Final Report

Fishery Resource Grants Program

Project Title:

Grow-out and marketability evaluation of triploid DEBY oysters

Project Investigator: Gustavo Calvo

Period Covered by this Report: October 1, 2002 – 31 December 2004

INDEX

Abstract

Triploid DEBY clearly outperformed their diploid counterparts in terms of survival, growth and condition over the course of this study at the site of Sweet Amalia Oyster Farm in Mathews, VA. By November 2004, when oysters were approximately 28 month old, cumulative mortality was 11% in triploids and 43% in diploids. At that time, 41% of the triploids were of market size (3 inch) whereas none of the diploids had even attained 2.5 inch. Condition of triploids and diploids similarly increased in fall 2003 and remained high over the winter and spring 2004. However, while condition decreased slightly in triploids a pronounced decrease was observed in diploids in summer 2004. *Perkinsus marinus* was nearly absent in August 2003 and its prevalence in triploids and diploids similarly increased to 90-97% in November 2004. In August 2003, prevalence of MSX was 4% in triploids and 40% in diploids whereas no MSX was present in November 2004. Based on the good performance of triploids oyster dealers started to purchase triploid stock at a wholesale price of $0.25 an oyster in fall 2004.

Introduction

The DEBY strain, a selectively bred eastern oyster with demonstrated resistance against the principal diseases in Chesapeake Bay (Ragone Calvo et al., 2003) has been emerging as the stock of choice for aquaculture in Virginia. DEBY oysters, developed at the Virginia Institute of Marine Science with funding from the Commonwealth of Virginia and NOAA Sea Grant, have demonstrated higher survival and faster growth than wild oysters and various other strains tested primarily at medium salinity sites in the lower Chesapeake Bay. For example, F4-DEBY experienced 30% lower cumulative mortality and had lower *Perkinsus marinus* infections than cohorts from putatively disease resistant Tangier Sound and Mobjack Bay brood stock concurrently deployed in the lower York River, Virginia, for 29 months. (Ragone Calvo et al., 2003). In a study with triploid and diploid oysters derived from Mobjack Bay brood stock, Barber and Mann (1991) showed that both were equally susceptible to *P. marinus* whereas triploids grew faster than diploids. At the time when Barber and Mann conducted their study DEBY oysters had not yet been developed.

On the basis of the demonstrated dual disease resistance in DEBY and fast growth in triploids, the objective of this project was to examine the comparative performance of diploid and triploid DEBY in an aquaculture setting at Sweet Amalia Oyster Farm. The farm operates a 1.5acre lease within inter-tidal sand flats flanked by wetlands in Mobjack Bay, Virginia. The site is located in waters approved for shellfish growing by Virginia Department of Health. A rack system is used to grow oysters in mesh cages approximately 1 foot above the bottom. It was hypothesized that triploid DEBY would have an advantage over their diploid counterparts especially during the summer when condition and marketability of diploids decreases as gonadal development and spawning occurs.

Methods

Diploid and triploid DEBY oyster seed, 4-8mm in shell height, was purchased from Middle Peninsula Aquaculture on 6 November 2002. Triploids were produced from brood stock maintained by VIMS Aquaculture Genetic and Breeding Technology Center Hatchery Manager Ann Arseniu. Triploid production was conducted at the VIMS

Gloucester Point Hatchery by chemical treatment of fertilized eggs with cytochalasin-B following the method of Allen et al. (1989). Subsequently, triploid eyed-larvae were set and juveniles reared at Middle Peninsula Aquaculture in Mathews, VA. Diploids were produced from F4-DEBY brood stock (Ragone Calvo et al. 2003) maintained by Sweet Amalia Oyster Farm in Mathews, VA. Diploid production was conducted by standard hatchery procedures at Middle Peninsula Aquaculture.

On 8 November 2002, fifty thousand triploid and the same number of diploid DEBY seed was deployed at Sweet Amalia Oyster Farm in Mathews, VA. Oysters were placed in 3mm mesh cages deployed on inter-tidal racks (3m x 1m x 3/4m) made of 12mm reinforced steel rods. Stocking density was approximately 5000 oysters per cage. To facilitate identification, cages containing triploid oysters were color coded with a black tag and cages with diploid oysters were color coded with a white tag. Due to extreme cold weather conditions following deployment, oysters were moved over the winter from the inter-tidal site to a sub-tidal location on the East River and then retuned to the original site. Over the course of the study cages were cleaned of fouling organisms as necessary to minimize obstruction to water flow. As oysters grew they were graded on a 25mm sieve. Animals retained on the sieve were placed in 16mm mesh cages. Animals not retained on the sieve were placed in 5mm mesh cages. Density was adjusted to approximately 1000 animals in the larger mesh cages and 250 animals in the smaller mesh cages.

Three replicate cages with 250 oysters were established to monitor survival, growth, disease and condition. Mortality was determined by counting all live and dead animals in each replicate cage. Growth was determined by measuring shell height (longest oyster dimension) of 50 animals in each replicate cage. Mortality and size frequency were assessed on 7 July, 9 August and 30 October 2003; and on 19 January, 20 April, 19 June and 1 November 2004. Condition index was determined by the method of Lawrence and Scott (1982). Briefly, whole oyster weight, shell weight and wet and dry meat weight was measured. Dry weight was determined after placing meats at 80°C overnight. Condition index was calculated by the formula: CI = meat dry weight/(whole oyster weight – shell weight). Condition was assessed in samples of 30 oysters collected from 9 August 2003 to 19 June 2004 concurrent with mortality and

size frequency assessments. *Perkinsus marinus* (Dermo) and *Haplosporidium nelsoni* (MSX) disease was assessed in oysters (n=25-30) collected in 9 August 2003 and 1 November 2004. *Perkinsus marinus* was diagnosed employing Ray's Fluid Thioglycollate Medium Assay and MSX was diagnosed employing standard paraffin histology methods. Disease diagnoses were conducted at VIMS Shellfish Pathology Laboratory.

Results

Over the course of the study mortality in triploids remained below 11%, whereas during summer and fall of 2004 mortality in diploids rapidly increased from 10% to 43% (Figure 1). During 2003 growth rate was similar in triploids and diploids, whereas during 2004 growth diminished in triploids and plateau in diploids (Figure 2). In 2004, triploid growth rate was relatively high during spring and rapidly increased at he end of the summer and into fall. The percentage of market size (3 inch or greater) triploids rapidly increased from 8% in 19 June 2004 to 41% in 1 November 2004. In that time frame, none of the diploids attained market size (Table 1). Condition in triploids and diploids similarly increased in the fall from August to November 2003, remained from November 2003 to April 2004 and subsequently declined at a faster rate in diploids than in triploids from April to August 2004 (Figure 3).

Perkinsus marinus was nearly absent in August 2003 and its prevalence in triploids and diploids similarly increased to 90-97% in November 2004. At that time, *P. marinus* prevalence in wild oysters in the vicinity of the farm was 63% (Table 2). In August 2003, prevalence of MSX was 4% in triploids and 40% in diploids. In November 2004, MSX was absent (Table 3).

Discussion

Triploid DEBY clearly outperformed their diploid counterparts in terms of survival, growth and condition over the course of this study at the site of Sweet Amalia Oyster Farm in Mathews, VA. By November 2004, when oysters were approximately 28 month old, cumulative mortality was 11% in triploids and 43% in diploids. At that time, 41% of the triploids were of market size (3 inch) whereas none of the diploids had even attained

2.5 inch. Condition of triploids and diploids similarly increased in fall 2003 and remained high over the winter and spring 2004. However, while condition decreased slightly in triploids a pronounced decrease was observed in diploids in summer 2004. Based on the good performance of triploids oyster dealers started to be interested in purchasing the triploid stock as oysters attained 2 years of age in summer 2004. Unfortunately, wholesale of triploids (in lots of approximately 500-1000 oysters) for the half-shell restaurant market started once the summer had ended and thus it was not possible to assess marketability over the summer as intended. Wholesale price was $0.25 an oyster. This is the same wholesale price commanded by diploid stocks harvested from the farm in the last 3 years. Given the poor performance of diploids in the present study, none of the stock has been marketable and the expectation for marketing any of the remaining stock is low. The very poor performance of diploids in this study was somewhat surprising because diploid DEBY had been employed at Sweet Amalia Oyster Farm in previous years with relative success. It is possible that genetic variability in DEBY brood stock employed for production of diploids may have been low and resulted in loss of fitness in diploid progeny employed in this study. As the expanding aquaculture industry increases demand for DEBY seed, it will be important to ascertain the genetic identity of the seed available from various commercial hatcheries in Chesapeake Bay and to continue monitoring the performance of various DEBY strains that are currently available in the marketplace.

ACKNOWLEDGEMENTS

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Table 1
Percentage of harvestable oysters by size and date

	Diploid DEBY			Triploid DEBY		
	<63.5mm	63.5-76.1mm	>76.1mm	<63.5mm	63.5-76.1mm	>76.1mm
6/19/04	89%	11%	0%	47%	45%	8%
11/1/04	85%	15%	0%	18%	41%	41%

Note: 76.2mm = 3.0 inch (market size for wild harvest in Virginia), 63.5mm = 2.5inch.

Table 2.
***Perkinsus marinus* prevalence and intensity**

	Diploid DEBY				Triploid DEBY			
	Prevalence (*)	H	M	L	Prevalence (*)	H	M	L
8/9/03	4% (1/25)	0	0	1	0% (0/30)	0	0	0
11/5/04	97% (29/30)	5	11	13	90% (27/30)	1	10	16

(*) = (Number infected / number examined).
Infection intensity categories: H = heavy, M = moderate, L = light.

Table 3.
***Haplosporidium nelsoni* prevalence and intensity**

	Diploid DEBY				Triploid DEBY			
	Prevalence (*)	H	M	L	Prevalence (*)	H	M	L
8/9/03	40% (1/25)	0	2	8	4% (1/24)	1	0	0
11/5/04	0% (0/30)	0	0	0	0% (0/30)	0	0	0

(*) = (Number infected / number examined).
Infection intensity categories: H = heavy, M = moderate, L = light.

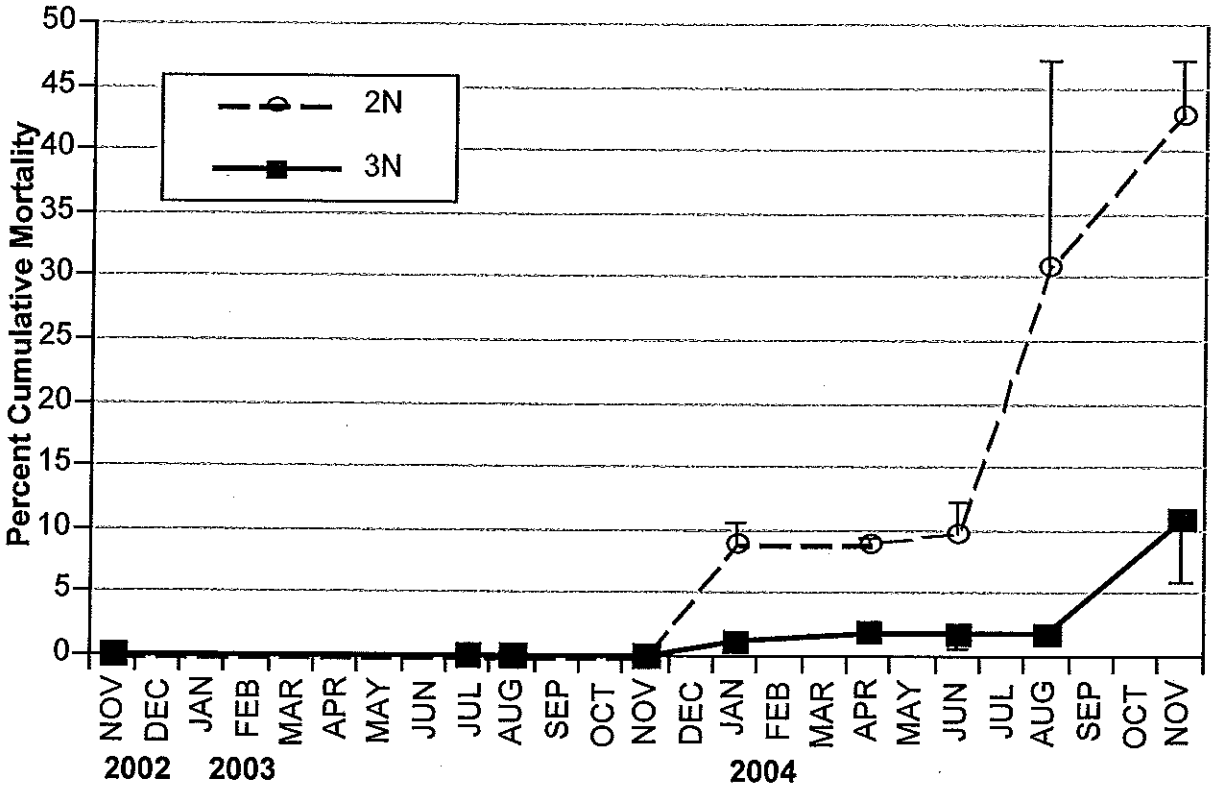

Figure 1. Mortality (Average +/-SD) in DEBY oysters. 2N = Diploids, 3N = Triploids

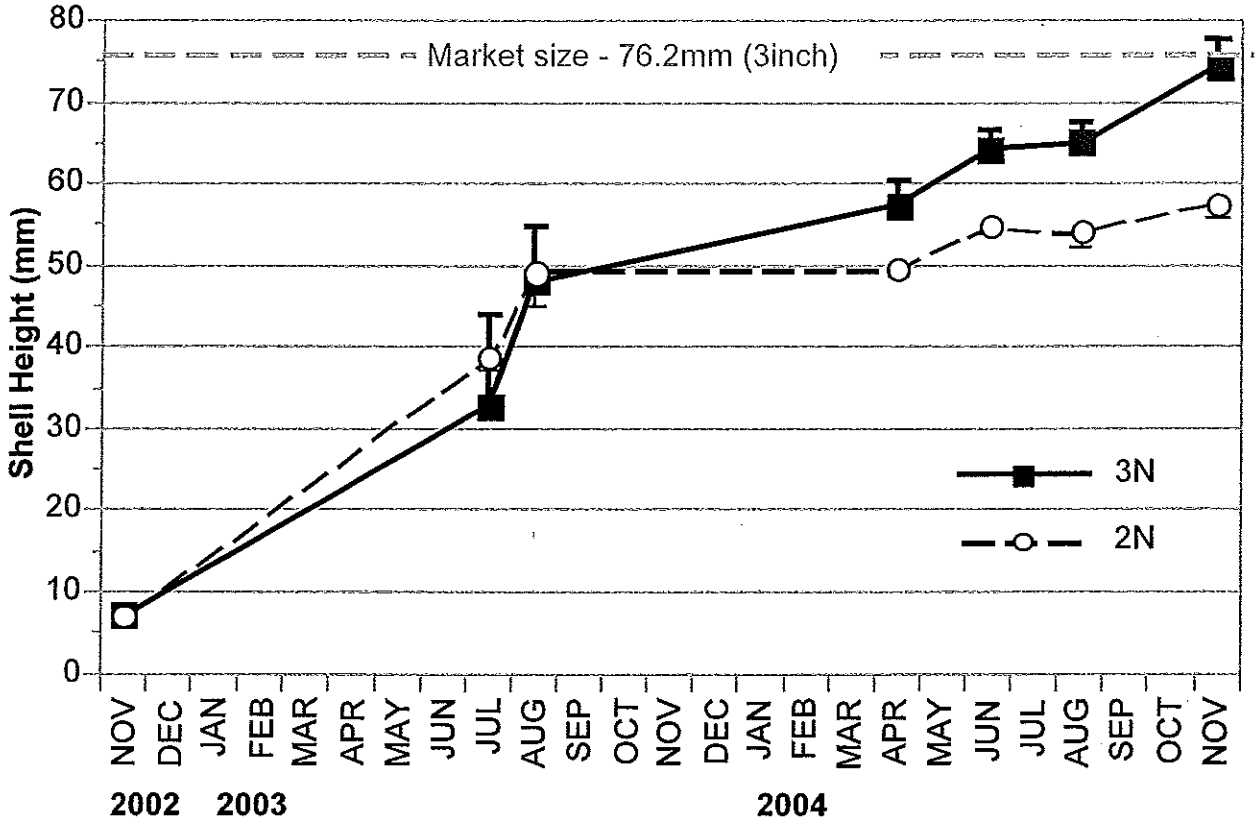

Figure 2. Growth (Average +/-SD) in DEBY oysters. 2N = diploids, 3N = triploids.

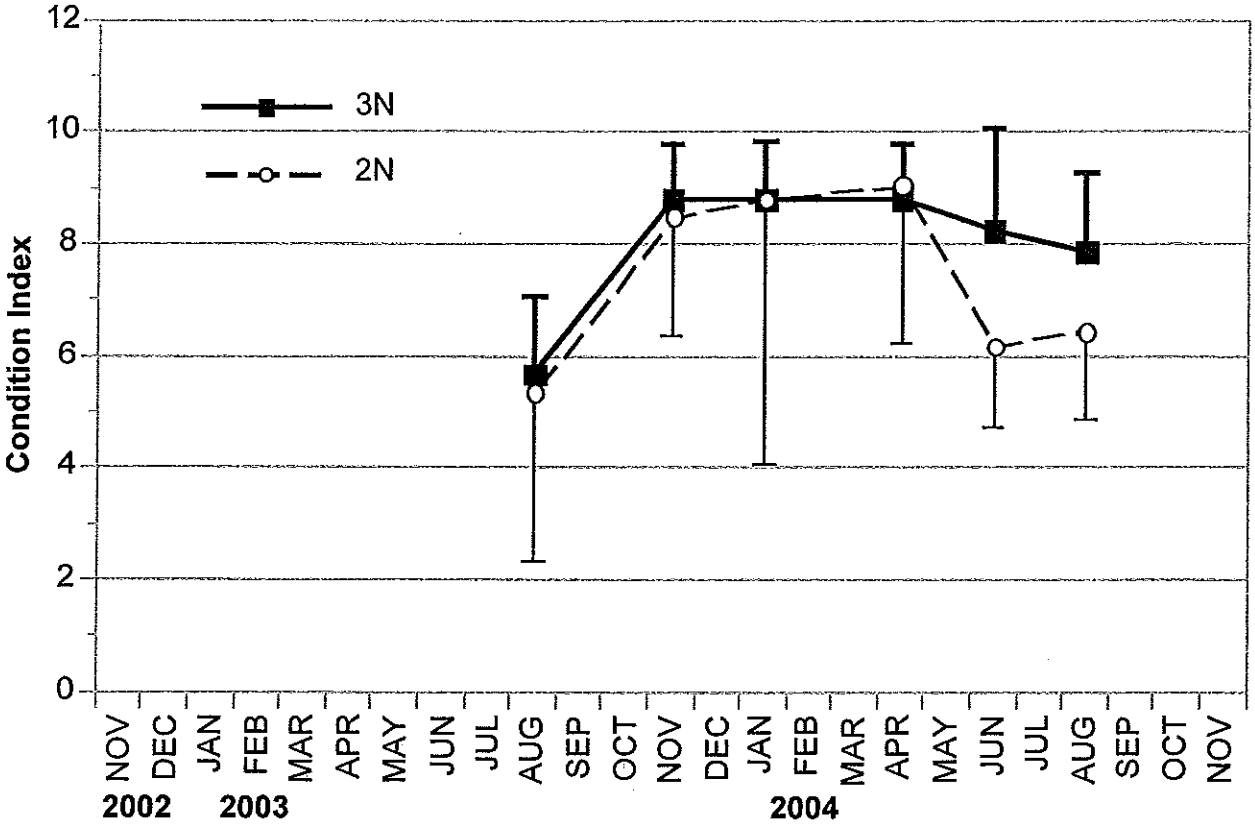

Figure 3. Condition index (Average +/-SD) in DEBY oysters. 2N = diploids, 3N = triploids.