	î
VZ106 ³	Ť	Ť	ţ	Ť	± ↓	N	↓	±↓	±↓	† †	† †	Ť	N	Ť	Ν	î
SW135 ²	-	-	-	-	-	N	I N	Ν	Ν	N	Ν	N	N	N	Ν	N
SW125 ²	Ν	Ν	N	Ν	N	N	I N	Ν	Ν	Ν	Ν	N	N	Ν	Ν	Ν
SW127 ²	Ν	Ν	N	N	¥	N	I N	N	Ν	Ν	Ν	N	N	N	Ν	N
VZ141 ¹	↓↓	Ν	Ν	Ť	±↓	î	ţ	Ν	Ν	† †	† †	† †	N	t	Ν	ſ
<u>UTTER#</u>	CRIT	VVBC	PHIL	Banus	CTIE G	LUC	PRUT ALI		JENITE	AIRA	PHUS	NA K		<u>. P</u>		
				Doodo					יט סדווא סר					-N		
			пт			D	тот		() C			A.	Δ1	K		

Table 7.4: Principal laboratory and clinical findings in lightly oiled sea otters dying acutely (within 10 days of presentation) at rehabilitation centers.

Key:	N = Normal	f f = Marked increase
	H = Moderate decrease	f = Moderate increase
	↓↓ = Marked decrease	$\pm \dagger$ = Marginal increase
	±↓ = Marginal decrease	

¹ Not available.

² Inadvertently killed at rehabilitation center through hyperthermia.

³ Black "oily" stool, seizures within 24 hours, then died.

⁴ Normal at presentation.

⁵ Geriatric poor doer at presentation. From presentation diarrhea and dyspnea. Anorexia developed at one week and animal died.

⁶ Nursing pup that nursed well for 8 days. Found on eighth day in shock with seizures.

TABLE 7.5

Summary by Group (Heavily, Moderately or Lightly Oiled) of Principal Clinical and Laboratory Findings

	Shock	Convul- sions	Anorexi a	Lympho- penia	Leukop- penia/ Increas- e d Bands	Anemia	Azptem- ia	Hper- kalemia	Hypo- glycemi a	Hepato- cellular Leakage	Hypopr o- teinemi a/ Hypoal- bumin- emia
Heavily Oiled	5/7	4/7	3/7	6/6	4/6	3/6	6/6	2/5	3/6	5/5	4/6
Moder- ately	4/5	4/5	2/5	4/5	2/5	1/5	3/4	4/5	2/5	3/4	2/5
Lightly Oiled	7/9	3/9	1/9	4/7	3/7*	5/7	6/9	6/9	3/9	5/9	4/9
										<u> </u>	
TOTAL	16/21	11/21	6/21	14/18	9/18	9/18	15/19	12/19	8/20	13/18	10/20

Summary by Group (Heavily, Moderately or Lightly Oiled) of Principal Clinical and Laboratory Findings

	Shock	Convulsions	Anorexia	Lymphopeni	ia Leukopenia/ incr bands	Anemia	Azotemia	Hyperkalemia	Hypoglycemia	Hepatocellular leakage	Hypoproteinemia/ Hypoalbuminemia
Heavily Oiled	5/7	4/7	3/7	6/6	4/6	3/6	6/6	2/5	3/6	5/5	4/6
Moderately Oiled	4/5	4/5	2/5	4/5	2/5	1/5	3/4	4/5	2/5	3/4	2/5
Lightly Oiled	7/9	3/9	1/9	4/7	3/7"	5/7	6/9	6/9	3/9	5/9	4/9
Total	16/21	11/21	6/21	14/18	9/18	9/18	15/19	12/19	8/20	13/18	10/20

Histopathologic Lesions Associated with Crude Oil Exposure

in Sea Otters

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Lesions in Oil Exposed Otters

Abstract. Following the Exxon Valdez oil spill in Prince William Sound, Alaska, sea otters (Enhydra lutris) that appeared oiled, were in danger of becoming oiled, or were behaving abnormally were captured and taken to rehabilitation centers. Oil exposure was assessed by visual examination on arrival at the centers. Tissues from 51 oiled sea otters and from 6 unoiled sea otters that died in rehabilitation centers were examined histologically. Pulmonary interstitial emphysema, gastric erosion and hemorrhage, centrilobular hepatic necrosis, and hepatic and renal lipidosis were common in oil exposed otters and were absent or uncommon in unoiled otters. Histologic examinations were performed on tissues from 5 sea otters found dead with external oil present shortly after the spill. Hepatic and renal lipidosis was common, and pulmonary interstitial emphysema was found. Necropsies were performed on 214 sea otters that had been collected and frozen in the period following the oil spill. Histologic examination was not performed. Pulmonary interstitial emphysema and gastric erosion and hemorrhage were common in animals with external oil present; these lesions were found much less frequently in animals without detectable external oil. Tissues from 6 apparently normal sea otters collected from an area not affected by the oil spill were examined histologically, and none of these lesions were found. We conclude that pulmonary interstitial emphysema, gastric erosion and hemorrhage, centrilobular hepatic necrosis, and hepatic and renal lipidosis were associated with exposure to crude oil in sea otters.

On March 23, 1989, the oil tanker Exxon Valdez ran aground on Bligh Reef in Prince William Sound, Alaska. The resulting spill of approximately 11 million gallons of North Slope crude oil was the largest in the history of the United States. In the months following the spill, over 1,000 sea otters from oil spill-affected areas are known to have died. The actual number dead was probably much greater. As part of an effort to determine the effects of the oil spill on sea otters, we examined tissues from otters that died in rehabilitation centers and that were found dead with and without external oil present. We also examined tissues from apparently normal sea otters from an area not contaminated by crude oil.

Materials And Methods

Following the oil spill, sea otters that appeared oiled, were in danger of becoming oiled, or were behaving abnormally were captured and taken to one of several rehabilitation centers. Oil exposure was assessed by visual examination on arrival at the centers. Degree of oiling was graded according to the following criteria: greater than 60% body coverage - heavily oiled; 30-60% body coverage - moderately oiled; less than 30% body coverage or light sheen on fur - lightly oiled. If there was no oil visible, otters were considered unoiled. Oiled otters that died in rehabilitation centers (Group 1), unoiled otters that died in rehabilitation centers (Group 2), and otters that were found dead with external oil present (Group 3) were necropsied by various veterinarians. This fact and the lack of a standard necropsy protocol during the first few weeks after the spill resulted in great variation in the tissues collected. Documentation of necropsy findings ranged from minimal to thorough. In some cases, no necropsy report was available. Only otters with documented oil-exposure assessment were included in this study. Otters that died in rehabilitation centers were collected from oil-contaminated areas of Prince William Sound from 30 March to 17 July 1989 and died Estween 3 April and 4 August 1989. Animals that were found dead and necropsied were collected from contaminated areas in early April 1989. During the summer of 1989, biologists

collected tissues from 6 apparently healthy sea otters (Group 4) that had been killed by gunshot in the waters surrounding the Kuril Islands, Union of Soviet Socialist Republics, as part of unrelated research. Pups were not included in the study because of the small number available.

Tissues were placed in 10% neutral buffered formalin and processed in paraffin. Tissues collected included adrenal gland, aorta, bone marrow, brain, esophagus, eye, heart, intestine, kidney, liver, lung, lymph node, mammary gland, ovary, pancreas, parathyroid, pituitary gland, skeletal muscle, skin, spinal cord, spleen, stomach, testis, thymus, thyroid, tongue, tonsil, trachea, urinary bladder, and uterus. Tissues collected from individual animals varied. Sections were cut at 5um and stained with hematoxylin and eosin for light-microscopic examination. Selected sections were stained with oil red O.

In July and August 1990, complete necropsies were performed on 214 sea otters that had been collected from oil spill affected areas, placed in plastic bags, and frozen in the period following the spill (Group 5). Presence of external and internal oil was noted in the necropsy reports. Histologic examination was not performed because the tissue had been frozen.

Results

Data on individual Group 1, 2, and 3 otters are presented in Table 1. Numbers of Group 1, 2, and 3 otters of each gender with each of the common lesions are presented in Table 2.

In Group 1 sea otters, pulmonary interstitial emphysema was the most prevalent lesion, being present in 11 of 15 (73%) heavily oiled, 5 of 11 (45%) moderately oiled, and 3 of 20 (15%) lightly oiled animals. Overall, the lesion was present in 19 of 46 (41%) Group 1 otters. It was common in heavily and moderately oiled otters that died within 8 days of arrival at the rehabilitation centers, being present in 16 of 22 (73%) animals. Histologically, the lesion appeared as expanded areas of clear space with rounded contours within the interlobular septa (Fig. 1). Occasionally, adjacent parenchyma was compressed.

Gastric erosions were seen in 2 of 14 (14%) heavily oiled, 7 of 9 (78%) moderately oiled, and 4 of 17 (24%) lightly oiled Group 1 sea otters. Among total Group 1 otters, 13 of 40 (32%) had the lesion. Histologically, there were focal areas of coagulative necrosis, measuring 1 to 3 mm, affecting superficial to mid-level gastric mucosa (Fig. 2). Variable amounts of hemorrhage and blood pigments were present in the necrotic areas. Small numbers of neutrophils were sometimes scattered along the margins of the erosions.

Hepatic lipidosis was present in 8 of 16 (50%) heavily oiled, 5 of 12 (42%) moderately oiled, and 1 of 19 (5%) lightly oiled Group 1 otters. Among total Group 1 otters, 14 of 47 (30%) had the lesion. The prevalence of renal lipidosis was somewhat less than that of hepatic lipidosis. Overall, 10 of 42 (24%) Group 1 otters had renal lipidosis. All Group 1 animals with hepatic or renal lipidosis were female. All animals with renal lipidosis also had hepatic lipidosis. Several of the affected animals were pregnant or lactating, but several others were not. Both lesions were common in heavily and moderately oiled animals that died within 8 days of arrival at the centers (13 of 22 (59%) animals with hepatic lipidosis and 10 of 22 (45%) animals with renal lipidosis) and did not occur in animals that died later. The liver lesion was characterized by the presence of usually multiple but occasionally single, round, unstained intracytoplasmic vacuoles in periportal to midzonal hepatocytes (Fig. 3). Affected proximal convoluted tubular epithelium of the kidney contained similar intracytoplasmic vacuoles that were usually single (Fig. 4). In oil red O-stained sections of liver and kidney, the intracytoplasmic vacuoles stained red, indicating the presence of lipid.

Centrilobular hepatic necrosis occurred in 4 of 16 (25%) heavily oiled, 3 of 12 (25%) moderately oiled, and 4 of 19 (21%) lightly oiled Group 1 otters. Among all Group 1 otters, 11 of 47 (23%) were affected. In affected livers, centrilobular hepatocytes had undergone coagulative necrosis (Fig. 5). Among all Group 1 otters, multifocal hepatic necrosis was present in 6 of 47 (13%) and focally extensive hepatic necrosis suggestive of infarction was present in 4 of 47 (8%). Multifocal hepatic necrosis occurred in an animal that died on the first day in captivity and in animals that died after 3, 4, 5, 26, and 27 days. Focally extensive hepatic necrosis was found in animals that died after 4, 6, 8, and 27 days.

Of the 6 unoiled otters that died in rehabilitation centers (Group 2), one (17%) had gastric erosions, 1 (17%) had hepatic lipidosis and multifocal hepatic necrosis, and 1 (17%) had focally extensive hepatic necrosis.

Of the 5 sea otters found dead with external oil present (Group 3), 1 had pulmonary interstitial emphysema and hepatic and renal lipidosis, and 2 others had hepatic and renal lipidosis. One of the otters with hepatic and renal lipidosis was male and the other 2 were female.

Of the 6 apparently previously healthy sea otters collected from an area that had not been affected by an oil spill (Group 4), none had pulmonary interstitial emphysema, gastric erosions, hepatic or renal lipidosis, or hepatic necrosis. Four were male and 2 were nonpregnant, nonlactating females.

Two hundred fourteen sea otter carcasses that were collected, placed in plastic bags, and frozen in the period following the spill (Group 5) were thawed and necropsied. One hundred fifty-two (71%) had detectable external oil present, and 62 (21%) had no detectable oil present. Pulmonary interstitial emphysema was present in 100 of 152 (66%) otters with external oil present and in 13 of 62 (21%) otters with no detectable external oil. Interlobular septa of affected lungs were expanded by bubbles of trapped air that ranged from 1 mm to β cm in diameter. The emphysema was frequently diffuse and severe. In such cases, large amounts of adjacent pulmonary parenchyma were compressed. Extension of the air into the mediastinum was common, and involvement of the pericardium and the subcutis of the neck and thorax were occasionally found. Gastric erosions were present in 83 of 152 (55%) otters with external oil present and in 4 of 62 (6%) otters with no detectable external oil. The erosions generally were 1 to 3 mm punctate mucosal defects with dark red bases. Occasionally, erosions were linear and up to 1 cm in length. In some cases only a few erosions were present in the pylorus, but often erosions were numerous and were scattered throughout the gastric mucosa. Occasionally, similar erosions were present in the proximal duodenum. Accompanying hemorrhage was always present and varied from scant in animals with few erosions to abundant in animals with numerous erosions. Internal oil was found in 32 of 152 (21%) otters with external oil present and in 1 of 62 (2%) otters with no detectable external oil. The oil appeared as multiple, small, usually less than 3 mm diameter, black or brown flecks and was found on the tracheal, bronchial, esophageal, and gastric mucosae. Exposure of the oil to ultraviolet light in a darkened room caused the oil to glow and appear yellow to green. Blood did not glow or change color under

these conditions. Specific liver and kidney lesions could not be confidently identified by gross examination.

Various incidental lesions were found infrequently in Groups 1, 2,3, and 5. Thyroid follicular ectasia was common in all groups examined histologically.

Discussion

Pulmonary interstitial emphysema was prevalent in oiled sea otters that died in rehabilitation centers and in sea otters with external oil present that were found dead, frozen, and later thawed and necropsied (Group 5). The incidence of the lesion correlated with degree of oiling in Group 1 otters. It was also present in 1 of 5 otters found dead with external oil present. Emphysema was not seen in unciled otters that died in rehabilitation centers nor in apparently normal otters; however, it was present in several Group 5 otters that did not have detected external oil. Interstitial emphysema was diagnosed by others in many oiled sea otters presented to rehabilitation centers.¹⁸ Although not recognized prior to the Exxon Valdez oil spill, it is clear that exposure to crude oil causes sea otters to develop emphysema. The pathogenesis of the lesion in this setting is unclear. Alveolar tears are the usual route by which air enters the pulmonary interstitium. Alveolar tears can occur when there is a combination of forced expiration or coughing and bronchiolar obstruction that produces sharply increased pressures within alveoli.^{6,7} In anatomically predisposed species such as cattle the lesion may occur agonally, presumably due to forced expiration combined with bronchiolar collapse.⁷ Pulmonary interstitial emphysema has been reported in sea otters with pneumonia 2.14 and has been seen rarely as a mild focal lesion in sea otters that died without evidence of respiratory disease or oil exposure (personal observation, TPL). Sea otters may have an anatomical predisposition to development of interstitial emphysema, but exposure to crude oil resulted in a remarkably high incidence of the lesion. During the early days of the spill, inhalation of volatile components of crude oil such as benzene might have damaged alveolar septa and caused the lesion, but neither interstitial pneumonia nor other lesions that might result from inhalation of an irritant vapor were found in affected sea otters. Aspiration of oil may have caused powerful forced expirations that could result in interstitial emphysema. Oiled sea otters attempt to remove oil by grooming, 15 which involves use of the mouth; this process provides ample opportunity for aspiration. However, aspiration pneumonia was not found. We speculate that sea otters may have a highly developed cough reflex that effectively prevents aspiration of oil into the lungs but promotes the development of interstitial emphysema. The Group 5 otters with emphysema and no detectable oil present may have successfully removed detectable oil prior to death, or the emphysema may have been caused by a different mechanism. Postmortem examination failed to indicate the cause of the emphysema in these otters.

Gastric erosions were common in oil-exposed sea otters that died in rehabilitation centers and in oil-exposed sea otters found dead and examined grossly but not histologically. They were also found in an unoiled otter that died in a center and in a few Group 5 otters with no detectable oil. An explanation for the relatively low frequency in heavily oiled Group 1 otters (Table 1) is not readily apparent. Only rarely was oil found in stomachs of otters with gastric erosions. Rapidly developing gastric erosions that appear following severe stress occur in humans and animals.^{1,5} Gastrointestinal erosion/ulceration and hemorrhage have been reported in sea otters that died in captivity and in the wild and have been attributed to stress.^{14,16} All of the gastric erosions seen in this study were acute; none showed signs of healing. Those present in otters that died shortly after arrival at the rehabilitation centers might have developed prior to capture because of stress associated with oil exposure, as a direct effect of oil on the gastric mucosa, or because of stress associated with capture and captivity. Erosions caused by ingestion of corrosive liquids are extensive⁸, but the erosions we encountered were small and relatively uniform. Those seen in otters that died several days or more after arrival at the centers clearly developed in captivity.

Hepatic lipidosis was common in oiled otters that died in rehabilitation centers and in oiled otters that were found dead. The incidence of the lesion correlated with the degree of oiling. It was also seen in an unoiled otter that died in a rehabilitation center. Renal lipidosis was somewhat less common and occurred only in otters that also had hepatic lipidosis. All animals with hepatorenal lipidosis were female except for 1 oiled male that was found dead. When the oil spill occurred, many females were in late gestation or had recently pupped and were lactating. Hepatic lipidosis is known to have various causes including toxins, mobilization of stored fats due to inadequate food intake, hepatocellular hypoxia, certain metabolic disorders such as diabetes mellitus, and obesity.¹¹ Causes of renal lipidosis include toxins, hypoxia, and decreased food intake.9,10 Studies of experimentally oiled otters report marked increases in activity and metabolic rate with unchanged or decreased time devoted to feeding.^{3,15} Animals with high energy demands like those that occur during peak lactation or late gestation are predisposed to hepatic lipidosis.¹³ Thus, hepatic and renal lipidosis may have been caused by an oil exposure-associated increase in energy demand with constant or decreased food intake resulting in mobilization of stored fat. Many of the affected otters were further predisposed because of high energy demands due to lactation or pregnancy. Hepatic lipidosis in pregnant and lactating females may have been "physiologic," as occurs in ruminants, although this phenomenon has not been reported in sea otters. A direct toxic effect is possible, but accompanying hepatic and renal necrosis was not regularly present, and it is unusual for toxins to preferentially affect one gender. The fatty liver of hepatocellular hypoxia primarily affects centrilobular hepatocytes," but lipid accumulation in these otters was periportal to midzonal. The high incidence of lipidosis in otters that died during the first few days of captivity, its absence in otters that died after captivity day 8, its presence in 3 of 5 oiled otters that were found dead in the wild, and the absence of reports of lipidosis in otters that died in captivity suggest that captivity was not the cause of the lesion in our otters.

Centrilobular hepatic necrosis was also relatively common in oiled otters that died in rehabilitation centers and was not found in unoiled otters that died in the centers. Causes of centrilobular hepatic necrosis include toxins and conditions that cause hepatic ischemia, such as anemia, shock, and heart failure.¹¹ Some oiled otters became anemic while at rehabilitation centers.¹⁹ Crude oil ingestion¹² and gastric erosion with hemorrhage are possible causes of anemia. However, gastric erosions and centrilobular hepatic necrosis were found in the same otter infrequently, so anemia due to gastric hemorrhage was not a common cause of centrilobular hepatic necrosis. It is likely that many otters experienced shock.¹⁹ Shock might result from oil exposure or captivity, but centrilobular hepatic necrosis has not been reported previously in otters that died in captivity. Multifocal hepatic necrosis and focally extensive hepatic necrosis suggestive of infarcts occurred at low frequency in both oiled and unoiled sea otters that died in rehabilitation centers. The causes of these lesions were not found.

Sea otters are largely dependent on the insulating properties of their pelage for protection from the cold waters they inhabit. It had

been suspected that hypothermia would be a major problem in oiled sea otters because oil markedly increases the thermal conductance of their coats.¹⁷ Hypothermia was a common problem in oiled sea otters presented to rehabilitation centers.¹⁸ Death caused by hypothermia can occur without distinctive gross or microscopic lesions.⁴ It is likely that stress and shock were significant medical problems.¹⁹ Both oil exposure and captivity are stressful to sea otters.^{15,16} We believe that hypothermia, stress, shock, respiratory compromise associated with interstitial emphysema, hemorrhage from gastric erosions, and hepatic necrosis contributed to the deaths of oiled sea otters.

In summary, pulmonary interstitial emphysema, gastric erosion and hemorrhage, hepatic and renal lipidosis, and centrilobular hepatic necrosis were common in oiled sea otters that died in rehabilitation centers and were absent or uncommon in the small group of unoiled sea otters that died in rehabilitation centers. Pulmonary interstitial emphysema and gastric erosion and hemorrhage were prevalent in oiled sea otters that were examined grossly but not histologically; these lesions were found much less commonly in sea otters without detectable oil that were examined grossly but not histologically. None of these lesions were seen in apparently normal, unoiled sea otters and, with the exception of gastric erosion and hemorrhage, have not been previously reported in association with death in captivity. Additionally, pulmonary interstitial emphysema and hepatic and renal lipidosis were present in a small group of oiled sea otters that were found dead in the wild. Pathologic examination of larger numbers of both oiled and unoiled sea otters not held in captivity would be useful in separating lesions resulting from exposure to crude oil and those resulting from effects of captivity.

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The opinions and assertions contained herein are those of the authors and are not to be construed as official or representing those of the Department of the Army or the Department of Defense.

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		GROUI	P 1 (oile	ed, died in	n cente	∋r)				
			Heavil	ly Oiled						
Otter		Arrival	Death	Days In						
Number	Gender	Date	Date	Center	EMP	GE	HL	RL	CLHN	
1	F	7Apr	7Apr	<1	X	X	X			
2	F	4Apr	5Apr	1		Х				
3	F	9Apr	10Apr	1	х		х	х		
4	F	6Apr	7Apr	1	х		х			
5	F	6Apr	7Apr	1					Х	
6	F	5Apr	8Apr	3	х		х	х		
7	F	31Mar	3Apr	3	X		Х			
8	F	4Apr	7Apr	3	Х					
9	F	19Apr	23Apr	4			х	х		
10	F	3Apr	7Apr	4	х					
11	F	5Apr	10Apr	5	х		х	х		
12	М	30Mar	5Apr	6	X					
13	М	2Apr	9Apr	7	Х				Х	
14	F	lApr	9Apr	8	Х		х	х	Х	
15	М	lApr	10Apr	9						
16	М	2Apr	28Jul	117					Х	
		•	Moder	ately Oile	d					
17	F	9Apr	10Apr	1			X	X		
18	F	4Apr	5Apr	1	Х		X	Х	X	
19	F	8Apr	-9Apr	1	Х	X	х	х		
20	F	7Apr	8Apr	1			X	X		
21	F	6Apr	8Apr	2	Х		X	X	X	
22	F	3Apr	6Apr	3	Х	X				
23	М	9Apr	13Apr	4		X				
24	F	4Apr	9Apr	5		Х			X	
25	М	18Apr	29Apr	11						
26	F	5Apr	18Apr	13	Х					
27	F	llMay	24May	13		X				
28	M	5Apr	5May	30		Х				
29	F	llMay	24Jul	74		Х				

•

EMP = emphysema GE = gastric erosion HL = hepatic lipidosis RL = renal lipidosis CLHN = centrilobular hepatic necrosis F = female M = male

			Ligl	ntly Oiled	1				
Otter		Arrival	Death	Days In					
Number	Gender	Date	Date	Center	EMP	_GE	HL_	RL	CLHN
30	F	20Apr	20Apr	<1					Х
31	F	5Jun	5Jun	<1					
32	F	6Apr	7Apr	1					
33	F	5Jun	6Jun	1					
34	м	13Jun	14Jun	1		х			
35	F	9Apr	llApr	2			х		
36	F	lApr	4Apr	3					
37	F	4Apr	7Apr	3	Х	Х			
38	м	6Apr	12Apr	6	Х				
39	F	10May	17MAy	7					
40	М	19Apr	27May	8					Х
41	F	25May	4Jun	10		Х			
42	М	20May	31May	11					
43	F	13Jun	27Jun	14					
44	F	8Apr	28Apr	20		Х			Х
45	М	8Apr	29Apr	21					
46	м	8Apr	1May	23					
47	м	6Apr	29Apr	23					
48	F	6Apr	30Apr	24	Х				
49	F	10Apr	6May	26					Х
50	F	llMay	7Jun	27					
51	F	20May	19Jun	30					
		GROUP	2 (unoil	Led, died	in cer	nter)			
52	F	29May	29May	<1					
53	м	- 13Apr	14Apr	1					
54	F	5Jul	6Jul	1			Х		
55	М	25Jun	27Jun	2					
56	F	19Jun	3Jul	14					
57	м	17Jul	4Aug	18		Х			
		G	coup 3 (f	ound dead	oiled)			
58	м		· · · · · · · · · · · · · · · · · · ·						
59	M						х	x	
60	M								
61	F						х	x	
62	F				х		x	x	
	-								

TABLE 1 (cont.)

TABLE 2

(15)73% 2	(10)200 (4) 0% (14)14%	8(12) 67% 0 (4) 0% 8(16) 50%	5(11) 45% 0(2) 0% 5(13) 38%	Necrosis 2(12)17% 2 (4)50% 4(16)25%
(8)62% 5 (3)0% 2 (11)45% 7	(6)83% (3)67% (9)78%	5 (9) 56% 0 (3) 0% 5(12) 42%	5 (9) 56% 0 (3) 0% 5(12) 42%	3 (9)33% 0 (3) 0% 3(12)25%
(14)14% 3 (6)17% 1 (20)15% 4	(13)23% (4)25% (17)24%	1(12) 8% 0 (7) 0% 1(19) 5%	0(12) 0% 0 (5) 0% 0(17) 0%	3(12)25% 1 (7)14% 4(19)21%
G	roup 1 To	tals		
(33)48% 10 (13)23% 3 (46)41% 13	(29)34% 1 (11)27% (40)32% 1	.4(33) 42% 1 0(14) 0% .4(47) 30% 1	0(32) 31% 0(10) 0% 0(42) 24%	8(33)24% 3(14)21% 11(47)23%
GROUP 2 (ur	noiled, di	ed in center	•)	
(3) 0% 0 (3) 0% 1 (6) 0% 1	(3) 0% (3)33% (6)17%	1(3) 33% 0(3) 0% 1(6) 17%	0(3) 0% 0(3) 0% 0(6) 0%	0 (3) 0% 0 (3) 0% 0 (6) 0%
GROUP 3 (fo	ound dead	oiled) Total	8	
(2)50% 0 (3) 0% 0 (5)20% 0	(2) 0% (3) 0% (5) 0%	2(2)100% 1(3) 33% 3(5) 60%	2(2)100% 1(3) 33% 3(5) 60%	0 (2) 0% 0 (3) 0% 0 (5) 0%
led le totaled ole for compa	arison; al	l of the rel	evant tissue	s were not
	(4)30% 0 (15)73% 2 (8)62% 5 (3)0% 2 (1)45% 7 (4)14% 3 (6)17% 1 20)15% 4 G 33)48% 10 13)23% 3 46)41% 13 GROUP 2 (ux) (3)0% 0 (3)0% 1 (6)0% 1 GROUP 3 (for (2)50% 0 (3)0% 0 (5)20% 0 .ed	(4)50% 0 (4)50% (5)73% 2 (14)14% (8)62% 5 (6)83% (3) 0% 2 (3)67% 11)45% 7 (9)78% 14)14% 3 (13)23% (6)17% 1 (4)25% 20)15% 4 (17)24% Group 1 To 33)48% 10(29)34% 1 13)23% 3 (11)27% 46)41% 13(40)32% 1 GROUP 2 (unoiled, di (3) 0% 0 (3) 0% (3) 0% 1 (6)17% GROUP 3 (found dead (2)50% 0 (2) 0% (3) 0% 0 (3) 0% (5)20% 0 (5) 0% .ed	<pre>(4) 50% 0 (4) 5% 0 (4) 5% (15) 73% 2(14) 14% 8(16) 50% (3) 0% 2 (3) 67% 0 (3) 0% 11) 45% 7 (9) 78% 5(12) 42% 14) 14% 3(13) 23% 1(12) 8% (6) 17% 1 (4) 25% 0 (7) 0% 20) 15% 4(17) 24% 1(19) 5% Group 1 Totals 33) 48% 10(29) 34% 14(33) 42% 1 13) 23% 3(11) 27% 0(14) 0% 46) 41% 13(40) 32% 14(47) 30% 1 GROUP 2 (unoiled, died in center (3) 0% 0 (3) 0% 1(3) 33% (3) 0% 1 (3) 33% 0(3) 0% (6) 0% 1 (6) 17% 1(6) 17% GROUP 3 (found dead oiled) Total (2) 50% 0 (2) 0% 2(2) 100% (3) 0% 0 (3) 0% 1(3) 33% (5) 20% 0 (5) 0% 3(5) 60%</pre>	<pre>(4) 50% 0 (4) 0% 0 (4) 0% 0 (4) 0% 0 (2) 0% (15) 73% 2(14) 14% 8(16) 50% 5(13) 38% (8) 62% 5 (6) 83% 5 (9) 56% 5 (9) 56% (3) 0% 2 (3) 67% 0 (3) 0% 0 (3) 0% (11) 45% 7 (9) 78% 5(12) 42% 5(12) 42% 14) 14% 3(13) 23% 1(12) 8% 0(12) 0% (6) 17% 1 (4) 25% 0 (7) 0% 0 (5) 0% 20) 15% 4(17) 24% 1(19) 5% 0(17) 0% Group 1 Totals 33) 48% 10(29) 34% 14(33) 42% 10(32) 31% 13) 23% 3(11) 27% 0(14) 0% 0(10) 0% 46) 41% 13(40) 32% 14(47) 30% 10(42) 24% GROUP 2 (unciled, died in center) (3) 0% 0 (3) 0% 1(3) 33% 0(3) 0% (3) 0% 1 (3) 33% 0(3) 0% 0(3) 0% (6) 0% 1 (6) 17% 1(6) 17% 0(6) 0% GROUP 3 (found dead ciled) Totals (2) 50% 0 (2) 0% 2(2) 100% 2(2) 100% (3) 0% 0 (3) 0% 1(3) 33% 1(3) 33% (5) 20% 0 (5) 0% 3(5) 60% 3(5) 60% et cotaled ble for comparison; all of the relevant tissue tters.</pre>

GROUP 1 (oiled, died in center)

Heavily Oiled

<pre># in Paper 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15</pre>	AFIP # 2270227 2270216 2269894 2269852 2269884 2269871 2270212 2269837 2269882 2269937 2269882 2269919 2269867 2270231 2269868 2269951	Rehab # VZ094 VZ054 VZ111 VZ077 VZ078 VZ070 VZ006 VZ053 VZ135 VZ042 VZ059 VZ001 VZ035 VZ023 VZ013
16	2275590	VZ029
Mod	erately Oiled	l
17 18 19 20 21 22 23 24 25 26 27 28 29 Li	2269892 2269862 2269895 2270229 2270233 2269847 2269949 2269993 2269920 2269946 2244498 2269927 2244493	VZ113 VZ049 VZ100 VZ085 VZ080 VZ043 VZ109 VZ045 VZ134 VZ060 SW076 VZ065 SW077
30	2269933	VZ141
31 32 33 34 35 36 37 38 39 40 41	2244226 2270214 2244228 2244233 2269888 2269864 2270209 2269956 2269914 2269913 2244241	SW127 VZ075 SW125 SW135 VZ106 VZ011 VZ047 VZ081 SW050 VZ136 SW120

42	2244217	SW104	
43	2244507	SW132	
44	2269875	VZ102	
45	2272018	VZ103	
46	2269955	VZ099	
47	2269923	VZ074	
48	2269930	VZ079	
49	2244497	SW115	
50	2244224	SW067	
51	2244227	SW103	
	Group 2		
50	2272017		
52	22/201/	VZIDO	
23	2269924	VZIZI	
54	2243617	SW163	
55	2244223	SWIGU	
56	2242106	SW149	
57	2244490	SW170	
	Group 3		
	ereab e		
58	2269855	VD123	
59	2269877	VD093	
60	2269857	VD110	
61	2269859	VD112	
62	2269874	VD097	

#

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Technical Report: Marine Mammals Study Number 6

Mortality of Sea Otter Weanlings in Eastern and Western Prince William Sound, Alaska, During the Winter of 1990-91

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August 1, 1991

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SUMMARY

A total of sixty-four dependent sea otters (control n = 24; oil-spill treatment n = 40) were captured, examined, instrumented with radio-transmitters, and monitored in Prince William Sound between September 1990 and present as part of a group of interrelated studies aimed at assessing the impact of the T/VExxon Valdez oil spill on sea otters in Prince William Sound, Alaska.

The peak of life history milestones such as birth, weaning, and mortality of young of the year may occur a month or more later in the southwestern (the oil spill area) than in the northeastern (the control area) portion of Prince William Sound. While the timing of instrumentation was similar for pups in the control and treatment areas, pups in the southwestern sound weighed significantly less at the time of capture than their counterparts in the northeastern sound. Subsequently, most pups in the northeastern sound became independent of their mothers in October, whereas mother-pup separation typically occurred in November and December in the oil spill area. Most mortality in the northeastern sound occurred during November and December of 1990, whereas most mortality in the southwestern sound occurred during January 1991.

Contingency chi-square analyses indicated that survival rates of weanlings over their first winter (analyses consider data until May 1, 1991) were significantly higher in the northeastern sound (the control) than in the southwestern Prince William Sound (the oil spill region). This result was the same regardless of whether missing animals were assumed to be dead (X^2 = 4.64, 1 D.F., p < 0.05) or were eliminated from the analyses (X^2 = 4.70, 1 D.F., p < 0.05).

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INTRODUCTION

On March 24, 1989, over 11 million gallons of crude oil were spilled in Prince William Sound, Alaska, due to the wreck of the T/V Exxon Valdez. The research discussed in this report was undertaken as part of Natural Resource Damage Assessment (NRDA) studies aimed at determining if the spill caused damage to the sea otter population(s) in the region, and, if so, the type, magnitude, and significance of the damage(s). The specific goal of this study was to test whether overwinter mortality rates were equivalent among weanling sea otters in southwestern and eastern areas of Prince William Sound. While this report is narrow in its focus on this question, this information is crucial to understanding the overall extent of damage to the sea otter population(s); to understanding whether the affected sea otter population(s) are in a recovery phase; to estimating the rate and pattern of recovery; and to formulating restoration and response policies for sea otters throughout their range.

OBJECTIVE

The specific objective of this study was originally defined in the corresponding statement of work as follows:

> To test the hypothesis that weanling survival at various age intervals is not different between ciled and unciled areas at alpha = 0.20.

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STUDY METHODOLOGY

Definitions

For consistency, the use of terms in this report follows that in previous Natural Resource Damage Assessment reports and study plans. Thus, "dependents" or "pups" are individuals accompanied by their mothers. For the purposes of this report, individuals were classified as "weanlings" when they were no longer accompanied by their mothers. This term is used only to make it comparable to previous NRDA sea otter literature. We have no information about the date at which individuals actually stopped nursing.

Status classifications are made based on consideration of data to May 1, 1991 (see below). Individuals classified as dead are known to be dead because their carcass or other remains were observed and, in some cases, recovered. "Missing" individuals are those whose radio signal cannot be detected by boat or aircraft radio searches within Prince William Sound or adjacent areas (see Monitoring section below). The classification of "alive" is based on visual observations of the individual.

The estimated weaning date is that date midway between the dates when the individual was last observed with its mother and when it was first observed to be independent. Estimated date of death is that date midway between the dates when the individual was last confirmed to be alive and when it was found dead.

Study Groups

Data from two groups of sea otters are compared: those captured as dependents in the fall of 1990 in the southwestern portion of Prince William Sound (sometimes referred to as the oil-spill area) and those captured as dependents in the fall of 1990 in castern Prince William Sound during 1990 (i.e. the control area).

Capture and Examination

Sea otters were captured at night in tangle nets and during the daytime with dip nets using methods previously described (Monnett et al. 1991). In most cases when dependent pups were being instrumented, mother-pup pairs were captured together and the mother was held in a floating net-pen while the pup was instrumented. However, some pups were captured with hand-held dip nets and brought aboard a 25 foot Boston Whaler for instrumentation. In those cases, the mother remained freeswimming while instrumentation was completed. However, for the purposes of calculating "take" (as defined under the 1972 Marine Mammal Protection Act of the United States) both the mother and the pup are counted in all pairs in which at least one of the pair was harassed or captured.

Anesthetized sea otters were placed in a nylon mesh bag and weighed on a hanging scale to the nearest 0.5 pound. Total body length was measured to the nearest 0.5 inch with a nylon tape. The tape was placed upon a flat surface. The otter was placed on

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its back and measured as it was stretched along the tape by two researchers.

Marking and Instrumentation

Following capture, otters were brought aboard various vessels for anesthesia, examination, tagging of the hind flippers, insertion of a transponder and instrumentation with implanted radio-transmitters following established protocols. Details of these protocols are found in Monnett (1988) and Monnett et al. (1991). Following completion of instrumentation, collection of biological samples, and other data collection procedures, otters were released near the site at which they were captured.

For this study, an individual was not considered to be suitable for instrumentation if: a) its weight was less than 18 lbs; b) it was in very poor physical condition; c) its mother was inattentive prior to injection of the anesthetic; d) its initial temperature was greater than $102^{\circ}F$ and could not be stabilized; or e) it did not respond to the anesthetic.

Monitoring

Radio-instrumented sea otters were monitored by observers in aircraft and Boston Whaler skiffs. Aircraft and skiffs were equipped with right- and left-mounted Yagi antennas and programmable, scanning FM receivers. Aircraft were flown at variable height depending upon whether observers were attempting to locate radio signals or make visual observation on individual sea otters. An attempt was made to find and visually examine each otter at least biweekly. All areas of Prince William Sound have been searched thoroughly by aircraft for missing animals. Additionally, a radio search of the nearcoastal areas between Prince William Sound and Homer, Alaska (to the west) and to Controller Bay (to the southeast) was made by aircraft, specifically to search for the missing individuals in this study. Data were recorded directly on xeroxed topographical maps and in "Rite-in-the-Rain books" for later entry into Lotus 123 on personal computers.

Comparison of Overwinter Mortality of Weanlings

Contingency chi-square analyses were used to compare the frequencies of weanling sea otters known to be alive, those known to be dead, and those that were missing as of May 1, 1991 in the control and oil-spill areas (see definitions above). Thus, a categorical statistical model was employed (rather than Kaplan-Meier survival estimation procedure as described in the corresponding study plan) because, given the nature of the study subjects and the short duration of the study, the biologically meaningful comparisons were the survival frequencies of weanlings to a specific life-history milestone: through the end of their first winter. The cutoff date of May 1, 1991 was used based on previous (Monnett 1988) information indicating that human causes of mortality, especially death associated with gillnet fisheries, become important in the eastern sound, but probably not in the southwestern sound, shortly thereafter due to the initiation of fishing activity.

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STUDY RESULTS AND DISCUSSION

Study Population and Monitoring

A total of 252 sea otters were captured or taken by harassment in Prince William Sound between 3 September 1990 and 15 October 1990 (Table 1). The locations of capture of instrumented sea otter pups are shown in Figure 1. Distribution of the date of instrumentation in the two general areas is shown in Figure 2. Twenty-four dependent pups in the eastern sound [16 males (67%) and 8 females (33%)]} and 40 pups in the oil spill area [25 males (60%) and 15 females (40%)] were instrumented with intraperitoncal radiotransmitters. Two individuals (one male and one female both from the eastern sound), whose deaths may have been related to capture/instrumentation activities, were excluded from all summaries and statistical comparisons (one was found dead with an infected incision, whereas the other individual died 2 weeks after instrumentation). A female pup in the southwestern sound that was still with its mother on May 1, 1991 (and hence, was not a weanling) was included in the comparison of body weight at the time of capture, and in summaries of the date of instrumentation, but eliminated from other comparisons. Her dependency length is much greater than those previously observed in Prince William Sound. Most importantly, since previous work (Monnett 1988) has shown that the probability of dying greatly increases after mother-pup separation, it would not be appropriate to include her in comparisons of outcome. Some pups that were last seen alive with their mothers were either dead at

the next observation or became missing. These pups were eliminated from the figure summarizing weaning date.

The sample size for this study differs markedly from that outlined in the study plan for this portion of the damage assessment research which called for the instrumentation of 100 pups split evenly between control and treatment areas. Too few radios were ordered to permit implantation of 100 pups.

Size at Capture. Timing of Weaning and Peak Mortality

There is some indication that in 1990-1991, the peaks of pupping, weaning, and weanling mortality may have occurred at least a month or two later in the oil-spill area than in the control region of Prince William Sound. However, at this time it cannot be ascertained whether these apparent differences in timing reflect normal differences, or whether the timing differences could be related to the spill.

The timing of instrumentation was similar for pups in the control and treatment areas (Figure 2). However, the mean body weight at capture of male and female dependent pups in the oil-spill treatment group was significantly less than that of pups in the control group [(MALES: west: X = 25.28, S.E. = 0.72, N = 25; east: X = 32.27, S.E. = 0.83, n = 15, t = 5.57, 38 D.F.., p < 0.001); FEMALES: west: X = 24.17, S.E. = 0.97, N = 15; east: X = 30.36, S.E. = 1.03, n = 7, t = 3.53, 20 D.F.., p < 0.003)]. These differences and unpublished data on the actual timing of pupping of instrumented adult females (Monnett and Rotterman, unpublished. data) indicate that the peak in pupping in 1990 was

later in southwestern Prince William Sound than in northeastern Prince William Sound, and thus, it is likely that the southwestern sound pups were younger at the time of capture. Dates of weaning tended to be a month to two months later in the oil-spill area than in the control region. At present it is not clear whether this later date of weaning is explained sufficiently by the temporal shift in pupping or whether dependency periods tended to be longer in the oil spill area. However, most pups in the northeastern sound became independent of their mothers in October, whereas mother-pup separation typically occurred in November and December in the oil spill area (Figure 2).

Since available data (Monnett and Rotterman, unpublished. data) suggest that mortality of large pups is very low (ie., while they are large but still with their mother), it is not surprising that with a shift in the peaks of weaning, the period of highest weanling mortality is also later. Most mortality in the northeastern sound occurred during November and December of 1990, whereas most mortality in the southwestern sound occurred during January 1991 (Figure 2).

Further effort needs to be focused on determining whether these apparent timing differences are persistent, whether the shifts in weaning and mortality dates are explained by a shift in the peak of pupping of the same magnitude, or whether they reflect longer dependency periods and/or differences in the length of survival after weaning. Integration of these data with

data from the reproductive behavior of instrumented adult females will be especially insightful in this regard.

Mortality Rate Comparisons

Contingency chi-square analyses indicated that survival rates of weanlings over their first winter (analyses consider data until May 1, 1991) were significantly higher in the northeastern sound (the control) than in the southwestern Prince William Sound (the oil spill region) (Table 2). This result was the same regardless of whether missing animals were assumed to be dead ($X^2 = 4.64$, 1 D.F., p < 0.05) or were eliminated from the analyses ($X^2 = 4.70$, 1 D.F., p < 0.05).

Figures 3 and 4 indicate the locations where the carcasses of instrumented weanlings were discovered.

As noted previously, the status of weanlings as of May 1, 1991 was compared because previous data (Monnett 1988) suggests that, after that time, fishing-related deaths are an important source of mortality to male weaning sea otters in eastern Prince William Sound. However, consideration of the current (as of July 15, 1991 status of the surviving weanlings does not alter the conclusions of this study. Between May 1 and July 15th, one individual in the eastern sound dropped out of the study due to radio failure and one additional weanling in the western sound became missing.

It is not the purpose nor the intent of this report to attempt to fully develop the significance of this difference in overwinter weanling survival to the larger question of damage to

sea otters from the T/V Exxon Valdez oil spill. Rather, the data presented here address a focussed and narrow objective: to test the hypothesis that overwintering mortality is similar between the defined control and oil-spill areas over the winter of 1990-1991. We reject that null hypothesis. Weanlings in the oil spill area died at higher frequency than did their control counterparts. A more complete interpretation of the significance of this finding to potential damage to the sea otter population(s) from the oil spill would be possible if integrated with existing data on: a) reproductive patterns of instrumented adult females (in order to determine the relationship between pupping peaks and weaning peaks); growth rates of pups in the eastern and western sound; movement patterns (to determine whether the putative mothers of the control group actually were less likely to have been exposed to crude oil from the spill than the mothers of pups caught in that region); data from the clinical exams of pups included in this study; data on the blood chemistry and hematology of pups and adult females; data on the kinds and levels of petroleum-derived hydrocarbon burdens of both adult females and pups caught in the control and oil-spill areas; and data from gross necropsies and histopathological examination of tissues of sea otters caught in Frince William Sound from September 1989 through present. However, data from future studies will be most useful with regards to understanding the effects of the oil spill if collected in conjunction with information on the outcomes of individuals rather than as

independent statistical analysis of data collected at unrelated locations and different times from the data on outcome.

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Table 1. Summary of Sea Otters Taken by Capture or Harrassment As Part of Capture Activities Between September 1, 1990 and October 15, 1990.

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AREA	AGE/SEX CATEGORY	NUMBER TAKEN	NUMBER IMPLANTED		
E PWS	Adult Male	8	0		
	Adult Female (indep)	37	4		
	Mother-Pup Pairs	106 (53 prs.)	24 pups		
	Unknown	2	0		
	Area Total	153	28		
W PWS	Adult Male	2	0		
	Adult Female (indep)	. 1	0		
	Mother-Pup Rairs	96 (46 prs.)	40 pups		
	Unknown	0	0		
	Area Total	99	40		
	Total Number of Pups	99	64		
	Grand Total	252	68		

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Table 2. Status of Radio-Instrumented Weanlings in Prince William Sound, Alaska, As of May 1, 1991. Two individuals whose death may have been related to capture/intrumentation activities were excluded from analyses, as was one pup that was still with its mother on May 1, 1991.

SEX	AREA	TOTAL	ALIVE	DEAD	MISSING
MALES	E PWS	15	5(.33)	8(.53)	2(.13)
	W PWS	25	2(.08)	17(.68)	6(.24)
FEMALES	E PWS	7	3(.43)	2(.29)	2(.29)
	W PWS	14	3(.21)	9(.64)	2(.14)
SEXES COMBINED	E PWS	22	8(.36)	10(.45)	4(.18)
	W PWS	39	5(.13)	26(.57)	8(.21)

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Figure 1. Capture locations of sea otter pups instrumented with implanted radio-transmitters during the fall, 1990, in Prince William Sound, Alaska. The numbers represent the number of individuals instrumented at each location.



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Figure 2. Summary of timing of instrumentation, weaning and mortality of sea otters in Prince William Sound, 1990-91.



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Figure 3. Locations at which remains of radio-instrumented weanling sea otters were recovered in Eastern Prince William Sound, Alaska, September 1990 - March 1991.



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Figure 4. Locations at which remains of radio-instrumented weanling sea otters were recovered in Western Prince William Sound, Alaska, November 1990 - April 1991.



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Technical Report: Marine Mammal Study Number 7

Mortality and Reproduction of Sea Otters Oiled and Treated as a Result of the Exxon Valdez Oil Spill

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August 15, 1991

ATTORNEY/CLIENT WORK PRODUCT

SUMMARY

Radio-instrumented sea otters (N = 45) that were released into eastern Prince William Sound during summer, 1989, following efforts to rehabilitate them at otter treatment centers, have been monitored regularly for approximately 2 years. During this time, survivorship and pupping rates were generally lower than that of sea otters in other study populations.

INTRODUCTION

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In response to the massive oil spill caused by the wreck of the T/V Exxon Valdez, a large number of sea otters (Enhydra lutris) were captured and brought into centers that were established in order to wash them, and to provide them with medical and other supportive treatment (e.g., see Williams et al. 1990). Many of the sea otters that survived such treatment were eventually released into wild populations in Prince William Sound and along the Kenai Peninsula. Of these survivors, forty-five were equipped with radio-transmitters and monitored during subsequent months. The goal of the study reported herein, was to provide data on the survival and reproduction of the radio-instrumented sea otters, and by doing so, to gain insights into both the damage done to the Prince William Sound sea otter population by that spill and into the efficacy of the "rehabilitation" strategy.

OBJECTIVES

The specific objectives of this study were originally defined in the corresponding statement of work as follows:

- 1. To test the hypothesis that survival of sea otters that underwent oiling, cleaning, treatment and release is not different from that of sea otters that were not affected by the oil spill.
- 2. To test the hypothesis that reproductive rates of female sea otters that underwent oiling, cleaning and treatment does not differ significantly from that of female sea otters that were not affected by the oil spill.

METHODS

Definitions

Status classifications are made based on consideration of data through July 31, 1991. Individuals classified as "dead" are known to be dead because their carcass or other remains were observed and, in some cases, recovered. "Missing" individuals are those whose radio signal cannot be detected by boat or aircraft radio searches within Prince William Sound or adjacent areas along the Kenai Peninsula and Copper River Delta. The classification of "alive" is based upon visual observations of the individual.

Females were classified as having pupped based upon visual observations that they were accompanied by a pup.

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Study Groups

Data from the treatment center otters were compared with concurrent data from otters that were captured in Prince William Sound.

Forty-five adult sea otters (28 females (TC FEMALES) and 17 males (TC MALES)) were selected as candidates for radio-instrumentation from individuals being held at the three treatment centers (see Haebler et al. 1990). Of these, 9 were captured in Prince William Sound, 34 along the Kenai Peninsula and 2 in the Kodiak Archipelago (Table 1). Capture/admission dates for this group were distributed: April = 17 otters; May = 21 otters; June = 5 otters; July = 2 otters. The eastern Prince William Sound grouping (EPWS FEMALES) consisted of 44 females that were instrumented during 1987, 22 females that were instrumented during 1989 and 23 females that were instrumented during 1990. The western Prince William Sound grouping (WPWS FEMALES) consisted of 9 females instrumented during 1989 and 42 females instrumented during 1990. The EPWS FEMALES and WPWS FEMALES groupings were combined into the ALL FEMALES grouping.

Data on survival were separated temporally into two groupings relative to the release of the otters from the treatment centers: year one (August 1989 - July 1990) and year two (August 1990 - July 1991). Survival analysis was completed on both temporal groupings. Analysis of pupping rates was completed on only the 1990 data set because the 1991 pupping season is still underway.

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TIGATION SENSITIVE

Instrumentation and Monitoring

Individuals in this study were anesthetized and radiotransmitters were surgically implanted in their peritoneal cavities (Garshelis and Siniff 1983; DeGange and Williams 1990). After a recovery period, individuals from the treatment centers were released in eastern Prince William Sound during July and August, 1989. Sea otters in the EPWS FEMALE and WPWS FEMALE study groupings were released at the location of capture immediately after recovering from their anesthesia. An attempt was made to locate each individual at least once each week, using aircraft or boats equipped with Yagi antennas. Additional methodological details are provided in Monnett et al. (1990).

Analyses

Probabilities of survival and 95% confidence intervals (CI's) are calculated using Pollock et al.'s (1989) staggered entry modification to the Kaplan and Meier (1958) product limit procedure. Differences in the probability of survival between study groups are tested using the procedure described by Cox and Oakes (1984; see also Pollock et al. (1989) and White and Garrott (1990)). Contingency Chi-squared analyses were used to test for differences in pupping rates between study groupings. Differences were judged to be significant if the probability of type II error was < 0.05.



RESULTS AND DISCUSSION

Results of this study through spring 1990 have been previously reported (see Appendix II).

Survival rates

Of the 45 sea otters from the treatment centers that were instrumented and released, as of 31 July 1991, 14 were known dead, 16 were missing and presumed to have died, 1 radio-transmitter had malfunctioned prematurely. Fourteen individuals were alive and being monitored. Locations of the last radio-locations of dead and missing sea otters from the treatment centers are shown in Figure 1. Last locations of live sea otters from the treatment centers are shown in Figure 2. Data on the fates of sea otters by groupings used in the following analyses are summarized in Table 2 and Table 3.

No differences were found between the survival rates of male and female sea otters from the treatment centers for either year of the study; Year 1 probability survival: males = 0.401, females = 0.445, X^2 = 0.02, 1 DF, N.S.; Year 2 probability survival: males = 0.714, females = 0.692, X^2 = 0.003, 1 DF, N.S. (Table 4).

Male treatment center otters were not included in further survival analysis due to lack of sufficient sample sizes and lack of biologically appropriate groups for comparison.

For year-one, female sea otters from the treatment centers exhibited lower survival rates than female sea otters from groupings EPWS FEMALES (missing individuals were assumed to be dead: $X^2 = 13.82$, 1 DF, <u>p</u> < 0.001; missing individuals were excluded: $X^2 = 8.36$, 1 DF, <u>p</u>

< 0.01) and ALL FEMALES (missing individuals were assumed to be dead: $X^2 = 12.97$, 1 DF, p < 0.001; missing individuals were excluded: $X^2 = 6.88$, 1 DF, p < 0.02) (Table 4). Insufficient females were available in the WPWS FEMALES grouping to warrant inclusion in analysis for year-one.

LITIGATION SERVICE ATTORNEY/CLIENT WORK

For year-two, probability of survival was not significantly different between female sea otters from the treatment centers and female sea otters in the EPWS FEMALES and ALL FEMALES groupings. However, the females in the WPWS FEMALES did exhibit a higher survival rate than females released from the treatment center during year-two when missing individuals were assumed to have died (missing individuals were assumed to be dead: $X^2 = 5.93$, 1 DF, p < 0.02; missing individuals were excluded: $X^2 = 1.03$, 1 DF, N.S.) (Table 4).

We suggest that the lack of difference in survival rates between the TC FEMALES grouping and the EPWS FEMALES grouping (probability survival, missing individuals assumed to be dead: TC FEMALES = 0.692 cf. EPWS FEMALES = 0.648) should not be construed to indicate that treatment center females are exhibiting a "normal" rate of survival. Either value is abnormally low for prime-aged sea otter females (cf. probability survival year-two, WPWS FEMALES = 0.934, missing individuals assumed to be dead; and year-one values Table 4). The question of the unusually low survival rates for females in eastern Prince William Sound will be treated in a future technical report on the survival of non-treatment center sea otters.

Summaries of survival data, rates and confidence intervals for various groupings are given in tabular form in Appendix 3.

Pupping

Three females that were resident near the western end of the Kenai Peninsula were not included in the analysis of pupping rates because monitoring was infrequent and unreliable during the summer of 1990 (see Appendix I).

None of the 28 females released from the treatment centers pupped following release during the summer or fall, 1989. Fourteen of the 28 females survived through the summer of 1990; 11 were monitored adequately for data to be included in analysis (Kenai otters excluded as explained above). Based upon body size, all 11 were mature individuals and should have been capable of pupping during 1990. However, only 2 of the females pupped. The proportion of females released from the treatment centers that pupped was lower than the proportions of females pupping during 1990 in both the EPWS FEMALES grouping ($X^2 = 3.29$ 1 DF, P < 0.08) and the WPWS FEMALES grouping (X^2 = 6.19 1 DF, P < 0.02). However, the former comparison was not significant by the criteria established in the methods section (ie. p < 0.05). Reproduction by instrumented females released from the treatment centers is summarized in Table 6.

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Figure 1. Locations of last radio-telemetry fixes of dead and missing instrumented sea otters released from otter treatment centers during summer, 1989.





Figure 2. Summary of recent locations of sea otters released from otter treatment centers during 1989.





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TABLE 1. INDENTIFICATION AND STATUS INFORMATION OF SEA OTTERS FROM TREATMENT CENTER.

TREATM	ENT	CAPTIRE	TREATMENT	DATE	DATE	CURRENT	DATE LAST	LOCATION LAST	TOTAL DAYS
ID	SE	LOCATION	CENTER	IMPLANTED	RELEASED	STATUS	SEEN	SEEN	OBSERVEL
S-002	f	Tonsina B	Valdez(Sew)	17-Jul-89	26-Jul-89	alive	05-Apr-91	Nuka Passage	618
S-124	£	Rocky B, KP	Seward	04-Aug-89	15-Aug-89	missing	26-May-90	Cape Cleare	284
S-069	f	Rocky B, KP	Jakolof	11-Aug-89	22-Aug-89	alive	27-Jul-91	Fairmont I	704
S-162	f	Long I, Tonsina	Seward	04-Aug-89	15-Aug-89	missing	22-Sep-89	Windy B., Hawk.I	38
V-123	m	Natoa I	Valdez	17-Jul-89	26-Jul-89	dead	31-Mar-90	s.Latouche I	248
S-015	f	Bootleg B	Valdez(Sew)	17-Jul-89	26-Jul-89	missing	17-Oct-89	Port Fidalgo	83
S-157	f	Rocky B, KP	Seward	04-Aug-89	15-Aug-89	missing	04-Oct-89	Orca Inlet	50
S-068	f	Rocky B, KP	Valdez(Sew)	17-Jul-89	26-Jul-89	TXfailure	01-Sep-89	Deep B	37
V-048	m	Flemming I	Valdez(Sew)	17-Jul-89	26-Jul-89	dead	12-May-90	Strawberry Hill	290
V-139	ш	Crab B, Evans	Valdez	17-Jul-89	26-Jul-89	alive	25-Jul-91	Makarka Pt	730
V-104	m	Ikuta B	Valdez	17-Jul-89	26-Jul-89	dead	17-Apr-90	s.Valdez	265
S-045	f	Picnic Hbr.	Valdez(Sew)	17-Jul-89	26-Jul-89	alive	05-Apr-91	Windy B	618
S-060	f	Windy B	Seward	11-Aug-89	22-Aug-89	dead	04-Mar-90	s.Latouche I	194
S-152		Rocky R, entr.	Seward	11-Aug-89	22-Aug-89	missing	18-Sep-89	Kings B	27
S-161	f	Long I, Tonsina	Seward	04-Aug-89	15-Aug-89	alive	27-Jul-91	Olsen Is	711
Ś-038	8	Windy B	Jakolof	11-Aug-89	22-Aug-89	dead	26-Jan-90	Cordova Channel	157
V-130	m	Natoa I	Valdez	17-Jul-89	26-Jul-89	missing	02-Jun-90	Orca Inlet	311
S-003	f	Tonsina B	Jakolof	11-Aug-89	22-Aug-89	dead	22-Feb-90	n.Simpson B	184
V-138	m	Crab B, Evans	Valdez	17-Jul-89	26-Jul-89	missing	03-Feb-90	Danger I	192
S-080	f	Rocky B. KP	Jakolof	11-Aug-89	22-Aug-89	dead	10-Feb-90	Boswell Rock	172
S-054	f	Windy B	Jakolof	11-Aug-89	22-Aug-89	dead	20-Jan-90	Constantine Hbr	151
V-029	m	Green I	Valdez	17-Jul-89	26-Jul-89	alive	23-Jul-91	Channel I	727
S-006	f	Tonsina B	Jakolof	11-Aug-89	22-Aug-89	missing	30-Jul-90	sw.Montague I	342
S-122	m	Kupreanof St.	Jakolof	11-Aug-89	22-Aug-89	missing	22-Aug-89	Nelson B	C
V-145	f	Tonsina B	Valdez	13-Jul-89	26-Jul-89	missing	13-Oct-90	Port Etches	444
S-114	f	Uyak B.Kodiak	Jakolof	11-Aug-89	22-Aug-89	missing	04-Jan-90	Jack B	135
S-044	m	Taylor B	Jakolof	11-Aug-89	22-Aug-89	dead	22-Feb-90	Beartrap B	184
S057	f	so.B.Natoa I	Jakolof	11-Aug-89	22-Aug-89	missing	16-Dec-89	Sheep B	116
S-043	f	Taylor B	Sewarci	15-Jul-89	22-Aug-89	dead	17-Apr-90	Galena B	238
V-152	73	Berger B, KP	Valdez	17-Jul-89	26-Jul-89	dead	20-Aug-90	Deep B	390
S-007	£	Tonsina B	Jakolof	11-Aug-89	22-Aug-89	missing	28-Feb-91	Nagahut, Kenai	555
S-053	f	Windy B	Jakolof	11-Aug-89	22-Aug-89	missing	03-Feb-90	Thunder B	165
S-155	£	Rocky B, KP	Seward	04-Aug-89	15-Aug-89	missing	18-Jul-91	Red Head	702
S-017	f	Bootleg B	Valdez(Sew)	14-Jul-89	26-Jul-89	alive/pup	07-Aug-91	Constantine	742
S-146	f	Windy B	Seward	04-Aug-89	15-Aug-89	alive/pup	26-Jul-91	Yelper C	710
S-059	f	Windy B	Valdez(Sew)	14-Jul-89	26-Jul-89	alive	05-Aug-91	Green I	740
V-062		Hogan B,Knight I	Valdez(Sew)	17-Jul-89	26-Jul-89	alive	04-Aug-91	Mummy I	739
S-035	f	Windy B	Valdez(Sew)	17-Jul-89	26-Jul-89	dead	12-May-91	Hanning B	655
V-137	m	Crab B, Evans	Valdez	13-Jul-89	26-Jul-89	dead	31-Jan-90	Knowles Head	189
S-128	f	Rocky B, KP	Seward	04-Aug-89	15-Aug-89	dead	26-Aug-89	Hells Hole	11
V-150	£	Tonsina B	Valdez	13-Jul-89	15-Aug-89	alive	08-Aug-91	e.Simpson B	723
V-146	m	Hardover P, NukaB	Valdez	13-Jul-89	26-Jul-89	alive	02-Aug-91	Cape Cleare	737
V-068	f	Herring B	Valdez	13-Jul-89	26-Jul-89	alive	27-Jul-91	Boulder B	731
V-140	m	Crab B, Evans	Valdez	13-Jul-89	26-Jul-89	alive	09-Aug-91	Mud B	744
V-148	m	Bainbridge	Valdez	14-Jul-89	15-Aug-89	missing	26-Nov-90	Orca Inlet	468

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Table 2a. Summary of the fates of female sea otters radio instrumented and released from the sea otter treatment centers.

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Month	≠ at Risk	= Dead =	Missing Tx	Expired =	Added
Jul 89	0	0	0	0	9
Aug 89	9	1	0	0	19
Sep 89	27	0	1	1	0
Oct 89	25	0	2	0	0
Nov 89	23	0	0	0	0
Dec 89	23	0	1	0	0
Jan 90	22	1	1	0	0
Feb 90	20	2	1	0	0
Mar 90	17	1	0	0	0
Apr 90	16	1	0	0	0
May 90	15	0	1	0	0
Jun 90	14	0	0	0	0
Jul 90	14	0	1	0	0
Aug 90	13	0	0	0	0
Sep 90	13	0	0	0	0
Oct 90	13	0	0	0	0
Nov 90	13	0	1	0	0
Dec 90	12	0	0	0	0
Jan 91	12	0	0	0	0
Feb 91	12	0	1	0	0
Mar 91	11	0	0	0	0
Apr 91	11	0	0	0	0
May 91	11	1	0	0	0
Jun 91	10	0	0	0	0
Jul 91	10	0	1	0	0
Aug 91	9				

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Table 2b. Summary of the fates of male sea otters radio instrumented and released from the sea otter treatment centers.

Month	≠ at Risk	≠ Dead #	Missing Tx	Expired =	Added
Jul 89	0	0	0	0	12
Aug 89	12	0	1	0	ō
Sep 89	16	0	1	0	0
Oct 89	15	0	0	0	0
Nov 89	15	0	0	0	0
Dec 89	15	0	0	0	0
Jan 90	15	2	0	0	0
Feb 90	13	1	1	0	0
Mar 90	11	1	0	0	0
Apr 90	10	1	0	0	0
May 90	9	1	0	0	0
Jun 90	8	0	1	0	0
Jul 90	7	0	0	0	0
Aug 90	7	1	0	0	0
Sep.90	6	0	0	0	0
Oct 90	6	0	0	0	0
Nov 90	6	0	0	0.	0
Dec 90	6	0	1	0	0
Jan 91	5	0	0	0	0
Feb 91	5	0	0	0	0
Mar 91	5	0	0	0	0
Apr 91	5	0	0	0	0
May 91	5	0	0	0	0
Jun 91	5	0	0	0	0
Jul 91	5	0	0	0	0
Aug 91	5				

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Table 3. Summary of the fates of radio-instrumented sea otters in study groupings used in survival analysis for comparison with sea otters released from sea otter treatment centers.

	1	987	St	udy		1	987	St	.udy			198	9-9	0			198	9-9	0			198	10_C	n	
	EP	WS	Fem	ale	S	E	PWS	Ma	les		EP	WS	Fem	ale	es	WP	WS	Fer	ale	S	Y	IPWS	i Ma	iles	
Month	R ₁	D	М	E	A	R	D	М	E	A	R	D	М	E	A	R	D	М	E	۸	R	D	М	E	A
Jul 89	39	1	0	0	0	12	0	0	0	0															
Aug 89	38	1	1	0	0	12	3	0	0	0															
Sep 89	36	0	0	0	0	9	0	1	0	0															
Oct 89	36	Ó	0	0	0	8	0	0	0	0					22										
Nov 89	36	1	0	1	0	8	0	0	0	0	22	0	0	0	0					я					0
Dec 89	34	0	1	1	0	8	0	1	0	0	22	0	2	0	0	8	0	0	0	ñ	2	Λ	Δ	0	0
Jan 90	32	0	1	0	0	7	1	0	0	0	20	0	0	0	Ő	Ř	õ	ŏ	õ	õ	2	ň	0	0	0
Feb 90	31	0	0	2	0	6	1	1	0	0	20	0	Ō	0	Õ	8	Ő	õ	ñ	ñ	2	0	0	0	0
Mar 90	29	0	0	1	0	4	1	0	0	0	20	ì	Õ	0	14	8	õ	ñ	0	0	2	0	0	0	0
Apr 90	28	0	0	0	0	3	0	0	0	0	33	0	Õ	Ő	4	8	õ	õ	ň	20	2	0	0	0	0
May 90	28	0	0	1	0	3	0	0	0	0	37	1	õ	0	0	47	ŏ	õ	0	0	2	0	0	0	1
Jun 90	27	0	0	4	0	3	0	0	1	0	36	Õ	0	Ő	Õ	47	õ	õ	ň	ñ	3	0	0	0	0
Jul 90	23	0	0	2	0	2	0	0	0	0	36	Õ	õ	õ	õ	47	ň	ñ	0	0	່ 3 3	0	0	0	0
Aug 90	21	0	0	3	0	2	0	0	0	0	36	Õ	õ	ŏ	õ	47	ň	ň	0	Ň	ວ 2	0	0	0	0
Sep 90	18	0	0	5	0	2	0	0	0	0	36	Õ	ĩ	ŏ	2	47	1	ň	0	0 0	2	0	0	0	0
Oct 90	13	0	0	0	0	2	0	0	2	0	37	Ĩ	1	õ	2	46	Ô	0	ñ	ñ	່ 3 2	0	0	0	0
Nov 90	13	0	0	5	0	0					37	0	1	Ő	õ	46	ň	ň	0	0	່ ງ	0	0	0	0
Dec 90	8	0	0	8	0						36	1	1	õ	õ	46	ň	0	0	0	3	0	0	0	0
Jan 91	0										34	Ô	Ō	õ	õ	46	0	0	0	0	ວ 2	0	1	0	0
Feb 91											34	õ	ň	ŏ	ň	44	ň	ň	0	0	ა ი	0	1	0	0
Mar 91											34	ĭ	ŏ	ŏ	ň	44	õ	0	0	0	4	0	0	0	0
Apr 91											33	Ô	ĩ	ŏ	ñ	44	ň	0	0	0	2	0	0	0	0
May 91											32	õ	Ô	ň	õ	44	ň	0	0	0	2	0	0	0	0
Jun 91											32	õ	4	õ	õ	44	.0	2	0	0	4	0	0	0	0
Jul 91											28	õ	3	õ	õ	42	0	ő	0	0	2	0	0	0	0
Aug 91											25	Ť	Ŭ	Ý	v	42	Ŷ	v	v	U	2	U	U	U	U

1KEY:

R: Number of sea otters at risk during month

D: Number of sea otters that died during month

M: Number of sea otters classified missing during month

E: Number of sea otters having transmitters expire during month

A: Number of sea otters added to study during month

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Table 4. Summary of statistics on survival of sea otters radioinstrumented and released in Prince William Sound. Study groupings include individuals released from otter treatment centers (T.C.), individuals from eastern Prince William Sound (EPWS) and individuals from western Prince William Sound (WPWS).

MISSING ASSUMED DEA	D		•		
1000 00	<u>p</u> Surviv	al C.I.	\mathbf{x}^2	D.F.	D
1383-30					
T.C. Females	0.445	(0.223 - 0.667)	0.02	1	N.S.
1.C. Mares	0.401	(0.133-0.010)			
1990-91					
T.C. Females T.C. Males	0.692 0.714	(0.421-0.964) (0.314-1.115)	0.003	1	N.S.

1989-90

MISSING	ASSUMED DEAD	₽	Survival	C.I.	x ²	D.F.	₽
	All Females T.C. Females		0.85 0.445	(0.757-0.943) (0.223-0.667)	13.82	1	0.001
	EPWS Females T.C. Females		0.834 0.445	(0.733-0.934) (0.223-0.667)	12.97	1	0.001
MISSING	ELIMINATED	₽	Survival	С.І.	x ²	D.F.	Ð
	All Females T.C. Females		0.932 0.674	(0.862-1.001) (0.412-0.935)	8.36	1	0.01
	EPWS Females T.C. Females		0.923 0.674	(0.847-0.999) (0.412-0.935)	6.88	1	0.02
	T.C. Females		0.674	(0.412-0.935)	U.00	+	0.02

1990-91

MISSING	ASSUMED DEAD						
		₽	Survival	. C.I.	x ²	D.F.	P
	All Females T.C.Females		0.798 0.692	(0.716-0.880) (0.421-0.964)	0.79	1	N.S.
	EPWS Females T.C.Females		0.648 0.692	(0.506-0.790) (0.421-0.964)	0.19	1	N.S.
	WPWS Females T.C.Females		0.934 0.692	(0.862-1.006) (0.421-0.964)	5.93	1	0.02
MISSING	ELIMINATED	₽	Survival	C.I.	x ²	D.F.	₽
	All Females T.C.Females		0.956 0.909	(0.915-0.998) (0.713-1.105)	0.3	1	N.S.
	EPWS Females T.C.Females		0.930 0.909	(0.848-1.011) (0.713-1.105)	0.19	1	N.S.
	WPWS Females T.C. Females		0.979 0.909	(0.936-1.021) (0.713-1.105)	1.03	1	N.S.

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TABLE 5. SUMMARY OF REPRODUCTION BY FEMALES FROM TREATMENT CENTERS VS. WILD CAPTURED FEMALES IN PRINCE WILLIAM SOUND 1990.

> Females Females Pupping Not Pupping

Treatment Center Females	2	9	$x^2 = 3.29$
1989-90 East PWS Females	14	14	$\underline{P} < 0.08$

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Females Females Pupping Not Pupping

Treatment Center Females	2	9	$x^2 = 6.19$
1989-90 West PWS Females	22	14	p < 0.02

Females Females Pupping Not Pupping

Treatment Center Females	2	9	$x^2 = 5.52$
1989-90 All PWS Females	36	28	<u>p</u> < 0.02

Table 6. Summary of reproduction by individual instrumented female sea otters following release from sea otter treatment centers.

ТΧ	Otter ID	July 1990 Status	Date Last Seen	Est. Date Pupped	Fate of Pup	July 1991 Status	Date Last Seen	Est.Date Pupped	Fate of Pup
4098	S-002	alive		didn't nun		aliyo	05 Apr 01		
4135	S-124	missing	26-May-90	didn't pup		anve	05-Apr-51	dian't pup	
4148	S-069	alive		19-Oct-90	weaned	alive	27-111-01	didn't num	
4176	S-162	missing	22-Sep-89	didn't pup		unve	21 Jul-31	ulun t pup	
4225	S-015	missing	17-Oct-89	didn't pup					
4238	S-157	missing	04-Oct-89	didn't pup					
4257	S-068	TX failure	01-Sep-89	didn't pup					
4340	S-045	alive	-	06-Apr-90	unknown	alive	05-Apr-91	didn't nun	
4355	S-060	dead	04-Mar-90	didn't pup			oo npr or	dian't pup	
4398	S-161	alive		didn't pup		alive	27-Jul-91	didn't pup	
4447	S-003	dead	22-Feb-90	didn't pup				aran e pap	
4478	S-080	dead	10-Feb-90	didn't pup					
4498	S-054	dead	20-Jan-90	didn't pup					
4547	S-006	alive		didn't pup		missing	30-Jul-90	didn't pup	
4593	V-145	alive		didn't pup		missing	13-Oct-90	didn't pup	
4608	S-114	missing	04-Jan-90	didn't pup				1.1	
4649	S-057	missing	16-Dec-89	didn't pup					
4696	S-043	dead	17-Apr-90	didn't pup					
1728	S-007	alive		didn't pup		missing	28-Feb-91	didn't pup	
1755	S-053	missing	03-Feb-90	didn't pup				•••	
4789	S-155	alive		didn't pup		missing	18-Jul-91	didn't pup	
4796	S-017	alive		31-Dec-89	weaned	alive/pup	07-Aug-91	07-Feb-91	w/mother
4815	S-146	alive		didn't pup		alive/pup	26-Jul-91	16-May-91	w/mother
4825	S-059	alive		didn't pup		alive	05-Aug-91	didn't pup	
4857	S-035	alive		didn't pup		dead	12-May-91	didn't pup	
4928	S-128	dead	26-Aug-89	didn't pup					
4935	V-150	alive		didn't pup		alive	08-Aug-91	didn't pup	
4300	v-008	anve		aidn't pup		alive	27-Jul-91	didn't pup	

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Technical Report: Marine Mammal Study Number 6

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Mortality and Reproduction of Female Sea Otters in Prince William Sound, Alaska

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SUMMARY

Ninety-six female sea otters were instrumented with implanted radio-transmitters in Prince William Sound, Alaska, during 1989-1990. Females in eastern Prince William Sound exhibited a lower survival rate than those in western Prince William Sound. No differences were observed between rates of pupping or between rates of survival of dependent pups for sea otters in the two areas.

INTRODUCTION

On March 24, 1989, over 11 million gallons of crude oil were spilled in Prince William Sound, Alaska, due to the wreck of the *T/V Exxon Valdes*. The research discussed in this report was undertaken as part of Natural Resource Damage Assessment studies aimed at determining if the spill caused damage to the sea otter population(s) in the region, and, if so, the type, magnitude, and significance of the damage(s). The goals of this study were to determine whether the mortality and reproductive rates of adult females were different in areas within or near the areas through which large amounts of crude oil were spilled than in areas in which no crude oil was known to have passed. This information is crucial to understanding the overall extent of damage to the sea otter population(s); to

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estimating the rate and pattern of recovery; and to formulating restoration and response policies for sea otters throughout their range.

OBJECTIVES

The specific objectives of this study were defined in the corresponding statement of work as follows:

- 1. To test the hypothesis that survival of adult female sea otters is not different in oiled and unoiled areas.
- 2. To test the hypothesis that pupping rates of adult female sea otters are not different between oiled and unoiled areas.
- 3. To test the hypothesis that pup survival pre-weaning is not different between oiled and unoiled areas.

METHODS

Definitions

Status classifications are made based on consideration of data through July 31, 1991. Individuals classified as "dead" are known to be dead because their carcass or other remains were observed and, in some cases, recovered. 4

detected by boat or aircraft radio searches within Prince William Sound or adjacent areas along the Kenai Peninsula and Copper River Delta. The classification of "alive" is based upon visual observations of the individual.

Females were classified as having pupped based upon visual observations that they were accompanied by a pup.

Study Groups

The eastern Prince William Sound grouping (EPWS) consists of 22 females that were instrumented during 1989 and 23 females that were instrumented during 1990. The western Prince William Sound grouping (WPWS) consisted of 9 females instrumented during 1989 and 42 females instrumented during 1990 (Table 1). Capture locations are summarized in Figure 1.

Instrumentation and Monitoring

Individuals in this study were anesthetized and radiotransmitters were surgically implanted in their peritoneal cavities (Garshelis and Siniff 1983; Monnett 1988). Sea otters were released at the location of capture immediately after recovering from their anesthesia. An attempt was made to locate each individual at least once biweekly, using aircraft or boats equipped with Yagi antennas.

Analyses

Probabilities of survival and 95% confidence intervals (CI's) are calculated using Pollock et al.'s (1989) staggered entry modification to the Kaplan and Meier (1958) product limit procedure. Differences in the probability of survival between study groups are tested using the procedure described by Cox and Oakes (1984; see also Pollock et al. (1989) and White and Garrott (1990)). Contingency Chisquared analyses were used to test for differences in pupping rates between study groupings. Differences were judged to be significant if the probability of type II error was < 0.05.

Analysis of survival of dependent pups was confined to pups during the first 60 days following birth. It has been shown that see otter pups in Prince William Sound may become independent and survive at less than 90 days of age (Monnett 1988). Thus, it was not possible to ascertain whether separation of older pups from mothers was due to weaning or mortality.

RESULTS AND DISCUSSION

Monitoring .

Intervals between radio-locations were on average: EPWS (1990) = 5.8 days (SD = 1.0), (1991) = 5.9 days (SD = 3.2); WPWS (1990) = 8.7 days (SD = 1.4), (1991) = 11.1 days (SD = 4.7). Intervals between visual observations were on $= 2^{-1} (1990) = 8.6 \text{ days}$ (SD = 1.8), (1991) = 7.3

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days (SD = 5.3); WPWS (1990) = 10.7 days (SD = 2.6), (1991) = 12.7 days (SD = 5.3).

Survival rates of adult females

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If females that were classified as missing are assumed to have died, the survival rate of females in WPWS was higher than that of females in EPWS (Table 2). If females classified as missing are excluded from the analysis, no differences exist (Table 2).

Pupping rates

No differences were found in pupping rates of adult females between EPWS and WPWS in either 1990 or 1991: (1990) EPWS = 13/28 (46%) females pupped versus WPWS = 21/36 (58%) females pupped (X^2 = 0.92, 1 DF, p > 0.50); (1991) EPWS = 21/30 (70%) females pupped versus WPWS = 29/37 (78%) females pupped (X^2 = 0.61, 1 DF, p > 0.50).

Pup survival

The survival rates of dependent pups for the first 60 days following birth were compared between EPWS and WPWS. No differences were found between the survival rates of dependent pups in either 1990 or 1991: (1990) EPWS = 9/13 (69%) pups survived versus WPWS = 15/21 (76%) pups survived $(X^2 = 0.19, 1 \text{ DF}, p > 0.70)$; (1991) EPWS = 17/21 (81%) pups survived versus WPWS = 28/29 (97%) pups survived $(X^2 = 3.29, 1 \text{ DF}, p < 0.08)$.

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Figure 1. Capture locations of female sea otters in Prince William Sound.



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Table 1. Data on instrumentation of female sea otters in Prince William Sound (PWS). Study groupings: E.P.W.S. = Eastern Prince William Sound; W.P.W.S. = Western Prince William Sound.

	Study	Date	Location
Otter ID	Grouping	Instrumented	Instrumented
99101	T. D. W. S.	08-Oct-89	Sheep Bay
00102	T D.W.S.	08-0ct-89	Sheep Bay
99102	E.P.W.S.	08-Oct-89	Sheep Bay
89103	F P.W.S.	08-Oct-89	Sheep Bay
03104		09-0ct-89	Sheep Bay
00106		12-0ct-89	North Island
03100	E E W C	12-Oct-89	North Island
00100	T.D.W.S.	12-0ct-89	North Island
20100	E.P.W.S.	12-Oct-89	North Island
99105	E.P.W.S.	12-Oct-89	North Island
20111	E.P.W.S.	12-Oct-89	North Island
20112	E.P.W.S.	13-Oct-89	North Island
00113	E.P.W.S.	13-Oct-89	North Island
89113	F. D. W. S.	13-Oct-89	North Island
20115	E.P.W.S.	20-Oct-89	North Island
99116 99116	E.P.W.S.	20-Oct-89	North Island
89117	E.P.W.S.	20-Oct-89	North Island
89118	E.P.W.S.	20-Oct-89	North Island
89121	E.P.W.S.	22-Oct-89	North Island
89122	E.P.W.S.	22-Oct-89	North Island
89124	E.P.W.S.	22-Oct-89	North Island
89125	E.P.W.S.	22-Oct-89	North Island
89126	E.P.W.S.	22-Oct-89	North Island
89127	W.P.W.S.	04-Nov-89	Chicken Island, Latouche P
89128	W.P.W.S.	06-Nov-89	Bainbridge Passage
89131	W.P.W.S.	07-Nov-89	Bainbridge Passage
89140	W.P.W.S.	12-Nov-89	Port Chalmers
89141	W.P.W.S.	13-Nov-89	Port Chalmers
89142	W.P.W.S.	13-Nov-89	Channel Island, Green 15.
89150	W.P.W.S.	15-Nov-89	Port Chaimers
89153	W.P.W.S.	15-Nov-89	Port Chaimers
89155	W.P.W.S.	15-Nov-89	Port Chaimers
90001	E.P.W.S.	16-Mar-90	North Island
90004	E.P.W.S.	16-Mar-90	North Island
90005	E.P.W.S.	10-M81-90	North Island
90006	E.P.W.S.	10-Mar-90	North Island
90008	E.P.W.S.	70-M81-90 70-M81-90	Quarry, Orca Inlet
90013	E.P.W.S.	22-Mar-90	Quarry, Orca Inlet
90014	5.2.W.S.	22-mar-30 94-Man-00	Quarry, Orca Inlet
90016	E.F.W.D.	24-Mar-90	Quarry, Orca Inlet
20010	$\Box \cdot F \cdot \overline{H} \cdot \Box \cdot$ $\nabla \cdot \Box U C$	26-Mar-90	Quarry, Orca Inlet
20019	EIFIMIDI F. P. W. S.	28-Mar-90	Quarry, Orca Inlet
90019	E.P.W.S.	26-Mar-90	Quarry, Orca Inlet
90022	E.P.W.S.	28-Mar-90	Quarry, Orca Inlet
00002	F.P.W.S.	27-Mar-90	Quarry, Orca Inlet

Table 1 (cont.). Data on instrumentation of female sca otters in Prince William Sound (PWS). Study groupings: E.P.W.S. = Eastern Prince William Sound; W.P.W.S. = Western Prince William Sound.

	Study	Date	Location
Otter ID	Grouping	Instrumented	Instrumented
			Chasm Past
90024	E.P.W.S.	04-Apr-90	Sheep Bay
90027	E.P.W.S.	05-Apr-90	Sheep Bay
90028	E.P.W.S.	05-Apr-50	Sheep Day
90029	E.P.W.S.	08-Apr-90	Sheep day Iiii a Green Taland
90031	W.P.W.S.	11 Apr 90	Bont Chalman
90033	W.P.W.S.	11-Apr-90	Port Chalmers
90034	$W_{1}P_{1}W_{1}D_{1}$	11-App-90	Port Chalmers
90035	W . T . W . D . U D W S	11-Apr-90	Port Chalmers
90036	W · F · W · O · M · D · M · C	$\frac{11-App-90}{11-App-90}$	Port Chalmers
89010		$\frac{11 - Apr - 90}{11 - Apr - 90}$	Little Green Island
90037	W P W S	11-App-90	Dost Chalmers
90038		13-Apr=90	Source Island, Knight Is.
90039	M.F.M.J. W D W S.	13 = Apr = 90	Squire Taland, Knight IS.
90040	W.P.W.S.	13-Apr-90	Squire Island, Knight Is.
90041	W.D.W.S.	13 - Apr = 90	Squire Island, Knight Is.
90042	W.P.W.S.	13-Apr-90	Squire Island, Knight Is.
00040	W.P.W.S.	13-Apr-90	Squire Island, Knight Is.
90044	W.P.W.S.	13-Apr-90	Squire Island, Knight Is.
90046	W.P.W.S.	13-Apr-90	Squire Island, Knight Is.
90047	W.P.W.S.	13-Apr-90	Squire Island, Knight Is.
90048	W.P.W.S.	13-Apr-90	Squire Island, Knight Is.
90049	W.P.W.S.	13-Apr-90	Squire Island, Knight Is.
90052	W.P.W.S.	22-Apr-90	Mummy Island, Knight Is.
90053	W.P.W.S.	22-Apr-90	Mummy Island, Knight Is.
90054	W.P.W.S.	22-Apr-90	Mummy Island, Knight Is.
90055	W.P.W.S.	22-Apr-90	Mummy Island, Knight Is.
90056	W.P.W.S.	23-Apr-90	Iktua Bay, Evans Island
90057	W.P.W.S.	24-Apr-90	Squire Island, Knight 18.
90058	W.P.W.S.	24-Apr-90	Squire Island, Knight 19.
90059	W.P.W.S.	24-Apr-90	Squire Island, Knight Is.
90061	W.P.W.S.	28-Apr-90	Squirret island, Anight Is,
90062	W.P.W.S.	20-Apr-90	Squirrei Island, Knight Is.
90063	W.P.W.S.	20-Apr-90	Squirrel Island, Knight Is.
90064	W. P. W. S.	20-Apr-90	Mummer Bay Reaf
90065	W.F.W.S.	27-A01-90 28-A01-90	Stockdale Harbor
90066	M, P, M, D, M D W S	28-Apr-90	Stockdale Harbor
90007	Π·Γ·Π:Ο· 4.0.4.5.	28-Apr-90	Stockdale Harbor
90000	W.P.W.S.	29-Apr-90	Stockdale Harbor
90071	W.P.W.S.	29-Apr-90	Stockdale Harbor
90072	W.P.W.S.	29-Apr-90	Stockdale Harbor
90073	W.P.W.S.	29-Apr-90	Stockdale Harbor
90074	W.P.W.S.	30-Apr-90	Little Green Island
90075	W.P.W.S.	30-Apr-90	Little Green Island

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