

**PRELIMINARY INVESTIGATION OF THE POTENTIAL FOR
INTEGRATED AQUACULTURE OF THE SAND WORM (*NEREIS
VIRENS*) WITH ATLANTIC SALMON.**

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Introduction

The Atlantic salmon industry in Maine is based on net pen culture and one of the constraints to growth is the availability of sites. The industry is now on a solid upswing and looks set to return to the levels of production last seen in the mid 1990's. Continued growth is dependant on a number of factors including the ability to permit new sites and the maintenance of existing ones. Those sites more prone to benthic impact would be more productive if effective ways to mitigate impact from organic enrichment were found. Modern feeding practices ensure that very little feed is wasted but fecal wastes and uneaten feed can still build up in low energy or shallow sites.

Integrated aquaculture methods are seen as a way to improve sustainability of fish production systems. This is a traditional concept familiar to aquaculturists that has a long history dating back as far as the second century BC in China. The wastes or byproducts from the culture of one species, instead of resulting in a waste stream, which is lost from the system, can be managed so as to contribute to the nutrient requirement of another. This often makes economic as well as environmental sense.

New forms or improvements in the practice are frequently reported. A number of models and systems have been proposed and tested for marine species both in sea-based net pen operations and for land based recirculating systems (Troell *et al.*, 2003; Neori *et al.*, 2004).

Cooke Aquaculture is actively pursuing collaborative research into integrated aquaculture and already cultures kelp and mussels alongside salmon net pens (Chopin *et al.*, 2001). However, these species do not contribute to the metabolism of deposited organic wastes. Organisms that might feed on these wastes include deposit feeding detritivores such as polychaete worms. Some of these worms are highly valued as bait or as aquaculture feeds and are cultured commercially (Olive, 1999). One such polychaete is the sand worm, *Nereis virens*. This species is farmed on a commercial scale in the UK and Holland and on a pilot scale in the USA at the University of Maine's Center for Cooperative Aquaculture Research (CCAR). This worm is a native of Maine waters and has long been an important harvest for the state's fisheries.

Recent research, funded by MAIC, has shown that this species can grow well on marine fish wastes collected from filters in a recirculation system (Brown *et al.*, 2010). The potential for another polychaete worm, *Capitella*, to ingest and assimilate fecal waste from Japanese flounder was shown by Honda and Kikuchi (2002) and field seeding of cultured *Capitella* was shown to accelerate recovery of impacted areas under net pens in the Japanese study by Kinoshita *et al.* (2008).

The role of benthic organisms, including polychaete worms, in the decomposition and mineralization of organic material has long been appreciated in an ecological context (Aller, 1988; Kristensen, 2001). It has been shown that these organisms influence the transport and metabolism of organic matter through irrigation and reworking of the sediments, a process known as bioturbation. The abundance of these worms can indicate the impact of coastal aquaculture operations (Tomassetti

et al., 2005) and their activity is highly conducive to the recovery of sediments under aquaculture sites (Heilskov and Holmer, 2001; Heilskov *et al.*, 2006).

Nereis virens is found on both coasts of the North Atlantic and as far south as Virginia off the east coast of the US. Sandworms are typically an intertidal species, with highest natural densities found in the lower portions of the intertidal zone. They have been reported to inhabit a range of sediments from sandy mud to fine sands. In Maine, larval development occurs in the spring, and there is only a brief period of around 15 hours when the trochophore larvae might enter the plankton. For the most part they are a benthic species, and the juveniles settle within the intertidal zone about 16 weeks after fertilization. They are considered omnivorous and they play an important role in nitrogen cycling and biogeochemical processes. These factors make them a good candidate for sediment bioremediation, but since they are not normally found in high numbers in deeper water, deliberate seeding is required for bioremediation under fish pens.

This proposal describes a collaborative project that tests an innovative form of integrated aquaculture that might help to improve the environmental and economic sustainability of near shore salmon aquaculture in Downeast Maine. The industry employs some 325 people and is a significant economic driver for the State,. This methodology has the potential to enhance current industry practices and improve profitability, which in turn may lead to job creation and retention.

Methods

Location

The project was conducted at Cooke Aquaculture's Broad Cove salmon net pen site off Estes Head, Eastport, Maine (Figure 1). The site consists of twenty 100 m circumference polyethylene ring cages (Polar Cirkel type). The cage selected for the study is located in the northeastern corner of the site at cage 18, shown in Figure 2.

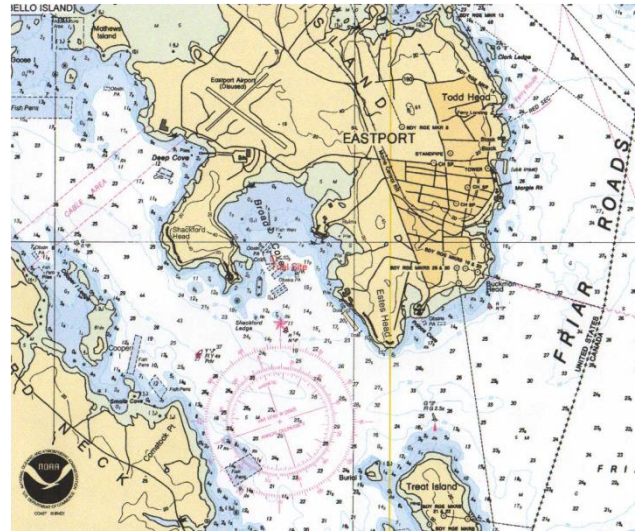


Figure 1. Chart showing Broad Cove lease site

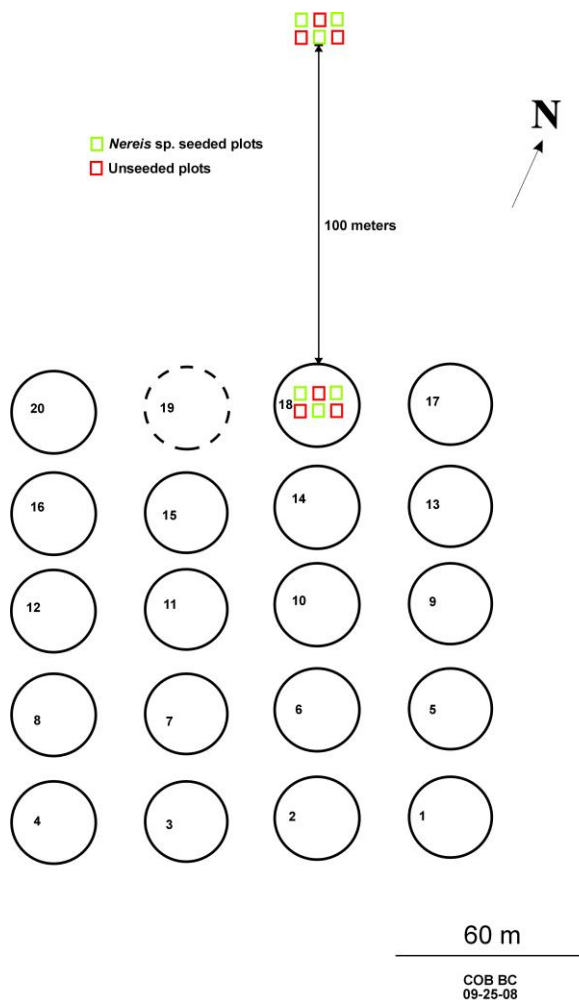


Figure 2. Broad Cove lease site and sampling plot layout.

Worm origin and transfer to site

The worms for this project were reared at the Center for Cooperative Aquaculture Research (CCAR). Just over 41,000 worms at an average weight of 0.74g were used for the trial. On October 1st 2009, these worms were harvested from the 1.8 m diameter, 20cm deep circular holding tanks and transferred to plastic trays (Figure 3). There were between 6230 and 7300 worms allocated to each of 6 sites, carried in 7 or 8 trays per site (see Table 1). Activated charcoal and calcium carbonate was added to the trays to maintain pH and to absorb metabolites.



Figure 3 Harvesting juvenile worms and placing into transfer trays

Table 1. Stocking schedule for worms

<i>Site</i>	<i># worms stocked</i>	<i>Av wt</i>	<i>Biomass (g)</i>	<i>Initial stocking density (kg/m²)</i>
RS1	6833	0.73	5006	1.25
RS2	7333	0.77	5643	1.41
RS3	7083	0.79	5593	1.40
PS1	7000	0.73	5090	1.27
PS2	7000	0.72	5010	1.25
PS3	6230	0.72	4472	1.12

On October 2nd 2009 the worms were transported overland by road and then by barge to the Broad Cove pen site in Eastport. Once on the barge, the containers were opened and fresh sea water was added.

Site layout and identification

Twelve 2m x 2m (4m²) frames were constructed using ¾ inch schedule 40 PVC pipe. Three of the corners were connected using a standard 90° elbow and the fourth corner connected using a 3-way 90° ell for insertion of a vertical pipe section to serve for attachment of a “flag” to ensure against loss of the frame in the event of burial in sediment or cage-related deposition (Figures 3). Small (~¼ inch) holes were drilled in the bottom of each frame member approximately 6 inches apart to allow the pipes to flood for placement on the bottom.

Six of the frames were designated as “Seeded” and spray painted yellow; the other six frames were designated as “unseeded” and spray painted red. A duct tape “flag” was placed on the upright pipe on each frame and secured in place with two standard 4 inch long plastic wire ties. Each flag was labeled according to its location (P- pen; R – reference), treatment (S – seeded; U- unseeded) and frame number (1 through 3) e.g. P U 3 for Pen, Unseeded number 3.

Three red frames and three yellow frames were placed under the approximate center of cage 18 arranged as shown in the Figure 2, i.e. alternating color pattern. The frames were anchored to the bottom with 2 to 3 approximately 6 inches long “u” shaped wire staples on each side of the frame. Once in place, two 4 inch diameter cores were taken randomly from within each frame for collection of sediment samples for sulfide and Total Organic Carbon measurement.

Nylon mesh pyramidal covers measuring slightly more than 2m x 2m were tied to metal reinforcing rod frames just slightly larger than 2m x 2m; two small buoys were attached to the middle inside surface of the cover to keep the mesh off of the bottom when in place. Covers were placed over each of the three yellow “seeded” frames to deter predation and prevent the worms from swimming away before burrowing (Figure 4).

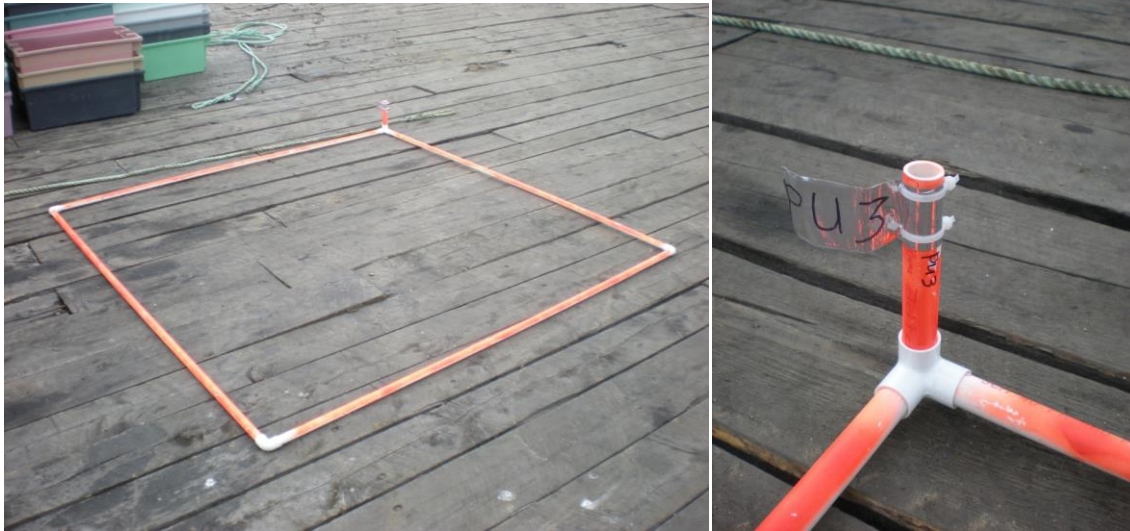


Figure 4. Typical 2m X 2m (4m²) PVC frame used for both seeded (yellow) and un-seed (red) plots. “Flag” incorporated into PVC frame to allow later detection of frames.

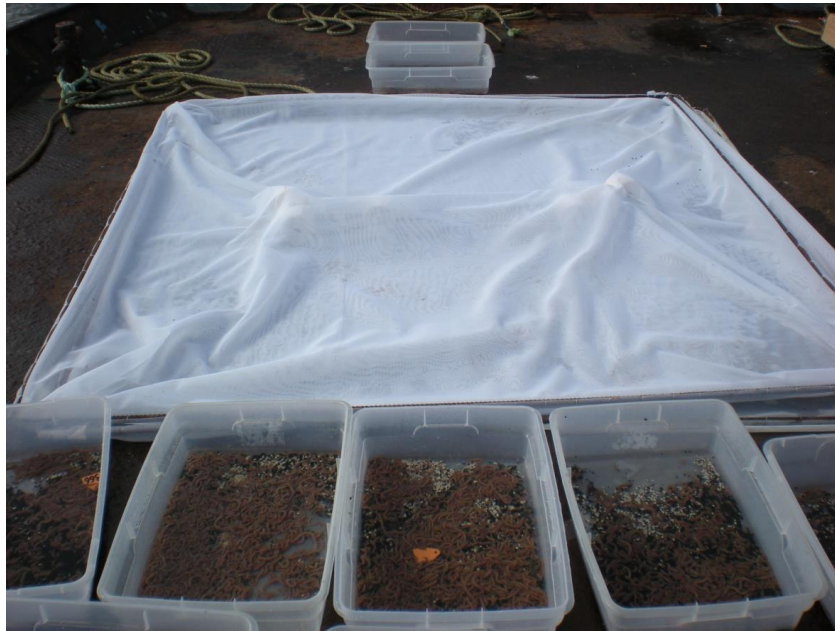


Figure 5. Mesh cover used to prevent predation and to keep worms from swimming off during initial seeding (background); trays with *Nereis* in foreground

Each group of sand worms allocated to a seeded plot was transported by divers from the surface in covered plastic trays and distributed over the bottom

within each of the covered frames. Once completely seeded, the covers were securely placed over the frames and the edges buried into the sediment.



Figure 6. Refreshing seawater in worm trays



Figure 7. Handing worms to divers

Sediment chemistry

Two sediment cores for sulfide measurement were taken at each plot using 4 in. diameter PVC pipe coring devices that were inserted to a depth of 10 cm or full resistance, whichever was greater. Sediment cores were removed from the corers by allowing the sediment column to slide out of the plastic corer so as not to disturb the sediment surface. Once exposed, the core surface material was removed down to a depth of 2 cm and the sediment placed in a small 125 ml plastic container and thoroughly mixed with a plastic spoon for approximately 1-2 minutes.

Sulfide measurement (*Method of Wildish et al., 1999*)

A 5 ml portion of the mixed sediment was removed with a modified 5 ml plastic syringe with the needle attachment end removed to form an open cylinder; the open end was immersed into the mixed sediment slurry and the sample extracted by pulling back on the plunger, thus obtaining a sample containing no bubbles. Immediately after obtaining the sample, the open end of the syringe was covered with plastic wrap insuring no air was trapped beneath the wrap. Aluminum foil was then placed over the end of the syringe to secure the plastic wrap in place. The syringe was then placed in a cooler with ice to maintain a temperature of <50C during transport to the laboratory for sulfide (S₂) analysis within >72 hrs. of sample collection.

Once at the lab, all syringes were allowed to warm to room temperature (≈200C) before analysis while the Accumet® AP63 pH/mV/Ion meter equipped with a Thermo Orion model 9616BN Combination Silver/Sulfide electrode filled with Thermo Orion Ionplus B Optimum Results™ Reference Electrode Filling Solution (900062) with standards prepared according to Wildish et al., 1999. The meter was standardized at 1.00 (100μM), 10.0 (1,000μM), and 100 (10,000μM). All samples were analyzed within a maximum of 3 hrs. Following analysis of all samples, measurements of the three standards were retaken and recorded on the calibration sheets. Actual S₂ μM values were calculated by multiplying the meter readings by 100.

Total organic carbon

Approximately 10-15 ml of sediment was taken as a subsample of the mixed sediment and placed in a labeled 7"x 3" 100 ml Nasco Whirl-Pak®; TOC samples were refrigerated until return to the lab, then frozen until delivered to the analyzing facility. TOC analysis was performed by the University of Maine, Ira C. Darling Center chemistry lab.

Nereis sampling

Two sediment cores for counting *Nereis* were taken at each plot as described above for sediment chemistry sampling. The contents of the cores were washed through a U.S. Standard No. 18 sieve (1mm mesh), all material retained on the screen was transferred into plastic sample jars, and the jars filled with 10% buffered formalin. Samples were subsequently transferred into 70% ETOH.

Counting and weighing of *Nereis* was done only for worms considered large enough to have been part of the original seeded population. Counting of *Nereis* was done by counting only heads. Worm weight was measured by initially separating whole worms from partial worms and the weights for each recorded separately. Worms were placed on absorbent towels and blotted just before weighing on a Denver Instrument Model APX-203 scale; worms were weighed quickly (within 1-2 minutes) to avoid excessive evaporation.

Results

Sampling synopsis

October 1-2, 2009 – established study layout beneath Cage 18 and at the Reference site. Cores taken for sulfide/TOC and baseline *Nereis* counts. Following sediment sampling, covers were placed over the frames and *Nereis* were distributed at approximately 7,000 per “seeded” plot within three plots at the cage and three plots at the Reference site. Covers were subsequently removed approximately 2 weeks later.

December 14, 2009 – sampling for *Nereis* was done in each of the three seeded plots at both the Cage and Reference locations. No sediment chemistry sampling was performed.

May 13, 2010 – sampling for *Nereis* was done in each of the (remaining) seeded plots at both the Cage and Reference locations. No sediment chemistry sampling was performed.

October 23, 2010 – Cores taken for sulfide/TOC and *Nereis* in remaining plots at the Cage; most plots were likely disturbed and moved from original location.

February 13, 2011 – sampling for *Nereis* within a presumed densely populated area based on holes seen by diver and on video. Twelve cores taken for *Nereis* counting; no sediment chemistry samples taken.

March 25, 2011 – returned final time to Cage to sample *Nereis* from within presumed populated area to determine weight of whole worms and test feasibility of suction sampling as a harvesting method.

Baseline sampling

Baseline counts of *Nereis* are shown in Table 2. All worms found were smaller than the hatchery reared worms used in the study. No worms were found at the Reference Sites.

Table 2. Baseline sand worm sampling at pen site, seeded (PS) and unseeded (PU), plots October 1, 2009.

Sample	Total whole <i>N.</i> weight (g)	# whole <i>N.</i> / sample	Mean wt./ <i>N.</i> (g)	Total # <i>Nereis</i> (heads)	Total # <i>Nereis</i> /0.1m ²	Density <i>Nereis</i> /m ²	Density <i>Nereis</i> /4m ² plot (by smpl)	Density <i>Nereis</i> /4m ² plot (mean)
PS 1-1	0.000	0	0.00	0	0.0	0	0	
PS 1-2	0.000	0	0.00	1	12.3	123	494	
								247
PS 2-1	0.000	0	0.00	3	37.0	370	1,481	
PS 2-2	0.000	0	0.00	4	49.4	494	1,975	
								1,728
PS 3-1	0.000	0	0.00	1	12.3	123	494	
PS 3-2	0.000	0	0.00	0	0.0	0	0	

Means	0.0	0.0	0.0	1.5	18.5	185		741

Sample	Total whole <i>N.</i> weight (g)	# whole <i>N.</i> / sample	Mean wt./ <i>N.</i> (g)	Total # <i>Nereis</i> (heads)	Total # <i>Nereis</i> /0.1m ²	Density <i>Nereis</i> /m ²	Density <i>Nereis</i> /4m ² plot (by smpl)	Density <i>Nereis</i> /4m ² plot (mean)
PU 1-1	0.000	0	0.00	0	0.0	0	0	
PU 1-2	0.000	0	0.00	0	0.0	0	0	
								0
PU 2-1	0.000	0	0.00	0	0.0	0	0	
PU 2-2	0.000	0	0.00	1	12.3	123	494	
								247
PU 3-1	0.000	0	0.00	0	0.0	0	0	
PU 3-2	0.000	0	0.00	0	0.0	0	0	
								0
Means	0.0	0.0	0.0	0.2	2.1	21		82

Growth and Survival

Figure 8 shows the decrease in survival over the trial period and change in average weight. On the first sampling after stocking out the worms, we found high survival (81%) and retrieved a good number of worms in all our core samples from the seeded plots at the pen site. Only one worm was present in samples taken at the reference site.

At the one-year sampling event, October 23 2010, only 4 frames were found. Of the four frames found, PU-1 appeared to be the only one remaining in place and undisturbed. PS-1 and PS-2 appeared to have moved, and PU-3 was upside down. Video recordings were made of the location of the frames and PU-3 was set right side-up. Four core samples were taken from each of the frames, two for sulfide/TOC measurement and two for *Nereis* counting. Only one large *Nereis* was found in the eight samples and it is uncertain whether this was from a seeded or unseeded location due to the uncertainty of frame location; however, based on size, we believe this could have been a seeded worm.

The disturbance to the frames appears to be the result of a dragger having caught a video transect line. This had been placed under the site, running under Pen 18, and appears to have caught the frames and pulled them out of position.

All six frames were found in place at the Reference Site. Four core samples were taken from each of the frames, two for sulfide/TOC measurement and two for *Nereis* counting, thus 12 sulfide/TOC and 12 *Nereis*; no *Nereis* were found in any of the samples.

Based on review of the video of October 2010 it was decided to return to Broad Cove on February 13 2011 to take numerous cores within what appeared to be an area of burrows possibly belonging to *Nereis*. A series of 12 cores were taken and sieved to determine presence of *Nereis*; no sediment chemistry sampling was done. In all but 2 cores, *Nereis* were found, though only 2 whole worms were recovered.

Based on the results of the 2/13/2011 sampling it was decided to return to Broad Cove yet again on March 25 2011 to take samples of larger areas to collect whole worms for weight and determine the feasibility of suction sampling.

A 2m x 2m frame was placed over an area considered a *Nereis* seeded area based on number of holes and the frame video recorded. A $\frac{1}{4}\text{m}^2$ frame was placed in three locations within the larger frame and 3 cores taken from each for sulfide sampling. The frames were suctioned sampled using a small airlift system and the effluent discharged through a mesh bag at the surface to collect any worms. Three 0.25 m^2 frames were placed in a non-seeded area and core sampled for sulfides; no suction sampling was done in the interest of keeping suctioning activity to a minimum. A total of 28 worms (heads) were recovered from the three 0.25 m^2 frames of which 12 were whole; worms were weighed at CCAR. The average weight was 5.1 g. Survival had decreased to an estimated 10% by that time.

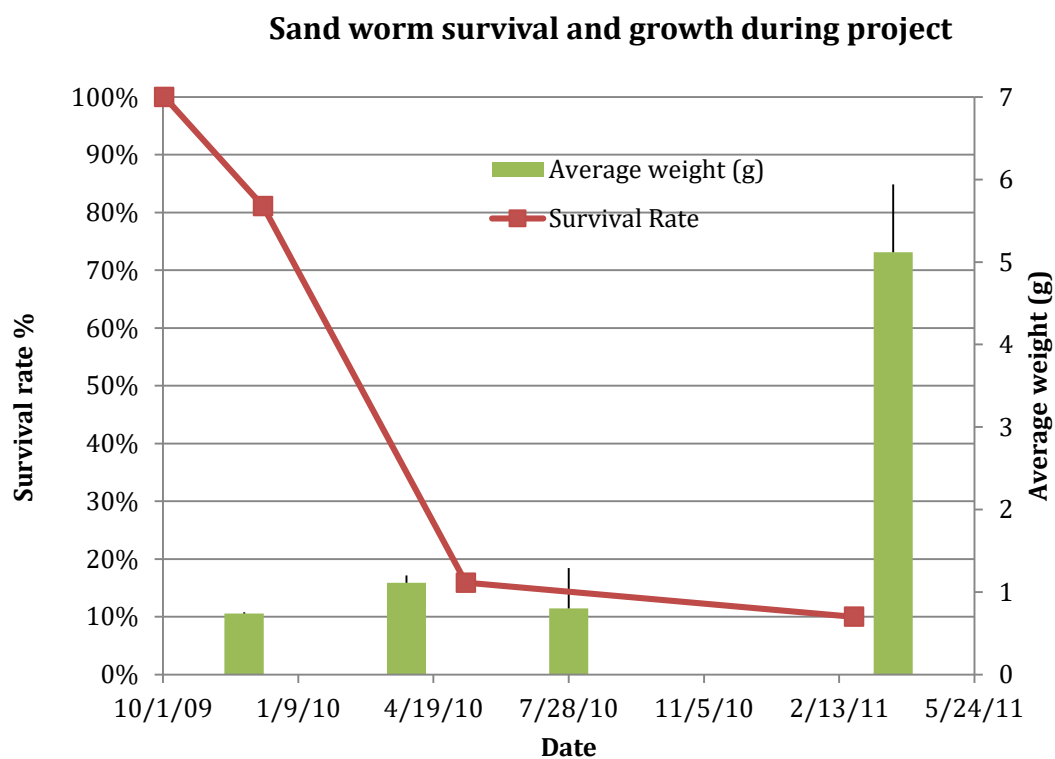


Figure 8. Graph of survival rate and average weight of sampled sand worms at each sample point. Error bars are S.E.M.

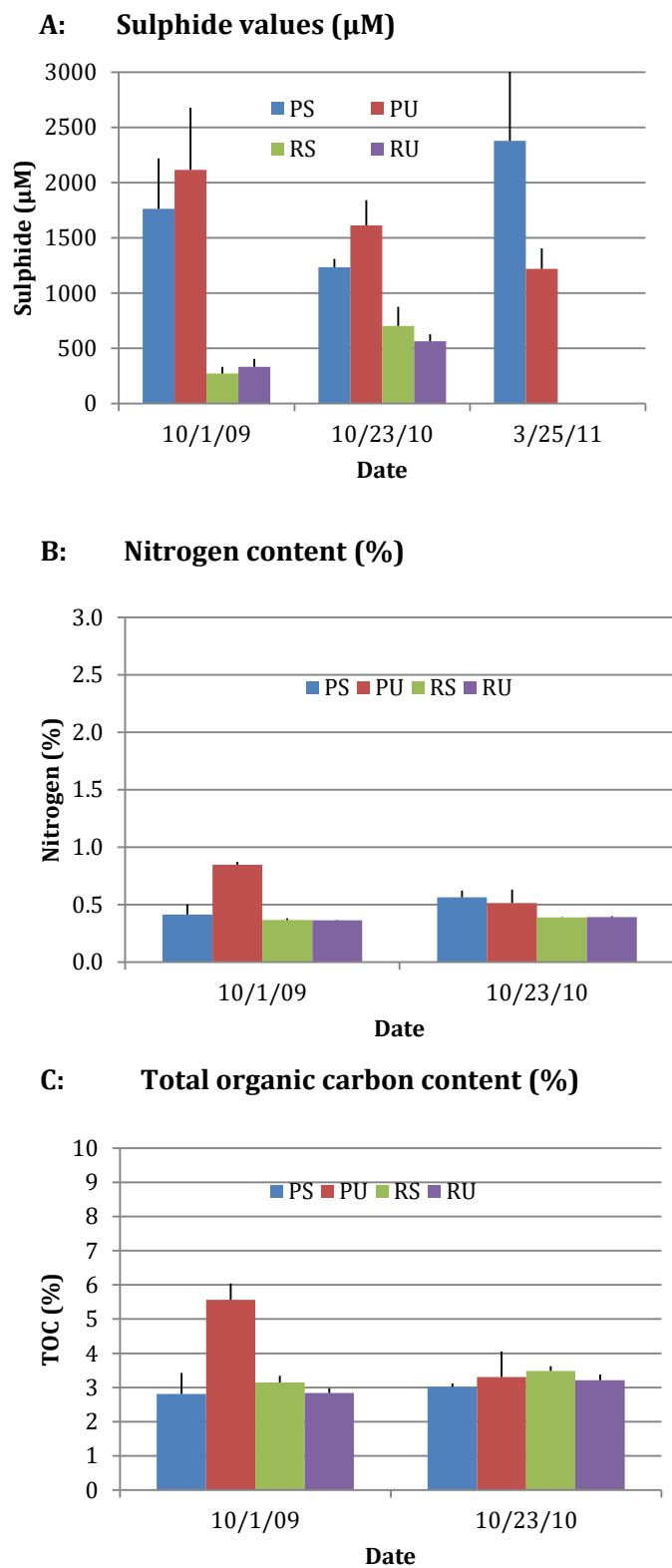


Figure 9. Values of sulphide, total organic carbon (TOC) and nitrogen from multiple core samples taken on 10/1/2009 (baseline all parameters), 10/23/2010 (all parameters) and 3/25/2011 (sulphide only). Error bars show S.E.M.

Changes in sulphide, total organic carbon and nitrogen are shown in Figure 9. For the first year of the project, sulphide levels were higher at the pen sites than at the reference sites. Sulphide decreased over this time and then at the last sampling 18 months into the project they increased at the (supposed) seeded location while showing a further decrease at the unseeded location.

There were few differences in TOC and nitrogen over time or between sites except that both were higher at the unseeded location in the baseline measurements.

Discussion

This project successfully demonstrated that it is technically feasible to stock out hatchery reared sandworms into the sediments under salmon pens. The high survival at 2 months demonstrated that the transport, predation or relocation of the worms themselves did not result in high losses.

The May 2010 sampling indicates a substantial decline in survival after 8 months; however, one of the seeded plots was not found suggesting some disturbance to the study site, which may have affected other plots.

The level of disturbance to the experimental site found in October 2010 significantly confounded proper sampling for the project. The remainder of the project was focused simply on finding evidence of persistence of the worms.

The recovery of worms in March 2011 with a mean weight of 5g, ranging as high as 10g, 1.5 years after seeding is encouraging and demonstrates the potential for *Nereis virens* to grow well in this environment.

Due to the problems encountered with the project it would be advisable to:

1. Establish the study site in the center of a site rather than under a perimeter cage
2. Ensure the location is free of any site video transect lines or other potential sources of interference
3. Ensure sufficient depth beneath the predator net to avoid any risk of net disturbance of the plots or bottom during extreme low tides.

Sampling for *Nereis* with corers results in numerous partial worms which makes both enumeration and true biomass estimation difficult since only heads are counted; other partial worm pieces not attached to counted heads are not counted, likely resulting in an underestimate of survival. Similarly, weighing of partial worms underestimates actual biomass. Also, the selection of worms considered large enough to have been part of the original seeded population may result in some underestimation of survival.

Some other sampling method needs to be developed that will allow whole animal sampling. Preserving samples on-site at the time of sampling for purposes of

enumeration is acceptable; however, for biomass determination worms should be kept live in sediment to be weighed whole live (wet weight), preferably on-site.

Airlift suctioning appears to be an effective way of removing *Nereis* from the bottom but results in excessive damage to the worms using conventional (urchin harvesting) equipment and techniques. Better results would likely be obtained with finer air input control, reduced air volume, finer bubbles, and a short travel distance through the airlift. However, the airlift distance needs to be sufficient to avoid elevating turbidity in the vicinity of the suctioning area.

The Reference site used for Broad Cove was established adjacent to a sunken ship to avoid the site being dragged by urchin and scallop draggers if placed elsewhere than near an obstruction. Sediment at the selected Reference site was substantially finer than at the experimental site beneath the cage and apparently unsuitable to *Nereis* since few persisted in the area following seeding. Locating a reference station not subject to dragging disturbance has often been difficult in Cobscook Bay.

It was hoped that some beneficial effect in terms of lower sulphide, TOC and nitrogen levels might result from seeding sand worms in the environment under salmon pens. However the analysis of the core samples did not show this. In view of the disturbance of the plot frames and our uncertainty over the actual location of the sampling areas, it is difficult to draw any firm conclusions from these results.

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Appendix 1

Benthic infauna analysis results of baseline 10/1/2011

PS-1

SPECIES level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	7	57	64	32.0	625.0
Abundance (organisms/0.1 m ²)	86	704	790	395.0	95249
Abundance-Caps (organisms/0.1 m ²)	86	691	778	388.9	91477
Species richness (No. species)	6	29	31	17.5	132.3
<i>Nereis</i> sp. (#/0.1m ²)	0	12	12	6.2	38.1
<i>Nereis</i> sp. (#/m ²)	0	123	123	61.7	3810
Distance in meters	0	0		0	
Rel. Diversity	0.976	0.890		0.933	0.002
% CAPITELLA	0.0	1.8		0.9	0.8

FAMILY level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	7	57	64	32.0	625.0
Abundance (organisms/0.1 m ²)	86	704	790	395.0	95249
Abundance-Caps (organisms/0.1 m ²)	86	691	778	388.9	91477
Family richness (No. families)	6	27	29	16.5	110.3
Distance in meters	0	0		0	
Rel. Diversity	0.976	0.894		0.935	0.002
% CAPITELLIDAE	0.0	1.8		0.9	0.8

PS-2

SPECIES level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	58	37	95	47.5	110.3

Abundance (organisms/0.1 m ²)	716	457	1173	586.4	16802
Abundance-Caps (organisms/0.1 m ²)	716	457	1173	586.4	16802
Species richness (No. species)	21	16	22	18.5	6.3
<i>Nereis</i> sp. (#/0.1m ²)	37	49	86	43.2	38.1
<i>Nereis</i> sp. (#/m ²)	370	494	864	432.1	3810
Distance in meters	0	0		0	
Rel. Diversity	0.875	0.914		0.894	0.000
% CAPITELLA	0.0	0.0		0.0	0.0

FAMILY level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	58	37	95	47.5	110.3
Abundance (organisms/0.1 m ²)	716	457	1173	586.4	16802
Abundance-Caps (organisms/0.1 m ²)	716	457	1173	586.4	16802
Family richness (No. families)	20	16	21	18.0	4.0
Distance in meters	0	0		0	
Rel. Diversity	0.881	0.914		0.897	0.000
% CAPITELLIDAE	0.0	0.0		0.0	0.000

PS-3

SPECIES level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	80	86	166	83.0	9.0
Abundance (organisms/0.1 m ²)	988	1062	2049	1024.6	1371
Abundance-Caps (organisms/0.1 m ²)	975	1049	2025	1012.3	1371
Species richness (No. species)	25	21	33	23.0	4.0
<i>Nereis</i> sp. (#/0.1m ²)	12	0	12	6.2	38.1
<i>Nereis</i> sp. (#/m ²)	123	0	123	61.7	3810
Distance in meters	0	0		0	
Rel. Diversity	0.849	0.770		0.810	0.002

% CAPITELLA	1.3	1.2		1.2	0.0
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FAMILY level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	80	86	166	83.0	9.0
Abundance (organisms/0.1 m²)	988	1062	2049	1024.6	1371
Abundance-Caps (organisms/0.1 m²)	975	1049	2025	1012.3	1371
Family richness (No. families)	23	19	29	21.0	4.0
Distance in meters	0	0		0	
Rel. Diversity	0.859	0.777		0.818	0.002
% CAPITELLIDAE	1.3	1.2		1.2	0.0

PU-1

SPECIES level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	19	128	147	73.5	2970.3
Abundance (organisms/0.1 m²)	235	1580	1815	907.4	452663
Abundance-Caps (organisms/0.1 m²)	136	839	975	487.6	123786
Species richness (No. species)	6	14	17	10.0	16.0
<i>Nereis</i> sp. (#/0.1m²)	0	0	0	0	0
<i>Nereis</i> sp. (#/m²)	0	0	0	0	0
Distance in meters	0	0		0	
Rel. Diversity	0.829	0.653		0.741	0.008
% CAPITELLA	42.1	46.9		44.5	5.7

FAMILY level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	19	128	147	73.5	2970.3
Abundance (organisms/0.1 m²)	235	1580	1815	907.4	452663
Abundance-Caps (organisms/0.1 m²)	136	839	975	487.6	123786

Family richness (No. families)	6	12	14	9.0	9.0
Distance in meters	0	0		0	
Rel. Diversity	0.829	0.664		0.746	0.007
% CAPITELLIDAE	42.1	46.9		44.5	5.7

PU-2

SPECIES level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	15	60	75	37.5	506.3
Abundance (organisms/0.1 m ²)	185	741	926	462.9	77152
Abundance-Caps (organisms/0.1 m ²)	111	679	790	395.0	80619
Species richness (No. species)	5	14	16	9.5	20.3
<i>Nereis</i> sp. (#/0.1m ²)	0	12	12	6.2	38.1
<i>Nereis</i> sp. (#/m ²)	0	123	123	61.7	3810
Distance in meters	0	0		0	
Rel. Diversity	0.893	0.781		0.837	0.003
% CAPITELLA	40.0	8.3		24.2	250.7

FAMILY level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	15	60	75	37.5	506.3
Abundance (organisms/0.1 m ²)	185	741	926	462.9	77152
Abundance-Caps (organisms/0.1 m ²)	111	679	790	395.0	80619
Family richness (No. families)	5	12	13	8.5	12.3
Distance in meters	0	0		0	
Rel. Diversity	0.893	0.773		0.833	0.004
% CAPITELLIDAE	40.0	8.3		24.2	250.7

PU-3

SPECIES level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	999	127	1126	563.0	190096
Abundance (organisms/0.1 m ²)	12333	1568	13901	6950.2	28970445
Abundance-Caps	8864	802	9666	4833.1	16246079

(organisms/0.1 m ²)					
Species richness (No. species)	10	8	12	9.0	1.0
<i>Nereis</i> sp. (#/0.1m ²)	0	0	0	0	0
<i>Nereis</i> sp. (#/m ²)	0	0	0	0	0
Distance in meters	0	0		0	
Rel. Diversity	0.378	0.708		0.543	0.027
% CAPITELLA	28.1	48.8		38.5	107.0

FAMILY level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	999	127	1126	563.0	190096.0
Abundance (organisms/0.1 m ²)	12333	1568	13901	6950.2	28970445
Abundance-Caps (organisms/0.1 m ²)	8864	802	9666	4833.1	16246079
Family richness (No. families)	9	7	10	8.0	1.0
Distance in meters	0	0		0	
Rel. Diversity	0.394	0.739		0.567	0.030
% CAPITELLIDAE	28.1	48.8		38.5	107.0

RS-1

SPECIES level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	21	18	39	19.5	2.3
Abundance (organisms/0.1 m ²)	259	222	481	240.7	343
Abundance-Caps (organisms/0.1 m ²)	259	222	481	240.7	343
Species richness (No. species)	8	9	13	8.5	0.3
<i>Nereis</i> sp. (#/0.1m ²)	0	0	0	0	0
<i>Nereis</i> sp. (#/m ²)	0	0	0	0	0
Distance in meters	0	0		0	
Rel. Diversity	0.842	0.902		0.872	0.001
% CAPITELLA	0.0	0.0		0.0	0.0

FAMILY level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	21	18	39	19.5	2.3
Abundance (organisms/0.1 m ²)	259	222	481	240.7	343
Abundance-Caps (organisms/0.1 m ²)	259	210	469	234.6	610
Family richness (No. families)	8	9	13	8.5	0.3
Distance in meters	0	0		0	
Rel. Diversity	0.842	0.902		0.872	0.001
% CAPITELLIDAE	0.0	5.6		2.8	7.7

RS-2

SPECIES level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	16	33	49	24.5	72.3
Abundance (organisms/0.1 m ²)	198	407	605	302.5	11011
Abundance-Caps	198	407	605	302.5	11011

(organisms/0.1 m ²)					
Species richness (No. species)	15	14	21	14.5	0.3
<i>Nereis</i> sp. (#/0.1m ²)	0	0	0	0	0
<i>Nereis</i> sp. (#/m ²)	0	0	0	0	0
Distance in meters	0	0		0	
Rel. Diversity	0.992	0.877		0.934	0.003
% CAPITELLA	0.0	0.0		0.0	0.0

FAMILY level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	16	33	49	24.5	72.3
Abundance (organisms/0.1 m²)	198	407	605	302.5	11011
Abundance-Caps (organisms/0.1 m²)	173	395	568	284	12344
Family richness (No. families)	14	12	18	13.0	1.0
Distance in meters	0	0		0	
Rel. Diversity	0.985	0.873		0.929	0.003
% CAPITELLIDAE	12.5	3.0		7.8	22.4

RS-3

SPECIES level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	16	43	59	29.5	182.3
Abundance (organisms/0.1 m ²)	198	531	728	364.2	27774
Abundance-Caps (organisms/0.1 m ²)	198	531	728	364.2	27774
Species richness (No. species)	10	15	20	12.5	6.3
<i>Nereis</i> sp. (#/0.1m ²)	0	0	0	0	0
<i>Nereis</i> sp. (#/m ²)	0	0	0	0	0
Distance in meters	0	0		0	
Rel. Diversity	0.964	0.832		0.898	0.004
% CAPITELLA	0.0	0.0		0.0	0.0

FAMILY level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	16	43	59	29.5	182.3
Abundance (organisms/0.1 m ²)	198	531	728	364.2	27774
Abundance-Caps (organisms/0.1 m ²)	198	531	728	364.2	27774
Family richness (No. families)	9	13	17	11.0	4.0
Distance in meters	0	0		0	
Rel. Diversity	0.956	0.832		0.894	0.004
% CAPITELLIDAE	0.0	0.0		0.0	0.0

RU-1

SPECIES level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	32	32	64	32.0	0.0
Abundance (organisms/0.1 m ²)	395	395	790	395.0	0.0
Abundance-Caps	395	395	790	395.0	0.0

(organisms/0.1 m ²)					
Species richness (No. species)	16	12	20	14.0	4.0
<i>Nereis</i> sp. (#/0.1m ²)	0	0	0	0	0
<i>Nereis</i> sp. (#/m ²)	0	0	0	0	0
Distance in meters	0	0		0	
Rel. Diversity	0.873	0.858		0.865	0.000
% CAPITELLA	0.0	0.0		0.0	0.0

FAMILY level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	32	32	64	32.0	0.0
Abundance (organisms/0.1 m²)	395	395	790	395.0	0.0
Abundance-Caps (organisms/0.1 m²)	358	370	728	364.2	38.1
Family richness (No. families)	14	12	18	13.0	1.0
Distance in meters	0	0		0	
Rel. Diversity	0.860	0.858		0.859	0.000
% CAPITELLIDAE	9.4	6.3		7.8	2.4

RU-2

SPECIES level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	49	31	80	40.0	81.0
Abundance (organisms/0.1 m ²)	605	383	988	493.8	12344
Abundance-Caps (organisms/0.1 m ²)	605	383	988	493.8	12344
Species richness (No. species)	14	14	21	14.0	0.0
<i>Nereis</i> sp. (#/0.1m ²)	0	0	0	0	0
<i>Nereis</i> sp. (#/m ²)	0	0	0	0	0
Distance in meters	0	0		0	
Rel. Diversity	0.708	0.847		0.778	0.005
% CAPITELLA	0.0	0.0		0.0	0.0

FAMILY level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	49	31	80	40.0	81.0
Abundance (organisms/0.1 m ²)	605	383	988	493.8	12344
Abundance-Caps (organisms/0.1 m ²)	605	346	951	475.3	16802
Family richness (No. families)	10	12	17	11.0	1.0
Distance in meters	0	0		0	
Rel. Diversity	0.733	0.853		0.793	0.004
% CAPITELLIDAE	0.0	9.7		4.8	23.4

RU-3

SPECIES level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	37	43	80	40.0	9.0
Abundance (organisms/0.1 m ²)	457	531	988	493.8	1372
Abundance-Caps	457	531	988	493.8	1372

(organisms/0.1 m ²)					
Species richness (No. species)	14	15	20	14.5	0.3
<i>Nereis</i> sp. (#/0.1m ²)	0	0	0	0	0
<i>Nereis</i> sp. (#/m ²)	0	0	0	0	0
Distance in meters	0	0		0	
Rel. Diversity	0.889	0.805		0.847	0.002
% CAPITELLA	0.0	0.0		0.0	0.0

FAMILY level analysis	Rep 1	Rep 2	Total	Mean	Var.
Total organisms	37	43	80	40.0	9.0
Abundance (organisms/0.1 m²)	457	531	988	493.8	1371
Abundance-Caps (organisms/0.1 m²)	432	518	951	475.3	1867
Family richness (No. families)	13	14	17	13.5	0.3
Distance in meters	0	0		0	
Rel. Diversity	0.871	0.796		0.834	0.001
% CAPITELLIDAE	5.4	2.3		3.9	2.4

