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# Ichthyoplankton Distribution And Assemblage Within And Around The Saco River Plume

Tracey Bauer
*University of New England*

# ICHTHYOPLANKTON DISTRIBUTION AND ASSEMBLAGE WITHIN AND AROUND THE SACO RIVER PLUME

BY

Tracey Calleen Bauer B.S. University of North Carolina Wilmington, 2013

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Thesis Director, Dr. James Sulikowski
Professor of Marine Science

Dr. Steve I. Zeeman
Professor of Marine Science

Dr. Jeri L. Fox
Associate Professor of Aquaculture and
Aquarium Sciences

8/25/15
Date

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Tracey Bauer

University of New England, August 2015

## Abstract

A majority of research has focused on the importance of large river plumes for ichthyoplankton survival and recruitment. However, the impacts of smaller, more ephemeral river plumes, such as those commonly found in the Gulf of Maine, on ichthyoplankton are far less understood. The purpose of the current study was to use a small river plume located in the southern Gulf of Maine as a model system to increase our understanding of their effects on ichthyoplankton distribution and diversity, and determine what biotic and abiotic factors may be influencing any differences observed. Plankton tow sampling revealed that although ichthyoplankton abundance was highest in the ocean habitat, species diversity was lowest within this region due to the dominance of one species. Chlorophyll α concentrations and zooplankton densities did not differ between plume or ocean waters, most likely due to the ephemeral nature of the river plume. Overall, compared to larger plume systems, the Saco River plume appeared to have minimal influence in Saco Bay. However, specific events of higher river discharge may be having the greatest effect on ichthyoplankton distribution through advection offshore, as well as downwelling at the front and subsequent entrainment into plume waters.

# Introduction

When rivers flow into coastal waters, the less dense riverine water will float on top of the denser ocean water, creating an estuarine plume that extends out across a bay or continental shelf (Bowman and Iverson 1978, Garvine 1987, Grimes and Kingsford 1996). These plumes are highly dynamic oceanographic features, and create a distinct environment from the surrounding marine water due to differences in salinity, temperature, nutrients, and turbidity (Grimes 2001, Kingsford and Suthers 1994, Grimes and Kingsford 1996). Plumes can be classified into small, medium, and large based on the amount of river discharge and their maximum offshore extent (Grimes and Kingsford 1996).

Large plumes (e.g. such as those created by the Mississippi, Columbia, or Amazon Rivers) have higher, more constant discharge, producing increased stability and high temporal persistence (Grimes and Kingsford 1996, Thorrold and McKinnon 1995). These characteristics make large river plumes much more convenient to study, thus, the majority of research has focused on such systems and their impacts on coastal environments (Grimes and Finucane 1991, Govoni 1993, Govoni and Grimes 1992, Govoni et al. 1989, Parnel et al. 2008, Morgan et al. 2005, Litz et al. 2014). Research suggests that larger river plumes dominate their coastal ecosystem with a wide area of influence (Wiseman and Garvine 1995), significantly affecting fish development and survival during the critical larval life stage (Grimes 2001, Warrick and Fong 2004) due to factors such as increased prey (Govoni et al. 1989, Grimes and Finucane 1991) and variable transport processes (Sabates 1990). Thus, the substantial effects of large plumes on the coastal ecosystem are relatively well understood.

In contrast, our knowledge on smaller river plume dynamics and their impacts on the coastal ecosystem is limited due to their high temporal and spatial variability (Saldías et al. 2012). Changes in discharge rates, winds, and tidal phase can all rapidly alter the size of smaller river plumes (Garvine 1987, Gelfenbaum and Stumpf 1993, Grimes and Kingsford 1996, Rodridgues et al. 2009), making them more difficult to study. Despite our limited knowledge, smaller river plumes have been shown to increase the productivity of the ecosystem (Connelly et al. 2009) and influence the community structure and abundance of ichthyoplankton (Thorrold and McKinnon 1995), though their influence may be limited due to their variable size (Gaston et al. 2006).

The Gulf of Maine, encompassing 25 separate watersheds, is an example of an area dominated by small- to medium-sized plumes. As the sixth largest watershed in Maine (NRC 2004), the Saco River plume is the first to be classified as small in this region due to its relatively low discharge and short average distance from shore (Tilburg et al. 2011, Grimes and Kingsford 1996). Research of this system, and of other river plumes in the Gulf of Maine, has mainly been limited to studying their physical processes (Hetland and MacDonald 2008, Tilburg et al. 2011).

Despite the demonstrated importance of plumes, no studies conducted within the Gulf of Maine (GOM) have focused on the effects of river plumes on ichthyoplankton. Current knowledge of ichthyoplankton in this region is limited to abundances and species composition in a few estuaries (Lazzari 2001, Lazzari and Tupper 2002, Lazzari 2002, Lazzari et al. 1999, Chenoweth 1973, Townsend 1984, Runge and Jones 2012). To date, only one study (Wargo et al. 2009), conducted in the Saco River watershed, has suggested that an abiotic factor (salinity) may be an influencing factor in ichthyoplankton distribution.

Based on this information, the goals of the current study were to use the Saco River as a model system for small river plumes within the GOM in order to: 1) determine how the plume affects ichthyoplankton horizontal and vertical distribution and diversity and 2) establish what abiotic and biotic factors may be influencing any observed differences in distribution and diversity.

## Materials and Methods

### Field sampling

#### Sampling area and site selection.

Ichthyoplankton were collected during weekly sampling trips in Saco Bay between the mouth of the Saco River (43.461 N, 70.355 W) and the eastern extent of the river plume (43.461 N, 70.238 W) (Figure 1) from June to September in 2013 and from May to November in 2014.

All sampling trips were conducted aboard the 7-m (23-ft) University of New England (UNE) research vessel *Llyr*, during daylight hours. Sampling trips typically began two hours before low tide and lasted approximately four hours (depending on sea conditions) in order to sample the plume at its maximum size. All sampling trips were highly dependent on weather and sea conditions.

Locations where the salinity ranged between 0 to 29 ppt were categorized as brackish water, and therefore considered "within" the plume, while locations where the salinities were greater than 29 ppt were categorized as marine water, and therefore

considered “outside” the plume (Ohrel and Register 2006, Litz et al. 2014). The outward edge of the plume, or the front, is the area of mixing between brackish and marine waters (~29 ppt) (Bowman and Iverson 1978, Garvine 1987, Garvine and Monk 1974, Pinckney and Dustan 1990). A blocked sampling design was utilized (Morgan et al. 2005) to control for the inherent spatial and temporal variability of the plume (Tilburg et al. 2011) by ensuring the plume and ocean stations were a constant distance from the front and each other every sampling trip.

In order to locate the plume front, surface water (0 – 0.5 m depth) salinity values were measured using an SBE 45 MicroTSG Thermosalinograph (Sea-Bird Electronics Inc., Bellevue, WA USA) (in 2013) and a YSI 556 MPS Handheld Multiparameter Instrument (YSI Incorporated, Yellow Springs, OH USA) (in 2014) beginning at the mouth of the Saco River and approximately every 10 m until the salinity approached 29 ppt. At this time, the actual physical location of the front was identified through visual observation as a clear demarcation with smoother water, foam, and floating debris (Morgan et al. 2005, Govoni and Grimes 1992, Kingsford and Suthers 1994). The sampling stations were then selected by driving the boat 200 to 400 m perpendicularly from the front into the plume for the “plume” station, or into marine waters for the “ocean” station (Rissik and Suthers 1996, Morgan et al. 2005, Kingsford and Suthers 1993). The “plume” station was sampled first during the outgoing tide in order to avoid the highly mobile plume contracting in size while sampling. The sampling locations selected in the current study were skewed to the north side of the plume due to limitations in how far the boat could sample offshore.

**Ichthyoplankton collection.**

Once the sampling stations were identified, ichthyoplankton were collected using a 1 m diameter, 333 μm mesh ring plankton net, equipped with a mechanical flowmeter (General Oceanics, Miami, FL USA). Subsurface and surface horizontal plankton nets were towed at each station for 10 minutes and at a speed of 1 – 1.3 m/s. Subsurface tows were fished at a depth of 3 m with the use of a heavy duty cast bronze double-trip mechanism (Aquatic Research Instruments, Hope, ID). Fishing depth was determined using an inclinometer, which provided the angle of the line in the water. Depth (D) of the net, was then calculated using the following equation (Sameoto et al. 2000):

$$D=(L)*COS(A) \quad (1)$$

Where (L) is the length and (A) is the angle of the line let out of the boat as the net is being towed.

Subsurface tows were always completed first at the "plume" station, and surface tows were always completed first at the "ocean" station. Immediately following the haul back, the net was elevated and thoroughly rinsed with seawater to eliminate the possibility of contamination between tows. Additionally, this ensured the entire plankton sample collected within the cod end, which was subsequently preserved in 70% ethanol.

**Environmental parameters.**

At the beginning and end of each plankton tow, a vertical profile (up to 30 m depth) of salinity, temperature, water density, and fluorescence was obtained using a SBE 25 Sealogger CTD (Sea-Bird Electronics Inc., Bellevue, WA USA).

Additionally, surface and subsurface water samples were collected at both stations every trip and immediately placed in a cooler filled with ice for later filtering and analysis

of chlorophyll α at the UNE Marine Science Center (MSC). The subsurface water sample was collected at 3 m depth (i.e. below the plume) using a Horizontal PVC Beta Water Sampler (Wildlife Supply Company, Yulee, FL).

Monthly means and daily mean values of Saco River discharge were obtained from the USGS gauging station in Cornish, Maine.

**Laboratory analysis**

Each plankton tow sample was examined using a Leica EZ4HD microscope (Leica Microsystems Inc., Buffalo Grove, IL USA) in order to separate, quantify, and identify ichthyoplankton larvae to the species-level. The abundances were standardized as number of ichthyoplankton per 100 $m^3$. Ichthyoplankton eggs in each sample were enumerated, but not further identified taxonomically.

In addition, six 1 mL subsamples were taken from each plankton tow sample and placed in separate vials of 70% ethanol for further analysis. These subsamples were examined using a Leica EZ4HD microscope (Leica Microsystems Inc., Buffalo Grove, IL USA) and zooplankton were identified down to family-level and quantified. The following equation was then used to estimate total zooplankton density in each sample, as well as densities for each individual family (#/100 $m^3$):

$$Whole\ sample\ density = \left[\frac{(n)(V_S)}{V_m}\right] * 0.01 \quad (2)$$

Where $n$ is the mean number of organisms in a 1 mL subsample, $V_S$ is the volume of plankton sample (mL), and $V_m$ is the volume of seawater sampled ($m^3$).

**Chlorophyll α extraction.**

Chlorophyll α extraction was conducted on water samples to determine if chlorophyll α concentrations varied between plume and ocean waters. Water samples were vacuum filtered through Whatman GF/F glass microfiber filters (25 mm) immediately upon return to lab on the day of sampling. Chlorophyll α extraction was performed based on methods described in "Fluorometric Determination of Chlorophyll α" (2015). Briefly, the filter was well-ground using a Pyrex pestle, treated with 10 mL of 90% acetone, and refrigerated for 24 hours in a 15 mL centrifuge tube. The tubes were shaken, and then centrifuged at 2400 rpm for 10 minutes, before being pipetted into a 13 mm round cuvette for reading on a TD700 fluorometer (Turner Designs, Sunnyvale, CA USA).

**Statistical Analysis**

**Environmental parameters.**

In order to determine if differences in environmental parameters (sea surface salinity, sea surface temperature, surface water density, surface fluorescence, salinity at 3 meters, temperature at 3 meters, water density at 3 meters, fluorescence at 3 meters) existed between plume and ocean waters, bootstrapped two-sample t-tests were run separately for each year (n = 5000) (SYSTAT v13). A bootstrapped two-sample t-test was also run for discharge to determine there were any annual differences. Means, standard error, and

coefficient of variation (CV) were calculated for all parameters. CV can be used to describe the dispersal of a variable, independent of the sampling measurement unit.

**Ichthyoplankton species composition.**

Species richness (S), or the number of species, was determined for each sampling location. In order to analyze species diversity, the Shannon index of diversity (H') and Pielou's J evenness (E) were calculated for each sample:

$$H' = -\sum_{i=1}^{S} p_i \ln(p_i) \quad (3)$$

$$E = \frac{H'}{\ln(S)} \quad (4)$$

Where $p_i$ is the proportion (n/N) of densities of one species found (n) divided by the total densities of individuals found (N). The higher the value of H and E, the more diverse and even the ichthyoplankton community (Gotelli and Ellison 2013).

One-way analysis of variance's (ANOVA) ($\alpha = 0.05$) were run to test the hypothesis of no significant difference in species diversity and evenness between stations (In surface, In subsurface, Out surface, Out subsurface). One-way ANOVAs were then run for each year separately to test the effects of station on species diversity and evenness. Post-hoc Tukey HSD tests were run after all analyses.

**Ichthyoplankton and zooplankton densities.**

Catches of ichthyoplankton larvae and eggs and total zooplankton densities in both years were highly skewed, and thus were fourth root transformed to minimize the influence of the few high catches.

A one-way ANOVA, using a Monte Carlo randomization test ($n = 5000$, $\alpha = 0.05$) to generate p-values, was run in R (v0.97.551, R Core Team 2013) There was no significant difference in ichthyoplankton, egg, and total densities between sampling years, and so data from both years were combined for all further analyses.

One-way ANOVAs, with the Monte Carlo randomization test, were then used to test for differences in ichthyoplankton densities sampled from the plume and ocean stations at the surface and subsurface, for a total of four sampling locations. When the randomization test indicated significance, post-hoc pairwise comparisons were performed for each combination of sampling locations.

For all post-hoc tests and bootstrapped t-tests, Holm's sequential Bonferroni adjustment of p-values (Holm 1979) was used in order to control for Type 1 error.

**Correlation analysis.**

Relationships between environmental parameters (Chlorophyll α concentration, Saco River discharge) and ichthyoplankton and zooplankton abundances were analyzed using nonparametric Spearman rank correlation for each sampling location.

## Results

**Environmental parameters**

Sea surface salinity ranged from 12 to 31 ppt in 2013 and 17 to 32 ppt in 2014. Unsurprisingly, sea surface salinity in the ocean habitat was significantly higher than in the plume habitat in both years (2013: $t = -9.303$, $df = 32$, $p < 0.001$; 2014: $t = -3.598$, $df = 48$, $p = 0.001$). No differences were observed in sea surface temperature between plume and ocean waters in either sampling year (Table 1), with values ranging from 13 to 22 °C in 2013, and 10 to 20 °C in 2014. Analysis of water density data indicated that ocean water at the surface was significantly more dense than plume water in 2013 ($t = -6.276$, $df = 28$, $p < 0.001$), but not in 2014. In both sampling years, salinity, temperature, and water density at 3 meters depth did not significantly differ between plume and ocean waters. Furthermore, analysis of chlorophyll α concentration and fluorescence indicated no difference between plume and ocean waters both in surface waters and at 3 meters depth.

Saco River discharge was relatively low on all sampling days during the study period, varying between 40 to 110 $m^3/s$ in 2013, and from 13 to 120 $m^3/s$ in 2014. There was much greater variability in discharge in 2014 (CV = 56%) than in 2013 (CV = 29%). However, average discharge was not significantly different between the two sampling years.

**Ichthyoplankton species composition**

Over the course of the study, 9000 ichthyoplankton larvae and 163,260 eggs were collected in 99 plankton tows from 11 sampling trips in 2013 and 16 in 2014. Twenty-two total ichthyoplankton species were observed over the two sampling years (Table 2).

Comparisons of the ichthyoplankton assemblage revealed differences between plume and ocean habitats, as well between the surface and subsurface. Overall, species diversity ($p = 0.001$, $F = 5.828$, $df = 3, 69$) and evenness ($p = 0.008$, $F = 4.259$, $df = 3, 69$) significantly differed between plume and ocean habitats (Figure 2). Surface plume waters and subsurface waters below the plume had significantly higher species diversity than any other habitat sampled. Surface ocean waters had the lowest species diversity (0.32) and evenness (0.31), with only two species comprising 94% of the catch in 2013 and three species comprising 98% of the catch in 2014.

**Ichthyoplankton and zooplankton distribution**

The one-way ANOVA revealed that there were significantly more ichthyoplankton larvae at the surface than in subsurface waters ($F = 11.578$, $df = 1, 83$, $p = 0.004$) (Figure 3). Additionally, the results suggest that significantly more ichthyoplankton larvae were found in ocean waters than in plume waters ($F = 9.232$, $df = 1, 83$, $p = 0.003$). Furthermore, significant differences in ichthyoplankton larvae densities between sampling locations were observed ($F = 7.191$, $df = 3, 81$, $p = 0.0002$). Post-hoc tests revealed that ichthyoplankton larvae densities in surface ocean waters were significantly greater than in surface plume waters ($p = 0.009$), in subsurface waters below the plume ($p = 0.0002$) and in subsurface ocean waters ($p = 0.012$). Ichthyoplankton larvae densities in surface plume waters were significantly greater than ichthyoplankton larvae densities in subsurface waters below the plume ($p = 0.014$). Subsurface ocean waters contained statistically similar ichthyoplankton larvae densities to surface plume waters and subsurface waters under the

plume. Densities in both years at the sampling locations, though, were highly variable. Overall for both years, coefficient of variation (CV) values ranged from 55 – 124%. In 2013, the highest variability of ichthyoplankton larvae densities was found in surface plume waters (CV = 87%), and in 2014, the highest variability in ichthyoplankton larvae density was found in subsurface waters below the plume (CV = 90%).

Ichthyoplankton egg densities were significantly greater at the surface than at depth ($F = 10.121$, $df = 1, 89$, $p = 0.002$). Significant differences in ichthyoplankton egg densities between stations were also observed ($F = 4.954$, $df = 3, 87$, $p = 0.02$). Of all four sampling locations, only ichthyoplankton egg densities collected in surface ocean waters were significantly greater than in subsurface waters below the plume ($p = 0.003$) (Figure 3). Ichthyoplankton egg densities were variable in all four sampling locations, similarly to ichthyoplankton densities. Overall, CV values ranged from 45 – 69%. The greatest variability was seen in subsurface waters below the plume (2013: CV = 67%; 2014: CV = 74%) in both years.

Total zooplankton densities did not differ by depth or between the four sampling locations. Variability of zooplankton densities between the three sampling locations were all relatively low compared to ichthyoplankton densities, with overall CV values ranging from 29 – 56%.

## Correlation analysis

Correlation analysis revealed that discharge was more frequently correlated with zooplankton and ichthyoplankton abundances than chlorophyll α concentration, and there

were differences between stations (Table 3). Additionally, correlations between chlorophyll α concentration and discharge revealed that chlorophyll α significantly decreased as Saco River discharge increased in surface plume and subsurface ocean waters. Furthermore, chlorophyll α was positively correlated with discharge in surface ocean waters, although the relationship was not significant. No correlations in 2013 were significant, most likely due to the smaller sample size and the smaller range of Saco River discharge values.

## Discussion

Over the course of the current study, many of the Saco River plume characteristics were found to be similar to those in other smaller watersheds (Grimes and Kingsford 1996). For example, relatively low discharge and short average distance from shore was also observed by Kingsford and Suthers (1994) for the Botany Bay plume and by Hetland and MacDonald (2008) for the Merrimack River plume.

### Ichthyoplankton distribution

In the current study, greater ichthyoplankton densities were found in the surface ocean habitat than within the plume. This phenomenon was first observed by Wargo et al. (2009) and appears to be a consistent feature of the watershed. While the plume dynamics of the Saco River (i.e. the lack of a well-defined frontal boundary, ephemerality; Bloodsworth et al. 2015) make the direct comparison to other systems difficult, this later

observation is in contrast to studies on larger systems that have suggested that larval organisms are transported by currents to the front, accumulating there in higher densities than in plume and ocean waters (Govoni 1993, Govoni and Grimes 1992, Morgan et al. 2005, Mackas and Louttit 1988, Harrison et al. 1991, Kingsford and Suthers 1994).

Previous research has also indicated the importance of adult spawning location to ichthyoplankton distribution (Grimes and Finucane 1991, Grioche and Koubbi 1997, Wong et al. 2013). For example, higher ichthyoplankton densities were attributed to spawning of adults within the plume waters of Botany Bay, Australia (Kingsford and Suthers 1994, 1996). Although not directly measured in the current study, the horizontal distributional pattern observed may be due to the spawning locations of the common ichthyoplankton species. The higher ichthyoplankton densities in surface ocean waters were principally due to large numbers of a single species (cunner, *Tautogolabrus adspersus*), which most likely was spawned in marine waters outside the plume (Collette and Klein-MacPhee 2002).

Ichthyoplankton densities in surface plume and ocean waters were, as a whole, higher than those found in subsurface waters. The observed vertical distributional pattern was most likely due to the dominance of pelagic ichthyoplankton species in this study (Sundby 1991, Conway et al. 1997). Although this is the first study to observe ichthyoplankton vertical distribution around a smaller river plume, similar observations have been observed within larger plumes. For example, Govoni et al's. (1989) study of the Mississippi River plume observed a similar pattern of greater densities of ichthyoplankton in surface water than at any depth or location in the plume.

**Ichthyoplankton diversity**

Another observation of the current study was that although ichthyoplankton densities were greatest in the ocean habitat, species diversity and evenness was lowest in this region, which was consistent with the observed dominance of *Tautogolabrus adspersus* (Ramos et al. 2006). Instead, the greatest ichthyoplankton diversity was found in the plume where there were lower *Tautogolabrus adspersus* densities and higher numbers of other marine, estuarine, and freshwater ichthyoplankton species. Throughout the current study, over 90% of the ichthyoplankton species were observed at some point within surface plume waters. In contrast, previous research on a small and a larger river plume has observed species diversity has been found to be highest at the front (Botany Bay plume; Kingsford and Suthers 1996) and in ocean waters (Mackenzie River plume; Wong et al. 2013). As no consistent pattern has been observed thus far, this may suggest that ichthyoplankton species diversity around river plumes is dependent on the system.

**Primary and Secondary Productivity**

Primary and secondary productivity were affected by the Saco River plume's ephemerality, as chlorophyll α concentrations, fluorescence, and zooplankton abundances were generally evenly distributed in and out of the plume. This is in contrast to the majority of results from larger plume systems, such as the Mississippi River plume (Grimes and Finucane 1991), Amazon River plume (Smith and Demaster 1996), and the Rhone River plume (Cadee 1978, Sabates 1990), where high nutrient concentrations have been known to enhance the productivity of local ecosystems. Unlike these larger plumes which persist over days and weeks, the Saco River plume is highly influenced by the tidal cycle, and

contracts towards the mouth of the river during flood tide (Tilburg et al. 2011). Thus, the area of the plume that was sampled in the current study is most likely not present in the Saco Bay for a long enough period of time for organisms to take advantage of higher nutrient concentrations before the tide changed (Morgan et al. 2005).

**Physical processes of the plume**

Higher river discharge was correlated with increased densities of ichthyoplankton in ocean waters, which has been similarly observed in both smaller and larger river plumes (Govoni 1993, Thiebaut 1996, Reiss and McConaugha 1999, Faria et al. 2006). Two possible explanations may be suggested for these results: (1) concentration of ichthyoplankton already in ocean waters as the area of the plume expanded across the bay, or (2) cross-frontal transport of ichthyoplankton from the plume into ocean waters (Reiss and McConaugha 1999). Cross-frontal transport of larvae may occur during periods of increased Saco River discharge, which has been shown to cause offshore advection and downshelf movement of the plume (Tilburg et al. 2011). At this time, any changes in the tidal phase or winds may suddenly force the plume back towards the river mouth, stranding a pocket of plume water, and associated ichthyoplankton, in offshore waters (Reiss and McConaugha 1999). Consequently, in larger river plumes, advection offshore has been suggested to delay recruitment of ichthyoplankton, increase their vulnerability to predation, and may result in permanent loss of ichthyoplankton to offshore waters (Govoni 1997, Reiss and McConaugha 1999). It is uncertain to what degree advection of ichthyoplankton may be occurring in the Saco River plume due to its ephemerality, but during extended

periods of higher river discharge, it may have profound impacts on ichthyoplankton survival and recruitment.

Downwelling currents at the plume front may have also been greater during periods of increased river discharge, causing higher ichthyoplankton densities to be observed in subsurface waters below the plume. Although not directly measured in the current study, downwelling is a common feature of plumes and their associated fronts (Garvine 1987, Garvine and Monk 1974, Bowman and Iverson 1978, Gelfenbaum and Stumpf 1993, Grimes and Kingsford 1996), and may have led to the introduction of marine ichthyoplankton species into plume waters. For example, Bloodsworth et al. (2015) hypothesized that the introduction of marine larvae into the Saco River plume was due to downwelling of organisms at the frontal boundary and subsequent vertical mixing between waters below and within the plume (Figure 4; St. John et al. 1992, Hetland 2010). Future research should focus on quantifying downwelling and entrainment that may be occurring around the Saco River plume. In addition, as low salinities in a plume have been shown to negatively affect the physiology, prey-capture ability, and growth of marine ichthyoplankton (Landaeta et al. 2012), the possibility of similar impacts to the condition of ichthyoplankton within smaller plumes needs to be further investigated.

Higher river discharge may have additionally influenced productivity in this region. Within the Gulf of Maine, nutrient concentrations are relatively low and stable, and input from plume systems has been suggested to be the primary influence on overall biological productivity along the coast (Salisbury et al. 2008). In the current study, increased Saco River discharge was correlated to lower chlorophyll α concentrations in surface plume waters and subsurface ocean waters, likely a result of dilution of chlorophyll α (O'Higgins

and Wilson 2005, Rodrigues et al. 2009, Maier et al. 2012). High riverine discharge also reduces the residence time of water within the estuary and may lead to higher turbidity in plume waters, which have been both correlated with lower chlorophyll concentrations (Lane et al. 2007). The current study only was limited to short-term effects of higher river discharge on productivity, but many studies have observed positive relationships between periods of higher river discharge and fishery production over a much longer period of time (Grimes 2001, Sutcliffe 1973).

## Conclusions

This study, through the use of the Saco River plume as a model system, provides valuable information of how the small river plumes within the Gulf of Maine may be affecting ichthyoplankton distribution and assemblage. Overall, compared to larger plume systems, the Saco River plume appeared to have minimal influence on ichthyoplankton in Saco Bay. However, specific events of higher river discharge may be having the greatest effect on ichthyoplankton and productivity by influencing physical processes around the Saco River plume, such as offshore advection, dilution, and downwelling at the front. These physical processes may be affecting ichthyoplankton distribution and provide an explanation for the presence of marine ichthyoplankton fish species in surface plume waters. This area within the plume had the highest species diversity, signifying that this environment, although lower in overall ichthyoplankton density, may be a crucial habitat for developing ichthyoplankton (Yoklavich et al. 1991).

Future studies within this system, and in other plume systems in the Gulf of Maine, should focus on further describing ichthyoplankton distributional and species compositional changes across tidal cycles, as well as evaluating overall condition of ichthyoplankton within and outside plume waters. As the effects of river plumes on ichthyoplankton survival and growth vary with the system, further research will be able to elucidate possible impacts of smaller plumes on ichthyoplankton recruitment within the Gulf of Maine.

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**Table 1.** Mean values for environmental parameters (±SE) in plume and ocean habitats in 2013 and 2014, with standard error. Grey boxes indicate significantly greater values, as determined by bootstrapped two-sample t-tests. Significance for each individual t-test was determined by the Bonferroni-Holm correction.

| Variable | Year | Habitat | |
|---|---|---|---|
| | | Plume | Ocean |
| SSS (ppt) | 2013 | 20.3 ± 0.85 | 29.5 ± 0.27 |
| | 2014 | 27.3 ± 0.58 | 29.8 ± 0.31 |
| SST (°C) | 2013 | 18.3 ± 0.54 | 16.8 ± 0.46 |
| | 2014 | 16.4 ± 0.45 | 16.3 ± 0.45 |
| Surface water density (kg/L) | 2013 | 1014.4 ± 0.78 | 1020.5 ± 0.57 |
| | 2014 | 1019.6 ± 0.58 | 1021.7 ± 0.26 |
| Surface fluorescence ($mg/m^3$) | 2014 | 7.1 ± 0.54 | 7.0 ± 0.71 |
| Surface chlorophyll α (μg/L) | 2014 | 2.0 ± 0.21 | 2.0 ± 0.20 |
| Salinity at 3m depth (ppt) | 2013 | 29.2 ± 0.39 | 30.5 ± 0.14 |
| | 2014 | 30.7 ± 0.13 | 30.7 ± 0.13 |
| Temperature at 3m depth (°C) | 2013 | 16.2 ± 0.40 | 16.1 ± 0.44 |
| | 2014 | 14.5 ± 0.51 | 15.1 ± 0.41 |
| Water density at 3m depth (kg/L) | 2013 | 1021.4 ± 0.42 | 1022.4 ± 0.38 |
| | 2014 | 1022.7 ± 0.13 | 1022.6 ± 0.08 |
| Fluorescence at 3m depth ($mg/m^3$) | 2014 | 7.3 ± 0.65 | 7.0 ± 0.70 |
| Chlorophyll α at 3 m depth (mg/L) | 2014 | 1.9 ± 0.17 | 2.0 ± 0.16 |

**Table 2.** Ichthyoplankton species collected in Saco Bay, ME in 2013 and 2014. Presence in a sampling year is denoted by "x".

| Scientific name | Common name | 2013 | 2014 |
|---|---|---|---|
| *Hippoglossoides platessoides* | American plaice | | x |
| *Ammodytes americanus* | American sand lance | | x |
| *Peprilus tricanthus* | Atlantic butterfish | x | x |
| *Clupea harengus* | Atlantic herring | x | x |
| *Scomber scombrus* | Atlantic mackerel | | x |
| *Liparis atlanticus* | Atlantic seasnail | | x |
| *Tautogus adspersus* | cunner | x | x |
| *Brosme brosme* | cusk | x | |
| *Enchelyopus cimbrius* | fourbeard rockling | x | x |
| *Hippoglossina oblonga* | fourspot flounder | | x |
| *Lophius piscatorius* | monkfish | | x |
| *Morone* spp. | temperate bass | x | |
| *Syngnathus fuscus* | Northern pipefish | x | x |
| *Pollachius virens* | pollock | | x |
| *Ulvaria subbifurcata* | radiated shanny | x | x |
| *Urophycis chuss* | red hake | x | x |
| *Merluccius billinearis* | silver hake | x | x |
| *Tautoga onitis* | tautog | x | x |
| *Gasterosteus aculeatus* | threespine stickleback | | x |
| *Urophycis tenuis* | white hake | | x |
| *Scophthalmus aquosus* | windowpane flounder | x | x |
| *Pseudopleuronectes americanus* | winter flounder | x | x |

**Table 3.** Summary of Spearman rank correlation analysis between fourth root transformed zooplankton, chlorophyll α, or ichthyoplankton (Eggs and Larvae) abundances and environmental parameters (discharge: D, chlorophyll α abundance: C) measured in Saco Bay in 2013 and 2014. Only correlation coefficients greater than ±0.4 are shown. The sign of the correlation coefficient indicates the relationship between the two variables (positive/negative). *, ** denotes $p < 0.05$ and $p < 0.01$ respectively. Chlorophyll α abundance was not measured in 2013, and thus is not included for that year in the table.

| **2013** | **In surface** | | **In subsurface** | | **Out surface** | | **Out subsurface** | |
|---|---|---|---|---|---|---|---|---|
| | D | | D | | D | | D | |
| Zooplankton | -0.67 | | -0.75 | | | | | |
| Eggs | 0.46 | | -0.49 | | | | | |
| Larvae | 0.52 | | | | | | | |
| **2014** | **In surface** | | **In subsurface** | | **Out surface** | | **Out subsurface** | |
| | D | C | D | C | D | C | D | C |
| Chlorophyll α | -0.61* | | | | | | -0.71* | |
| Zooplankton | 0.52 | | | 0.43 | | 0.55 | 0.49 | |
| Eggs | 0.61* | -0.67* | 0.82** | | 0.41 | | 0.59 | |
| Larvae | | -0.43 | 0.79* | | 0.50 | | | |

## Figure Legends

Figure 1. Map of the study area and sampling locations at the Plume and Ocean stations for both sampling years.

Figure 2. Mean Shannon index of diversity (H) and evenness (E) for each sampling location (in surface = 1, in subsurface = 2, out surface = 3, out subsurface = 4). Bars denote standard error. Different letters denote significance between groups, as determined by a post-hoc Tukey HSD test. Note the differences in scale between the two axis's.

Figure 3. Fourth root transformed densities of ichthyoplankton and zooplankton in surface and subsurface ("Sub") waters outside of and within the plume. Length of each box represents the range within which 50% of values fall, and whiskers represent minimum and maximum values. Different letters denote significance between groups as indicated by post-hoc pairwise comparisons.

Figure 4. Conceptual model of downwelling and subsequent mixing into plume waters. (A) Ocean and plume currents converge at the front (B) causing downwelling and subduction of ocean waters under the plume, transporting organisms with it. (C) Vertical mixing between plume, front, and ocean waters causes entrainment of ocean water into the plume, introducing marine organisms into plume waters.

**Figure 1**

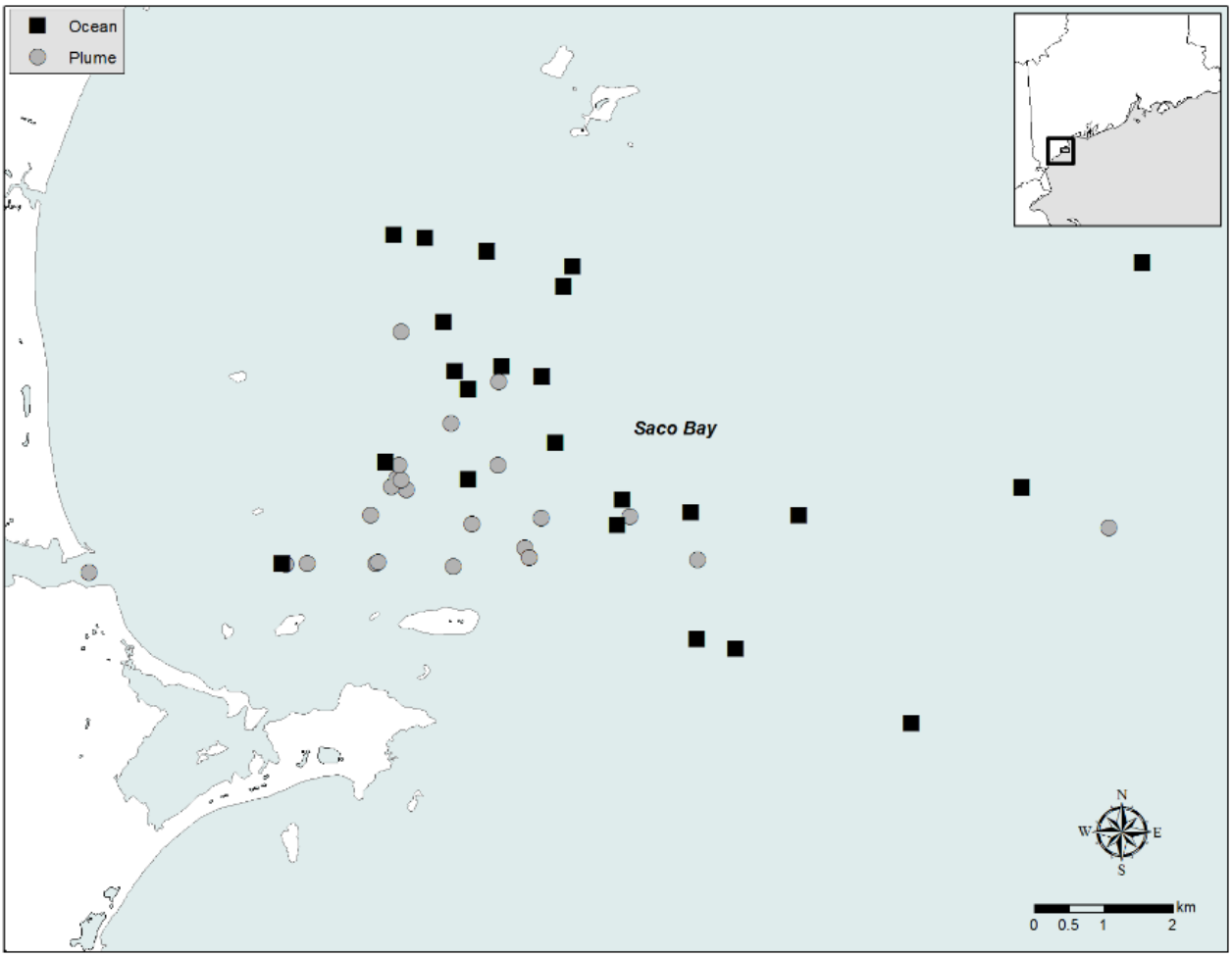


**Figure 2**

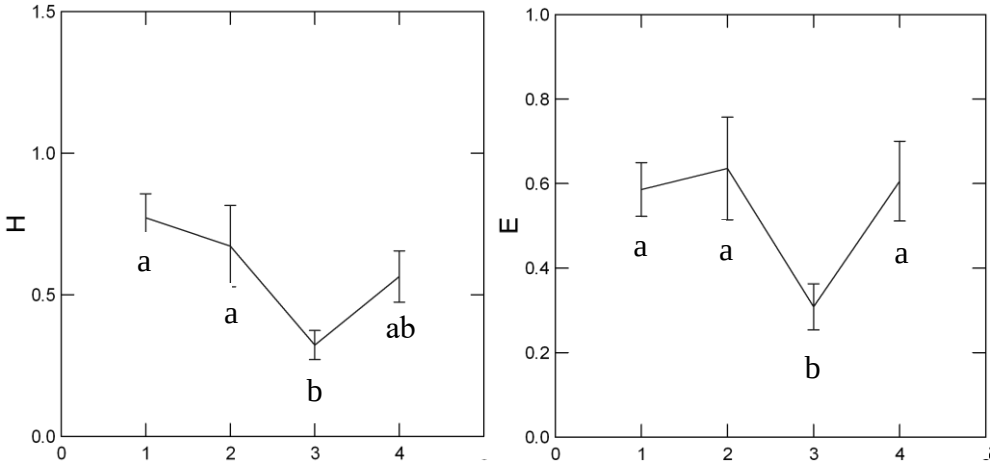


**Figure 3**

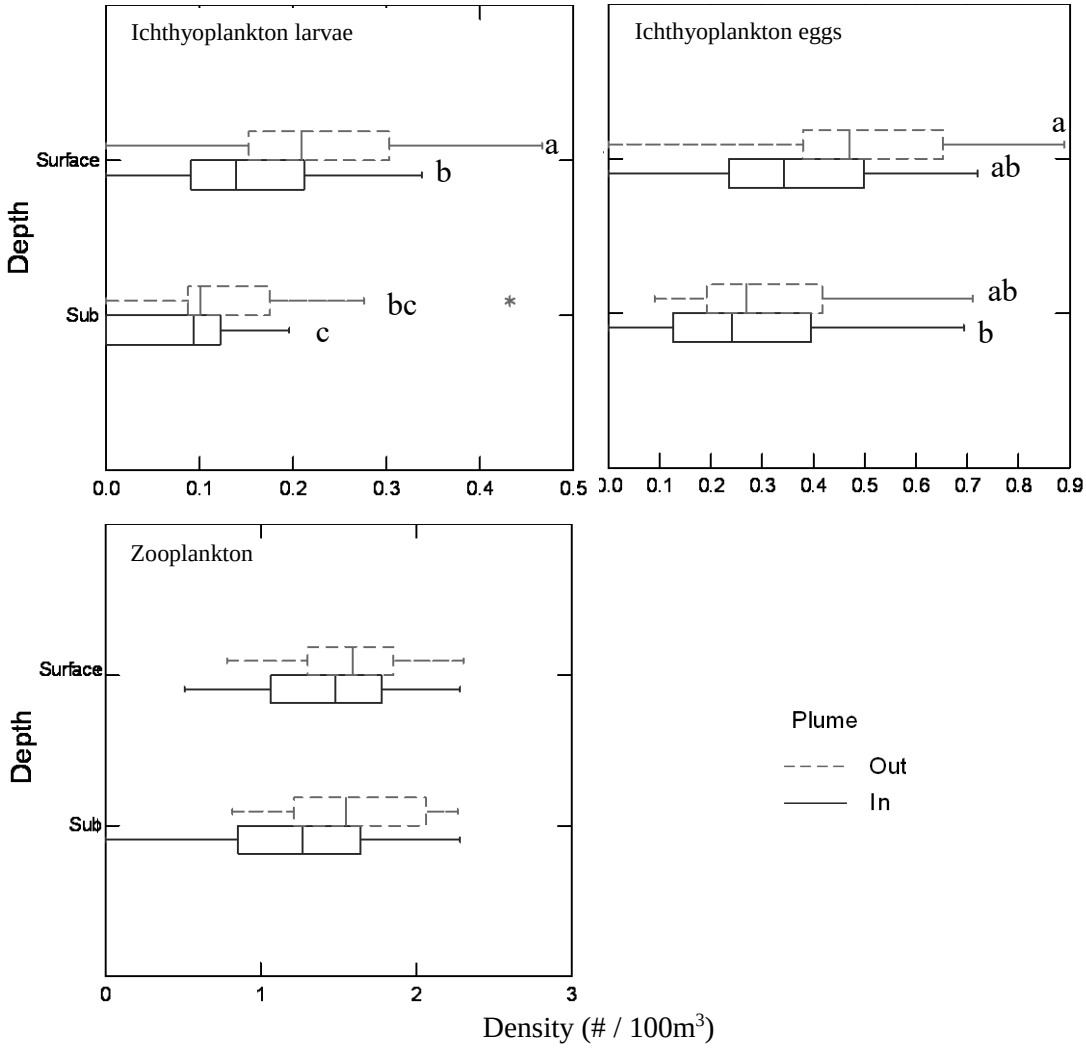
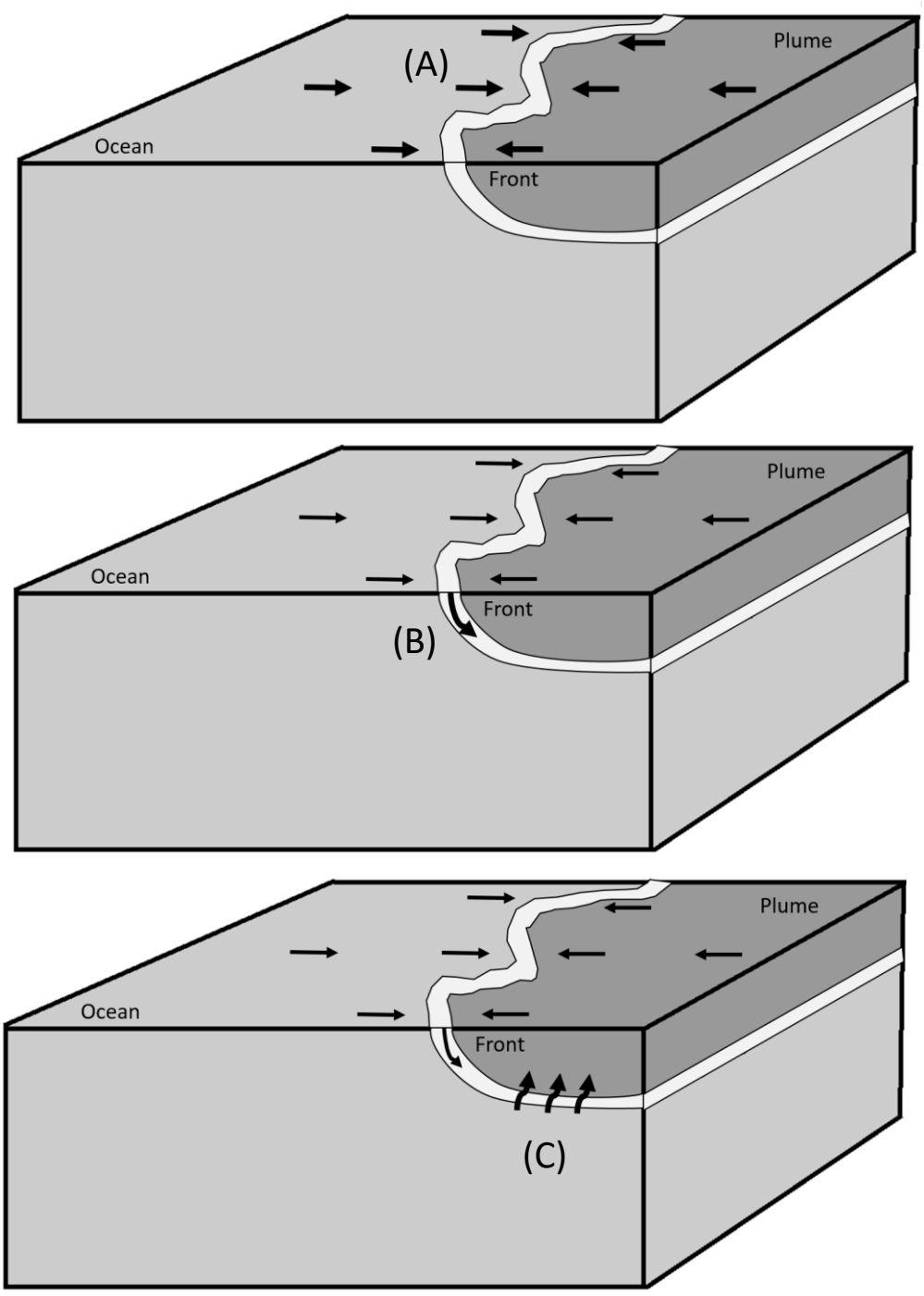

Ichthyoplankton larvae
Ichthyoplankton eggs
Zooplankton
Surface
Sub
Depth
a
b
bc
c
ab
*
0.0
0.1
0.2
0.3
0.4
0.5
0.6
0.7
0.8
0.9
0
1
2
3
Plume
Out
In
Density (# / 100m3)

**Figure 4**



# APPENDIX

## SAMPLING ICHTHYOPLANKTON IN SACO BAY WITH STATIONARY PLANKTON NETS

### Introduction

While towed plankton nets have been effectively used to collect ichthyoplankton in Saco Bay (Wargo et al. 2009), sampling thus far has been highly weather-dependent and limited by the geography of the bay, which is relatively shallow. An alternative sampling gear, stationary plankton nets, has several advantages, including the ability to sample in locations where it may be unsafe for towed plankton nets (Dovel 1964, Graham and Venno 1967).

The purpose of the current study was to test the effectiveness of stationary plankton nets as a new gear in Saco Bay, Maine.

### Methods

Stationary plankton nets were fished opportunistically from July to October 2014 at two fixed stations for, on average, six daylight hours. These stations were selected based on similar depths profiles (~10 m) and because they were generally located in or out of the plume. The stationary plankton nets (0.5 m diameter, 333 um mesh), were constructed based on the methods of Graham and Venno (1968) and were equipped with a triangular vane (positioned on the mainline) to facilitate the positioning of the net's opening into the current, thus maximizing the amount of water flowing through it. Each stationary plankton net device consisted of two individual plankton nets, one positioned at the surface and equipped with a mechanical flowmeter (General Oceanics, Miami, FL USA), and a second

plankton net positioned at a depth of 3 m (below the plume), allowing for simultaneous fishing. This device was anchored to the ocean floor with a Danforth Anchor #22 Super Hooker and buoyed at the surface. In order to reduce the chance of fishing during retrieval, the stationary plankton net device was vertically hauled to the boat. The nets were then thoroughly rinsed with seawater to ensure all sample collected within the cod end to be preserved in 70% ethanol.

Each stationary net sample was examined using a Leica EZ4HD microscope in order to separate, quantify, and identify ichthyoplankton to the species-level. The abundances were standardized as number of ichthyoplankton per 100 m3. Fish eggs in each sample were enumerated, but not further identified taxonomically.

**Results**

The stationary plankton nets were set a total of 9 times out in Saco Bay, from July 21st to October 31th, 2014 (Table 1). Throughout this study period, 147 ichthyoplankton larvae and 3218 ichthyoplankton eggs were collected, with a total of 10 ichthyoplankton species observed (Table 2). The top five most abundant species were cunner, windowpane, fourbeard rockling, silver hake, and red hake, comprising 41%, 16%, 16%, 9%, and 4% of the total catch, respectively (Figure 1).

**Discussion**

Although there was sufficient sampling effort (9 sampling trips) in the current study, stationary plankton nets were not successful as a sampling gear in Saco Bay. For example, average ichthyoplankton densities obtained from stationary plankton nets were 95% lower than towed plankton nets, most likely due to a much lower volume of water

filtered (Dovel 1964 and Graham and Venno 1967). Thus, towed plankton nets are a much more efficient and effective sampling gear of ichthyoplankton in this region.

## References

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**Table 1.** The dates and tidal phase the stationary plankton nets were sampled, as well as how long the nets were soaked.

| Sampling date | Tide | Soak time |
|---|---|---|
| 7/21/2014 | Flood | 2 |
| 8/8/2014 | Ebb | 4 |
| 8/11/2014 | Ebb | 4.5 |
| 8/21/2014 | Ebb | 5 |
| 8/25/2014 | Ebb | 5.5 |
| 9/4/2014 | Mix | 8.25 |
| 9/12/2014 | Flood | 6.25 |
| 9/26/2014 | Flood | 6 |

**Table 2.** Ichthyoplankton species collected in the stationary plankton nets.

| Scientific name | Common name |
|---|---|
| *Peprilus tricanthus* | Atlantic butterfish |
| *Scomber scombrus* | Atlantic mackerel |
| *Tautogus adspersus* | cunner |
| *Enchelyopus cimbrius* | fourbeard rockling |
| *Hippoglossina oblonga* | fourspot flounder |
| *Syngnathus fuscus* | Northern pipefish |
| *Urophycis chuss* | red hake |
| *Merluccius billinearis* | silver hake |
| *Tautoga onitis* | tautog |
| *Urophycis tenuis* | white hake |
| *Scophthalmus aquosus* | windowpane flounder |

## Figure Legends

Figure 1. Ichthyoplankton species that comprise 1% or greater of the total catch collected from plankton tows (both sampling years) and stationary nets.

## Figure 1