EVOSTC FY17-FY21 INVITATION FOR PROPOSALS
FY20 CONTINUING PROJECT PROPOSAL SUMMARY PAGE

Project Number and Title

20120111-E

Herring Disease Program

Primary Investigator(s) and Affiliation(s)

Paul K. Hershberger, USGS – Marrowstone Marine Field Station

Maureen K. Purcell, USGS – Western Fisheries Research Center

Date Proposal Submitted

August 16, 2019

Project Abstract

We will investigate fish health factors that may be contributing to the failed recovery of Pacific herring populations in Prince William Sound. Field samples will provide infection and disease prevalence data from Prince William Sound and Sitka Sound to inform the age structured assessment (ASA) model, serological data will indicate the prior exposure history and future susceptibility of herring to viral hemorrhagic septicemia virus (VHSV), and diet information will provide insights into the unusually high prevalence of *Ichthyophonus* that occurs in juvenile herring from Cordova Harbor. Laboratory studies will validate the newly developed plaque neutralization assay as a quantifiable measure of herd immunity against VHS, provide further understanding of disease cofactors including salinity, and investigate possible routes of transmission for *Ichthyophonus*. Information from the field and laboratory studies will be integrated into the current ASA model and inform a novel ASA-type model that is based on the immune status of herring age cohorts.

EVOSTC Funding Requested* *(must include 9% GA)*

FY17	FY18	FY19	FY20	FY21	TOTAL
197,800	228,900[a]	236,700[a]	243,300[a]	251,100[a]	1,157,900[a]

Non-EVOSTC Funds to be used, please include source and amount per source: (see Section 6C for details)

FY17	FY18	FY19	FY20	FY21	TOTAL
61,600	63,600	64,000	65,200	66,900	321,400

[a]Totals in FY18-21 include additional annual requests of $22,500 that will be used for processing additional herring plasma samples: results will be incorporated into a revised ASA model.

1. PROJECT EXECUTIVE SUMMARY

A better understanding of the epidemiological principles governing herring diseases in Prince William Sound (PWS) is necessary for the development of adaptive management strategies intended to account for and mitigate the effects of diseases to wild herring populations. The incorporation of laboratory-based manipulations and observations in the herring disease program (HDP) has led to the realization that some of our prior assumptions of these diseases were incorrect. For example, in a typical herring population, the prevalence of viral hemorrhagic septicemia virus (VHSV) generally falls below the realistic detection threshold obtained from 60-fish subsamples of a population. Even though the endemic prevalence is typically extremely low, an epizootic can occur very quickly because of changing host and environmental conditions. As such, the incorporation of VHSV prevalence data into the age structured assessment (ASA) model is inconsequential from both population forecasting and epidemiological perspectives. For example, a prevalence of 0% (0/60) in a pre-spawn herring population provides no indication of whether the population previously experienced a viral hemorrhagic septicemia (VHS) epizootic, or whether an epizootic is likely to occur in the future. For this reason, we have developed a serological assay (50% plaque neutralization assay [PNT]) that can be used to determine whether herring have survived previous exposure to VHSV. Once fully validated and vetted, these serological results will elucidate the prior VHSV exposure history and future disease potential; further, these serological results will replace the current VHSV infection prevalence inputs in the ASA model with biologically and ecologically meaningful values. Serological samples for this assay have been incorporated into the annual fish health assessments.

<u>Summary and Highlights since Feb. 2018</u>

1) Results from the 2019 health assessments of pre-spawn herring were consistent with previous years. Overall *Ichthyophonus* infection prevalence was 19% (34/179) in in PWS and 14% (25/179) in Sitka Sound; VHSV was not detected in in any samples from PWS or Sitka Sound; viral erythrocytic necrosis (VEN) was detected at very light levels in 1.7% (3/176) of the samples from PWS but none (n = 180) from Sitka Sound.
2) We are continuing a long-term study to document the kinetics of the VHSV antibody response; particularly, we are assessing how long circulating antibodies remain detectable after herring survive a single exposure to the virus. We are currently 18 months into the experiment (post-virus exposure) and we continue to detect neutralizing antibodies in 70% of the previously exposed herring (Fig. 1).

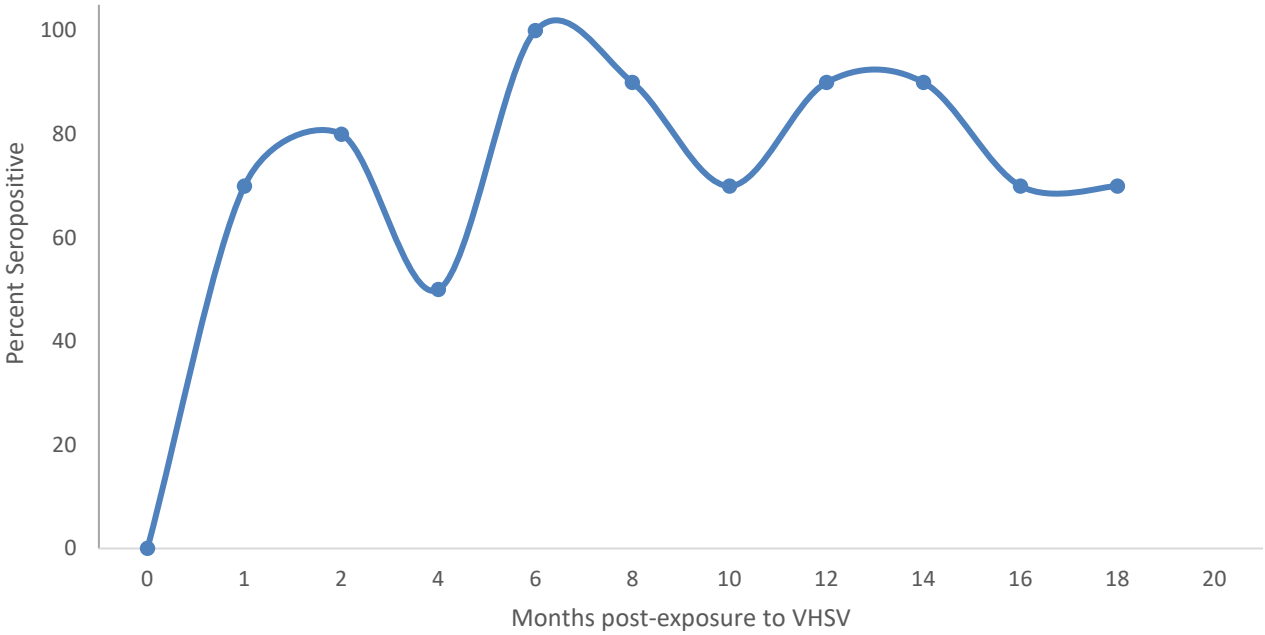


Figure 1. Monthly profile of neutralizing antibodies in Pacific herring that survived viral hemorrhagic septicemia virus exposure. Each data point represents the percent seropositive from 10 subsampled herring. None of the specific pathogen-free negative controls (n = 10 / sampling date) tested positive for neutralizing antibodies. The experimental subsampling is ongoing (April 2019 is represented by month 18). Note: these antibody values may change after the samples are re-processed using the updated plaque neutralization assay methods (see explanation in the text).

3) The final study designed to compare the relative VHS susceptibilities of oiled and unoiled herring has been completed. Like the preliminary studies performed in 2018 (Hershberger and Purcell 2018), cumulative mortality after VHSV exposure was similar between oiled and unoiled herring from all three stocks (PWS, Sitka Sound, and Puget Sound (Figs. 2-4); gene expression results are pending further processing at U.C. Davis.

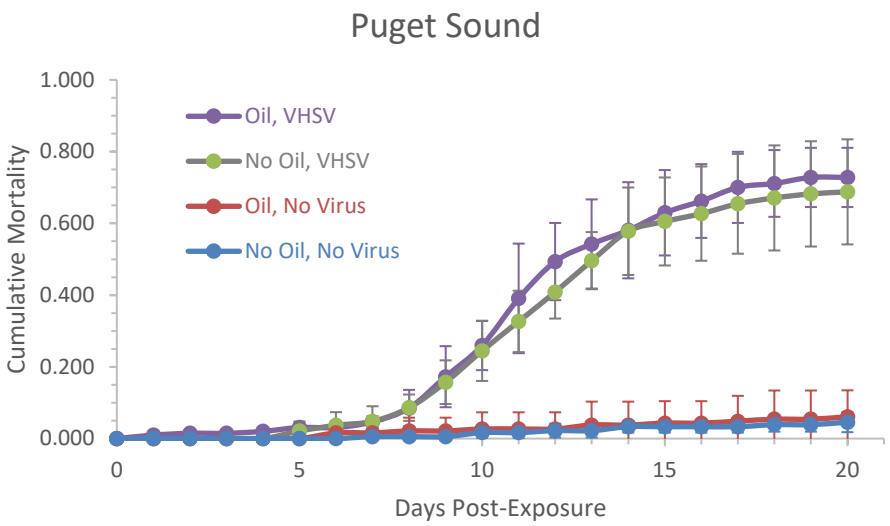


Figure 2. Relative susceptibility to viral hemorrhagic septicemia among Puget Sound herring that survived early life stage exposures to polycyclic aromatic hydrocarbons.

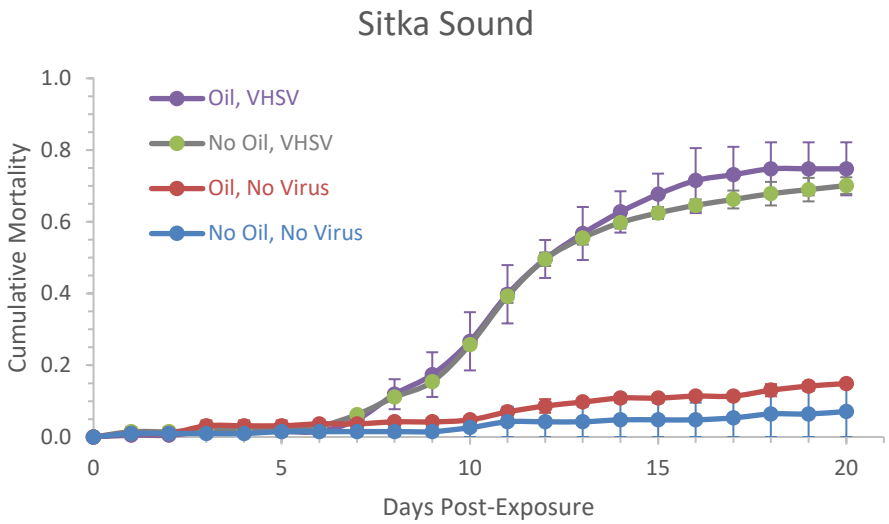


Figure 3. Relative susceptibility to viral hemorrhagic septicemia among Sitka Sound herring that survived early life stage exposures to polycyclic aromatic hydrocarbons.

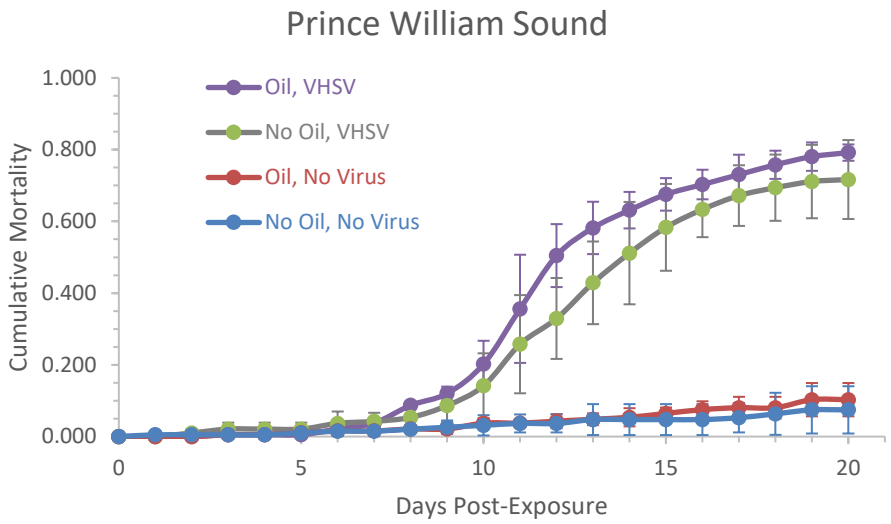


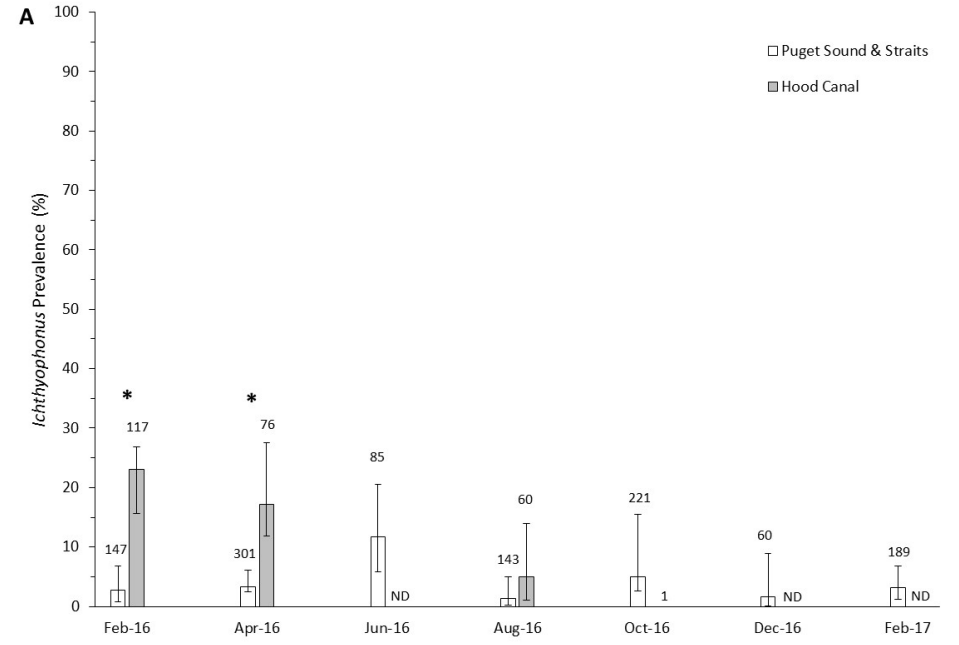
Figure 4. Relative susceptibility to viral hemorrhagic septicemia among Prince William Sound herring that survived early life stage exposures to polycyclic aromatic hydrocarbons.

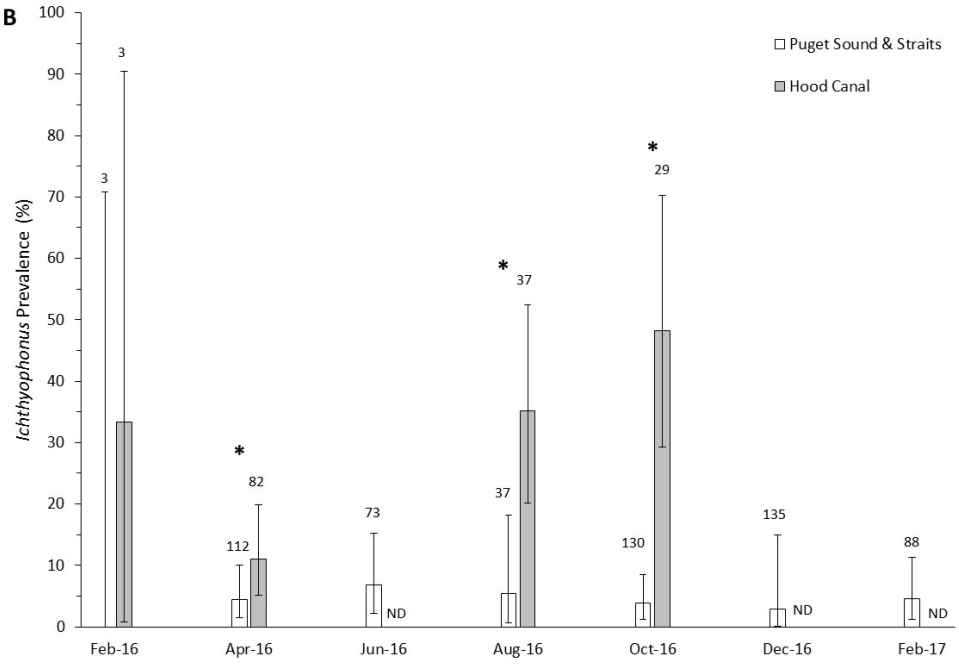
4) Optimization and adjustments to the plaque neutralization test continue. We anticipate proceeding with the validation of the newest methods, beginning July 31, 2019.

As no-cost additions, EVOS TC funding for the Herring Disease Program was leveraged to learn more about the pathogens of concern to Pacific herring. The following products, although not necessarily identified in the study plan, provide new insights into these pathogens and would not have been possible without the core support of the EVOS TC.

- Hershberger, P.K., A.H. MacKenzie, J.L. Gregg, A. Lindquist, T. Sandell, M.L. Groner, D. Lowry. *Accepted*. A Geographic Hot Spot of *Ichthyophonus* infection in the Southern Salish Sea, USA. Diseases of Aquatic Organisms.

The prevalence of *Ichthyophonus* infection in Pacific herring *Clupea pallasii* was spatially heterogeneous in the Southern Salish Sea, WA, USA. Over the course of 13 months, 2,232 Pacific herring were sampled from 38 midwater trawls throughout the Salish Sea. Fork length was positively correlated with *Ichthyophonus* infection at all sites. After controlling for the positive relationship between host size and *Ichthyophonus* infection, the probability of infection was approximately 6X higher in North Hood Canal than in Puget Sound and the northern Straits (12 % versus 2 % predicted probability for a 100 mm fish and 30 % versus 7% predicted probability for a 180 mm fish; Fig. 5A-C). Temporal changes in *Ichthyophonus* infection probability were explained by seasonal differences in fish length, owing to Pacific herring life history and movement patterns. Reasons for the spatial heterogeneity remain uncertain but may be associated with density dependent factors inherent to the boom-bust cycles that commonly occur in Clupeid populations.


A
Puget Sound & Straits
Hood Canal
Ichthyophonus Prevalence (%)
100
90
80
70
60
50
40
30
20
10
0
*
*
117
76
85
60
221
60
189
147
301
143
1
ND
ND
ND
Feb-16
Apr-16
Jun-16
Aug-16
Oct-16
Dec-16
Feb-17


B
Puget Sound & Straits
Hood Canal
Ichthyophonus Prevalence (%)
100
90
80
70
60
50
40
30
20
10
0
3
3
*
29
*
37
*
82
73
37
135
88
130
112
ND
ND
ND
Feb-16
Apr-16
Jun-16
Aug-16
Oct-16
Dec-16
Feb-17

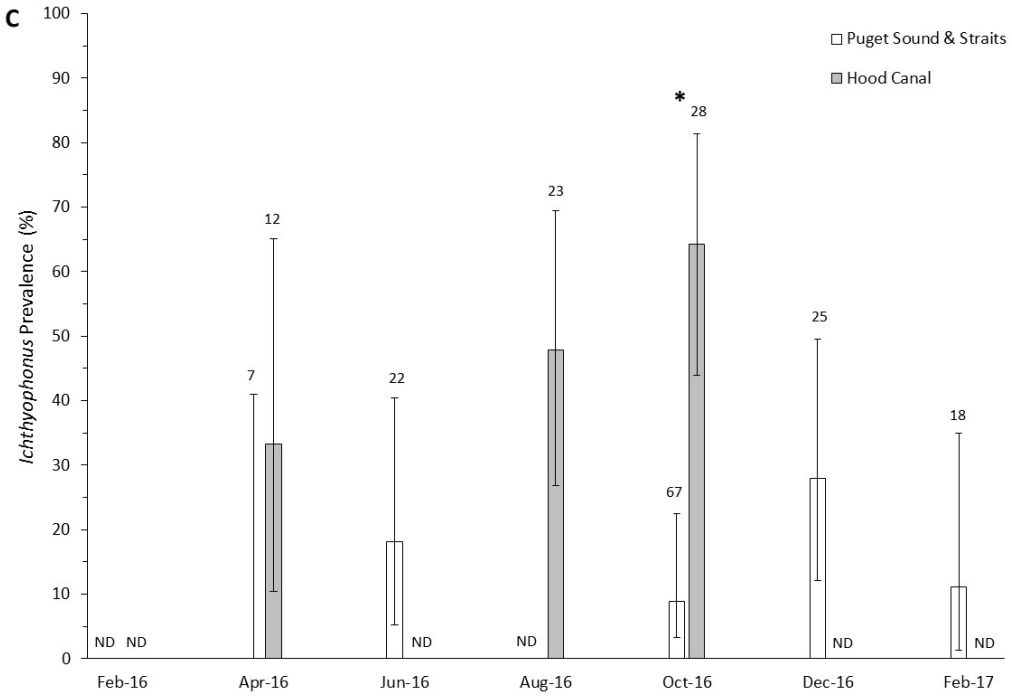


Figure 5. (A-C) Bi-monthly *Ichthyophonus* infection prevalence among the smallest (<150 mm; 5A), medium size (150 – 179 mm; 5B) and largest (<180 mm; 5C) herring. Error bars represent 95% confidence intervals and numerals above the error bars indicate n. "ND" (no data) indicates sampling blocks where no representatives were sampled. * indicates bimonthly sampling blocks where infection prevalence was significantly higher ($P < 0.05$; χ^2) in Hood Canal than in Puget Sound / Straits.

- New Book Chapter Accepted:

Burge, C.A., P.K. Hershberger. Chapter 9: Climate Change and Marine Disease. *In:* Marine Disease Ecology. B. Sillman, K. Lafferty, and D. Behringer (Eds.)

- New Manuscript Submitted:

Gross, L., J. Richard, P. Hershberger, K. Garver. *In Review*. Low susceptibility of sockeye salmon (*Oncorhynchus nerka*) to viral hemorrhagic septicemia virus genotype IVa. Diseases of Aquatic Organisms.

The susceptibility of Atlantic salmon and sockeye salmon to VHSV-IVa was evaluated using exposure routes including injection, static immersion, and cohabitation with diseased Pacific herring *Clupea pallasii*. Exposed fish were monitored for mortality and external pathology, mortalities were tested by cell culture, and live fish were regularly sampled and screened for infection. Among injected sockeye, susceptibility was extremely low, as VHSV was detected in one mortality (n = 195) and two sub-sampled fish (n = 30); further, sockeye exposed by immersion and cohabitation did not experience mortality nor was systemic infection indicated by tissue screening. Injection and cohabitation exposure routes confirmed the susceptibility of Atlantic salmon to VHSV. Neither sockeye nor Atlantic salmon surviving the cohabitation served as a reservoir of VHSV, but Pacific herring did. The results suggest that VHSV-IVa poses low risk to sockeye salmon under natural routes of exposure and confirmed the results of

previous studies that Pacific herring and Atlantic salmon demonstrate high and moderate susceptibility, respectively.

- New Manuscript Submitted:

Sitkiewicz, S.E., N. Wolf, P.K. Hershberger, T. Scott Smettz, S.R. Webster, B.P. Harris. *In Review*. Temporal changes in *Ichthyophonus* infection prevalence in Pacific halibut provide evidence for a stable host pathogen paradigm. Journal of Fish Diseases.

The prevalence of *Ichthyophonus* infections in sport-caught Pacific halibut landed at Homer, AK increased from 19-63% from 2011-2018 (Fig. 6). Additionally, a shift in demographic patterns occurred during 2017-2018, when infection prevalence increased among the youngest and decreased among the oldest age cohorts. Despite the high infection prevalence in halibut from the waters around Homer, there was no indication that the parasite caused damage to the host, as significant relationships were not detected between host infection status and sex, age, length-at-age, or fish condition. Preliminary assessments of infection severity using several methods, including histopathology, bioelectrical impedance, and parasite density, indicated uniformly light infections. These results, when combined with analogous observations of rapid changes in infection prevalence throughout the NE Pacific Ocean, provide evidence for a stable host/pathogen paradigm characterized by light infections that are fully or partially cleared by the host.

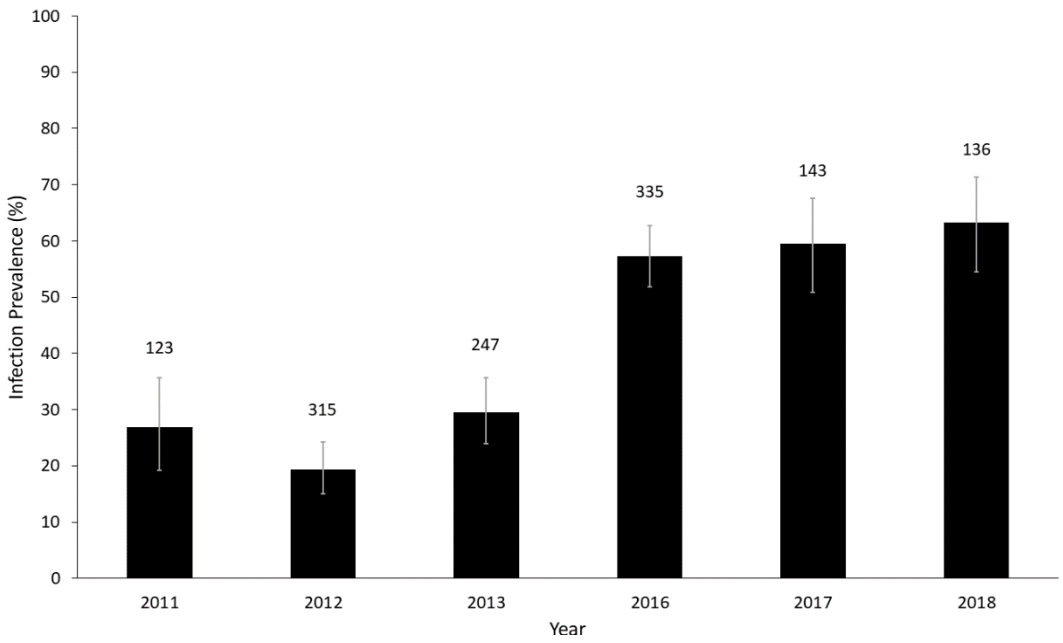


Figure 6. Inter-annual changes in *Ichthyophonus* infection prevalence in Pacific halibut captured near Homer, AK.

2. PROJECT STATUS OF SCHEDULED ACCOMPLISHMENTS

A. Project Milestones and Tasks

Table 1. Project milestones and task progress by fiscal year and quarter, beginning February 1, 2017. Yellow highlight indicates proposed fiscal year workplan. Additional milestones and/or tasks have been added in red. C = completed, X = not completed or planned. Fiscal year quarters: 1 = Feb 1 – April 30; 2 = May 1 – July 31; 3 = Aug. 1 – Oct. 31; 4 = Nov. 1 – Jan. 31.

	FY17				FY18				FY19				FY20				FY21			
Milestone/Task	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
Fieldwork																				
Collect herring eggs for rearing SPF colonies	C				C				C				X				X			
Collect adult herring to assess annual infection and disease prevalence	C				C				C				X				X			
Collect zooplankton for investigation of possible *Ichthyophonus* intermediate host	C				C				X				X				X			
Collect herring from Cordova Harbor to assess *Ichthyophonus*-infected offal in the stomach bolus		C				X*			C											
Captive Study																				
Vibrio challenge experiments	C	C	C	C																
SPF herring metamorphosed to juveniles			C				C				C				X				X	
Assess effects of salinity on fish-to-fish transmission of *Ichthyophonus*					C	C	C	C												
Assess the effect of temperature on VHSV shedding									C	C	C	X								
Lab Analysis																				
Process spring adult herring to determine infection and disease prevalence		C				C				C				X				X		
Validate the plaque neutralization assay using wild herring		C	C			C	C			C	C			X	X			X	X	
Analysis of plasma samples			C				C				C				X				X	
Analysis of archived plasma samples from 2012-16				C																
Histological processing of herring from Cordova Harbor to assess *Ichtyophonus*-infected offal in the stomach bolus																X				
PCR and CISH analyses of zooplankton samples for *Ichtyophonus*																			X	
Data																				
Share 2012-16 plasma sample data with modelers				C																
Upload data and metadata to Workspace				C				C				X				X				X

Milestone/Task	FY17				FY18				FY19				FY20				FY21			
	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
Reporting																				
Annual report					C				C				X							
FY work plan (DPD)			C								C				X					
FY17-21 Final Report																			X	X
Meetings & Conferences																				
Annual PI meeting				C				C				X				X				X
Publications			C	C		C	C	C			C									

B. Explanation for not completing any planned milestones and tasks

2019 Zooplankton collections: Collections were discontinued: see explanation in section 'C' below.

2018 Cordova Harbor herring: Juvenile herring were not present in Cordova Harbor during 2018. Therefore, although sampling was attempted, none were collected. However, juvenile herring did occur in Cordova Harbor during 2019, and monthly samples (including fish, sediment, and water) were collected. These samples are currently being processed; we anticipate submission of a manuscript describing novel molecular methods to detect *Ichthyophonus* in sediment samples by the end of FY19. An additional manuscript describing the changes in infection prevalence in fish is expected to be ready for submission in spring 2020.

C. Justification for new milestones/tasks

To evaluate the possibility of an intermediate host in the *Ichthyophonus* life cycle, we processed numerous zooplankton samples using *Ichthyophonus*-specific qPCR primers. To date, these efforts have not returned consistent results, and we do not feel confident reporting any true positives. Additionally, to focus our surveillances on a copepod with a high likelihood of exposure to *Ichthyophonus*, we sampled sea lice from the flanks of herring demonstrating external ulcers from ichthyophoniasis (Fig. 7); these lice were grazing on mucus and *Ichthyophonus* life stages that occurred on the external surfaces of heavily infected wild herring. These lice samples also returned inconclusive results using the *Ichthyophonus* primers. Therefore, until we have more convincing evidence that an intermediate host is involved with this parasite life history, we will be investigating possible alternative routes of *Ichthyophonus* transmission to Pacific herring (described in Section 4B).

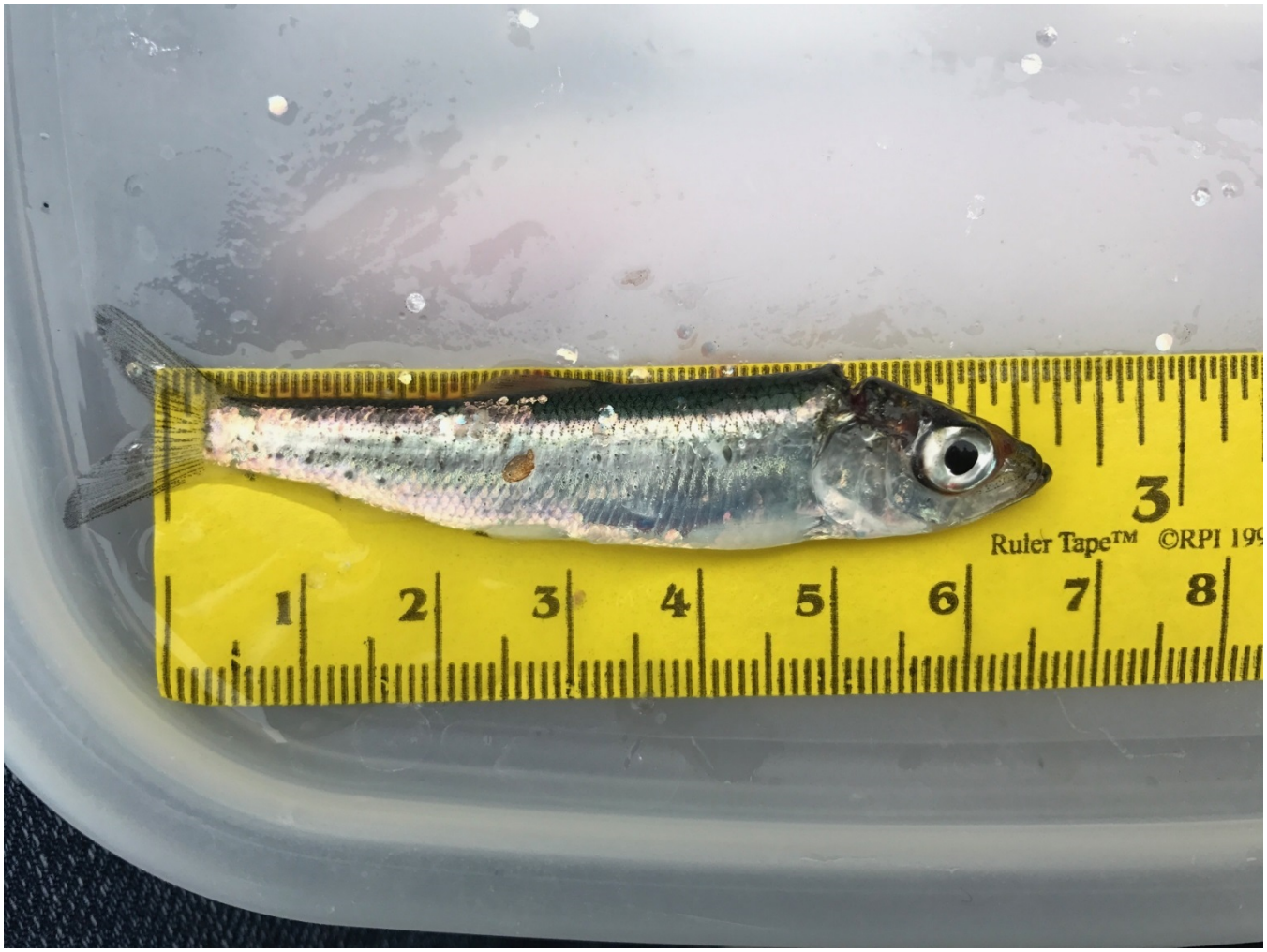


Figure 7. Sea louse (*Caligus clemensi*) grazing on the flank of a wild herring demonstrating external ulcers from ichthyophoniasis. Analysis of this and other lice using *Ichthyophonus*-specific primers returned ambiguous results that could not confirm the presence of the parasite in association on the lice.

3. PROJECT COORDINATION AND COLLABORATION

A. Within an EVOSTC-funded Program

Herring Research and Monitoring

- We are working closely with PWS Science Center (#19120111-G) and the Alaska Department of Fish and Game (19160111-F) to collect herring tissue and plasma samples during the spring herring cruises (shared research platforms). Additionally, ADF&G continues to provide age data for the fish health samples.
- As In-Kind contributions to Dr. Andrew Whitehead's project (#19170115), we provided samples for their reference genome, completed a series of experimental exposures of Specific Pathogen -Free (SPF) herring to VHSV, and initiated an experimental study to assess the relative susceptibility of oil-exposed herring to *Ichthyophonus*.
- Serum neutralization results, to assess herd immunity by quantifying VHSV neutralizing titer, will be shared with Dr. Trevor Branch (#19120111-C). These results will be used to create a novel age-structured assessment model that incorporates herd immunity by herring age class.

- As In-Kind contributions to Dr. Maya Groner's project (#19120111-A), several experiments were initiated and are currently underway at the U.S. Geological Survey (USGS) - Marrowstone Marine Field Station.
    - An *in vivo* study was initiated to evaluate the histological infection threshold associated with herring mortality from ichthyophoniasis.
    - Archived histology samples were processed from PWS and Sitka Sound, dating to 2007. All culture-positive samples have now been embedded, sectioned, and stained. We are in the final stages of creating composite images of the sections (anticipated completion Aug 31, 2019); data analysis is expected to begin in the 4th quarter of FY19.

Gulf Watch Alaska

Researchers in Gulf Watch Alaska (GWA) Nearshore project (20120114-H; Jim Bodkin, Heather Coletti, Brenda Ballachey, and Elizabeth Bowen) have been very kind to provide samples of razor clams from Katmai and Kenai Fjords. These Alaskan razor clams are extremely valuable for us in the development of molecular tools to detect the bacterium responsible for nuclear inclusion-X (NIX) disease. Although this NIX project is not directly part of the HDP, obtaining these samples would not have been possible without the professional relationships we were fortunate to build between the Herring Research and Monitoring (HRM) program and GWA.

Data Management

Infection prevalence data from PWS and Sitka Sound are provided to the data management team annually.

B. With Other EVOSTC-funded Projects

Nothing to report.

C. With Trustee or Management Agencies

- We continue to partner with ADF&G – Cordova to collect herring infection and disease data onboard the shared ADF&G seining platform.
- We continue to partner with ADF&G – Sitka to collect herring infection and disease data from pre-spawn aggregations in Sitka Sound.
- We continue to partner with ADF&G – Juneau to provide consistent virologic methods between all *Exxon Valdez* Oil Spill Trustee Council (EVOSTC) funded herring disease projects between 1994 – present.
- We have partnered with ADF&G – Sitka & Juneau to assess whether temporal changes in the severity of *Ichthyophonus* infections may be responsible for recent declines in the spawning herring biomass and age structure. Data and archived samples from the past 10 years of this EVOSTC-funded project were leveraged to obtain supplemental funding from the North Pacific Research Board (NPRB; # 1807: *Ichthyophonus* in Pacific Herring).
- We have partnered with Drs. John Incardona and Nat Sholtz (National Oceanic and Atmospheric Administration – Northwest Fisheries Science Center) to provide herring for their NPRB project investigating the long-term effects of embryonic oil exposure on herring cardiac morphology. Further, we are investigating the long-term impacts of these cardiac abnormalities on the health and survival of juvenile herring.

4. PROJECT DESIGN

A. Overall Project Objectives

FY20 Objectives

I. Provide pathogen and disease prevalence data to inform the ASA model

II. Produce specific pathogen-Free (SPF) Pacific herring for laboratory experiments

III. Process new and archived herring plasma samples for indications of prior VHSV exposure

IV. Validate the novel plaque neutralization assay using wild herring

V. Changed from: Investigate the possibility of an invertebrate host for *Ichthyophonus*

Changed to: Investigate the possibility of *Ichthyophonus* transmission through the consumption of fish eggs containing the parasite

VI. Determine the causes for abnormally high *Ichthyophonus* prevalence among juvenile Pacific herring that establish temporary residency in Cordova Harbor

B. Changes to Project Design and Objectives

Change in Objective V. Investigate the possibility of an invertebrate host for *Ichthyophonus*.

The goal of this objective was to identify the mechanism(s) by which *Ichthyophonus* transmission occurs in Pacific herring. We have committed considerable effort towards examining the possibility of intermediate hosts; however, these efforts have not produced consistent results, even after selecting for copepods (i.e., sea lice) with known exposures to the parasite. Therefore, a more productive approach is warranted for investigating possible *Ichthyophonus* transmission routes (described below).

A new parsimonious transmission hypothesis, based on several observations, has been developed and is ready for testing. During our spring sampling trip to Sitka Sound in 2019, Drs. Groner and Hershberger were fortunate to meet with Dr. Ron Heintz to discuss some his recent findings relating to overwintering herring diets and bioenergetics. We were excited to learn that a large percentage of overwintering herring in PWS and the Gulf of Alaska are ovivorous (consume fish eggs). Although the species of fish eggs observed in the herring stomachs was not conclusively determined (the stomach samples and eggs are no longer available), they were most likely pelagic walleye pollock eggs. Because pollock represent a common host for *Ichthyophonus*, we are interested in investigating whether consumption of these eggs may represent an effective transmission route for Pacific herring. We will begin exploring this hypothesis by discussing it with research partners in GWA and HRM at the 2019 PI meeting in Homer, where we will ask for assistance with field sampling from existing projects and platforms. Ideal samples would include:

- Gravid *Ichthyophonus*-infected pollock to determine whether the parasite occurs in association with the eggs
- Overwintering herring with fish eggs in their stomachs to conclusively determine the egg species and assess whether *Ichthyophonus* occurs in association with the consumed eggs
- January – March plankton tows to determine what eggs are available in the water column

Further support for this ovivory hypothesis is provided by consistent observations of herring collected from active spawning aggregations containing large quantities of herring eggs in their stomachs. We will also

investigate the possibility of parasite transmission via consumption of herring eggs by feeding eggs from *Ichthyophonus*-infected pre-spawn female herring to our colonies of laboratory-reared specific-pathogen-free (SPF) herring. We will perform analogous exposures using eggs from infected pollock if they become available.

5. PROJECT PERSONNEL – CHANGES AND UPDATES

No staffing changes to report.

6. PROJECT BUDGET

A. Budget Forms (See GWA FY20 Budget Workbook)

EXXON VALDEZ **OIL SPILL TRUSTEE COUNCIL**
PROGRAM PROJECT BUDGET PROPOSAL AND REPORTING FORM

Budget Category:	Proposed FY 17	Proposed FY 18	Proposed FY 19	Proposed FY 20	Proposed FY 21	TOTAL PROPOSED	ACTUAL CUMULATIVE
Personnel	$122.4	$140.9	$148.1	$154.1	$161.3	$726.8	
Travel	$20.1	$20.1	$20.1	$20.1	$20.1	$100.5	
Contractual	$0.0	$0.0	$0.0	$0.0	$0.0	$0.0	
Commodities	$39.0	$49.0	$49.0	$49.0	$49.0	$235.0	
Equipment	$0.0	$0.0	$0.0	$0.0	$0.0	$0.0	
SUBTOTAL	$181.5	$210.0	$217.2	$223.2	$230.4	$1,062.3	
General Administration (9% of subtotal)	$16.3	$18.9	$19.5	$20.1	$20.7	$95.6	
PROJECT TOTAL	$197.8	$228.9	$236.7	$243.3	$251.1	$1,157.9	
Other Resources (Cost Share Funds)	$61.7	$63.6	$64.0	$65.2	$66.9	$321.4	

B. Changes from Original Project Proposal

An additional $22,500 / year (FY 19-21) is requested to enable the processing of additional herring plasma samples from PWS. This supplement will provide funds for additional plaque neutralization supplies ($10,000), 2.5 months of support for a seasonal technician to assist with the processing of field samples ($12,500), and 9% General Administration charges ($2,000).

These expanded samples (in addition to the 180 random samples from the AWL collections) will be size-selected and are intended to provide more robust sample sizes for each available herring age class. The resulting reduction in variability around the age-specific immunity is needed by Dr. Branch for the revised ASA modelling.

C. Sources of Additional Project Funding

- USGS provides matching funds for PIs and support staff as part of their base appropriation.
- The HDP was leveraged to obtain NPRB funding (Groner and Hershberger) to evaluate the possible involvement of *Ichthyophonus* in the recent herring population trends occurring in Sitka Sound.

NORTH PACIFIC RESEARCH BOARD
SUBAWARD AGREEMENT

1. Scope of Work

(a) The North Pacific Research Board (NPRB) and USGS listed below, jointly and severally agree to the provisions herein and to perform the work described in Appendix 1, which contains a statement of work, schedule, and budget for the following project recommended for approval by NPRB on May 9, 2018, and approved by the Secretary of Commerce on May 14, 2018:

PROJECT NUMBER:	1807
PROJECT TITLE:	Is shifting epidemiology of Ichthyophonus increasing mortality in Pacific herring?
RECIPIENT:	U.S. Geological Survey Western Fisheries Science Center 6505 NE 65th St. Seattle, Washington, 98115
RECIPIENT DUNS:	Exempt
PRINCIPAL INVESIGATOR:	Paul K Hershberger phershberger@usgs.gov
PERIOD:	September 1, 2018 – August 31, 2020
TOTAL SUBAWARD:	$94,928 (Uniform Guidance 2.C.F.R Part 200 applies)

Project Partners Under Separate Agreement

Prince William Sound Science Center:	$43,708
Project TOTAL:	**$138,636**

7. FY17-19 PROJECT PUBLICATIONS AND PRODUCTS

Publications

Hart, L.M., M.K. Purcell, R. Powers, A. MacKenzie, P.K. Hershberger. 2017. Optimization of a plaque neutralization test to identify the exposure history of Pacific herring to viral hemorrhagic septicemia virus (VHSV). Journal of Aquatic Animal Health 29: 74-82.

Hart, L.M., N. Lorenzen, K. Einer-Jensen, M. Purcell, P.K. Hershberger. 2017. Influence of temperature on the efficacy of homologous and heterologous DNA vaccines against viral hemorrhagic septicemia (VHS) in Pacific herring. Journal of Aquatic Animal Health 29: 121-128.

Hershberger, P.K., J.L. Gregg, C. Dykstra. 2018. High-prevalence and low-intensity *Ichthyophonus* infections in Pacific Halibut (*Hippoglossus stenolepis*). Journal of Aquatic Animal Health 30:13-19.

Harris, B.P., S.R. Webster, J.L. Gregg, P.K. Hershberger. 2018. *Ichthyophonus* in sport-caught groundfishes from southcentral Alaska. Diseases of Aquatic Organisms 128: 169-173.

Lowe, V.C., P.K. Hershberger, C.S. Friedman. 2018. Analytical and diagnostic performance of a qPCR assay for *Ichthyophonus* spp. compared to the tissue explant culture 'gold standard'. Diseases of Aquatic Organisms 128: 215-224.

Hershberger, P.K., A.H. MacKenzie, J.L. Gregg, A. Lindquist, T. Sandell, M.L. Groner, D. Lowry. *Accepted*. A Geographic Hot Spot of *Ichthyophonus* infection in the Southern Salish Sea, USA. Diseases of Aquatic Organisms.

Gross, L., J. Richard, P. Hershberger, K. Garver. *In Review*. Low susceptibility of sockeye salmon (*Oncorhynchus nerka*) to viral hemorrhagic septicemia virus genotype IVa. Diseases of Aquatic Organisms.

Sitkiewicz, S.E., N. Wolf, P.K. Hershberger, T. Scott Smettz, S.R. Webster, B.P. Harris. *In Review*. Temporal changes in *Ichthyophonus* infection prevalence in Pacific halibut provide evidence for a stable host pathogen paradigm. Journal of Fish Diseases.

Four additional manuscripts are currently in preparation; submission to the journals is anticipated prior to the end of FY'19.

Published and updated datasets

Metadata and data describing infection prevalence results from herring health surveillance have been provided to Axiom annually.

Presentations

Hershberger, P.K., R.L. Powers, B.L. Besijn, J. Rankin, M. Wilson, B. Antipa, J. Bjelland, A.H. MacKenzie, J.L. Gregg, M.K. Purcell. June 17-20, 2019. Platform. Intra-annual variability in waterborne *Nanophyetus salmincola*. AFS Fish Health Section Annual Meeting and 60th Western Fish Disease Workshop. Ogden, UT.

Groner, M., E. Bravo, C. Conway, J. Gregg, P. Hershberger. January 28-31, 2019. Poster. A quantitative histological index to differentiate between endemic and epidemic ichtyhophoniasis in Pacific herring. Alaska Marine Science Symposium. Anchorage, AK.

Wendt, C., P. Hershberger, C. Wood. January 28-31, 2019. Poster. Patterns of *Ichthyophonus* sp. infection in age zero Pacific herring. Alaska Marine Science Symposium. Anchorage, AK.

Cypher, A.D., P. Hershberger, N. Scholz, J.P. Incardona. January 3-7, 2019. Larval cardiotoxicity and juvenile performance are likely contributors to the delayed fishery collapse of Pacific herring after the *Exxon Valdez* oil spill. Society for Integrative & Comparative Biology Annual Meeting. Tampa, FL.

Bravo, E., C. Conway, P. Hershberger, J. Gregg, M. Groner. October 11-13, 2018. Poster. Do histological analyses of herring infected with *Ichthyophonus* sp. suggest a shift from endemic to epidemic disease? Society for the Advancement of Chicanos / Hispanics and Native Americans in Science. San Antonio, TX.

Sitkiewicz, S., P. Hershberger, N. Wolf. B. Harris. January 22-26, 2018. Poster. Effects of the parasite *Ichthyophonus* (spp.) on Pacific halibut (*Hippoglossus stenolepis*) growth and condition. Alaska Marine Science Symposium. Anchorage, AK.

MacKenzie, A.H., J.L. Gregg, M.D. Wilmot, T. Sandell, D. Lowry, P.K. Hershberger. June 20-22, 2017. Poster. Temporal and spatial patterns of *Ichthyophonus* in Pacific herring throughout the southern Salish Sea. 58th Western Fish Disease Workshop. Suquamish, WA.

Sitkiewiz, S.E., B.P. Harris, P.K. Hershberger, N. Wolf. June 20-22, 2017. Poster. Effects of the parasite *Ichthyophonus* on groundfish growth and condition. 58th Western Fish Disease Workshop. Suquamish, WA.

Hershberger, P.K., A.H. MacKenzie, J.L. Gregg, M.D. Wilmot, R. Powers, M.K. Purcell. June 20-22, 2017. Platform. Long term shedding of viral hemorrhagic septicemia virus from Pacific herring. 58th Western Fish Disease Workshop. Suquamish, WA.

Sitkiewicz, S., B. Harris, P. Hershberger, N. Wolf. March 19-23, 2017. Poster. Impacts of the Parasite *Ichthyophonus* (sp.) on Groundfish Growth and Condition. Joint Meeting of the American Fisheries Society, Alaska Chapter American Water Resources Association, Alaska Section. Fairbanks, AK.

Hershberger, P.K., L. Hart, A. MacKenzie, R, Powers, M. Purcell. January 23-27, 2017. Poster. Quantifying the potential for disease impacts to Pacific Herring. Alaska Marine Science Symposium. Anchorage, AK.

Sitkiewicz, S., B. Harris, P. Hershberger, N. Wolf. January 23-27, 2017. Poster. Effects of the parasite *Ichthyophonus* (sp.) on groundfish growth and condition. Alaska Marine Science Symposium. Anchorage, AK.

Outreach

Information about the herring disease program is included on the USGS - Marine Field Station (https://www.usgs.gov/centers/wfrc/science/marrowstone-marine-field-station-mmfs?qt-science_center_objects=0#qt-science_center_objects) and Prince William Sound Science Center (https://pwssc.org/herring/) web pages.

LITERATURE CITED

Hershberger, P.K., M.K. Purcell. 2018. EVOSTC Annual Report – Herring Disease Program 18120111-E. 25 pp.