Petroleum Hydrocarbons in Alaskan Invertebrate Subsistence Foods Following the 1989 Exxon Valdez Oil Spill


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The Exxon Valdez ran aground on Bligh Reef, Prince William Sound, Alaska on 24 March, 1989, spilling millions of gallons of Prudhoe Bay crude oil (PBCO). During the weeks following the spill, large amounts of oil flowed toward southwestern Prince William Sound, and as a result, many shorelines were oiled. The spreading of spilled oil raised many concerns, and among them was the concern that subsistence seafoods (e.g., fish and invertebrates) were contaminated (see abstracts by J. Field and J. Stein et al.) by the spilled petroleum.

The objectives of this paper are to describe the extent and duration of contamination of invertebrates by the spilled oil. The data from this study together with those described by Stein et al. (in this session) were used by the Alaskan Oil Spill Health Task Force and the U.S. Food and Drug Administration to address the issue of potential risk to native Alaskans of eating seafood from their traditional collection sites.

Crude petroleum contains many hundreds of individual chemicals which are generally divided into classes such as aliphatic hydrocarbons, aromatic hydrocarbons and other constituents containing sulfur, oxygen and nitrogen (Clark and Brown, 1977). Among these compounds are dibenzothiophene and alkyl-substituted dibenzothiophenes, which are significant components of PBCO and can serve as useful markers in discerning PBCO. Among the constituents of petroleum, the aromatic hydrocarbons are considered the most toxic. The lower molecular weight aromatic compounds (LACs), those that contain three or fewer benzene rings, have been found to be the more acutely toxic, whereas the higher molecular weight aromatic compounds (four to six rings, HACs) are generally considered the more chronically toxic components.

A majority (99%) of the ACs in fresh PBCO measured in this study consisted of LACs. However, as the oil weathered, the concentrations of many of the more volatile and water soluble LACs decreased rapidly. Sixteen non-substituted ACs and 17 groups of alkyl-substituted ACs comprising some 270 individual chemicals present in PBCO were determined in subsistence seafood samples to assess the extent of contamination, to evaluate temporal changes, and to compare to possible sources.

More than a thousand samples of fish, marine mammals, and invertebrates were collected between 1989 and 1991 and were analyzed for ACs by gas chromatography/mass spectrometry according to Krahn et al. (1988). These samples included a variety of invertebrates from 80 sampling stations used by the residents of 17 villages for collecting subsistence seafoods. The focus of this paper will be on the four target invertebrate taxa; mussels (Mytilus edulis), butter clams...
(Saxidomus giganteus), littleneck clams (Protothaca staminea), and chitons (order Neoloricata).

For discussion purposes, invertebrate samples (wet weight basis) are divided into four categories on the basis of the sum of concentrations of the ACs:
1. Not contaminated - <10 ng/g;
2. Minimally contaminated - from 10 ng/g to 100 ng/g;
3. Moderately contaminated - from 100 ng/g to 1000 ng/g;
4. Heavily contaminated - >1000 ng/g.

Invertebrates from most of the 17 villages and the two reference areas, Angoon and Yakutat, fell into the first two categories: not contaminated or minimally contaminated. Invertebrates from some stations at two sampling areas, Chenega Bay and Windy Bay, and from two stations near Kodiak Island were moderately or heavily contaminated with ACs.

Following the spill, oil was observed in Chenega Bay in the vicinity of station CHE1 (the beach below the village of Chenega Bay — used by villagers as a harvesting area). A tar mat about 1 m wide extended the length of the beach at the high tide line at CHE10 (the southern end of Erlington Island). Invertebrates from eight of eleven stations at Chenega Bay were not contaminated or were minimally contaminated. Invertebrates from three stations at Chenega Bay from one or more sampling events were moderately or heavily contaminated. Specifically, the mussels collected from CHE1 (1989) and CHE7 (at Port Ashton-1990) were moderately contaminated and those from CHE10 (1990) were heavily contaminated.

However, mussel samples collected from CHE1 in 1990 and 1991, and mussels collected from CHE7 and CHE10 in 1991 were minimally contaminated. Butter clams collected from CHE7 (1990 and 1991) and littleneck clams collected in 1990 were moderately contaminated. Butter clams from CHE1 (1989, 1990, and 1991) and from CHE10 (1990) were minimally contaminated as were littleneck clams from CHE1 (1990 and 1991) and from CHE10 (1990, 1991). Butter clams from CHE10 (1991) and chitons from several stations were not contaminated.

Based on mean concentrations of ACs, invertebrate samples from two of the five stations sampled at Windy Bay (stations WNB4 and WNB5) were not contaminated or were minimally contaminated. Two stations, WNB1 and WNB3, located on two adjacent small islands in the mouth of Windy Bay, were both observed to be moderately to heavily oiled.

The degree of contamination of invertebrates from these two stations varied with sampling year and by species. Specifically, mussels from Windy Bay station WNB1 (1989) and from WNB3 (1990) were heavily contaminated, whereas the concentrations of ACs in mussels from these stations in 1991 were much lower (minimally to moderately contaminated).

Mussels were not collected at WNB1 in 1990 or at WNB3 in 1989. Only one sample was collected at WNB2, littleneck clams sampled in 1989, which were almost in the heavily contaminated category, with 960 ng/g ACs.

Invertebrate samples from two sampling stations on Kodiak Island (KOD3 and OHA4) also contained elevated concentrations of ACs. Station KOD3 was located near the city of Kodiak and adjacent to the boat harbor. An oily sheen was observed on one occasion at KOD3 by the field party while digging for clams. Mussels, butter clams, littleneck clams, and chitons from station KOD3 from both
1989 and 1990 were moderately or heavily contaminated.

Station OHA4 at the village of Old Harbor was also near a boat harbor. Oil was not observed to be present during sample collection at this site. Butter clams and littleneck clams collected at Old Harbor station OHA4 in 1989 and mussels collected in 1990 were just within the moderately contaminated category. Five of the six mussel samples and three of six butter clams (collected in 1989) were minimally contaminated.

The relative amounts of the ACs can be useful in evaluating temporal changes and to compare to potential sources. The relative amounts of the parent and alkyl-substituted ACs are useful for comparing patterns of these ACs among petroleums, petroleum products, related materials, and ACs in invertebrate samples. Dibenzothiophene and alkyl-substituted dibenzothiophenes (which comprise 27 discernible components in PBCO) were particularly useful in making comparisons.

For example, these chemicals were mostly absent in a sample of Cook Inlet crude oil but were much more prominent in a sample of Kuwaiti crude oil than PBCO. Graphs of these patterns from various oils and from certain moderately and highly contaminated invertebrate samples were compared and the same data were treated using principal components analysis and hierarchical cluster analysis. The relative amounts of these analytes in oil collected nine days after the Exxon Valdez spill were similar to those in the fresh oil except for a loss of the more volatile and water soluble components, particularly, naphthalene, C1- and C2-naphthalenes.

The pattern of ACs in a sample of oil collected from the beach at Snug Harbor 15 months after the spill was similar to that of the nine-day weathered oil. The patterns of ACs in mussels from Windy Bay station WNB3 collected in 1989 were similar to the oil sample from Snug Harbor except for additional losses of the more volatile and water soluble components of LAC. The temporal changes of the patterns of AC in mussels collected from Windy Bay from 1989 to 1991 showed increasing losses of the more volatile and water soluble components of LAC. Evaluation of the patterns of ACs from mussel samples from CHE7 implies the presence of PBCO plus ACs indicative of combustion products.

ACs present in samples from Kodiak Island stations OHA4 and KOD3 were indicative of petroleum or petroleum products and the pattern of ACs was very similar to that of the nine-day weathered PBCO. The pattern of the ACs in a mussel sample from KOD3 was essentially the same as in butter clams from the same site collected in March 1990, April 1990, and September 1990, and in a littleneck clam sample collected April 1990 at KOD3. There was little evidence of temporal change in the pattern of ACs in these clam samples from KOD3 which could imply a continual exposure to the same source of ACs. It is possible that there is a local and continual source of hydrocarbons in the area of these two stations that would have a pattern of ACs similar to the somewhat weathered PBCO.

In summary, invertebrate samples from most stations, including those from the two reference areas, were not contaminated or were only minimally contaminated with ACs typical of petroleum. Invertebrates from a few stations in the Chenega Bay sampling area and Windy Bay were moderately or heavily
contaminated with ACs. Invertebrates from one station (KOD3) on Kodiak Island were moderately to heavily contaminated with ACs and the concentrations did not appear to decrease significantly from 1989 to 1990 (samples were not collected in 1991) suggesting the presence of other possible sources of ACs not related to the Exxon Valdez spill. Mean concentrations of ACs in mussels from the more contaminated sites at Chenega Bay and Windy Bay generally decreased with time. However, because of sample variability, it is difficult to draw conclusions about temporal trends.

References
Investigations of Crude Oil Contamination in Intertidal Archaeological Sites Around the Gulf of Alaska
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The State of Alaska initiated a brief study during 1991 aimed at investigating the presence of crude oil in archaeological sites which occur in the intertidal zone. The concern was that cultural deposits subjected to oiling retained crude oil contaminants that would affect radiocarbon dates obtained from the sites. Designed to complement a larger, area-wide study funded through the U.S. Forest Service, the State study concentrated on 13 intertidal sites suspected to contain intact cultural deposits. Site selection was based on documented presence of cultural remains in the intertidal zone and evidence of beach oiling during the Exxon Valdez oil spill. Preliminary field examinations eliminated 10 sites from further examination because they lacked intact cultural deposits. One site above the high tide line was added to test presence of wind or storm wave-borne oil contaminants.

Laboratory studies about the effect of incorporation of crude oil on radiocarbon dating with datable samples suggested that significant skewing of dates occurred (Mifflin and Associates, 1991). The goals of the project therefore were to test the selected sites for presence of oil and to concurrently test the reliability of radiocarbon dates from the deposits. Comparison of time diagnostic artifacts from the tested sites with similar artifacts from well dated nearby sites was the method chosen to check validity of the oiled site dates. Sites were partially excavated to obtain large enough collections and adequate radiocarbon samples to accomplish the comparison.

Sites were tested to reveal stratigraphy of deposits in the middle to upper intertidal zone and sediment samples collected to test for presence of subsurface oil. Selected sediment samples were submitted to the National Oceanic and Atmospheric Administration/National Marine Fisheries Service laboratory in Seattle for analysis by high performance liquid chromatography with ultraviolet detection (HPLC/UV). One sample from the AFG-098 Site on Shuyak Island and a sample from the SEL-215 Site on Nuka Island contained minute traces of petroleum hydrocarbons.

The AFG-098 Site on Shuyak Island contains cultural deposits which yielded artifact collections typical of Koniag Phase from the region and provides the best opportunity to test the hypothesis that contamination affected radiocarbon dating. Two cultural levels are clearly separate in the stratigraphic profile of the site. The Lower Component is an early Koniag Phase collection containing stemmed, ground slate, projectile points with barbs. Cross-section of the points is generally a flattened biconvex form although some points have a medial ridge on one face. Another form of point, triangular in outline with a flat basal facet, was found in the lower level of the site. Ground slate ulus from the component have straight, bifacially ground cut-
ting edges. The collection contains planing adzes of the variety usually inset into a bone socket and then hafted. Other diagnostic artifacts in the Lower Component include a slate fragment with an etched face, a stone saw, and chipped slate preforms for subsequent grinding into projectile points.

Similar, early Koniag Phase collections at well dated sites in the region have assigned ages of between A.D. 1350 and A.D. 1500. Age estimates for the Koniag Phase in the Kodiak Archipelago cite a beginning around A.D. 1100 to A.D. 1200. Age of the incised slate figurines or faces is particularly well defined during the A.D. 1350 to A.D. 1500 period. Triangular ground slate points with a flat ground basal facet typically date from A.D. 1300 to A.D. 1500. Four radiocarbon samples stratigraphically associated with the Lower Component range from A.D. 787 ±110 to A.D. 1143 ±65. The four dates are acceptably within the earliest estimated range of the Koniag Phase and may indicate that the developmental stage of the phase occurred earliest in the northern Kodiak area.

The AFG-098 Upper Component collection provided an artifact collection which is clearly related to Koniag Phase materials elsewhere. Ground slate projectile points of several forms were recovered. Stemmed points, some with barbs and some with rounded shoulders, were the most common forms recovered. Medial ridges were present on some barbed forms but a flattened biconvex cross-section was most common among the stemmed points. A very distinctive ground slate projectile point with a triangular outline and a sharply defined flute or butt facet was found in the Upper Component. Several ground slate ulu forms occur in the component. Both are bifacially ground and have straight to slightly convex cutting edges. One form has a distinctive notch or offset at the back of the blade which suggests hafting at the back edge near one end with the other end extending out of the handle. Other stone artifacts from the Upper Component include a stone saw, sawn slate fragments, whetstones, planing adzes, a quartz crystal, a hematite nodule for firestarting, and chipped slate preforms. Fragments of several bone or ivory dart heads with unilateral barbs, a barbed hook fragment, and bone awls are some of the organic artifacts recovered. A small jet labret was also found. Fragile artifacts carved from spruce wood, bark and grass matting, and fragments of a birch bark container were recovered from the saturated deposits. More than a dozen species of seeds were found in floor deposits of the Upper Component.

Comparison of the Upper Component collection with Koniag Phase or related site collections on Kodiak Island, both sides of the Alaska Peninsula, and the outer coast of the Kenai Peninsula demonstrate close similarities. The triangular point form with a basal flute consistently occurs in Koniag Phase or related collections dating from between A.D. 1500 and Historic times (A.D. 1750-1800). Other point and ulu forms are consistent with later Koniag Phase ages as well. Six radiocarbon dates obtained from the Upper Component levels of AFG-098 range from A.D. 1343 ±60 to A.D. 1490 ±125.

The AFG-082 Site is an upland site eroding slowly into the intertidal zone which was tested for contamination from storm wave or wind deposited oil. The ground slate ulus, chipped bifaces, planing adze, and small single chamber
houses compare reasonably well with middle age range Kachemak Tradition remains, approximately 2,000 years old. The ratio of ground slate to chipped stone remains also supports a Kachemak Tradition comparison. Unfortunately, the AFG-082 collection is too small and typologically limited to provide a very accurate age estimate.

Two radiocarbon samples obtained from the site date to A.D. 203 ±65 and A.D. 288 ±65. The AFG-082 dates fall well within the expected age range determined from artifact typology. No evidence of oil was found in the site and no sediment samples were submitted for analysis.

The SEL-215 Site on Nuka Island contains intact cultural remains within a peat deposit in the intertidal zone. Time specific traits in the collection are meager but grooved splitting adzes suggest an age between A.D. 1000 and A.D. 1500. Inclusion of a glass trade bead in the collection is interpreted as an intrusive historic element.

Radiocarbon samples from the site yielded seven dates ranging from A.D. 1142 ±60 to A.D. 1442 ±105. A trace of oil was detected in one of the two sediment samples submitted for HPLV/UV screening. However, the radiocarbon determinations compare well with the expected age of the deposits.

The SEW-068 Site consists of a peaty intertidal deposit containing cultural remains which relate to Kachemak Tradition collections elsewhere. Grooved splitting adzes located nearby may not be associated however such tools have been dated to that early time from other sites in Prince William Sound. A general estimate of age for the peaty deposits, based on artifact typology, is 1000-2000 years ago. Geological age estimates based on rates of isostatic rebound from the 1964 Earthquake and from long term regional subsidence indicate the deposits should be at least 1500 years old. Two wood samples from the saturated deposits provided radiocarbon dates of A.D. 10 ±65 and A.D. 391 ±65. The ages obtained from the culturally modified wood fragments agree roughly with the expected age of the cultural deposits.

Conclusions drawn from this study include:

(1) Intertidal archaeological deposits at three of the sites investigated demonstrate that useful and important information is preserved in some intertidal sites even though there is sometimes no surface evidence of buried remains.

(2) Traces of petroleum hydrocarbons in subsurface remains do occur although the origin of the contaminants in the sites tested is unknown.

(3) Reliable radiocarbon dates can be obtained from oiled deposits. It is uncertain, however, whether that results in the tested sites from cleaning of the samples or simply lack of actual contamination. Examination of samples in the radiocarbon laboratory for oil contaminants should be routine and cleaning methods should be modified if necessary to remove identified contaminants.

(4) Dating of intertidal or even exposed upland archaeological remains needs to involve every possible approach to dating, not just reliance on a single method. Archaeologists investigating chronological questions in the area of the Exxon Valdez oil spill need to be especially critical with their conclusions. No evidence was found in the sites examined that the Exxon Valdez
oil spill adversely affected the radiocarbon dating results. Damage to sites appears to be from erosion or vandalism rather than direct oiling.

References
Generating Damage Restoration Costs for Archaeological Injuries of the *Exxon Valdez* Oil Spill
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This paper summarizes the results of a monetary assessment of damage for injuries to archaeological sites documented in the *Exxon Valdez* oil spill response records. Injuries attributable to the oil spill at 35 archaeological sites in Prince William Sound and the Gulf of Alaska were analyzed to estimate restoration costs for use by the *Exxon Valdez* Trustee Council in planning restoration of damages for archaeological resources. The damage assessment was accomplished in two steps.

First, a damage assessment panel, chaired by the author, met to consider restoration costs based on the archaeological injury data. Second, working from the findings of the panel, additional analyses were conducted by the author to calculate specific restoration costs for the archaeological injuries. Two levels of restoration costs were produced by the damage assessment process. First, site-specific restoration costs were developed for the archaeological sites identified as having substantive injuries. Second, gross restoration costs were estimated for projected numbers of sites injured by the oil spill.

The procedures employed by the damage assessment panel were carried out in nine steps:

1. The data on the 35 archaeological sites with injuries attributable to the oil spill and other relevant data sources were reviewed.
2. A conceptual framework on which restoration costs would be based was developed from the damage assessment model contained in the Archaeological Resources Protection Act of 1979, as amended (ARPA).
3. Documented oil spill injuries to archaeological sites were analyzed and grouped into two major categories, those resulting from oiling and those resulting from oil spill response activities.
4. Two restoration options were developed, one for ten years of oil effect monitoring and one for direct physical restoration. These were used to formulate specific restoration measures appropriate for the two categories of injuries.
5. Ten injured sites were eliminated from further consideration because appropriate site restoration work had already been accomplished or the site damage was not severe enough to require restoration. Damage at one site was determined to be unrestorable.
6. Three categories of site specific restoration proposals were developed for the remaining 24 sites: sites recommended for monitoring only, sites recommended for direct physical restoration only, and sites with both types of measures recommended.
7. Standard levels of effort were formulated for direct physical restoration in year one and in years two through ten for oil effect monitoring. Appropriate site-specific salary estimates...
were generated using standard levels of effort as a guide.

8. Site-specific support costs necessary to carry out direct physical restoration measures and oil effect monitoring were estimated.

9. Restoration costs were estimated for four different injury scenarios, each with a different number of injured sites, using average per site costs for direct physical restoration measures and oil-effect monitoring.

Two principal sets of findings were produced. The first consists of the proposals for site restoration measures and costs for the 24 archaeological sites with substantive injuries. The second consists of restoration costs for projected numbers of sites injured by the oil spill.

Site-specific restoration proposals are based on the oil effect monitoring and direct physical restoration measures. Using the above criteria, there are five sites in the oil effect monitoring only category, 14 sites in the direct physical restoration only category, and five sites at which both types of measures are recommended.

The first element of the restoration cost proposals for the 24 sites are personnel salaries. The salary figures for 17 of the 24 sites are based on the three standard levels of effort defined above. Due to special circumstances, seven sites have salary figures based on variations of the standard computations.

Five sites have cost proposals involving only salaries for ten years of monitoring. The standard salary figure for year one oil effect monitoring is $2,904.51, and $2,202.17 per year for years two through ten or $19,819.53 for 9 years. Therefore, the total salary figure for each of these sites is $22,724.04.

Salary for direct physical restoration only is proposed for 11 sites. The total salary figure for each of these sites is $3,155.79.

Only one site is in the oil effect monitoring and direct physical restoration salary category. The total salary figure for this site consists of the standard salaries for ten years oil-effect monitoring, plus costs for direct physical restoration—$25,879.83.

Four sites have salary figures for direct physical restoration measures above the standard level of effort, as well as oil effect monitoring. One has disinterred human remains which required the addition of eight days of project supervisor time for consultation with Native Corporations. The result is a salary increase of $1,804.24 over the standard salary for direct physical restoration. The total salary figure for the site consists of the amount for the combined standard salaries shown in the preceding paragraph and the extra cost for consultation, $25,879.83 + $1,804.24, or $27,684.07. At the other three sites, a test excavation is proposed to fully assess the magnitude of damage. This required the addition of two person days for fieldwork and one person day each for analysis and report preparation. The result is a salary increase of $527.76 over the standard salary for direct physical restoration. The total salary figure for these sites consists of the amount for the combined standard salaries and the extra cost for the test excavation, $25,879.83 + $527.76, or $26,407.59.

One other site is proposed for direct physical restoration salary above the standard level, but not for oil effect monitoring. Because this site has disinterred human remains, eight days of project supervisor time were added for consultation with Native Corporations at the
cost of $1,804.24. Also, the large volume of disturbance at this site required the addition of four person days for fieldwork and two person days each for analysis and report preparation. The result is a salary increase of $1,055.52 over the standard salary for direct physical restoration. The total salary figure for this site consists of the standard salary for direct physical restoration and the extra costs for consultation and restoration measures, $3,155.79 + $1,804.24 + $1,055.52, or $6,015.55.

Finally, two sites in the direct physical restoration only category have salary figures below the standard level because their injuries require measures different from those proposed for most other sites. For one site, the total salary figure of $2,480.83 is for repatriation or reinterment administration and consultation. The direct physical restoration measures proposed for the othersite allowed the elimination of two person days for field work and one person day each for analysis and report preparation. The result is a salary decrease of $892.08 from the standard salary for direct physical restoration to a total salary figure of $2,263.71.

The other elements of the restoration cost proposals are support costs. Basic support costs are for: fieldwork per diem, transportation, supplies and equipment, and processing and duplication. Recovery of items requiring expenditures for curation is anticipated at all but two of the sites proposed for direct physical restoration measures. The proposals for three sites involve repatriation or reinterment costs. (At one site, the only support costs are for repatriation or reinterment.)

The support cost amounts vary by site. The average support cost figures are: for year one oil effect monitoring, $4,086.83; for years two through ten oil effect monitoring, $33,159.60; and for direct physical restoration, $10,920.33.

The total figures for the restoration cost proposals for the 24 sites under consideration are as follows:

- Year one oil effect monitoring: $69,913.40
- Years two through ten oil effect monitoring: $529,791.30
- Ten years of oil effect monitoring: $599,704.70
- Direct physical restoration measures: $272,126.49
- Total restoration costs: $871,831.19

Average costs per site for oil effect monitoring and direct physical restoration were calculated from the total cost figures by dividing them by the number of sites for which the measures are proposed. The average costs are $59,970.47 per site for oil effect monitoring and $14,322.45 per site for direct physical restoration.

The projections for the numbers of sites injured on which the estimates of gross restoration costs are based were derived from four sets of figures:

1. The projected numbers of injured sites in the draft State University of New York report entitled Exxon Valdez Oil Spill Archaeological Damage Assessment by Albert J. Dekin et al. (1992).
2. The Dekin et al. figures reduced by the percentages corresponding to the number of sites eliminated from consideration by the damage assessment panel because the injuries were not severe enough to require restoration.
3. The number of sites proposed by the panel for oil effect monitoring and...
direct physical restoration as percentages of the total number of sites now included in the Alaska Heritage Resource Survey site inventory records for the oil spill area.

4. The number of sites proposed by the panel for oil effect monitoring and direct physical restoration as percentages of the number of sites actually located and examined by Exxon crews during oil spill response activities.

Each of these four figures was multiplied by the average per site costs for oil effect monitoring and direct physical restoration. The resulting gross restoration costs estimates for oil effect monitoring are:

   (1)  $31,844,319.57  
   (2)  $28,126,150.43  
   (3)  $1,439,291.28  
   (4)  $3,598,228.20

For direct physical restoration measures, they are:

   (1)  $4,840,988.10  
   (2)  $3,867,061.50  
   (3)  $658,832.70  
   (4)  $1,618,436.85

The total gross restoration cost estimates are:

   (1)  $36,685,307.67  
   (2)  $31,993,211.93  
   (3)  $2,098,123.98  
   (4)  $5,216,665.05

The gross cost estimates based on the number of sites actually field checked by Exxon (number four above) are seen as reliable indicators of the overall magnitude of archaeological restoration needs resulting from the oil spill.

Two important conclusions are drawn from the results of the work summarized in this paper. First, the ARPA damage assessment model was used successfully to generate credible restoration cost determinations for the documented archaeological injuries of the Exxon Valdez Oil Spill. Second, the ARPA model should be the damage assessment and restoration cost determination standard for archaeological injuries resulting from future oil spills or other similar situations.

References
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Long-Term Social Psychological Impacts of the Exxon Valdez Oil Spill

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During the last decade scientific studies have been conducted on the community impacts of technological disasters (Baum et al., 1982, 1983; Omohundro, 1982; Couch and Kroll-Smith, 1985; Gill and Picou, 1989). Most recently, a number of studies provide empirical evidence which demonstrates that, in contrast to natural disasters, technological disasters produce long-term patterns of stress. These patterns of stress appear to be related to issues of “uncertainty” of extent of contamination (Vyner, 1988), protracted litigation activities (Brown and Mikkelsen, 1989; Picou and Rosebrook, 1992), and general sociocultural disruption (Freudenberg and Jones, 1991).

Social impact assessments of the Exxon Valdez oil spill have been relatively limited. Studies suggest that subsistence activities of Alaskan natives were initially disrupted by the spill (Fall, 1990; Restoration Planning Work Group, 1990; Dyer et al., 1992). Furthermore, research also reveals the existence of social impacts some 18 months following the spill (Picou et al., 1992). The nature of these impacts included relatively high levels of family, work and personal disruption, as well as continuing patterns of personal stress (Picou et al., 1992).

Given this recent interest in identifying long-term social impacts of technological disasters and the paucity of longitudinal data on this subject, this study will present and evaluate a causal model which depicts long-term social psychological impacts.

A disaster impact assessment design structured the methodological procedures of this research (Gill and Picou, 1991; Picou and Rosebrook, 1992). This approach included standardized indicators of social impacts, random sampling procedures, and a control community comparison. The survey instrument included demographic, social and psychological indicators used in previous disaster research. This research design used stratified random sampling techniques and included personal, telephone and mail surveys over the three year data collection period.

Impact and Control Community Description

Cordova was selected as an impacted community because of its economic dependency on commercial fishing and its cultural heritage of subsistence activities. It is located in the southeastern region of Prince William Sound. The community is isolated from other communities by mountains, glaciers, rivers and the sea. No roads have connected Cordova to the outside world since 1964. Incorporated in 1909, the city became an export center for copper mined in the Wrangell mountains to the north. The closing of the copper mines in 1939 led to increasing involvement in commercial fishing. The community population currently fluctuates from 2,500 during the winter to over 4,500 during the summer commercial fishing season.

Cordova fishermen own almost one-half (44%) of all Prince William Sound
herring permits and 55% of all Prince William Sound salmon fishery permits (Stratton, 1989). Subsistence activities (i.e., harvesting, giving, receiving fish, moose, deer, berries, etc.) characterize 90% of Cordova’s households (Stratton, 1989) and Alaskan natives make up 18 percent of the population. These and other data classify Cordova as a natural resource community (Dyer et al., 1992). The commercial fishing industry and numerous related businesses link this community directly to seasonal harvests of renewable natural resources.

Petersburg, Alaska was selected as the control community for this study. Petersburg is located on an island in the southeastern part of the state and is relatively isolated with no road connections outside Mitkof Island. Petersburg has a population of 3,500 people, with Alaskan natives comprising approximately 20%. Petersburg’s economic base is based on commercial fishing. Petersburg residents also engage in subsistence activities at a rate similar to Cordova (Smythe, 1988; Stratton, 1989).

Data Collection: A stratified random household sample was selected in the Cordova community. In August, 1989, personal interviews with 86 respondents were conducted, reflecting 70 households. Random digit dialing telephone interviews were conducted in Petersburg and Cordova to complete data collection activities for the first year. In 1990, follow-up interviews were conducted by mail and telephone surveys. In 1991, respondents in Cordova were reinterviewed by personal interviews and respondents in Petersburg were once again contacted by telephone.

Indicators and Measures: Information was collected from respondents on demographics, social attitudes, work, family and personal disruption and social psychological stress. Social psychological stress was measured by the “impact of events scale,” which identifies two stress components—intrusion and avoidance (Horowitz et al., 1979; Horowitz, 1986).

Statistical Analysis: A series of path models, or structural equation models, were calculated for data available from 1989 to 1991 (Duncan, 1966; Birnbaum, 1981). In general, these models attempt to identify causal relationships between social structural characteristics, social disruption and psychological stress.

Results

Higher levels of intrusive stress and avoidance behavior were observed for the impacted community in 1989, 1990 and 1991. These differences were found to be statistically significant (Pr < .05) when t-tests were applied to compare impact and control community mean scores on the stress indicators. Mean stress scores were found to decline from 1989 to 1991 in the impact community suggesting that, over time, a reduction in the intensity of the social psychological impacts.

The evaluation of the models provided limited support for the hypothesis that social structural characteristics influence social and psychological reactions of victims of technological disasters. The primary predictors of intrusive stress in 1991 included intrusive stress in 1990 and 1989, work disruption in 1989 and continuing social disruption in 1991. Attitudes toward the effectiveness of cleanup operations were found to predict long-term stress. That is, respondents who were most pessimistic about cleanup effectiveness in 1990 tended to be more stressed in 1991. In general, respondents who were male and who
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experienced both work and family disruption in 1989 held the most pessimistic views of cleanup effectiveness.

The long-term patterns of social psychological stress found in previous studies of a variety of technological disasters were also observed for residents of the impact community in this research. Higher levels of intrusive stress and avoidance behavior were found to exist in 1989, 1990 and 1991 in the impact community. Over time, levels of stress were found to be declining. For example, mean intrusive stress scores fell from 24.47 in 1989 to 19.32 in 1990 and then further declined to 17.74 in 1991. This general trend suggests a pattern of return to community equilibrium.

Attempts to develop and evaluate causal models of long-term stress resulting from the Exxon Valdez oil spill were modestly successful. Although some intervening variables were found to predict long-term stress, initial stress levels were the most important predictors of later stress levels in the models. Intrusive stress existing some 20 months after the spill was found to be related to perceptions that the cleanup was ineffective. However, the relative effects of both attitudes toward the cleanup and problems experienced from litigation on intrusive stress were small when compared to effects of previous stress levels generated from the spill.

In conclusion, alternatives to linear, additive models may be required to fully understand the complex patterns of long-term stress created by technological disasters in general and the Exxon Valdez oil spill, in particular. Models utilizing interaction terms may provide more accurate explanations. Future analyses of these data will evaluate the utility of this approach.

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The Economic Impacts of the Exxon Valdez Oil Spill on Southcentral Alaska’s Commercial Fishing Industry
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The potential of natural disasters to generate short-term economic benefits for impacted individuals and communities has become an accepted social science notion (Dacy and Kunreuther, 1969; Rogers, 1970; Cochrane, 1975; Wright et al. 1979; Friesema, et al. 1979; Rossi et al. 1983). Unfortunately, the economic dimensions of human-made disasters have not received similar treatment despite the considerable attention that has recently been focused on these events and their sequelae (e.g., Erickson, 1976, 1991; Levine, 1982; Couch and Kroll-Smith, 1985; Shrivastava, 1987).

In the case of oil spills, economic research has been largely confined to the estimation of comprehensive damage assessments (Mead and Sorensen, 1970; Burrows et al. 1974a, 1974b; Brown et al. 1983; Grigalunas, 1982; Grigalunas et al. 1986; Assaf et al. 1986). With the exception of work completed by Nelson (1981) and Restrepo (1982) there has been little consideration of the perturbing influences that technological disruptions can have on local and regional economies.

Within the context of recent technological accidents (e.g., Bhopal, Three Mile Island, Love Canal), the Exxon Valdez oil spill is distinguished by its widespread physical damage of a highly valued natural environment and its extraordinary economic bonanza (Cohen, 1993). In order to contain the spilled cargo, collect contaminated debris, and clean oiled shorelines a massive emergency response operation was assembled that eventu-ally employed 11,000 local residents and transient laborers at wages exceeding $16 per hour.

However, while these ephemeral windfall profit opportunities were being exploited, the fundamental component of the regional economy was experiencing a downward realignment. In an effort to measure the magnitude of this adjustment, this analysis derives ex post estimates of the oil spill’s economic impacts on southcentral Alaska’s commercial fishing industry during the years 1989 and 1990 in isolation of the considerable financial benefits imparted by the emergency response operation. A three-phase methodology is employed to determine the ex-vessel revenue that would have been earned for each of southcentral Alaska’s eleven major commercial fishery products (chinook, sockeye, coho, pink and chum salmon, king crab, tanner crab, Dungeness crab, Pacific herring sac roe, Pacific halibut, and sablefish) during each of these two years had the oil spill not occurred.

First, estimates of the accident’s harvest volume impacts are constructed using data reported by the Alaska Department of Fish and Game (ADF&G), International Pacific Halibut Commission (IPHC), and the National Marine Fisheries Service (NMFS). The pre-season harvest expectations for each commercial fishery in southcentral Alaska (Prince William Sound, Lower Cook Inlet, and Kodiak Island) were compared to actual yields. In order to ascertain the extent of
contamination from the oil spill, physical monitoring and organoleptic test results were examined for each regulatory jurisdiction that evidenced a harvest deficit in either of the two years under consideration. Harvest volume impacts for each commercial fishery were then apportioned to the accident according to these data. This method led to the conclusion that the oil spill's harvest volume impacts were confined principally to Pacific herring and pink and chum salmon during the 1989 season. No harvest volume reductions attributable to the accident were experienced in 1990.

Second, the Exxon Valdez oil spill was hypothesized to have motivated a fundamental shift in ex-vessel demand for southcentral Alaska's commercial fishery products as retail consumers became fearful of tainted seafood. This effect was estimated with a price-dependent demand model that was variously adapted to the specific characteristics of each of the region's major fish and shellfish species. Independent variables included in these specifications were the seasonal quantity of landed product, national income of major consuming countries, frozen and canned inventory holdings, price of substitutes, and exchange rates between the United States and major consuming countries.

The structural equations derived from this model were estimated by a biased least squares fitting procedure with data for the period 1964-1988. The subsequently derived coefficients were then used to generate ex post forecasts of the ex-vessel prices that would have prevailed for southcentral Alaska's commercial fishery products during 1989 and 1990 in the absence of the oil spill. The predicted ex-vessel prices were then contrasted with their corresponding actual values. On the basis of this technique, the largest ex-vessel price impacts were sustained in both years by Pacific herring sac roe and coho and chum salmon. Noteworthy is the observation that actual ex-vessel prices for Pacific halibut and sablefish exceeded predicted values during both seasons, raising the possibility that some demand substitution of these products least threatened by oil contamination may have occurred.

Finally, the full extent of these estimated harvest volume and ex-vessel price impacts cannot be attributed to the Exxon Valdez oil spill as the commercial fishing industry was concurrently perturbed by several other biological and economic influences in addition to the accident. On the biological side, commercial fishery yields are generally subject to considerable stochastic variability due to numerous environmental factors. For instance, fluctuations in rates of predation, disease mortality, and water temperature can alter interseasonal commercial fishery harvest volumes. Additionally, various forms of human intervention, including regulatory measures and artificial cultivation, can result in harvest volume adjustments.

On the economic side, ex-vessel prices in 1989 and 1990 were influenced by several perturbations that occurred simultaneously to the oil spill. After attaining unprecedented levels in 1988, ex-vessel prices for most Alaskan commercial fishery products began to erode in 1989. This trend was motivated by substantial increases in the volume of salmon produced internationally. Other factors contributing to this decline included excessive wholesale inventories, reduced consumer spending, unfavorable exchange rate adjustments, and suspended
speculation among Tokyo fishery product wholesalers. In order to isolate the ex-vessel revenue impacts of the Exxon Valdez oil spill from these confounding biological and economic factors several alternative scenarios were simulated.

Though each scenario was based on different assumptions regarding the magnitude of the Exxon Valdez oil spill relative to the array of confounding factors, a consistent conclusion emerges. The economic boom that swept over southcentral Alaska as a result of the oil spill obscured the decline in the profitability of the region’s commercial fishing industry and exacerbated the deterioration of international market conditions. Specifically, the accident reduced ex-vessel revenue for southcentral Alaska’s commercial fishers during 1989 by an amount ranging between $6.4 million and $41.8 million. Ex-vessel revenue reductions were greatest for the region’s commercial harvest of sockeye and pink salmon while increased ex-vessel values for Pacific halibut and sablefish marginally mitigated these declines. This analysis indicates that the oil spill’s ex-vessel revenue impacts in 1990 were between $11.1 million and $44.5 million.

Employing 1988 as a baseline, these amounts represent 3-19 percent of the ex-vessel value of southcentral Alaska’s commercial fishing economy. Given the considerable imprecision inherent in economic impact analysis of complex perturbations such as the Exxon Valdez oil spill, more explicit evaluation is not readily possible. In spite of this indeterminacy, this analysis provides a bounded interval in which one measure of the accident’s economic dimensions can be considered.

The interpretation of these results requires the consideration of at least four factors. First, the harvest volume impact methodology lacks scientific rigor and is further undermined by the uncertainty that exists regarding hydrocarbon-based toxicity on individual marine species. Even under relatively static circumstances the determination of fishery harvest volumes on the basis of historical trends and biometric forecasting models is an imprecise exercise.

Second, the ex-vessel price model provides only a partial explanation of demand behavior and its empirical specification relies on commercial fishery product data not generally noted for a high degree of reliability.

Third, changes in the market characteristics of many of southcentral Alaska’s fishery products over the course of the specification period raises concerns as to parameter stability in the ex-vessel price model.

Finally, many of southcentral Alaska’s commercial fishers were employed in various capacities by the cleanup operation mobilized after the oil spill. Remuneration for boat rentals and services exceeded by several orders of magnitude the ex-vessel revenue lost due to the inability to conduct scheduled commercial fisheries, and overall economic performance must be viewed in this light.

References:


Detecting Population Impacts from Oil Spills: A Comparison of Methodologies.
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The simplest response of a population to an oil spill is an immediate kill. A certain proportion of the population is killed by the spill, and then the population gradually recovers. A second possibility is that there is continued post-spill impact on the population due to reduced reproduction, growth or some other mortality, but this impact disappears over time. A third possibility is that there is some permanent impact on the population, so that it never would recover to its pre-spill abundance. This could be due to permanently lost habitat, change in the community composition, or permanent changes in survival rates.

There are four ways that the impacts on a population can be detected. These are (1) direct body counts of the number of animals killed, (2) pre vs. post spill comparison of population sizes, (3) oiled vs. unoiled spatial comparisons of abundance after a spill, and (4) direct measurement of vital rates in oiled vs. unoiled sites.

(1) Body counts are the number of individuals killed by the spill and can sometimes be directly measured, or estimated. Such counts are available for several bird and marine mammal species. However, body counts do not provide, by themselves, any evidence of population level impact. The body counts must be considered in relation to population abundance, natality and mortality rates. Thus any quantitative assessment of population level impacts will require some form of population dynamics model, combined with several types of data.

(2) When abundance surveys are available, comparison of pre and post spill numbers may be made to assess the change in population. The statistical power of such comparison will depend upon the reliability of the census method, the natural variability of the population, and the magnitude of change induced by the spill. This method can clearly not be used when no pre-spill abundance data are available, as was the case in many fish species. Pink salmon exhibit such high year-to-year variability that only the most severe of impacts could have been detected by this method. We show that the good returns of pink salmon to Prince William Sound in 1990 and 1991 do not provide strong evidence that there was no significant impact of the oil spill.

(3) When the populations can be spatially stratified into oiled and unoiled sites, it may be possible to assess the impact of the spill even when pre and post spill data are not available. The abundance of individuals in oiled sites is compared to the abundance in unoiled sites. Again the power of this method will depend on the reliability of the census method, the natural variability from site to site, and the magnitude of the change induced by the spill. A key problem posed by this approach is the fact that treatments (oiled vs. unoiled) were not
assigned randomly, and post spill differences may reflect underlying habitat differences rather than the impacts of oiling. The study of impacts on Dolly Varden and cutthroat trout illustrates how such oiled vs. unoiled comparisons can work (Hepler et al., 1993). This is contrasted to pink salmon studies where oiled vs. unoiled abundance data provide little statistical power (Sharr et al., 1993).

(4) A final approach is to measure life history parameters in oiled and unoiled sites. The estimated parameters are used in a life history model to estimate population level impacts. The differences in growth observed in oiled vs. unoiled sites (Hepler et al., 1993), and in egg survival for pink salmon (Bue and Sharr, 1993) are examples of how this approach can provide evidence of damage even where population level damages may be difficult to measure directly.

The weakness of this approach is it depends on the validity of the population dynamics models used, and in most cases the extent of damage depends on the level of compensatory mortality in the life history after the damage. If there is strong density dependent mortality then the population level impacts will be much less than the raw mortality caused by oiling. The potential for compensatory mortality significantly decreases the power of this approach.

A final issue we consider is the statistical framework for analysis of damage studies. An obvious question encountered in any study of oil spill impacts is what level of significance should be used. We argue that traditional hypothesis testing statistics should not be used, but rather the statistical product of an analysis should be a Bayesian posterior distribution on the intensity of oil spill impact. Hypothesis tests have little if any meaning in testing for impacts, and should be abandoned.

References


Coastal Habitat Studies: The Effect of the Exxon Valdez Oil Spill on Shallow Subtidal Fishes in Prince William Sound.

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Fish communities were monitored in shallow (<20 m) depths of paired oiled and unoiled (control) study sites within Prince William Sound in 1990, using diving transect techniques. Habitats studied were defined by dominant macrophytes (macroscopic plant), including eelgrass and Agarum/Laminaria.

There were four pairs of oiled and unoiled (control) eelgrass study sites. At each site, we established three 30 m long transects running parallel to shore, in approximately the middle of the depth range of eelgrass and at randomly selected locations along a 200 m section of shoreline selected for sampling. Two divers counted fishes along each transect within 1 m on either side and within 3 m of the bottom.

Agarum/Laminaria habitats were subdivided into bay and point categories, and further stratified into shallow (2-11 m) and deep (11-20 m) depth zones. There were three pairs of bay and point oiled and control sites. In each depth zone three 30 m long transects were randomly located and run parallel to the shoreline. Fish were counted as in the eelgrass habitat.

The null hypotheses of no difference among oiled and control sites was tested with a blocked ANOVA, with oil category as the main effect and pair as a blocking factor. Homogeneity of variance was tested with Levene's test (Milliken and Johnson, 1984). If data failed to meet the assumption of homogeneity, and if the significance level for the ANOVA was less than 0.10, we tested the significance of the ANOVA F statistic with a randomization procedure (Manly, 1991).

Over 15 species of fishes were observed in the eelgrass transects, although numbers were dominated by young of the year (YOY) Pacific cod (Gadus macrocephalus), which were the most abundant species in both oiled (0.84 of all fishes) and control sites (0.46 of all fishes). Other important taxa were gunnels (Polliidae), greenlings (Hexagrammidae), and Arctic shanny (Stichaeus punctatus). The number of all fishes per transect (60 m2) ranged from 5.3 to 105.0, with highest numbers in the oiled sites in each of the four site pairs. The abundance of all fishes, and of YOY Pacific cod, were significantly (p<0.01) higher at oiled sites.

Over 30 species of fishes were observed in Agarum/Laminaria habitats, and the community was dominated by Arctic shanny and a mixed group of sculpins. For the purposes of our study we divided the sculpins into small and large species categories. In Agarum/Laminaria bays, Arctic shanny, most of which were YOY juveniles, were the most abundant species in shallow depths (0.63 of all fish in oiled sites and 0.31 in control sites) and were significantly more abundant at oiled sites. Total fish abundance was also higher at oiled sites as a result of the high numbers of Arctic shanny. In the deep depths of bays, small sculpins (mainly Ariedius sp.) were the most abundant fishes. There were no differences in fish
abundances in the deep strata of bays.

At _Agarum/Laminaria_ study sites on points, small sculpins were the most abundant fishes in shallow depths and were significantly more abundant at oiled sites. Greenlings were more abundant at control sites (p < 0.05). In the deep strata of the point habitats greenlings were significantly more abundant at oiled sites, as was the abundance of all fishes combined (p = 0.03).

In both eelgrass and _Agarum/Laminaria_ habitats, oiled sites had overall higher abundances of fishes than did control sites due to the higher numbers of the dominant YOY fishes. Ebeling et al. (1972) also found greater numbers of larval and young of the year fishes in oiled sites relative to control sites in the Santa Barbara Channel after a large oil spill there in 1969. Higher abundances of YOY fishes may have resulted from greater settlement of pelagic larvae into oiled areas, from lowered mortality of YOY juveniles after settlement, or from post-settlement migration of juveniles into oiled sites or away from control sites. Our data do not provide a basis for distinguishing among these hypotheses. However, other components of the overall subtidal program measured the abundances of many invertebrates at the same locations where fishes were counted. Consequently, we conducted a feeding study of YOY Pacific cod from eelgrass study sites to determine if prey use patterns were influenced by availability of invertebrate prey taxa.

We examined the feeding habits and condition (based on Fulton's length/weight index) of YOY Pacific cod from oiled and control eelgrass sites. Fish from oiled sites tended to have higher volumes of stomach contents, and had eaten proportionally more mollusk larvae than fish from control sites, where crustaceans were more common in diets. These differences in diet appear related to the relative availability of prey in oiled versus control eelgrass study sites. Fulton's condition index did not differ in any consistent pattern between oiled and control study sites. Our results suggest that in eelgrass habitat the increased abundance of YOY Pacific cod at oiled study sites is related to the increased diversity and abundance of epifaunal suspension-feeding prey taxa, especially mussels.

**References**


Survey of Oil Exposure and Effects in Subtidal Fish Following the Exxon Valdez Oil Spill: 1989-1991
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Petroleum and its components have the potential to damage fishery resources. Of special concern in Prince William Sound and areas in the Gulf of Alaska affected by the Exxon Valdez oil spill, are species such as Dolly Varden char and adult salmon, which inhabit or pass through the littoral zones, and benthic fish species which live in subtidal areas in close association with bottom sediments. Numerous laboratory studies have demonstrated that exposure of fish to petroleum hydrocarbons can result in a variety of adverse effects.

However, because of the high rate of metabolism of petroleum hydrocarbons by many species of fish, direct measurement of tissue concentrations of parent compounds does not generally provide a useful indicator of exposure of fish to petroleum hydrocarbons. Therefore, it has been difficult to document exposure of fish to petroleum following oil spills.

In recent years, however, methods have been developed for determining such exposure, based on our knowledge that many of the metabolites of aromatic petroleum hydrocarbons are fluorescent, and a primary route of excretion of these metabolites is through the bile. Thus we have demonstrated that the measurement of fluorescent aromatic compounds (FACs) in fish bile serves as a useful indicator of petroleum exposure in field-sampled animals (Krahn et al., 1992; Krahn et al., this volume). Such measurements are now a mainstay of many assessment and monitoring programs, including those following the Exxon Valdez oil spill. Additionally, it is known that certain forms of cytochrome P450 can increase dramatically following exposure to a variety of exogenous compounds. Of most interest for aquatic monitoring programs is the finding that an increase in hepatic cytochrome P4501A (P4501A) appears to be a useful indicator of exposure of fish species to a wide variety of organic contaminants, including many compounds contained in petroleum (Payne et al., 1986; Collier and Varanasi, 1991).

In this paper we present summaries of our assessments of exposure of several species of fish for three years after the Exxon Valdez oil spill, using these two methods (biliary FACs and hepatic P4501A). Data are presented for Dolly Varden char (Salvelinus malma), yellowfin sole (Limanda aspera), rock sole (Lepidopsetta bilineata), flathead sole (Hippoglossoides elassodon), Pacific halibut (Hippoglossus stenolepis), and pollock (Theragra chalcogramma).

Our studies of exposure of adult Pacific salmon species are presented by Stein et al. in this volume. In addition to assessing petroleum exposure, samples were also taken for examination of histological alterations and assessment of reproductive function in two species, Dolly Varden and yellowfin sole, because of the potential for petroleum and related compounds to adversely affect tissue
structure and reproduction. The data reported here are in units of ng phenanthrene equivalents/mg bile protein, or ng/mg, for levels of FACs in bile, and pmol benzo[a]pyrene metabolized/mg microsomal protein•minute, or pmol/mg, for P4501A-associated enzyme activity. Methodology used for obtaining these results can be found in the detailed study plans prepared for the Natural Resources Damage Assessment Trustee Council. More detailed presentations and analyses of the data are being prepared for publication in peer-reviewed scientific journals.

Summaries of findings by species

Dolly Varden char, sampled in the littoral zone by either beach seines or gill nets run perpendicular to the shore, showed some of the highest levels of exposure to oil (as measured by levels of FACs in bile, up to mean values of ~15,000 ng/mg) of any fish sampled in 1989. However, by 1990, these levels had dropped markedly (to <5,000 ng/mg) at heavily oiled sites such as Tonsina Bay and Snug Harbor. Hepatic P4501A activities were also elevated (>200 pmol/mg/min) in Dolly Varden at these oiled sites in 1989, and had dropped by 1990 (<100 pmol/mg/min). These results suggest that the heavy oiling of the intertidal area seen in 1989 affected fish in the very nearshore subtidal area, such as Dolly Varden char, and by 1990 the levels of petroleum hydrocarbon contamination of these areas were substantially reduced. In 1990, Dolly Varden tissues were also analyzed histologically, but no increases in prevalences of histopathological conditions of liver, kidney, gonad, or gill were seen in conjunction with apparent oil exposure. Additionally in 1990, measurements of plasma estradiol concentrations and ovarian maturation in female Dolly Varden captured at 12 sites showed little evidence of reproductive impairment in this species, although plasma estradiol levels tended to be lower in the animals most heavily exposed to petroleum. Because there was a substantial decrease in petroleum exposure between 1989 and 1990, Dolly Varden were not sampled in 1991.

Exposure to petroleum was readily discernible in the benthic flatfish species, yellowfin sole, though the levels of FACs in bile (up to ~10,000 ng/mg) were less than for Dolly Varden, suggesting less exposure to oil. However, levels did not drop markedly between 1989 and 1990 at oiled sites. By 1991, however, substantially decreased exposure was evident at Snug Harbor. In both 1990 and 1991 there was evidence of increased P450 activities in yellowfin sole at oiled sites. Similar to Dolly Varden, there was little evidence of reproductive dysfunction in the female yellowfin sole from the oiled sites in 1990, although there was again a trend toward lower levels of plasma estradiol in the most heavily exposed fish.

Levels of biliary FACs and hepatic P4501A activities were determined in rock sole from several sites in 1989, 1990, and 1991, and rock sole were also examined for the presence of histopathological lesions in 1990. Similar to yellowfin sole, there was evidence of exposure to petroleum in rock sole sampled near oiled sites in 1989 and 1990, again up to ~10,000 ng/mg. Decreased exposure was observed at oiled sites in the limited sampling done in 1991. The results of histological analyses of tissues collected from this benthic species in 1990 showed no alterations in liver, kidney, or gonad histology. However, there was a significantly (p<0.005) increased prevalence of respiratory epithelial hyperplasia (REH)
of the gill at three sites where the biliary FAC data suggested that oil exposure was greatest (Tonsina Bay, Snug Harbor, and Sleepy Bay) as compared to the prevalence in fish collected from Olsen Bay and Rocky Bay. Additionally, the severity of gill REH was significantly greater (p<0.05) at the three more impacted sites.

Increased levels of biliary FACs and induced hepatic P4501A activities were measured in flathead sole from heavily oiled sites in 1989 (FACs only) and 1990 (FACs and P4501A). In 1991 these same measures were slightly elevated in sole from Snug Harbor compared to a less impacted site, Olsen Bay. These results are similar to observations in yellowfin sole and rock sole, which is consistent with all three of these flatfish species being captured from similar habitats. No histological analyses of tissues from flathead sole have been done.

Pacific halibut, captured generally at depths of >30 m, showed some evidence of increased oil exposure in 1989, as determined by levels of FACs in bile, but levels were substantially less (<7000 ng/mg) than for other flatfish species captured at shallower depths. By 1990 these levels of FACs had dropped considerably (to <2500 ng/mg) at Tonsina Bay and Snug Harbor.

Pollock were not sampled until the late winter of 1990, and then were only sampled for levels of biliary FACs. At that time increased levels of FACs (up to ~8000 ng/mg) were evident, especially in pollock from inside Prince William Sound, and by 1991 these levels had dropped quite substantially, though it was still possible to detect increased levels at some sites inside FWS, compared to animals collected from unimpacted or distant sites. Assessment of reproductive function in female pollock collected in 1991 did not show any substantial effects that could be positively ascribed to increased oil exposure.

The results of analyses of oil exposure in several species of subtidal fish following the Exxon Valdez oil spill definitively point to the necessity of monitoring the subtidal environment following major oil spills. The littoral zone appeared to be heavily impacted in the months immediately following the spill, but a year later, after a winter storm cycle, exposure of fish inhabiting this zone (i.e. Dolly Varden) was substantially decreased. In contrast, nearshore benthic fish species (up to ~30 m depth, species sampled were rock sole, yellowfin sole, and flathead sole) showed continuing exposure through the first two field seasons after the spill, and even after more than two years there was still some evidence of increased exposure of fish from these habitats.

Beyond this depth (>30m), the degree of exposure of Pacific halibut appeared to be less than in benthic fish residing at shallower depths. A surprising finding was the evidence of exposure of pollock to petroleum approximately one year after the spill, at a site (e.g. Tugidak Island) more than 400 miles from the grounding site. Pollock, a major fisheries resource in Alaskan waters, are bathypelagic fish which feed in the water column. Thus these results suggest that the spilled oil affected either the water column or food supply of these fish at great distances from the spill, and for some time after the spill.

What remains to be determined are the potential impacts on fishery resources of long-term exposure to petroleum, albeit at moderate to low levels. To date, our studies have not shown any profound effects in the species studied following
the *Exxon Valdez* oil spill, but this finding is tempered by the delay in initiating studies of serious effects such as reproductive function, and the relatively short time over which such analyses have been conducted.

Although it will always be difficult to sample subtidal fishery resources comprehensively and rapidly following an oil spill, a better understanding of the potential impacts of petroleum exposure on fishery resources can be obtained through careful and realistic laboratory exposure of fish to petroleum. Such studies will need to go beyond the relatively straightforward short-term exposure studies which have been commonly done in the past. However, recent advances in methodology for assessing oil exposure in fish, together with current knowledge of the biological processes involved in reproduction, immune function, and growth and survival of fish species, make this an appropriate course of action.

**References**


Impact of Oil Spilled from the Exxon Valdez on Survival and Growth of Dolly Varden and Cutthroat Trout in Prince William Sound, Alaska

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From 1989 through 1991, the impact of spilled crude oil from the Exxon Valdez on growth and survival of anadromous Dolly Varden (Salvelinus malma) and cutthroat trout (Oncorhynchus clarki) in Prince William Sound were studied in accordance with the Clean Water Act and the Comprehensive Environmental Response, Compensation, and Liability Act. At the time of the spill, anadromous Dolly Varden and cutthroat trout were in lakes around Prince William Sound. Past studies on the behavior of these fish have shown that Dolly Varden and cutthroat trout spend their winters in lakes, emigrate to the sea during late spring, and return to the same lakes the following fall. Survival and growth rates in emigrating populations could be calculated from fish recaptured during the next spring emigration.

Five such populations of each species were intercepted with weirs in 1989, 1990, and 1991 during their annual seaward emigration to Prince William Sound in the spring. Two populations emigrated into the wake of the spill, while three emigrated into waters free of spilled crude oil. Our study populations were comprised of tagged adults and subadults of both species. Growth was measured directly on recaptured fish. Survival rates were estimated with log-linear models of capture histories of tagged fish.

A two-stage simulation based on bootstrapping and Monte Carlo techniques was used to compare average survival rates between study populations that were and were not associated with spilled oil.

Ten thousand eight hundred fifty seven Dolly Varden and 1,086 cutthroat trout were tagged and released in 1989; and 41,510 and 2,496, respectively, in 1990; 46,286 Dolly Varden and 2,701 cutthroat trout were inspected for tags in 1990; and 28,657 Dolly Varden and 5,062 cutthroat trout were inspected in 1991. Fewer fish were intercepted in 1989 because weirs were installed after the larger, older fish had emigrated.

Three hundred thirty-six fish were recaptured and reported by persons working or recreating around Prince William Sound during the summers of 1989 and 1990; few of these fish (18 Dolly Varden and no cutthroat trout) had been caught across the boundaries of the spill. Ninety to 100% of recaptured emigrants were recaptured at the same weirs in 1990 and 1991 at which they were released a year earlier. Of those fish that had strayed (298), few Dolly Varden and no cutthroat trout had moved across boundaries marking the extent of spilled crude oil. On average an unexpected 47% of survivors of both species evaded recapture in 1990 even though three of the five weirs were fish-tight that year. This evasion was incorporated into the log-linear models used to estimate survival rates.

Growth and survival rates were significantly lower in study populations associated with spilled oil. Growth from 1989-1990 was on average less in study
populations that emigrated into the wake of the spill: 24% and 22% less for recaptured subadult and adult Dolly Varden and 36% and 43% less for subadult and adult cutthroat trout.

This difference persisted through 1991 for cutthroat trout, but not for Dolly Varden; growth during 1990-91 of recaptured Dolly Varden in populations not associated with oil slowed. Averages of estimated survival rates from 1989 to 1990 were less in study populations associated with spilled oil: 36% and 40% less for subadult and adult Dolly Varden and 28% less for adult cutthroat trout. Bioaccumulation of petrogenic hydrocarbons in the food chain or chronic starvation from its collapse were hypothesized as the pathways that spilled crude oil had slowed growth and accelerated mortality of Dolly Varden and cutthroat trout. Other studies in Prince William Sound in the wake of the spill have shown a collapse in some of the populations of inter-tidal and subtidal invertebrates that are common prey for both Dolly Varden and cutthroat trout.

Our results are consistent with the occurrence of a deleterious impact on growth and survival of those Dolly Varden and cutthroat trout emigrating into the wake of the oil spilled from the Exxon Valdez. Because our results come from observation and not from experiment, our study can not confirm that impact. No information on growth and survival rates of fish in these populations was available prior to our study, nor could we control which populations were exposed to spilled oil.

However, preponderance of evidence, and not confirmation, is the usual rule of judgment for assessing impacts in accordance with the Clean Water Act and with the Comprehensive Environmental Response, Compensation, and Liability Act. Survival and growth rates in those study populations of Dolly Varden and cutthroat trout not associated with spilled oil in Prince William Sound were typical of published rates for these species elsewhere, while rates for fish associated with spilled oil were atypically lower.
Assessment of Damage to Demersal Rockfish in Prince William Sound Following the Exxon Valdez Oil Spill

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Populations of demersal rockfish in Prince William Sound were studied from 1989 through 1991 to assess potential injury due to the Exxon Valdez oil spill. Injury was evaluated by determining the concentrations of hydrocarbons and histopathological alterations in rockfish that inhabit reefs located in oiled and unoiled sites.

Oil spilled from the Exxon Valdez was the probable cause of death for demersal rockfish killed in Prince William Sound immediately after the spill. Approximately 20 dead rockfish were brought to the collection centers in Valdez and Cordova from sites of reported fish kills. Five of these rockfish were necropsied and it was concluded that crude oil was the probable cause of death of all five.

These results prompted additional testing for hydrocarbons in living rockfish. The 1989 hydrocarbon analyses showed that at least 11 fish of 30 rockfish tested from treatment sites had been exposed to oil within the two weeks prior to collection, while none of the 13 fish in control sites were exposed to oil. These two pieces of information prompted the 1990 and 1991 studies to sample and test rockfish for continued exposure to hydrocarbons.

Tissues were collected from rockfish for analysis at four sites (two oiled and two control) in Prince William Sound in both 1990 and 1991. In 1990, four sites were sampled along the outer Kenai Peninsula. Tissues from several species of demersal rockfish: yelloweye (Sebastes ruberrimus), quillback (S. maliger), and copper (S. caurinus), were collected for histopathological evaluation. Rockfish tissues, as well as stomach contents, unconsolidated benthic sediments, and sessile suspension feeders were collected at each study site for analysis of hydrocarbons.

The proportion of samples from oiled sites with aromatic hydrocarbons and their metabolites in the bile was compared to the proportion of samples from control sites with contaminated bile. Evidence of exposure to hydrocarbons was indicated by elevated concentrations of phenanthrene and naphthalene-equivalent compounds in the bile in concert with chromatographic patterns characteristic of hydrocarbon contamination. Results indicate a significantly higher incidence of hydrocarbons in the bile of fish from oiled areas than control areas in 1989 (P=0.005), however there were no differences in 1990 (P=0.9332) or 1991 (P=.8438).

In 1990, nine histopathologic lesions were scored by pathologists. However only four—liver lipidosis, liver sinusoidal fibrosis, liver, and kidney macrophage aggregates—were compared statistically. These lesions were selected because they were the most likely to be caused by exposure to toxins. In 1991, 26 different lesions were scored by the pathologists, however only the same four that were tested in 1990 were tested in 1991. Results from the statistical analysis of these lesion scores from rockfish in
Prince William Sound in 1990 and 1991 indicated that rockfish were exposed to toxic agents. There were differences between control and treatment sites in Prince William Sound in two of the four liver lesion scores (liver lipidosis \(P=0.0016\) and liver sinusoidal fibrosis \(P=0.0118\)) in 1990 and one of four (liver lipidosis \(P=0.008\)) in 1991. No differences in lesion scores were seen between sites on the outer Kenai Peninsula in 1990.

The histopathologic evaluation was conducted blind, that is, pathologists did not know if the tissues were from fish from oiled or unoiled sites. Upon completion of the histopathologic examination the pathologists “predicted” which sites were oiled based on qualitative analysis of lesion scores. The speculated exposure history was accurate for all four sites in Prince William Sound.

Subsequent statistical analysis using Principal Component Analysis was able to determine differences in oiled and unoiled sites in both 1990 and 1991. Differences were more significant in 1991 than in 1990 using this analysis.
Histopathologic Analysis of Chronic Effects of the Exxon Valdez Oil Spill on Alaska Fisheries
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To determine the long-term effects of the Exxon Valdez oil spill on fish resources in Prince William Sound, Alaska, sampling was begun in 1990, more than 12 months after the spill, on four types of fish: (1) Dolly Varden char (Salvelinus malma) adults; (2) Pacific herring (Clupea harengus) larvae and adults; (3) several rockfish species (Sebastes spp.); and (4) pink salmon (Oncorhynchus gorbuscha) larvae and adults. Organs chosen for histopathologic examination were those most likely to contain chronic or residual rather than acute lesions. Whole larvae, or adult liver, trunk kidney, spleen, and olfactory nares were sampled and preserved in Bouin’s or 10% neutral buffered formalin solution.

Fixed tissues shipped to the Aquatic Toxicology Program, University of California, Davis, were labeled by site of origin (e.g., Rocky Bay), but not by type of exposure history (i.e., oiled vs. clean/control). To eliminate bias during histopathologic examination, tissues from each fish were assigned a random number used only to identify a given study but not a site of origin. Tissues were embedded in paraffin, sectioned at 5 μm, and read in ascending numerical order. All lesions were semiquantitatively ranked (none = 0, mild = 1, moderate = 2, and severe = 3). After lesions were scored, significant lesions were identified using principal components analysis (PCA), and analysis of variance (ANOVA) of scale values derived from PCA was used to identify site differences. After histopathologic results were reported, actual exposure history was revealed. Multivariate ANOVA was used to differentiate oiled from clean sites, and also to account for other variables such as age or sex. Sampling was repeated in 1991 only if statistically significant differences between fish from oiled vs. clean sites were demonstrated in 1990.

Dolly Varden char in Prince William Sound congregate over winter (November—May) in a few freshwater lakes, but then split up and return to the mouths of their home streams to spawn and feed during the rest of the year (Andy Hoffman, personal communication). Some of these stream outlets were heavily impacted by the Exxon Valdez oil spill. In June 1990, livers were sampled from 12 Dolly Varden from each of five sites; in October 1990, liver, spleen, kidney, and olfactory nares were sampled from 14 to 20 fish from four of the five sites sampled in June. For the June samples, the first principal component explained 34% of the variability in the lesion scores, giving significant weight to hepatic lipodosis and hepatocellular megalocytosis.

Hepatic lipodosis is a common response associated with exposure of fish to a variety of different agents (Meyers et al., 1985) including petroleum hydrocarbons (Fletcher et al., 1979; McCain et al., 1978); however, other studies have shown a decrease in hepatic lipid stores in fish from oiled sites (Haensly et al., 1982). Megalocytosis is a marker of hepatocyte damage from a variety of insults (Hinton
et al., 1992), but has not previously been associated with exposure to oil. Scores from the two oiled sites were significantly different from three clean sites (ANOVA, \(P = 0.001\)), and were ranked in decreasing order for fish from Eshamy Creek weir (oiled, score = 1.7), Green Island weir (oiled, score = -0.17), Rocky Bay weir (clean, score = -0.43), Makarka Creek (clean, score = -0.47), to Boswell Bay (clean, score = -0.54). Only scores from Eshamy Creek weir were significantly different from other scores with Tukey's Studentized range test. For the October samples, the first principal component explained 16% of the variability, and oiled vs. clean differences were still highly significant (\(P = 0.0001\)).

Herring in Prince William Sound concentrate into a few select near-shore areas and spawn in April. Eggs hatch and movement patterns of resultant larvae are mostly unknown until they appear in bait fish industry catches approximately 18 months after hatch (Evelyn Biggs, personal communication). Movement of adult fish between annual spawns and potential for mixing of stocks are also unknown. Three types of histopathologic studies were completed with herring.

First, naturally spawned eggs were collected from oiled and clean sites, brought to the laboratory to hatch, and resultant hatchlings were examined for lesions (1989, 309 larvae; 1990, 189 larvae). Several of the larvae sampled in 1989 had cranial masses that resembled tumors; however, histopathologic examination revealed these to be a result of autolysis and not neoplasia. Vacuolation of the lens, a common finding in 1989 (both in oiled and clean larvae) but not 1990 larvae, was also a result of postmortem autolysis. None of the herring larvae had significant histologic lesions.

In the second group of herring studies, tissues from adults were sampled in April 1989 (ten fish each from two oiled and two clean sites), Fall 1990 (50 fish each from one oiled and one clean site), and April 1991 (20 fish each from one oiled and two clean sites). In 1989, severe hepatic necrosis in 20% of herring from the two oiled sites (Naked Island and Rocky Point) clearly differentiated these fish from control fish (none of the fish from control sites had severe hepatic necrosis).

Interestingly, PCA failed to demonstrate significant differences based on exposure history of the herring sampled in 1989. Hepatocellular necrosis was described in wild fish sampled nine months after the 1978 Amoco Cadiz oil spill (Haensly et al., 1982). In analysis of lesion scores in 1990 samples, the first principal component explained 18% of the variability, and placed substantial weight on liver glycogen, liver macrophages, liver single cell necrosis, splenic macrophages, and kidney macrophages. Hepatocellular megalocytosis, though present, did not contribute to site variability. Macrophage aggregates, including those in liver, spleen, and kidney, have been used as indicators of contaminant exposure and more often as a generalized nonspecific response to several stressful stimuli (e.g., starvation, heat stress) in many studies (Blazer et al., 1987; Herraez et al., 1986; Wolke et al., 1985) and numbers of hepatic macrophage aggregates were increased in fish exposed to oil (Haensly et al., 1982).

Studies have shown that macrophage aggregates increase with age in healthy fish but are independent of age in stressed fish (Blazer et al., 1987; Hinton et al., 1992). Age of herring from the oiled site (Green Island, 5.6 ± 0.16 years)
was significantly greater than the clean site (Knowles Head, 2.4 ± 0.13 years). Multiple analysis of variance (MANOVA) revealed that site differences were highly significant (P = 0.01), whereas age differences did not contribute significantly to lesion scores (P = 0.29). Differences associated with potential oil exposure were no longer detectable in herring sampled in the spring of 1991. In the third study, adult herring were exposed to crude oil in the water (20 or 60% water soluble fraction) or per os (force-fed in gelatin capsules) in the laboratory (220 exposed fish, 20 controls) for up to ten days. Exposed fish developed hepatic necrosis, providing strong evidence that oil was the cause of hepatic necrosis in wild-caught herring in 1989, and supporting evidence that macrophage aggregates in herring captured in 1990 resulted from oil exposure.

Rockfish in Prince William Sound establish home ranges and tend to stay on the same rock bed for years (Andy Hoffman, personal communication). Therefore, they are potentially an excellent group of fish to use for monitoring effects of perturbations such as the Exxon Valdez oil spill. In 1990, 121 rockfish (8 different species) were collected from four sites in Prince William Sound and four sites off the Kenai Peninsula. Nine lesions in the liver, spleen, and kidney were scored and analyzed using PCA; an orthogonal transformation was done to put more weight on macrophage scores in Factor 1. When species differences were ignored in ANOVA of Factor 1, oiled vs. clean differences were not significant (P = 0.21); however, when species differences were included, oiled vs. clean differences were significant (P = 0.035).

In 1991, 107 rockfish (either quillback, yelloweye, or copper rockfish) were collected from four sites in Prince William Sound. Twenty-six lesions in the liver, spleen, kidney, gill, and heart were scored; analysis was the same as for 1990 rockfish. When species differences were ignored in ANOVA, oiled vs. clean differences were not significant for Factor 1 (P = 0.09) but were significant for Factor 3 (P = 0.001); when species differences were included, oiled vs. clean differences were significant for Factor 1 (P = 0.001) but not for Factor 3 (P = 0.27).

Multivariate analysis of variance on the 1991 data showed significant oiled vs. clean differences whether species differences were ignored (P = 0.005) or considered (P = 0.02). Lesions contributing most to variability in 1991 included hepatic megalocytosis and
macrophage aggregates, and splenic macrophage aggregates. Rockfish were collected in 1991 nearly 2.5 years after the Exxon Valdez oil spill, but histopathologic analysis indicates that lesion differences were more significant in 1991 than in 1990. Therefore, additional sampling is proposed for 1993 to determine if toxic effects continue.

Acknowledgments - Neil Willits performed the statistical analysis. Histopathologic analysis was made possible by efforts of Alaska personnel to identify sampling sites, collect fish, and coordinate hydrocarbon analysis and population studies. We particularly acknowledge the following: Joseph Sullivan, project coordinator; Evelyn Biggs, Tim Baker, and Adam Mole, herring; Samuel Sharr, Henry Yuen, Mark Fink, Mike Wiedmer, and Brian Bue, pink salmon; and Kelly Hepler and Andy Hoffman, rockfish and Dolly Varden char.

References
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Embryolarval development of Prince William Sound herring was evaluated from 1989 to 1991 to determine possible adverse effects from the Exxon Valdez oil spill. Herring embryos were collected from three replicate sites within each oiled or unoiled location. Because herring did not spawn at the same sites every year, sites were generally different for each year.

Fairmont Bay, an unoiled location, was sampled every year as was Rocky Bay, an oiled location on Montague Island. Naked Island, an oiled location, was sampled only in 1989 and 1990. Eggs were transported to the laboratory and incubated to hatch. Larvae were assessed for two types of sublethal damage, morphologic malformations and genetic effects. Relationships between the sublethal endpoints and hydrocarbon measurements from caged mussels were examined. Hydrocarbon measurements were ranked using principal components (PCA) specific to Exxon Valdez crude oil.

In 1989, larval malformations were significantly (p<0.05) more severe at the two oiled locations (Rocky Bay and Naked Island) than at Fairmont Bay. Malformation scores at Naked Island were 55% higher than those from Fairmont Bay; differences were significant at all three depths. At Rocky Bay, scores were elevated by a mean of 37% but only the middle and high depths were significantly higher. The observed malformations included skeletal curvatures, craniofacial defects, reduced cephalic differentiation, and finfold reductions. Larval defects are a standard manifestation of embryonic stress, whether from a toxic event or extreme environmental conditions such as high temperature (von Westernhagen, 1988; Weis and Weis, 1989).

Both morphologic endpoints, the severity of the malformations and the percentage of malformed larvae, ranked consistently with the level of oiling in 1989 by location. At Fairmont Bay, both endpoints showed the least damage and the mean PCA value was 2.0 (no to low oiling). Rocky Bay had intermediate values for sublethal damage and its mean PCA score (6.7, not high oiling). Naked Island had the greatest sublethal damage and the highest mean PCA value (9.8, low to high oiling). The chemistry results reflected variability in the oiling levels within areas. Neither of the morphologic endpoints was significantly correlated with site-specific PCA scores.

Mitotic configurations in the pectoral fins were enumerated and evaluated for evidence of chromosome breakage (anaphase aberrations). Fairmont Bay larvae averaged 8.5 mitoses per fin and 10.4% aberrant anaphase configurations, values within normal limits. Larvae from the two oiled locations had reduced cell division and elevated anaphase aberration rates relative to Fairmont Bay larvae. At Rocky Bay, the aberration rate was 33.9%, 2.3 times higher than at the
Herring: Effects on Herring Embryos and Larvae

At the unoiled site at Naked Island, the aberration rate was 45.7%, 3.4 times higher. More individuals from the oiled locations thus exhibited genetic damage capable of reducing their subsequent survival. Reduced cell division and chromosome breakage result from exposure to genotoxic agents, including petroleum hydrocarbons (Longwell and Hughes, 1980).

Similar cytogenetic effects have been previously documented in fish eggs following oil spills (Longwell, 1977). All three cytogenetic endpoints (number of mitoses, % aberrations and % cytogenetically abnormal larvae) were ranked consistent with PCA scores among the three locations. Fairmont Bay larvae exhibited the least cytogenetic damage, Rocky Bay larvae were intermediate, and Naked Island larvae were most affected. Skeletal malformations and the anaphase aberration rate were significantly correlated with site-specific PCA scores. Skeletal and finfold defects, the anaphase aberration rate and the percentage of cytogenetically abnormal larvae were correlated with concentrations of aromatic hydrocarbons.

Subsequent sampling in 1990 and 1991 demonstrated that sublethal damage was undetectable. The extent of sublethal damage in Prince William Sound herring larvae declined from 1989 to 1990. The reduction in morphologic damage resulted in part from the improved incubation technique in 1990 but cytogenetic measurements were unaffected by the change. In 1990, morphologic endpoints were within baseline levels at all three locations but both the severity of malformations and the percentage of malformed larvae were significantly higher at Naked Island than at Fairmont Bay. Cytogenetic endpoints were within baseline levels at Fairmont Bay and Naked Island, consistent with the decrease in hydrocarbon measurements at all sites evaluated in 1990. The mean PCA scores for Fairmont Bay and Naked Island were less than zero and that of Rocky Bay was 0.6. At Rocky Bay, the aberration rate and the percentage of cytogenetically abnormal larvae were significantly higher compared to the unoiled site. Because the 1990 hydrocarbon levels were also low at Rocky Bay, this residual genotoxicity might reflect sustained damage to the adults and thus be unrelated to mussel uptake measurements. To resolve this complicated picture, sublethal effects were more intensively studied the next year.

In 1991, morphologic and cytogenetic measurements were similar between Fairmont Bay and Rocky Bay. All endpoints were slightly higher than in 1990, probably reflecting the colder 1991 water temperatures. Whereas the aberration rate and percentage of cytogenetically abnormal larvae were significantly elevated at Rocky Bay in 1990, 1991 values were virtually identical to those from Fairmont Bay. Aberration rates were both 21.5%, slightly above the upper normal limit of 20%. At Fairmont Bay, 58% of the larvae were cytogenetically abnormal compared to 57% at Rocky Bay. These data suggest that oil-related toxicity was ameliorated by 1991 at Rocky Bay.

Herring did not spawn on Naked Island in 1991, so embryos were placed at three of the 1989 sites and allowed to develop in situ. Morphologic and cytogenetic analyses detected significant toxicity present at only one site which had been moderately oiled in 1989. Two 1989 sites at Rocky Bay were similarly evaluated, and consistently adverse sublethal
effects were not found. To summarize, these data suggest that the Exxon Valdez oil spill transiently elevated the incidence of sublethal damage in larval herring within certain affected areas but significant damage did not persist into 1991.

In the laboratory, herring embryos exposed to low concentrations of Prudhoe Bay water soluble fraction (WSF) exhibited sublethal damage similar to that observed in the field. The morphologic endpoints were significant at 0.10 to 0.48 mg/L WSF and the cytogenetic endpoints significant at 0.01 to 0.24 mg/L WSF. The anaphase aberration rate proved to be the most highly sensitive endpoint with rates elevated above the control at the lowest dose tested, 0.01 mg/L WSF. Significant differences at 0.10 mg/L WSF were detected with three other endpoints: craniofacial malformation severity, finfold malformation severity, and the percentage of cytogenetically abnormal larvae. All endpoints were significantly correlated with the log WSF dose. This presentation will also discuss relationships between the sublethal effects observed in the field and in the laboratory and measurements of Exxon Valdez crude oil throughout Prince William Sound.

Sublethal impacts are an important part of toxicity assessments since they can be used to predict long-term damage or estimate effects on life stages that are difficult to study. For instance, sublethal effect data from herring can be integrated with measurements of egg abundance, embryo survival and larval densities to obtain estimates of embryo/larval success. Such estimates are essential to define potential toxic effects on fish populations.

References
The purpose of this study was: (1) To determine if short-term exposure to water soluble fraction (WSF) adversely affected developing embryos to different degrees depending on their developmental stage; (2) To establish an EC$_{50}$ for Prudhoe Bay water soluble fraction following continuous embryo exposure for the entire incubation period; and (3) To evaluate previously oiled and unoiled sites in situ for embryo toxicity.

To minimize inter-female variability, eggs from 8-10 females were randomly distributed to 100 slides at a density of 40-60 eggs/slide. Sperm from three to four males was then pooled and used to fertilize the eggs. After one hour, several slides were examined to verify fertilization success. At least 90% of the eggs had to be fertile in order to consider the spawning successful.

Slides containing fertile eggs were placed into a Plexiglas carrier to prevent slide-to-slide contact during transit, then submerged in seawater, gassed with O$_2$ and placed into a cooler containing wet ice for transport back to the University of Washington by commercial airliner. The total elapsed time from fertilization in Prince William Sound to arrival in Seattle was eight hours. Once at the University, the embryos were incubated at 8°C in 29 ppt seawater in an environmental chamber for the duration of the experiments. Embryos were maintained in 300 ml of seawater and gently aerated with approximately 60 bubbles/minute for the entire incubation period. The combination of a high humidity and low rate of aeration resulted in minimal evaporative loss. Dissolved O$_2$ remained constant at 10-11 mg/L/L.

Transport Effects

In order to control for possible transport effects, ripe Puget Sound herring (PS) were transported to the University of Washington and spawned in the environmental chamber as described above. These were then incubated in parallel with the Prince William Sound embryos, exposed similarly to WSF and compared for differences in survival time and developmental abnormalities. The experiment was designed to detect any effect of prolonged transport.

Preparation of WSF

To test the toxicity of water soluble components of oil, a water soluble fraction was prepared by shaking 40 ml of oil with one liter of 29 ppt synthetic seawater in a 2L separator funnel at 8°C for 15 minutes, then allowing the mixture to separate in the funnel for 18 hours at 8°C. The funnel was tapped lightly several times to release oil droplets adhering to the glass, then the water layer was drained off into a glass-stoppered bottle and used as a stock for exposure dilutions. The seawater removed from the separator funnel was designated 100% WSF, and used as a stock from which dilutions down to 0.1% were made. A new stock was prepared every 48 hours.
for the duration of the embryo exposures (approx. 21 days).

Chemical analyses of WSF were based on the total peaks observed for low molecular weight (C6-C12) gasoline range hydrocarbons, and high molecular weight (C12-C28) diesel range hydrocarbons. Low molecular weight (LMW) samples were analyzed by GC/FID (Purge & Trap) and the high molecular weight (HMW) samples by GC/FID, both modified U.S. Environmental Protection Agency method 8015. Because LMW values decreased with time, the HMW samples were used to convert "%" WSF to real values corresponding to mg/L (ppm) of dissolved petroleum hydrocarbons. Analysis of three replicate extractions demonstrated that 100% WSF contained 9.67 mg/L (ppm) of HMW hydrocarbons.

Developmental Stage Sensitivity

To establish specific developmental stage sensitivity to WSF, 24 hours after fertilization, and every 24 hours thereafter, a different group of embryos was exposed to WSF for 36 hours. This resulted in four groups of embryos being exposed to WSF at 24, 48, 72 and 96 hours post fertilization. Three concentrations of WSF were used (25%, 50% & 100% WSF), which corresponded to 2.4, 4.8 and 9.7 ppm Prudhoe Bay crude oil. After the exposure period was complete, the embryos were washed free of adhering oil and incubated in flowing natural seawater until they hatched. The exchange rate was 6-8 times per hour, ensuring adequate aeration and removal of excreted metabolites.

Upon arrival at the University of Washington the Prince William Sound embryos were placed into 300 ml of WSF ranging from 100% to 0.1% (9.67-0.009 ppm). The WSF was changed every 48 hours for 18 days. Larvae were then allowed to hatch into clean seawater so that only embryotoxic effects would be measured.

Genotoxic Damage

Chromosome and mitotic damage was evaluated by examining mitotically active tissue from newly hatched larvae which had been exposed during the EC₉₀ determinations. The number of mitoses and the number of abnormal anaphase-.telophase cells in each of the treatment groups were evaluated from germinal layer between the muscle cells and developing ray structure of the larval pectoral fin.

In Situ Embryo Exposures

Field deployment occurred in late April of 1991 in Prince William Sound. Slides containing newly fertilized eggs were placed into PVC cassettes and placed at two depths each of two oiled and two unoiled sites. The cassettes were retrieved from the field 10-12 days later and returned to the University of Washington. Embryos were in the environmental chamber in clean aerated seawater within 8 hours after being retrieved. Each exposure site received 250-350 eggs. Exposures occurred at 10 sites within Prince William Sound designated "C" and "O", with deployments below the mean low water mark at 5 ft and 15 ft.

(In Vitro) Embryonic Stage Sensitivity

The earliest exposure periods and highest WSF concentrations had the greatest effect on hatching success of herring embryos. The 24 and 48 hour developmental stages were the most severely affected with a 20-45% embryo mortality relative to the controls. By 96 hours post fertilization, embryo survival
increased and was 80-100% of the control levels.

Increasing concentrations of WSF produced an increase in the percent of abnormal larvae, ranging from 10% at 2.5 ppm to 65% in 10 ppm WSF. The exposed embryonic stage however, had no detectable influence on normal larval development. Abnormalities evaluated were scoliosis, lordosis, cranial malformations and optic deformities.

**EC₉₀ Determination for WSF**

Observations on the mean-hatching-day post fertilization revealed that embryo exposure to concentrations >0.242 mg/L WSF resulted in embryos hatching 4 to 5 days earlier (mean = 15 days) than did the controls and lower WSF concentration exposures (mean = 19 to 20 days). This concentration is one half the abnormality EC₉₀.

Continuous embryo exposure to WSF of crude oil had little or no effect on embryo survival or live hatch, but did significantly increase the percent of physically defective larvae. Physical deformities included spinal deformity (scoliosis or lordosis), optic malformations, mandibular malformations and an enlarged pericardial region. These defects appeared not to be pathognomonic so were all considered together for the purpose of this study (e.g. total abnormal larvae).

The abnormality data were analyzed using EPA’s Probit Analysis Program for data from Acute and Short-Term toxicity tests with aquatic organisms (EPA Biological Methods Branch, Cincinnati, O.). An EC₁₅ to EC₉₀ curve was generated which shows an EC₅₀ of 0.432 mg/L of the high molecular weight components of WSF. The EC₁₅ was 0.078 mg/L and the EC₅₀ 2.39 mg/L. By comparing these values to those observed in the field, it should be possible to predict the number of larvae which would be affected following exposure to a specific concentration of WSF in situ.

Normal untreated larvae were 71% heavier (2.4 mg/20 larvae) than the untreated abnormal larvae (1.4 mg/20 larvae). Normal larval weights decreased as the WSF concentration increased, but abnormal larval weights remained constant as the WSF concentration increased. Because many of the larvae were so severely deformed, it was not possible to obtain accurate lengths. Consequently, this measurement was abandoned in favor of the more consistent dry weight measurement.

**Genotoxic Damage**

Examination of mitotically active cells revealed that mitotic activity was significantly reduced at 0.24 ppm WSF and greater. Chromosome abnormalities micronuclei were significantly increased at and above WSF concentrations of 0.01 ppm, well below that which produced grossly visible physical abnormalities.

The mean number of embryos hatching in the control (C) group was significantly lower than in the oiled (O) group (p < 0.01; t Test). There was also a significantly greater number of normal larvae hatching from the “C” group (63.3%) than from the “O” group (51.3%) (p << 0.01; t Test). This response is similar to what was seen in the in vitro embryo exposures. It will be necessary to obtain analytical data on the chemical contaminants on site at the time of exposure before any correlation can be made between in vitro and in situ responses.

A dry weight analysis of larvae hatching from the two exposure groups showed that the mean weight of both normal and abnormal larvae from the
"C" group were significantly higher than the weights of the "O" group (p <=< 0.01; t Test).

The mean % hatch was 73 +/- 11 for Prince William Sound and 65 +/- 9 for Puget Sound embryos, and identical values for gross abnormalities, indicating nonsignificant effect resulting from transport of the fertilized eggs from Alaska to Washington.

Conclusions

1. Early developmental stages (24-48 hrs post fertilization) appear to be the most sensitive to the effects of Prudhoe Bay WSF. The concentration of WSF and not developmental stage however, influenced the production of grossly abnormal larvae. Consequently, under natural exposure conditions it might be expected that early exposure of embryos would result in higher mortality while the concentration to which they were exposed would result in increased numbers of abnormal (non-viable larvae).

2. EC_{50}: The experimental EC_{50} of Prudhoe Bay crude oil WSF for Pacific herring embryos exposed for their entire incubation period was 0.43 ppm. By comparing this value with the concentration of WSF found following the Exxon Valdez oil spill, it should be possible to determine the total loss of herring embryos from oiled sites within Prince William Sound.

3. Genetic Damage: Significant damage to chromosomes and mitotically active cells in newly hatched herring larvae occurred at concentrations well below the EC_{50}, indicating that sublethal long-term damage had occurred and may not become apparent for several generations.

4. Hatching dynamics: Precocious hatching occurred at concentrations of WSF at or above 5% (0.484 ppm), with larvae hatching 4 to 5 days early. Alteration of mean hatching day frequently occurs in fish exposed to a widerange of pollutants. Early hatching may produce weak ill-prepared larvae which are more vulnerable to predation than those hatching after a normal incubation period.

5. Physical defects: The most obvious effect of WSF on developing herring embryos was the induction of physical defects in live-hatched larvae. Specific defects were not pathognomonic so were not reported separately. The LC_{50} for total abnormalities was 0.432 ppm with no normal larvae being present at or above 0.96 ppm.

6. Larval weights: A reduction in dry weight appeared to be related to exposure to WSF only in normal larvae. There was no correlation between abnormal larval weights and exposure to WSF in vitro. Not enough data is available to properly interpret the differences in larval weights observed at the oiled and unoiled sites in Prince William Sound.
Egg-larval Mortality of Pacific Herring in Prince William Sound, Alaska, After the Exxon Valdez Oil Spill
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The Exxon Valdez oil spill of March 24, 1989, was followed 2.5 weeks later by spawning of the local stock of Pacific herring (Clupea pallasi). The effect of the spill on herring may have been restricted to eggs because growth and mortality of herring larvae captured 1 to 5 km offshore of the herring egg beds in 1989 were not significantly different between oiled and non-oiled areas (McGurk et al. in press). This study tested the hypothesis of an egg effect by comparing in situ egg-larval mortality between two oil-exposed areas and two non-exposed areas.

Two of the four major herring spawning sites in the Sound, the North area centered on Fairmount Island and the Northeast area centered on Tatitlek Narrows, were classified as non-oiled because the oil slick never contacted them. The other two spawning areas, the Naked Island archipelago and the northern tip of Montague Island, were classified as oiled.

Egg-larval mortality (Z, day⁻¹) was the ln-transformed ratio of larval density at hatch (NL, number m⁻² sea surface) to mean egg density (NE, number m⁻² spawning bed), divided by the duration of the egg-larval period (t, days), i.e. Z = -(1/t)ln(NL/NE).

Density of larval herring at each of the four areas was measured by weekly oblique plankton tows to 30 m depth from May 7 to June 22, 1989. Herring larvae were sorted from the zooplankton, counted and density was corrected for net avoidance by larvae. A linear regression of ln(NL) on age was fit to the descending right-hand limb of the catch curve for the major cohort at each plankton station. Larval density at hatch was the intercept of the regression at t = 0 days.

Egg density at each area was measured by SCUBA divers as part of ADF&G's annual herring spawn survey. Mean egg density was the geometric mean of all transects that could have reasonably been expected to contribute larvae to the pool sampled by the plankton nets.

Aerial surveys by ADF&G found that mid-points of the spawning period in the four areas ranged from April 11 to 13. SCUBA surveys of egg density were conducted about 10-16 days later. Hatch dates of the four major cohorts of larvae occurred 7 to 12 days after the mid-dates of the SCUBA surveys.

Z was significantly different between areas; the greatest Z, 0.598 day⁻¹, was measured at an oiled site on Montague Island and the lowest Z, 0.065 day⁻¹, was measured at a control area in the North area. Mean Z in the two oiled areas, 0.410 day⁻¹, was three times higher than mean Z in the two non-oiled areas, 0.123 day⁻¹.

Unfortunately, spatial distribution of herring eggs differed between the four areas in such a way as to confound the interpretation of egg-larval mortality. For example, mean width of herring spawn was significantly greater at Montague
Island than at the North area, which implied that the Montague Island site had a lower beach gradient than that of the North area. Since egg loss due to scouring by waves decreases with increasing depth, low gradient beaches will tend to have greater scouring than high gradient beaches, all other factors being equal. Also, the distribution of herring spawn ranged from 48.8% subtidal in the Northeast area to 75.4% subtidal at Bass Harbor. Since intertidal eggs are more vulnerable to desiccation and bird predation than subtidal eggs, egg-larval mortality decreases with increasing percent subtidal eggs.

We conclude that this study offers tentative support to the hypothesis that the Exxon Valdez oil spill increased egg-larval mortality in oiled areas of Prince William Sound. It is not conclusive evidence for an oil effect because an unknown portion of the between-area differences in egg-larval mortality may have been caused by between-area differences in natural egg mortality. We cannot eliminate this possibility because we have no independent information on natural egg loss rates in 1989.

To the best of our knowledge, this study is the first to estimate herring egg-larval mortality by combining measurements of larval density with direct measurements of egg density. It is valuable because it provides estimates of egg mortality that are independent of the mode of mortality.

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There is no documentation of larval fish species and distribution in Prince William Sound prior to the Exxon Valdez oil spill. In response to the 1989 oil spill, six 1-week cruises in Prince William Sound (April-November) and four 4-day cruises in Resurrection Bay (May-July) were conducted. Both oiled and control unoiled sites were sampled. Plankton tows were taken with a 1 m³ NIO Tucker trawl or a 1 m³ MOCNESS with 505 μm mesh nets. Discrete depth increments were sampled to 100 m in Prince William Sound and to 250 m in Resurrection Bay.

Over 40,000 larvae were collected in Prince William Sound and in Resurrection Bay. Over the 8-month sampling season, the greatest proportion of fish larvae were captured during May in both Prince William Sound and Resurrection Bay. Of the larvae captured in May, walleye pollock (Theragra chalcogramma) was the major species, comprising 80% of the fish captured. Pollock larvae were well distributed around the Sound.

In May concentrations of pollock larvae were collected in both oiled (Montague Island, Knight Island, Main Bay) and unoiled sites (Orca Bay, Port Valdez). Most of the pollock larvae were captured in the upper 50 m of the water column. Size distribution of pollock larvae captured within Prince William Sound in 1989 ranged from 2.8 mm to 11.3 mm and ranged in age from 1 to 40 days. A bell-shaped distribution of lengths probably indicates only one cohort. Larvae on the western side of the Sound were slightly larger, hence older, than those found in the middle and eastern portions of the Sound, perhaps indicating movement through the region.

Pacific herring (Clupea harengus pallasi) were captured in May, June and July, but not April, September or October. In May, 321 herring larvae were captured. Most (197) were at one oiled station south of Naked Island. The others were taken in unoiled sites (Knight Island) and unoiled sites (east side of Hinchinbrook Entrance). In June, 1349 herring larvae were collected. Of those, 1175 were in oiled areas around Montague and Knight Island, with 765 larvae taken at one station near Green Island.

Very few larvae were collected in unoiled areas near Ester Island and eastern Prince William Sound near Hinchinbrook Island. In July there were few (56) herring larvae evenly distributed in oiled and unoiled areas. Like pollock, most herring larvae were in the upper 50 m of the water column. The herring larvae ranged in size from 6.0 to 23.2 mm.

We will relate the distribution of larval fish to the distribution of the oil spilled. We will use length/frequency analysis to follow cohorts over time and space in relation to the distribution of oil. A modified Graded Severity Index (GSI) will be used to measure morphological deformities in herring larvae due to oil.
exposure.

This study demonstrates that at least minimal baseline data are needed for species' presence/absence seasonally so that in the event of an oil spill or other catastrophic event we will be able to assess the effect on larval fish, and the ultimate effect on the fisheries.