FEASIBILITY OF HARD CLAM, *MERCENARIA MERCENARIA*, CULTURE IN

GRAND BAY, ALABAMA.

Except where reference is made to the work of others, the work described in this thesis is my own or was done in collaboration with my advisory committee. This thesis does not include proprietary or classified information.

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FEASIBILITY OF HARD CLAM, *MERCENARIA MERCENARIA*, CULTURE IN GRAND BAY, ALABAMA.

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THESIS ABSTRACT

FEASIBILITY OF HARD CLAM, *MERCENARIA MERCENARIA*, CULTURE IN GRAND BAY, ALABAMA.

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The northern hard clam, *Mercenaria mercenaria*, has proven to be a profitable aquaculture species along the Eastern seaboard as well as the west coast of Florida. The potential for *M. mercenaria* aquaculture in the North Central Gulf of Mexico has not been thoroughly investigated. The biological and production feasibility of clam culture was analyzed using two bivalve grow-out systems in the coastal waters of Grand Bay, Alabama. Clams were stocked into mesh bags in a belt system placed on the bottom and in bags suspended from an adjustable long-line system. The stocking densities used for each treatment were 188/m^2, 375/m^2, 750/m^2, and 938/m^2. Each bag density was replicated three times for the adjustable long-line system and five times for the belt system on bottom. The mesh bags placed on the bottom allowed clams to burrow into the sediment as they would naturally. The clams placed in the adjustable long-line system were suspended mid-water column approximately 0.5 m from the bottom. The belt system and the long-line system were stocked with clams of an average size of 15.3 mm and 17.9 mm, respectively. Over a period of seven months clams in the belt system showed a 25.0% increase in size with 42.6% survival compared to only a 14.6% increase in size for clams in the long-line system with a 24.3% survival. While the belt system performed significantly better than the adjustable long-line system, both system's growth and survival were lower when compared to published data for similar periods of time along the east coast of Georgia, Florida and South Carolina. The low growth rate and survival may be due to excessive stresses from tunicate and polycheate bio-fouling on the belt system and siphon nipping by fish and crabs in the suspended long-line system.

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I. INTRODUCTION

Northern hard clams, *Mercenaria mercenaria,* have a wide distribution ranging from the Gulf of St. Lawrence down the east coast of the United States and extending into the Gulf of Mexico as far south as the Texas coastline (Abbott 1974; Manzi and Castagna 1989). Dillon and Manzi (1989) found a similar distribution, however limited the northern extent of range to New Brunswick, Canada.

The closely related southern hard clam, *M. campechiensis*, naturally occurs from Mexico, throughout the Gulf of Mexico, and up the East Coast to New Jersey (Eversole 1987; Abbott 1974; Manzi and Castagna 1989). The southern hard clam is typically found in deeper and higher salinity water than the northern hard clam (Eversole 1987). Limited hybridization occurs in areas where the two species of hard clams overlap producing a recognized sub species, *M. mercenaria texanus* (Menzel 1964; Menzel and Menzel 1965).

Hard clams are marine ectotherms of the family Veneridae. Water temperature limits the range of northern hard clam distribution at the latitudinal edges of its distribution in North America. Williams (1970) determined the minimum lethal temperature for adult hard clams to be -6°C and found that adult hard clams survived freezing of up to 64% of the body tissue. Henderson (1929) found 45°C to be the maximum lethal temperature for an adult hard clam. The optimum temperature for hard clam growth is 20°C with growth occurring in a range of 10°C to 30°C (Pratt and

Campbell 1956; Loosanoff 1939; Menzel 1963, 1964; Van Winkle et al. 1976).

The optimum salinity range for hard clams ranges from 12.8 to 35 ppt (Castagna and Chanley 1973; Menzel 1961; Woodburn 1961; Van Winkle et al. 1976). The survival limitation of hard clams based on salinity is highly variable as the length of exposure is very critical. Burrell (1977) found a mortality of less than 5 percent in clams exposed to salinities lower than 10 ppt for periods of 2 and 3 weeks. The hard clam protects itself from low salinity by closing its shell (Burrel 1977). This behavior makes it difficult to assign the hard clam with a set lethal salinity level. Salinity tolerances, therefore, should be treated more like a range not a specific point.

Hard clams are a protandric species and spawn polymodally from the spring to the fall as long as water temperatures are at or above 20°C (Loosanoff and Davis 1950; Manzi et al. 1985; Eversole 1987). Hard clams become sexually mature at a shell length of 33 mm (Eversole 1987). Bricelj and Malouf (1980) found that clams are sexually mature by two years of age. Gamete maturity is the deciding factor that determines whether an individual clam can be triggered to spawn once conditions are suitable (MacKenzie et al. 2002). The males spawn first, releasing sperm and pheromones which in turn induces surrounding female clams to spawn. A single adult female clam may release up to 16.8 million eggs (Bricelj and Malouf 1980), with fertilization occurring in the water column (Eversole 1987).

Larvae develop through straight-hinged veliger, umboed veliger, and pediveliger stages in 10 days at 30°C (Carriker 1961; Davis and Calabrese 1964; Eversole 1987). Davis (1958) found that larvae developed best at salinities at or above 20 ppt. Lower salinities slow development and death occurs at salinities 17.5 ppt or less. Hard clam larvae typically reach the trochophore stage after a period of 12 hours at a temperature of 22°C (Loosanoff and Davis 1950). Larvae take 24 to 36 hours to reach the straight-hinged veliger stage and reach the umboed veliger stage in six days. The larvae develop a foot by day 10 becoming pediveliger larvae. After transformation to a plantigrade juvenile clam, pedal feeding may be used briefly until gills can be developed for filter feeding (Reid et al. 1992). An adult hard clam may reach up to 40 years of age (Jones et al. 1989).

Adult Clam Behavior

Adult hard clams are capable of filtering on average 7.6 liters of water per hour to obtain single celled phytoplankton for food (Hadley et al. 1997). High levels of suspended sediment can impede the clam's ability to feed. Bricelj et al. (1984) and Pratt and Campbell (1956) found that high levels of suspended sediment decreased hard clams feeding efficiency. With high levels of suspended sediment, the clam produces pseudofeces at a higher rate than normal (Eversole 1987; Pratt and Campbell 1956). Pseudofeces production increases energy costs and hinders feeding, thereby decreasing growth.

Hard clams naturally burrow in bottom sediments. Pratt and Campbell (1956) found that clams would burrow up to 1 cm deep in mud and up to 2 cm in sand. This relationship of burrowing depth to sediment type may be due to the previously mentioned energy loses attributed to higher suspended sediments. As clams burrow deeper, water is filtered closer to the area of mixing at the water-sediment boundary. Hard clams are generally thought of as a sessile organism but are capable of vertical movement at a rate of 44 cm per hour (Kranz 1974).

A number of predators are known to consume hard clams. Hard clam larvae are commonly preyed upon by larval fish and jellyfish (MacKenzie et al. 2002). Birds, fish, and crabs are known to prey on juvenile and larger hard clams. Bird predation is limited to periods of low water. Fish and crabs, however, are able to prey on clams located in deeper water. Clams have demonstrated defensive behaviors in response to predators. Clams were observed to increase their depth in sediment at low tide providing protection from predation by herring gulls (Roberts et al. 1989). Doering (1982) found that clams would burrow deeper and decrease oxygen consumption in response to sea star, *Asterias forbesi,* stimulus. Crabs have been found to be a major predator of hard clams with blue crabs, *Callinectes sapidus,* responsible for most consumption (Menzel and Sims 1964).

Hard Clam Disease

Hard clams have a relatively low risk of disease. Although diseases have devastated northern populations of oysters, these diseases have had no significant impact on clam populations (MacKenzie et al. 2002). Diseases associated with hard clams include, Chlamydia, *Perkinsus sp*., and QPX (quahog parasite unknown) (Sunila 2004). Mortalities from QPX were first noted in the 1960's at New Brunswick, Canada (MacKenzie et al. 2002). Although QPX has not historically had a major impact on adult hard clam populations, occurrence increased starting in the early 1990s (Ford et al. 2002; Smolowitz et al. 1998). Ford et al. (2002) and Sunila (2004) pointed out that QPX, which is caused by a protozoan parasite, is the major disease of concern for hard clams (MacKenzie et al. 2002; Bugge and Allam 2007). Ford et al. (2002) has shown that imported seed clams are more vulnerable than local seed, but that the hatchery is not the likely source of infection. QPX is naturally occurring and widespread, however QPX densities increase with high clam densities, such as a clam farm (Ford et al. 2002; Calvo et al. 2007). Hard clams inability to tightly close its valves was described as a clinical sign of QPX by Smolowitz et al. (1998) with many farmers reporting clams migrating to the sediment surface before death. There have been no documented QPX outbreaks south of Virginia (Ford et al. 2002; Calvo 2007). The mechanism that limits QPX in southern waters has not been determined.

Hard Clam Exploitation

The northern hard clam has a long history of being exploited as food, tools, and currency in the eastern United States. America's relationship with hard clams began hundreds of years ago with Native Americans who first utilized this resource (MacKenzie et al. 2002). Native Americans are credited with initiating clam bakes. They would travel to the coast to celebrate harvests of green corn and would have huge bakes of clams, corn, and seaweed. Native Americans also used clam shells for tools such as hoes, knifes, spoons, and many other useful tools. Native Americans would fashion beads of white and purple out of the clam shells. These beads were referred to as "wampum" and used as currency for trading (MacKenzie et al. 2002).

European settlers interacted with Native Americans and followed their example of use for the hard clam. European settlers traded supplies to Native Americans for strings of wampum (MacKenzie et al 2002). The settlers in turn, used the wampum to trade back to Native Americans in exchange for furs. Wampum was also used among the settlers as a form of currency. The Europeans followed Native Americans' tradition of clam bakes with continued popularity today. Hard clams remain a highly recognized seafood product with over 80 millions dollars of combined sales in 2005 from wild harvest and farmed hard clams in the United States (U.S.D.A. 2005; NMFS 2005) .

Hard clams were initially harvested from the wild before the development of clam aquaculture. The simplest form of harvesting hard clams is probing around in the mud with a person's foot until clams are located and removed by hand (Mackenzie et al. 2002). A simple metal garden rake may also be used to sift through the sediment in search of clams (MacKenzie et al. 2002). Clam rakes were modified to have a collection chamber on the back so that any clams that are dug up are trapped in the chamber (Mackenzie et al. 2002). A hydraulic dredge is often used for a larger scale commercial operation. This dredge is pulled along the bottom, with jets of water that suspend the sediment and clams, allowing for the clams to be caught in a collection chamber (MacKenzie 2002). Harvested clams are sorted into 4 classifications based upon size: the "littleneck" (25 mm shell thickness), the "topneck" (30 mm shell thickness), the "cherrystone" (38 mm shell thickness), and the "chowder" (>38 mm shell thickness) (Hadley et al. 1997).

Aquaculture

While commercial landings of hard clams have decreased, aquaculture production has increased. Commercial harvest of hard clams has decreased from 9,436 metric tons in 1950, to 3,877 metric tons in 2006. Aquaculture production of hard clams has increased from 5,176 metric tons in 1998 (USDA 1998) to 38,635 metric tons in 2005 (USDA 2005).

The simplest form of clam culture consists of stocking a natural bottom with seed clams then covering the area with a layer of mesh to protect the clams from predation

(Fernandez et al. 1999; Grabowski, Powers, and Hooper 2000). However, predators that penetrate the mesh at an early life stage then remain under the mesh are able to prey on the clams once the predator is large enough to consume the clams (Whetstone and Eversole 1978). This extensive method can be inefficient in that all clams may not be found in the sediment at harvest. A more intensive form of culture consists of planting seed clams in sediment filled trays usually constructed of wood covered with protective mesh (Pfeiffer and Rusch 2001; Eldridge 1979; Harding 2007). The use of trays allows the entire crop to be harvested. The tray method requires removal of the trays from the water which can be labor intensive. Another method allowing complete harvest is stocking seed clams in large flat cages that are then partially buried in the sediment (Walker 1983; Walker 1984; Walker and Heffernan 1990). A similar method uses nylon mesh bags (Mojica and Nelson 1993; Grabowski, Powers, and Hooper 2000). The belt-bag system (Vaughan et al. 1988) is the most complex form of clam culture requiring slightly more equipment. This system uses a series of mesh bags attached to a bridle of heavy rope or similar material anchored at both ends. This production system allows total harvest, reduces labor required to retrieve bags, and limits loss due to storms.

The grow-out period for hard clams fluctuates with changes in water quality conditions such as water temperature. Therefore, the location of a production site has a major impact on the length of time required to grow a clam to market size. The smallest size class of clams, known as "littlenecks", receives the highest price, and as a result a shell thickness of 25 mm is the preferred target size. Clams in Cedar Key, Florida, planted at a 4-6 mm size are typically harvested after a 10 to 14 month grow-out period (Adams and Sturmer 2004). Farmers in Sapelo Island, Georgia, start with 9 mm seed clams and have a grow-out period of 15 to 18 months (Hurley and Walker 2000). Progressing further north, the grow-out period for clams in South Carolina is 18 to 30 months for seed planted at 8-10 mm (Hadley et al. 1997), followed by 24 to 28 months for 5 mm seed in Massachusetts (Kraus 1989). These data demonstrate a strong relationship between grow-out period and latitude. Since the Alabama and Mississippi coasts lie between Cedar Key, Florida, and Sapelo Island, Georgia in respect to latitude, it would seem reasonable to expect a similar grow-out period for hard clams. Therefore, the grow-out period would likely be 18 months as a conservative estimate to reach a 25 mm littleneck size for the Alabama and Mississippi coasts. A small-scale production operation could feasibly harvest and market clams, providing income by the second year after stocking. This approximate two-year grow out coupled with the ability to utilize seasonal labor could lead to the creation of new busninesses.

Hard clams have emerged as one of Florida's most important aquaculture species. Hard clams have a relatively short grow-out period, small grow-out size, high market value, and low disease risk, making it a suitable aquaculture species. The culture of hard clams was promoted by the Florida Department of Labor to ease the socio-economic difficulty created from the loss of jobs caused by the closing of the commercial gill-net fishery (Sturmer and Vaughan 1997). The use of monofilament entangling nets was banned in all inshore and nearshore waters of Florida effective July of 1995 (Article X 1995). Many net fishermen unemployed by the net-ban were retrained by the State of Florida Department of Labor to grow hard clams.

The Florida hard clam industry has been a great success and is now the state's most rapidly increasing aquaculture segment (Baker et al. 2005). The farm gate value of farm raised clams in Florida has increased from $3.7 million in 1993, to $15.9 million in 1999 (Phillippakos et al. 2001). In a study by Adams and Van Blokland (1998), a 0.8 hectare grow-out facility planting 600,000 clams per year had the potential to earn $30,000 per year at a market price of $0.10-0.12 cents per clam. With an average price per pound of $0.66/lb (USDA 2005), low costs and high returns have allowed clam aquaculture to be a profitable small-scale business.

Hard clam aquaculture could potentially be utilized to provide the same socio-economic benefits to the Alabama and Mississippi region for the seafood industry which has become economically depressed due to rising fuel costs, hurricane damage, and resource allocation between commercial and recreational fisheries. Currently, there have been no efforts to culture hard clams in the North Central Gulf of Mexico and only a small intermittent recreational harvest (personal communication with Mark Van Hoose of Alabama Marine Resources Division, July 12, 2005).

The belt-bag system (BBS) and the adjustable long-line system (ALS) were evaluated during this study. The BBS is widely used for hard clam production. This technique employs a row of bags attached to a bridle that is anchored at the ends. The BBS is stretched out flat across the bottom and both anchors are placed into the sediment to hold the line in place. The ALS was developed in Australia for oyster production. The ALS is gaining popularity for oyster production, but there is no data available on its use for hard clam production. The ALS method allows for very easy access to the product so that growth, health, and fouling may be closely monitored.

The presented study examines the production feasibility of hard clam, *M. mercenaria*, culture in the coastal waters of the North Central Gulf of Mexico.

Experiments evaluated growth and survival for hard clams in two different bivalve production systems stocked at four different stocking densities. The following hypotheses were tested:

1. H_O: No significant difference in final size exists between clams grown at four stocking densities in the belt-bag system.

   H_A: Final size of clams grown at four stocking densities in the belt-bag system exhibits significant differences.

2. H_O: No significant difference in growth rate exists between clams grown at four stocking densities in the belt-bag system.

   H_A: Growth rate of clams grown at four stocking densities in the belt-bag system exhibits significant differences.

3. H_O: No significant difference in survival exists between clams grown at four stocking densities in the belt-bag system.

   H_A: Survival of clams grown at four stocking densities in the belt-bag system exhibits significant differences.

4. H_O: No significant difference in condition index exists between clams grown at four stocking densities in the belt-bag system.

   H_A: Condition index of clams grown at four stocking densities in the belt-bag system exhibits significant differences.

5. H_O: No significant difference in final size exists between clams grown at four stocking densities in the adjustable long-line system.

   H_A: Final size of clams grown at four stocking densities in the adjustable long-line system exhibits significant differences.

6. H_O: No significant difference in growth rate exists between clams grown at four stocking densities in the adjustable long-line system.

   H_A: Growth rate of clams grown at four stocking densities in the adjustable long-line system exhibits significant differences.

7. H_O: No significant difference in survival exists between clams grown at four stocking densities in the adjustable long-line system.

H_A: Survival of clams grown at four stocking densities in the adjustable long-line system exhibits significant differences.

8. H_O: No significant difference in condition index exists between clams grown at four stocking densities in the adjustable long-line system.

H_A: Condition index of clams grown at four stocking densities in the adjustable long-line system exhibits significant differences.

II. MATERIALS AND METHODS

Field experiments were conducted in Grand Bay, Alabama to investigate the biological and production feasibility of culturing *M. mercenaria* along the North Central Gulf of Mexico (Figure 1). Experiments analyzed four clam stocking densities in each of two bivalve production systems, the belt-bag system (BBS) and the adjustable long-line system (ALS). The production systems were located in Sandy Bay, a small bay on the east side of larger Grand Bay (Figure 1). Permission was obtained from private landowner, Mr. Steve Crocket, for access to the site through his property. As part of the agreement, Auburn University agreed to facilitate the appropriate permitting for the experimental apparatus. Mr. Crockett agreed to allow Auburn University to apply for a riparian lease and an Army Corp of Engineers permit for the construction of the ALS on his property. The BBS was removed once the study was completed.

Permitting

The construction of the ALS in navigable waters required a permit from the Army Corp of Engineers. Application for the permit was made my submitting an Engineer Form 4345 (Appendix 1) obtained from the Army Corp of Engineers. The completed form and diagrams of the planned structures were submitted with the application. The Army Corp of Engineers approved the application and issued a permit for the construction of the ALS through the General National Permit 4 (Appendix 4) on

October 26, 2006. Permitting was expedited through approval under the General National Permit which does not require a public hearing.

The placement of the BBS on the submerged bottom required obtaining a riparian lease from the State of Alabama. A riparian lease, as stated in the Alabama Code, allows landowners of waterfront property, to express possession of oysters on submerged bottom that have been planted provided there are no naturally occurring oysters located there which would be considered a public resource. This area of possession extends 549 meters from the landowner's shore unless circumstances apply where boundaries would intersect with a neighboring landowner's boundary. To establish the bounds of the leased bottom a registered surveyor must survey and mark the boundary with markers spaced no more than 183 meters apart from each (State of Alabama).

Lawler and Company (Mobile, AL) performed a riparian survey of the property. The riparian boundary was marked with 3.05 meter (m) sections of PVC pipe painted fluorescent orange. Alabama Marine Resources Division was presented with a plat of the survey (Appendix 2) accompanied with a written letter of permission (Appendix 3) by the landowner for approval. The riparian lease was executed by the Alabama Marine Resources Division upon certification that the site met the requirements for a riparian lease on June 20, 2007.

Grow-out Systems

<u>Belt-Bag Systems</u>

The BBS consists of a series of mesh bags (Figure 2) filled with clams strung between parallel lines and placed on bottom between two anchoring poles (Figure 3). The mesh bags are allowed to settle into the bottom providing clams with a substrate in which to burrow. The system can be easily removed from the water for maintenance and ease of harvest.

Four BBS units were constructed for the current study. Each BBS consisted of seven black polyethylene bags (Figure 2) strung between two parallel Bayco® monofilament lines (5mm diameter). Mesh bags (9.52 mm square mesh) measured 100 cm in length by 51 cm in width were purchased from Aquatic Ecosystems, Inc. The open ends of each bag were folded over the Bayco® line and secured with a PVC closure (Figure 4) and a cable tie. The PVC closures were constructed by making a single cut down a 45 cm length of 3.81 cm schedule 20 PVC pipe (Figure 4). A triangular notch was cut into the end of each closure to facilitate attachment to the bag and monofilament. PVC risers (Figure 2) were utilized to prevent bags from collapsing and limiting the ability of clams to burrow and orient. The risers were constructed of 5 cm diameter schedule 40 PVC cut to lengths of 5 cm and secured with three 21.6 cm cable ties equidistant on the centerline along the length of each bag (Figure 2). The bags on each end of the belt were left empty to prevent bias from the possibility of end-bags not fully burying.

Spreader bars were placed between each bag to hold the bags in place and keep the Bayco® lines spread apart. The spreader bars were constructed of 86 cm length of 5 cm diameter PVC pipe. A hole was drilled 5 cm from each end of the spreader bar to thread the Bayco® line through. Half hitch knots were used to secure the spreader bars and provide stops to hold the mesh bags in place.

The system was anchored at each end by attaching the Bayco® line to a 3 m long schedule 40 PVC pipe (Figure 3). The pipes were driven into the sediment 152 cm. A 10 mm hole was drilled through each anchor 152 cm from the top end as a point of attachment for the Bayco® line. The two ends of the Bayco® line were secured with two galvanized 0.95 cm cable clamps. A second hole was drilled 35 cm above the attachment point to drain off rain water.

The belt-bag systems were oriented approximately 200 yards from shore in approximately 1 m of water (Figure 5). The BBS was oriented perpendicular to the shoreline (Figure 5). The four belt-bag systems were placed parallel to each other with 2.5 m spacing between belts.

Adjustable Long-line System Construction

The ALS consists of a series of mesh bags suspended mid-water column from a monofilament line anchored to pilings at each end and supported off bottom at regular intervals by support poles (Figure 6). Each bag can easily be removed from the system for maintenance and harvest without disturbing the rest of the line. The ALS can be raised out of the water for short periods of time to desiccate bio-fouling organisms.

The adjustable long-line system (ALS) was constructed by jetting two pilings three meters long, 1.5 m into the sediment and 17.5 m apart (Figure 6). Six 3.05 m sections of schedule 40 PVC pipe were jetted into the sediment in line with the anchor posts to serve as riser posts. Riser clips (Figure 7) were attached to the riser posts with #8 panhead stainless steel screws at 15 cm intervals with the bottom clip 35 cm above the bay bottom. Bayco® monofilament line (5 mm diameter) was tied onto one of the anchor pilings at a height of 0.9 m from the bay bottom. Pre-cut sections of polyethylene dripper tubing (9.5 mm diameter) were threaded onto the Bayco® line. Groups of three 80 cm sections were alternated with one 20 cm section along the length of the line. The 80 cm sections of tubing acted as a sleeve bearing to allow bags attached to the line to rotate. The 20 cm sections of tubing were used as a spacer for clipping the line into the riser clips. The system was designed to hold three bags consecutively between poles. After completing the pattern to the other anchor piling, the line was pulled tight by hand and tied to the opposing anchor piling. Another section of tubing was cut to the necessary length, split lengthwise, and used to cover any Bayco® line that remained exposed between the last riser post and anchor piling. Bags were attached to the long-line by clipping the bags to the line and using a 21.6 cm cable tie to secure the clip to the dripper tubing. The long-line bags (mesh size 11.7 mm) measured 73 cm long by 22 cm wide by 21 cm tall (Figure 8).

The adjustable long-line can be easily raised above the water to control bio-fouling through desiccation of smaller more susceptible fouling organisms. After a brief period of dessication, the line is returned to the water. This dessication of bio-fouling organisms increases the time interval between pressure washing of bags to remove bio-fouling organisms (Rikard 1997).

Clam Sourcing

Approximately 7,000 juvenile northern hard clams, *M. mercenaria*, were purchased from the Bay Shellfish Company located in Terra Ceia, Florida to use in the study. The clams were shipped to the Auburn University Shellfish Lab (AUSL) located in Dauphin Island, Alabama on June 13, 2007. Upon arrival the clams were placed into a 310 L acclimation tank (2.43 m long x 0.56 m wide x 0.23 m deep). The tank was supplied with 26.6 ppt salinity water chilled to match the clam's shipping temperature of 18°C. The clams were acclimated to 29.6°C, the ambient temperature of the flow-through water from the Gulf of Mexico over a period of 24 hours. After acclimation, the clams were transferred to 910 L flow-through tanks (2.43 m long x 0.92 m wide x 0.41 m deep). The water was continuously filtered through a 200 μ bag filter over the inflow pipe. The clams remained in the flow-through tanks until stocking into field grow-out systems.

Clam Stocking

Four treatment densities were used in the belt-bag system (BBS) and adjustable long-line system (ALS) system (Table 1). A baseline density of 750 clams/m² was established based on optimum stocking density determined by Walker (1984) in Georgia. Additional treatment densities were set at one-fourth of baseline (188 clams/m²), one-half of baseline (375 clams/m²), and one and one-fourth of baseline (938 clams/m²).

The bags used in the belt-bag system had a bottom surface area of 0.40 m² which dictated bag densities of 75, 150, 300, 375 clams/m² based on the above treatment densities (Table 1). Each treatment density was replicated in five bags on the BBS.

The ALS bags had a bottom surface area of 0.13 m² which dictated bag densities of 24, 49, 98, and 121 clams/m² based on the above treatment densities (Table 1). Each treatment density was replicated in three bags on the ALS. Lower replicate numbers in the ALS were due to inadequate supply of clams for this study. The large mesh size also restricted the number of clams available for use in the system.

Clams were graded into groups based on the mesh size of bags used in each system. Clams retained on a 12.2 mm square mesh sieve were used in the ALS. Clams retained on a 10.0 mm square mesh sieve were used in the BBS. A random sub-sample of 225 clams from each system was measured for shell length (longest anterior-posterior measurement). Clams used for the BBS had a mean shell length 15.3 mm. Clams used for the ALS had a mean shell length of 17.9 mm.

The first two treatment densities in the BBS of 188 clams/m² and 375 clams/m² were stocked by directly counting each individual clam into the bags. The last two density treatments of 750 clams/m² and 938 clams/m² were stocked using a volumetric count. The volumetric count was performed by filling a one liter beaker with clams then counting the number of clams in the beaker. This was repeated three times to get the average number of clams per liter. The number of clams to be stocked was then divided by the average of the number of clams in a liter. This provided an estimate of the number of liters required to stock the appropriate number of clams. All densities of the long-line system were stocked by directly counting each individual clam into the bag.

The grow-out bags for each system were stocked on June 26, 2008, and placed back into flow through tanks until transport to the field. Bags covered with wet burlap and were transported to the field location on June 28, 2008. Once on site, the bags were attached to randomly assigned positions on each of the four BBS and on the ALS. Individual bags for the BBS were assigned by randomly selecting from all possible bags for a specific location on the belt. This was repeated until all of the belts had been filled. Individual ALS bags were assigned by randomly selecting from all possible bags for the first open position starting on the southwest end of the ALS and continuing to the end.

Data Collection

<u>Belt-Bag System</u>

Clams stocked into field grow-out systems on June 28, 2007 were monitored for growth and mortality on a monthly basis for the BBS until termination of the experiment on February 4, 2008. The BBS was sampled by removing each bag from a particular belt and clipping it to a rope attached to a boat so that bags could remain in the water until sampled. Once all the bags from one belt were removed, one bag at a time was brought onboard and emptied into a sampling tray. Dead clams were removed and the number dead recorded. Dead clams were placed in a labeled Ziploc bag for transport back to the laboratory and stored in a freezer until shell length measurements could be recorded to the nearest 0.1 mm using vernier calipers. Twenty-five live clams were randomly selected and shell length measured and recorded to the nearest 0.1 mm using Vernier calipers and recorded. The clams were stocked back into the bag, clipped onto the rope, and placed back into the water until each successive bag was sampled. After sampling for a particular system was complete, the bags were reattached to the belt in their original order. The bags were attached to the bridle so that the bio-fouled side was on the bottom to suffocate any bio-fouling organisms. Mesh bags were replaced every other month if bio-fouling became excessive. This process was repeated until all four belt-bag systems had been sampled.

<u>Adjustable Long-line System</u>

Growth and mortality were also monitored for the clams in the ALS until February 4, 2008. Three bags were removed from the ALS at a time and placed on the deck of the boat. Each bag was emptied into a sampling tray. Dead clams were removed and the number dead recorded. Dead clams were placed in a labeled Ziploc bag for transport back to the laboratory and stored in a freezer until shell lengths measured and recorded to the nearest 0.1 mm with Vernier calipers. Fifteen live clams were randomly selected from each bag and shell length measured and recorded using Vernier calipers.

After sampling the clams were returned to the bag. Once clams in all three bags were sampled, the bags were clipped back onto the ALS and cable ties replaced on the clips. This process was repeated until all the bags had been sampled. Mesh bags were replaced every other month if bio-fouling became excessive.

Environmental Data

Water temperature (°C), salinity (ppt), and dissolved oxygen levels (mg/l and % saturation) were recorded biweekly from July 5, 2007 to February 4, 2008 using YSI ™ 85D handheld electronic water quality meter. Water quality was measured monthly on the same day and prior to sampling clams. At two week intervals between dates of clam samplings, water quality was measure at the end of a pier approximately 30 m from the experimental site.

Condition Indices

At the conclusion of the experiment, thirty clams from each bag in both the BBS and ALS were randomly selected for condition analysis. All clams available were used if a bag did not contain a population of 30 or more clams. Live weight was recorded for the clams to the nearest 0.01 g. The soft tissue of a clam was shucked onto an aluminum weighing dish. The empty shell was placed into a second weighing dish. The soft tissue and the shell were dried in an oven at 100°C temperature for 24 hours. After drying, a dry meat weight and dry shell weight were recorded to the nearest 0.01 g. The following formula was used to determine the condition index (CI) for each clam (Crosby and Gale 1990):

$$CI = \frac{\text{Dry soft tissue wt (g) x 1000}}{\text{Internal shell cavity capacity (g)}}$$

Internal shell cavity capacity = Total whole live weight – Dry shell weight

Data Analysis

The statistical software package Minitab® 15.1.1.0 (Minitab Inc. 2007) was used to perform all statistical analyses. Significant difference occurrs when $P \leq 0.05$. Mean final size, mean growth rate per month, mean percent mortality, mean percent mortality per month, and mean condition index were calculated for each treatment density in the BBS and ALS. Percent survival data for the BBS and ALS was arcsine square root transformed before analysis. An Analysis of Variance (ANOVA) was used to test for significant differences among treatment densities within a system unless assumptions of an ANOVA were violated. When assumptions were violated the Kruskal-Wallis test was used (Table 2 and 3). A Tukey's Studentized Range (HSD) was used to identify treatment densities with a significant difference within a system following an ANOVA showing a significant difference.

Table 1. Clam stocking densities used per replicates based on clams/m².

Treatment	Clams/m²	Number/BBS Bag (.4m²)	Number/ALS Bag (.13m²)
1/4X	188	75	25
1/2X	375	150	49
X	750	300	98
1 1/4X	938	375	121

Table 2. Statistical tests used for BBS analysis were selected based upon the characteristics of the dataset.

Data set	Test
Final size among densities	ANOVA
Growth rate per month (188/m²)	Kruskal-Wallis
Growth rate per month (375/m²)	ANOVA
Growth rate per month (750/m²)	Kruskal-Wallis
Growth rate per month (938/m²)	ANOVA
Percent mortality among densities	ANOVA
Percent mortality per month (188/m²)	ANOVA
Percent mortality per month (375/m²)	Kruskal-Wallis
Percent mortality per month (750/m²)	ANOVA
Percent mortality per month (938/m²)	Kruskal-Wallis
Condition index among densities	Kruskal-Wallis

Table 3. Statistical tests used for ALS analysis were selected based upon the characteristics of the dataset.

Data set	Test
Final size among densities	ANOVA
Growth rate per month (188/m^2)	Kruskal-Wallis
Growth rate per month (375/m^2)	Kruskal-Wallis
Growth rate per month (750/m^2)	Kruskal-Wallis
Growth rate per month (938/m^2)	ANOVA
Percent mortality among densities	ANOVA
Percent mortality per month (188/m^2)	ANOVA
Percent mortality per month (375/m^2)	Kruskal-Wallis
Percent mortality per month (750/m^2)	ANOVA
Percent mortality per month (938/m^2)	ANOVA
Condition index among densities	ANOVA

Figure 1. The study site is located on the west side of Pointe Aux Pines in Grand Bay, AL.

Figure 2. BBS bag (9.5 mm mesh) with PVC risers used to prevent collapse of the bag.

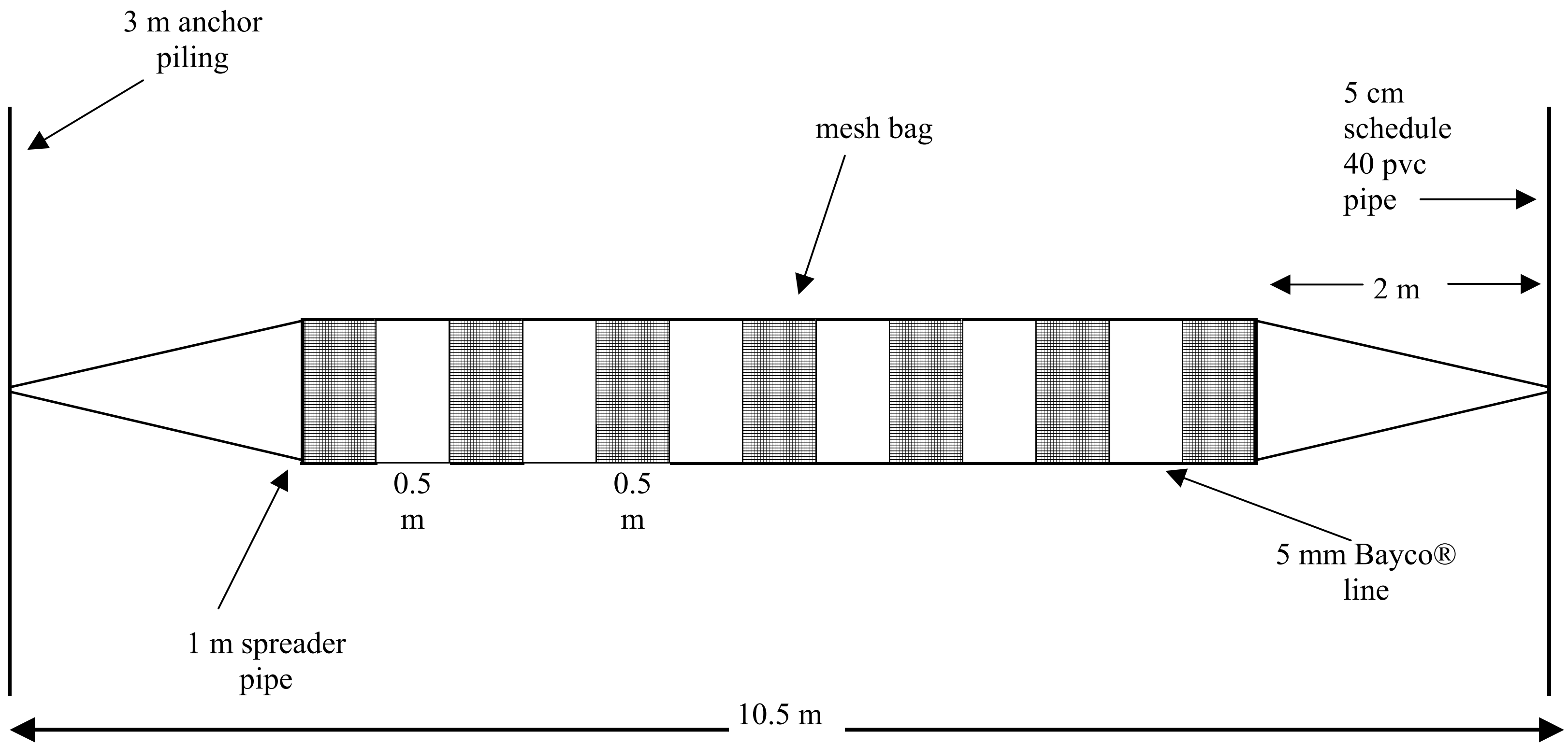

Figure 3. The BBS utilizes a bridle of 5 mm black Bayco® attached to two PVC anchors to secure the mesh bags flat on

Figure 4. PVC closure used to hold the folded edge of the BBS bags over the Bayco® monofilament bridle of the BBS.

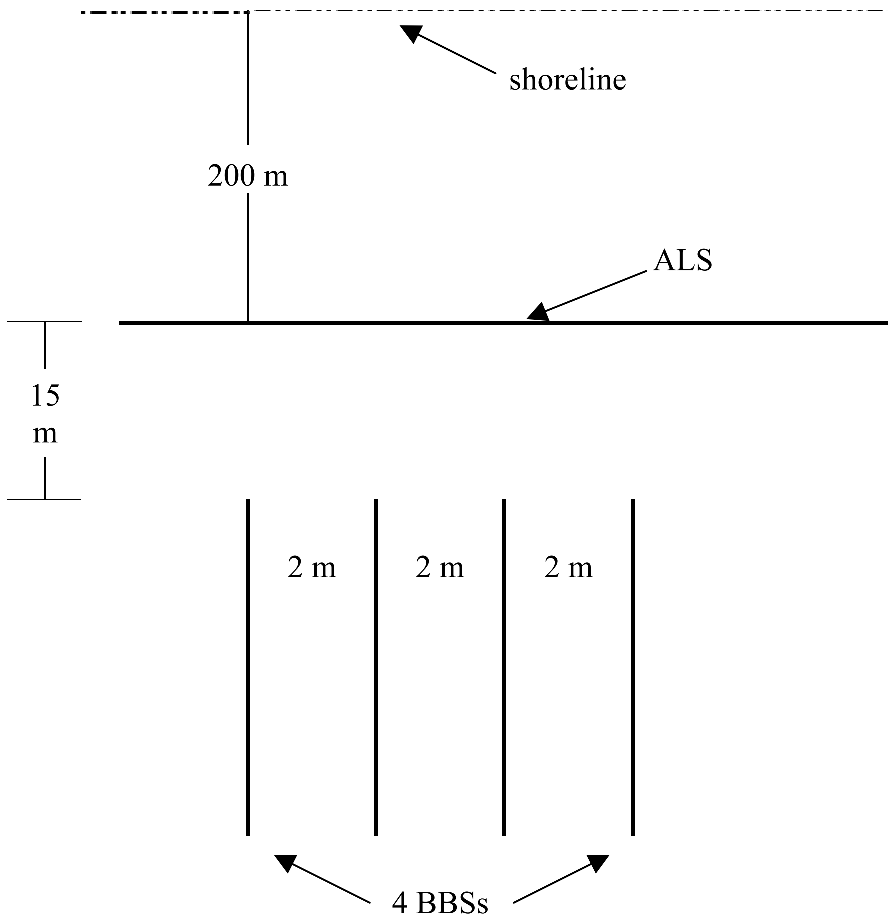

Figure 5. The 4 BBSs were placed 15 m offshore from the ALS and perpendicular to both the ALS and the shoreline.

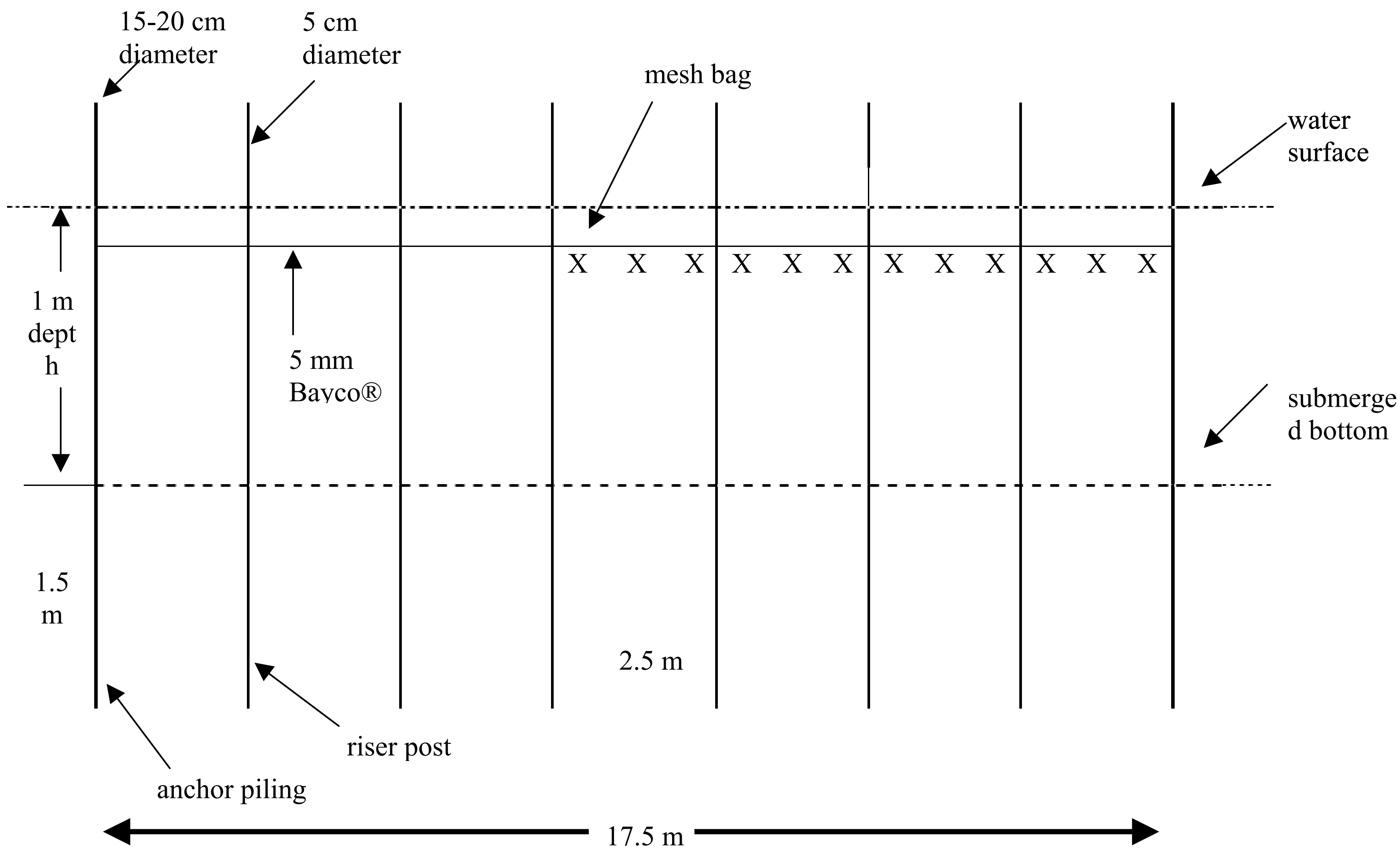

Figure 6. The ALS was constructed using wooden pilings for the anchor pilings and PVC pipes for riser posts. The bags are suspended in the water column and do not come into contact with the bottom.

Figure 7. Photograph of a riser clip attached to a riser post used to support the 5 mm Bayco® line covered with dripper tubing on the ALS

Figure 8. Mesh bag used in the adjustable long-line system with the end open.

III. RESULTS

Environmental Data

Water quality parameters were within favorable ranges for hard clam growth with the exception of water temperature during an 8 week period. Salinity ranged from 24.3 ppt to 31.4 ppt (Figure 9). Dissolved oxygen levels ranged from 4.4 mg/l up to 8.9 mg/l (Figure 10). Percent dissolved oxygen levels ranged from 67.5 up to112.2 (Figure 11). Water temperature ranged from 14.3°C to 33.1°C (Figure 12). From July 19 to September 12, 2007, water temperatures were recorded at or above 30°C.

Shell Length

Hard clams were stocked at a mean shell length of 15.3 mm ± 0.1 in the BBS. At the end of the experiment clams grown in the BBS ranged from 13.6 mm to 32.8 mm shell length. Clams grown in the BBS at the four treatment densities (188, 375, 750, 938 clams/m^2) reached a mean shell length of 20.3 mm ± 0.3, 20.3 mm ± 0.3, 21.5 mm ± 0.3, and 21.1 mm ± 0.3 respectively (Figure 13) (Table 6). The final mean shell length of clams grown at the 750 clams/m^2 was significantly larger ($P = 0.020$) than clams from other densities.

Three bags of clams stocked in the ALS were lost due to storms. One of these bags was a replicate of the 188 clams/m^2 treatment density and two were replicates for the 375 clams/m^2 treatment density. The loss of these replicates prevented any sound statistical inferences being drawn from the ALS data. Hard clams were stocked at a mean shell length of 17.9 mm ± 0.1 in the ALS. Clams grown in the ALS had a range of 17.8 mm to 24.6 mm shell length. Clams grown in the ALS at the four treatment densities (188, 375, 750, 938 clams/m²) had a mean shell length at the end of the study of 21.0 mm ± 1.4, 21.5 mm ± 1.0, 19.6 mm ± 0.3, and 20.7 mm ± 0.7 respectively. (Figure 14) (Table 6). No significant difference was detected for final size of any of the stocking densities.

Growth Rate

The highest mean growth rate per month for clams grown in the BBS, 1.6 ± 0.4 mm/month, was achieved by clams at a density of 938 clams/m² during the month of November (Figure 15) (Table 7). The lowest mean growth rate for clams grown in the BBS, 0.3 ± 0.2 mm/month, was recorded for clams grown in the 375 clams/m² during July. A significant difference was not detected for growth rates between treatment densities.

Clams grown at 375 clams/m² density had the highest growth rate, 2.4 ± 0.0 mm/month, for all densities in the ALS during the month of October (Figure 16) (Table 8). Several densities recorded negative growth rates in the ALS. This was caused by a small sample size due to mortalities. With a small sample size, the effect of a loss of a larger clam can significantly skew mean growth rates. A significant difference was not detected for mean growth rates among treatment densities at specific sampling periods.

Mortality

Mean percent mortality for the clams grown in the BBS at the four densities (188, 375, 750, 938 clams/m²) was 74.4 ± 6.6, 66.1 ± 6.1, and 65.5 ± 6.3, and 55.0 ± 3.9 respectively (Figure 17) (Table 9). There was no significant difference detected among mean percent mortality for the treatment densities in the BBS.

The mean percent mortality for the clams grown in the ALS at the four densities (188, 375, 750, 938 clams/m²) was 50.0 ± 10.0, 61.2 ± 0.0, 80.6 ± 6.4, and 98.1 ± 15.4 respectively (Figure 18) (Table 9). There was no significant difference detected among mean percent mortality for the treatment densities in the ALS.

Percent mortality of the BBS was further analyzed by calculating each month's mean percent mortality (Figure 19) (Table 10). For clams grown at the treatment density 188 clams/m², the month of October was found to have a significantly higher (P = 0.011) mean percent mortality of 32.5 ± 5.2, from all other monthly mean percent mortalities at this treatment density. For clams grown at 375 clams/m², the month of October was found to have a higher mean percent mortality of 37.7 ± 6.0 from all other monthly mean percent mortalities at this treatment density (P = 0.004). Clams grown at 375 clams/m², had significantly lower mean percent mortality from all other monthly mean percent mortalities at this treatment density during the months of August (P = 0.004) and February (P = 0.004), 9.0 ± 0.6 and 8.3 ± 3.4 respectively. The 750 clams/m² treatment density had significantly higher mean percent mortality during the months of September (P = 0.008) and October (P = 0.008) respectively 22.9 ± 3.6 and 23.8 ± 4.8, from all other monthly mean percent mortalities at this treatment density. Mean percent mortality for clams in the 750 clams/m² treatment density were significantly lower during the months of August (P = 0.008), 10.2 ± 0.9 and February (P = 0.008) 8.3 ± 3.9 from all other monthly mean percent mortalities at this treatment density. The highest treatment density of 938 clams/m², had a significantly higher (P = 0.010) mean percent mortality from all other monthly mean percent mortalities at this treatment density during the month of October with 11.9 ± 2.4.

The clams grown in the ALS at the 188 clams/m² and 375 clams/m² treatment densities did not have any significant difference in percent mortality from all other monthly mean percent mortalities at these respective treatment densities. The clams grown in the ALS at the treatment density of 750 clams/m² showed that the month of September had significantly higher ($P = 0.00$) mean percent mortality of 61.9 ± 8.9 from all other monthly mean percent mortalities at this treatment density. The month of November for clams in the ALS at the treatment density of 750 clams/m² had a significantly lower mean percent mortality ($P = 0.00$) of 1.0 ± 1.0 from all other monthly mean percent mortalities at this treatment density (Figure 20) (Table 11). The clams grown in the treatment density of 938 clams/m² had a significantly higher mean percent mortality ($P = 0.008$) of 127.3 ± 64.0 during September from all other monthly mean percent mortalities at this treatment density. July ($P = 0.008$) and February ($P = 0.008$) for clams grown in the treatment density of 938 clams/m² had significantly lower mean percent mortalities of 24.0 ± 4.4 and 2.1 ± 2.1 respectively from all other monthly mean percent mortalities at this treatment density.

Condition Index

Condition indices were calculated and compared between treatment densities within a grow-out system. Clams in the belt-bag system had a range of 42.4 to 125.0. Clams grown in the BBS at the four treatment densities (188, 375, 750, 938 clams/m²) had mean condition index values of 104.1 ± 1.4, 102.8 ± 1.2, 99.5 ± 1.3, and 91.9 ± 1.3 respectively (Figure 21) (Table 12). The clams from the first stocking density of 188 clams/m² had a significantly higher ($P < 0.001$) mean condition index, 104.1, than the other treatment densities. Clams grown at the density of 938 clams/m² had a significantly lower ($P < 0.001$) mean condition index, 91.9, than the other treatment densities.

The condition index values for clams grown in the ALS ranged from 71.4 to 118.9. Clams grown in the ALS at the four treatment densities (188, 375, 750, 938 clams/m²) had mean condition index values of 118.0 ± 2.8, 109.9 ± 9.3, 105.6 ± 4.0, and 90.2 ± 4.1 respectively (Figure 22) (Table 12). The treatment density with 188 clams/m² had a condition index of 118.0 and was significantly higher (P value = 0.018; 3 outliers removed) than the other treatment densities.

Table 4. Mean final size (mm) (± SE) of clams grown in the BBS and the ALS at Grand Bay, AL from June 28, 2007 through February 4, 2008.

Treatment Density	BBS	ALS
188*	20.3 ± 0.3	21.0 ± 1.4
375**	20.3 ± 0.3	21.5 ± 1.0
750	21.5 ± 0.3	19.6 ± 0.3
938	21.1 ± 0.3	20.7 ± 0.7

*** only two remaining replicate ALS bags**
**** only one remaining replicate ALS bag**

Table 5. Mean growth rate per month (mm) (± SE) of clams grown in the BBS at Grand Bay, AL from June 28, 2007 through February 4, 2008.

Treatment Density	Jul	Aug	Sept	Oct	Nov	Jan
188	0.6 ± 0.1	0.6 ± 0.1	1.1 ± 0.3	0.4 ± 0.1	1.1 ± 0.5	0.7 ± 0.2
375	0.3 ± 0.2	1.0 ± 0.2	1.0± 0.3	0.3 ± 0.2	1.0 ± 0.2	0.7 ± 0.3
750	0.6 ± 0.1	0.4 ± 0.1	1.4± 0.5	0.5 ± 0.4	0.7 ± 0.4	1.3 ± 0.5
938	0.4 ± 0.0	0.6 ± 0.3	1.1 ± 0.4	0.7 ± 0.3	1.6 ± 0.4	0.7 ± 0.2

Table 6. Mean growth rate per month (mm) (± SE) of clams grown in the ALS at Grand Bay, AL from June 28, 2007, through February 4, 2008.

Treatment Density	Jul	Aug	Sept	Oct	Nov	Jan
188*	0.6 ± 0.1	1.1 ± 1.0	0.3 ± 0.0	0.7 ± 0.3	0.5 ± 0.1	0.0 ± 0.0
375**	0.4 ± 0.0	1.0 ± 0.0	-0.1 ± 0.0	2.4 ± 0.0	-0.4 ± 0.0	0.0 ± 0.0
750	0.5 ± 0.2	1.4 ± 0.5	-0.8 ± 0.3	0.7 ± 0.3	0.2 ± 0.6	-0.1 ± 0.1
938	0.7 ± 0.1	0.9 ± 0.5	0.1 ± 0.3	0.4 ± 0.3	0.1 ± 0.6	0.3 ± 0.1

*** only two remaining replicate bags**
**** only one remaining replicate bag**

Table 7. Mean percent mortality of clams grown in the BBS and the ALS at Grand Bay, AL from June 28, 2007 through February 4, 2008.

Treatment Density	BBS	ALS
188*	74.4 ± 6.6	50.0 ± 10.0
375**	66.1 ± 6.1	61.2 ± 0.0
750	65.5 ± 6.3	80.6 ± 6.4
938	54.9 ± 4.0	98.1 ± 15.4

*** only two remaining replicate ALS bags**
**** only one remaining replicate ALS bag**

Table 8. Mean percent mortality per month of clams grown in the BBS at Grand Bay, AL from June 28, 2007 through February 4, 2008.

Treatment Density	Jul	Aug	Sept	Oct	Nov	Jan
188	21.1 ± 2.5	11.3 ± 2.0	25.2 ± 8.0	32.5 ± 5.2	20.9 ± 5.9	9.0 ± 2.7
375	15.1 ± 1.3	8.9 ± 0.6	16.5 ± 3.2	37.7 ± 6.0	17.7 ± 3.8	8.6 ± 3.4
750	15.5 ± 2.2	10.2 ± 0.9	22.8 ± 3.6	23.8 ± 4.8	18.2 ± 5.0	8.3 ± 3.9
938	11.6 ± 0.7	11.8 ± 2.4	14.3 ± 3.3	16.4 ± 1.1	11.9 ± 2.4	4.6 ± 0.2

Table 9. Mean percent mortality per month of clams grown in the ALS at Grand Bay, AL from June 28, 2007 through February 4, 2008.

Treatment Density	Jul	Aug	Sept	Oct	Nov	Jan
188*	12.0 ± 8.0	6.3 ± 6.3	21.5 ± 16.6	6.5 ± 1.2	0.0 ± 0.0	8.3 ± 0.0
375**	4.1 ± 0.0	10.6 ± 0.0	42.9 ± 0.0	16.7 ± 0.0	0.0 ± 0.0	2.5 ± 0.0
750	15.0 ± 1.5	19.0 ± 5.9	61.9 ± 8.9	14.1 ± 5.1	1.0 ± 1.0	7.0 ± 5.6
938	24.0 ± 4.4	40.4 ± 18.2	127.3 ± 64.0	5.6 ± 9.1	0.4 ± 2.0	2.1 ± 2.1

*** only two remaining replicate bags**
**** only one remaining replicate bag**

Table 10. Mean condition indices of clams grown in the BBS and the ALS at Grand Bay, AL sampled February 4, 2008.

Treatment Density	BBS	ALS
188*	104.1 ± 1.4	118.0 ± 2.8
375**	102.8 ± 1.2	109.9 ± 9.3
750	99.5 ± 1.3	105.6 ± 4.0
938	91.9 ± 1.3	90.2 ± 4.1

*** only two remaining replicate ALS bags**

**** only one remaining replicate ALS bag**

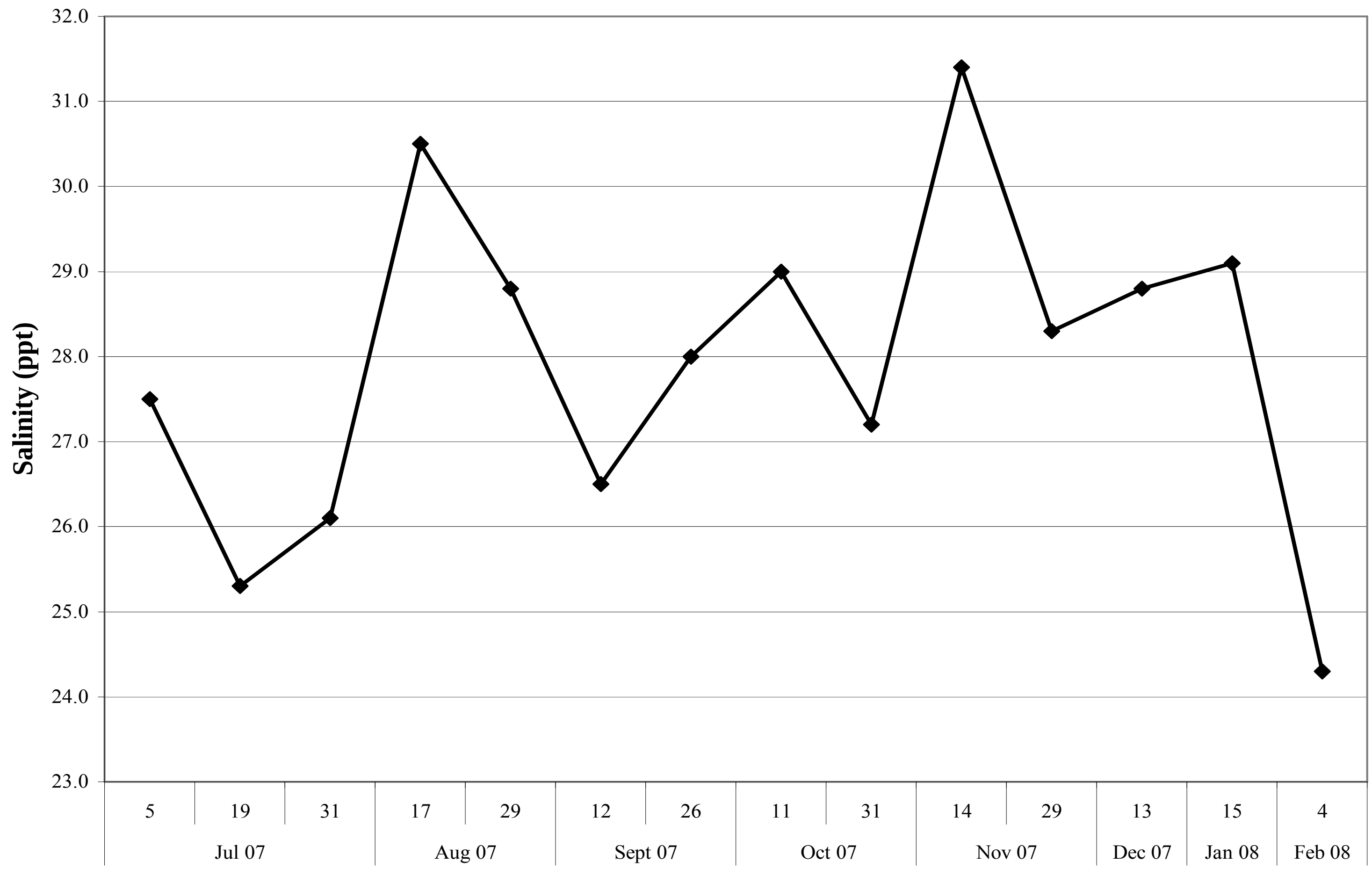

Figure 9. Salinity (ppt) levels recorded at the study site in Grand Bay, AL from June 28, 2007 through February 4, 2008.

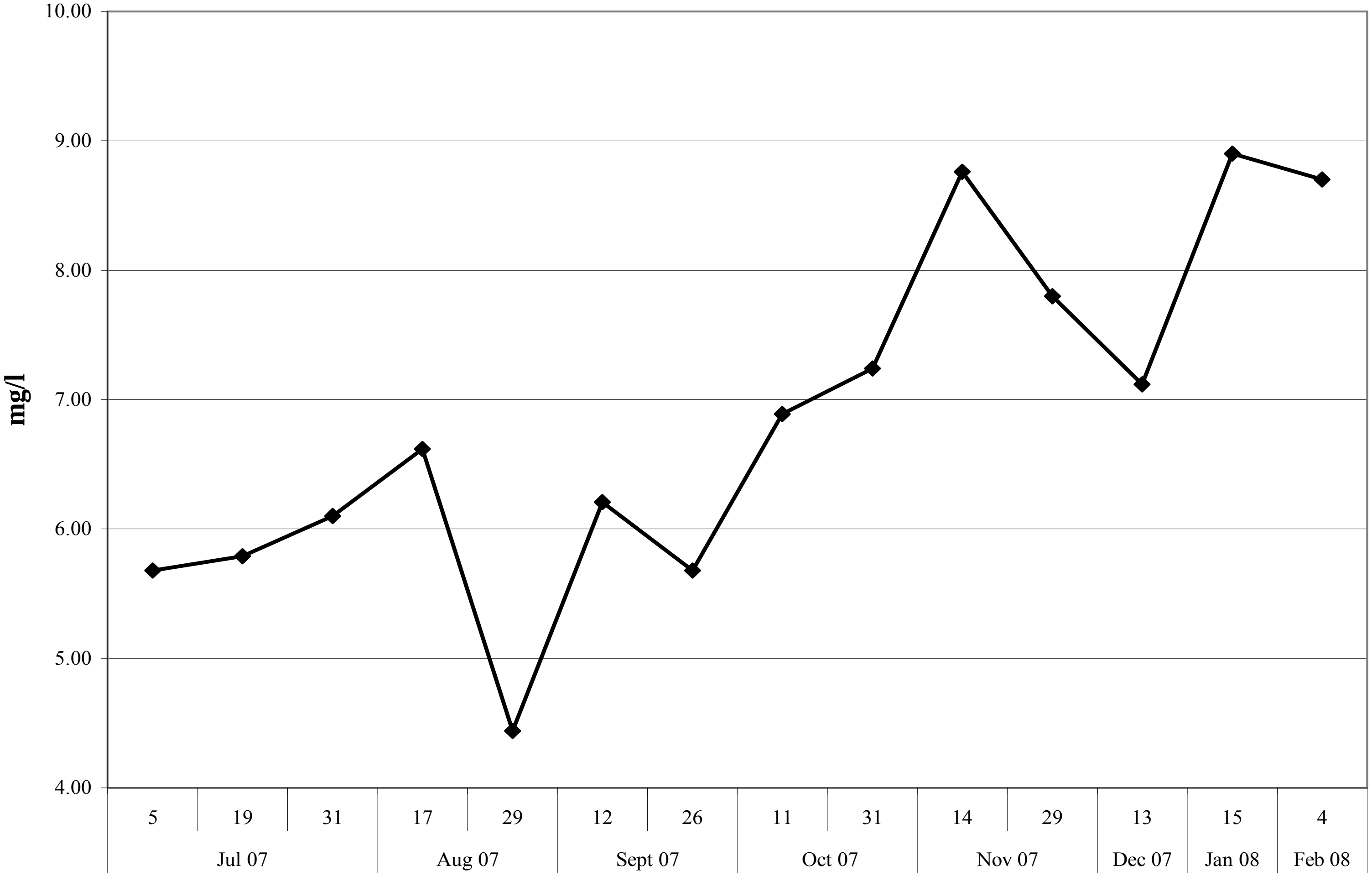

Figure 10. Dissolved oxygen (mg/l) levels recorded at the study site in Grand Bay, AL from June 28, 2007 through February 4, 2008.

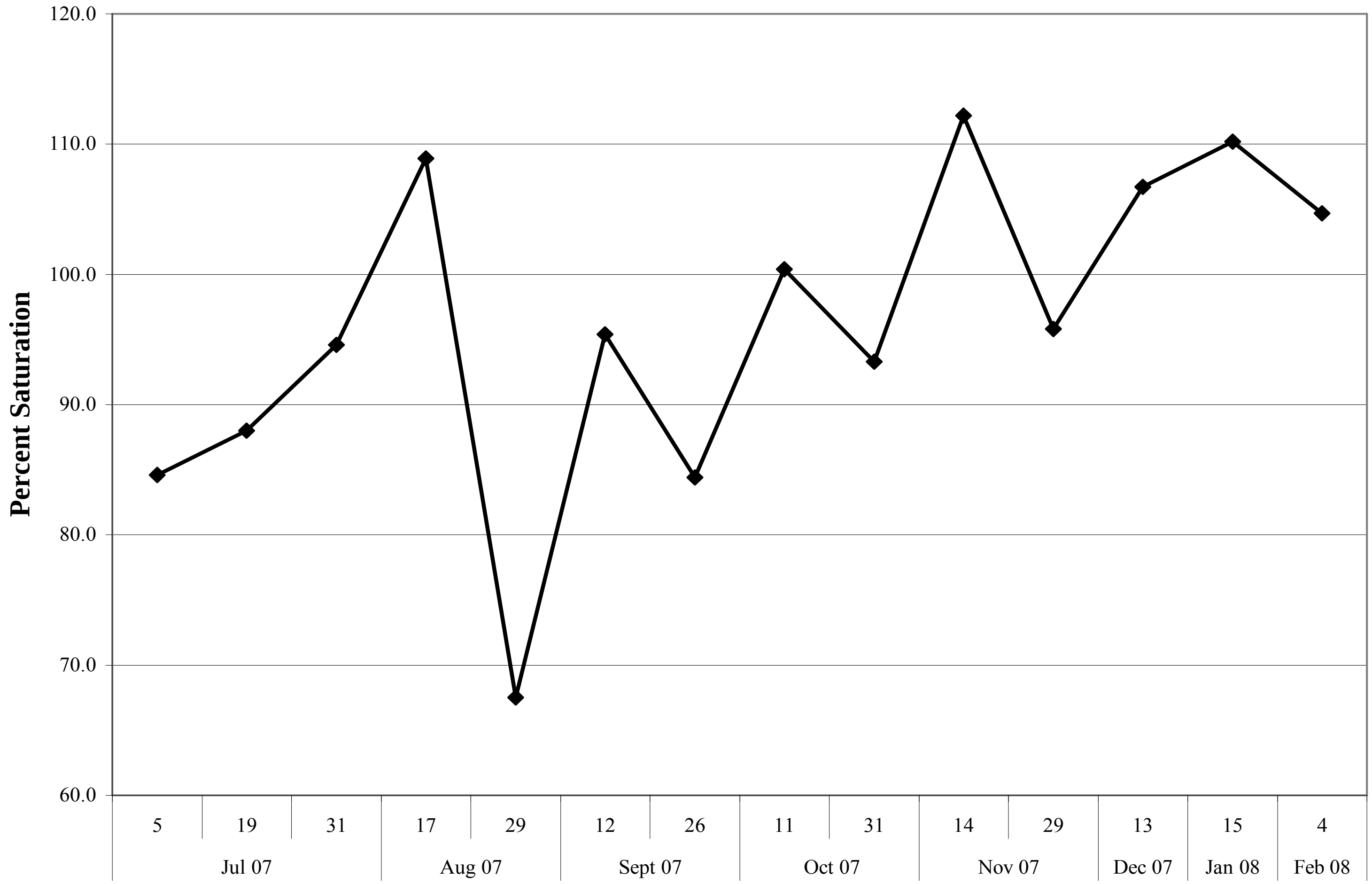

Figure 11. Percent dissolved oxygen saturation levels recorded at the study site in Grand Bay, AL from June 28, 2007 through February 4, 2008.

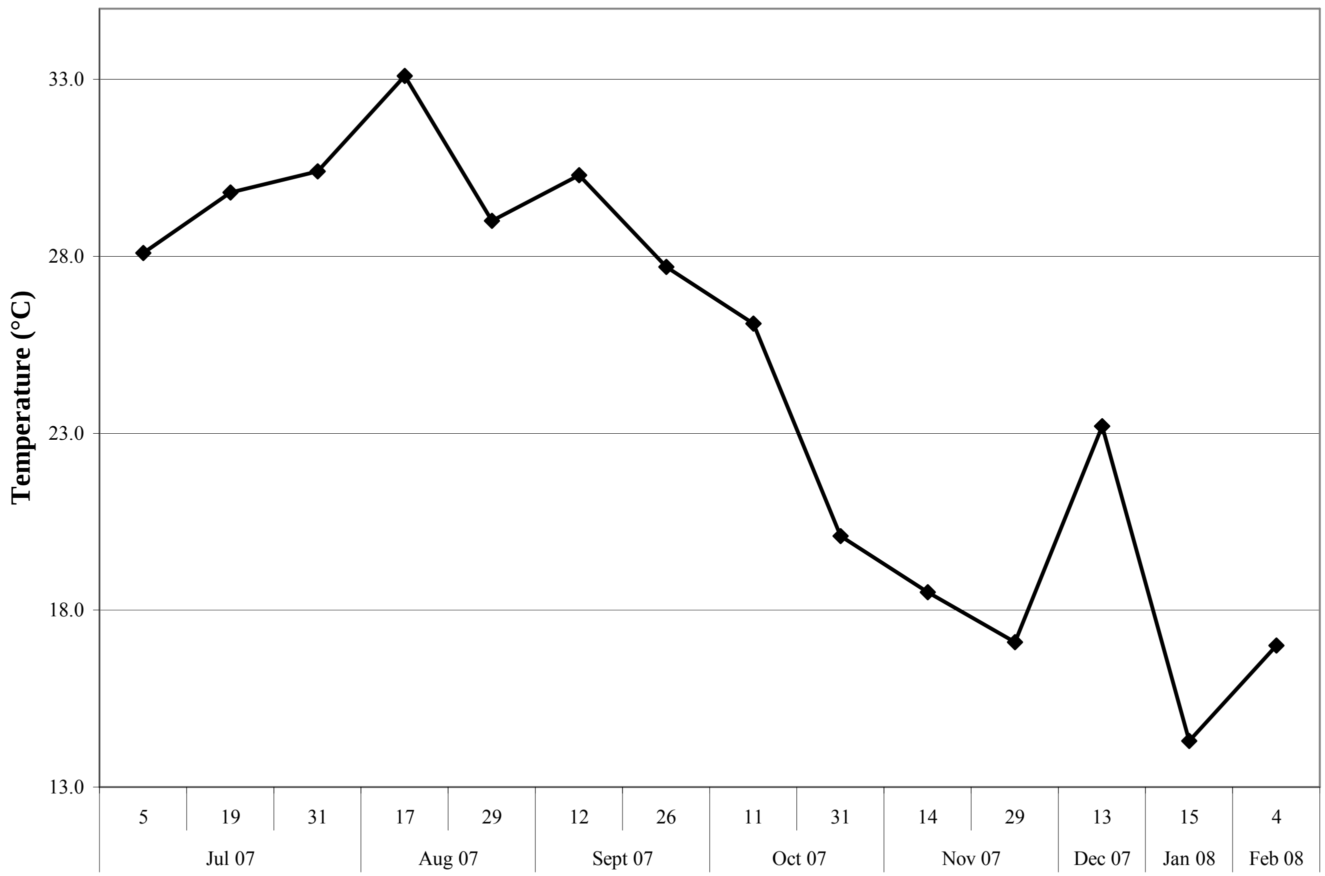

Figure 12. Water temperature (°C) levels recorded at the study site in Grand Bay, AL from June 28, 2007 through February 4, 2008.

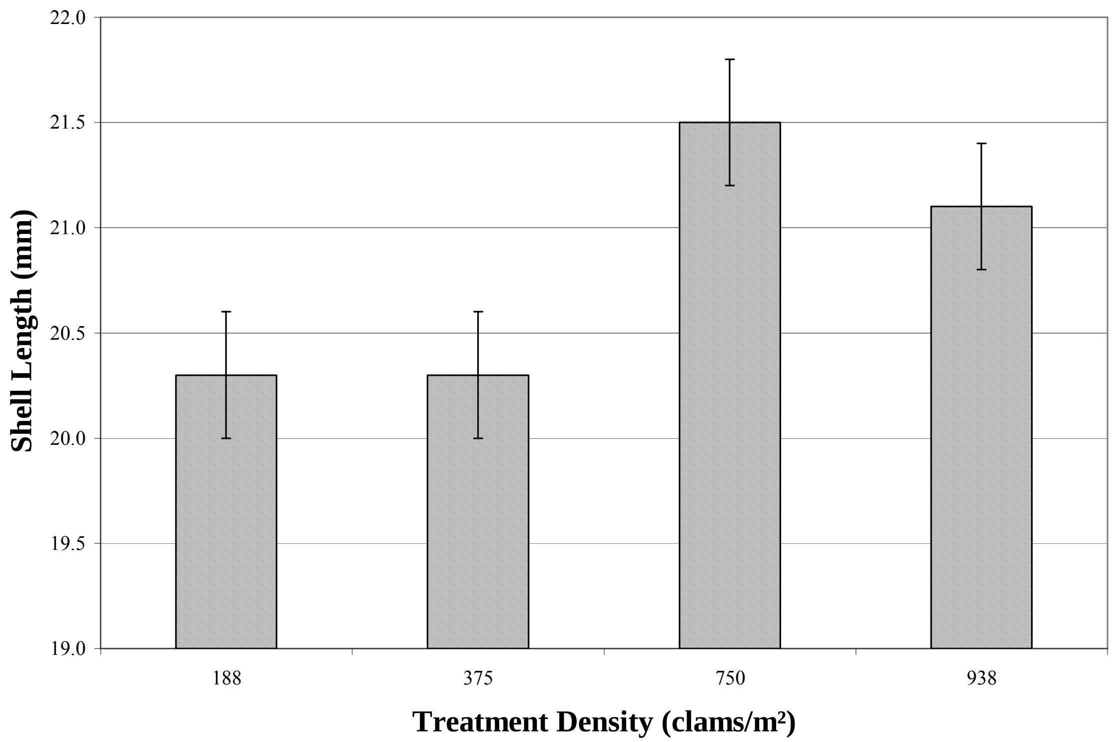

Figure 13. Mean final size (mm) of clams grown in the BBS by treatment density at Grand Bay, AL from June 28, 2007 through February 4, 2008.

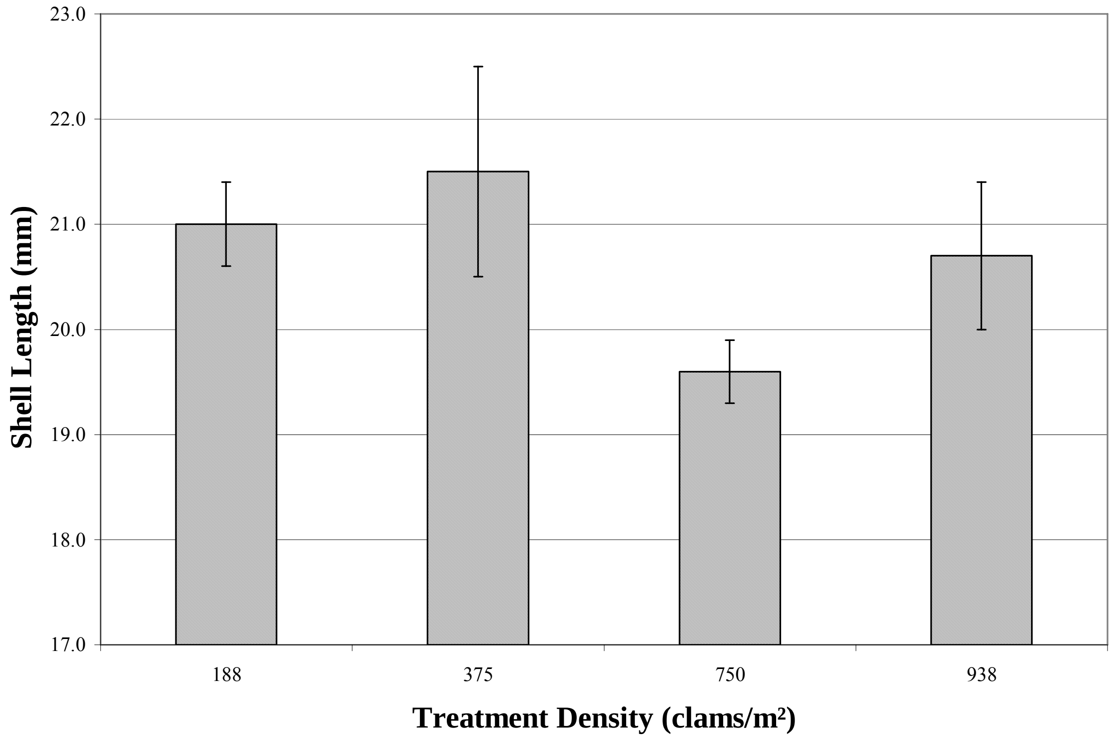

Figure 14. Mean final size (mm) of clams grown in the ALS by treatment density at Grand Bay, AL from June 28, 2007 through February 4, 2008.

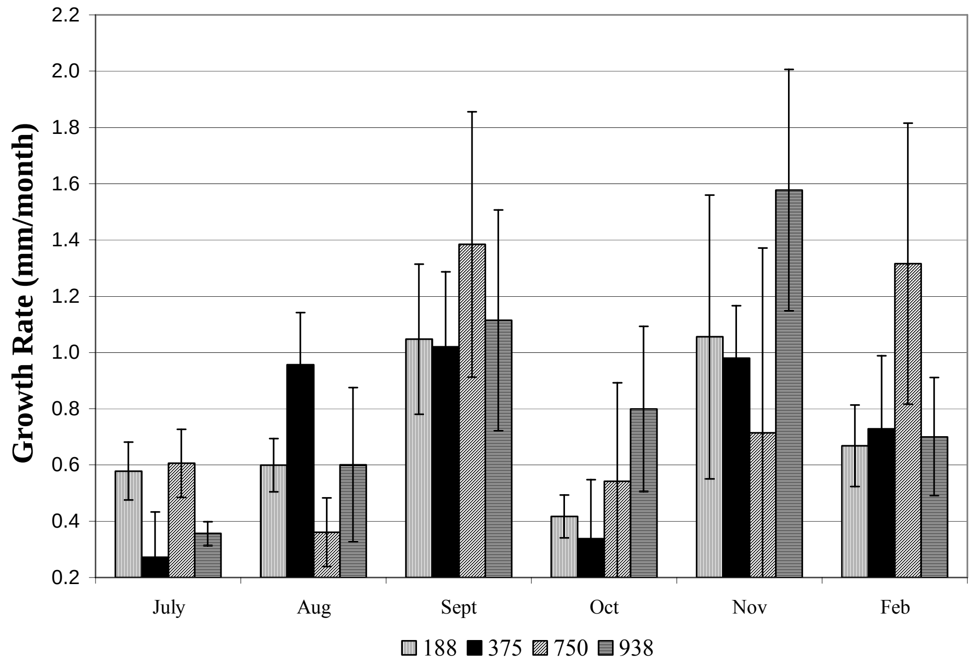

Figure 15. Mean growth rate (mm/month) for clams grown in the BBS by treatment density at Grand Bay, AL from June 28, 2007 through February 4, 2008.

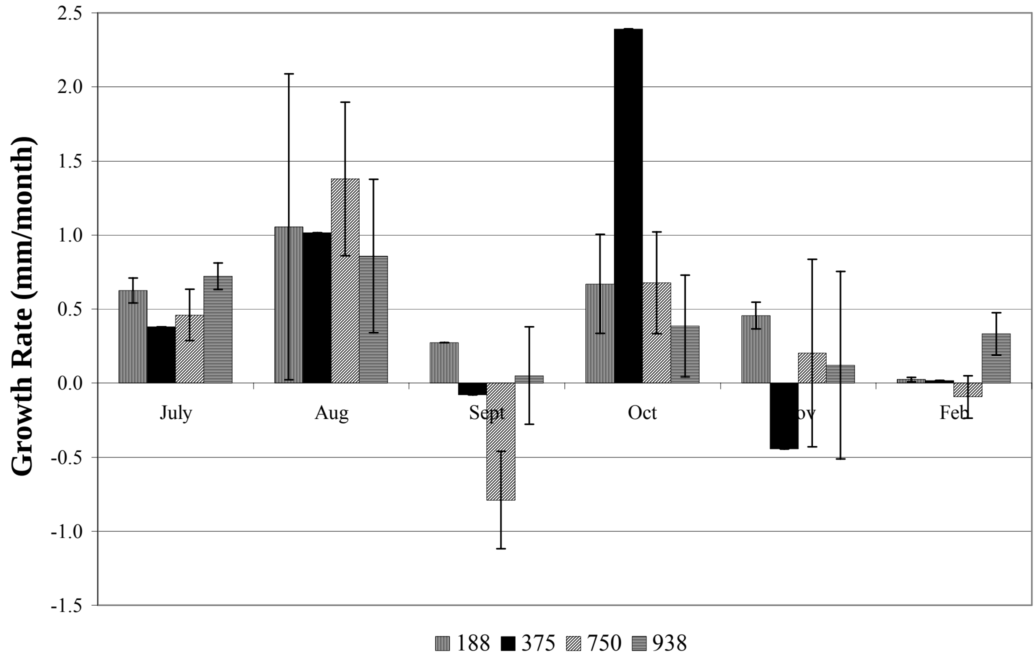

Figure 16. Mean growth rate (mm/month) for clams grown in the ALS by treatment density at Grand Bay, AL from June 28, 2007 through February 4, 2008.

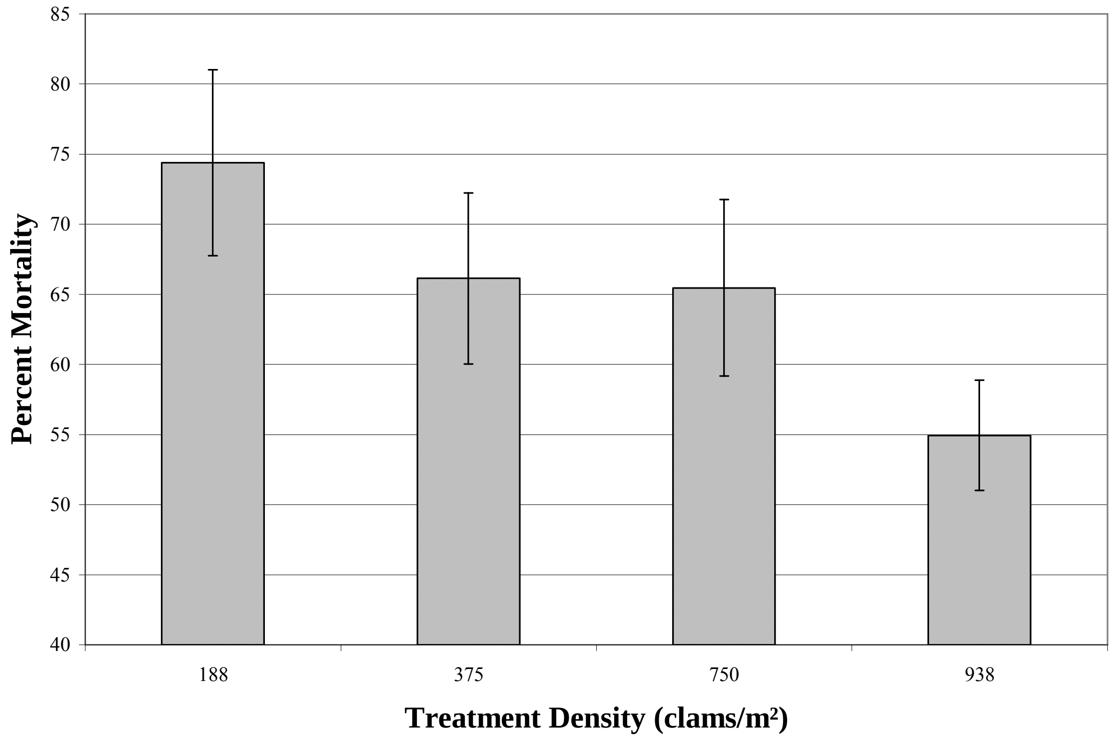

Figure 17. Mean percent mortality of clams grown in the BBS per treatment density at Grand Bay, AL from June 28, 2007 through February 4, 2008.

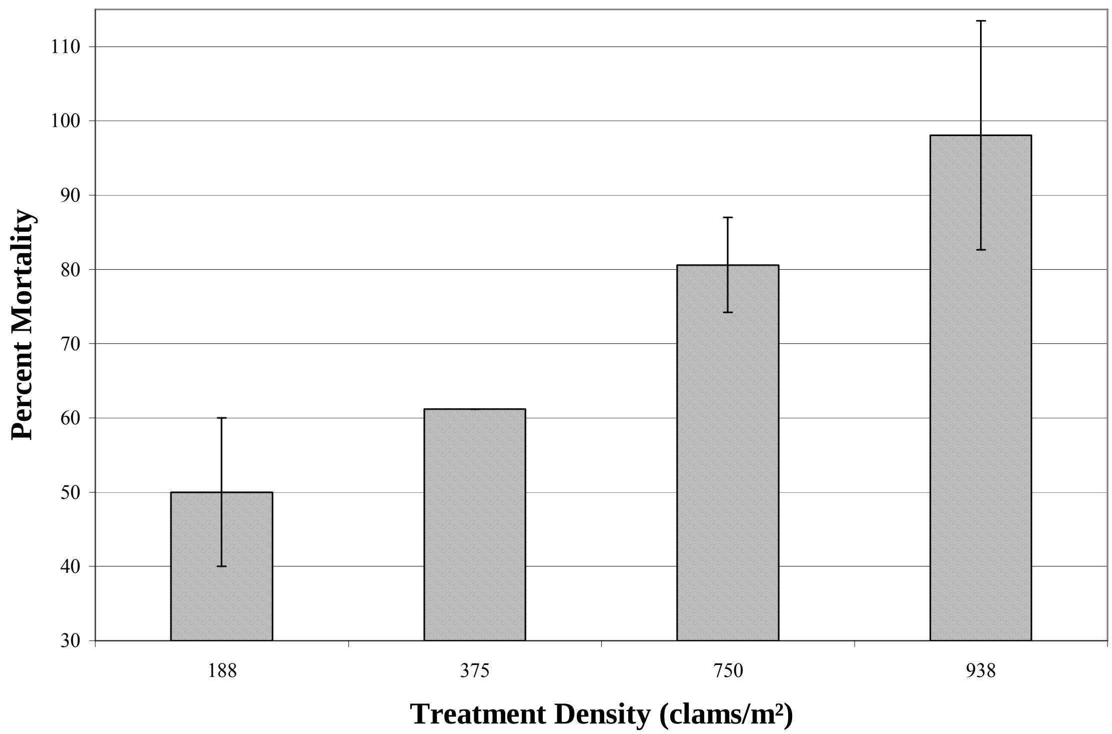

Figure 18. Mean percent mortality of clams grown in the ALS per treatment density at Grand Bay, AL from June 28, 2007 through February 4, 2008.

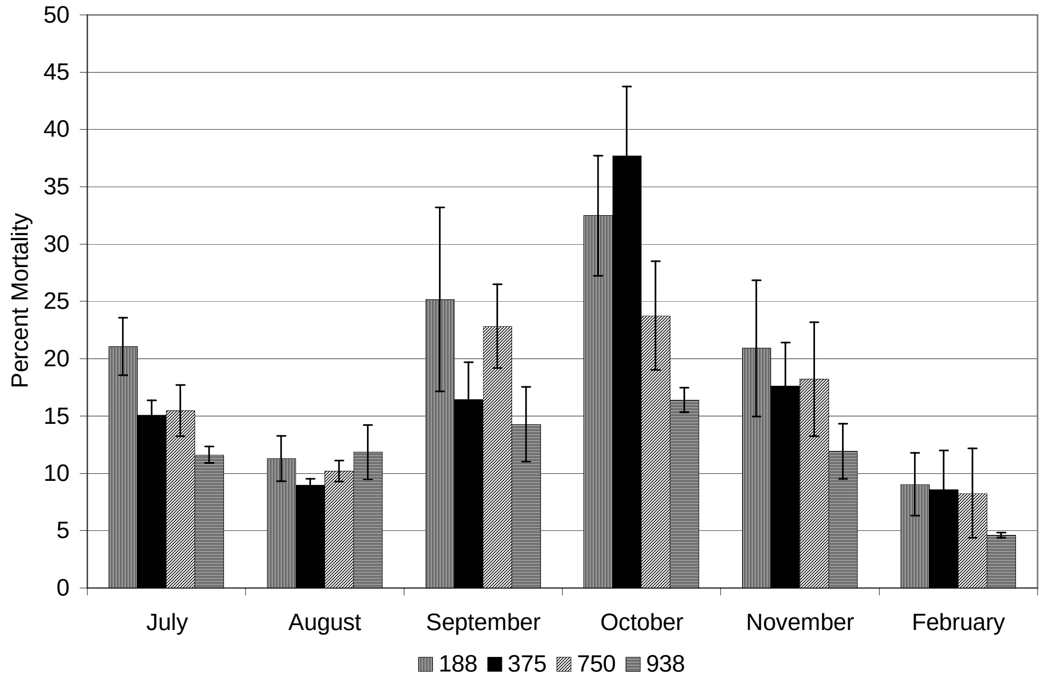

Figure 19. Mean percent mortality per month of clams grown in the BBS per treatment density at Grand Bay, AL from June 28, 2007 through February 4, 2008.

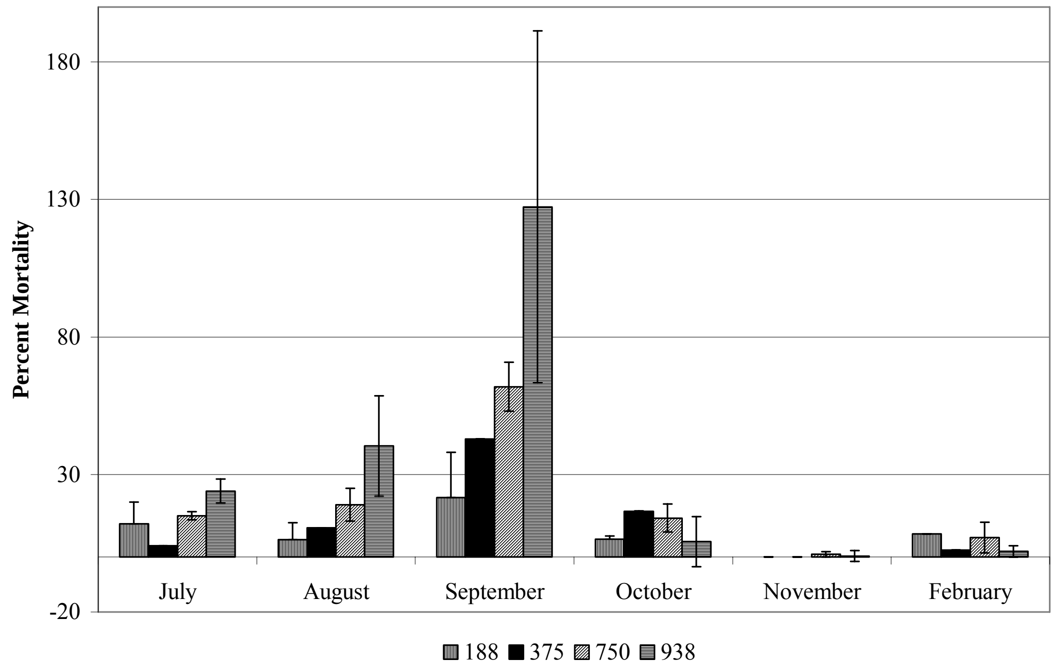

Figure 20. Mean percent mortality per month of clams grown in the ALS per treatment density at Grand Bay, AL from June 28, 2007 through February 4, 2008.

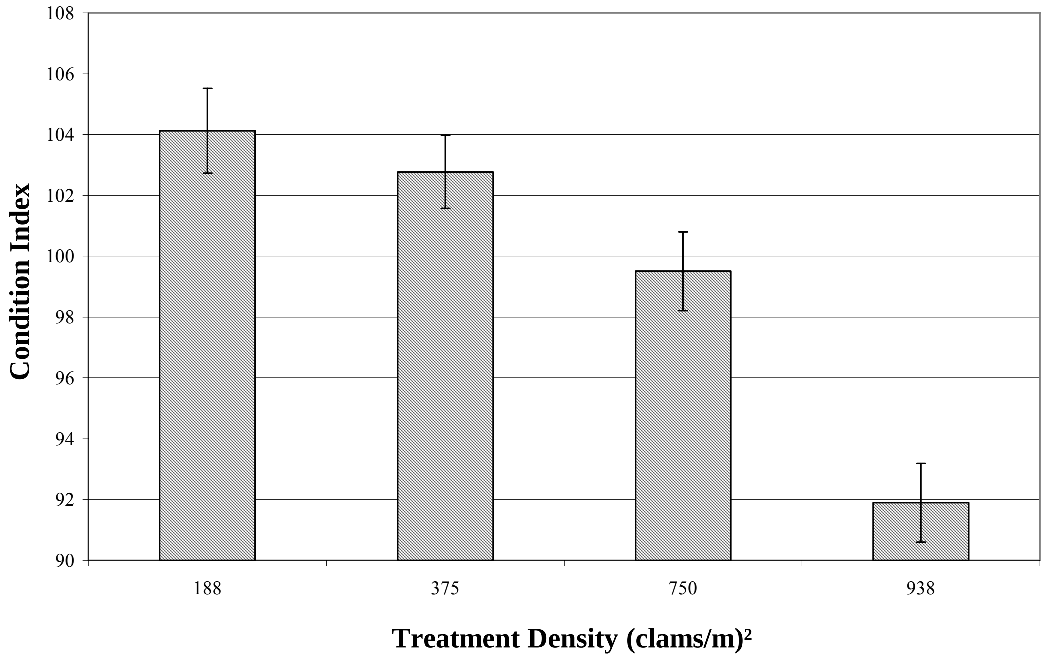

Figure 21. Mean condition index values for clams grown in the BBS per treatment density at Grand Bay, AL sampled on February 4, 2008.

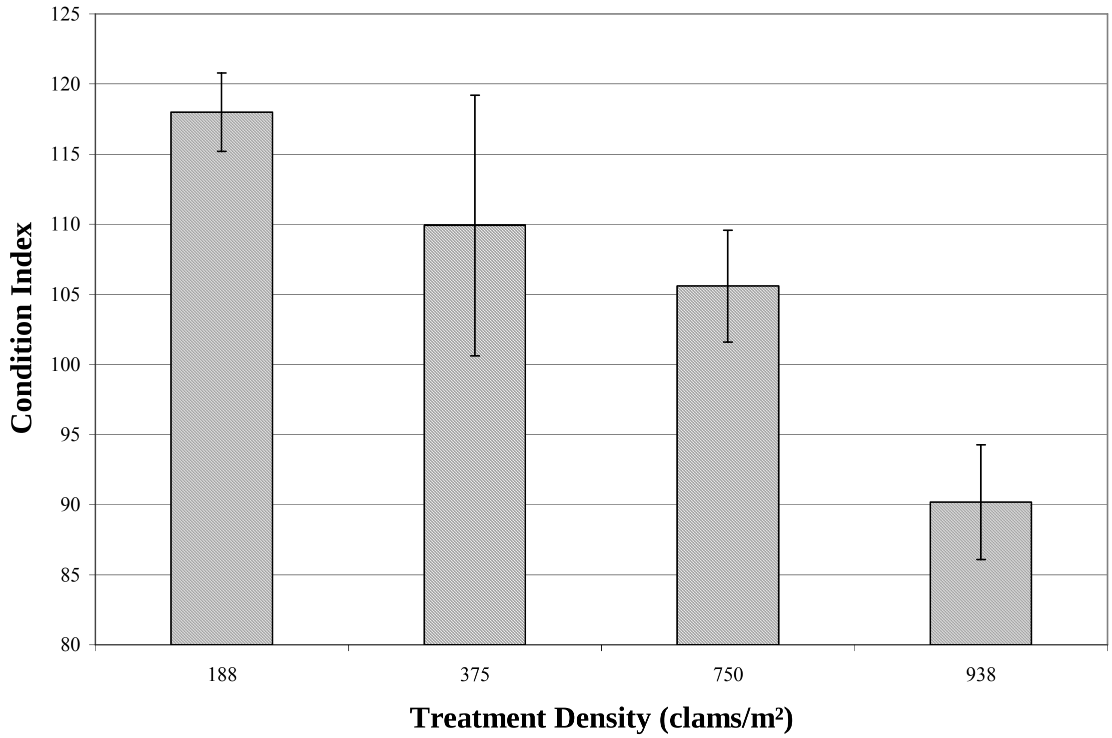

Figure 22. Mean condition index values for clams grown in the ALS per treatment density at Grand Bay, AL sampled on February 4, 2008.

IV. DISCUSSION

Belt-Bag System

Salinity and dissolved oxygen were within favorable range for hard clam growth (Figures 9 and 10). Salinity ranged from 24.3 ppt to 30.5 ppt. Salinity should not have had any negative effect on clam growth or survival as salinity levels stayed well within the hard clam's optimal salinity range of 12.8 to 35 ppt. (Menzel 1963; Woodburn 1961; Van Winkle et al. 1976; Castagna and Chanley 1973). Dissolved oxygen levels ranged from 4.44 mg/l to 8.90 mg/l. Dissolved oxygen levels should not have had a negative effect on clam growth either, with clam growth being shown to decrease at levels of 1 mg/l (Savage 1976; Huntinton and Miller 1989).

Water temperature departed from optimum conditions for a period of 8 weeks during which the water temperatures recorded were near or above 30°C (Figure 12). Water temperatures above 30°C have been shown to limit growth of northern hard clams (Pratt and Campbell 1956; Loosanoff 1939; Menzel 1963, 1964; Van Winkle et al. 1976). Despite high water temperatures, growth rates were not significantly different for sampling periods during the study.

Mean growth rates per month for clams grown in BBS ranged from 0.72 mm/month to 0.86 mm/month (Figure 15). Walker (1984) reported growth rates ranging from 1.41 mm to 2.29 mm mm per month in coastal Georgia. Fernandez et al. (1999) and Walker and Heffernan (1990) both reported average growth rates of 2.6 mm per month.

These reports show that mean clam growth rates per month from the BBS were low. Low growth rates may show that the clams were experiencing a stressor(s) that limited the growth rate.

Treatment density showed no significant effect on mean percent mortality or mean percent mortality by month (Figures 17 and 19). A significantly larger mean final size was observed with the 750 clams/m² treatment density than the mean final size of the other treatment densities (Figure 13). The cause of difference in mean final size in the 750 clams/m² treatment density is not clear. The mean final size measurements did not indicate density dependent growth. Eldridge et al. (1979) showed that clams stocked at lower densities grew faster than clams stocked at higher densities. The findings of Walker (1984) and Peterson and Beal (1989) both support the claims of Eldridge.

The condition index shows clams from the lowest density (188 clams/m²) being in better condition than clams from the highest density (938 clams/m²) (Figure 21). The findings of the condition index show a density dependent condition. It is reasonable to expect a difference in condition index to be translated into a difference in final size. Therefore, it can be speculated that the differences in the mean condition indices represent a recent change in the stressor(s) on the clams in the BBS.

Water quality did not depart from conditions suitable for hard clam growth from late September to early February. Therefore changes in measured water quality parameters are not a contributor to the change in stressor(s) on the clams.

The first likely cause of the change in stressor(s) could be reduced interspecific competition by reduction of bio-fouling organisms. During the warmer months the bags were experiencing a high level of bio-fouling in between sampling periods. During the cooler months the degree of bio-fouling was observed to have decreased with relatively no bio-fouling during the last sampling period. It is possible that the high level of bio-fouling organisms was creating a micro-habitat inside the bags that was limiting food, causing poor water quality, or a combination of both stresses. Paul and Davies (1986) described how bio-fouling organisms restrict the flow of water through mesh. Peterson and Black (1987) and Wildish and Kristmanson (1984) found that suspension feeders are capable of limiting food resources at sufficiently low flow velocities. This effect of limiting food resources and flow velocity may have reduced the growth rate for clams in the BBS.

The other feasible scenario for a decrease in clam stress could be due to a longer period between bag disturbances. The bags were sampled at a one month period except the last sample, which was sampled after a two month sample period. The longer period would have allowed the bags to become buried deeper into the sediment. In a one month period the bags may not have become sufficiently buried to allow for clam burrowing. Clam growth may have slowed because energy was being expended burrowing. To alleviate this situation, a longer period between bag disturbances, or actively burying the bags upon deployment would allow for better growth.

The effects of the bio-fouling organisms and human disturbances of bags may have attributed to limited clam growth. The solution would be difficult to implement because combating bio-fouling requires increased bag disturbances. By lengthening the period between bag disturbances, the accumulation of bio-fouling organisms would likely increase.

During the laboratory analysis for the condition index, shipworms, small burrowing clams of the family Teredinidae, were found in the clams shells. Shipworms typically burrow into wood, however it was found that the shipworms were burrowing into the shell of the clam and eventually burrowing through the shell leading to death. This phenomenon was not found in the literature. The settlement of shipworms on the clams may be a sign that clams were not able to sufficiently burrow to protect themselves.

The treatment density of 750 clams/m² was determined to be the optimum stocking density for clams grown in the BBS during this experiment. This density had a significantly higher mean final size and the mean condition index value was not significantly lower than other treatment densities. Therefore, this is the highest density that did not show any signs of density dependent growth. However, it could not be determined from project data as to whether active bag burial is required and if shorter intervals between bag rotation and replacement are needed during times of high bio-fouling.

Adjustable Long-line System

The adjustable long-line is a non-traditional form of aquaculture for hard clams but may hold some potential and would allow flexibility between crops of oysters and clams. Growth was slow and mortality was high in the ALS. Hard clams are a naturally burrowing animal; therefore being suspended in the water column may act as a stress on the clams. Three bags were lost during storms prior to the first sampling. Afterwards, cable ties were used to secure the bags to the line preventing the loss of bags. The loss of bags resulted in one treatment density (188 clams/m²) with two bags remaining, and another with one bag (375 clams/m^2) remaining. The limited replicates severely restricted the scope of conclusions that could be drawn from the ALS study. The system did experience high mortality and a low rate of growth.

No significant differences were detected for mean final size, mean growth rate per month, mean percent mortality, or mean percent mortality per month (Figures 14, 16,18, and 20). The lowest stocking density of 188 clams/m^2 had significantly higher mean condition index value than the other treatment densities in the ALS (Figure 22). The condition indices did show a trend of decreasing condition with increasing stocking density. The mean percent mortality of clams in the ALS showed a similar trend of increasing percent mortality as density increased.

The ALS experienced higher growth rates during the months of July and August (Figure 16). The BBS had higher growth rates during the months of November and February (Figure 15). The two systems appear to have contrasting high growth periods. A significant difference in growth rate per month was not detected for any treatment density for either system. A trend does appear to exist though. The ALS contained a total of 77 clams by the end of October.

DeCrescenzo (2001) found that hard clams did not grow as well as eastern oysters and had a higher mortality rate when they were grown in Taylor Floats in estuarine conditions. The Taylor Float is a floating cage constructed by building a cage out of mesh then attaching a float such as sealed PVC tubes to the sides of the cage to provide flotation (Goldsborough and Meritt 2001). Coen and Heck (1991) provided data that siphon nipping can significantly decrease growth in hard clams. This likely occurred with clams grown in the ALS bags suspended in the water column. Based on this one experiment, it would be difficult to produce hard clams in the ALS.

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APPENDIX 1. Engineer Form 4345 for application to the U.S. Army Corp of Engineers for a construction permit.

JOINT APPLICATION AND NOTIFICATION
U. S. DEPARTMENT OF ARMY, CORPS OF ENGINEERS
ALABAMA DEPARTMENT OF ENVIRONMENTAL MANAGEMENT

This form is to be used for proposed activities in waters of the United States within the political boundaries of the State of Alabama.

PLEASE TYPE OR PRINT IN INK

1. Date: ____________ month day year	APPLICATION NUMBER (TO BE ASSIGNED BY CORPS)
2. Applicant: Name and Address: Telephone Number during business hours: A/C ( ) ____________Residence A/C ( ) ____________Office	Official Use Only Coe____________ ADEM____________ State Clearinghouse____________ Date Received____________
3. Designation of Agent, Statement of Authorization, I hereby designate and authorize ____________ ____________ to act on my behalf in the processing of this permit application and to furnish, upon request, supplemental information in support of the application. ____________ Signature of Applicant Date	Agent: Name and Address: Telephone Number during business hours: A/C ( ) ____________ A/C ( ) ____________

4. Project Location:
Street Address____________ City/Community____________
Name of Waterway____________ Latitude____________ Longitude____________
Geographic Location: Section ____Township___Range____________ County____________
Loran C coordinates (if applicable)____________

5. Project Description, including all aspects of the project. Describe completely and in detail. Include any structures such as piers, wharfs, bulkheads, pipelines, boathouses, boatramps, groins, jetties, and appurtenances, as well as any dredging, excavation, or fill activities. Attach additional sheets if necessary.

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APPENDIX 1. Continued.

5. Project Description (continued)

6. Dredging Project Specifications (Show locations and dimensions of proposed dredge areas on attached plans. Include existing and proposed depths.).

New Work______ Maintenance Work_______________
Cubic yards of material to be removed________________ Type of material_______________
Surface area (square feet) impacted________________
Method of excavation_______________
Nature of area to be dredged (check one) Upland________ Wetland______ Waterbottom_______________
Other (explain)_______________

7. Specifications for Discharge of Dredged or Fill Material (Show locations and dimensions of all disposal or fill areas on attached plans.).
Cubic yards of fill______________________ Type of fill_______________
Surface area (square feet) impacted_______________
Source of fill material (check one) Commercially obtained________ Dredged material_______ Borrowed on-site_______________
Other (explain)_______________
How will discharged material be contained? (Include erosion control measures, levees, etc.)_______________

Nature of disposal/fill areas (check one) Upland________ Wetland ________ Waterbottom_______________
Other (explain)_______________

8. Additional information relating to the proposed activity.
Are oyster reefs located within or near the project area? Yes______ No_________ If yes, explain:_______________

Will this project result in the siting, construction, and/or operation of an energy-related facility? Yes________ No________
Is the project area greater than 25 acres in size? Yes_______ No_________
Is any portion of the activity for which authorization is sought now complete? Yes________ No_________ If yes, explain:

_______________ Month and year activity took place_______________
If project is for maintenance work of existing structures or existing channels, describe legal authorization for the existing work. Provide permit number, dates or other form of authorization_______________

9. Describe the purpose and public benefit, if any, of the project. Describe the relationship between the project and any secondary or future development the project is designed to support. _______________

Intended use: Public____________ Private _________ Commercial ___________Other (explain)_______________

10. Project Schedule:
Proposed start date _______________ Proposed completion date_______________

11. Names and address of adjoining property owners, lessees, etc. whose property also adjoins the waterway. Also identify the owners on the plan views in Attachment.

12. List all authorizations or certifications received or applied for from federal, state or local agencies for any Structures, construction, discharges, deposits or other activities described in or directly related to this application. Note that the signature in Item 13 certifies that application has been made to or that permits Are not required from the following agencies. If permits are not required place NA in space for Type Approval.

Agency	Type Approval	Identification No.	Date of Application	Date of Approval	Date of Denial
AL Dept. of Environmental Management					
U. S. Army Corps of Engineers					
Alabama State Docks					
City/County____________					
Other_________________					

APPENDIX 1. Continued.

13. Application is hereby made for authorization to conduct the activities described herein. I agree to provide any additional information/data that may be necessary to provide reasonable assurance or evidence to show that the proposed project will comply with the applicable state water quality standards or other environmental protection standards both during construction and after the project is completed. For projects within the coastal area of Mobile and Baldwin Counties, I certify that the proposed project for which authorization is sought complies with the approved Alabama Coastal Area Management Program and will be conducted in a manner consistent with the program. I agree to provide entry to the project site for inspectors from the environmental protection agencies for the purpose of making preliminary analyses of the site and monitoring permitted works. I certify that I am familiar with responsible for the information contained in this application, and that to the best of my knowledge and belief such information is true, complete and accurate. I further certify that I posses the authority to undertake the proposed activities or I am acting as the duly authorized agent of the applicant.
(SIGNATURE OF APPLICANT OR AGENT REQUIRED BELOW)

Signature of Applicant or Agent

Date

18 U.S.C. Section 1001 provides that: Whoever, in any manner within the jurisdiction of any department or agency of the United States knowingly and willingly falsifies, conceals, or covers up by any trick, scheme or device a material fact or make any false, fictitious or fraudulent statements or representations or makes or uses any false writing or document knowing same to contain any false, fictitious or fraudulent statement or entry, shall be fined not more than $10,000 or imprisoned not more than five years or both.

14. **In addition to the completed application, the following attachments are required:**

Provide a vicinity map showing the location of the proposed site along with a written description of how to reach the site from major highways or landmarks. Provide accurate drawings of the project site with existing structures and proposed activities shown in detail. All drawings must be to scale or with dimensions noted on drawings and must show a plan view and across section or elevation. **All plans and attachments must be of reproducible quality on 8 ½ inch *11 inch paper. FEES ARE REQUIRED IN CONJUCTION WITH ADEM CERTIFICATION:**
ADEM WILL CONTACT APPLLICANT WITH FEE REQUIREMENTS.

15. APPLICATION SUBMISSION INFORMATION

Contact the Corps of Engineers prior to submittal with any questions or to request acceptable alternate content/format. An instruction package, example PAP and SPCC plans, and other information are available upon request. Complete this form, attach additional information as necessary, and submit signed original to:

(Statewide, Except Tennessee River Watershed)
District Engineer, Attn: Regulatory Branch
U.S. Army Corps of Engineers - **Mobile District**
Post Office Box 2288
Mobile, Alabama 36628-0001
Phone: (251) 690-2658 Fax: (251) 6902660
WebPage: www.sam.usace.army.mil/

OR

(Tennessee River Watershed Only)
District Engineer, Attn: Regulatory Branch
U.S. Army Corps of Engineers - **Nashville District**
3701 Bell Road
Nashville, Tennessee 37214
Phone: (615) 369-7500 Fax: (615) 3697501
WebPage: www.orn.usace.army.mil/

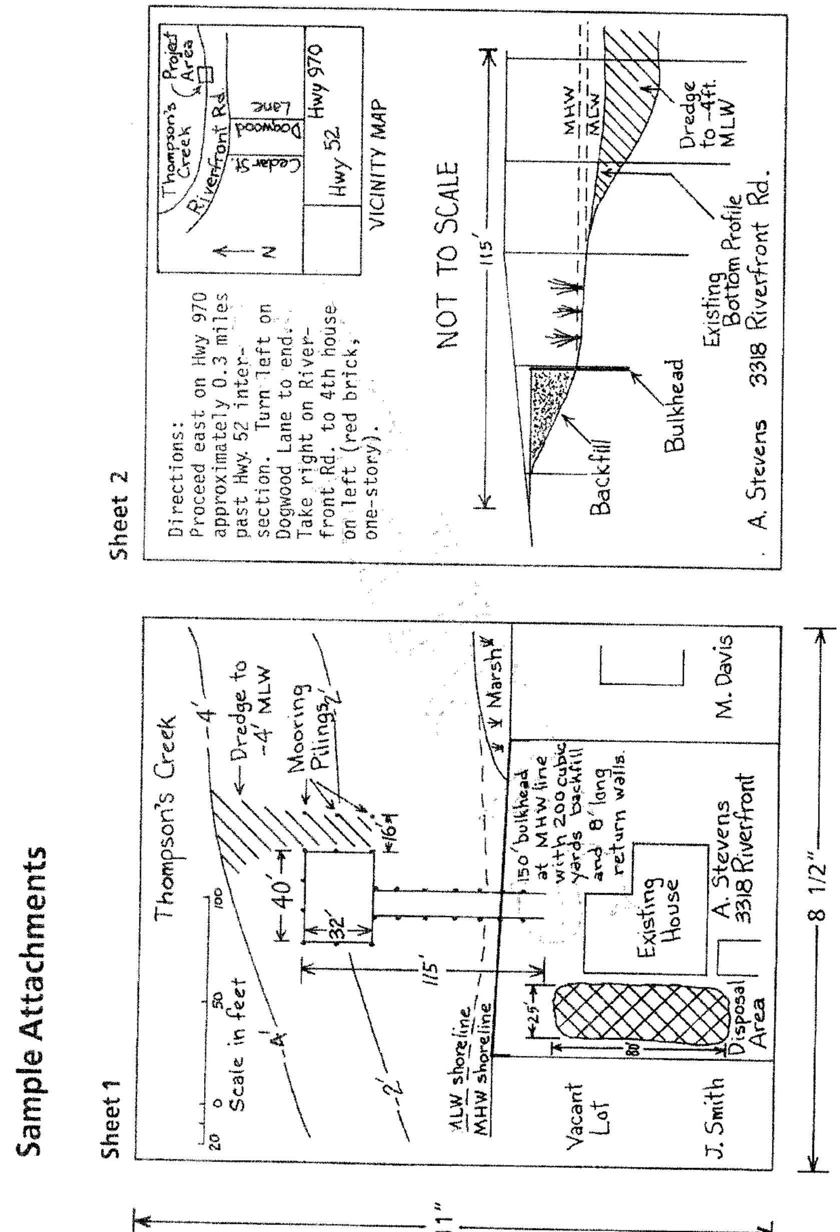
Submit signed copy of application and attachments to:

(Statewide)
Mining & Nonpoint Source Section
Field Operations Division, ADEM
PO Box 301463
Montgomery, AL 36130-1463 or
1400 Coliseum Boulevard
Montgomery, AL 36110-2059
Phone: (334) 394-4311 Fax: (334) 394-4326
Email: mnps@adem.state.al.us
WebPage: www.adem.state.al.us

(Mobile, Baldwin, & Washington Counties Only)
Coastal Section - Mobile Branch
Field Operations Division, ADEM
4171 Commander's Drive
Mobile, AL 36615
Phone: (251) 432-6533 Fax: (251) 4326598
Email: coastal6@adem.state.al.us
WebPage: www.adem.state.al.us

(Statewide)
Alabama State Port Authority
Environmental, Health & Safety
1400 ASD Boulevard Room 216
Mobile, AL 36602
Phone: (251) 441-7085
Fax: (251) 441-7255
WebPage: www.asdd.com

APPENDIX 1. Continued.



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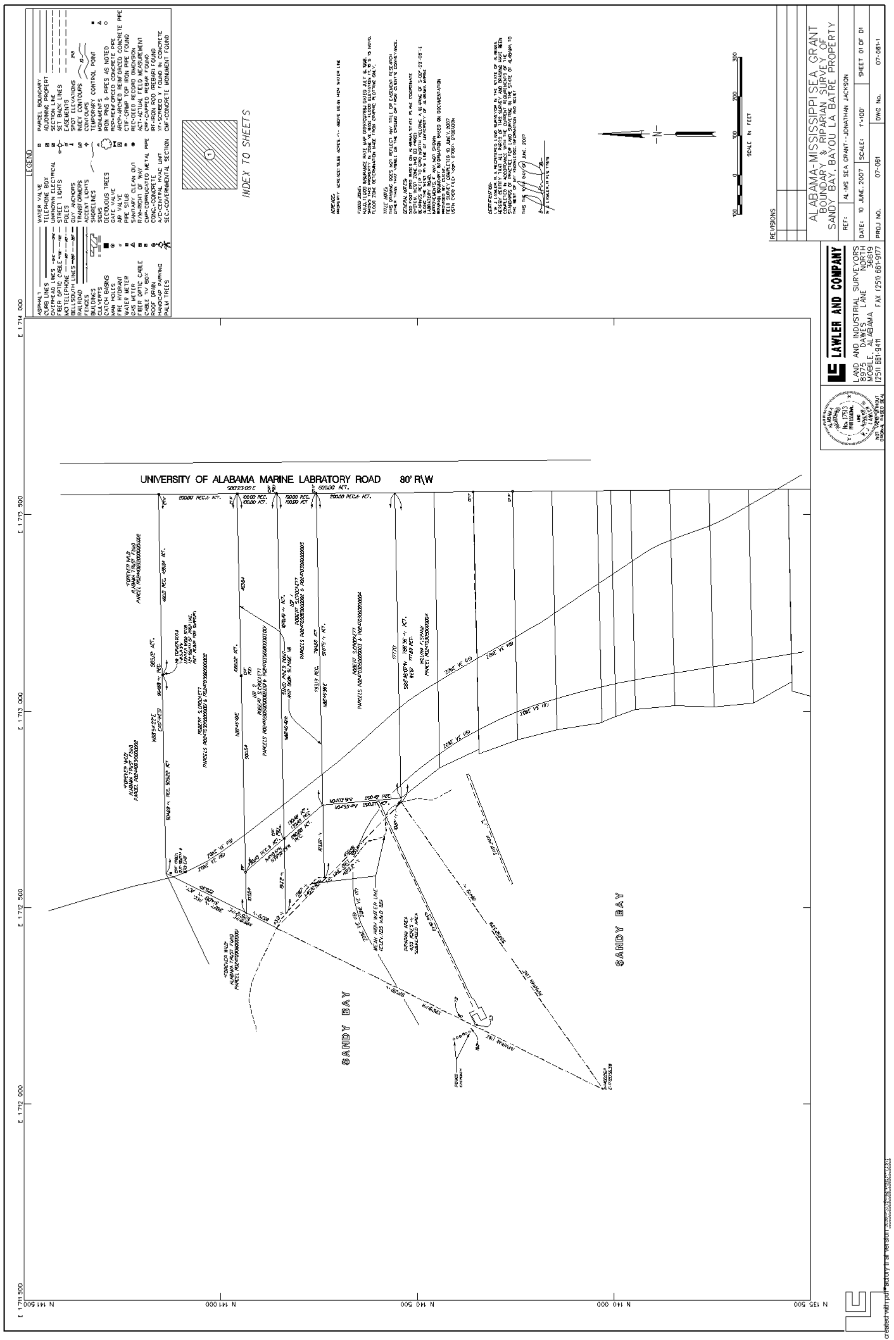
APPENDIX 2. Plat of riparian survey for application of a riparian lease from the state of Alabama.



APPENDIX 3. Letter of permission from the landowner, Steve Crockett, to Auburn University Marine Extension and Research Center.

Memorandum

To: Jonathan Jackson

CC:

From: Steve Crockett

Date: 6/13/2007

Re: Permission to access property

This document gives Mr. Jonathan Jackson permission to use my property located on Sandy Bay, pier, deck, and the adjacent water to which attach riparian rights as delineated by survey for the purposes of clam aquaculture.

APPENDIX 4. Nationwide Permit 4 used by the U.S. Army Corp of Engineers for classifying construction permits.

DECISION DOCUMENT
NATIONWIDE PERMIT 4

This document discusses the factors considered by the Corps of Engineers (Corps) during the issuance process for this Nationwide Permit (NWP). This document contains: (1) the public interest review required by Corps regulations at 33 CFR 320.4(a)(1) and (2); (2) a discussion of the environmental considerations necessary to comply with the National Environmental Policy Act; and (3) the impact analysis specified in Subparts C through F of the 404(b)(1) Guidelines (40 CFR Part 230). This evaluation of the NWP includes a discussion of compliance with applicable laws, consideration of public comments, an alternatives analysis, and a general assessment of individual and cumulative impacts, including the general potential effects on each of the public interest factors specified at 33 CFR 320.4(a).

1.0 Text of the Nationwide Permit

Fish and Wildlife Harvesting, Enhancement, and Attraction Devices and Activities. Fish and wildlife harvesting devices and activities such as pound nets, crab traps, crab dredging, eel pots, lobster traps, duck blinds, and clam and oyster digging, and small fish attraction devices such as open water fish concentrators (sea kites, etc.). This NWP does not authorize artificial reefs or impoundments and semi-impoundments of waters of the United States for the culture or holding of motile species such as lobster, or the use of covered oyster trays or clam racks. (Sections 10 and 404)

1.1 Requirements

General conditions of the NWPs are in the Federal Register notice announcing the issuance of this NWP. Pre-construction notification requirements, additional conditions, limitations, and restrictions are in 33 CFR part 330.

1.2 Statutory Authority

- Section 10 of the Rivers and Harbors Act of 1899 (33 U.S.C. 403)
- Section 404 of the Clean Water Act (33 U.S.C. 1344)

1.3 Compliance with Related Laws (33 CFR 320.3)

1.3.1 General

NWPs are a type of general permit designed to authorize certain activities that have minimal adverse effects on the aquatic environment and generally comply with the related laws cited in 33 CFR 320.3. Activities that result in more than minimal adverse effects on the aquatic environment, individually or cumulatively, cannot be authorized by NWPs. Individual review of each activity authorized by an NWP will not normally be performed, except when preconstruction notification to the Corps is required or when an applicant requests verification that an activity complies with an NWP. Potential adverse impacts and compliance with the laws cited in 33 CFR 320.3 are controlled by the terms and conditions of each NWP, regional and case-specific conditions, and the review process that is undertaken prior to the issuance of NWPs.

The evaluation of this NWP, and related documentation, considers compliance with each of the following laws, where applicable: Sections 401, 402, and 404 of the Clean Water Act; Section 307(c) of the Coastal Zone Management Act of 1972, as amended; Section 302 of the Marine Protection, Research and Sanctuaries Act of 1972, as amended; the National Environmental Policy Act of 1969; the Fish and Wildlife Act of 1956; the Migratory Marine Game-Fish Act; the Fish and Wildlife Coordination Act, the Federal Power Act of 1920, as amended; the National Historic Preservation Act of 1966; the Interstate Land Sales Full Disclosure Act; the Endangered Species Act; the Deepwater Port Act of 1974; the Marine Mammal Protection Act of 1972; Section 7(a) of the Wild and Scenic Rivers Act; the Ocean Thermal Energy Act of 1980; the National Fishing Enhancement Act of 1984; and the Magnuson-Stevens Fishery and Conservation and Management Act. In addition, compliance of the NWP with other Federal requirements, such as Executive Orders and Federal regulations addressing issues such as floodplains, essential fish habitat, and critical resource waters is considered.

1.3.2 Terms and Conditions

Many NWPs have notification requirements that trigger case-by-case review of certain activities. Two NWP general conditions require case-by-case review of all activities that may adversely affect Federally-listed endangered or threatened species or historic properties (i.e., general conditions 17 and 18). General condition 15 restricts the use of NWPs for activities that are located in Federally-designated wild and scenic rivers. None of the NWPs authorize artificial reefs. General condition 24 prohibits the use of an NWP with other NWPs, except when the acreage loss of waters of the United States does not exceed the highest specified acreage limit of the NWPs used to authorize the single and complete project.

In some cases, activities authorized by an NWP may require other federal, state, or local authorizations. Examples of such cases include, but are not limited to: activities that are in marine sanctuaries or affect marine sanctuaries or marine mammals; the ownership, construction, location, and operation of ocean thermal conversion facilities or deep water ports beyond the territorial seas; activities that result in discharges of dredged or fill material into waters of the United States and require Clean Water Act Section 401 water quality certification; or activities in a state operating under a coastal zone management program approved by the Secretary of Commerce under the Coastal Zone Management Act. In such cases, a provision of the NWPs states that an NWP does not obviate the need to obtain other authorizations required by law. [33 CFR 330.4(b)(2)]

Additional safeguards include provisions that allow the Chief of Engineers, division engineers, and/or district engineers to: assert discretionary authority and require an individual permit for a specific activity; modify NWPs for specific activities by adding special conditions on a case-by-case basis; add conditions on a regional or nationwide basis to certain NWPs; or take action to suspend or revoke an NWP or NWP authorization for activities within a region or state. Regional conditions are imposed to protect important regional concerns and resources. [33 CFR 330.4(e) and 330.5]

1.3.3 Review Process

The analyses in this document and the coordination that was undertaken prior to the issuance of the NWP fulfill the requirements of the National Environmental Policy Act (NEPA), the Fish and Wildlife Coordination Act, and other acts promulgated to protect the quality of the environment.

All NWPs that authorize activities which may result in discharges of dredged or fill material into waters of the United States require water quality certification. NWPs that authorize activities within, or affecting land or water uses within a state that has a Federally-approved coastal zone management program, must also be certified as consistent with the state's program. The procedures to ensure that the NWPs comply with these laws are described in 33 CFR 330.4(c) and (d), respectively.

1.4 Public Comment and Response

For a summary of the public comments received in response to the September 26, 2006, Federal Register notice, refer to the preamble in the Federal Register notice announcing the reissuance of this NWP. The substantive comments received in response to the September 26, 2006, Federal Register notice were used to improve the NWP by changing NWP terms and limits, notification requirements, and/or NWP general conditions, as necessary.

We proposed to remove the provision for shellfish seeding, since we proposed to modify NWP 27 to authorize this activity. No comments were received in response to the September 26, 2006, Federal Register notice.

2.0 Alternatives

This evaluation includes an analysis of alternatives based on the requirements of NEPA, which requires a more expansive review than the Clean Water Act Section 404(b)(1) Guidelines. The alternatives discussed below are based on an analysis of the potential environmental impacts and impacts to the Corps, Federal, Tribal, and state resource agencies, general public, and prospective permittees. Since the consideration of off-site alternatives under the 404(b)(1) Guidelines does not apply to specific projects authorized by general permits, the alternatives analysis discussed below consists of a general NEPA alternatives analysis for the NWP.