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RESEARCH ARTICLE

A comparative study on the nursery culture of hatchery-reared sub-adult cupped oyster, *Crassostrea iredalei* (Faustino, 1932), in an earthen pond and a mangrove canal

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Abstract. A comparative study on the nursery culture of the spat of the tropical oyster, *Crassostrea iredalei*, in an earthen pond and a mangrove canal was conducted over two months. The results revealed no differences in the absolute growth rate determined by shell width between the two culture sites ($P < 0.05$). Sub-adult oysters cultured in the mangrove canal showed a higher absolute growth rate in shell length and a higher daily growth rate than the oysters cultured in the earthen pond ($P < 0.05$). The mean survival rate of sub-adult oysters cultured in the earthen pond ($99.8 \pm 0.2\%$) was significantly higher than for those cultured in the mangrove canal ($66.7 \pm 31.4\%$). Decreased density from the loss of sub-adult oyster nursery culture in the mangrove canal led to higher growth performance than in the earthen pond. However, no difference was found for the fraction of oysters larger or smaller than 5 cm for the two culture sites ($P < 0.05$). A significant difference was noted in the Condition Index (CI) between the two culture sites ($P < 0.05$). The high primary productivity in mangroves is a major supporter of higher CI in sub-adult oysters cultured in mangrove canals versus in earthen ponds. Water exchange in the earthen pond to maintain calcium and magnesium concentrations resulted in no differences in the shell compressibility of sub-adult oysters compared with those cultured in the mangrove canal.

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Introduction

Thailand has abundant oyster resources from natural beds located in the shallow coastal waters of intertidal mangrove areas. Among the indigenous oysters of Thailand, *Crassostrea belcheri*, *C. iredalei*, and *Saccostrea cucullata* are economically important and widely cultured species (Chalermwat et al. 2003). The cupped oyster or black-scar oyster, *C. iredalei*, is one of the most commercially important species thanks to its high marketability, fast growth, and good flesh appearance. A large number of oyster seeds from natural sources are collected and supplied to grow-out farms, but the amount of oyster seed produced from these sources is limited and insufficient. Recently, decreases in water quality and increases in pollution have greatly influenced the depletion of oyster populations by hindering larval development. To resolve this problem, oyster seed production in hatcheries has been developed and is a subject of great interest in Thailand. Artificial seed production technology has succeeded in producing hatchery techniques for *C. belcheri* (Tan and Wong 1996, Tanyaros et al. 2008, Tanyaros and Kitt 2011, 2012, Tanyaros et al. 2015) and *C. iredalei* (Devakie and Ali 2000a, 2000b, 2002, Teh et al. 2012, 2016). However, the high cost associated with the production of microalgae still poses problems for oyster hatchery operations as well as for the post-setting stage.

The requirements for a microalgal diet for hatchery-reared oyster spat varies depending on their size. Those of smaller size need a small amount of microalgae, but large volumes are needed for those of larger sizes. Microalgae culture is labor intensive and large-scale production is often accompanied by various technical difficulties. Suitable sites with abundant natural food sources are necessary for the mass culture of oyster spat in order to reduce the cost of producing microalgae for hatchery operations. A number of previous studies have reported on aspects of the nursery culture of oyster spat in the sea (Holliday et al. 1991) and in mangrove canals (Tanyaros et al. 2008, Tanyaros and Tarangkoon 2014). Nursery culture of sub-adult oysters in earthen ponds is a potential solution since it is easy to maintain food sources by adding fertilizer to stimulate the growth of microalgae. The aim of this study was to compare the growth performance of hatchery-reared sub-adult *C. iredalei* in nursery culture in an earthen pond and in a mangrove canal. These research findings will benefit the nursery culture of sub-adult oysters produced in hatcheries and help in providing more effective oyster farming practices.

Materials and methods

Experimental oysters

The sub-adult *C. iredalei* used in this experiment were produced in the hatchery at the Marine Shellfish Breeding Research Unit, Faculty of Science and Fisheries Technology, Rajamangala University of Technology Srivijaya, Trang Campus, Trang, Thailand. The hatchery-reared oyster spats (mean shell length 0.5 cm) were cultured in a flow-through 1,000 L recirculating tank that was part of a land-based nursery system for two months until the oysters had achieved the sub-adult stage with a mean shell length of 3-5 cm. The oysters were graded for minimum size variation to prevent possible growth retardation, and thirty oysters were sampled randomly for shell width and length measurements. The mean (± SD) shell widths (dorsal-ventral) of 3.66 ± 0.37 cm and 3.57 ± 0.34 cm and shell lengths (anterior-posterior) of 4.70 ± 0.55 cm and 4.54 ± 0.48 cm were recorded for oysters in the nursery culture in the earthen pond and the mangrove canal, respectively. The animals were kept for two days in a flow-through 1,000 L recirculating system for acclimation before the start of the experiment. An electric pump (2 HP and 2 inches of water intake) pumped sea water from the earthen pond supply into the culture tank.

Experimental design

A horizontal suspension method was designed to investigate the effects on the growth performance of hatchery-reared *C. iredalei* sub-adults in plastic mesh nets (mesh diameter size 2.5 cm) during the two-month-long growth trial. Green plastic mesh was used in this study since previous research showed a low degree of fouling by filamentous algae (Tanyaros and Kitt 2012). Each plastic mesh net (dimension 40 cm × 90 cm × 5 cm) was divided into three sections by inserting pieces of hard wood (5 cm × 30 cm × 5 cm) at 30-cm intervals to prevent the oysters from accumulating in the centre of the plastic nets as their weight increased. Homogenous groups of sub-adult oysters were distributed randomly onto the plastic mesh nets at densities of 200 individuals per net. Four plastic mesh nets were tied horizontally to each rectangular PVC frame (45 cm × 100 cm × 100 cm) using aluminum wire. The frames were suspended from a long line in the earthen pond and the mangrove canal at 30 cm below the water surface. Four replicates were used in each treatment.

A blower was used to create water movement throughout the nursery culture area in the earthen pond. It was controlled by an automatic timer that switched the blower on and off every 30 min throughout the study period. Natural tidal flow provided water movement for the nursery culture unit in the mangrove canal so there was no need for aeration. The tray was cleaned at 5-day intervals by manually scrubbing the outside tray surfaces *in situ* with brushes, and washing them with sea water pumped in at low pressure by a submersible pump. Water quality parameters over the study period in the earthen pond and the mangrove canal were as follows: water salinity 26.1 ± 2.0 and 26.4 ± 2.1 PSU; dissolved oxygen 5.1 ± 0.5 and 5.21 ± 0.4 mg l^{-1}; total ammonia nitrogen 0.061 ± 0.021 and 0.069 ± 0.020 mg l^{-1}; water temperature 30.0 ± 1.7 and 29.1 ± 1.2°C; and pH 7.1 ± 0.1 and 6.8 ± 0.2, respectively.

Sample collection and analysis

Thirty animals from each replicate were sampled randomly biweekly for measurements of shell width, shell length, and wet weight. Growth was expressed as absolute growth rates of shell length (AGRL) and width (AGRW) was as follows: AGRL (cm/month) = (mean final shell length – mean initial shell length)/culture period; AGRW (cm/month) = (mean final shell width – mean initial shell width)/culture period. The daily growth rate (DGR) was then calculated for individuals with the method by Coutteau et al. (1990). The experiment was conducted for two months. At the end of the experiment, all sub-adult oysters from each experimental unit were graded using sieves with mesh sizes of 5 cm in diameter, and then counted. The survival rates and size fractions were calculated and expressed as percentages. Five oysters from each replicate were selected for Condition Index (CI) analysis. The CI was calculated for selected oysters for dry meat weight (DMW) and shell weight (SW) using the equation by Royer et al. (2007). The right shell of oysters selected from each replicate was used for CI analysis to determine shell compressibility using a Compression machine (Triaxial tester T400 Digital).

Statistical analysis

A Mann–Whitney U test (M–W) was used to test for differences in growth variables between the nursery culture in an earthen pond and mangrove canal using SPSS (version 17.0). Differences were considered significant at $P < 0.05$.

Results

The growth performances for hatchery-reared sub-adult *C. iredalei* grown in plastic mesh nets suspended horizontally for two months in the earthen pond and for those grown in the mangrove canal are presented in Table 1. No significant effects ($P < 0.05$)

Table 1
Growth performance for hatchery-reared oyster spat, *Crassostrea belcheri*, grown in plastic mesh nets for two months in different areas. AGRL = Absolute growth rates of shell length, AGRW = Absolute growth rates of shell width, and DGR = Daily growth rate

	Nursing areas		
Parameters	Earthen pond	Mangrove canal	P – value
AGRL (mm d^{-1})	0.17 ± 0.78	0.29 ± 0.28	P < 0.05
AGRW (mm d^{-1})	0.17 ± 0.04	0.21 ± 0.14	P > 0.05
DGR (% d^{-1})	12.0 ± 0.6	29.1 ± 4	P < 0.05
Size fraction >5 cm (%)	43.8 ± 9.0	47.4 ± 7.9	P > 0.05
Size fraction <5 cm (%)	56.2 ± 9.0	52.6 ± 7.9	P > 0.05
Survival rate (%)	99.8 ± 0.2	66.7 ± 31.4	P < 0.05
Condition Index (CI)	3.1 ± 0.2	5.4 ± 0.8	P < 0.05
Shell Compressibility (kg)	25.5 ± 10.5	29.9 ± 8.7	P > 0.05

on AGRW were found between the oysters cultured in the earthen pond and those cultured in the mangrove canal. However, nursery culture in the mangrove canal produced a significantly higher AGRL and DRG for sub-adult oysters than did that in the earthen pond over the study period ($P < 0.05$). No differences in the proportions of sub-adult oysters > 5 cm and < 5 c m were found between nursery culture in the earthen pond and in the mangrove canal. The mean size fraction for sub-adult oysters was 43.8 ± 9.0% (>5 cm) and 56.2 ± 9.0% (<5 cm) for nursery culture in the earthen pond and 47.4 ± 7.9% (>5 cm) and 52.6 ± 7.9 (<5 cm) for nursery culture in the mangrove canal, respectively. Significant differences were noted for the final survival rates and Condition Indexes (CI) in the oysters from the two culture sites. The mean survival rates for oyster nursery culture in the earthen pond and in the mangrove canal were 99.8 ± 0.2% and 66.7 ± 31.4%, respectively. However, the difference in shell compressibility of the sub-adult oysters from the two culture areas was not significant. The mean weight for shell compressibility was 25.5 ± 10.5 kg and 29.9 ± 8.7 kg for the oyster nursery culture in the earthen pond and the mangrove canal, respectively.

Discussion

The development of hatchery production techniques for the cupped oyster, *Crassostrea iredalei,* (Devakie and Ali 2000a, 2000b, 2002, Teh et al. 2012, 2016) has permitted oyster culture to become independent of the inherent variability associated with the collection of spat or adults from the wild. To capitalize on these developments, however, appropriate nursery culture protocols are required. The oyster spat must be transferred to an appropriate nursery culture site to reduce operating costs for producing microalgal feed. Transferring spat and juvenile oysters from the hatchery to the sea is a critical step, and the methods employed affect subsequent growth and survival. Transferring hatchery-reared oyster spat (*C. belcheri*) of a size larger than 1.2 cm in length to nursery culture in the sea has shown a zero mortality rate and high growth rates (Tanyaros and Tarangkoon 2014). The higher survival rates for sub-adult oysters in the earthen pond than for those reared in the mangrove canal was attributed in this study to the damage that crabs and puffer fish inflicted on the plastic mesh that allowed predators access to the oysters, which lead to the loss of seed during the nursery culture period. This often occurs in natural waters and is difficult to control. The loss of seed during oyster nursery culture in the mangrove canal led to decreased density that led to higher growth performance (AGRL and DGR) in sub-adult oyster nursery culture in the mangrove canal in comparison with that in the earthen pond. There is an inverse relationship between density and growth, and individuals held at lower densities have faster growth rates, greater overall individual weight, higher individual meat weight, and increased production compared with those held at higher stocking densities (Roland and Albrecht 1990, Holliday et al. 1991, 1993, Parsons and Dadswell 1992). In this study, the results reveal a higher Condition Index for sub-adult oyster culture in the mangrove canal than for oysters cultured in the earthen pond since this habitat provides rich food sources. Within estuarine ecosystems, mangroves play important roles in biodiversity and energy flow and in maintaining functioning food chains, and phytoplankton plays a vital role as a primary producer (Sridhar et al. 2006). Mangroves also have high potential as locations to nurse and rear hatchery-produced juvenile oysters. The nursery culture of sub-adult oysters in the mangrove canal did not result in higher shell compressibility than it did in an earthen pond because water was exchanged to maintain calcium and magnesium concentrations during the culture period. However, increased amounts of excretory products and CO_2 dissolved in pond water that are caused by high stocking densities result in decreased pH levels and changes in seawater carbonate chemistry, with a reduction in the saturation states of aragonite and calcite, which are the most prevalent shell-forming carbonates (Orr et al. 2005). Therefore, frequent water exchange may be required in ponds to maintain calcium and magnesium concentrations when stocking density is increased.

In summation, both earthen ponds and mangrove canals are potential sites for culturing sub-adult oysters. When conducting oyster nursery culture in earthen ponds it is easy to control predators and stimulate the growth of microalgae by adding fertilizer, but water exchange must be managed properly to maintain appropriate water quality and aeration, and pond water circulation increases electricity costs. Oyster nursery culture in mangrove canals requires methods to prevent predator access to the oysters.

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Author contributions. P.C. and S.T. designed and performed the research; S.T. and W.T. analyzed the data; S.T. wrote and corrected the paper.

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