2010

Habitat complexity and habitat function of native (Crassostrea virginica) and non-native (C. ariakensis) oysters in the Chesapeake Bay region

Heather D. Harwell
College of William and Mary - Virginia Institute of Marine Science

Recommended Citation

Harwell, Heather D., "Habitat complexity and habitat function of native (Crassostrea virginica) and non-native (C. ariakensis) oysters in the Chesapeake Bay region" (2010). *Dissertations, Theses, and Masters Projects.* Paper 1539616687.
https://dx.doi.org/doi:10.25773/v5-vkb6-qz52

Habitat Complexity and Habitat Function of Native (*Crassostrea virginica*) and Non-native (*C. ariakensis*) Oysters in the Chesapeake Bay Region

---

A Dissertation
Presented to
The Faculty of the School of Marine Science
The College of William and Mary in Virginia

In Partial Fulfillment
Of the requirements for the Degree of
Doctor of Philosophy

By
Heather Diane Harwell
2010

APPROVAL SHEET

This dissertation is submitted in partial fulfillment of

The requirements for the degree of

Doctor of Philosophy

Heather D. Harwell

Heather D. Harwell

Approved, by the Committee, May 2010

Standish K. Allen, Jr., Ph.D.
Co-Advisor

Mark Luckenbach

Mark W. Luckenbach, Ph.D.
Co-Advisor

Roger Mann

Roger Mann, Ph.D.

John Brubaker

John Brubaker, Ph.D.

Martin Posey, Ph.D.
University of North Carolina at Wilmington
Wilmington, North Carolina

TABLE OF CONTENTS

ACKNOWLEDGEMENTS

This dissertation would not have been possible without the patience, guidance and support of my co-advisors, Mark Luckenbach and Stan Allen. I especially thank Stan for graciously providing me the opportunity to travel to South Korea to observe the Suminoe oyster in its native habitat, and Mark for preventing me from getting lost among the trees while looking for the forest. I also thank my additional committee members: Roger Mann, for taking time out of his busy schedule as Director of Research and Advisory Services to teach his amazing malacology course to a classroom of one, and for his insightful opinions and support; John Brubaker, for presenting concepts of physical oceanography in such a way that even a biologist like myself could understand them; and Martin Posey, who gave me my start as an undergraduate intern in his lab at UNCW and has been there every step of the way since.

I would also like to thank the many, many people who helped make this work happen. Thanks especially to Peter Kingsley-Smith and Lisa Kellogg, the post-docs who were largely responsible for bringing the experimental design of the field study to fruition. Lisa's impeccable organizational skills were instrumental in the efficient transfer of samples and data between institutions. Peter provided constant support and encouragement throughout my time at VIMS, and was instrumental in editing manuscripts along the way.

Every member of the VIMS Eastern Shore Laboratory has contributed to this study in multiple ways. Thanks to B. Barnes, A. Birch, R. Bonniwell, S. Bonniwell, A. Curry, S. Fate, P. Ross, E. Smith, and L. Ward for all of their help, including field sampling, data collection, sorting samples, filing paperwork, building and maintaining mesocosm equipment, and just making me feel like a part of the crew. VIMS summer interns B. Hammer, W. Hart, S. Mallette, A. Makin, T. Wagner, and C. Vaughn assisted in sampling processing.

Thanks also to Karen Hudson and everyone at the VIMS Aquaculture Breeding and Technology Center for their assistance, as well as Don Merrit's crew at the Horn Point Laboratory. I would also like to thank Ken Paynter for his helpful comments and his lab members at the University of Maryland, M. Chen, J. Goodwin, M. Sherman, and N. Ward, who were instrumental in sample processing.

None of this would have been possible without the undying support and advice from my family. My parents have always been my greatest cheerleaders, especially during the hard times. I am also grateful to Rosia, my best friend and confidant, who has helped keep me grounded all of these years. Finally, thank you to Justin for his patience and love during the writing process. He has taught me so much in such a short time and has become my rock.

LIST OF TABLES

CHAPTER 2

CHAPTER 3

CHAPTER 4

LIST OF FIGURES

CHAPTER 2

CHAPTER 3

CHAPTER 4

ABSTRACT

Complex oyster reefs created by the Eastern oyster, *Crassostrea virginica*, though once prominent features of the Chesapeake Bay region, have become relatively scarce in comparison to historic descriptions. This decline, caused by a combination of overfishing, disease, habitat destruction and pollution, this decline continues despite substantial restoration efforts that have spanned decades. In response to this decline, the states of Virginia and Maryland considered the intentional introduction of the non-native Suminoe oyster, *C. ariakensis*. Previous studies questioned the reef-building capability of this *Crassostrea* species, which may affect its habitat function. Through a combination of field and mesocosm studies, I examined the possibility that this non-native oyster species would provide an ecologically-functional equivalent of the native oyster species if introduced into Chesapeake Bay.

Habitat complexity and associated benthic communities of experimental triploid *C. virginica* and *C. ariakensis* reefs were investigated at four sites of varying salinity, tidal regime, water depth, predation intensity and disease pressure in the Chesapeake Bay region (Virginia and Maryland). Four experimental treatments were established at each site: *C. virginica*; *C. ariakensis*; 50:50 of *C.virginica*: *C. ariakensis*; and shell only. Abundance, biomass, species richness, evenness, dominance and diversity of reef-associated fauna were evaluated in relation to habitat location and oyster species over a period of 21 months.

Habitat complexity varied spatially, although no differences among complexity indices were associated with oyster species. Increases in vertical reef heights and surface rugosity were observed over time for all experimental reefs, and treatment effects were observed after 19 months of development, when *C. ariakensis* reefs exceeded the vertical heights of *C. virginica* reefs, removing any doubt regarding the Suminoe oyster's reef-building capability. Spatial comparisons of reef-associated macrofauna suggested functional equivalency between oyster species with respect to habitat at intertidal locations (where *C. ariakensis* survival was low), and at subtidal sites of low salinity. At subtidal locations of higher salinities, however, the numbers of organisms associated with *C. virginica* reefs per unit of oyster biomass were significantly greater than the numbers of organisms associated with *C. ariakensis*. Multivariate analyses of data from subtidal high salinity sites also revealed unique communities associated with *C. virginica* treatments, while mixed oyster species assemblages were functionally equivalent to mono-specific *C. ariakensis* experimental treatments. Temporal comparisons at one mesohaline subtidal site revealed that the observed effects of oyster species on habitat function in higher salinity locations are inconsistent over time and likely overshadowed by seasonal larval recruitment dynamics and local hydrodynamics.

Though a common oyster reef trophic cascade between juvenile oysters, *C. virginica*; mud crabs, *Panopeus herbstii*; and oyster toadfish, *Opsanus tau* was successfully replicated during mesocosm trials, specific trophic interactions were not significantly affected by oyster substrate species or habitat complexity (as it was defined within the constraints of the experiment). Together, these experiments represent the first effort to quantify the potential habitat function of *C. ariakensis* in Chesapeake Bay, and provide evidence of species-specific similarities and differences in reef-associated communities.

Habitat complexity and habitat function of native (*Crassostrea virginica*) and non-native (*C. ariakensis*) oysters in the Chesapeake Bay region

CHAPTER 1: INTRODUCTION

The decline of the eastern oyster

Named Chesepiooc, or "great shellfish bay," by the Algonquin Native Americans of the region, Chesapeake Bay was once one of the world's greatest sources of oyster (*Crassostrea virginica*) (Gmelin) production (Woods et al. 2005). Following the development of the United States' railroad system, national and international markets were solidified for Chesapeake Bay oysters (Wennerston 1981), and annual oyster harvests in Virginia alone ranged from 5 to 7.5 million bushels from 1894 to 1912 (Hargis and Haven 1988). Once a prominent feature of the ecosystem, complex oyster reef systems have now been reduced to a small fraction of historic levels (Hargis and Haven 1988, Newell 1988, MacKenzie 1996). The continued decline in oyster populations is not unique to the Chesapeake, and has been documented along the entire eastern seaboard, following peak total landings of 27 million bushels from Rhode Island through South Carolina in 1890 (MacKenzie 2007). By 1940, landings from Rhode Island through South Carolina had decreased almost 60% to 11.5 million bushels; by 2004, harvests did not even reach 1 million bushels.

The decline of the native oyster has been attributed in part to increases in mechanical harvesting using dredges and hydraulic-powered tongs, a practice which not only extracts the live oysters, but also the attached shell matrix beneath, leading to reef degradation (Hargis and Haven 1988, Mann 2000). Furthermore, large-scale harvesting results in the destruction of the very structure and habitat on which the oyster population depends (Coen 1995). Other factors contributing to the demise of *C. virginica* are over-fishing (Gross and Smyth 1946, Rothschild et al. 1994), deterioration in water quality (Lenihan and Peterson 1998), and increases in disease pressure, particularly in the past 50 years, from *Haplosporidium nelsoni* (MSX) and *Perkinsus marinus* (Dermo) (Ford and Tripp 1996, Fisher 1996, Lenihan et al. 1999, Mann 2000).

The role of multiple interacting stressors in the decline of the native oyster has complicated the management of this species (Lenihan and Peterson 1998). Substantial state and federal resources have been directed towards efforts to restore the fishery and recreate habitat, both subtidally and intertidally in many of the Atlantic coast's estuaries (Ortega and Sutherland 1992, Luckenbach et al. 1996, Coen et al. 1997, Mann and Powell 2007), often with limited success.

Ecological importance of native oyster reefs

Although *C. virginica* has long been recognized as an important economic estuarine species because of its direct fisheries value, the ecological value of the habitat oysters create and the influence of oysters on estuarine function has been frequently overlooked. Only within the last couple of decades have resource managers begun to consider oyster reefs as critical estuarine habitat (Lenihan and Peterson 1998, Meyer and Townsend 2000). The native oyster provides several critical ecosystem functions, including reduction of water turbidity through active filtration (Newell 1988, Nelson et al. 2004) and decreased water flow (Dame et al. 1984), stabilization of substrate, erosion amelioration (Meyer et al. 1997), habitat provision for many other marine organisms (Coen et al. 1999), and improved benthic-pelagic coupling through the facilitation of the transfer of energy from the benthos to higher trophic levels (Peterson et al. 2003).

Throughout its geographic range, *C. virginica* provides hard substrate and generates 3-dimensional structure in an otherwise 2-dimensional soft substrate environment. Such structure supports a diverse assemblage of organisms generally not found in surrounding soft-bottom habitats (Dame 1979, Zimmerman et al. 1989, Coen et al. 1999, Posey et al. 1999). For example, densities of grass shrimp, xanthid crabs, blue crabs, and benthic fishes associated with oyster reefs have been shown to be enhanced compared to those found in open sand areas (Meyer and Townsend 2000). Many of the organisms found in association with oyster reefs are also known to be important food items for several commercially and recreationally important finfish species in Chesapeake Bay, including croaker, spot, weakfish, sheepshead, blue crabs, and striped bass (Chao and Musick 1997, Rodney and Paynter 2006).

Proposed Exotic Introduction

Since the decline of *C. virginica* in Chesapeake Bay continues despite substantial restoration efforts, the introduction of an exotic oyster species that is resistant to known diseases of the native oyster was until very recently under consideration (Rickards and Ticco 2002). In response to a mandate from the Virginia Legislature, the Virginia Institute of Marine Science initiated field research on the Suminoe oyster, *Crassostrea ariakensis,* in 1998, following unpromising results from investigations of another non-native oyster species, *Crassostrea gigas*. All field trials of *C. ariakensis* to date have employed sterile triploids that have exhibited greater disease resistance, as well as higher growth rates, compared to the native oyster over a variety of salinities (Calvo et al. 2001). There are, however, some caveats to the findings of Calvo et al. (2001), in that triploid non-native oysters were compared to diploid native oysters, and experimental oysters were deployed as cultchless individuals in off-bottom predator-exclusion cages. These caveats likely affected both the survival and growth comparisons of the two species under investigation and did not serve to compare their habitat provision capabilities.

C. ariakensis has long been utilized in aquaculture activities in Japan and China, and, together with *C. honkongensis*, is the most economically important marine shellfish species cultured in South China (Zhang et al. 1995). The Suminoe oyster is thought to occur naturally over a wide latitudinal geographic range (12°N to 34°N), from southern Japan to southern India (Kuroda and Habe 1952). Due to morphologic uncertainty, there is a general lack of information on the basic biology and ecology of this oyster within its native habitat, which makes predicting the ecological impacts of an introduction within Chesapeake Bay much more difficult.

Impacts of previous oyster introductions

The premeditated movement of aquatic species for aquaculture and fishery enhancement purposes has occurred for over 2000 years (Mann et al. 1991). The primary stimuli for the introduction of nonendemic species include economic pressures in the presence of diminishing wild fisheries resources, destruction of a fishery because of disease, and the original nonexistence of a native fishery (Mann 1979). Perhaps the most pervasive examples of aquatic introductions have been oysters, which have been introduced worldwide to 73 countries, and have been permanently established outside their native range in at least 24 (Ruesink et al. 2005).

The Pacific oyster, *Crassostrea gigas*, is the most commonly introduced oyster, and is now established on all major coasts of the Northern Hemisphere, with the exception of the Atlantic Coast of North America. (Shatkin et al.1997). Known for its ability to adapt to a wide range of environmental and hydrographic conditions (Chew 1990), it has become the most ubiquitous oyster in the world and its harvest represents a large percentage of total world production of edible oysters (Ayres 1991). *C. gigas* was first introduced to the West Coast of the United States to supplement dwindling stocks of the native Olympic oyster, *Ostrea conchaphila*. At its peak in 1890, the harvest of wild *O. conchaphila* was over 130,000 bushels prior to its rapid decline (Clark and Langmo 1970), which has been attributed to several factors, including over-harvesting, poor management, disease, and adverse winter weather (Chew 1979). Adult *C. gigas* from Japan were first imported to Puget Sound, Washington in 1902 (Kincaid 1951), following the unsuccessful introduction of the Eastern oyster, *C. virginica* (Chew 1979). The growth rates of the Pacific oyster were much higher than those of *O. conchaphila*, which requires 4 years to reach their maximum size of only 50 mm (Andrews 1980). Large-scale culture in Washington was then established via imports of seed from Miyagi and Kumamoto prefectures (Chew 1979). Plantings in Oregon, California, and British Columbia soon followed (Chew 1987). By 1940, the production of shucked meats from Willapa Bay, Washington, exceeded 3.8 million liters (Sparks and Chew 1961).

Presently, *C. gigas* in Willapa Bay yields about four times more shucked meat weight annually than that of the West Coast native oyster at peak production in the late 1800's (Ruesink et al. 2005). The ease with which Pacific oysters can now be produced on the West Coast has revolutionized the fishery. In fact, the increased availability of larvae and small seed oysters from commercial hatcheries, as well as efficiency improvements in shipping, has resulted in recent transfers of *C. gigas* from the Northwest to other areas worldwide, for both experimental and commercial purposes (Chew 1990).

Whether or not the introduction of *C. gigas* contributed to the decline of the Olympic oyster remains difficult to determine (Beattie 1983). Competition between West Coast native and introduced oysters is expected to be most intense if they share similar habitat (Ruesink et al. 2005). The West Coast native oyster tends to occur at lower depths with less temperature stress than that of *C. gigas*. When overlap of the two species does occur, *C. gigas* grows five times faster than the Olympic oyster, possibly because of its higher per-area filtration rate (Baker 1995). The Pacific oyster is more resistant to environmental stresses and disease, possibly enabling it to out-compete *O. conchaphila* for space in environments suitable for both species (Dinamani 1981). Competition between oyster species can also occur indirectly via habitat modification. In Willipa Bay, *C. gigas* inhabits both feral oyster reefs and planted aquaculture beds, largely in the intertidal zone (Feldman et al. 2000). Neither of these habitat types provides a functional replacement for the dominantly subtidal accumulations of shell where the native oyster previously occurred (Townsend 1896).

In addition, chemicals released by *C. gigas* may inhibit the recruitment of the Olympic oyster (Chew 1979). Native oyster larvae disproportionately settle in areas with large accumulations of shell. Intertidal *C. gigas* comprises most of the shell habitat in the bay. Therefore, the West Coast native oysters predominantly recruit to zones where immersion times are too short for survival (Ruesink et al. 2005). Thus, the introduced oyster has caused a recruitment sink for natives, particularly in the absence of remnant subtidal native oyster reefs. Competition may also occur with species other than oysters. For example, on wave-exposed shores of California, mussels are known to be dominant competitors that reduce growth rates of *C. gigas* by more than 30% (Ruesink, unpublished data).

Introduced oysters are often vectors for other non-native species, and the *C. gigas* introduction to the Pacific Northwest is no exception (Ruesink et al. 2005). Additional species shipped from Japan, often referred to as "piggy-backed introductions," or "hitchhikers," present problems for the continued production of oysters in addition to potentially interacting with native species and altering the structure and function of surrounding communities (Wilson et al. 1988).

In some cases, hitchhikers out-compete and eventually displace native species. For example, the Asian snail *Battilaria attramentaria* out-competes the native mud snail *Cerithidea californica*, causing local extinction of the endemic snail in several West Coast estuaries (Wasson et al. 2001). Other hitchhikers provide structural habitats than can be utilized by various other species, as is the case for *Caulacanthus ustulatus,* an Asian, turf-forming red algae that supports both native and introduced invertebrates (Neto 2000). Parasites are also commonly introduced along with oysters. The shell-boring sabellid polychaete, *Terebrasabella heterouncinata,* introduced in California along with *C. gigas*, infested cultured red abalone, causing large economic losses prior to its eradication (Kuris and Culver 1999). Other invasive species shipped along with the Pacific oyster that have had negative impacts on the oysters themselves include the Japanese oyster drill *Ceratostoma inornatum,* the turbellarian flatworm *Pseudostylochus ostreophagus,* the macrophyte algae *Sargassum muticum,* and the parasitic copepod *Mytilicola orientalis.* (Chew 1990). The Japanese oyster drill is particularly damaging to newly-seeded Pacific oyster crops, and has interfered with attempts to restore native oyster beds (NRC 2004). *M. orientalis*, which originated in Japan, occurs in the lower intestine of oysters and could greatly affect the condition or marketability of several species of bivalves (Chew 1990).

Identification of the problem

Federal Cooperating Agencies (EPA, FWS, and NOAA) prepared a summary of research needs for *C. ariakensis,* identifying seven primary topics for which basic biological information on this species is needed (National Research Council 2002). One of these topics focused on the ecosystem services and functions that *C. ariakensis* may or may not provide if introduced. Whether or not *C. ariakensis* would exhibit a similar level of ecosystem service to that of *C. virginica*, is of particular concern, especially if the introduced species were to out-compete the native species, leaving only the introduced species with ecologically relevant population sizes. Given the now accepted habitat value of *C. virginica*, an evaluation of the provision of habitat by *C. ariakensis* is certainly needed before an introduction should be considered. Current knowledge of growth forms and reef-forming capabilities of *C. ariakensis*, however, remains in question (Zhou and Allen 2003).

In the Ariake Sea, in southern Japan, *C. ariakensis* were found growing in small clumps in mud and on rocks, or growing singly in mud (Luckenbach, pers. comm.). The only reef mounds were found to be predominately aggregates of *C. gigas,* with individual *C. ariakensis* interspersed throughout the reef. Short-term laboratory trials also raise doubts over the ability of *C. ariakensis* to form the dense aggregations observed in *C.*

virginica (Luckenbach, unpublished data); however, long-term trials have yet to be conducted because of quarantine system constraints.

Previous studies of *C. virginica* have shown that certain aspects of reef morphology (e.g., shape, size, and vertical complexity) may influence the degree to which reefs are utilized as habitat by other species (Posey et al., unpublished data). If *C. virginica* and *C. ariakensis* do indeed differ in their reef-forming capabilities, this difference in reef morphology could have dramatic effects on the reef-associated fauna of Chesapeake Bay.

Importance of habitat complexity

Biological structures that have been recognized as providing crucial habitats in marine systems include coral reefs (e.g., Genin et al. 1986), seagrass beds (e.g., Heck and Orth 1980), salt marshes (e.g., Kneib 1984), kelp beds (e.g., Estes and Duggins 1995), foliose aglae (e.g., Kelaher and Rouse 2003), reef-building polychaetes (e.g., Schwindt and Iribarne 2000), mussel beds (e.g., Seed 1996), and oyster reefs (e.g., Coen et al. 1997, Posey et al. 1999). The physical structure of biogenic habitat, including its size, location, and architectural complexity, may influence its ecological function (Bell et al. 1991). For example, the topography, morphology, and structural heterogeneity of oyster reefs often control recruitment, persistence, and diversity of the species inhabiting them, including many commercially important fish and decapod species (Lenihan and Peterson 1998).

Predator-prey interactions can change dramatically in response to habitat complexity (Crowder and Cooper 1982, Grabowski and Powers 2004).

Macroinvertebrate densities and species richness are generally positively correlated with structurally complex habitats (Crowder and Cooper 1982, Diehl 1992, Posey et al. unpublished data), allowing for coexistence of competitors while providing refuge for prey species (Hixon and Menge 1991). Structurally complex areas provide habitat patches where predators are no longer capable of accessing prey resources (Hughes and Grabowski 2006), and predator foraging efficiency generally varies inversely with habitat heterogeneity (Diehl 1992, Beukers and Jones 1997), with individual predators consuming fewer prey in more structurally complex habitats (Grabowski 2004, Warfe and Barmuta 2004). In fact, greater abundances and species diversity found in structured as opposed to unstructured bottom habitats are often attributed to reduced predation within such habitats (Summerson and Peterson 1984, Lenihan et al. 2001, Grabowski et al. 2005). Even if habitat complexity does not completely remove the risk of predation, the created structure can decrease the foraging efficiency of predators by interfering with the ability of the predator to locate and handle its prey (Crowder and Cooper 1982, Summerson and Peterson 1984). For example, in structurally complex shell and sand mixtures, crabs spend more time handling shell fragments, thereby reducing their overall foraging efficiency on clams (Sponagule and Lawton 1990). Alternatively, in systems where competitive interactions between predators are strong, habitat complexity can increase predator foraging efficiency by decreasing encounter rates among predators and reducing interference behavior (Grabowski 2004).

Predator-prey dynamics and trophic cascades

Although foraging in structurally complex environments may be more difficult for intermediate predators, such as xanthid crabs on oyster reefs, the added structure may also reduce the foraging efficiency of higher-order consumers, thereby increasing survivorship of intermediate predators (Diehl 1992, Grabowski 2004). Some studies have shown that top predators motivate many intermediate predators to seek shelter and forage in less than optimal conditions (Wahle 1992, Spanier et al. 1998). Other studies, however, have found that foraging efficiency of top predators is maximized by intermediate or high levels of structural complexity, presumably due to increased prey densities or decreased predator detection within more complex habitats (Crowder and Cooper 1982).

The direct effects of predators on prey populations (i.e., the reduction of prey abundance or biomass) are well documented in marine systems (Paine 1966, Connell 1972). Indirect effects, whether trait-mediated or density mediated, have been posited by some to be as, if not more, important than direct effects in structuring communities (Paine 1966, Wootton 1993, Menge 1995). Density-mediated indirect interactions are those in which one species influences the abundance or biomass of one or several other species. Trait-mediated indirect interactions are those in which a species can mediate trophic interactions through behavioral, chemical, and environmental pathways (Wootton 1993, Menge 1995, Grabowski 2004). Prior to Grabowski (2004), little attempt had been made to determine the relative contributions of density versus trait-mediated indirect interactions to community structure.

Trophic cascades, in which a top predator indirectly benefits basal prey through its effects on an intermediate predator, have been documented in a wide range of systems and are particularly strong in freshwater and marine benthic communities (Carpenter et al. 1985, Shurin et al .2002). Grabowski (2004) examined trophic cascades on oyster reefs and found that habitat complexity weakened the strength of component interactions within a trophic cascade, whereas predator-induced modifications in prey behavior reinforced the cascade. This author conducted a tri-trophic mesocosm study utilizing juvenile oysters (*C. virginica*); mud crabs (*Panopeus herbstii*), a prevalent intermediate predator and important consumer of juvenile oysters (McDermott and Flower 1952), and oyster toadfish (*Opsanus tau*), a common consumer of mud crabs (Wilson et al. 1982). Grabowski (2004) found that on simple reefs, toadfish reduced mud crab densities and induced greater hiding in mud crabs, thereby reducing the magnitude of mud crab predation on juvenile oysters. On more complex reefs, increased habitat complexity inhibited toadfish from feeding on mud crabs, yet toadfish-induced effects on mud crab behavior maintained the indirect effect of toadfish on juvenile oysters. The results of Grabowski (2004) demonstrate that habitat complexity reduces mud crab predation on oysters, thereby further releasing juvenile oysters from predation pressures.

Overview of this research

The research described in the following chapters addresses several of the previously unanswered questions regarding the potential ecological function of habitat which would likely result from the introduction of *C. ariakensis*. Chapter 2 describes a large-scale field experiment in which triploid *C. virginica* and triploid *C. ariakensis* were deployed in cages at four sites in Virginia and Maryland. The physical structure and habitat complexity of these experimental reefs and the community structure of the resident macrofaunal community were then compared for treatments composed of the native species, the non-native species, a mix of the two and shell only controls across the four sites. Chapter 3 investigates the temporal trends in reef complexity and community structure within each of these treatments at a single site in the Patuxent River, Maryland. In combination, these two chapters provide the first available information on how reef complexity and reef-associated communities might compare between *C. virginica* and *C. ariakensis* reefs in Chesapeake Bay, should the latter species be introduced.

Chapter 4 details a series of mesocosm experiments that investigated the roles of differing oyster species and differing levels of reef complexity in affecting a trophic cascade previously described for native oyster reefs. Specifically, the tri-trophic system in which the oyster toadfish, *Opsanus tau*, mediates predation rates of mud crabs, *Panopeus herbstii*, on juvenile oysters via density-mediated and trait-mediated interactions was examined in experimental reefs composed of either *C. virginica* or *C. ariakensis*.

LITERATURE CITED

Andrews, J. 1980. A review of introductions of exotic oysters and biological planning for new importations. Marine Fisheries Review 42: 1-11.

Ayres, P. 1991. Introduced Pacific oysters in Australia. In *The Ecology of* Crassostrea gigas *in Australia, New Zealand, France and Washington State,* ed. M.C. Greer, J.C. Leffler, pp. 3-8. College Park, MD: Maryland Sea Grant Coll. Prog.

Baker, P. 1995. Review of ecology and fishery of the Olymia oyster, *Ostrea conchaphila* with annotated bibliography. Journal of Shellfish Research 14: 501-518.

Beattie, J. 1983. An overview of oyster culture on the Pacific coast. Proceedings of the North American Oyster Workshop. Special Publications 1:11-15.

Bell, S.S., E.D. McCoy, and H.R. Mushinsky, editors. 1991. Habitat structure: the physical arrangement of objects in space. Chapman and Hall, London, U.K.

Beukers, J.S. and G.P. Jones. 1997. Habitat complexity modifies the impact of piscivores on a coral reef fish population. Oecologia 114:50-59.

Calvo, G.W., M.W. Luckenbach, S.K. Allen, Jr. and E.M. Burreson. 2001. A comparative field study of *Crassostrea ariakensis* (Fujita 1913) and *Crassostrea virginica* (Gmelin 1791) in relation to salinity in Virginia. Journal of Shellfish Research 20:221-229.

Carpenter, S.R., J.F. Kitchell, and J.R. Hodgson. 1985. Cascading trophic interactions and lake productivity. BioScience 35:634-639.

Chao, L.N. and J.A. Musick. 1977. Life history, feeding habits and functional morphology of juvenile Sciaenid fishes in the York River estuary, Virginia. U.S. National Marine Fisheries Bulletin 72:657-702.

Chew, K. 1979. The Pacific oyster (*Crassostra gigas*) in the West Coast of the United States. Pp. 54-82. In: R. Mann (Ed.). Exotic Species in Mariculture. The MIT Press, Cambridge, MD.

Chew, K. 1987. Oyster aquaculture in the Pacific Northwest. Fourth Alaska Aquaculture Conference. Sitka, AK. 8 pp.

Chew, K.K. 1990. Global bivalve shellfish introductions: in the global shell game of bivalve introductions there have been some big winners and some big losers. World Aquaculture 21(3): 9-22.

Clark, J. and R. Langmo. 1970. Oyster seed hatcheries of the U.S. West Coast: an overview. Manuscript Report, 28 pp.

Coen, L.D. 1995. A review of the potential impacts of mechanical harvesting on subtidal and intertidal shellfish resources. Report of North Carolina Department of Natural Resources, Marine Resources, Marine Research Institute, Beaufort.

Coen, L.D., E.L. Wenner, D.M. Knott, M.Y. Bobo, N.H. Hadley, D.L. Richardson, B. Stender, and R. Giotta. 1997. Intertidal oyster reef habitat use and function: what have we learned after two years? Journal of Shellfish Research 16:336. Abstract.

Coen, L.D., M.W. Luckenbach, and D.L. Breitburg. 1999. The role of oyster reefs as essential fish habitat: a review of current knowledge and some new perspectives. American Fisheries Society Symposium 22:438-454.

Connell, J.H. 1972. Community interactions on marine rocky intertidal shores. Annual Review of Ecology and Systematics 3:169-192.

Crowder, L.B. and W.E. Cooper. 1982. Habitat structural complexity and the interaction between bluegills and their prey. Ecology 63:1802-1813.

Dame, R.F. 1979. The abundance, diversity and biomass of macrobenthos on North Inlet, South Carolina, intertidal oyster reefs. Proceedings of the National Shellfisheries Association. Vol. 68, pp.6-10.

Dame, R.F., R.G. Zingmark, and E. Haskin. 1984. Oyster reefs as processors of estuarine materials. Journal of Experimental Marine Biology and Ecology 83:239-247.

Diehl, S. 1992. Fish predation and the benthic community structure: the role of omnivory and habitat complexity. Ecology 73:1646-1661.

Dinamani, P. 1991. The Pacific oyster, *Crassostrea gigas* (Thunberg, 1973) in New Zealand. Pp. 343-352. In: W. Menzel (Ed.). Estuarine and marine Bivalve Mollusk Culture. CRC Press, Inc. Boca Raton, FL.

Estes J.A. and D.O. Duggins. 1995. Sea otters and kelp forests in Alaska: generality and variation in a community ecological paradigm. Ecological Monographs 65: 75-100.

Feldman, K.L., D.A. Armstrong, B.R. Dumbauld, T.H. DeWitt, D.C. Doty. 2000. Oysters, crabs, and burrowing shrimp: review of an environmental conflict over aquatic resources and pesticide use in Washington state's (USA) coastal estuaries. Estuaries 23: 141-176.

Fisher W.S. 1996. Preface. Journal of Shellfish Research 15: 1.

Ford, S.E. and M.R. Tripp. 1996. Diseases and defense mechanisms. Pp. 581-660 *In:* V.S. Kennedy, R.I.E. Newll, and A.F. Eble (eds.) The eastern oyster, *Crassostrea virginica.* Maryland Sea Grant, College Park, Maryland, USA.

Genin, A., P.K. Dayton, P.F. Lonsdale, and F.N. Spiess. 1986. Corals on seamounts: evidence of current accelerations over deep-sea topography. Nature 332: 59-61.

Grabowski, J.H. 2004. Habitat complexity disrupts predator-prey interactions but not the trophic cascade on oyster reefs. Ecology 85:995-1004.

Grabowski, J.H. and S.P. Powers. 2004. Habitat complexity mitigates trophic transfer on oyster reefs. Marine Ecology Progress Series 277:291-295.

Grabowski, J.H., A.R. Hughes, D.L. Kimbro, and M.A. Dolan. 2005. How habitat setting influences restored oyster reef communities. Ecology 86:1926-1935.

Gross, F. and R.C. Smyth. 1946. The decline of oyster populations. Nature 157:540-542.

Hargis, W.J., and D.S. Havin. 1988. The imperiled oyster industry of Virginia. VIMS Special Report 290 in Applied Marine Science and Ocean Engineering, Virginia Institute of Marine Science, Gloucester Point,Virginia. 130 pp.

Heck, K. L., Jr. & R. J. Orth. 1980. Structural components of eelgrass (*Zostera marina*) meadows in the lower Chesapeake Bay – Decapod Crustacea. *Estuaries* 3:289-295.

Hixon, M.A., and B.A. Menge. 1991. Species diversity: prey refuges modify the interactive effects of predation and competition. Theorectical Population Biology 39: 178-200.

Hughes, A.R. and J.H. Grabowski. 2006. Habitat context influences predator interference interactions and the strength of resource partitioning. Oecologia 149:256-264.

Kelaher. B.P. and G.W. Rouse. 2003. The role of colonization in determining spatial patterns on *Proscoloplos bondi* sp. nov (Orbiniidae: Annelida) in coralline algal turf, Marine Biology 143:909-917.

Kincaid, T. 1951. The oyster industry of Willapa Bay, Washington. The Tribute, Ilwaco, Washington. 55 pp.

Kneib, R.T. 1984. Pattern of invertebrate distribution and abundance in the intertidal salt marsh: causes and questions. Estuaries 7:392-412.

Kuris, A.M. and C.S. Culer. 1999. An introduced sabellid polychaete pest infesting cultured abalones and its potential spread to other California gastropods. Invertebrate Biology 118: 391-403.

Kuroda, T. and T. Habe. 1952. Check List and Bibliography of the Recent Marine Mollusca of Japan. Tokyo: Leo W. Stach. 210 pp.

Lenihan, H.S. 1999. Physical-biological coupling on oyster reefs: how habitat structure influences individual performance. Ecological Monographs 69:251-275.

Lenihan, H.S., and C.H. Peterson. 1998. How habitat degradation through fishery disturbance enhances impacts of hypoxia on oyster reefs. Ecological Applications 8(1):128-140.

Lenihan, H.S., F. Micheli, S.W. Shelton, and C.H. Peterson. 1999. The influence of multiple environmental stressors on susceptibility to parasites: an experimental determination with oysters. Limnology and Oceanography 44:910-924.

Lenihan, H.S., C.H. Peterson, J.E. Byers, J.H. Grabowski, G.W. Thayer, and D.R. Colby. 2001. Cascading of habitat degradation: oyster reefs invaded by refugee fishes escaping stress. Ecological Applications 11:764-782.

Luckenbach, M.W., J.A. Nestlerode, and G.M. Coates. 1996. Oyster reef restoration: developing relationships between structure and function. Journal of Shellfish Research 16:270-271. Abstract.

MacKenzie, C.L. 1996. Management of natural populations. Pages 707-721 in V.S. Kennedy, R.I.E. Newell, and A.F. Ebele, editors. The eastern oyster, *Crassostrea virginica*. Maryland Sea Grant, College Park, Maryland, USA.

MacKenzie, C.L. 2007. Causes underlying the historical decline in eastern oyster (*Crassostrea virginica* Gmelin, 1791) landings. Journal of Shellfish Research 26 (4):927-938.

Mann, R, ed. 1979. *Exotic Species in Mariculture.* Cambridge, MA: MIT Press. 363 pp.

Mann, R., E.M. Burreson, and P.K. Baker. 1991. The decline of the Virginia oyster fishery in Chesapeake Bay: considerations for introduction of a non-endemic pecies, *Crassostrea gigas* (Thunberg, 1793). Journal of Shellfish Research 10(2): 379-388.

Mann, R. 2000. Restoring the oyster reef communities in the Chesapeake Bay: a commentary. Journal of Shellfish Research 19(1): 335-339.

Mann, R. and E. Powell. 2007. Why oyster restoration goals in the Chesapeake Bay are not and probably cannot be achieved.

McDermott, J.J. and F.B. Flower. 1952. Preliminary studies of the common mud crabs on oyster beds of Delaware Bay. National Shellfisheries Association Convention Address 1952:47-50.

Menge, B.A. 1995. Indirect effects in marine rocky intertidal interaction webs: patterns and importance. Ecological Monographs 65:21-74.

Meyer, D.L., E.C. Townsend, and G.W. Thayer. 1997. Stabilization and erosion control value of oyster cultch for intertidal marsh. Restoration Ecology 5(1):93-99.

Meyer, D.L. and E.C. Townsend. 2000. Faunal utilization of created intertideal eastern oyster (*Crassostrea virginica)* reefs in the southeastern United States. Estuaries 23:34-45.

Nelson, K.A., L.A. Leonard, M.H. Posey, T.D. Alphin, and M.A. Mallin. 2004. Transplanted oyster (*Crassostrea virginica*) beds as self-sustaining mechanisms for water quality improvement in small tidal creeks. Journal of Experimental Marine Biology and Ecology 298:347-368.

Neto, A.I. 2000. Ecology and dynamics of two intertidal algal communities on the littoral of the island of Sao Miguel (Azores). Hydrobiologia 432: 135-147.

Newell, R.I.E. 1988. Ecological changes in Chesapeake Bay: are they the result of overharvesting of the american oyster, *Crassostrea virginica*?, p. 536-546. In: M.P. Lynch and E.C. Knome, editors, Understanding the Estuary: Advances in Chesapeake Bay Research. Chesapeake Bay Research Consortium Publication 129, Solomons, Maryland.

Ortega, S. and J.P. Sutherland. 1992. Recruitment and growth of the eastern oyster, *Crassostrea virginica*, in North Carolina. Estuaries 15:158-170.

Paine, R.T. 1966. Food web complexity and species diversity. American Naturalist 100:65-75.

Peterson, C.H., R.T. Kneib, and C.A. Manen. 2003. Scaling restoration actions in the marine environment to meet quantitative targets of enhanced ecosystem services.

Posey, M.H., T.D. Alphin, C.M. Powell, and E. Townsend. 1999. Use of oyster reefs as habitat for epibenthic fish and decapods. Pp 229-237. In: M.W. Luckenbach, R.Mann, and J.A. Wesson (editors). Oyster reef habitat restoration: a synopsis and synthesis of approaches. Virginia Institute of Marine Science Press.

Rickards, W.L. and P.C. Ticco. 2002. The Suminoe oyster, *Crassostrea ariakensis,* in Chesapeake Bay: current status and near-term research activities. Virginia Sea Grant Publication VSG-02-23. Charlottesville, Virginia. 10p.

Rodney, W.S. and K.T. Paynter. 2006. Comparisons of macrofaunal assemblages on restored and non-restored oyster reefs in Maryland. Journal of Experimental Marine Biology and Ecology 335:39-51.

Rothschild, B.J., J.S. Ault, P. Goulletquer, and M. Heral. 1994. Decline of the Chesapeake Bay oyster population: a century of habitat destruction and overfishing. Marine Ecology Progress Series 111: 29-39.

Ruesink, J.L., H.S. Lenihan, A.C. Trimble, K.W. Heiman, F. Micheli, J.E. Byers, and M.C. Kay. 2005. Annual Review of Ecological and Evolutionary Systems 36: 643-689.

Schwindt, E. and O.O. Iribarne. 2000. Settlement sites, survival and effects on benthos of an introduced reef-building polychaete in a SW Atlantic coastal lagoon. Bulletin of Maine Science 67:73-82.

Seed, R. 1996. Patterns of biodiversity in the macro-invertebrate fauna associated with mussel patches on rocky shores. Journal of the Marine Biological Association of the U.K. 76:203-221.

Shatkin, G., S.E. Shumway, and R. Hawes. 1997. Considerations regarding the possible introduction of the Pacific oyster (Crassostrea gigas) to the Gulf of Maine: a review of global experience. Journal of Shellfish Research 16(2): 463-477.

Shurin, J.B., E.T. Borer, E.W. Seabloom, K. Anderson, C.A. Blanchette, B. Broitman, S.D. Cooper, and B.S. Halpern. 2002. A cross-ecosystem comparison of the strength of trophic cascades. Ecology Letters 5:785-791.

Spanier, E., T.P. McKenzie, J.S. Cobb, and M. Clancy. 1998. Behavior of juvenile American lobsters, *Homarus americanus,* under predation risk. Marine Biology 130:397-406.

Sparks, K. and K. Chew. 1961. Preliminary report on growth and survival of the Pacific oyster in Washington waters. University of Washington, College of Fisheries. Seattle. 8 pp.

Sponaugle, S. and P. Lawton. 1990. Portunid crab predation on juvenile hard clams: effects of substrate type and prey density. Marine Ecology Progress Series 67:43-53.

Summerson, H.C. and C.H. Peterson. 1984. Role of predation in organizing benthic communities of a temperate-zone seagrass bed. Marine Ecology Progress Series 15:63-77.

Townsend, C.H. 1896. *The Transplanting of Eastern Oysters to Willapa Bay, Washington, with Notes on the Native Oyster Industry.* Rep. US Comm. Fish. 1895. Washington DC: Gov. Print. Off.

Wahle, R.A. 1992. Body-size dependent anit-predator mechanisms of the American lobster. Oikos 65:52-60.

Warfe, D.M. and L.A. Barmuta. 2004. Habitat structural complexity mediates foraging success of multiple predator species. Oecologia 141:171-178.

Wasson, K., C.J. Zabin, L. Bediger, D.M. Diaz, J>S. Pearse. Biological invasions of e stuaries without international shipping: the importance of intraregional transport. Biological Conservation 102: 143-153.

Wennerston, J. R. 1981. The oyster wars of the Chesapeake Bay. Centerville, MD: Tidewater Publishers. 147 pp.

Wilson, C.A., J.M. Dean, and R. Radtke. 1982. Age, growth rate, and feeding habits of the oyster toadfish, *Opsanus tau* (Linnaeus) in South Carolina. Journal of Experimental Marine Biology and Ecology 62:251-259.

Wilson, E.A., E.N. Powell, M.E. White, S.M Ray. 1988. The effect of the ectoparasitic snail, *Boonea impressa* on oyster growth and health in the field with comments on patch formation in snail populations (Abstr.) Journal of Shellfish Research 7: 137-138.

Woods, H., W. J. Hargis, Jr., D. H. Hershner and P. Mason. 2005. Disappearance of the natural emergent 3-dimensional oyster reef system of the James River, Virginia, 1871-1948. Journal of Shellfish Research 24:139-142.

Wootton, J.T. 1993. Indirect effects and habitat use in an intertidal community: interaction chains and interaction modifications. American Naturalist 141:71-89.

Zhang, Y., B.L. Munday and H. Handlinger. 1995. Mass mortality of flat oysters (*Ostrea rivularis*) associated with a bloom of *Prorocentrum* sp. in the port of Zhanjiang, South China. Bulletin of the European Association of Fish Pathology 15:61-63.

Zhou, M. and S.K. Allen, Jr. 2003. A review of published work on *Crassostrea ariakensis*. Journal of Shellfish Research 22(1):1-20.

Zimmerman, R., T. Minello, T. Baumer, and M. Castiglione. 1989. Oyster reef as habitat for estuarine macrofauna. Technical Memorandum NMFS-SEFC-249. National Oceanic and Atmospheric Adiministration, Springfield, Virginia.

CHAPTER 2: A COMPARISON OF *CRASSOSTREA VIRGINICA* AND *C. ARIAKENSIS* IN CHESAPEAKE BAY: DOES OYSTER SPECIES AFFECT HABITAT FUNCTION?

Heather D. Harwell [a*], Peter R. Kingsley-Smith [a,b], M. Lisa Kellogg [c], Steven M. Allen [d,e], Standish K. Allen Jr. [f], Donald W. Meritt [d], Kennedy T. Paynter Jr. [d,g], Mark W. Luckenbach [a]

[a] Eastern Shore Laboratory, Virginia Institute of Marine Science,
College of William and Mary, P.O. Box 350, Wachapreague, VA 23480

[b] Current address: Marine Resources Research Institute, South Carolina
Department of Natural Resources, P.O. Box 12559, Charleston, SC 29422-2559

[c] Horn Point Laboratory, 2020 Horns Point Road, University of Maryland Center
for Environmental Science, Cambridge, MD 21613

[d] Department of Biology, University of Maryland, College Park, MD 20742

[e] Current address: Oyster Recovery Partnership, 1805A Virginia Street,
Annapolis, MD 21401 (Main Office)

[f] Aquaculture Genetics and Breeding Technology Center, Virginia Institute of
Marine Science, College of William & Mary, P.O. Box 1346, Gloucester Point,
VA 23062

[g] Chesapeake Biological Laboratory, University of Maryland Center for
Environmental Science, Solomons, MD 20688

In press, *Journal of Shellfish Research*, April 2010

ABSTRACT

We examined the possibility that a non-native oyster species would provide an ecologically-functional equivalent of the native oyster species if introduced into the Chesapeake Bay. Habitat complexity and associated benthic communities of experimental triploid *Crassostrea virginica* and *C. ariakensis* reefs were investigated at four sites of varying salinity, tidal regime, water depth, predation intensity, and disease pressure in the Chesapeake Bay region (Maryland and Virginia). Four experimental treatments were established at each site: *C. virginica*; *C. ariakensis*; 50:50 of *C. virginica*: *C. ariakensis*; and shell only. Abundance, biomass, species richness, evenness, dominance and diversity of reef-associated fauna were evaluated in relation to habitat location and oyster species. Although habitat complexity varied with location, no differences among complexity were associated with oyster species. Similarly, differences in faunal assemblages were more pronounced between sites than within sites. Our results show functional equivalency between oyster species with respect to habitat at the intertidal site and the low salinity, subtidal location. At subtidal sites of higher salinity, however, the numbers of organisms associated with *C. virginica* reefs per unit of oyster biomass were significantly greater than the numbers of organisms associated with *C. ariakensis* reefs. Multivariate analyses of data from subtidal high salinity sites revealed unique communities associated with *C. virginica* treatments, while mixed oyster species assemblages were functionally equivalent to mono-specific *C. ariakensis* experimental treatments. Our study represents the first effort to quantify the potential habitat function of *C. ariakensis*, which has been proposed for an intentional introduction into

Chesapeake Bay, and provides evidence of species-specific similarities and differences in reef-associated communities.

KEY WORDS: *Crassostrea ariakensis*; reef-associated fauna; habitat complexity

INTRODUCTION

Biogenic reefs constructed by the Eastern oyster, *Crassostrea virginica,* provide complex, three-dimensional structural habitats in soft sediment marine systems analogous to those provided by coral reefs (e.g., Genin et al. 1986), seagrass beds (e.g., Heck & Orth 1980), salt marshes (e.g., Kneib 1984), kelp beds (e.g., Estes & Duggins 1995), foliose algae (e.g., Kelaher & Rouse 2003), reef-building polychaetes (e.g., Schwindt & Iribarne 2000), and mussel beds (e.g., Seed 1996). The physical structure of these biogenic habitats, including their size, location, and architectural complexity, may influence ecological function (Bell et al. 1991). For example, the morphology, structural heterogeneity, and vertical complexity of oyster reefs often control the recruitment, persistence and diversity of their inhabitants (e.g., Lenihan & Peterson 1998). Furthermore, macroinvertebrate densities and species richness are generally positively correlated with habitat structural complexity (Crowder & Cooper 1982, Diehl 1992, Posey et al., in prep), which often allows for the coexistence of competitors while providing refuge for prey species (Hixon & Menge 1991).

Dramatic declines in the abundance of *C. virginica* populations in Chesapeake Bay and other localities along the US eastern seaboard have been observed over the past 50 years as a result of the combined stresses of disease (*Haplosporidium nelsoni* [MSX] and *Perkinsus marinus* [Dermo]; Ford & Tripp 1996, Fisher 1996, Lenihan et al. 1999, Mann 2000), over-fishing (Gross & Smyth 1946, Rothschild et al. 1994), deterioration in water quality (Lenihan & Peterson 1998), and reef degradation (Hargis & Haven 1988, Coen 1995, Lenihan & Peterson 1998, Mann 2000). In addition to the loss of a once valuable oyster fishery, these declines have reduced many of the ecological functions once provided by this species.

The native Eastern oyster, *C. virginica,* provides several critical ecosystem services, including reduction of water turbidity through active filtration (Newell 1988, Nelson et al. 2004) and decreased water flow (Dame et al.1984), stabilization of substrate, erosion amelioration (Meyer et al. 1997), habitat provision for many other marine organisms (Coen et al. 1999), and enhanced benthic-pelagic coupling through the transfer of nutrients from the water column to the benthos (Dame, 1999, Dame et al. 2001, Porter et al. 2004) and the facilitation of the transfer of energy from the benthos to higher trophic levels (Peterson et al. 2003). As a result of the three-dimensional structure provided by *C. virginica* that supports a diverse assemblage of organisms generally not found in surrounding soft-bottom habitats (Dame 1979, Zimmerman et al. 1989, Coen et al. 1999, Posey et al. 1999), oyster reefs are now broadly recognized as ecosystem engineers (Luckenbach et al. 1999, Gutiérrez et al. 2003, ASMFC 2007).

Substantial efforts to restore both the fishery resource and habitat value of oyster reefs in many Atlantic coast estuaries (Ortega & Sutherland 1992, Luckenbach et al. 1996; Coen et al. 1997, Mann & Powell 2007) have often been limited in their success. As a result of the continued decline of *C. virginica* in Chesapeake Bay, the introduction of a non-indigenous oyster species (the Suminoe oyster, *C. ariakensis*) that is resistant to known diseases of the native oyster has been under consideration by the states of Maryland and Virginia for most of the last decade (e.g., Rickards & Ticco 2002).

Proposals of deliberate introductions of exotic species raise many concerns. In this case, for example, issues of controversy included possible competitive interactions with the native species for food and space, the possible co-introduction of other non-native species, including pathogens, and the general lack of information on the biology and ecology of the Suminoe oyster, *C. ariakensis* in its native environment. Although an introduction has been tabled at present, the proposal was far from novel, and we expect similar proposals to arise in the future. The pre-meditated movement of aquatic species for aquaculture and fishery enhancement purposes has occurred for over 2000 years (Mann et al. 1991), with oysters being perhaps the most pervasive example (Ruesink et al. 2005). Thus, despite the present abandonment of the proposal in Chesapeake Bay, results of research investigating this proposed introduction may be far-reaching.

Given the accepted habitat value of *C. virginica* reefs, the functional equivalency of *C. ariakensis* reefs should be of concern, particularly given that the growth forms and reef-forming capabilities of *C. ariakensis* remain in question (Zhou & Allen 2003). Functional equivalency, often used as a predictor of restoration success in marine systems (Lockwood & Pimm 2001, Peterson & Lipcius 2003, Peyre et al. 2007), may be especially important if the introduced species were to out-compete the native species in some areas, leaving only the non-native species with ecologically relevant population sizes. Due to morphologic and genetic uncertainties surrounding species identifications in the genus *Crassostrea*, there is a general lack of information on the basic biology and ecology of *C. ariakensis* within its native habitat, making the prediction of the ecological impacts of an introduction of *C. ariakensis* within Chesapeake Bay more difficult. Short-term laboratory trials have also raised doubts over the ability of *C. ariakensis* to form the dense aggregations observed in *C. virginica* (Luckenbach, unpubl. data); however, long-term trials have yet to be conducted due to quarantine system constraints.

Previous studies of *C. virginica* have shown that certain aspects of reef morphology (e.g., shape, size and vertical complexity) may influence the degree to which reefs are utilized as habitat by other species (Posey et al., in prep). If *C. virginica* and *C. ariakensis* differ in their reef-forming capabilities, this could have dramatic effects on the reef-associated fauna of Chesapeake Bay if *C. ariakensis* were to be introduced. While there have been several previous field studies investigating the survival and growth of *C. ariakensis* in Virginia and North Carolina waters, (e.g., Calvo et al. 2001, Grabowski et al. 2005), these trials provide little information on the growth form and reef-building potential of *C. ariakensis*, or the potential competitive interactions between the two *Crassostrea* species. Although several recent studies using diploid *C. ariakensis* and *C. virginica* in quarantined systems (Kingsley-Smith & Luckenbach 2008, Newell et al. unpubl. data, Allen et al. unpubl. data.) have addressed some of these issues, they do not duplicate conditions in natural bottom habitats and therefore have limitations in their applications.

In a recently-completed large-scale field study, Kingsley-Smith et al. (2009) examined the comparative survival, growth and disease dynamics of *C. virginica* and *C. ariakensis* in bottom environments in the Chesapeake Bay region. This project provided the first opportunity for an on-bottom comparison of reef formation, habitat provision and habitat function in *C. virginica* and *C. ariakensis*. Given the ecological importance of habitat provision by the native oyster, *C. virginica* (Luckenbach et al. 2005b, Rodney & Paynter 2006), there is an obvious need to evaluate the functional equivalency of a non-native species prior to an intentional introduction. The objective of the present study was to provide a quantitative comparison of the habitat structure of *C. virginica* and *C.*

ariakensis experimental reefs and of their utilization as habitat by other marine organisms throughout the course of reef development. Our results suggest habitat function may vary between species at some locations within the Chesapeake Bay region. These findings should be included among future considerations of the advantages and concerns surrounding the potential environmental impacts of non-native species introductions.

MATERIALS AND METHODS

Experimental Design

Details of the experimental design are given in Kingsley-Smith et al. (2009), which compared the survival, growth, and disease dynamics of triploid *C. virginica* and triploid *C. ariakensis* in bottom environments across a range of environmental conditions in the Chesapeake Bay region. Briefly, four field sites within the Chesapeake Bay region were selected to encompass a range of tidal environments, predicted salinities, disease pressures, and relative predator abundances (Table 1, Fig. 1).

In late October – early November 2005, four experimental treatments were established at each of the four sites; each site included two blocks with one treatment replicated per block. Experimental triploid oyster treatments were as follows: *C. virginica* only, *C. ariakensis* only, and a 50:50 mixture of the two oyster species. A tray control with no live oysters was also included, comprised of clean *C. virginica* shell. In-depth descriptions of triploid oyster production, setting, and biosecurity precautions can be found in Kingsley-Smith et al. (2009).

Treatment replicates (henceforth referred to as reefs) were established as 5 x 5 arrays of plastic oyster grow-out trays. Each tray (58.4 cm W x 58.4 cm L x 7.3 cm H) was evenly ventilated with 0.6 cm diameter holes (Buckhorn Inc, a Meyers Industries Company). Prior to the start of experiments, all trays were lined with 2 mm fiberglass window screen and filled with a base layer of clean *C. virginica* shell. For live oyster treatments, juvenile oysters were added to achieve a target density of ~400 oysters m^{-2} (= 136 animals $tray^{-1}$). The tray control received oyster shell but no live oysters. Realized initial densities differed slightly across sites and between treatments (Virginia sites: *C. virginica* = 358.1 oysters m^{-2}, *C. ariakensis* = 325.9 oysters m^{-2}, mixed-species treatments = 342.0 oysters m^{-2}; Maryland sites: all treatments = 353.1 oysters m^{-2}). Mean shell heights of *C. virginica* and *C. ariakensis* at deployment were 12.80 mm (n = 1362, SD = 5.68) and 13.85 mm (n = 1272, SD = 5.45), respectively.

As two of our treatments contained non-native oysters, it was necessary to enclose all of our experimental reefs in cages as a biosecurity measure to protect against disturbances, redistributions, and losses of oysters from the experimental plots by extreme weather events and anthropogenic activities. Each array of 25 trays was surrounded by a large metal cage constructed from 3.8 cm diameter galvanized steel pipe and chain-link fence with 5 cm openings. Cages were placed on the seabed at least 1m apart. The 5 cm mesh prevented disturbances by large epibenthic predators, such as cownose rays, while permitting access to the oysters by small benthic predators such as gobies, blennies, and xanthid crabs.

Sampling occurred one month post-deployment and again in spring, summer, and fall of the following two years (2006 and 2007). Using the risk-averse sampling design described by Kingsley-Smith et al. (2009), three trays were removed from each cage at each site during each sampling event, and were replaced with trays filled with clean shell to maintain the spatial integrity of each experimental reef. All 24 trays (3 trays cage^{-1} x 4 treatments x 2 blocks) from a site were sampled on a single day and transported to the laboratory for processing. Due to unforeseen complications discussed at length in Kingsley-Smith et al. (2009), July 2006 was the last sampling period for which all treatment replicates were intact across all sites, and results from that sampling event are to be reported here. Temporal comparisons of habitat complexity and faunal communities will be reported elsewhere (Harwell et al. in prep).

Habitat Complexity

Upon returning to the laboratory, each tray was photographed from the side, maintaining a pre-determined, consistent distance between the camera and each tray. The software program Image-J was used to quantify habitat complexity by obtaining measurements of maximum vertical reef height, average reef height, and surface rugosity from each digital image. Maximum vertical height was defined as the greatest distance between the top of the tray and the growing margin of an oyster protruding upwards from the tray. In addition to the maximum vertical height, measurements were taken for the next nine oyster growing margins judged to be at the greatest perpendicular distance from the upper level of the tray. Average reef heights were calculated as the means of these sets of ten measurements. A unit-less surface rugosity measurement was obtained from digital images of each tray by calculating the ratio of a contoured outline of the oysters within a tray to the linear length of the tray. This was a modified adaptation of the 'chain-length' method, widely used to assess surface topography of coral reefs (Rogers et al. 1983, Aronson & Precht 1995), in which rugosity (R) was calculated as $R = 1\text{-}d/l$, where d is the horizontal distance covered by the chain when conformed to the substratum and l is the length of the chain when fully extended (Aronson & Precht 1995).

Associated Fauna

After the removal of experimental oyster clumps and all *C. virginica* shell material, the remaining contents of each tray were rinsed on a 1-mm mesh sieve, then fixed in 10% buffered formalin for a minimum of 48 hrs prior to sorting, identification, and enumeration of organisms at the lowest practical taxonomic level. Organisms were then preserved in 70% ethanol prior to drying, weighing, and combustion to determine ash-free dry weights. In addition to abundance and biomass data, species richness, Pielou's evenness and Shannon-Weiner diversity were calculated for each sample using the PRIMER software package.

Statistical analyses

Prior to analyses, all data from July 2006 were log-transformed to meet the assumptions of normality (Shapiro-Wilk) and homogeneity of variance (F-max test). Three-way, fixed factor ANOVA models, with site, treatment and block (nested with site) as factors, were used to analyze data for each index of habitat complexity (maximum reef height, average reef height, and surface rugosity) and each of the community metrics (total number of individuals per sample, species richness, Pielou's evenness and Shannon-Weiner diversity). Due to the high prevalence of significant site-treatment interactions, a series of two-way, fixed factor ANOVAs for each main effect (site, treatment) were run within appropriate subsets of the data. In two-way ANOVA models, block effects were generally not significant (block effects were rare) so we removed block from the model and ran one-way ANOVA models. Pair-wise comparisons were made using Tukey's tests when ANOVA indicated a significant site or treatment effect.

A similar series of tests (three-way, fixed-factor ANOVA models followed by reduced two-way and one-way models) were run on the total abundance and biomass of all reef-associated fauna, as well as for individual dominant species. Species comprising at least one percent of the total abundance or biomass of associated fauna at a site were considered dominant. All abundance and biomass data for live oyster treatments were standardized by oyster biomass prior to further analyses and met assumptions of normality (Shapiro-Wilk) and homogeneity of variance (F-max test). When block was not significant, the fixed-factor ANOVA model was reduced and Tukey's test was used to conduct pair-wise comparisons among sites and treatments if ANOVA indicated a significant effect of a main factor.

To further evaluate variations in community structure between treatments, multivariate approaches in the PRIMER statistical software package were also used. Similarity matrices were calculated using non-transformed abundance and biomass data standardized by oyster biomass. These similarity matrices were used to create non-metric multi-dimensional scaling (MDS) plots of each sample at a given site. Analysis of Similarity (ANOSIM), which takes both species composition and abundance into account, was then performed on the similarity matrices in order to determine whether treatment differences were present.

RESULTS

Habitat Complexity

Maximum reef heights for *C. ariakensis* treatments were lowest at the intertidal, high salinity site (Machipongo River, VA), averaging 2.42 cm above the top of the trays (F=16.79, p<0.0001) (Fig. 2). Non-native oysters grown at the subtidal sites exhibited a positive relationship between maximum reef height and salinity, with averages of 3.75 cm, 5.15 cm, and 5.97 cm at the low (Severn River, MD), mid (Patuxent River, MD), and high salinity (York River, VA) subtidal sites, respectively. Statistically, values at the Severn were lower than those at the York, while the intermediate heights observed at the Patuxent were similar to the other two subtidal sites. *C. virginica* maximum heights were also lowest at the intertidal site (mean = 2.19 cm). At subtidal locations, the native species (*C. virginica*) displayed similar maximum reef heights, regardless of salinity (F=11.64, p=0.0001), with average values of 3.78 cm (Severn), 4.74 cm (Patuxent), and 3.47 cm (York). Site effects on maximum reef height for mixed oyster species treatments were similar to those of *C. ariakensis*, with lowest values at the intertidal site (1.83 cm) and increasing heights with increasing salinity at subtidal locations (F=25.51, p<0.0001). Similar heights were observed at all four sites for shell only treatments.

Site effects were also observed for average reef height, where once again, intertidal reefs were shorter than all subtidal reefs, irrespective of oyster treatment (F=47.87, p<0.0001). *C. ariakensis* and mixed species treatments displayed a pattern similar to that observed for maximum height, in that greater average heights were observed at the high salinity subtidal site than at the low salinity site (F=40.07, p<0.0001 and F=31.68, p<0.0001). *C. virginica* reefs, however, achieved higher average heights at the mid salinity subtidal site (Patuxent) than at the high salinity subtidal site (York), with intermediate, overlapping heights found at the low salinity site (Severn) ($F=19.61$, $p<0.0001$). Average heights of shell only treatments were similar across all sites.

Site effects were not found for mean surface rugosity of shell only treatments, nor of native oyster treatments (Fig. 3). In the cases of *C. ariakensis* and mixed reefs, however, at both the York and Severn River sites, higher rugosity values were found than at the Machipongo River site ($F=8.30$, $p<0.001$ and $F=16.4$, $p<0.0001$, respectively), with intermediate, overlapping (i.e., non-significant) values at the Patuxent River site.

Habitat complexity indices differed between live oyster treatments and controls but did not differ among the three live oyster treatments. With the exception of maximum reef height at the Machipongo River, values from live oyster treatments were significantly higher than those without live oysters (Table 2), for all three habitat complexity indices (maximum reef height, average reef height, and surface rugosity), at all four sites. Significant differences between live oyster treatments were not observed for any of the habitat complexity indices used in this study, regardless of location in the Chesapeake Bay region.

Associated Fauna: Community Metrics

Of the community metrics tested in this study, significant treatment effects were rare but site effects were common. The total number of organisms collected and identified from the July 2006 samples was 94,434 individuals with a total biomass of reef-associated fauna in all samples reaching 983.9 g ash-free dry weight (Table 3). The greatest abundances of organisms were found at the two higher salinity subtidal sites, the

York (40,695) and Patuxent (32,419) Rivers. Intermediate numbers were found at the Severn River (17,009), with lowest overall abundances occurring at the Machipongo (4,311) (F=180, $p<0.0001$). This trend of increased total abundances with increased salinity in subtidal sites was seen across all experimental treatments. Without taking oyster biomass into account, the total number of reef-associated organisms was significantly higher in the live oyster treatments than in the shell only treatment (F=6.00, p=0.0011), but similar across oyster treatments, regardless of oyster species.

For *C. ariakensis* reefs, species richness was highest (33.5 species) at the York River (F=24.78, $p<0.0001$) with similar values found at the remaining sites (Severn=11.2, Patuxent=15.0, Machipongo=16.7). Species richness for *C. virginica* reefs was also highest at the York (37.8), with intermediate values in the Patuxent (16.5) and Machipongo (15.5), and lowest values at the Severn site (10.7) (F=114.21, $p<0.0001$). Mixed oyster species reefs exhibited a trend similar to that of the *C. ariakensis* oyster reefs, with highest species richness at the York (33.5) and similar values for the remaining sites (Severn=11.8, Patuxent=16.3, Machipongo=15.5, F=17.86, $p<0.0001$). The numbers of species found in the shell only treatment were highest in the York (34.8), lowest in the Severn (12.8) and Machipongo (12.5), and intermediate in the Patuxent (18.2) (F=73.02, $p<0.0001$).

Pielou's species evenness for *C. ariakensis* treatments was similar across the Machipongo, Severn, and York River sites, with lowest values at the Patuxent site (F=9.36, p=0.0005) (Table 4). Lowest species evenness was also observed at the Patuxent site for all other treatments ($p<0.0001$ in all cases). Species evenness for *C. virginica* reefs was higher at the Machipongo than at the York (F=14.46, $p<0.0001$), with intermediate, overlapping values at the Severn. Mixed species reefs had highest species evenness at the Machipongo River site (F=18.23, p<0.0001) and intermediate values at the York and Severn, while the shell only treatment had the highest species evenness at both the Machipongo and the Severn sites, with intermediate values occurring at the York.

For both single oyster species treatments (*C. virginica* only and *C. ariakensis* only), dominance differed significantly among sites, with the highest values at the York, followed in decreasing order by the Machipongo, then the Patuxent, and finally the Severn (p<0.0001 in all cases). In the mixed treatment, where both oyster species co-existed, a similar trend was observed, with highest values recorded at the York (F=28.76, p<0.0001), although dominance at the Patuxent overlapped values seen at both the Machipongo and Severn River sites. In the absence of live oysters (i.e., shell only treatment) dominance was significantly higher at the York River site (F=45.16, p<0.0001) than at all other sites.

Site effects on Shannon-Weiner diversity, which takes species richness, dominance, and evenness into account, were similar across all live oyster treatments, with the highest species diversity found at the sites of higher salinities, the York and Machipongo (p<0.0001 in all cases). Similarly, lower values of diversity were observed at the Severn and Patuxent across live oyster treatments. The shell only treatment had lower Shannon-Weiner diversity at the Patuxent River site than at all other site (F=12.48, p<0.0001).

Of the community metrics tested in this study, significant treatment effects were rare, and only observed at the low salinity site (Table 4). At the Severn River site, dominance was higher in the shell only treatment than for reefs comprised of only the native oyster, *C. virginica* (F=4.47, p=0.0148). Non-native (*C. ariakensis*) and mixed oyster species reefs exhibited intermediate, overlapping values for dominance. The only other treatment effect on a community metric was higher Shannon-Weiner diversity at the Severn River site in the shell only treatment compared to *C. ariakensis* reefs (F=4.10, p=0.0202).

Associated Fauna: Total Standardized Abundance and Biomass

Oyster survival and growth differed significantly between sites (see Kingsley-Smith et al. 2009 for details). To more accurately compare the effects of oyster species, not oyster survival, on reef-associated communities across sites, the total abundances and ash-free dry weights of reef-associated organisms per sample were standardized by the oyster biomass present (methods described in Kingsley-Smith et al. 2009). ANOVAs for log-transformed data revealed site and treatment effects that differ from the ANOVA results for raw abundance data. Correcting for oyster biomass removed all site effects on the total abundance and biomass of reef-associated fauna. Oyster species did not affect the total number of reef-associated organisms at either the Machipongo River or the Severn River site (Table 5). At the Patuxent and York River sites, however, the average standardized number of organisms associated with *C. virginica* reefs was significantly greater than the number associated with *C. ariakensis* reefs (Patuxent: F=7.77, p=0.0048; York: F=8.42, p=0.0025, Fig. 4). When oyster species coexisted in mixed assemblages, standardized abundances at the Patuxent River were similar to those found in *C. virginica* reefs, whereas those in the York River were similar to *C. ariakensis* reefs.

Standardized total ash-free dry weights of reef-associated fauna also showed treatment effects at the Patuxent and the York River sites but not at the Machipongo or the Severn River sites (Figure 5). Once again, values were higher for *C. virginica* reefs than for *C. ariakensis* reefs (Patuxent: F=4.23, p=0.0350; York: F=5.43, p=0.0169). Mixed oyster species reefs had values that were intermediate, yet overlapping.

Associated Fauna: Species Composition

A total of 78 different species were found throughout this study, including polychaete worms (28 spp.), bivalves (11 spp.), amphipods (10 spp.), crabs (7 spp.), fishes (6 spp.), shrimps (2 spp.), cnidarians (1 sp.) and isopods (1 sp.). A complete list of all species and the sites at which they occurred can be found in Table 6. Standardized abundance and biomass data was used to determine which species dominated samples from each site (Table 7).

Dominant species (defined as those comprising at least one percent of the total abundance of organisms) found at the subtidal site of lowest salinity (Severn) in July 2006 included an errant polychaete (*Neanthes succinea),* three amphipods (*Apocorophium lacustre, Apocorophium simile* and *Melita nitida*), the white-fingered mud crab (*Rhithropanopeus harrisii)*, unidentified juvenile xanthid crabs (all < 5 mm carapace width, CW), and the naked goby, *Gobiosoma bosc.* At this site, standardized abundances of all dominant species, with the exception of the juvenile xanthids, were similar across live oyster treatments. Once standardized by total oyster biomass, *C. virginica* reefs supported higher numbers of the juvenile xanthids than their non-native counterparts (*C. ariakensis*), with intermediate, overlapping values found on reefs of mixed oyster species (F=5.45, p=0.0166).

Dominant species at the Patuxent River site included *N. succinea,* the mud crab *Eurypanopeus depressus,* the amphipods *Gammarus palustris* and *M. nitida,* and the bivalves *Ischadium recurvum, Macoma balthica* and *Mya arenaria.* Here, four of the seven dominants displayed significant treatment effects. Standardized abundances of *N. succinea*, *E. depressus*, *G. palustris*, and *M. arenaria* were all higher in *C. virginica* cages than in *C. ariakensis* cages ($p<0.0196$ in all cases). Mixed oyster species cages also contained significantly lower numbers of *N. succinea*, *E. depressus*, and *G. palustris* compared to *C. virginica* cages, although numbers of *M. arenaria* did not differ from the other live oyster treatments.

At the York River site, dominant species included polychaete worms (*Demonax microphthalmus, Heteromastus filiformis, Loimia medusa,* and *N. succinea*), amphipods (*Caprella penantis, A. lacustre, Elasmopus levis,* and *M. nitida*), the mud crab, *E. depressus*, unidentified juvenile xanthid crabs (all < 5mm CW), the naked goby, *G. bosc,* and the gastropod *Crepidula fornicata*. *C. virginica* reefs at this site supported greater numbers of all dominant species per gram of oyster biomass than *C. ariakensis* reefs ($p\leq0.015$ in all cases). With one exception (*C. fornicata*), when oyster species coexisted, those reefs also supported lower numbers of dominant species per gram of oyster biomass than reefs comprised only of *C. virginica.*

Using standardized abundance data at the high salinity, intertidal Machipongo River site, sixteen dominant species were identified. These included polychaetes (*Leitoscoloplos fragilis*, *Mediomastus ambiseta*, *N. succinea*, *Streblospio benedicti*, and *Tharyx acutus*), the amphipod *M. nitida*, the isopod *Cyathura burbancki*, gastropods (*Boonea impressa, Nassarius vibex*), crabs (*E. depressus*, *Panopeus herbstii*, unidentified juvenile xanthid crabs (all < 5mm CW), and *Uca* spp. (all < 5mm CW)), nemerteans and two insect species. At this site, similar standardized abundances were seen across treatments for all dominant species.

The species comprising at least one per cent of the standardized total biomass at each site were different than dominants calculated using abundance data. At the Severn River site, dominant species for standardized biomass data included three fishes, four crabs, one polychaete, and one bivalve (see Table 7 for species list). At the Patuxent River site, 17 species dominated ash-free dry weights: 6 bivalves, 2 crabs, 2 fishes, 4 amphipods, 1 polychaete, 1 gastropod, and 1 cnidarian. Biomass dominants at the York site included 13 different species: 5 fishes, 3 crabs, 4 polychaetes, and 1 bivalve. At the intertidal site, only 5 species dominated the biomass of reef-associated fauna. These included 3 crabs and 2 gastropods.

Standardized ash-free dry weights of biomass dominants were all similar across live oyster treatments at the Severn, Patuxent, and Machipongo sites. At the York River location, three species exhibited treatment effects: the polychaete *Demonax microphthalmus*, the mud crab *E. depressus*, and the skillet fish *Gobiesox strumosus*. As previously observed for dominant species by abundance, *C. virginica* reefs supported higher biomass of these species than did either *C. ariakensis* or mixed oyster species reefs ($p \leq 0.007$).

Multivariate Analyses

Non-metric MDS plots and Analysis of Similarity (ANOSIM) of standardized abundances of associated fauna highlight treatment effects at both the York and Patuxent

River sites (Fig. 6). The benthic communities associated with *C. virginica* experimental reefs were different from those associated with both *C. ariakensis* and mixed oyster reefs at both the Patuxent and York River sites. Communities associated with the mixed species reefs did not differ from the *C. ariakensis* reefs. Reef-associated communities at the Machipongo and Severn River sites did not differ between treatments. Multivariate analyses utilizing the biomass of associated organisms standardized by oyster biomass yielded results similar to those obtained from abundance data, although treatments effects were only observed at the York River site, where once again, *C. virginica* reef communities differed from those supported by *C. ariakensis* and mixed oyster species reefs (Fig. 7).

DISCUSSION

Although results from short-term laboratory trials and anecdotal observations of *C. ariakensis* in its native range have raised doubt over the species' ability to form the dense aggregations observed in *C. virginica*, a growing body of evidence suggests that the Suminoe oyster is a reef-building *Crassostrea* species. After approximately eight months of deployment, comparisons of three distinct habitat complexity indices (maximum vertical reef height, average reef height and surface rugosity) revealed no significant differences between native and non-native experimental reefs, regardless of location within the Chesapeake Bay region. Despite evidence of negative effects of interspecific competition on the growth of *C. ariakensis* at the low and mid salinity subtidal sites (Kingsley-Smith et al. 2009), experimental reefs containing a mixture of both oyster species had similar measures of habitat complexity when compared to monospecific reefs.

Site effects on complexity indices were common, particularly for treatments containing *C. ariakensis*. Due to its relative intolerance of intertidal exposure (Luckenbach et al. 2005a, Kingsley-Smith & Luckenbach 2008, Wang et al. 2008, Yoon et al. 2008) survival of *C. ariakensis* at our intertidal site, the Machipongo River, VA was markedly low (Kingsley-Smith et al. 2009), negatively affecting all indices of habitat complexity for treatments containing Suminoe oysters at this location. Maximum and average vertical reef heights among native oyster treatments, however, were also significantly lower at this intertidal site. To explore the limits of *C. ariakensis* reef formation, we intentionally placed our experimental reefs near the upper limit of native oyster reefs and acknowledge that this resulted in harsher physical conditions than those occurring on many natural intertidal reefs.

Among subtidal sites, reefs containing *C. ariakensis* (both monospecific and mixed species assemblages) had significantly higher complexity indices at the higher salinity site (York River). We largely attribute this to the positive relationship between salinity and the growth rate of *C. ariakensis* (Calvo et al. 2001, Grabowski et al. 2004, Hudson et al. 2005, Paynter et al. 2008).

Although qualitative differences in reef morphologies are apparent when visually comparing native and non-native treatments, particularly those grown at the higher salinity subtidal sites, such observations did not translate to quantitative differences in any of the complexity indices measured in this study. While treatment effects on maximum and average vertical reef heights were not expected, as there were no discernable visual differences in these characteristics during sampling events, differences in rugosity indices were anticipated. Though the overall amount of interstitial space present within reefs may have been similar among oyster species, the arrangement of that space varied. *C. virginica* reefs contained a tight arrangement of individuals, resulting in a high number of small crevices; *C. ariakensis* reefs grown at high salinity subtidal sites more frequently had growth trajectories in a more horizontal direction, leading to greater angles between individuals. In retrospect, the 'chain link' method of assessing habitat heterogeneity may not have been the most appropriate choice, given the nature of the visual discrepancies in morphology between oyster treatments. Despite its widespread use and general acceptance as an indicator of habitat complexity in aquatic systems, the rugosity index cannot discriminate between the shape and size of components relative to the scale of the topography under investigation (Roberts & Ormond 1987, Shumway et al. 2007). For example, a complex, small-scale topography would have the same rugosity value as a simple, large-scale topography (Roberts & Ormond 1987). We recommend future studies of habitat complexity in oyster reefs include more novel approaches to complexity comparisons, such as measuring the distance between individual oysters, or the angles at which individual oysters meet one another.

Though necessary for biosecurity reasons, the cages in which the oysters were deployed may have indirectly affected oyster survival (Kingsley-Smith et al. 2009), as their presence likely reduced predation rates by limiting the access of larger predators such blue crabs, *Callinectes sapidus*, and cownose rays, *Rhinoptera bonasus*. Particularly at small sizes, *C. ariakensis* shells are structurally weaker than those of the native species, allowing for greater susceptibility to predation (Bishop & Peterson 2006, Newell et al. 2007). Therefore, our use of cages in this study may have disproportionately decreased mortality rates of juvenile *C. ariakensis*, which in turn may have affected habitat complexity. It is also possible that observed similarities in habitat complexity may begin to diverge as the reefs mature beyond the age at which they were assessed in the present study. To address this, additional analyses of reef complexity at later time points (e.g., 2007 sampling events described in Kingsley-Smith et al. 2009) are underway (Harwell et al. in prep).

As was the case for habitat complexity, differences among reef-associated faunal assemblages were more pronounced between sites than between treatments within sites. Lowest overall abundances were found at the intertidal site, reflecting low oyster survival, growth, and habitat complexity. At subtidal sites, there was a trend of increasing total abundance with increasing salinity across experimental treatments. Similarly, the high salinity subtidal site (York River) supported higher species richness, dominance, and diversity values. Our observation of a positive relationship between species richness and salinity in oyster reef communities was first suggested by Wells (1961), who documented that a majority of oyster reef inhabitants were limited in their upstream distribution by a reduction in salinity. A more recent study by Tolley et al. (2005) also revealed that several community metrics (organism abundance, biomass, and diversity) increased downstream in a Florida estuary. Furthermore, those authors observed that salinity appeared to be more important than abundance of living oysters as a predictor of increased community metrics (Tolley et al. 2005). Similarly, our findings suggest that, at least at the scale addressed here, salinity-driven effects on reef biota are greater than those of the reef-forming species in question.

Significant treatment effects on community metrics were rare, even when comparing live oyster treatments with the shell only treatment. Although average total abundance per sample was higher for live oyster reefs than for the shell only treatment, no significant differences in species richness or evenness were detected. At the low salinity location (Severn River, MD), Shannon-Weiner diversity was actually higher for the treatment without live oysters (shell only). The importance of the biological properties of live bivalves in determining the structure of associated macro-invertebrate assemblages has been assessed by several previous studies, many of which have documented similar colonization of mimics, live, and dead bivalves (Crooks & Khim 1999, Tolley & Volety 2005), although that is not always the case (Boudreaux et al. 2006, Norling & Kautsky 2007). Most dead oysters differ structurally from their live counterparts, however, as the valves of dead oysters typically disarticulate within 12 months (Ford et al. 2006). This decrease in vertical height compared to live oyster reefs has been thought to lead to a decrease in habitat function. Summerhayes et al. (2009), however, observed that epibiota were generally more abundant in treatments containing half shells than in those with whole oysters, suggesting that the shells offered additional interstitial space and greater surface area for initial colonization. Whether this relationship changes over time is not known. The results of the present study, as well as previous research, indicate that the effects of live oyster presence on community structure remain poorly understood, largely due to the coupling of oyster presence with increased habitat complexity.

Without taking oyster biomass into account, the total numbers of reef-associated organisms were similar among live oyster treatments, suggesting habitat functional equivalency of *C. ariakensis* and *C. virginica*. However, both oyster survival and growth varied across and within sites in this study (Kingsley-Smith et al. 2009). This analysis simply compares the habitat function of the *surviving* oysters, not the overall capability of the species. By normalizing abundance and biomass of reef-associated fauna in each sample by the oyster biomass we were better able to examine the functional equivalency of the two oyster species with regard to habitat provision. The results support functional equivalency with respect to habitat between oyster species at the intertidal site, as well as the low salinity, subtidal location. At subtidal sites with higher salinities (York and Patuxent), however, habitat function varies between oyster species once the data are normalized. Here, the mean number and biomass of organisms associated with *C. virginica* reefs was significantly greater than the number associated with *C. ariakensis* reefs. This suggests that, if introduced, the non-native oyster may have less potential for habitat provision than the native oyster in subtidal, high and mid salinity regions of Chesapeake Bay. The decreased habitat potential of *C. ariakensis* in these areas may be reinforced by increased predation due to its weaker shell (Bishop & Peterson 2006, Newell et al. 2007), or may be offset by increased growth rates (Calvo et al. 2001, Grabowski et al. 2004, Hudson et al. 2005, Paynter et al. 2008).

Over 75 species were identified during the present study, which is the first investigation of benthic community composition on experimental Suminoe oyster (*C. ariakensis*) reefs in the Chesapeake Bay region. Dominant species varied among locations and included amphipods, bivalves, fishes, decapod crustaceans, gastropods, and polychaete worms. The assemblages collected during this study were similar to those previously reported on restored and natural reefs from temperate waters (Coen et al. 1999, Posey et al. 1999, Rodney & Paynter 2006). Similar to results for community metrics, the number of dominant species generally increased with increasing salinity. Patterns of standardized abundances of individual dominant species largely mirrored those observed for overall abundance, in that no differences were seen at the intertidal site between live oyster treatments, and only one group (juvenile xanthid crabs) had increased numbers on native reefs compared to non-native treatments at the low salinity site. Significant increases in individual species abundances associated with *C. virginica* treatments were seen predominantly at the subtidal site of highest salinity (York), with some occurrences at the upper mesohaline location (Patuxent). We observed increased abundances of all dominant species on native oyster reefs at the York River site and increased abundances of 50% of dominants at the Patuxent River site, further supporting a greater potential for habitat provision by native oysters in subtidal areas of high salinity.

Unlike standardized abundance data for individual dominant species, standardized biomass data for dominant reef-associated fauna revealed very few treatment effects. We observed increased biomass on native oyster treatments for only three dominant species at the high salinity subtidal site: *D. microphthalmus*, a polychaete worm; *E. depressus*, a xanthid crab; and *G. strumosus*, the skillet fish. In all other cases, standardized biomasses of species comprising at least 1% of the total biomass were similar regardless of oyster species. At the York and Patuxent sites, where differences in habitat complexity were visually observed but not quantitatively detected, oyster species had a greater influence on reef-associated species that were dominant in abundance, rather than biomass. In other words, it was mostly the smaller, more prolific organisms that were significantly affected by oyster species. This suggests that oyster species may have significantly impacted the size of organisms able to utilize the reefs as habitat. It appears that *C. virginica* reef communities at this location were comprised of a greater number of relatively smaller individuals, and that those found on *C. ariakensis* reefs, while lower in standardized abundances, were larger in size.

Results from multivariate ANOSIM comparisons, which take into account both species composition and relative abundance, revealed similar patterns to those observed from univariate ANOVA comparisons. Once again, oyster species did not affect community structure at the low salinity (Severn) or intertidal (Machipongo) site. At both subtidal sites of higher salinity (York and Patuxent), the benthic communities associated with *C. virginica* reefs were unique among live oyster treatments, although the mechanism behind this difference remains unclear. Although community differences may have been related to subtle differences in complexity that we were unable to quantify using the rugosity index, larval recruitment dynamics, chemical cues, or other unknown factors may also have been involved.

Although site and treatment effects of oyster species on reef-associated fauna were observed, we acknowledge the limitation of this study in estimating abundance and biomass of faunal assemblages occurring on natural oyster reefs. Cage presence likely reduced predator-prey interactions through the exclusion of larger predators, such as large blue crabs, cownose rays, striped bass (*Morone saxatilis*), sheepshead (*Archosargus probatocephalus*), and oyster toadfish (*Opsanus tau*). A lack of larger predators may have resulted in increased abundances of prey species. It may have also increased the effectiveness of intermediate predators via trait-mediated effects (Grabowski 2004). Although our results should not be directly compared to other studies estimating tertiary production on oyster reefs, the relative comparisons made between our experimental oyster treatments remain valid.

Should an introduction of *C. ariakensis* occur, it is most likely that the two species would co-occur on some reefs. In this regard our results from the mixed species treatment are informative. Where significant differences were found between the benthic communities supported by native and non-native experimental reefs, mixed oyster species treatments most often displayed patterns similar to those of monospecific *C. ariakensis* reefs. Although the mechanism behind this remains unknown and may be unrelated to reef morphology, more robust measures of habitat complexity, such as distance between individual oysters, or the angles at which they meet, may aid in the elucidation of this mechanism.

This study indicates that, if introduced to the Chesapeake Bay region, the ability of *C. ariakensis* to serve as a functional equivalent of the native oyster with respect to habitat provision is likely to vary with location. Poor survival of *C. ariakensis* in intertidal areas suggests that *C. virginica* would be a better provider of habitat in such areas, although differences in intertidal reef communities were not detected here. A degree of functional equivalency is more likely in low salinity subtidal areas, where the growth of the two oyster species is most similar. Pronounced differences in benthic community structure are most likely to occur in the lower reaches of Chesapeake Bay.

ACKNOWLEDGMENTS

The authors thank B. Barnes, A. Birch, R. Bonniwell, S. Bonniwell, R. Bushnell, M. Chen, A. Curry, S. Fate, J. Goodwin, B. Hammer, W. Hart, A. Makin, P. Ross, M.

Sherman, E. Smith, C. Vaughn and N. Ward for assistance in data collection, and D. Gillett for taxonomic and statistical assistance. This research was funded by grants from the NOAA Chesapeake Bay Office (#NA05NMF4571240), Virginia Marine Resources Commission, Maryland Department of Natural Resources, and the Keith Campbell Foundation for the Environment.

LITERATURE CITED

Aronson, R. B. & W. F. Precht. 1995. Landscape patterns of reef coral diversity: A test of the intermediate disturbance hypothesis. *J. Exp. Mar. Biol. Ecol.* 192:1-14.

ASMFC. 2007. The importance of habitat created by shellfish and shell beds along the Atlantic Coast of the U.S. Prepared by L.D. Coen and R. Grizzle with contributions by J. Lowery and K.T. Paynter Jr. 108 pp.

Bell, S. S., E. D. McCoy & H. R. Mushinsky, editors. 1991. Habitat structure: the physical arrangement of objects in space. Chapman and Hall, London, U.K.

Bishop, M. J. & C. H. Peterson. 2006. When *r*-selection may not predict introduced-species proliferation: Predation of a nonnative oyster. *Ecol. Appl.* 16:718-730.

Boudreaux, M. L., J. L. Stiner & L. J. Walters. 2006. Biodiversity of sessile and motile macrofauna on intertidal oyster reefs in Mosquito Lagoon Florida. *J. Shellfish Res.* 25:1079-1089.

Calvo, G. W., M. W. Luckenbach, S. K. Allen, Jr. & E. M. Burreson. 2001. A comparative field study of *Crassostrea ariakensis* (Fujita 1913) and *Crassostrea virginica* (Gmelin 1791) in relation to salinity in Virginia. *J. Shellfish Res.* 20:221-229.

Coen, L. D. 1995. A review of the potential impacts of mechanical harvesting on subtidal and intertidal shellfish resources. *Report of North Carolina Department of Natural Resources, Marine Resources.* Marine Research Institute, Beaufort.

Coen, L. D., E .L. Wenner, D. M. Knott, M. Y. Bobo, N. H. Hadley, D. L. Richardson, B. Stender & R. Giotta. 1997. Intertidal oyster reef habitat use and function: what have we learned after two years? *J. Shellfish Res.*16:336. (Abstract).

Coen, L. D., M. W. Luckenbach & D. L. Breitburg. 1999. The role of oyster reefs as essential fish habitat: a review of current knowledge and some new perspectives. *Am. Fish. Soc. Symp.* 22:438-454.

Crooks, J. A. & H. S. Khim. 1999. Architectural vs. biological effects of a habitat-altering, exotic mussel, *Musculista senhousia*. *J. Exp. Mar. Biol. Ecol.* 240:53-75.

Crowder, L. B. & W. E. Cooper. 1982. Habitat structural complexity and the interaction between bluegills and their prey. *Ecology* 63:1802-1813.

Dame, R.F. 1979. The abundance, diversity and biomass of macrobenthos on North Inlet, South Carolina, intertidal oyster reefs. Proceedings of the National Shellfisheries Association. Vol. 68, pp.6-10.

Dame, R. F., R. G. Zingmark & E. Haskin. 1984. Oyster reefs as processors of estuarine materials. *J. Exp. Mar. Biol. Ecol.* 83:239-247.

Dame, R. F. 1999. Oyster reefs as components in estuarine nutrient cycling: Incidental or controlling? In: M. W. Luckenbach, R. Mann, and J. A. Wesson (Eds). *Oyster reef habitat restoration: a synopsis and synthesis of approaches.* Virginia Institute of Marine Science, Gloucester Point, USA, pp. 267–280.

Dame, R., Bushek, D. & T. Prins. 2001. Benthic suspension feeders as determinants of ecosystem structure and function in shallow coastal waters. In: K. Reise (Ed.). Ecological comparisons of sedimentary shores. *Ecol. Stud.* 151:11-37. Springer-Verlag Berlin Heidelberg, Germany.

Diehl, S. 1992. Fish predation and the benthic community structure: the role of omnivory and habitat complexity. *Ecology* 73:1646-1661.

Estes, J. A. & D. O. Duggins. 1995. Sea otters and kelp forests in Alaska: generality and variation in a community ecological paradigm. *Ecol. Monogr.* 65:75-100.
Fisher W. S. 1996. Preface. *J. Shellfish Res.* 15:1.

Ford, S. E. & M. R. Tripp. 1996. Diseases and defense mechanisms. *In:*
V. S. Kennedy, R. I. E. Newell & A. F. Eble (eds.). The Eastern oyster, *Crassostrea virginica.* Maryland Sea Grant, College Park, Maryland, USA, pp. 581-660.

Ford, S. E., M. J. Cummings & E. N. Powell. 2006. Estimating mortality in natural assemblages of oysters. *Estuar. Coast.* 29:361-374.

Genin, A., P. K. Dayton, P. F. Lonsdale & F. N. Spiess. 1986. Corals on seamounts: evidence of current accelerations over deep-sea topography. *Nature* 332:59-61.

Grabowski, J.H. 2004. Habitat complexity disrupts predator-prey interactions but not the trophic cascade on oyster reefs. Ecology 85:995-1004.

Grabowski, J. H., C. H. Peterson, S. P. Powers, D. Gaskill & H. C. Summerson. 2004. Growth and survivorship of nonnative (*Crassostrea gigas* and *Crassostrea ariakensis*) versus native oysters (*Crassostrea virginica*). *J. Shellfish. Res.* 23:781-793.

Grabowski, J. H., A. R. Hughes, D. L. Kimbro & M. A. Dolan. 2005. How habitat setting influences restored oyster reef communities. *Ecology* 86:1926-1935.

Gross, F. & R. C. Smyth. 1946. The decline of oyster populations. *Nature* 157:540-542.

Gutiérrez, J. L., C. G. Jones, D. L. Strayer & O. O. Iribarne. 2003. Mollusks as ecosystem engineers: the role of shell production in aquatic habitats. *Oikos* 101:79-90.

Hargis, W. J. & D. S. Havin. 1988. The imperiled oyster industry of Virginia. *VIMS Special Report 290 in Applied Marine Science and Ocean Engineering,* Virginia Institute of Marine Science, Gloucester Point, Virginia. 130 pp.

Heck, K. L., Jr. & R. J. Orth. 1980. Structural components of eelgrass (*Zostera marina*) meadows in the lower Chesapeake Bay – Decapod Crustacea. *Estuaries* 3:289-295.

Hixon, M. A., & B. A. Menge. 1991. Species diversity: prey refuges modify the interactive effects of predation and competition. *Theor. Popul. Biol.* 39:178-200.

Hudson, K. L., A. Kozlowski, A. J. Erskine & S. K. Allen, Jr. 2005. Comparative field trials of triploid *Crassostrea ariakensis* with *C. virginica* at eight field sites in the Chesapeake Bay: Growth, mortality, condition, reversion and gametogenesis. *J. Shellfish. Res.* 24:658-659.

Kelaher, B. P. & G. W. Rouse. 2003. The role of colonization in determining spatial patterns on *Proscoloplos bondi* sp. nov (Orbiniidae: Annelida) in coralline algal turf, *Mar. Biol.* 143:909-917.

Kingsley-Smith, P. R. & M. W. Luckenbach. 2008. Post-settlement survival and growth of the Suminoe oyster, *Crassostrea ariakensis*, exposed to simulated emersion regimes. *J. Shellfish Res.* 27:609-618.

Kingsley-Smith P. R., H. D. Harwell, M. L. Kellogg, S. M. Allen, S. K. Allen, Jr., D. W. Meritt, K. T. Paynter, Jr. & M. W. Luckenbach. 2009. Survival and growth of triploid *Crassostrea virginica* (Gmelin, 1791) and *C. ariakensis* (Fujita, 1913) in bottom environments of Chesapeake Bay: implications for an introduction. *J. Shellfish Res.* 28:169-184.

Kneib, R. T. 1984. Pattern of invertebrate distribution and abundance in the intertidal salt marsh: causes and questions. *Estuaries* 7:392-412.

Lenihan, H. S. & C. H. Peterson. 1998. How habitat degradation through fishery disturbance enhances impacts of hypoxia on oyster reefs. *Ecol. Appl.* 8(1):128-140.

Lenihan, H. S., F. Micheli, S. W. Shelton & C. H. Peterson. 1999. The influence of multiple environmental stressors on susceptibility to parasites: an experimental determination with oysters. *Limnol. Oceanogr.* 44:910-924.

Lockwood, J. L. & S. L. Pimm. 2001. When does restoration succeed? In: E. Weiher & P. Keddy (eds.), Ecological Assembly Rules: Perspectives, Advances, Retreats, 1st paperback edition. Cambridge University Press, Cambridge, U.K., pp. 363-392.

Luckenbach, M. W., R. Mann & J.A. Wesson (Eds.) 1999. *Oyster reef habitat restoration. A Synopsis and synthesis of approaches.* Virginia Institute of Marine Science Press, Gloucester Point, VA, 358 pp.

Luckenbach, M. W., R. B. Carnegie, L. D. Coen, K. T. Paynter, Jr. & K. S. Reece. 2005a. Observations on the ecology of *Crassostrea ariakensis*. *J. Shellfish Res.* 24:326-327.

Luckenbach, M. W., L. D. Coen, P. G. Ross & J. A. Stephen. 2005b. Oyster reef habitat restoration: relationships between oyster abundance and community development based on two studies in Virginia and South Carolina. *J. Coast. Res.* 40:64-78.

Luckenbach, M. W., J. A. Nestlerode & G. M. Coates. 1996. Oyster reef restoration: developing relationships between structure and function. *J. Shellfish Res.* 16:270-271. (Abstract).

Mann, R. 2000. Restoring the oyster reef communities in the Chesapeake Bay: a commentary. *J. Shellfish Res.* 19:335-339.

Mann, R., E. M. Burreson & P. Baker. 1991. The decline of the Virginia oyster fishery in Chesapeake Bay: considerations for introduction of a non-endemic species, *Crassostrea gigas* (Thunberg, 1973). *J. Shellfish Res.* 10:379-388.

Mann, R. & E. Powell. 2007. Why oyster restoration goals in the Chesapeake Bay are not and probably cannot be achieved. *J. Shellfish Res.* 26:905-917.

Meyer, D. L., E. C. Townsend & G. W. Thayer. 1997. Stabilization and erosion control value of oyster cultch for intertidal marsh. *Restor. Ecol.* 5:93-99.

Nelson, K. A., L. A. Leonard, M. H. Posey, T. D. Alphin & M. A. Mallin. 2004. Transplanted oyster (*Crassostrea virginica*) beds as self-sustaining mechanisms for water quality improvement in small tidal creeks. *J. Exp. Mar. Biol. Ecol.* 298:347-368.

Newell, R. I. E. 1988. Ecological changes in Chesapeake Bay: are they the result of overharvesting of the American oyster, *Crassostrea virginica*?, In: M. P. Lynch & E.C. Knome, editors, Understanding the Estuary: Advances in Chesapeake Bay Research. *Chesapeake Bay Research Consortium Publication 129,* Solomons, Maryland, pp. 536-546.

Newell, R. I. E., V. S. Kennedy, K. S. Shaw & S. Kristi. 2007. Comparative vulnerability to predators, and induced defense response, of eastern oysters *Crassostrea virginica* and nonnative *Crassostrea ariakensis* oysters in Chesapeake Bay. *Mar. Biol.* 152:449-460.

Norling, P. & N. Kautsky. 2007. Structural and functional effects of *Mytilus edulis* on diversity of associated species and ecosystem functioning. *Mar. Ecol. Prog. Ser.* 351:163-175.

Ortega, S. & J. P. Sutherland. 1992. Recruitment and growth of the eastern oyster, *Crassostrea virginica*, in North Carolina. *Estuaries* 15:158-170.

Paynter, K. T., Jr., J. D. Goodwin, M. E. Chen, N. J. Ward, M. W. Sherman, D. W. Meritt & S. K. Allen, Jr. 2008. *Crassostrea ariakensis* in Chesapeake Bay: growth, disease and mortality in shallow water environments. *J. Shellfish Res.* 27:509-515.

Peterson, C. H. & R. N. Lipcius. 2003. Conceptual progress towards predicting quantitative ecosystem benefits of ecological restorations. *Mar. Ecol. Prog. Ser.* 264:297-307.

Peterson, C. H., J. H. Grabowski & S. P. Powers. 2003. Estimated enhancement of fish production resulting from restoring oyster reef habitat: quantitative valuation. *Mar. Ecol. Prog. Ser.* 264:249-264.

Peyre, M. K., B. Gossman & J. A. Nyman. 2007. Assessing functional equivalency of nekton habitat in enhanced habitats: comparison of terraced and unterraced march ponds. *Estuar. Coast.* 30:526-536.

Porter, E. T., J. C Cornwell, L. P. Sanford & R. I. E. Newell. 2004. Effect of oysters *Crassostrea virginica* and bottom shear velocity on benthic-pelagic coupling and estuarine water quality. *Mar. Ecol. Prog. Ser.* 271:61-75.

Posey, M. H., T. D. Alphin, C. M. Powell & E. Townsend. 1999. Use of oyster reefs as habitat for epibenthic fish and decapods. In: M. W. Luckenbach, R. Mann & J. A. Wesson (editors). *Oyster reef habitat restoration: a synopsis and synthesis of approaches.* Virginia Institute of Marine Science Press, Gloucester Point, USA, pp 229-237.

Rickards, W. L. & P. C. Ticco. 2002. The Suminoe oyster, *Crassostrea ariakensis,* in Chesapeake Bay: current status and near-term research activities. *Virginia Sea Grant Publication VSG-02-23.* Charlottesville, Virginia, 10pp.

Rogers, C. S., M. Gilnack & H. C. Fitz. 1983. Monitoring of coral reefs with linear transects: A study of storm damage. *J. Exp. Mar. Biol. Ecol.* 66:285-300.

Rothschild, B. J., J. S. Ault, P. Goulletquer & M. Heral. 1994. Decline of the Chesapeake Bay oyster population: a century of habitat destruction and overfishing. *Mar. Ecol. Prog. Ser.* 111:29-39.

Ruesink, J. L., H. S. Lenihan, A. C. Trimble, K. W. Heiman, F. Micheli, J. E. Byers & M. C. Kay. 2005. Introduction of nonnative oysters: ecosystem effects and restoration implications. *Annu. Rev. Ecol. Evol. Syst.* 36:643-689.

Roberts, C. M. & R. F. G. Ormond. 1987. Habitat complexity and coral reef fish diversity and abundance on red sea fringing reefs. *Mar. Ecol. Prog. Ser.* 41:1-8.

Rodney, W. S. & K. T. Paynter. 2006. Comparisons of macrofaunal assemblages on restored and non-restored oyster reefs in mesohaline regions of Chesapeake Bay in Maryland. *J. Exp. Mar. Biol. Ecol.* 335:39-51.

Schwindt, E. & O. O. Iribarne. 2000. Settlement sites, survival and effects on benthos of an introduced reef-building polychaete in a SW Atlantic coastal lagoon. *Bull. Mar. Science* 67:73-82.

Seed, R. 1996. Patterns of biodiversity in the macro-invertebrate fauna associated with mussel patches on rocky shores. *J. Mar. Biol. Assoc. U.K.* 76:203-221.

Shumway, C. A., H. A. Hofmann & A. P. Dobberfuhl. 2007. Quantifying habitat complexity in aquatic ecosystems. *Freshwater Biol.* 52:1065-1076.

Summerhayes, S. A., M. J. Bishop, A. Leigh, B. P. Kelaher. 2009. Effects of oyster death and shell disarticulation on associated communities. *J. Exp. Mar. Biol. Ecol.* 379:60-67.

Tolley, S. G. & A. K. Volety. 2005. The role of oysters in habitat use of oyster reefs by resident fishes and decapod crustaceans. *J. Shellfish Res.* 24:1007-1012.

Tolley, S. G., A. K. Volety & M. Savarese. 2005. Influence of salinity on the habitat use of oyster reefs in three Southwest Florida Estuaries. *J. Shellfish Res.* 24:127-137.

Wang, H., G. Zhang, X. Liu & X. Guo. 2008. Classification of common oysters from North Carolina. *J. Shellfish Res.* 27:495-503.

Wells, H. W. 1961. The fauna of oyster beds, with special reference to the salinity factor. *Ecol. Monogr.* 31:239-266.

White, M. E. & E. A. Wilson. 1996. Predators, pests, and competitors. In: V. S. Kennedy, R. I. E. Newell & A. F. Eble, editors. The eastern oyster, *Crassostrea virginica*. Maryland Sea Grant. College Park, MD. Pp 559-579.

Yoon, H. S., H. T. Jung & S. D. Choi. 2008. Suminoe oyster (*Crassostrea ariakensis*) culture in Korea. *J. Shellfish Res.* 27:505-508.

Zhou, M. & S. K. Allen, Jr. 2003. A review of published work on *Crassostrea ariakensis*. *J. Shellfish Res.* 22:1-20.

Zimmerman, R., T. Minello, T. Baumer, & M. Castiglione. 1989. Oyster reef as habitat for estuarine macrofauna. *National Oceanic and Atmospheric Administration, Technical Memorandum NMFS-SEFC-249.*

Table 1. Field site characteristics, predicted disease pressures and relative predator abundances.

Site	Tidal Regime	Depth (m)	Salinity (psu) (Avg; Range)	Predicted Disease Pressure[a]	Predicted Relative Predator Abundance[b]
Severn	subtidal	3-4	Low (9.6; 3-14)	No Dermo, No MSX	Low
Patuxent	subtidal	3-4	Mid (11.6, 8-16)	Low Dermo, No MSX	Moderate
York	subtidal	1-2	High (16.5; 8-22)	High Dermo, High MSX	High
Machipongo	intertidal	0-2	High (25.8; 3-34)	High Dermo, High MSX	Highest

[a] Supporting citation for *a priori* prediction of disease patters across sites: Calvo et al. (1999).

[b] Supporting citation for *a priori* prediction of predation patterns across sites: White & Wilson (1996).

Table 2. Mean (and standard deviation) of habitat complexity indices (maximum reef height, average reef height, surface rugosity) for each treatment by site. F- and p-values from reduced model one-way ANOVAs on the effect of treatment with site. Different letters within a row indicate significantly different values ($p<0.05$, Tukey's test).

	Treatment					
Complexity Index	***C. ariakensis***	***C. virginica***	**Mixed species**	**Shell only**	**F**	**p**
Maximum reef height (cm)						
Severn	3.75 (0.42)^{A}	3.78 (0.73)^{A}	3.25 (0.34)^{A}	2.49 (0.62)^{B}	8.04	0.0010
Patuxent	5.15 (0.87)^{A}	4.74 (0.84)^{A}	4.37 (0.47)^{A}	1.94 (0.90)^{B}	17.24	<0.0001
York	5.97 (1.90)^{A}	3.47 (0.61)^{A}	6.15 (2.46)^{A}	1.81 (0.92)^{B}	11.74	0.0001
Machipongo	2.42 (0.52)^{A}	2.19 (0.45)^{A}	1.83 (0.30)^{A}	1.79 (0.61)^{B}	2.29	0.1112
Average reef height (cm)						
Severn	2.77 (0.28)^{A}	2.72 (0.15)^{A}	2.59 (0.18)^{A}	1.19 (0.29)^{B}	43.87	<0.0001
Patuxent	3.82 (0.46)^{A}	3.42 (0.68)^{A}	3.20 (0.22)^{A}	1.05 (0.82)^{B}	23.65	<0.0001
York	4.03 (1.01)^{A}	2.38 (0.54)^{A}	4.07 (1.33)^{A}	0.59 (0.48)^{B}	30.72	<0.0001
Machipongo	1.36 (0.19)^{A}	1.45 (0.16)^{A}	1.07 (0.25)^{A}	0.59 (0.33)^{B}	14.88	<0.0001
Surface rugosity index						
Severn	1.36 (0.14)^{A}	1.29 (0.15)^{A}	1.35 (0.10)^{A}	1.13 (0.15)^{B}	4.98	0.0096
Patuxent	1.30 (0.04)^{A}	1.29 (0.08)^{A}	1.29 (0.07)^{A}	1.11 (0.09)^{B}	8.50	0.0090
York	1.46 (0.10)^{A}	1.36 (0.15)^{A}	1.50 (0.13)^{A}	1.13 (0.06)^{B}	13.09	<0.0001
Machipongo	1.20 (0.03)^{A}	1.22 (0.04)^{A}	1.13 (0.07)^{A}	1.12 (0.10)^{B}	3.35	0.0408

Table 3. Summary of associated fauna found across all sites in July 2006.

	Severn	Patuxent	York	Machipongo
Number of species	22	35	63	48
Number of individuals	17,009	32,419	40,695	4,311
Biomass of assocaited fuana (g)	167.95	571.05	213.2	31.71
Biomass of oysters (g)	456.11	781.05	1,371.05	22.59
Biomass index for associated fauna*	0.37	0.73	0.16	1.4

*(ash-free dry weight of associated fauna / ash-free dry weight of oysters)

Table 4. Mean (and standard deviation) of community metrics (species richness, Peilou's evenness, Shannon-Weiner diversity, dominance) for each treatment by site. F- and p-values from reduced model one-way ANOVAs on the effect of treatment within site. Different letters within a row indicate significantly different values ($p<0.05$, Tukey's test).

	Treatment					
Community Metric	***C. ariakensis***	***C. virginica***	**Mixed species**	**Shell only**	**F**	**p**
Species Richness (S)						
Severn	11.2 (1.0)	10.7 (1.2)	11.8 (1.2)	12.8 (1.8)	2.91	0.0597
Patuxent	15.0 (3.6)	16.5 (1.5)	16.3 (2.9)	18.2 (1.5)	1.50	0.2444
York	33.5 (6.3)	37.8 (1.9)	33.5 (3.3)	34.8 (6.5)	1.03	0.3995
Machipongo	16.7 (4.9)	15.5 (3.1)	15.5 (6.5)	12.5 (1.6)	0.77	0.5246
Pielou's evenness (J')						
Severn	0.60 (0.90)	0.63 (0.12)	0.61 (0.11)	0.73 (0.05)	2.26	0.1123
Patuxent	0.45 (0.11)	0.42 (0.08)	0.45 (0.08)	0.47 (0.10)	0.22	0.8784
York	0.58 (0.05)	0.58 (0.07)	0.64 (0.05)	0.59 (0.05)	1.58	0.2263
Machipongo	0.72 (0.09)	0.73 (0.06)	0.78 (0.07)	0.76 (0.03)	1.24	0.3230
Shannon-Weiner diversity (H')						
Severn	1.44 (0.18)^{B}	1.50 (0.28)^{AB}	1.50 (0.24)^{AB}	1.85 (0.15)^{A}	4.10	0.0202
Patuxent	1.21 (0.39)	1.18 (0.21)	1.26 (0.29)	1.36 (0.32)	0.36	0.7813
York	2.04 (0.12)	2.08 (0.25)	2.24 (0.19)	2.10 (0.25)	1.20	0.3346
Machipongo	1.98 (0.25)	1.99 (0.11)	2.06 (0.31)	1.90 (0.16)	0.44	0.7245
Dominance						
Severn	1.56 (0.11)^{AB}	1.47 (0.21)^{B}	1.62 (0.21)^{AB}	1.91 (0.28)^{A}	4.47	0.0148
Patuxent	1.92 (0.52)	2.13 (0.18)	2.14 (0.41)	2.48 (0.31)	2.14	0.1276
York	4.32 (0.73)	4.84 (0.31)	4.51 (0.43)	4.66 (0.75)	0.90	0.4569
Machipongo	2.97 (0.65)	2.70 (0.46)	3.04 (0.99)	2.44 (0.28)	0.98	0.4227

Table 5. Mean (and standard deviation) of total abundance and biomass for each treatment by site. F- and p-values from reduced model one-way ANOVAs on the effect of treatment within site. Data for each oyster treatment were standardized by oyster biomass. Different letters within a row indicate significantly different values ($p<0.05$, Tukey's test).

	Treatment				
Associated Fauna	*C. ariakensis*	*C. virginica*	**Mixed species**	**F**	**p**
Standardized Total Abundance					
Severn	23.8 (8.0)	38.9 (12.9)	34.6 (15.1)	2.37	0.1273
Patuxent	22.6 (8.3)^{B}	99.0 (59.3)^{A}	38.3 (13.4)^{A}	7.77	0.0048
York	16.8 (5.8)^{B}	154.1 (116.2)^{A}	15.7 (3.23)^{B}	8.42	0.0035
Machipongo	2296 (3250.0)	191.8 (148.5)	418.0 (546.0)	2.21	0.1447
Standardized Total Biomass					
Severn	0.31 (0.11)	0.35 (01.6)	0.31 (0.12)	0.17	0.8491
Patuxent	0.35 (0.29)^{B}	1.40 (1.04)^{A}	0.57 (0.37)^{AB}	4.23	0.0350
York	0.09 (0.04)^{B}	0.48 (0.36)^{A}	0.15 (0.11)^{AB}	5.43	0.0169
Machipongo	21.6 (32.2)	2.58 (3.14)	3.15 (3.98)	1.97	0.1733

Table 6. Complete list of all species found in July 2006 samples across all sites.

Taxonomic Group	**Species**	**Severn**	**Patuxent**	**York**	**Machipongo**
Amphipods					
	Apocorophium lacustre		X	X	X
	Apocorophium simile	X			
	Caprella equilibra			X	
	Caprella penantis			X	X
	Cymadusa compta			X	
	Elasmopus levis	X		X	X
	Gammarus mucronatus		X	X	
	Gammarus palustris		X	X	X
	Melita nitida	X	X	X	X
	Paracaprella tenuis			X	X
Arthropoda					
	Limulus polyphemus			X	
	Unidentified insect				X
	Unidentified insect larva				X
Bivalves					
	Anadora transversa			X	
	Gemma gemma		X	X	X
	Geukensia demissa		X	X	X
	Ischadium recurvum	X	X		
	Macoma balthica	X	X	X	
	Macoma mitchelli		X		
	Macoma tenta			X	
	Mercenaria mercenaria			X	X
	Mulinia lateralis	X	X	X	
	Mya arenaria	X	X	X	
	Mytilus edulis		X		
Cnidarian					
	U/I jelly			X	X
Decapod Crustaceans					
	Alpheus heterochaelis			X	X
	Callinectes sapidus		X	X	
	Dyspanopeus sayi		X	X	X
	Eurypanopeus depressus	X	X	X	X
	Hexapanopeus angustifrons	X		X	
	Palaemonetes pugio			X	X
	Panopeus herbstii	X	X	X	X
	Rhithropanopeus harrisii	X	X	X	
	Uca spp.				X
Fishes					
	Anguilla rostrata	X		X	
	Chasmodes bosquianus		X	X	
	Gobiesox strumosus	X	X	X	
	Gobiosoma bosc	X	X	X	X
	Hypsoblennius hentz			X	

Table 6 cont'd. Complete list of all species found in July 2006 samples across all sites.

Taxonomic Group	**Species**	**Severn**	**Patuxent**	**York**	**Machipongo**
Fishes					
	Opsanus tau			X	
Gastropods					
	Astyris lunata		X	X	
	Acteocina canaliculata		X	X	
	Boonea bisuturalis		X	X	
	Boonea impressa		X	X	X
	Crepidula convexa			X	
	Crepidula fornicata			X	X
	Crepidula plana			X	
	Nassarius vibex		X	X	X
	Neverita duplicata				X
	Rictaxis punctostriatus	X	X	X	
	Urosalpinx cinerea			X	X
Isopods					
	Cyathura burbancki				X
Nemerteans				X	X
Polychaetes					
	Capitella capitata				X
	Clymenella torquata			X	
	Cyrtopleura costata			X	
	Demonax microphthalmus			X	
	Edotia triloba		X		
	Eteone heteropoda		X		X
	Glycera dibranchiata		X		
	Hemipodus roseus			X	
	Heteromastus filiformis		X	X	X
	Hobsonia florida				X
	Hydroides dianthus			X	
	Leitoscoloplos fragilis			X	X
	Lepidontus sublevis			X	
	Loimia medusa			X	X
	Scoletoma tenuis				X
	Lysidice ninetta				X
	Mediomastus ambiseta				X
	Neanthes succinea	X	X	X	X
	Parahesione luteola			X	X
	Pectinaria gouldii		X	X	
	Petriocolaria pholadiformis			X	
	Piromis eruca				X
	Podarke obscura				X
	Polydora websteri				X
	Sabellaria vulgaris			X	
	Streblospio benedicti			X	X
	Stylocus sp.	X	X		
	Tharyx acutus				X

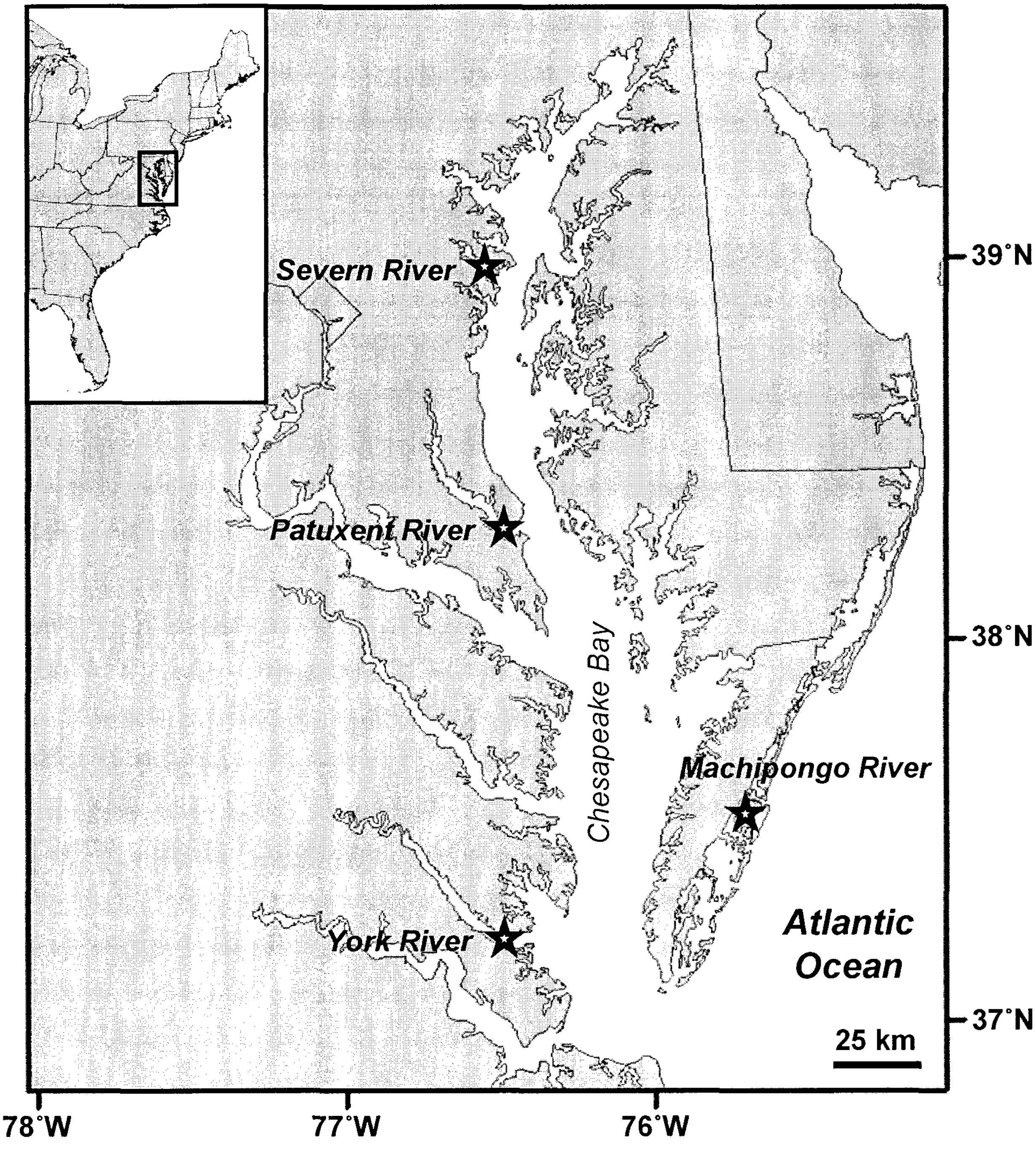
Table 7. Reduced-model one-way ANOVA results for the effect of live oyster treatment on individual species abundances comprising at least one per cent of total standardized abundance at each site. Treatments are ranked in descending order; different letters within a row indicate significantly different values (Tukey's test).

Site	Species	F	p	Ranking
Severn				
	Apocorophium lacustre	2.52	0.1140	
	Apocorophium simile	1.66	0.2230	
	Gobiosoma bosc	0.72	0.5033	
	Melita nitida	1.34	0.2908	
	Neanthes succinea	0.35	0.7101	
	Rhithropanopeus harrisii	1.91	0.1823	
	U/I juvenile xanthid	5.45	0.0166	*C. v.*^{A} mixed^{AB} *C. a.*^{B}
Patuxent				
	Eurypanopeus depressus	10.35	0.0015	*C. v.*^{A} mixed^{B} *C. a.*^{B}
	Gammarus palustris	8.61	0.0032	*C. v.*^{A} mixed^{B} *C. a.*^{B}
	Ischadium recurvum	2.64	0.1039	
	Macoma balthica	0.82	0.4583	
	Melita nitida	0.99	0.3951	
	Mya arenaria	5.17	0.0196	*C. v.*^{A} mixed^{AB} *C. a.*^{B}
	Neanthes succinea	13.02	0.0005	*C. v.*^{A} mixed^{B} *C. a.*^{B}
York				
	Apocorophium lacustre	10.30	0.0015	*C. v.*^{A} mixed^{B} *C. a.*^{B}
	Caprella penantis	5.63	0.0150	*C. v.*^{A} mixed^{B} *C. a.*^{B}
	Crepidula fornicata	4.97	0.0221	*C. v.*^{A} mixed^{B} *C. a.*^{B}
	Demonax microphthalmus	28.22	<0.0001	*C. v.*^{A} mixed^{AB} *C. a.*^{B}
	Elasmopus levis	10.30	0.0015	*C. v.*^{A} mixed^{B} *C. a.*^{B}
	Eurypanopeus depressus	18.12	<0.0001	*C. v.*^{A} mixed^{B} *C. a.*^{B}
	Gobiosoma bosc	7.03	0.0070	*C. v.*^{A} mixed^{B} *C. a.*^{B}
	Heteromastus filiformis	19.69	<0.0001	*C. v.*^{A} mixed^{B} *C. a.*^{B}
	Loimia medusa	7.71	0.0050	*C. v.*^{A} mixed^{AB} *C. a.*^{B}
	Melita nitida	22.06	<0.0001	*C. v.*^{A} mixed^{B} *C. a.*^{B}
	Neanthes succinea	37.01	<0.0001	*C. v.*^{A} mixed^{B} *C. a.*^{B}
	U/I juvenile xanthid	12.17	0.0007	*C. v.*^{A} mixed^{B} *C. a.*^{B}
Machipongo				
	Boonea impressa	0.02	0.9805	
	Cyathura burbancki	2.76	0.0955	
	Eurypanopeus depressus	0.31	0.7372	
	Leitoscoloplos fragilis	0.35	0.7117	
	Mediomastus ambiseta	1.96	0.1758	
	Melita nitida	0.19	0.8260	
	Nassarius vibex	0.74	0.4946	
	Neanthes succinea	1.63	0.2294	
	Nemertean	0.63	0.5460	
	Panopeus herbstii	1.39	0.2797	
	Streblospio benedicti	1.00	0.3898	
	Tharyx acutus	0.08	0.9260	
	Uca spp.	1.12	0.3510	
	U/I insect	2.71	0.0988	
	U/I insect larva	2.64	0.1041	
	U/I juvenile xanthid	0.51	0.6093	

Table 8. Reduced-model one-way ANOVA results for the effect of live oyster treatment on individual species biomass comprising at least one per cent of total standardized biomass at each site. Treatments are ranked in descending order; different letters within a row indicate significantly differer values ($p<0.05$, Tukey's test).

Site	Species	F	p	Ranking
Severn				
	Anguilla rostrata	0.62	0.5513	
	Eurypanopeus depressus	2.37	0.1276	
	Gobiesox strumosus	1.11	0.3563	
	Gobiosoma bosc	0.20	0.8178	
	Mya arenaria	0.80	0.4688	
	Neanthes succinea	0.04	0.9566	
	Panopeus herbstii	0.79	0.4708	
	Rhithropanopeus harrisii	1.95	0.1777	
	U/I juvenile xanthid	2.50	0.1154	
Patuxent				
	Apocorophium lacustre	2.52	0.1141	
	Boonea bisuturalis	0.51	0.6090	
	Eurypanopeus depressus	1.81	0.1982	
	Gammarus mucronatus	1.29	0.3039	
	Gammarus palustris	2.14	0.1524	
	Gemma gemma	1.23	0.3190	
	Gobiesox strumosus	1.04	0.3762	
	Gobiosoma bosc	0.91	0.4229	
	Ischadium recurvum	1.23	0.3192	
	Macoma balthica	1.54	0.2457	
	Macoma mitchelli	1.30	0.3021	
	Melita nitida	0.40	0.6756	
	Mulinia lateralis	0.86	0.4423	
	Mya arenaria	2.45	0.1201	
	Neanthes succinea	0.18	0.8339	
	Panopeus herbstii	1.79	0.2005	
	U/I cnidarian	1.17	0.3375	
York				
	Anguilla rostrata	0.97	0.4003	
	Callinectes sapidus	1.04	0.3770	
	Chasmodes bosquianus	2.02	0.1666	
	Demonax microphthalmus	10.97	0.0012	*C. v.* ^{A} mixed^{B} *C. a.* ^{B}
	Eurypanopeus depressus	10.73	0.0013	*C. v.* ^{A} mixed^{B} *C. a.* ^{B}
	Gobiesox strumosus	6.94	0.0074	*C. v.* ^{A} mixed^{B} *C. a.* ^{B}
	Gobiosoma bosc	2.14	0.1527	
	Heteromastus filiformis	0.81	0.4642	
	Hypsoblennius hentz	0.68	0.5226	
	Loima medusa	0.99	0.3935	
	Mya arenaria	1.58	0.2394	
	Neanthes succinea	3.51	0.0562	
	Panopeus herbstii	0.59	0.5654	
Machipongo				
	Astyris lunata	1.00	0.3911	
	Boonea impressa	1.09	0.3628	
	Dyspanopeus sayi	0.78	0.4779	
	Eurypanopeus depressus	0.70	0.5117	
	Panopeus herbstii	0.88	0.4370	

Figure 2-1. Study site locations throughout the Chesapeake Bay region.



Severn River
Patuxent River
Chesapeake Bay
Machipongo River
York River
Atlantic Ocean
25 km
39°N
38°N
37°N
78°W
77°W
76°W

Figure 2-2. Mean maximum (black bars) and average (white bars) 'reef' heights of experimental treatments at each site in July 2006: (A) Severn, (B) Patuxent, (C) York and (D) Machipongo.

Data are expressed as mean reef height (cm) as measured from the top of each tray. Error bars represent the standard error of each mean, and different letters over bars indicate significantly different values ($p<0.05$, Tukey's test).

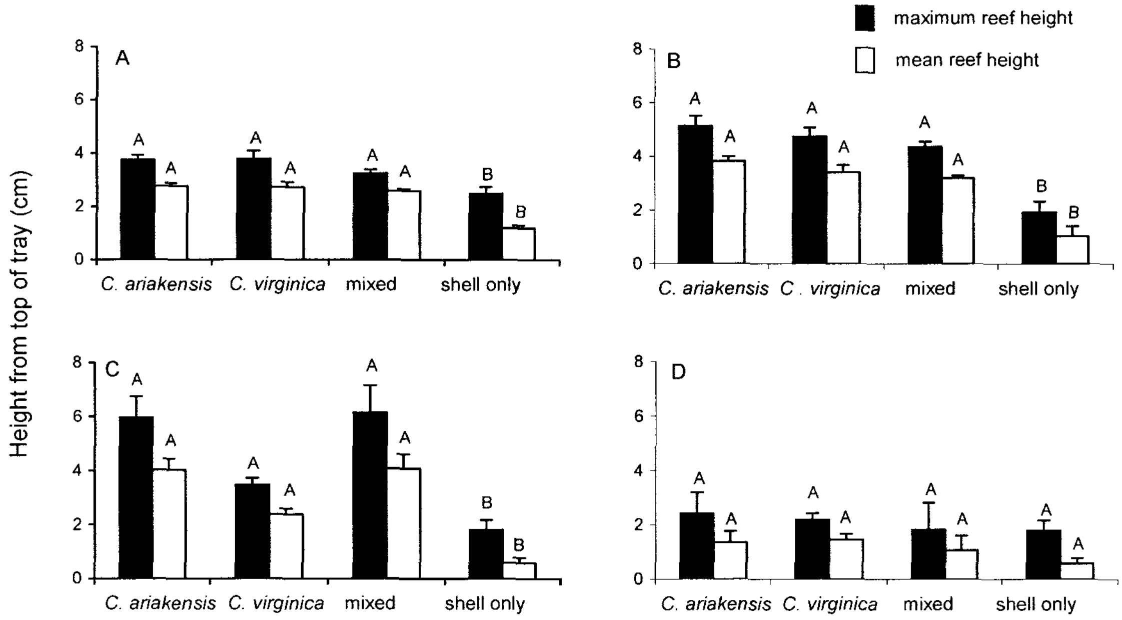

maximum reef height
mean reef height
Height from top of tray (cm)
A
B
C
D
0
2
4
6
8
C. ariakensis
C. virginica
mixed
shell only
A
B

Figure 2-3. Mean surface rugosity index values of experimental treatments at each site in July 2006: (A) Severn, (B) Patuxent, (C) York and (D) Machipongo.

Unit-less surface rugosity measurements were obtained from digital images by calculating the ratio of a contoured outline of the oysters within a tray to the linear length of the tray. Error bars represent the standard error of each mean, and different letters over bars indicate significantly different values ($p<0.05$, Tukey's test).

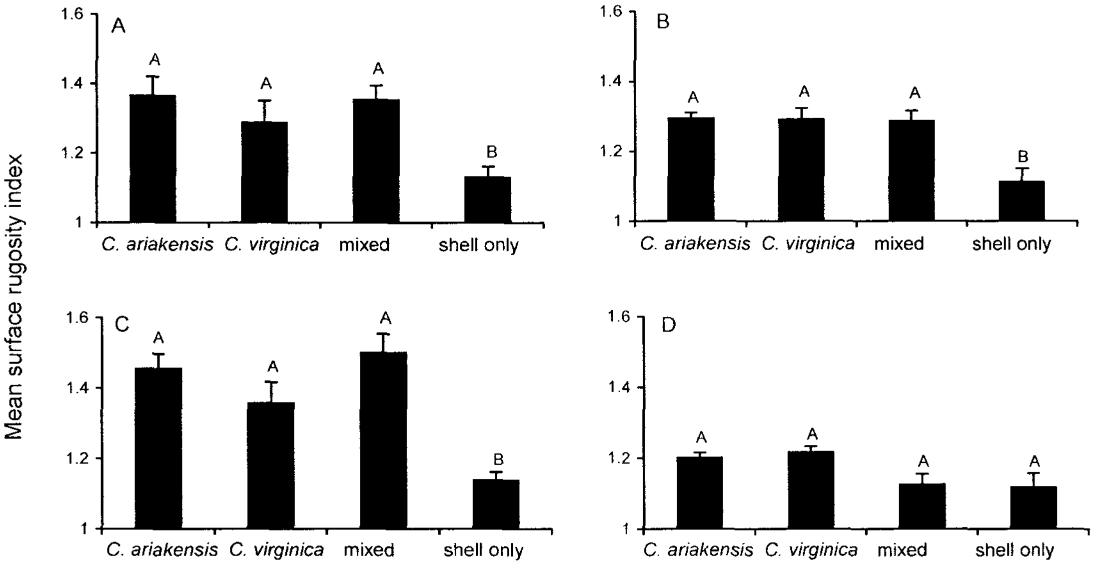
Mean surface rugosity index
A
1.6
1.4
1.2
1
A
A
A
B
C. ariakensis
C. virginica
mixed
shell only
B
1.6
1.4
1.2
1
A
A
A
B
C. ariakensis
C. virginica
mixed
shell only
C
1.6
1.4
1.2
1
A
A
A
B
C. ariakensis
C. virginica
mixed
shell only
D
1.6
1.4
1.2
1
A
A
A
A
C. ariakensis
C. virginica
mixed
shell only

Figure 2-4. Mean total abundance of associated fauna per sample standardized by oyster biomass for all treatments containing live oysters across all sites: (A) Severn, (B) Patuxent, (C) York and (D) Machipongo.

Significant within-site treatment effects are indicated by differing letters above the standard error bars.

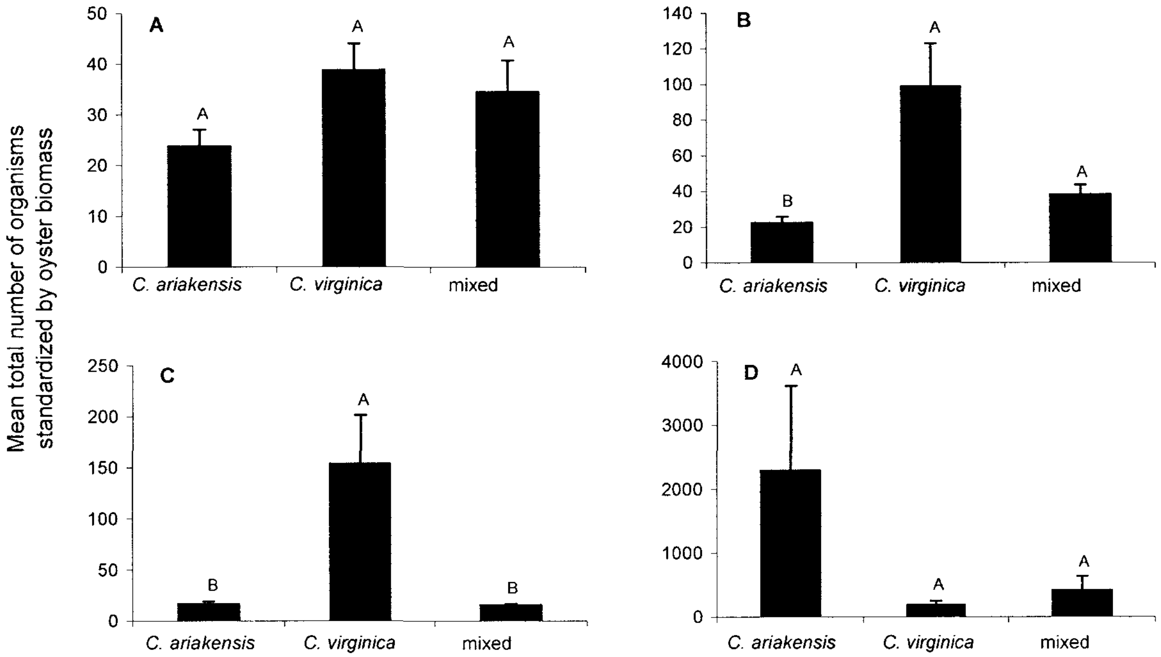

Mean total number of organisms
standardized by oyster biomass
A
50
40
30
20
10
0
A
A
A
C. ariakensis
C. virginica
mixed
B
140
120
100
80
60
40
20
0
B
A
A
C. ariakensis
C. virginica
mixed
C
250
200
150
100
50
0
B
A
B
C. ariakensis
C. virginica
mixed
D
4000
3000
2000
1000
0
A
A
A
C. ariakensis
C. virginica
mixed

Figure 2-5. Mean total biomass of associated fauna per sample standardized by oyster biomass for all treatments containing live oysters across all sites: (A) Severn, (B) Patuxent, (C) York and (D) Machipongo.

Significant treatment effects are indicated by differing letters above the standard error bars.

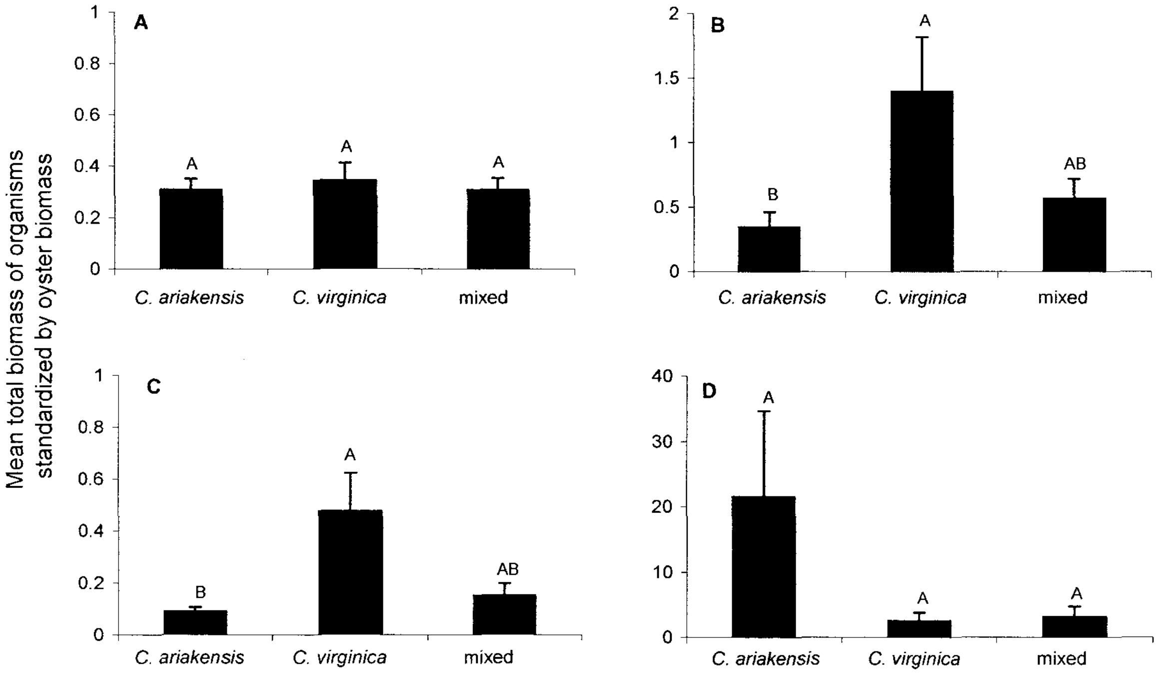

Mean total biomass of organisms standardized by oyster biomass
A
B
C
D
C. ariakensis
C. virginica
mixed
0
0.2
0.4
0.6
0.8
1
0.5
1.5
2
10
20
30
40
A
B
AB

Figure 2-6. Non-metric MDS plots for standardized abundances of associated fauna at all sites: (A) Severn, (B) Patuxent, (C) York and (D) Machipongo.

Significant ANOSIM results were found only at the Patuxent and York River sites, where the *C.virginica* treatment differed significantly from both the *C.ariakensis* only and mixed species treatments.

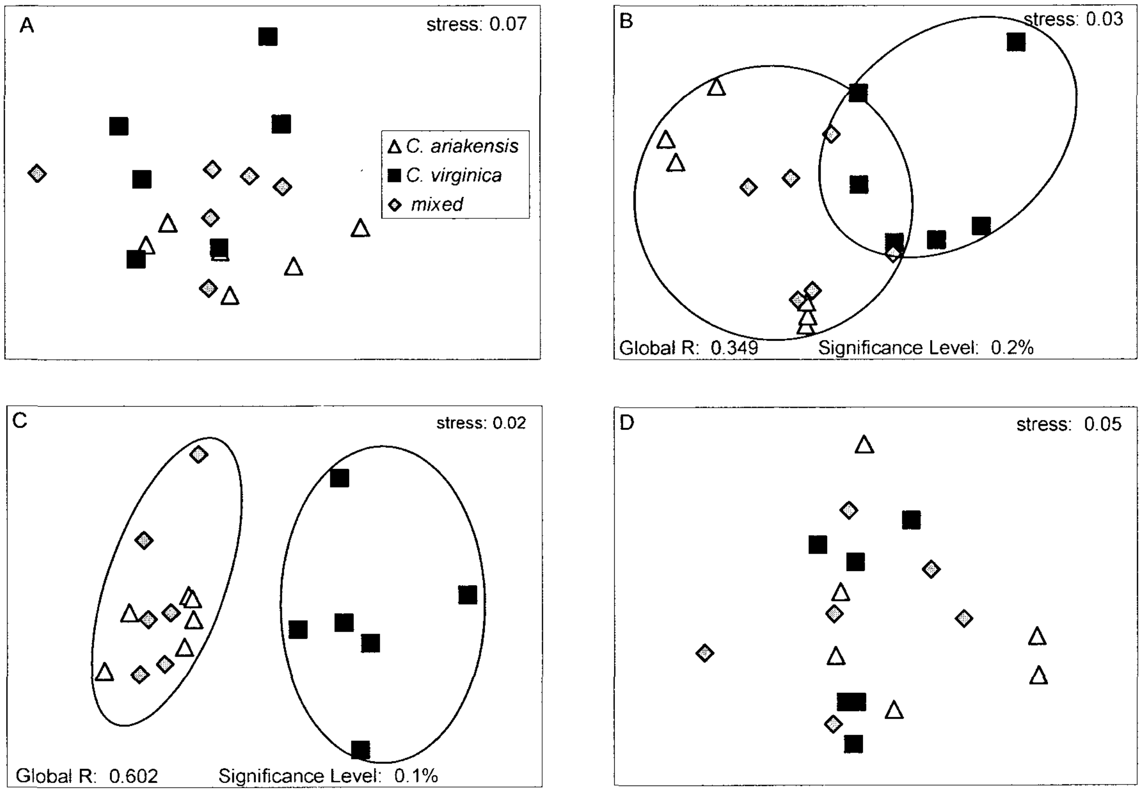
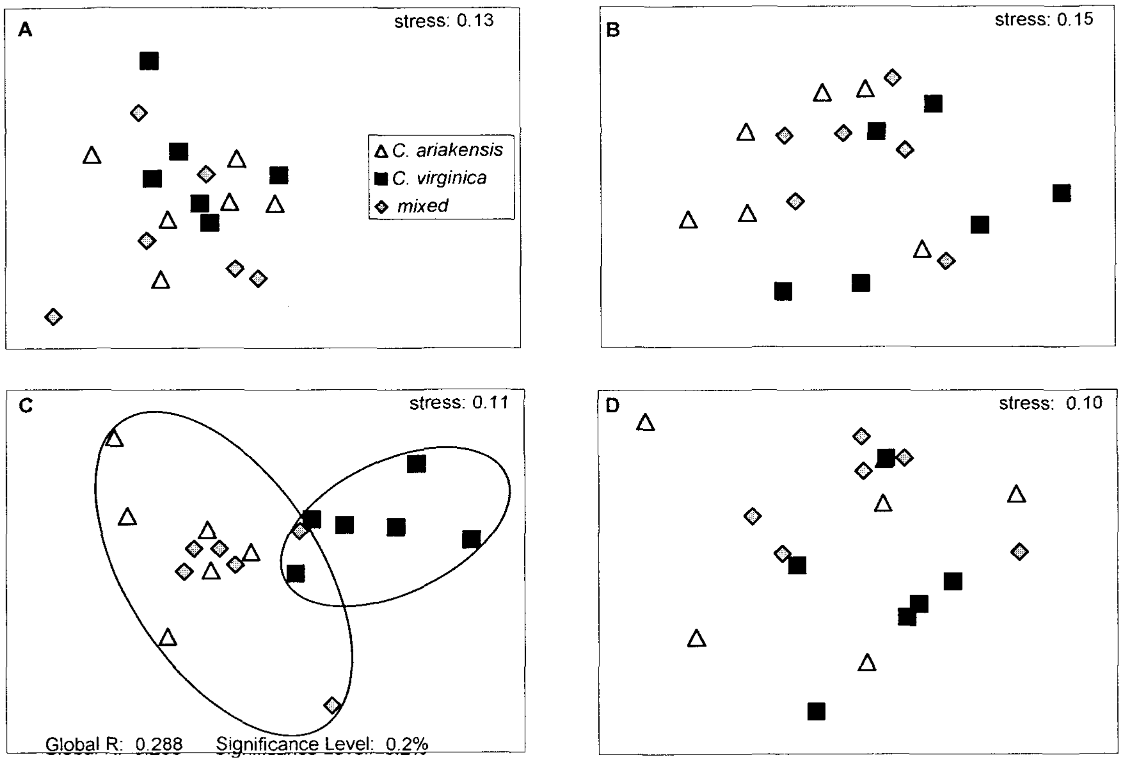

A
stress: 0.07
Δ C. ariakensis
■ C. virginica
◇ mixed
B
stress: 0.03
Global R: 0.349
Significance Level: 0.2%
C
stress: 0.02
Global R: 0.602
Significance Level: 0.1%
D
stress: 0.05

Figure 2-7. Non-metric MDS plots for standardized biomass of associated fauna at all sites: (A) Severn, (B) Patuxent, (C) York and (D) Machipongo.

Significant ANOSIM results were found only at the York, where the *C. virginica* treatment differed significantly from both the *C.ariakensis* only and mixed species treatments.


A
stress: 0.13
△ C. ariakensis
■ C. virginica
◇ mixed
B
stress: 0.15
C
stress: 0.11
Global R: 0.288
Significance Level: 0.2%
D
stress: 0.10

CHAPTER 3: OYSTER REEF DEVELOPMENT AND ASSOCIATED FAUNAL COMMUNITIES: A TEMPORAL COMPARISON OF *C. VIRGINICA* AND *C. ARIAKENSIS* IN THE PATUXENT RIVER, MARYLAND, USA

Heather D. Harwell [a*], Peter R. Kingsley-Smith [a,b], M. Lisa Kellogg [c], Steven M. Allen [d,e], Standish K. Allen Jr. [f], Donald W. Meritt [d], Kennedy T. Paynter Jr. [d,g], Mark W. Luckenbach [a]

[a] Eastern Shore Laboratory, Virginia Institute of Marine Science,
College of William and Mary, P.O. Box 350, Wachapreague, VA 23480

[b] Current address: Marine Resources Research Institute, South Carolina Department of Natural Resources, P.O. Box 12559, Charleston, SC 29422-2559

[c] Horn Point Laboratory, 2020 Horns Point Road, University of Maryland Center for Environmental Science, Cambridge, MD 21613

[d] Department of Biology, University of Maryland, College Park, MD 20742

[e] Current address: Oyster Recovery Partnership, 1805A Virginia Street, Annapolis, MD 21401 (Main Office)

[f] Aquaculture Genetics and Breeding Technology Center, Virginia Institute of Marine Science, College of William & Mary, P.O. Box 1346, Gloucester Point, VA 23062

[g] Chesapeake Biological Laboratory, University of Maryland Center for Environmental Science, Solomons, MD 20688

[*] Corresponding author: Tel.: + 1 757 787 0959; fax + 1 757 787 5831
E-mail address: harwell@vims.edu

ABSTRACT

A temporal comparison of developing triploid *Crassostrea virginica* and *C. ariakensis* oyster reefs was carried out in a Chesapeake Bay tributary in Maryland to examine the possibility of functional equivalency between native and non-native oysters. Habitat complexity and reef-associated benthic communities of experimental reefs were investigated over a 21 month period. One replicate of each of four experimental treatments (*C. virginica*; *C. ariakensis*; 50:50 of *C. virginica*: *C. ariakensis*; and shell only) were established in two blocks at the study site. Abundance, biomass, species richness, evenness, dominance and diversity of reef-associated fauna were evaluated in relation to date and oyster species, as were three indices of habitat complexity (maximum reef height, mean reef height, and surface rugosity). Habitat complexity varied with date as the experimental reefs developed over time, although little difference among oyster species was detected until approximately 21 months after deployment, when *C. ariakensis* experimental reefs achieved greater vertical heights than *C. virginica* reefs. Like habitat complexity indices, differences in benthic community structure were more common between sampling dates than between oyster species, indicating strong effects of both seasonality and reef development. Treatment effects on community metrics and standardized abundances of reef-associated fauna were not consistent among species over time and differed between treatment blocks, possibly indicating effects of flow and sedimentation rates on recruitment rates. Though univariate analyses results were largely inconsistent over time, multivariate analyses of standardized abundance data indicate that benthic community structure is affected by oyster species.

KEY WORDS: *Crassostrea ariakensis*; reef development; reef-associated fauna

INTRODUCTION

The Eastern oyster, *Crassostrea virginica*, historically played a large role in shaping the physical, chemical and biological systems of estuaries within the mid-Atlantic region of the United States (Kennedy 1996). With its continued decline, due to the combined stresses of disease (*Haplosporidium nelsoni* [MSX] and *Perkinsus marinus* [Dermo]; Ford & Tripp 1996, Fisher 1996, Lenihan et al. 1999, Mann 2000), over-fishing (Gross & Smyth 1946, Rothschild et al. 1994), deterioration in water quality (Lenihan & Peterson 1998), and reef degradation (Hargis & Haven 1988, Coen 1995, Lenihan & Peterson 1998, Mann 2000), there have been significant ecological and economic impacts throughout the region (Kennedy 1996). In response to these losses, substantial efforts have been made to restore both the fishery resource and habitat value of oyster reefs in many Atlantic coast estuaries (Ortega & Sutherland 1992, Luckenbach et al. 1996; Coen et al. 1997, Mann & Powell 2007). Due to the limited success of native oyster restoration efforts, the introduction of a non-indigenous oyster species (the Suminoe oyster, *C. ariakensis*) that is resistant to known diseases of the native oyster has been under consideration by the states of Maryland and Virginia for most of the last decade (e.g., Rickards & Ticco 2002). The pre-meditated movement of aquatic species for aquaculture and fishery enhancement purposes has occurred for over 2000 years (Mann et al. 1991), with oysters being perhaps the most pervasive example (Ruesink et al. 2005). Thus, despite the present abandonment of the proposal in Chesapeake Bay, results of research investigating this proposed introduction may be far-reaching.

As a result of the three-dimensional structure provided by *C. virginica* that supports a diverse assemblage of organisms generally not found in surrounding soft-bottom habitats (Dame 1979, Zimmerman et al. 1989, Coen et al. 1999, Posey et al. 1999), oyster reefs are now broadly recognized as ecosystem engineers (Luckenbach et al. 1999, Gutiérrez et al. 2003, ASMFC 2007). Given the accepted habitat value of *C. virginica* reefs, the functional equivalency of *C. ariakensis* reefs should be of concern, particularly given that the growth forms and reef-forming capabilities of *C. ariakensis* remain in question (Zhou & Allen 2003). Functional equivalency, often used as a predictor of restoration success in marine systems (Lockwood & Pimm 2001, Peterson & Lipcius 2003, Peyre et al. 2007), may be especially important if the introduced species were to out-compete the native species in some areas, leaving only the non-native species with ecologically relevant population sizes.

Previous studies of *C. virginica* have shown that certain aspects of reef morphology (e.g., shape, size and vertical complexity) may influence the degree to which reefs are utilized as habitat by other species (Posey et al., in prep, Breitburg 1999). For example, Breitburg (1999) suggested that enhancing topographical relief within reefs might attract oyster reef fish larvae by creating downcurrent low flow zones that allow larvae to remain on reefs and metamorphose to the benthos. If *C. virginica* and *C. ariakensis* differ in their reef-forming capabilities, this could have dramatic effects on the reef-associated fauna of Chesapeake Bay if *C. ariakensis* were to be introduced.

While there have been several previous field studies investigating the survival and growth of *C. ariakensis* in Virginia, Maryland, and North Carolina waters, (e.g., Calvo et al. 2001, Grabowski et al. 2005, Paynter et al. 2008), these trials provide little information on the reef morphology of *C. ariakensis* or the potential competitive interactions between the two *Crassostrea* species. Although several recent studies using diploid *C. ariakensis* and *C. virginica* in quarantined systems (Kingsley-Smith & Luckenbach 2008, Newell et al. unpubl. data, Allen et al. unpubl. data.) have addressed some of these issues, they do not duplicate conditions in natural bottom habitats and therefore have limitations in their applications. Additionally, the subsequent macrofuanal communities that might be associated with *C. ariakensis* reefs if introduced into Chesapeake Bay have yet to be examined.

Regardless of composition, structure, or size, the creation of oyster reefs results in the placement of new, un-colonized habitat into the natural environment (Osman and Whitlach 1999). Throughout the colonization process, the macrobenthic communities associated with the reefs remain dynamic, with continuous immigration of new individuals and new species, as well as the mortality of existing individuals and the local loss of species over time (Osman 1982). Such colonization is controlled by both regional and local processes, including the temporal and spatial distributions of larvae, as well as the life history of organisms that make up the associated reef community (Osman and Whitlach 1999). It is possible that recruitment dynamics on *C. virginica* and *C. ariakensis* reefs may also differ, possibly driven by physical, chemical, and/or biological mechanisms.

In a recently-completed large-scale field study, Kingsley-Smith et al. (2009) examined the comparative survival, growth and disease dynamics of *C. virginica* and *C. ariakensis* in bottom environments in the Chesapeake Bay region. This project provided the first opportunity for an on-bottom comparison of reef formation, habitat provision and habitat function in *C. virginica* and *C. ariakensis* across a range of habitat locations. Results from our spatial comparison suggested functional equivalency between oyster species with respect to habitat at an intertidal site and at a low salinity, subtidal location (Chapter 2, Harwell et al. 2010). At subtidal sites of higher salinity, however, the numbers of organisms associated with *C. virginica* reefs per unit of oyster biomass were significantly greater than the numbers of organisms associated with *C. ariakensis* reefs. Multivariate analyses of data from subtidal high salinity sites also revealed unique communities associated with *C. virginica* treatments, while mixed oyster species assemblages were functionally equivalent to mono-specific *C. ariakensis* experimental treatments (Chapter 2, Harwell et al. 2010). The results of this spatial comparison, however, were based on a single season of data. Given the dynamic nature of oyster reef communities throughout the process of reef development (Osman 1982), a comparison of the communities associated with native and non-native oyster reefs over time is needed for more complete investigation into the effects of oyster species on habitat function. Therefore, the objective of the present study was to provide a quantitative comparison of the habitat structure of *C. virginica* and *C. ariakensis* experimental reefs and of their utilization as habitat by other marine organisms throughout the course of reef development.

MATERIALS AND METHODS

Experimental Design

This study was part of a larger collaborative research effort comparing the survival, growth, and disease dynamics of triploid *C. virginica* and triploid *C. ariakensis* in bottom environments across a range of environmental conditions in the Chesapeake Bay region (Kingsley-Smith et al. 2009). To encompass the array of tidal environments, salinities, disease pressures, and relative predator abundances at which the native oyster, *C. virginica*, can be found in the region, one field site was selected in each of four tributaries: the Severn, Patuxent, York, and Machipongo Rivers.

In late October – early November 2005, four experimental treatments were established at each of the four sites; each site included two blocks with one treatment replicated per block.

Experimental triploid oyster treatments were as follows: *C. virginica* only, *C. ariakensis* only, and a 50:50 mixture of the two oyster species. A tray control with no live oysters was also included, comprised of clean *C. virginica* shell. In-depth descriptions of triploid oyster production, setting, and biosecurity precautions can be found in Kingsley-Smith et al. (2009).

Treatment replicates (henceforth referred to as reefs) were established as 5 x 5 arrays of plastic oyster grow-out trays. Each tray (58.4 cm W x 58.4 cm L x 7.3 cm H) was evenly ventilated with 0.6 cm diameter holes (Buckhorn Inc, a Meyers Industries Company). Prior to the start of experiments, all trays were lined with 2 mm fiberglass window screen and filled with a base layer of clean *C. virginica* shell. For live oyster treatments, juvenile oysters were added to achieve a target density of ~400 oysters m^{-2} (= 136 animals $tray^{-1}$). The tray control received oyster shell but no live oysters. Realized initial densities differed slightly across sites and between treatments (Virginia sites: *C. virginica* = 358.1 oysters m^{-2}, *C. ariakensis* = 325.9 oysters m^{-2}, mixed-species treatments = 342.0 oysters m^{-2}; Maryland sites: all treatments = 353.1 oysters m^{-2}). Mean shell heights of *C. virginica* and *C. ariakensis* at deployment were 12.80 mm (n = 1362, SD = 5.68) and 13.85 mm (n = 1272, SD = 5.45), respectively.

As two of our treatments contained non-native oysters, it was necessary to enclose all of our experimental reefs in cages as a biosecurity measure to protect against disturbances, redistributions, and losses of oysters from the experimental plots by extreme weather events and anthropogenic activities. Each array of 25 trays was surrounded by a large metal cage constructed from 3.8 cm diameter galvanized steel pipe and chain-link fence with 5 cm openings. Cages were placed on the seabed at least 1m apart. The 5 cm mesh prevented disturbances by large epibenthic predators, such as cownose rays, while permitting access to the oysters by small benthic predators such as gobies, blennies, and xanthid crabs.

Sampling occurred one month post-deployment and again in spring, summer, and fall of the following two years (2006 and 2007). Using the risk-averse sampling design described by Kingsley-Smith et al. (2009), three trays were removed from each cage at each site during each sampling event, and were replaced with trays filled with clean shell to maintain the spatial integrity of each experimental reef. All 24 trays (3 trays $cage^{-1}$ x 4 treatments x 2 blocks) from a site were sampled on a single day and transported to the laboratory for processing.

Several unforeseen complications during the course of these experiments, however, necessitated a reduction in experimental design. First, in late August 2006 Tropical Storm Ernesto caused measurable redistribution of oysters between trays within cages occurred, limiting the value of future data collected from the York River site. Although no oysters were released from the cages, raised concern about biosecurity led to the early termination of the experiment at this location in October 2006. At the Machipongo River site, differing elevations between blocks with respect to mean low water led to dramatic declines in *C. ariakensis* survival at the higher block (Kingsley-Smith et al. 2009) which led to a lack of comparable data at this site beyond July 2006. Finally, cages at the Severn River site were damaged in June to July 2007 as a result of illegal fishing activity. Extensive search and recovery efforts were made to remove all oysters within and around the damaged cages; however, the remaining scheduled sampling events were compromised.

July 2006 was the last sampling period for which all treatment replicates were intact across all sites. A spatial comparison of results from that sampling event across all sites is reported elsewhere (Chapter 2, Harwell et al. 2010). We chose to limit the scope of our temporal comparisons of habitat complexity and faunal communities to include only samples taken from the Patuxent River site, as this was the only location that remained uncompromised throughout the duration of the experiment. Results from those temporal comparisons are reported herein.

Habitat Complexity

Upon returning to the laboratory, each tray was photographed from the side, maintaining a pre-determined, consistent distance between the camera and each tray. The software program Image-J was used to quantify habitat complexity by obtaining measurements of maximum vertical reef height, average reef height, and surface rugosity from each digital image. Maximum vertical height was defined as the greatest distance between the top of the tray and the growing margin of an oyster protruding upwards from the tray. In addition to the maximum vertical height, measurements were taken for the next nine oyster growing margins judged to be at the greatest perpendicular distance from the upper level of the tray. Average reef heights were calculated as the means of these sets of ten measurements. A unit-less surface rugosity measurement was obtained from digital images of each tray by calculating the ratio of a contoured outline of the oysters within a tray to the linear length of the tray. This was a modified adaptation of the 'chain-length' method, widely used to assess surface topography of coral reefs (Rogers et al. 1983, Aronson & Precht 1995), in which rugosity (R) was calculated as $R = 1\text{-}d/l$, where d is the horizontal distance covered by the chain when conformed to the substratum and l is the length of the chain when fully extended (Aronson & Precht 1995).

Associated Fauna

After the removal of experimental oyster clumps and all *C. virginica* shell material, the remaining contents of each tray were rinsed on a 1-mm mesh sieve, then fixed in 10% buffered formalin for a minimum of 48 hrs prior to sorting, identification, and enumeration of organisms at the lowest practical taxonomic level. Organisms were then preserved in 70% ethanol prior to drying, weighing, and combustion to determine ash-free dry weights. In addition to abundance and biomass data, species richness, Pielou's evenness and Shannon-Weiner diversity were calculated for each sample using the PRIMER software package.

Statistical analyses

Prior to analyses, all data from the Patuxent River site were log-transformed to meet the assumptions of normality (Shapiro-Wilk) and homogeneity of variance (F-max test). Three-way, fixed factor ANOVA models, with date, treatment, and block as factors were used to analyze data for each index of habitat complexity (maximum reef height, average reef height, and surface rugosity). Due to the high prevalence of significant date-treatment interactions, a series of two-way, fixed factor ANOVAs for each main effect (date, treatment) were run within appropriate subsets of the data. In two-way ANOVA models, block effects were not significant so we removed block from the model and ran one-way ANOVA models. Pair-wise comparisons were made using Tukey's tests when ANOVA indicated a significant date or treatment effect.

Three-way, fixed factor ANOVAs also were run for each of the community metrics (total number of individuals per sample, species richness, Pielou's evenness and Shannon-Weiner diversity). Although date-treatment-block interactions were not significant, there was a high prevalence of significant block effects. Therefore, a series of two-way, fixed factor ANOVAs for each main effect (date, treatment) were run separately for each block. Subsequent reduced-model one-way ANOVAs were run for each main effect (date, treatment) within each block. Pair-wise comparisons were made using Tukey's tests when ANOVA indicated a significant date or treatment effect.

A similar series of tests (three-way, fixed-factor ANOVA models followed by reduced two-way and one-way models for each block) were run on the total abundance and biomass of all reef-associated fauna, as well as for individual dominant species. Species comprising at least one percent of the total abundance or biomass of associated fauna during a sampling event were considered dominant. All abundance and biomass data for live oyster treatments were standardized by oyster biomass prior to further analyses and met assumptions of normality (Shapiro-Wilk) and homogeneity of variance (F-max test). Tukey's test was used to conduct pair-wise comparisons among dates and treatments if ANOVA indicated a significant effect of a main factor.

Although they may not have been defined as dominant species with respect to numbers or biomass, resident oyster reef fish species, including the striped blenny, *Chasmodes bosquianus*; the skilletfish, *Gobiesox strumosus*; the naked goby, *Gobiosoma bosc*; and the oyster toadfish, *Opsanus tau*, are important predators that can shape benthic community structure. Therefore, standardized abundances of these demersal fishes were also analyzed through a similar series of 3-way and reduced model 2-way and 1-way ANOVAs, using Tukey's test to conduct pair-wise comparisons among dates and treatments.

To further evaluate variations in community structure between treatments, multivariate approaches in the PRIMER statistical software package were used. Similarity matrices were calculated using non-transformed abundance and biomass data standardized by oyster biomass. These similarity matrices were used to create non-metric multi-dimensional scaling (MDS) plots of each sample at a given date. Analysis of Similarity (ANOSIM), which takes both species composition and abundance into account, was then performed on the similarity matrices in order to determine whether treatment differences were present.

RESULTS

Habitat Complexity

Maximum reef heights for all live oyster treatments were similar during initial sampling in December 2005, and did not significantly increase until July 2006 (Fig. 1A, $p<0.0001$ in all cases). Subsequent increases in maximum reef height occurred through July 2007 for *C. ariakensis*, *C. virginica*, and mixed oyster species reefs ($p<0.0001$ in all cases), while measurements taken from September 2007 samples were similar to those from July 2007. All trays without live oysters had similar vertical heights until July 2007, when a significant increase occurred ($p<0.0001$). Significant treatment effects on maximum reef heights were also present (Table 1). Though measurements of trays without live oysters were initially similar to maximum heights of some live oyster treatments, by July 2006 significant differences emerged ($F=17.24$, $P<0.0001$) and were maintained throughout the remainder of the experiment ($p<0.0001$ in all cases). Differences between live oyster treatments were less common. Initially, reefs comprised of a mix of both oyster species achieved higher maximum heights (mean = 2.19 cm) than monospecific *C. ariakensis* reefs (mean = 1.89 cm), while *C. virginica* reef measurements (mean = 2.11 cm) were intermediate and overlapping ($F=3.20$, $p=0.0484$). This pattern did not persist, however, and by July 2007 the maximum vertical heights of non-native oyster reefs (mean = 17.17 cm) were greater than those of native reefs (mean = 15.08 cm), while mixed species reefs (mean = 16.47 cm) were intermediate and overlapping (F=722.80, p<0.0001). Similar observations were made in September 2007, when mean reef heights reached 19.17 cm (*C. ariakensis*), 15.87 cm (*C. virginica*) and 18.58 cm (mixed) (F=178.0, p<0.0001).

Date effects on mean reef heights were identical to those for maximum reef heights, with similar heights amongst live oyster treatments in December 2005 and April 2006, followed by subsequent increases beginning in July 2006 and continuing through July 2007 (Fig. 1B, p<0.0001 in all cases). Aside from differences between shell only and live oyster reefs, treatment effects on mean reef heights were rare, only occurring in July 2007, when average reef heights were significantly lower for *C. virginica* reefs (mean = 13.62 cm) than for those containing *C. ariakensis* (mean = 14.93 cm for single species, mean = 14.95 for mixed treatments,). This pattern, however, was not maintained through the end of the experiment.

Surface rugosity was also similar among live oyster treatments until July 2006, when index values significantly increased for native, non-native, and mixed oyster species reefs (Fig. 1C, p<0.001 in all cases). Another increase was observed for the subsequent sampling event in October 2006, (p<0.0001 in all cases). After the October 2006 sampling date, rugosity measurements of *C. virginica* reefs (mean = 1.61) remained statistically unchanged through the end of the study. The surface rugosity of *C. ariakensis* reefs, however, increased again in April 2007 from 1.49 to 1.66, where the measurements reached a plateau. Mixed species reefs had similar rugosity measurements from October 2006 (mean = 1.56) through July 2007, which a final increase in September 2007 (mean = 1.85). Treatment differences in surface rugosity were only present between live oyster and shell only treatments throughout the entire study.

Associated Fauna: Community Metrics

A total of 137,985 individual organisms were collected from the Patuxent River site over the course of this study, resulting in a combined ash-free dry weight of 2.5 kg. Seasonal patterns were present, with highest overall abundances occurring in summer (32,419 in July 2006 and 28,869 in July 2007) and lowest abundances recorded during fall / winter (9,574 in October 2006; 14, 029 in December 2005; and 14,147 in September 2007) (Table 2). Intermediate totals were present in spring samples (17, 680 in April 2006 and 21,267 in April 2007). A similar pattern was observed for overall biomass of associated macrofauna, ranging from 29.05 g in December 2005 to 716.95 g in July 2007. Due to significant block effects, all community metrics data was analyzed separately within each block (Table 3). The only treatment effect within block 1 occurred in July 2007, when the average number of organisms found per tray on *C. ariakensis* reefs (mean = 1,839) was significantly higher than that of *C. virginica* reefs (mean = 1,156) and trays without live oysters (mean = 975), with overlapping, intermediate values on mixed reefs (mean = 1417) (F=9.18, p=0.0057). Differences between treatments were more prevalent in block 2, although these differences were not similar across sampling dates (Fig. 2). Initially, *C. virginica* (mean = 768) and *C. ariakensis* treatments (mean = 686) supported higher numbers of associated fauna than shell only trays (mean = 317), while reefs containing both oyster species (mean = 418) had intermediate, overlapping numbers (F=7.88, p=0.009). In April 2006, macrofaunal abundances were higher on native oyster reefs than those associated with any other treatment

(F=11.30, p<0.003). Treatment differences observed in July 2006 were similar to those seen initially in block 2 (F=4.56, p=0.0383). Treatment effects were not observed again until July 2007, when *C. ariakensis* and mixed oyster species reefs supported greater numbers of organisms than shell only trays, with overlapping, intermediate values present on *C. virginica* reefs (F=5.90, p=0.02).

As with the total number of individuals, a significant block effect occurred for all remaining community metrics, with block 1 having significantly higher numbers than block 2. Species richness (Fig. 3), Peilou's evenness (Fig. 4), dominance (Fig. 5), and Shannon-Weiner diversity (Fig. 6) did not differ between treatments at any time within block 1 (Table 4), though they varied significantly with sampling date. Similarly, within block 2 no treatment effects were present at any point during the study for species richness or dominance. A single significant effect occurred for Peilou's evenness, which was lower among *C. virginica* samples than for any other treatment in October 2006 (F=21.33, p<0.0004). Treatment effects within block 2 were most common for Shannon-Weiner diversity, occurring in December 2005, April 2006, October 2006, and July 2007. The direction of these effects was not consistent over time (Table 4).

Associated Fauna: Total Standardized Abundance and Biomass

Oyster survival and growth differed significantly with sampling date (see Kingsley-Smith et al. 2009 for details). To more accurately compare the effects of oyster species, not oyster survival and growth, on reef-associated communities over time, the total abundances and ash-free dry weights of reef-associated organisms per sample were standardized by the oyster biomass present (methods described in Kingsley-Smith et al. 2009). ANOVAs for log-transformed data revealed date and treatment effects that differ from the ANOVA results for raw abundance data (Fig.7), although the block effect was consistent (block 1 > block 2). The effect of date was large, with highest numbers of associated organisms per gram of oyster biomass occurring in July 2006, followed by July 2007. (F=68.07, p<0.0001).

Once again, treatment effects were more common within block 2; July 2006 was the only time at which oyster species affected standardized abundance of associated macrofauna within block 1 (Table 5). At this time, *C. virginica* reefs (mean = 107.1) supported high standardized abundances than *C. ariakensis* reefs (mean = 15.3), with intermediate, overlapping numbers found on reefs comprised of both oyster species (36.7) (F=8.96, p=0.016). Treatment effects within block 2 were intermittent and inconsistent over time. Initially, native reefs supported marginally higher numbers of associated organisms per gram of oyster biomass than mixed reefs, with intermediate values found on *C. ariakensis* reefs (F=5.17, p=0.05). Similar standardized abundances were observed among treatments the following spring, yet in July 2006, reefs containing *C. ariakensis*, both single species (mean = 29.8) and mixed species treatments (mean = 39.9), supported lower numbers of associated organisms than did monospecific *C. virginica* reefs (mean = 90.8) (F=10.75, p=0.01). By April 2007, however, the trend of increased standardized abundances on native reefs was reversed, with *C. ariakensis* reefs supporting higher numbers of individuals than the other live oyster treatments (F=13.35, p=0.006). Similarly, September 2007 standardized abundances were higher on *C. ariakensis* reefs than *C. virginica* reefs (F=8.16, p=0.019).

Total standardized ash-free dry weight of associated macrofauna also varied with date (Fig. 8); highest values were observed in April 2006 and July 2006 (F=20.22, p<0.0001). The effect of block also was significant (F=7.79, p=0.0065), with values within block 1 greater than those within block 2. Standardized biomass was similar among live oyster treatments in block 1 for most dates, although *C. virginica* reefs did have higher total ash-free dry weight than *C. ariakensis* and mixed species treatments in April 2006. A similar pattern was seen in block 2 in December 2005, April 2006, and July 2006 (Table 5). Beyond that date, however, no significant treatment effects were present.

Associated Fauna: Species Composition

A total of 45 different species were found throughout this study, including polychaete worms (9 spp.), bivalves (9 spp.), amphipods (6 spp.), decapod crustaceans (7), fishes (6 spp.), gastropods (6), and cnidarians (2 spp.). A complete list of all species and the dates at which they occurred can be found in Table 6. Standardized abundance and biomass data was used to determine which species dominated samples from each date (Tables 7 & 8). Dominant species were defined as those comprising at least one percent of the total abundance or biomass of organisms, respectively. Initially, dominant species with respect to abundance included 3 polychaetes (*Eteone heteropoda*, *Neanthes succinea*, and *Polydora websteri*), 2 amphipods (*Apocorophium lacustre* and *Melita nitida*), 2 bivalves (*Mya arenaria* and *Mulinia lateralis*) and 1 decapod crustacean (juvenile xanthid crabs under 2 mm carapace width). Treatment effects for these species were only seen in block 2, where 3 of the 8 species (*M. nitida*, *M. lateralis*, and juvenile xanthid crabs) had higher standardized abundances on *C. virginica* reefs than on mixed reefs ($p<0.0136$). Abundances of dominant organisms were comparable between monospecific native and non-native reefs in all but one case (*M. lateralis* numbers were higher on native reefs, ($F=14.58$, $p=0.005$)).

The following spring, 10 species were dominant, including 2 polychaetes, 2 amphipods, 4 bivalves, 1 decapod crustacean, and 1 gastropod (Table 7). Once again, standard abundances of individual species within block 1 were generally similar between live oyster treatments, with the exception of *M. nitida*, which had higher number on *C. ariakensis* reefs than mixed reefs, with intermediate numbers on *C. virginica* reefs (F=5.15, p=0.0499). Individual dominant species abundances for block 2 were similar among treatments for 6 of the 10 species. Two of the bivalves (*Macoma balthica* and *M. arenaria*) were more abundant on *C. virginica* reefs than on both *C. ariakensis* and mixed reefs ($p<0.0365$). This pattern was also observed for juvenile xanthid crabs (F=12.17, p=0.0077), and *A. lacustre* abundances in April 2006 were also higher on native reefs than non-native reefs, though intermediate numbers were present on mixed species reefs (F=6.16, p=0.0351).

Treatment effects were most prevalent among individual species abundances in July 2006 samples, when 5 of the 7 dominant species in block 2 had higher standardized abundances on *C. virginica* reefs than on *C. ariakensis* reefs, with intermediate, overlapping number on mixed reefs ($p\leq0.0378$ in all cases). These species included the bivalves *M. balthica* and *M. arenaria*, the amphipod *Gammarus palustris*, the polychaete *N. succinea*, and the xanthid crab *Eurypanopeus depressus*. Treatments effects were also seen in block 1, though they were much less prevalent. Here, both *E. depressus* and *N. succinea* abundances were higher on native reefs than on non-native reefs. No differences between live oyster treatments were present for *M. nitida* or the bivalve *Ischadium recurvum* in either block.

In October 2006, only 3 of the 12 dominant species showed preferential utilization of a specific treatment. *M. balthica* abundances within block 1 were higher on monospecific *C. ariakensis* reefs than on mixed reefs (F=6.94, p=0.0275). In block 2 the naked goby, *Gobiosoma bosc*, was more common on mixed oyster species and *C. virginica* reefs than on *C. ariakensis* reefs (F=17.01, p=0.0034), whereas the amphipod, *M. nitida*, had higher numbers on *C. ariakensis* reefs than on other live oyster treatments (F=9.61, p=0.0135).

Across sampling events in 2007, only 2-4 of the 7-10 dominant species within a given date exhibited preferential habitat use of a particular oyster species. In these cases, *C. ariakensis* reefs were preferred over *C. virginica* reefs, with mixed reefs often having intermediate values (Table 7). Such treatment effects were seen for *I. recurvum*, *N. succinea*, juvenile xanthid crabs, *M. balthica*, *A. lacustre* and *G. bosc*.

Dominant species as determined by biomass displayed different patterns than those observed for abundance dominants. In December 2005, 9 species comprised at least 1% of total standardized biomass, including the blue crab, *Callinectes sapidus*; the flat mud crab, *E. depressus*; juvenile xanthid crabs; the skillet fish, *Gobiesox strumosus*; the naked goby, *G. bosc*; the polychaetes *E. heteropoda* and *N. succinea*; the amphipod *M. nitida*; and the grass shrimp, *Palaemonetes pugio* (Table 8). As with abundance patterns, treatment effects were only seen in block 2: 6 of the 9 dominant species had greater biomass on *C. virginica* reefs than on mixed reefs ($p \leq 0.0475$).

Only 5 species were considered dominant by standardized biomass in April 2006, and *G. bosc*, *M. arenaria*, and *N. succinea* all had greater ash-free dry weights in samples from *C. virginica* reefs than for any other live oyster treatment ($p \leq 0.0005$ in all cases). The following summer, however, treatment effects on individual species biomass were rare, and only present for 2 of the 14 dominant species (the bivalve, *M. arenaria* and the gastropod, *Boonea bisuturalis*). Patterns were similar in throughout the remainder of the experiment, with few species exhibiting preferential usage of a particular oyster species treatment. When a preference *was* present, it was always for native reefs over non-native reefs (Table 8), although the particular species exhibiting this preference varied with date.

Standardized abundances of mobile predatory fish species, including the striped blenny, *C. bosquianus*; the skilletfish, *G. strumosus*; the naked goby, *G. bosc*; and the oyster toadfish, *O. tau*, generally increased over time (Table 11). This pattern was especially evident for the naked goby, *G. bosc*, the most common resident fish species found throughout the study (Fig. 9).

Multivariate Analyses

Non-metric MDS plots and Analysis of Similarity (ANOSIM) of standardized abundances of associated fauna indicated significant treatment effects in July 2006, April 2007, July 2007, and September 2007(Fig. 10). The benthic communities associated with *C. virginica* experimental reefs were different from those associated with *C. ariakensis* reefs during the aforementioned sampling dates. Communities associated with the mixed species reefs were similar to those of *C. ariakensis* reef in July 2006, yet in April 2007 and September 2007, they did not differ from either monospecific oyster reef treatment. During the summer of 2007, each of the three live oyster treatments supported its own distinct community. Multivariate analyses utilizing the biomass of associated organisms standardized by oyster biomass yielded results different from those obtained from abundance data; treatment effects were only observed in April 2006, when each live oyster treatment supported unique community assemblages (Fig. 11).

DISCUSSION

Results from short-term laboratory trials and anecdotal observations of *C. ariakensis* in its native range have raised doubt over the species' ability to form the dense aggregations observed in *C. virginica*. However, we have recently reported no significant differences between native and non-native experimental reefs with regard to three distinct habitat complexity indices (maximum vertical reef height, average reef height and surface rugosity) after approximately eight months of deployment, regardless of location within the Chesapeake Bay region (Chapter 2, Harwell et al. 2010). Despite evidence of negative effects of interspecific competition on the growth of *C. ariakensis* at the low and mid salinity subtidal sites (Kingsley-Smith et al. 2009), experimental reefs containing a mixture of both oyster species had similar measures of habitat complexity when compared to monospecific reefs (Chapter 2, Harwell et al. 2010). The present temporal comparison of habitat complexity over 21 months of reef development revealed similar results for surface rugosity index, for which no significant differences were found between live oyster treatments throughout the duration of the experiment. Average initial values of surface rugosity ranged from 1.12 to 1.16 between live oyster treatments, which is similar to values reported from unrestored native oyster bars in Maryland (mean = 1.15) (Rodney and Paynter 2006). By the end of the experiment, mean rugosity index measurements for live oyster treatments ranged from 1.78 to 1.85, resembling estimates for restored native oyster bars in the aforementioned study (mean = 1.84).

The observed similarity in surface rugosity index between oyster species throughout the study was surprising, as we observed qualitative differences in reef morphologies when visually comparing native and non-native treatments. Though the overall amount of interstitial space present within reefs may have been similar among oyster species, the arrangement of that space varied. *C. virginica* reefs contained a tight arrangement of individuals, resulting in a high number of small crevices; *C. ariakensis* reefs more frequently had growth trajectories in a more horizontal direction, leading to greater angles between individuals. In retrospect, the 'chain link' method of assessing habitat heterogeneity may not have been the most appropriate choice, given the nature of the visual discrepancies in morphology between oyster treatments. Despite its widespread use and general acceptance as an indicator of habitat complexity in aquatic systems, the rugosity index cannot discriminate between the shape and size of components relative to the scale of the topography under investigation (Roberts & Ormond 1987, Shumway et al. 2007). For example, a complex, small-scale topography would have the same rugosity value as a simple, large-scale topography (Roberts & Ormond 1987). We recommend future studies of habitat complexity in oyster reefs include more novel approaches to complexity comparisons, such as measuring the distance between individual oysters, or the angles at which individual oysters meet one another.

Although measures of maximum and mean reef heights were also initially similar, divergence between oyster species was detected in July 2007, after approximately 19 months of growth (Fig. 1A and 1B). By this time, *C. ariakensis* reefs achieved greater heights than *C. virginica* reefs, while mixed species reefs had intermediate heights. For maximum reef heights, this pattern persisted during the final sampling event in September 2007. We largely attribute this to well-documented differential growth rates between *C. ariakensis* and *C. virginica* in higher salinities (Kingsley-Smith et al. 2009, Paynter et al. 2008, Calvo et al. 2001).

Though necessary for biosecurity reasons, the cages in which the oysters were deployed may have indirectly affected oyster survival (Kingsley-Smith et al. 2009), as their presence likely reduced predation rates by limiting the access of larger predators such blue crabs, *Callinectes sapidus*, and cownose rays, *Rhinoptera bonasus*. Particularly at small sizes, *C. ariakensis* shells are structurally weaker than those of the native species, allowing for greater susceptibility to predation (Bishop & Peterson 2006, Newell et al. 2007). Therefore, our use of cages in this study may have disproportionately decreased mortality rates of juvenile *C. ariakensis*, which in turn may have affected habitat complexity. Despite these caveats, our results suggest that, at least within the mesohaline region of Chesapeake Bay tributaries, *C. ariakensis* reefs would achieve, or possibly exceed, the vertical reef heights that native oysters assume.

Of the 45 different species of reef-associated macrofauna collected during the study, 27 have been recently found on restored and non-restored portions of four historic Maryland natural oyster bars, one of which was located in the Patuxent River (Rodney and Paynter 2006). The vast majority of species we determined to be dominant were also reported as dominant in Rodney and Paynter's assessment (2006). Species present during our experiment but not found in the aforementioned study included 3 amphipods (*Cymadusa compta*, *E. levis*, and *G. palustris*); 2 bivalves (*Ensis directus* and *Geukensia demissa*); the mud crab *Dyspanopeus sayi*; 5 species of gastropods (*Acteocina canaliculata*, *Boonea bisuturalis*, *Boonea impressa*, *Crepidula fornicata*, and *Nassarius vibex*); 3 fishes (*Anguilla rostrata*, *Gobiesox strumosus*, and *Syngnathus focus)*; and 3 polychaete species (*Eteone heteropoda, Leitoscoloplos fragilis, and Mediomastus ambiseta*).

The colonization of new habitat is controlled by both regional and local processes, although the regional pool of available species sets the upper limit on colonization (Osman and Dean 1978). The temporal and spatial distributions of larvae produced within the region cause variations in the availability of these motile stages and thus determine both the initial order and abundances of colonizing species (e.g. Grosberg 1982, Olson 1985, Roughgarden et al. 1985, Gaines and Roughgarden 1987, Gotelli 1987, Todd et al. 1988, Farrell et al. 1991, Carlon and Olson 1993). These in turn can determine dominant interactions within and among species and set both short- and long-term patterns of abundance (Osman and Whitlach 1999). Timing of deployment can also determine order of early colonization and thus the development of the community (Osman 1977, Sutherland and Karlson 1977).

As was the case for habitat complexity, differences among reef-associated faunal assemblages were more pronounced between dates than between treatments. Unlike complexity measurements, however, overall abundances of associated fauna, standardized abundance and biomass of macrofauna, as well as several community metrics (species richness, Peilou's evenness, dominance, and Shannon-Weiner diversity), varied seasonally. Highest overall abundances were found during summer sampling events in both July 2006 and July 2007; lowest total numbers of associated fauna occurred in December 2005 and October 2006, and intermediate values occurring in spring. Seasonal variations in relative and absolute larval abundances among species can cause very different patters of larval settlement (e.g. Osman 1977, 1978, Sutherland and Karlson 1977), and seasonal effects have been documented in other oyster reef macrobenthic communities (e.g., Posey et al. 1999, Nestlerode 2004, Tolley et al. 2005, Boudreaux et al. 2006). Annual cycles with an increase in number and biomass in the summer followed by large winter mortality are also common in soft-bottom benthic communities (Buchanan et al .1978), driven primarily by greater larval recruitment in the summer months (Gray 1981). This may be due in part to the strong seasonal cycles of phytoplankton abundance, productivity, and composition in temperate estuaries, which is caused by changes in the availability of light and nutrients, as well as temperature variations (Tenore 1988). Water column food supplies drop to minimum levels in winter months, which may help regulate reef productivity and community structure (Nestlerode 2004). Thus, it is not surprising that the most common patters found in benthic communities are those associated with seasonal changes (Gray 1981).

Habitat stability, or stage of succession, also has been shown to affect species abundance distributions in benthic communities (Death 1996). In freshwater streams, unstable and very stable stream communities were dominated by one or two taxa, with a large number of rare species, while communities of intermediate stability had relatively uniform species abundances, or low dominance values. If we apply this paradigm to the current study, we expect the earliest sampling date (when succession stage is low) to have high values of dominance, followed by low dominance values as the reefs increase in stability, and finally, we would expect another increase in dominance as reef development continues. This pattern is indeed present (Fig. 5) from December 2005 through April 2007, at which points dominance begins and continues to decrease until the end of the experiment. A similar trend can be seen in species richness, which was initially high, then decreased dramatically, then increased in the summer of 2006 and decreased after April 2007 (Fig. 3). Although Death's (1996) paradigm does not account for the observed decreases in dominance in July 2007, it may be that abundances of benthic predators, such as the demersal fishes *G. bosc*, *G. strumosus*, *C. bosquianus*, and *O. tau*, was high during that time period, thus affecting the abundances of amphipod and polychaete prey species, which made up the majority of the total abundances observed on the experimental reefs. Standardized abundances of demersal fishes generally increased over time (Fig. 9) until the end of the experiment. This may in part be due to an increased amount of interstitial space that occurs as reefs develop over time, which the fishes use as habitat. Interestingly, the naked goby, *G. bosc*, which are typically either the first or second most abundant fish larvae in mesohaline areas of Chesapeake Bay tributaries during summer (Breitburg 1999), had highest standardized abundances in September 2007 (Table 9), indicating high recruitment levels in July 2007. Thus, benthic community structure over time reflected the influence of reef development and stability, larval recruitment, and seasonality.

Total standardized abundance of all reef-associated macrofauna also varied with date, with highest numbers found in July 2006 samples, followed by July 2007. This is likely due to seasonal increases in recruitment. Resident oyster reef fish species within the mesohaline region of Chesapeake Bay (*G. bosc*, *G. strumosus*, *C. bosquianus*, and *O. tau*) have peak recruitment during mid-summer (Breitburg 1999), as do many benthic invertebrates (Gray 1981). Although some treatment differences were detected for other sampling dates, they were small in comparison to the treatment effect seen in July 2006, when abundances on *C. virginica* experimental reefs were much greater than on either remaining live oyster treatment. This is also the time period at which treatment effects on individual species standardized abundances were most prevalent; 5 of the 7 dominant species occurred in greater numbers on native reefs than on non-native reefs. This relationship, as was the case for most measures of associated fauna, was only observed for block 2. The cages comprising block 2 were situated in a row closer to the channel than the cages within block 1 (approximately 9.1 m further from shore than block 1) (Fig. 12). This design may have inherently impacted the relative flow rates of water passing through the cages of each block (i.e., cages in block 2 may have been exposed to higher flow rates than cages in block 1). Recruitment often increases with current speed in the marine benthic environment due to enhanced larval supply (e.g., Mullineaux and Garland 1993). Within block 2, relative to the direction of outgoing tidal flow, passive planktonic larvae would encounter the *C. virginica* cage first, increasing the likelihood that larvae would encounter chemical cues from current reef inhabitants of that specific cage and settle there. Alternatively, it is possible that these block effects are related to differences in bedload sediment transport, which may also have influenced oyster physiology and growth at this location (Kingsley-Smith et al. 2009).

The preferential utilization of *C. virginica* reefs over *C. ariakensis* reefs by associated macrofauna within block 2 may, therefore, be explained by factors other than oyster species. However, multivariate analyses (MDS and ANOSIM) of standardized abundances of reef-associated macrofauna indicated overall treatment effects on benthic community structure during the majority of sampling dates, regardless of block. In addition to the July 2006 sampling event, structure was different between native and non-native oyster reef communities for all of 2007. As reefs continue to develop past one year of age, the communities associated with *C. ariakensis* (with respect to abundance and composition) were unique compared to those associated with *C. virginica*. Interestingly, communities supported by mixed oyster species reefs were similar to those supported by monospecific reefs of both oyster species during most of study. In summers, however, their communities were either similar to *C. ariakensis* communities (2006) or completely unique (2007). Results from the mixed oyster treatment are of particular interest, since it is most likely that the two oyster species would co-occur on some reefs, should an introduction of *C. ariakensis* occur.

This study indicates that, if introduced to mesohaline areas within the Chesapeake Bay region, the ability of *C. ariakensis* to serve as a functional equivalent of the native oyster with respect to habitat provision is likely to change over time. Though initial equivalency is likely, some differences in habitat complexity and benthic community structure may occur as reefs mature.

ACKNOWLEDGMENTS

The authors thank B. Barnes, A. Birch, R. Bonniwell, S. Bonniwell, R. Bushnell, M. Chen, A. Curry, S. Fate, J. Goodwin, B. Hammer, W. Hart, A. Makin, P. Ross, M. Sherman, E. Smith, T. Wagner, C. Vaughn and N. Ward for assistance in data collection, and D. Gillett for taxonomic and statistical assistance. This research was funded by grants from the NOAA Chesapeake Bay Office (#NA05NMF4571240), Virginia Marine Resources Commission, Maryland Department of Natural Resources, and the Keith Campbell Foundation for the Environment.

LITERATURE CITED

Aronson, R. B. & W. F. Precht. 1995. Landscape patterns of reef coral diversity: A test of the intermediate disturbance hypothesis. *J. Exp. Mar. Biol. Ecol.* 192:1-14.

ASMFC. 2007. The importance of habitat created by shellfish and shell beds along the Atlantic Coast of the U.S. Prepared by L.D. Coen and R. Grizzle with contributions by J. Lowery and K.T. Paynter Jr. 108 pp.

Bishop, M. J. & C. H. Peterson. 2006. When *r*-selection may not predict introduced-species proliferation: Predation of a nonnative oyster. *Ecol. Appl.* 16:718-730.

Boudreaux, M. L., J. L. Stiner & L. J. Walters. 2006. Biodiversity of sessile and motile macrofauna on intertidal oyster reefs in Mosquito Lagoon Florida. *J. Shellfish Res.* 25:1079-1089.

Breitburg, D. L. 1999. Are three-dimensional structure and healthy oyster populations the keys to an ecologically interesting and important fish community? In: M. W. Luckenbach, R. Mann & J. A. Wesson (editors). *Oyster reef habitat restoration: a synopsis and synthesis of approaches.* Virginia Institute of Marine Science Press, Gloucester Point, USA, pp 239-250.

Buchanan, J. B., M. Sheader & P. R. Kingston. 1978. Sources of variability in the benthic macrofauna off the south Northumberland coast, 1971-1976. *J. Mar. Biol. Ass. UK* 58:191-210.

Calvo, G. W., M. W. Luckenbach, S. K. Allen, Jr. & E. M. Burreson. 2001. A comparative field study of *Crassostrea ariakensis* (Fujita 1913) and *Crassostrea virginica* (Gmelin 1791) in relation to salinity in Virginia. *J. Shellfish Res.* 20:221-229.

Carlon, D. B. & R. R. Olson. 1993. Larval dispersal distance as an explanation for adult spatial patterns in two Caribbean reef corals. *J. Exp. Mar. Biol. Ecol.* 173:247-263.

Coen, L. D., E .L. Wenner, D. M. Knott, M. Y. Bobo, N. H. Hadley, D. L. Richardson, B. Stender & R. Giotta. 1997. Intertidal oyster reef habitat use and function: what have we learned after two years? *J. Shellfish Res.*16:336. (Abstract).

Coen, L. D., M. W. Luckenbach & D. L. Breitburg. 1999. The role of oyster reefs as essential fish habitat: a review of current knowledge and some new perspectives. *Am. Fish. Soc. Symp.* 22:438-454.

Death, R. G. 1996. The effect of habitat stability on benthic invertebrate communities: utility of species abundance distributions. *Hydrobiologia* 317:97-107.

Dame, R. F. 1979. The abundance, diversity and biomass of macrobenthos on North Inlet, South Carolina, intertidal oyster reefs. Proceedings of the National Shellfisheries Association. Vol. 68, pp.6-10.

Farrell, T., M. D. Bracher & J. Roughgarden. 1991. Cross-shelf transport causes recruitment to intertidal populations in central California. *Limnol. Oceanogr.* 36:279-288.

Fisher W. S. 1996. Preface. *J. Shellfish Res.* 15:1.

Ford, S. E. & M. R. Tripp. 1996. Diseases and defense mechanisms. *In:*
V. S. Kennedy, R. I. E. Newell & A. F. Eble (eds.). The Eastern oyster, *Crassostrea virginica.* Maryland Sea Grant, College Park, Maryland, USA, pp. 581-660.

Gaines, S. D. & J. Roughgarden. 1987. Fish in offshore kelp forests affect recruitment to intertidal barnacle populations. *Science* 235:479-481.

Gotelli, N. J. 1987. Spatial and temporal patterns of reproduction, larval settlement, and recruitment of the compound ascidian *Aplidium stellatum. Mar. Biol.* 94:45-51.

Grabowski, J. H., A. R. Hughes, D. L. Kimbro & M. A. Dolan. 2005. How habitat setting influences restored oyster reef communities. *Ecology* 86:1926-1935.

Gray, J. S. 1981. The ecology of marine sediments. An introduction to the structure and function of benthic communities. Cambridge Univ. Press, Cambridge, U. K., 185 p.

Grosberg, R. K. 1982. Intertidal zonation of barnacles: the influence of planktonic zonation of larvae on vertical distribution of adults. *Ecology* 63:894-899.

Gross, F. & R. C. Smyth. 1946. The decline of oyster populations. *Nature* 157:540-542.

Gutiérrez, J. L., C. G. Jones, D. L. Strayer & O. O. Iribarne. 2003. Mollusks as ecosystem engineers: the role of shell production in aquatic habitats. *Oikos* 101:79-90.

Harwell, H.D, P. R. Kingsley-Smith, M. L. Kellogg, S. M. Allen, S. K. Allen, Jr., D. W. Meritt, K. T. Paynter, Jr. & M. W. Luckenbach. 2010. A comparison of *Crassostrea virginica* and *C. ariakensis* in Chesapeake Bay: Does oyster species affect habitat function? *J. Shellfish Res.,* accepted.

Kingsley-Smith, P. R. & M. W. Luckenbach. 2008. Post-settlement survival and growth of the Suminoe oyster, *Crassostrea ariakensis*, exposed to simulated emersion regimes. *J. Shellfish Res.* 27:609-618.

Kingsley-Smith P. R., H. D. Harwell, M. L. Kellogg, S. M. Allen, S. K. Allen, Jr., D. W. Meritt, K. T. Paynter, Jr. & M. W. Luckenbach. 2009. Survival and growth of triploid *Crassostrea virginica* (Gmelin, 1791) and *C. ariakensis* (Fujita, 1913) in bottom environments of Chesapeake Bay: implications for an introduction. *J. Shellfish Res.* 28:169-184.

Lenihan, H. S. & C. H. Peterson. 1998. How habitat degradation through fishery disturbance enhances impacts of hypoxia on oyster reefs. *Ecol. Appl.* 8(1):128-140.

Lenihan, H. S., F. Micheli, S. W. Shelton & C. H. Peterson. 1999. The influence of multiple environmental stressors on susceptibility to parasites: an experimental determination with oysters. *Limnol. Oceanogr.* 44:910-924.

Lockwood, J. L. & S. L. Pimm. 2001. When does restoration succeed? In: E. Weiher & P. Keddy (eds.), Ecological Assembly Rules: Perspectives, Advances, Retreats, 1st paperback edition. Cambridge University Press, Cambridge, U.K., pp. 363-392.

Luckenbach, M. W., R. Mann & J.A. Wesson (Eds.) 1999. *Oyster reef habitat restoration. A Synopsis and synthesis of approaches.* Virginia Institute of Marine Science Press, Gloucester Point, VA, 358 pp.

Luckenbach, M. W., J. A. Nestlerode & G. M. Coates. 1996. Oyster reef restoration: developing relationships between structure and function. *J. Shellfish Res.* 16:270-271. (Abstract).

Mann, R. 2000. Restoring the oyster reef communities in the Chesapeake Bay: a commentary. *J. Shellfish Res.* 19:335-339.

Mann, R., E. M. Burreson & P. Baker. 1991. The decline of the Virginia oyster fishery in Chesapeake Bay: considerations for introduction of a non-endemic species, *Crassostrea gigas* (Thunberg, 1973). *J. Shellfish Res.* 10:379-388.

Mann, R. & E. Powell. 2007. Why oyster restoration goals in the Chesapeake Bay are not and probably cannot be achieved. *J. Shellfish Res.* 26:905-917.

Mullineaux, L. S. & E. D. Garland. 1993. Larval recruitment in response to manipulated field flows. Marine Biology 116: 667-683.

Nestelrode, J. A. 2004. Evaluating restored oyster reefs in Chesapeake Bay: how habitat structure influences ecological function. College of William and Mary, Virginia. Dissertation.

Newell, R. I. E., V. S. Kennedy, K. S. Shaw & S. Kristi. 2007. Comparative vulnerability to predators, and induced defense response, of eastern oysters *Crassostrea virginica* and nonnative *Crassostrea ariakensis* oysters in Chesapeake Bay. *Mar. Biol.* 152:449-460.

Olson, R. R. 1985. The consequences of short-distance larval dispersal in a sessile marine invertebrate. *Ecology* 66:30-39.

Ortega, S. & J. P. Sutherland. 1992. Recruitment and growth of the eastern oyster, *Crassostrea virginica,* in North Carolina. *Estuaries* 15:158-170.

Osman, R. W. 1977. The establishment and development of a marine epifaunal community. *Ecol. Monog.* 47:37-63.

Osman, R. W. 1978. The influence of seasonality and stability on the species equilibrium. *Ecology* 59: 383-399.

Osman, R. W. 1982. Artificial substrates as ecological islands. In J. Carins, ed. Artificial Substrates. Ann Arbor Science Publishers, Inc., Massachusetts. pp. 71-114.

Osman, R. W. & T. A. Dean. 1987. Intra-and inter-regional comparisons of equilibrium numbers of species on marine hard substrate islands. *J. Biogeogr.* 14:53-67.

Osman, R. W. & R. B. Whitlach. 1999. Processes controlling local and regional patterns of invertebrate colonization: applications to the design of artificial oyster habitat. In: M. W. Luckenbach, R. Mann & J. A. Wesson (editors). *Oyster reef habitat restoration: a synopsis and synthesis of approaches.* Virginia Institute of Marine Science Press, Gloucester Point, USA, pp 179-197.

Paynter, K. T., Jr., J. D. Goodwin, M. E. Chen, N. J. Ward, M. W. Sherman, D. W. Meritt & S. K. Allen, Jr. 2008. *Crassostrea ariakensis* in Chesapeake Bay: growth, disease and mortality in shallow water environments. *J. Shellfish Res.* 27:509-515.

Peterson, C. H. & R. N. Lipcius. 2003. Conceptual progress towards predicting quantitative ecosystem benefits of ecological restorations. *Mar. Ecol. Prog. Ser.* 264:297-307.

Peyre, M. K., B. Gossman & J. A. Nyman. 2007. Assessing functional equivalency of nekton habitat in enhanced habitats: comparison of terraced and unterraced march ponds. *Estuar. Coast.* 30:526-536.

Posey, M. H., T. D. Alphin, C. M. Powell & E. Townsend. 1999. Use of oyster reefs as habitat for epibenthic fish and decapods. In: M. W. Luckenbach, R. Mann & J. A. Wesson (editors). *Oyster reef habitat restoration: a synopsis and synthesis of approaches.* Virginia Institute of Marine Science Press, Gloucester Point, USA, pp 229-237.

Rickards, W. L. & P. C. Ticco. 2002. The Suminoe oyster, *Crassostrea ariakensis,* in Chesapeake Bay: current status and near-term research activities. *Virginia Sea Grant Publication VSG-02-23.* Charlottesville, Virginia, 10pp.

Roberts, C. M. & R. F. G. Ormond. 1987. Habitat complexity and coral reef fish diversity and abundance on red sea fringing reefs. *Mar. Ecol. Prog. Ser.* 41:1-8.

Rodney, W. S. & K. T. Paynter. 2006. Comparisons of macrofaunal assemblages on restored and non-restored oyster reefs in mesohaline regions of Chesapeake Bay in Maryland. *J. Exp. Mar. Biol. Ecol.* 335:39-51.

Rogers, C. S., M. Gilnack & H. C. Fitz. 1983. Monitoring of coral reefs with linear transects: A study of storm damage. *J. Exp. Mar. Biol. Ecol.* 66:285-300.

Rothschild, B. J., J. S. Ault, P. Goulletquer & M. Heral. 1994. Decline of the Chesapeake Bay oyster population: a century of habitat destruction and overfishing. *Mar. Ecol. Prog. Ser.*111:29-39.

Roughgarden, J., Y. Iwasa & C. Baxter. 1985. Demographic theory for an open marine population with space-limited recruitment. *Ecology* 66:54-67.

Ruesink, J. L., H. S. Lenihan, A. C. Trimble, K. W. Heiman, F. Micheli, J. E. Byers & M. C. Kay. 2005. Introduction of nonnative oysters: ecosystem effects and restoration implications. *Annu. Rev. Ecol. Evol. Syst.* 36:643-689.

Shumway, C. A., H. A. Hofmann & A. P. Dobberfuhl. 2007. Quantifying habitat complexity in aquatic ecosystems. *Freshwater Biol.* 52:1065-1076.

Sutherland, J. P. and R. H. Karlson. 1977. Development and stability of the fouling community at Beaufort, North Carolina. *Ecol. Monogr.* 47:425-446.

Tenore, K. R. 1988. Nitrogen in benthic food chains. In: Nitrogen Cycling in Coastal Marine Environments. T. H. Blackburn & J. Sorenson (eds.), Wiley, Chichester.

Todd, C. D., J. N. Havenhand & J. P. Thorpe. 1988. Genetic differentiation, pelagic larval transport and gene flow between local populations of the intertidal marine mollusk *Adalaria proxima* (Alder & Hancock). *Functional Ecol.* 2:441-451.

Tolley, S. G., A. K. Volety & M. Savarese. 2005. Influence of salinity on the habitat use of oyster reefs in three southwest Florida estuaries. *J. Shellfish Res.* 24:127-137.

Zhou, M. & S. K. Allen, Jr. 2003. A review of published work on *Crassostrea ariakensis. J. Shellfish Res.* 22:1-20.

Zimmerman, R., T. Minello, T. Baumer, & M. Castiglione. 1989. Oyster reef as habitat for estuarine macrofauna. *National Oceanic and Atmospheric Administration, Technical Memorandum NMFS-SEFC-249.*

Table 1. Results from reduced model one-way ANOVAs on the effect of treatment within each combination of block and date on habitat complexity indices. Different letters within a row indicate significantly different means ($p<0.05$, Tukey's test).

Complexity Index	**F**	**p**	**Ranking**			
Maximum reef height (cm)						
Dec. '05	3.20	0.0484	mixed^{A}	*C. virginica*^{AB}	*C. ariakensis*^{AB}	shell^{B}
Apr. '06	3.11	0.0495	*C. ariakensis*^{A}	*C. virginica*^{AB}	mixed^{AB}	shell^{B}
Jul. '06	17.24	<0.0001	*C. ariakensis*^{A}	*C. virginica*^{A}	mixed^{A}	shell^{B}
Oct. '06	38.72	<0.0001	*C. ariakensis*^{A}	*C. virginica*^{A}	mixed^{A}	shell^{B}
Apr.'07	69.92	<0.0001	*C. ariakensis*^{A}	mixed^{A}	*C. virginica*^{A}	shell^{B}
Jul. '07	722.80	<0.0001	*C. ariakensis*^{A}	mixed^{AB}	*C. virginica*^{B}	shell^{C}
Sep. '07	178.02	<0.0001	*C. ariakensis*^{A}	mixed^{AB}	*C. virginica*^{B}	shell^{C}
Mean reef height (cm)						
Dec. '05	12.69	0.0001	mixed^{A}	*C. virginica*^{A}	*C. ariakensis*^{A}	shell^{B}
Apr. '06	11.13	0.0002	*C. ariakensis*^{A}	*C. virginica*^{A}	mixed^{A}	shell^{B}
Jul. '06	23.65	<0.0001	*C. ariakensis*^{A}	*C. virginica*^{A}	mixed^{A}	shell^{B}
Oct. '06	103.63	<0.0001	*C. virginica*^{A}	*C. ariakensis*^{A}	mixed^{A}	shell^{B}
Apr.'07	62.48	<0.0001	*C. ariakensis*^{A}	mixed^{A}	*C. virginica*^{A}	shell^{B}
Jul. '07	1277.7	<0.0001	mixed^{A}	*C. ariakensis*^{A}	*C. virginica*^{B}	shell^{C}
Sep. '07	131.73	<0.0001	*C. ariakensis*^{A}	mixed^{A}	*C. virginica*^{A}	shell^{B}
Surface Rugosity						
Dec. '05	3.24	0.0463	*C. virginica*^{A}	*C. ariakensis*^{AB}	mixed^{AB}	shell^{B}
Apr. '06	4.08	0.0206	*C. ariakensis*^{A}	mixed^{AB}	*C. virginica*^{AB}	shell^{B}
Jul. '06	8.50	0.0009	*C. ariakensis*^{A}	mixed^{A}	*C. virginica*^{A}	shell^{B}
Oct. '06	22.27	<0.0001	*C. virginica*^{A}	mixed^{A}	*C. ariakensis*^{A}	shell^{B}
Apr.'07	14.49	<0.0001	*C. ariakensis*^{A}	mixed^{A}	*C. virginica*^{A}	shell^{B}
Jul. '07	46.50	<0.0001	*C. ariakensis*^{A}	*C. virginica*^{A}	mixed^{A}	shell^{B}
Sep. '07	31.63	<0.0001	mixed^{A}	*C. ariakensis*^{A}	*C. virginica*^{A}	shell^{B}

Table 2. Summary of associated fauna found across all sites in July 2006.

	Dec. '05	Apr. '06	Jul. '06	Oct. '06	Apr. '07	Jul. '07	Sep. '07
Number of species	33	33	35	28	26	23	29
Number of individuals	14,029	17,680	32,419	9,574	21,267	28,869	14,147
Biomass of assocaited fuana (g)	29.05	145.72	571.05	141.11	400.64	716.9503	494.33
Biomass of oysters (g)	69.48	126.67	781.05	1605.031	3282.077	3868.178	3793.366
Biomass index for associated fauna*	0.42	1.15	0.73	0.09	0.12	0.19	0.13

*(ash-free dry weight of associated fauna / ash-free dry weight of oysters)

Table 3. Results from reduced model one-way ANOVAs on the effect of treatment within each combination of bloc and date on the total number of individuals present. Different letters within a row indicate significantly different me (p<0.05, Tukey's test).

Total Number of Individuals	**F**	**p**	**Ranking**
Block 1			
Dec. '05	2.19	0.1673	
Apr. '06	1.10	0.4021	
Jul. '06	0.37	0.7763	
Oct. '06	0.16	0.9194	
Apr.'07	1.23	0.3604	
Jul. '07	9.18	0.0057	*C. ariakensis*^{A} mixed^{AB} *C. virginica*^{B} shell^{B}
Sep. '07	2.43	0.1406	
Block 2			
Dec. '05	7.88	0.0090	*C. virginica*^{A} *C. ariakensis*^{A} mixed^{AB} shell^{B}
Apr. '06	11.25	0.0030	*C. virginica*^{A} mixed^{B} *C. ariakensis*^{B} shell^{B}
Jul. '06	4.56	0.0383	*C. ariakensis*^{A} *C. virginica*^{A} mixed^{AB} shell^{B}
Oct. '06	2.47	0.1363	
Apr.'07	0.72	0.5700	
Jul. '07	5.90	0.0200	*C. ariakensis*^{A} mixed^{A} *C. virginica*^{AB} shell^{B}
Sep. '07	1.59	0.2655	

Table 4. Results from reduced model one-way ANOVAs on the effect of treatment within each combination of block and date on community metrcis (species richness and Pielou's evenness). Different letters within a row indicate significant different means ($p<0.05$, Tukey's test).

	F	p	Ranking
Species Richness			
Block 1			
Dec. '05	1.32	0.3348	
Apr. '06	2.87	0.1034	
Jul. '06	0.20	0.8911	
Oct. '06	0.87	0.4960	
Apr.'07	0.31	0.8193	
Jul. '07	0.71	0.5731	
Sep. '07	2.24	0.1615	
Block 2			
Dec. '05	1.46	0.2970	
Apr. '06	3.72	0.0608	
Jul. '06	3.11	0.0887	
Oct. '06	0.80	0.5291	
Apr.'07	1.42	0.3074	
Jul. '07	1.44	0.3018	
Sep. '07	2.11	0.1779	
Species Evenness			
Block 1			
Dec. '05	1.35	0.3266	
Apr. '06	0.61	0.6246	
Jul. '06	1.86	0.2145	
Oct. '06	0.98	0.4507	
Apr.'07	0.73	0.5610	
Jul. '07	2.26	0.1581	
Sep. '07	1.14	0.3907	
Block 2			
Dec. '05	2.50	0.1332	
Apr. '06	2.74	0.1130	
Jul. '06	1.02	0.4338	
Oct. '06	21.33	0.0004	*C. ariakensis*^{A} shell^{A} mixed^{A} *C. virginica*^{B}
Apr.'07	1.12	0.3979	
Jul. '07	3.81	0.0579	
Sep. '07	2.01	0.1910	

Table 4 cont'd. Results from reduced model one-way ANOVAs on the effect of treatment within each combination of block and date on community metrics (dominance and Shannon-Weiner diversity). Different letters within a row indicate significar different means ($p<0.05$, Tukey's test).

	F	p	Ranking
Dominance			
Block 1			
Dec. '05	2.38	0.1453	
Apr. '06	3.08	0.0906	
Jul. '06	0.36	0.7836	
Oct. '06	0.75	0.5518	
Apr.'07	0.14	0.9361	
Jul. '07	0.43	0.7402	
Sep. '07	0.26	0.8508	
Block 2			
Dec. '05	2.30	0.1544	
Apr. '06	3.51	0.0690	
Jul. '06	3.76	0.0596	
Oct. '06	0.43	0.7380	
Apr.'07	1.50	0.2858	
Jul. '07	1.82	0.2210	
Sep. '07	1.44	0.3014	
Shannon-Weiner Diversity			
Block 1			
Dec. '05	3.31	0.0780	
Apr. '06	0.46	0.7196	
Jul. '06	3.33	0.0773	
Oct. '06	1.06	0.4181	
Apr.'07	0.39	0.7635	
Jul. '07	1.69	0.2466	
Sep. '07	1.35	0.3255	
Block 2			
Dec. '05	5.23	0.0274	shell^{A} *C. virginica*^{AB} mixed^{AB} *C. ariakensis*^{B}
Apr. '06	5.38	0.0254	*C. virginica*^{A} *C. ariakensis*^{AB} shell^{AB} mixed^{B}
Jul. '06	1.64	0.2567	
Oct. '06	14.11	0.0015	*C. ariakensis*^{A} mixed^{A} shell^{A} *C. virginica*^{B}
Apr.'07	1.46	0.2958	
Jul. '07	4.69	0.0357	shell^{A} mixed^{AB} *C. virginica*^{AB} *C. ariakensis*^{B}
Sep. '07	1.88	0.2118	

Table 5. Results from reduced model one-way ANOVAs on the effect of treatment within each combination of block and date on standardized total abundance. Different letters within a row indicate significantly different means ($p<0.05$, Tukey's test).

	F	p	Ranking		
Standardized Total Abundance					
Block 1					
Dec. '05	0.96	0.434			
Apr. '06	0.50	0.627			
Jul. '06	8.96	0.016	*C. virginica*^{A}	mixed^{AB}	*C. ariakensis*^{B}
Oct. '06	0.55	0.6050			
Apr.'07	2.43	0.168			
Jul. '07	0.68	0.54			
Sep. '07	1.96	0.221			
Block 2					
Dec. '05	5.17	0.05	*C. virginica*^{A}	*C. ariakensis*^{AB}	mixed^{B}
Apr. '06	3.06	0.122			
Jul. '06	10.75	0.01	*C. virginica*^{A}	mixed^{B}	*C. ariakensis*^{B}
Oct. '06	0.35	0.721			
Apr.'07	13.35	0.006	*C. ariakensis*^{A}	mixed^{B}	*C. virginica*^{B}
Jul. '07	8.16	0.019	*C. ariakensis*^{A}	mixed^{AB}	*C. virginica*^{B}
Sep. '07	2.10	0.204			
Standardized Total Biomass					
Block 1					
Dec. '05	0.17	0.851			
Apr. '06	8.7	0.017	*C. virginica*^{A}	mixed^{B}	*C. ariakensis*^{B}
Jul. '06	2.69	0.147			
Oct. '06	3.58	0.095			
Apr.'07	1.34	0.331			
Jul. '07	0.88	0.461			
Sep. '07	1.53	0.29			
Block 2					
Dec. '05	7.96	0.021	*C. virginica*^{A}	mixed^{B}	*C. ariakensis*^{B}
Apr. '06	10.85	0.01	*C. virginica*^{A}	*C. ariakensis*^{B}	mixed^{B}
Jul. '06	11.12	0.01	*C. virginica*^{A}	mixed^{B}	*C. ariakensis*^{B}
Oct. '06	0.38	0.697			
Apr.'07	1.37	0.324			
Jul. '07	5.01	0.053			
Sep. '07	1.70	0.26			

Table 6. Complete list of all species found during each sampling event.

Taxonomic Group	Species	Dec. '05	Apr. '06	Jul. '06	Oct. '06	Apr. '07	Jul. '07	Sep. '07
Amphipods								
	Apocorophium lacustre	X	X	X	X	X	X	X
	Cymadusa compta	X	X			X		X
	Elasmopus levis	X	X		X	X	X	
	Gammarus mucronatus	X	X	X		X	X	
	Gammarus palustris	X	X	X				
	Melita nitida	X	X	X	X	X	X	X
Bivalves								
	Ensis directus						X	
	Gemma gemma	X	X	X	X		X	X
	Geukensia demissa		X	X				X
	Ischadium recurvum	X	X	X	X	X	X	X
	Macoma balthica	X	X	X	X	X	X	X
	Macoma mitchelli			X				
	Macoma tenta	X						X
	Mulinia lateralis	X	X	X	X	X	X	X
	Mya arenaria	X	X	X	X	X	X	X
Cnidarian								
	U/I anemone	X	X	X	X	X	X	X
	U/I jelly	X	X	X	X	X	X	X
Decapod Crustaceans								
	Callinectes sapidus	X	X	X				
	Dyspanopeus sayi			X	X			
	Eurypanopeus depressus	X	X	X	X	X	X	X
	Palaemonetes pugio	X	X		X	X		
	Panopeus herbstii			X	X		X	X
	Rhithropanopeus harrisii	X	X	X				
	U/I juvenile xanthid	X	X		X	X	X	X
Fishes								
	Anguilla rostrata		X				X	
	Chasmodes bosquianus			X	X	X	X	X
	Gobiesox strumosus	X	X	X	X	X	X	X
	Gobiosoma bosc	X	X	X	X	X	X	X
	Opsanus tau			X	X	X	X	X
	Syngnathus fucus				X			
Gastropods								
	Acteocina canaliculata	X	X	X	X			X
	Boonea bisuturalis	X	X	X				
	Boonea impressa	X	X	X	X	X	X	X
	Crepidula fornicata				X			
	Nassarius vibex		X	X	X	X		
	Rictaxis punctostriatus	X		X				
Nemerteans		X						X
Polychaetes								
	Eteone heteropoda	X	X			X		X
	Heteromastus filiformis	X	X	X	X	X	X	X
	Leitoscoloplos fragilis		X					X

Table 6 cont'd. Complete list of all species found during each sampling event.

Taxonomic Group	**Species**	**Dec. '05**	**Apr. '06**	**Jul. '06**	**Oct. '06**	**Apr. '07**	**Jul. '07**	**Sep. '07**
Polychaetes								
	Mediomastus ambiseta				X			
	Neanthes succinea	X	X	X	X	X	X	X
	Pectinaria gouldii	X	X	X		X		
	Polydora websteri	X	X	X	X	X		X
	Streblospio benedicti	X						X
	Stylocus sp.	X	X					

Table 7. Reduced-model one-way ANOVA results for the effect of live oyster treatment on individual species abundances comprising at least one per cent of total standardized abudances for each sampling period. Treatments are ranked in descending order; different letters within a row indicate significantly different values ($p<0.05$, Tukey's).

Date	Block	Species	F	p	Ranking
Dec. '05					
	1				
		Apocorophium lacustre	4.19	0.0726	
		Eteone heteropoda	0.09	0.9181	
		Melita nitida	2.20	0.1920	
		Mulinia lateralis	3.02	0.1236	
		Mya arenaria	1.84	0.2382	
		Neanthes succinea	1.20	0.3647	
		Polydora websteri	2.17	0.1955	
		U/I juvenile xanthid	0.08	0.9251	
	2				
		Apocorophium lacustre	1.40	0.3161	
		Eteone heteropoda	4.81	0.0567	
		Melita nitida	9.58	0.0136	*C. virginica*^{A} *C. ariakensis*^{A} mixed^{B}
		Mulinia lateralis	14.58	0.0050	*C. virginica*^{A} mixed^{B} *C. ariakensis*^{B}
		Mya arenaria	0.02	0.9796	
		Neanthes succinea	0.11	0.8940	
		Polydora websteri	2.26	0.1852	
		U/I juvenile xanthid	14.85	0.0048	*C. virginica*^{A} *C. ariakensis*^{A} mixed^{B}
Apr. '06					
	1				
		Acteocina canaliculata	0.29	0.7595	
		Apocorophium lacustre	2.71	0.1453	
		Gemma gemma	0.40	0.6899	
		Macoma balthica	0.10	0.9053	
		Melita nitida	5.15	0.0499	*C. ariakensis*^{A} *C. virginica*^{AB} mixed^{B}
		Mulinia lateralis	1.50	0.2957	
		Mya arenaria	0.09	0.9177	
		Neanthes succinea	0.62	0.5671	
		Polydora websteri	0.41	0.6837	
		U/I juvenile xanthid	2.59	0.1549	
	2				
		Acteocina canaliculata	0.50	0.6295	
		Apocorophium lacustre	6.16	0.0351	*C. virginica*^{A} mixed^{AB} *C. ariakensis*^{B}
		Gemma gemma	1.00	0.4219	
		Macoma balthica	6.04	0.0365	*C. virginica*^{A} mixed^{B} *C. ariakensis*^{B}
		Melita nitida	4.28	0.0698	
		Mulinia lateralis	3.45	0.1005	
		Mya arenaria	9.91	0.0125	*C. virginica*^{A} mixed^{B} *C. ariakensis*^{B}
		Neanthes succinea	0.82	0.4848	
		Polydora websteri	2.86	0.1341	
		U/I juvenile xanthid	12.17	0.0077	*C. virginica*^{A} *C. ariakensis*^{B} mixed^{B}

Table 7 cont'd. Reduced-model one-way ANOVA results for the effect of live oyster treatment on individual species abundances comprising at least one per cent of total standardized abudances for each sampling period. Treatments are ranked in descending order; different letters within a row indicate significantly different values (p<0.05, Tukey's).

Date	Block	Species	F	p	Ranking		
Jul. '06							
	1						
		Eurypanopeus depressus	14.73	0.0048	*C. virginica*^{A}	mixed^{B}	*C. ariakensis*^{B}
		Gammarus palustris	4.89	0.0550			
		Ischadium recurvum	2.31	0.1204			
		Macoma balthica	1.56	0.2851			
		Melita nitida	1.15	0.3773			
		Mya arenaria	5.04	0.0519			
		Neanthes succinea	11.93	0.0081	*C. virginica*^{A}	mixed^{AB}	*C. ariakensis*^{B}
	2						
		Eurypanopeus depressus	7.14	0.0259	*C. virginica*^{A}	mixed^{B}	*C. ariakensis*^{B}
		Gammarus palustris	7.80	0.0214	*C. virginica*^{A}	mixed^{AB}	*C. ariakensis*^{B}
		Ischadium recurvum	2.62	0.1025			
		Macoma balthica	5.94	0.0378	*C. virginica*^{A}	mixed^{AB}	*C. ariakensis*^{B}
		Melita nitida	2.65	0.1493			
		Mya arenaria	22.64	0.0016	*C. virginica*^{A}	mixed^{B}	*C. ariakensis*^{C}
		Neanthes succinea	8.57	0.0174	*C. virginica*^{A}	mixed^{AB}	*C. ariakensis*^{B}
Oct. '06							
	1						
		Apocorophium lacustre	0.70	0.5337			
		Eurypanopeus depressus	0.27	0.7734			
		Gobiosoma bosc	3.35	0.1054			
		Heteromastus filiformis	4.50	0.0640			
		Ischadium recurvum	0.93	0.4437			
		Macoma balthica	6.94	0.0275	*C. ariakensis*^{A}	*C. virginica*^{AB}	mixed^{B}
		Melita nitida	2.73	0.1437			
		Mya arenaria	1.21	0.3611			
		Neanthes succinea	0.01	0.9876			
		U/I anemone	1.68	0.2638			
		U/I jelly	2.38	0.1735			
		U/I juvenile xanthid	0.04	0.9628			
	2						
		Apocorophium lacustre	1.39	0.3200			
		Eurypanopeus depressus	2.54	0.1589			
		Gobiosoma bosc	17.01	0.0034	mixed^{A}	*C. virginica*^{A}	*C. ariakensis*^{B}
		Heteromastus filiformis	0.80	0.4904			
		Ischadium recurvum	0.78	0.4992			
		Macoma balthica	1.00	0.4219			
		Melita nitida	9.61	0.0135	*C. ariakensis*^{A}	*C. virginica*^{B}	mixed^{B}
		Mya arenaria	0.92	0.449			
		Neanthes succinea	2.10	0.2031			
		U/I anemone	0.40	0.6897			
		U/I jelly	0.54	0.6102			
		U/I juvenile xanthid	0.08	0.9272			

Table 7 cont'd. Reduced-model one-way ANOVA results for the effect of live oyster treatment on individual species abundances comprising at least one per cent of total standardized abudances for each sampling period. Treatments are ranked in descending order; different letters within a row indicate significantly different values ($p<0.05$, Tukey's).

Date	Block	Species	F	p	Ranking
Apr. '07					
	1				
		Apocorophium lacustre	1.04	0.4109	
		Eteone heteropoda	0.94	0.4421	
		Eurypanopeus depressus	2.41	0.1707	
		Ischadium recurvum	2.09	0.2043	
		Macoma balthica	6.28	0.0338	*C. ariakensis* ^{A} mixed^{AB} *C. virginica* ^{B}
		Melita nitida	2.85	0.1346	
		Mya arenaria	0.47	0.6438	
		Neanthes succinea	2.48	0.1642	
		U/I anemone	3.48	0.0995	
		U/I juvenile xanthid	5.09	0.0511	
	2				
		Apocorophium lacustre	1.24	0.3539	
		Eteone heteropoda	1.27	0.3470	
		Eurypanopeus depressus	3.47	0.0996	
		Ischadium recurvum	5.97	0.0375	*C. ariakensis* ^{A} *C. virginica* ^{AB} mixed^{B}
		Macoma balthica	0.01	0.9896	
		Melita nitida	1.61	0.2753	
		Mya arenaria	1.61	0.2753	
		Neanthes succinea	12.19	0.0077	*C. ariakensis* ^{A} mixed^{B} *C. virginica* ^{B}
		U/I anemone	2.21	0.1906	
		U/I juvenile xanthid	13.34	0.0062	*C. ariakensis* ^{A} *C. virginica* ^{B} mixed^{B}
Jul. '07					
	1				
		Apocorophium lacustre	15.57	0.0042	*C. ariakensis* ^{A} mixed^{A} *C. virginica* ^{B}
		Eurypanopeus depressus	0.39	0.6903	
		Ischadium recurvum	0.74	0.5153	
		Macoma balthica	0.13	0.8793	
		Melita nitida	0.28	0.7652	
		Mya arenaria	0.49	0.6339	
		Neanthes succinea	0.27	0.7741	
	2				
		Apocorophium lacustre	2.71	0.1450	
		Eurypanopeus depressus	1.12	0.3852	
		Ischadium recurvum	0.54	0.6103	
		Macoma balthica	1.13	0.3840	
		Melita nitida	1.26	0.3503	
		Mya arenaria	2.95	0.1280	
		Neanthes succinea	9.10	0.0153	*C. ariakensis* ^{A} mixed^{AB} *C. virginica* ^{B}

Table 7 cont'd. Reduced-model one-way ANOVA results for the effect of live oyster treatment on individual species abundances comprising at least one per cent of total standardized abudances for each sampling period. Treatments are ranked in descending order; different letters within a row indicate significantly different values ($p<0.05$, Tukey's).

Date	Block	Species	F	p	Ranking
Sep. '07					
	1				
		Apocorophium lacustre	28.37	0.0009	*C. ariakensis* ^{A} mixed^{A} *C. virginica* ^{B}
		Eurypanopeus depressus	0.14	0.8695	
		Gobiosoma bosc	8.21	0.0192	*C. ariakensis* ^{A} *C. virginica* ^{B} mixed^{B}
		Ischadium recurvum	1.36	0.3261	
		Macoma balthica	1.07	0.4013	
		Melita nitida	9.02	0.0155	*C. ariakensis* ^{A} mixed^{AB} *C. virginica* ^{B}
		Mya arenaria	0.73	0.5219	
		Neanthes succinea	0.04	0.9620	
		U/I anemone	0.15	0.8620	
		U/I juvenile xanthid	0.85	0.4737	
	2				
		Apocorophium lacustre	4.56	0.0626	
		Eurypanopeus depressus	0.24	0.7960	
		Gobiosoma bosc	0.53	0.612	
		Ischadium recurvum	1.57	0.2826	
		Macoma balthica	1.23	0.3578	
		Melita nitida	0.22	0.8068	
		Mya arenaria	0.22	0.8098	
		Neanthes succinea	3.37	0.1045	
		U/I anemone	0.73	0.5187	
		U/I juvenile xanthid	0.76	0.5070	

Table 8. Reduced-model one-way ANOVA results for the effect of live oyster treatment on individual species biomass comprising at least one per cent of total standardized biomass for each sampling period. Treatments are ranked in descending order; different letters within a row indicate significantly different values ($p<0.05$, Tukey's).

Date	Block	Species	F	p	Ranking
Dec. '05					
	1				
		Callinectes sapidus	0.31	0.7434	
		Eteone heteropoda	0.76	0.5067	
		Eurypanopeus depressus	1.00	0.4219	
		Gobiesox strumosus	0.54	0.6086	
		Gobiosoma bosc	2.74	0.1430	
		Macoma tenta	1.01	0.4200	
		Melita nitida	0.09	0.9145	
		Neanthes succinea	0.11	0.9003	
		Palaemonetes pugio	0.22	0.8084	
		U/I juvenile xanthid	1.94	0.2238	
	2				
		Callinectes sapidus	0.37	0.7042	
		Eteone heteropoda	8.13	0.0196	*C. ariakensis*^{A} *C. virginica*^{AB} mixed^{B}
		Eurypanopeus depressus	6.67	0.0299	*C. virginica*^{A} *C. ariakensis*^{AB} mixed^{B}
		Gobiesox strumosus	2.20	0.1916	
		Gobiosoma bosc	7.35	0.0244	*C. virginica*^{A} *C. ariakensis*^{AB} mixed^{B}
		Melita nitida	9.99	0.0123	*C. virginica*^{A} *C. ariakensis*^{AB} mixed^{B}
		Neanthes succinea	9.40	0.0141	*C. virginica*^{A} mixed^{B} *C. ariakensis*^{B}
		Palaemonetes pugio	2.32	0.1788	
		U/I juvenile xanthid	5.28	0.0475	*C. virginica*^{A} *C. ariakensis*^{AB} mixed^{B}
Apr. '06					
	1				
		Eurypanopeus depressus	0.96	0.4339	
		Gobiosoma bosc	0.71	0.5274	
		Ischadium recurvum	1.17	0.3721	
		Mya arenaria	5.14	0.0501	
		Neanthes succinea	17.79	0.0030	*C. virginica*^{A} mixed^{B} *C. ariakensis*^{B}
	2				
		Eurypanopeus depressus	5.98	0.0373	*C. ariakensis*^{A} *C. virginica*^{AB} mixed^{B}
		Gobiosoma bosc	157.25	<0.0001	*C. virginica*^{A} *C. ariakensis*^{B} mixed^{B}
		Ischadium recurvum	1.38	0.3211	
		Mya arenaria	34.61	0.0005	*C. virginica*^{A} mixed^{B} *C. ariakensis*^{B}
		Neanthes succinea	34.98	0.0005	*C. virginica*^{A} mixed^{B} *C. ariakensis*^{B}

Table 8 cont'd. Reduced-model one-way ANOVA results for the effect of live oyster treatment on individual species biomass comprising at least one per cent of total standardized biomass for each sampling period. Treatments are ranked in descending order; different letters within a row indicate significantly different values ($p<0.05$, Tukey's).

Date	Block	Species	F	p	Ranking
Jul. '06					
	1				
		Boonea bisuturalis	0.93	0.4432	
		Eurypanopeus depressus	0.72	0.5253	
		Gammarus mucronatus	0.72	0.5231	
		Gammarus palustris	1.27	0.3474	
		Gemma gemma	0.58	0.5883	
		Gobiesox strumosus	2.48	0.1642	
		Gobiosoma bosc	0.78	0.5010	
		Ischadium recurvum	1.2	0.3644	
		Macoma balthica	1.79	0.2456	
		Macoma mitchelli	0.82	0.4844	
		Mya arenaria	6.82	0.0286	*C. virginica* ^{A} mixed^{AB} *C. ariakensis* ^{B}
		Neanthes succinea	0.03	0.9719	
		Panopeus herbstii	1.32	0.3360	
		U/I anemone	1.03	0.4114	
	2				
		Apocorophium lacustre	2.52	0.1141	
		Boonea bisuturalis	23.40	0.0015	*C. virginica* ^{A} mixed^{B} *C. ariakensis* ^{B}
		Eurypanopeus depressus	1.63	0.2723	
		Gammarus mucronatus	0.50	0.6292	
		Gammarus palustris	1.41	0.3138	
		Gobiesox strumosus	0.90	0.4556	
		Gobiosoma bosc	0.92	0.4469	
		Ischadium recurvum	1.56	0.2848	
		Macoma balthica	0.60	0.5795	
		Macoma mitchelli	0.75	0.5140	
		Mya arenaria	0.90	0.4560	
		Neanthes succinea	0.83	0.4799	
		Panopeus herbstii	3.37	0.1044	
		U/I anemone	0.91	0.4505	
Oct. '06					
	1				
		Chasmodes bosquianus	1.64	0.2699	
		Elasmopus levis	1.08	0.3981	
		Eurypanopeus depressus	2.03	0.2127	
		Gobiesox strumosus	1.22	0.3599	
		Gobiosoma bosc	0.99	0.4243	
		Ischadium recurvum	0.31	0.7469	
		Mya arenaria	0.76	0.5077	
		Neanthes succinea	0.50	0.6297	
		Opsanus tau	1.48	0.3002	
		Panopeus herbstii	1.00	0.4219	

Table 8 cont'd. Reduced-model one-way ANOVA results for the effect of live oyster treatment on individual species biomass comprising at least one per cent of total standardized biomass for each sampling period. Treatments are ranked in descending order; different letters within a row indicate significantly different values ($p<0.05$, Tukey's).

Date	Block	Species	F	p	Ranking
Oct. '06					
	2				
		Chasmodes bosquianus	0.50	0.6300	
		Elasmopus levis	1.00	0.4219	
		Eurypanopeus depressus	0.82	0.4917	
		Gobiesox strumosus	0.92	0.4475	
		Gobiosoma bosc	6.70	0.0296	*C. virginica*^{A} mixed^{AB} *C. ariakensis*^{B}
		Ischadium recurvum	0.73	0.5206	
		Mya arenaria	2.33	0.1778	
		Neanthes succinea	5.62	0.0421	*C. virginica*^{A} mixed^{AB} *C. ariakensis*^{B}
		Opsanus tau	0.16	0.8579	
		Panopeus herbstii	1.00	0.4219	
Apr. '07					
	1				
		Chasmodes bosquianus	7.69	0.0221	
		Eurypanopeus depressus	13.34	0.0062	*C. virginica*^{A} mixed^{A} *C. ariakensis*^{B}
		Gobiesox strumosus	2.99	0.1255	
		Gobiosoma bosc	6.30	0.0336	*C. virginica*^{A} mixed^{AB} *C. ariakensis*^{B}
		Ischadium recurvum	0.98	0.4271	
		Macoma balthica	1.02	0.4159	
		Mya arenaria	0.28	0.7625	
		Neanthes succinea	0.40	0.6867	
		Opsanus tau	0.90	0.4541	
		Palaemonetes pugio	1.39	0.3187	
	2				
		Chasmodes bosquianus	0.91	0.4530	
		Eurypanopeus depressus	1.87	0.2333	
		Gobiesox strumosus	0.00	0.9962	
		Gobiosoma bosc	1.25	0.3528	
		Ischadium recurvum	1.75	0.2515	
		Macoma balthica	1.01	0.4203	
		Mya arenaria	0.5	0.6282	
		Neanthes succinea	1.28	0.3434	
		Opsanus tau	1.08	0.3961	
		Palaemonetes pugio	11.70	0.0085	*C. virginica*^{A} mixed^{B} *C. ariakensis*^{B}

Table 8 cont'd. Reduced-model one-way ANOVA results for the effect of live oyster treatment on individual species biomass comprising at least one per cent of total standardized biomass for each sampling period. Treatments are ranked in descending order; different letters within a row indicate significantly different values ($p<0.05$, Tukey's).

Date	Block	Species	F	p	Ranking
Jul. '07					
	1				
		Eurypanopeus depressus	10.69	0.0105	*C. virginica*^{A} mixed^{AB} *C. ariakensis*^{B}
		Gobiosoma bosc	1.40	0.3178	
		Ischadium recurvum	1.14	0.3808	
		Macoma balthica	0.25	0.7885	
		Mya arenaria	0.38	0.6989	
		Neanthes succinea	1.66	0.2667	
		Opsanus tau	0.30	0.7524	
	2				
		Eurypanopeus depressus	7.73	0.0218	*C. virginica*^{A} mixed^{AB} *C. ariakensis*^{B}
		Gobiosoma bosc	2.53	0.1597	
		Ischadium recurvum	0.03	0.9704	
		Macoma balthica	0.80	0.4901	
		Mya arenaria	3.11	0.1186	
		Neanthes succinea	24.39	0.0013	mixed^{A} *C. virginica*^{A} *C. ariakensis*^{B}
		Opsanus tau	0.97	0.4328	
		Panopeus herbstii	1.65	0.2682	
Sep. '07					
	1				
		Chasmodes bosquianus	0.10	0.9057	
		Eurypanopeus depressus	1.25	0.3506	
		Gobiesox strumosus	0.50	0.6296	
		Gobiosoma bosc	2.41	0.1703	
		Ischadium recurvum	0.23	0.7988	
		Macoma balthica	1.53	0.2905	
		Mya arenaria	1.12	0.3854	
		Neanthes succinea	10.61	0.0107	*C. virginica*^{A} mixed^{AB} *C. ariakensis*^{B}
		Opsanus tau	0.45	0.6599	
	2				
		Chasmodes bosquianus	0.58	0.5873	
		Eurypanopeus depressus	1.57	0.2827	
		Gobiesox strumosus	0.86	0.4679	
		Gobiosoma bosc	1.54	0.2884	
		Ischadium recurvum	0.75	0.5110	
		Macoma balthica	1.20	0.3637	
		Mya arenaria	0.25	0.7865	
		Neanthes succinea	1.41	0.3156	
		Opsanus tau	4.72	0.0587	

Table 9. Reduced-model one-way ANOVA results for the effect of sampling date on individual species abundances of predatory demersal fishes. Dates are ranked in descending order; different letters within a row indicate significantly different values ($P<0.05$, Tukey's). ($p<0.05$, Tukey's).

Species	Block	F	p	Ranking
Chasmodes bosquianus				
	1	2.97	0.0137	Sep. 07^{A} Apr. 07^{AB} Jul. 07^{AB} Oct. 06^{AB} Jul. 06^{AB} Dec 05^{B} Apr. 06^{B}
	2	8.31	<0.0001	Apr. 07^{A} Sep. 07A Oct. 06^{AB} Jul. 07^{AB} Jul. 06^{B} Dec 05^{B} Apr. 06^{B}
Gobiesox strumosus				
	1	4.65	0.0007	Jul. 06^{A} Sep. 07^{A} Apr. 07^{AB} Oct. 06^{AB} Jul. 07^{AB} Dec 05^{AB} Apr. 06^{B}
	2	7.66	<0.0001	Jul. 06^{A} Sep. 07^{A} Oct. 06^{A} Apr. 07^{AB} Jul. 07^{BC} Dec 05^{BC} Apr. 06^{C}
Gobiosoma bosc				
	1	42.7	<0.0001	Sep. 07^{A} Jul. 07^{B} Oct. 06^{BC} Jul. 06^{C} Apr. 07^{CD} Dec 05^{D} Apr. 06^{D}
	2	49.8	<0.0001	Sep. 07^{A} Jul. 07^{B} Oct. 06^{B} Jul. 06^{BC} Apr. 07^{CD} Dec 05^{D} Apr. 06^{D}
Opsanus tau				
	1	7.91	<0.0001	Oct. 06^{A} Jul. 07^{A} Sep. 07^{A} Apr. 07^{AB} Jul. 06^{B} Apr. 06^{B} Dec 05^{B}
	2	22.3	<0.0001	Jul. 07^{A} Sep. 07^{AB} Oct. 06^{AB} Apr. 07^{B} Jul. 06^{C} Apr. 06^{C} Dec 05^{C}

Figure 3-1. Habitat complexity indices of experimental treatments over time: (A) maximum reef height, (B) mean reef height, and (C) surface rugosity.

Error bars represent the standard error of each mean and (*) over bars indicate significantly different values between treatments for that date ($p<0.05$, Tukey's test).

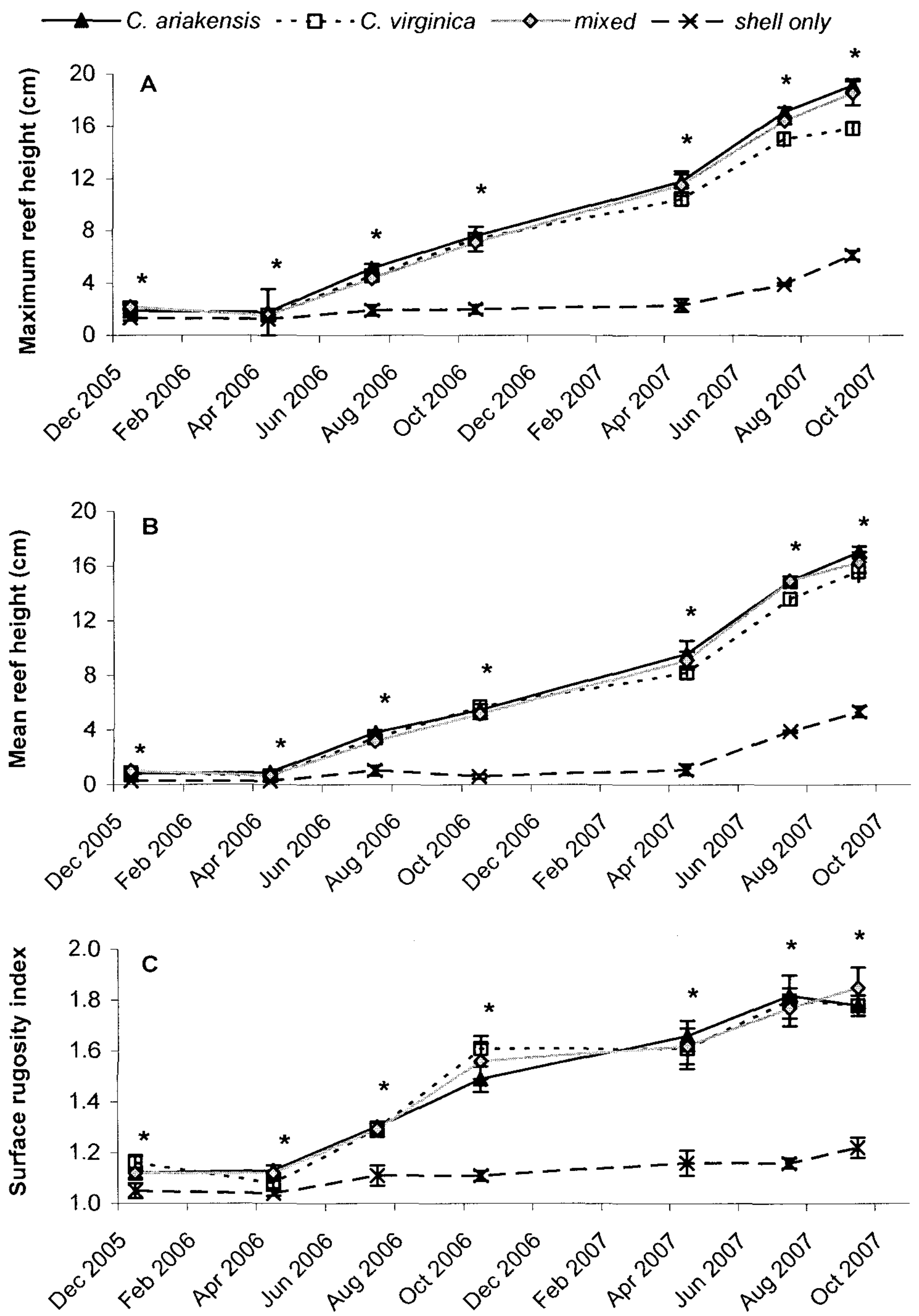

C. ariakensis
C. virginica
mixed
shell only
A
Maximum reef height (cm)
0
4
8
12
16
20
B
Mean reef height (cm)
0
4
8
12
16
20
C
Surface rugosity index
1.0
1.2
1.4
1.6
1.8
2.0
Dec 2005
Feb 2006
Apr 2006
Jun 2006
Aug 2006
Oct 2006
Dec 2006
Feb 2007
Apr 2007
Jun 2007
Aug 2007
Oct 2007
*

Figure 3-2. Mean total number of associated fauna per sample (un-standardized) for all treatments within each block (1 & 2) over time.

Error bars represent the standard effort of each mean and (*) over bars indicate significant within-date treatment effects.

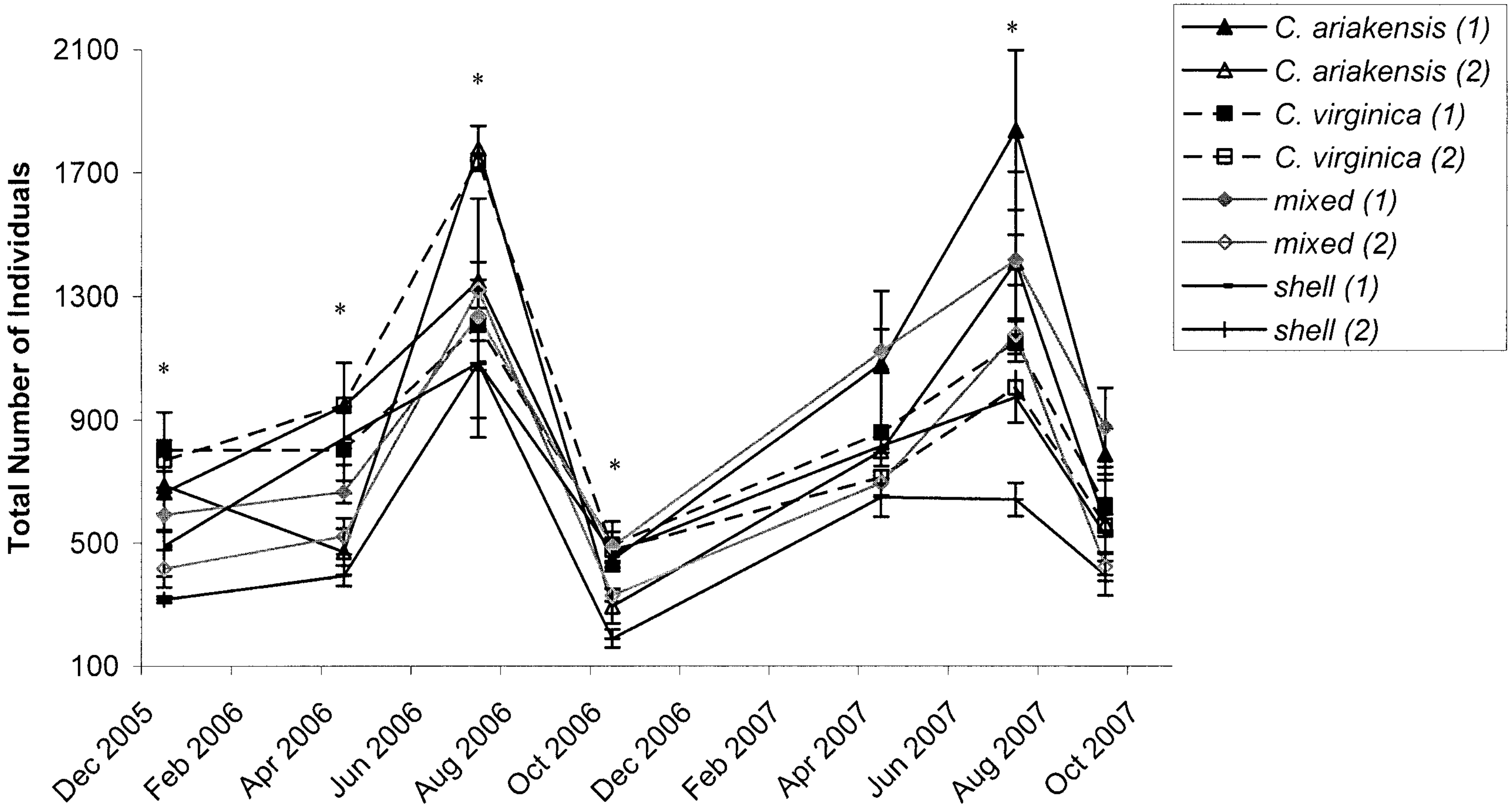

Total Number of Individuals
2100
1700
1300
900
500
100
Dec 2005
Feb 2006
Apr 2006
Jun 2006
Aug 2006
Oct 2006
Dec 2006
Feb 2007
Apr 2007
Jun 2007
Aug 2007
Oct 2007
*
C. ariakensis (1)
C. ariakensis (2)
C. virginica (1)
C. virginica (2)
mixed (1)
mixed (2)
shell (1)
shell (2)

Figure 3.3. Mean species richness per sample for all treatments within each block (1 & 2) over time.

Error bars represent the standard effort of each mean and (*) over bars indicate significant within-date treatment effects.

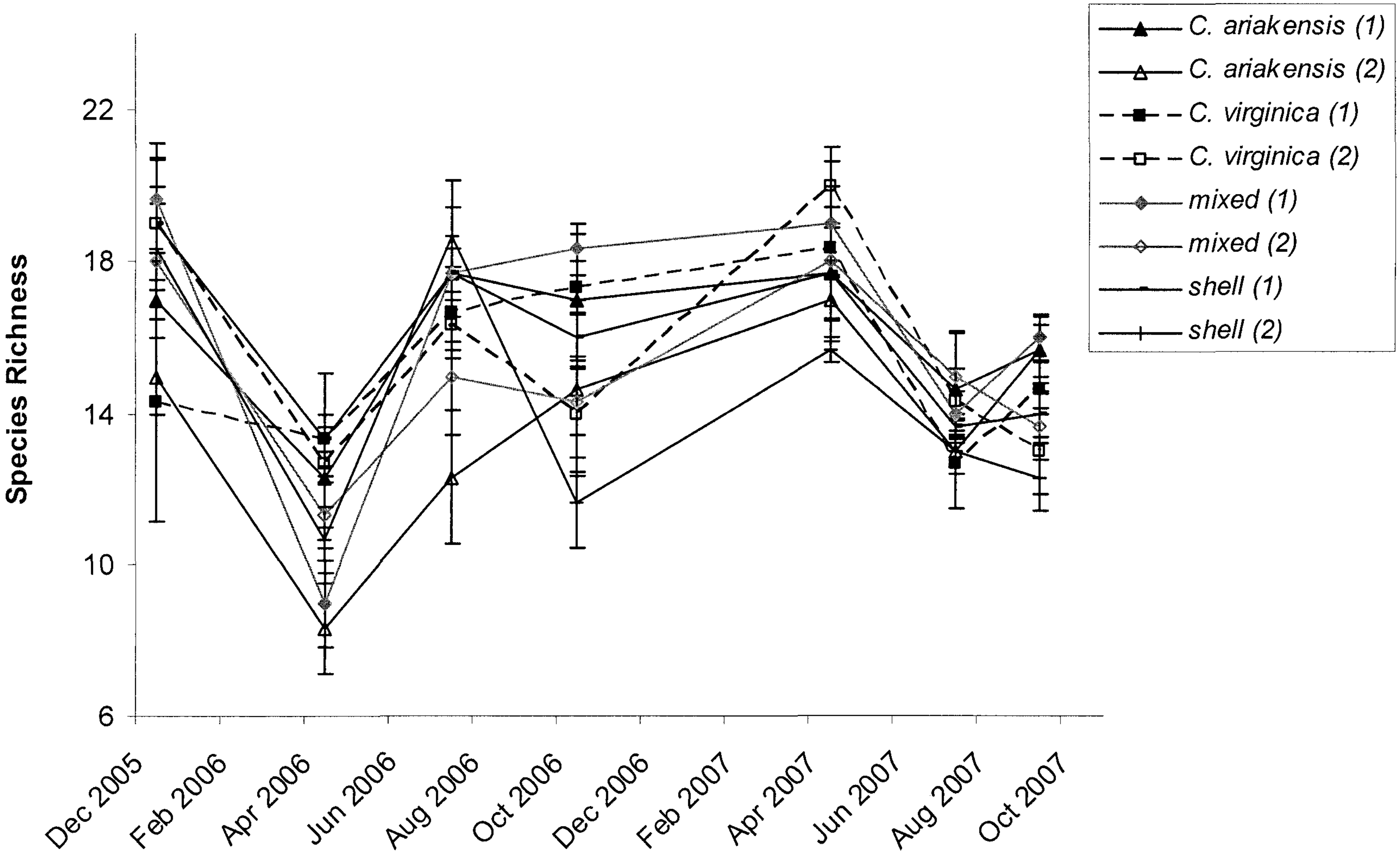

Species Richness
22
18
14
10
6
Dec 2005
Feb 2006
Apr 2006
Jun 2006
Aug 2006
Oct 2006
Dec 2006
Feb 2007
Apr 2007
Jun 2007
Aug 2007
Oct 2007
C. ariakensis (1)
C. ariakensis (2)
C. virginica (1)
C. virginica (2)
mixed (1)
mixed (2)
shell (1)
shell (2)

FIGURE 3-4. Mean values for Peilou's evenness per sample for all treatments within each block (1 & 2) over time.

Error bars represent the standard effort of each mean and (*) over bars indicate significant within-date treatment effects.

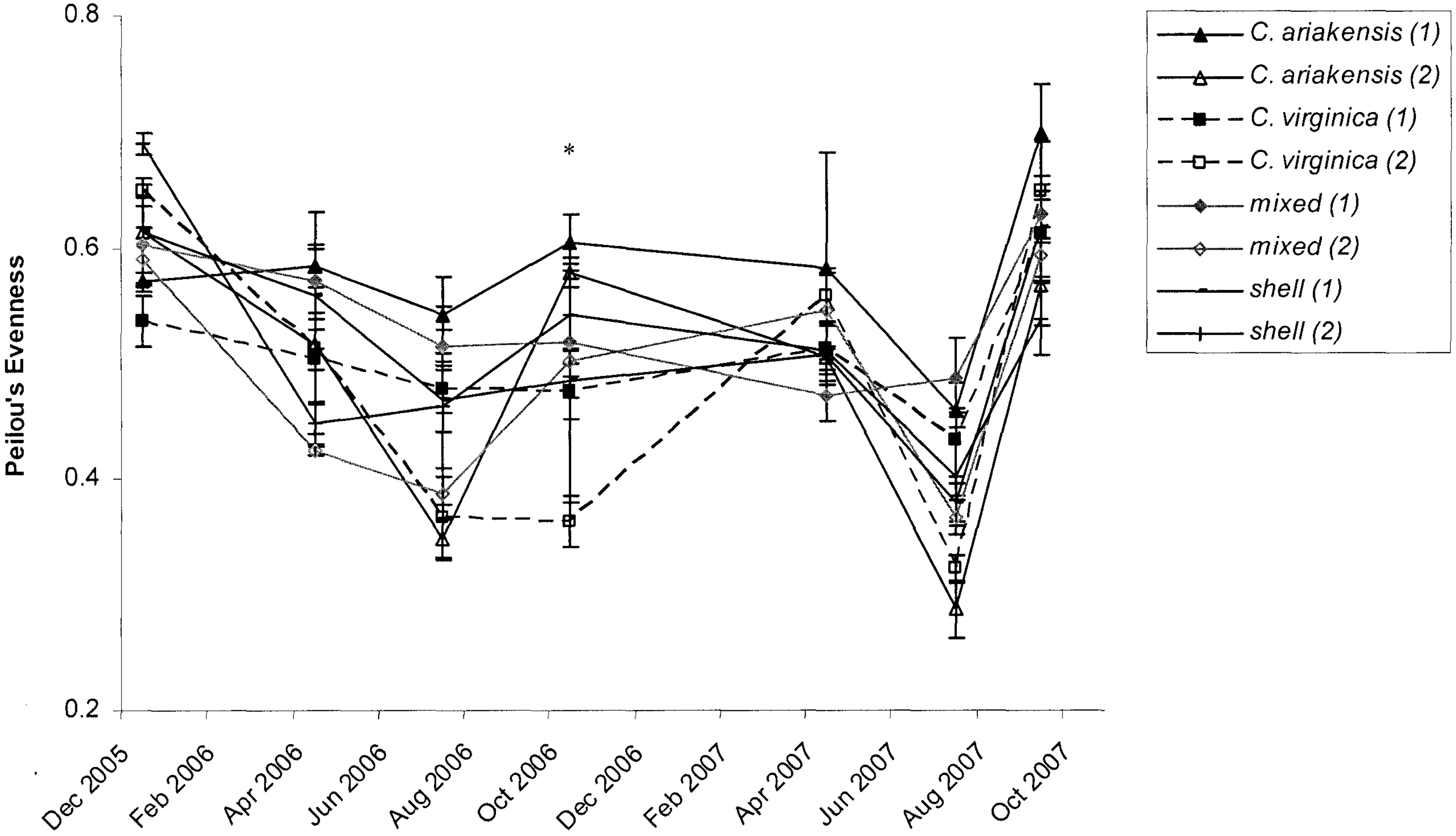

Pielou's Evenness
0.8
0.6
0.4
0.2
Dec 2005
Feb 2006
Apr 2006
Jun 2006
Aug 2006
Oct 2006
Dec 2006
Feb 2007
Apr 2007
Jun 2007
Aug 2007
Oct 2007
*
C. ariakensis (1)
C. ariakensis (2)
C. virginica (1)
C. virginica (2)
mixed (1)
mixed (2)
shell (1)
shell (2)

Figure 3-5. Mean values for dominance per sample for all treatments within each block (1 & 2) over time.

Error bars represent the standard effort of each mean and (*) over bars indicate significant within-date treatment effects.

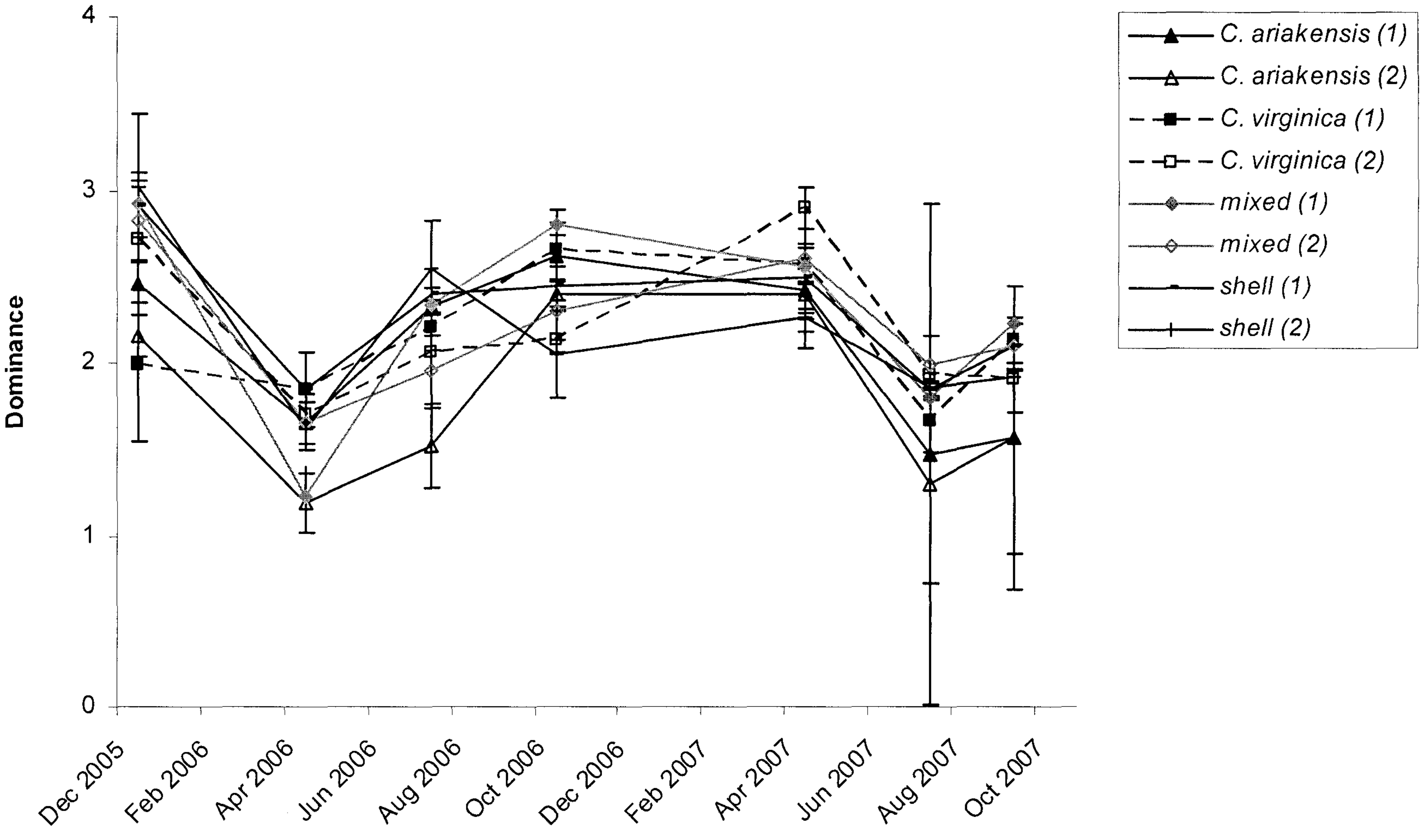

Dominance
4
3
2
1
0
Dec 2005
Feb 2006
Apr 2006
Jun 2006
Aug 2006
Oct 2006
Dec 2006
Feb 2007
Apr 2007
Jun 2007
Aug 2007
Oct 2007
C. ariakensis (1)
C. ariakensis (2)
C. virginica (1)
C. virginica (2)
mixed (1)
mixed (2)
shell (1)
shell (2)

Figure 3-6. Mean values for Shannon-Weiner diversity per sample for all treatments within each block (1 & 2) over time.

Error bars represent the standard effort of each mean and (*) over bars indicate significant within-date treatment effects.

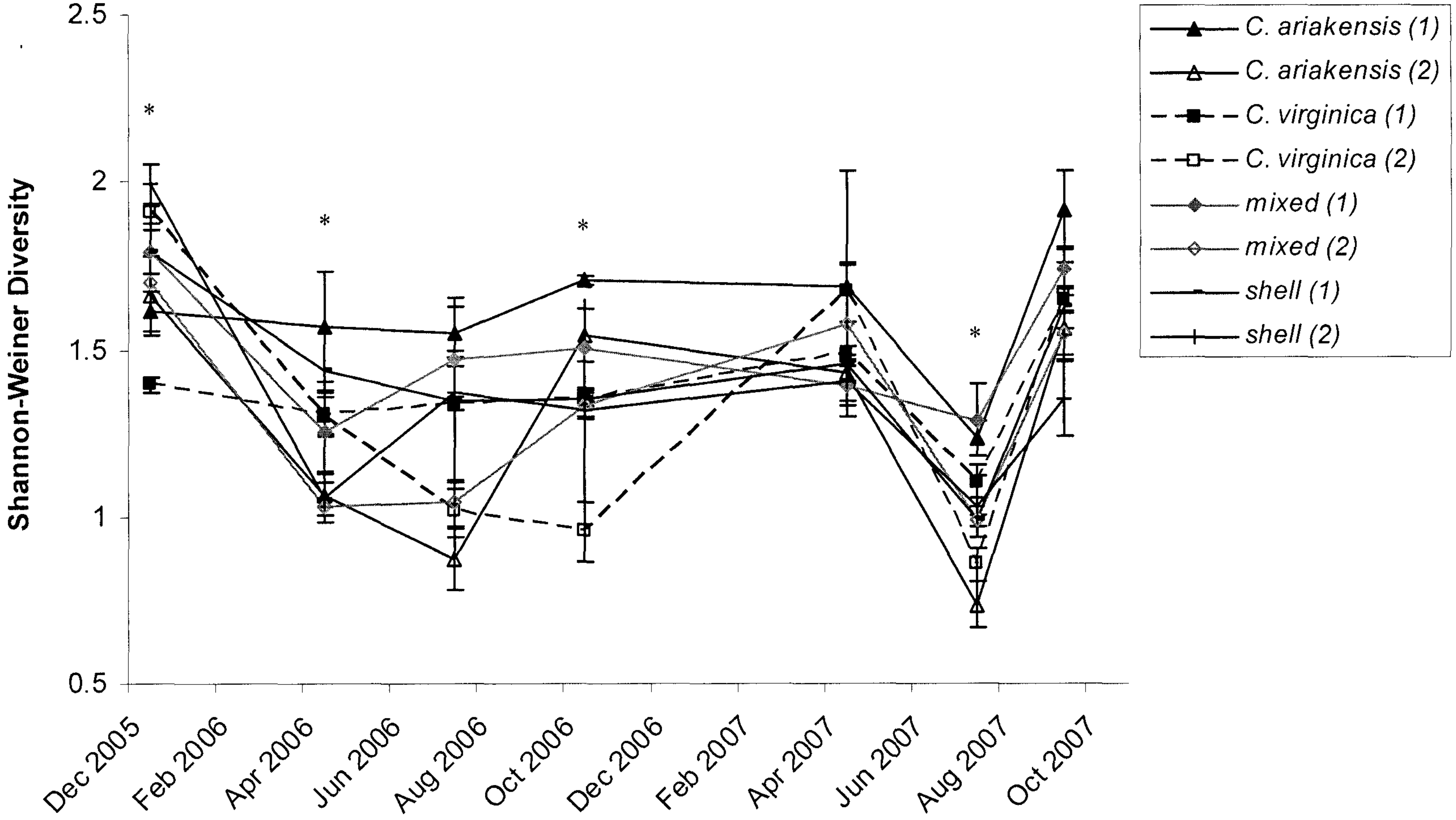

Shannon-Weiner Diversity
2.5
2
1.5
1
0.5
Dec 2005
Feb 2006
Apr 2006
Jun 2006
Aug 2006
Oct 2006
Dec 2006
Feb 2007
Apr 2007
Jun 2007
Aug 2007
Oct 2007
*
*
*
*
C. ariakensis (1)
C. ariakensis (2)
C. virginica (1)
C. virginica (2)
mixed (1)
mixed (2)
shell (1)
shell (2)

Figure 3-7. Mean total abundance of associated fauna per sample standardized by oyster biomass for all live oyster treatments within each block (1 & 2) over time.

Error bars represent the standard effort of each mean and (*) over bars indicate significant within-date treatment effects.

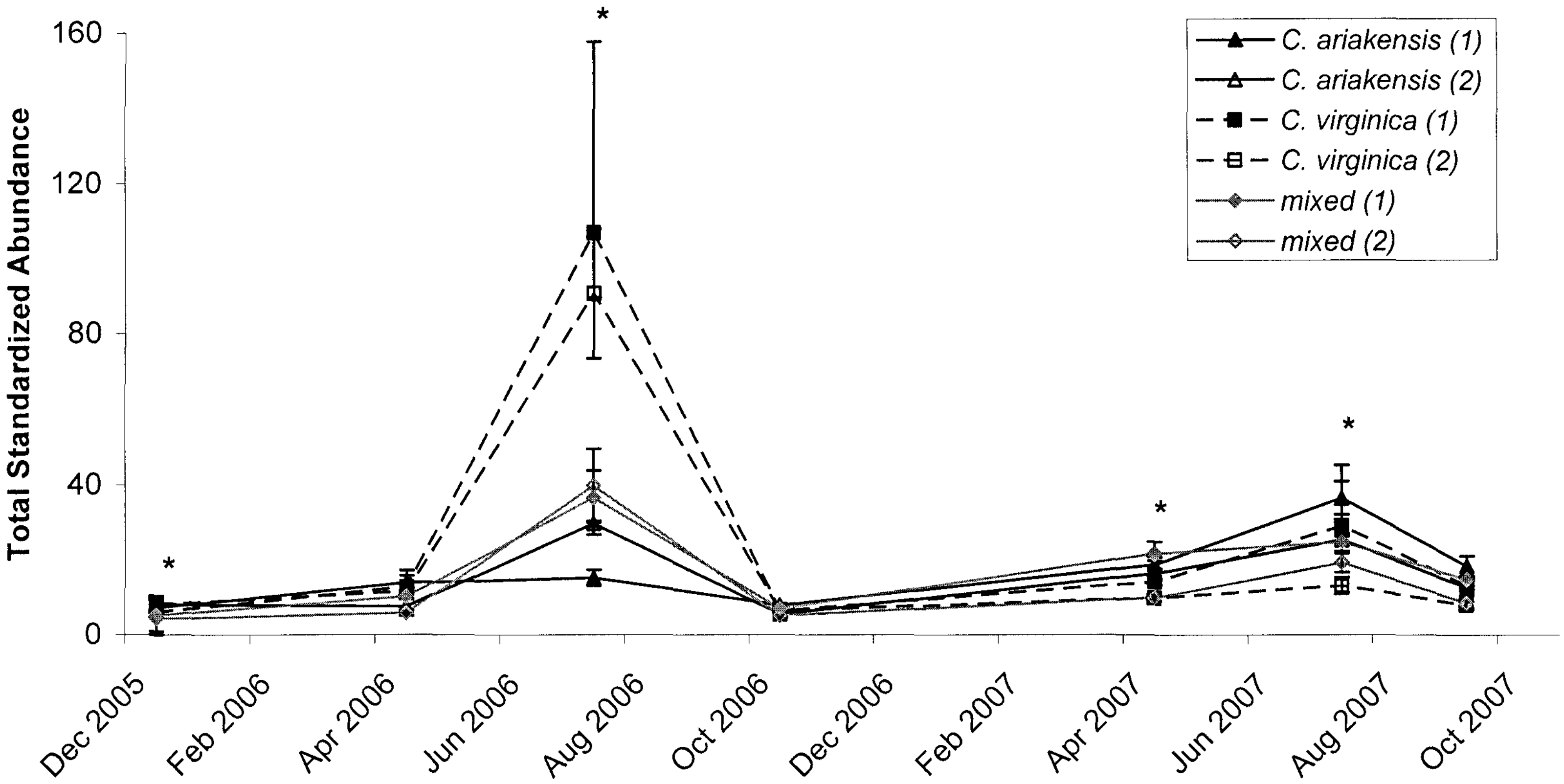

Total Standardized Abundance
160
120
80
40
0
Dec 2005
Feb 2006
Apr 2006
Jun 2006
Aug 2006
Oct 2006
Dec 2006
Feb 2007
Apr 2007
Jun 2007
Aug 2007
Oct 2007
*
*
*
*
C. ariakensis (1)
C. ariakensis (2)
C. virginica (1)
C. virginica (2)
mixed (1)
mixed (2)

Figure 3-8. Mean total biomass of associated fauna per sample standardized by oyster biomass for all live oyster treatments within each block (1 & 2) over time.

Error bars represent the standard effort of each mean and (*) over bars indicate significant within-date treatment effects.

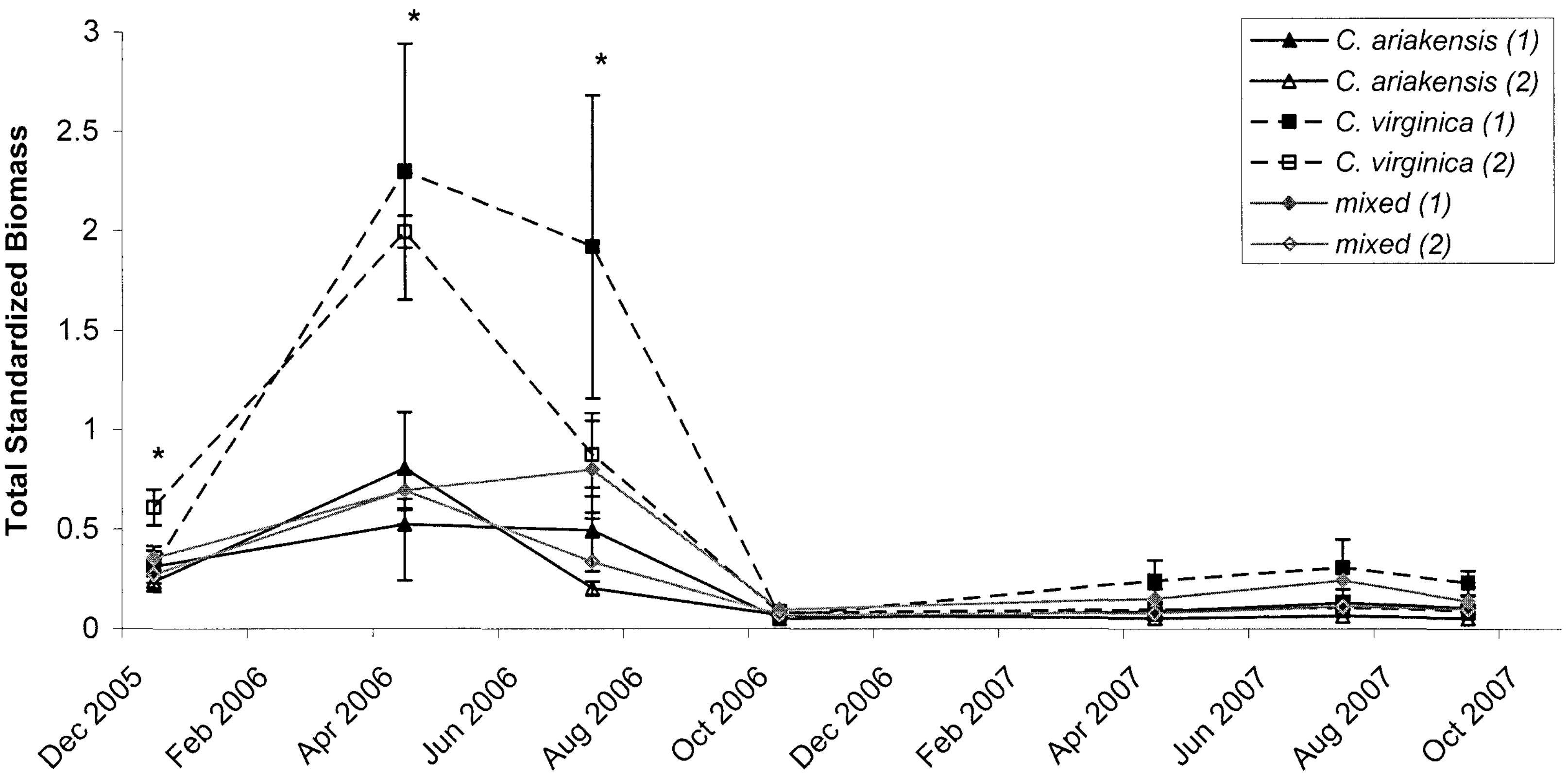
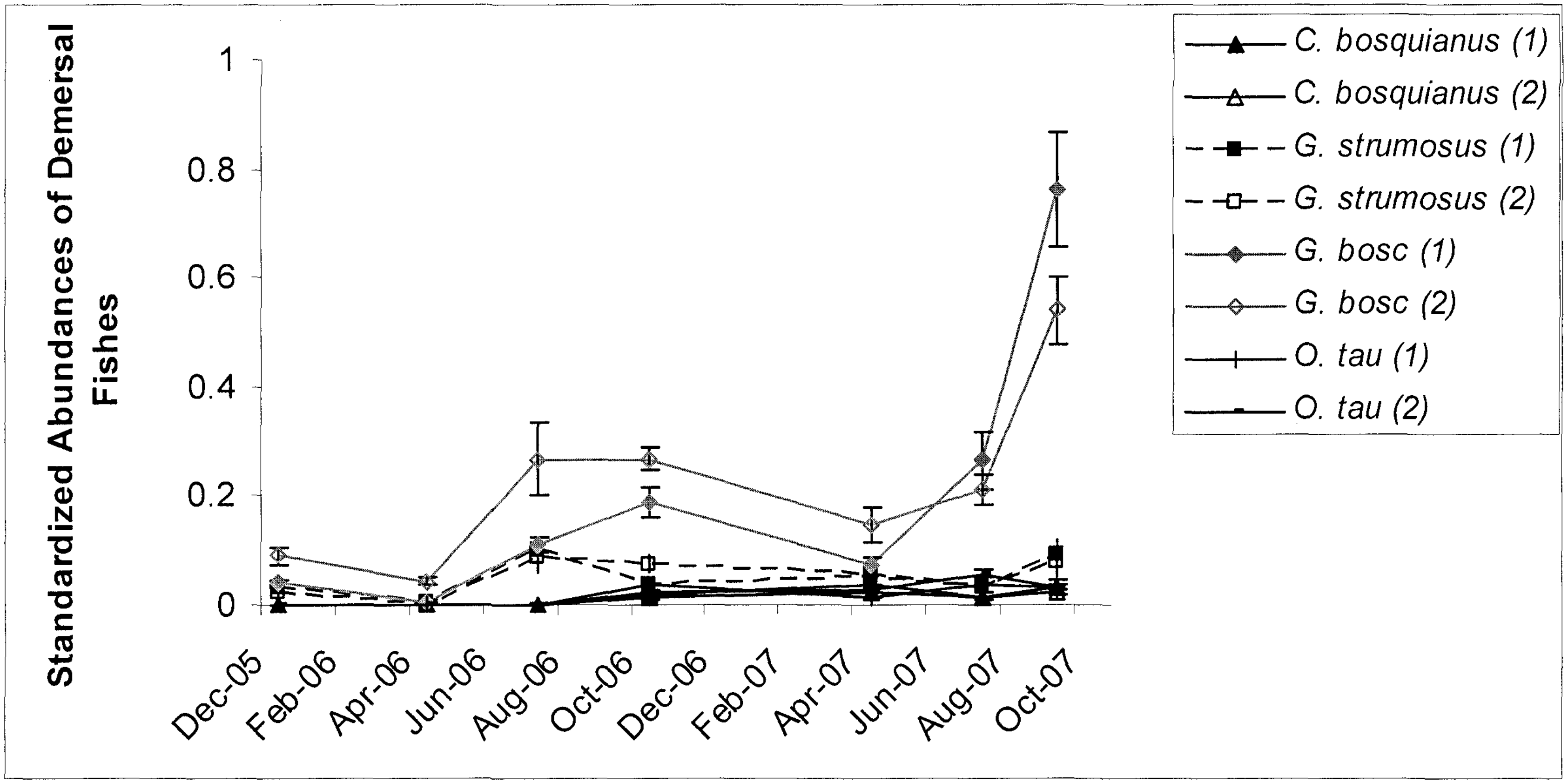

Total Standardized Biomass
3
2.5
2
1.5
1
0.5
0
*
*
*
Dec 2005
Feb 2006
Apr 2006
Jun 2006
Aug 2006
Oct 2006
Dec 2006
Feb 2007
Apr 2007
Jun 2007
Aug 2007
Oct 2007
C. ariakensis (1)
C. ariakensis (2)
C. virginica (1)
C. virginica (2)
mixed (1)
mixed (2)

Figure 3-9. Mean total abundance of demersal fishes (*C. bosquianus*, *G. strumosus*, *G. bosc*, and *O. tau*) per sample standardized by oyster biomass for all live oyster treatments within each block (1 & 2) over time.

Error bars represent the standard effort of each mean and (*) over bars indicate significant within-date treatment effects.



Standardized Abundances of Demersal Fishes
1
0.8
0.6
0.4
0.2
0
Dec-05
Feb-06
Apr-06
Jun-06
Aug-06
Oct-06
Dec-06
Feb-07
Apr-07
Jun-07
Aug-07
Oct-07
C. bosquianus (1)
C. bosquianus (2)
G. strumosus (1)
G. strumosus (2)
G. bosc (1)
G. bosc (2)
O. tau (1)
O. tau (2)

Figure 3-10. Non-metric MDS plots for standardized abundances of associated fauna over time: (A) April 2006, (B) July 2006, (C) October 2006, (D) April 2007, (E) July 207 (F) September 2007.

Circles around treatment clusters indicate significant ANOSIM results, and significant global r values are listed on the corresponding graphs.

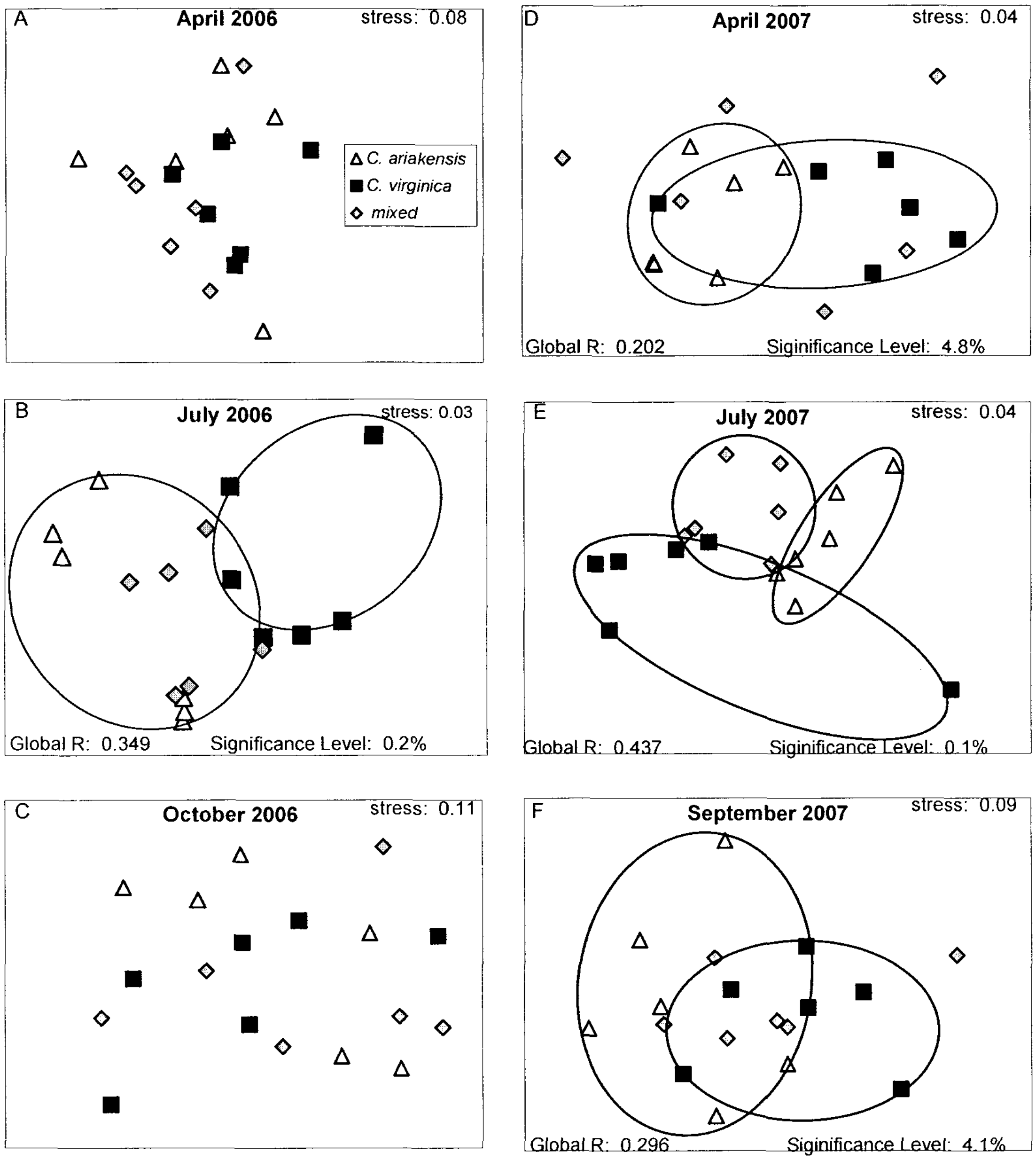

A
April 2006
stress: 0.08
C. ariakensis
C. virginica
mixed
D
April 2007
stress: 0.04
Global R: 0.202
Siginificance Level: 4.8%
B
July 2006
stress: 0.03
Global R: 0.349
Significance Level: 0.2%
E
July 2007
stress: 0.04
Global R: 0.437
Siginificance Level: 0.1%
C
October 2006
stress: 0.11
F
September 2007
stress: 0.09
Global R: 0.296
Siginificance Level: 4.1%

Figure 3-11. Non-metric MDS plots for standardized biomass of associated fauna over time: (A) April 2006, (B) July 2006, (C) October 2006, (D) April 2007, (E) July 207 (F) September 2007.

Circles around treatment clusters indicate significant ANOSIM results, and significant global r values are listed on the corresponding graphs.

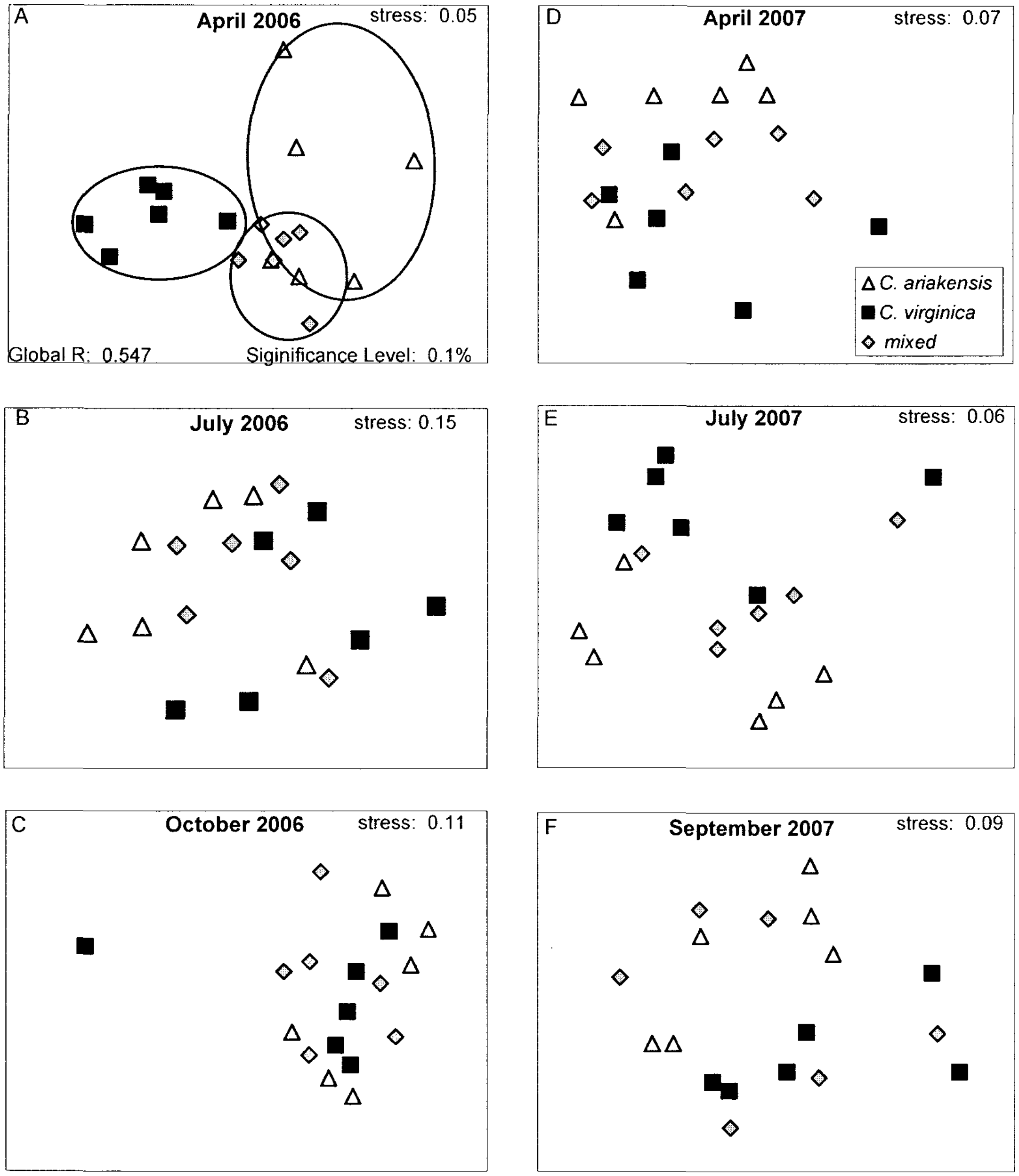
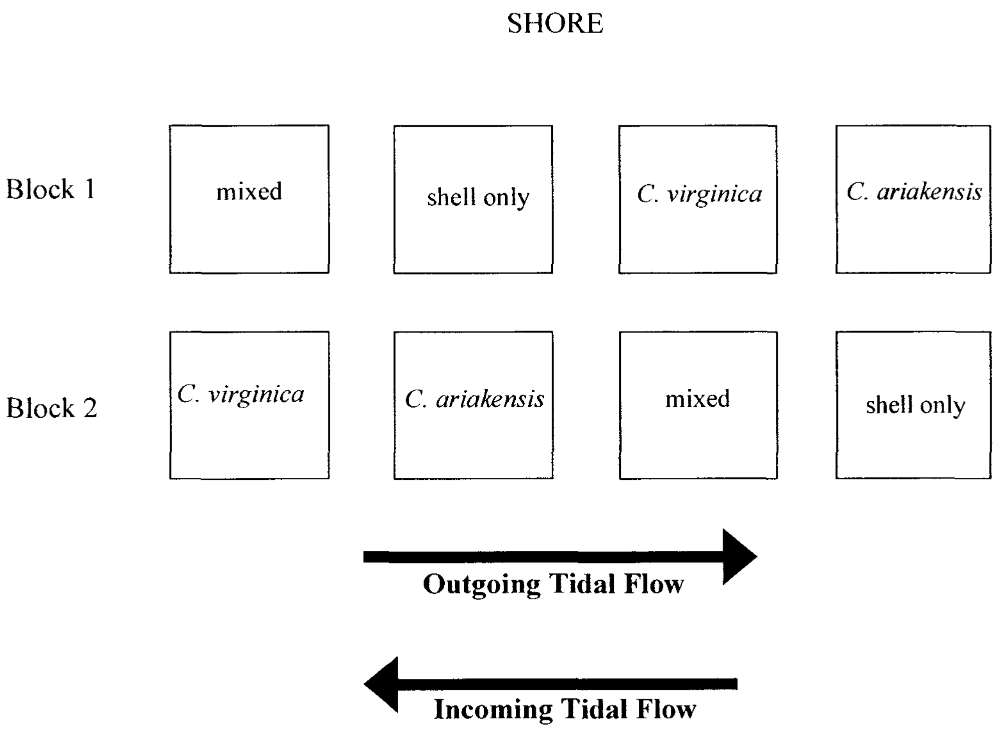

A
April 2006
stress: 0.05
Global R: 0.547
Siginificance Level: 0.1%
D
April 2007
stress: 0.07
△ C. ariakensis
■ C. virginica
◇ mixed
B
July 2006
stress: 0.15
E
July 2007
stress: 0.06
C
October 2006
stress: 0.11
F
September 2007
stress: 0.09

Figure 3-12. Placement of replicate cages containing experimental treatments at the study site with respect to block and flow.


SHORE
Block 1
mixed
shell only
C. virginica
C. ariakensis
Block 2
C. virginica
C. ariakensis
mixed
shell only
Outgoing Tidal Flow
Incoming Tidal Flow

CHAPTER 4: EFFECTS OF OYSTER SPECIES (*CRASSOSTREA VIRGINICA* VS *C. ARIAKENSIS*) AND HABITAT COMPLEXITY VARIATIONS ON AN OYSTER REEF TROPHIC CASCADE

Heather D. Harwell and Mark W. Luckenbach

Eastern Shore Laboratory, Virginia Institute of Marine Science,
College of William and Mary, P.O. Box 350, Wachapreague, VA 23480

[*] Corresponding author: Tel.: + 1 757 787 0959; fax + 1 757 787 5831
E-mail address: harwell@vims.edu

ABSTRACT

Investigations of trophic interactions between common oyster reef inhabitants have shown that the oyster toadfish, *Opsanus tau*, indirectly benefits native oysters, *Crassostrea virginica*, by affecting the foraging behavior of an intermediate predator, the mud crab *Panopeus herbstii.* Increased habitat complexity has been shown to attenuate the strength of the cascade on *C. virginica* oyster reefs (Grabowski 2004). Deliberations over a potential introduction of a non-native oyster species (*Crassostrea ariakensis*) to Chesapeake Bay, USA, have raised questions regarding the ecological functional equivalency of the native and non-native species. Given the possible differences in growth form between the two species and the uncertainty about the reef-forming capability of *C. ariakensis,* we investigated the effects of oyster species and subtle variance in habitat complexity on this trophic cascade. We conducted a mesocosm study using the aforementioned tri-trophic system of oyster reef inhabitants and four reef types: low complexity, *C. virginica*; high complexity, *C. virginica*; low complexity, *C. ariakensis*; and high complexity, *C. ariakensis.* Although toadfish presence significantly increased mud crab mortality in both native and non-native oyster reef treatments, habitat complexity differences associated with oyster size or species did not alter on trophic interactions within this system. The proportion of oysters released from predation as a function of toadfish-induced modification in mud crab behavior (trait-mediated indirect interaction, TMII) was far greater than the proportion of oysters released via direct predation of toadfish on mud crabs (density-mediated indirect interaction, DMII), regardless of oyster species or habitat complexity level, contributing to a growing number of studies emphasizing the relative importance of the influence of TMIIs on community structure.

KEY WORDS: *Crassostrea ariakensis*; habitat complexity; trophic cascade

INTRODUCTION

Indirect effects of predators on ecological community organization have long been recognized (e.g., Hairston et al. 1960, Paine 1966), and have more recently been posited to be as, if not more, important than direct effects in structuring species assemblages (Paine 1966, Wootton 1993, Menge 1995, Grabowski 2004). Trophic cascades, in which a top predator benefits basal prey through effects on an intermediate predator, have been shown to indirectly sustain several important basal species, such as hardwood trees (Ripple & Beschta 2007), kelps (Estes & Palmisano 1974), salt-marsh plants (Silliman & Bertness 2002), scallops (Myers et al. 2007), and oysters (Kimbro et al. 2009). Such cascading indirect effects are initiated through consumption of a mesopredator (density-mediated) or alteration in its foraging behavior (trait-mediated) (Carpenter et al. 1985, Kerfoot & Sih 1987, Wootton 1993, Abrams 1995, Menge 1995), and are particularly strong in marine benthic communities (Shurin et al. 2002).

Habitat complexity also has been shown to influence predation and community structure in marine environments (Crowder & Cooper 1982, Grabowski & Powers 2004). Macroinvertebrate densities and species richness are generally positively correlated with structurally complex habitats (Crowder & Cooper 1982, Diehl 1992) allowing for coexistence of competitors while providing refuge for prey species (Hixon & Menge 1991). Structurally complex areas provide habitat patches where predators are incapable of accessing prey resources (Hughes & Grabowski 2006), and predator foraging efficiency generally varies inversely with habitat heterogeneity (Diehl 1992, Beukers & Jones 1997), with individual predators consuming fewer prey in more structurally complex habitats (Grabowski 2004, Warfe & Barmuta 2004). In fact, greater abundances and species diversities found in structured as opposed to unstructured bottom habitats are often attributed to reduced predation within such habitats (Summerson & Peterson 1984, Lenihan et al. 2001, Grabowski et al. 2005). Even if habitat complexity does not completely remove the risk of predation, structure can decrease the foraging efficiency of predators by interfering with the ability of the predator to locate and handle its prey (Crowder & Cooper 1982, Summerson & Peterson 1984). In addition to influencing predator-prey interactions, increased habitat complexity often decreases encounter rates among predators, thereby reducing the strength of interference interactions and intraguild predation (Swisher et al. 1998, Grabowski & Powers 2004, Hughes & Grabowski 2006, Grabowski et al. 2008).

Previous studies of trophic interactions between common oyster reef inhabitants have shown that the oyster toadfish, *Opsanus tau*, indirectly benefits native oysters, *Crassostrea virginica*, by inducing an intermediate predator, the mud crab *Panopeus herbstii*, to occupy a deeper portion of the reef matrix and move less frequently (Grabowski 2004, Grabowski & Kimbro 2005). These investigations revealed that habitat complexity weakened the strength of component interactions within the trophic cascade, whereas top predator-induced modifications in intermediate predator behavior reinforced the cascade. Increased habitat complexity inhibited toadfish from feeding on mud crabs, yet toadfish-induced effects on mud crab behavior maintained the indirect effect of toadfish on juvenile oysters. Thus, trait-mediated indirect effects had a greater influence on the cascade than density-mediated effects.

Though our understanding of trophic cascades has been refined in recent years, the ongoing movement of species beyond their natural ranges has increased interactions between native and non-native species, the consequences of which remain largely unknown (Ruiz et al. 2000, Sax & Gaines 2003), including effects on trophic cascades. Since historical exposure may influence predator and prey behaviors (Vermeij 2001, Strauss et al. 2008), introduced species may interfere with or fail to recreate historically important native trophic cascades (Kimbro et al. 2009). The premeditated movement of aquatic species for aquaculture and fishery enhancement purposes has occurred for over 2000 years (Mann et al. 1991), with oysters being perhaps the most pervasive example (Reusink et al. 2005). In response to a decline in *C. virginica* abundance over the past 50 years, an intentional introduction of a non-indigenous oyster species, *Crassostrea ariakensis*, to Chesapeake Bay, USA, has been under consideration for most of the past decade (Rickards & Ticco 2002) with the intent of enhancing both fishery and ecosystem services. Though this proposal has recently been rejected (USACOE 2009), such an action would not be unprecedented and similar future proposals are anticipated. Short-term laboratory trials (Kingsley-Smith et al. 2008) and anecdotal evidence from its native range (Luckenbach pers. obs.) have raised questions concerning *C. ariakensis'* ability to form the dense, structurally complex reefs comparable to those of *C. virginica.*

Within the context of examining the potential ecological consequences of introducing this non-indigenous oyster to Chesapeake Bay and the U.S. Atlantic coast, we investigated how the biogenic habitats formed by this species might affect predator-prey interactions in the toadfish-mud crab-oyster trophic cascade. In doing so, we examined subtler effects of habitat complexity, resulting from differences in oyster species and oyster sizes, on these trophic interactions than previous studies.

MATERIALS AND METHODS

Experimental Design

Experiments were conducted in mesocosms at the Virginia Institute of Marine Science's Eastern Shore Laboratory (VIMS ESL) in Wachapreague, Virginia, USA, between August and November of 2007 and 2008 to determine the effects of oyster species and habitat complexity on specific predator-prey interactions. Surplus oyster clusters removed during April 2007 from a field study comparing growth, survival, and reef form and function of triploid (sterile) *C. ariakensis* and *C. virginica* in Chesapeake Bay (Kingsley-Smith et al. 2009, Harwell et al. 2010). These clusters were air dried for a period of three months to ensure that no living oyster tissue remained, reducing biosecurity risk and quarantine conditions needed to carry out the experiment. To remove any pre-existing bias between treatments, all remnants of epiphytes such as sponges, bryozoans, mussels, and barnacles were removed, leaving only the physical structure provided by the oyster clusters. All opened and disarticulated shells were resealed with a non-toxic marine epoxy to produce mimic reef habitats. Using measures of vertical relief and the number of individual oysters present, oyster clusters of each species were designated as complex or simple, numbered, and pooled for randomized use within treatments (Table 1). For *C. virginica* mean number of oysters in complex clusters was 3.8 X that in the simple clusters and the mean maximum vertical height in complex clusters was 2.7 X that in the simple clusters. Comparable values for *C. ariakensis* were 3.9 X for numbers and 2.4 X for maximum height (see Table 1).

To test the effects of reef-forming species, habitat complexity and fish presence on crab survival and oyster survival, we used an experimental design similar to that of Grabowski (2004), with the additional factor of oyster species. A 2 X 2 X 2 X 2 factorial design, consisting of two levels of complexity (simple or complex), two levels of reef-forming species (*C. virginica* or *C. ariakensis*), two levels of toadfish (present or absent), and two levels of mud crabs (present or absent), was employed (Table 2).

Sixteen cylindrical fiberglass tanks (height = 60 cm, diameter = 57.4 cm) were supplied with sand-filtered seawater at ambient temperature and salinity in a flow-through system (0.29 – 0.32 m/s). The bottom of each tank was covered with a thin layer of clean sand, followed by a single layer of disarticulated *C. virginica* shell. Oyster clusters of a designated species and pre-determined complexity level (simple or complex) were placed in each tank to form integrated reef-like structures covering similar bottom surface areas across all treatments. Clusters were randomly assigned (within proper categories) until the designated bottom surface areas in all tanks were filled (total clusters used ranged from 16 to 26 for low complexity treatments and 5 to 11 for high complexity treatments). Once arranged within each mesocosm, two independent measures of surface rugosity were taken using an adaptation of the 'chain length' method (Rogers et al. 1983, Aronson & Precht 1995), in which a ratio of the length of a chain molded to a surface and the distance between the chain's start and endpoint is created.

Compared to native oysters, *C. ariakensis* shells are structurally weaker, allowing for greater susceptibility to predation (Bishop & Peterson 2006, Newell et al. 2007). To avoid confounding the effects of predation rates and habitat complexity on oyster survivorship, a single basal prey species, *C. virginica*, was used across all treatments. Thirty live juvenile *C. virginica* (10-15 mm SH) obtained from the VIMS Aquaculture Genetics and Breeding Technology Center were glued within the interstices of the cluster in each tank using non-toxic marine epoxy. For the eight treatments with mud crabs present, 10 adult *Panopeus* *herbstii* of similar carapace width (25-30 mm) were released within each tank (~38 crabs m^{-2}), which is consistent with mud crab densities seen in a field study with both reef-forming species (Harwell et al. 2010, *unpublished data*). One adult toadfish (mean standard length = 215 mm, SD = 21.6 mm) was then added to eight tanks, creating treatments containing (1) only oysters, (2) oysters and mud crabs, (3) oysters and toadfish, and (4) oysters, mud crabs, and toadfish. Mud crabs used throughout the experiment were collected from a nearby natural oyster reef, and oyster toadfish were caught on-site through hook-and-line sampling and held in separate flow-through tanks under ambient conditions for up to one week prior to the start of an experimental trial. To avoid starvation-induced effects, hard clam tissue (*Mercenaria mercenaria*) was placed daily in each holding tank prior to each trial start date.

One replicate of each of the 16 treatments (the 4 species combinations listed above x 2 reef-forming species x 2 habitat complexity levels) was established per trial and 6 replicate trials lasting 5 days each were conducted (2 in 2007 and 4 in 2008). Water temperature, salinity, flow speed, and ammonium concentration were monitored throughout the duration of the experiment. Once during each day of the trial, the number of mud crabs visible on the reef surface was recorded for each treatment to determine whether the presence of toadfish induced avoidance behavior in mud crabs. At the end of the 5th day, living, dead, and missing mud crabs and oysters were quantified. Following each run, all surviving mud crabs and toadfish were released, and newly captured organisms were used for each subsequent trial. Due to limited availability of oyster clusters from the field experiment, replacement of all oyster clusters with new ones for each run was not possible. Instead, individual oyster clusters from the existing pools of simple and complex *C. virginica* and *C. ariakensis* were randomly assigned to new treatments after each trial.

Partitioning of direct and indirect effects

To further evaluate effects of an apex predator on basal prey, we used mud crab consumption rates of oysters to partition the indirect effects of toadfish presence (direct removal of crabs versus predator-avoidance behavior) on mud crab consumption of oysters within simple and complex reefs of each oyster species (Table 3). Crab consumption rates of oysters were calculated by determining the average number of oysters eaten by crabs per day during each experimental trial, described by Grabowski (2004). Toadfish effects were then partitioned between direct removal of crabs by toadfish (DMIIs) and predator-avoidance behavior of crabs (TMIIs). The effect of direct removal (DMII) was estimated by determining the daily rate of oysters consumed by crabs in the absence of toadfish and multiplying it by the average number of crabs consumed by toadfish during an experimental trial. This yielded the expected number of oysters per day that should have been released from mud crab predation as a consequence of toadfish consumption of mud crabs. Using expected versus actual oyster release, we quantified the proportion of oysters released from mud crab predation as a consequence of toadfish removal of mud crabs (toadfish effect 1, DMII = expected/actual oyster release) and the proportion of oysters released as a function of toadfish induced modification in mud crab behavior (toadfish effect 2, TMII = [actual-expected]/actual oyster release).

Statistical analyses

To test the assumptions of normality and homogeneity of variance, Sharipo-Wilke's test and the F-max test were performed on all main effects in each analysis. Oyster mortality data required arcsin transformation to remove heteroscedasticity, although geometric means are reported here. To test for differences in surface rugosity across treatments, we performed a five-way blocked ANOVA with reef-forming species, complexity level, crab presence, toadfish presence, and experimental run (block) as fixed factors. We conducted a four-way blocked ANOVA on percentage crab mortality, with oyster species, complexity, toadfish presence, and experimental run (block) as fixed factors. To test for the variation of oyster mortality with reef-forming species, complexity level, toadfish presence, and mud crab presence, a five-way ANOVA was performed, blocked by experimental run. For all tests, block effects were not significant ($p > 0.05$), so we re-ran reduced models without the block factor. Student-Newman-Keuls (SNK) tests were conducted when ANOVA indicated a significant effect. We conducted a three-way repeated measure ANOVA on percentage crab visibility for reef-forming species, habitat complexity, and toadfish presence, within each replicate mesocosm. To determine whether variance in toadfish standard length had an influence on main effects, regressions of percentage mud crab mortality and juvenile oyster mortality versus toadfish length were also performed.

RESULTS

Surface rugosity index did not vary significantly between oyster species, although significant differences were present between complexity categories (Fig. 1). Complex reef treatments had rugosity indices that were approximately 29.8% (*C. virginica*) and 26.3% (*C. ariakensis*) greater than their simple reef counterparts, ($F = 267.52$, $p < 0.0001$). No other differences in surface rugosity among treatments were detected, and there were no significant interactions between fixed factors.

Mud crab mortality in the absence of toadfish ranged from 0% to 10% and was similar across treatments containing only oysters or oysters and toadfish. Toadfish presence significantly increased mud crab mortality in both native and non-native oyster reef treatments (F = 20.59, $p < 0.0001$) (Fig. 2). On simple native oyster reefs, the presence of a toadfish increased mud crab mortality from 5% to 15%. Similarly, mud crab mortality rates on simple non-native oyster reefs increased from 1.7% to 13.3% in the presence of toadfish. Mud crab mortality in the presence of toadfish was not reduced on the complex reefs relative to the simple reefs (Fig. 2). We observed increased mud crab mortality rates of 16% (*C. virginica* reefs) and 8.4% (*C. ariakensis* reefs) in the presence of toadfish on our complex reefs, similar to the effect seen on simple oyster reef treatments. In addition, ANOVA failed to detect effects of reef-forming species on percent mud crab mortality, and interaction terms were not significant.

In treatments without mud crabs, oyster mortality was similar and ranged from 0% to 13.3%. Oyster mortality varied with mud crab presence and toadfish presence, but was not affected by oyster reef-forming species or complexity level (Fig. 3). When mud crabs were present but under no predation pressure from oyster toadfish, average oyster mortality was significantly higher than in treatments without mud crabs (F = 536.86, $p < 0.0001$), yet similar across oyster reef-forming species and complexity treatments. When toadfish were not present, mud crabs consumed 99.4% and 95.6% of the juvenile oysters present on the low complexity *C. virginica* and *C. ariakensis* reefs, respectively, and 93.9% (*C. virginica*) and 93.3% (*C. ariakensis*) on the high complexity reefs. The presence of toadfish significantly decreased oyster mortality (F = 36.97, $p < 0.0001$) regardless of oyster reef-forming species or habitat complexity level. Mud crab consumption of juvenile oysters on native oyster reefs decreased in the presence of toadfish by 44.4% on simple reefs and 46.7% on complex reefs, with non-native reefs having comparable decreases of 38.9% and 32.2% on simple and complex reefs, respectively. Increased habitat complexity did not reduce mud crab predation on oysters in either *C. virginica* or *C. ariakensis* treatments.

The proportion of mud crabs visibly foraging on experimental reefs varied with experimental day, with significantly more crabs visible on day 1 (25%) than on subsequent days (11.7%-15.4%) during the 5-day trials (F=6.67, p<0.0001) (Fig. 4A). Neither oyster reef-forming species nor habitat complexity level affected the percentage of visible mud crabs within treatments (Fig. 4B), with an average of 17% of all mud crabs visible on simple native oyster reefs, 15% visible on complex *C. virginica* and simple *C. ariakensis* treatments, and 14.3% seen on complex non-native reefs. Toadfish presence, however, significantly decreased mud crab visibility (F=92.26, p<0.0001) by 8.6% in *C. virginica* treatments and 15.3% in *C. ariakensis* treatments (Fig. 4C). Regressions of toadfish length versus percentage mud crab mortality and juvenile oyster mortality were not significant in any case (Fig. 5).

Partitioning the indirect effects (DMIIs versus TMIIs) of toadfish presence on mud crab consumption of oysters yielded similar results for both *C. virginica* and *C.ariakensis* treatments. In all cases, the trait-mediated effect of reduced mud crab foraging was responsible for at least 90% of the reduction in oyster mortality provided by toadfish presence. Direct predation of mud crabs by toadfish only explained 6.7% of oyster release on simple native oyster reefs, and only 1.1% on simple *C. ariakensis* reefs. Increased complexity slightly increased the importance of DMIIs to 9.9% for *C. virginica* and 8.5% for *C. ariakensis* treatments.

DISCUSSION

Toadfish presence provided an indirect benefit to juvenile oysters in this study by decreasing the predation activity of mud crabs, indicating the successful replication of a trophic cascade which has been the focus of previous research efforts (Grabowski 2004, Grabowski et al. 2008). Similar cascading effects of predator-prey interactions were observed on both *C. virginica* and *C. ariakensis* reef treatments, suggesting functional equivalency in this regard between the two oyster species with respect to habitat, at least for the simplified reefs in this study. Habitat functional equivalency is further supported by similar surface rugosity index measurements between *C. virginica* and *C. ariakensis* experimental treatments.

Grabowski (2004) found that habitat complexity weakened the strength of this trophic cascade by disrupting predator-prey interactions on multiple levels. Not only did increased complexity significantly decrease mud crab mortality in the presence of toadfish, but it also decreased mud crab predation of juvenile *C. virginica*, whether or not toadfish were present. Contrasting results were seen in the present study, in which habitat complexity did not affect any of the predator-prey interactions of the trophic cascade: similar levels of predation were seen on simple and complex reefs for both *O. tau* – *P. herbstii* and *P. herbstii* – *C. virginica* interactions, regardless of oyster reef-forming species.

Structure in complex habitats can interfere with a predator's ability to see or otherwise sense prey (Savino & Stein 1982, Main 1987, Bartholomew 2002), and it can interfere with a predator's ability to maneuver through the spaces in the habitat in search of, or in pursuit of, prey that are usually smaller than the predator (Ryer 1988, Bartholomew et al. 2000, Ryer et al .2004). Accordingly, predator size has been shown to influence the effect of habitat complexity on predator-prey interactions in some environments (Attrill et al. 2000, Bartholomew & Shine 2008). To assess whether variance in toadfish size affected predator-prey interactions observed in our experiment, regressions of toadfish standard length and percent oyster and crab mortality were performed and were not be significant in any case (Fig. 5). Although toadfish size did not influence effects of habitat complexity on predator-prey dynamics over the range of standard lengths incorporated in our study, the average SL (215 ± 21.6 mm) was almost twice the size of toadfish used in Grabowski's study (147 ± 14.3 mm). Therefore, the relative increase in predator body size may have made foraging and maneuvering within the interstices of the reefs more difficult, contributing to contrasting results.

We primarily attribute these different findings to varying definitions of 'simple' and 'complex' reef treatments between the two studies. Grabowski (2004) used un-aggregated oyster shell with < 5 cm of vertical relief for his simple reefs, mimicking the highly degraded natural reefs he has observed in coastal North Carolina, which contain few living oysters and little to no vertical relief. Based on results from our comparative field study of *C. virginica* and *C. ariakensis* survival and growth in Maryland and Virginia waters (Kingsley-Smith et al. 2009, Harwell et al. 2010), we chose to construct our simple reef mimics using aggregated oyster clusters with some live oysters and moderate vertical height, reflecting slow growing, immature or partly degraded reefs. Our complex reef category designation also differed from Grabowski's experiments in which complex treatments contained aggregated oyster clusters with vertical heights between 10 and 30 cm, whereas the height ranges of our complex oyster reef treatments were more narrowly constrained between approximately 12 and 13 cm. Although Grabowski did not quantify any differences in surface topography between reef categories, we assume that this difference would have been demonstrable, and likely greater than the 29.8 % difference (*C. virginica*) observed here. Thus, the divergence between Grabowski's simple and complex treatments was markedly greater than that of present study, which may explain why we did not observe a weakening of the trophic cascade with increased habitat complexity.

As was the case in Grabowski's experiment, there was an initial peak in the percentage of crabs visibly foraging on reefs on day 1 of each experiment, suggesting that crabs were still acclimating to experimental conditions at that time (Fig. 3A). Grabowski (2004) also observed a greater proportion of visible mud crabs on complex reefs, suggesting that prey might be more mobile or hide less within complex habitats because they are either less susceptible to predation or less capable of recognizing predators. No such increase in visible mud crabs on complex reefs was observed in the present study, further suggesting that our reefs may represent intermediates between Grabowski's simple and complex treatment definitions. Toadfish presence significantly reduced the proportion of visible mud crabs regardless of oyster species or habitat complexity (Fig. 4C), supporting previous findings that toadfish induce a predator-avoidance behavior in mud crabs, or trait-mediated effect, which results in reduced predation of oysters (Grabowski 2004, Grabowski et al. 2008).

Several empirical studies have demonstrated that trait-mediated indirect interactions (TMIIs) not only occur in natural and experimental systems (reviewed in Werner & Peacor 2003), but that these effects can be greater than density-mediated indirect interactions (DMIIs) (Soluk &Collins 1988, Huang & Sih 1990, Schmitz 1998, Diehl et al. 2000, Peacor & Werner 2001). Here, the proportion of oysters released from predation as a function of toadfish-induced modification in mud crab behavior (TMII) was far greater than the proportion of oysters released via direct predation of toadfish on mud crabs (DMII), regardless of oyster species or habitat complexity level (Table 3). Thus, our results join a growing body of evidence that suggests that the mere presence of a top predator may be more important than the removal of intermediate predators as a determinant of community structure (Werner & Peacor 2003, Schmitz et al. 2004, Grabowski & Kimbro 2005).

Effects of habitat complexity have also been shown to be density-dependent through alterations in both predator and prey densities (Grabowski & Powers 2004). At relatively low densities (11 and 22 mud crabs m^{-2}), Grabowski & Powers (2004) found that habitat complexity did not affect foraging rates of mud crabs on juvenile hard clams, *M. mercenaria.* At higher predator densities (44 crabs m^{-2}), however, crabs consumed 51% more clams on complex reefs than on simple reefs. Mud crab densities in the present study were ~38 m^{-2}, which should have been sufficient to detect any influences that habitat complexity may have had on predation rates. The fact that we did not observe effects of habitat complexity on mud crab foraging rates on oysters suggests again that the complexity differences afforded by our treatments, though statistically significant (see Table 1 & Fig. 1) and ecologically realistic (see Kingsley-Smith et al. 2009), were not sufficient to substantially alter the species interactions in this tri-trophic system.

As biogenic oyster reefs have declined globally at an unprecedented rate, there is a pressing need to conserve existing reefs and develop effective restoration approaches (Beck et al. *in review*). Determining how specific reef characteristics affect ecological processes can help to inform both conservation practices and evaluation of restoration success (Coen & Luckenbach 2000, Luckenbach et al. 2005). Our study suggests that even small aggregated clusters of oysters associated with simple reefs can affect multi-level trophic interactions.

Though some introduced species may fail to recreate historically important native trophic cascades (Kimbro et al. 2009), this comparison of the effects of *C. virginica* and *C. ariakensis* as reef substrate on an oyster reef trophic cascade suggests functional equivalency with respect to habitat function, at least for the species-specific interactions evaluated herein. Moreover, our results suggest low complexity reefs, which may be characteristic of younger restored reefs or those whose age structure is truncated by disease impacts, may still afford some ecosystem functions similar to those of more complex reefs.

ACKNOWLEDGEMENTS

We thank I. Anderson, E. Canuel, and A. Hardison for graciously providing tanks and plumbing equipment and R. Bonniwell and S. Fate for system organization and maintenance. Oysters were provided by the VIMS Aquaculture Breeding and Genetics Technology Center, thanks especially to S. Allen, Jr. and K. Hudson. Brian Barnes, Alan Birch, Sean Fate, Peter Kingsley-Smith, and Edward Smith provided assistance with field work and data collection, and Stephanie Bonniwell was instrumental in rearing oysters prior to trial start dates. This research was funded by the Keith Campbell Foundation for the Environment.

LITERATURE CITED

Abrams, P. A. 1995. Implications of dynamically variable traits for identifying, classifying, and measuring direct and indirect effects in ecological communities. *Amer. Nat.* 146:112-134.

Aronson, R. B. & W. F. Precht. 1995. Landscape patterns of reef coral diversity: A test of the intermediate disturbance hypothesis. *J. Exp. Mar. Biol. Ecol.* 192:1-14.

Attrill, M. J., J. A. Strong & A. A. Rowden. 2000. Are macroinvertebrate communities influenced by structural complexity? *Ecography* 23:114-121.

Bartholomew, A. 2002. Faunal colonization of artificial seagrass plots: the importance of surface area versus space size relative to body size. *Estuaries* 25:1045-1052.

Bartholomew, A. & R. L. Shine. 2008. Space size relative to prey width (Sp/Py) influences macrofaunal colonization of artificial structures. *Mar. Ecol. Prog. Ser.* 358:95-102.

Beck, M.W., R. D. Brumbaugh, L. Airoldi, A. Carranza, L. D. Coen, C. Crawford, O. Defeo, G. J. Edgar , B. Hancock, M. Kay, H. Lenihan, M. W. Luckenbach, C. L. Toropova, G. Zhang. 2010. Shellfish Reefs at Risk Globally and Recommendations for Ecosystem Revitalization. *BioSci.* (*in review*).

Beukers, J. S. & G. P. Jones. 1997. Habitat complexity modifies the impact of piscivores on a coral reef fish population. *Oecologia* 114:50-59.

Bishop, M. J. & C. H. Peterson. 2006. When *r*-selection may not predict introduced-species proliferation: Predation of a non-native oyster. *Ecol. Appl.* 16:718-730.

Carpenter, S. R., J. F. Kitchell & J. R. Hodgson. 1985. Cascading trophic interactions and lake productivity. *BioSci.* 35:634-639.

Coen, L. D. and M. W. Luckenbach. 2000. Developing success criteria and goals for evaluating shellfish habitat restoration: ecological function or resource exploitation? Ecological Engineering. 15:323-343.

Crowder, L. B. & W. E. Cooper. 1982. Habitat structural complexity and the interactions between bluegills and their prey. *Ecology* 78:1802-1813.

Diehl, S. 1992. Fish predation and benthic community structure: the role of omnivory and habitat complexity. *Ecology* 73:1646-1661.

Diehl, S., S. D. Cooper, K. W. Kratz, R. M. Nisbet, S. K. Roll, S. W. Wiseman & T. M. Jenkins. 2000. Effects of multiple, predator-induced behaviors on short-term producer-grazer dynamics in open systems. *Amer. Nat.* 156:293-313.

Estes, J. A. & J. F. Palmisano. 1974. Sea otters-their role in structuring nearshore communities. *Science* 185:1058-1060.

Grabowski, J. H. 2004. Habitat complexity disrupts predator-prey interactions but not the trophic cascade on oyster reefs. *Ecology* 85:995-1004.

Grabowski, J. H., A. R. Hughes, D. L. Kimbro & M.A. Dolan. 2005. How habitat setting influences restored oyster reef communities. *Ecology* 86:1926-1935.

Grabowski, J. H. & D. L. Kimbro. 2005. Predator-avoidance behavior extends trophic cascades to refuge habitats. *Ecology* 86:1312-1319.

Grabowski, J. H. & S. P. Powers. 2004. Habitat complexity mitigates trophic transfer on oyster reefs. *Mar. Ecol. Prog. Ser.* 277:291-295.

Grabowski, J. H., A. R. Hughes & D. J. Kimbro. 2008. Habitat complexity influences cascading effects of multiple predators. *Ecology* 89:3413-3422.

Hairston, N. G., F. E. Smith & L. B. Slobodkin. 1960. Community structure, population control, and competition. *Amer. Nat.* 94:421-425.

Harwell, H.D, P. R. Kingsley-Smith, M. L. Kellogg, S. M. Allen, S. K. Allen, Jr., D. W. Meritt, K. T. Paynter, Jr. & M. W. Luckenbach. 2010. A comparison of *Crassostrea virginica* and *C. ariakensis* in Chesapeake Bay: Does oyster species affect habitat function? *J. Shellfish Res.,* accepted.

Hixon, M. A., & B. A. Menge. 1991. Species diversity: prey refuges modify the interactive effects of predation and competition. *Theo. Pop. Biol.* 39:178-200.

Huang, C. & A. Sih. 1991. Experimental studies on direct and indirect interactions in a three trophic-level stream system. *Oecologia* 85:530-536.

Hughes, A. R. & J. H. Grabowski. 2006. Habitat context influences predator interference interactions and the strength of resource partitioning. *Oecologia* 149:256-264.

Kerfoot, C. & A. Sih. 1987. Predation: direct and indirect impacts on aquatic communities. *University Press of New England*, Hanover, New Hampshire, USA.

Kimbro, D. L., E. D. Grosholz, A. J. Baukus, N. J. Nesbitt, N. M. Travis, S. Attoe & C. Coleman-Hulbert. 2009. Invasive species cause large-scale loss of native California oyster habitat by disrupting trophic cascades. *Oecologia* 160:563-575.

Kingsley-Smith, P. R. & M. W. Luckenbach. 2008. Post-settlement survival and growth of the Suminoe oyster, Crassostrea ariakensis, exposed to simulated emersion regimes. *J. Shellfish Res.* 27:609-618.

Kingsley-Smith P. R., H. D. Harwell, M. L. Kellogg, S. M. Allen, S. K. Allen, Jr., D. W. Meritt, K. T. Paynter, Jr. & M. W. Luckenbach. 2009. Survival and growth of triploid *Crassostrea virginica* (Gmelin, 1791) and *C. ariakensis* (Fujita, 1913) in bottom environments of Chesapeake Bay: implications for an introduction. *J. Shellfish Res.* 28:169-184.

Lenihan, H. S., C. H. Peterson, J. E. Byers, J. H. Grabowski, G. W. Thayer & D. R. Colby. 2001. Cascading of habitat degradation: oyster reefs invaded by refugee fishes escaping stress. *Ecol. Appl.* 11:764-782.

Luckenbach, M. W., L. D. Coen, P. G. Ross, Jr. and J. A. Stephen. 2005. Oyster reef Habitat Restoration: Relationship between oyster abundance and community development based on two studies in Virginia and South Carolina. J. Coast. Res. Special Issue No. 40:64-78

Main, K. L. 1987. Predator avoidance in seagrass meadows: prey behavior, microhabitat selection, and cryptic coloration. *Ecology* 68:170-180.

Mann, R., E. M. Burreson & P. Baker. 1991. The decline of the Virginia oyster fishery in Chesapeake Bay: considerations for introduction of a non-endemic species, *Crassostrea gigas* (Thunberg, 1973). *J. Shellfish Res.* 10:379-388.

Menge, B. A. 1995. Indirect effects in marine rocky intertidal interaction webs: patterns and importance. *Ecol. Mon.* 65:21-74.

Myers, R. A., J. K. Baum, T. D. Shepherd, S. P. Powers & C. H. Peterson. 2007. Cascading effects of the loss of apex predatory sharks from a coastal ocean. *Science* 315:1846-1850.

Newell, R. I. E., V. S. Kennedy, K. S. Shaw & S. Kristi. 2007. Comparative vulnerability to predators, and induced defense response, of eastern oysters *Crassostrea virginica* and non-native *Crassostrea ariakensis* oysters in Chesapeake Bay. *Mar. Biol.* 152:449-460.

Paine, R. T. 1966. Food web complexity and species diversity. *Amer. Nat.* 100:65-75.

Peacor, S. D. & E. E. Werner. 2001. The contribution of trait-mediated indirect effects to the net effects of a predator. *Proc. Nat. Acad. Sci.* 98:3904-3908.

Reusink, J. L., H. S. Lenihan, A. C. Trimble, K. W. Heiman, F. Micheli, J. E. Byers & M. C. Kay. 2005. Introduction of non-native oysters: ecosystem effects and restoration implications. *Annu. Rev. Ecol. Evol. Syst.* 36:643-689.

Rickards, W. L. & P. C. Ticco. 2002. The Suminoe oyster, *Crassostrea ariakensis,* in Chesapeake Bay: current status and near-term research activities. *Virginia Sea Grant Publication VSG-02-23.* Charlottesville, Virginia. 10p.

Ripple, W. J. & R. L. Beschta. 2007. Hardwood tree decline following large carnivore loss on the Great Plains, USA. *Front. Ecol. Environ.* 5:241-246.

Risk, M. J. 1972. Fish diversity on a coral reef in the Virgin Islands. *Atoll Res. Bull.* 193:1-6.

Rogers, C. S., M. Gilnack & H. C. Fitz. 1983. Monitoring of coral reefs with linear transects: A study of storm damage. *J. Exp. Mar. Biol. Ecol.* 66:285-300.

Ruiz, G. M., P. W. Fofonoff, J. T. Carlton, M. J. Wonham & A. H. Hines. 2000. Invasion of coastal marine communities in North America: apparent patterns, processes, and biases. *Annu. Rev. Ecol. Syst.* 31:481-531.

Ryer, C. H. 1988. Pipefish foraging: effects of fish size, prey size and altered habitat complexity. *Mar. Ecol. Prog. Ser.* 48:37-45.

Ryer, C. H., A. W. Stoner & R. H. Titgen. 2004. Behavioral mechanisms underlying the refuge value of benthic habitat structure for two flatfishes with differing anti-predator strategies. *Mar. Ecol. Prog. Ser.* 268:231-243.

Savino, J. F. & R. A. Stein. 1982. Predator-prey interaction between largemouth bass and bluegills as influenced by simulated, submerged vegetation. *Trans. Am. Fish. Soc.* 111:255-266.

Sax, D. F. & S. D. Gaines. 2003. Species diversity: from global decreases to local increases. *Trends Ecol. Evol.* 18:561-566.

Schmitz, O. J. 1998. Direct and indirect effects of predation and predation risk in old-field interaction webs. *Amer. Nat.* 151:327-342.

Schmitz, O. J., V. Krivan & O. Ovadia. 2004. Trophic cascades: the primacy of trait-mediated indirect interactions. *Ecol. Let.* 7:153-163.

Shurin, J. B., E. T. Borer, E. W. Seabloom, K. Anderson, C. A. Blanchette, B. Broitman, S. D. Cooper & B. S. Halpern. 2002. A cross-ecosystem comparison of the strength of trophic cascades. *Ecol. Let.* 5:785-791.

Silliman, B. R. & M. D. Bertness. 2002. A trophic cascade regulates salt marsh primary production. *Proc. Natl. Acad. Sci. USA* 99:10500-10505.

Soluk, D. A. & N. C. Collins. 1988. Synergistic interactions between fish and stoneflies: facilitation and interference among stream predators. *Oikos* 52:94-100.

Strauss, S. Y., J. A. Lau, T. W. Schoener & P. Tiffin. 2008. Evolution in ecological field experiments: implications for effect size. *Ecol. Lett.* 11:199-207.

Summerson, H. C. & C. H. Peterson. 1984. Role of predation in organizing benthic communites of a temperate-zone seagrass bed. *Mar. Ecol. Prog. Ser.* 15:63-77.

Swisher, B. J., D. A. Soluk & D. H. Wahl. 1998. Non-additive predation in littoral habitats: influences of habitat complexity. *Oikos* 81:30-37.

USACOE 2009. Final Programmatic Environmental Impact Statement for Oyster Restoration in Chesapeake Bay. U. S. Army Corps of Engineers, Norfolk, VA.

Vermeij, G. J. 2001. Community assembly in the sea: geologic history of the living shore biota. *In* Berness M. D., S. D. Gaines, M. E. Hay (Eds.), Marine community ecology. Sinauer Associates, Sunderland, pp 39-60.

Warfe, D. M. & L. A. Barmuta. 2004. Habitats structural complexity mediates the foraging success of multiple predator species. *Oecologia* 141:171-178.

Werner, E. E. & S. D. Peacor. 2003. A review of trait-mediated indirect interactions in ecological communities. *Ecology* 84:1083-1100.

Wootton, J. T. 1993. Indirect effects and habitat use in an intertidal community: interaction chains and interaction modifications. *Amer. Nat.* 141:71-89.

Table 1. Explanation of characteristics used to assign complexity categories for *C. ariakensis* and *C. virginica* surplus oyster clusters recovered from the Kingsley-Smith et al. (2009) field study for use in mesocosm trials.

Species	Complexity	n	Individual Oysters Present Per Cluster (Mean ± SD)	Maximum Vertical Height (cm) (Mean ± SD)
C. virginica	simple	203	1.9 ± 1.0	4.5 ± 2.3
C. ariakensis	simple	204	1.5 ± 0.8	5.3 ± 2.0
C. virginica	complex	74	7.4 ± 3.3	12.3 ± 2.1
C. ariakensis	complex	49	5.9 ± 2.2	13.2 ± 2.0

Table 2. 2X2X2X2 factorial experimental design used during each of six experimental trials: two levels of complexity (low or high) two levels of oyster substrate species (*C. virginica* or *C. ariakensis*), two levels of toadfish (present or absent), and two levels of mud crabs (present or absent).

Reef Substrate Species	Habitat Complexity Level	Basal Prey Presence (*C. virginica*)	Intermediate Predator Presence (*P. herbstii*)	Apex Predator Presence (*O. tau*)
C. virginica				
	simple	present	absent	absent
	simple	present	present	absent
	simple	present	absent	present
	simple	present	present	present
	complex	present	absent	absent
	complex	present	present	absent
	complex	present	absent	present
	complex	present	present	present
C. ariakensis				
	simple	present	absent	absent
	simple	present	present	absent
	simple	present	absent	present
	simple	present	present	present
	complex	present	absent	absent
	complex	present	present	absent
	complex	present	absent	present
	complex	present	present	present

Table 3. Effects of toadfish on the daily rates of oyster mortality induced by mud crabs (mud crab feeding rates) across treatments. Means are presented with standard errors in parentheses.

Toadfish presence	Oyster species	Habitat complexity	Mud crab feeding rate (oysters • $crab^{-1}day^{-1}$)	Expected oyster release[a] (oysters/d)	Actual oyster release[b] (oysters/d)	Toadfish effect 1: removal of crabs DMII[c] (expected/actual oyster release)	Toadfish effect 2: crab behavior change TMII[d] ([actual-expected]/ actual osyter release)
No toadfish							
	C. virginica	simple	0.48 (0.01)				
	C. virginica	complex	0.45 (0.01)				
	C. ariakensis	simple	0.47 (0.01)				
	C. ariakensis	complex	0.46 (0.02)				
Toadfish							
	C. virginica	simple	0.23 (0.07)	0.45 (0.22)	13.3 (4.0)	6.7% (5.6%)	93.3% (5.6%)
	C. virginica	complex	0.26 (0.06)	0.43 (0.18)	12.3 (3.8)	9.9% (6.3%)	90.1% (6.3%)
	C. ariakensis	simple	0.24 (0.08)	0.22 (0.08)	13.3 (4.3)	1.1% (0.3%)	98.9 (0.3%)
	C. ariakensis	complex	0.29 (0.04)	0.36 (0.11)	9.7 (3.3)	8.5% (3.5%)	91.5% (3.5%)

[a] Expected oyster release estimates the expected decrease in the average number of oysters consumed per day by mud crabs as a function of lowered crab densities in the presence of fish. Expected oyster release is calculated by multiplying the daily rate of crab removal of oysters in the absence of fish by the reduction in crab density induced by toadfish during the trial run.

[b] Actual oyster release calculations measure the actual number of oysters per day released from mud crab predation in the presence of toadfish. Actual release is calculated by subtracting the number of oysters consumed by mud crabs with toadfish present from the number consumed by mud crabs in the absence of toadfish.

[c] Toadfish effect 1: This is the percentage of toadfish indirect effects on oyster mortality explained by toadfish removal of mud crabs.

[d] Toadfish effect 2: This is the percentage of toadfish indirect effects on oyster mortality explained by toadfish-inducted modifications in mud crab foraging behavior.

Figure 4-1. The effects of oyster species, habitat complexity level, mud crab presence, and toadfish presence on surface rugosity index.

Mean surface rugosity index is equal to the average index value calculated from six individual experimental trials. Error bars indicate +1 standard error, and significant SNK post hoc results ($p < 0.05$) are represented by different letters above the error bars. Although presented in two separate graphs, data for each oyster species were analyzed in one single ANOVA; an 'a' over a bar from the *C. virginica* graph is similar to an 'a' over a bar from the *C. ariakensis* graph.

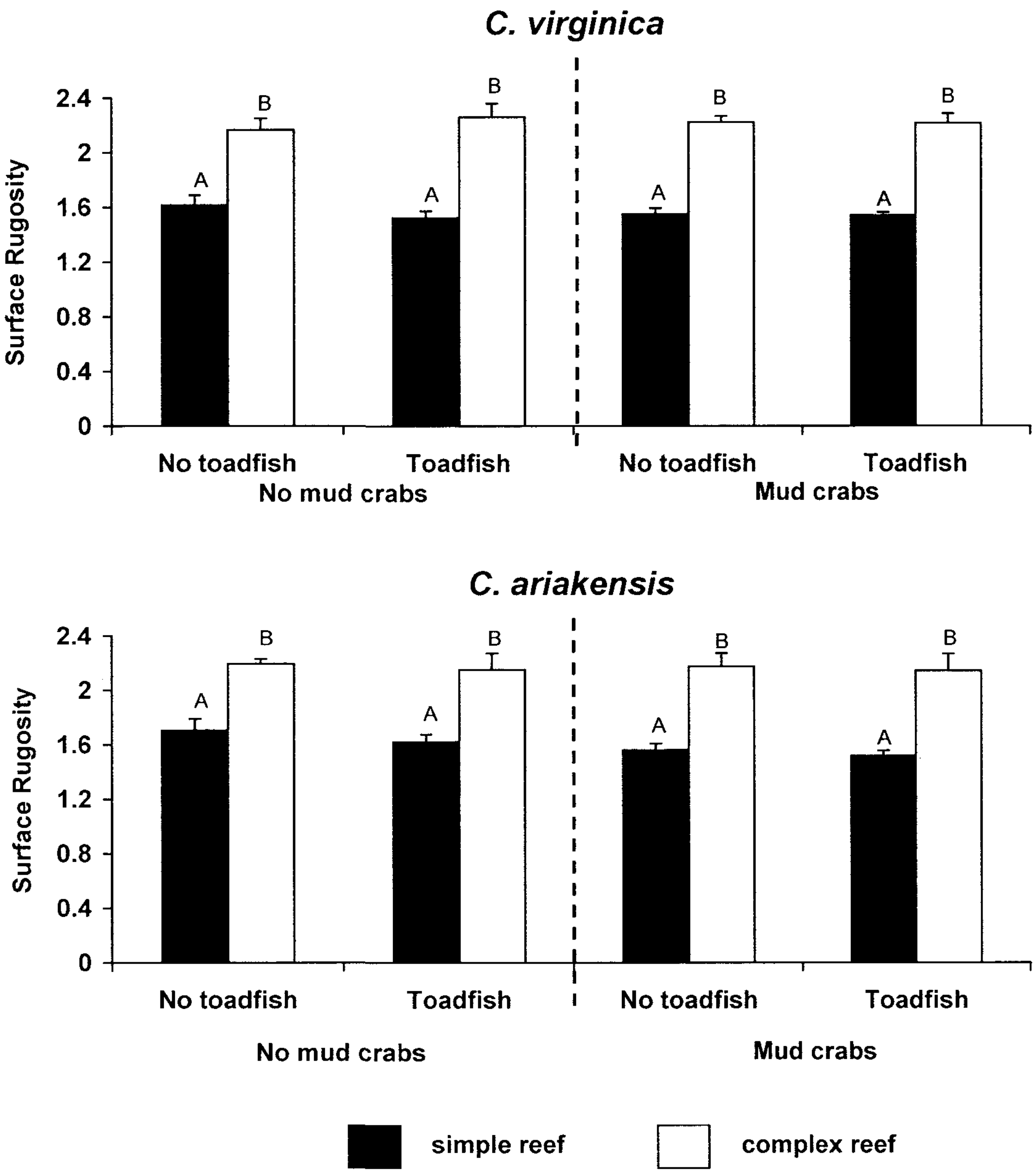

C. virginica
Surface Rugosity
2.4
2
1.6
1.2
0.8
0.4
0
A
B
A
B
A
B
A
B
No toadfish
Toadfish
No toadfish
Toadfish
No mud crabs
Mud crabs
C. ariakensis
Surface Rugosity
2.4
2
1.6
1.2
0.8
0.4
0
A
B
A
B
A
B
A
B
No toadfish
Toadfish
No toadfish
Toadfish
No mud crabs
Mud crabs
simple reef
complex reef

Figure 4-2. The effects of oyster species, habitat complexity, and toadfish presence on percentage mud crab mortality.

Crab mortality (%) is equal to the mean percentage of crabs per experimental run (from the original 10 crabs) that did not survive to the end of each 5-day trial. Error bars indicate +1 standard error, and significant SNK post hoc results ($p < 0.05$) are represented by different letters above the error bars. Although presented in two separate graphs, data for each oyster species were analyzed in one single ANOVA; an 'a' over a bar from the *C. virginica* graph is similar to an 'a' over a bar from the *C. ariakensis* graph.

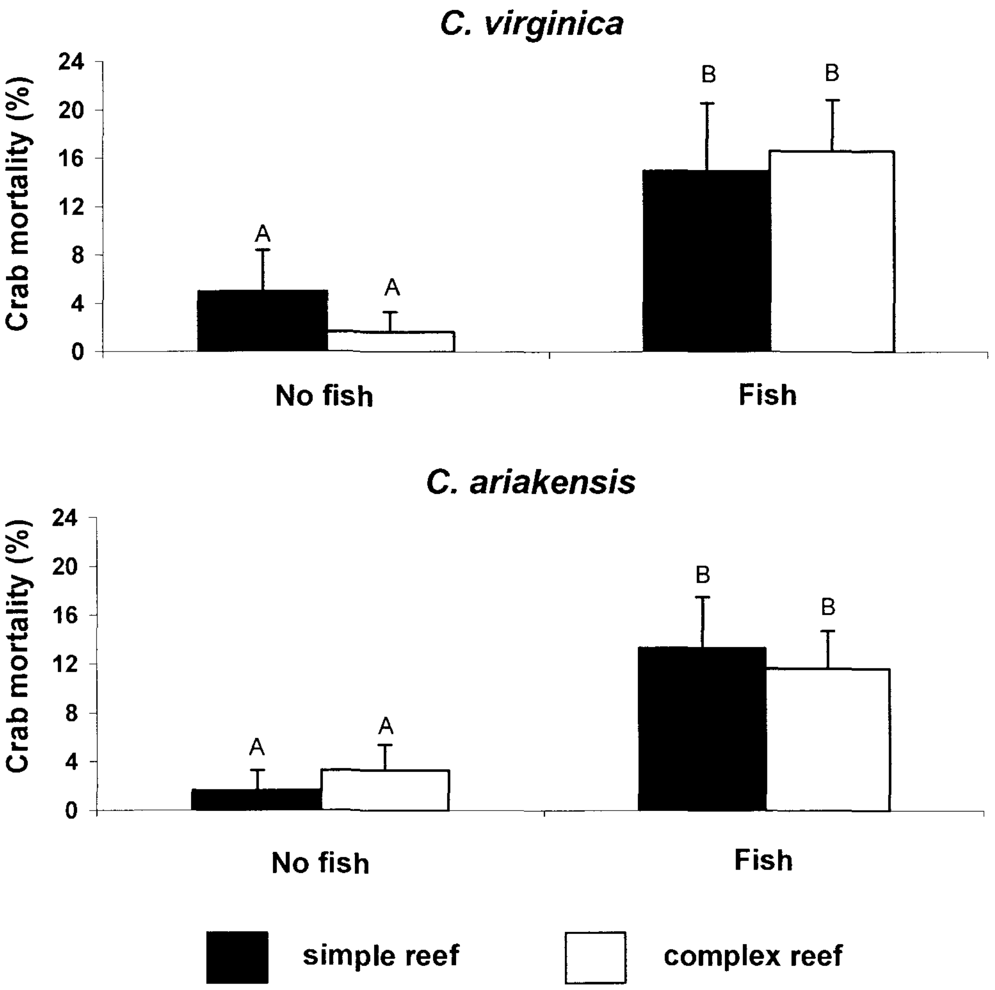

C. virginica
Crab mortality (%)
24
20
16
12
8
4
0
A
A
B
B
No fish
Fish
C. ariakensis
Crab mortality (%)
24
20
16
12
8
4
0
A
A
B
B
No fish
Fish
simple reef
complex reef

Figure 4-3. The effects of oyster species, habitat complexity, mud crab presence, and toadfish presence on juvenile *C. virginica* mortality.

Oyster mortality (%) is equal to the mean percentage of oysters per experimental run (from the original 30 oysters) that did not survive to the end of each 5-day trial. Error bars indicate +1 standard error, and significant SNK post hoc results ($p < 0.05$) are represented by different letters above the error bars. Although presented in two separate graphs, data for each oyster species were analyzed in one single ANOVA; an 'a' over a bar from the *C. virginica* graph is similar to an 'a' over a bar from the *C. ariakensis* graph.

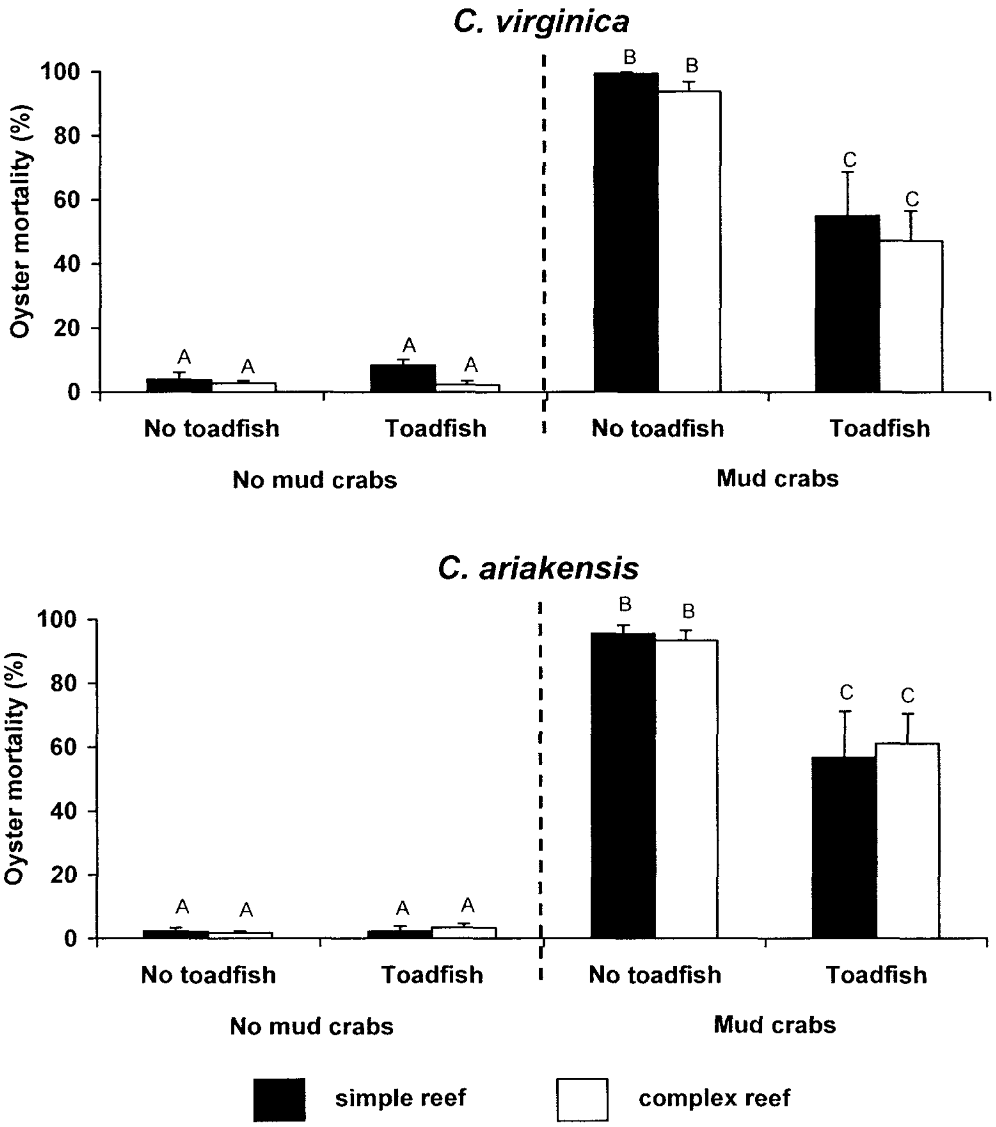

C. virginica
Oyster mortality (%)
100
80
60
40
20
0
A
A
A
A
B
B
C
C
No toadfish
Toadfish
No toadfish
Toadfish
No mud crabs
Mud crabs
C. ariakensis
Oyster mortality (%)
100
80
60
40
20
0
A
A
A
A
B
B
C
C
No toadfish
Toadfish
No toadfish
Toadfish
No mud crabs
Mud crabs
simple reef
complex reef

Figure 4-4. The mean percentage of visible mud crabs:

(A) during each of the five experimental days, (B) observed for each oyster species and habitat complexity combination, and (C) illustrating the effects of toadfish presence for each oyster species. Error bars indicate +1 standard error.

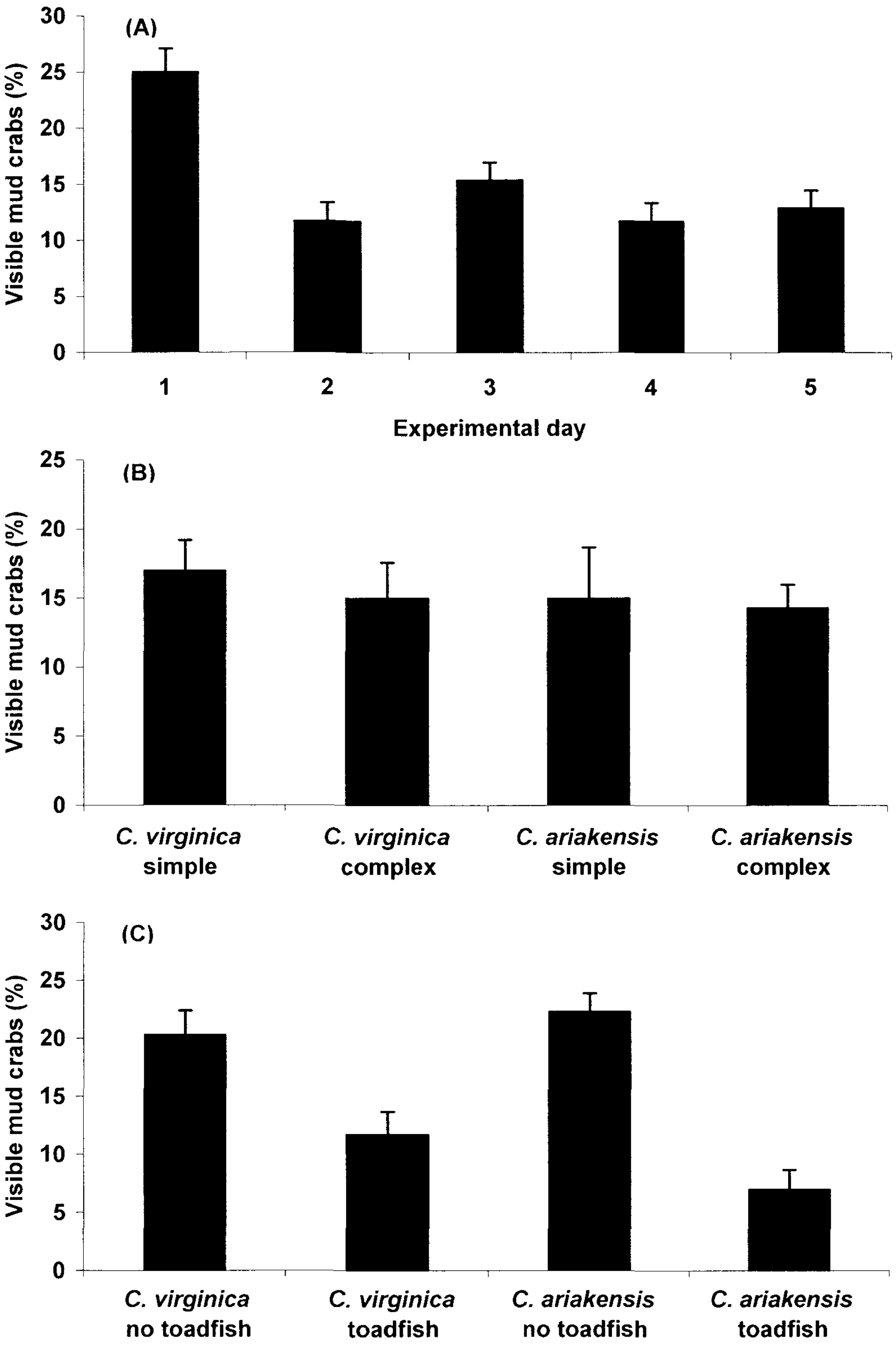
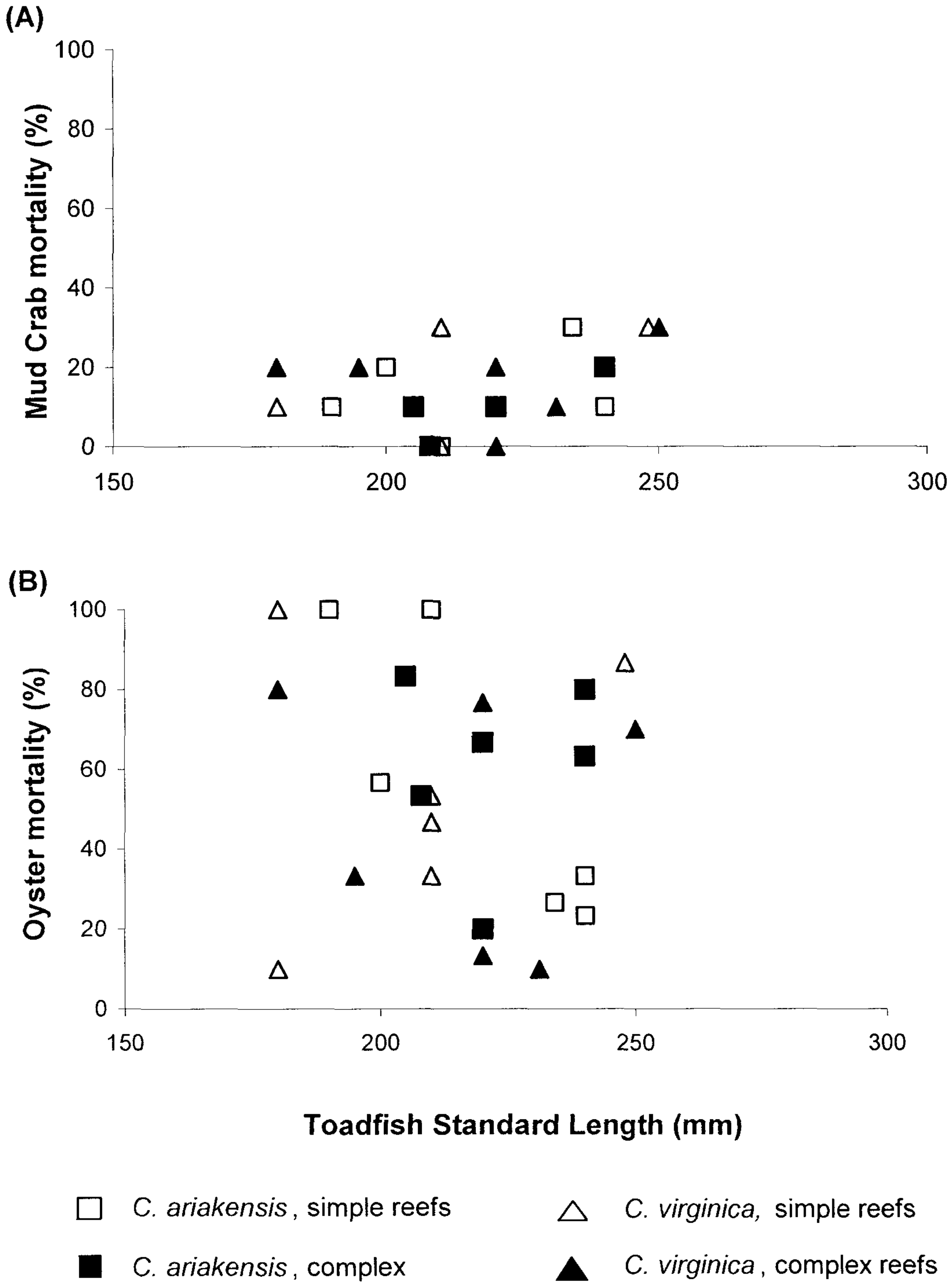

(A)
Visible mud crabs (%)
30
25
20
15
10
5
0
1
2
3
4
5
Experimental day
(B)
Visible mud crabs (%)
25
20
15
10
5
0
C. virginica simple
C. virginica complex
C. ariakensis simple
C. ariakensis complex
(C)
Visible mud crabs (%)
30
25
20
15
10
5
0
C. virginica no toadfish
C. virginica toadfish
C. ariakensis no toadfish
C. ariakensis toadfish

Figure 5-5. Percentage mud crab mortality (A) and juvenile *C. virginica* mortality (B) versus toadfish standard length (mm) for all experimental trials and treatments. All regressions were non-significant.


(A)
Mud Crab mortality (%)
100
80
60
40
20
0
150
200
250
300
(B)
Oyster mortality (%)
100
80
60
40
20
0
150
200
250
300
Toadfish Standard Length (mm)
C. ariakensis, simple reefs
C. ariakensis, complex
C. virginica, simple reefs
C. virginica, complex reefs

VITA

Heather Diane Harwell

Heather was born in Norfolk, Virginia on October 19, 1980. After graduating in 1998 from Frank W. Cox High School in Virginia Beach, Virginia, she went on to earn a B.S. in Marine Biology (*magna cum laude*, with a minor in Chemistry and departmental honors in Biology), from the University of North Carolina at Wilmington. In 2004, Heather received a Masters Degree in Marine Science from the University of North Carolina at Wilmington for her work under graduate advisor Dr. Martin Posey. Upon completion of her thesis, she enrolled in the Doctoral program at the Virginia Institute of Marine Science, College of William and Mary, working under Drs. Stan Allen and Mark Luckenbach.