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Application of Triploidy to an Emergent Oyster Aquaculture Industry on the West Coast of Florida

Investigators:
Leslie N. Sturmer, Extension Service, Florida Sea Grant, IFAS University of Florida
Charles M. Adams, Florida Sea Grant Extension Program, Food Resource and Economics, University of Florida,
Charles A. Sims, Food Science and Human Nutrition Department, University of Florida,
Huiping Yang, School of Forest Resources and Conservation, Fisheries and Aquatic Sciences Program, University of Florida IFAS
Susan Laramore, Aquaculture and Stock Enhancement, Harbor Branch Oceanographic Institute, Florida Atlantic University

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RESEARCH PROJECT STATUS

The overall goal of this project was to provide the necessary infrastructure via a public-private partnership to commercialize the eastern oyster *Crassostrea virginica* through demonstration and evaluation of a breeding technique to local conditions on Florida's west coast. The planned scope of work did not change and all project objectives were completed.

ATTAINMENT OF RESEARCH PROJECT OBJECTIVES

OBJECTIVE 1. Document the production performance of diploid and triploid oysters under commercial conditions and quantify effects of different growing locations, culture techniques, and seasonal harvests.

Seed Production - Spring 2016

Single-set triploid oyster seed were produced in April by crossing west coast Florida stocks with sperm from tetraploid stocks obtained from the Louisiana Sea Grant oyster hatchery. Dr. John Supan with Louisiana State University (LSU) delivered the tetraploid stocks and assisted in the spawn conducted at a commercial hatchery in Cedar Key. Diploids were also produced by using the same Florida stocks. Ploidy of tetraploids, triploid larvae and post-set was confirmed using flow cytometry. Seed were nursed at a commercial facility using identical upwellers and barrel drum systems.

Grower Trials 1, July 2016 – March 2017

When the oyster seed reached 20-22 mm in shell height (SH), 2,500 of each ploidy type were distributed in July to certified growers, who were approved to culture oysters on their shellfish aquaculture leases. Grower selection was based on willingness to participate, location of their lease, and type of culture gear used. Ten growers in four west coast counties (Charlotte, Franklin, Levy, and Wakulla) used a variety of culture systems (floating bags, bottom cages, and adjustable long lines), which allowed for evaluation of site and gear interaction on ploidy type.

The large seed size provided to growers limited the number of bag changes over the growout period and minimized possible mixing of diploid (2N) and triploid (3N) stocks during the transfer to larger mesh bags. Growers maintained the seed using their own gear until harvest. Color-coded tags and zip-ties were provided to identify ploidy stocks throughout the culture period and to distinguish the stocks from other oysters being cultured on the lease. To document production performance diploid and triploid oysters under commercial conditions, oysters from three replicate bags (four baskets if using the adjustable longline system) for each ploidy type (a total of 6-8 culture units per grower) were supplied to the UF project team at harvest. Large color-coded tags were provided to growers to mark the replicate bags. These bags were stocked at a final density used by the grower and maintained similarly as other culture bags except they were not sorted or graded for harvest. When most of the oysters reached harvest size, the sample oysters were collected by the UF team.

In addition to seed, each participating grower received a notebook containing information on the project, seed documentation (e.g., disease report, ploidy verification), culture activity forms (see Objective 4), and selected references. To document water quality conditions, a refractometer was provided so each grower could measure salinity on their lease. In addition, a data logger, which measured water temperatures continuously, was provided and placed inside a culture bag. At harvest, the logger was retrieved and returned to UF for downloading of data.

In March 2017 after almost eight months in growout and just 11 months from spawning, oysters were collected by UF staff from seven of the ten participating growers. Three growers in Levy County lost most of their oysters due to the impacts of Hurricane Hermine so no information was obtained. Due to a misunderstanding of how to set aside oysters for the UF samples, it was decided not to include information from one of the Wakulla County growers. Several growers noted that oysters had reached market size one to two months earlier and would have typically been sold then, which may have affected their results. To document growth, samples from each bag were measured for shell height, length, and width. Oysters were also weighed for total and meat (wet) weights. Survival was determined by counting the number of live and dead oysters in each bag. Samples were collected per bag per grower for determining presence of internal and external parasites, verifying ploidy, and assessing biofouling.

Triploid oysters were larger, and meats weighed more than diploid oysters at each site with the exception of oysters cultured using the adjustable long line system; however, growth was similar for both ploidy types at this farm site in Oyster Bay (Wakulla County). Again, except for this site, average shell height of triploid oysters ranged from 73 mm at Alligator Harbor (Franklin County) to 98 mm at Cedar Key (Levy County), confirming growers' observations that oysters exceeded market size. Both Alligator Harbor growers and one of the Cedar Key growers used the floating bag system. In contrast, average shell height of diploid oysters ranged from 65 mm at Alligator Harbor to 83 mm at Cedar Key. Although smaller, these oysters had also reached market size in this time frame. Meat weights (MW) followed the same trends for ploidy type and farm location. It is interesting to note that at one of the Cedar Key farms, average meat weight of triploid oysters (14 grams) doubled that of diploid oysters (7 grams).

Survival of diploid and triploid oysters was more difficult to address as survival was only determined for oysters in the sample culture units, not for the entire crop. With the exception of oysters grown at Charlotte Harbor (Charlotte County), where high mortality was observed in bottom cages due to the prolonged red tide bloom, survival of both ploidy types was commercially acceptable, ranging from 89-99%. Differences in fouling was observed by gear type and farm location. The most heavily fouled oysters were from floating bags at Cedar Key, with naturally occurring oyster spat being the dominant organism. Interestingly, the bottom cages used at a different lease area in Cedar Key resulted in little fouling; bags were pressure washed routinely. Oysters grown at Charlotte Harbor and Oyster Bay had some oyster spat and barnacles on the shells. The least amount of fouling was observed on oysters grown at Alligator Harbor, where both growers employed biofouling control methods by flipping and shaking bags on a routine basis.

UF Field Trials 1, August 2016 - April 2017

Triploid and diploid oyster stocks were planted in August by project investigators at the UF experimental lease located within a commercial lease area off Cedar Key. Detailed information was collected to document practices (for example, biofouling control, bag transfers), production, and labor. Approximately 5000 seed of each ploidy type were placed into four 9 mm mesh Vexar bags, resulting in a stocking density of 1250 oysters per bag. Shell measurements were similar for both ploidy stocks (26 mm SH), but total weights (TW) differed (3N, 2.5 grams; 2N, 1.9 grams). The gear used to culture oysters was the floating bag. Two 4.5" square floats were used per bag; bags were attached to a longline secured by screw anchors. After 25 days, oysters were measured to estimate growth. Triploid oysters averaged 40 mm SH and 8.1 grams TW, while diploid oysters averaged 37 mm SH and 5.6 grams TW. To control fouling, bags were flipped weekly and allowed to aerial dry for at least 6 hours. Very little fouling was noted during this time period.

During September and October, the eight culture bags were retrieved twice from the lease and juvenile oysters were held temporarily in the UF shore-based nursery system in anticipation of hurricanes (Hermine and Matthew). After each hurricane, the lease was inspected and longlines were intact. On October 20, replicated field trials were initiated to document the effects of ploidy, stocking density, and float design on oyster production. About 2700 oysters of both ploidy types were stocked into eighteen 12 mm mesh Vexar bags at 125, 150, and 175 oysters/bag (6 bags/density/ploidy type) for a total of 36 bags. Each bag was supported by two 4.5" square floats. The average size of diploids was 51 mm SH and 18 grams TW, whereas triploids averaged 54 mm SH and 26 g TW. In addition, six bags with cylindrical or "bullet" floats attached on both sides of bag were stocked each with 150 3N oysters. With this float design, the bags were flipped routinely, but the placement of the floats allowed the oysters to always be submerged. Thus, the bags did not have to be "flipped back" after drying as was required of bags supported by square floats attached to the top of bags. This allowed for evaluation of two management practices for biofouling control.

Oysters were sampled every 6-8 weeks to determine growth and survival. Three bags from each of the density and ploidy combinations, a total of 18 bags, were sampled each period. In December, oysters in half of the replicate bags were measured and counted. At that time, there were no differences ($P>0.05$) in stocking densities or float designs for both ploidy types. Shell length of triploid (61.0 mm) and diploid (59.1 mm) oysters was similar. However, significant differences were observed for total weight (3N, 44.8 grams; 2N, 30.9 grams; $P=0.003$). Survival was high and similar for both plody types (2N, 99.6%; 3N, 98.1%; $P=0.07$). Overset of natural oyster spat was pronounced in oysters of both ploidy types, and a ranking scheme was developed to estimate degree of biofouling. No other fouling organisms were noted. Oysters in the other half of the replicate bags were sampled in February 2017. During this sample period, growth did not differ between ploidy types (3N, 71 mm SH; 2N, 68 mm SH). Average weight (total) of triploid oysters (69 grams) continued to exceed diploids (51 grams) ($P=0.004$). Survival was found to be significantly higher ($P=0.0005$) in the diploids (100%) compared to triploids (95%). As in the first sampling period, float design did not appear to influence oyster growth or survival. Stocking density also had minimal effect on shell height as triploid oysters averaged 72 mm in high density bags, 72 mm in medium density bags, and 70 mm in low density bags. No differences in total weight due to stocking densities were observed.

After eight months, all oysters were harvested in April when the majority had reached or exceeded 75 mm SH, one year from spawning. Growth and survival were determined following the same methods used in the growers' trials. No significant differences ($P>0.05$) were found in growth or survival of diploid and triploid oysters at the three stocking densities evaluated. Thus, measurements were averaged by ploidy type across stocking densities. The shell height of triploid oysters averaged 83 mm, which was significantly higher ($P=0.001$) than diploid oysters (76 mm). Total weights were also significantly different ($P=0.004$) with triploid oysters averaging 143 grams and diploid oysters averaging 109 grams. Likewise, both wet meat weights and dry meat weights differed with triploid oysters averaging 13 grams wet MW ($P=0.003$) and 3.1 grams dry MW ($P=0.0001$), and diploid oysters averaging 9 grams wet MW and 1.7 grams dry MW. Condition index, an estimate of the meat yield, is a relationship between dry MW, wet MW, and shell weight. Consequently, condition index was significantly higher ($P=0.0001$) in triploid oysters (12.9) than diploid oysters (9.7). In contrast, survival was higher ($P=0.0001$) in diploid oyster bags with an average of 97%, while triploid oyster bags had an average survival of 90%. Nonetheless, results obtained for both ploidy types over a "winter" growing period (August through April) were commercially acceptable.

At harvest, triploid oysters grown in bags with bullet floats attached to the sides were significantly larger (88 mm SH, $P=0.002$) than triploid oysters grown in bags with square floats attached to the tops of bags (80 mm SH). However, survival of oysters differed significantly ($P=0.002$) in bags supported with square floats (91%) when compared to oysters in bags supported with bullet floats (84%). As the oysters grew and bags became heavier, the bullet floats did not have enough buoyancy to support the weight. During the last month of the culture period, all bullet float-supported bags were found vertical in the water column, which resulted in oysters being crowded at one end of the bag. This most likely contributed to increased mortality.

Shell measurements were used to calculate fan and cup ratios to determine if external shell appearance was appealing for the half shell market. Shell measurements are used to calculate fan ratio (shell length/shell height) and cup ratio (shell width/shell height). Fan ratios of 0.66 and above and cup ratios of 0.33 and above are considered favorable by industry experts for half shell oysters designated for raw bars. Fan ratios differed among ploidy types (2N, 0.68; 3N, 0.72; P=0.0001) as did cup ratios (2N, 0.33; 3N, 0.36; P=0.02). Nonetheless, ratios for both ploidy types were within or exceeded the favorable ranges. There was a distinct difference between the two ploidy types, with triploids retaining their characteristic black "strike" on top of the shells.

The warm water temperatures in Florida allow for accelerated and year-round growth of oysters; however, these conditions can also increase the presence of biofouling organisms. In these field trials, oyster spat naturally recruited from "wild" oysters were the predominant fouling organism. Spat were counted on cultured oysters from each bag to determine the percent fouling. Only 20% of the diploids and 23% of the triploids did not have any spat on their shells. Regardless of ploidy, density, or float type, 28% of the oysters had one spat per shell, and 19-21% had two spat per shell. Thus, approximately 50% of the oysters had at least one spat per shell but they were considered marketable. Oysters with two or more spat per shell required cleaning prior to marketing.

Seed Production - Fall 2016

To quantify the effects of seasonal harvests on ploidy, several spawns using tetraploid oysters held from stocks delivered in April 2016 were attempted at the participating commercial hatchery; however, viable gametes were not obtained. Availability of triploid seed was limited, and it was decided to purchase seed directly from the LSU oyster hatchery and Auburn University shellfish hatchery. Disease certifications and ploidy verification required by the Florida state agency were obtained. A similar number of diploid seed were purchased from the Florida commercial hatchery from a spawn that occurred within a similar time period as the triploid spawns. Seed shipped in November were small (about 2 mm SH) and were nursed at the commercial facility in Cedar Key over the winter. All three stocks reached 20-25 mm SH in March 2017.

Grower Trials 2, March 2017 – November 2017

The second phase of the demonstration project evaluated the performance of diploid and triploid oysters planted in early spring, as opposed to the first phase in which oysters were planted in late summer. Diploid and triploid seed (2,500 of each) were distributed to eight growers in four counties during March. Some of the same growers who participated in the first trial did so in the second. Locations and gear types included bottom cages at Pine Island (Lee County), floating bags at Cedar Key, floating cages and adjustable longlines at Oyster Bay, and floating bags at Alligator Harbor. Oysters grown at the Pine Island location were harvested after four months in growout as biofouling on the bottom cages was difficult to maintain. At the other sites, oysters began reaching harvest size after eight months; in November, sample culture bags were collected from growers. Data from two growers was excluded from this summary due to confusion of how to maintain the samples. Further, information from one of the farm sites in Alligator Harbor was not obtained. The same protocol used in the previous growers' trials was used to document growth and survival.

Triploid oysters were larger and meats weighed more than diploid oysters at each site except for oysters cultured in floating cages at Oyster Bay. As in the first trials, this was the only location where diploid and triploid oyster growth was similar (52 mm SH, 3g MW). As opposed to the first trials, diploid oysters at the other sites had also not reached market size, ranging from 54 mm SH at Alligator Harbor to 61-63 mm SH at Cedar Key. Triploid oysters at these sites ranged from 67 mm SH at Alligator Harbor to 73-75 mm SH at Cedar Key. Meat weights followed a similar trend with diploid oysters averaging 3 grams at all locations and triploid oysters ranging from 5 grams at Alligator Harbor to 6-7 grams at Cedar Key.

Relevant observations of survival from sample bags at each participating farm were documented. Survivals were overall lower than those in the first set of trials regardless of ploidy, farm location, or gear type. At the Cedar Key farms, survivals for both ploidies were similar (2N: 68-70%, 3N: 57-68%). At the Oyster Bay farm where floating cages were used, survivals were higher than those at Cedar Key but, again, similar for both ploidy types (2N: 82%, 3N: 85%). Differences in survival were observed between ploidy types at the Alligator Harbor farm site. Survival of triploid oysters was 92%, the highest obtained at all farm sites in this set of trials, whereas survival of diploid oysters was 67%. Oyster survival for the Pine Island location was not estimated due to the early termination of the trial.

Differences in fouling were observed by ploidy type and farm location. As in the first trials, oysters grown in Cedar Key exhibited the most biofouling. This time, barnacles were the dominant organism. Oysters grown in Oyster Bay and Alligator Harbor had substantially less fouling, consisting primarily of barnacles. To quantify the amount of fouling on the oysters, organisms were scraped from the shells of both diploid and triploid oysters and weighed. The oysters were shucked, and shells weighed. Biofouling was expressed as a percentage of the fouling weight to the corresponding shell weight. At Cedar Key farms, fouling was lower for triploid oysters (43-57%) than diploid oysters (81-143%), as opposed to the other two locations where fouling amounts were overall lower but similar for both ploidy types. At Oyster Bay, fouling was estimated at 22% for triploids and 7% for diploids, whereas at Alligator Harbor, fouling was 9% for triploids and 26% for diploids.

Differences in oyster production between the two seasonal trials were apparent at all farm locations. Growth was substantially slower during the second set of trials when most of the culture period occurred during warmer water temperatures (farm averages: 80-82ºF) as compared to the first set of trials, which were conducted at lower water temperatures (farm averages: 71-81ºF). Warm water temperatures, characteristic of shallow coastal waters in Florida, enable year-round growth and shorter crop periods; however, excessively high temperatures in the summer months may inhibit growth and increase stress, resulting in increased mortalities.

UF Field Trials 2, April - November 2017

In addition to seed provided to growers, oysters of each ploidy type were reared by the UF project team at their experimental lease off Cedar Key. In April, diploid oysters (29 mm SH) and triploid oysters (34 mm SH) were stocked into 9 mm mesh Vexar bags at densities of 700 per bag. After seven weeks, diploids averaged 39 mm SH and 9 grams TW, while triploids averaged 48 mm SH and 17 grams TW. On June 7, diploid and triploid juveniles were restocked into forty 14 mm mesh Vexar bags at a density of 150 per bag. To further examine the effects of float type (square versus cylindrical or bullet) and float placement (square floats on sides of bags versus tops of bags) on oyster production and fouling control, replicated (n=4) trials were conducted. In addition, two commercially available, biocide-free, antifouling coatings were applied to four replicate bags supported by the square floats and stocked with triploid oysters.

Weekly, all oyster bags were flipped so that one side of the bag was exposed to air for drying of fouling organisms. During the summer, bags were flipped in late afternoon, so that oysters were exposed over night when air temperatures were cooler. Bags in which the 4.5” square floats were placed on top of the bags required being flipped back after exposure. Bags supported by floats, both square and bullet, attached on the sides of bag did not have to be flipped back after drying, resulting in labor requirements being reduced by half.

Two of the four replicate bags from each treatment were sampled to estimate oyster growth and survival after seven weeks, while the other two replicate bags were sampled after 14 weeks. In July, shell height of triploid oysters (56 mm) was greater than diploid oysters (47 mm). Similarly, triploid oysters (35 grams TW) weighed more than diploid oysters (19 grams TW). However, survival of diploid (92%) and triploid (94%) oysters was similar. Regardless of ploidy type, oysters in bags supported with square floats attached to the top of bags were larger and heavier (2N, 49 mm SH, 23 g TW; 3N, 59 mm SH, 40 g TW) than oysters in bags supported by square floats on the sides of bags (2N, 46 mm SH, 17 g TW; 3N, 54 mm SH, 32 g TW). Oysters in bags supported by bullet floats (2N, 46 mm SH, 17 g TW; 3N, 56 mm SH, 32 g

TW) were similar to those oysters in bags supported by square floats on the sides of bags. Survival of triploid oysters (91-94%) was similar across float designs, whereas the lowest and highest survivals obtained in this sample period were of diploid oysters in bags supported by square floats placed on top of bags (88%) and placed on sides of bags (97%). Antifouling coatings did not influence oyster production at this time.

In September, shell height of triploid oysters (70 mm) continued to exceed that of diploid oysters (53 mm). Likewise, total weight of triploid oysters (88 g) was greater than and doubled that of diploid oysters (40 g). As in the first sample period, average survival was similar in diploid (82%) and triploid oysters (82%). However, mortalities of 9-10% occurred between the first and second sample periods. With respect to float design, both diploid and triploid oysters in bags supported with square floats attached to the top of bags were slightly larger than oysters in bags supported by square floats on the sides of bags and oysters in bags supported by bullet floats. Weight of oysters was variable across float design treatments. The heaviest oysters (3N, 94 g) and lightest oysters (2N, 31 g) were in bags supported with square floats attached to the sides of the bags. Survival of oysters was similar across float design treatments. Interestingly, triploid oysters cultured in bags without an antifouling coating were larger (72 mm SH) and heavier (91 g TW), than those cultured in bags with antifouling coatings (coating A, 68 mm SH, 79 g TW; coating B, 66 mm SH, 75 g TW); survival of triploid oysters did not seem to be affected by coating treatments.

The replicated growout trials were concluded after six months (a total of eight months in the field), and the two ploidy types were compared. Measurements were averaged by ploidy type across the three gear types. Triploid shell height averaged 77 mm, which was significantly higher (P=0.0003) than diploid oysters (56 mm). Total weights were also significantly different (P=0.0001) with triploids averaging 123 grams and diploids averaging 55 grams. Likewise, wet meat weights differed (P=0.0002) with triploids averaging 8.7 grams and diploids averaging 5.0 grams. Further, dry meat weights differed (P=0.0001) with triploids averaging 1.97 grams and diploids averaging 1.25 grams. Despite the better growth observed in the triploids, condition index was significantly higher (P=0.02) in diploid oysters (2N, 9.3; 3N, 7.8). Although meats of diploid oysters weighed less than triploids, their shells also weighed much less than triploid shells. External appearances were distinctive between ploidy types with diploid shells exhibiting a much smoother, worn appearance. It is possible that the smaller size of the diploid oysters made them more susceptible to wave action, thus the associated shell shaping. Survival was similar between ploidy types, averaging 77% among triploids and 74% among diploid oysters.

No significant differences were observed in growth or survival among triploid oysters cultured using the three float designs. However, float design did have a significant effect (P=0.0002) on biofouling accumulation of the gear. Bags supported with square floats attached to the top of the bag had the most fouling at an average of 17.9 lbs. The same floats attached to the sides of the bags resulted in minimal fouling accumulation, averaging only 1.3 lbs. These results were surprising as the square floats placed on top of the bags aided in supporting bags and oysters out of the water during weekly aerial drying, whereas bags supported with square floats on the sides did not during weekly flipping. Given the culture conditions, the control method of flipping bags with floats attached to the sides of the bags was adequate in reducing fouling on the gear (but not necessary on the oysters), which reduced labor input in half as the bags did not need to be flipped back after the drying period. Bags supported with the smaller bullet floats placed on the sides of the bag resulted in an intermediate level of biofouling (7.3 lbs).

Application of either biocide-free antifouling coating (A or B) did not significantly affect oyster production. When applied to bags with square floats attached to the tops of bags, antifouling coatings did appear to reduce biofouling. Bags treated with Coating B had 8.6 lbs of biofouling and those treated with coating A had 14.1 lbs, whereas untreated bags had 17.9 lbs of biofouling. However, these differences were not statistically significant. Altering gear design, specifically attaching square floats to the sides of bags, was more effective in reducing biofouling. Uncoated bags with this float placement had significantly less (P=0.0007) biofouling weight (1.3 lbs) than those bags with square floats placed on top of the bags. Biofouling organisms consisted primarily of barnacles, hooked mussels, macroalgae, and some oyster spat.

During this trial, the predominant fouling organism on oysters was barnacles. To quantify biofouling, oysters were weighed before and after removal of all fouling organisms from the shells to determine percentage of fouling weight (fouling weight/shell weight). Neither ploidy, float design, nor antifouling coating affected the amount of biofouling determined using this method. Among treatments, biofouling weight ranged from 67-105% of shell weight. Shell shape was also considered and shell measurements were used to calculate fan ratio and cup ratio. Fan ratios were similar for both ploidy types (2N, 0.66; 3N, 0.64) as were cup ratios (2N, 0.38; 3N, 0.36) and were within or exceeded the favorable ranges.

The results of both UF field trials were compared to examine seasonal effects on oyster production. Growth and survival of both ploidy types were higher during the first trial (August 2016-April 2017) when compared to the second trial (April-November 2017). Data loggers were used to measure water temperature in culture bags during both trials. Temperatures were lower during the first trial (avg 68°F) than the second trial (avg 83°F) with temperatures periodically exceeding 90°F in the second trial. It is possible these summer temperatures were stressful, which could inhibit growth and increase mortalities.

<u>OBJECTIVE 2. Assess the health of diploid and triploid cultured oysters harvested during summer and winter produced under various conditions as outlined in the first objective.</u>

Dermo is an oyster disease caused by the protozoan parasite, *Perkinsus marinus*. The parasite occurs in oysters located along the east coast of the US and throughout the Gulf of Mexico and high prevalence and intensity are associated with mass mortality. High temperatures and salinities are associated with increased parasite prevalence and intensity. Dermo disease impacts are described in three ways: prevalence, infection intensity, and weighted prevalence. The standard method for identification of the parasite is culture in Ray's fluid thioglycollate medium (FTM). After five to seven days of culture of affected tissues in FTM, trophozoites enlarge and become hypnospores that are readily seen after staining with Lugol's iodine. Infection intensity is scored using a 0-to-5 Mackin scale, where 0 is no infection and 5 is an infection in which the oyster tissue is riddled with hyponospores. Prevalence is defined as the percentage of infected individuals at a site and may range from o to 100%. Infection intensity (II) is calculated as the sum of infection rankings on a 0-5 Mackin scale divided by the number of infected oysters. Weighted prevalence (WP), or mean abundance, is the infection intensity multiplied by the prevalence and indicates the relative severity of Dermo infection in a population. A WP of 1.5 indicates that disease-related mortalities are occurring in a population. A weighted prevalence of 2.0 in a population constitutes an intense epidemic with and more than half of the population in advanced stages of the disease and all individuals likely infected.

Grower Trials 1, July 2016 – March 2017

Diploid and triploid oyster seed spawned in spring (April 2016) were distributed to participating growers in four west coast locations (Charlotte Harbor, Charlotte County; Cedar Key, Levy County; Alligator Harbor, Franklin County; Oyster Bay, Wakulla County) during July/August (2016). Eight months later, oysters were harvested in March 2017. At which time, samples (N=10) from three replicate bags or baskets were obtained from seven growers for assessment of internal and external parasites and pests.

<u>Gear Type</u>: The impact of both on-bottom and off-bottom gear types was compared. On-bottom culture participants (N=2) used cages. Off-bottom culture participants (N=5) used two gear types - adjustable long lines (N=2) or floating bags (N=3). There was no differences in Dermo prevalence or intensity between on-bottom culture (18%) and off-bottom culture systems (17%). There was a significant difference ($P<0.0001$) between gear types with floating bags having significantly less Dermo prevalence (2%) than longlines (36%) or bottom cages (18%). Infection intensity was also significantly lower ($P=0.025$) with floating bags (0.12) than with longlines (0.88) or bottom cages (0.84). Weighted prevalence was likewise significantly lower ($P=0.001$) with floating bags (0.03) than longlines (0.45) or bottom cages (0.17). No significant differences were seen with respect to ploidy between gear types. Diploids had higher prevalence, but similar intensity with long line gear, while triploids had higher prevalence but lower intensity with on-bottom cages. Dermo was only found in triploids when floating bags were used.

Location: The location of participating growers included Alligator Harbor (N=2), Oyster Bay (N=2), Cedar Key (N=2) and Charlotte Harbor (N=1). There was a significant difference (P=0.0012) in Dermo prevalence between locations with Oyster Bay having the highest prevalence (36%) and Alligator Harbor the lowest (3%). Infection intensities were highest at Oyster Bay (0.84) and lowest at Alligator Harbor (0.18), but differences were not significant. Weighted prevalence showed the same trend. No significant differences were seen with respect to ploidy between locations. Diploids had higher prevalence at Oyster Bay and Charlotte Harbor, while triploids had higher prevalence at Alligator Harbor and Cedar Key. Higher infection intensities were seen in diploids at Charlotte Harbor, triploids at Cedar Key and Alligator Harbor, with no differences at Oyster Bay.

Salinity: Oysters were cultured in higher salinity waters (>25 ppt) at Alligator Harbor, Charlotte Harbor and one of the Cedar Key sites than at Oyster Bay (20-25 ppt) and the second Cedar Key site. Dermo prevalence was significantly lower (P<0.0001) at the high salinity sites (4%) than at sites with medium salinity (33%). Infection intensity (P=0.0008) and weighted prevalence (P=0.0002) were likewise significantly different between the two salinities, with lower infection intensities and weighted prevalence at the high salinity sites. Prevalence and intensity were similar between diploids and triploids.

Summary: For oysters planted in the late summer and cultured mostly over the colder months (referred to as “winter trials”), Dermo prevalence and intensity was similar between on- and off-bottom culture methods, but significantly different between gear types, with long lines having higher prevalence and intensity. Location and salinity likewise significantly impacted Dermo prevalence and intensity. The Oyster Bay sites had the highest Dermo prevalence and intensity and Alligator Harbor sites the lowest. Higher salinity (>25 ppt) sites had lower Dermo prevalence and intensity. No significant differences were seen with respect to ploidy.

Grower Trials 2, March 2017 – November 2017

The second phase of the demonstration project evaluated the performance of diploid and triploid oysters planted in early spring (March 2017), as opposed to the first phase in which oysters were planted in late summer. The majority of the culture period in this phase occurred during the warmer months (referred to as “summer” trials). Diploid and triploid seed were distributed to eight growers in four counties with a grower participating in Pine Island (Lee County). About eight months later, oysters were harvested in November. At which time, samples (N=10) from three replicate bags or baskets were obtained from seven growers for assessment of internal and external parasites and pests.

Gear Type: The impact of both on-bottom and off-bottom gear types was compared. On-bottom culture participants (N=1) used cages. Off-bottom culture participants (N=6) used three gear types – adjustable long lines (N=2), floating bags (N=3) or floating cages (N=1). Dermo prevalence was significantly less (P<0.0001) with on-bottom culture systems (0%) than with off-bottom culture systems (28%). There was a significant difference (P<0.0001) between gear types with floating cages (58%) having a higher prevalence of Dermo than long lines (43%), floating bags (11%) or bottom cages (0%). There was no significant difference in ploidy between on-bottom and off-bottom culture methods. Dermo was more prevalent in diploids with both long line (63%) and floating cages (69%) than in triploids (30%, 48%), but the difference was significant (P=0.0007) only with long lines. There was a significant difference in dermo intensity (P<0.0001 II, P<0.0001 WP) between on-bottom and off-bottom culture systems, and gear types (P=0.016 II, P<0.0001 WP). Off-bottom culture had higher infection intensity (0.87) and weighted prevalence (0.33) than on-bottom culture (0 II, 0 WP). Long lines had the highest infection intensity (1.19), while floating cages had highest weighted prevalence (0.54); however, differences were not significant between these two gear types. There was no significant difference with respect to ploidy. Infection intensity was higher in triploids with floating cages and floating bags, but similar to diploids with long lines and bottom cages. Weighted prevalence was higher in diploids with floating cages and long lines and lower with floating bags.

Location: The location of participating growers included Alligator Harbor (N=1), Oyster Bay (N=3), Cedar Key (N=2) and Pine Island (N=1). There was a significant difference between locations (P<0.0001) with highest Dermo prevalence at Oyster Bay (50%), and lowest dermo prevalence at

Alligator Harbor (3%) and Pine Island (0%). In Oyster Bay, prevalence was significantly higher (P=0.002) in diploids (67% versus 38%), but not at Cedar Key (18% triploids, 10% diploids). There was no prevalence difference seen between diploids and triploids at Alligator Harbor or Pine Island. There was a significant difference in both infection intensity (P=0.007) and weighted prevalence (P=0.0003), with Oyster Bay and Cedar Key having the highest infection intensities (0.92) and Alligator Harbor and Pine Island the lowest (0.17, 0). Oyster Bay, however, had a higher weighed prevalence (0.42) than Cedar Key (0.2). Although not significant, triploids had higher infection intensities (1.2) at Oyster Bay and Cedar Key than diploids (1.0, 0.67). The only significant difference with respect to intensity was seen at Oyster Bay, where diploids (0.68) had a significantly higher weighted prevalence (P=0.014) than triploids (0.39).

Salinity: Oysters were cultured in higher salinity waters (>25 ppt) at Alligator Harbor, Cedar Key and Pine Island, than at Oyster Bay (20-25 ppt). Dermo prevalence was significantly lower (P<0.0001) at the high salinity sites (8%) than at Oyster Bay sites (50%). There was a significant difference between triploids and diploids at medium salinity sites (P=0.002), but not at high salinity sites. Infection intensities (P=0.03) and weighted prevalence (P<0.0001) were significantly lower at the higher salinity sites (0.5 II, 0.10 WP) than at Oyster Bay (1.1 II, 0.52 WP) sites.

Summary: For oysters cultured over the warmer months, Dermo was less prevalent and intense in bottom culture than off-bottom culture, with floating cages and long lines having the highest prevalence and intensity. Location and salinity also impacted Dermo prevalence and intensity. The Oyster Bay sites had the highest Dermo prevalence and intensity. Higher salinity sites had lower Dermo prevalence and intensity. There were no differences with respect to ploidy and culture methods or gear types, but differences were seen with respect to location and salinity with diploids having significantly higher Dermo prevalence and intensity at the medium salinity Oyster Bay sites.

Dermo Summary Trials 1 & 2: Prevalence appeared to be higher in the second set of trials ("summer" reared oysters). However, that seemed to be driven more by location/salinity than gear type. It is hard to compare bottom cages with regards to seasonal impact. Although it appears that Dermo is less prevalent in the "summer" reared oysters, the opposite effect was seen with bottom cages. However, it should be noted that the one grower who did participate pulled his oysters a few months earlier than the other participants, which likely impacted results. Prevalence was higher in oyster grown over the summer compared to oysters grown over the winter for long line culture; however, growers that used long lines were situated in the same location and experienced the same salinity. Oysters from off-bottom cages (not in trial 1) and long lines in the second trial likewise were situated in the same location and experienced the same salinity. Therefore, it appears that the over-riding factor was environmental in nature. Impact on ploidy was inconclusive. Prevalence and intensity were generally similar, except for with long line culture where diploids were more negatively affected. Again, this seems to be as much a location factor as due to gear type as this was seen with both off-bottom long line and cages in the second set of trials. Temperature is notably more of a factor in Dermo proliferation than salinity. Interestingly although salinity had an impact in Dermo in "winter" reared oysters, no differences were seen with respect to ploidy, while differences were seen in "summer" reared oysters with a higher prevalence and intensity seen in diploids at Oyster Bay where salinity was lower (20 ppt).

Oyster pests can potentially impact oyster health as well as decrease market value. Oysters were examined for the presence of the polychaete worm (*Polydora websteri*), and the boring sponge (*Cliona* spp.). *P. websteri* burrows into oyster shells and egests muddy wastes causing irritation. The oyster secretes new shell material over the burrows forming unsightly mud blisters. Infestations can result in distorted shaped shells, weakened shells and can cause physiological stress. It has been suggested that off-bottom culture might reduce mudworm infestations. Low temperature and hypo- or hyper-salinity have been shown to reduce *P. websteri* infestations. *Cliona* spp. also burrows into oyster shells weakening them and making the oyster more susceptible predation. Sponge prevalence is associated with increased salinities (>15 ppt) and dipping oysters in brine solution is one method of decreasing infestation.

Grower Trials 1, July 2016 – March 2017

Gear type - Boring sponge: Although prevalence was higher with bottom culture (19%) than with off-bottom culture (6%), the difference was not significant. No significant differences were seen between gear types with long lines (4%) and floating bags (7%) having similar prevalence. No significant differences were seen with respect to ploidy although slightly more triploid oysters were affected by boring sponge, regardless of gear type.

Gear type - Mud blisters: Gear type had no significance with respect to mud blisters, and prevalence was similar between on-bottom (9%) and off-bottom (12%) culture. Long lines had a higher prevalence (15%) of mud blisters than did floating bags (9%), but differences were not significant. In contrast to boring sponge, mud blisters were more prevalent in diploids regardless of gear type though prevalence was not significant.

Location - Boring sponge: Location significantly affected boring sponge prevalence ($P<0.0001$) with highest prevalence at Charlotte Harbor (38%) compared to the other three locations (4-7%). A higher percentage of triploids were affected by boring sponge than diploids at all locations, but the difference was not significant.

Location - Mud blisters: Location did not influence mud blister prevalence. Cedar Key locations had the lowest prevalence (3%) while prevalence in other locations was similar (13-15%). Mud blisters were more prevalent in diploids than in triploids at all locations, but the difference was not significant.

Salinity - Boring sponge: Locations with higher salinity had a significantly higher ($P=0.013$) prevalence (15%) of boring sponge compared to sites with medium salinity (3%). Triploids tended to be more negatively impacted (11.5%) by boring sponge than diploids (6.5%) although differences were not significant.

Salinity - Mud blisters: Salinity did not impact mud blister prevalence. Prevalence was similar at both high (11%) and medium (12%) salinities. Diploids had a higher percentage (15.5%) of mud blisters than triploids (7.5%), but differences were not significant.

Trial 1 Summary: Boring sponge prevalence was higher with on-bottom culture at Charlotte Harbor and at sites with higher salinity. Triploids were more likely to be affected by boring sponge than diploids. Mud blisters prevalence was similar between on- and off-bottom culture, although more prevalent with long lines. Neither site nor location impacted mud blister prevalence. Diploids were more likely to be affected by mud blisters than triploids.

Grower Trials 2, March 2017 – November 2017

Gear type - Boring sponge: Oysters cultured on-bottom had higher (46%) prevalence compared to those cultured off-bottom (23%), but the difference was not significant. Triploids cultured on-bottom had a higher prevalence (60%) than those cultured off-bottom (25%), but differences were not significant and diploid prevalence was similar. Although floating cages had a higher prevalence (46%) than other gear types, the difference was not significant ($P=0.057$). Triploids had a higher percentage of boring sponge than diploids in both floating cages and bottom cages, but not in floating bags or long lines; however, differences were not significant.

Gear type - Mud blisters: Oysters cultured on-bottom had significantly lower ($P<0.0001$) prevalence of mud blisters (6%) than those cultured off bottom (54%). Diploids had higher prevalence than triploids, but differences were not significant. Significant differences (P<0.0001) were seen between gear types, with floating cages having highest prevalence (100%). No mud blisters were found on oysters cultured on long lines. No difference was seen with regards to ploidy.

Location - Boring sponge: Prevalence was significantly different (P=0.043) between locations and was highest in Alligator Harbor (52%) and lowest in Cedar Key (8%). Differences were seen with regards to location when triploids were compared (P=0.016) but not diploids (P=0.82). Prevalence was higher for triploids than diploids at all locations except Cedar Key, but differences were not significant.

Location - Mud blisters: Prevalence between sites was significantly different (P=0.0003). Mud blister prevalence was the opposite of boring sponge prevalence, with lowest prevalence at Pine Island (6%) and highest prevalence at Cedar Key (82%). Prevalence was higher in diploids at all sites except Cedar Key, but differences were not significant.

Salinity - Boring sponge: No difference in prevalence was seen between sites with medium (24%) and high (28%) salinity. Prevalence was higher in triploids than in diploids, but differences were not significant.

Salinity - Mud blisters: No difference in prevalence was seen between sites with medium (46%) and high (49%) salinity. Prevalence was higher in diploids, but differences were not significant.

Trial 2 Summary: Boring sponge prevalence was impacted by gear type and location but not salinity. Prevalence was higher in this trial ("summer" reared) than in the first trial ("winter" reared) but similar to that trial triploids were more negatively impacted than diploids. Mud blister prevalence was also impacted by gear type and location but not salinity. Similar to boring sponge, prevalence was higher for oysters in this trial than in the first trial. Diploids were more likely to be negatively impacted by mud blisters than triploids.

Pest Summary Trials 1 & 2: Boring sponge was more prevalent in "summer" cultured oysters regardless of gear type. Boring sponge prevalence increased in all locations and at all salinities in these oysters. Triploids were more negatively impacted than diploids, particularly those cultured in cages, whether on- or off-bottom. Mud blisters were also more prevalent in "summer" cultured oysters. Salinity did not appear to have an impact on prevalence whereas gear type and location did. This was especially notable in Cedar Key where both growers used floating bags. Diploids appeared to be more negatively impacted than triploids in "winter" cultured oysters, but this did not carry over to "summer" cultured oysters. With gear and in areas where prevalence was heavy, no ploidy differences were seen.

UF Field Trials 1, August 2016 - April 2017

Replicated field trials were conducted to document the effects of ploidy and stocking density on oyster production. About 2700 oysters of both ploidy types were stocked in October 2016 into eighteen 12 mm mesh Vexar bags at 125, 150, and 175 oysters/bag (6 bags/density/ploidy type) for a total of 36 bags. At harvest in April 2017, five replicates per treatment group (each replicate consisting of five oysters) were examined for oyster diseases. Prevalence and intensity of *Perkinsus marinus* (Dermo) and prevalence of pests (*Cliona* spp, *Polydora websteri*) were compared in diploid and triploid oysters cultured at three different densities.

Dermo: There was a significant difference (P<0.0001) seen in Dermo prevalence between oysters cultured at high (60%) versus medium (2.5%) and low (2%) densities. This was seen with both diploids (P=0.0172) and triploids (P=0.0006). However, there was no difference with regards to ploidy. Prevalence in diploids grown at high densities was 55%, and in triploids was 65%. Infection intensity (0.77) was also significantly higher (P=0.008) when oysters were cultured at high densities compared to oysters cultured at medium (0.0625) and low (0.0125) densities. This difference was due to the higher infection intensity (1.03) seen with triploids grown at high densities, which was twice that of diploids (0.525), however differences were not significant. Similarly, weighted prevalence was also significantly higher (P=0.0003) in oysters cultured at high densities. Only one bag of triploid oysters at the medium density and one bag diploid oysters at the low density contained Dermo.

Boring Sponge: There was no significant difference with respect to boring sponge prevalence in oysters cultured at high (16%), medium (3%) and low (23%) densities. Although diploid oysters had a higher prevalence (18%) than triploid oysters (6%), significant differences were only seen at high densities (P=0.0114). At high densities, 32% of diploid oysters showed evidence of boring sponge and 0% of triploids were affected by boring sponge.

Mud Blisters: There was no significant difference with respect to mud blister prevalence in oysters cultured at high (20%), medium (17%) and low (11%) densities. However, diploids had an overall significantly higher prevalence of mud blisters (P=0.005). However, the difference was only significant in oysters cultured at medium densities (P=0.008). At medium densities, 22% of diploid oysters showed evidence of mud blisters and 0% of triploids were affected.

UF Field Trials 2, April - November 2017

Oysters of each ploidy type were reared by the UF project team at their experimental lease off Cedar Key. In April 2017, diploid and triploid oysters were stocked into 9 mm mesh Vexar bags for field nursing. On June 7, juveniles were restocked into forty 14 mm mesh Vexar bags at a density of 150 per bag. To further examine the effects of float type (square versus cylindrical or bullet) and float placement (square floats on sides of bags versus tops of bags) on oyster production and fouling control, replicated (n=4) trials were conducted. In addition, two commercially available, biocide-free, antifouling coatings (A, B) were applied to four replicate bags supported by the square floats and stocked with triploid oysters. The replicated growout trials were concluded after six months (a total of eight months in the field) in November. At which time, samples from four replicates per treatment group were collected, each replicate consisting of five oysters, for examination of oyster diseases.

Dermo: No significant difference was seen between float types (P=0.06) with regards to Dermo prevalence, although oysters in bags supported by square floats placed on the sides of the bags had the least Dermo (5%), while oysters in bags supported by bullet floats had the most (25%), regardless of ploidy. A ploidy difference was seen with oysters in bags with square floats attached on top (P=0.032) with triploids having greater Dermo prevalence (35%) compared to diploids (0%), but not with other float types. Likewise, there was no significant difference seen between float types with regard to intensity, although it was higher in oysters cultured in bags supported by bullet floats (0.53II,0.25WP) and lowest in oysters cultured in bags supported by square floats on the sides of the bags (0.19II, 0.04WP). A ploidy difference in oysters was seen in bags supported by square floats placed on tops of bags (P=0.037) with regards to infection intensity. Triploids exhibiting higher II (0.45) than diploids (0), but not with other float types. No significant ploidy differences were seen with weighted prevalence, even with oysters in bags supported by floats place don tops of the bags. Although triploid oysters in non-coated bags tended to have higher Dermo (27%) prevalence than in coated bags (A-10%, B-12%), the difference was not significant. Likewise, triploid oysters in non-coated bags tended to have higher intensities (0.41II, 0.21 WP) than in coated bags (0.125-0.38II, 0.05-0.13WP).

Boring Sponge: There was no significant difference in boring sponge prevalence between float types or coating treatments. No differences were seen with respect to ploidy. Prevalence was generally low (0-8%) in all treatments.

Mud Blisters: There was no significant difference in mud blister prevalence between float types or coated and non-coated groups. No differences were seen with respect to ploidy. Prevalence was high (55-86%) in all treatments.

Summary of UF Trials 1 and 2

A seasonal comparison of both trials in which diploid and triploid oysters were grown at medium density shows that Dermo and mud blister prevalence increased in oysters reared mostly over the summer months (Trial 2) while boring sponge prevalence was similar in both trials. Although it is not known what densities growers conducted their trials, it is interesting to compare the UF trials to Cedar Key growers, who participated in the grower trials. A comparison of UF trial results to oyster growers located in Cedar Key showed that Dermo prevalence was lower at UF (2%) than at growers' locations (13%) for oysters reared primarily over the winter months (Trial 1), but similar (14%, 16%) in Trial 2, during which oysters were reared mostly over the summer months. Boring sponge prevalence was similarly low in both UF trials (6%, 4%) and for Cedar Key growers (4%, 8%) regardless of culture seasons. Trials conducted by UF and with Cedar Key growers both showed a marked increase in mud blister prevalence in oysters reared over the summer in Trial 2 (17% to 68% UF; 3% to 85% CK growers). The similarities are particularly striking when one notes that one of the Cedar Key growers in that trial used floating bags (also used by UF) while the other used bottom cages. Once again this points to location and environmental conditions as being more a driving factor than gear type in oyster health assessment.

<u>OBJECTIVE 3. Evaluate the quality and sensory attributes of cultured triploid oysters compared to diploid oysters harvested seasonally.</u>

Cultured oysters produced by the UF project team from both sets trials were subjected to consumer evaluation for acceptability. In May 2017, diploid and triploid oysters were delivered under refrigerated conditions to the UF/IFAS Food Science and Human Nutrition Department. All state regulations pertaining to harvesting and handling of oysters were followed. Triploid oysters averaged 81 mm SH and 120 grams TW, while the diploids averaged 76 mm and 74 grams. Although the two oyster types were similar in size, the triploids were obviously heavier than the diploids.

Panelists (n=75) were recruited to participate in the sensory evaluation; all indicated in a pre-screening test that they were consumers of raw oysters. Oysters were shucked and two oysters from each ploidy type were provided to panelists in individual booths. Panelists rated appearance, texture, flavor, and overall acceptability on a 9-point hedonic scale (1=dislike extremely, 5=neither like nor dislike, 9=like extremely). The triploid oysters rated higher in overall acceptability and appearance than the diploid oysters (P=0.10); however, there were no statistical differences in flavor and texture between the two oyster samples. Likewise, there was no difference between the two samples for preference. There was a trend that the triploid oysters were favored as the averages for all attributes were higher than those for the diploid oysters. Diploid oysters are typically developing gametes or spawning during this time of year, which may affect meat quality, but this was not reflected in the sensory results.

A portion of the oysters harvested by the UF project team in November 2017 were placed back on the lease and held until February 2018; at which time, they were subjected to a second consumer evaluation for determination of seasonal differences. Panelists (n=94) were recruited to participate in the sensory evaluation. The same protocols were followed as in the first evaluation. Again, triploid oysters rated higher in overall appearance than the diploid oysters (P=0.10). However, for overall liking, flavor, texture, and preference, there were no differences between the two ploidy types. One might expect these results as diploid oysters are full of glycogen (“fat) during the winter months, allowing sensory characteristics and consumer preference to be similar to triploid oysters.

Oysters typically acquire their taste from their growing environment. Consumers can expect variations in sensory attributes that influence preferences, value, and reputation. These attributes and acceptability can make the difference in market success of Florida cultured oysters.

<u>OBJECTIVE 4. Examine the basic financial characteristics of the triploid oyster culture process by addressing production characteristics and comparing those with diploid oyster production, focusing on providing insight into risks and profitability that accompany potential respective costs of investment and operation.</u>

Oyster farming, like any aquaculture or agriculture operation, has major risk sectors that are beyond the grower’s control. Normal risk is related to occurrences, such as inclement weather, predation, fouling, or other variables, that typically can occur during production. Oyster mortality and costs associated with normal risk are factored into the farm’s potential profitability and generally are considered acceptable at a certain level. Environmental risk in oyster farming can include events such as hurricanes or extreme changes in water quality, particularly salinity due to drought, excessive rainfall, or flooding. As environmental risk increases, potential economic impacts include increases in oyster mortality, labor time, and repair or loss of gear and equipment. Economic risk is associated with factors that directly affect the farmer's revenue, such as varying market prices or changes in input costs. The financial characteristics of off-bottom oyster culture along the west coast of Florida were determined in order to assess the effects of risk on farm profitability.

Information on the costs of operating an oyster farm was obtained several ways. Ten farmers in four counties were provided each with 5,000 triploid and diploid oyster seed and a logbook to document time (labor) associated with activities in culturing these oysters, such as planting, fouling control, transferring bags, culling/sorting, and harvesting (see Objective 1). Three of these farmers were interviewed to provide greater background farm information to estimate the effects various risks may have on their businesses, including potential increases in oyster mortality, labor, and capital costs. In addition, field trials replicating industry practices using the same seed amount as the participating growers were conducted at the UF experimental lease off Cedar Key. On the experimental farm, labor time associated with gear set-up and all culture activities was recorded, as well as seasonal oyster growth and mortality rates documented. Labor, operational and capital costs were derived from these data collection methods and applied to a hypothetical oyster farm.

Using this information, an oyster farm budget model was simulated over a five-year planning period incorporating effects of normal risk. The following assumptions were made pertaining to a small-scale farm operation. In year one, the farm was assumed to be new with 10,000 oyster seed planted. Each year the number of seed planted increased reaching 250,000 in year five. Triploid seed were purchased at a cost of $25 per 1000. The culture system used on the hypothetical farm was floating bags in three mesh sizes. The average culture unit cost was estimated at $33 for the bag, floats, PVC pipe, rope, anchors, clips, and cable ties. Based on growers' input, the final stocking density was set at 250 oysters per bag. An average survival of 80% from plant to harvest was used, and of those, 90% were marketable. The model also included the purchase of a boat and motor at a cost of $32,000 over a 10-year loan at 7% interest. In year 1, the owner/operator did not need part-time labor. As oyster plantings increased annually, average labor increased from 9 hours in year 2 to 126 hours in year 5 at an hourly wage of $12. The values for each assumption used in the farm budget model, as well as the average, minimum, and maximum values, affected by the normal risk variable over a 5-year planning period were calculated.

Estimates of oyster production and profitability for a range of environmental and economic risk scenarios were then generated. To do so, long-term databases available from federal and state agencies were used to predict environmental risks for oyster growing areas on the west coast of Florida. The probability of a major storm affecting an area was determined by reviewing historical data from the National Oceanic and Atmospheric Administration (NOAA). Monthly salinity data since the 1980s were obtained for water quality stations located adjacent to oyster farms monitored by the Florida Department of Agriculture and Consumer Services (FDACS). Historical market prices of cultured oysters were not available for Florida, so prices collected by the Virginia Institute of Marine Science from 2005 to 2016 for Virginia oysters were used as a proxy. The probability of an environmental risk event affecting a given Florida west county could then be calculated, whereas the probability of market risk was the same for each county.

The environmental and economic data were analyzed under six risk scenarios to generate financial predictions for oyster growing areas in Alligator Harbor (Franklin County), Cedar Key (Levy County), Oyster Bay (Wakulla County), and Pensacola Bay (Escambia County). In Alligator Harbor and Cedar Key, farms are also suitable for culturing hard clams. Thus, start-up costs were not applied in the financial model for these two areas. Further, a partial budgeting model for boat payments and fuel costs was used assuming that 50% of the costs were associated with the clam operation. Risks were independently observed within each scenario. In addition, one scenario considered all environmental and economic risks. For all scenarios, normal risk was applied. The analysis utilized Simetar software, which is a Microsoft Excel Add-on, providing the ability to model and simulate an industry with numerous risk variables. Simetar was used to simulate each oyster farm risk variable by county for each scenario. The program randomly selected a value from each potential impact for each risk scenario 1,000 times by growing location. This provided outputs of a distribution of profitability estimates for each county on an annual basis for a five-year period. The expected net returns were the average farm profitability before taxes and owner/operator salary was taken based on the simulation results derived from each considered risk distribution.

Scenario 1: Baseline (Normal) In this scenario, no abnormal environmental or economic risks were applied, allowing for comparison between other scenarios. There were no differences in the expected net returns between the two growing areas located in Alligator Harbor and Cedar Key. These farms sustained the same normal risk, and partial budgeting was also applied. The expected net return was estimated at $66,500 with a 50% range of $62,500 to $70,250 after five years of operation. The growing areas in Oyster Bay and Pensacola Bay had the same return of $63,200, which was lower than the other two counties as clam farming was not considered.

Scenario 2: Hurricanes/Tropical Storms From 2015 to 2017 (the study period), oyster farms experienced three events within 60 miles of their locations. The occurrence of similar events was determined by searching the NOAA database. In turn, the probability of one or two of these events affecting an oyster growing location in any given year could be calculated. There is the potential for other hurricanes with varying strengths and directions to impact oyster farms. However, only those storms that occurred during the study were considered. Both Alligator Harbor and Cedar Key had the highest probability (19%) of a category 1 hurricane or tropical storm impacting the growing area on an annual basis. This additional risk lowered the expected net return by year 5 to $62,440. The higher probability of a storm striking an oyster farm affects labor time and costs due to storm preparation time, clean-up, and repairs, as well as potential increased oyster mortalities. The probability of one of these storms striking Oyster Bay or Pensacola Bay was reduced to 16% and 11%, respectively. However, Oyster Bay had the highest probability (3%) of two major storms striking within a year. The expected net return for farms located in Oyster Bay was reduced to $59,015 at the end of five years, which incorporated the additional risk from multiple storms affecting an oyster farm in a given year.

Scenario 3: Low Salinities Salinity levels that fall below 10 ppt over two consecutive months were considered in this scenario. Salinity can be affected by excessive rainfall and runoff and/or increased flows from a freshwater source, such as a river. The timing and duration of low salinities may dramatically affect growth and mortality of different oyster sizes. The FDACS water quality database was searched to determine the probability of low salinities occurring at monitoring stations located adjacent to oyster growing areas. The highest probability of a low salinity event was determined to be 50% for Pensacola Bay at any given year. During this study, an actual event did occur at an oyster farm in Escambia County with resulting mortalities. This risk probability resulted in the expected net value by year 5 to decrease to $56,290 with a range of values of $54,960 to $65,770 occurring 50% of the time. However, 2% of the time the possibility existed that the farm operation could lose money in year 5 due to this risk. In Oyster Bay, there was an 11% probability of a low salinity event occurring in a production year, which would reduce the expected net value to $61,850 in year 5. At the other oyster growing locations, probabilities of a low salinity event occurring were zero.

Scenario 4: High Salinities Extreme high salinity events can affect oyster production due to mortalities from increased predation, pests, and diseases. Events which resulted in salinities above 35 ppt for a period of two consecutive months were considered. High salinities usually develop as a result of prolonged droughts. Again, the FDACS water quality database was used to determine probabilities of this type of event occurring at any of the oyster growing locations. The highest probability (30%) of high salinities affecting oyster crops was in Alligator Harbor, which decreased the expected net value to $61,850, a loss of about $5,000 in revenue from the baseline scenario. Probabilities of a high salinity event occurring at the other growing locations were less than 1%.

Scenario 5: Market Prices This scenario did not include environmental risk only normal and market risks based on the variation of prices from projected increases over a five-year period. Historical market prices of Virginia cultured oysters from 2005 through 2016 were used to create a linear regression model for predicting the expected market price per year from 2018 to 2022. The average market price at the end of year five was projected to be $0.48. This created an expected net return of $66,490 for farms located in Alligator Harbor and Cedar Key, and $63,220 for farms located in Oyster Bay and Pensacola Bay.

Scenario 6: All Risks All normal, environmental and market risks within an oyster growing area were considered in this scenario. Since it is possible that any one of these risks could occur within a given year, this is a more realistic scenario. For each county, the expected average and potential ranges of annual net returns, or profitability to the owner/operator were generated. Given all the risks, potential net returns at year 5 for growers in Alligator Harbor was $57,959. There was a 50% probability of earning an income between $47,930 and $68,730. However, there was a 5% probability that net returns could exceed $77,890 or fall below $27,850. The expected net return of $62,440 in year 5 for farms in Cedar Key was higher than Alligator Harbor. The higher return is based on a lower probability of all risks affecting farms in that county (23%) compared to Franklin County (51%). The highest probability of all risks affecting oyster farms occurred in Pensacola Bay (61%), which resulted in an expected net return of $53,850 in year 5. This was a difference of about $10,000 in revenue from the county's baseline projection. The average net return in year 5 for farms in Oyster Bay was $58,300 based on a 32% probability that any risk could affect farms in that county.

In summary, there was an upward trend in expected net returns for years 1 through 5 as seed planting increased and expected quantity of oysters sold into the market increased. However, the risk associated with net returns increased. All counties had a 100% probability of negative net returns at the end of year 1 as revenue did not exceed start-up costs. However, there was a greater than 98% chance that farms in all counties were profitable by the end of year 5 when all risk variables were considered. Based on results from the simulation, oyster farms in Levy County had the greatest expected net income each year, followed by farms in Franklin, Wakulla, and Escambia Counties. The analysis assumed that Levy County oyster growers shared expenses with a clam culture operation reducing total costs associated with the oyster operation, and this diversification largely caused the higher expected net returns. Escambia County had the lowest expected net returns and greatest variance among counties due to the high probability of a sustained low salinity event, which had the greatest effect on mortality of the environmental risks considered.

Problems Encountered

Funding for this project was released in September 2015. There was not adequate time to plan and conduct a fall spawn. Thus, the first spawn was conducted in April 2016; it was anticipated that these oysters would be harvested by growers and UF during March through May 2017, approximately one year from spawning. To evaluate seasonal effects, diploid and triploid oyster seed (2-4 mm shell height) were procured in the fall of 2016 and were distributed to growers in February 2017. A no-cost extension was requested in order to complete the project objectives.

After meandering around the Gulf of Mexico as a tropical depression, Hurricane Hermine gathered steam and headed straight for the Big Bend coast on September 2, 2016. The first hurricane to hit Florida in 11 years, Hermine did most of its damage with a 7-9 foot tidal surge and waves driven by 70-80 knot winds. In anticipation of the hurricane, several growers in Cedar Key participating in the project were able to retrieve their oyster gear and hold juvenile oysters in either nursery systems or protected waters. One grower left his bottom cages on his lease and all were lost. Another grower lost the majority of his seed as the bags temporarily tied up to his floating dock were damaged. In the Panhandle, little damage was reported by growers in Oyster Bay (Wakulla County) using the adjustable longline system or by growers in Alligator Harbor (Franklin County) using the floating bag system. A little over a month later, Hurricane Matthew, a powerful category 5 storm, took aim at Florida, but its path kept it off the west coast. However, several growers moved their oyster bags in anticipation of potential impacts. One grower kept his oysters on his boat for over a day and noted that about half of the diploids died as a result.

RESEARCH PROJECT ACCOMPLISHMENTS

Sensory Characteristics of Cultured Oysters

Relevance

Oysters, both wild harvested and cultured, typically acquire their taste from their environment. Off-bottom oyster culture is a new industry on the west coast of Florida. Prior to this project, the sensory characteristics of diploid and triploid oysters cultured under subtropical conditions had not yet been evaluated. Oyster growers and shellfish wholesalers need to be able to distinguish their product by location, season, or growout process, which can aid in developing appellations that may stimulate and attract consumer interest.

Response

Cultured oysters produced by the UF project team in Cedar Key were harvested seasonally and subjected to consumer evaluation for acceptability. Diploid and triploid oysters were delivered to the UF/IFAS Food Science and Human Nutrition Department in May 2017 and, again, in February 2018. Panelists were recruited to participate in a sensory evaluation. Panelists rated appearance, texture, flavor, overall acceptability, and preferences on a 9-point hedonic scale.

Results

Triploid oysters rated higher in overall acceptability and appearance than the diploid oysters in both evaluations; however, there were no statistical differences in flavor, texture, or preference between the two oyster samples. There was a trend that the triploid oysters were favored as the averages for all attributes were higher than those for the diploid oysters. Consumers can expect variations in sensory attributes that influence preferences, value, and reputation. These attributes and acceptability can make the difference in market success of Florida cultured oysters.

Health Assessment of Cultured Oysters

Relevance

Off-bottom oyster culture is a new industry on the west coast of Florida. Prior to this study, the health of diploid and triploid oysters produced under various culture conditions had not yet been documented. Site conditions that may affect health status include those that vary with respect to environmental conditions (e.g., temperature, salinity), culture methods (e.g., off-bottom, on-bottom), planting and harvest times. Understanding how these factors affect the prevalence and intensity of internal and external parasites can lead to management practices to improve survival and quality of oysters cultured in subtropical conditions.

Response

Diploid and triploid oyster seed were distributed to participating growers in four west coast locations (Charlotte Harbor, Charlotte County; Cedar Key, Levy County; Alligator Harbor, Franklin County; Oyster Bay, Wakulla County) during July/August (2016). Eight months later, oysters were harvested in March 2017. At which time, samples from three replicate bags or baskets obtained from seven growers were shipped to the FAU-HBOI Aquatic Animal Health Lab for examination of internal parasites, in particular *Perkinsus marinus* (Dermo), as well as associated oyster pests, such as *Cliona* sp. (boring sponge) and mud blisters (*Polydora websteri)*. To assess seasonal differences, samples were also obtained from participants, who harvested diploid and triploid oysters in November (2017); these oysters were planted in March and were cultured primarily over the warmer summer months.

Results

This was the first health assessment conducted for oyster growers on Florida's west coast. The assessment included growers in four counties and examined the impact of gear, location, ploidy and season on prevalence and intensity of Dermo and external pests. Location and season appeared to have more of an impact on both parasites and pests than gear type, with increased prevalence of both in oysters cultured during the warmer summer months. Oysters in Oyster Bay (Wakulla County) tended to be more significantly impacted by Dermo than other locations regardless of season, while oysters in southwest Florida tended to be more significantly impacted by boring sponge than other locations regardless of season. There was no clear-cut health impact with regards to ploidy. Although triploids fared better overall with respect to Dermo, differences were not significant due to the low number of participants and replicates. Interestingly, triploids appeared to be more adversely affected by boring sponge while diploids appeared to be more adversely affected by mud blisters. Findings offer insight as to how various culture practices and farm site locations may affect overall health of diploid and triploid oysters.

Oyster FARM (Financial And Risk Model) Calculator

Relevance

Risk in oyster farming, as in all agriculture and aquaculture production, is any event that a grower cannot control but affects the grower's operation. However, various types of risk can be forecasted based on historical events and trends. To understand the risks associated with off-bottom oyster aquaculture, information on the financial characteristics of operating an oyster farm on the west coast of Florida was documented in order to assess the effects of risk on farm profitability.

Response

Information on the costs of operating an oyster farm was obtained from ten growers participating in the demonstration project. Three of these farmers were interviewed to provide greater background information to estimate the effects various risks may have on their farms. Using this information, an oyster farm budget model was simulated over a five-year planning period incorporating effects of normal risk. Estimates of oyster production and profitability for a range of environmental and economic risk scenarios were then generated. To do so, long-term databases available from federal and state agencies were used to predict risk probabilities for four oyster growing areas. The environmental and economic data were analyzed under six risk scenarios to generate financial predictions. The expected net returns, or farm profitability, was based on the simulation results derived from each considered risk distribution.

Results

A financial and risk assessment tool, *Oyster Financial And Risk Model Calculator*, was developed using these findings. The Microsoft Excel spreadsheet allows oyster growers to generate predictive costs and revenues for their individual farms and investment situation. The Calculator can be accessed at the website, *Online Resource Guide for Shellfish Aquaculture*, http://shellfish.ifas.ufl.edu (click on Oyster Culture Resources). Financial findings are illustrated using a fan graph and stoplight chart, which determines the probability of a farm's net income falling between a range of values determined by the grower. Net income is simulated based on the farm inputs provided and probability of risk events by county. Growers can change the lower and upper bound after the simulation has occurred to determine the probability of achieving the stated net returns in the presence of all environmental and market risks. The Calculator allows oyster growers to better understand how risk affects profitability.

Evaluation of Triploidy by Oyster Growers along Florida's West Coast

Relevance

Natural triploids of the Eastern oyster have been available only since the early 2000s elsewhere in the United States, and not until 2014 in Florida. The techniques should work well for Florida oyster growers and provide the benefit of limiting natural reproduction. Yet, site-to-site variability in production benefits of triploidy to oyster aquaculture, as well as gear type-interaction, have been noted in other coastal states. There was a need to fully understand whether the application of triploidy in the development of an emergent oyster culture industry on Florida's west coast would result in increased oyster growth and survival and therefore beneficial economic gains. Florida represents the southernmost limit of natural distribution for *Crassostrea virginica* in the United States. In contrast to other states, Florida's subtropical water temperatures result in a prolonged spawning season for oysters, with intermittent spawning and redevelopment throughout the year. The effects of triploidy on the performance of cultured oysters produced by tetraploid technology under these conditions had not yet been documented.

Response
To document the production performance of diploid and triploid oysters under commercial conditions and quantify effects of different culture techniques (on-bottom versus off-bottom), salinity regimes (medium, high), and seasonal harvests (summer versus winter), a large-scale demonstration and evaluation was conducted at various oyster farms on Florida's west coast. Single-set diploid and triploid seed were produced and provided to ten growers in the following counties: Charlotte (first trial), Lee (second trial), Levy, Wakulla, and Franklin. In the first set of growers' trials, seed were planted during July/August 2016 and harvested eight months later in March 2017. Since most of the culture period was over cooler months, these were referred to as "winter" trials. In the second set of growers' trials, seed were planted in March 2017 and, again, harvested eight months later in November. These were referred to as "summer" trials, since most of the culture period was over the warmer months. At the harvest of both trials, samples from three replicate bags or baskets (depending on gear type used) per ploidy type were collected by the UF project team to document growth, survival, condition index and biofouling.
Results
In both sets of trials, triploid oysters were larger, and meats weighed more than diploid oysters at each growing location except for oysters cultured in Wakulla County, regardless of gear type used; however, growth was similar for both ploidy types at these farm sites. In the winter trials, survival of both ploidy types was commercially acceptable, ranging from 89-99%, while survival in the summer trials were overall lower (57-92%), regardless of ploidy, farm location or gear type. Differences in fouling was observed by gear type and farm location, but not ploidy type. In both set of trails, oyster grown in Levy County exhibited the most biofouling; however, fouling was significantly lower for triploid oysters than for diploids. Fouling on oysters in other counties were overall lower for each growing season but similar for both ploidy types. Differences in oyster production between the two seasonal trials were apparent at all farm locations. Overall, the demonstration project established multiple sites at commercial farms in four counties. Potential advantages of triploidy, such as faster growth, shorter crop times and less biofouling, were documented. The project also provided information on oysters cultured using different gear types at different farm locations with varying environmental regimes.

Evaluation of Management Practices for Off-Bottom Oyster Culture
Relevance
Oyster production is an important cultural and socio-economic component of many coastal communities throughout the southern United States, yet the region (with the notable exception of Virginia) has only recently embraced the production of hatchery-reared, single-set oysters using off-bottom techniques to target the premium, half-shell market. Beginning in 2014, a small number of commercial off-bottom oyster farms were created in several Florida west coast counties. These oyster farms have helped establish the potential and economic feasibility for off-bottom oyster farming in the state. This new industry, however, relies on consistent production of top-quality oysters to maintain market demand. While methods have been adopted that produce oysters, there was a pressing need for the area's new oyster farmers to understand the effects of different culture decisions on the production of their shellfish products. By honing husbandry techniques, oyster growers may optimize production, thereby increasing profit margins while improving product consistency.
Response
To develop management strategies specific to off-bottom oyster production on Florida's west coast, replicated field trials with robust experimental designs were conducted by the University of Florida team at an experimental lease located within a commercial aquaculture use zone off Cedar Key (Levy County). Strategies included evaluation of stocking densities in floating bag systems, float design (type and placement), and biocide-free, antifouling coatings for controlling the development of biofouling organisms that negatively affect the performance of oysters grown using off-bottom methods. As in the growers' trials, both diploid and triploid oysters were cultured over two growing seasons to document the effects of ploidy and seasonal harvests. In both sets of trials, growth and survival were determined at eight-week intervals. At harvest, oysters from each replicate bag per treatment were measured for shell

height, length, and width and weighed (total, wet meat). Pictures of meats and shells were taken to document appearance. In addition, samples per bag were collected for health assessment, ploidy verification, and analysis for dry meat weight, condition index, and biofouling. Survival was also determined per replicate bag. These data were compiled and statistically analyzed to determine the significance of ploidy and management practices on oyster production.

Results

In the first trial, stocking density had no significant effects on survival or growth of diploid or triploid oysters cultured at 125, 150, and 175/bag; however, growth at each density was consistently higher in the triploids than diploids, whereas survival of each density treatment was higher for diploids. These results suggested that densities as high as 175/bag over a "winter" growing period can result in viable production and may be increased. In the second trial, triploid oysters were significantly larger and heavier than diploids, but no significant differences were observed in growth or survival among triploid oysters cultured using three float designs. Float type and placement did affect biofouling accumulation of the gear with the control method of flipping bags with floats attached to the sides of bags more effective. Application of coatings did not affect oyster production but did appear to reduce biofouling; however, altering gear design was more effective. Results of both trials were examined for seasonal effects. Growth and survival of both ploidy types were higher during the "winter" trial when compared to the second "summer" trial. Results were shared with industry, allowing new and perspective oyster growers to make informed decisions about production strategies and management.

Information Exchange for Oyster Growers along Florida's West Coast

Relevance

Off-bottom oyster culture is a new industry on the west coast of Florida. Oyster farms are located primarily in isolated rural coastal communities, which limits exchange of information among growers located in the various production areas. To assist in the sustainable development of an off-bottom oyster aquaculture industry, ongoing information exchange on results of applied research and demonstration efforts was facilitated.

Response

To inform new and prospective oyster growers about results of this demonstration project, including the growers' trials, UF replicated trials on selected management practices, consumer evaluations, health monitoring, and risk assessment, a news blog was developed on the *Online Resource Guide for Florida Shellfish Aquaculture* website, http://shellfish.ifas.ufl.edu/oyster-demo-project/. Fifteen articles were posted providing detailed continuous updates about the various facets of this project. Grower workshops were conduction in three locations with one workshop broadcasted live via the Zoom app. A video of the workshop along with all presentations were posted to the news blog. In addition, information about the project was presented at annual Oyster South Symposiums to inform growers in the southeastern U.S. Presentations were also given at a national shellfish conference. Fact sheets on the application of triploidy and financial risk assessment of oyster culture were developed and were also made available at the website. Further, an Excel-based tool, Oyster Financial And Risk Model Calculator, was developed allowing oyster growers to generate predictive costs and revenues for their individual farms and investment situation.

Results

Through presentations at national conferences, regional symposiums, and local workshops, along with news blog posts, fact sheets, and published abstracts, over 600 people, consisting of industry members, general public and academia, were informed about the results of this integrated applied research and demonstration project. Evaluations at each of the local workshops were conducted to better understand if the project results and presentations were helpful to the attendees. Of the 50 or more who attended the workshops and completed a survey, average responses of 4.2 or greater, based on a scale of 1 ("strongly disagree") to 5 ("strongly agree"), were obtained for all questions, indicating that the workshops were worth the time to attend, new and helpful information was presented, presentations were informative, and sufficient information was presented to better understand the use of triploidy in oyster culture. There were many responses to the question of what was liked best about the workshops; these included profitability analysis, valuable information for newcomers, fouling issues, differences in gear types and locations, learning about biofouling and economics, information, being able to ask questions and answers to questions.

Project Students Supported

Name: Russel Dame
Institution: UF IFAS
Department: Food and Resource Economics Department
Major Professor: Kelly Grogan
Status: Masters
Anticipated Graduation: May 2018
Thesis Title: A Risk Assessment on Triploid Oysters along the West Coast of Florida
Students email: rstyjj@ufl.edu
Graduate student assistantship from UF/IFAS Nature Coast Biological Station, 2016-18, $43,365

Name: Nick Brandimarte
Institution: Florida Atlantic University
Department: Harbor Branch Oceanographic Institute
Major Professor: Susan Laramore
Status: Masters
Anticipated Graduation:
Thesis Title:
Students email:

New Extramural Funding Based on the Project

Title Project: Initiation of a Program to Produce Florida Tetraploid Founder Stocks for the Gulf Oyster Industry
Funding Amount: $73,308
Project Completion Date: 31 December 2017
Project Sponsor: Gulf States Marine Fisheries Commission

Title Project: Tetraploid technology for *Crassostrea virginica* in Florida: A public-private partnership to create west coast Florida-specific tetraploid brood stock
Funding Amount: $56,753
Project Completion Date: 30 June 2016
Project Sponsor: Florida Department of Agriculture and Consumer Services, Florida Aquaculture Project

OUTREACH EVENTS AND ACTIVITIES

These informational resources have allowed new and perspective oyster growers to make informed decisions about production strategies and management. Through presentations at national conferences, regional symposium, and local workshops, along with published abstracts, newsletter articles and website posts, have informed over 600 people, consisting of industry members, general public and academia, about the results of this integrated applied research and demonstration project. Further, this information allows new and perspective oyster growers to make informed decisions about production strategies and management practices.

- An additional topic page linked to the *Online Resource Guide for Florida Shellfish Aquaculture* website, http://shellfish.ifas.ufl.edu/oyster-demo-project/, was designed and produced. A news blog with 17 articles and project updates, as well as other informational sources, can be accessed. The topic page and news blog serve as web-based tools for growers, wholesalers, and individuals.

- A poster, which was updated with current information about the project, was presented at the following symposiums: OysterSouth in Auburn, AL; Big Bend Science in Cedar Key; and, Florida Sea Grant Coastal in Gainesville, FL.
- The following activities were conducted to provide results of both phases of the demonstration project, including the growers' trials, UF replicated trials on selected management practices, consumer evaluations, animal health monitoring, and risk assessment to a variety of audiences:
    - A PowerPoint presentation providing information on the growers and UF trials was presented at a regional symposium, Oyster South.
    - Three abstracts were submitted and a PowerPoint presentation on the UF trials and two poster presentations on the growers' trials and health monitoring were presented at a national shellfish conference.
    - Workshops, in which five presentations were given by the various project team members, were held on Florida's west coast in three locations (Franklin, Dixie, and Levy Counties) for growers to learn about project results.
    - Evaluations of each of the local workshops were conducted to better understand if the project results and presentations were helpful to the attendees. Following is a summary of results from each workshop:
        - Of the 25 participants, who attended the workshop in Carrabelle, Franklin County, 19 completed the survey.Overall, average responses of 4.6 or greater, based on a scale of 1 ("strongly disagree") to 5 ("strongly agree"), were obtained for all questions, indicating that the workshop was worth the time to attend, new and helpful information was presented, presentations were informative, and sufficient information was presented to better understand the use of triploidy in oyster culture. There were many responses to the question of what was liked best about the workshop; these included profitability analysis, valuable information for newcomers, fouling issues, differences in gear types and locations.
        - Of the 20 participants, who attended the workshop in Cedar Key, Levy County (13 attended remotely), four completed the survey. Average responses of 4.25 or greater, based on the same scale, were obtained for all but one question. A score of 3.75 was obtained on the questions asked if presentation were informative. Data on gear was the response on what was liked best about the workshop.
        - Of the 5 new oyster growers, who attended the workshop in Cross City, Dixie County, four completed the survey. Average responses were 5, based on the same scale, were obtained for all questions. Responses on what was liked best about the workshop were learning about biofouling and economics, information, being able to ask questions, and answers to questions.
    - The Cedar Key workshop was broadcasted live via the Zoom app. A video of the workshop, along with all PowerPoint presentations were posted on the news blog at the *Online Resource Guide for Florida Shellfish Aquaculture* website, http://shellfish.ifas.ufl.edu/news/workshops-application-of-triploidy-to-oyster-culture/.
- An EDIS manuscript on the financial risk assessment of oyster culture was developed and submitted for review.
- An excel-based tool, *Oyster Financial And Risk Model Calculator*, was developed and is being beta tested.
- An article was published in the Florida Aquaculture Association's quarterly newsletter.

PRESENTATIONS

Sturmer, L. (presenter), C. Cyr, and R. Markham. 2017. Application of triploidy to an emergent oyster culture industry on Florida's west coast. Oyster South Symposium, Auburn, Alabama, 27-28 January 2017.

Sturmer, L. (presenter), C. Cyr, and R. Markham. 2017. Application of triploidy to an emergent oyster culture industry on Florida's west coast. Florida Sea Grant Coastal Science Symposium, Gainesville, Florida, 26 September 2017.

Sturmer, L.(presenter), C. Cyr, R. Markham, H. Yang, and S. Laramore. 2017. Application of triploidy to an emergent oyster culture industry on Florida's west coast. Big Bend Science Symposium, Cedar Key, Florida, 1-3 February 2017.

Dame, R. (presenter), K. Grogan, C. Adams, and L. Sturmer. 2017. Risk assessment on triploid eastern oysters along the Nature Coast of Florida. Big Bend Science Symposium, Cedar Key, Florida, 1-3 February 2017.

Carter, C. (presenter), Sturmer, L., Markham, R. and Sims, C. 2018. Application of triploidy to an emergent culture industry on Florida's west coast. 110th annual meeting of the National Shellfisheries Association, Seattle, Washington, 18-22 March 2018.

Sturmer, L. (presenter) and Cyr, C. 2018. Application of triploidy to an emergent culture industry on Florida's west coast. Oyster South Symposium, Charleston, South Carolina, February 2018.

Sturmer, L. (presenter). 2018. Clamelot: the story and significance of shellfish farming in Cedar Key, Florida. South Atlantic Regional Sea Grant Meeting, Cedar Key, Florida, 31 July – 1 August 2018.

Dame, R. (presenter), Adams, C. Sturmer, L. 2019. Financial risk analysis of off-bottom oyster aquaculture along the west coast of Florida. Aquaculture 2019, New Orleans, Louisiana, 7-11 March 2019.

Simon, N. (presenter), Sturmer, L., and Markham, R. 2019. Gear type comparison for off-bottom oyster aquaculture in Florida, USA. Aquaculture 2019, New Orleans, Louisiana, 7-11 March 2019.

Dame, R. and Sturmer, L. (presenter). 2019. Financial risk in oyster culture on the Florida west coast. Oyster South Symposium, Auburn, Alabama, 22-23 February 2019.

PUBLICATIONS

Sturmer, L., C. Cyr, R. Markham, H. Yang, and S. Laramore. 2017. Application of triploidy to an emergent oyster culture industry on Florida's west coast. Big Bend Science Symposium Book of Abstracts:26.

Sturmer, L., C. Cyr, and R. Markham. 2017. Application of triploidy to an emergent oyster culture industry on Florida's west coast. Florida Sea Grant Coastal Science Symposium Book of Abstracts:42.

Dame, R., K. Grogan, C. Adams, and L. Sturmer. 2017. Risk assessment on triploid eastern oysters along the Nature Coast of Florida. Big Bend Science Symposium Book of Abstracts:33.

Sturmer, L. (presenter). 2017. Oyster Aquaculture on Florida's West Coast. Oyster South Symposium, Auburn, Alabama, 27-28 January 2017.

Sturmer, L., Cyr, C., Markham, R., Brandimarte, N., and Laramore, S. 2018. Application of triploidy to am emergent oyster culture industry on the Florida west coast: results of grower trails. Program and Abstracts of the 110th Annual Meeting of the National Shellfisheries Association: 169.

Cyr, C., Sturmer, L., Markham, R., and Sims, C. 2018. Application of triploidy to am emergent oyster culture industry on the Florida west coast: results of grower trails. Program and Abstracts of the 110th Annual Meeting of the National Shellfisheries Association: 57.

Brandimarte, N., Sturmer, L., and Larmore, S. 2018. Health evaluation of triploid and diploid eastern oysters under various culture conditions. Program and Abstracts of the 110th Annual Meeting of the National Shellfisheries Association: 152.

Sturmer, L. 2018. Florida Aquaculture Association Newsletter. Spring issue: 5-6.
Yang, H. Simon, N, and Sturmer, L. 2018. Production and Performance of Triploid Oysters for Aquaculture. UF/IFAS Extension, EDIS fact sheet FA208. 9 pp. http://edis.ifas.ufl.edu/fa208
Dame, R., Sturmer, L., Adams, C., Weldon, R., and Grogan, K. 2018. Financial Risk in Off-bottom Oyster Culture along Florida's West Coast. UF/IFAS EDIS manuscript submitted to Food and Resource Economics Department (in review).
Dame, R., Adams, C. Sturmer, L. 2019. Financial risk analysis of off-bottom oyster aquaculture along the west coast of Florida. Program and Abstracts of the Aquaculture 2019 Triennial Conference

COMMUNICATIONS PRODUCTS

- Three PowerPoint slideshows on application of triploidy to an emergent oyster culture industry on Florida's west coast – results of UF replicated field trials, results of grower's trials, and overall project results.
    - Sturmer, L., Cyr, C., Markham, L., Yang, H., and Laramore, S. 2017. Application of triploidy to an emergent oyster culture industry on Florida's west coast
    - Sturmer, L. 2017. Status of oyster culture on Florida's west coast
    - R. Dame, K. Grogan, C. Adams, and L. Sturmer. 2017 Risk Assessment on Triploid Eastern Oysters along the Nature Coast of Florida
- Two posters on application of triploidy to an emergent oyster culture industry on Florida's west coast – results of grower's trials and health evaluation of oysters.
- Oyster Farming Demonstration Webpages - News blog and information on oyster culture demonstration project, posted to http://shellfish.ifas.ufl.edu/oyster-demo-project.

TOOLS/WEBSITE

- Dame, R. and Sturmer, L. 2018. Oyster FARM (Financial And Risk Model) Calculator. An Excel spreadsheet which allows oyster growers to generate predictive costs and revenues for their individual farms and investment situation. Net income is simulated based on the farm inputs provided and probability of risk events by county. http://shellfish.ifas.ufl.edu/news/oyster-farm-financial-and-risk-model-calculator/
- Video of oyster culture workshop posted to news blog on website, http://shellfish.ifas.ufl.edu/news/workshops-application-of-triploidy-to-oyster-culture/.
- Web topic page linked to the *Online Resource Guide for Florida Shellfish Aquaculture* website, http://shellfish.ifas.ufl.edu/oyster-demo-project/, with a news blog and three new articles posted.

PROGRAMMATIC OUTCOMES AND IMPACTS

The primary goal of this project was to allow for large-scale demonstration and evaluation of an oyster breeding process to local conditions on Florida's west coast by new and perspective oyster growers. Adoption of tetraploid technology to produce natural triploid oysters may increase the prospects for commercially viable, small-scale oyster farms, create jobs, and provide a sustainable domestic oyster supply. Additionally, the project provided the impetus needed for the Florida clam farming industry to advance the production and distribution of another molluscan shellfish species, thus revitalizing an industry that is currently based exclusively on one species, the hard clam *Mercenaria mercenaria*.

Natural triploids of the Eastern oyster *Crassostrea virginica* have been available only for the past 15 years elsewhere in the U.S., and not until 2013 in Florida. The techniques should work well for Florida oyster growers and provide the benefit of limiting natural reproduction. Overall, the demonstration project established multiple sites at commercial shellfish aquaculture leases in four counties on Florida's west coast. This project documented potential advantages of triploidy, such as faster growth, shorter crop times, less risk of disease, and availability of year-round quality oyster meat. The project also provided information on diploid and triploid oysters grown using different gear types at different farm locations with varying environmental regimes.

The project brought together faculty from two universities (UF and FAU), a growers' association, and industry partners to provide the necessary infrastructure via a public-private partnership to commercialize the Eastern oyster through large-scale demonstration and evaluation of a breeding technique to local conditions on Florida's west coast. The production (growth, survival) performance of diploid and triploid oysters was documented under commercial conditions and effects of different culture techniques (on-bottom versus off-bottom), salinity regimes (medium, high), and seasonal harvests (summer versus winter) were quantified. The health of diploid and triploid cultured oysters harvested during summer and winter produced under various conditions was assessed. The quality and sensory attributes of cultured triploid oysters compared to diploid oysters harvested from different growing areas and harvest seasons were evaluated. The basic financial characteristics of the triploid oyster culture process were examined by addressing production characteristics and comparing those with diploid oyster production, focusing on providing insight into break-even production levels and market prices that will accompany potential respective costs of investment and operation.

Impact: Application of Triploidy to an Emergent Oyster Aquaculture Industry in Florida

Relevance

Aquaculture of alternative bivalve species is an opportunity for the shellfish aquaculture industry in Florida to diversity and expand, providing an economic boost both to growers as well as supporting industries. Revitalization of an industry based on one bivalve species may be achieved by facilitating technology transfer to the various industry sectors of the established hard clam industry and geographically diversifying culture areas. In 2014, renewed interest in intensive oyster culture was brought about by an oyster fishery failure and removal of a regulatory restriction limiting water column use on shellfish aquaculture leases. As a result, many clam growers and others began investing in oyster culture. Though newly established and relatively small-scale, these farms helped to establish the potential of off-bottom oyster culture on Florida's west coast.

Response

To address increased interest in oyster culture on Florida's west coast, a large-scale demonstration project was conducted, which allowed for evaluation of an oyster breeding process (triploidy) to local conditions by new oyster growers. Both diploid and triploid oyster seed were provided to 10 growers in four coastal counties during 2016-17 to document production performance under commercial conditions. Several culture systems used by growers also provided for evaluation of site and gear interaction. In addition, replicated field trials were conducted by the University of Florida project team to quantify effects of stocking densities, gear modifications, and seasonal harvests. The integrated project also allowed for evaluation of sensory attributes of cultured triploid oysters compared to diploid oysters, health assessment of oysters harvested seasonally and produced under various conditions, and examination of financial risk characteristics focusing on environmental and market risks.

Results

Project results were presented to growers and others through workshops, symposiums, demonstrations, and a news blog. Several tools (e.g., Oyster FARM Calculator) and fact sheets were developed from project findings. Resulting educational materials and outreach efforts reached over 100 people, allowing shellfish growers and others to make informed decisions about the prospects of off-bottom oyster culture and the application of triploidy. Development of alternative culture species and farming technology represents an important gain over the present reliance of a single species crop and should have a positive impact by increasing productivity, creating jobs, and providing a healthful local seafood supply. In summary, through ongoing information exchange, demonstrations, and applied research efforts, the sustainable development of a multi-species shellfish aquaculture industry may become a reality.