# Off bottom oyster (*Crassostrea virginica* Gmelin) culture in Prince Edward Island: an evaluation of seed sources and stocking density

Luc A. Comeau, Garth Arsenault and Jeffrey Davidson

Fisheries and Oceans Canada Gulf Fisheries Centre P.O. Box 5030 Moncton, New Brunswick Canada, E1C 9B6

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Luc A. Comeau, Garth Arsenault<sup>1</sup> and Jeffrey Davidson<sup>1</sup>

Fisheries and Oceans Canada Gulf Fisheries Centre, Oceans and Science Branch P.O. Box 5030, Moncton, New Brunswick E1C 9B6

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<sup>&</sup>lt;sup>1</sup> Department of Health Management, Atlantic Veterinary College, University of Prince Edward Island, 550 University Ave., Charlottetown, Prince Edward Island, Canada,

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#### **ABSTRACT**

Comeau, L.A., Arsenault, G., and Davidson, T.J. 2011. Off bottom oyster (*Crassostrea virginica* Gmelin) culture in Prince Edward Island: an evaluation of seed sources and stocking density. Can. Tech. Rep. Fish. Aquat. Sci. 2976: viii + 13 p.

In Prince Edward Island (PEI), off bottom culture of oysters (Crassostrea viginica) has become the method of choice in at least a portion of the oyster aquaculture production cycle. The purpose of this study was to examine the effects of seed source and stocking density on the productivity of oysters cultivated off the bottom. Seed oysters were collected from five estuaries in PEI and subsequently transferred to a single study site. Oysters were placed in polyethylene bags and, using steel racks, suspended at a distance of approximately 50 cm above the bottom in the intertidal zone. Results show that, over the three-year period, there was no significant difference in mortality rates between the source estuaries. However, oysters originating from two of the five estuaries had significantly better growth rates compared to the remaining three source estuaries. Also, in the first year, oysters stocked at densities of 500, 750 and 1000 individuals per bag had significantly greater shell growth than those stocked at 2000 individuals per bag. Similarly, in the second year, oysters stocked at 250 and 500 individuals per bag had significantly greater shell growth than those stocked at 750 individuals per bag. In the third year, however, no significant difference in growth or mortality was found between stocking densities of 100, 200, 300 and 500 individuals per bag. Nonetheless, bags holding 500 oysters in the third year had significantly more commercial (lower) grade oysters than those stocked at lower densities.

#### **RÉSUMÉ**

Comeau, L.A., Arsenault, G., and Davidson, T.J. 2011. Off bottom oyster (*Crassostrea virginica* Gmelin) culture in Prince Edward Island: an evaluation of seed sources and stocking density. Can. Tech. Rep. Fish. Aquat. Sci. 2976: viii + 13 p.

L'élevage sur table de l'huître Crassostrea virginica devient une pratique courante à l'Île-du-Prince-Édouard (IPE). Notre étude a examiné les effets de la source de naissain et la densité d'élevage sur la productivité de l'huître sur table. Du naissain fut recueilli dans cinq estuaires de l'IPE et transféré dans un seul estuaire où leur croissance et mortalité ont été suivies au cours d'une période de trois ans. Les huîtres ont été maintenues dans des sacs en polyéthylène posés sur des tables ostréicoles, de sorte que les huîtres reposaient à environ 50 cm du fond dans la zone intertidale. L'analyse des données montre un taux de mortalité similaire entre les cinq sources de naissain. Toutefois, les huîtres de deux sources ont eu de meilleurs taux de croissance comparés aux trois autres sources. Aussi, au cours de la première année de l'étude, les huîtres maintenues à une densité de 500, 700 et 1000 individus par sac ont montré de meilleurs taux de croissance comparés aux huîtres maintenues à une densité de 2000 individus par sac. De la même façon, au cours de la deuxième année, les huîtres maintenues à une densité de 250 et 500 individus par sac ont connu de meilleurs taux de croissance comparés aux huîtres maintenues à une densité de 750 individus par sac. Au cours de la troisième année, aucune différence significative de la croissance n'a été enregistrée entre les densités d'élevage. Néanmoins, une qualité inférieure au niveau de la forme de la coquille fut notée chez les huîtres maintenues à une densité de 500 individus par sac.

#### INTRODUCTION

Off bottom culture is a relatively new production method for the farming of the Eastern Oyster, *Crassostrea virginica*, in Prince Edward Island (PEI) (Lavoie, 1996). The trend in the oyster aquaculture industry is towards off bottom culture in at least a portion of the oyster aquaculture production cycle and the use of bags mounted on racks has gained favor among oyster aquaculturists. Growth rate, survival and shell shape are generally considered important traits to determine the quality and grade of cultivated oysters (Medcof, 1961; Mahon, 1983; Lavoie, 1996).

In addition, an important consideration for any off bottom aquaculture endeavor is the selection of a reliable source for seed. The oyster industry in Eastern Canada depends on the collection of wild seed as opposed to hatchery sources. Studies with several bivalve species have demonstrated that animals from different sources can exhibit differences in rates of growth and survival (Haley & Newkirk, 1977; Newkirk, 1980; Pérez Comacho *et al.*, 1995; Labarta *et al.*, 1997). The effects of seed source are, however, difficult to extrapolate from one study to another (Newkirk, 1980). There are several sites on PEI where oyster seed is collected commercially, but there are no published studies comparing the rates of growth and mortality in a common site. Thus, the first objective of this study was to compare the growth and mortality rates of oysters obtained from five seed collection areas on PEI and maintained in one growing area.

There is often great variability in the size of individuals from the same year-class within an oyster population (Medcof, 1961; Newkirk, 1981). For example, larger individuals of the Manila clam (*Tapes philippinarum*) (St.-Félix *et al.*, 1984) and quahog (*Mercenaria mercenaria*) (Walker & Hurley, 1995) have been shown to maintain above-average growth rates compared to average-sized siblings. In some species, such as the oyster *Saccostrea cucullata*, growth rate is considered a heritable trait (Jarayabhand & Thavornyutikam, 1995) and superior-growth individuals are routinely used as part of a selective breeding program to improve growth rate and resistance to mortality (Newkirk, 1980). The second objective of this study was to compare growth and mortality rates of oysters demonstrating superior growth traits from four of the five seed collection areas on PEI.

An important consideration in bivalve aquaculture is how many individuals of a certain species can be grown before growth is limited or before mortality becomes unacceptable (Honkoop & Bayne, 2002). In the aquaculture environment, bivalves are often maintained in relatively confined spaces, and intraspecific competition becomes a limiting factor to growth possibly due to food limitation at higher densities (Hadley & Manzi, 1984; Jarayabhand & Newkirk 1989; Fréchette & Lefaivre, 1990; Parsons & Dadswell 1992; Fréchette, 1998). Beyond a certain threshold, growth rates tend to decrease with increasing stocking density (St.-Félix *et al.*, 1984). Increasing stocking density has also been shown, on occasion, to increase mortality rates (Fréchette & Lefaivre, 1990; Taylor *et al.*, 1997a,b) and to have a negative impact on shell shape in several bivalve species (Adams *et al.* 1994, Taylor *et al.* 1997a). It has also been demonstrated that a low stocking density can also increase the variability in body mass, as some individuals grow faster than others (Kautsky, 1982; Jarayabhand, 1988). Even at a small surface coverage, i.e. only a small part of the available surface area being occupied by individuals, and consequently, the distance between individuals is relatively

large, the decrease of growth can be substantial (Taylor *et al.*, 1997a, b). The third objective of this study was to examine the effects of stocking density on oyster growth, mortality and shell shape (grade).

#### MATERIALS AND METHODS

#### **Experiment A – Comparison of Oyster Growth from Different Seed Sources**

Oyster seed from five different spawning areas on PEI was acquired in May 1997 from a commercial oyster company for experiments spanning a three-year period. The five sources were: Cascumpec Bay, Bideford River, Vernon River, East River and Malpeque Bay (Figure 1). All seed was transferred to a single oyster lease in Covehead Bay to ensure that they were subjected to the same environmental conditions during the study period.

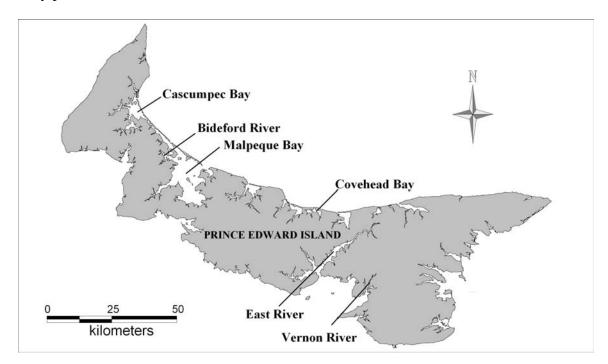


Figure 1. Map of Prince Edward Island indicating the location of study sites.

At the start of the study in May 1997 and subsequently in May of each study year, a random sample of 100 oysters, from each seed source, were measured for shell length. Shell length was defined as the distance from umbo to apex. The mean of these initial measurements was used as a baseline from which to compare growth for that study year. Oysters from each of the five locations were placed in 14 mm mesh bags at a density of 1000 (first year), 500 (second year) and 275 (third year) oysters per bag. These densities correspond to standard practices in the industry. The volumes approximated two liters. Bag replicates ranged from six to eleven as determined by oyster availability. In November of each year, a random sample of 25 oysters from each bag was measured for

shell length. Mortality estimates were expressed as a percentage of the initial oysters in the bag. Mortality was defined as a dead oyster or an empty shell.

#### Experiment B – Comparison of Oyster Growth from a Superior Seed Source

Oysters that exhibited superior growth in shell length during their first year were selected for this 2-year experiment. These oysters represented the top 10 % in shell length. Oyster seed originating from four different spawning areas on PEI were used for this experiment: Cascumpec Bay, Bideford River, Vernon River and Malpeque Bay. The experiment was conducted in an identical fashion to Experiment A, with the exception that the stocking density was 400 (first year) and 275 (second year), again approximating a volume of two liters.

#### Experiment C – Comparison of Oyster Growth at Different Stocking Densities

Oyster seed originating from the Bideford River site was used for the experiment investigating stocking density that spanned a three year period. The seed was supplied from a single oyster collection lease in the Bideford River. The experiment was initiated in May 1997 and subsequently, in May of each study year, a random sample of 100 oysters was removed and measured for initial shell length. The mean of these initial measurements was used as a baseline from which to compare growth for this experiment. Table 1 outlines the stocking density for each of the three years. Each study group consisted of ten replicates. In November of each year, a random sample of 25 oysters from each bag was measured for shell length. Mortality estimates were defined as a percentage of the initial oysters in the bag.

Table 1. Stocking Density during growth experiment.

Year	Stocking Density (oysters per bag)			
One (1997)	500	750	1000	2000
Two (1998)	250	500	750	
Three (1999)	100	200	300	500

#### Oyster Shape (grade)

The quality of Canadian Maritime oysters is based solely on shape. During the third year of Experiment C shell length and width were used to determine the relationship between oyster shape and stocking density. In late November, 50 randomly selected oysters from three bags of each stocking density were measured for shell length and

width. Oyster shape was determined by calculating length to width ratios (Millewski & Chapman, 2002) and assigning each oyster to a quality criterion: <1.5 = Fancy, <1.75 = Choice, <2.0 = Standard and  $\ge 2.0$  = Commercial. The lower ratios indicate a better grade.

#### Husbandry

The following protocols and equipment were the same for each study year in all of the experiments described above. Numbered tags were attached to each bag to ensure continuity of identification and to allow the experiments to be conducted as a blinded study. Bags were randomly placed on steel racks (oyster tables) located in the intertidal zone. Bags were flipped and cleaned every two to three weeks throughout the study period and husbandry practices were identical for all bags. Racks were constructed of rebar steel, holding five bags on one level, approximately one-half meter above the benthos. The bags were made of extruded polyethylene, measuring  $90 \text{ cm} \times 50 \text{ cm}$  with 1.4 cm, diamond shaped, mesh size.

#### **Statistical analysis**

Following ANOVA, Tukey and Bonferroni analyses were employed to compare shell length, the percentage mortality and shape. Homogeneity of variance was verified with Bartlett's test for equal variances. Statistical significance was set at the 0.05 probability level.

#### **RESULTS**

#### **Experiment A – Comparison of Oyster Growth from Different Seed Sources**

No significant difference in shell growth rate was observed between oysters from the five seed sources during the first year of growth. At the end of the second year, oysters from Cascumpec Bay had significantly (P < 0.05) better shell growth than oysters from the Bideford River and Malpeque Bay. During the third year of growth, oysters from Cascumpec Bay again, had significantly better (P < 0.05) shell growth than oysters from East River. Overall, for the three-year growth period, oysters from Cascumpec Bay and the East River had significantly better (P < 0.05) shell growth than those from the Vernon River, the Bideford River and Malpeque Bay (Table 2). There were no significant differences in mortality rates between the groups during the first and second study years. During the third year of growth, oysters from the Bideford River had significantly higher (P < 0.05) mortality rates than those from East River and Malpeque Bay. Despite this, there was no significant difference in the mortality rates between the seed sources over the combined study years (Table 2).

Table 2. Table showing mean  $(\pm SE)$  shell growth and mortality rates from five different sources of spawning. Total growth and mortality were obtained by adding yearly means.

	Growth (mm)				Mortality (%)			
Source	First year	Second year	Third year	Total	First year	Second year	Third year	Total
Cascumpec	20.8±3.9	23.5°±1.9	17.0°±1.0	61.3 <sup>e</sup>	23.2±18.1	2.2±1.8	$3.5^{gh} \pm 1.2$	28.9
Bideford	23.4±4.0	19.8 <sup>b</sup> ±2.3	16.7 <sup>cd</sup> ±2.1	59.9 <sup>f</sup>	11.5±10.8	1.1±1.4	$5.1^{\mathbf{g}} \pm 2.8$	17.7
Vernon	21.1±2.9	20.7 <sup>ab</sup> ±2.5	15.1 <sup>cd</sup> ±1.4	56.9 <sup>f</sup>	16±11.8	2.5±1.8	$3.4^{\text{gh}} \pm 1.8$	21.9
East River	24.8±3.4	21.9 <sup>ab</sup> ±1.3	14.5 <sup>d</sup> ±1.9	61.2 <sup>e</sup>	15.7±10.8	2.6±4.0	2.1 <sup>h</sup> ±1.6	20.4
Malpeque	20.5±4.1	20.3 <sup>b</sup> ±1.9	16.2 <sup>cd</sup> ±1.6	57.0 <sup>f</sup>	10.2±5.8	2.0±2.0	1.7 <sup>h</sup> ±0.7	13.9

 $(a\,,b)\,(c\,,d)\,(e\,,f)\,(g\,,h)$  significant statistical difference (P<0.05)

#### Experiment B – Comparison of Oyster Growth from a Superior Seed Source

No significant difference (P > 0.05) in shell growth was observed between any of the four groups in the first year of growth. During the second year of growth, oysters from the Bideford River and Malpeque Bay had significantly better (P < 0.05) shell growth compared to those from Vernon River. There was no significant difference in shell growth between any of the locations over the combined study period (Table 3).

There were no significant differences in mortality rates among all seed sources in individual years or over the combined study period (Table 3).

Table 3. Comparison of mean (±SE) shell growth and mortality rates of oysters from Superior Seed Sources. Totals were obtained by adding yearly means.

	Growth (mm)			Growth (mm) Mortality (%)			
Source	First year	Second year	Total	First year	Second year	Total	
Cascumpec	$16.4 \pm 4.3$	$19.8^{\mathbf{ab}} \pm 0.7$	36.2	$9.5 \pm 14.1$	$3.2 \pm 2.5$	12.7	
Bideford	$17.8 \pm 4.5$	$21.2^{\mathbf{b}} \pm 2.6$	39.0	$6.6 \pm 8.4$	$5.5 \pm 0.6$	12.1	
Vernon River	$21.2 \pm 2.7$	$17.9^{a} \pm 1.3$	39.1	$3.6 \pm 3.1$	$3.2 \pm 2.9$	6.8	
Malpeque Bay	$17.8 \pm 3.1$	$21.8^{\text{b}} \pm 0.9$	39.6	$4.2 \pm 4.7$	$4.6 \pm 4.6$	8.8	

 $<sup>^{(</sup>a,b)}$  significant statistical difference (P < 0.05)

#### **Experiment C – Comparison of Oyster Growth at Different Stocking Densities**

In year 1, bags stocked at 500, 750 and 1000 oysters had significantly greater (P < 0.05) shell growth than those stocked at 2000 oysters per bag (Table 4). There was no significant difference in mortality rates found among the four groups held for one year (Table 4).

Table 4. Comparison of mean (±SE) shell growth and % mortality at different stocking densities in the first year of growth.

Density	Growth* (mm)	Mortality (%)
500	$28.2^{\mathbf{a}} \pm 2.2$	$6.8 \pm 5.8$
750	$28.8^{a} \pm 2.1$	$6.4 \pm 4.5$
1000	$28.5^a \pm 2.3$	$5.3 \pm 3.9$
2000	$25.1^{\mathbf{b}} \pm 2.4$	$4.1 \pm 3.2$

 $<sup>^{</sup>a,b}$  significant statistical difference (P < 0.05)

In year 2, bags stocked at densities of 250 and 500 oysters had significantly greater (P < 0.05) shell growth than those stocked at 750 oysters per bag (Table 5). At a density of 500 oysters per bag the mortality rate was significantly lower (P < 0.05) than in bags stocked at 250 oysters (Table 5).

<sup>\*</sup>initial mean shell length was 23.3 mm  $\pm$  2.24.

Table 5. Comparison of mean (±SE) shell growth and % mortality at different stocking densities in year 2.

Density	Growth* (mm)	Mortality (%)
250	$16.8^{a} \pm 2.1$	$5.1^{\rm d}\pm2.5$
500	$16.8^{\mathbf{a}} \pm 3.2$	$1.4^{\rm c}\pm1.7$
750	$12.4^{\mathbf{b}} \pm 3.9$	$3.9^{\text{d}} \pm 3.5$

 $^{(a, b) (c, d)}$  significant statistical difference (P < 0.05)

In year 3, no significant difference in growth or mortality was found between stocking densities of 100, 200, 300 and 500 oysters per bag (Table 6).

Table 6. Comparison of mean (±SE) shell growth and % mortality at different stocking densities in year 3.

Density	Growth* (mm)	Mortality (%)
100	$11.1 \pm 2.9$	$2.6 \pm 1.7$
200	$12.4 \pm 3.3$	$3.2 \pm 3.2$
300	$13.2 \pm 3.0$	$3.9 \pm 5.0$
500	$13.6 \pm 2.6$	$3.4 \pm 3.4$

no significant statistical difference (P > 0.05)

<sup>\*</sup>initial mean shell length was 49.1 mm  $\pm$  2.03.

<sup>\*</sup>initial mean shell length was 63.7 mm  $\pm$  0.6.

#### **Shell Shape**

There was significantly (P < 0.05) more commercial grade oysters in bags stocked at 500 oysters per bag than at lower stocking densities. In oysters stocked at densities of 100, 200, and 300 there were between 5.3 % and 8.0 % more choice oysters than those stocked at 500 oysters per bag (Table 7).

Table 7. Table showing the mean (±SE) proportion of oyster grade for each stocking density.

Density	Fancy (%)	Choice (%)	Standard (%)	Commercial (%)
100	$32.7 \pm 4.2$	$54.0 \pm 0.0$	$12.7 \pm 5.0$	$0.7 \pm 1.2^{\mathbf{a}}$
200	$37.3 \pm 5.0$	$46.7 \pm 5.0$	$16.0 \pm 0.0$	$0.0 \pm 0.0^{\rm a}$
300	$41.3 \pm 11.0$	$45.3 \pm 9.9$	$13.3 \pm 2.3$	$0.0 \pm 0.0^{\mathbf{a}}$
500	$34.0 \pm 5.3$	$44.7 \pm 3.1$	$16.0 \pm 6.9$	$5.3 \pm 3.1^{\mathbf{b}}$

 $<sup>^{(</sup>a,b)}$  significant statistical difference (P < 0.05)

#### **DISCUSSION**

The overall growth suggests that oysters from Cascumpec Bay and East River grow to market size faster than those from the Bideford River, Vernon River and Malpeque Bay. Other studies have shown bivalve growth rates to be source-dependant (Newkirk, 1980; Pérez Comacho *et al.*, 1995). Moreover, growth rate has been shown to be heritable in several Bivalve species including in *C. virginica* (Haley & Newkirk 1977; Losee, 1977).

It is unlikely that the populations of *C. virginica* used in this study are genetically different and the differences observed between growth rates may be linked to heterozygosity. Indeed, gene flow in the marine environment is generally strong for species with planktotrophic stages. Although it may seem advantageous for the aquaculturist in PEI to use oyster seed from Cascumpec Bay and East River, consideration should be given to the possibility of genetic-environment interactions in the results of this study. Certain characteristics, or *phenotypes*, including growth rate, may vary according to the environment (Rawson & Hilbish, 1991; Labarta *et al.*, 1997) and seldom will a phenotype have superior performance in all environments (Rawson & Hilbish, 1991). In fact, phenotypic performance in *C. virginica* has been linked to location (Mallet & Haley, 1983). It is important, therefore, to test the performance of seed from a given source in the anticipated aquaculture setting, prior to large-scale production.

Significant differences in the mortality rates were only observed in the three year growth period, between seed obtained from the Bideford River, East River and Malpeque

Bay. These mortality rates do not correlate with significant differences in the observed growth rates for seed from these sources during the three-year growth period. There is no obvious indication as to why mortality rates were greater in seed from the Bideford River during that year.

Unlike results from Experiment A, where "regular" seed was used, there was no significant difference in the total growth of different seed sources selected for superior growth characteristics. There was, however, a significant difference in growth observed over the two-year growth period, between superior growth seed from the Bideford River and Malpeque Bay compared with superior seed from the Vernon River. Superior seed from Cascumpec Bay grew at an intermediate rate. It is worthwhile to note that over a two year growth period, the comparable, "regular" seed from the Bideford River and Malpeque Bay used in Experiment A had significantly inferior growth compared to the "regular" seed from Cascumpec Bay. This raises the question of why the performances were not similar for both "regular" seed and superior-growth seed from the Bideford River and Malpeque Bay in the two-year growth period. One possibility is that the superior-growth individuals from those sources were better suited (e.g. resistant fringe, thicker shell) to environmental conditions experienced during that year than the "regular" seed, resulting in better growth (phenotype) performance.

Stocking density appeared to have an effect on growth rate during the first year of growth, but had no significant effect on mortality. This is in agreement with other studies on oysters and other bivalves that failed to demonstrate a positive correlation between stocking density and mortality (Hilbish, 1986; Holliday *et al.*, 1991; Mgaya & Mercer, 1995; Hurley & Walker, 1996; Taylor *et al.*, 1997b). Optimal stocking density for one-year old oysters in the present study was between 500 and 1000 oysters per bag. Future experiments with intermediate densities between 1000 and 2000 oysters per bag may pinpoint the optimum stocking density, which would allow the aquaculturist to maximize the productivity of rack and bag culture during the first year of oyster growth.

During the second year of growth, optimal stocking density was between 500 and 750 oysters per bag. It is not known why mortality was highest in the lowest density bags stocked at 250 oysters per bag. Generally, mortality increases at higher stocking densities (Holliday *et al.* 1993, Fréchette *et al.* 2000).

There was no significant difference in the growth rates or in the mortality rates at the four densities tested in the three-year growth experiment. Given the results of the growth comparison at different stocking densities in the two year growth experiment, the most cost-effective stocking density during two and three year growth spans would be 500 oysters per bag. Maintaining stocking densities from one year to the next eliminates any handling necessary for redistribution to lower densities. The aquaculturist, however, also needs to consider shell shape which affects the commercial value of the oysters when determining stocking density in their production methods. In this study, there were significantly more commercial (low value) oysters in bags stocked at 500 oysters per bag than in bags stocked at the three lower densities. Depending on marketing objectives, the aquaculturist may therefore decide to restock bags to lower densities during the third year growth.

#### **CONCLUSION**

The results obtained in this study are uniquely applicable to oyster culture in PEI. These experiments can, however, be reproduced elsewhere to meet specific requirements of other geographic areas. It may be worthwhile to monitor the general health and performance of the source populations in parallel to field experiments. Information on the source population might allow a more detailed interpretation of seed performance under experiment conditions. Testing the condition index of seed prior to the start of the experiment may also be a useful tool in interpreting data (Lucas & Beninger, 1985).

The fine-tuning of off bottom culture will enable the aquaculturist to maximize profits while decreasing the risk involved with mortalities and decreased production. Determining the best areas for spat collection will hopefully allow the industry to take advantage of natural selection for growth of oysters in off bottom culture. Identifying superior individuals for growth in off bottom culture and retaining those individuals for husbandry purposes could form a basis for a hatchery-based industry if the need arises.

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