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Aquaculture Collaborative Research and Development Program (ACRDP) Fact Sheet

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Basket Cockle (*Clinocardium nuttallii*): Candidate Commercial Aquaculture Species in British Columbia

Summary

The Aquaculture Collaborative Research and Development Program (ACRDP) has funded three research projects, with Evening Cove Oysters Ltd., focusing on various hatchery, nursery and grow-out aspects of the development of Basket Cockle (*Clinocardium nuttallii*) aquaculture in British Columbia (BC). There is a great desire to move this species toward commercialization for both the diversification of the aquaculture industry in Canada, as well as for the creation of much needed jobs in coastal and First Nations communities. The Basket Cockle is native to BC and although commonly found on the shore, it is generally present in low abundances. They are a culturally-significant species for coastal Aboriginal peoples in BC and interest has been expressed by some First Nation communities in commercializing the species in their territories. Advances in hatchery and out-planting research have moved this species one step closer to commercialization. The outcomes and recommendations of these projects have been documented in various publications and should be referred to for a complete account of methodologies and results for the various projects. The following provides a general summary of the research to date.

The Aquaculture Collaborative Research and Development Program (ACRDP) is a Fisheries and Oceans Canada (DFO) initiative to increase the level of collaborative research and development activity between the aquaculture industry and DFO.

Introduction

The Basket Cockle (*Clinocardium nuttallii*) is native to the low intertidal and shallow subtidal zones of coastal British Columbia (BC) (Liu *et al*., 2008a). It is a traditional food of many First Nations in BC, but is not found in great abundance nor harvested commercially in the province. There is, however, a limited commercial capture fishery in the United States for this species and in 2010, 179 tonnes were harvested (FAO, 2010).

Interest in diversifying the BC shellfish aquaculture industry to include this potentially high-value clam species, in addition to interest expressed by coastal First Nations, prompted the collaborative research and development projects on cockles funded between 2005 and 2011. Three projects addressing various hatchery, nursery and grow-out issues, and one ongoing project (not discussed here) on the genetic diversity and health status of various populations of Basket Cockles, have been funded through the Aquaculture Collaborative Research and Development Program (ACRDP).

Five phases of research have been undertaken, each one building on previous production practices to both maximize growth and increase survival of larval and juvenile Basket Cockles as well as to increase the reproductive potential of adult Basket Cockles. These phases were: 1) Broodstock and embryos, 2) Hatchery rearing: larvae to pediveligers, 3) Hatchery rearing: post settlement, 4) Nursery rearing, and 5) Outplanting.

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These five phases of research have contributed to the understanding of Basket Cockle production requirements and the determination of its feasibility as an aquaculture species.

Research Summary

Phases 1 – 4 were undertaken at the Centre for Shellfish Research (CSR) at Vancouver Island University (VIU) in Nanaimo, BC. Phase 5 was conducted at CSR's Deep Bay Marine Field Station in Baynes Sound, BC. Summaries of the methods and results for each of these phases are provided below.

Figure 1.
Basket Cockles (*Clinocardium nuttallii*).

1. Broodstock and Embryos

Detailed methods pertaining to the broodstock and embryonic research of Basket Cockles are described in Liu *et al*. (2008 a,b). Three experiments were undertaken.

Sperm motility and temperature

The first experiment examined the effect of storage temperature on sperm motility. For this, pooled sperm samples from animals held at 16°C were diluted and held at two temperatures (4 and 19°C) for two hours with sperm motility measured at six intervals therein.

Results showed that sperm samples held at 19°C progressively decreased in mobility, with only 24% being weakly motile after two hours. In contrast, a high level of sperm motility was maintained for two hours in samples held at 4°C, indicating that sperm could be held at standard refrigeration temperatures until sufficient eggs were released for fertilization.

Fertilization rate and sperm-to-egg ratio

The second experiment examined the effect of various sperm-to-egg ratios on fertilization rate. Pooled samples of eggs were fertilized with pooled samples of sperm at various ratios ranging from 10:1 to 10,000:1 (sperm:egg). Controls of eggs without addition of sperm were used to determine if self-fertilization was occurring.

Fertilization success was determined after two hours incubation at 19°C (the temperature which yielded the fastest fertilization and development rate). Greatest fertilization success was observed at a sperm-to-egg ratio of 10,000:1. Self-fertilization was observed to occur, which is not uncommon with hermaphroditic species such as the Basket Cockle.

Broodstock conditioning, temperature and diet effects

The third experiment examined the effect of various microalgal diets on broodstock conditioning. Animals were fed four microalgal species [*Isochrysis galbana* (Tahitian strain) (TISO), *Thalassiosira pseudonana* (3H), *Tetraselmis suecica* (TS) and *Chaetoceros gracilis* (CG)] in four combinations [TISO/3H, TISO/CG, TS/3H and TS/CG] with a starved control. Diets were provided over 20-hour periods for thirteen weeks at 16°C. An additional experiment was run at the same time in which cockles were held at 2.5°C, and fed with the same concentrations as the higher temperature. Condition indices of the cockles were determined at the end of both experiments.

These conditioning trials found that it was difficult to bring immediately post-spawned (collected in June) animals into condition when held at high temperatures (16°C) (*i.e.*, egg/oocyte development was not noted at this temperature). However, those that did increase soft-tissue weight had been fed diets containing TISO. In contrast, cockles exposed to low temperatures (2.5°C) showed signs of oocyte (or egg) development in all specimens examined ten and thirteen weeks after the onset of the experiment, suggesting that a simulated winter period may be needed to initiate the ability for reproduction following a spawning event.

2. Hatchery Rearing: Larvae to Pediveligers

Two primary publications resulted from this work (Liu *et al*., 2009, 2010), which can be referred to for specific information on methodologies and detailed results.

Effect of microalgal diet on larval growth and survival

The first set of experiments was aimed at determining growth and survival of larval and early post-larval cockles fed different microalgal diets. Single-species and multiple-species diets of TISO, *Chaetoceros muelleri* (CM), *Pavlova lutheri* and *Thalassiosira pseudonana* (3H) were provided over fourteen days. Larval shell length, survival and metamorphosis were measured over time.

Single and mixed-species diets containing TISO resulted in the greatest larval shell growth and metamorphic rates. Post-larvae stages had the greatest shell growth with single- and mixed-species diets containing CM (algae).

Effect of stocking density on larval growth and survival

The goal of the second set of experiments was to determine the effect of stocking density on larval growth and survival. The combined effect of larval stocking density and algal density were examined using nine treatment combinations of three larval stocking densities (2, 4, and 8 larvae/mL) and three algal densities (10, 25, and 50x10³ cells/mL). Single factor effects of TISO algal density, with a fixed stocking density of 2 larvae/mL, were also examined. Growth and survival of larvae were monitored for nine days.

When feeding bi-algal diets of CM and TISO, optimal conditions for growth and survival were: initial stocking density of 2 larvae/mL with an algal density of 25 or 50x10³ cells/mL or initial stocking density of 4 larvae/mL with an algal density of 50x10³ cells/mL.

Effect of temperature on larval growth and survival

A third experiment examined the effect of six temperatures (range of 5.9 to 26.3°C) on larval shell length and survival from the veliger stage to the pediveliger (mobile) stage. All larvae were fed a diet of TISO, increasing in concentration as the experiment proceeded. Shell length was measured every second day and survival rate determined. The experimental time varied from six to fourteen days, depending on temperature treatment, as the endpoint was reaching the pediveliger stage.

Within experimental parameters, larval growth rate was greater with increasing temperature, but the ability of the larvae to survive to the settlement stage was not significantly affected by temperature within the range examined.

3. Hatchery Rearing: Post Settlement

Stocking density, food ration and temperature were manipulated to identify optimal hatchery and rearing conditions for post-settled Basket Cockles. Methodologies and results are described in detail in Liu *et al*. (2011).

Effect of stocking density on early seed growth and survival

Stocking density experiments were performed on both pediveligers and 1-mm seed. Animals were held in downwelling systems and metamorphic rate and post-settlement growth were measured. Pediveligers were stocked at 20, 40, 80, 160 and 320 individuals/cm² and were sampled at two and three weeks, resulting in ten treatment combinations. The 1-mm seed were stocked at 10, 20, 40, 60 and 80 individuals/cm^2 and sampled at two and four weeks, also resulting in ten treatment combinations. At the end of each experimental period, animals were measured for shell length and height. Survival rate and metamorphic rate were subsequently calculated.

The effect of stocking density on seed growth was found to be size-dependent, with smaller individuals allowing for more seed to be stocked on the unit bottom without negative effects. Optimal seed growth (regardless of size) was observed when less than 100% of the rearing unit's bottom area was covered with seed.

Effect of diet on early seed growth and survival

The effect of a bi-algal diet (CM + TISO) fed at different rations (range: 0 – 51.2x10^6 equivalent TISO cells/individual/day) on seed growth was examined in five experiments using different seed size classes (0.74±0.01, 1.59±0.02, 2.10±0.05, 2.60±0.05 and 3.00±0.04 mm shell lengths). Conditions ranged from underfeeding to overfeeding, to determine optimal rations with changing seed size. Optimal rations were determined as the minimum amount of food offered beyond which no significant increase in shell length was observed. At each sample period, shell length, shell height, wet weight, dry weight and organic content were measured. The duration of each experiment varied, depending on the initial stocking size, but generally ranged from one to two weeks.

The optimal rations of a bi-algal diet (CM + TISO) were determined for several size classes. In general, the rations increased with increasing shell length, but the opposite trend was observed for weight-specific rations. Increasing food rations, based on cockle weight, did not have a positive effect on growth and resulted in slower growing seed, which produced more fecal matter.

Effect of temperature on early seed growth and survival

The effect of temperature on shell length was tested at six temperatures (range of 5.9 to 26.4°C). All treatments were fed the same algae (CM + TISO) with daily rations increasing with time. Weekly measurements of shell length and weight were performed for the duration of the three-week experiment.

When considering such factors as heating water costs, the optimal growing temperature was determined to be approximately 18°C.

4. Nursery Rearing

Four laboratory experiments were undertaken in order to refine nursery and outplanting practices of Basket Cockles, with methodologies and results described in detail in Epelbaum *et al*. (2011).

Effect of stocking density and substratum on seed survival, growth and condition, and burrowing rate

The presence/absence of substrate (fine sand and no sand) and seed stocking densities (50 and 150% bottom cover in a monolayer) were evaluated to determine the effect on seed survival, growth and condition, as well as burrowing rate at outplanting. Weekly mortality rates, wet weights, shell lengths and shell widths were measured over a four-week period.

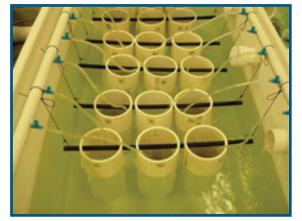

Figure 2.
Nursery rearing set-up for Basket Cockles at the Centre for Shellfish Research (CSR) at Vancouver Island University (VIU) in Nanaimo, BC.

There was no significant effect of substrate (present/absent) on seed survival or growth, but significantly higher rates of shell abnormality and lower burrowing rates at outplanting were observed for seed raised without substratum when compared to animals grown in substrate. The lower stocking density of 50% bottom cover resulted in an increase in shell length from 3 to 7 mm over four weeks. The higher stocking density (150% cover) negatively impacted growth, but was not found to significantly impact seed survival.

5. Outplanting

The effect of two culture systems, intertidal (in sediment) and suspended, and cockle stocking density were tested in the field in grow-out Years 1 and 2. Methodologies and results are described in detail in Dunham *et al*. (in press a,b).

Effect of culture mode, depth and initial stocking density on survival, growth and condition during the first and second years of grow-out

Year 1: The effect of culture mode and initial stocking density on cockle seed survival, growth and condition (including marketability-affecting qualities) were assessed in Year 1. The intertidal grow-out system consisted of a series of plastic containers with mesh bottoms and removable lids, partially buried in the sediment in the low intertidal zone. The suspended system consisted of traditional pearl nets arranged in vertical stacks of three and suspended from longlines. Seed were stocked at initial densities of 1500, 3000, 10500 and 21000 individuals/m^2, representing 5, 10, 35 and 70% bottom cover, in both culture systems. Physical parameters such as temperature and chlorophyll were measured to determine the productivity potential of both culture systems. A random sample of 20 cockles was measured from each sampling net and sampling tote every month with shell lengths and wet weights being measured. After five months, the experiment was terminated and final measurements on all animals were made.

Cockles grown in the suspended culture system had high survival rates at all stocking densities; in contrast those grown intertidally had poor growth and survival. Highest growth rates and condition parameters in the suspended system were achieved at densities of 1500 and 3000 individuals/m^2; meat yield was greater than 40% of the total wet weight at all stocking densities. Minimal shell deformities (0 – 2.9%) were observed in suspended culture.

Year 2: As with Year 1, the effect of culture mode and initial stocking density on cockle seed survival, growth, and condition were assessed, although in this year the effects on various size classes of seed were also examined. The same stocking densities as Year 1 were used to stock both intertidal and suspended culture systems with animals of three size classes (16, 22, and 32 mm shell length; these size classes represented the range that a shellfish farmer could expect to obtain after the first year of growing *C. nuttallii* in suspended culture at stocking densities of ≤3000 individuals/m^2). In addition, the effect of culture depth in the suspended system was examined. The same variables were measured in Year 2 as in Year 1 over a time period of approximately four months.

Cockles in all size classes in the suspended culture system had significantly higher soft-tissue biomass, growth rates and condition indices than those in the intertidal system. The suspended culture system did have its drawbacks, however, with sub-optimal levels of shell deformities (8 – 40%), likely due to decreasing space and the increased amounts of biofouling noted. In both culture systems, stocking density had a significant effect on growth and culture indices. Recommended initial stocking density is 10% cover for all size classes tested. Culture depth did not have a significant effect on growth or condition indices. Depending on the grow-out scenario, stocking density and harvestable size chosen, the cumulative harvestable percentage after Year 2 of grow-out constituted 15.5 to 63.1% of the seed planted.

Conclusions

Over the past six years, research efforts have shown that the Basket Cockle (*C. nuttallii*) is a promising candidate for aquaculture in BC. Experiments to determine optimal hatchery, nursery and outplanting practices have provided a strong foundation for the development of this industry. Breeding techniques have been developed and refined, allowing for the determination of optimal diets/temperatures for broodstock conditioning, temperatures for sperm storage and sperm-to-egg ratios. Optimal algal diets, rations and temperatures have been determined for critical stages of larval and early seed development.

Figure 3.
Long-line (suspended culture) system used for Basket Cockle outplanting experiments.

The effect of stocking density and presence/absence of substratum on survival and burrowing rate of larger juveniles in the nursery phase have been examined as they affect cockle size, stocking density and grow-out method on survival and growth of outplanted seed.

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For further information on this and other ACRDP projects, visit: **http://www.dfo-mpo.gc.ca/science/aquaculture/acrdp-pcrda/main_e.htm**

Information on the cockle research can also be found in Aquaculture Update, issues 103 and 104 at **http://www.pac.dfo-mpo.gc.ca/science/aquaculture/update-misesajour-eng.htm**

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