

EVOSTC ANNUAL PROJECT REPORT

Recipients of funds from the *Exxon Valdez* Oil Spill Trustee Council must submit an annual project report in the following format by Sept. 1 of each fiscal year for which project funding is received (with the exception of the final funding year in which a final report must be submitted). Please help ensure that continued support for your project will not be delayed by submitting your report by Sept. 1. Timely receipt of your report allows more time for court notice and transfer, report review and timely release of the following year's funds.

Satisfactory review of the annual report is necessary for continuation of multi-year projects. Failure to submit an annual report by Sept. 1 of each year, or unsatisfactory review of an annual report, will result in withholding of additional project funds and may result in cancellation of the project or denial of funding for future projects. **PLEASE NOTE:** Significant changes in a project's objectives, methods, schedule, or budget require submittal of a new proposal that will be subject to the standard process of proposal submittal, technical review, and Trustee Council approval.

Project Number: 080811

Project Title: Prince William Sound Herring Forage Contingency

PI Name: Thomas Kline

Time period covered: FY08

Date of Report: August 28 2008

Report prepared by: Dr. Tom Kline

Project website (if applicable):

Work Performed: Summarize work performed during the reporting period, including any results available to date and their relationship to the original project objectives. Explain deviations from the original project objectives, procedural or statistical methods, study area or schedule. Also describe any known problems or unusual developments, and whether and how they have been or can be overcome. Include any other significant information pertinent to the project.

- Funding of year 2 commenced in March, retroactive to include attendance at 2008 Alaska Marine Science Symposium.
- The P.I. and post-doc (Dr. Campbell) attended the 2008 Alaska Marine Science Symposium.
- The P.I. and post-doc participated in the herring workshops held in Cordova during March 2008, July 2008, and August 2008.
- Post-doc is on the herring plan writing committee.
- Four cruises were conducted, two to collect herring (November 2007 and March 2008) and two to collect zooplankton (Sept-Oct 2007 and May 2008). The scope of these cruises was described in the Interim Progress Report July 2007 and in the Powerpoint presentation shown during the October 2007 workshop. Preliminary results were shown in the latter. These previously shown results are not presented here so that new information can be instead.
- Further preliminary results are presented below (these are draft figures, some may be included in the presentation the P.I. will make at the Annual Herring Workshop, which is expected to take place in October 2008).
- Further lab processing of samples is in progress.

Plankton Energetics

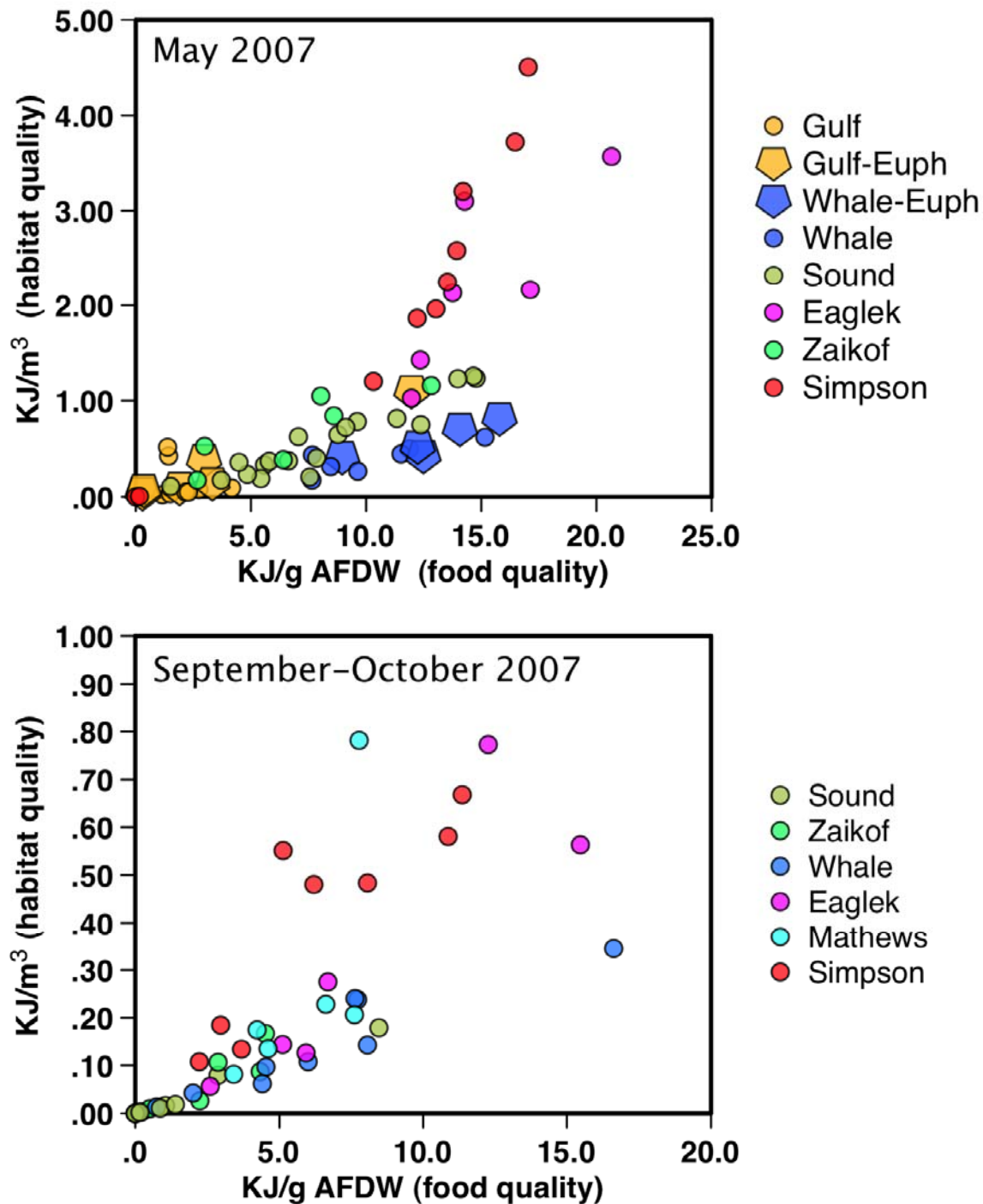


Figure 1. Plankton energy; May (top) and September-October (bottom) 2007.

Figure 1 summarizes zooplankton energy content data. Energy is expressed in terms of density with respect to water volume within listed habitats as a measure of habitat quality and in terms of energy density per unit mass (ash-free dry weight = AFDW) of zooplankton as a measure of food quality. The Prince William Sound (PWS) herring nursery bays (Whale, Eaglek, Zaikof, and Simpson) were each quite

different with respect to both parameters. There was approximately one order of magnitude less energy available per unit volume of water in the fall compared to spring, however, food quality was generally comparable. Spring samples with a number of euphausiids are depicted as pentagons; this may have been important as winter forage for herring (as it has in Sitka Sound, J.J. Vollenweider, pers. comm.) and may be a source of good quality food where available such as in Whale Bay. Two of the bays had overall more energy per unit volume than the greater Sound or offshore in the Gulf. Each point in the figure above reflects one net sample from 50 to 100 m³ of water (sample volume based on flow meter reading). These results confirm that plankton energy availability is highly variable in space and time and that not all herring nursery habitats are equal. This information will need to be considered for herring population supplementation projects, especially those involving juvenile life stages.

Euphausiids in Whale Bay, May 2007

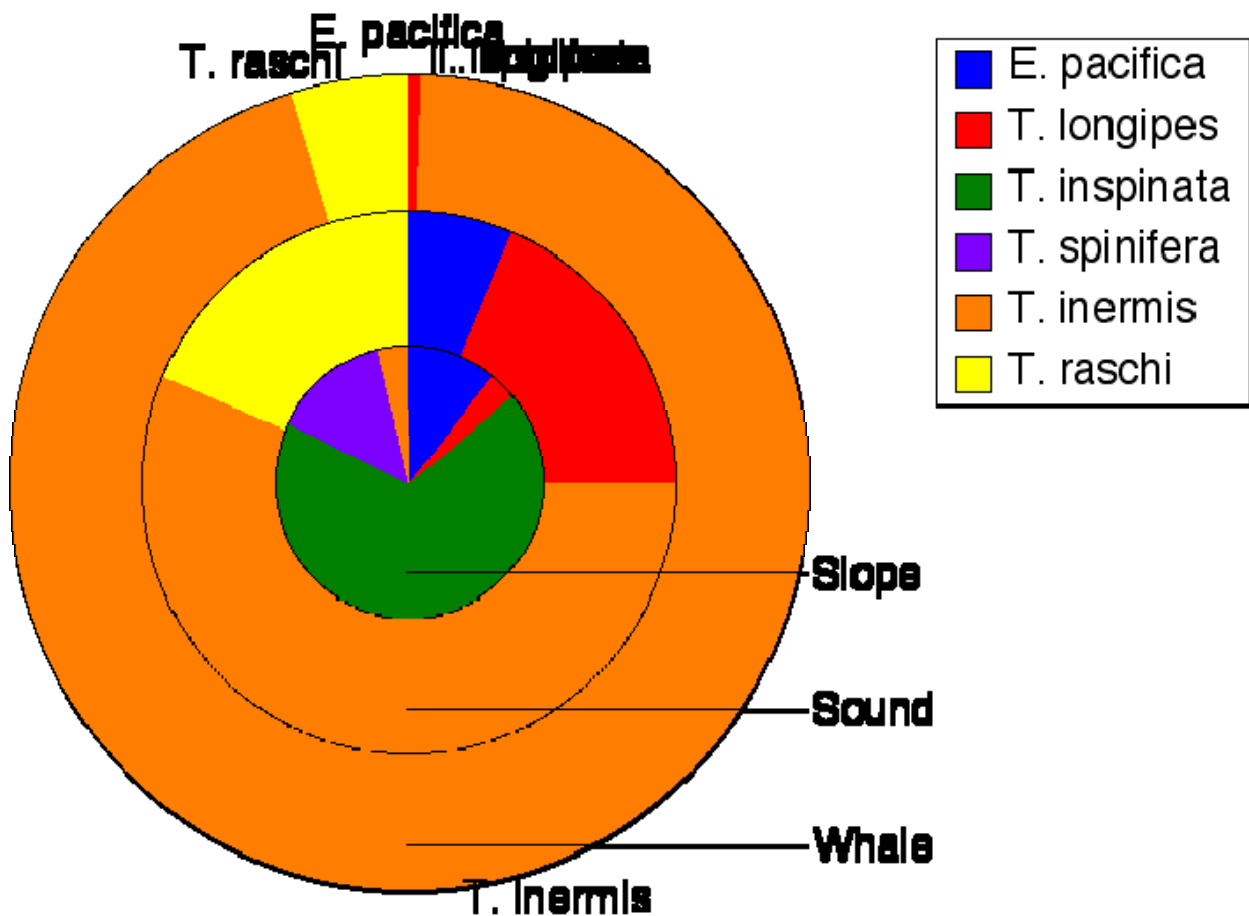


Figure 2. Species composition of Euphausiids, May 2007.

Whale Bay was the most euphausiid rich area in May 2007. *Thysanoessa inermis* dominated the euphausiid species composition in Whale Bay (Fig. 2). They contributed a smaller fraction of those found elsewhere in the Sound and less still on the slope. They may have been a contributing factor to the higher energetic content of herring during the preceding winter (see below). Because *T. inermis* is a shelf species that is more abundant in colder years (Pinchuk et al. 2008), there is potential for inter-annual variation.

Herring as plankton

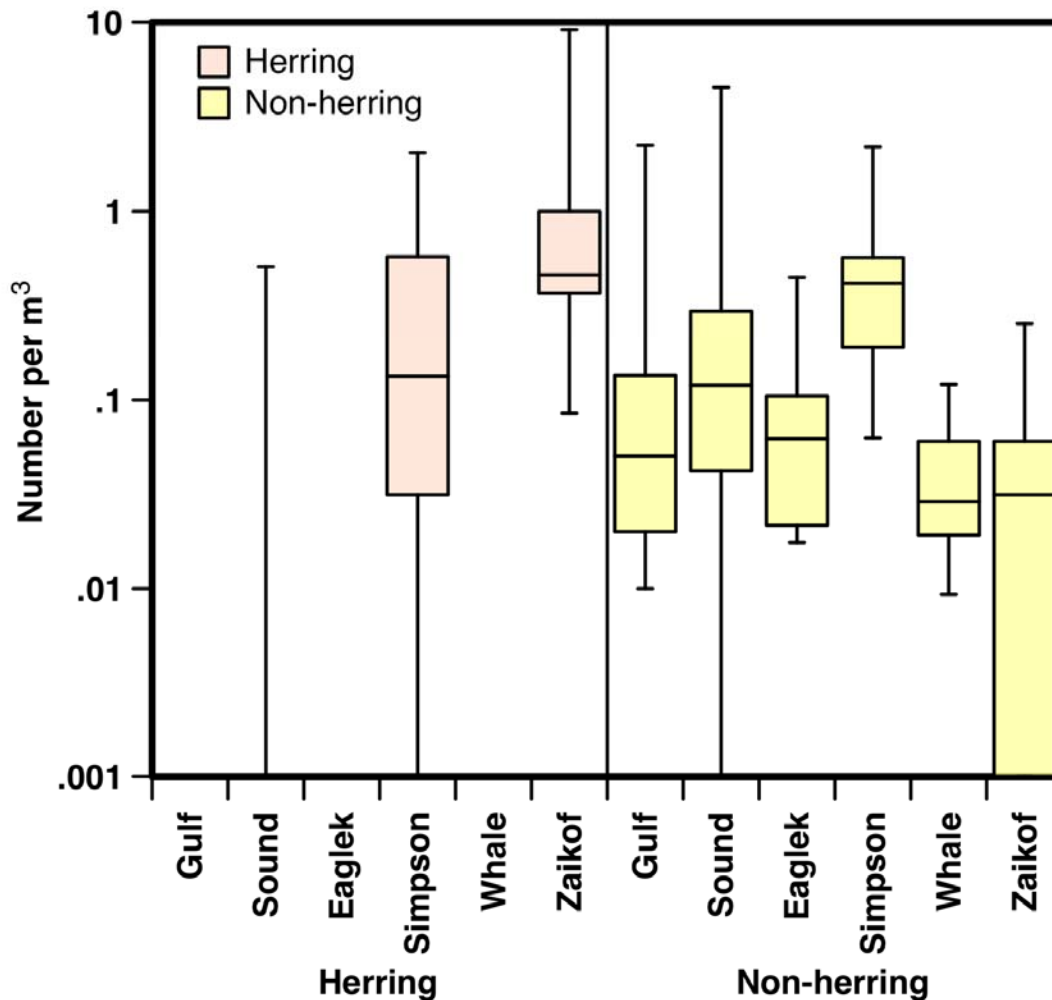


Figure 3. Herring larvae and other ichthyoplankton in PWS, May 2007.

Figure 3 summarizes the fish larval composition of zooplankton from the May 2007 plankton survey as box and whisker plots in terms of herring and non-herring (all other fish spp.). There was about one fish larva per 1 to 10 m³ of water. Two to three week old (age consensus derived at October 2007 workshop) herring larvae contributed to significant portions of the ichthyoplankton composition in two of the nursery bays. Herring larvae were found in all tows made in Zaikof Bay and all but one tow made in Simpson Bay in May 2007. A similar pattern is expected for the 2008 survey based upon samples picked for SIA by the P.I. during the cruise. Herring larvae were found in just two of the tows made in the greater Sound in May 2007. Herring larvae were also observed in certain Sound tows made in May 2008. **NOTE: Sampling herring larvae in our tows was not anticipated in the proposal (DPD); these serendipitous results, however, are critical to understanding the herring recruitment process and thus herring restoration in PWS.** These results show the extreme spatial variability of herring recruitment processes that should be taken into account for intervention. Assessment of herring larval distribution like that shown here is a new tool that was not previously available to PWS herring restoration.

Plankton community structure (this section prepared by Dr. Rob Campbell)

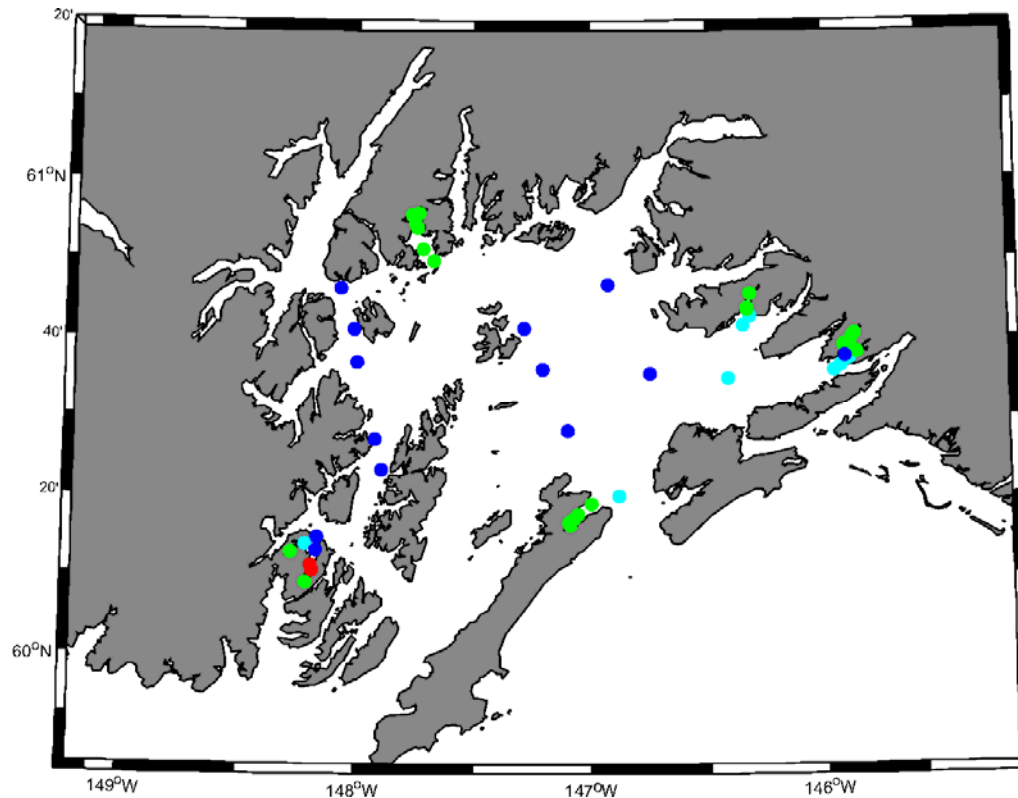


Figure 4. Hierarchical clustering analysis of plankton taxa in PWS September-October 2007. Map of the stations indicating the cluster identity of each station. Colors correspond to the identified clusters (Fig. 5; Green = Bay cluster; Light blue = Eastern PWS cluster; Dark blue: Central PWS cluster; Red = Whale Bay cluster).

Patterns in the structure of the plankton community were analyzed by hierarchical clustering and indicator species analysis (ISA; Figs. 4 and 5). Rare species (occurring in <10 % of stations) were not included in the analysis, and data was $\log_{10}(n+1)$ transformed prior to analysis. Clustering was done on the Euclidean distance matrix from the species \times station matrix, using Ward's linkage method. The resulting clusters broke down fairly well into geographic areas (Fig. 4), with well-defined clusters for three of the bays (Simpson, Eaglek and Zaikof), and an open water cluster that could be further subdivided into central and eastern PWS clusters. Stations in Whale Bay fell into all of the clusters, as well as two stations that fell into a cluster unique to Whale Bay (Fig. 5).

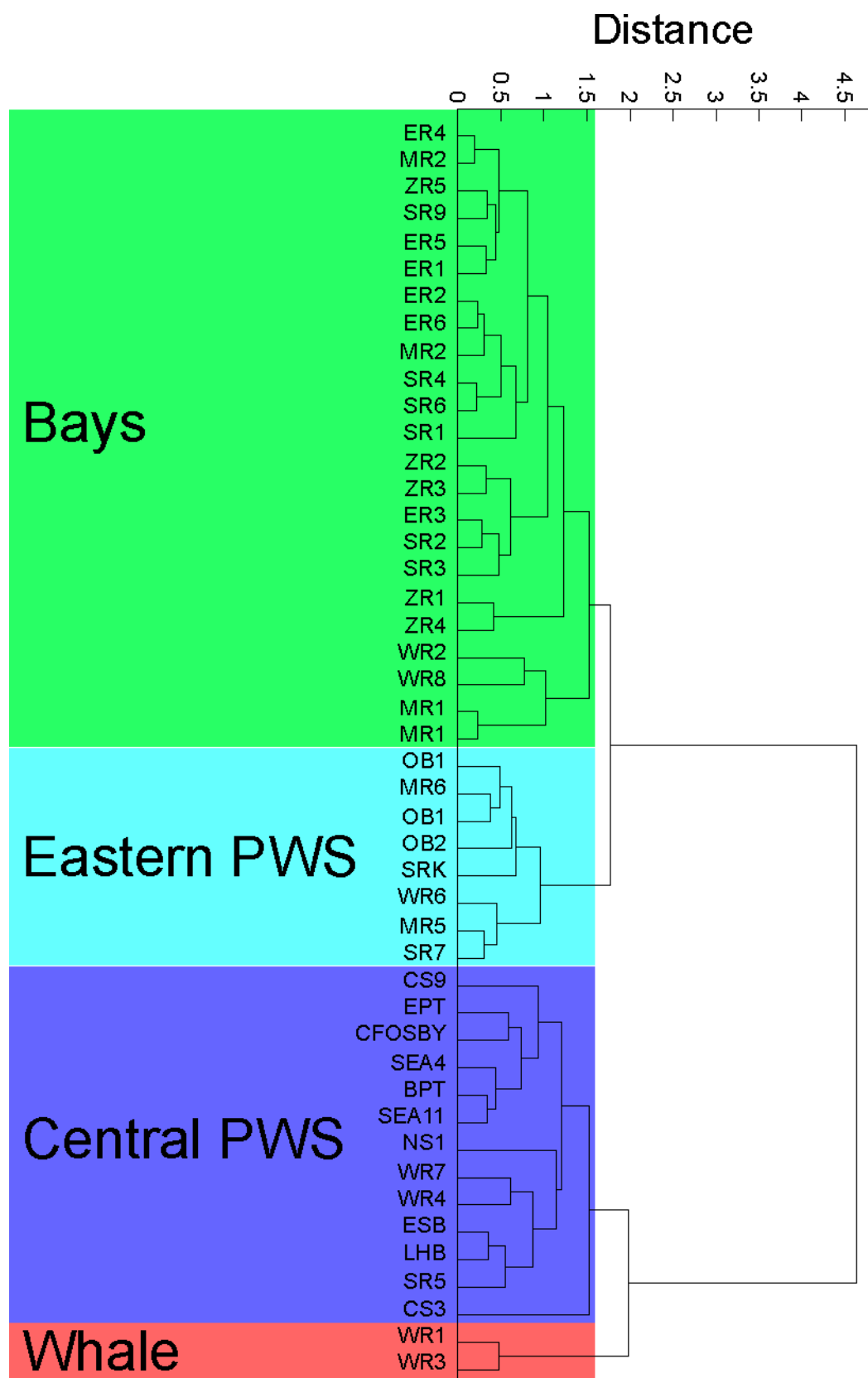


Figure 5. Hierarchical clustering analysis of plankton taxa in PWS, September-October 2007. Dendrogram showing the resulting station groupings.

Indicator species analysis (Dufrene and Legendre, 1997) was used to identify the important plankton taxa in the station groups identified by the cluster analysis. The ISA statistic for each group is the product of the relative abundance of the taxa in the group (scaled to all groups) and the frequency of occurrence of species in the group. An associated probability was calculated for each ISA statistic by Monte Carlo simulation (10000 iterations), and a critical value of 0.05 used to diagnose significant groups. The results of the ISA were sorted within each group to produce a list of taxa ranked by their relative importance in each cluster (Table 1). There were several taxa common to the different clusters, particularly large copepods (<2 mm), small pteropods (>0.5 mm), and cnidarian medusae. The differences between the clusters were thus primarily caused by differences in the abundances of the different taxa. Abundances of the common taxa identified by ISA (Fig. 6) varied considerably between areas, with larger numbers of small copepods occurring in the bay stations, and larger copepods and pteropods (which are more oceanic species) occurring in the open water stations. Euphausiid calyptopis (immature life stage) were also common at the bay stations.

Table 1. Indicator species analysis (ISA) for the station groups identified by hierarchical cluster analysis. The taxa are vertically sorted by relative importance within each cluster, with more important taxa above less important taxa.

Bays	Eastern PWS	Central PWS	Whale Bay
Copepods <2 mm	Pteropods <0.5 mm	Copepods <2 mm	Pteropods <0.5 mm
Pteropods <0.5 mm	Medusae	Medusae	Copepods <2 mm
Medusae	Ctenophora	Pteropods <0.5 mm	Pteropods >0.5 mm
Larvacea	Copepods <2 mm	Cladocera	Euphausiid calyptopis
Euphausiid calyptopis	Larvacea	Larvacea	Medusae

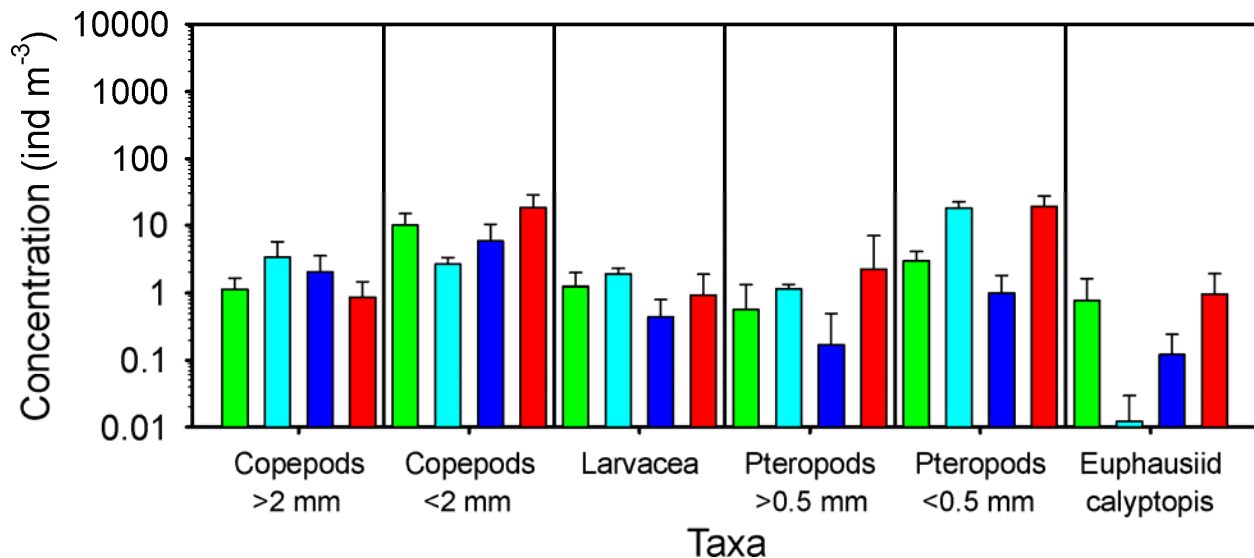


Figure 6. Abundance (mean \pm standard deviation) of the more common plankton taxa identified by ISA, arranged by the station groups identified by hierarchical cluster analysis. Color indicates the cluster group, and corresponds to the colors used in fig. X (Green = Bay cluster; Light blue = Eastern PWS cluster; Dark blue: Central PWS cluster; Red = Whale Bay cluster).

Characterization of herring food sources using stable isotope analysis (SIA)

Validation of stable isotope analysis diagnostics

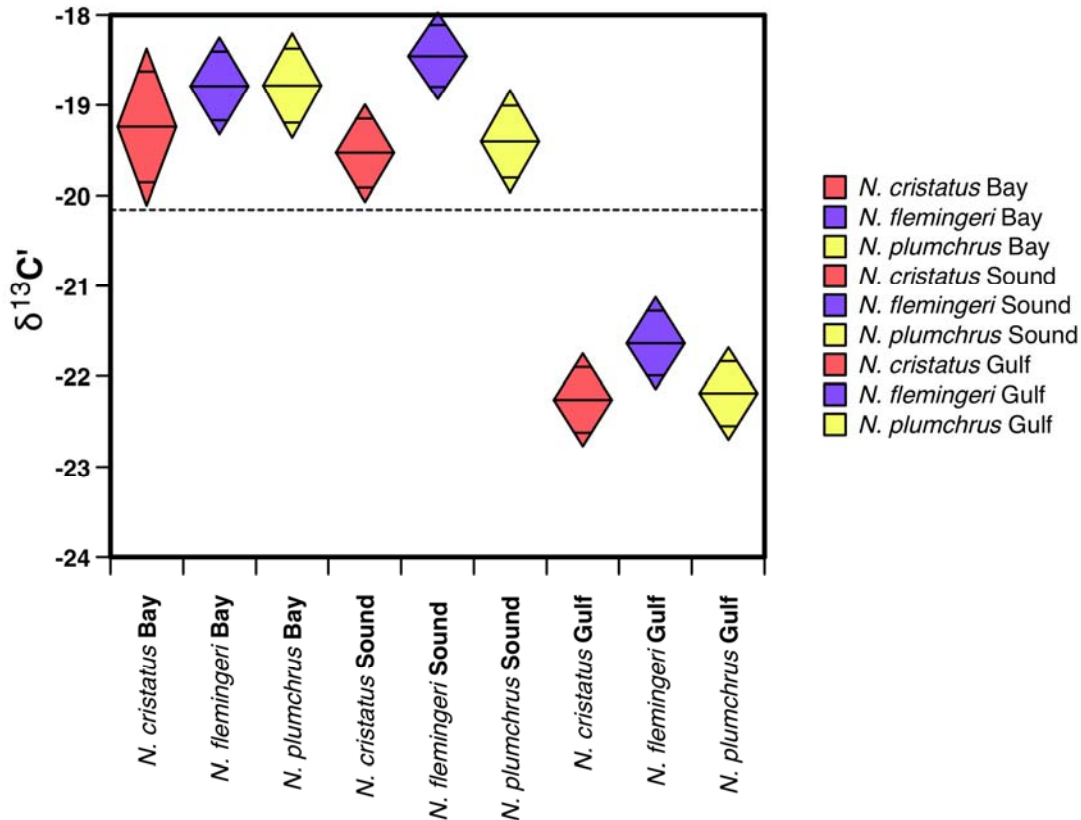


Figure 7. Stable isotope diagnostics.

Figure 7 summarizes the May 2007 $\delta^{13}C'$ data of the three *Neocalanus* spp. by habitat type (herring nursery bays = Bay, other parts of PWS = Sound, offshore in the Gulf of Alaska = Gulf) as 'diamond mean comparison plots' to validate SIA as a tracer of Gulf of Alaska (GOA) carbon in PWS by confirming previous observations (e.g., Kline et al. 2008). The dashed line is the grand mean of all sample groups. The diamonds show group means by their widest line and 95% confidence intervals at the vertices. The short horizontal lines within the diamonds indicate where groups are significantly different from each other by non-overlap. Rationale for the SIA single species approach was given in Kline (1999) and Kline (2009). PWS carbon, including that from within herring nursery bay habitats, continues to be observed within a narrow $\delta^{13}C'$ range of -20 to -18 whereas that of the GOA is < -21. These data are critical for interpreting the isotope shifts measured in herring, below. Planktivorous fish such as herring consuming carbon exclusively of PWS origin are expected to have $\delta^{13}C'$ values of -19 to -17 based on these observations and trophic fractionation effects (e.g., Kline et al. 2008). Values more negative than ~ -20 range can thus be inferred to reflect incorporation of GOA or oceanic carbon.

Neocalanus is an oceanic copepod taxon requiring water deeper than 300m for part of its life history. The mere occurrence of *Neocalanus* in the relatively shallow PWS herring nursery bays (Fig. 7) quite distant from this deep water speaks volumes for the connectivity of these habitats with the greater oceanic system.

Herring food sources and whole-body energy composition (WBEC) during 2006-7

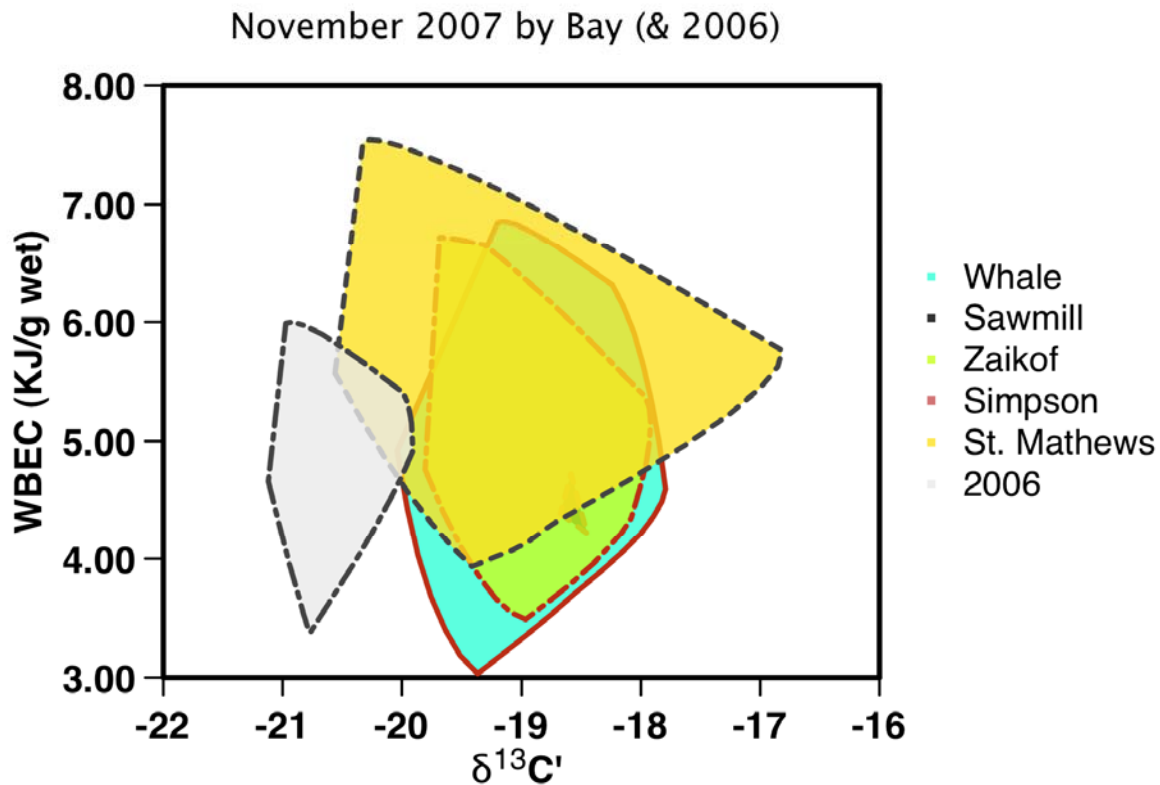


Figure 8. Fall 2006 and 2007 herring energy in relation to SIA.

Figure 8 relates the whole body energy content (WBEC) of age-0 herring (Y-axis) to their $\delta^{13}\text{C}'$ values (x-axis) observed during November 2006 (limited samples) and November 2007 as a reflection of pre-winter condition. Data are presented as 'convex hulls' that show the range for each parameter by data group. Herring in 2007 were relatively less dependent on GOA than 2006. A number herring from St. Mathews had relatively greater GOA carbon (values more negative than -19) and these tended towards higher WBEC. The relatively high overlap in $\delta^{13}\text{C}'$ values among bays within a given sampling period is similar to that reported by Kline (1999). These results confirm that herring energy sources are variable in time on inter-annual time scales (2006 and 2007 was quite different compared to differences among bays at a given time; parallels previous observations e.g., Kline 2007). This inter-annual variability was hypothesized to reflect variability in ocean climate processes driving exchange between PWS and GOA (Kline 1999) and is a consideration for intervention projects on juvenile life stages.

March & April (where indicated) 2007 by Bay

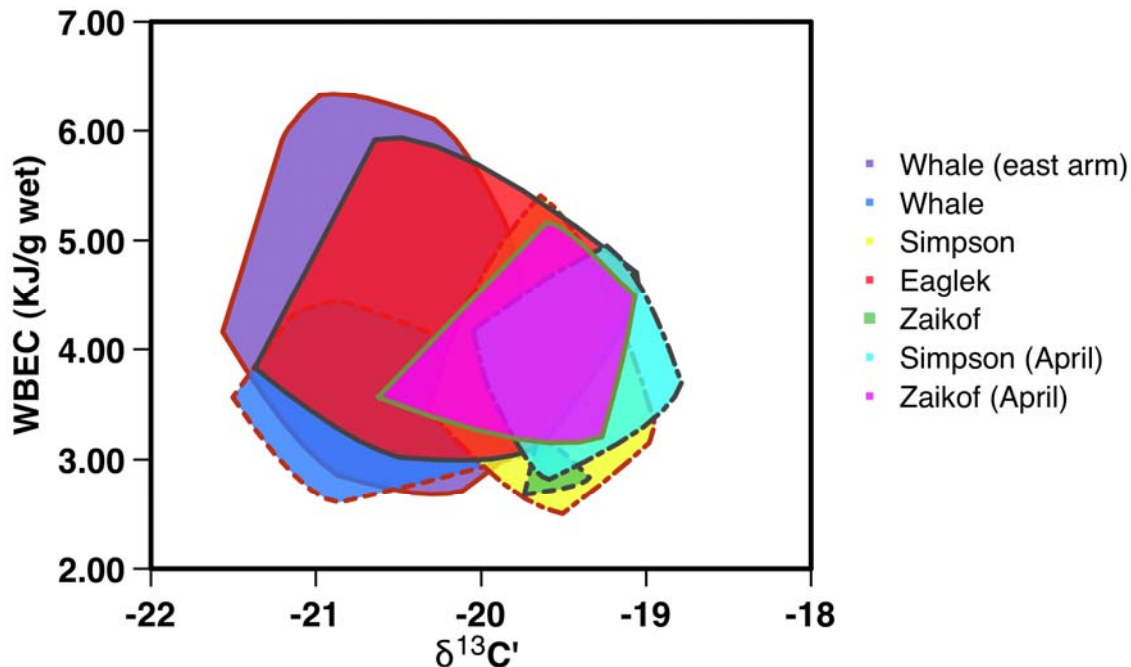


Figure 9. Spring 2007 herring energy in relation to SIA.

Figure 9 relates the whole body energy content (WBEC) of age-0 herring in relation to $\delta^{13}\text{C}'$ values observed during March to April 2007 among PWS herring nursery bays as a reflection of post-winter condition. A fortuitous second set of samples from April provided by Steve Moffitt (ADFG) lacked the lowest WBEC values observed in March. The $\delta^{13}\text{C}'$ values overlap in part with those observed in the fall of 2006 so are more negative than those observed later in 2007 (Fig. 8). The East and main arm of Whale Bay tended to have herring with higher energy content as well as lower $\delta^{13}\text{C}'$ values that may have resulted from feeding on euphausiids there (see Fig 2).

Interaction with other fishes

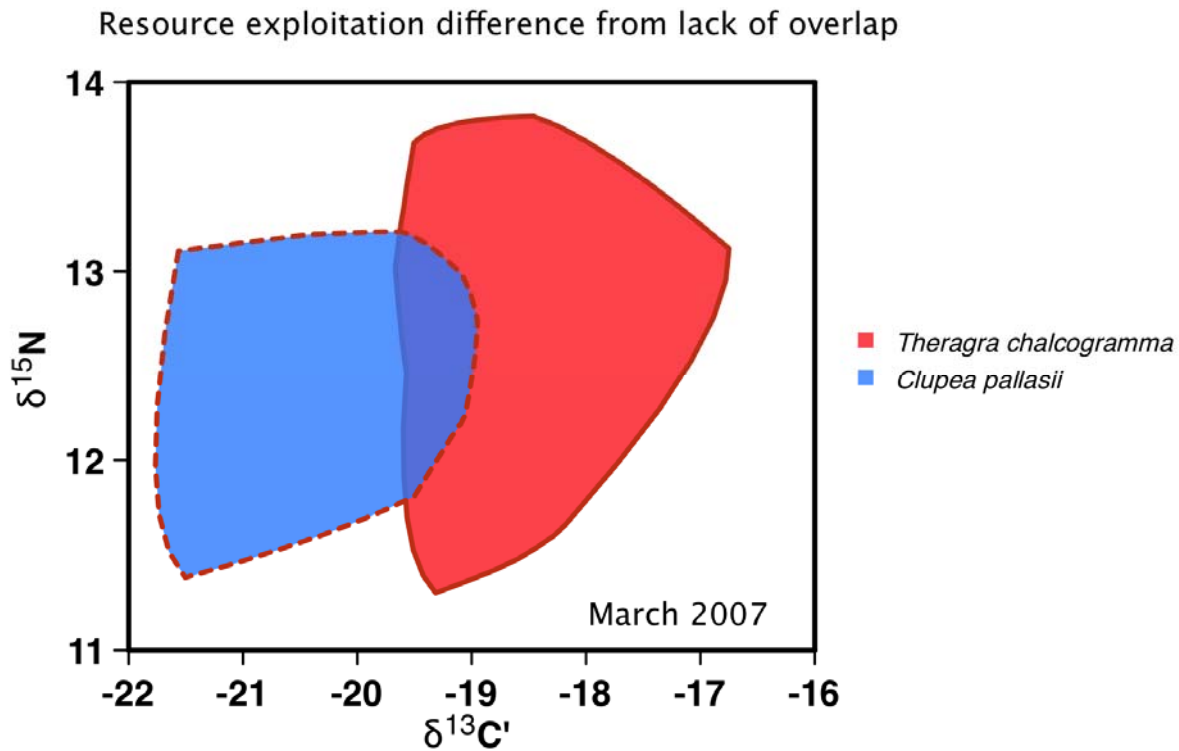


Figure 10. Overwintering dietary (carbon source) difference between herring and pollock.

Stable isotope analysis of age 0 and 1 walleye pollock (*Theragra chalcogramma*) sampled incidentally with herring in March 2007 plotted in Figure 10 provides a measure of over-winter dietary overlap. Data are not inconsistent with predation on herring by a few of the pollock (these have higher $\delta^{15}\text{N}$ values). Lower $\delta^{13}\text{C}'$ values of herring are similar to the values observed in November 2006 (Fig. 8), which is consistent with little feeding during winter, the intervening period. The much higher $\delta^{13}\text{C}'$ values in pollock suggest feeding on PWS carbon during winter as inferred by Kline (2008). Pollock are thus able to maintain or increase energy content during the winter while herring cannot (Kline 2008). The upshot of this is that some food resources that are available to pollock in winter are apparently not available herring. There is far less overlap between herring and pollock in March compared to November (Fig. 11).

Resource competition suggested by dietary overlap

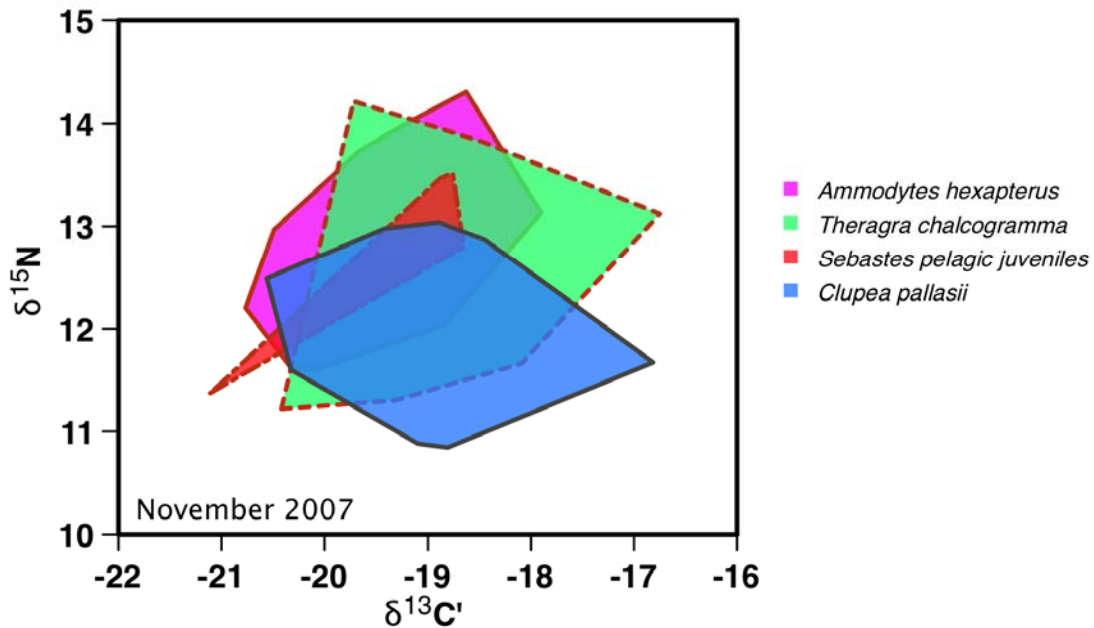


Figure11. Dietary overlap between herring and other species, November 2007 based on SIA.

Center of the $\delta^{13}\text{C}'$ value distribution near -19 suggests that mostly PWS carbon was important for fall 2007 herring food webs, which contrasts with 2006 observations and some of those from the 1990's (Fig. 11; Kline 1999, 2007). There is relatively high concordance in both stable isotope values among fish taxa. Nevertheless herring $\delta^{15}\text{N}$ values were slightly lower suggesting that not all resources available to competitors were also available to herring. This could be explained by herring not able to consume higher trophic plankters, which can have higher $\delta^{15}\text{N}$ values such as amphipods, decapods or carnivorous coepods (Kline 1999).

Preliminary results from the May 2008 plankton survey

Massive phytoplankton bloom observed in Zaikof Bay

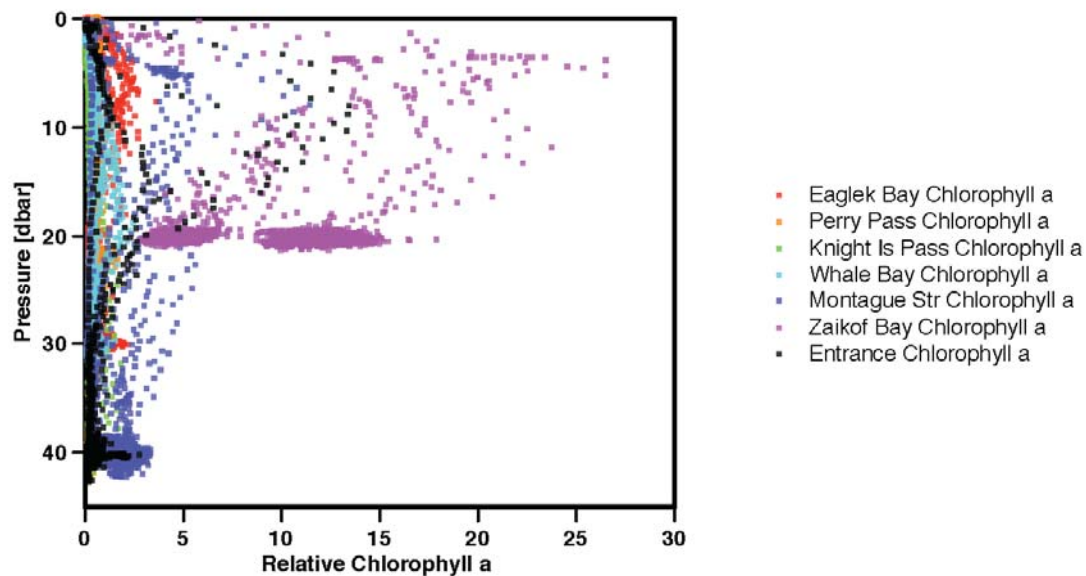


Figure 12. Distribution of chlorophyll with depth (1 dbar = 1 m) in PWS in late May 2008 indicating very high concentrations down to 20m depth in Zaikof Bay. Data from the fluorometer that is integrated into the Multinet zooplankton sampling device.

There was a very noticeable bloom of diatoms in Zaikof Bay based on surface water color and microscopic observations made during the May 2008 plankton survey. This bloom extended in depth to at least 20m (Fig. 12). The Hinchinbrook Entrance transect from Porpoise Rocks to Naked Island had the second highest concentrations of chlorophyll which was followed by Montague Strait transect from Northwest Passage to Montague Point.

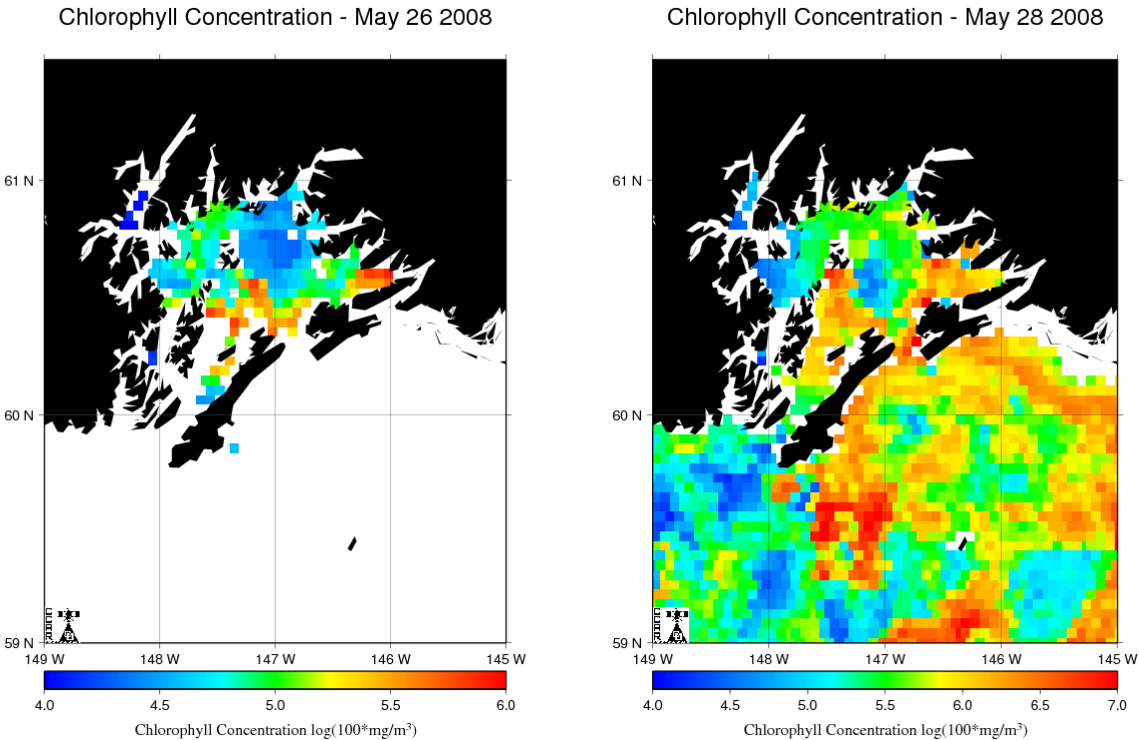


Figure 13. Surface chlorophyll distribution at end of May 2008 cruise and two days later. Data obtained from Colorado Center for Atmospheric Research website: <http://argo.colorado.edu/~realtime/welcome/>. Note how there is almost no coverage within actual herring nursery bays (only Gravina).

Phytoplankton can be detected from space because they contain photosynthetic chlorophyll pigments that naturally fluoresce. However, this fluorescence can only be seen from a satellite orbiting in space when skies are clear. The cloudiness that prevails in our area often precludes remote sensing from space. This happened during the May 2008 plankton survey; the first day with clear skies was the last day of the cruise, 26 May. Figure 13 (left) is a satellite-derived image showing surface chlorophyll in PWS on the last day of the cruise. This was just the beginning of a clearing trend so that two days later there was even better coverage, which is shown in Figure 13 (right). Figure 13, left and right collectively, shows that (1), chlorophyll was most concentrated in parts of PWS bordering the Gulf, (2) that the bloom was even stronger out in the Gulf and (3), that the bloom can shift in distribution in matter of a few days. A limitation of satellite-derived images is that only the surface of the ocean is looked at and that smaller spatial scales features such as the chlorophyll in herring nursery bays (Fig. 12) within PWS are not seen. Chlorophyll standing stock as a reflection of primary productivity is an important consideration since Perry and Schweigert (2008) showed that northeast Pacific herring carrying capacity is correlated to primary productivity.

Evidence for late bloom from copepod development

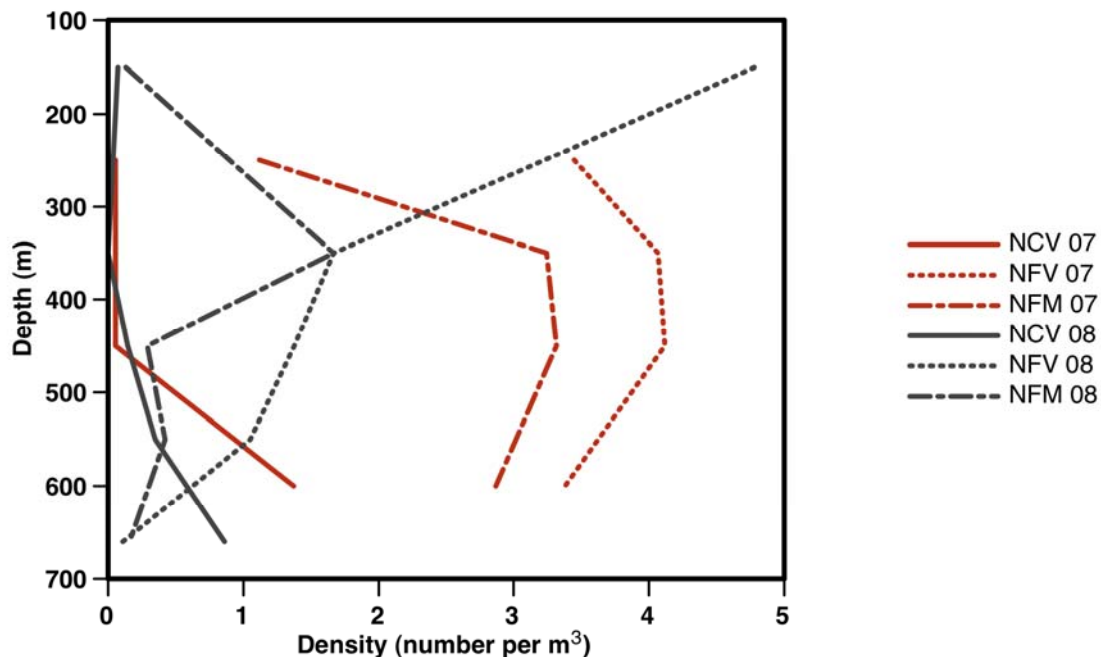


Figure 14. Comparison of vertical distributions of late developmental stages of key copepod species from the deep part of PWS in May 2008 with that of May 2007. There were less stage copepodite-V *N. cristatus* in late May 2008 compared to late May 2007 (NCV in Fig. 14). We observed far fewer male *N. flemingeri* in 2008 compared to 2007 (NFM in Fig. 14). Females were all but absent. There were more copepodite-V stage *N. flemingeri* in shallow water but less in deep water 2008 than 2007 (NFV in Fig. 14). My interpretation is that neither species had yet progressed as far in its maturation and migration pattern in 2008 compared to 2007. Hence the bloom in 2008 was relatively late. The sampling and data analysis shown in this figure is due to synergistic funding provided by the Oil Spill Recovery Institute and the Murdock Charitable Trusts.

Recent Pacific Decadal Oscillation Index (PDOI) trends: 2008 has been a cold year

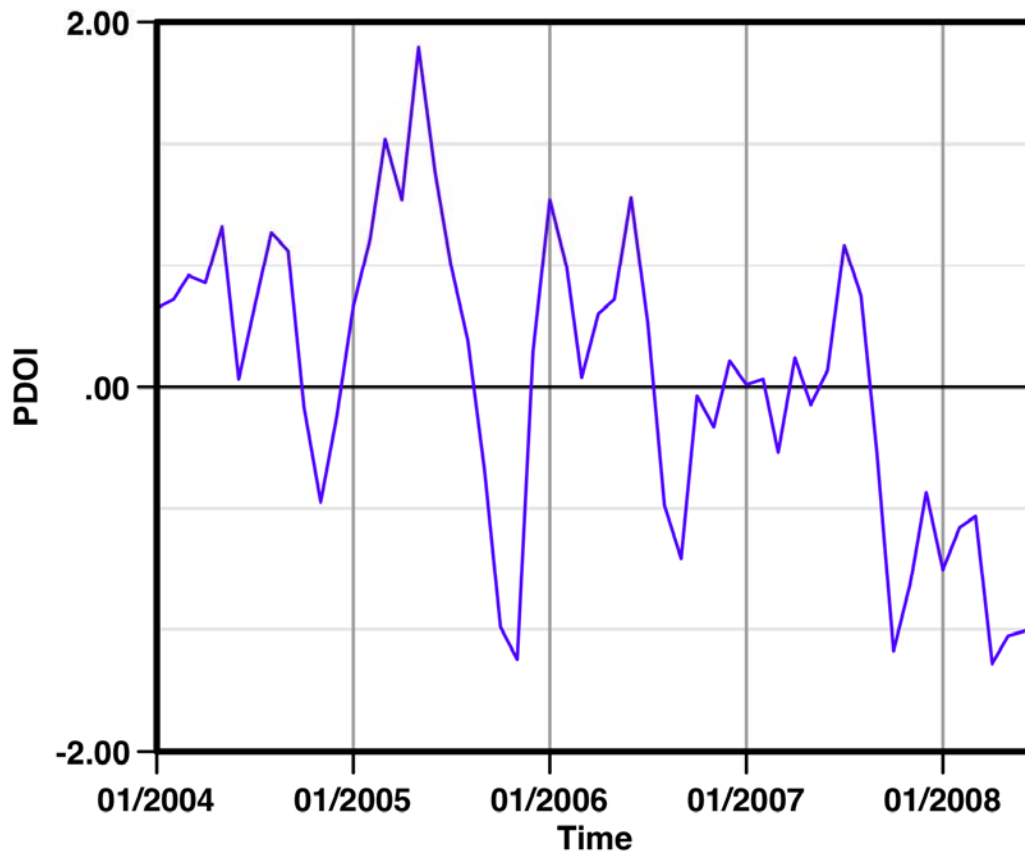


Figure 15. PDOI trend of the last 4.5 years showing continuous negative anomaly during the first half of 2008. Data from PDOI webpage: <http://jisao.washington.edu/pdo/>.

The PDOI, a measure of the north Pacific sea temperatures, slipped into a negative or cool phase in late 2007 that has persisted through 2008 until the present (Fig. 15). Cooler temperatures may slow development rates (Q10 law), which may provide an explanation for the apparent late bloom. Herring recruited well in the 1980's when the PDOI was positive; thus quite different from present ocean conditions.

Settled volume analysis

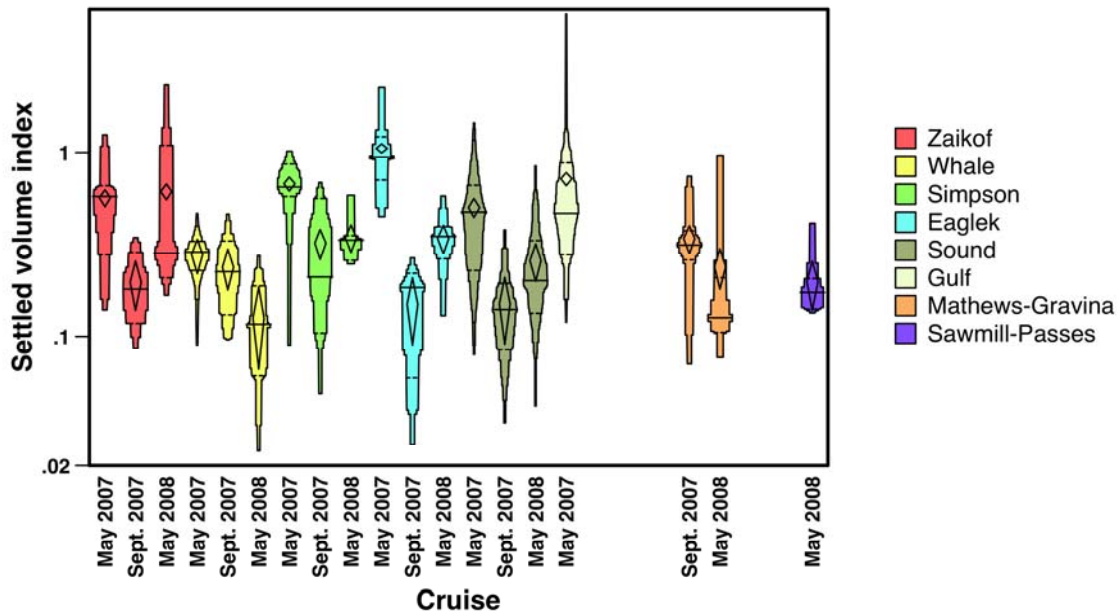


Figure 16. Spatial and temporal variability of net plankton volume per unit volume water sampled shown as distributional box and whisker plots.

Relative settled volume (Fig. 16) provides a synoptic comparison of the plankton populations from the three completed plankton surveys. The four key herring nursery bays are shown on the left. Sawmill Bay and nearby passes and St. Mathew Bay and adjacent Port Gravina were also sampled when time allowed. These supplemental sites were also sampled for juvenile herring (Fig. 8). May 2008 included extreme high and low plankton densities. The high values from Zaikof in May 2008 were probably influenced by phytoplankton contributing to the sample.

Literature cited

Dufrene, M. and P. Legendre 1997. Species assemblages and indicator species: the need for a flexible asymmetrical approach. *Ecol. Monogr.* 67:345-366

Kline, T.C. Jr. 1999. Temporal and Spatial Variability of $^{13}\text{C}/^{12}\text{C}$ and $^{15}\text{N}/^{14}\text{N}$ in Pelagic Biota of Prince William Sound, Alaska. *Can. J. of Fish. Aquat. Sci.* 56 (Supplement 1): 94-117

Kline, T.C. Jr. 2007. Rockfish trophic relationships in Prince William Sound, Alaska based on natural abundance of stable isotopes. *In: J. Heifetz, J. Dicosimo, A.J. Gharrett, M.S. Love, V.M. O'Connell, and R.D. Stanley (eds), Biology, Assessment, and Management of North Pacific Rockfishes, Alaska Sea Grant College Program, AK-SG-07-01.*

Kline, T.C. Jr. 2008. Ontogenetic, temporal, and spatial variation of feeding niche in an unexploited population of walleye Pollock (*Theragra chalcogramma*). *In: G.H. Kruse, K. Drinkwater, J.N. Ianelli, J.S. Link, D.L. Stram, V. Wespestad, and D. Woodby (eds), Resiliency of gadid stocks to fishing and climate change, AK-SG-08-01.*

Kline, T.C. Jr., J.L. Boldt, E.V. Farley, Jr., L.J. Haldorson, and J.H. Helle. 2008. Pink salmon (*Oncorhynchus gorbuscha*) marine survival rates reflect early marine carbon source dependency. *Progr. Oceanogr.* 77:194-202.

Kline, T.C. Jr. 2009. Characterization of carbon and nitrogen stable isotope gradients in the northern Gulf of Alaska using terminal feed stage copepodite V *Neocalanus cristatus*. Deep-Sea Research. Provisionally Accepted, June 2008.

Perry, R.I. and J.F. Schweigert. 2008. Primary productivity and the carrying capacity for herring in NE Pacific marine ecosystems. Progr. Oceanogr. 77:233-240.

Pinchuk, A.I., K.O. Coyle, and R.R. Hopcroft. 2008. Climate-related variability in abundance and reproduction of euphausiids in the northern Gulf of Alaska 1998-2003. Progr. Oceanogr. 77:203-216.

Completed and Future Measurable Project Tasks

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

November	Complete juvenile herring survey #2, November 2007: COMPLETED
December	Complete analysis of juvenile herring survey #1, March 2007: COMPLETED
December	Complete analysis of zooplankton survey #1, May 2007: COMPLETED
December	Complete analysis of zooplankton survey #2, September 2007: COMPLETED

FY08 2nd Quarter (January 1, 08 to March 31, 08)

January	Annual Marine Science Symposium: COMPLETED
March	Complete juvenile herring survey #3, March 2008: COMPLETED

FY08 3rd Quarter (April 1, 08 to June 30, 08)

May	Complete zooplankton survey #3, May 2008: COMPLETED
June	Complete analysis of juvenile herring survey #2, November 2007: COMPLETED

FY08 4th Quarter (July 1, 08 to September 30, 08)

September	Complete zooplankton survey #4, September 2008 (TO BE DONE)
August	Submit Annual Report (THIS REPORT)

FY09 1st Quarter (October 1, 08 to December 31, 08)

November	Complete juvenile herring survey #4, November 2008
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FY09 2nd Quarter (January 1, 09 to March 31, 09)

January	Annual Marine Science Symposium
March	Complete juvenile herring survey #5, March 2009

FY09 3rd Quarter (April 1, 09 to June 30, 09)

June	Complete analysis of zooplankton survey #3, May 2008
June	Complete analysis of juvenile herring survey #3, March 2008

FY09 4th Quarter (July 1, 09 to September 30, 09)

September*	Complete analysis of zooplankton survey #4, September 2008
September*	Complete analysis of juvenile herring surveys #4, November 2008 and #5, March 2009
April, 2010	Submit Final Report, to the Trustee Council Office

* the NOAA contract is expected to extend beyond the end of FY09; accordingly the work will be completed one year after the anticipated start of the FY09 contract (March 2009) and not the calendar date given here.

Future Work: Summarize work to be performed during the upcoming year, if different from the original proposal. Describe any proposed changes in objectives, procedural or statistical methods, study area or schedule. *NOTE: Significant changes in a project's objectives, methods, schedule or budget*

require submittal of a new proposal subject to the standard process of proposal submittal, technical review and Trustee Council approval.

No changes other than addition of herring larval analysis see. Fig. 3.

Coordination/Collaboration: Describe efforts undertaken during the reporting period to achieve the coordination and collaboration provisions of the proposal, if applicable.

- Field work sampling for herring involved collaboration with the project being led by Dr. Thorne
- Collected live materials for Dr. Hershberger's project
- Herring length frequency data requested by Dr. Bishop transmitted to Herring P.I.'s via email
- These data were provided to Dr. Bishop also as XL spreadsheets
- Preliminary graphs transmitted to Herring P.I.'s via email
- Samples were provide to this project by other projects; from Steve Moffitt (ADFG) and J.J. Vollenweider (NOAA)

Community Involvement/TEK & Resource Management Applications: Describe efforts undertaken during the reporting period to achieve the community involvement/TEK and resource management application provisions of the proposal, if applicable.

- Contributed to community herring planning effort
- Conducted special plankton outreach laboratory sessions for Cordova High School students
- Gave lectures on plankton to the community
- Preliminary results sent to collaborators for use in resource management

Information Transfer: List (a) publications produced during the reporting period, (b) conference and workshop presentations and attendance during the reporting period, and (c) data and/or information products developed during the reporting period. *NOTE: Lack of compliance with the Trustee Council's data policy and/or the project's data management plan will result in withholding of additional project funds, cancellation of the project, or denial of funding for future projects.*

a)

Kline, T.C. Jr. 2007. Rockfish trophic relationships in Prince William Sound, Alaska based on natural abundance of stable isotopes. *In:* J. Heifetz, J. Dicosimo, A.J. Gharrett, M.S. Love, V.M. O'Connell, and R.D. Stanley (eds), *Biology, Assessment, and Management of North Pacific Rockfishes*, Alaska Sea Grant College Program, AK-SG-07-01.

Includes herring data.

Kline, T.C. Jr. 2008. Ontogenetic, temporal, and spatial variation of feeding niche in an unexploited population of walley Pollock (*Theragra chalcogramma*). *In:* G.H. Kruse, K. Drinkwater, J.N. Ianelli, J.S. Link, D.L. Stram, V. Wespestad, and D. Woodby (eds), *Resiliency of gadid stocks to fishing and climate change*, AK-SG-08-01.

Compares pollock with herring; analyses competition and over-wintering energy interactions.

Kline, T.C. Jr., J.L. Boldt, E.V. Farley, Jr., L.J. Haldorson, and J.H. Helle. 2008. Pink salmon (*Oncorhynchus gorbuscha*) marine survival rates reflect early marine carbon source dependency. *Progr. Oceanogr.* 77:194-202.

Provided evidence that oceanic subsidies affects PWS food webs sufficient to drive 50% of pink salmon marine survival. Summarizes carbon source diagnostics presently being used.

Prepared draft article for the PWSSC Breakwater newsletter.

b)

Poster presentation at the 2008 Alaska Marine Science Symposium

Poster presentation at 2008 Ocean Sciences meeting

c)

Herring length frequency data requested by Dr. Bishop transmitted to Herring P.I.'s via email. These data were provided to Dr. Bishop also as XL spreadsheet.

Preliminary graphs transmitted to Herring P.I.'s via email.

Incorporated results of Sitka herring project (using results presented by Jeep Rice in April and from email exchange with J.J. Vollenweider) herewith.

Interacted with an external (to the herring program) researcher Dr. Barb Block on the present climatic state of the Sound vis a vis the PDOI.

Synergistic funding aided in explaining the delayed nature of the 2008 bloom.

Budget: Explain any differences and/or problems between actual and budgeted expenditures, including any substantial changes in the allocation of funds among line items on the budget form. Also provide any new information regarding matching funds or funds from non-EVOS sources for the project. *NOTE: Any request for an increased or supplemental budget must be submitted as a new proposal that will be subject to the standard process of proposal submittal, technical review, and Trustee Council approval.*

none

We can accept your annual report as a digital file (Microsoft Word or WordPerfect), with all figures and tables embedded. Acrobat Portable Document Format (PDF) files (version 4.x or later) are also acceptable; please do not lock PDF files or include digital signatures.

Please submit reports electronically to science_director@evostc.state.ak.us. Also, please be sure to post your annual report on your own website, if you have one.



*We appreciate your prompt submission of your annual report
and thank you for your participation.*

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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PROPOSAL SUMMARY PAGE

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

Project Period: FY 07-FY 09

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 Received:	FY 08	\$ 353.7K	
	EVOS Funding Requested:	FY 09	\$ 249.7K	TOTAL: \$865.4K
	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: 2 Sept. 2008

PROJECT PLAN

NEED FOR THE PROJECT

Statement of Problem

Pacific herring (*Clupea pallasii*) 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 kJ/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 kJ/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 kJ/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, 50% 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. 2008). In late 1995, a strong pulse of oceanic carbon subsidies was detected concurrently using $^{13}\text{C}/^{12}\text{C}$ 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, 2007). Oceanic subsidies were driven by influx of Gulf of Alaska origin zooplankton, which is based on the diagnostic value of $^{13}\text{C}/^{12}\text{C}$ 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 $^{13}\text{C}/^{12}\text{C}$ content (Fig. 2). Herring with low $^{13}\text{C}/^{12}\text{C}$ 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 September, 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.

The diagnostic value of SIA to infer oceanic subsidies is based on comparing $^{13}\text{C}/^{12}\text{C}$ values of zooplankton from the continental slope with those in PWS (Fig. 2). A cross-shelf $^{13}\text{C}/^{12}\text{C}$ 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, Kline et al. 2008). 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 September. Continued sampling from GAK10 to GAK13 will enable collecting May end-members exactly as was done on GLOBEC (Kline et al. 2008) furthering the existing time series. Two days of the May and September 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.

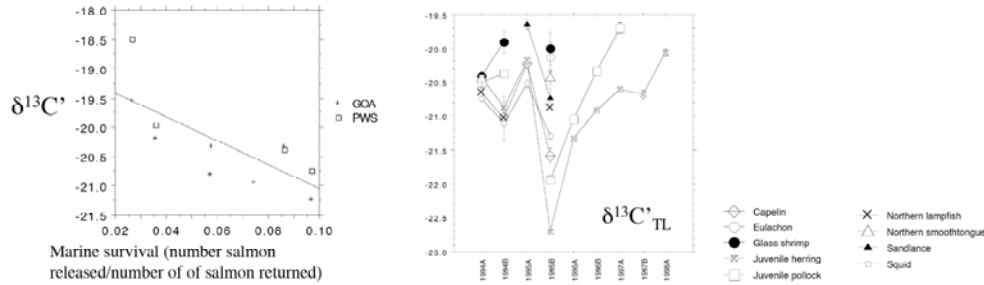


Figure 1. Left. Relationship between stable carbon isotope composition and marine survival rate for PWS hatchery pink salmon (line shows regression for combined sampling areas ($R^2 = 0.58$, symbols by sampling area ($R^2 = 0.63$ (PWS), $R^2 = 0.73$ (GOA = Gulf of Alaska))). **Right.** Concomitant shifts in $^{13}\text{C}/^{12}\text{C}$ among PWS forage trophic level taxa during SEA suggested a large oceanic carbon pulse in late 1995. Years were split so that January through September was 'A' and September through December was 'B.' Data from, respectively, Kline et al. (2008) and Kline (2007).

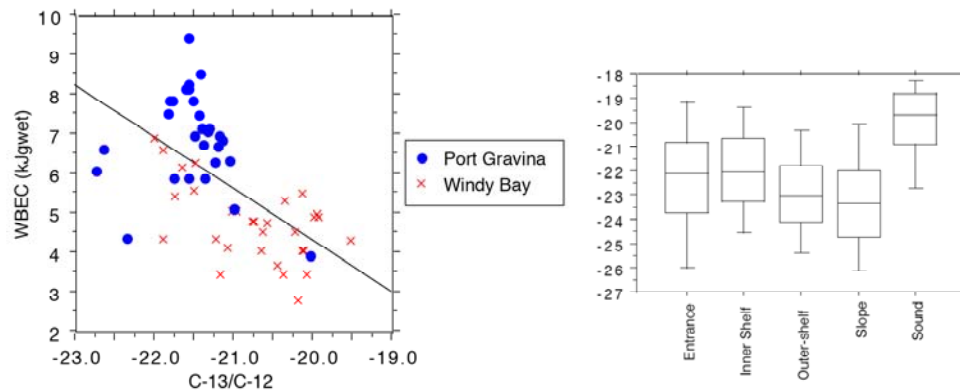


Figure 2. Left. Oceanic subsidies led to higher WBEC. Inverse relationship between $^{13}\text{C}/^{12}\text{C}$ and WBEC of YOY from fall of 1994 suggested oceanic carbon subsidies increased WBEC and the potential for over-winter survival for Port Gravina herring. In contrast, many if not most herring 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 $^{13}\text{C}/^{12}\text{C}$ data (left axis). Carbon measured in feeding stage *Neocalanus* from slope stations (Slope) was ^{13}C -depleted, had low $^{13}\text{C}/^{12}\text{C}$ values with a mean of -23 , whereas feeding stage *Neocalanus* from PWS (Sound) was ^{13}C -enriched, had high $^{13}\text{C}/^{12}\text{C}$ 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 A.J Paul (pers. comm.) and Kline (1999); right panel from Kline et al. (2008).

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 $^{13}\text{C}/^{12}\text{C}$, a large but variable fraction (can be $\gg 50\%$) 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. 2008). 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.

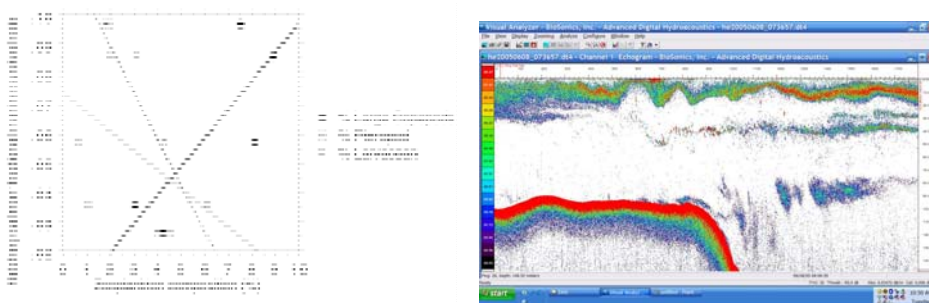


Figure 3. **Left.** Relationship between taxonomic composition of net plankton and salmon marine survival rate. Marine survival rate = number of adult salmon returning/number of salmon released. The percent of net samples comprised of large copepods, mainly *Neocalanus* spp, was directly correlated with 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 2001, 2007, 2008). 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 November. 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 2008) 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 than 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, 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. 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 dry mass of bulk zooplankton samples from the Multinet for three mesh sizes, 150, 335, and 500 microns. Zooplankton density will be calculated by dividing the mass by 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.

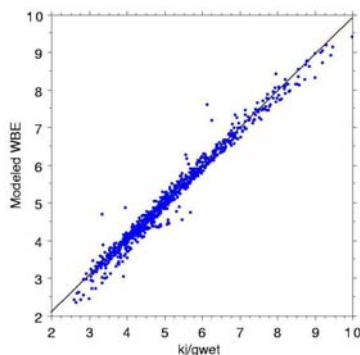


Figure 4. Modeled WBEC versus (vertical axis) measured WBEC (horizontal) content of juvenile herring from PWS sampled during the SEA project (units are kilo-Joules per gram wet mass). The close fit to the 45° line indicates that the model provides a good estimate of WBEC. The WBEC was measured using 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 in November. Two zooplankton cruises per calendar year will be funded by this project, one in May

and one in September. 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).

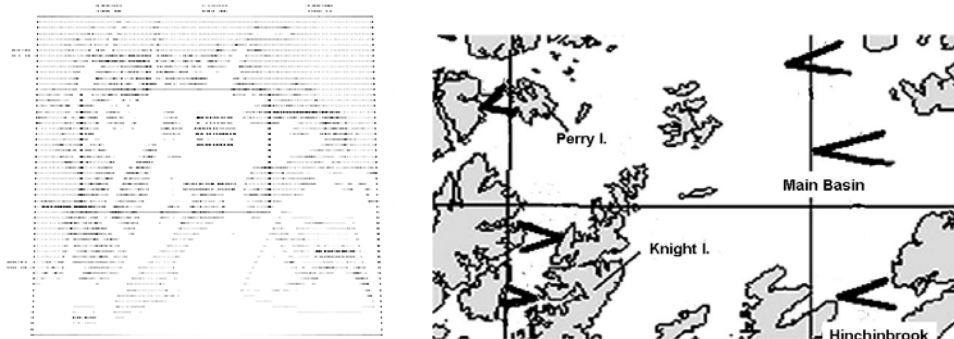


Figure 5 Left. The PWS study showing the four SEA bays and central area expounded in right panel. Right. Six standardized zooplankton acoustic transects from Thorne (2006).

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 200m. Transects in the main basin are the nearest deepwater areas to Orca Bay.

Coordination and Collaboration with Other Efforts

This project is coordinated and managed within the larger Prince William Sound Science Center program on juvenile herring research. There are 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 are 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 starting date was October 1, 2006. The 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 be performed during March and one in November of each year of the project. All herring survey vessel charter time is funded on other projects (NOAA funded) or Dr. Thorne’s EVOS projects. **Zooplankton** surveys take place in May and September of each year in years one and two. This project is funding the vessel charter for one cruise each May and September each year for a total of four cruises over the duration of the project. It is estimated to take one year following the final cruise (which will be herring survey #5 in March 2009) to complete the laboratory work and analysis of samples.

Measurable Project Tasks (FY09)

FY09 1st Quarter (October 1, 08 to December 31, 08)

November Complete juvenile herring survey #4, November 2008

FY09 2nd Quarter (January 1, 09 to March 31, 09)

January Annual Marine Science Symposium

March Complete juvenile herring survey #5, March 2009

FY09 3rd Quarter (April 1, 09 to June 30, 09)

June Complete analysis of zooplankton survey #3, May 2008

June Complete analysis of juvenile herring survey #3, March 2008

FY09 4th Quarter (July 1, 09 to September 30, 09)

September*	Complete analysis of zooplankton survey #4, September 2008
September*	Complete analysis of juvenile herring surveys #4, November 2008 and #5,
March 2009	
April, 2010	Submit Final Report, to the Trustee Council Office

* the NOAA contract is expected to extend beyond September, 2009; accordingly the work will be completed one year after the anticipated start of the FY09 contract (March 2009) and not the calendar dates given here.

RESPONSIVENESS TO KEY TRUSTEE COUNCIL STRATEGIES

Community Involvement and Traditional Ecological Knowledge (TEK)

This project is community based. It is 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 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 be 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

1994-2008	Research Scientist, Prince William Sound Science Center
1995-2008	Director, Prince William Sound Science Center Scientific Diving Program
1995-2008	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 acknowledge EVOS-TC funding)

- 2008 Kline, T.C. Jr., J.L. Boldt, E.V. Farley, Jr., L.J. Haldorson, and J.H. Helle. Pink salmon (*Oncorhynchus gorbuscha*) marine survival rates reflect early marine carbon source dependency. *Progr. Oceanogr.* 77:194-202.
- 2008 Kline, T.C. Jr. Ontogenetic, temporal, and spatial variation of feeding niche in an unexploited population of walley Pollock (*Theragra chalcogramma*). *In*: G.H. Kruse, K. Drinkwater, J.N. Ianelli, J.S. Link, D.L. Stram, V. Wespestad, and D. Woodby (eds), Resiliency of gadid stocks to fishing and climate change, AK-SG-08-01.
- 2007 Kline, T.C. Jr. Rockfish trophic relationships in Prince William Sound, Alaska based on natural abundance of stable isotopes. *In*: J. Heifetz, J. Dicosimo, A.J. Gharrett, M.S. Love, V.M. O'Connell, and R.D. Stanley (eds), Biology, Assessment, and Management of North Pacific Rockfishes, Alaska Sea Grant College Program, AK-SG-07-01.
- 2001 Kline, T.C., Jr. The trophic position of Pacific herring in Prince William Sound Alaska based on their stable isotope abundance. *In*: F. Funk, J. Blackburn, D. Hay, A.J. Paul, R. Stephenson, R. Toresen, and D. Witherell (eds.), Herring: Expectations for a New Millennium. University of Alaska Sea Grant, AK-SG-01-04, Fairbanks. p. 69-80.
- 1999 Kline, Thomas C., Jr. Temporal and Spatial Variability of $^{13}\text{C}/^{12}\text{C}$ and $^{15}\text{N}/^{14}\text{N}$ in pelagic biota of Prince William Sound, Alaska. *Can. J. Fish. Aquat. Sci.* 56 (Suppl. 1) 94-117.

Recent Collaborators

Coyle, K., Cooney, R., Haldorson, L., Hopcroft, Weingartner, T., Whitley, 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 (FY09)

This project is essentially an integration of separate three projects of the SEA program. These were projects 320I and 311, which were led by T. Kline, project 320U, which was led by A.J. Paul and project 320H, which was led by Ted Cooney. The on-going project has the additional goal of determining zooplankton energy density that was not part of either 311, 320H, 320I or 320U. This is thus a very economical budget given the annual costs of the separate projects in SEA.

Personnel – \$145.8K

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.

The P.I. is dedicating 100% (7 out of 7 months from March through September 2009) of his time to PWS herring research. This time is needed 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 post-doc is leading zooplankton taxonomic analysis, and is collaborating with the PI during data analysis and synthesis. The post-doc will spend 4 out of the 7 months working on this project. The technician is assisting in both the field and laboratory and will be working on the project for 7 out of the 7 months. The P.I. is leading field sampling, performing data interpretation, supervising other personnel, and performing administrative functions. The project is labor intensive in both the field and in the lab. There is considerable handling of individual specimens, which takes up time. Hence personnel costs contribute significantly to the total project costs.

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.

Travel – 2.2K

Travel Funds are being requested for attendance at the January 2009 Alaska Marine Science Symposium.

Contractual – \$24.9K

A major cost in the contractual category is the actual mass spectrometric (isotopic) analysis of the samples, which is outsourced to UAF (see QA-QC section). There was a per sample price increase of \$3 that took effect in 2008 that is reflected in the FY 09 budget. Furthermore there was also an increase of \$1 per sample charge for lyophilization for FY09. 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 \$50 per person-month network charge.

Equipment & Commodities- \$4.7K

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. Annual costs for software updates are based on past experience.

Funds from non-EVOS sources, including in-kind contributions: Private foundation grant (M.J. Murdock) provided 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 funded the Steller sea lion project (Thorne lead PI) that provided comparison data as described in the proposal. This project was funded at \$350K in FY 07 and \$250K in FY 08. The Oil Spill Recovery Institute is contributed \$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 contributed one month of PI time so that data from this project can be

incorporated into the Observing System (FY 07 contribution about 15K). There will be support for three months of support for the post-doc in from the PWSOS in FY09. 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.

A Sample type.

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))

B Locations.

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	<u>March</u>	<u>Early May</u>	<u>Mid-May</u>	<u>September</u>	<u>November</u>
	2006	existing	existing			existing
	2007	completed		completed	completed	completed
	2008	completed		completed	pending	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
	<u>Task</u>					
	Adult Herring acoustics	Thorne				
	Zooplankton acousites		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
	<u>Geographic scope</u>					
	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)	
	<u>Vessel-days</u>					
	Vessel Charter (Thorne)	16				16
	Vessel Charter (Hopcroft)		5			
	Vessel Charter (Kline - this project)			10	12	

C Study design with sample numbers.

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 are 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:

The four bays (Zaikof, Whale, Eaglek, and Simpson Bays; both May and September)

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.

Slope stations (due south of PWS where is $Z > 300$ m) in September

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).

Central area in six transects (Thorne's existing transects; both May and September)

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.

LTOP cruise (existing stations; early May)

A sub-sample of the MOCNESS drogue (Kline et al. 2008) 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 et al. (2008).

The number of zooplankton samples for combined SIA and ZED per May/September 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 zooplankton SIA per year is 150.

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:

The four bays (Zaikof, Whale, Eaglek, and Simpson Bays; both May and September)

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.

Slope stations (due south of PWS where is $Z > 300$ m) in September

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.

Central area in six transects (Thorne's existing transects; both May and September)

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 September cruise. The total number of SED samples per year will thus be 126.

D Statistical tests.

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).

E Essential metadata.

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 ten samples; blanks are run after 20 samples.

7. Procedures for data reduction and reporting.

A single isotopic analysis generates the following data: $^{13}\text{C}/^{12}\text{C}$ and $^{15}\text{N}/^{14}\text{N}$ ratios expressed in standard delta units, $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$, 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 Pee Dee belemnite (VPDB) for carbon. By definition, the isotope standards have delta values of zero, i.e. $\delta^{15}\text{N} = 0$ ‰ 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 $\delta^{13}\text{C}$, $\delta^{15}\text{N}$ 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 $\delta^{13}\text{C}_{\text{TL}}$ following Kline (1999). The $\delta^{13}\text{C}_{\text{TL}}$ will reflect source, either GOA or PWS, which have end-member values of -23.0 and -19.5, respectively (Kline 1997, 1999, 2001, Kline et al. 2008).

Trophic level will be determined by comparing $\delta^{15}\text{N}$ values to a reference value (Vander Zanden et al. 1997). The $\delta^{15}\text{N}$ 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: $\text{TL}_i = (\delta^{15}\text{N}_i - \delta^{15}\text{N}_H / 3.4) + 2$, where TL_i is the trophic level of organism *i*, $\delta^{15}\text{N}_i$ is the mean $\delta^{15}\text{N}$ value of organism *i*, and $\delta^{15}\text{N}_H$ is the mean reference herbivore $\delta^{15}\text{N}$ 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: $\text{WBEC} (\text{kJg}^{-1} \text{ wet mass}) = 0.103 \times \text{C/N} (\text{carbon-nitrogen atom ratio}) + 32.60 \times \text{D/W} (\text{dry-wet ratio}) - 2.904$. 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 be computed by dividing direct measures with sample volumes obtained using the Multinet flowmeters.

**EXXON VALDEZ OILSPILL TRUSTEE COUNCIL
DETAILED BUDGET FORM FY 07 - FY 09**

Budget Category:	Approved FY 07	Approved FY 08	Pending FY 09		TOTAL PROPOSED
Personnel	\$103.4	\$167.8	\$145.8		\$417.0
Travel	\$3.2	\$2.2	\$2.2		\$7.6
Contractual	\$58.5	\$72.3	\$24.9		\$155.7
Commodities	\$13.7	\$13.2	\$4.7		\$31.6
Equipment	\$10.5	\$0.0	\$0.0		\$10.5
Subtotal	\$189.3	\$255.5	\$177.6		\$622.4
Indirect (rate will vary by proposer)	\$51.1	\$69.0	\$51.5		\$171.6
Project Total	\$240.4	\$324.5	\$229.1		\$794.0
Trustee Agency GA (9% of Project Total)	\$21.6	\$29.2	\$20.6		\$71.4
Total Cost	\$262.0	\$353.7	\$249.7		\$865.4
Other Resources:					
<p>Comments: FY 07 was approved at the Trustee Council meeting of 11/14/06; FY 08 was approved in 2007; FY 09 is pending: Actual calendar dates to be covered in FY09 are anticipated as being from March 09 through September 09 (7 mo.)</p> <p>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</p>					

FY 07-09

Project Number: 070811
 Project Title: Prince William Sound Herring Forage Contingency - Submitted under the BAA
 Proposer: T.Kline, PWSSC

Date Prepared: 1 August 2006

**EXXON VALDEZ OILSPILL TRUSTEE COUNCIL
DETAILED BUDGET FORM FY 07 - FY 09**

Contractual Costs:		Contract
Description		Sum
Vessel Charter (purse seiner)		33.6
Photocopying		0.3
PWSSC Network charge (computer-months)		1.4
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		1.6
If a component of the project will be performed under contract, the 4A and 4B forms are required.		Contractual Total
		\$58.5
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
Replacment 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		2.0
		Commodities Total
		\$13.7

FY 07

Project Number: 070811
 Project Title: Prince William Sound Herring
 Forage Contingency - Submitted under the BAA
 Name: T.Kline, PWSSC

**EXXON VALDEZ OILSPILL TRUSTEE COUNCIL
DETAILED BUDGET FORM FY 07 - FY 09**

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

Project Number: 070811
 Project Title: Prince William Sound Herring
 Forage Contingency - Submitted under the BAA
 Proposer: T.Kline, PWSSC

**EXXON VALDEZ OILSPILL TRUSTEE COUNCIL
DETAILED BUDGET FORM FY 07 - FY 09**

Contractual Costs:				Contract
Description	unit cost	units	total cost	Sum
Photocopying				0.0
PWSSC Network charge (computer-months)	\$50	18	\$900	0.5
** Stable isotope analytical (freeze-dry)	\$25	600	\$15,000	0.9
Stable isotope lyophilizer useage	\$4	600	\$2,400	15.0
** Stable isotope analytical (oven-dry)	\$25	200	\$5,000	2.4
Shipping include hazmat				5.0
Communications(fax&phone)				0.4
				0.7
				0.0
(** There was a \$3 increase per sample from 1 July 2008; from \$22 to \$25)				
Contractual Total				\$24.9
Commodities Costs:				Commodity
Description				Sum
calorimeter spare parts kit				0.3
ignition unit				0.4
				0.0
				0.0
				0.0
				0.0
				0.0
Lab supplies- chemicals, gases, vials, bags				2.0
Office supplies				0.5
Computer supplies - software, upgrades				1.5
Commodities Total				\$4.7

FY 09

Project Number: 070811
 Project Title: Prince William Sound Herring
 Forage Contingency - Submitted under the BAA
 Proposer: T.Kline, PWSSC

