25 July 2007

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Exxon Valdez Oil Spill Trustee Council
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From: Thomas Kline, Principal Investigator
Project 070811/PWS Herring Forage Contingency
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Subject: Cover sheet detailing any project components or budget items that have changed from original submission pre request Re: Project 070811/PWS Herring Forage Contingency

Thank you for this opportunity to provide a progress report and revised DPD. In addition to this note, please find three documents attached, a revised DPD, a revised budget and an interim progress report. Please let me know if you have questions about any of these materials.

Hardcopies of these documents are being mailed today.

DPD changes:

As discussed in the progress report, we were not able to participate in the LTOP (Long Term Observation Program) cruise because an ocean class research vessel was not available. Instead, two days were added to the length of our May cruise to fulfill sampling needs. We assume that this will be the case in 2008 and are planning accordingly. Thus travel to Seward has been dropped for 2008 and the same length charters planned for both May and September. Vessel charter rates have increased due to soaring fuel costs. We are estimating that each 2008 cruise will cost 24K, which is an increase of 5K per cruise from that estimated last year. To make up part of the added charter costs, the air charter to board the fish sampling cruise in the Sound was dropped from the budget as this was not needed in 2007 and assumed it will be not needed in 2008. The dropped travel to Seward and dropped air charter are not enough to compensate for the added fuel costs, thus the FY08 budget is about 10K higher than originally anticipated.

The following changes to the DPD text reflect the changed cruise logistics:

Original:

LTOP systematic sampling will continue on this project but on a more limited scope compared to that done during GLOBEC (Global Ocean Ecosystem Dynamics; http://www.globec.org/). *Current LTOP zooplankton sampling, consisting of the 13 stations comprising the 'Seward Line', GAK1 to GAK13, and three in Knight Island Passage in western PWS, is being lead by R. Hopcroft (UAF) with cruises funded by NPRB* (North Pacific Research Board). *The PI (Kline) is presently participating as a guest investigator on LTOP cruises to ensure continuity of collections since GLOBEC, to furnish the Steller sea lion project with SIA* (Stable Isotope Analysis)*'end-members.'*

Replaced with:
Systematic SIA sampling previously done LTOP will continue on this project but on a more limited scope compared to that done during GLOBEC in terms of sampling on the Seward Line. LTOP zooplankton sampling, which consisted of the 13 stations comprising the 'Seward Line', from GAK1 to GAK13, will be reduced to the Slope stations GAK10 to GAK13.

Original:
Continued participation in LTOP cruises will enable collecting May end-members exactly as was done on GLOBEC (Kline submitted to Deep-Sea Res. II). Two days of the August cruise will be dedicated to sampling out to the continental slope south of PWS to furnish a summer oceanic end-member. The summer PWS end-member data will come from the four bays as well as the central area.

Replaced with:
Continued sampling from GAK10 to GAK13 will enable collecting May end-members exactly as was done on GLOBEC (Kline submitted to Deep-Sea Res. II) furthering the existing time series. Two days of the May and August cruises will be dedicated to sampling out to the continental slope south of PWS to furnish oceanic end-members. PWS end-member data will come from the four bays as well as the central area. Intrinsic to the stable isotope methodology, which is a major focus of the project, is the determination of end-members (e.g., Kline et al. 1990). Getting the Gulf of Alaska end-member is crucial and a particularly difficult task in the Gulf of Alaska because of the effect of weather on logistics, i.e., the need to sample zooplankton at the continental slope, which is a significant sailing distance from shore.

Deleted:
Additionally, the PI will participate in the LTOP cruises funded by NPRB (Hopcroft PI). K. Coyle will be leading zooplankton sampling on these cruises. The PI has worked with Coyle for over a decade on MOCNESS sampling, which will be used to obtain zooplankton on this project as described by Kline (submitted).

Deleted from budget justification (parts added to DPD main body shown above):
Intrinsic to the stable isotope methodology, which is a major focus of the project, is the determination of end-members (e.g., Kline, T.C. Jr., J.J. Goering, O.A. Mathisen, P.H. Poe, and P.L. Parker. 1990. Recycling of elements transported upstream by runs of Pacific salmon: I. ^{15}N *and* ^{13}C *evidence in Sashin Creek, southeastern Alaska. Can. J. Fish. Aquat. Sci. 47:136-144. Getting the Gulf of Alaska end-member is crucial and a particularly difficult task in the Gulf of*

Alaska because of the effect of weather on logistics, i.e., the need to sample zooplankton at the continental slope, which is a significant sailing distance from shore.

Deleted from budget justification:
Drs. Hopcroft and Coyle (UAF) and the PI were GLOBEC collaborators on LTOP cruises, which are being continued here. Vessel support for LTOP cruises comes from NPRB.

… one round-trip to Seward for the PI and post-doc to participate on LTOP cruises to collect samples for this project, and other travel costs including subsistence. LTOP purpose:

The following DPD changes are mainly updates:

Deleted: old CV

Replaced with: new CV

Deleted (Crawford's project was not funded so references to it have been removed):

Fine scale analysis is focused on eastern PWS in a project being led by R. Crawford (PWSSC). This project will provide Crawford with zooplankton analyses (SIA, ZED, and SED) that are analogous to those done in the four bays and central area.

There will also be two zooplankton cruises funded as part of the fine scale study (Crawford PI) in May and August. The PI's will be coordinating these two cruises each month as they will be participating in both.

Dr. R. Crawford ("Characterization of Pacific herring nursery habitat in Prince William Sound")

Updates to references cited:
Hay, 2007 added
Kline, T.C. Jr. 2006 updated
Kline, 2007 added
Kline et al. 1990, added

Changes to FY08 budget:
On P. 5 of 10 'Cruise travel' is now 0 K
On p. 6 of 10 'Vessel Charter' is now 48.0 K (see beginning of this document for details narrative)
On p. 6 pf 10 '2 R/T air charter …' is now 0 K

INTERIM PROGRESS REPORT

July 2007

Project Title: Prince William Sound Herring Forage Contingency

Project Number: 070811

Principal Investigator: Thomas C. Kline, Jr., Ph. D.

Affiliation: Prince William Sound Science Center

Introduction

The Herring Forage Contingency project is addressing how energy flow through the Prince William Sound (PWS) pelagic ecosystem is limiting herring recovery. In particular, we want to know why, based on their low whole body energy content, herring are apparently not getting enough zooplankton food in what is assumed to be a rich zooplankton ecosystem. To address this question we are sampling zooplankton, age-0 herring, and other zooplanktivorous fishes that are potential competitors of age-0 herring for their energy content, stable isotope abundance, and in the case of zooplankton, their abundance using nets and acoustics. Fish sampling is taking place in Eaglek, Simpson, Whale, and Zaikof Bays because these were the four bays sampled a decade ago in the EVOS-TC-funded Sound Ecosystem Assessment project (Fig. 1). This will enable assessing whether herring are better or worse off now compared to then.

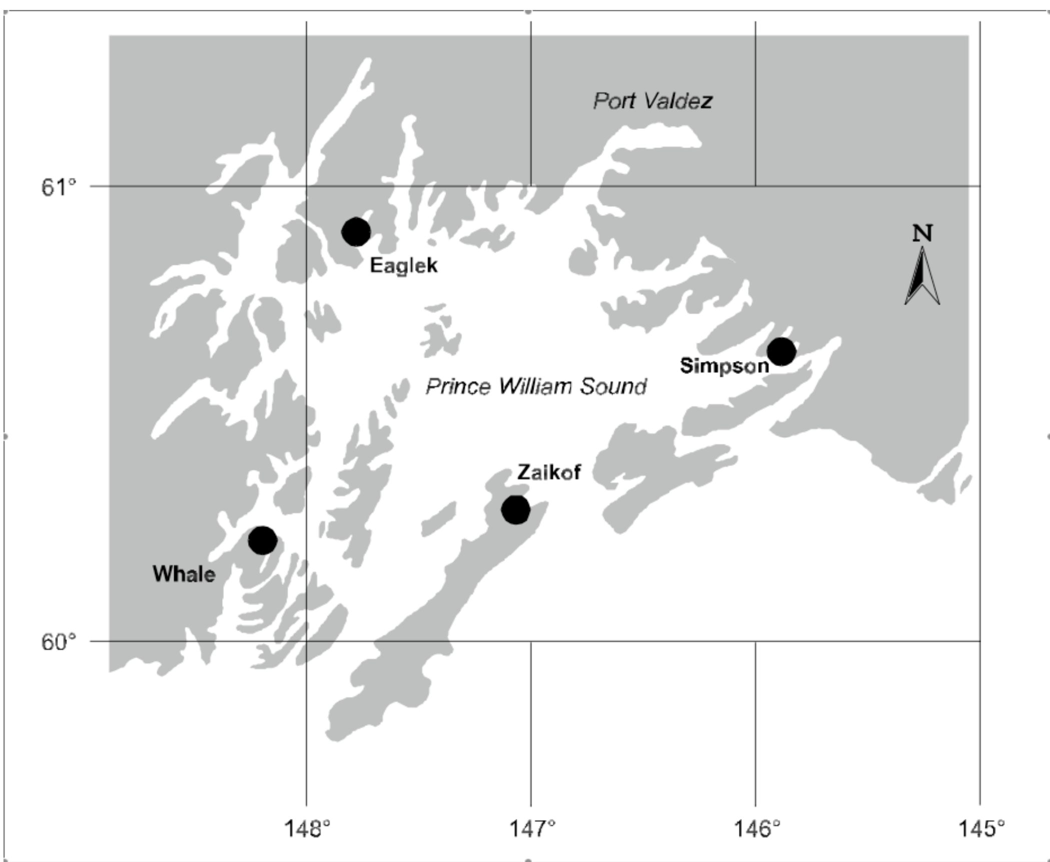

Figure 1. The four Prince William Sound herring nursery bays where age-0 herring, potential competitors and zooplankton forage are being sampled.

When zooplankton are transported into PWS from the Gulf of Alaska they will subsidize the forage of juvenile herring. These oceanic subsidies are driven in part by the dynamic nature of the physical environment, and in part by the need for deepwater habitat over part of the life cycle of many zooplankton species. The rapid spread of the *Exxon Valdez* oil spill out of PWS and into the Gulf of Alaska (GOA) after one week provides an example of the dynamic nature of the environment. There is also exchange of water into PWS from the Gulf, and this exchange of water is hypothesized to drive zooplankton exchange, which in turn is hypothesized to drive oceanic food subsidies. As well, copepods of the genus *Neocalanus*, the dominant zooplankton of the sub-Arctic Pacific Ocean, require water more than 300 m deep for over-wintering. Whereas the western portion of PWS has sufficiently deep water, the extent of deep-water habitat in the open ocean is far greater (Fig. 2), thus the potential supply of zooplankton from the open ocean is far greater. There are other copepod species, as well as other zooplankton types such as krill (euphausiids), that also require deep-water habitat. PWS herring nursery areas are in comparatively shallow water, depth is generally < 50m. Thus the occurrence of certain zooplankton in herring nursery habit is a result of advection, which may include subsidies of oceanic origin. For example, *Neocalanus* copepods are found in Simpson Bay, which is a substantial distance from their over-wintering habitat (compare Figs 1 and 2). A goal of the project is to assess how important oceanic food subsides are for herring.

Oceanic zooplankton food subsidies may be detected using stable isotope analysis (SIA). The SIA technique involves characterizing the isotopic composition of GOA and PWS zooplankton carbon by direct analysis of representative zooplankton samples from these locations. SIA is also performed on tissues of age-0 herring. Because there are predictable relationships between the stable isotope values of consumers (such as fish) with their diet, one may use SIA of biological samples to reconstruct food webs. A recurrent regional stable carbon isotope gradient between PWS and the adjacent GOA distinguishes organic carbon generated in PWS from organic carbon generated in the GOA. The SIA values of herring can thus be compared to the SIA values of zooplankton from the GOA and PWS to assess the relative contribution of each source. During the

Sound Ecosystem Assessment program period herring dependence on GOA subsidies ranged from about half to almost 90%.

The Herring Forage Contingency Project is assessing the current status of how zooplankton forage may be limiting recovery of herring populations in PWS. The relative proportion of GOA subsidies and relative energy content of age-0 herring are hypothesized to be contingent on occurrence of high-density patches and on occurrence of species suitable as herring food. The energy content of zooplankton must also be sufficiently concentrated so that herring foraging activity results in a net gain in energy. This is critical because herring in PWS have been shown to vary in whole-body energy content and that this energy content may not be sufficient for over-winter survival. Accordingly, the energy content of zooplankton, which is likely to vary in space and time, is also being assessed.

Climate change is hypothesized to drive long time scale changes in zooplankton populations. Long time scale population changes may occur as change in species composition as well as total numerical abundance. Certain species are favored in colder conditions while others are favored in warmer conditions. It is presently unknown how these changes affect the fishes that feed upon them. Closely monitoring zooplankton populations will address this need, which will further our understanding of how and why populations of herring and other fishes that depend upon zooplankton fluctuate.

The Herring Forage Contingency Project commenced in March 2007 with a PWS fish-sampling cruise. A project technician was hired. This was followed by a zooplankton sampling cruise in May to capture the spring bloom. Presently (July 2007) samples are undergoing preparation for analysis. Details of the two cruises and some preliminary results are presented in the following sections.

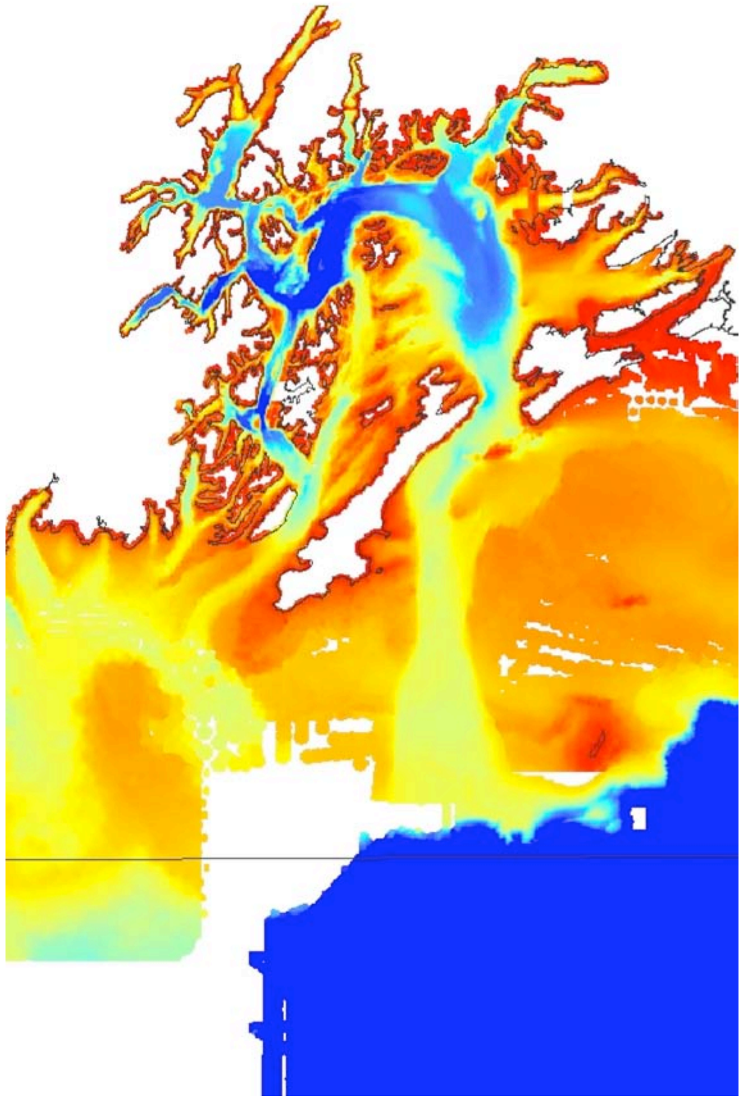

Figure 2. Study area bathymetry. Areas deeper than 300 m are shown as blue. A notable aspect of PWS is the deep area (up to 800 m deep) north of Knight Island and east of Lone Island known as the 'Black Hole.' Note also the submarine canyon extending from the northern end of Hinchinbrook Island south across the relatively wide continental shelf towards deep waters of the North Pacific Ocean. This image, provided by Dr. Carl Schoch, reflects a 'work in progress' and thus is incomplete as indicated by the white areas. Nevertheless it provides a good representation of the locations of deep areas.

Fish Sampling

The Project's PI and post-doc sampled fish by participating in a fish-sampling cruise led by a sister project under the direction of Dr. R. Thorne. Sampling took place in the aforementioned four bays shown in Fig. 1. Fish were caught with a trawl designed by Dr. Thorne's associate, Dr. R. Crawford (Fig. 3). In addition to age-0 herring, catches and thus collections included juvenile walleye pollock, including those that were age-0. Additional fish samples collected under the direction of Steve Moffitt of the Alaska Department of Fish and Game were provided to this project. Fish samples were frozen and are presently in preparation for SIA.

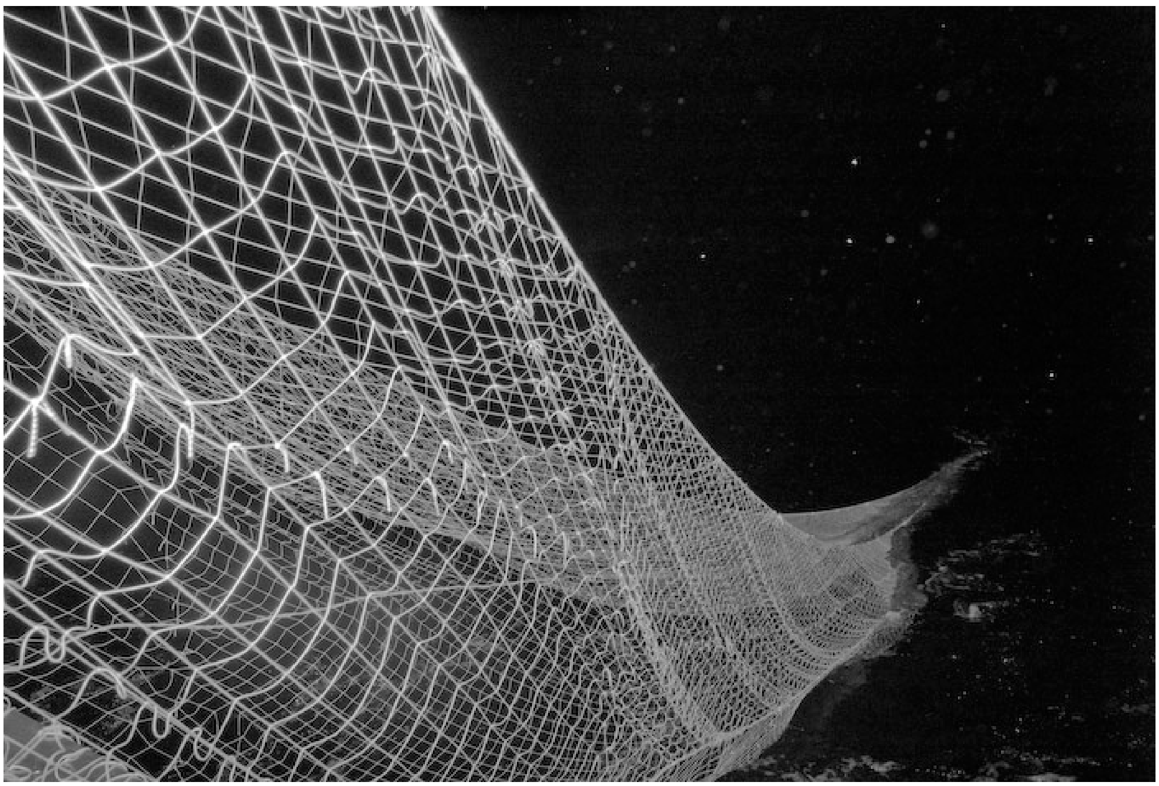

Figure 3. Deployment of the trawl used to sample age-0 herring and other fishes, which was done at night, from the F/V Kyle David in PWS during March 2007. The white dots are reflections from snow flakes of the flash illumination used for this photo.

Zooplankton sampling

A broad scale spring bloom zooplankton cruise was conducted during May. Originally GOA samples were to be collected by the PI and post-doc on a Long-Term Observation Program (LTOP) cruise. However, unlike previous years a UNOLS ocean class vessel was not available for the LTOP cruise. The goals of this project could not be met by participating on the alternate, smaller vessel used instead for the LTOP. In order to address project needs, two days were added to the planned May cruise to collect GOA samples. GOA sampling was constrained by this two-day limit, but met the goal to collect samples from the slope stations of the Seward Line stated in the original DPD. These additional vessel charter costs approximately balanced out with the budgeted travel cost to Seward to participate on the LTOP cruise. Consolidating spring zooplankton sampling to one cruise simplifies logistics and provides direct project control over the sampling. The principal disadvantage is that GOA sampling is weather-limited. It is not be possible to sample in the GOA from a small vessel (58 feet long) unless conditions are ideal. Our success during May 2007 suggests that it is indeed feasible, given good weather.

The sampling design used for GOA sampling during the May cruise is as was stated in the DPD for the late summer cruise, which is still to be done this year. Sampling thus included stations GAK10 to GAK13 of the Seward Line (previously sampled on LTOP cruises) and additional samples needed to cover GOA zooplankton spatial variability. Sampling was done on three transect lines approximately 1 degree longitude apart in order to get a representative isotope 'end-member' for GOA zooplankton. The end-member is the isotope value needed for interpreting SIA of herring. The line south of Montague Island is situated over the submarine canyon shown in Fig. 2. The other two lines are located on the 146 and 148° west longitude meridians. All but two of the GOA stations were situated where the water is much deeper than 300 m.

Figure 4 illustrates the distribution of ring-net samples made during the May cruise. We used a 335 micron mesh, 0.5 m diameter ring net, deployed to 50m, as was done by T. Cooney and R. Thorne in previous studies. These samples are used for species composition analysis. A preliminary analysis consisted of comparing the settled volume to provide an approximation of zooplankton density. Because the net was deployed consistently at all sampling stations the data can be compared directly as shown in Fig. 5. To the extent possible, ring net stations were made at the same locations (i.e., hydrographic stations) as previous projects, especially in PWS. This will facilitate comparison at particular locations with past data.

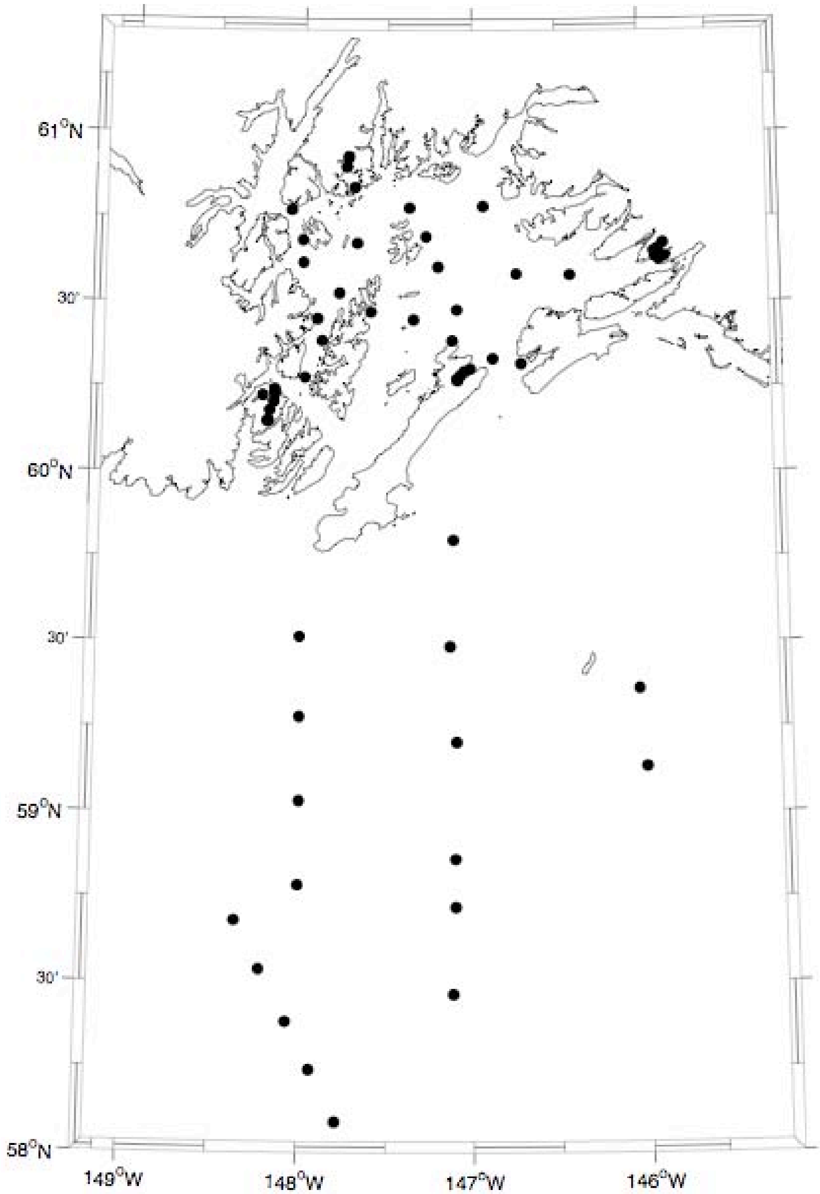

Figure 4. Distribution of ring-net sampling stations made during the May 2007 zooplankton-sampling cruise. At each symbol a 335 micron mesh, 0.5 m diameter ring net was used to sample the upper 50 m of the water column for zooplankton. The south-most station is GAK13 of the Seward Line, which is oriented approximately on a northwest-southeast axis, rather than a north-south axis. Depth at GAK13 is about 2000 m, which is a continental slope depth.

Ring net station settled volumes were stratified into three sampling areas: the four herring nursery bays, PWS other than the four nurseries, and the GOA (Fig. 5). Box and whisker plots of these data illustrate how the four nursery bays are relatively poor habitats in terms of zooplankton forage compared to surrounding waters. The box and whiskers (Fig. 5) suggest that the highest 25 percentile of zooplankton density observed in the herring bays is comparable to the range of densities represented by mean and mode values in the Sound and the lower 25 percentile observed in the GOA. More than half of the zooplankton samples from the four herring bays had zooplankton densities equivalent to the lowest ten percentile from the GOA. The densities observed in the GOA and PWS are highly skewed, reflecting occasionally very dense patches. Zooplankton is about three times as dense in these patches as compared to the mean value of zooplankton in herring bays.

The analysis based upon settled volume should be considered a preliminary analysis; the energetic differences may be influenced by species composition differences among the areas. For example, high-energy content copepods are expected to comprise a larger portion of GOA zooplankton compared to the herring nursery bays. Additionally, this first survey represents the spring bloom. Zooplankton densities are expected to be lower during the late summer cruise.

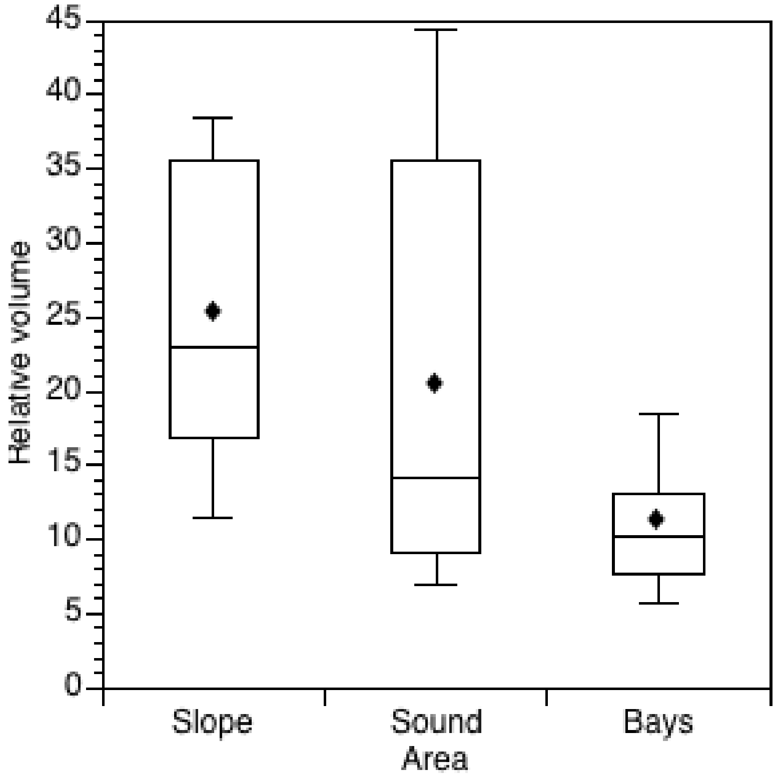

Figure 5. Box and whisker plots of settled volumes data from ring net samples collected during May 2007. The lines cross each box at the 25, 50 and 75th percentile. The upper and lower whiskers indicate 10 and 90th percentiles. The central symbols are the mean values.

Herring are probably not in bay habitats because of the forage that is there. Possibly they are in shallow water to avoid predation by whales and other predators. Nevertheless, two juvenile humpback whales were observed in the innermost section of Zaikof Bay during May, near where herring were surfacing and where we sampled herring during March. Humpback whales were observed near all the herring bays during May except for Whale Bay. For example, there was a calf and another whale, presumably its mother, next to Eaglek Island as we entered Eaglek Bay (forms part of the mouth of Eaglek Bay). Seven whales were sighted in the eastern end of Orca Bay as we approached Simpson bay (Simpson Bay is a tributary of Orca Bay).

Multinet Sampling

A Hydro-Bios Multinet Mini was used to sample the larger volumes of water needed to collect energy and isotope samples. The Multinet consists of 5 electronically controlled nets, which can be remotely activated via a conducting cable from the deck control unit on the surface vessel. The Multinet was deployed horizontally and obliquely (Fig. 6).

Figure 6. Multinet deployment from the F/V Kyle David in PWS during May 2007. One can see one of the white flow meters in the V within the square opening of the sampling device. The other flow meter is on outboard side of the Multinet and not visible in the photograph. The five nets can be seen dangling below, each with a separate bucket or cod-end, which are skating on the water surface in the photograph. The post-doc, Dr. R. Campbell is holding ropes leading to a V-depressor, which is behind the gunwale.

Each Multinet deployment lasted about one hour. Each net was towed until 100 m^3 of water was filtered or there was evidence of net clogging, whichever came first. The volume filtered, which is displayed on the deck unit, is based on data transmitted from flow meters located in (Fig. 6) and out of the mouth of the Multinet. Flow meter data are also used to ascertain if there is net clogging. In general two samples from each Multinet deployment were saved for energy analysis and two samples were saved for species composition. The fifth sample was sorted immediately following retrieval for SIA samples. Energy and SIA samples were frozen whereas the species composition sample was preserved in formalin. Samples are presently being prepared for analyses by freeze and oven drying.

Multinet samples parallel ring net samples so the distribution of Multinet tracks (Fig. 7) is similar to points where the ring net was deployed (Fig. 4). However, the fewer Multinet deployments in PWS and GOA reflect that the Multinet has five nets; there were five samples per deployment. Intensive Multinet sampling was done in the four herring nursery bays. There was at least one deployment in each bay done in the part of the bay where age-0 herring were sampled in March.

Eaglek Bay

Age –0 herring were successfully sampled next to the steep drop-off on the western side of the northern end of Eaglek Bay in March. Accordingly, one Multinet tow was made in the same location (Fig. 8). Additional tows were made along the central axis, north and south of a central shoal.

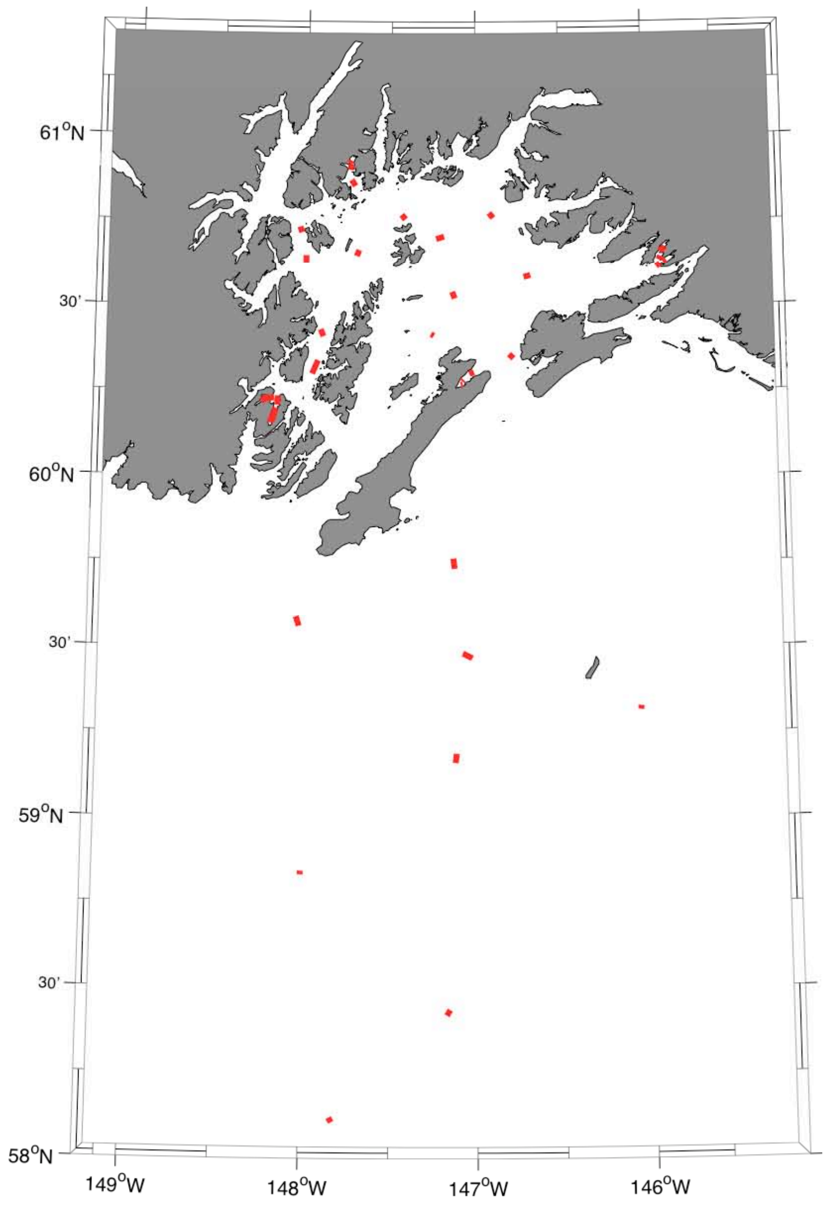

Figure 7. Figure 4. Distribution of Multinet tows made during the May 2007 zooplankton-sampling cruise. Each trace, shown in red, represents a separate deployment. The tows made in the four herring nursery bays are shown in greater detail in the following four figures.

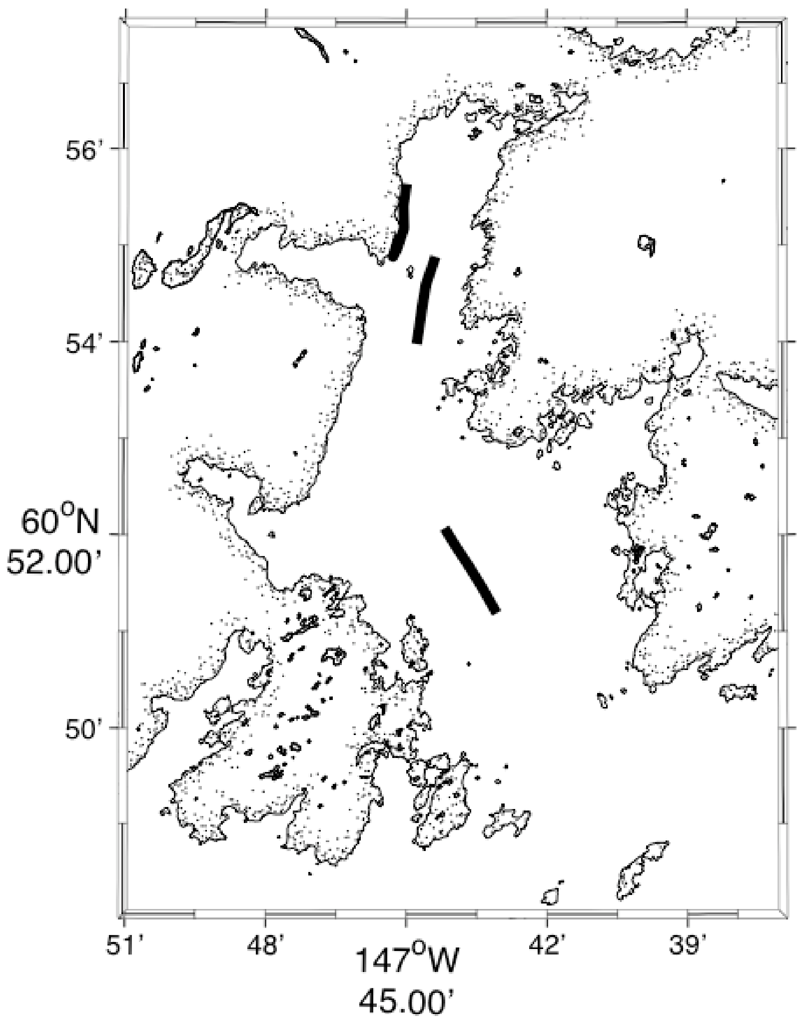

Figure 8. Multinet tows made in Eaglek Bay, May 2007. The track near the shore at the northern end of the bay was where age-0 herring were sampled in March.

Simpson Bay

Age-0 herring were successfully sampled in the northern arm of Simpson Bay in March. Accordingly, two Multinet tows were made in the same location (Fig. 9). Additional tows were made in the relatively deep mouth of the northern arm, in the southern arm, and at the mouth of Simpson Bay. There were a notable number of herring larvae and *Neocalanus* in the net sorted for SIA samples from the tow made at the mouth of the northern arm.

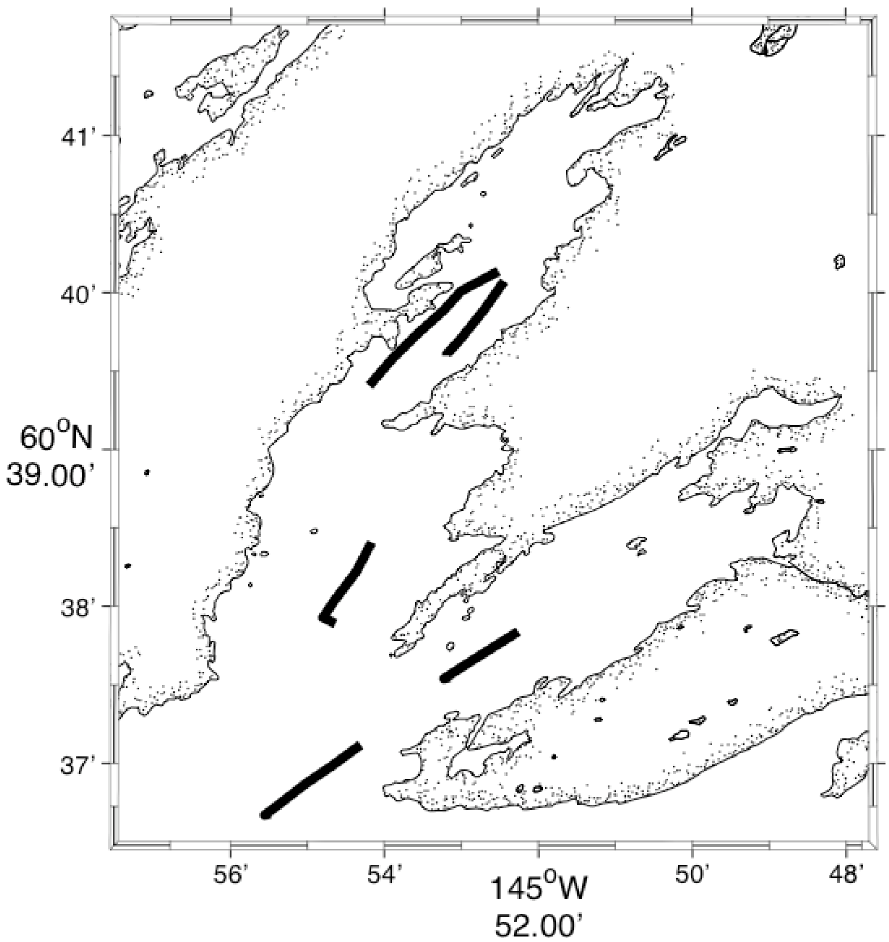

Figure 9. Multinet tows made in Simpson Bay, May 2007. The tracks in the northern arm of the bay reflect where herring were sampled in March.

Whale Bay

Herring were successfully sampled in upper portions of both arms of Whale Bay during March. Accordingly, Multinet tows were made in these locations as well as in the mouth of Whale bay (Fig. 10).

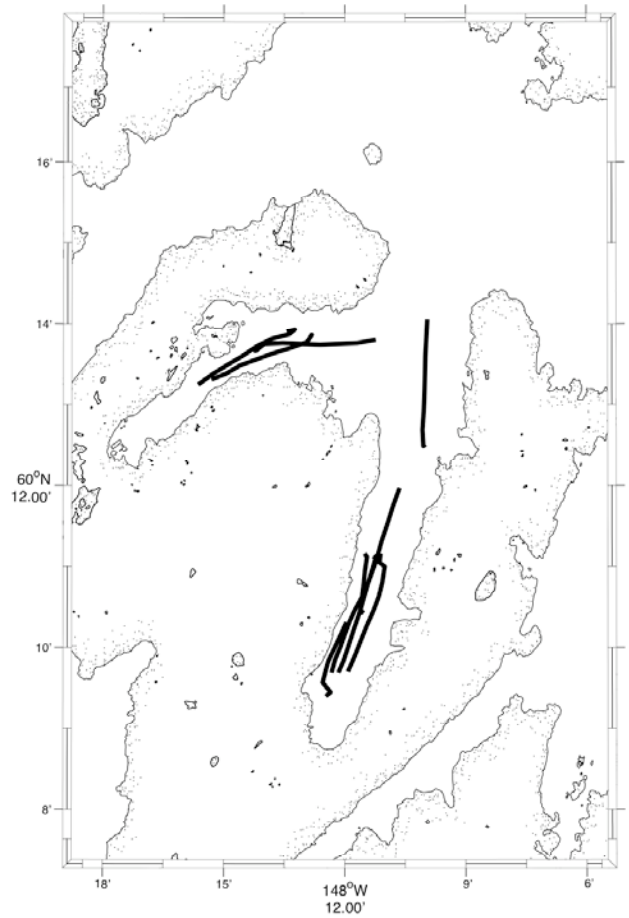

Figure 10. Multinet tows made in Whale Bay, May 2007. Herring were sampled both arms in March.

Zaikof Bay

Age-0 herring were sampled in uppermost portion of Zaikof Bay during March. Accordingly, one Multinet tow was made there as well as locations towards the mouth of Whale bay (Fig. 11).

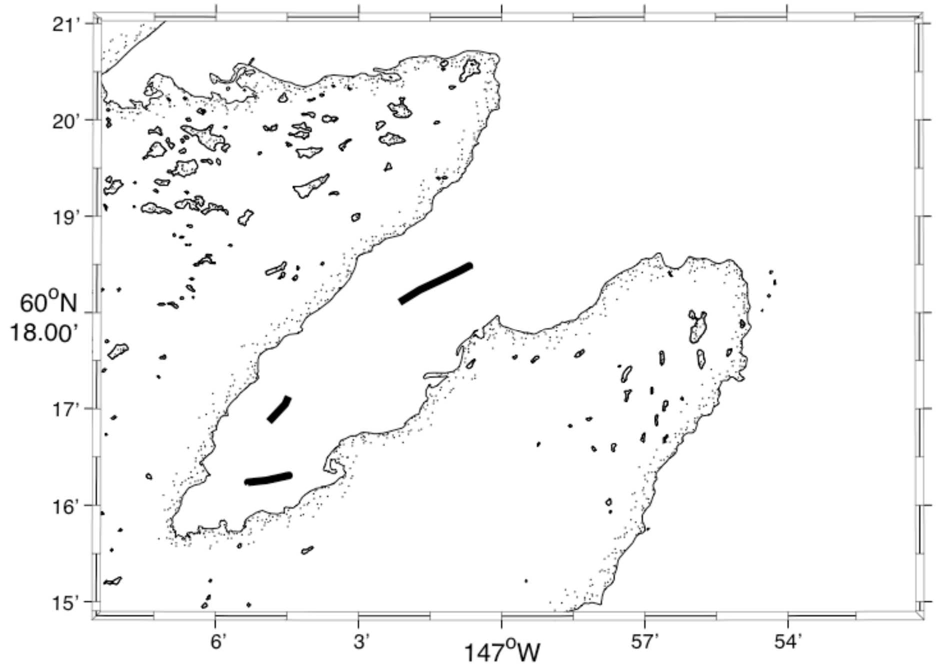

Figure 11. Multinet tows made in Zaikof Bay, May 2007. Age-0 herring were sampled in western-most part of Zaikof Bay in March.

Acoustics transects

Acoustic methods complement physical sampling of zooplankton done with nets. We used an acoustic system consisting of a two-frequency BioSonics DT 4000 deck unit with 6-degree transducers at 120 kHz and 420 kHz that were mounted on a tow-body that was deployed along the side the vessel opposite of the Multinet. The 120 kHz is the primary frequency used in euphausiid assessments whereas the 420 kHz frequency is optimally matched to the large copepods and is commonly used in zooplankton research. Acoustics were deployed during all Multinet tows and while transiting between stations in PWS and the GOA (Fig. 12). An example of acoustics record is shown in Figure 13.

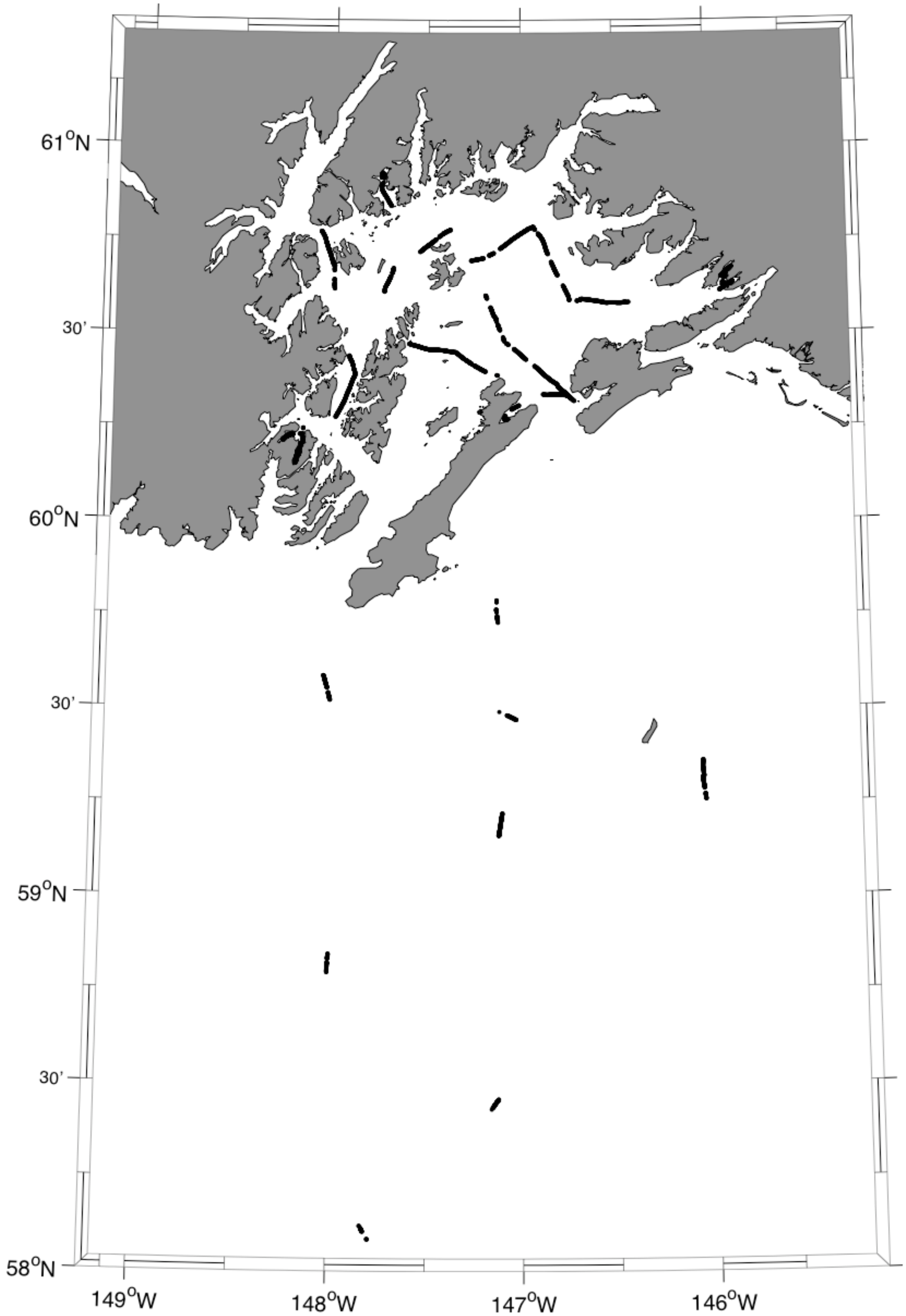

Figure 12. Acoustic transects made during May 2007 cruise.

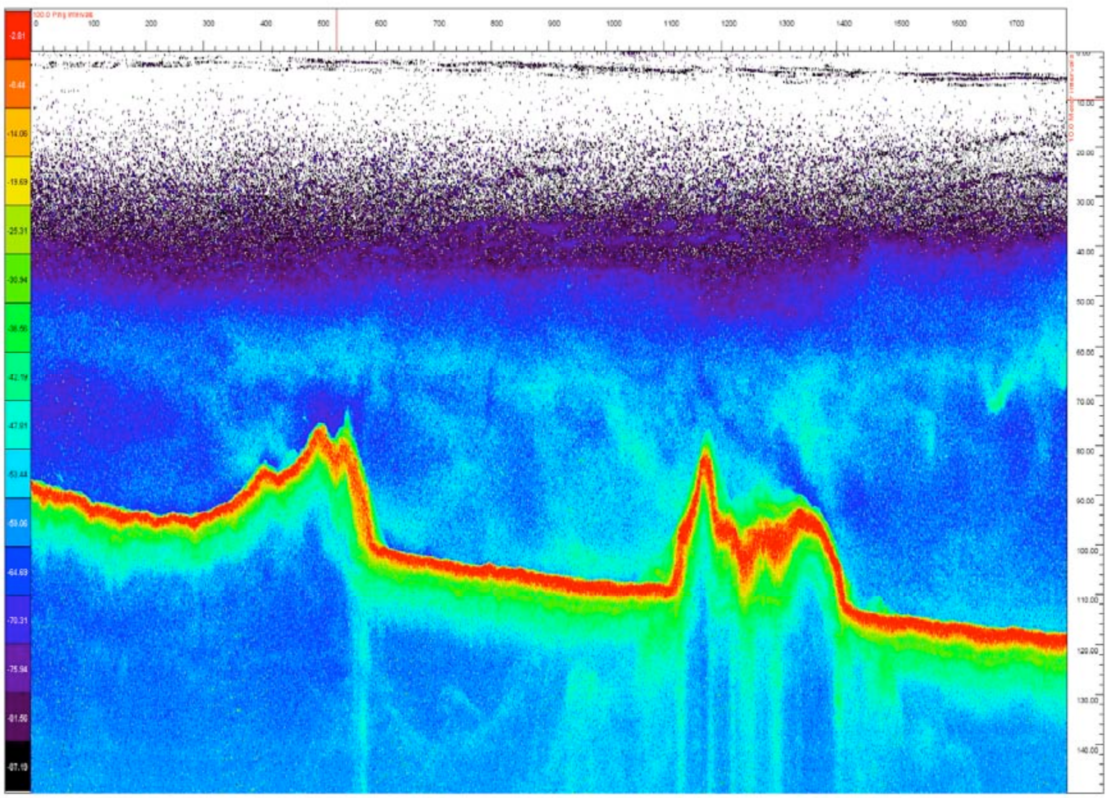

Figure 13. An acoustic record from Whale bay from the 120 kHz transducer showing some water column structure below ~ 40m. The bottom, which has the strong signal (red), while trending deeper going from left to right, has two shoals probably reflecting rocky outcroppings.

PROPOSAL SIGNATURE FORM

By submission of this proposal, I agree to abide by the Trustee Council's data policy (*Trustee Council Data Policy**, adopted July 9, 2002) and reporting requirements (*Procedures for the Preparation and Distribution of Reports***, adopted July 9, 2002).

PROJECT TITLE: Prince William Sound Herring Forage Contingency, submitted under the BAA

Printed Name of P.I. Thomas C. Kline, Jr. Ph. D.

Signature of P.I. ______________________________ Date __________________

* **www.evostc.state.ak.usk/Policies/data.htm**

** **www.evostc.state.ak.us/Policies/Downladables/reportguidelines.pdf**

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Date Received: ________________

PROPOSAL SUMMARY PAGE

Project Title - Prince William Sound Herring Forage Contingency, submitted under the BAA

Project Period: October 1, 2007 – September 30, 2008

Proposer(s): Thomas C. Kline Jr., Ph. D.
Prince William Sound Science Center

Study Location: Prince William Sound and Adjacent Gulf of Alaska
Key Words: Juvenile Herring, Zooplankton, Prince William Sound, Energy, Subsidies

Abstract

Prince William Sound (PWS) herring recruitment is hypothesized to be contingent on young of the year herring attaining from zooplankton sufficient whole body energy content (WBEC) to survive their first winter. PWS recruitment is presently variable, having changed since the Trustee Council funded Sound Ecosystem Assessment (SEA) project ended. Juvenile herring will be sampled and analyzed for WBEC and natural stable isotope abundance (SIA) for comparison with SEA data. The PI has direct familiarity with WBEC and SIA done during SEA enabling duplication. Oceanic subsidies (detected with SIA) are hypothesized to augment zooplankton energy density, which varies in time and locations. High zooplankton energy density is hypothesized to enable herring to acquire high WBEC in certain areas at certain times. To test these hypotheses, herring forage will be assessed in terms species composition and density, SIA, and energy density, which will be related to herring WBEC by location and time.

Funding:	EVOS Funding Received:	FY 07	$ 262.0K	
	EVOS Funding Requested:	**FY 08**	**$ 353.7K**	
		FY 09	$ 167.3K	**TOTAL: $783.0K**
	Non-EVOS Funds Used:	FY 07	$765.0K	
	Non-EVOS Funds to be Used:	FY 08	$250.0K	
		FY 09	$ 0	TOTAL: $1,015K

Date: 25 July, 2007

PROJECT PLAN

NEED FOR THE PROJECT

Statement of Problem

Pacifc herring (*Clupea pallasi*) populations in Prince William Sound, Alaska (PWS) have remained low since crashing in the early 1990's. The 1999-year class performed well, by having a strong recruitment at age three (R. Thorne, PWSSC, pers. comm.). However, this recruitment event was insufficient to restore herring populations to the levels of the 1980's. Overall, recruitment of herring in PWS remains low. The *Exxon Valdez* Oil Spill Trustee Council funded Sound Ecosystem Project (SEA), which made observations during the 1994 to 1998 period, postulated that herring recruitment in PWS is driven by early life history processes (Norcross et al. 2001). This project will address how have these processes may have changed since SEA ended. The project will describe manifestations of this change for PWS herring recruitment in terms of the energy content of herring and zooplankton, the role of oceanic zooplankton subsidies for herring and herring competitors, and the species of zooplankton available as forage.

The SEA project provides a foundation for this investigation. The Principal Investigator (PI) participated as a SEA project leader working closely with other SEA herring PIs. In particular, the PI collaborated closely with A.J. Paul, now retired. We paired our analyses using the same herring samples, exchanged results as well as specimens, and co-published results. An important issue is to determine if conditions in herring nursery habits in PWS have changed since SEA ended in 1998. To make this assessment without systematic error requires close duplication of the sampling design and methods used then so that the results will be comparable. The PI's direct familiarity with the methods used and results obtained during SEA will facilitate making this a successful project. Results of the SEA project, subsequent projects, and other proposed project results will be integrated in the proposed study as described below.

Role of herring whole body energy content (WBEC)

Herring recruitment in PWS is hypothesized to be contingent on forage during their first year. Good forage enables young-of-the-year (YOY) herring to acquire sufficient energy for surviving winter until good forage is available again in spring. Experiments on herring raised in captivity demonstrate that good growth rate depends on forage availability (Hay, 2007). Whole-body energy content (WBEC) of young-of-the-year (YOY) herring at the end of their first growing season in PWS may not be sufficient to last the whole winter (Paul and Paul 1998a). Those that actually survive to March may have less than a month's energy reserve left, which was estimated to be not sufficient to survive until good forage levels resume during the spring bloom (Paul and Paul 1998a). There can be significant spatial and inter-annual variability of WBEC measured during the fall in PWS (Paul and Paul 1998b, Paul et al. 1998). For example, the 6 kJ/g (wet) found in Simpson Bay (eastern PWS) was the highest observed mean WBEC value of YOY herring among the bays compared in October 1995 (Paul and Paul 1999). However, with a mean value of 4 klJ/g (wet), Simpson Bay had the lowest WBEC in October 1996, when the mean WBEC at Whale Bay (southwestern PWS) was about double that (Paul and Paul 1999). A WBEC value of 4 klJ/g (wet) was within the range of starting values in a laboratory study that led to herring mortality (Paul and Paul 1998a). It is likely that a herring with a WBEC of 4 klJ/g (wet) will die during the winter period, being more susceptible to predation, disease, and starvation (Paul and Paul 1998a). Winter survival can be the primary determinant of recruitment (Norcross and Brown 2001).

Since the SEA project ended, herring populations have slowly rebounded but only in certain areas in PWS. R. Thorne (PWSSC, pers. comm.) has observed high recovery rates for the herring that over-winter in northeastern PWS (includes Simpson Bay), where the population increased to over 12,000 metric tons in 2004 from 1,800 metric tons in 2000. Very little recovery has occurred in the Montague Island population. There is evidence that these populations are separate (Norcross et al. 2001, Brown 2003). To understand PWS herring recruitment, it is necessary to make observations on herring systematically at several locations in PWS since any one location is likely not to be representative of PWS as a whole. To facilitate comparison with SEA project data, a minimal sampling design should incorporate the same four bays sampled during that four-year project (see ‘study area’).

Herring form a critical component of the diet of many marine mammals and birds (Hardy 1924; Agler et al. 1999; Matkin et al. 1999; Irons et al. 2000; NAS 2002). Herring predators may be competing for a very limited resource. Herring predators including Steller sea lions, baleen whales (Mysticeti), orcas (*Orcinus orca*), harbor seals (*Phoca vitulina*), and several piscivorous marine birds have declined after the *Exxon Valdez* oil spill (Frost et al. 1999; Matkin et al. 1999). The effects of the *Exxon Valdez* oil spill on populations of several marine birds may have lasted longer than expected because of reduced forage fish abundance including herring (Irons et al 2000).

Recent results suggest that foraging intensity by Steller sea lions (*Eumetopias jubatus*) has had substantial impact on herring mortality (Thorne 2006). Using the dietary requirements from Winship and Trites (2002), in conjunction with forage intensity observations from herring surveys during 2000-2002, Thorne (2006) indicated that winter-period Steller sea lion foraging could remove as much as 12% of the herring biomass. Certain herring populations more susceptible to predation by sea lions and other predators may be more attractive because they are in weaker condition. This hypothesis will be answered from data to be collected on this project synergistically with results of a separate NOAA-funded study where the PI is measuring juvenile herring WBEC near sea lion fall foraging activity (Thorne et al. ‘Steller Sea Lion Winter Food Limitation Research’). By comparing the WBEC of YOY herring from the NOAA project with that being proposed it will be possible to test the hypothesis that sea lions are preferentially feeding on populations of ‘easy prey’, i.e., those with lower WBEC.

During the growing season (spring and summer) the WBEC of juvenile herring increases as a function of body length. The slope of the increase, however, varies by location within PWS and from year to year (Paul and Paul 1998b), suggesting temporal and spatial variability in the ability of YOY herring to forage successfully in PWS. In PWS, herring growth rate, spatial distribution, and WBEC are highly variable (Stokesbury et al. 1999, 2000, Paul and Paul 1999). These SEA project observations were made on year classes of poor recruitment. WBEC observations were not made of 1999 year-class YOY herring. Observations need to be made for years covering a range of WBEC values, and must include years when it is relatively high in the fall, sufficient for over-winter survival, to test the hypothesis that PWS herring is contingent on fall WBEC levels. Analogous measurements to those made during SEA will enable assessing whether growing conditions have changed in the last decade. Therefore, herring WBEC observations will be made in October and March (cf. Paul et al. 1998, Paul and Paul 1999). If growing conditions inferred from WBEC have improved since SEA, in particular in the eastern areas where herring appears to be rebounding, resulting in higher WBEC than during SEA, the expectation is that the slope of WBEC with respect to herring length measured in October will have increased since the SEA project observations reported by Paul and Paul (1999). Presently, the March WBEC values should also be higher compared to those observed during SEA. This could reflect either better fall WBEC conditions or that winter period habitat conditions are such that less energy is lost (cf. Foy and Paul 1999). A herring over-wintering model was developed during SEA to address this issue. The WBEC proximate data required for the over-winter period (Vince Patrick, pers. comm.) will be provided by this project to herring modelers.

Role of zooplankton forage for determining herring WBEC

Relative foraging success on zooplankton can determine herring year class strength because herring need to acquire sufficient energy from their food to survive the long high latitude winter (Blaxter et al. 1963). Herring feed opportunistically on a wide range of zooplankton species (Foy and Paul 1999, Foy and Norcross 1999, 2001). Good herring recruitment may be contingent on the presence of zooplankton populations enabling herring to achieve high fall WBEC values. Zooplankton must be studied to understand this source of WBEC variability. Zooplankton may vary in quality (origin and composition – species and stages of species) and quantity (zooplankton population density) and these aspects may be related to each other.

Oceanic zooplankton subsidies play a key role in the PWS marine ecosystem (Cooney et al. 2001). Oceanic zooplankton is comprised of large calanoid copepods (especially *Neocalanus* spp.) and krill (especially *Euphausia pacifica* and *Thysanoessa* spp.). Subsidies of oceanic zooplankton can be detected in PWS by stable carbon isotope abundance (Kline 1999). For example, > 60% of the variability of marine survival rate of hatchery pink salmon with a history of recruitment co-variation was explained by the relative proportion of oceanic carbon, measured as stable carbon isotope ratio, consumed by early marine stages (Fig. 1, Kline et al. submitted). In late 1995, a strong pulse of oceanic carbon subsidies was detected concurrently using $^{13}C/^{12}C$ in several taxa including herring and pollock juveniles, other forage taxa (fishes and invertebrates), as well as *Neocalanus* copepods resting (diapause stage) in the deep area of PWS (Fig. 1, Kline 1999, 2006). Oceanic subsidies were driven by influx of Gulf of Alaska origin zooplankton, which is based on the diagnostic value of $^{13}C/^{12}C$ values via stable isotope analysis (SIA; Fig. 2, Kline 1999). Oceanic zooplankton subsidies, however, may only be part of story. For example, WBEC of herring in 1995 was generally low although subsidies were high. However, when PWS YOY herring had high WBEC (Paul and Paul 1999), there was an inverse correlation between WBEC and YOY herring $^{13}C/^{12}C$ content (Fig. 2). Herring with low $^{13}C/^{12}C$ values in 1994 were due to incorporation of oceanic carbon. Thus in 1994 oceanic subsidies played a role yielding YOY herring of higher WBEC, when WBEC was above average. We did not measure zooplankton energy densities during SEA. It is possible that oceanic subsidies enabled better WBEC in 1994, if background values were better to start with. If ambient (without subsidies) zooplankton populations were very low in 1995, the relative contribution by subsidies would have been high by default. It is thus important to estimate zooplankton energy as well as source.

Oceanic subsidies may affect herring recruitment in two ways, first by providing more zooplankton food for herring and second, by reducing predation pressure on herring. Certain herring predators such as walleye pollock (*Theragra chalcogramma*) and baleen whales are facultative zooplanktivores. When zooplankton densities are above threshold levels, predators may switch from fish prey to zooplankton sparing juvenile salmon and herring (Willette et al. 1999). The relative proportion of zooplankton size classes will be compared by sampling with three different mesh sizes since YOY herring prefer zooplankton such as that sampled with finer mesh sizes (Sturdevant 2001). Thorne (2006) has observed the importance of specific kinds of zooplankton for salmon survival (Fig. 3). It is thus important to account for taxonomic composition, i.e., species enumeration and density (SED). Density is based upon quantitative sampling. Larger oceanic zooplankton species, such as calanoid copepods are consumed by juvenile herring in the spring (Foy and Norcross 1999, 2001). Oceanic subsidies may be more important for the spring recovery of WBEC.

Zooplankton energy density may be driving herring WBEC. Accumulated energy stores in herring are a result of the balance between that gained through foraging and the losses incurred during the foraging processes as well as metabolic losses. Higher zooplankton energy density will be a better quality food source if herring are able to increase WBEC with lower expenses. Herring WBEC differences within PWS may be explained by differences in zooplankton energy density (ZED) during feeding. Accordingly, ZED will be determined at the same sites where herring will be sampled for WBEC. Herring WBEC will be measured at specific points in time to correspond to those done in the SEA program and to correspond to starting (October) and ending points (March) of the herring over-wintering model, when there is very little zooplankton forage. ZED needs to be determined at times that are more representative of zooplankton abundance and herring feeding on zooplankton. Accordingly, ZED will be determined twice, during the May spring-bloom zooplankton peak and August, which is a mid-point during the initial growth phase of YOY herring (Foy and Norcross 2001). This is an economically driven minimal sampling strategy. Sampling throughout their initial growth as well as their spring recovery would be cost-prohibitive.

Zooplankton sampling will take place on three spatial scales: the central area of PWS, the four SEA bays (Zaikof, Whale, Eaglek, and Simpson Bays), and on a fine scale in eastern PWS. The relatively deep central area is an important zooplankton corridor for, and is located between, the four study bays of SEA and this study (see 'study area'). Standard acoustic transects established in the central area (Thorne 2006) will continue (see 'study area'). The continuity of sampling the four bays used in SEA will enable inter-decadal comparison of WBEC. Comparable methods for ZED, SIA, and SED will enable relating data across these three spatial scales.

The diagnostic value of SIA to infer oceanic subsidies is based on comparing $^{13}C/^{12}C$ values of zooplankton from the continental slope with those in PWS (Fig. 2). A cross-shelf $^{13}C/^{12}C$ gradient has now been observed for 10 years. This was confirmed by the PI systematically sampling on U.S. GLOBEC program long-term observational program (LTOP) cruises (Fig. 2). Systematic SIA sampling previously done LTOP will continue on this project but on a more limited scope compared to that done during GLOBEC in terms of sampling on the Seward Line. LTOP zooplankton sampling, which consisted of the 13 stations comprising the 'Seward Line', from GAK1 to GAK13, will be reduced to the Slope stations GAK10 to GAK13. An 'end-member' is the stable isotope value expected of an organism exclusively dependent on a given source of a given element. The oceanic carbon end-member of, e.g., -23, was based on samples from the continental slope whereas the coastal carbon end-member of, e.g., -19.5, was based on samples from PWS (Fig. 2). End-members need to be derived for both May and August. Continued sampling from GAK10 to GAK13 will enable collecting May end-members exactly as was done on GLOBEC (Kline submitted to Deep-Sea Res. II) furthering the existing time series. Two days of the May and August cruises will be dedicated to sampling out to the continental slope south of PWS to furnish oceanic end-members. PWS end-member data will come from the four bays as well as the central area. Intrinsic to the stable isotope methodology, which is a major focus of the project, is the determination of end-members (e.g., Kline et al. 1990).

Getting the Gulf of Alaska end-member is crucial and a particularly difficult task in the Gulf of Alaska because of the effect of weather on logistics, i.e., the need to sample zooplankton at the continental slope, which is a significant sailing distance from shore.

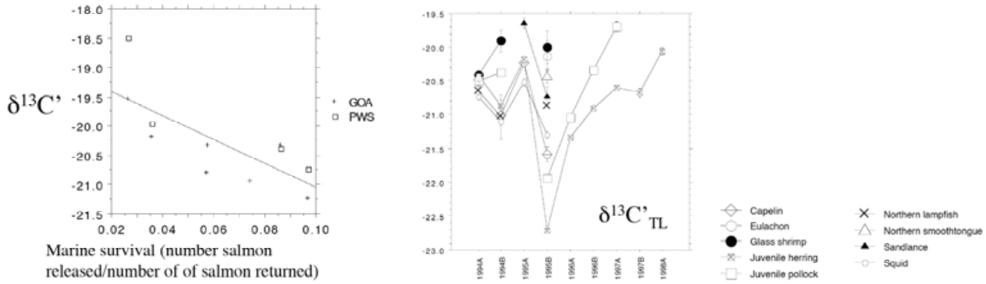

Figure 1. Left. Relationship between stable carbon isotope composition and marine survival rate for PWS hatchery pink salmon (line shows regression for combined sampling areas (R^2 =0.58, symbols by sampling area (R^2 = 0.63 (PWS), R^2 = 0.73 (GOA = Gulf of Alaska)). **Right.** Concomitant shifts in $^{13}C/^{12}C$ among PWS forage trophic level taxa during SEA suggested a large oceanic carbon pulse in late 1995. Years were split so that January through August was 'A' and September through December was 'B.' Figures from Kline et al. (submitted) and Kline 2006.

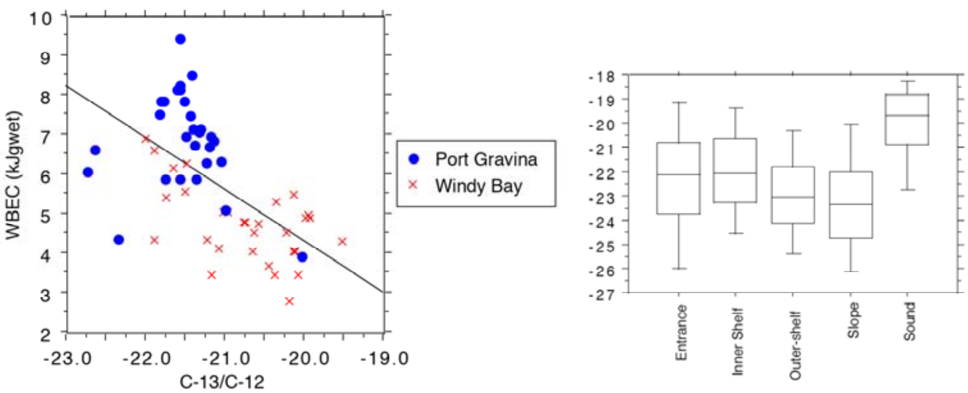

Figure 2. Left. Oceanic subsidies led to higher WBEC. Inverse relationship between $^{13}C/^{12}C$ and WBEC of YOY from fall of 1994 suggested oceanic carbon subsidies increased WBEC and the potential for over-winter survival for Port Gravina herring. In contrast, many if not most herring from Windy Bay were destined to die, R^2 = 0. 37. Paired analyses enabled this graph since both SIA and WBEC were made on the same individual fish. Similar sampling and analysis is proposed here. **Right.** Combined data from GLOBEC years 1998–2004 illustrates diagnostic value of $^{13}C/^{12}C$ data (left axis). Carbon measured in feeding stage *Neocalanus* from slope stations (Slope) was ^{13}C-depleted, had low$^{13}C/^{12}C$ values with a mean of –23, whereas feeding stage *Neocalanus* from PWS (Sound) was ^{13}C-enriched, had high $^{13}C/^{12}C$ with a mean of –19.5. Those from intermediate locations, Hinchinbrook Entrance, and the inner and outer shelf, had intermediate values reflecting mixing of these sources. Data for left panel from Paul and Paul () and Kline (1999); right panel from Kline (submitted).

A novel aspect of this study is determining ZED. Is ZED related to oceanic subsidies? A separate project funding by the Oil Spill Recovery Institute and a private foundation is sampling *Neocalanus* in deep areas of PWS during their resting phase (diapause). Based on their $^{13}C/^{12}C$, a large but variable fraction (can be >> 50%) of the *Neocalanus* diapausing in PWS come from the Gulf and not PWS (Kline 1999). About 90% of the *Neocalanus* from the 1995 year class diapausing in PWS came from the Gulf. This was the same year when herring and other forage taxa received strong Gulf subsidies. This concomitant relationship will be tested using the combined results of both projects. Multiple years of such a relationship would support the hypothesis that oceanic subsidies are a driving force for inter-annual variability in PWS.

Need for time series

By sampling early marine pink salmon over 7 years, encompassing poor to good marine survival rates, it was possible to show a relationship between oceanic food subsidies and marine survival rate (Kline et al. submitted). Accordingly, a long-term goal is to make concerted fisheries-oceanographic observations of herring over a sufficient period in order to find environmental linkages for good, as well as poor herring recruitment and answer the simple question: What does it take in terms of environment to make a good herring year class in PWS? Relationships between oceanic subsidies and zooplankton species composition with salmon survival rate was based on time-series observations. If the SEA program had run for six instead of four years and had ended in the spring of 2000 instead of the spring of 1998, it would have encompassed the first year of the stronger herring recruiting 1999 year class.

This project will address three over-wintering periods, 2006 to 2007, 2007 to 2008 and 2008 to 2009. Herring from the 2006 year will recruit as age 3 fish during 2009. WBEC data will provide an immediate performance parameter to partially overcome the inability to assess recruitment within the period of this project. Although sampling for this project will end in early 2009, the expectation is that it will continue in a second phase, albeit with sampling and analysis improvements, and will eventually lead to a time series analysis so that the question posed above can be answered.

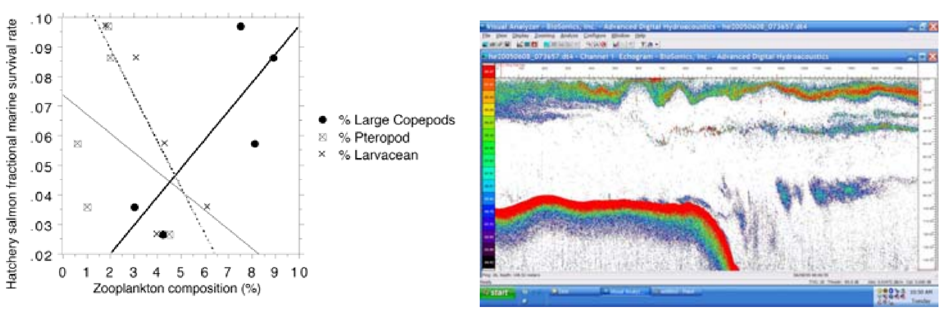

Figure 3. Left. Relationship between taxonomic composition of net plankton and salmon marine survival rate. Marine survival rate = number of adult salmon returning/number of salmon released. The percent of net samples comprised of large copepods, mainly *Neocalanus* spp, was directly correlated with survival $R^2 = 0.66$ (heavy line) whereas the percent composition of larvaceans (broken line) and pteropods (thin line) was inversely correlated with marine survival, $R^2 = 0.64$ and 0.10, respectively. These results illustrate how zooplankton composition may be as important as zooplankton abundance. **Right.** An example of an acoustic echogram collected by Thorne (2006) showing a strong zooplankton layer at ~ 10m depth, ranging from near the surface to ~ 20m. The lower layers are fish (Thorne pers. comm.). Acoustical data such as this will enable us to tow the Multinet horizontally through a layer with net openings and net closings conducted so as to sample particular features. Data and echogram figure from Thorne (2006).

Relevance to 1994 Restoration Plan Goals and Scientific Priorities

Herring remains in the non-recovered status. There is growing information on the culpability of EVOS in its decline, as well as indirect damage to the PWS ecosystem as a result of the herring collapse. PWSSC research shows that herring are an important winter-period food supply for at least three of the five other remaining non-recovered resources, including cormorants. The PWS herring crash is also implicated in the decline of the endangered western stock of Steller sea lions.

The effort proposed herein is relevant to most of the 8 categories for herring proposals outlined in the Invitation. It is most directly an oceanography effort, but the information is important for planning, modeling, predation and intervention. Oceanic subsidy assessment, WBEC and proximate data as part of the program would be useful to research in other areas such as modeling.

PROJECT DESIGN

Objectives

This is a three-year project that is designed to fill the information gaps on critical stage/seasonal data as outlined in Tables 1 and 2 in the QA-QC section. Listed within are objectives in terms 'cruise tasks' (Table 1) and sample particulars (Table 2).

Procedural and Scientific Methods

This project addresses the role of herring forage as a determinant of herring WBEC by leveraging existing studies and complementing other proposed studies. These other projects are focused on other ecosystem questions in the same time and space context. This project will provide support to these other projects through the zooplankton and herring analyses and by sharing vessel time. This project will enable a more complete picture by synergizing with an existing program that is specifically addressing herring as sea-lion forage. The same analyses are being performed by the PI.

This project deals with analysis of two trophic levels of the PWS ecosystem. One trophic level is the 'forage fish' trophic level of herring and similar trophic level fish competitors (Kline 2006, 2007). They are often referred to as forage fish since many organisms, mammals and birds in particular, eat them. The other trophic level is that of zooplankton, which are the forage of zooplanktivores such as herring and other forage fish. This project will perform three types of analysis: natural stable isotope analysis (SIA), energy content, and species identification. SIA will be performed on individual fish and selected individual zooplankters to match what was done on previous studies (e.g., Kline 1999). SIA will also be done on bulk (the contents of a given net sample comprising a mixture a species and individuals) zooplankton samples collected quantitatively from layers in the water column in order to be matched to zooplankton energy density (ZED) measurements that will be performed on the same sample. Bulk sampling will ensure there is sufficient material to perform both analyses. Energy content will be determined on whole herring to be comparable with whole-body energy content (WBEC) determinations resulting from the SEA program. As was done during the SEA program, WBEC and SIA will be performed on the same individual fish.

Herring and other fish sampling

Herring and other fish sampling will be performed in a separate proposal titled "Trends in adult and juvenile herring distribution and abundance in PWS," PI Thorne. The PI and technician will participate, assist with sampling while collecting fish samples for WBEC and SIA on Thorne's herring cruises in March and October. Thorne will be locating herring schools acoustically which then be sampled with nets. Sub-samples of catches of herring and other forage fish taxa caught incidentally (e.g., pollock; Kline, 2007) will be saved (frozen). Freezing was also the method used during SEA. Fish will be thawed in the laboratory and weighed. Herring otoliths will be extracted and sent to UAF (to Brenda Norcross and Nate Bickford). Their primary use of the otolith will be to determine the particular bay of origin of a given herring using geochemical techniques. Otoliths were also removed during SEA. The herring will be oven dried (60°C) to a constant weight (same technique as SEA). Dried sub-samples from each fish will be assessed for whole body energy content (WBEC) and stable isotope analysis (SIA) using mass spectrometry. Ten percent of the herring will also have WBEC determined using bomb calorimetry to validate the WBEC model described below. This will be more economical then doing bomb calorimetry on all samples, since relatively few, about 10 to 12 bomb calorimetric determinations, can be completed in an 8-hour day.

Zooplankton sampling

Zooplankton will be sampled in the four SEA bays (Zaikof, Whale, Eaglek, and Simpson Bays) and in the open area using two types of plankton nets, a Hydro-Bios Multinet Mini and ring nets. The Multinet is an electronically controlled multiple-net sampling device that can be towed vertically, horizontally, and obliquely. It has five nets that are deployed sequentially effecting vertically and horizontally stratified samples, according to how the net is towed. Nets are triggered by remote control via conducting cable or can be pre-programmed to specific depths (our Multinet can operate by both methods). Stratified horizontal tows will enable sampling horizontal features in the zooplankton distribution (Fig. 3). It has internal and external flow meters to determine the sample volumes. Nets are interchangeable. We have three mesh sizes, 150, 335 and 500 microns, respectively corresponding to the best size needed to collect herring forage (Sturdevant 2001), the standard mesh size used during SEA (Kline 1999), and the standard mesh size used during GLOBEC to sample meso-zooplankton (Coyle and Pinchuk 2005). Additionally we have ring nets of 0.5 and 1.0-meter diameter of 335 and 500 micron mesh nets equipped with flow meters that will be used to collect larger and rarer taxa needing greater sample volumes. The Multinet will be deployed horizontally at depths at which zooplankton layers are detected using hydro-acoustics. Ring nets will be towed vertically.

The Multinet unit is equipped with pressure (used to calculate depth), temperature, conductivity (used to calculate salinity), chlorophyll fluorescence, and dissolved oxygen sensors. It is effectively a plankton net combined with a CTD. The CTD capability will be used to assess relationships between zooplankton layers and other parameters, in particular where the phytoplankton is in relationship to the zooplankton. This will be useful for anticipated modeling activities to be done on other projects.

We will use acoustics to find zooplankton layers and then will deploy the Multinet in these layers. The acoustics will be run simultaneously so that the zooplankton abundance data can be used for calibration. Finite samples will be collected in the layers using the three mesh sizes. The fifth net of the Multinet must remain open and so will sample the water column from the layer to the surface. It will be used to sample live materials for sorting for SIA and for experiments. The other four nets of each Multinet deployment will yield four ‘closed’ (closed within zooplankton layers) samples, which will either be frozen (for combined SIA and ZED analysis) or preserved in formaldehyde (for identification and enumeration). The sample volume of the four closed samples will be known from the Multinet software, which uses the two flow meters. This sample volume will be the numerator for calculated ZED and zooplankton abundance per unit volume within zooplankton layers. This information will be provided to the acoustics PIs (collaborators Thorne and Crawford) so they can extrapolate the data to local area (fine scale project), bay, and Sound-wide according to acoustics data.

Bulk frozen zooplankton samples will be thawed, a wet weight determined, then oven dried (60°C) to a constant dry weight. Sub-samples of each dry zooplankton sample will be combusted in a Parr plain oxygen bomb calorimeter model 1341. Energy content will be reported as KJ/g wet and dry. The energy content of the sub-sample will be extrapolated to the whole sample and to in-situ ZED (KJm^{-3}) values using the sample volume. SIA will be performed on another sub-sample. SIA methods are described below.

Individual zooplankters will be sampled during cruises, sorted to species and stage (under microscopic examination) and preserved individually frozen in vials as described by Kline 1999.

The zooplankton samples preserved in formalin will be analyzed in the laboratory. Analyses will include enumeration after identification to species and stage based on microscopic examination. Sample volumes will be used to extrapolate to number, by species, per unit volume (m^3) in the zooplankton layers like the ZED data. These data will be furnished to acoustics PIs (collaborators Thorne and Crawford) so they can extrapolate the data to local area (fine scale project), bay, and Sound-wide according to their acoustics data.

SIA and WBEC

From the analysis of a single age-0 herring we will simultaneously determine whole-body energy content (WBEC) content and the carbon and nitrogen stable isotope composition via stable isotope analysis (SIA). WBEC predicted by a model using data from SIA (SIA protocols described in QA-QC section) compared favorably with that measured directly with a bomb calorimeter (Fig. 4). The advantage here is that one analysis will reduce costs and will serve a dual purpose. WBEC will be a by-product of SIA at no additional cost.

The energy content of bulk zooplankton samples will be determined using a Parr oxygen bomb calorimeter. Zooplankton energy density (ZED) will be calculated from the energy content of bulk zooplankton samples and the in situ zooplankton density. We will measure the wet and dry weight of bulk zooplankton samples from the Multinet for three mesh sizes, 150, 335, and 500 microns. Zooplankton density will be calculated by dividing these weights (dry and wet for each mesh size) with the volume sampled (from the Multinet's flowmeter). For each bulk sample, the ZED will be calculated by dividing the bulk energy content by the in situ zooplankton density. This will be done for each mesh size. The ZED values for each mesh size will be matched with acoustics data (projects of Crawford and Thorne) to extrapolate ZED values the zooplankton populations at each site (bay or transect according to survey type) to assess zooplankton forage energy.

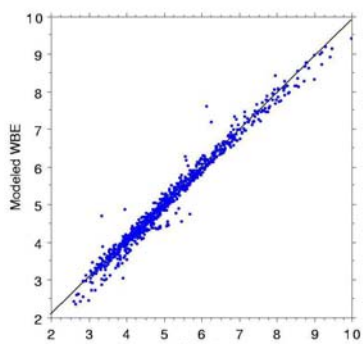

Figure 4. Modeled WBEC versus (vertical axis) measured WBEC (horizontal) content of juvenile herring from PWS sampled during the SEA project (units are kilo-Joules per gram wet mass). The close fit to the 45° line indicates that the model provides a good estimate of WBEC. The WBEC was measured using a bomb calorimeter which is being compared to modeled values based on C/N ratio and wet-dry ratio ($R^2 = 0.98$; $P < 0.01$); N = 833).

Cruises and Sampling

Cruises

Cruise are designated herring or zooplankton according to their primary sampling goal (Table 1 in QA-QC section). Herring cruises will be funded on a separate project (Thorne PI). There will be two juvenile herring sampling cruises per calendar year, one in March and one October. Two zooplankton cruises per calendar year will be funded by this project, one in May and one in August. Which bays will be sampled first, second and so forth will be weather determined. This is an absolute requirement based on the PI's more than 25 years of aquatic research in Alaska. The weather is the limiting factor! Table 1 also lists existing cruise funded by NOAA and the time course to early 2009.

The rationale for sample timing and geographic scope of cruises was given earlier. These rationales and tasks are listed in Table 1 (QA-QC section). The length of cruises is given in the lower block along with the funding source. Because this project will only be funding 22 days per year (see row labeled 'Kline – this project') this project will benefit substantially from cruises funded by other projects, from various sources, in collaboration with other PI's listed in this block.

Study area

The study encompasses PWS but with two foci, the 'four bays' studied during the SEA program and central Sound 'open areas' that have been monitored for zooplankton using acoustics since 2000 (Fig. 5), as well as the continental slope south of PWS. This sampling will provide continuity with previous research enabling systematic across-time comparisons with similar methodology.

The 'four bays'

During the SEA program four bays were selected and sampled for herring over the four-year course of the observational program (Norcross et al. 2001; Fig. 5). These are Whale Bay, in southwestern PWS; Eaglek bay in northwestern PWS, Zaikof Bay, west side of Hinchinbrook Entrance in southern PWS; and Simpson bay in eastern PWS. These bays should not be considered as replicates but instead as four rather different bays with many differences in geomorphology and oceanography (Gay and Vaughan 2001). It was necessary and will continue to be necessary to sample in four disparate bays such as these to encompass the range in habitat found in PWS, given that there were significant differences found in properties of herring from them (Norcross et al. 2001).

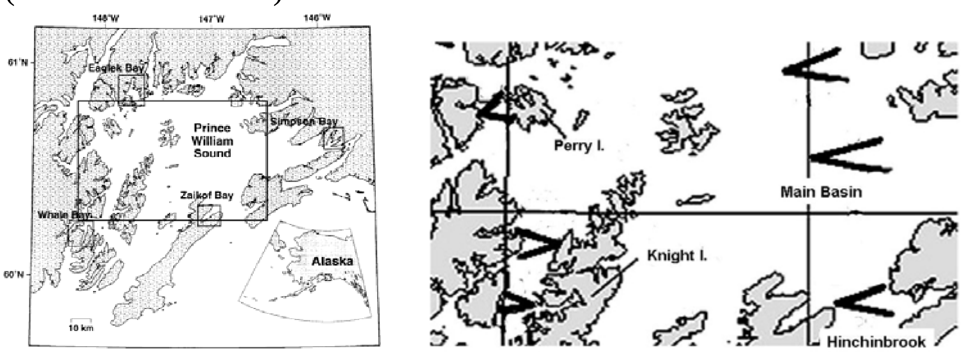

Figure 5 Left. The PWS study showing the four SEA bays and central area expounded in right panel. **Right.** Six standardized zooplankton acoustic transects from Thorne (2006).

The 'open areas'

The open areas in central PWS have been routinely sampled for zooplankton using six standardized acoustic transects shown in Figure 5. These areas are important for conveyance of plankton into the four bays from the Gulf and PWS. Sampling here will maintain continuity with a previous monitoring project, which is ending in 2006. Transects are located in deep water, which is important as zooplankton habit, especially for species that migrate vertically, either daily or as part of their life history (e.g., *Neocalanus*). The four bays are tributaries of this central area. The eastern end of PWS, Orca bay, and its tributary bays such as Simpson bay, is shallower than 200mm. Transects in the main basin are the nearest deepwater areas to Orca Bay.

Coordination and Collaboration with Other Efforts

This project will be coordinated and managed within the larger Prince William Sound Science Center program on juvenile herring research. There will be collaborators from outside the institution including, Alaska Department of Fish and Game, the University of Alaska (UAF), and the Prince William Sound Fisheries Research Applications Group. Samples and data from this project can be made be available for other uses including disease, marking and modeling. Prince William Sound Science Center program on juvenile herring research investigators leading separate projects include Dr. R. Thorne ("Trends in adult and juvenile herring distribution and abundance in PWS"), Dr. M. Bishop ("Sea bird predation on juvenile herring"), and Doctoral candidate S. Gay ("Physical oceanographic factors affecting productivity in juvenile pacific herring nursery habitats"). This project will support these projects by providing data and results described in the proposal. Dr. Thorne's project will provide vessel and sampling support for this project.

There will effectively be a quid pro quo arrangement with the projects being proposed by Thorne to the same announcement. This project will be providing WBEC and SIA data on herring for Thorne's herring cruises. Combining cruise-days for both projects will optimize our ability to use weather windows for sampling more challenging sites (Gulf, central Sound, and Zaikof Bay – based on experience).

SCHEDULE

Project Milestones

The study will be conducted over a three-year period. The proposed starting date is October 1, 2006. The provisional cruise schedule is detailed in Table 1. It includes five juvenile herring surveys in PWS and four zooplankton surveys in PWS and the adjacent Gulf (see Table 1, QA-QC section). One juvenile survey will added during March of each year of the project, two in years one and two. All herring survey vessel charter time is funded on other projects (NOAA funded) or proposed projects (EVOS funded). Zooplankton surveys will be added in May and August of each year in years one and two. Zooplankton surveys will be of multiple legs comprised of cruises funded by separate projects. This project will be funding the vessel charter for one cruise each May and August each year for a total of four cruises over the duration of the project.

Measurable Project Tasks

FY08 1st Quarter (October 1, 07 to December 31, 07)

November	Complete juvenile herring survey #2
December	Complete analysis of juvenile herring survey #1 (March 2007)
December	Complete analysis of zooplankton survey #1 (March 2007)
December	Complete analysis of zooplankton survey #2 (September 2007)

FY08 2nd Quarter (January 1, 08 to March 31, 08)
January		Annual Marine Science Symposium
March		Complete juvenile herring survey #3

FY08 3rd Quarter (April 1, 08 to June 30, 08)
May		Complete zooplankton survey #3
June		Complete analysis of juvenile herring survey #2

FY08 4th Quarter (July 1, 08 to September 30, 08)
August		Complete zooplankton survey #4
August		Submit Annual Report

FY09 1st Quarter (October 1, 08 to December 31, 08)
November		Complete juvenile herring survey #4
December		Complete analysis of juvenile herring survey #3
December		Complete analysis of zooplankton survey #3
December		Complete analysis of zooplankton survey #4

FY09 2nd Quarter (January 1, 09 to March 31, 09)
January		Annual Marine Science Symposium
February		Complete analysis of juvenile herring survey #4
March		Complete juvenile herring survey #5

FY09 3rd Quarter (April 1, 09 to June 30, 09)
June		Complete analysis of juvenile survey #5

FY09 4th Quarter (July 1, 09 to September 30, 09
September		Submit Final Report, which will consist of a draft manuscript for publication, to the Trustee Council Office

RESPONSIVENESS TO KEY TRUSTEE COUNCIL STRATEGIES

Community Involvement and Traditional Ecological Knowledge (TEK)

This project is community based. It will be conducted out of the Prince William Sound Science Center (PWSSC) in Cordova, a community-based research and education center. The PWSSC is an existing community asset, since it consists of scientists and interested lay people and is in a community that depended on herring. We will employ local fishing vessels to do sampling. We will use their local knowledge of weather patterns.

The PI is a member of the Cordova-based PWS community research planning effort being led by Ross Mullins and Ken Adams (funded by the EVOS Trustee Council). The PI will continue to participate with this planning group and will exchange findings of the project as they develop.

Other community involvement consists of participation through the PWSSC education program. At the PWSSC, the PI has conducted well-received plankton exercises where students sample plankton with a small net in the Cordova harbor. They are then shown their own plankton collection and a variety of plankton from collections made on research cruises through microscopes. The PI also provides guest lectures and class exercises for the Cordova High School.

Resource Management Applications

Information from this project can incorporated into management models that are used to make decisions on herring fisheries management. The addition of juvenile herring information provides a potential forecast of recruitment. We may be able to advise best how and where to enhance populations through intervention from our assessment of the bays as good rearing habitat. The data will be made available to modelers of PWS herring (see modeling references in body of proposal).

PUBLICATIONS AND REPORTS

No costs for publications are specifically requested in this proposal beyond those for annual and final reports.

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Resume

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Education

1991	Ph.D. in Oceanography, University of Alaska, Fairbanks
1983	M.S. in Fisheries, University of Washington, Seattle
1979	B.S. in Fisheries, University of Washington, Seattle
1976	B.S. in Oceanography, University of Washington, Seattle
1972-74	Coursework at Sophia University, Tokyo

Research Accomplishments

- Dr. T. Kline developed natural stable isotope abundance techniques that provided the first direct evidence of the significant role of anadromous-salmon-marine-derived nutrients in freshwater ecosystems
- Dr. T. Kline developed natural stable isotope abundance techniques for detecting amphidromous fish migrations on the Alaska North Slope
- Dr. T. Kline developed natural stable isotope abundance techniques for providing evidence that production derived in the Gulf of Alaska plays a significant role subsidizing Prince William Sound food webs
- Dr. T. Kline discovered the existence of large inter-annual variations in natural stable isotope abundance in the Gulf of Alaska and their probable cause by meso-scale eddies
- Dr. T. Kline determined that oceanic subsidies enhance the marine survival rate of Prince William Sound pink salmon populations

Professional Appointments

1994-2006	Research Scientist, Prince William Sound Science Center
1995-2006	Director, Prince William Sound Science Center Scientific Diving Program
1995-2006	Diving Safety Officer, Prince William Sound Science Center Scientific Diving Program
1992-93	Instructor, University of Alaska Fairbanks
1991-94	Postdoctoral Fellow, University of Alaska Fairbanks
1985-91	Research Assistant, University of Alaska Fairbanks
1984-85	Teaching Assistant, University of Washington
1977-83	Research Assistant, University of Washington

Five Related Recent Research Papers (all based on EVOS Trustee Council funded projects)

2007 Kline, T.C. Jr. Ontogenetic, temporal, and spatial variation of feeding niche in an unexploited population of walleye pollock (*Theragra chalcogramma*). *In:* Resiliency of Gadid Stocks to Fishing and Climate Change University of Alaska Sea Grant, Fairbanks. ACCEPTED MS.

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Recent Collaborators

Coyle, K., Cooney, R., Haldorson, L., Hopcroft, Weingartner, T., Whitledge, T. (Univ. Alaska Fairbanks); Bishop, M., Schoch, C., Thorne, R. (P.W.S. Science Center); Moffitt, S. (Alaska Dept. Fish and Game); Knudsen, E., Woody, C. (U.S.G.S.); Cheng, L. (Univ. Calif. San Diego); Boldt, J., Hermann, A., Hinckley, S. (NOAA)

BUDGET JUSTIFICATION

This project is essentially like a combination of two separate sub-projects of the SEA project and a separate isotope project. These were project 320I and 311, which were led by PI Kline, and project 320U, which was led by PI Paul. The collective budget of these prior projects was about $2million. The proposed project has additional goals that were not part of either 311, 320I or 320U, that of zooplankton species enumeration and their density and that of zooplankton energy density. This is thus a very economical budget.

Personnel – Total $377.5.
The employment period is annual, therefore salary is shown on budget forms under "calendar months." Salaries requested are consistent with the PWSSC's regular practices.
Three project personnel will be required to perform tasks during zooplankton sampling cruises – a winch operator, a deck unit operator and a primary net handler. Once the deck unit is shut off during a net recovery, the computer operator will assist the primary net handler in bringing the net onto the vessel. We will require that vessel personnel operate the vessel's boom. Personnel will consist of the P.I. (part time), one post-doc, and a technician. The P.I. will need to dedicate about nine months per year to PWS herring research in order to successfully integrate across the three disciplines of isotope chemistry, fish biology, and zooplankton biology, all of which are in the P.I.'s background. The portion of the P.I.'s time not being funded directly by this project will be being subsidized by other sources as described in the proposal. The Post-doc will be entirely subsidized by other sources during year 1. The project is labor intensive in both the field and in the lab. There will be considerable handling of individual specimens, which takes up time. Hence personnel costs contribute significantly to the total project costs. The post-doc will serve as co-PI, will lead zooplankton taxonomic analysis, assist in sampling and lead the laboratory processing of samples in preparation for SIA. The technician will assist in both the field and laboratory. The P.I. will lead field sampling, perform data interpretation, supervise other personnel, and perform administrative functions. Actual sampling of the target bays is expected to take 8 of the 16 days of each cruise. All project personnel (3) are needed for the zooplankton cruises to deploy the Multinet (105 kg) so there was no reduction for those cruises. Entire zooplankton cruises will be devoted to sampling for this project.
Fringe Benefits – Fringe benefits are incorporated into the average monthly cost. It is the PWSSC's usual accounting practice that contributions to employee benefits (social security, retirement, etc.) are treated as direct costs. Workman's compensation for anticipated sea-days varies per year according to the anticipated number of day on cruises. The PI's average monthly rate is less in FY09 because of the reduced number of sea-days (only the March cruise in 2009).
Travel – Total $8.6K
Travel Funds are being requested for attendance at the annual January Alaska Marine Science Symposium.
Contractual – Total $138.5K
A major cost in the contractural category is the actual mass spectrometric (isotopic) analysis of the samples which is outsourced (see QA-QC section). We will contract a local vessel (28 days per year), cost is based upon recent experience with the type of vessel needed. A vessel of ~ 20m length will be needed to provide enough deck space for equipment. We also will need to be able to bunk four scientists. Shipping costs anticipate having to pay hazardous material (a.k.a. hazmat) delivery charges for oxygen, benzoic acid, and formalin. The PWSSC presently levies a $100 per person-month network charge. In order to reduce cruise time, there will be one round-trip small airplane charter per juvenile herring cruise. We will need to get another ride per cruise from one of the other projects also rotating people and will provide space for them in exchange (two R/Ts anticipated per cruise).
Equipment & Commodities- Total $41.1 (all items under $5K)
Major laboratory supplies each year include expendables such as micro-centrifuge tubes, LSC vials, plankton jars, chemicals (e.g., formalin), sample bags, a spare parts kit, benzoic acid pellets, ignition wire and oxygen for the Parr bomb.

We will use the old deep-sea (~ 800m) winch bought on the SEA project. A new one will be purchased only if needed.
Each year we anticipate having to replace plankton nets (total of 3 at $1.05K), plankton net buckets (3 at 1.6K), the ‘canvas part’ (one at $1K), and one of the impellers of the Multinet, replacement rate, based on experience. Annual maintenance also requires buying a Multinet maintenance kit ($2.9K). Each year we anticipate having to replacement 1 ring-net ($0.5K), also based on experience.

Funds from non-EVOS sources, including in-kind contributions: Private foundation grant (M.J. Murdock) provides support for post-doc position during Year 1 only; this grant also bought the HydroBios Multinet ($80K) and provides some lab analysis and vessel charter support. Total Murdock funding is $260K (FY 07). NOAA is funding the Steller sea lion project (Thorne lead PI) that will provide comparison data as described in the proposal. This project is funded at $350K in FY 07 and $250K in FY 08. The Oil Spill Recovery Institute is contributing $40K in FY 07 to support sampling the diapausing *Neocalanus* project funded through Murdock, which will be compared to herring as described in the proposal. The PWS Ocean Observing System is contributing one month of PI time so that data from this project can be incorporated into the Observing System (FY 07 contribution about 15K). Additional support from the PWSOS in FY08 is probable but not yet appropriated. Non-EVOS funds thus far amount to $765K in FY 07 and $250K in FY 08, for project total of $1,015K.

Our federal cognizant agency is NOAA. Future estimates of the negotiated rate were provided by the contracting officer, P. Oswalt, who should be contacted for more details.

DATA MANAGEMENT QA/QC

1. Study design, sample type(s) and location requirements, statistical analyses, types and numbers of physical samples.

<u>A Sample type.</u>

1. Fish (frozen – for stable isotope analysis (SIA) and whole body energy content (WBEC) analysis)
2. Zooplankton (frozen – for SIA and zooplankton energy density (ZED))
3. Zooplankton (preserved in formalin for identification to species and stage, enumeration and density (SED))

<u>B Locations.</u>

The area study encompasses Prince William Sound but with two foci, the 'four bays' studied during the SEA program and central Sound 'open areas' that have been monitored for zooplankton using acoustics since 2000 (Fig. 1 in main body of proposal), as well as the continental slope south of PWS. This sampling will provide continuity with previous research enabling systematic across-time comparisons with similar methodology.

Table 1. Outline of cruise schedule.

Timing	March	Early May	Mid-May	August	October
2006	existing	existing			existing
2007	proposed	proposed	proposed	proposed	existing
2008	proposed	proposed	proposed	proposed	proposed
2009	proposed				
Rationale	Herring 0-age post-winter	LTOP spring Bloom SEWARD LINE	Spring zooplk bloom PWS	Herring YOY growth	Herring YOY pre-winter
Designation	Herring	Zooplankton	Zooplankton	Zooplankton	Herring
GLOBEC time series		X		X	
SEA comparison time series	X	X	X		X
Task					
Adult Herring acoustics	Thorne				
Zooplankton acousitcs		Coyle	Post-doc	Post-doc	
Zooplankton species		Coyle	X	X	
Zooplankton SIA		X	X	X	
Zooplankton energy		X	X	X	
Juv. Herring acoustics	Thorne				Thorne
Juv. Herring SIA & WBEC (fish capture)	X				X
Herring competitors SIA (fish capture)	X		June-ADFG		X
Avian aggregation	Bishop				Bishop
Geographic scope					
Sound-wide survey	X (NOAA)		X (new)	X (new)	X (NOAA)
Four fjords	X (new)		X (new)	X (new)	X (new)
GOA shelf & slope		X (NPRB)		X (new)	
Vessel-days					
Vessel Charter (Thorne)	16				16
Vessel Charter (Hopcroft)		5			
Vessel Charter (Kline - this project)			10	12	

<u>C Study design with sample numbers.</u>

Sampling design is categorized according to sample type (fish or zooplankton) and cruise type (Table 1). Zooplankton is further categorized into samples that will be combusted and bombed (for SIA and ZED) and those that will be enumerated (SED), which are outlined in Table 2.

Herring and other fishes. A sample size of 25 herring per bay will be adequate for statistics (Paul and Paul 1998a, Kline 1999). Four bays will be sampled on each herring cruise. We will sample up to 100 YOY herring per bay. We will select randomly from each bay 25 YOY herring for SIA. There will thus be a total of 100 herring for SIA per survey. Ten of these will be randomly selected for bomb calorimetry. On each herring cruise, forage trophic level fish (similar to herring, thus competitors, which area expected to shift concomitantly in SIA with herring, will be selected randomly from each bay for SIA. Fifteen forage fish will be selected per bay for a total of 60 per cruise. There will thus be 160 fish per cruise. One hundred additional herring competitors, mainly YOY salmon, will be collected by ADFG in PWS in June-July. The more than half billion salmon released into PWS each spring may be the most significant competitors of herring for zooplankton resources. Furthermore, the relative proportion of oceanic subsidies in YOY salmon in a given year was independent of month during the July - October period (Kline et al. submitted) suggesting they were not a short-lived phenomenon within a year. Therefore, concomitant oceanic subsidies are expected among all planktivorous fish taxa. Thus each 420 fish will undergo SIA. Twenty of the herring will also be bombed.

Table 2. Outline of project zooplankton samples.

Zooplankton SIA and ZED. Sample design for SIA and ZED varies by sample area-cruise as described below:

<u>The four bays (Zaikof, Whale, Eaglek, and Simpson Bays; both May and August)</u>

There will be two Multinet tows per bay. Each tow will consist of two each 150micron, one each 335 and 500 micron samples for a total of 4 samples per tow. There will thus be 8 SIA and ZED samples per bay, thus 32 samples per cruise.

<u>Slope stations (due south of PWS where is Z > 300 m) in August</u>

There will be four Multinet tows with four samples per tow like in the four bays for a total of 16 samples per cruise. There will be six ring net tows for a total of 6 samples per cruise. 50 individual *Neocalanus* will be picked for SIA only, consistent with Kline (1999) and Kline (submitted).

<u>Central area in six transects (Thorne's existing transects; both May and August)</u>

There will be one Multinet tow per transect with four samples per tow like in the four bays for a total of 24 samples per cruise. There will be one ring net per transect for a total of 6 samples per cruise.

<u>LTOP cruise (existing stations; early May)</u>

A sub-sample of the MOCNESS drogue (Kline submitted) will be saved from each LTOP station for combined SIA and ZED for a total of 16 samples per cruise. 100 individual *Neocalanus* will be picked for SIA only, consistent with Kline (1999) and Kline (submitted).

The number of zooplankton samples for combined SIA and ZED per May/August is 32 + 30 + 24 + 16 = 102. Total zooplankton combined SIA and ZED per year is 204. Total number of bomb samples per year is thus 204 + 20 (fish) = 224. The total number of individual zooplankter SIA per year is150.

Zooplankton samples to be identified and enumerated by species and stage for SED. Sample design for SED varies by sample area-cruise as described below:

<u>The four bays (Zaikof, Whale, Eaglek, and Simpson Bays; both May and August)</u>

There will be one Multinet tows per bay. Each tow will consist of two each 150micron, one each 335 and 500 micron samples for a total of 4 samples per tow. There will thus be 4 SED samples per bay, thus 16 samples per cruise.

<u>Slope stations (due south of PWS where is Z > 300 m) in August</u>

There will be two Multinet tows with four samples per tow like in the four bays for a total of 8 samples per cruise. There will be two ring net tows for a total of 2 samples per cruise. Total slope station SED samples will thus be 10.

<u>Central area in six transects (Thorne's existing transects; both May and August)</u>

There will be one Multinet tow per transect with four samples per tow like in the four bays for a total of 24 samples per cruise. There will be one ring net per transect for a total of 6 samples per cruise. Total slope station SED samples will thus be 30 per cruise.

There will be a total of 16 + 30 + 12 = 58 SED samples per May cruise: 16 + 30 + 12 = 58, and a total of 16 + 10 + 30 +12 = 68 SED samples per August cruise. The total number of SED samples per year will thus be 126.

<u>D Statistical tests.</u>

Data from this project will be comparable with the historical data from the mid 1990's as well as among the sampling sites and years of the proposed project. SIA and WBEC will be done with ANOVA statistical procedures in mind. ANOVAs will be possible to test for year (e.g., 2007 vs. 1995), and site (e.g., one of the bays) effects. Same seasons will be compared among years, e.g., one Fall versus another Fall as was done by Kline (1999). Ontogenetic effects will be determined by plotting WBEC and SIA as dependent variables using standard length as the independent variable (linear regression). This will be done for data aggregates well as by sampling cruise. Previously, there was no significant difference in SIA between synoptically sampled cohorts of age-0 herring and age-1 herring, and virtually no ontogenetic effect in SIA above 70 mm standard length, which is also about the size of herring when recruited into bays as very few smaller than this were sampled during 1994 - 1998 (Kline 1999, Kline 2001).

<u>E Essential metadata.</u>

Name of bay (location), Month, Year (to be recorded) – factors to be use in ANOVA

Fish standard length, wet weight, dry weight – to be measured and used as independent variables in regressions for WBEC and stable isotope values

WBEC and stable isotope values will also be regressed against each (e.g., see Fig. 2 in main body of proposal).

2. Criteria for determining acceptable data quality in terms of the activities to be performed or hypotheses to be tested.

Criteria = significance values for standard statistical tests: Alpha = 0.05 and P = 0.05

3. Metadata 'lite' information.

Title: Prince William Sound Herring Forage Contingency

Date: 1 August 2006

Contacts: Dr. Thomas C. Kline, Jr.
Access: via web and e-mail
Use and Security Policies: open via web
Contacts and Credits: Dr. Thomas C. Kline, Jr.
Geographical Boundaries: Northern Boundary: 60.8° N Latitude, Southern Boundary: 55.0° N Latitude, Eastern Boundary: 145.5 W Longitude, Western Boundary: 148.5 W Longitude
b. species-specific measurements- stable C and N isotope value and WBEC
taxonomic sampling—identification to species and stage (e.g. copepodite 5 for copepods, e.g., juvenile for fishes)

4. Algorithms.

Isotopic analysis is outsourced to labs using continuous flow isotope ratio mass spectrometers (CFIRMS). All algorithms are internal to the machine. Isotope abundance data are reported to the PI in conventional delta units.

5. Handling and custody of samples.

Samples are frozen, held frozen until freeze dried then stored dry. Identification to species and stage takes place in the field. Samples remain in the custody of the P.I. at all times except for when being shipped in which case Express Mail is used for tracking. Isotopic analyses will be outsourced to either the University of Alaska Stable Isotope facility or the Colorado Plateau Stable Isotope Laboratory at Northern Arizona University according to backlog status and price at each lab. This approach will expedite analysis while maintaining good quality. Both labs use similar equipment, Finnegan Delta Plus continuous flow isotope ratio mass spectrometers (CFIRMS), and employ similar quality assurance and quality control protocols. Dr. Kline has a history of outsourcing to these labs. Preserved samples will be preserved in the field and disposed of after counting in accordance to direction given by the sewage treatment authority (1 gallon per day allowed).

6. Calibration and evaluation of instrumentation.

Quality assurance and quality control protocols include analyses of laboratory standards before and after every five samples as well as periodic (1 in every 5 samples) sample duplication.

7. Procedures for data reduction and reporting.

A single isotopic analysis will generate the following data: $^{13}C/^{12}C$ and $^{15}N/^{14}N$ ratios expressed in standard delta units, $\delta^{13}C$ and $\delta^{15}N$, respectively, and %C and %N. The delta notation used to express stable isotope ratios is reported as the per mil (‰) deviation relative to an international standard, air for nitrogen, and Vienna Peedee belemnite (VPDB) for carbon. By definition, the isotope standards have delta values of zero, i.e. $\delta^{15}N = 0$ ‰ for atmospheric N_2. Instrument replication is typically within 0.2 ‰. The %C and %N data will be used to calculate C/N atom ratios. The data will consist of $\delta^{13}C$, $\delta^{15}N$ and C/N.

Stable carbon isotope ratios will be normalized for lipid content following the methods of McConnaughey and McRoy (1979) and trophic level following the method of Kline et al. (1998) and expressed as $\delta^{13}C_{TL}$' following Kline (1999). The $\delta^{13}C_{TL}$' will reflect source, either GOA or PWS, which have end-member values of –23.0 and –19.5, respectively (Kline 1997, 1999, 2001).

Trophic level will be determined by comparing $\delta^{15}N$ values to a reference value (Vander Zanden et al. 1997). The $\delta^{15}N$ of higher trophic levels will be calculated by adding the trophic enrichment factor, 3.4 (Minagawa and Wada 1984, Checkley and Miller 1989, Kline 1997), to the reference value. The herbivorous, i.e., trophic level = 2, copepod *Neocalanus cristatus*, will be used as the reference (Kline and Pauly 1998, Kline 1999). *N. cristatus* was chosen as the reference herbivore based upon observations that their carbon isotope values corresponded with those of PWS fishes (Kline 1999). Furthermore, *N. cristatus* will be sampled and SIA will be performed on them in the companion zooplankton project being submitted simultaneously to NPRB. The following formula will be used to calculate trophic level: $TL_i = (\delta^{15}N_i - \delta^{15}N_H/ 3.4) + 2$, where TL_i is the trophic level of organism i, $\delta^{15}N_i$ is the mean $\delta^{15}N$ value of organism i, and $\delta^{15}N_H$ is the mean reference herbivore $\delta^{15}N$ value.

The following applies to herring:

From the analysis of a single age-0 herring we will simultaneously determine whole-body energy content (WBEC) and the carbon and nitrogen stable isotope composition via stable isotope analysis (SIA). Wet and dry weight will be measured using an electronic balance and with carbon to nitrogen ratio data that are a product of SIA. WBEC will be determined using the following empirical model: WBEC (kJg^{-1} (wet mass)) = 0.103 x C/N (carbon-nitrogen atom ratio) + 32.60 x D/W (dry-wet ratio) –2.904. This model was determined empirically from measurements of herring from PWS (Kline unpublished; Paul et al. 2001). WBEC predicted by the above model compared favorably with that measured directly with a bomb calorimeter (Fig. 4 in main body of proposal). SED and ZED will computed by dividing direct measures with sample volumes obtained using the Multinet flowmeters.

EXXON VALDEZ OILSPILL TRUSTEE COUNCIL
DETAILED BUDGET
October 1, 2007 to September 30, 2008

Budget Category:	Approved FY 07	Requesting FY 08	Pending FY 09	TOTAL PROPOSED
Personnel	$103.4	$167.8	$106.3	$377.5
Travel	$3.2	$2.2	$3.2	$8.6
Contractual	$58.5	$72.3	$7.7	$138.5
Commodities	$13.7	$13.2	$3.7	$30.6
Equipment	$10.5	$0.0	$0.0	$10.5
Subtotal	$189.3	$255.5	$120.9	$565.7
Indirect (rate will vary by proposer)	$51.1	$69.0	$32.6	$152.7
Project Total	$240.4	$324.5	$153.5	$718.4
Trustee Agency GA (9% of Project Total)	$21.6	$29.2	$13.8	$64.6
Total Cost	$262.0	$353.7	$167.3	$783.0
Other Resources:				

Comments:

Revised header for FY 08, Added FY 08 PJ #; Changed FY 08 total on DPD Proposal Summary

Funds from non-EVOS sources, including in-kind contributions: Private foundation grant (M.J. Murdock) provides support for post-doc position during Year 1 only; this grant also bought the HydroBios Multinet ($80K) and provides some lab analysis and vessel charter support. Total Murdock funding is $260K (FY 07). NOAA is funding the Steller sea lion project (Thorne lead PI) that will provide comparison data as described in the proposal. This project is funded at $350K in FY 07 and $250K in FY 08. The Oil Spill Recovery Institute is contributing $40K in FY 07 to support sampling the diapausing Neocalanus project funded through Murdock, which will be compared to herring as described in the proposal. The PWS Ocean Observing System is contributing one month of PI time so that data from this project can be incorporated into the Observing System (FY 07 contribution about 15K). Additional support

FY 08 Request of FY 07 - FY 09

Project Number: 080811
Project Title: Prince William Sound Herring Forage Contingency - Submitted under the BAA
Proposer: T.Kline, PWSSC

Date Revised: 7/26/07

EXXON VALDEZ OILSPILL TRUSTEE COUNCIL
DETAILED BUDGET
October 1, 2007 to September 30, 2008

Personnel Costs:			Months	Monthly		Personnel
Name	Description		Budgeted	Costs	Overtime	Sum
T. Kline	PI		5	11.8		59.0
R. Campbell	Post-doc		9	6.2		55.8
TBN	Tech		10	5.3		53.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
		Subtotal	24.0	23.3	0.0	
					Personnel Total	$167.8

Travel Costs:	Ticket	Round	Total	Daily	Travel
Description	Price	Trips	Days	Per Diem	Sum
Annual meeting	0.3	2	8	0.2	2.2
Cruise travel	0.0	0	0	0.0	0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
				Travel Total	$2.2

FY 08

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Date Revised: 7/26/07

EXXON VALDEZ OILSPILL TRUSTEE COUNCIL
DETAILED BUDGET
October 1, 2007 to September 30, 2008

Contractual Costs:	Contract
Description	Sum
Vessel Charter (purse seiner)	48.0
Photocopying	0.3
PWSSC Network charge (computer-months)	2.4
Stable isotope analytical (freeze-dry)	13.7
Stable isotope lyophilizer useage	1.7
Stable isotope analytical (oven-dry)	4.9
Shipping include hazmat	1.0
Communications(fax&phone)	0.3
2 R/T (one per Juv Herring cruise) aircharter CDV-western PWS	0.0
If a component of the project will be performed under contract, the 4A and 4B forms are required. **Contractual Total**	$72.3

Commodities Costs:	Commodity
Description	Sum
calorimeter spare parts kit	0.3
ignition unit	0.4
Multinet maintenance kit	2.9
Replacement Multinet nets (3)	1.0
Replacement Multinet buckets (3)	1.5
Replacement Multinet canvas part (1)	1.0
Replacement impeller	0.2
Replacement ring net	0.5
Lab supplies- chemicals, gases, vials, bags	3.5
Office supplies	0.4
Computer supplies - software, upgrades	1.5
Commodities Total	$13.2

FY 08

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Date Revised: 7/26/07

EXXON VALDEZ **OILSPILL TRUSTEE COUNCIL**
DETAILED BUDGET
October 1, 2007 to September 30, 2008

New Equipment Purchases: Description	Number of Units	Unit Price	Equipment Sum
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
		New Equipment Total	$0.0

Existing Equipment Usage: Description	Number of Units	Inventory Agency
Winch	1	PWSSC
Balance	1	PWSSC

FY 08

Date Revised: 7/26/07

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