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*"Our mission: To protect and restore marine and terrestrial ecosystems through scientific research and local community, place based partnerships."*

### **Observations of sea lice on juvenile forage fish along the northwest Salish Sea**

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## Abstract

We summarize 22 years of observations of juvenile forage fish and sea lice from north coastal Washington state. Only juvenile herring were observed over multiple years with sea lice beginning in 2004, after which the prevalence varied but trended to increase annually. Larger schools of juvenile herring were observed to have proportionally more lice and singletons never had lice. The findings documented in this study raise a number of important questions including sea lice species, infection source, and impact of sea lice on young of the year herring fitness and survival. These initial observations provide important and unique information that can only be attained from behavioral data collected underwater. Given documented impacts of sea lice on fitness and mortality of juvenile salmon, and the importance of herring for Northeast Pacific marine systems, more detailed study, including species and source of herring sea lice infection, is a priority.

## Introduction

Herring (*Cupea pallasii*), sand lance (*Ammodytes personatus*), and surf smelt (*Hypomesus pretiosus*), collectively known as ‘forage fish’ are critical components of Northeast Pacific marine systems. These forage fish have complex life histories that include seasonal migratory patterns to and from nearshore spawning grounds.

Sea lice are a guild of isopod and copepod marine ectoparasites, colloquially known as ‘sea lice’ that occur on salmon. Historically their prevalence on wild and juvenile salmon in the northeast Pacific was low, but has increased significantly over the last quarter of a century (Costello 2009; Morton et al 2004). Sea lice infestations on juvenile salmon compromise the young fish’s health and fitness, and ultimately, survival (Webster et al 2007, Costello 2009). Some estimate salmon population collapse with parasite density of 1.5 parasites per fish (Krkošek et al 2007). Historically these parasites have not been a concern for wild juvenile fish, however sea lice have been observed to reach lethal densities with increasing frequency specifically and significantly related to net pens. Fish farms, or net pens, are now considered a disease source for them (Morton et al 2008, Costello 2009;).

Herring are a critical component to coastal ecosystems of the north east Pacific, and an important recreational and commercial resource. However, little is known about sea lice on forage fish. Morton et al. (2008) noted sea lice, (*Lepeophtheirus salmonis* and *Caligus Spp*) . on herring larvae along the coast of British Columbia, Canada. Beamish et al (2009) noted high concentrations of ectoparasites on herring in the Strait of Georgia. We have observed juvenile forage fish, including those with sea lice infestations, as a component of our long term surveys of two sites along the central Strait of Juan de Fuca over the last 22 years. In this paper, we summarize our long-term observations of ectoparasites on forage fish from 1996-2017 along Washington coast. We provide recommendations on priorities to further understand the relationship and risks of salmon net pens to forage fish, and herring in particular.

## Methods and materials

Data were extracted from snorkeling dive notes recorded from two transect sites 1996-2017. The transects were set depth that parallel the contour 0-20 feet along the Crescent Bay and Freshwater Bay shorelines (Figure 1). Transects sampling occurred during daylight hours, with a visibility of at least 3 meters, and were completed within 1-1.5 hours. During surveys visual observations of forage fish species, abundance, and size were noted along the transect by general nomenclature and school size of four general categories (Table 1), and fish sizes visually estimated and binned into 10 cm categories. Immediately after the visual survey was concluded summary notes were recorded including visibility, forage fish species, adult/juvenile, approximate size, and general abundance. When observed, percent of fish with parasites sea lice was also recorded. Transects were conducted for at least once a year from 1996-2004, and at least quarterly from 2004-2017 (Table 2). Fish were present than in summer months but not in winter, so many more transects were conducted during summer. Data were therefore averaged by month and year. No sea lice were observed before 2004 (Table 4)

We used generalized linear models to create a time series describing the presence of lice on herring. In these models, the unit of replication was a school of herring, the response variable was the presence of lice on any herring in a school, and we parameterized (1) year as a categorical fixed effect to create the time series, (2) site as a fixed effect, and (3) the log-transformed school size as an offset to account for the premise that larger schools are more likely to include an individual with lice. We did not include an intercept parameter so that the year

parameter alone described variance in lice presence among years. Sites are typically treated as random effects, but should be treated as fixed effects when there are fewer than 5-6 replicates as models poorly account for random effects with few replicates (Bolker et al. 2009). We fit these models using a binomial distribution and a log-link and used backwards selection and AIC comparisons to refine models (Zuur et al. 2009). To visualize the time series, we predicted the presence of lice each year using the mean school size of herring for input into the offset parameter.

## Results

A total of 237 visual transects were conducted over the 22-year period. Due to seasonal presence of fish and field conditions, the majority of surveys were conducted during spring and summer months. Forage fish were, with only rare exception, observed from May to September, with the majority observed during June through August. (Table 2). Juvenile herring and sand lance were by far the most prevalent forage fish observed over the course of the study (Table 2). Juvenile and adult surf smelt were also observed seasonally but in much smaller numbers. Juvenile gadids were also seen some years. Juvenile herring and sand lance size also varied with year (Table 3).

Sea lice were not observed on juvenile herring before 2004. After 2004 sea lice prevalence varied quite a bit by year (Figure 2). Only one sand lance was observed with sea lice over the course of the study (Figure 2). No juvenile surf smelt were observed with sea lice over the course of the study.

Models quantified an increase in lice presence on herring over time. Backwards selection and comparison of AICs identified the parsimonious model including year as the only fixed effect ( $\Delta AIC$  between models including and omitting site = 2.9). Quantitative model predictions of the time series were similar to observations of the raw values (Table 4, Fig. 3). Lice were absent from 1996-2000 but, assuming an average school size of 6,395 fish, were present on at least one fish in about half of the schools after 2010.

## Discussion

Juvenile forage fish abundance in the nearshore is very seasonal. Juvenile herring abundance and timing in general is consistent with predictions of Snauffer (2013) model of larval herring and hake (*Merluccius productus*) distribution, who concluded that wind patterns are the driving factor for larval herring and hake distribution in the Salish Sea. Sea lice presence and percent cover varied significantly by year. Sea lice were not observed before 2004, and then consistently, and sometimes heavily, in subsequent years. This may be the result of the multiple environmental factors of herring spawning time, larval emergence and environmental factors driving larval distribution. It may also reflect an increase in sea lice density in the environment. As larval herring migrate away from their natal spawning beds and migrate along shore they are exposed to sea lice, including high densities of sea lice associated with net pens along their migration route, including the San Juan Islands and Port Angeles Harbor (Figure 4-5). Rees et al 2015 found that sea lice infection pressure from Atlantic salmon farms on wild juvenile salmon extended throughout most of the study region, was highest near the fish farms, and that sea lice infections on wild juvenile salmon were predicted to be highest in main channels. Patanasatienkul et al 2013 determined a relationship between size of juvenile salmon and parasite intensity, along with large interannual variability in sea lice infestations.

Such infestations are well documented to have a significant effect on juvenile salmon. Webster et al. (2007) and Godwin et al. (2015) documented that sea lice can decrease fitness in juvenile salmon by inducing physiologic stress, invoking behavioral changes, and decreasing competitiveness. The decreased fitness may in turn result in high mortality. Assuming that forage fish experience the same effects as salmon, possibly amplified by their strong schooling life history strategy, such exposure and mortality may play a role in the observed declines in juvenile herring stocks in the region such as those observed along the Strait of Juan de Fuca and Cherry Point/ Admiralty Inlet (Stick et al 2014).

Details on the species of the sea lice observed is important. Unfortunately, it is necessary to examine sea lice closely to determine species, and so not possible thru these field observations. Further, sea lice will often drop from their host fish when handled, making consistent observations from herring catches using standardized fishery sampling techniques impossible.

While challenging, such detailed information is important to acquire. Some species of sea lice are global in distribution, others are specifically associated with Atlantic salmon (*S. salar*), and so introduced to the west coast through net pens. Our work indicates that sea lice are a new/recent phenomenon on juvenile forage fish, and their prevalence on juvenile herring is appears to be growing.

It is very important to define the source of sea lice infestations for young of the year forage fish documented in this study. Costello (2009) provides an informative overview of sea lice, and states that “Persistent infestations on farms increase the risk of lice transferring to wild fishes” including levels that could extirpate wild pink salmon populations in four generations (eight years). In Norway, ectoparasites are credited with wiping out 10% of total Norwegian wild salmon populations annually (Thorstad and Finstad 2018). In British Columbia researchers have documented linkages between net pens and sea lice prevalence (Krkošek et al. 2005).

Our documentation of persistent, multi-year prevalence of sea lice on young of the year herring, combined with the published linkage between net pens and sea lice prevalence, and the impact sea lice have on survival of young fish, gives strong evidence that sea lice are important to consider not only for juvenile salmon, but also for forage fish, and herring in particular. In addition to further research, given the importance of herring for the region, prudent management actions are also important to consider specifically for forage fish conservation. Specifically, it is important to identify industry activities specific to herring exposure to sea lice, and increase management focus on developing and implementing net pen technologies that eliminate this risk to critical forage fish resources. Upland contained fish farming is the only method proven to eliminate these ectoparasites.

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## Tables

Table 1. Bins for relative estimated abundance of juvenile forage fish school size (number of fish), by species, recorded during visual transect surveys 2004-2017.

<u>Herring</u>	<u>Smelt</u>	<u>Sand lance</u>
0-10	0-10	0-10
11- 75	11-100	11- 75
75-500	100-500	75-500
1000	500	1000
10000	-	10000

Table 2. Average school size of juvenile forage fish groups, by month, observed 1996-2017 and total number of transects by year. Only months with observed juvenile fish are presented. Note that there were no surveys between 2001-2003, or 2006-2009.

<b>Year and month</b>	<b>Total transects</b>	<b>Herring</b>	<b>Sand lance</b>	<b>Gadids</b>	<b>Surf smelt</b>
<b>1996</b>	<b>1</b>	<b>1000</b>	<b>2</b>	<b>0</b>	<b>0</b>
8	1	1000	2	0	0
<b>1997</b>	<b>1</b>	<b>10000</b>	<b>0</b>	<b>0</b>	<b>0</b>
8	1	10000	0	0	0
<b>1998</b>	<b>1</b>	<b>10000</b>	<b>0</b>	<b>0</b>	<b>0</b>
7	1	10000	0	0	0
<b>1999</b>	<b>3</b>	<b>3333</b>	<b>3333</b>	<b>0</b>	<b>0</b>
6	1	0	10000	0	0
10	1	10000	0	0	0
<b>2000</b>	<b>8</b>	<b>13</b>	<b>2516</b>	<b>0</b>	<b>22</b>
4	1	0	10000	0	0
5	1	0	10000	0	50
6	1	100	75	0	75
7	2	0	0	0	25
<b>2004</b>	<b>2</b>	<b>10000</b>	<b>50</b>	<b>0</b>	<b>505</b>
7	1	10000	0	0	1000
9	1	10000	100	0	10
<b>2005</b>	<b>2</b>	<b>25</b>	<b>500</b>	<b>0</b>	<b>0</b>
3	1	50	1000	0	0
4	1	0	0	0	0
<b>2010</b>		<b>1692</b>	<b>3667</b>	<b>0</b>	<b>183</b>
4	1	0	10000	0	0
7	2	5000	5000	0	550
8	3	50	667	0	0
<b>2011</b>	<b>4</b>	<b>5075</b>	<b>5000</b>	<b>500</b>	<b>3</b>
8	2	10000	5000	500	0
9	1	300	10000	1000	10
<b>2012</b>	<b>17</b>	<b>3682</b>	<b>6494</b>	<b>59</b>	<b>1</b>
8	1	10000	10000	0	10

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9	6	5217	3367	167	2
10	7	1614	10000	0	0
11	3	3333	3400	0	0
<b>2013</b>	<b>33</b>	<b>2473</b>	<b>2709</b>	<b>156</b>	<b>53</b>
6	10	20	5180	315	0
7	1	0	1000	1000	100
8	9	3478	4000	111	43
9	4	10000	153	0	288
10	3	3367	0	0	33
<b>2014</b>	<b>41</b>	<b>5939</b>	<b>1139</b>	<b>0</b>	<b>25</b>
4	2	0	50	0	5
5	5	0	420	0	0
6	1	0	1000	0	0
7	6	8500	1690	0	0
8	12	7600	1100	0	78
9	10	7110	2010	2	10
10	4	7550	0	0	0
11	1	0	0	0	0
<b>2015</b>		<b>4105</b>	<b>1683</b>	<b>0</b>	<b>60</b>
4	1	0	200	0	0
5	5	0	500	0	10
6	8	7500	1263	0	143
7	8	2641	6875	0	64
8	5	2212	480	0	100
9	9	1790	11	0	33
10	8	7500	25	0	0
<b>2016</b>	<b>40</b>	<b>4954</b>	<b>1256</b>	<b>1</b>	<b>53</b>
4	2	0	0	0	35
5	4	0	125	5	0
6	9	0	2489	0	182
7	6	8333	0	0	45
8	7	8600	371	0	19
9	7	10000	3061	0	0
10	4	5750	825	0	0
11	1	0	0	0	0
<b>2017</b>	<b>36</b>	<b>1452</b>	<b>2262</b>	<b>0</b>	<b>1</b>
5	2	0	50	0	0

6	9	5667	133	0	0
7	8	145	6750	0	0
8	6	0	2170	0	3
9	6	17	2183	0	0

Table 3. Average size (mm) of herring and sand lance 1999-2017. NA denotes that fish sizes were not recorded.

<u>Year</u>	<u>Herring</u>	<u>Sand lance</u>	<u>Percent herring with parasites</u>
1996	na	na	0
1997	na	na	0
1998	na	na	0
1999	70	70	0
2000	70	65	0
2004	na	na	80
2005	na	na	0
2010	75	85	80
2011	70	80	15
2012	79	75	93
2013	59	73	36
2014	83	161	89
2015	83	88	46
2016	70	89	44
2017	66	93	88

Table 4. Parameter estimates of time series describing presence of sea lice on herring over time. As years are categorical variables, their summary statistics are shown in contrast to a baseline, which in this model is the year 1995 when no sea lice were observed.

<u>Year</u>	<u>Estimate</u>	<u>Std.Error</u>	<u>Statistic</u>	<u>P value</u>
1996	-20.566	3956.180	-0.005	0.996
1997	-21.566	3956.180	-0.005	0.996
1998	-21.566	3956.180	-0.005	0.996

1999	-21.566	3956.180	-0.005	0.996
2000	-19.566	3956.180	-0.005	0.996
2004	-4.000	1.414	-2.828	0.005
2005	-19.265	3956.180	-0.005	0.996
2010	-3.396	1.389	-2.445	0.014
2011	14.197	2113.905	0.007	0.995
2012	-4.032	0.654	-6.165	0.000
2013	-3.620	0.587	-6.164	0.000
2014	-4.536	0.393	-11.549	0.000
2015	-4.769	0.480	-9.937	0.000
2016	-4.644	0.463	-10.035	0.000
2017	-3.033	0.579	-5.240	0.000

### Figure Captions

Figure 1 Aerial views of the study area and transect sites 1996-2017 Crescent and Freshwater Bays, central Strait of Juan de Fuca Washington state. Figure by Dave Parks, Washington Department of Natural Resources.

Figure 2. Mixed school of young of the year (yoy) juvenile herring infected with sea lice at Freshwater Bay, Strait of Juan de Fuca, August 2010, and sand lance with sea lice (circled in red), Crescent Bay Strait of Juan de Fuca 29 July 2012. Photos by Anne Shaffer

Figure 3. Top and middle: Average herring school size and percent of fish with parasites, by year. Panels on the left show proportional estimates to facilitate comparisons of portions. Panels on the right show absolute estimates to visualize sample sizes. Bottom: Model-predicted probabilities that lice were present in a school of herring. Predictions are based on the average school size of 6,395 fish and blue lines indicate one standard error above and below predictions. Note that standard errors are infinite for years when no lice were observed.

Figure 4. Left, Location of Atlantic Salmon net pens, Washington state (Seattle Times/DoE), Right, herring spawning grounds (Stick et al 2014). Cherry Point and Strait of Juan de Fuca herring stocks are listed as critical. Red star (\*) denotes this study site.

Figure 5. Graphic presentation of modeled trajectory of surface drifting herring spawn (Snauffer et al 2013)

Figure 1. Aerial views of the study area and transect sites 1996-2017 Crescent and Freshwater Bays, central Strait of Juan de Fuca Washington state. Figure by Dave Parks, Washington Department of Natural Resources.

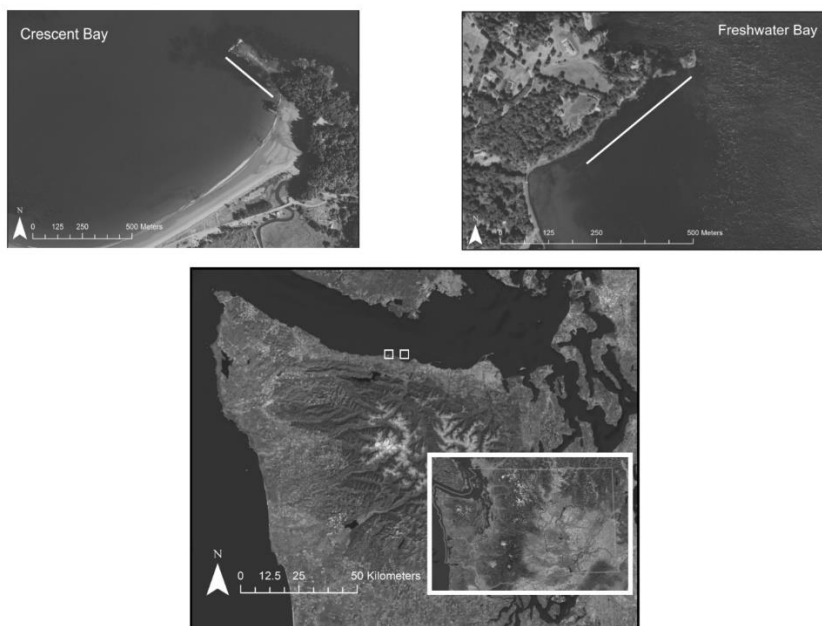


Figure 2. Mixed school of young of the year (yoy) juvenile herring infected with sea lice at Freshwater Bay, Strait of Juan de Fuca, August 2010 (left), and sand lance with sea lice (circled in red, right), Crescent Bay Strait of Juan de Fuca 29 July 2012. Photos by Anne Shaffer



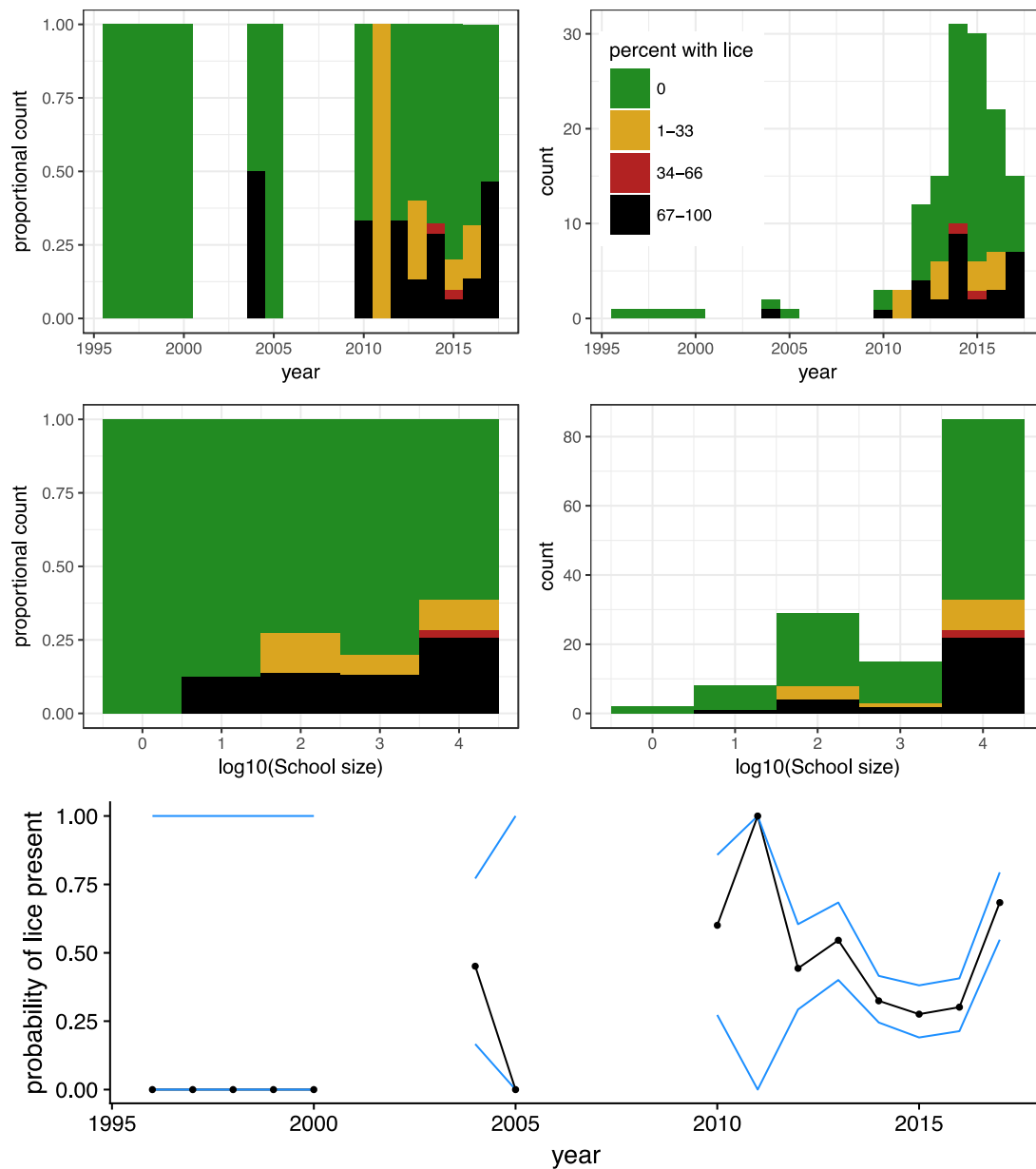


Figure 3. Top and middle: Average herring school size and percent of fish with parasites, by year. Panels on the left show proportional estimates to facilitate comparisons of portions. Panels on the right show absolute estimates to visualize sample sizes. Bottom: Model-predicted probabilities that lice were present in a school of herring. Predictions are based on the average school size of 6,395 fish and blue lines indicate one standard error above and below predictions. Note that standard errors are infinite for years when no lice were observed.



Figure 4. Right, herring spawning grounds (Stick et al 2014). Cherry Point and Strait of Juan de Fuca herring stocks are listed as critical Red star (\*) denotes this study site.

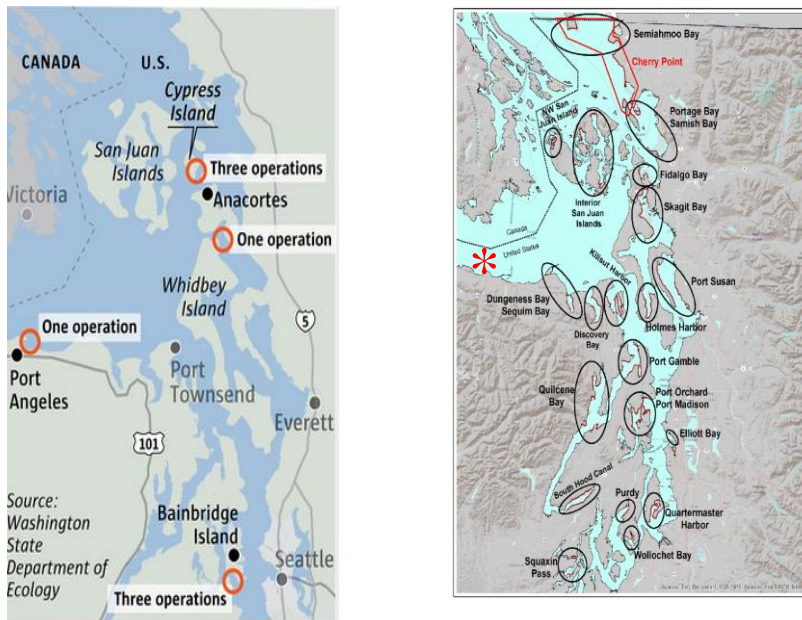


Figure 5. Graphic presentation of modeled trajectory of surface drifting herring spawn (Snauffer et al 2013). Red arrow in first box indicates study location.

