Project Completion Report
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PROJECT TITLE: Assessment of grow-out strategies for the green sea urchin

DATES OF WORK: July 1, 2009 to July 30, 2013 (includes 1 year no-cost extension)

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REASON FOR TERMINATION: Objectives completed.

PROJECT OBJECTIVES:
This project demonstrated and evaluated sea and land based aquaculture methods for green sea urchins, using hatchery seed. The project objectives were:

1) Compare two nursery strategies (sea cages vs. land based tanks) for growing green sea urchin seed (*Strongylocentrotus droebachiensis*) to a size (10-15mm) suitable for out-planting on sea bottom leases.
2) Demonstrate a land based recirculating seawater aquaculture system to on-grow green sea urchins to market size. Test different feeds, feeding strategies, culture densities, and husbandry methods.

3) Test and demonstrate sea ranching (out planting) for growing green sea urchins to harvest. Compare two ocean lease sites in terms of recovery rates and growth of out-planted sea urchins.
4) Compare growth, survival/retrieval, and economic costs/returns of sea ranching vs. tank farming.
5) Develop and disseminate extension and outreach materials on suitable techniques and the economic viability of green sea urchin aquaculture in the Northeast Region.

ANTICIPATED BENEFITS:

- Increase hatchery and nursery capacity for green sea urchin seed and develop more efficient methods
- Increase yields and economic opportunities for the sea urchin fishery using sea ranching
- Develop cost-effective methods for tank farming of sea urchins
- Encourage industry stakeholders to use aquaculture tools to revitalize the fishery

Sea urchin fishermen and dealers in Maine have expressed interest in using hatchery seed to reseed depleted grounds, as done in Japan. Unlike Japan, minimal public funds are available for reseeding so these efforts would have to be privatized. One way to do this is with sea ranching, where hatchery seed is released on bottom aquaculture leases to roam and feed at will, until capture after several years growth. Industry has been reluctant to try this due to concerns about seed costs; doubts that released seed would survive out-planting, grow, and remain within lease boundaries; and unwillingness to privatize fishing grounds. An alternative to sea ranching is tank farming, which offers potential for better survival and growth. However, the economic viability of land based aquaculture is uncertain due to higher costs and the lack of demonstrated methods. It was anticipated that this project might stimulate industry interest in sea urchin aquaculture by addressing these and other related questions. Adoption of aquaculture tools by the sea urchin industry could help revitalize a once economically significant fishery.

PRINCIPAL ACCOMPLISHMENTS:

1. Hatchery Production and Nursery Strategies

Altogether, ≈150,000 green sea urchin juveniles were produced during the course of this project and about 100,000 were used in the project itself. Green sea urchin seed production commenced prior to project funding in February 2009 at the Harris/Hill (Portsmouth, NH) and the Center for Cooperative Aquaculture Research (CCAR, Franklin, ME). Both hatcheries produced additional seed in 2010 and 2011. Sea and land based nursery methods were used to grow the hatchery seed to a size suitable for out-planting (≥ 10-15 mm test diameter). Simple mesh panels were shown to protect seed urchins from predation in the sea based nursery. In the land based nursery seed was reared in hydroponic plant baskets in a recirculating aquaculture seawater system. Seed reared through the nursery stage using either approach was used for field studies and on-growing.

2. Land-based On-growing of Green Sea Urchins to Market

We reared 9,200 green sea urchins from hatchery seed to near market size in a land based recirculating seawater aquaculture system. During the three year culture period trials were conducted to evaluate feeds, feeding strategies, stocking densities, and husbandry methods.

These trials provided valuable information for economic modeling and useful for any future tank farming efforts. As of July 2013 about 4,700 urchins (260 kg biomass) were large enough (≥ 45 mm TD) for market analysis, to proceed in 2013-2015 in a separately funded project.

3. Sea Ranching of Green Sea Urchins

We out-planted 21,000 hatchery reared sea urchins at two bottom leases and showed that the out-planted urchins will remain for extended periods within the release area. This has not been previously documented with green sea urchins and it has positive implications for sea ranching. However, growth data was equivocal; only a small number of out-planted urchins were close to the legal minimum harvest size after 27 months, and overall growth was relatively poor at both sites compared to tank farming. Nonetheless, this holds promise that sea ranching, if done at sufficient scale and on suitable grounds, can add value to the fishery over time.

4. Comparison of Sea Ranching with Tank Culture

This is the first time that sea ranching has been compared with tank culture using the same hatchery cohort. Methods, survival and growth, and costs were assessed and compared between the two methods. Tank farming resulted in better growth and survival (recovery) of the urchins, but high operating and feed costs and the long time to market (3+ years) are issues that must be addressed with innovation and research before this can be fully developed. Sea ranching is lower cost and presents less of an entry barrier to industry, but slow growth and losses of out-planted seed from natural causes or poaching remain as concerns.

5. Extension and Outreach

Industry stakeholders were involved throughout the project and kept informed of goals, methods and results. Steve Eddy and Larry Harris attended numerous Sea Urchin Zone Council (SUZC) meetings and both are voting members and are on the SUZC research sub-committee. Recent discussions at SUZC meetings have included reseeding and sea ranching as topics, along with consideration of creating one or more zones operated by fishing coops, where reseeding and other intensive management methods could be practiced. Paul Anderson and Dana Morse of Maine Sea Grant facilitated a panel discussion on sea urchin aquaculture for the 2012 Maine Fishermen's Forum and a class session on sea urchin aquaculture for the 2013 "Aquaculture in Shared waters" course. Steve Eddy gave a presentation at the 2013 Maine Fishermen's Forum on sea urchin aquaculture, attended by around 50 individuals. Graduate student Pamelia Fraungruber gave several poster and oral presentations, including at the Northeast Aquaculture Conference and Exposition 2010 and at the National Shellfisheries Association Meeting 2012. Over 400 pamphlets describing sea urchin aquaculture were disseminated at these and other forums, and the Maine Public Broadcasting Network did a segment on the project in 2011. The project will also be described in detail in a text on sea urchin aquaculture, currently in progress.

IMPACTS:

1. Hatchery and nursery production demonstrated to industry that there are reliable sources of green sea urchin seed available for commercial projects and reseeding. Several recent industry inquiries regarding hatchery capacity and seed costs can be attributed to the success of these efforts and to our outreach.
2. The project showed that out-planted sea urchins will remain within lease boundaries, resulting in three Maine-based companies to apply for bottom leases to culture green sea

urchins. Successful sea ranching could revitalize a fishery annually valued at around 5 million dollars in Maine.

3. We produced market sized sea urchins with tank farming, enabling us to obtain further funding to evaluate intensive land-based gonad enhancement aquaculture. The economic benefits of gonad enhancement aquaculture are readily appreciated and accessed by industry, and if successful could double the final market value of processed roe.

RECOMMENDED FOLLOW-UP ACTIVITIES:
The sea ranching trials showed promise but our ability to evaluate their true potential was limited by the small scale of the releases (10,500 seed at each of the two leases) and the logistical/funding constraints restricting the quadrat sampling spatially and temporally (we only sampled within the release areas and the surveys ended after 27 months). We recommend that further sea ranching trials occur but at a larger scale of at least 150,000 seed/acre and that sampling to evaluate tag absence/presence take place for at least three years post-release. We also recommend funding for studies to determine the longevity of fluorescent tagging beyond the 27 months we saw in this project.

We saw wide variation in green sea urchin growth rates at all life stages, and this likely has some genetic basis. We recommend a selective breeding program to see if time to market size (≥45 mm) in tank culture can be reduced from three years to 18 months or less. Green sea urchins can reach reproductive maturity at 25 mm (12-18 months), so it should be possible to produce three generations within 36-54 months. Improvements in hatchery methods and infra-structure are also needed to reduce seed production costs. Areas that should be addressed include survival through metamorphosis, more efficient settlement methods, and development of micro-diets for larval feeding.

Finally, we recommend that low cost formulated on-growing feeds be commercially developed. Formulated diets can significantly increase growth rates but the lack of available diets and their high cost currently make their use impractical. In addition, the effects of formulated feeds on roe culinary quality must be evaluated. This topic was beyond the project scope because most of the tank farmed urchins did not reach market size until year 4. The CCAR has obtained additional funding for 2013-2015 to conduct gonad enhancement trials, quality evaluation, a Taste Panel, and a market analysis of cultured sea urchin roe.

SUPPORT:

YEAR	NRAC-USDA FUNDING	OTHER SUPPORT					TOTAL SUPPORT
		UNIVER-SITY	INDUSTRY	OTHER FEDERAL	OTHER	TOTAL	
2009/2010	77,696	94,531					172,227
2010/2011	45,524	61,728					107,252
2011/2012	58,813	69,662					128,475

2012/2013	0	21,500 (est.)					21,500
TOTAL	182,033	247,421					429,454

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED

Publications in Print

Eddy, S. D., Brown, N.P., Watts, S. A., and A. Kling. 2012. Growth of juvenile green sea urchins *Strongylocentrotus droebachiensis* fed formulated feeds with varying protein levels compared with a macroalgal diet and a commercial abalone feed. Journal of the World Aquaculture Society. 43(2):159-173.

Manuscripts

Eddy, S.D. 2011. *Sea Urchin Aquaculture in the Gulf of Maine.* Tri-fold pamphlet describing the project and green sea urchin aquaculture efforts. Distributed at Maine Fishermen's Forum, Sea Urchin Zone Council meetings and other forums.

Fraungruber, P. 2013 (*in progress*). An Assessment of Growout Strategies for the Green Sea Urchin (*Strongylocentrotus droebachiensis*). MsC Dissertation. University of Maine, Orono, Maine.

Papers Presented

Eddy, S., Brown, N., Lawrence, A.L., Watts, S., and A. Kling. 2010. Effects of varying protein and carbohydrate levels on somatic growth of the green sea urchin *Strongylocentrotus droebachiensis.* Aquaculture 2010, San Diego, CA, 1-5 March 2010.

Eddy, S., Brown, N., Harris, L., and P. Fraungruber. 2010. Assessment of grow-out strategies for the green sea urchin *Strongylocentrotus droebachiensis*. Aquaculture 2010, San Diego, CA, 1-5 March 2010.

Fraungruber, P., Eddy, S., Brown, N., and L. Harris. 2010. Assessment of grow-out strategies for the green sea urchin *Strongylocentrotus droebachiensis*. Northeast Aquaculture Conference and Exposition 2010, Plymouth, MA, 1-3 December 2010.

Eddy, S. 2011. Feeds for sea urchin aquaculture and gonad enhancement. Cultivo Comercial de Erizo (Commercial Cultivation of Sea Urchins), Universidad Católica del Norte, Coquimbo, Chile, 30 June 2011.

Eddy, S.D., Wadsworth, J., Harris, L., Fraungruber, P., and G. Mouzakitis. 2012. From Capture to Culture: Opportunities for the Sea Urchin Fishery in Maine. Panel Discussion, 37th Maine Fishermen's Forum, Rockland, ME, 1-3 March 2012.

Eddy, S., Brown, N.P, and P. Fraungruber. 2012. Growth of hatchery reared green sea urchins *Strongylocentrotus droebachiensis* under various culture conditions. National Shellfisheries Association 104th Annual Meeting, Seattle, WA, 24-29 March 2012.

Eddy, S. 2012. Green sea urchin aquaculture in the Gulf of Maine: Can aquaculture help sustain a fishery? Aquaculture Research Institute Open House, University of Maine, Orono, ME, 24 August 2012.

Eddy, S. 2013. Prospects for green sea urchin aquaculture in the Gulf of Maine. 38th Maine Fishermen's Forum, Rockland, ME, 28 February-2 March, 2013.

Eddy. S. 2013. Green sea urchin aquaculture in the Gulf of Maine. Aquaculture in Shared Waters, Maine Sea Grant. Class session at the Center for Cooperative Aquaculture Research, Franklin, ME, 11 April 2013.

PART II

Objective 1) Compare two nursery strategies (sea cages vs. land based tanks) for growing green sea urchin seed (*Strongylocentrotus droebachiensis*) to a size (10-15mm) suitable for out-planting on bottom leases.

Improved survival and returns to the fishery are possible when seed is grown to 10-15 mm TD (test diameter) prior to out-planting. The period of growth from settlement to release size is the nursery stage, and for *S. droebachiensis* this may require 3-10 months. Nursery rearing can be done in sea based caging systems or land based tank systems. Sea based nurseries are less costly and may produce hardier urchins. Land-based nurseries, although costlier, allow for more intensive management to improve survival and growth. A sea based caging system located at the New Hampshire lease site and an intensive tank system located at the CCAR were used to demonstrate, improve, and compare nursery methods for green sea urchin seed production.

Methods

All of the Harris/Hill hatchery seed were released on-bottom or reared in sea based caging systems at the two Portsmouth Harbor sites in New Hampshire. The majority of the CCAR hatchery seed (≈60,000) were reared in tank-nursery systems at the CCAR, with the exception of 9,850 seed that were transferred to New Hampshire. Most of the CCAR seed urchins (36,100) came from the 2009 hatchery cohort and the remainder from 2010 and 2011. The land based nursery methods described here apply to the 2009 cohort.

Methods, Tagging and tag identification

Hatchery seed juveniles destined to be out-planted for nursery caging trials or release onto the bottom leases were tagged in the hatchery when they were 5-10 mm TD with a fluorescent dye. All urchins were tagged by two day immersion in a dye bath, either with Alizarin Red at 50 mg/l (Sigma-Aldrich Alizarin complexone Alizarin-3-methyliminodiacetic acid A3882) or tetracycline at 100 mg/l (Sigma-Aldrich Tetracycline T3258). Feeding continued throughout dying to ensure growth and incorporation of the dye into the calcareous structural skeleton. Fluorescence microscopy was used to visualize the tags at time of sampling. Sampled urchins were measured for weight and diameter in the laboratory. The jaws of each urchin were removed using forceps and placed in a sodium hypochlorite bleach solution to remove the soft tissue from the calcareous skeleton. Some individuals were too small to remove the mouthparts without causing damage by crushing. In these cases the entire body was immersed in bleach. The bleach solution does not degrade the fluorescent tags, and several days after jaws were immersed only the demipyramid structures and calcite teeth, which combine to form the jaws, remained. Samples were then rinsed with fresh water and dried. The samples were taken to the University of Maine campus in Orono, where a Zeiss stereo discovery microscope was used to visualize the fluorescent tags, which show clearly under a broad spectrum light with filters for red or green pigments (Figure 1a and 1b).

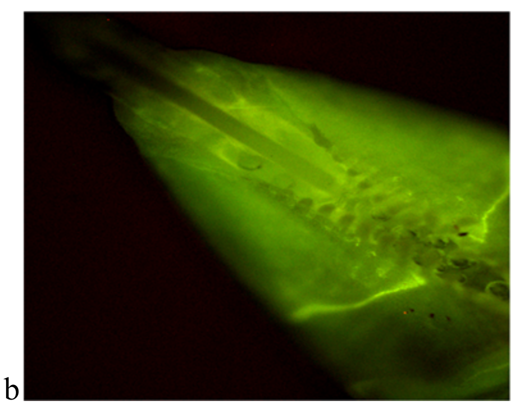
a  b 

Figure1 Sea urchin jaws dyed using Alizarin Red (a, left) and Oxytetracycline (b, right)

Methods, Sea based nursery

Two stations within the Portsmouth Harbor sea urchin research site were chosen. The first is a side channel off the main channel that connects Little Harbor to Portsmouth Harbor. There is less current and the bottom is a combination of cobble and shell fragments, primarily of the horse mussel *Modiolus modiolus*, both of which are encrusted with coralline algae. The second site is in the main channel adjacent to the bridge connecting Portsmouth to NewCastle. The current is stronger here during the incoming and outgoing tides and is most readily accessed during the periods near slack water. The bottom is also composed of cobble and shell fragments, but is less silty than the side channel. Three trials were undertaken at the first site and the second site was established for the third experiment. The first trial assessed the possibility of utilizing bottom cages for initial grow out. On 12 November, 2009, seven cages with 5,250 urchins from the CCAR and tagged with Alizarin red were deployed (750 per cage). The cages were all 50 x 50 x 15 cm in dimension and contained 15 (15 x 50 cm) fiberglass panels suspended vertically. The cages were elevated approximately 8 cm off the bottom by four bricks attached to the undersides of the cages. The second trial involved seeding 10 sets of 750 juvenile urchins without cages in defined areas marked by bricks left on bottom. The urchins were deployed on 14 June 2010. The third trial involved placing urchins under 0.5 m^2 square sheets of plastic coated wire mesh (1/2” x 1/2”) anchored by 4 bricks. Five sets of urchins left over from the first caging study were placed under wire sheets in the side channel on 27 June 2011 and another five sets of 920 urchins per set (4,600) from CCAR were out planted under sheets at the bridge site on the same date.

In addition to urchins supplied by CCAR, urchins produced in the Portsmouth Hatchery were out planted on two occasions. The first trial involved adding approximately 1000 1 mm juveniles to each of five cages that were placed on top of flat wire mesh sheets to provide a refuge under the cages for urchins that might leave the cages. The first trial began on 10 August 2010. The second trial involved placing 100 10 to 15 mm juvenile urchins in five cages and another 100 urchins each under five wire sheets at the main channel site on 14 April 2011. Other than the initial caging trial, all experiments are still in place and monitored at least twice a year.

Methods, Land based nursery

Tank-nursery culture at the CCAR occurred in two overlapping stages. The first stage began after metamorphosis/settlement onto plastic panels or floating media in a pair of 570L raceways (244 cm x 76 cm x 38 cm). About 770,000 competent larvae, as calculated with 1 ml volumetric counts, were transferred for metamorphosis from the larval rearing tanks into the settlement raceways. The raceways were supplied with flow-through filtered seawater in a refrigerated room tempered to maintain temperature between 9-14 °C. The newly settled urchins ('pinheads') began grazing upon surface diatoms and other microbes after jaw formation, or about 7 days post-metamorphosis. After 45 days the survivors were ≥2 mm TD and locally harvested kelp *Saccharina* sp. was added to the raceways. Larger urchins (≥4 mm) that moved on to the kelp for feeding were removed and hand counted into perforated hydroponic plant baskets (16 cm x 16 cm x 10 cm) in groups of up to 250 urchins per basket. As larger urchins were removed from the raceways they were replaced by others, and after another 30-60 days (90-120 days post-settlement) most of the seed had been transferred into baskets for the second nursery stage. A total of 38,000 urchins were transferred from the raceways into the baskets.

The baskets were suspended in foam floats or placed on plastic grating in shallow round tanks or raceways in a seawater recirculating aquaculture system (RAS). Aeration was provided at intervals between baskets to improve water circulation. Rearing temperatures were held between 9-14 °C, and the sea urchins were fed locally harvested kelp *Saccharina* sp. to satiation every 3-5 days. Growth was monitored by sampling 30 animals per basket at 2-3 month intervals. Test diameter was measured to 0.1mm using electronic digital calipers (Mitutoyo model CD-6"PMX) and blotted wet weight was measured to 0.1g using an electronic balance (A&D GF200, e=0.01g). Daily mortality records were kept to monitor the performance of the culture system, animals, and husbandry methods. Periodic hand grading and sorting was done to maintain uniform size ranges in the baskets.

Results

Tagging

A number of tagged urchins were held in the lab and sampled immediately and then at one month to determine tagging rates. These samples indicated 100% tagging rates. At the lease sites, fluorescent tagging bands were detected in sampled urchins at every dive sample up to the last in May 2012. This shows that the fluorescent tags can persist and be detected for up to 27 months after tagging.

Results, Sea based nursery

The survival of out planted urchins varied significantly from trial to trial with the greatest survival observed with small urchins protected by the flat wire sheets. Habitat type also appeared to make a difference with the greatest survival in structurally complex substrates dominated by shell fragments. The first caging trial was terminated on 14 June, 2010 for four cages and 20 July, 2010 for the final three. The cages were opened and all urchins were collected to be measured and data sent to CCAR for analysis. There was survival in six out of the seven cages, though the percentage of survivors varied greatly and growth was limited. Overgrowth of the cages by invasive (non-indigenous) algae and colonial tunicates (*Botrylloides violaceus* and *Didemnum vexillum*) reduced light and water flow within the cages. The second out planting experiment is still being monitored, but survival has been very low, with only 16 total urchins still present over the 10 quadrats. None of the urchins has reached the minimum legal size of 52 mm. In the third trial the original caged urchins have done very well and have remained associated with the protective wire sheets. The sheets in the side channel were turned over on 9 March 2012 and numbers have increased with larger urchins attracted to the structure and epibiotic growth on the wire mesh. Survival of the urchins at the main channel site is also good, but there appears to be limited growth without turning over of the sheets to create open space for more movement. On 18 July 2012, there were respectively 94, 84,118, 55 and 10 urchins associated with the five sheets in the side channel and a number of the urchins were of legal size.

The caging experiments using Portsmouth Hatchery urchins were not as successful. The survival of 1 mm urchins was poor and only a few individuals have been observed, though the cages are still in place and yet to be destructively sampled. The attempt at caging juveniles in the main channel was hampered by loss of cage tops due to drag from algal growth and strong tidal currents, which was not an issue in the side channel. The survival of urchins under the sheets appears to be similar for both the urchins raised at the CCAR and those from the Portsmouth Hatchery.

Results, Land based nursery
The end of the tank-nursery period came when ≈85% of the population was ≥10 mm TD, at about 10 months following metamorphosis/settlement. Survival through the entire nursery period was estimated as 4.7%. Most of the mortality likely occurred very early with urchins that failed to successfully make the transition from metamorphosis to the onset of exogenous feeding. Of the estimated 770,000 competent larvae stocked into the settlement raceways only 38,000 seed (4+ mm) were hand counted out into the nursery baskets for the second nursery stage. Survival through the second nursery stage in the plant baskets was 95%, and 36,100 viable juveniles were produced for the out-planting and tank trials.

The mean test diameter at the end of nursery culture was 11.0 ± 3.7 mm. 12.1% of the population was smaller than the minimum recommended release size of 10 mm, 82.6% was between 10-15 mm, and 5.3% was 15.5-36 mm. The population was randomly mixed together in February 2010 and 21,000 were randomly chosen for release at the Penobscot Bay leases. Another 4,600 were transferred to the Gingrich lease site for use in nursery caging trials, and 10,500 were kept in the CCAR facility to be used in the land based on-growing trials.

Discussion of hatchery and nursery production
The low survival (4.9%) we observed at the CCAR following settlement/metamorphosis to a size of 4-5 mm has been seen with other sea urchin species. Investigators have proposed several causes, including insufficient or inappropriate diatom species available for feed in the settlement tanks, harmful microbes, predation by copepods or nematodes, and poor maternal egg quality or larval nutrition. Japanese researchers have seen post-settlement survival rates as high as 60-70%, but it is unclear as to whether this is the rule or the exception. Improving post-settlement survival would increase hatchery efficiency and reduce seed production costs, and this is clearly an area requiring further work.

The hydroponic plant baskets proved to be efficient and effective nursery containers. The perforated baskets allowed water and wastes to pass through. The baskets facilitated feeding and minimized direct handling of the animals, and the side walls increased the total surface area available for urchin attachment relative to the surface area provided by the tank itself. Survival in the baskets over the course of 8 months as the juveniles grew from ≈4 mm to 10+ mm TD was 95%. This compares favorably with sea cage nurseries, where survival rates can be variable and subject to unpredictable natural events. In a previous study where we used mesh tubes attached to on-bottom oyster cages as a nursery, survival ranged from 56% to 89%.

At this point we cannot recommend sea based caging systems such as those tested at the Portsmouth Harbor site over land based nursery systems such as those used at the CCAR. Survival in the initial caging study at the Portsmouth Harbor site was highly variable and it is unlikely that utilizing a field-based cage system for the nursery phase of juvenile urchins is a viable approach. If done at a larger scale than in this project the amount of bottom gear and the extended length of time that it needs to remain on bottom before the urchins are at release size (9-12 months) might make on-bottom or floating cage systems unworkable. Additionally, when gear is included in an aquaculture lease application it complicates the process and potentially creates opposition to the lease. This could be especially true at sites where other types of fishing occur, such as dragging or lobstering. There are three alternatives to sea based nursery cages that may be viable. The first option is to seed very young juveniles directly into structurally complex habitats in the winter when most predators are not active. The Portsmouth Hatchery can produce more than 2 million juveniles in a single run and out-planting newly settled urchins in the right habitat may be the most cost effective approach. The second approach is to use a land based initial grow-out phase (nursery) as has been utilized by the CCAR. Land based nurseries offer some advantages, such as the ability to cull out slow growing seed while at the same time improving overall growth rates and survival. Adding lights and a flow through seawater source so that natural epibiont growth provides the food source could help reduce labor and feed costs. In an earlier study we showed that formulated feeds with about 20% protein can significantly increase juvenile growth rates. This shortens the nursery phase but it might not reduce costs due to the high expense of formulated diets vs. natural feeds such as field collected kelp or epibiont growth. Also, it is not known if survival might differ between hatchery seed reared on formulated feeds vs. seed reared on natural feeds once the seed is released on bottom. The third option is to combine juvenile grow out with another species, such as oysters, scallops or mussels to provide a value added and complementary culture species. This takes advantage of existing culture methods and equipment to reduce costs and increase value. Ultimately, it will be critical to be selective about where and when to utilize hatchery-produced juveniles. Larval cultivation to settled juveniles is well understood, but there is still much to learn about how to maximize field-based production of out planted juveniles, and this will be the most cost effective strategy for most markets for the foreseeable future. Sea ranching of urchins after the initial growth phase is likely the most economical approach for a species that requires high volume and shows relatively slow growth, such as the green sea urchin in the Gulf of Maine.

Objective 2) Demonstrate a land based recirculating seawater aquaculture system to on-grow green sea urchins to market size. Test different feeds, feeding strategies, culture densities, and husbandry methods.

Land-based aquaculture of sea urchins offers several potential advantages over sea based aquaculture methods such as sea ranching. Because temperatures can be controlled and feeding optimized in a land based system it can be possible to have improved growth and survival. Husbandry operations such as grading and culling can be carried out to focus on the best performing animals. However, energy, labor, and real estate costs can be much higher for a land based operation and these costs may negate any gains. With this objective we sought to test and demonstrate efficient methods for tank farming sea urchins and quantify growth, survival, and some of the associated costs.

Methods

Methods, Tank culture system

The CCAR designed and built a tank system and RAS for sea urchin growout and stocked it in November 2010 with 9,200 juveniles from the 2009 hatchery cohort. Rearing tanks were configured to maximize internal surface area available for attachment, allow for efficient feeding and waste removal, and permit observation and easy access to the urchins. Ultimately we wanted to provide a low cost do-it-yourself design that could be built by fishermen and start-up companies. V-shaped troughs with perforated floor plates were fabricated out of dimensional lumber and plywood covered with fiberglass.   Each set of paired troughs was assembled with three sheets of 1.2 m x 2.4 m (4 ft x 8 ft) exterior grade plywood supported with 5 cm x 10 cm (2 in x 4 in) boards and plywood ribs.   The side walls were 61 cm (2 ft) wide and 2.4 m (8 ft) long and sloped at 45°. A perforated PVC plate over a half-round 10 cm (4 in) diameter PVC pipe formed a drainage channel to capture wastes, which were flushed by pulling a pipe on an external standpipe. Six pairs of V-troughs were piped into a recirculating aquaculture system (RAS) equipped with a parabolic filter for solids removal, moving bed biofilter, foam fractionator with oxygen injection, and UV sterilizer (Figure 2).   The urchins were reared in this system up to the date of this report (July 2013). Growth and survival were monitored over the course of three years, and water quality parameters were measured either daily (oxygen, temperature) or weekly (total and un-ionized ammonia, nitrite, nitrate, pH, alkalinity, and CO2). Three trials were carried out to assess feeds, feeding regimes and stocking densities.

Figure 2: Urchin grow out system built for NRAC project.

Methods, Feeds and feeding of sea urchins in the tank system

Two production scale feeding trials were carried out to evaluate different feeds and feeding rates. An earlier trial (2008) with small juveniles (mean = 5.5 mm TD) demonstrated that tank reared urchins have better growth when fed formulated diets compared to kelp fed urchins. Eight diets formulated for sea urchins by the Texas A&M Feed Labs and varying in protein (16% to 40% protein) and carbohydrate (29% to 49% carbohydrate) were compared to each other and to a commercially available abalone diet and the kelp *Saccharina latissima*. Diets with about 20% protein gave the best results in that study.

However, the Texas A&M diets were not available in sufficient quantity (>100 kg) to support a commercial scale demonstration project, so a sea urchin feed imported from Norway was used (Nofima). The Nofima diet was formulated for green sea urchins and had a proximate composition of 21.3% protein, 46.2% carbohydrate, 7.5% fat, 14.2% ash, and a carotenoid pigment. A sinking pelleted catfish feed manufactured by Cargill was also tested as a low cost alternative. The Cargill diet was 32% protein, 5% fat, and 10% fiber. The bulk of the protein was from cereal grains and some offal, and the diet did not include a carotenoid pigment source or any marine derived lipids. In these respects the catfish feed appeared to be less than optimal for sustaining sea urchin health and growth, but other researchers have used maize (corn) based diets with satisfactory results to feed the European sea urchin. The Nofima and Cargill diets were compared at production scale using the entire population. The urchins were sorted by three size grades into seven V-troughs and each tank/size grade was fed either the Nofima or Cargill diet at ≈2% biomass once every three days for 281 days. Weights and test diameters were measured (as described previously) for a random sample of thirty urchins from each tank at days 0, 184, and 281. The daily specific growth rate (SGR) was calculated for each tank as SGR (%) = [((Ln whole wet weight (t2)) - (Ln (whole wet weight (t1))) / ((t2) - (t1))] x 100. At day 281 all groups on the Cargill diet were switched to the Nofima diet and the trial was continued for another 56 days to look for evidence of compensatory growth in the Cargill fed urchins.

During the Nofima/Cargill feed trial it was observed that all of the feed was generally consumed within 2-3 days, but it was unclear if 2% ration at 1x/3days maximized both somatic growth and economic return. A second trial was conducted to investigate the effects of different feeding frequencies on growth and feed conversion. The urchins were size graded into twelve V-troughs; three tanks held 'small' urchins (30-34 mm TD at 12-15 g), five held 'medium' urchins (35-40 mm at 18-23 g) and four held 'large' urchins (>40 mm at 28-60 g). All tanks and size grades were fed Nofima at a ration of 1% biomass at varying frequencies; five tanks were fed 1x/3 days, three were fed 1x/7days, and four were fed 1x/14 days. Size measurements (weight and diameter) were done as previously described on a random sample of thirty urchins per tank at 2-3 month intervals, and the feed ration was re-adjusted to account for growth. Specific growth rates (SGR) and feed conversion ratios (FCR) were calculated over the course of the 9-month trial.

Methods, Stocking densities of sea urchins in the tank culture system

Previous studies have recommended a maximum tank stocking density of about 6 kg/m^2 for green sea urchins reared in raceways with 90° vertical walls. We were interested in determining if higher stocking densities could be achieved in the slanted wall V-troughs, without affecting growth or survival. Each V-trough had 2.67 m^2 of submerged surface available for urchin attachment . Seven of the twelve troughs were initially stocked with ≈9,200 juvenile urchins graded into three overlapping size ranges (9-18 mm; 15-26 mm; 22-33 mm) at densities ranging from 0.5 to 2.9 kg/m^2. The urchins were size sampled at 2-3 months throughout the course of the project and stocking densities were re-calculated at each sampling interval. The population was graded following one year of growth (Nov. 2011) into three additional tanks at densities ranging between the ten tanks from 5.1 to 14.8 kg/m^2. Growth and survival were then measured for another year, until Oct. 2012.

Results

Results, Performance of the tank culture system

The tank culture system operated continuously from December 2010 to July 2013 with no significant failures. Critical water chemistry parameters (NH_3, NO_2, CO_2) were always below the threshold levels reported as harmful to green sea urchins by other investigators (un-ionized ammonia ≤ 0.016mg/l, nitrite ≤ 0.5 mg/l, and CO2 ≤ 18mg/l) (see papers by Siikavupoio *et al*, 2004-2007). Oxygen typically ranged from 7.0 to 10.0 mg/L. The temperature declined to as low as 2°C in the winter to as high as 19°C in the summer, but only for brief periods (1-2 weeks), and for the most part it remained within the optimum range of 8-14°C. Mortality rates increased when temperatures exceeded 16°C, but despite this the total mortality during the first two years was less than 5% (425 urchins, or 4.6%). After 25 months of on-growing (or 44 months post-settlement) 56% of the population was ≥ 40 mm, but less than 5% of the population was at or near the legal minimum harvest size of 52 mm (Figure 3).

During spring of the third year (March-April 2013) the population experienced a 2-month chronic mortality event triggered by handling and grading. In affected tanks the urchins developed purple lesions with subsequent spine loss and mortality. Diagnostics indicated that the prevalent bacterial isolate was *Vibrio vulnificus.* After a period of aggressive culling of symptomatic urchins the mortality abated, but total mortality from this event was about one-third of the entire tank population. As of July 2013 4,620 remaining urchins were large enough (≥ 45 mm) to be included in a separately funded market enhancement and quality evaluation project. Approximately 1,500 remained too small for market but will be used in future out-planting studies.

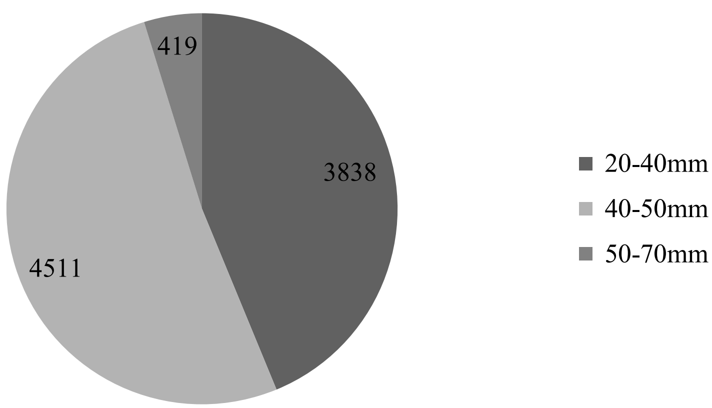


Figure 3. Number of tank urchins in three size categories after 25 months growth in an RAS.

Results, Feeds and feeding

Feeding trial #1: Comparison of Nofima sea urchin diet vs. Cargill catfish diet

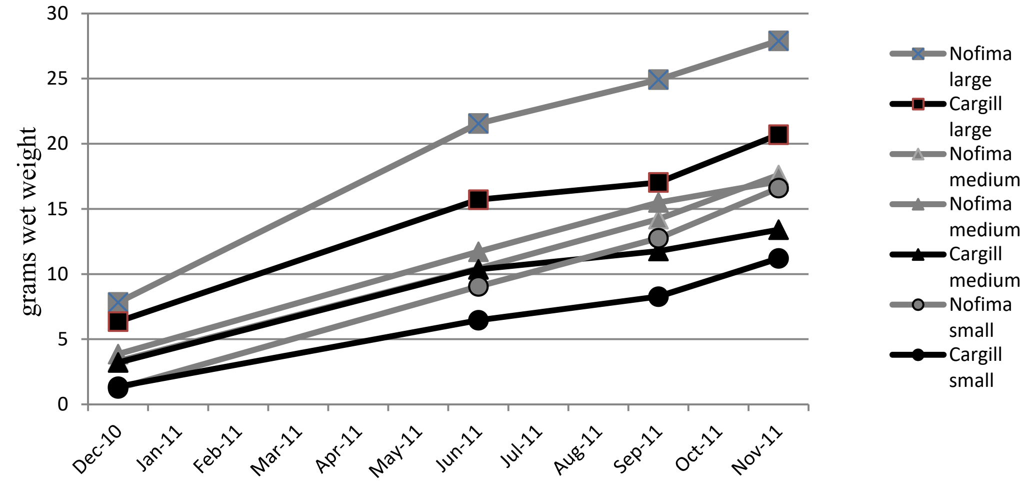


Figure 4. Growth of green sea urchins of different size categories over an 11 month period when fed the Nofima urchin diet or the Cargill catfish diet. Small=10-18mm, avg. 1.3g; medium=16-24mm, avg. 3.4g; large=22-30mm, avg. 7.1g. **Growth rates were better for urchins fed the Nofima diet vs. those fed the Cargill diet, for urchins in all size categories.**

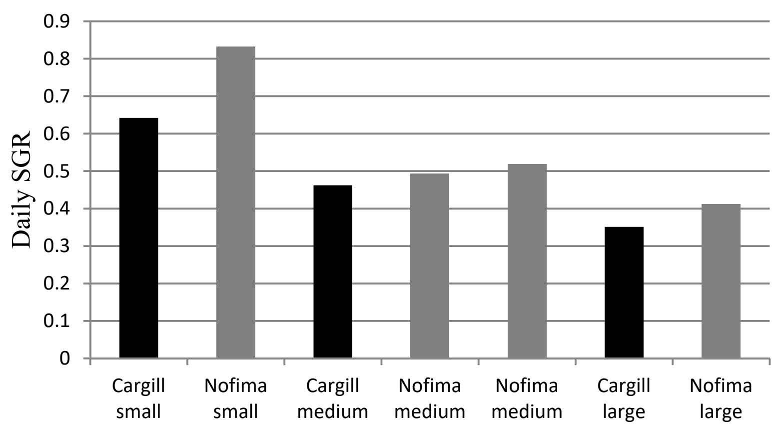


Figure 5. Daily specific growth rates for different size classes of green sea urchins fed either the Nofima sea urchin diet or the Cargill catfish diet. Small=10-18mm, avg. 1.3g; medium=16-24mm, avg. 3.4g; large=22-30mm, avg. 7.1g. Each bar represents one tank and significance levels were not calculated due to the lack of replication between treatments. **The Nofima diet outperformed the Cargill diet in all urchin size categories.**

Feeding trial #2: Effects on growth and feed conversion when Nofima was fed at different rates

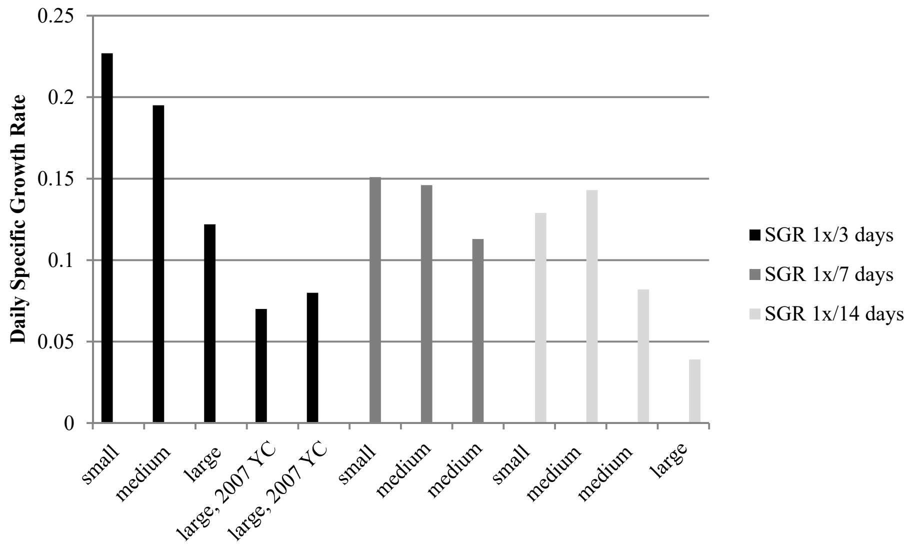


Figure 6. Specific growth rates (SGR) of green sea urchins of different size categories fed at frequent (1x/3 days), less frequent (1x/7 days) and infrequent (1x/14 days) intervals. Small=30-34 mm TD and 12-15 g; medium=35-40 mm and 18-23 g; large>40 mm and 28-61 g. Each bar represents one tank and significance levels were not calculated due to the lack of replication between treatments. **Growth was improved with more frequent feeding, but only for urchins <40 mm.**

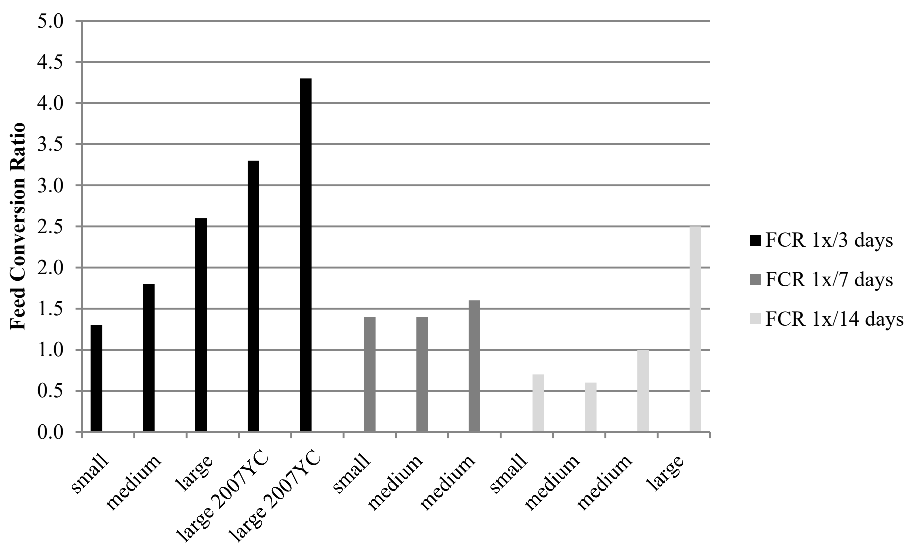


Figure 7. Feed conversion ratios (FCR) of green sea urchins of different size categories fed at frequent (1x/3 days), less frequent (1x/7 days) and infrequent (1x/14 days) intervals. Small=30-34mm TD and 12-15g; medium=35-40mm and 18-23g; large>40mm and 28-61g.

Each bar represents one tank and significance levels were not calculated due to the lack of replication between treatments. **Feed conversion was more efficient at reduced feeding levels, but only for urchins <40mm.**

Results, Stocking densities

Tank	# of urchins	Avg. wt (g)	Start density (kg/m^2)	Avg. wt (g)	Final density (kg/m^2)	# of urchins	Avg. wt (g)	Start density (kg/m^2)	Avg. wt (g)	Final density (kg/m^2)
DATE	*12/11/10*	*12/11/10*	*12/11/10*	*11/8/11*	*11/8/11*	*1/10/12*	*1/10/12*	*1/10/12*	*10/3/12*	*10/3/12*
1A	1,500	1.4	0.8	11.2	6.3	1,198	14.9	6.7	27.1	11.6
1B	1,500	3.3	1.8	17.6	9.9	989	19.5	7.2	28.6	10.1
2A	1,000	6.4	2.4	20.7	7.8	669	34.0	8.5	37.7	9.1
2B	1,500	3.2	1.8	13.4	7.5	1,184	12.9	5.7	18.1	8.0
3A	1,012	7.8	2.9	27.9	10.6	1,000	18.2	6.8	30.4	10.2
3B	1,505	3.9	2.2	17.1	9.6	666	28.5	7.1	39.3	9.1
4A						680	34.8	8.9	42.5	10.3
4B	1,176	1.2	0.5	16.6	7.3	772	22.8	6.6	30.7	8.8
5A						649	60.7	14.8	73.9	16.3
5B						791	20.0	5.9	29.1	8.6
6A						834	22.4	7.0	27.8	8.6
6B						961	14.2	5.1	21.1	7.6

Table 1. Green sea urchin starting and ending weights and tank stocking densities before and after grading (shaded area). **There was no difference in growth or mortality seen at any stocking density. The highest stocking density we achieved was 16.3 kg/m^2, at which point there was 20% or less open surface area available for urchin movement and growth.**

Discussion of land based tank culture

Tank culture system

Our previous experience using flat bottom raceways with 90° walls showed that sea urchins strongly preferred attaching to the walls and did not utilize the floor area. This made it impossible to evenly distribute feed to all of the animals other than those along the top row. The flat bottom collected wastes, which could only be removed with laborious siphoning. The 45° wall design mitigated these issues and in particular was extremely effective at allowing even distribution of formulated feed onto a tightly packed population of surface attached animals with limited capacity for movement. As the feed rained down on the animals their tube feed rapidly grasped onto the feed pellets and moved them to the aboral mouth for consumption, and within 5-10 minutes most of the feed was no longer visible. Fecal pellets excreted out of the anal/genital pore located on top of the animal readily drifted down onto the perforated plate. Unfortunately, the perforated plates did not altogether eliminate the need for tank siphoning, but the narrow trough greatly reduced the area to be siphoned and hence the time required to do it. We also discovered, to our chagrin, that sea urchins will eat holes into textured areas of fiberglass gel coat, and this required some repair efforts in year 3 of tank on-growing. This appeared to be less of a problem in tanks where the gel coat was applied to give a smoother finish.

The tank design could be improved while still retaining the basic elements of slanted walls and a perforated or screened bottom. Lightweight molded plastic or fiberglass tanks that can be stacked at two or more levels will make more efficient use of floor space and reduce space heating/cooling costs. These stacked troughs would be similar to those previously used by Mick Devin *et al*., but deeper and wider. The floor channel should include a series of evenly spaced sump drains along the length (rather than a single drain at the end). We found that the single drain caused wastes to build up underneath the plate due to the lack of current. The wastes did not readily flow down the end drain when it was purged, requiring that a hose be snaked underneath the plate to free up the wastes. A series of drain sumps along the length of the channel should mitigate this problem.

We showed that green sea urchins can thrive in a seawater RAS. The 2.4 m^3 capacity moving bed biofilter supported a sea urchin biomass of as much as 600 kg with a seawater make-up flow of less than 10%, while holding ammonia and nitrites below harmful levels (un-ionized ammonia $\leq$ 0.016mg/l and nitrite $\leq$ 0.5 mg/l). The use of RAS technology gave us better control over temperature than we would have had in a single pass flow through system, which is crucial for optimizing growth and survival rates. The RAS had a waste discharge flow of <8 l/m on average. This is important given that land-based aquaculture requires NPDES discharge permitting, which can be costly depending upon discharge rates.

Feeds and feeding

Previous studies have shown that when fed *ad libitum* most sea urchin species consume more feed than needed for somatic growth, putting the excess into gonad production or excretion. Sea urchins do not respond to satiation and feeding rates are influenced by factors such as feed quality, season, and size of the animal. Since feed costs and feed conversion can significantly affect production costs of land-based aquaculture, determining the optimal feed regime for green sea urchins is crucial for economic success.

The results of our feeding trials (Figures 4 & 5) show that green sea urchins can be grown for a lengthy period on terrestrial grain based feeds (Cargill) used for other species such as catfish. The lack of marine proteins, lipids and pigments in this diet did not compromise survival but did appear to compromise growth compared to sea urchins grown with a high quality specialty diet (Nofima). The price differential between the two diets (≈ $0.55US/kg for Cargill vs. $7.45/kg for the Nofima) indicates room for improvement in terms of balancing feed costs with quality. Low cost sea urchin feeds could be developed by supplementing existing cereal grain diets used to grow catfish or tilapia with some minimal level of marine based ingredients. What that level is remains a subject for further study and the issue of gonad quality will in all likelihood need to be addressed with a different feed regime to prepare the urchins for market. The use of seaweed as a 'finishing diet' for urchins reared on formulated feeds is currently under investigation at the CCAR.

Our investigation into the effects of different feeding frequencies on growth and feed conversion (Figures 6 & 7) revealed some interesting findings, but more in-depth study is needed. Urchins at 35-40 mm appeared to be past the period of fast somatic growth, and therefore should be fed with the goal of improving gonad quality rather than maximizing growth (Figure 6). A reduced feeding frequency (perhaps 1x/week) might be appropriate for these market size urchins, whereas more frequent feeding (1x/3 days) seems to be required for juveniles (Figure 6). There was, however, a trade-off between increased juvenile growth at higher rations and feed conversion. Juveniles fed more frequently grew faster but at the expense of feed conversion efficiency (Figure 7). The proper balance between the two will undoubtedly depend on the diet, but in any case it appears that larger urchins may only require 1 feed per week, which will help to reduce labor and feed costs.

Stocking densities

Stocking densities are a key determinant affecting profitability of sea urchin tank culture. Using tanks with slanting walls we grew urchins at a range of densities from 0.5 to 16.3 kg/m^2, with no adverse effects on survival or growth observed at the highest densities (Figure 8), which were well above the 6 kg/m^2 recommended for this species in previous studies by Siikavuopio et al. (2007). At the highest density of 16.3 kg/m^2 a set of paired troughs held 87 kg of urchins within 4.5 m^2 of floor area, for a footprint density of 19.3 kg/m^2 (Figure 8). By comparison the stacked system reported by Devin *et al* (2002) held 200 kg within a 7 m^2 footprint, or 28.6 kg/m^2. Vertically stacked tanks reduce the real estate footprint and may be more economically efficient. However, stacked tanks require platforms or ladders for access and might increase pumping costs due to the additional head. Another way to maximize holding efficiency is to use cage systems within tanks, as with the UrchinPlatter™ developed by the Irish company Gourmet Marine. This system reportedly holds from 50 to 90 kg/m^2 of sea urchins, and it is currently being trialed by the CCAR for gonad enhancement of *S. droebachiensis*. Another option is tank/cage polyculture systems that hold sea urchins, sea cucumbers and fish. This approach is reportedly being trialed by the Japanese at this time.

Objective 3) Test and demonstrate sea ranching for growing green sea urchins to harvest. Compare two ocean lease sites in terms of recovery rates and growth of out-planted sea urchins.

Sea ranching is a low cost extensive culture method that may be well suited for use by existing fishery participants. It can be done with or without an aquaculture lease and the major expenses are seed and out-planting costs, any lease rental fees, and harvesting costs. A major concern with sea ranching at an aquaculture lease is that very few if any of the out-planted urchins will survive the transfer or remain within lease boundaries. Growth rates of out-planted hatchery seed are also of concern. Primary project objectives were to demonstrate sea ranching at aquaculture leases and gain information on site criteria, survival rates (or more accurately, recovery rates), and growth while looking for differences between sites. Two lease sites were acquired for this project in Penobscot Bay, Maine by industry partner Friendship International. The sites were known as Job and Sloop after nearby islands.

Methods

Prior to out-planting an initial transect dive was done to estimate the extent of existing sea urchins, predators, and bottom cover. Predators were not found in abundance at either site, with only one large Jonah crab (*Cancer borealis*) observed. Though a large number of naturally occurring urchins were not found at the Job site, reports from local urchin divers and the suitable nature of the habitat indicated that both sites could support sea ranching. The habitat consisted mostly of mussel shell cobble at the Sloop Island site and rock cobble at the Job Island site, ideal for out-planting seed. Crustose coralline algae covered most of the cobble, providing an ample food source. The leases were marked with buoys to indicate that harvesting urchins by dragging nets across the bottom was prohibited.

In February of 2010 21,000 juvenile green sea urchins reared in the CCAR hatchery were tagged with tetracycline (Figure 1b) and 10,500 were out-planted at each site. The urchins were about 9 months post-settlement and ranged from 5 to 20 mm TD (average 10.6 mm at Job; 11.3 mm at Sloop). They were distributed along transect lines laid out to 15m in all four compass directions, encompassing a total area of 400m^2. Between 1,000 and 1,500 juveniles were released at 4m and 10m markers along the transects to ensure an even distribution. A post-release dive survey was conducted in April of 2010 to estimate survival of out-planted juveniles. At each lease site a baseline was laid out in a North-South orientation and five transect lines were laid out on a perpendicular (East-West) bearing extending to 10m. Sample quadrats consisting of a 1m^2 PVC frame were placed at the 10m marker in each direction, at the center of the transect, and just over the baseline (0 m on transect), for a total of 15 quadrats per site (Figure 8). All urchins within the quadrat were enumerated and those between 4-30 mm TD were collected in numbered mesh tubes to be taken to the laboratory for measurement and identification (presence/absence of fluorescent dye tags). The out-planted areas were subsequently dive surveyed at regular intervals (4-5 months) on six more occasions over the course of two years. Urchins smaller or much larger than the original release size were not collected in early surveys but during later surveys larger urchins were collected with the assumption that growth had occurred. This sampling regime allowed some inferences regarding survival and growth to be made, to be compared with the 10,000 urchins from the same hatchery cohort grown in the land based tank system.

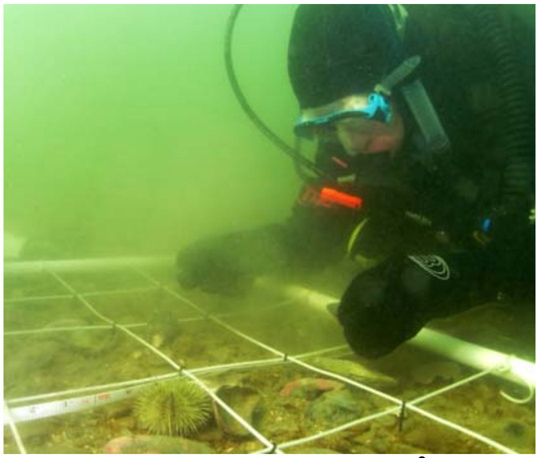


Figure 8. Sampling quadrat ($1m^2$)

Results of Sea Ranching trials

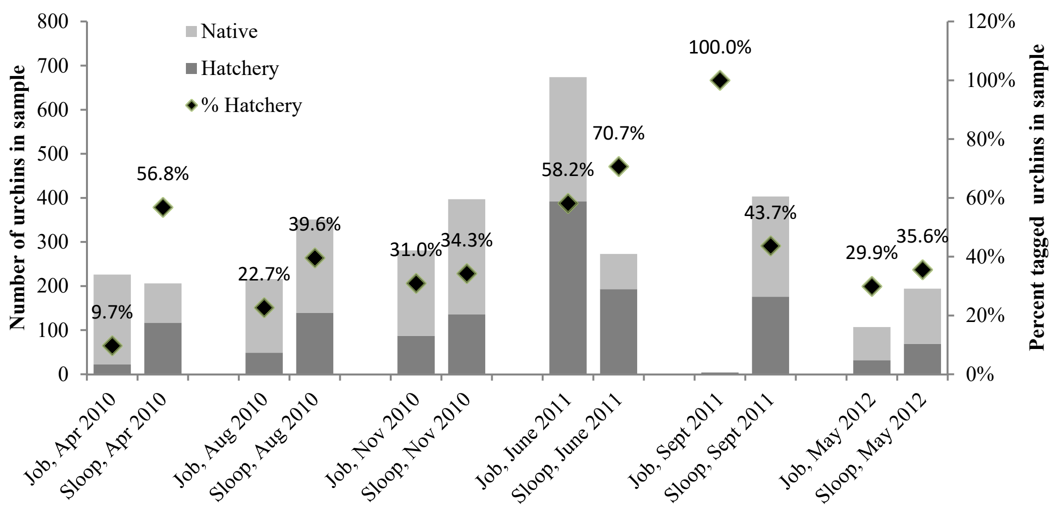


Figure 9. Total numbers of native and tagged (hatchery origin) green sea urchins collected during six dive surveys from the Job and Sloop lease sites. Percentages show percent of tagged

urchins in sample. **In general, the Sloop site had more total urchins and a higher percentage of tagged urchins in the population. A notable exception was the June 2011 survey, when the Job site showed an unexpected spike in total urchin numbers. A sharp reduction in numbers at Sloop was evident at the last survey in May 2012 and was accompanied by evidence that the site had been recently fished by a dragger vessel.**

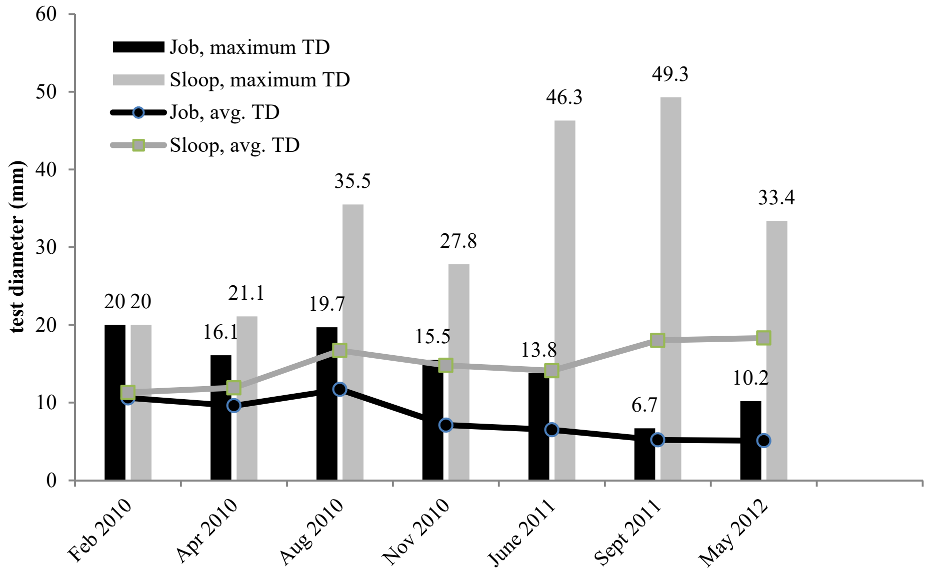


Figure 10. Average and maximum size (mm test diameter) of tagged hatchery origin green sea urchins at the initial out-planting in February, 2010 and at each of the subsequent dive samples from the Job and Sloop lease sites. **The Sloop site showed a pattern of increasing growth over the course of the project, and much larger tagged urchins were regularly recovered there than at Job. At the Job site the average size of tagged urchins actually declined over the course of the project.**

Discussion of Sea Ranching

The results indicate that out-planting success can vary between sites (Figures 9 & 10). The Sloop site appeared to be more favorable for out-planting than the Job site; in general more total urchins were found at Sloop and there was a higher percentage of tagged hatchery source urchins in the sampled population (Figure 6), and the released urchins showed greater growth at Sloop over the course of the study (Figure 7). Green sea urchin growth rates can be highly variable in the natural environment, primarily in response to feed availability and type. Growth can be very slow and rates of ≤ 0.25 mm per year have been documented for urchins found in tide pools. However, the persistent numbers of small tagged urchins found at both sites and especially at the Job site over the course of the two years might also be attributed to emigration of larger urchins from the site. Over time, sea urchins can move considerable distances in response to feed availability or to avoid predation. The degree and extent of movement in green sea urchins is a function of size. At about 15 mm and smaller they are cryptic and movement is minimal, but above this size movement patterns become more widespread and they can move greater distances to avoid predators or in search of food. These movement patterns could explain why the average size of tagged urchins at the Job site actually declined from the release size. Larger urchins might have left the area in response to predation, feed availability or other factors, leaving the smaller urchins behind. At the Sloop site a greater percentage of the released urchins grew and remained within the release area. 6.25% of the tagged urchins found at Sloop were >30 mm at the final survey and the largest tagged urchin recovered there was 49.3 mm (Figure 7). The site characteristics favorable for out-planting need further study but at a minimum it appears that a hard bottom comprised of shell hash and cobble, presence of feed, and a recent history of sea urchins populating the site are all favorable criteria.

It would be premature to recommend or discourage sea ranching based on the results from this study. Logistical considerations and other constraints did not allow the survey area to be extended beyond the release area, and factors for optimal site selection need to be better defined. It is possible, if not likely, that many of the released urchins moved beyond the release/survey area but still remained within lease site boundaries. Larger urchins would have been more likely to move and therefore the surveys may have underestimated both recovery and growth of the released urchins. Despite these unknowns some encouraging conclusions can be drawn. Released urchins were found at both sites after two years (Figure 6) and at the Sloop site some of those urchins showed enough growth after 27 months to indicate that legal harvest sized urchins (>52 mm) could be obtained from released seed after 3-4 years (Figure 7). On the other hand, the lack of evidence that seeded urchins grew at the Job site and the evidence indicating that the Sloop site had been subject to fishing activity (dragging) present significant barriers to sea ranching. Enforcement of legal protections to prevent poaching at lease sites is a difficult topic complicated by sociological and political considerations. The concept of individuals or companies leasing public fishing grounds for exclusive use is controversial in the Gulf of Maine region, at least for the present.

Objective 4) Compare growth, survival/retrieval, and economic costs/returns of sea ranching vs. tank farming.

Comparing sea ranching with tank farming and collecting data for a preliminary economic analysis were important project objectives. It was unknown prior to the project if green sea urchins out-planted onto an aquaculture bottom lease would survive, grow and remain in sufficient numbers to justify the expenses associated with hatchery/nursery production and lease fees. Similarly, it was unknown if survival and growth in a land-based tank system would be sufficient to justify the high costs associated with land-based aquaculture. In his project we were able to use the same hatchery cohort of green sea urchins to address these questions while comparing sea ranching with tank farming. To our knowledge, this is the first time such a comparison has been attempted.

Methods

All 31,500 green sea urchins used for the out-planting and tank culture trials came from the same CCAR 2009 hatchery cohort reared to the minimum out-planting size of 10 mm in the same nursery system. In February 2010 they were haphazardly mixed together and randomly assigned to be either out-planted at the two lease sites or reared in the tank system, using the methods described above for both. Returns in terms of tagged sea urchin numbers and biomass were estimated for the sea ranching from the dive surveys and for tank culture from the size and census samples. Main costs associated with each activity were tracked or estimated throughout the project period. Seed costs were based on 2009 prices for similar sized oyster seed plus 20%.

The leaseholders' cost and return at each lease site from out-planted urchins was based on the last dive surveys in May 2012 (after 27 months growth) and extrapolated to the entire 2-acre lease using three assumptions. These were: 1) the total urchin population (wild + tagged) was evenly distributed throughout each 400m^2 study area and could be estimated from the numbers collected in 15 m^2; 2) tagged urchins were evenly distributed throughout each 400m^2 study area and could be estimated from the percentage of tagged urchins collected in 15 m^2; 3) the average size of tagged urchins recovered at the last survey was representative of the average size of all remaining tagged urchins within each study area. Recovery rates and a unit cost per kilogram and per urchin were then estimated for each 2-acre lease site given out-planting and recovery numbers proportional to those for the study areas (Table 2).

For the tank culture analysis we used the growth, survival, density and feeding data from the project to set production targets that might be achieved with intensification within the same real estate footprint (2,400 ft^2) as that used for the project. Similarly, we used our known and estimated production costs to set target production costs, assuming that efficiencies could be gained with an insulated building, more efficient pumps and chilling, lower cost feed, better feed conversion, faster growth, etc. The following assumptions and cost targets were used for the model farm: 1) in the project we only used 2/3 of the available greenhouse space for 12 tanks. Intensification could be realized by using all of the floor area and stacking the tanks to total 36 tanks; 2) we assumed a maximum final density for all tanks of 16.3 kg/m^2, which was the highest density we observed in the project; 3) seed costs were reduced to $25 per thousand; 4) improved growth rates and feed conversion could be realized; 5) electrical costs could be reduced by ≈1/3 even at a greater production scale by using more efficient 3 phase pumps and chillers; 4) fuel oil costs could be reduced by half in an insulated building; 6) average daily labor time would increase only marginally at increased intensification, primarily because tanks with better self-cleaning ability would be used; and 7) the target feed cost was set at $1.10/kg, double the price we paid for the Cargill catfish feed but ≈90% less than the price paid for the premium Nofima diet. The production targets and costs were used to estimate market values and a unit cost per kilogram and per urchin for the model farm. Sea urchin value per kg was based on the average 2012-2013 ex-vessel price of $3/lb reported by the Maine DMR, and the value per sea urchin (piece) on the assumption that live 70 gram urchins could be sold to specialty markets at twice the value of bulk product. Uni prices were based on data from the National Marine Fisheries Service and from 2013 prices for 100g trays of uni as listed on the internet by a Maine processor/dealer. These and other assumptions are summarized in Tables 3 and 4.

Results, Costs and Returns

Sea ranching costs and returns

The main costs of sea ranching for the period of February 2009 to May 2012 were for seed, out-planting (boat and diver costs), equipment (marker buoys, ropes, and anchors), and lease fees (real estate). Out-planting the full 2-acre lease site (8093.7 m^2) on a scale proportional to the study area would require that 212,460 hatchery seed be out-planted. At $35 per 1000 the seed would cost $7,436. Fees for a three year experimental lease would total $700, out-planting would require two boat and diver days for an estimated $2,000, and equipment to mark the lease site and out-plant the urchins would cost another estimated $2,500. Thus, the total costs to out-plant one 2-acre lease site with 212,460 urchins for three years would be $12,636. Based on the last study area surveys done in May 2012 the estimated returns and unit costs for the leaseholder from each site can be summarized in Table 2.

SITE	# of urchins collected in 15 m^2 May 2012	% tagged	average wt tagged seed May 2012	total urchin population projected to lease area	# tagged urchins projected to lease area	biomass loss or gain projected for lease area	Unit cost per kg	Unit cost per urchin
JOB	107	29.9%	0.09 g	57,735	17,263	-104.7 kg	NA	$0.72
SLOOP	194	35.6%	4.03 g	104,679	37,266	22.7 kg	$547.74	$0.33

Table 2 Projected returns and costs to lease holder from two 2-acre lease sites with out-planting numbers, returns and growth proportional to those seen for the study areas in May 2012 at each site. Total operating cost per site is estimated as $12,636 for three years

Tank farming costs and returns

The main costs associated with tank farming for the period from December 2010 to October 2012 were for seed, equipment, real estate, electricity, fuel oil for heating, labor, and feed. To simplify the tank farming analysis we excluded real estate and equipment costs. These costs will vary a great deal depending upon where, how and by whom the system is constructed, whereas the lease fees and equipment costs for sea ranching are far more predictable and consistent between sites. Production scale and actual or projected costs are summarized for tank farming in Table 3. Estimated returns for the model farm described in Table 3 are summarized in Table 4.

	CCAR Tank System (2 year period)	Model farm (3 year period)
# of tanks	12	36
# of seed urchins	9,193	53,048
survival rate	95.4%	90.0%
years to market	4	3
# remaining	8,770	47,743
final avg. density, kg/m^2	9.2	16.3
feed conversion	1.42	1.1
seed cost per 1000	$35	$25
total seed cost	$322	$1,326
feed cost per kg	$14.77	$1.10

total cost of feed	$3,552	$3,758
Electricity	$11,183	$8,569
Fuel oil for heating	$9,997	$4,999
average minutes daily labor	39	45
Labor cost @ $12/hr	$5,047	$9,864
TOTAL COST	**$30,100**	**$28,516**

Table 3 Production and cost parameters and targets for the CCAR tank system and the model farm. CCAR tank system data is based on measured observations. Model farm data is based on assumptions described in the methods.

MODEL FARM	# of urchins	Total weight kg	Cost or value per kg	Total cost or value kg weight basis	Cost or value per sea urchin (piece)	Total cost or value piece basis	production cost per unit kg	production cost per piece
Seed stocked @ 0.5 g	53,048	26.5	$50.05	$1,326	$0.025	$1,326	No data	No data
Harvest @ 70 g	44,743	3132.0	$6.50	$20,358	$0.90	$40,269	$9.10	$0.64
roe yield	20%	626.4	$90.00	$56,376	$1.26	$56,376	$45.52	$0.64

Table 4 Projected returns with 90% survival from the model farm after three years of tank culture. 2013 ex-vessel prices for whole sea urchins averaged $3/lb while uni routinely sold for ≈$200/lb.

Discussion of sea ranching and tank farming costs/returns

The case can be made that the returns from sea ranching we observed during the project represent a worst case scenario. The dive surveys likely underestimated the average size of out-planted urchins over time, mainly because larger urchins would have been more likely to move away from the study area, as discussed earlier. For similar reasons the surveys may have underestimated the total numbers of tagged urchins that remained within the lease but were no longer within the study area. There was also evidence that the more productive Sloop site had been dragged just before the last dive survey. In defense of the dragger it must be noted that 3 out of the 4 buoys marking the lease site corners were missing, probably due to winter storms, so therefore the dragger may not have realized that he/she was fishing on an aquaculture lease. It must also be acknowledged that in reality sea urchins are unlikely to be evenly distributed throughout a lease area or even a study area, and more likely will be found in patches of varying densities. A more sophisticated statistical analysis of our survey data is required to elucidate distribution patterns, currently in progress as a component of the MS thesis.

Given these limitations and qualifiers, it is nonetheless encouraging that the unit cost per sea urchin at the Sloop site was 33¢, even at the low rate of return we projected for a scaled up release. At a decent harvest size of around 70 g there are 6.5 urchins per pound, and at the current ex-vessel price of $3/lb the profit on 6.5 urchins with a production cost of $2.145 is

$0.855. If the site had not been dragged it is likely that the projected return would have been higher, with a lower production cost per urchin. This shows that sea ranching can potentially be profitable, but that a great deal depends upon the site because at the Job site the data projected a net biomass loss, with a unit cost of 77¢ per urchin. It is also important to note that the greatest single expense by far was the seed cost, which amounted to 58.8% of the total. The cost of $35 per 1000 (3.5¢ per seed) we used for the sea ranching analysis was based on oyster seed pricing and may or may not reflect the true cost of urchin seed production. According to Yuichi Sakai of the Mariculture Fisheries Research Institute, in Hokkaido, Japan it costs 5-10 JPY (6.2 to 12.5 US cents) to produce one seed of 5 mm test diameter, the majority of which is wage (20%) and energy (40-60%) costs. The true costs of producing green sea urchin seed in Maine need to be determined, with the objective of reducing the cost per thousand to $25 or less. This may require out-planting larger numbers at a smaller size, which will increase the scale and therefore efficiency of hatchery production while reducing the time spent in costly land-based systems. However, this may come at the cost of lower survival rates for out-planted urchins.

Our projections show that the economic prospects for tank farming are more uncertain than those for sea ranching.
Total costs to operate the model tank farm for 3 years were estimated at $28,516 (Table 3), but the ex-vessel value of the harvested biomass was only $20,358 (Table 4). The production cost per kg was much lower for tank farming than for sea ranching (Tables 2 and 4), due to faster growth in tank culture and poor growth data for the study areas. However, as noted earlier the poor growth data we saw for the study areas may be attributable to emigration of larger tagged urchins, skewing the surveys towards smaller urchins and resulting in a higher production cost per kg. In addition, the tank culture production cost per sea urchin was almost twice that seen for sea ranching at the Sloop site (Tables 2 and 4). To obtain more value urchin growers could process and market the uni directly rather than selling whole urchins at ex-vessel prices to a third party processor/dealer. Good quality uni sells for $90 or more per kg, netting $56,376 for the model farm if it does its' own processing and marketing (Table 4). This doesn't take into account processing costs, but on the other hand uni can sell for as much as $150 per kg, and roe yields from sea urchins conditioned for market can be as high as 25-30%. It's also possible that with good marketing a grower could sell individual 70 g live sea urchins to restaurants, academia, internet customers or other specialty markets for 90¢ or more per urchin, which would net $40,269 for the model farm (Table 4).

In any event, given the high value of uni it is clear that further investigation into sea urchin tank farming is justified. Two important areas that need to be addressed are development of low cost formulated diets and selective breeding for fast growing strains. A small percentage (<2%) of the urchins were ≥45 mm TD after just seven months in tank culture (27 months post-settlement), and we saw large differences in growth rates within the same hatchery cohort reared under the same conditions. Growth rates can also vary a great deal with green sea urchins in the wild, and although environmental factors no doubt play a large role researchers have hypothesized that there may be a genetic basis as well. Chinese researchers have shown that there is enough genetic variation in sea urchin growth rates to justify selective breeding efforts. If a fast growing strain of green sea urchins were developed specifically for tank culture the culture time could potentially be reduced to two years or less. This would greatly improve the prospects for land based tank farming of green sea urchins in the Gulf of Maine region.