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INJURY TO PINK SALMON EMBRYOS IN PRINCE WILLIAM SOUND - FIELD MONITORING

EXXON VALDEZ RESTORATION PROJECT 97191A-1

by

Andrew K. Craig,
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Exxon Valdez Oil Spill
Restoration Project Annual Report

Injury to Pink Salmon Embryos in Prince William Sound - Field monitoring

Restoration Project 97191A-1
Annual Report

This annual report has been prepared for peer review as part of the *Exxon Valdez* Oil Spill Trustee Council restoration program for the purpose of assessing project progress. Peer review comments have not been addressed in this annual report.

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Injury to Pink Salmon Embryos in Prince William Sound

Restoration Project 97191
Annual Report

Study History: This study originated in March of 1989 and continued through February of 1991 as Natural Resource Damage Assessment Fish/Shellfish Study 2. The project consisted of embryo sampling in the fall and preemergent fry sampling in the spring at oil-contaminated and unimpacted reference streams to determine if the *Exxon Valdez* oil spill affected incubating pink salmon. This work continued in 1992 as Restoration Study R60C. At that time the project was expanded to include (1) laboratory evaluation of field results through the controlled incubation of pink salmon embryos on oiled substrate (NOAA); (2) an experiment designed to determine if the results observed in the field were due to environmental factors (ADFG); and (3) a search for evidence of genetic damage (ADFG). This work was continued as Restoration Study 93003, Restoration Study 94191, Restoration Study 95191, Restoration Study 96191, and this project, Restoration Study 97191. Final reports have been written for Fish/Shellfish Study 2 and Restoration Studies R60C, 93003, 94191, 95191 and 96191.

Abstract: We examined pink salmon embryo mortality in intertidal and upstream areas of both oil-contaminated and reference streams in Prince William Sound. Embryo mortality was elevated in oil-affected streams during the falls of 1989 through 1993 ($P<0.028$ for all years). However; no statistical difference was observed in the fall of 1994, 1995 or 1996 ($P>0.473$). We also tested the hypothesis that differences in embryo mortality observed in the field were due to naturally occurring environmental variables that differed systematically between the oil-contaminated and reference streams. Gametes were collected from adults in spawning condition from eight oil-contaminated and eight reference streams, and matings were conducted at a hatchery. The resulting embryos were incubated in controlled environmental conditions. Embryos originating from oil-contaminated streams showed elevated mortalities when compared to the embryos from reference streams in 1993 ($P=0.012$), but not in 1994 ($P=0.343$). Results from the controlled incubation study support the results from the field study.

Key Words: crude oil, embryo mortality, embryos, *Exxon Valdez*, flow cytometry, genetic damage, *Oncorhynchus gorbuscha*, pink salmon, preemergent fry, Prince William Sound.

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EXECUTIVE SUMMARY

This study was designed to monitor recovery of pink salmon *Oncorhynchus gorbuscha* populations in Prince William Sound that were impacted by the *Exxon Valdez* oil spill. Embryo mortality and embryo to preemergent fry survival have been examined in intertidal and upstream areas of oil-contaminated and unaffected (reference) streams since the spring of 1989, and the potential of crude oil to induce genetic damage has been examined since the fall of 1992. This report focuses upon work performed between October 1, 1996 and September 30, 1997.

Embryo mortality was significantly greater in oiled streams in 1989 and 1990 with the differences observed in all intertidal areas in 1989 and in the highest intertidal area in 1990. These results were consistent with other studies regarding observations of intertidal oiling. Among oiled streams, all intertidal areas were contaminated in 1989 whereas in 1990 visible oil remained only in the upper intertidal zone.

The 1991 evaluation demonstrated a significant difference in embryo mortality between oil-contaminated and reference streams in both the intertidal and upstream zones. This finding was unexpected, as the presence of oil was dramatically reduced in all areas. The result led investigators to hypothesize that one or more of the following scenarios were at work:

(1) oil-induced damage to the 1989 and 1990 broods included deleterious mutations in the germline;
(2) incubating embryos continued to be damaged in a physiological manner by an oiled environment even after visible oil was absent, and this impact was expressed as functional sterility;
(3) the difference in embryo mortality was due to naturally-occurring environmental factors that differed uniformly between oiled and non-oiled streams.

The genetic-damage and physiological-damage hypothesis seemed credible because oil is a known clastogenic substance (breaks chromosomes), and it also influences endocrine function. Pink salmon have an obligate two-year life cycle, and those fish that spawned during the fall of 1991 incubated in oiled gravels during the fall of 1989 and spring of 1990. Also, a pattern of embryo mortality similar to, but not as extreme as that found in 1991, was observed in 1992 and 1993. No statistical difference in embryo mortality was detected in 1994 and 1995.

The 1993 embryos were two generations removed from oil exposure in 1989. The environmental-difference hypothesis seemed credible because, in fact, it was environmental factors (wind and currents) that determined the fate of the oil. Such environmental factors might also influence the survivability of salmon embryos incubating intertidally.

We tested the hypothesis that differences in pink salmon embryo mortality observed in recent years were due to environmental differences between oiled and reference streams. Gametes were collected from adults in spawning condition as they amassed on or near the spawning grounds from eight oil-contaminated and eight reference streams during the 1993 spawning season. The gametes were flown to the Armin F. Koernig hatchery in southwest Prince William Sound where intrastream crosses were made. The resulting embryos from each stream were placed in a common incubator. The pink salmon embryos from oil-contaminated streams showed elevated mortalities when compared to the embryos from reference streams. This finding clearly indicated that the elevated embryo mortalities observed in the field monitoring portion of the study were not due to systematic differences between the incubating environments of oiled and reference streams. This embryo incubation experiment was repeated in 1994, but no significant difference in embryo mortality between oil-contaminated and reference streams was detected. This result is consistent with results obtained from the field monitoring portion of the project in 1994. This study was attempted in 1995 but was incomplete due to lack of spawners in some study streams.

Lack of a significant difference in pink salmon embryo mortality in 1994, 1995 and 1996 between oiled and reference streams demonstrates a possible recovery of the populations that have been monitored since 1989.

INTRODUCTION

Wild salmon play a major role in the Prince William Sound (PWS) ecosystem while also contributing to the region's commercial fisheries. Migrating salmon fry are an important food source in the spring for various mammals, birds, and fishes. Marine mammals prey on the ocean life stages of Pacific salmon while terrestrial mammals and birds, such as bears, river otters, eagles, and gulls depend on salmon for a large portion of their summer diet. Salmon also provide a pathway for transferring nutrients from marine ecosystems to near-shore and terrestrial ecosystems. In recent years, commercial catches of wild salmon in Prince William Sound have ranged from 10 to 15 million pink salmon and from 0.8 to 1.5 million chum salmon.

Up to 75% of spawning pink *Oncorhynchus gorbuscha* and chum *O. keta* salmon in PWS use intertidal areas (Helle et al. 1964). These areas are highly susceptible to contamination from marine oil spills. Moles et al. (1987) and Rice et al. (1975) found that pink salmon embryos and preemergent fry were adversely affected by exposure to crude oil and that the affect was most acute in intertidal environments. The March 24, 1989 oil spill from the *T\V Exxon Valdez* contaminated many intertidal spawning areas in central and southwest PWS just prior to the spring migration of salmon fry.

Embryo mortality was significantly greater in all intertidal areas of oiled streams in 1989 and in the highest intertidal area of oiled streams in 1990 (Figure 1) (Sharr et al. 1994a, Bue et al. 1996). These results were consistent with observations of intertidal oil-contamination (Wolfe et al. 1996). Among oiled streams, all intertidal areas were contaminated in 1989 whereas in 1990 oil remained only in the upper intertidal zone.

The 1991 evaluation demonstrated a very significant and large difference in embryo mortality between oil-contaminated and reference streams (Figure 1; Sharr et al. 1994a). This finding was unexpected and raised several questions about the source of the elevated mortality in oiled streams, including the possibility that oil-induced damage was transmitted genetically. Petrochemicals have been shown to damage chromosomes (Longwell 1977; McBee and Bickham 1988; Hose et al. *in press*). The pink salmon which spawned during the fall of 1991 were from the 1989 brood year. These fish incubated in oiled gravels during the fall of 1989 and spring of 1990. A pattern of embryo mortality similar to but not as extreme as 1991 was observed in 1992 and 1993 (Figure 1; Sharr et al. 1994b and Sharr et al. 1994c). Field sampling in 1994 and 1995 however, showed no statistical difference between oiled and reference streams (Figure 1; Craig et al. 1996).

Three hypotheses have been proposed to explain differences in embryo mortality between oil-contaminated and reference streams: (1) environmental differences between oiled and reference streams are responsible, (2) transmission of genetic damage is responsible, and (3) incubating embryos continued to be damaged in a physiological manner by an oiled environment that affected later reproduction.

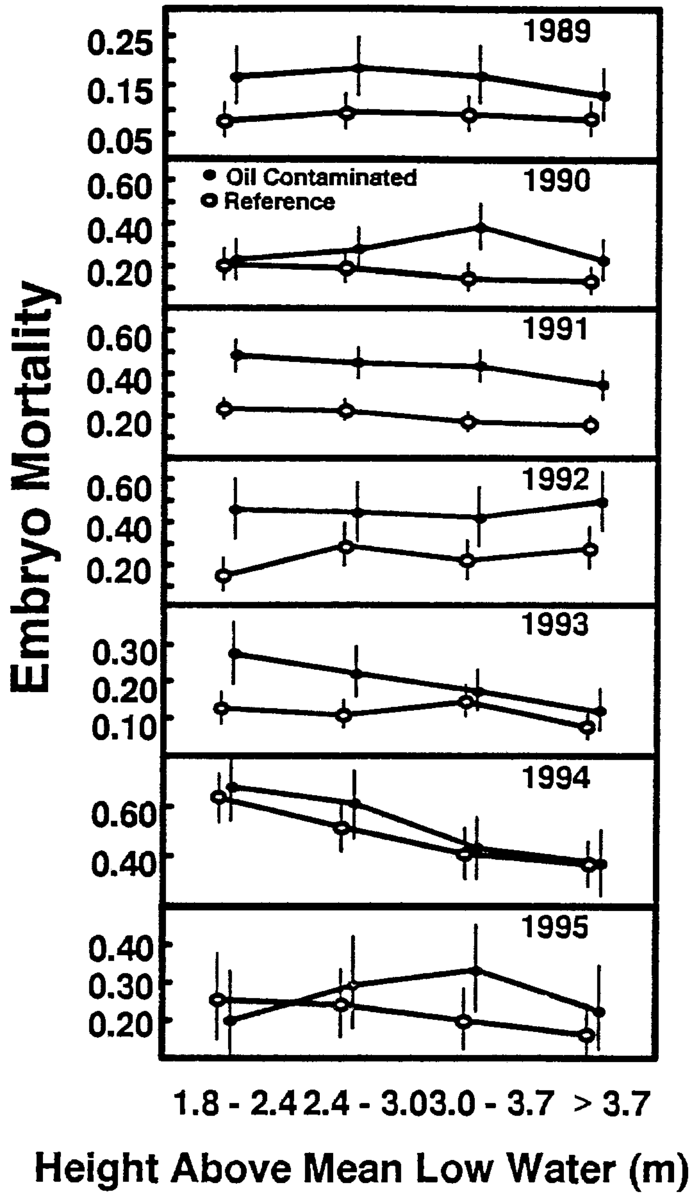

Figure 1. Pink salmon embryo mortality observed during field sampling. Solid circles indicate the means for oil-contaminated streams (n=10) while open circles identify reference streams (n=15). Note: Y-axes differ among years.

Observed differences in embryo mortality may have been due to differences in environmental conditions between oil-contaminated and reference streams. This embryo mortality study is based on observational data, and as such, we were unable to randomize stream oiling to account for environmental differences between streams. We attempted to address this concern in our original experimental design by selecting reference streams in close proximity to oil-contaminated streams; however, there is a definite oiling pattern in southwest PWS where streams on points which faced northeastward were heavily oiled. Likewise, streams which faced west and southwest were most likely not oiled. Environmental differences between oil-contaminated and reference streams do not seem to be a confounding factor because results from the controlled incubation experiment in 1993 and 1994 mirrored the results of the field study (Bue et al. 1996).

Both hypotheses of genetic damage and physiological damage are consistent with previous field observations and laboratory experiments on the effects of crude oil on early life stages of fish. Long term intra-gravel oil exposures (7-8 months) to freshly fertilized eggs provide embryos sufficient time to accumulate polycyclic aromatic hydrocarbons (PAH's) from very low aqueous concentrations of crude oil. Moles et al. (1987) confirmed that pink salmon embryos take up PAH's and demonstrated that the uptake was much greater in an intertidal environment than in strictly freshwater conditions. PAH's are abundant in crude oil and are capable of inducing chromosomal lesions (McBee and Bickham 1988) and influencing endocrine function (Thomas and Budiantara 1995) and later reproduction (Trustcott et al. 1983). Mironov (1969) observed reduced survival of fish embryos and larvae exposed to very low aqueous doses (1 ul oil/l seawater) of oil. Longwell (1977) reported genetic damage in pelagic embryos affected by the *Argo Merchant* oil spill. It is logical that the same type of damage may have occurred in pink salmon, and this damage could have affected the germline of exposed individuals (cf., Malkin 1994). Pink salmon have an obligate two-year life cycle which results in two genetically isolated lineages, one produced during odd years and one in even years. Therefore, genetic or physiological damage induced in one brood year would be expressed in that lineage two years later.

This study was initially designed to monitor the effect of intertidal oiling on pink salmon embryo mortality and embryo to preemergent fry survival. The project was amended during the summer of 1992 to evaluate the systematic environmental difference and genetic damage hypotheses. At that time, experiments were initiated to: (1) incubate embryos from oiled and reference streams in a common environment to evaluate the environmental difference hypothesis (administered by Alaska Department of Fish and Game); (2) verify the field findings that oil affected embryo survival through controlled oiling (administered by National Marine Fisheries Service); and (3) test for genetic damage using flow cytometry and androgenesis screens (administered by Alaska Department of Fish and Game).

Finally, after initiation of these studies, an additional hypothesis was proposed by project reviewers that suggested that the differential mortalities we observed in the field studies were caused by the genetic effects of outbreeding depression. Under this hypothesis, elevated rates of straying of non-locally-adapted adults into oiled streams would result in reduced embryo survival through the introgression of the non-locally-adapted genes. This hypothesis grew in part from inferences drawn from NRDA Fish/Shellfish Study 1 and Study 3 (F/S 1 and F/S 3) which suggested that large numbers of pink salmon were straying into streams in or near our study area. We agreed to investigate this hypothesis by (1) further examining the validity of using extrapolations of coded-wire-tag recoveries to infer rates of straying, and (2) testing for the effects of population mixing through analysis of genotype data.

No difference in embryo to preemergent fry survival between oil-contaminated and reference streams has been observed since the initiation of the study in 1989 (Figure 2; Sharr et al. 1994a, 1994b, and 1994c). We expected embryo to preemergent fry survival to be reduced in oiled streams given that an increase in embryo mortality had been detected. This result can potentially be explained by (1) compensation in the environment, or (2) problems in the experimental design. Geiger et al. (1996) found no evidence to suggest that compensation in the intragravel life stages played a role in determining the number of emerging fry for the years in the study. We believe that the experimental design is inadequate for detecting differences in embryo to preemergent fry survival. The power analysis for the test of differences in survival from embryo to preemergent fry indicated statistical power was adequate to detect a biologically meaningful difference. However, unexpected changes in stream characteristics may have prevented sampling the same areas for embryos in the fall and fry in the spring. Also, stream channels in PWS are not well defined in intertidal areas. Some intertidal stream segments were found to migrate along the beach, especially if the beach is usually exposed to winter storms. The magnitude of these changes was unexpected when this study was designed and initiated.

OBJECTIVES

Recovery Monitoring of Injury to Pink Salmon Embryos in Prince William Sound

1. Estimate the density by tide zone of embryos in 30 streams using numbers of live and dead embryos and fry.

2. Estimate embryo mortality of pink salmon embryos in ten oiled and 15 reference streams.

METHODS

Recovery Monitoring of Injury to Pink Salmon Embryos in Prince William Sound

Study Sites

This project concentrated on populations inhabiting the southwestern portions of PWS; although, streams from Montague Island and eastern PWS were sampled to provide a sound-wide perspective (Figure 2).

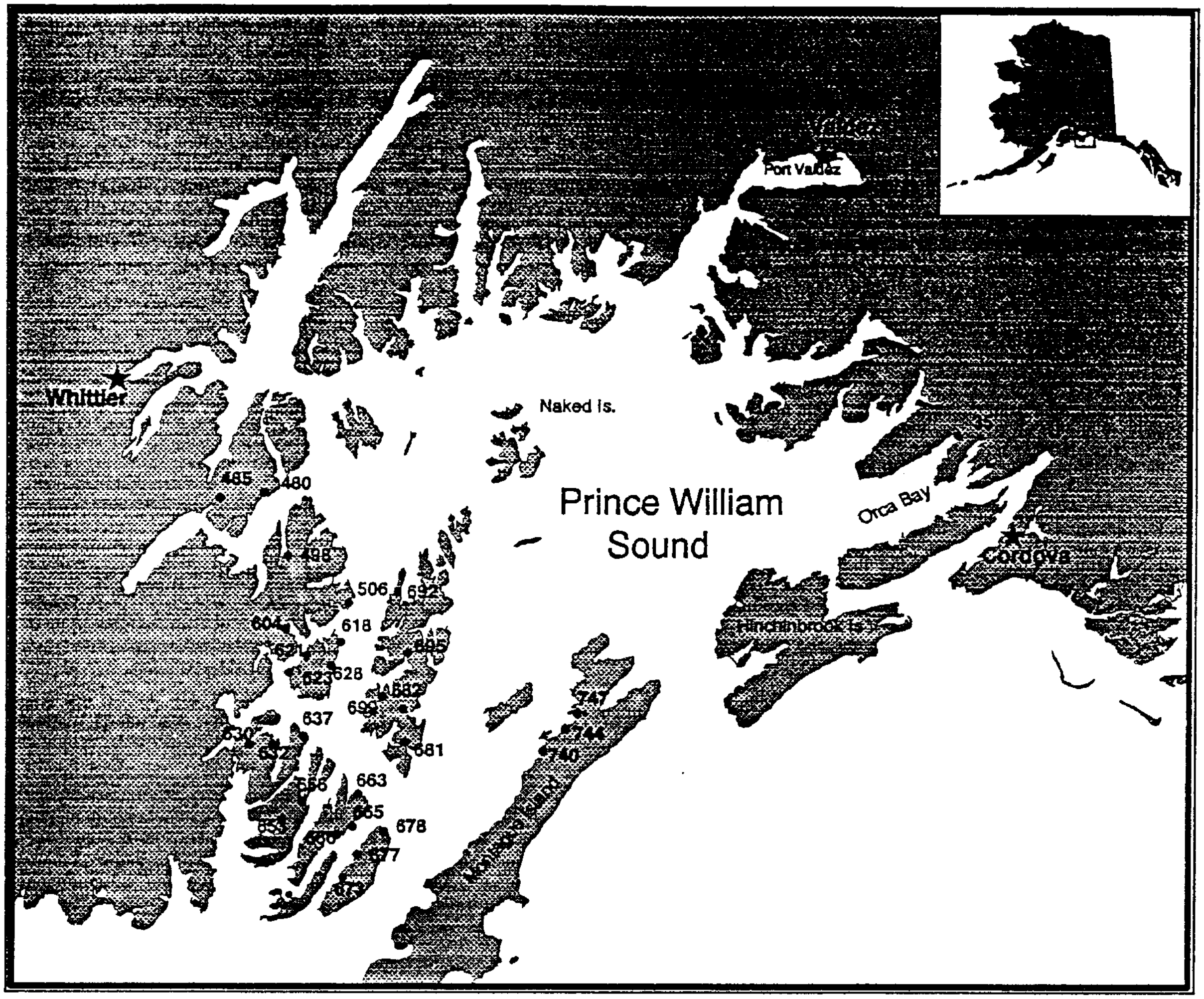

Figure 2. Streams examined during the 1989-1996 pink salmon preemergent fry and egg deposition surveys.

These streams were selected for the following reasons:

1. They have significant spawning populations in both odd and even years.
2. They are accessible for sampling in most years.
3. They are representative of oiled or reference sites in the oil-impacted area.

Sample Design

The methods used for embryo sampling are described by Craig et al. (1995) and Sharr et al. (1994a and 1994b). Sampling was stratified by tide zone to control for possible differences in salinity, temperature, predation, or a combination of these factors. Zone boundaries were established with a surveyor's level and stadia rod and staked prior to sampling. Four zones were sampled in each stream whenever possible: 1.8 - 2.4 m, 2.4 - 3.0 m, 3.0 - 3.7 m above mean low water, and upstream of mean high tide (3.7 m). No sampling was done below the 1.8 - 2.4 m zone because mortality was expected to be high (Helle et al. 1964).

Separate linear transects were established for each zone on the embryo surveys. Although most transects were 30.5 m long, some were shorter due to steep stream gradients. Transects were placed in riffle areas where spawning was observed during escapement surveys conducted for F/S 1. Transects ran diagonally across the river starting downstream against the left bank and moved upstream to the right bank. A map drawn for each stream indicated the tide zones and transect locations in relation to major landmarks. Each embryo transect was photographed and marked with surveyor's flagging to insure that future transects could be located in the same area of the stream.

Fourteen circular areas (0.186 m^2) were systematically sampled along each transect. The number of digs was a compromise between reducing variance and the practicality of conducting the study. Fewer digs were completed on narrow stream channels to avoid excessive sampling of the stream. Streams that split into two or more channels within a zone were sampled either by allocating digs among channels based on spawner distribution observed during F/S 1 or, where spawner distribution was unknown, by an equal allocation.

The following data were collected for each tide zone transect during both embryo and fry sampling:

1. Sample date.
2. Sample tide zone.
3. Start and stop time for the tide zone transect.
4. Numbers of live and dead fry and embryos for each species in each dig.

Pink salmon embryos were separated from chum *O. keta* and coho *O. kisutch* salmon embryos by their smaller size. Chum salmon embryos were separated from coho salmon embryos by their greater development and different coloration. An embryo was considered dead if it was opaque or discolored with coagulated lipids. Fry were considered dead only if decomposition was evident, because sampling often killed fry.

Data Analysis

Numbers of live and dead embryos and fry were summarized by date, stream, level of hydrocarbon impact, and stream zone. The density of live embryos for each stream zone in m^2 (E_{ij}) was estimated by:

$$\hat{E}_{ij} = \frac{\Sigma LE_{ijk}}{0.3n_{ij}} , \tag{1}$$

where LE_{ijk} is the number of live embryos found in the k^{th} dig, in stream i, zone j, and n_{ij} is the number of digs from stream i, zone j. Densities of dead embryos were calculated using the same estimator with appropriate substitutions.

Pink salmon embryo mortality was estimated for each stream using the following relationship:

$$\hat{M}_{ij} = \frac{\Sigma(DE_{eijk} + DF_{eijk})}{\Sigma(LE_{eijk} + DE_{eijk} + LF_{eijk} + DF_{eijk})} , \tag{2}$$

where DE_{eijk}, DF_{eijk}, LE_{eijk}, and LF_{eijk} are the number of dead embryos, dead fry, live embryos, and live fry for the k^{th} dig from stream i, zone j, collected during embryo dig e, respectively.

The Arcsin square root transformation was examined as well as the Logit transform of embryo mortality [ln (odds)], i.e.

$$Logit_{ij} = \ln\left[\frac{\Sigma(DE_{eijk} + DF_{eijk})}{\Sigma(LE_{eijk} + LF_{eijk})}\right] \tag{3}$$

Differences in embryo mortality were examined using a mixed effects two-factor experiment with repeated measures on one factor (Neter et al. 1990):

$$Y_{ijk} = \mu_{...} + O_i + Z_j + (OZ)_{ij} + S_{k(i)} + e_{(ijk)}. \tag{4}$$

The two treatments were level of oiling, (O_i, 2 levels; oiled and reference), and height in the intertidal zone (Z_j, 4 levels; 2.1, 2.7, and 3.4 m above mean low water, and upstream) both fixed effects. The data were blocked by stream ($S_{k(i)}$), a random effect nested within level of oiling. The interaction of level of oiling and height in the intertidal zone was also examined.

Equality of variances was tested using the F_{max}-test (Sokal and Rohlf, 1969), while normality of error terms was visually assessed using normal quantile-quantile and box plots (Chambers et al. 1983). Arcsin square root, logit, log, and square root transforms were examined if the data indicated non-constant variances or non-normal error terms. Tests of homogeneity of-between treatment covariance matrices and the degree of sphericity of the pooled covariance matrix were effected. Four contrasts (oil vs. reference for the four stream zones) and corresponding Bonferroni family confidence intervals ($\alpha = 0.10$ overall) were estimated if a significant difference due to oiling was detected. The SAS (SAS Institute Inc. 1988) General Linear Models Procedure was used to analyze the data.

Stream oiling was assessed through visual observations of the stream and the adjacent area. The observations were supported by photographs, observation maps, and hydrocarbon analysis of mussels *Mytilus sp.* collected near stream mouths. These data were collected as part of another Natural Resource Damage Assessment study (Sharr et al. 1994a).

RESULTS

Recovery Monitoring of Injury to Pink Salmon Embryos in Prince William Sound

Thirty streams were sampled between October 3 and October 22, 1996 for embryos. Mean embryo densities were 985 eggs per m^2 in the intertidal zones and 1090 eggs per m^2 in the upstream zones (Appendix A). Analysis the 1996 embryo mortality data indicated no significant difference between the oil-contaminated and reference streams ($P=0.473$; Figure 3). No significant zone effect ($P=0.352$) or oil-by-zone interaction was found ($P=0.274$). The overall mean embryo mortalities for the oil contaminated and reference streams were 0.254 and 0.189.

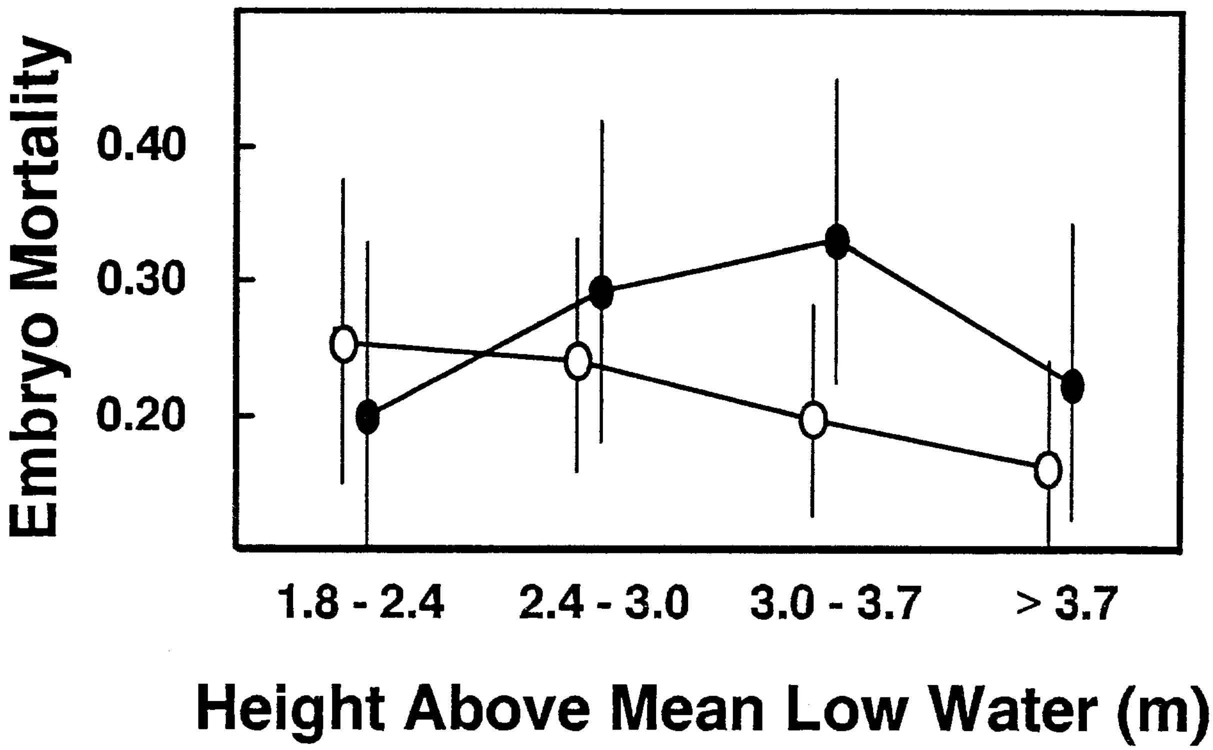

Figure 3. Mean pink salmon embryo mortality and corresponding 90% confidence bounds by tide zone for oil-contaminated and reference streams in Prince William Sound, 1996. Solid circles represent data from ten oil-contaminated streams, and open circles represent data from 15 reference streams.

DISCUSSION

Pink salmon embryos that incubated in oil-contaminated spawning areas in PWS appeared to have been adversely affected by the *Exxon Valdez* oil spill up until 1994 (Sharr et al. 1994a, 1994b and 1994c; Figure 1). However, embryo mortality was not significantly different between oil-contaminated and reference stream in 1994-1995 (Figure 1) and 1996 (Figure 3). We believe that the elevated mortalities observed in 1989 and 1990 were due to direct exposure to oil; elevated mortalities observed in 1991, 1992 and 1993 may have been due to genetic or physiological damage sustained in the parental lines during embryonic development in 1989 and 1990. Lack of a significant difference in 1994, 1995 and 1996 between oiled and reference streams demonstrates a possible recovery of the populations that have been monitored since 1989.

The pink salmon that spawned during the fall of 1991 incubated in oil-contaminated streams during winter of 1989-1990, the first winter after the spill. Likewise, pink salmon that spawned during the fall of 1992 incubated in oiled stream gravel during the fall of 1990 and spring of 1991. Sharr et al. (1994a) found significantly elevated embryo mortalities in oil-contaminated streams during the fall of 1989 and 1990, and the surviving embryos may have sustained sublethal genetic or physiological damages which could have been manifested in the form of functional sterility in 1991, 1992 and 1993. Chromosome damage was observed in other taxa exposed to petrochemicals (Longwell 1977, McBee and Bickham 1988, Hose et al. 1995), although documentation of germline damage is yet to be reported. Likewise, Thomas and Budiantara (1995) showed hydrocabon exposure affected reproductive success by influencing endocrine function and later reproduction in mature fish. In 1994 the returning adults were two generations removed from the 1990 brood and no statistical mortality difference was found in the field or the controlled incubation experiments. Similarly, no statistical difference was found in the field studies of 1995 and 1996. Mortality differences in oiled and reference streams seem to be returning to normal levels, lending credit to a post-spill damage hypothesis with later recovery.

An alternative to the genetic or physiological damage hypothesis is that observed differences in embryo mortality were due to differences in environmental conditions between oil-contaminated and reference streams. This embryo mortality study is based on observational data, and as such, we were unable to randomize stream oiling to account for environmental differences between streams. We attempted to address this concern in our original experimental design by selecting reference streams in close proximity to oil-contaminated streams; however, there is a definite oiling pattern in southwest PWS where streams on points which faced northeastward were heavily oiled. Likewise, streams which faced west and southwest were most likely not oiled.

Environmental differences between oil-contaminated and reference streams does not seem to be a confounding factor because results from the controlled incubation experiment in 1993 and 1994 mirrored the results of the field study (Bue et al. 1996). In 1995, although oiled and reference stream mortalities again look similar (Figure 1), the controlled oiling experiment was incomplete due to few spawners in some study streams. The gametes used in these studies were never in direct contact with a stream; although, the adults which produced them had incubated in the natal streams.

These data do not prove that the observed differences in embryo mortality between oil-contaminated and reference streams were caused by exposure to hydrocarbons. Some have suggested that the streams which were oiled also historically had lower embryo survival. We believe this to be unlikely because mortality between oil-contaminated and reference streams was not significantly different in 1994, 1995 or 1996. The controlled oiling experiment conducted by the National Marine Fisheries Service will provide laboratory evidence to further clarify interpretation of these field data.

Finally, another alternative suggested to explain the differential mortality observed in the field was that elevated straying following the oil spill resulted in outbreeding depression that affected embryo survival in the oil-contaminated streams. The controlled incubation study does not address this hypothesis. Analyzing outbreeding depression and obtaining straying rates of hatchery salmon into all study streams was outside the scope of this project. A proposed project to collect thermal marked otoliths from pink salmon in wild streams should provide further insight into this hypothesis.

CONCLUSIONS

Embryo mortalities were elevated in oil contaminated streams from 1989-1993, but no statistical difference between oil-contaminated and reference streams was observed in 1994 through 1996.

Mortalities in an incubation experiment were elevated for embryos from oiled streams in 1993, but no statistical difference between embryos from oil-contaminated and reference streams was detected in 1994.

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Appendix A. Summary of pink and chum salmon egg dig data from Prince William Sound, 1996.

Appendix A. Summary of pink and chum salmon egg dig data from Prince William Sound, 1996.

					Pink Salmon						Chum Salmon						
					Embryos		Live Embryos/m^2		Fry		Embryos		Live Embryos/m^2		Fry		
Stream #	Stream Name	Date	Height in Tidal Zone(ft)	Loc	Dead	Live	Mean	SE	Dead	Live	Dead	Live	Mean	SE	Dead	Live	No. Samples
35	Koppen Creek	10 4 96	9.0	30	6537	9366	3595.88	709.59	0	0	56	614	235.73	166.19	0	0	14
			11.0	40	13327	10531	4043.15	845.32	0	0	13	15	5.76	4.66	0	0	14
			Upstream	60	13928	6676	2563.11	566.63	0	0	93	273	104.81	63.93	0	0	14
		Total Intertidal			19864	19897	3819.51	543.22	0	0	69	629	120.75	84.52	0	0	28
		Total Upstream			13928	6676	2563.11	566.63	0	0	93	273	104.81	63.93	0	0	14
480	Mink Creek	10 7 96	7.0	20	658	4804	1844.39	438.59	0	74	23	0	.00	.00	0	0	14
			9.0	30	706	3579	1374.08	328.83	0	120	66	43	16.51	16.51	0	83	14
			11.0	40	661	2676	1027.39	275.63	0	51	30	0	.00	.00	0	22	14
			Upstream	60	623	4921	1889.31	443.94	0	5	1	10	3.84	3.84	0	0	14
		Total Intertidal			2025	11059	1415.29	206.21	0	245	119	43	5.50	5.50	0	105	42
		Total Upstream			623	4921	1889.31	443.94	0	5	1	10	3.84	3.84	0	0	14
485	W. Finger Creek	10 6 96	7.0	20	197	3855	1480.04	392.66	0	65	5	34	13.05	12.25	0	0	14
			9.0	30	1044	6198	2379.59	550.83	1	101	71	0	.00	.00	0	40	14
			11.0	40	1152	11220	4307.68	728.62	4	1046	321	162	62.20	59.34	15	206	14
			Upstream	60	580	4331	1662.79	320.82	0	1145	86	41	15.74	11.77	0	0	14
		Total Intertidal			2393	21273	2722.44	372.03	5	1212	397	196	25.08	20.14	15	246	42
		Total Upstream			580	4331	1662.79	320.82	0	1145	86	41	15.74	11.77	0	0	14
498	McClure Creek	10 6 96	7.0	20	434	1975	758.26	242.90	0	0	0	0	.00	.00	0	0	14
			9.0	30	1330	4927	1891.62	483.65	0	0	0	0	.00	.00	0	0	14
			11.0	40	1881	6176	2371.14	589.87	0	146	0	0	.00	.00	0	0	14
			Upstream	60	147	842	323.27	159.60	0	3	0	0	.00	.00	0	0	14
		Total Intertidal			3645	13078	1673.67	280.87	0	146	0	0	.00	.00	0	0	42
		Total Upstream			147	842	323.27	159.60	0	3	0	0	.00	.00	0	0	14

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					Pink Salmon						Chum Salmon						
					Embryos		Live Embryos/m^2		Fry		Embryos		Live Embryos/m^2		Fry		
Stream #	Stream Name	Date	Height in Tidal Zone(ft)	Loc	Dead	Live	Mean	SE	Dead	Live	Dead	Live	Mean	SE	Dead	Live	No. Samples
506	Loomis Creek	10 8 96	7.0	20	948	776	297.93	146.68	0	0	0	0	.00	.00	0	0	14
			9.0	30	3623	2790	1071.16	223.18	0	0	4	0	.00	.00	0	0	14
			11.0	40	6225	2267	870.37	245.94	0	0	0	0	.00	.00	0	0	14
			Upstream	60	2491	4427	1699.65	431.74	0	0	0	0	.00	.00	0	0	14
		Total Intertidal			10796	5833	746.49	128.64	0	0	4	0	.00	.00	0	0	42
		Total Upstream			2491	4427	1699.65	431.74	0	0	0	0	.00	.00	0	0	14
604	Erb Creek	10 12 96	7.0	20	25	584	448.43	356.18	0	2	0	0	.00	.00	0	0	7
			7.0	23	282	882	677.25	647.85	0	0	0	0	.00	.00	0	0	7
			9.0	30	78	1530	587.41	285.89	0	5	0	0	.00	.00	0	0	14
			11.0	40	47	89	34.17	31.71	0	1	0	0	.00	.00	0	0	14
			Upstream	60	15	0	.00	.00	0	0	0	0	.00	.00	0	0	14
		Total Intertidal			432	3085	394.81	154.18	0	8	0	0	.00	.00	0	0	42
		Total Upstream			15	0	.00	.00	0	0	0	0	.00	.00	0	0	14
618	Junction Creek	10 9 96	7.0	20	10	0	.00	.00	0	0	0	0	.00	.00	0	0	12
			9.0	30	58	1581	708.16	264.15	0	0	0	0	.00	.00	0	0	12
			11.0	40	107	1810	810.73	188.44	0	0	0	0	.00	.00	0	0	12
			Upstream	60	118	1301	582.74	403.18	0	0	0	0	.00	.00	0	0	12
		Total Intertidal			175	3391	506.30	121.42	0	0	0	0	.00	.00	0	0	36
		Total Upstream			118	1301	582.74	403.18	0	0	0	0	.00	.00	0	0	12
621	Totemoff Creek	10 12 96	7.0	20	148	723	277.58	156.35	0	1	0	0	.00	.00	0	0	14
			9.0	30	415	1530	587.41	178.94	0	62	0	0	.00	.00	0	0	14
			11.0	40	723	3842	1475.05	295.64	0	209	0	0	.00	.00	0	0	14
			Upstream	60	90	238	91.38	49.77	0	0	0	0	.00	.00	0	0	14
		Total Intertidal			1286	6095	780.01	146.58	0	272	0	0	.00	.00	0	0	42
		Total Upstream			90	238	91.38	49.77	0	0	0	0	.00	.00	0	0	14

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Stream #	Stream Name	Date	Height in Tidal Zone(ft)	Loc	Pink Salmon Embryos Dead	Pink Salmon Embryos Live	Pink Salmon Live Embryos/m² Mean	Pink Salmon Live Embryos/m² SE	Pink Salmon Fry Dead	Pink Salmon Fry Live	Chum Salmon Embryos Dead	Chum Salmon Embryos Live	Chum Salmon Live Embryos/m² Mean	Chum Salmon Live Embryos/m² SE	Chum Salmon Fry Dead	Chum Salmon Fry Live	No. Samples
623	Brizgaloff Creek	10 12 96	7.0	20	183	549	210.78	154.77	0	0	0	0	.00	.00	0	0	14
			9.0	30	111	1736	666.50	337.08	0	0	3	0	.00	.00	0	0	14
			11.0	40	67	459	176.22	155.66	0	0	0	0	.00	.00	0	0	14
			Upstream	60	295	1333	511.78	267.69	0	0	0	0	.00	.00	0	0	14
		Total Intertidal			361	2744	351.17	135.35	0	0	3	0	.00	.00	0	0	42
		Total Upstream			295	1333	511.78	267.69	0	0	0	0	.00	.00	0	0	14
628	Chenega Creek	10 9 96	7.0	20	14	436	167.39	165.74	0	0	0	0	.00	.00	0	0	14
			9.0	30	374	1133	434.99	215.77	0	0	0	0	.00	.00	0	0	14
			11.0	40	2089	2821	1083.06	486.68	0	0	0	0	.00	.00	0	0	14
			Upstream	60	983	5532	2123.89	356.09	0	0	0	0	.00	.00	0	0	14
		Total Intertidal			2477	4390	561.82	190.95	0	0	0	0	.00	.00	0	0	42
		Total Upstream			983	5532	2123.89	356.09	0	0	0	0	.00	.00	0	0	14
630	Bainbridge Creek	10 11 96	7.0	20	867	3812	1463.54	438.96	0	0	0	0	.00	.00	0	0	14
			9.0	30	941	6134	2355.02	493.27	0	203	0	0	.00	.00	0	0	14
			11.0	40	443	7203	2765.44	523.50	10	1349	6	0	.00	.00	0	1	14
			Upstream	60	1550	4471	1716.54	316.20	0	325	2	0	.00	.00	0	0	14
		Total Intertidal			2251	17149	2194.66	286.79	10	1552	6	0	.00	.00	0	1	42
		Total Upstream			1550	4471	1716.54	316.20	0	325	2	0	.00	.00	0	0	14
632	Claw Creek	10 11 96	7.0	20	0	0	.00	.00	0	0	0	0	.00	.00	0	0	14
			9.0	30	10	0	.00	.00	0	0	0	0	.00	.00	0	0	14
			11.0	40	249	1782	684.16	270.01	0	0	0	0	.00	.00	1	0	14
			Upstream	60	3	45	34.55	33.67	0	0	0	0	.00	.00	0	0	7
		Total Intertidal			259	1782	228.05	101.20	0	0	0	0	.00	.00	1	0	42
		Total Upstream			3	45	34.55	33.67	0	0	0	0	.00	.00	0	0	7

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Stream #	Stream Name	Date	Height in Tidal Zone(ft)	Loc	Pink Salmon Embryos Dead	Pink Salmon Embryos Live	Pink Salmon Live Embryos/m² Mean	Pink Salmon Live Embryos/m² SE	Pink Salmon Fry Dead	Pink Salmon Fry Live	Chum Salmon Embryos Dead	Chum Salmon Embryos Live	Chum Salmon Live Embryos/m² Mean	Chum Salmon Live Embryos/m² SE	Chum Salmon Fry Dead	Chum Salmon Fry Live	No. Samples
637	Pt. Countess	10 10 96	7.0	20	1634	1892	726.39	208.18	0	0	0	0	.00	.00	0	0	14
			9.0	30	844	2039	782.83	245.90	0	0	0	0	.00	.00	0	0	14
			11.0	41	803	1385	1063.48	577.20	0	0	0	0	.00	.00	0	0	7
			11.0	42	1072	1306	1002.82	469.78	0	0	0	0	.00	.00	0	0	7
			Upstream	61	2269	82	62.96	57.67	0	0	0	0	.00	.00	0	0	7
			Upstream	62	348	1581	1213.98	445.13	0	0	0	0	.00	.00	0	0	7
		Total Intertidal			4353	6622	847.46	157.86	0	0	0	0	.00	.00	0	0	42
		Total Upstream			2617	1663	638.47	268.27	0	0	0	0	.00	.00	0	0	14
653	Hogg Creek	10 20 96	7.0	20	35	103	39.54	19.97	0	15	1	0	.00	.00	0	0	14
			9.0	31	6	5	3.84	1.93	0	1	0	0	.00	.00	0	0	7
			9.0	32	7	3	2.30	1.60	0	0	1	0	.00	.00	0	0	7
			11.0	40	38	758	291.02	208.67	0	1230	0	0	.00	.00	0	0	14
			Upstream	60	162	1175	451.12	198.92	0	87	0	0	.00	.00	0	0	14
		Total Intertidal			86	869	111.21	71.02	0	1246	2	0	.00	.00	0	0	42
		Total Upstream			162	1175	451.12	198.92	0	87	0	0	.00	.00	0	0	14
656	Halverson Creek	10 21 96	7.0	20	693	904	347.07	180.87	0	0	0	0	.00	.00	0	0	14
			9.0	30	12	470	180.45	112.78	0	0	0	0	.00	.00	0	0	14
			11.0	40	630	3381	1298.06	262.26	0	0	0	0	.00	.00	0	0	14
			Upstream	60	576	5558	2133.88	569.99	0	12	0	0	.00	.00	0	0	14
		Total Intertidal			1335	4755	608.53	134.10	0	0	0	0	.00	.00	0	0	42
		Total Upstream			576	5558	2133.88	569.99	0	12	0	0	.00	.00	0	0	14
663	Shelter Bay	10 18 96	7.0	20	60	857	383.86	170.19	0	0	0	0	.00	.00	0	0	12
			9.0	30	73	1386	620.81	287.64	0	0	0	0	.00	.00	0	0	12
			11.0	40	651	1405	629.32	180.62	0	0	0	0	.00	.00	0	0	12
			Upstream	60	1464	6609	2960.28	847.07	0	0	0	0	.00	.00	0	0	12
		Total Intertidal			784	3648	544.67	124.46	0	0	0	0	.00	.00	0	0	36
		Total Upstream			1464	6609	2960.28	847.07	0	0	0	0	.00	.00	0	0	12

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Stream #	Stream Name	Date	Height in Tidal Zone(ft)	Loc	Pink Salmon Embryos Dead	Pink Salmon Embryos Live	Pink Salmon Live Embryos/m^2 Mean	Pink Salmon Live Embryos/m^2 SE	Pink Salmon Fry Dead	Pink Salmon Fry Live	Chum Salmon Embryos Dead	Chum Salmon Embryos Live	Chum Salmon Live Embryos/m^2 Mean	Chum Salmon Live Embryos/m^2 SE	Chum Salmon Fry Dead	Chum Salmon Fry Live	No. Samples
665	Bjorne Creek	10 18 96	7.0	20	3608	3328	1277.71	333.43	0	0	0	0	.00	.00	0	0	14
			9.0	30	2766	2707	1039.29	319.76	0	0	0	0	.00	.00	0	0	14
			11.0	40	1602	613	235.35	101.81	0	0	0	0	.00	.00	0	0	14
			Upstream	60	836	5231	2008.33	464.82	0	0	0	0	.00	.00	0	0	14
		Total Intertidal			7976	6648	850.79	168.83	0	0	0	0	.00	.00	0	0	42
		Total Upstream			836	5231	2008.33	464.82	0	0	0	0	.00	.00	0	0	14
666	O'Brien Creek	10 19 96	7.0	20	88	5	1.92	1.55	0	0	0	0	.00	.00	0	0	14
			9.0	30	719	1615	620.04	146.25	0	0	0	0	.00	.00	0	0	14
			11.0	40	253	3224	1237.79	423.74	0	0	0	0	.00	.00	0	0	14
			Upstream	60	506	2412	926.04	356.39	0	0	0	0	.00	.00	0	0	14
		Total Intertidal			1060	4844	619.92	165.67	0	0	0	0	.00	.00	0	0	42
		Total Upstream			506	2412	926.04	356.39	0	0	0	0	.00	.00	0	0	14
673	Falls Creek	10 20 96	7.0	20	76	1039	398.90	210.51	0	0	0	0	.00	.00	0	0	14
			9.0	30	13	853	327.49	155.87	0	0	0	0	.00	.00	0	0	14
			11.0	40	41	251	96.37	46.17	0	0	0	0	.00	.00	0	0	14
			Upstream	60	69	536	205.79	102.09	0	0	0	0	.00	.00	0	0	14
		Total Intertidal			130	2143	274.25	88.79	0	0	0	0	.00	.00	0	0	42
		Total Upstream			69	536	205.79	102.09	0	0	0	0	.00	.00	0	0	14

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					Pink Salmon						Chum Salmon						
					Embryos		Live Embryos/m^2		Fry		Embryos		Live Embryos/m^2		Fry		
Stream #	Stream Name	Date	Height in Tidal Zone(ft)	Loc	Dead	Live	Mean	SE	Dead	Live	Dead	Live	Mean	SE	Dead	Live	No. Samples
677	Hayden Creek	10 19 96	7.0	21	41	2080	1597.14	692.07	0	0	0	0	.00	.00	0	0	7
			7.0	22	173	817	627.34	407.11	0	0	0	0	.00	.00	0	0	7
			9.0	31	310	1904	1462.00	398.75	0	0	0	0	.00	.00	0	0	7
			9.0	32	67	1326	1018.18	425.83	0	0	0	0	.00	.00	0	0	7
			11.0	41	338	1499	1151.02	802.78	0	1	0	0	.00	.00	0	0	7
			11.0	42	90	1031	791.66	458.45	0	0	0	0	.00	.00	0	0	7
			Upstream	61	145	911	699.52	231.63	0	0	0	0	.00	.00	0	0	7
			Upstream	62	29	651	499.88	330.48	0	0	0	0	.00	.00	0	0	7
		Total Intertidal			1019	8657	1107.89	218.49	0	1	0	0	.00	.00	0	0	42
		Total Upstream			174	1562	599.70	195.83	0	0	0	0	.00	.00	0	0	14
678	Sleepy Bay	10 19 96	7.0	20	0	4	1.79	.76	0	0	0	0	.00	.00	0	0	12
			9.0	30	2	169	75.70	74.24	0	0	0	0	.00	.00	0	0	12
			11.0	40	168	423	189.47	156.47	0	0	0	0	.00	.00	0	0	12
			Upstream	60	23	163	73.01	48.83	0	0	0	0	.00	.00	0	0	12
		Total Intertidal			170	596	88.99	57.55	0	0	0	0	.00	.00	0	0	36
		Total Upstream			23	163	73.01	48.83	0	0	0	0	.00	.00	0	0	12
681	Hogan Bay	10 17 96	7.0	20	54	2483	953.29	567.89	0	0	0	0	.00	.00	0	0	14
			9.0	30	446	6600	2533.93	649.00	0	7	0	0	.00	.00	0	0	14
			11.0	40	828	6053	2323.92	646.69	0	1	0	0	.00	.00	0	0	14
			Upstream	60	594	4130	1585.63	345.20	0	0	0	0	.00	.00	0	0	14
		Total Intertidal			1328	15136	1937.05	367.13	0	8	0	0	.00	.00	0	0	42
		Total Upstream			594	4130	1585.63	345.20	0	0	0	0	.00	.00	0	0	14

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Stream #	Stream Name	Date	Height in Tidal Zone(ft)	Loc	Pink Salmon Embryos Dead	Pink Salmon Embryos Live	Pink Salmon Live Embryos/m^2 Mean	Pink Salmon Live Embryos/m^2 SE	Pink Salmon Fry Dead	Pink Salmon Fry Live	Chum Salmon Embryos Dead	Chum Salmon Embryos Live	Chum Salmon Live Embryos/m^2 Mean	Chum Salmon Live Embryos/m^2 SE	Chum Salmon Fry Dead	Chum Salmon Fry Live	No. Samples
682	Snug Harbor	10 17 96	7.0	20	1458	4138	1588.70	400.54	0	295	0	0	.00	.00	0	0	14
			9.0	30	1033	5872	2254.43	578.32	14	1450	0	0	.00	.00	0	0	14
			11.0	40	1111	5566	2136.95	257.88	1	603	0	0	.00	.00	0	0	14
			Upstream	60	1463	3947	1515.37	369.78	0	745	0	0	.00	.00	0	0	14
		Total Intertidal			3602	15576	1993.36	247.76	15	2348	0	0	.00	.00	0	0	42
		Total Upstream			1463	3947	1515.37	369.78	0	745	0	0	.00	.00	0	0	14
692	Herring Bay	10 8 96	7.0	20	831	3056	1173.29	431.06	0	0	0	0	.00	.00	0	0	14
			9.0	30	484	4370	1677.77	456.20	0	0	0	0	.00	.00	0	0	14
			11.0	40	520	2681	1029.31	263.56	0	0	0	0	.00	.00	0	0	14
			Upstream	60	340	5160	1981.07	421.57	0	0	0	0	.00	.00	0	0	14
		Total Intertidal			1835	10107	1293.46	225.53	0	0	0	0	.00	.00	0	0	42
		Total Upstream			340	5160	1981.07	421.57	0	0	0	0	.00	.00	0	0	14
695	Port Audrey	10 10 96	7.0	21	12	180	138.21	86.85	0	0	0	0	.00	.00	0	0	7
			7.0	22	49	847	650.38	381.01	0	17	0	0	.00	.00	0	0	7
			9.0	30	566	2548	978.25	327.15	0	19	0	0	.00	.00	0	0	14
			11.0	40	363	1134	435.38	190.41	0	18	3	0	.00	.00	0	0	14
			Upstream	60	114	958	367.80	196.92	0	128	0	0	.00	.00	0	0	14
		Total Intertidal			990	4709	602.64	145.36	0	54	3	0	.00	.00	0	0	42
		Total Upstream			114	958	367.80	196.92	0	128	0	0	.00	.00	0	0	14
699	Cathead Bay	10 9 96	7.0	20	31	100	38.39	38.39	0	0	0	0	.00	.00	0	0	14
			9.0	30	4	0	.00	.00	0	0	0	0	.00	.00	0	0	14
			11.0	40	13	24	9.21	9.21	0	0	0	0	.00	.00	0	0	14
			Upstream	60	173	0	.00	.00	0	0	0	0	.00	.00	0	0	14
		Total Intertidal			48	124	15.87	13.09	0	0	0	0	.00	.00	0	0	42
		Total Upstream			173	0	.00	.00	0	0	0	0	.00	.00	0	0	14

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Stream #	Stream Name	Date	Height in Tidal Zone(ft)	Loc	Pink Salmon Embryos Dead	Pink Salmon Embryos Live	Pink Salmon Live Embryos/m² Mean	Pink Salmon Live Embryos/m² SE	Pink Salmon Fry Dead	Pink Salmon Fry Live	Chum Salmon Embryos Dead	Chum Salmon Embryos Live	Chum Salmon Live Embryos/m² Mean	Chum Salmon Live Embryos/m² SE	Chum Salmon Fry Dead	Chum Salmon Fry Live	No. Samples
740	Kelez Creek	10 21 96	7.0	20	268	1043	400.44	189.83	0	0	0	0	.00	.00	0	0	14
			9.0	30	1003	3395	1303.44	366.31	0	0	0	0	.00	.00	0	0	14
			11.0	40	212	1171	449.58	187.97	0	5	0	0	.00	.00	0	0	14
			Upstream	60	196	481	184.67	160.07	0	0	0	0	.00	.00	0	0	14
		Total Intertidal			1483	5609	717.82	160.99	0	5	0	0	.00	.00	0	0	42
		Total Upstream			196	481	184.67	160.07	0	0	0	0	.00	.00	0	0	14
744	Wilby Creek	10 22 96	7.0	20	412	1266	486.05	176.95	0	0	0	0	.00	.00	0	0	14
			9.0	31	16	303	116.33	95.83	0	0	0	0	.00	.00	0	0	14
			11.0	40	0	0	.00	.00	0	0	0	0	.00	.00	0	0	0
			Upstream	60	0	0	.00	.00	0	0	0	0	.00	.00	0	0	0
		Total Intertidal			428	1569	301.19	104.95	0	0	0	0	.00	.00	0	0	28
		Total Upstream			0	0	.00	.00	0	0	0	0	.00	.00	0	0	0
747	Cabin Creek	10 22 96	7.0	20	239	997	382.78	147.74	0	1	0	0	.00	.00	0	0	14
			9.0	30	350	3704	1422.07	358.37	0	813	0	0	.00	.00	0	0	14
			11.0	40	1703	3250	1247.77	312.82	0	17	0	0	.00	.00	0	0	14
			Upstream	60	821	1001	384.31	97.41	0	35	0	0	.00	.00	0	0	14
		Total Intertidal			2292	7951	1017.54	176.81	0	831	0	0	.00	.00	0	0	42
		Total Upstream			821	1001	384.31	97.41	0	35	0	0	.00	.00	0	0	14

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Stream #	Stream Name	Date	Height in Tidal Zone(ft)	Loc	Pink Salmon Embryos Dead	Pink Salmon Embryos Live	Pink Salmon Live Embryos/m² Mean	Pink Salmon Live Embryos/m² SE	Pink Salmon Fry Dead	Pink Salmon Fry Live	Chum Salmon Embryos Dead	Chum Salmon Embryos Live	Chum Salmon Live Embryos/m² Mean	Chum Salmon Live Embryos/m² SE	Chum Salmon Fry Dead	Chum Salmon Fry Live	No. Samples
861	Bernard Creek	10 3 96	7.0	20	3003	1909	732.92	237.73	0	0	0	0	.00	.00	0	0	14
			9.0	30	3544	3285	1261.21	279.33	0	0	0	0	.00	.00	0	0	14
			11.0	40	3416	7835	3008.08	784.90	0	0	0	0	.00	.00	0	0	14
			Upstream	60	1101	4978	1911.20	364.81	0	0	0	0	.00	.00	0	0	14
		Total Intertidal			9963	13029	1667.40	319.98	0	0	0	0	.00	.00	0	0	42
		Total Upstream			1101	4978	1911.20	364.81	0	0	0	0	.00	.00	0	0	14
Prince William Sound Summary																	
		Total Intertidal			84846	222368	984.54	44.26	30	7928	603	868	3.84	75.95	16	352	1214
		Total Upstream			32052	79681	1089.78	75.95	0	2485	182	324	4.43	93.80	0	0	393

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