WHAT LIMITS AN INVASIVE? BIOTIC AND ABIOTIC EFFECTS ON THE DISTRIBUTION OF THE INVASIVE MUSSEL *MYTILUS GALLOPROVINCIALIS* ON THE SOUTH AFRICAN COASTLINE.

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Abstract

Invasive species are negatively affecting communities and ecosystems worldwide, and rates of invasion are expected to increase. The potential for negative impacts from invasive species makes it important to understand invasion dynamics, particularly how novel species relate to their host communities and what factors control their distributions. This thesis investigates the factors controlling the distribution of an invasive mussel, *Mytilus galloprovincialis*, on the shores of South Africa. The distributional limit for the invasive coincides with a biogeographic break on the coast and a phylogeographic break in the native mussel, *Perna perna,* leading to two competing hypotheses. The hypotheses are: (1) The distribution of the invasive is determined by abiotic factors associated with the biogeographical break, and (2) the distribution of the invasive is determined by biotic interactions with different lineages of the native species. In order to determine what drives the large-scale pattern, I examined species survival across a range of biotic and abiotic conditions and tested for larval supply of the invasive. A competition experiment revealed that the eastern lineage of *P. perna* is competitively superior to the invasive, preventing the expansion of the invasive onto the east coast of South Africa. Additionally, a transplant of *M. galloprovincialis* on the east coast demonstrated that adult mussels can tolerate the abiotic conditions there. While *M. galloprovincialis* larvae were present in the waters off the east coast, more investigation is required into other aspects of larval supply (e.g. larval delivery and density) and the fitness of invasive settlers and recruits before it can be concluded that the invasive is not limited by supply-side factors there. The results of this thesis provide strong evidence for the role of biotic interactions and niche mechanisms in limiting the spread of the invasive mussel *M. galloprovincialis* in South Africa.

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Chapter 1: General Introduction

Pattern and Complexity in the Environment:

Across disciplines, from population biology to theoretical physics, the search for unifying laws in nature is fraught with the fundamental conceptual problem of scale (Levin 1992, Nottale 1996). Science strives to illuminate organizational mechanisms in order to predict and understand the natural world, yet patterns and the responses of systems and organisms can appear vastly different depending on the resolution and ranges of the experimental design (Levin 1992). For example, Rose and Leggett (1990) demonstrated that densities of predator and prey are positively correlated on the largest scale and simultaneously negatively correlated on the smallest scale. Experiments must be designed at scales that are ecologically relevant to the organisms being considered and, ideally, they should be conducted across the full range of scales, allowing them to incorporate all significant bio-physical interactions and effects (Rose and Leggett 1990, Fahrig 1992). This is, however, atypical as experimental design is caught in the balance between idealism and logistics (Rose and Leggett 1990, Levin 1992). As a result, ecological studies are often conducted on spatial and temporal scales that are orders of magnitude smaller than the scale at which the organizational processes of interest function (Levin 1992, Russell *et al.* 2006), potentially creating experimenter bias and flawed conclusions (Rose and Leggett 1990).

The environment is made up of patterns existing in both space and time that vary from regular to stochastic, from heterogeneous to homogeneous (Addicott 1987). The interaction of time and scale in determining pattern is complicated by the relative nature of these descriptors. Environments can simultaneously exhibit different patterns at different scales (Krebs 1978, Wiens 1989, Rose and Leggett 1990, Bar and Babbitt 2002). Consequently, patterns can be simultaneously long-lasting and ephemeral depending on the scale at which they are observed, the organisms under study and the ecological hierarchy involved (Southwood 1977, Connell and Sousa 1983). Furthermore, the existence of patterns in the environment does not necessarily mean that the described patterns are ecologically important to the organisms involved or ecological processes existing in that system (Addicott 1987). The nature of environmental patterns complicates their interpretation and can veil the importance of organismal responses in ecology.

Problems of pattern and scale plague scientists across disciplines, and those who attempt to understand important ecological drivers and responses in the marine environment are not spared! Abiotic and biotic pressures are non-uniform in space and time and vary according to species, traits (e.g. behavioural, physiological, reproductive and larval), the taxonomic resolution of the study and density (Breton and Addicott 1992, Hixon and Jones 2005, Freckleton and Lewis 2006, Diaz and McQuaid 2011). The different combinations and strengths of these interacting factors produce the diversity that is characteristic of most communities (Quinn and Dunham 1983, Diaz and McQuaid 2011). The non-uniform nature of the physical and biological environment makes generalizations about the forces that structure communities elusive at large spatial scales (Connolly and Roughgarden 1998). The fact that intertidal ecology has often focused on community interactions that operate at relatively small scales (Connell 1961, Paine 1966, Dayton 1971) is perhaps due to the heterogeneous nature of the system or the inherently intangible drivers of broad patterns. Strong physical gradients and biotic interactions drastically alter the environment, often inconsistently, at a variety of scales (Paine 1966, Menge *et al.* 1994, Guichard *et al.* 2003, Russell *et al.* 2006) making intertidal ecosystems highly variable and dynamic. As a result, the common forces that drive patterns at much larger scales, such as positive community interactions, are often neglected in intertidal ecology (Bertness and Leonard 1997, Bertness *et al.* 1999, Bruno *et al.* 2003). This thesis will, in part, address the more neglected drivers of large-scale ecological patterns by examining the forces that structure intertidal communities across biogeographical regions.

Community Composition

There are disparate perspectives when attempting to identify the factors that structure ecological communities (Hutchinson 1959, Hubbell 2001). A niche-based, equilibrium perspective of species coexistence and ecological communities places importance on biotic interactions and niche differentiation (Chesson 2000). Contrastingly, a neutral perspective postulates that species do not coexist at equilibrium due to niche partitioning; rather, species can be treated as competitively equivalent (Hubbell 2001). Under the simplifying assumptions of functional and ecological equivalency of species, neutral theory states that ecological drift and random processes structure ecological communities (Hubbell 2001, 2005, 2006). These two perspectives are strikingly different in the manner in which they approach understanding community structure; one relies on differences between species while the other relies on similarities, and much debate has ensued over which theory better describes natural systems (Leigh 1993, Chase and Leibold 2003, Guichard *et al.* 2003, McGill 2003, Ricklefs 2003, Chave 2004, Alonso *et al.* 2006, McGill *et al.* 2006). Despite the fundamentally different perspectives of these two theories on community ecology, there is merit to each approach (Hubbell 2001, Leibold and McPeek 2006), and empirical investigations have revealed that community composition in most environments appears to be shaped by a combination of niches and neutrality (Adler *et al.* 2007). The processes shaping natural communities can be thought of as lying on a continuum between niche and neutrality where few communities are determined exclusively by either niche or neutral processes, and the majority lie intermediately on the continuum (Gravel *et al.* 2006). Recent ecological thought has focused on determining where on this continuum a given community is located and the degree to which neutral and niche processes determine diversity in a community (Gravel *et al.* 2006, Adler *et al.* 2007). Empirical results from North and South America suggest that intertidal communities lie closer to the niche end of the spectrum than the neutral one (Menge 1976, Wootton 2005, Caro *et al.* 2010). For example, a large-scale study on intertidal community composition in South America showed that, at equilibrium, post-settlement processes were more influential than neutral mechanisms (Caro *et al.* 2010). In intertidal ecosystems, the concept of ecological equivalence on which neutral theory is based is not well supported; the differences between species on which niche theory is based appear to be critical to ecological structure and dynamics (Wootton 2005).

Community assembly in the intertidal is strongly affected by dispersal, recruitment and the availability of space (Connell 1961, Paine 1966, Dayton 1971, Connell 1972, Sale 1974, Menge 1976, Schoener 1982, Paine 1984, Underwood and Fairweather 1989, Connolly and Roughgarden 1999). For intertidal species that are broadly equivalent in resource requirements, unsynchronized dispersal and recruitment rates coupled with environmental heterogeneity can prevent competitive exclusion. Additionally, the interaction between environmental heterogeneity of the system and ecological trade-offs across species can be important for selective pressure on a small scale (Hubbell 2005). Differences among species may include ecophysiological trade-offs, such as responses to desiccation and wave stress, and biotic interactions, both intraspecific and interspecific. Moreover, biotic interactions can change in intensity with changes in species density and environmental stress (Breton and Addicott 1992, Hixon and Jones 2005, Freckleton and Lewis 2006, Eranen and Kozlov 2008, Rius and McQuaid 2009, Diaz and McQuaid 2011). Abiotic and biotic factors are highly heterogeneous in intensity, space and time, but important in shaping selection regimes and community structure. Therefore, in order to understand the important forces that structure communities on the continuum from niche to neutrality, we must examine the survival of species with similar requirements across a wide range of biotic and abiotic conditions. This is especially important as the changing global climate intensifies the environmental pressure on local species, and the number of new interspecific interactions increases through the spread of non-native species. The experiment at hand attempts to elucidate the roles of niche and neutrality by examining the survival rates of two competing mussel species, one invasive and one native, in the South African intertidal.

Theoretical Implications

The reality of invasion events by nonindigenous species has changed the conversation in community ecology (Shea and Chesson 2002, Coulatti and MacIsaac 2004, MacDougall and Turkington 2005). There is evidence that invasion events can alter community structure, composition and functioning with potentially devastating consequences (Porter and Savignano 1990, Kourtev *et al.* 2002, Strayer *et al.* 2006). The effects of invasive species are already expected to increase with time, giving urgency to understanding invasion biology and ecology (Carlton 1996, Shea and Chesson 2002, Gurevitch and Padilla 2004). Despite the danger posed by non-indigenous species to global biodiversity levels and the global economy, scientific opinion is mixed on the actual threats posed to biodiversity and the health of non-native environments (Wilson 1992, Carlton and Geller 1993, Lovei 1997, Wilcove *et al.* 1998, Stachowicz *et al.* 1999, Rosenzweig 2001, Pimentel *et al.* 2005, Davis *et al.* 2011). While contentious, the potential for negative impacts from invasive species makes it important to understand their ecological dynamics, including how they spread and what controls their distributions (Carlton and Geller 1993). From a management perspective, it is essential to understand the ecological relationship between invasive species and the native species in their host environments in order to mitigate negative impacts (King and Tschinkel 2006).

This thesis explores which factors limit the distribution of an invasive species across a biogeographical break on the South African coast. Specifically, what factors prevent an invasive species from expanding into seemingly suitable habitat across a given boundary? The distributional limit for the invasive species in this experiment coincides with a break in biogeographic regions, leading to the hypothesis that abiotic factors control species distribution and neutral mechanisms are key. There is a competing interpretation, however. The species against which the introduced animal must compete for space is an indigenous mussel. The distributional limit for the introduced species coincides with a phylogeographic break in the indigenous species with distinct genetic lineages having different physiological tolerances on either side. Based on the fundamentals of niche theory we can develop an alternative hypothesis: biotic factors, such as competition, will be more important than abiotic factors in determining the distribution of the invasive. The primary aim of this thesis is to identify whether biotic interactions or abiotic factors are more important in determining the distribution of the invasive. Additionally, this thesis will address whether the important factors transcend scale, and if the factors controlling species distribution change with species density.

Chapter 2: Effects of Abiotic Factors and Competition on *Mytilus galloprovincialis* and *Perna perna.*

Introduction

Invasion Biology

Worldwide, trade and tourism have provided an opportunity for the intentional or inadvertent transfer of invasive species between regions, and the rate of establishment of foreign marine species in ports has increased dramatically in recent decades (Arroyo *et al.* 2000, Wonham *et al.* 2000, Gurevitch and Padilla 2004). The United Nations defines invasive species as “organisms (usually transported by humans) which successfully establish themselves in, and then overcome, otherwise intact, pre-existing native ecosystems” (United Nations system-wide earthwatch; www.un.org/earthwatch). The most significant pathway of inadvertent species transfer depends on the type of organism (McQuaid and Arenas 2009). For marine organisms, the most effective dispersal mechanism is the ballast water of commercial vessels (Carlton and Geller 1993, Wonham 2000). Ballast water is taken from a harbour in one location and may be discharged elsewhere, allowing an opportunity for the introduction of a range of species surviving in the ballast water (Carlton and Geller 1993, Wonham 2000). At any given time up to 10,000 species are believed to be in transit between geographic regions in ballast tanks; ballast water surveys have revealed everything from the planktonic larvae of invertebrates to post-larval and adult fishes, some of which will establish in non-native regions and compete with native species (Carlton and Geller 1993, Wonham 2000, Bax *et al.* 2003). Invasive marine species have been implicated in the decrease of biodiversity, the collapse of numerous fisheries, and many negative social and economic impacts around the world (Gurevitch and Padilla 2004). Invasive species are severely affecting many communities and ecosystems worldwide, and the rates of marine invasions are expected to continue to increase particularly rapidly in coastal areas (Carlton 1996, Gurevitch and Padilla 2004). The breakdown of geographic barriers through invasion and the ensuing homogenization of global biota could precipitate a catastrophic loss of biodiversity (Lovei 1997, Rosenzweig 2001).

The introduction of non-native species poses great danger to the goals of conservation and currently costs billions to the global economy (Wilson 1992, Carlton and Geller 1993, Wilcove *et al.* 1998, Pimentel *et al.* 2005). The danger due to invasion by alien species is perceived to be so severe by ecologists and conservationists that it was named as one of the “mindless horsemen of the environmental apocalypse” by Harvard professor E. O. Wilson (1992), alongside habitat loss, overexploitation, and the spread of diseases carried by alien species. Invasive species are considered to be second only to habitat loss among the threats posed to biodiversity and conservation (Wilcove *et al.* 1998). For example, in the United States 400 of the 958 species listed as endangered or threatened by the Endangered Species Act are thought to have been imperiled by competitive or predatory interactions with non-native species (Wilcove *et al.* 1998). In addition, the 50,000 documented invasive species in the United States are estimated to cost the US economy $120 billion per year, $2 billion of this figure being associated with damage from invasive mollusks (Pimentel *et al.* 2005, Davis *et al.* 2011). Moreover, this figure does not quantify damages to biodiversity, ecosystem services, or resultant species extinctions, which would exponentially drive up the costs of damage associated with non-native species (Pimentel *et al.* 2005). Invasive plants have been known to disrupt ecosystem services through impeding essential plant-pollinator mutualisms as well as changing essential disturbance regimes like fire frequency (Mack and Antonio 1998, Traveset and Richardson 2006, Sax *et al.* 2007). In New Zealand, invasions by predatory mammals proved damaging through subsequent declines in native avifauna (Blackburn *et al.* 2005). There is, however, an increasingly popular opinion that invasive species must be assessed individually, that they do not necessarily threaten species diversity, that it is impossible to restore habitats to purely native species, and that not all invasives are dangerous to their adopted environments (Stachowicz *et al.* 1999, Rosenzweig *et al.* 2001, Davis *et al.* 2011). Along those lines, evidence shows that invasive predators and pathogens have been disproportionately implicated in native extinctions, whereas there is currently little or no evidence of species extirpations by invasive birds, plants or marine invertebrates (Sax *et al.* 2007, McQuaid and Arenas 2009). This controversial view suggests that much of our opinion on invasive species is simply opinion and not based on data and fact (Davis *et al.* 2011). The current understanding of invasive species is contentious, yet aliens are often viewed as a problem for global biodiversity and the global economy, making it all the more important to understand their ecology and any controlling factors in their distribution (Carlton and Geller 1993). Understanding the ecological relationships among invasive and native species and what controls the distribution and spread of introduced species in the colonized environment is essential for managing potentially negative impacts (King and Tschinkel 2006).

Interspecific Competition

Since the derivation of mathematical proofs defining the dynamics of competition by Lotka (1920) and Volterra (1928), scientists have fiercely debated the theory of competition; from the very definition of the word “competition” itself to the relative importance of competition as a driving force of natural selection in nature (Andrewartha and Birch 1954, Elton and Miller 1954, Birch 1957, Connell 1961, Connell 1972, Diamond 1978, Armstrong and McGehee 1980, Schoener 1982, Roughgarden 1983, Connell 1983, Grace 1991). Volterra’s 1928 paper *Variations and Fluctuation of the Number of Individuals in Animal Species Living Together* describes interspecific competition as: “Biological associations… established by many species which live in the same environment [where] the various individuals of such an association contest for the same food… The quantitative character of this phenomenon is manifested in the variations in the number of individuals which constitute the various species” (Volterra 1928 pg 4). Notwithstanding many decades of argument, it is recognized that competition is undoubtedly an important process that plays a central role in ecological theory and widely affects the structure and composition of a range of natural communities (Hairston *et al.* 1960, Connell 1961, Grant and Grant 1982, Paine 1984, Roughgarden 1983, Underwood and Fairweather 1989, McCann *et al.* 1998, Aerts 1999, Munday *et al.* 2001, Grant and Grant 2006). The mathematical models created by Lotka and Volterra and confirmed experimentally by Gause (1934) led to Gause’s principle, which states that species requiring resources that are too similar cannot coexist indefinitely as one will competitively exclude the other and species that naturally coexist accomplish this by requiring resources that are sufficiently different from one another (Volterra 1928, Gause 1934, MacArthur and Levins 1967, May and MacArthur 1972, Hairston

1980, Schoener 1982). Yet the relationship between levels of resource use and the occurrence of interspecific competition is not easily predictable or definable, as studies have shown strong interspecific competition is associated with both low and high levels of overlap in resource use (Birch 1957, Colwell and Futuyma 1971, Armstrong and McGehee 1980, Hariston 1980, Roughgarden 1985, Munday 2001). Despite this, numerous studies have shown that interspecific competition is extraordinarily strong in the case of sessile species that compete for space, a fact that holds true across intertidal and terrestrial ecosystems (Hairston *et al.* 1960, Schoener 1982, Sebens 1982, Branch 1984). Because primary space for attachment is often very limited in the rocky intertidal zone, space can be the essential limiting resource needed for survival and interspecific competition for space can become the critical factor in determining intertidal community structure (Connell 1961, Paine 1966, Dayton 1971, Connell 1972, Sale 1974, Menge 1976, Schoener 1982, Paine 1984).

The aim of this study was to examine the factors determining the distribution of the two species of mussels that dominate South African shores: the invasive *Mytilus galloprovincialis* and the native *Perna perna*. *M. galloprovincialis* is the most significant marine invader on South African coasts, where it competes for intertidal space with *P. perna* (McQuaid and Phillips 2000, Branch and Steffani 2004, Robinson *et al.* 2005, Bownes and McQuaid 2006). It is suspected that *M. galloprovincialis* was introduced to the shores of South Africa in the mid 1970's by shipping, whereupon it spread at a rate of approximately 115 kilometers per year (Branch and Steffani 2004). In the 1980's and 1990's, *M. galloprovincialis* was used for mariculture in Saldhana Bay and Port Elizabeth harbour (Pitcher and Calder 1998, Robinson *et al.* 2005). The population used for the mussel culture industry was subsequently removed but natural spread from the west coast populations had already begun (Phillips 1994, Robinson *et al.* 2005). Brought to South Africa in the ballast tanks of ships, there is cause for concern that *M. galloprovincialis* will continue to spread from South Africa in this manner to places as remote as subAntarctic Marion Island (Lee and Chown 2007). Currently *M. galloprovincialis* occurs on the South African coast from Luderitz in southern Namibia to East London on the eastern coast of South Africa (Figure 2.1) (Zardi *et al.* 2007). *M. galloprovincialis* now dominates the west coast of South Africa where it has caused declines in several indigenous species and constitutes 74% of the intertidal mussel biomass (Griffiths *et al.* 1992).

Species Distribution and Ecology

Competition is an evolutionary force powerful enough to select for adaptations leading to niche diversification in the form of differing resource use and distribution ranges amongst competing species, including habitat segregation (Hairston *et al.* 1960, Schoener 1982, Grant and Grant 2006). Vertical zonation in the distribution of intertidal species is globally documented and well supported in the literature, and zonation has been shown to play a major role in the competitive outcomes of many interspecific interactions in the intertidal (Baker 1909, Connell 1961, Dayton 1971, Connell 1972, Lubchenco 1980, Bownes and McQuaid 2010). The upper and lower limits of zonation in most intertidal species are likely determined by different forces: the lower limits are generally understood as being set by biotic factors, such as interspecific competition, and the upper limits by physical factors (Baker 1909, Connell 1961, Lubchenco 1980). There is some evidence that the reverse can also occur; upper limits can be determined by biotic factors (Bertness and Leonard 1997) and lower limits can be determined by physical factors such as mortality due to immersion (Lubchenco 1980). Despite this, most evidence supports the idea that biotic factors such as interspecific competition determine the lower limits of intertidal species as physical conditions, with the exception of wave action, are less harsh on the low shore (Baker 1909, Connell 1961, 1972).

In the case of the study species, the indigenous species, *P. perna* coexists with *M. galloprovincialis* along the south coast of South Africa, where the two species show partial habitat segregation. *P. perna* dominates the low mussel zone, *M. galloprovincialis* dominates the high zone, and the two species coexist in the mid zone (Bownes and McQuaid 2006, Rius and McQuaid 2006, Zardi *et al.* 2007). Of the four major South African intertidal mussel species examined in a 1992 study by Van

Erkom Schurink and Griffiths (*M. galloprovincialis*, *A. ater*, *C. meridionalis*, and *P. perna*), *M. galloprovincialis* showed the highest rates of water filtration and food particle absorption, while *P. perna* possessed intermediate levels. All four species showed faster growth rates at high water circulation sites and slower growth rates with increasing tidal elevation, yet each species had different tolerances to aerial exposure (Van Erkom Schurink and Griffiths 1993). Further studies of energy balance revealed that *M. galloprovincialis* also possesses the highest positive energy balance of the four species considered, which is mirrored by field observations that *M. galloprovincialis* can simultaneously maintain high growth and high reproductive output (Van Erkom Schurink and Griffiths 1992).

A study by Bownes and McQuaid (2006) demonstrated that habitat segregation by vertical zonation on the south coast of South Africa is responsible for the coexistence of *M. galloprovincialis* and *P. perna* and will likely prevent *M. galloprovincialis* from dominating this shoreline as it has on the west coast of South Africa (Griffiths *et al.* 1992). In areas of the coast where *M. galloprovincialis* is not present, *P. perna* ranges in continuous beds from the low-shore to the mid-shore and decreases in abundance on the high-shore (McQuaid *et al.* 2000). Recruitment limits colonization of both species on the higher mussel zone to some extent, however increased desiccation rates in the high zone are tolerated by adult *M. galloprovincialis* while excluding *P. perna* individuals (Rius and McQuaid 2006, Porri *et al.* 2007). Post-settlement effects such as wave action and interference competition with *P. perna* prevent *M. galloprovincialis* from spreading to the low-shore (Bownes and McQuaid 2006), although there is empirical evidence that larval transport and delivery may be also be important (e.g. Porri *et al.* 2006). In mixed beds, *P. perna* initially facilitates *M. galloprovincialis* in the low zone by protecting it from wave action before ultimately competitively excluding it (Rius and McQuaid 2006). Weak byssal attachment and a broader shell lead to *M. galloprovincialis* experiencing higher lift forces , which makes this species more vulnerable to dislodgement by wave action in the low zone (Rius and McQuaid 2006, Zardi *et al.* 2006). Farther upshore, stress from wave action decreases, while heat and desiccation stress increase. *M. galloprovincialis* is more tolerant of heat and desiccation than *P. perna,* and consequently *M. galloprovincialis* is dominant in the high mussel zone (Rius and McQuaid 2006). In the mid zone where both species occur, weak facilitation has been shown, indicating that the presence of *M. galloprovincialis* facilitates *P. perna* survival by providing a physical matrix where *P. perna* is partially protected from desiccation (Rius and McQuaid 2009).

Recent evidence has shown that two distinct genetic lineages of *P. perna* exist along the South African coast (Zardi *et al.* 2007). While the mechanism creating this genetic divergence remains unclear, the two would appear not to be sister-lineages (Cunha *et al.* 2014). The western *P. perna* lineage occurs on the south coast from Haga Haga to the Cape of Good Hope, and also extends north from Luderitz to the Mediterranean Sea (Zardi *et al.* 2007). The eastern *P. perna* lineage overlaps with the western lineage for 200 km from Kenton-on-Sea to Haga Haga, and continues eastward up the South African coast (Zardi *et al.* 2007, Fig. 1). Although the origins of the two lineages are unclear, it has been suggested that the lineages are maintained by a combination of selective pressure and limitations of dispersal (Zardi *et al.* 2011). The present study was designed to assess the roles of competition and abiotic factors in setting the distributions of *M. galloprovincialis* and the two lineages of *P. perna.*

Study Sites

The experiment was set up in December 2009 among mussel beds in the mid and low mussel zones at four sites on the coast of South Africa. Two sites were chosen in the Subtropical East Coast Province where the *P. perna* eastern lineage (*P. perna* East) is native and two in the Warm Temperate South Coast Province where the *P. perna* western lineage (*P. perna* West) is native. In the Subtropical East Coast Province (the east) these sites were Haga Haga (32°45'59.32"S, 28°14'35.72"E) and Kei Mouth (Kei Mouth A [32°42'33.35"S , 28°21'50.70"E], Kei Mouth B [32°42'30.79"S, 28°21'33.73"E]), and on the west side of the biogeographical break, the Warm Temperate South Coast Province (the west), they were Skoenmakerskop (34° 2'31.99"S, 25°32'40.00"E) and Kini Bay (34° 1'19.95"S, 25°22'48.39"E).

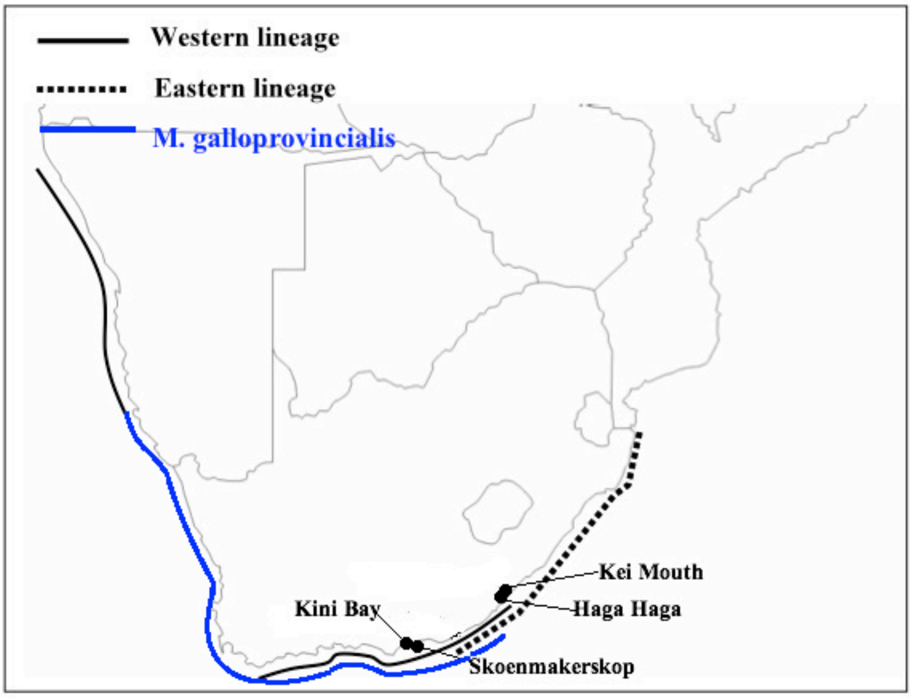


Figure 2.1: Map of southern Africa showing distribution of* P. perna *lineages,* M. galloprovincialis *distribution, and study sites.

Experimental Design

Methods for the competition experiments follow an adjustment of the procedure of Rius and McQuaid (2006) and are described below. The experiment is designed to account for both intraspecific and interspecific competition, in accordance with Underwood (1986). In order to examine the effects of abiotic factors and competition on the two *P. perna* lineages separately, they were treated as different species in the experimental setup.

Due to the partial habitat segregation seen in the intertidal distributions of *M. galloprovincialis* and *P. perna* on the South African coast, it was important to examine the effects of zone on species/lineage survival, condition, and total growth. The intertidal zone was divided vertically into two mussel zones, the low and the mid, in accordance with procedures adapted from previous intertidal studies (see Rius, 2004). The methodology used to divide these zones in this study was as follows: the border between the subtidal and low zones was delineated by the presence of the ascidian *Pyura stolonifera* in the subtidal, the low mussel zone was defined by the presence of encrusting coralline algae (*Spongites yendoi*), colonial polychaetes (*Gunnarea capensis*) and large mussel patches; the mid mussel zone where mussel beds predominate; and the high mussel zone characterized by barnacles (*Tetraclita serrata, Chthamalus dentatus,* and occasional *Octomeris angulosa*) and sparse mussel patches.

The experimental design involved nine treatments replicated four times in each of the mid zone and the low zone at each study site (Table 1). This design resulted in a total of 288 experimental quadrats and a total of 3,200 individuals per lineage/species for an overall total of 9,600 individuals across both zones at each of the four study sites. Small mussels between 20 and 30 mm were collected from Plettenberg Bay (*M. galloprovincialis* and *P. perna* West) and Kei Mouth (*P. perna* East) and placed at the study sites in December 2009. Following collection, the mussels were placed in large ziplock bags and covered with seawater. The bags of mussels were then put into cooler boxes, kept chilled with frozen freezer blocks, and covered with paper towels soaked in sea water for transport back to the laboratories at the Department of Zoology and Entomology at Rhodes University, Grahamstown, South Africa. In preparation, buckets of seawater were placed in a controlled environment room at 19° C and circulated for 24 hours to homogenize and oxygenate. Once at the laboratory, the mussels were placed in the circulating seawater in controlled environment rooms for one day at 19° C with a 12 hour: 12 hour light:dark cycle. Each mussel was then tagged for growth and species identification and separated into treatment bags for placement. The tagging involved attaching 2 mm paper triangles to the growing edge of each mussel, securing the tags with superglue. *P. perna* West and *M. galloprovincialis* were both marked with red growth tags as *P. perna* and *M. galloprovincialis* are morphologically distinguishable by shell shape and color (Van Erkom Schurink and Griffiths 1990). Additionally, *P. perna* East was marked with blue growth tags in order to discriminate between the two lineages which have not been shown to be morphologically distinguishable.

	Treatment	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**
Species										
PE		20	40	-	-	-	-	20	-	20
PW		-	-	20	40	-	-	20	20	-
Mytilus		-	-	-	-	20	40	-	20	20

Table 2.1: Experimental Treatments.

List of competition experiment treatments addressing both intra and interspecific competition, following Underwood (1986). Mussels were placed in monospecific and mixed cages in varying densities to assess competitive effects of intraspecific and interspecific competition, respectively. Values indicate number of individuals in each experimental quadrat. PE= P. perna East. PW= P. perna West.

Comparison	**Intraspecific Competition**	
	PE	1 vs 2
	PW	3 vs 4
	Mytilus	5 vs 6

Table 2.2a. Intraspecific Competitive Comparisons.

Description of which treatment comparisons listed in Table 2.1 *address intraspecific competition. Evaluation of intraspecific competition is accomplished by comparing two monospecific cages with different animal densities for each species/lineage.*

	Interspecific Competition	
Comparison	PW on PE	1 vs 7
	PE on PW	3 vs 7
	M on PW	3 vs 8
	PW on M	5 vs 8
	M on PE	1 vs 9
	PE on M	5 vs 9

Table 2.2b: Interspecific Competitive Comparisons.

Description of which treatment comparisons listed in Table 2.1 *address interspecific competition. Evaluation of interspecific competition is accomplished by comparing variables (mortality, growth, condition) in a monospecific cage with variables for cages containing a mixture of two species, with the subject species at the same starting density. For example, comparing survival, condition or growth of PE in treatments 1 and 7 tests for effects of PW on PE.*

Experimental Setup

Metal quadrats, measuring 0.1 m X 0.1 m, were attached to the shore in the mid and low mussel zone using screws. Treatments were haphazardly placed in quadrats, covering the mussels tightly with a soft plastic mesh (2 mm) to allow them to form new byssal threads and adequately attach to the substratum. After four weeks, the mesh was removed and lids with mesh were placed on top of each quadrat using cable ties. After a total of three months from the start of the experiment, the mesh lids were removed and new lids formed of looser, coarser mesh (4 mm) were placed over each quadrat. After eight months of running time, the lids and loose mesh were completely removed (Figure 2.2) and the experiment continued with the quadrats uncovered for three additional months for a total run time of 11 months.

Figure 2.2: A fully occupied cage. *Blue tags indicate the growing edge when the animals were initially tagged when placed in the cage.*

Experimental Re-set

Following the set-up of the experiment in December 2009, the first monthly mortality monitoring in January 2010 revealed devastating losses at both zones at Kei Mouth. In cases where 50% or more of replicates of a particular treatment had been lost, new replicates were placed in March 2010 to supplement the surviving replicates. The first site, Kei Mouth A, was an exposed site located at Kei point. To avoid extreme losses a second time, a second Kei Mouth site was chosen for the re-set of replicates. This site, Kei Mouth B, was 400 m West of Kei Mouth A and less exposed. Methods of collection, transport, tagging, and placement of mussels followed the first experimental set-up described above.

Survivorship: Measurements and Calculations.

Mortality measurements were taken monthly for the duration of the experiment. Empty shells and tagged mussels displaced outside of the quadrat boundary were counted as dead. The number of surviving mussels at the end of the experiment was measured for each quadrat and used to calculate the % Survivorship.

$$\%\ \text{Survivorship} = \frac{(\#\ \text{surviving mussels})}{(\#\ \text{initial mussels})}\ \text{X}\ 100$$

Growth and Condition Index

Upon collection of the experiment in November 2010, the surviving mussels from each cage were placed in plastic jars and preserved in 70% ethanol until processed in the laboratory for growth and condition measurement. Growth assessment involved measuring initial shell length (marked by growth tags placed at initial collection) and final shell length using Vernier calipers accurate to 0.1 mm. Condition index (CI) was measured by removing the soft tissue from each mussel, drying the shell and accompanying tissue for 24-48 hours at 60° C in a desiccating oven, then using the following formula from Davenport and Chen (1987):

$$\text{CI} = \frac{\text{Dry meat weight}}{\text{Shell weight}}\ \text{X}\ 100$$

Genetic Verification of *P. perna* Lineages

Upon collection, it became clear that genetic verification was necessary for individuals in the *P. perna* East vs. *P. perna* West treatment, as many of their lineage tags had fallen off and the lineages have not been shown to be morphologically distinguishable. A 0.1 g biomass sample was taken from the mantle of each unidentified mussel and preserved in absolute ethanol for lineage verification by DNA extraction, amplification and sequencing. DNA was extracted from the tissue samples using an Invisorb Spin Tissue Mini Kit, following the manufacturer's protocol. DNA in the extractions was then amplified using a polymerase chain reaction (PCR) using primers LCO 1490 (5’-GGT CAA CAA ATC ATA AAG ATA TTG-3’) and HCO 2198 (5’-TAA ACT TCA GGG TGA CCA AAA AAT CA-3’; Folmer *et al.* 1994) to amplify a portion of the mitochondrial cytochrome oxidase subunit I gene (MtDNA COI).

Analysis

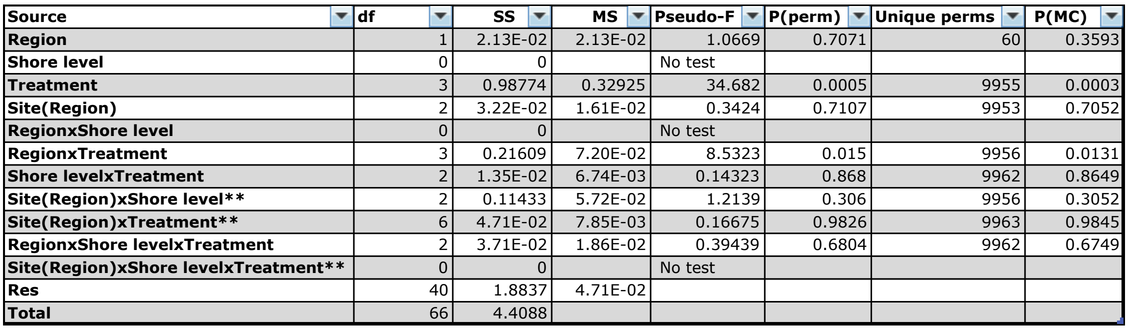
Percent survivorship, condition index, and growth were analyzed separately using PERMANOVA. There were four factors: region, shore level and treatment were all fixed factors, with site as a random factor nested in region. The % survivorship data were ArcSin transformed. Data were used to generate a resemblance matrix utilizing the D1 Euclidean distance before analysis. Because of the large number of possible permutations (9 groups, 4 replicates, 36!= 3.7e41 total possible permutations), the Monte Carlo P-values and the permutation P-values converge (Anderson 2005). Within levels of the factor “region”, however, the number of possible unique permutations is greatly reduced (Table 2.3), therefore it is advantageous to use the Monte Carlo P-values over the permutation P-values (Anderson 2005) and the Monte Carlo P-values are reported hereafter.

Results

Competition Experiment

Wave induced losses and sand inundation disproportionately affected the mid zone at Kini Bay and the low zone at Haga Haga, and thus PERMANOVA was not able to test for effects of Shore level (Table 2.3).For survival, the only significant effects were Treatment (P = 0.0003) and the interaction of Treatment with Region (P = 0.0131, Table 2.3). *M. galloprovincialis* showed lower survival when in combination with *P. perna* East in both regions (M20 > PE/M, P=0.0426, P=0.0239 for East and West respectively). *P. perna* West did not affect survival of *M. galloprovincialis* in either region.   In the Western region, *M. galloprovincialis* exhibited intraspecific competition (M40 < M20; P=0.0062), but this effect was not seen in the east (P<0.093; Table 2.4 and Figure 2.2).

With the exception of a strong effect of Site (P=0.0003 and P=0.0006 for growth and CI, respectively) there were no significant main effects or interactions for growth or CI for any treatment combination.



Source	df	SS	MS	Pseudo-F	P(perm)	Unique perms	P(MC)
Region	1	2.13E-02	2.13E-02	1.0669	0.7071	60	0.3593
Shore level	0	0		No test			
Treatment	3	0.98774	0.32925	34.682	0.0005	9955	0.0003
Site(Region)	2	3.22E-02	1.61E-02	0.3424	0.7107	9953	0.7052
RegionxShore level	0	0		No test			
RegionxTreatment	3	0.21609	7.20E-02	8.5323	0.015	9956	0.0131
Shore levelxTreatment	2	1.35E-02	6.74E-03	0.14323	0.868	9962	0.8649
Site(Region)xShore level**	2	0.11433	5.72E-02	1.2139	0.306	9956	0.3052
Site(Region)xTreatment**	6	4.71E-02	7.85E-03	0.16675	0.9826	9963	0.9845
RegionxShore levelxTreatment	2	3.71E-02	1.86E-02	0.39439	0.6804	9962	0.6749
Site(Region)xShore levelxTreatment**	0	0		No test			
Res	40	1.8837	4.71E-02				
Total	66	4.4088					

Table 2.3: PERMANOVA results table. *Results from the PERMANOVA comparison of ArcSin transformed survivorship data. df= degrees of freedom, SS= sum of squares, MS= mean squared, Psuedo-F= pseudo F-ratio, P(perm)= p value from permutations, Unique perms= p value from unique permutations, P(MC)= Monte Carlo p-value*

Region: West				
Groups	**t**	**P(perm)**	**Unique perms**	**P(MC)**
M20, PE/M	27.23	0.0001	6	0.0239
M20, M40	3.6974	0.0072	9804	0.0062
M20, PW/M	1.0949	0.4958	6	0.6212
PE/M, M40	4.6451	0.0015	9825	0.0015
PE/M, PW/M	3.1011	0.339	6	0.1968
M40, PW/M	3.1295	0.0091	9866	0.009
Region: East				
Groups	**t**	**P(perm)**	**Unique perms**	**P(MC)**
M20, PE/M	13.596	0.1028	30	0.0426
M20, M40	8.7864	0.0337	28	0.093
M20, PW/M	1.2434	0.2266	9801	0.2422
PE/M, M40	No test, df = 0			
PE/M, PW/M	1.8508	0.2972	28	0.3668
M40, PW/M	1.1439	0.2789	9863	0.2753

Table 2.4: Post-hoc PERMANOVA comparisons of survivorship at the Region X Treatment level. *P(perm)= p value from permutations, Unique perms= p value from unique permutations, P(MC)= Monte Carlo p-value.*

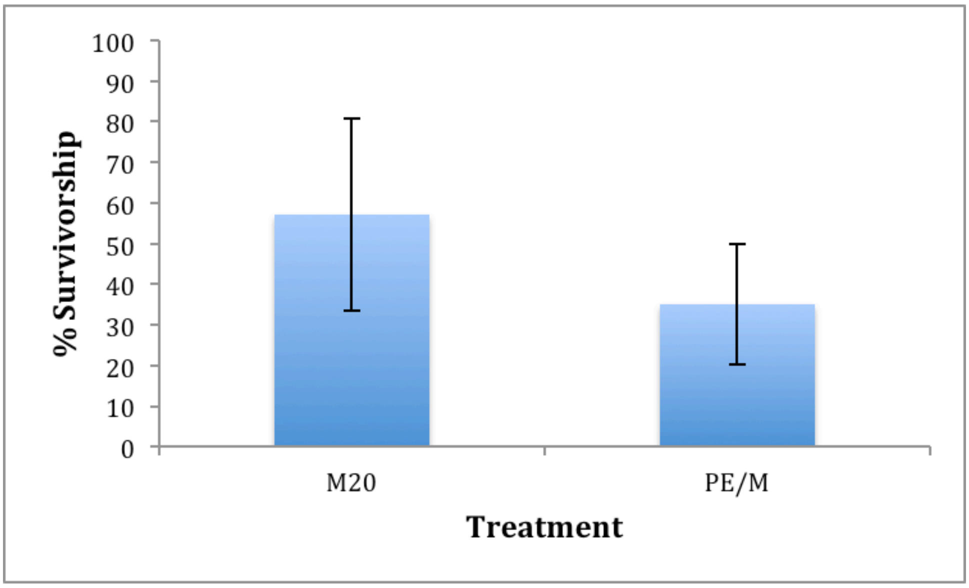


Figure 2.3a: Mean percent survivorship of* M. galloprovincialis *in monospecific (M20), and mixed (PE/M) treatments at the eastern sites (Haga Haga, Kei Mouth). Error bars represent one standard deviation.

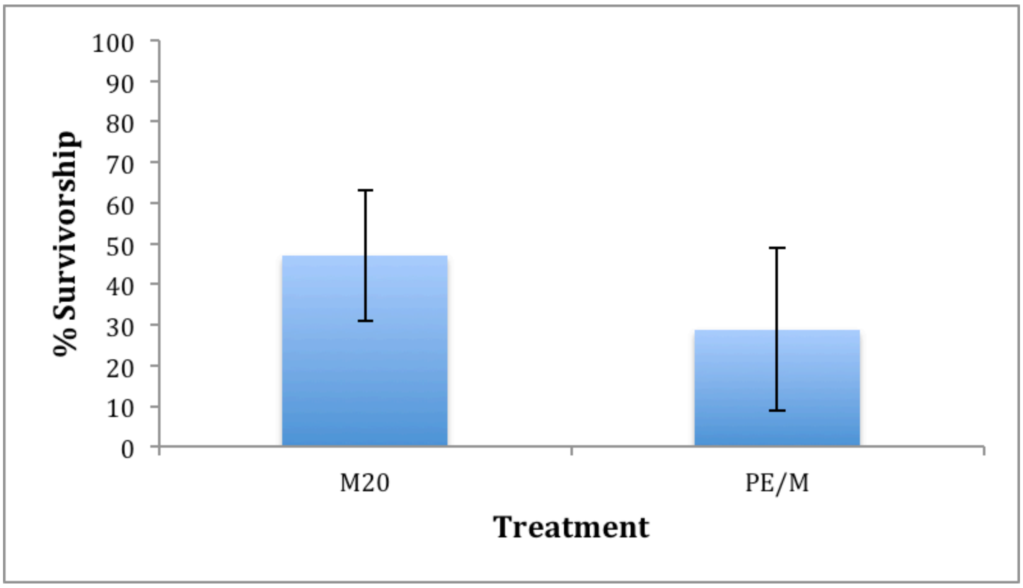


Figure 2.3b: Mean Percent survivorship of* M. galloprovincialis *in monospecific (M20), and mixed (PE/M) treatments at the western sites (Skoenmakerskop, Kini Bay). Error bars represent one standard deviation.

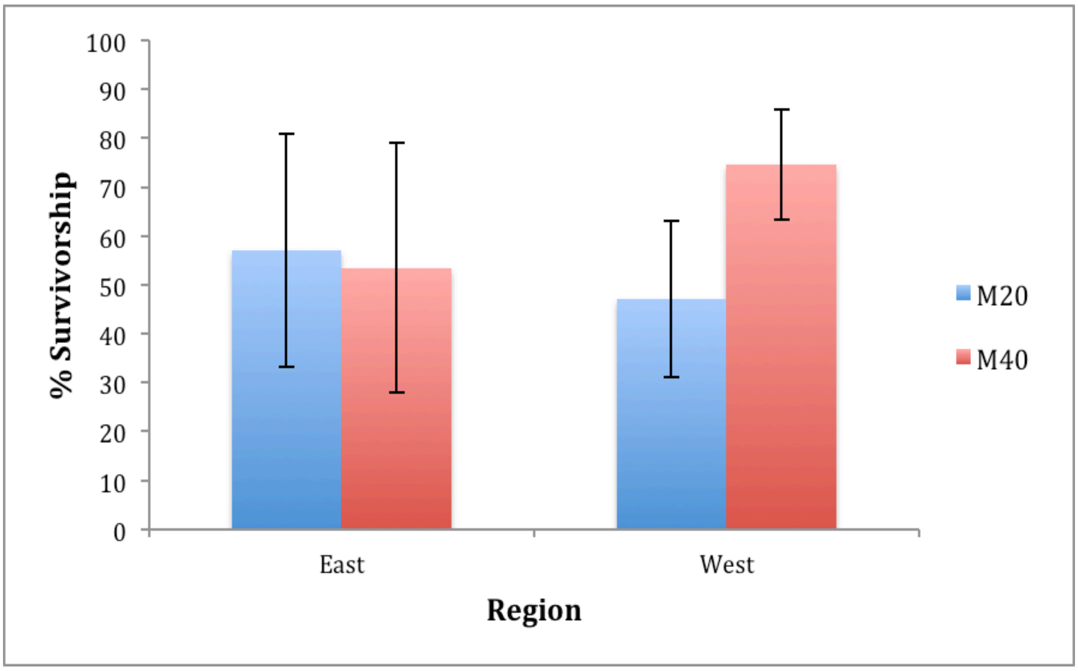


Figure 2.4: Mean percent survivorship of* M. galloprovincialis *in the two treatments M20 and M40 in the east (Haga Haga, Kei Mouth) and in the west (Skoenmakerskop, Kini Bay). Error bars represent one standard deviation.

Genetic Verification of *P. perna* Lineages

There was incomplete DNA amplification for the unknown individuals in the PE/PW treatments. This may have resulted from DNA degradation when the mussels were preserved in 70% alcohol, perhaps a higher concentration of alcohol should have been used. As a result, this treatment was excluded from the analysis.

Discussion

Individual *M. galloprovincialis* adults have been found in the subtropical Eastern Province, but only as isolated individuals in *P. perna* beds (B. Mostert, A. Ludford, pers. comm.; M. Hall pers. obs.). In addition, *M. galloprovincialis* settlers have been recorded in Coffee Bay on South Africa's Wild Coast, east of the biogeographical boundary of East London where adult *M. galloprovincialis* beds do not occur (unpub. data). These results would suggest that *M. galloprovincialis* larvae are reaching the Eastern region despite the oceanographic challenges posed by the Agulhas Current (Zardi *et al.* 2007, 2011) but that they very rarely survive to adulthood.

The results for survivorship of *M. galloprovincialis* in the M20 and M40 treatments by region revealed that the survivorship of *M. galloprovincialis* in the M40 treatment is significantly higher than in M20 for the Western region but not the Eastern region (Figure 2.4). Positive density-dependent survivorship is common in species of sessile organisms (Bertness and Leonard 1997) as they can provide each other with structural protection that enhances survival. This has been demonstrated initially for mixed beds of *M. galloprovincialis* and *P. perna*, where, on the low shore, *P. perna* initially protects the comparatively weakly attached *M. galloprovincialis*, though *P. perna* later competitively displaces *M. galloprovincialis* (Rius and McQuaid 2006). On the midshore, the roles are reversed and *M. galloprovincialis* can reduce the effects of desiccation stress on the more sensitive *P. perna* (Rius and McQuaid 2009). The lack of significant positive density-dependent survivorship in the east can be interpreted as indicating that environmental stress in the subtropical Eastern region was so strong as to override density-dependent intraspecific facilitation in *M. galloprovincialis.* This accords with the suggestion of Rius and McQuaid (2009) that facilitation need not increase with environmental stress, but can be overridden when such stress becomes so great as to override the beneficial effects of facilitation. Thus, regardless of density, *M. galloprovincialis* suffers reduced survival in the east, possibly because of abiotic environmental pressure overcoming the facilitation and protection offered by increasing densities. Some of this stress can be attributed to temperature, as maximum temperatures have been shown to be significantly higher in the eastern than in the western region (Zardi *et al.* 2011).

Rius and McQuaid (2006) demonstrated a negative effect of *P. perna* on *M. galloprovincialis* survival. Although there was no distinction made as to the lineage of *P. perna* in that experiment as the genetic structure of the species was then unknown, the mussels are presumed to be *P. perna* West as they were collected at a western site where the study was carried out. In the present study, this effect was not found to be significant in the *post-hoc* comparison of PW/M and M20 treatments for the same region as in the Rius and McQuaid study (P>0.60). Separating the two lineages, however, has clarified their order of competitive strength in relation to region. Survivorship levels of *M. galloprovincials* were significantly lower in the PE/M treatement than the M20 treatment regardless of region (P<0.0426 and P<0.0239 for the east and west respectively), indicating that *P. perna* East is competitively superior to *M. galloprovincialis* even in the west where *P. perna* East does not normally occur. This indicates that *P. perna* East can outcompete *M. galloprovincialis* on both side of the biogeographical boundary between the warm temperate South Coast Province and the subtropical East Coast Province. The fact that *P. perna* East can outcompete *M. galloprovincialis* on the western side of the boundary, which is not its region of origin, indicates that *P. perna* East is an especially strong competitor in these intertidal interactions. This adds to the results of Zardi and co-authors (2011), which concluded that *P. perna* East is physiologically more robust than its western counterpart. Restricted gene flow between populations allows for adaptation of novel traits to fulfill local ecological requirements, some of which may result in competitive advantages (Mayr 1983, Garcia-Ramos and Kirpatrick 1997, Bekessy *et al*. 2003). There is evidence that the phylogeographic structure of *P. perna* is due to the Agulhas Current restricting gene flow between the two lineages and the presence of sharp environmental clines in the east and west (Zardi *et al.* 2011). Either local adaptation of the *P. perna* East lineage or its phylogenetic history has resulted in a trait or traits that translate into competitive superiority of the lineage across regions. Still, at this point, the mechanism that results in the competitive superiority of the *P. perna* East lineage remains unclear.

Spatial dominance of mussels in the intertidal is dictated by the summation of growth, recruitment, and mortality rates (Petratis 1995). The absence of significant treatment effects for growth and CI could partially be interpreted as cumulative effects of competition and environmental stress that are detrimental but only reflected in mortality (Michalek-Wagner and Willis 2001, Petes *et al.* 2007, Zardi *et al.* 2011). Growth rates in mussels result from a range of factors that were not measured across sites during this experiment, including overcrowding, temperature, light, and food availability (Petratis 1995). One or more of these factors could have some potentially confounding impact that has disguised any significant competitive effects on growth rates and condition. *M. galloprovincialis* is known to possess a faster growth rate than *P. perna*, at least seasonally (Van Erkom Schurink and Griffiths 1993, Bownes and McQuaid 2010), therefore the absence of a significant difference in growth rates suggests some levels of stress in *M. galloprovincialis*. This would be consistent with the results of previous studies on *M. galloprovincialis* that demonstrated decreased growth under high environmental stress levels despite the species' characteristic resilience to thermal stress and desiccation (Petes *et al.* 2007). These results suggest that mortality is the most important, extreme and integrated element characterizing interspecific competition between *M. galloprovincialis* and *P. perna.* Gamete release can, however, lead to losses of 50% of wet flesh mass of the animal and represents a large energetic investment for pelagic and benthic filter-feeders (Griffiths 1977, Van Erkom Schurink and Griffiths 1991). Secondary production by mussels in the form of gamete release can be significant input to surrounding systems, as high as 10.4 kg m $^{-2}$ yr $^{-1}$ (Van Erkom Schurink and Griffiths 1991). While males spawn a higher raw number of gametes than females, the genders tend to spawn similar amounts of gametes by weight, and CI cannot be distinguished between sexes (Griffiths 1977, Griffiths and King 1979, Thompson 1979, Van Erkom Schurink and Griffiths 1991). Condition index relies absolutely on dry flesh weight, which can vary by a factor of 2-3 over a period of days with gamete release, depending on the species (Van Erkom

Schurink and Griffiths 1992). *M. galloprovincialis* possesses the greatest capacity to accumulate and release gonad material within a spawning cycle (Van Erkom Schurink and Griffiths 1991). These fluctuations in animal mass make CI measurements noisy, complicated, and somewhat unreliable, especially when one considers that the timing of gametogenesis varies with species (Van Erkom Schurink and Griffiths 1991, Zardi *et al.* 2007).

Since there was no significant difference in growth rate between the two species, and since *M. galloprovincialis* can survive the stressful environmental conditions in the East, interspecific competition is the only remaining variable capable of explaining the distribution of *M. galloprovincialis*. These results suggest that the competitive superiority of the *P. perna* East lineage is the driving factor preventing establishment of *M. galloprovincialis* beds on the east coast of South Africa. This conclusion is confirmed by the significantly higher mortality rates of *M. galloprovincialis* when competing with *P. perna* East in comparison to the mortality rates of *M. galloprovincialis* when competing with *P. perna* West.

This raises the intriguing possibility that interspecific competition with *P. perna* East is therefore the critical factor determining the distribution limit of *M. galloprovincialis* and preventing the expansion of the species into the subtropical eastern region of South Africa. The high performance of *P. perna* in very hot, nutrient poor conditions (Van Erkom Schurink and Griffiths 1992) is supported by the general field observations that *P. perna* is successful on the east coast and the success of the *P. perna* East lineage in this experiment. Future work should look at the effects of higher temperatures on energetic balances in the eastern and western lineages of *P. perna.*

Chapter 3: The Effect of Abiotic Factors on Survival of *Mytilus galloprovincialis* in the Eastern Region

Introduction

The nature of biological invasion is highly dependent on physical, environmental factors. The spread of invasive species is not only shaped by the physical characteristics of the donor and recipient regions, but the ultimate fate of an inoculation event in a recipient region can be driven by physical factors and environmental stochasticity (terminology from Carlton 1996). For instance, a flooding event that suppressed native biota has been implicated in the subsequent invasion of San Francisco Bay by an invasive clam (Nichols *et al*. 1990). The intensity and frequency of physical environmental changes can be affected by human activities, which can influence the likelihood of an invasion event in space and time (Carlton 1996, Ruiz *et al.* 1997). For example, industrial discharges into the Rhine River in The Netherlands changed water temperatures and salinity levels, priming the environment for invasion by a brackish water amphipod (Van den Brink *et al.* 1993). Human activities not only change the physical environment but are responsible for inoculation events themselves. The best example of this is shipping, as the frequency and nature of shipping activity has been implicated in numerous invasion events, including marine bioinvasions (Carlton *et al.* 1995, Carlton 1996, Ruiz *et al.* 1997, Ruiz *et al.* 2000). Thus, the interplay between natural environmental stochasticity and human effects on the physical environment can help drive the establishment of species in non-native regions. In the case of *Mytilus galloprovincialis,* spread of the species was initiated by human activity, and seems to have been both facilitated and hindered by different environmental characteristics of the South African coast.

M. galloprovincialis is a globally successful invader with nearly all points of introduction coinciding with large shipping ports in temperate regions (Branch and Steffani 2004). This description matches the limited number of potential introduction points in South Africa, and thus experts have retrospectively been less than surprised that *M. galloprovincialis* was successfully introduced to Saldhana Bay, South Africa in the 1970s (Branch and Steffani 2004). The presumed vector of invasion for *M. galloprovincialis* to Saldhana Bay was a combination of ship fouling and ballast water; these modes of transport of invasives have been responsible for 86% of marine bioinvasions in South Africa to date (Mead *et al.* 2011). After its initial introduction, *M. galloprovincialis* spread rapidly, the direction of spread dictated mainly by wind-driven current directions, aided on the south coast by mariculture (McQuaid and Phillips 2000, Branch and Steffani 2004, Robinson *et al.* 2005). However, the invasion seems to have reached a limit at East London in the Eastern Cape, which coincides with the warm temperate South Coast/subtropical East Coast biogeographic boundary (Emanuel *et al.* 1992). Currently, the distribution of *M. galloprovincialis* ranges from Luderitz in southern Namibia to East London (Zardi *et al.* 2007). Central Namibia and East London represent the southern boundaries of subtropical provinces, and Mytilid species are characteristically anti-tropical (Hilbish *et al.* 2000). This leads to the hypothesis that the distribution will remain constant, based on the idea that *M. galloprovincialis* has reached its biogeographic limits in Luderitz (on the west coast) and East London (on the east coast), with abiotic subtropical conditions preventing further spread of the species.

In other areas of South Africa, *M. galloprovincialis* has been very successful. The invasive mussel constitutes 74% of coastal mussel biomass on South Africa's west coast, where it has competitively replaced *Aulacomya ater* as the dominant mussel, and coexists with *P. perna* through partial habitat segregation on the south coast (Griffiths *et al.* 1992, Bownes and McQuaid 2006, Zardi *et al.* 2007). Primary production rates on South Africa's east coast are very low when compared to the south and west (Bustamante *et al.* 1995), but interestingly, all four of the dominant south African intertidal mussel species (*A. ater, Choromytilus meridionalis, M. galloprovincialis,* and *P. perna*) show surprisingly high positive energy balances even at very low food concentrations (1 mg l^{-1}; Van Erkom Schurink and Griffiths 1992). Positive energy balances, and thus scope for mussel growth, only marginally increase with increasing food particle concentrations (Van Erkom Schurink and Griffiths 1992). Both *M. galloprovincialis* and *P. perna* show increases in scope for growth with increasing temperature and can maintain growth with increasing aerial exposure, but *P. perna* displays rapid increases with exposure temperature and clearly performs best at high temperatures (Van Erkom Schurink and Griffiths 1992, 1993). This leads to predictions that at temperatures above 20° C, *P. perna* is capable of outperforming *M. galloprovincialis* (Van Erkom Schurink and Griffiths 1992) and we have seen that under field conditions, this is true for the eastern lineage of *P perna*.

Reduced attachment strength combined with a broad shell shape makes *M. galloprovincialis* vulnerable to dislodgement from the hydrodynamic forces characteristic of the low intertidal (Rius and McQuaid 2006, Zardi *et al.* 2006). Increased stress from wave action in the low zone combined with interspecific competition with *P. perna* excludes *M. galloprovincialis* from the low intertidal (Bownes and McQuaid 2006). In the high zone of the intertidal, both *P. perna* and *M. galloprovincialis* are somewhat limited by recruitment (Porri *et al.* 2007). However, the decreased hydrodynamic stress and increasing desiccation rates in this zone favor *M. galloprovincialis*, as the invasive is much more tolerant to air exposure than any indigenous intertidal mussel species (Branch and Steffani 2004, Rius *et al.* 2006, Rius and McQuaid 2009). This helps to explain the partial habitat segregation seen in the two species on the south coast of South Africa where *P. perna* dominates the low zone, *M. galloprovincialis* dominates the high zone, and the two species coexist in the mid zone (Bownes and McQuaid 2006, Rius *et al.* 2006, Zardi *et al.* 2007). As in other areas of the coast, *P. perna* does not occupy the high zone in the eastern region due to increased desiccation rates. This leaves available habitat for *M. galloprovincialis* in the high mussel zone of the East, yet the invasive mussel has not yet been able to successfully spread to or establish itself there. This leads to the hypothesis that physical conditions of the subtropical region hinder the spread of the invasive species.

There are differences between the west, south, and east coasts of South Africa in the types and numbers of invasive species that have become established. For example, within South Africa, the subtropical biogeographical region (i.e. east of East London) is second only to the west coast in the number of invasive species found there. There are also differences in the origins of the invasive species found on the different coasts of the country. Temperate species from the northern hemisphere have been most successful on the west to southeast coasts, whereas southern hemisphere species have established more on the east to northeast coasts, strongly implicating environmental control of species distributions (Mead *et al.* 2011). *M. galloprovincialis* is a temperate species originating from the northern hemisphere that has established itself from the west to southeast coasts, in line with the previous observation. This leads to the hypothesis that physical conditions, namely high temperatures in the high intertidal zone, are the reason that *M. galloprovincialis* has not been successful on the east coast. The aim of this chapter was to test the prediction that *M. galloprovincialis* could not survive the subtropical abiotic conditions across the biogeographical break at East London.

Study Sites

In order to determine the effects of abiotic factors on the survival of *M. galloprovincialis* on the east coast of South Africa, a transplant experiment was conducted at two sites to the east of East London, where *M. galloprovincialis* occurs only sporadically and in very low numbers. This experiment was conducted in the high mussel zone as previous studies have shown that *M. galloprovinicialis* tends to dominate this zone on the South African coastline (Bownes and McQuaid 2006, Bownes *et al.* 2008, Rius *et al.* 2006, Zardi *et al.* 2007). These two sites were Haga Haga (32°45'59.32"S, 28°14'35.72"E) and Kei Mouth (Kei Mouth A [32°42'33.35"S , 28°21'50.70"E]).

Experimental Design

Due to the partial habitat segregation seen in the intertidal distributions of *M. galloprovincialis* and *P. perna* on the South African coast, it was important to examine the effects of zone on species survival. This experiment was designed to complement the competition experiment, with the goal of assessing the role of abiotic factors in the subtropical region on the distribution of *M. galloprovincialis.* The intertidal zone was divided vertically into three mussel zones in accordance with procedures adapted from previous intertidal studies (see Rius 2004) and the criteria used to identify the different zones are described in detail in the previous chapter.

The experimental design involved two treatments replicated four times in the high mussel zone at each of two sites. The treatments consisted of *M. galloprovincialis* at two different densities (20, 40) to account for intraspecific competition. This design resulted in 16 quadrats and 480 *M. galloprovincialis* individuals. Small mussels between 20 and 30 mm were collected from Plettenberg Bay and the experimental set up was placed on the shore in March 2010. Following collection, mussels were treated as described in the previous chapter. Individual mussels were tagged for growth and separated into treatment bags for deployment in the field.

Field Component

Transplant

Metal quadrats were prepared and set up as in Chapter 2. After eight months, the lids and mesh were completely removed and the experiment continued with the quadrats uncovered for two additional months for a total run time of 10 months. The aim was to have this transplant experiment run for 11 months as for the competition experiment (see Chapter 2), but the experiment was vandalized at Haga Haga after the January monthly monitoring and, following that, the last month of data could not be collected. As a result, all data on mussel growth and condition were lost from that site.

Maximum wave force

Dynamometers (Bell and Denny 1994) were deployed to measure maximum wave force simultaneously at both sites (Kei Mouth A, Haga Haga) in November 2010, December 2010, and January 2011. Prior to deployment, each device was individually calibrated using several known masses (2, 3, 4 and 5 kg), which were then multiplied by the force of acceleration due to gravity (9.81 m/s^2) to convert the recorded values from mass (kg) into force (N). Five replicates were placed in each intertidal zone (low, mid, high; N=30). The devices were attached to the shore using stainless steel eye-bolts and collected after 24 hours.

Data Analysis

Survivorship data from the transplant experiment was collected in January 2011 and analyzed using a two-way-ANOVA with two fixed factors (site, treatment).

The goal of the dynamometers was to assess whether wave forces were significantly different between the two eastern sites. Since these sites and shore levels were based on the experimental design from the competition experiment (Chapter 2), and the same eye bolts were used to deploy the dynamometers every month, the dynamometer data were analyzed using a repeated-measures PERMANOVA for the effect of three fixed factors (site, shore height, and time) on maximal wave force. Data were used to generate a resemblance matrix utilizing the D1 Euclidean distance before analysis, and the number of permutations was set to 9999.

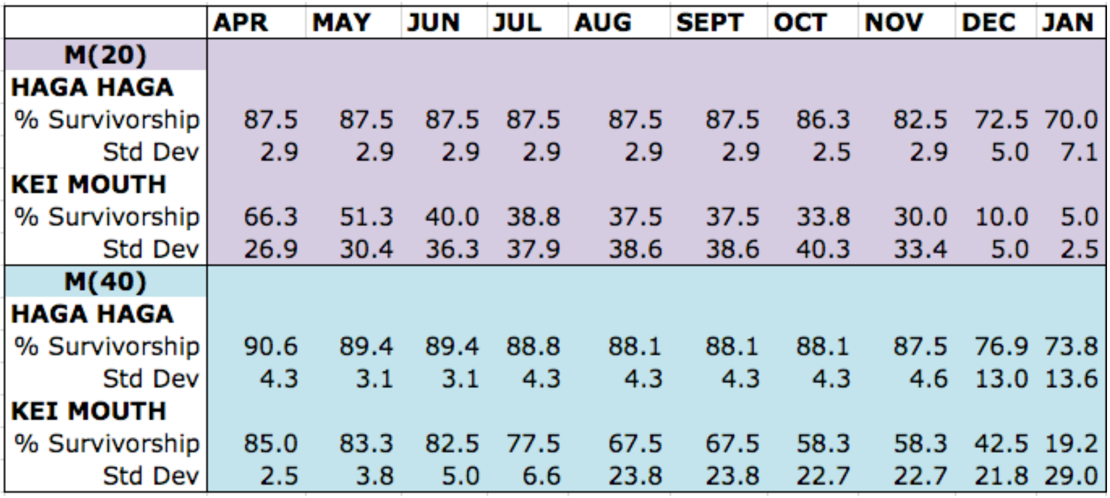
Results

Maximum wave force

There were significant effects of site ($P < 0.001$) and time ($P < 0.0001$), and a significant interaction of site and shore height ($P = 0.04$). Pair-wise tests for the interaction of site and shore height revealed significant differences between maximum wave force at Kei Mouth and Haga Haga for the low ($P < 0.0001$), and high ($P = 0.0079$), but not the mid intertidal zones ($P = 0.9939$). A storm in December 2010, drastically increased maximum wave forces (Figure 3.1) and wave-related mortality (Figure 3.2, Figure 3.3) and drove the significant effect of time ($P < 0.0001$). These results reveal significant differences between the two sites in the maximum wave forces in the high and low intertidal zones. The results also confirm that the transplant experiment, which was conducted in the high zone, experienced significantly higher wave forces at Kei Mouth than at Haga Haga.

Transplant

The transplant experiment demonstrated that *M. galloprovincialis* adults can survive the abiotic conditions across the biogeographical break at East London (Table 3.1). The survivorship rates diverged greatly between sites (Table 3.1) with site having a significant effect on survivorship ($P < 0.0001$, Table 3.2) and explaining most of the variation in the survivorship data ($\omega^2=0.82$, Table 3.2). No significant effect of Treatment and no significant interactions were detected. The large storm that occurred between November and December disproportionately affected the cages at Kei Mouth, resulting in wave-induced mussel mortality (Figure 3.2, Figure 3.3). At Kei mouth, the storm drove survivorship in the M(20) treatment down 25% (survivorship fell from 30% to 5%), and 38% for the M (40) treatment (survivorship fell from 58% to 20%). In comparison, losses at Haga Haga from the same storm were 12% for M(20) and 14% M(40).



	APR	MAY	JUN	JUL	AUG	SEPT	OCT	NOV	DEC	JAN
M(20)										
HAGA HAGA										
% Survivorship	87.5	87.5	87.5	87.5	87.5	87.5	86.3	82.5	72.5	70.0
Std Dev	2.9	2.9	2.9	2.9	2.9	2.9	2.5	2.9	5.0	7.1
KEI MOUTH										
% Survivorship	66.3	51.3	40.0	38.8	37.5	37.5	33.8	30.0	10.0	5.0
Std Dev	26.9	30.4	36.3	37.9	38.6	38.6	40.3	33.4	5.0	2.5
M(40)										
HAGA HAGA										
% Survivorship	90.6	89.4	89.4	88.8	88.1	88.1	88.1	87.5	76.9	73.8
Std Dev	4.3	3.1	3.1	4.3	4.3	4.3	4.3	4.6	13.0	13.6
KEI MOUTH										
% Survivorship	85.0	83.3	82.5	77.5	67.5	67.5	58.3	58.3	42.5	19.2
Std Dev	2.5	3.8	5.0	6.6	23.8	23.8	22.7	22.7	21.8	29.0

Table 3.1. Monthly mean survivorship and standard deviation by Site and Treatment.

Source	**Df**	**SS**	**MS**	**Pseudo-F**	**P (perm)**	**Unique perms**	**P (MC)**
Site	1	364.46	364.46	11.436	0.001	9846	0.0009
Shore height	2	96.068	48.034	1.5072	0.2292	9951	0.2266
Time	2	10940	5470.1	171.64	0.0001	9946	0.0001
Site X Shore height	2	202.92	101.46	3.1837	0.0396	9943	0.0479
Res	82	2613.3	31.869				
Total	89	14217					

Table 3.2. PERMANOVA results table. *Results from PERMANOVA comparison of maximum wave force at Haga Haga and Kei Mouth.*

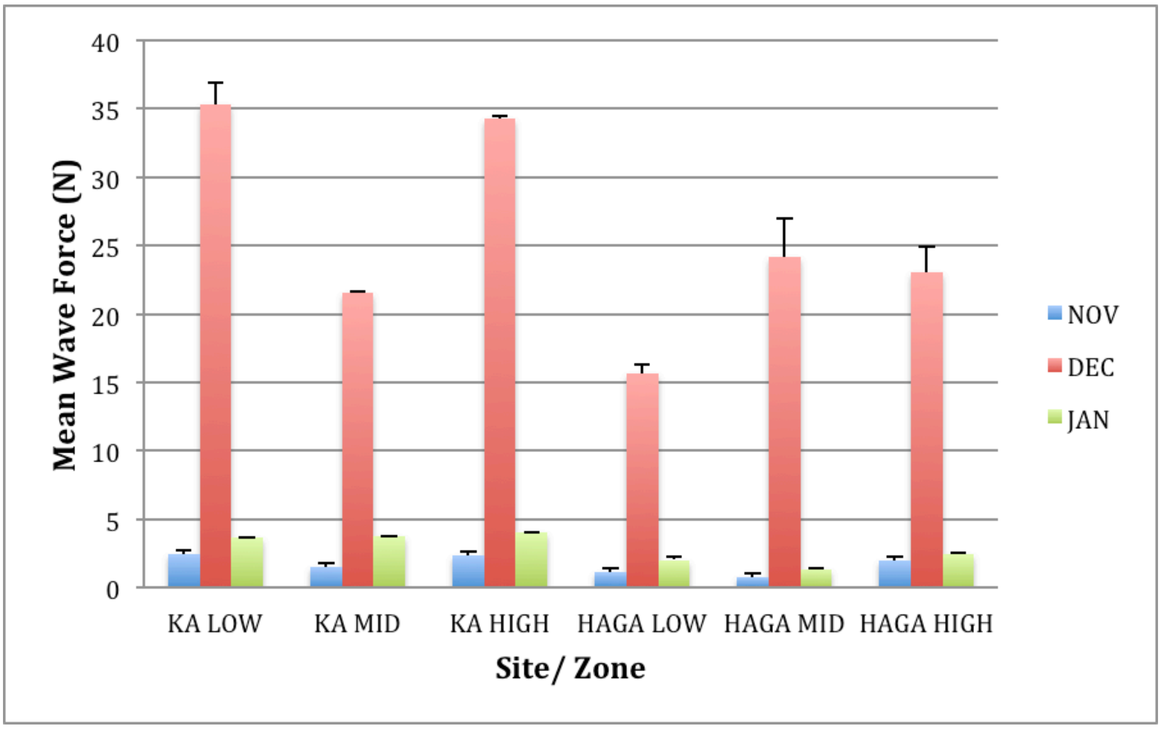


Figure 3.1. Maximum wave force by site, zone and month. *Mean (+St Dev, n=30) 'snap shot' maximum wave force (N) for November 2010, December 2010, and January 2011.*

Source of Variation	SS	d.f.	MS	F	p-level	F crit	Omega Sqr.
Treatment	284	1	285	1.28	0.28086	7.19	0.00312
Site	16,416	1	16416	73.51	1.83693E-06	7.19	0.82231
Treatment x Site	88	1	88	0.39	0.54217	7.19	0.E+0
Within Groups	2,680	12	223				
Total	19,468	15	1298				
Omega sqr- combined effect	1						

Table 3.3: ANOVA results. *Results table from the two-way-ANOVA on the M. galloprovincialis high-zone transplant survivorship data. Omega squared is an unbiased statistic to describe the % variance explained.*

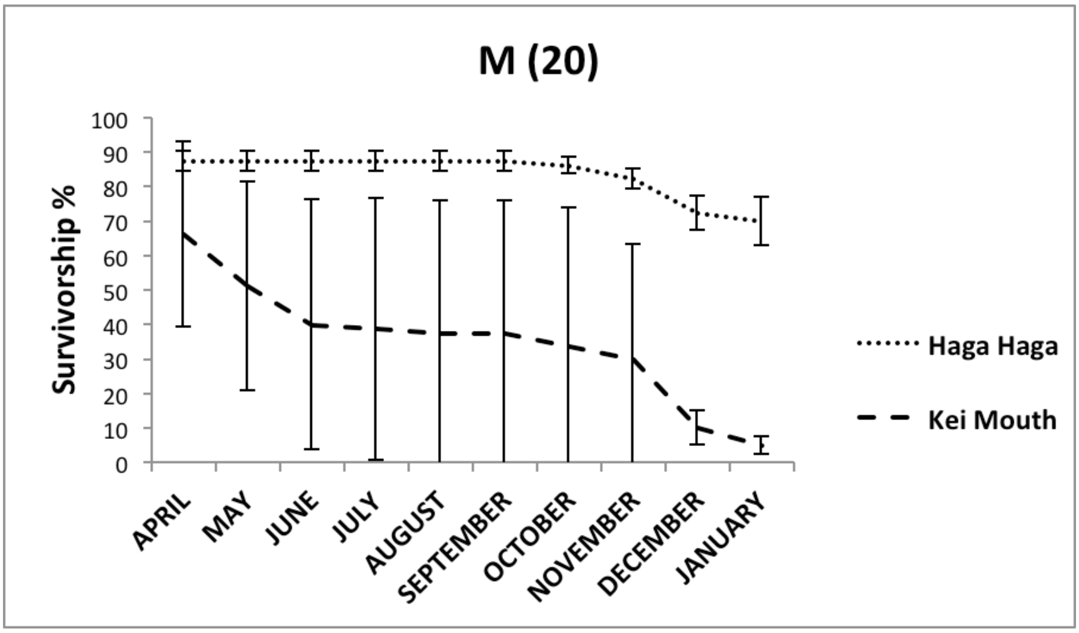


Figure 3.2. Mean survivorship of M(20) transplant treatments by month. *Error bars represent 1 standard deviation from the mean. A storm occurred between November and December. Subsequent wave-induced mortality was higher at Kei Mouth, as that site is significantly more exposed and experiences higher maximal wave force.*

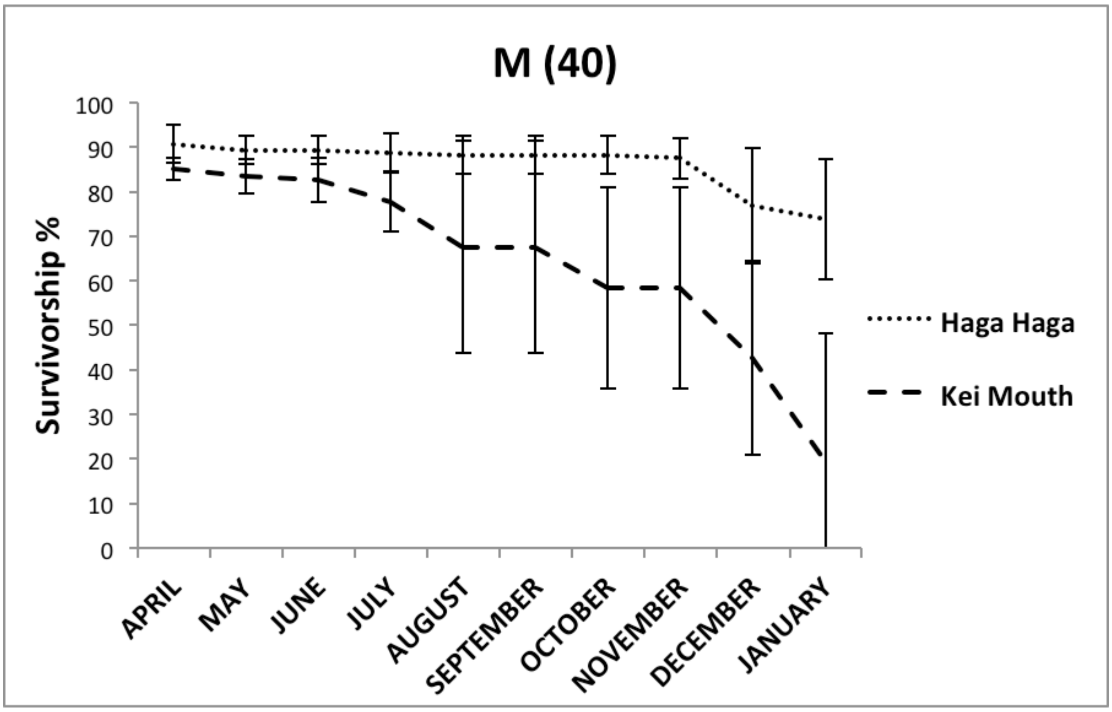


Figure 3.3.Mean survivorship of M(40) transplant treatments by month. *Error bars represent 1 standard deviation from the mean. A storm occurred between November and December. Subsequent wave-induced mortality was higher at Kei Mouth, as that site is significantly more exposed and experiences higher maximal wave force.*

Discussion

The goal of this transplant experiment was to assess if *M. galloprovincialis* can survive environmental conditions in the Eastern region. Due to logistical constraints associated with the already large number of mussels in this project (>13,000), I could not repeat this transplant experiment in the high zone to the west of the biogeographical break at East London, which left many statistical limitations in the data. Prior evidence has however shown that *M. galloprovincialis* successfully inhabits the high zone in the western region (Bownes and McQuaid 2006, Rius *et al.* 2006, Zardi *et al.* 2007). The results of this experiment show that it is possible for *M. galloprovincialis* to survive some abiotic conditions, but not severe wave action in the eastern region (See Table 3.1 though, where mortality was high at one site because of wave action). Although the survivorship rates differed greatly between sites, this can be explained by the wave-exposed nature of Kei Mouth vs. the protected nature of Haga Haga. The dynamometer data revealed that the wave action between sites was significantly different ($p<1.83E-6$), driving the Site effect (Table 3.3, Figure 3.1). Due to several factors, including shell shape and the energetic demands of reproduction, *M. galloprovincialis* is vulnerable to dislodgement by wave action (Rius and McQuaid 2006, Zardi *et al.* 2006, 2007). During the December mortality monitoring, patches of byssal threads were found in the transplant quadrats, consistent with dislodgement by waves. *M. galloprovincialis* has fewer, thinner byssal threads than *P. perna*, and is naturally subjected to higher wave forces than *P. perna* due to a wider shell shape (Zardi *et al.* 2006). While *P. perna* can cope with higher wave forces on the open coast without sacrificing reproductive output, *M. galloprovincialis* cannot (Zardi *et al.* 2007, Nicastro *et al.* 2010). Populations of *M. galloprovincialis* located near Cape Town were shown to spawn once in summer (October to February) and once in winter (April to July) (Van Erkom Schurink and Griffiths 1991); spawning cycles however vary by site/population and attachment strength is negatively correlated with sea surface temperature (Van Erkom Schurink and Griffiths 1991, Zardi *et al.* 2007). Thus, further exploration into the spawning habits of mussel populations on the east coast is required at this point, especially in light of the genetic structure of *P.*

perna on this coast and the potential for lineage specific energy allocation, reproductive cycles, and attachment strengths. The December storm may have hit the experiment just as the mussels were spawning, at a time when attachment strength was weakest. Attachment strength and gametogenesis are highly demanding and conflicting energetic processes; high gamete production in *M. galloprovincialis* paradoxically allows the invader to colonize and prevents colonization under exposed conditions (Zardi et al. 2007). Both species are less abundant on the open coast in comparison to protected bays, but the bay effect is stronger for *M. galloprovincialis* as a result of its greater vulnerability to wave forces (von der Meden *et al.* 2008). The sheltering effect of bays becomes clear when the two sites are compared. The dynamometers recorded significantly higher maximal wave forces for Kei Mouth than for Haga Haga across the sampling period (Table 3.3, Figure 3.2), which explains the poor performance of the transplant treatments at the exposed site of Kei Mouth. These results echo previous studies, confirming that *M. galloprovincialis* is a weaker competitor in wave exposed environments (von der Meden *et al*. 2008, Nicastro *et al.* 2010). Nevertheless, *M. galloprovincialis* demonstrated impressive survivorship rates at the more protected site of Haga Haga, indicating that abiotic conditions, such as desiccation (Zardi *et al.* 2007) in the east do not have dramatic negative effects on adult survivorship. These results lead to the confident conclusion that increased desiccation in the east does not affect adult survival. Significantly higher temperatures typify the Eastern region (Zardi *et al.* 2007), but *M. galloprovincialis* is characteristically robust to desiccation (Rius *et al.* 2006) and the results from this chapter show that adults can cope with such abiotic conditions.

It is possible that the eastern lineage of *P. perna* can exclude *M. galloprovincialis* from the Eastern region simply because it is a more effective competitor than its western conspecific. However, the biphasic life history of mussels presents a special problem to the analysis of these results, as the larval phase may possess a distinct suite of ecological and physiological requirements and experience different selective pressures from the adult phase, and this in turn may affect adult population dynamics (Grosberg and Levitan 1992, Richmond and Woodin 1996, Pechenik *et al.* 1998, see Chapter 4 for details).

Therefore, although we can decisively say that abiotic conditions in the east are not insurmountable for *M. galloprovincialis* adults, we cannot extrapolate these results to larvae and, most importantly, recruits. Perhaps abiotic stress in the east weakens the larvae, resulting in less-fit adults incapable of competing with the *P. perna* East lineage in the mid-intertidal, or conceivably, incapable of tolerating abiotic conditions. *M. galloprovincialis* is recruitment limited in the high-intertidal zone (Porri *et al.* 2007), so the inability to compete with *P. perna* East and invade the mid-zone in the east may prove critical for species distribution. Perhaps the invasive species is supply, settlement and/or recruitment limited in the eastern region. More information on the subtropical abiotic effects on early stage survivorship, condition, and metamorphosis in *M. galloprovincialis* is needed in order to adequately and generally assess the effects of physical conditions on overall species distribution.

Chapter 4: Assessment of Supply-side ecology as a Driver of *Mytilus galloprovincialis* Distribution in South Africa

Introduction

Marine and intertidal organisms employ a variety of reproductive strategies that result in varying degrees of dispersal potential (Vance 1973, Palmer *et al.* 1996, Grantham *et al.* 2003). Most sessile and sedentary intertidal animals employ a planktotrophic phase for which long-distance dispersal is possible but largely passive (Strathmann 1974, Palmer *et al.* 1996). The high dispersal capabilities of planktotrophic larvae suggest intertidal assemblages are demographically open and under regional rather than local control (Palmer *et al.* 1996). Evidence concerning oceanographic transport mechanisms and larval behaviour indicates high local retention of larvae in the inshore, resulting in communities being demographically more closed than previously assumed (Swearer *et al.* 1999, Cowen *et al.* 2000, Levin 2006). In either case, the rate of larval supply for marine animals is not continuous, which has consequences for recruitment in community assemblages (Underwood and Fairweather 1989). The inherent variability in larval transport processes has been shown to affect the intensity of intertidal community interactions in space and time as gradients in larval supply produce gradients in the interaction strength of competitive and predator-prey interactions (Connolly and Roughgarden 1999). Supply-side ecology must therefore be considered when examining forces that structure intertidal communities.

The supply of planktonic larvae of intertidal species influences adult populations (Connell 1985, Underwood and Fairweather 1989, Palmer *et al.* 1996) and has been suggested as a contributing factor of the geographic distribution of mussels in South Africa. There is a phylogeographic break in the native species *Perna perna* between the subtropical East Coast and the warm temperate South Coast of South Africa (Zardi *et al.* 2007). It has been hypothesized that the powerful, southwesterly flowing Agulhas Current helps to maintain the divergence of the two lineages of *P. perna* by advecting larvae of the eastern lineage away from the coast so they cannot colonize sites farther west (Zardi *et al.* 2007, 2011). It follows that the Agulhas Current may contribute to the distribution of *Mytilus galloprovincialis* by limiting its eastwards extension through the same mechanism. Powerful currents and other oceanographic features have the capacity to restrict species ranges, even when suitable habitat is abundant outside that range (Gaylord and Gaines 2000). This would appear to describe the distribution for *M. galloprovincialis,* with the Agulhas Current preventing the easterly spread of the invasive species despite readily available rocky-shore habitat on the east coast of South Africa.

There is a strong biogeographical break in intertidal communities on the South African coast (Emanuel *et al.* 1992) that coincides with the eastern limit of the range of *M. galloprovincialis*. Intertidal air temperatures across this break are significantly higher than in the region where *M. galloprovincialis* currently exists (Zardi *et al.* 2011). *M. galloprovincialis* is nevertheless characteristically resistant to desiccation (Rius *et al.* 2006) and previous results from Chapter 3 have shown that adults of *M. galloprovincialis* can survive environmental conditions in the Eastern region (see Chapter 3 for details), although information on condition was lost. *M. galloprovincialis* does not occur in beds on the east coast of South Africa (Robinson *et al.* 2005, Zardi *et al.* 2007), but *M. galloprovincialis* individuals have been found in *P. perna* beds between Dwesa Nature reserve and Kei Mouth (B. Mostert, A. Ludford, pers. comm.; M. Hall pers. obs.) and *M. galloprovincialis* settlers have been discovered in the Eastern region, beyond where adults have been found (unpub. data). This would imply that *M. galloprovincialis* is not excluded from the east coast of South Africa by the inability of larvae to reach that coast, but by other biological processes, among which interspecific competition is a strong candidate.

The patterns of dispersal of *M. galloprovincialis* have been shown to be predictable on the basis of hydrographic data; the dispersal of mussel larvae is highly passive and larval dispersal ranges are relatively limited in the nearshore region (McQuaid and Phillips 2000, Shanks *et al.* 2003, Shanks and Brink 2005, Morgan *et al.* 2009; Porri et al 2014). Studies on larval supply and delivery show significant effects of time and space for larvae and settlers (Porri *et al.* 2006). Additionally, Harris *et al.* (1998) found highly significant differences between localities and between sites only a few kilometers apart, indicating strong influences of larval dispersal on very small scales. It is difficult to ignore the possibility that dispersal limits the range of *M. galloprovincialis* on the south coast of South Africa, preventing it from colonizing the east coast. However, the role that larval dispersal plays in the distribution of *M. galloprovincialis* on the east coast of South Africa has not been explored. The aim of this chapter was to test the prediction that larval transport drives the distribution of adults of *M. galloprovincialis* in South Africa. Absence of *M. galloprovincialis* larvae from the water column in inshore waters east of East London (the subtropical East Coast province) would support this hypothesis (although multiple explanations would be possible), whereas the presence of *M. galloprovincialis* larvae in those waters would reject it.

Material and Methods

Sample Collection

Larval samples were collected in September- October 2009 from the FRS *Algoa*. Sampling was performed from Haga Haga north to the Mozambican border, a stretch of coastline outside the range of *M. galloprovincialis* beds (Zardi *et al.* 2007, Figure 2.1). Sampling was performed as close to the surf zone as possible to maximize the potential of sampling competent larvae. The samples processed for this chapter were collected at two sites to the east of Haga Haga ([30°46’ 63” S, 28°15’58”E], [32°29’76” S 28°40’1”E]). A submersible 2.2 KC Denmark 23.580 plankton pump with a net of 60 µm mesh was used to sample surface water (<2 m) at a rate of approximately 200L/min for 10 min at each site. The mesh of the plankton pump was then rinsed with seawater into a sieve to collect the sample. Collected samples were then rinsed into jars with 70% ethanol and stored for processing.

Species Identification

Mussel larvae were isolated from other animals and debris in the samples using a dissecting stereomicroscope. Mytilid larvae were then grouped according to shell morphology. Potential *M. galloprovincialis* larvae were soaked in a 5-10%

hypochloric acid solution to dissolve tissue and aid the disarticulation of valves. Once completed, disarticulated valves were rinsed with distilled water and prepared for scanning electron microscopy (SEM). Final species identification was based on hinge morphology (Table 1), following Siddall (1980) and Bownes *et al.* (2010). The latter study explored morphological characteristics of post-larvae whereas this study sampled larvae, however, the methodology was adopted for this study as sizes of the larvae and post-larvae were comparable.

	Shell morphology	Hinge morphology
Perna perna	• Rounded, central umbo (d) • Red mark on umbo (d) • Reddish hinge	• Presence of primary lateral teeth (d) • 22-24 provincular teeth (d)
Mytilus galloprovincialis	• Wide, flat umbo towards dorsal margin • Ventral shell margin asymmetrically elongated	• 30-36 provincular teeth
Choromytilus meridionalis	• Wider, rounder umbo • Bluish hinge • Ventral shell margin less elongate • High height:length ratio	• Early development of secondary lateral and dysodont teeth (350-400 µm) • 28-32 provincular teeth

Table 4.1: Morphological characters used for species identification. *From Siddall (1980) and Bownes et al. (2010). (d) represents a diagnostic character.*

Results

Larvae of *M. galloprovincialis* were positively identified from samples, but they were rare at a density of 1 *M. galloprovincialis* larvae per 902.8 L seawater sampled (examples shown in Figure 4.2). A total of 16 larvae were examined as potential *M. galloprovincialis* larvae, all were between 200-450 µm. Of the 16 potential *M. galloprovincialis* larvae, only five were positively identified. These larvae are certainly not *P. perna* due to the absence of diagnostic shell characters

(Bownes *et al.* 2010 Table 4.1). Additionally, no hinge pigmentation was observed in any of these larvae, which is consistent with post-larval *M. galloprovincialis* individuals, but not with *P. perna* (Bownes *et al.* 2010). *P. perna* individuals from the same samples were positively identified, confirming the effectiveness of the methodology in sampling intertidal species (also see Porri et al. 2014 for large numbers of larvae of both species collected on the western side of Cape Agulhas using the same methodology).

SEM results revealed the possible *M. galloprovincialis* larvae were missing lateral hinge teeth (Figure 4.2). The presence of lateral hinge teeth is a reliable characteristic used in identification that is unique to *P. perna* larvae (Siddall 1980). Distinguishing between post-larval individuals of *M. galloprovincialis* and *Choromytilus meridionalis* is more difficult due to the lack of diagnostic shell characteristics in post larvae of either species. Larval and early post-larval stages of the two species are especially hard to differentiate as neither species possess distinguishing colour markings and the hinge morphology is similar (Bownes *et al.* 2010). However, the highly asymmetrical growth in *M. galloprovincialis* contrasts with the straight dissoconch growth in *C. meridionalis*, making the two species distinguishable by shell morphology (Bownes *et al.* 2010). The longest distance in *C. meridionalis* is usually a straight line from umbo to tip resulting in a higher height to length ratio, whereas elongation on the ventral shell margin is much more pronounced in *M. galloprovincialis* (Bownes *et al.* 2010). It is, however, unlikely any larvae of the sampled would be *C. meridionalis* as this species is rare east of Cape Agulhas, and most abundant on the highly productive west coast (Van Erkom Schurink and Griffiths 1990).

The hinge of mytlids consists of small denticles in a narrow, central provinculum flanked on both sides by large denticles, about 30-36 provincular teeth in total (Semenikhina *et al.* 2008, Bownes *et al.* 2010). The shape and size of the provinculum and the number of teeth is similar in *C. meridionalis* and *M. galloprovincialis.* However, *C. meridionalis* develops secondary lateral and dysodont teeth earlier, at a smaller size (350-400 µm) than *P. perna* and *M. galloprovincialis* (450-500 µm) (Bownes *et al.* 2010). While the small denticles in the inner provinculum cannot be counted accurately in some cases (Figure 4.2, all panels), the potential *M. galloprovincialis* larvae appear to possess the correct number of provincular teeth. These larval shells also lacked primary lateral, secondary lateral and dysodont teeth, which is appropriate for *M. galloprovincialis* larvae of this size, but not *P. perna* or *C. meridionalis*. This confirms the presence of *M. galloprovincialis* larvae in the waters off the east coast of South Africa.

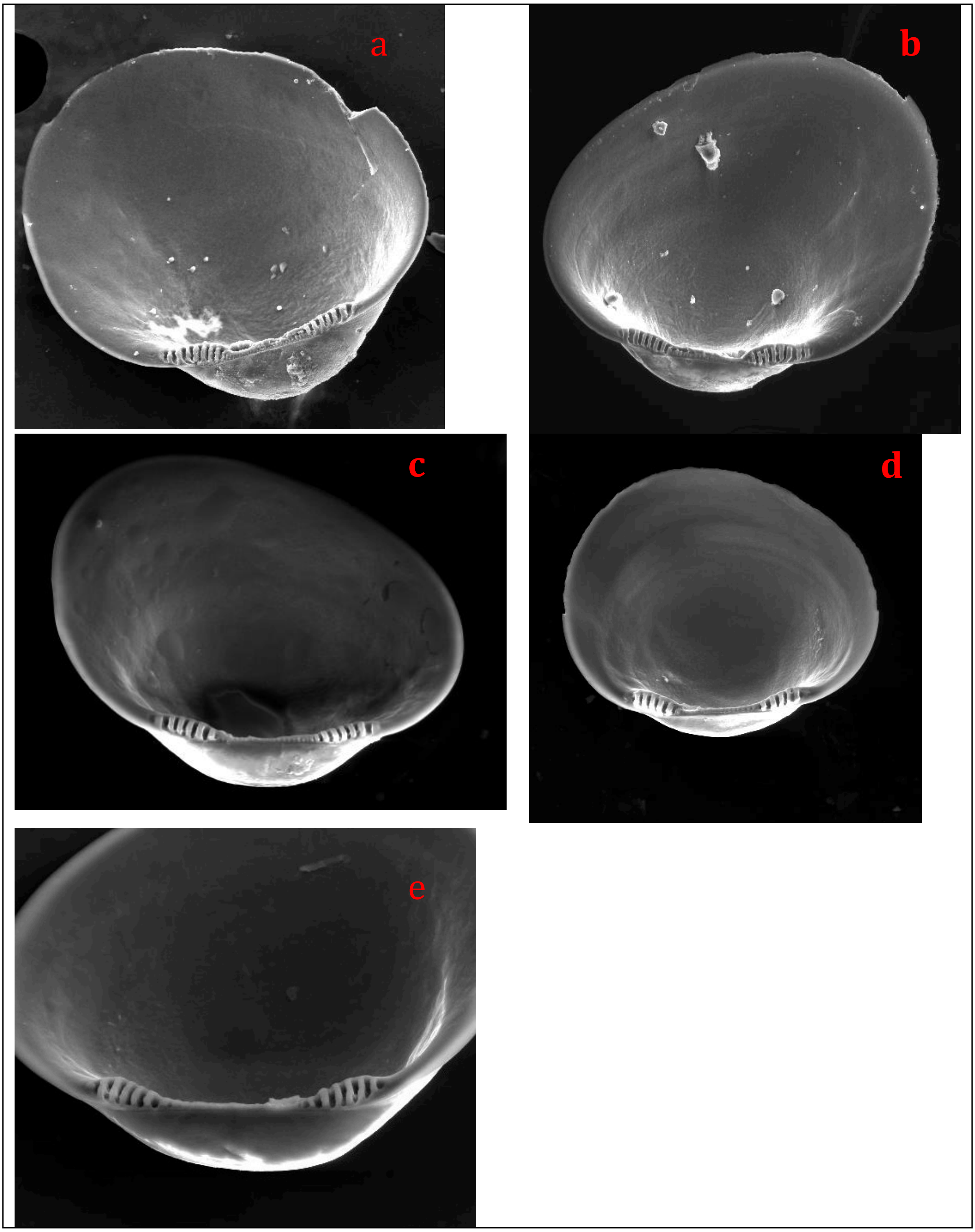


Figure 4.2: SEM results confirmed positive species identification of *M. galloprovincialis* larvae. Panels (a-c) show larvae larger than 300 μm. Panel (d) shows a larva <300 μm, Panel (e) shows a closer view of the larval hinge morphology of *M. galloprovincialis*

Discussion

The presence of *M. galloprovincialis* larvae east of the biogeographical break at East London suggests that larval supply is not entirely responsible for the absence of *M. galloprovincialis* from the east coast of South Africa. The hypothesis that supply-side ecology drives the distribution of adults of *M. galloprovincialis* in South Africa was therefore tentatively rejected. While the dynamics of the Agulhas Current likely play a role in the genetic structure of *P. perna* (Zardi *et al.* 2011), it is hard to reconcile the theory that larval supply is a decisive factor with the fact that the *P. perna* western lineage occurs on either side of a 1000 km break on the west coast.

The results of this chapter confirm that *M. galloprovinicialis* larvae are capable of reaching the east coast of South Africa against the main flow of the Agulhas Current, confirming that larval supply is possible. Previous studies on spawning cycles of *M. galloprovincialis* on the southern coast showed that spawning generally occurs once in summer (September/October- February) and once in winter (April/May- July) (Van Erkom Schurink and Griffiths 1991); however, information on east coast populations is lacking and more research is needed into specific spawning cycles there. Spawning cycles of *M. galloprovincialis* populations near Cape Town revealed summer spawning bursts from September to December, depending on the population (Van Erkom Schurink and Griffiths 1991). Therefore, while south coast populations may in fact differ in temporal spawning cycles from previously measured populations, it is completely within the realm of possibility that I could detect *M. galloprovincialis* larvae in eastern waters from September to October if larvae from other areas of the coast can overcome the main westward flow of the Agulhas current. This chapter of my thesis focused purely on presence/absence information of *M. galloprovincialis* larvae in east coast waters, however, future work should focus on specific spawning cycles of both eastern and western lineages of *P. perna*, and of *M. galloprovincialis*, on the east coast of South Africa.

The presence of *M. galloprovincialis* larvae in these samples means that the larvae may be exploiting northeastwardly flowing countercurrents inshore of the Agulhas driven by wind or the periodic Natal Pulse (McQuaid and Phillips

2000, Roberts *et al.* 2010). Still, *M. galloprovincialis* adults are not found in the east in great numbers. Perhaps the larvae are missing chemical cues from conspecifics needed to settle (Crisp and Meadows 1962, von der Meden *et al.* 2010), as there are no *M. galloprovincialis* beds on this stretch of shoreline. However, individual adults have been spotted in *P. perna* beds, albeit rarely, and settlers have been detected in Coffee Bay (unpub. data), which is beyond the biogeographic break on the east coast. Nevertheless, my initial process-driven criterion for rejection of the hypothesis that supply side dynamics are driving the distribution of *M. galloprovincialis* is, in fact, not adequate. The presence of larvae alone does not explain the supply side ecology of a species, and other physical factors such as current speed and direction as well as larval density need to be taken into account.

While settlement is indeed a function of larval supply (Minchinton and Scheibling 1991), adequate supply does not equate to recruitment or couple settlement and recruitment (Pineda *et al*. 2010). Studies on larval supply and delivery show significant effects of time and space for larvae and settlers; larval supply seems to be highly variable with location exercising a strong influence on settler dynamics (Porri *et al.* 2006). Harris *et al.* (1998) found highly significant differences between sites only a few kilometers apart, and the same has been shown for sites separated by 100s m (Porri et al. 2008), indicating strong influences of larval dispersal on very small scales. Furthermore, if the larvae in eastern waters were to successfully recruit, that would not guarantee their survival to adulthood (Hunt *et al.* 1997). While some of larval supply is evident, I do not have information on larval delivery *per se*, as it would be possible for larvae to be detected in the water column without successful delivery to the shore. Moreover, I do not have information on the larval density in the water column, or the densities at which larvae arrive to settle at the intertidal and this is likely to be variable in space and time. In fact, studies have shown that invertebrate stock and recruit density may be more closely tied and supply distances may be much shorter than previously assumed (Harris *et al.* 1998; Shanks et al 2003). The lack of *M. galloprovincialis* adults on the east coast may therefore further limit the ability for *M. galloprovincialis* larvae to settle on eastern shores through pre-settlement mechanisms. Evidence from *P. perna* in the east suggest that regions with low adult supply experience recruitment limitation (Harris *et al.* 1998, Reaugh *et al.* 2007). There may also be a density-dependent effect where larvae from other regions are successfully transported to eastern waters but not in adequate numbers due to the predominance of oceanographic transport mechanisms that carry larvae away from the coast. The transport of larvae will be highly dependent on the interaction of oceanographic mechanisms, which are highly dynamic and region-specific in South African waters (Porri *et al.*, 2014), as well as larval behaviour and physiology. Temporal cycles of recruitment seem to be irregular and episodic, which may have repercussions for adult mussel dynamics (Harris *et al.* 1998). In the East, the periodic Natal Pulse (Roberts *et al.* 2010) can sweep larvae offshore into the Agulhas meander (Porri *et al.* 2014), limiting the arrival of larvae to eastern shores. If the Natal Pulse were to happen concurrently with spawning events, there would be a massive loss of larvae to offshore waters. The Natal Pulse could intermittently allow some larvae to be pushed northeasterly up the coast of Africa, but the frequency of these events and the subsequent larval delivery and densities need to be explored.

Further, the stress experienced by *M. galloprovincialis* larvae in eastern waters has yet to be quantified. Larval life-history stages can be especially susceptible to environmental stresses (Richmond and Woodin 1996) and stress experienced during this larval phase can negatively impact larval quality and adult performance (Pechenik *et al.* 1998). In fact, previous empirical energetic measurements on adult mussels have predicted that *P. perna* would outcompete *M. galloprovincialis* on the east coast where temperatures exceed 20°C (Van Erkom Schurink and Griffiths 1992). Despite the ability of *M. galloprovincialis* adults to survive abiotic conditions in the East (as the results of Chapter 3 show), the larvae could conceivably not be equally robust to conditions associated with the biogeographic break. There is some evidence that *M. galloprovincialis* individuals are capable of successfully settling in the eastern region (unpub. data). This is difficult to reconcile with the fact that mytilids are characteristically anti-tropical in distribution (Hilbish *et al.* 2000). Perhaps the larvae are behaving according to the "desperate larvae hypothesis" (Knight-Jones 1951, 1953), becoming less selective in habitat choice as environmental stresses increase in the east. The larvae in eastern waters are likely to be stressed by food scarcity, as there is a productivity gradient in both intertidal and offshore waters along the South African coast where productivity is high in the west and low in the east (Brown 1992, Bustamante *et al*. 1995). Larvae that are starved during the planktotrophic stage decrease in quality, become less selective/more desperate to settle and simultaneously less likely to survive (Elkin and Marshall 2007). Therefore, any *M. galloprovincialis* larvae that settle and survive such stresses may produce less-fit adults, incapable of outcompeting the already competitively dominant *P. perna* East lineage (Chapter 2). Because the occurrence of *M. galloprovincialis* adults is rare in the eastern region, it is likely that population-level bottleneck constraints occur at early life-stages. These constraints may stem from limited larval delivery, early stage physiological restrictions, competitively weak settlers, or a combination of the three.

The pressure of selective forces on each stage of the biphasic life history of *M. galloprovincialis* may be different in intensity. Thus, the survival of adults in the east does not provide sufficient evidence that the larvae are also capable of surviving the abiotic stress associated with the subtropical region. It is most likely that a combination of physical, abiotic, and biotic factors is actually inhibiting the eastwardly spread of *M. galloprovincialis.* Further research on the competitive biology and ecology of early stage and juvenile mussels is required.

Chapter 5: General Discussion

Niche Theory and Invasion Biology

Niche theory has been important to the development of major ideas in community ecology and invasion biology. In order for a species to invade a community there must be an opportunity to take advantage of freely available niche space, partition niche space with a native species, or make unavailable niche space available to itself by force through competition (Elton 1958, Hutchinson 1959). From these ideas Elton formed the biotic resistance hypothesis, which states that more diverse communities utilize the dimensions of a niche more effectively, decreasing freely available niche space and the potential for invasion (Elton 1958, Stachowicz *et al.* 1999). Therefore, highly diverse communities should be naturally resistant to invasion because available niche space is rare. Evidence on the hypothesis of biotic resistance seems however to differ, depending on the scale of the observations made (Stohlgren *et al.* 1999, Fridley *et al.* 2007, McQuaid and Arenas 2009). Empirical evidence shows a negative correlation between native and invasive species richness on a small scale and a positive correlation on a large scale, called the invasion paradox, where highly diverse regions are more heavily invaded (Levine and D'Antonio 1999, Shea and Chesson 2002, Fridley *et al.* 2004, Fridley *et al.* 2007). Stochastic niche theory rose in response to the invasion paradox, arguing that invasibility is low in more diverse communities because of uniformly low resources, not because of high diversity *per se*; invasibility directly depends on how native species utilize heterogeneity in limiting resources, and successful invaders will exploit underused resources (Tilman 2004). This theory fits well in intertidal dynamics, where many species rely on one key limiting resource, primary attachment space (Connell 1961, Paine 1966, Dayton 1971, Connell 1972, Sale 1974, Menge 1976, Schoener 1982, Paine 1984). Indeed, the hypothesis of biotic resistance and other concepts based in niche dynamics have been empirically demonstrated for intertidal systems (Stachowicz *et al.* 1999, Byers 2002, Hunt and Yamada 2003, Jensen *et al.* 2007). The success of niche-based theories in describing intertidal ecology is strong evidence that intertidal communities fall closest to the niche end on the continuum from niche to neutral systems. If one thinks of niche theory as focusing on the differences between species and neutral theory as focusing on similarities between species, the differences between species seem to control dynamics in intertidal communities.

This thesis sought to investigate whether niche or neutral mechanisms determined the success and distribution of an invasive mussel in South Africa, *Mytilus galloprovincialis*. The invasive species exists through partial habitat partitioning, dominating the upper mussel zone, where the native *Perna perna* cannot survive, evidence that *M. galloprovincialis* is exploiting an underused component of a limiting resource in line with stochastic niche theory. While the distribution of the invasive *M. galloprovincialis* stops at a biogeographic break at East London, underutilized primary attachment space is available on the high-shore farther east. What prevents the invasive from taking advantage of this resource east of the biogeographic break? Do niche or neutral mechanisms control the distribution of the invasive? These questions are especially intriguing considering the success of *M. galloprovincialis* on the west and south coasts of South Africa.

While neutral perspectives rely on ecological equivalency to explain community assembly, niche-based perspectives place importance on biotic interactions and niche differentiation (Chesson 2000, Hubbell 2001). The results of this thesis show that the distribution of *M. galloprovincialis* is not necessarily limited by abiotic factors (Chapter 3), instead, biotic interactions with the eastern lineage of *P. perna* seem to be important (Chapter 2). The results presented here support the idea that niche mechanisms are indeed responsible for limiting the distribution of the invasive *M. galloprovincialis* on the South African coast.

Abiotic Effects

In order to assess the effects of abiotic factors on the distribution of the invasive, I transplanted adult *M. galloprovincialis* to two sites on the eastern side of the biogeographic break. Results of this transplant experiment (Chapter 3) showed that *M. galloprovincialis* performed significantly better at the more protected site than the more exposed site, echoing previous results on the invasive's inability to withstand high levels of wave action (von der Meden *et al*. 2008, Nicastro *et al.* 2010). The high survivorship rates at the more protected site of Haga Haga demonstrate that adult *M. galloprovincialis* can survive the increased temperatures and abiotic conditions associated with the eastern side of the biogeographic break. More investigation is nevertheless required into the abiotic effects on larval and recruit survivorship, as this may be partially responsible for the absence of adults in the east. It should also be noted that there was a region-specific change in the density-dependent intraspecific dynamics of *M. galloprovincialis,* where abiotic stress negated facilitative intraspecific protection for the invasive in the east.

Results from Chapter 4 showed that *M. galloprovincialis* larvae are present in the waters east of the biogeographical break around East London, indicating potential larval supply to the eastern region. The mere presence of larvae in the water is only the initial step in the supply side ecology of an intertidal species; more information is needed on larval delivery and density. Because the occurrence of *M. galloprovincialis* adults is rare in the eastern region, it is likely that population-level constraints occur at early life-stages. These constraints may stem from limited larval delivery, competitively weak settlers, intolerance of settlers to abiotic conditions, or a combination of these factors. The invasive could potentially be supply, settlement, or recruitment limited on the eastern side of the biogeographic break. The fact that *M. galloprovincialis* adults can survive the environmental conditions in the east in a location where it is not naturally found, is in accordance with previous results indicating that adult *M. galloprovincialis* individuals are resistant to high temperatures and desiccation (Rius and McQuaid 2006, 2009)

Biotic Effects

In order to assess the effects of biotic factors on the distribution of the invasive, a competition experiment was performed. Results from this experiment (Chapter 2) showed that *P. perna* East individuals significantly outcompete the invasive on both the western and eastern side of the biogeographical break. If *P. perna* East is a superior competitor on both sides of the geographic break, what prevents it from spreading west across the barrier into the warm temperate South Coast province? This question was left unanswered by the ambiguous genetic results of Chapter 2 PE/PW comparison. In the absence of results from a competition experiment, we can look to the concepts of niche conservatism and vicariance. Niche conservatism limits the habitat breadth of a species in niche space; vicariance and subsequent local adaptation leads to novel traits, but also increases the effectiveness of geographic barriers (Mayr 1983, Garcia-Ramos and Kirpatrick 1997, Bekessey *et al*. 2003, Wiens 2004, Wiens and Graham 2005). This maintains and reinforces the separation of genetic lineages, and prevents a given species from adapting across geographic barriers and spreading into the range of a sister species (Wiens 2004). There is evidence of a geographic barrier associated with the Agulhas Current that could lead to reproductive isolation of the two lineages of *P. perna* (Zardi *et al.* 2011). There is also evidence of local adaptation to environmental conditions; significantly higher maximum temperatures characterize the eastern side of the biogeographic break and *P. perna* East has been shown to be physiologically more robust than *P. perna* West (Zardi *et al.* 2011). These two components combined can explain why *P. perna* East is a superior competitor, yet does not spread across to the western side of the biogeographical break around East London. Biotic effects, namely interspecific competition between *M. galloprovincialis* and *P. perna* East, are therefore significant within-site competitive interactions that contribute to determining the large-scale distribution of the invasive.

M. galloprovincialis individuals are sporadically present but extremely rare in the mussel beds of the East, and *M. galloprovincialis* larvae are present in the water east of the biogeographical break. These results suggest that the competitive superiority of *P. perna* East (in combination with the unexplored potential supply side limitation) is capable of explaining the large scale distribution of *M. galloprovincialis*, as this experiment found that adults are capable of surviving the abiotic conditions characteristic of the subtropical eastern region. These results are in accordance with Levine's idea of biotic containment (2004), where interactions with native species can limit, constrain, and reduce the performance of the invasive without being able to exclude it. Further investigation is required to identify the specific characteristic or characteristics of the *P. perna* East lineage that confer this competitive advantage and the effects of abiotic conditions on *M. galloprovincialis* larvae and recruits.

In addition to the interspecific interactions, the competition experiment in Chapter 2 aimed to address questions about whether the effects of intraspecific biotic interactions change with density. The results showed significant intraspecific facilitation in *M. galloprovincialis* with increasing density in the area where it naturally occurs, the western side of the biogeographic break, but not the eastern side. In other words, on a large biogeographic scale, there was an interaction of the consequences of positive effects in *M. galloprovincialis* and biogeographical region. These sorts of positive interactions are common for sessile species (Bertness and Leonard 1997) and some positive, facilitative interactions have been previously demonstrated between *M. galloprovincialis* and *P. perna*. Initially *P. perna* protects *M. galloprovincialis* beds from wave force on the low shore and *M. galloprovincialis* protects *P. perna* from desiccation on the mid shore (Rius and McQuaid 2006). The results from the current experiment show that, intraspecifically, *M. galloprovincialis* shows facilitation with increasing density, but only when abiotic stresses are within a tolerable range (i.e. in the western region). When environmental stress increases beyond this range, the effects of facilitation are overwhelmed, preventing positive density-dependent survivorship. This is in agreement with the results from a previous study on facilitation and competition between *M. galloprovincialis* and *P. perna* over a gradient of physical stress. The relationship between environmental stress and facilitation was hump-shaped; facilitation was unimportant in environmentally benign conditions, significant under intermediate levels of stress, and outweighed under conditions of high stress (Rius and McQuaid 2009). The results from the present study show that the relationship between stress and intraspecific facilitation is in line with the previously observed interspecific relationship.

Conclusion

The results from this thesis show that abiotic factors do not appear to be as important as biotic factors in determining the eastern limit to the distribution of the invasive *M. galloprovincialis* in South Africa. Romanuk and Kolasa (2005) empirically demonstrated that community invasibility in rock pools is related to both abiotic conditions (resource availability) and biotic conditions (community diversity). Their study showed that invasion success was highest when resource levels were high and diversity levels were low and stated that the presence of particular species can have an effect on invasion success through competition (Romanuk and Kolasa 2005). In South Africa, there is a gradient in intertidal primary productivity, with high primary production on the west coast that progressively declines to the south and then the east coast (Brown 1992, Bustamante *et al.* 1995). While higher levels of productivity are more likely to exhaust limiting nutrients (Tilman *et al.* 1997) limiting niche space and preventing invasion (Elton 1958, Stachowicz *et al.* 1999), upwelling on the west coast is so intense that nutrient supply is rarely limiting (Bustamante *et al.* 1995). This upwelling supports much higher biomass levels for west coast communities, despite the fact that western communities contain only half the species richness of the south or east coast communities (Bustamante and Branch 1996). The west is different from the other two biogeographic coastal regions in that it is simultaneously biomass rich and species poor, yet communities on the south and west coasts are more similar to each other than either is to the east coast. In fact, presence/absence data of intertidal species revealed that species composition in east coast communities differed from south and west coast communities by 70% (Bustamante and Branch 1996). The fact that the east coast is distinct in community composition and depleted in productivity with respect to the other two biogeographic regions helps to explain the lack of invasion success in the east. By the same token, the high resource availability coupled with species poor communities on the west coast helps to explain the success of *M. galloprovincialis* there. While resource levels and abiotic conditions undoubtedly contribute to the distribution of *M. galloprovincialis,* this thesis supports the hypothesis that competition with *P. perna* East is particularly important in keeping *M. galloprovincialis* from successfully invading South Africa's subtropical east coast province.

Interspecific biotic interactions with *P. perna* East through competition for primary attachment space appear to exclude the invasive from the east coast of South Africa. Primary attachment space is the defining limiting resource for the survival of many species in intertidal communities, and interspecific competition for this resource is critical in determining community composition (Connell 1961, Paine 1966, Dayton 1971, Connell 1972, Sale 1974, Menge 1974, Schoener 1982, Paine 1984). The fact that interspecific competition is so important for intertidal communities explains why competition with *P. perna* East is so important for determining the distribution of *M. galloprovincialis*. The invasive can survive the abiotic conditions in the east, as is shown in Chapter 3, making interspecific competition with *P. perna* East the only remaining variable in the experiment that is capable of explaining the distribution of *M. galloprovincialis*. This is probably not the end of the story though. Abiotic stress on multiple early stages of *M. galloprovincialis* (from larvae to recruits) may either limit recruitment, or produce less-fit adults that are incapable of competing with the superior *P. perna* East lineage. This idea echoes the results of a previous study, that biotic and abiotic effects in concert are more effective at resisting invasion than either process by itself (Levine *et al.* 2004). The results of this thesis support biotic interactions as the primary driver of the present distribution of this invasive species, which is in line with the previous suggestion that intertidal communities are closer to the niche end on the spectrum from niche to neutrality (Menge 1976, Wootton 2005, Caro *et al.* 2010). Nevertheless, intertidal communities are probably not determined by niche-based mechanisms alone (Gravel *et al.* 2006), and much further investigation into this specific case is required. There must be further study of larval supply to the eastern region and the effectiveness of the Agulhas Current as a barrier to gene flow must be quantified. Also, the effects of abiotic conditions on larvae and recruits in the east should be investigated before we can conclude that *M. galloprovincialis* is generally not affected by the increased abiotic stress. While the adults can survive the abiotic conditions there, we cannot extend that result to other life stages of the organism. Lastly, the specific trait or traits that confer a competitive advantage in *P. perna* East require further study. Nonetheless, this thesis provides strong evidence for the role of biotic interactions and niche mechanisms in limiting the spread of the invasive mussel *M. galloprovincialis* in South Africa.

References

Addicott, J. F., et al. (1987). "Ecological neighborhoods: scaling environmental patterns." Oikos **49**: 340-346.

Adler, P. B., et al. (2007). "A niche for neutrality." Ecology Letters **10**(2): 95-104.

Aerts, R. (1999). "Interspecific competition in natural plant communities: mechanisms, trade-offs and plant-soil feedbacks." Journal of Experimental Botany **50**(330): 29-37.

Alonso, D., et al. (2006). "The merits of neutral theory." Trends in Ecology & Evolution **21**(8): 451-457.

Anderson, M. (2005). "PERMANOVA: a FORTRAN computer program for permutational multivariate analysis of variance." Department of Statistics, University of Auckland, New Zealand **24**.

Andrewartha, H. G. and L. C. Birch (1954). The distribution and abundance of animals. Chicago, University of Chicago Press.

Armstrong, R. A. and R. McGehee (1980). "Competitive exclusion." American Naturalist **115**(2): 151-170.

Arroyo, M. T. K., et al. (2000). Plant invasions in Chile: present patterns and future predictions. Washington, D.C., Island Press.

Baker, S. M. (1909). "On the causes of the zoning of brown seaweeds on the seashore." New Phytologist **8**(5 - 6): 196-202.

Barr, G. E. and K. J. Babbitt (2007). "Trout affect the density, activity and feeding of a larval plethodontid salamander." Freshwater Biology **52**(7): 1239-1248.

Bax, N., et al. (2003). "Marine invasive alien species: a threat to global biodiversity." Marine Policy **27**(4): 313-323.

Bekessy, S. A., et al. (2003). "Neutral DNA markers fail to detect genetic divergence in an ecologically important trait." Biological Conservation **110**(2): 267-275.

Bell, E. C., and M. W. Denny (1994). "Quantifying 'wave exposure': a simple device for recording maximum velocity and results of its use at several field sites." Journal of Experimental Marine Biology and Ecology **181**: 9-29.

Bertness, M. D. and G. H. Leonard (1997). "The role of positive interactions in communities: lessons from intertidal habitats." Ecology **78**(7): 1976-1989.

Bertness, M. D., et al. (1999). "Climate-driven interactions among rocky intertidal organisms caught between a rock and a hot place." Oecologia **120**(3): 446-450.

Birch, L. C. (1957). "The meanings of competition." American Naturalist **91**(856): 5-18.

Blackburn, T., et al. (2005). "Functional diversity of mammalian predators and extinction in island birds." Ecology **86**(11): 2916-2923.

Bownes, S., et al. (2008). "Morphological identification of primary settlers and post-larvae of three mussel species from the coast of South Africa." African Journal of Marine Science **30**(2): 233-240.

Bownes, S. J. and C. D. McQuaid (2006). "Will the invasive mussel Mytilus galloprovincialis replace the indigenous Perna perna on the south coast of South Africa?" Journal of Experimental Marine Biology and Ecology **338**(1): 140-151.

Bownes, S. J. and C. D. McQuaid (2010). "Mechanisms of habitat segregation between an invasive (Mytilus galloprovincialis) and an indigenous (Perna perna) mussel: adult growth and mortality." Marine Biology **157**(8): 1799-1810.

Branch, G. (1984). "Competition between marine organisms: ecological and evolutionary implications." Oceanography and Marine Biology Annual Review **22**: 429-593.

Branch, G. M. and C. N. Steffani (2004). "Can we predict the effects of alien species? A case-history of the invasion of South Africa by Mytilus galloprovincialis." Journal of Experimental Marine Biology and Ecology **300**(1): 189-215.

Breton, L. M. and J. F. Addicott (1992). "Density-dependent mutualism in an aphid-ant interaction." Ecology **73**(6): 2175-2180.

Brown, P. (1992). "Spatial and seasonal variation in chlorophyll distribution in the upper 30 m of the photic zone in the southern Benguela/Agulhas ecosystem." South African Journal of Marine Science **12**(1): 515-525.

Bruno, J. F., et al. (2003). "Inclusion of facilitation into ecological theory." Trends in Ecology & Evolution **18**(3): 119-125.

Bustamante, R. and G. Branch (1996). "Large scale patterns and trophic structure of southern African rocky shores: the roles of geographic variation and wave exposure." Journal of Biogeography **23**(3): 339-351.

Bustamante, R. H., et al. (1995). "Gradients of intertidal primary productivity around the coast of South Africa and their relationships with consumer biomass." Oecologia **102**(2): 189-201.

Byers, J. E. (2002). "Physical habitat attribute mediates biotic resistance to non-indigenous species invasion." Oecologia **130**(1): 146-156.

Carlton, J. T. and J. B. Geller (1993). "Ecological roulette: the global transport of nonindigenous marine organisms." Science **261**(5117): 78-82.

Carlton, J. T., et al. (1995). "The role of shipping in the introduction of nonindigenous aquatic organisms to the coastal waters of the United States (other than the Great Lakes) and an analysis of control options." United States Coast Guard and The National Sea Grant Program/Connecticut Sea Grant. United States Coast Guard, Marine Environment Protection Division, Washington, DC.

Carlton, J. T. (1996). "Biological invasions and cryptogenic species." Ecology **77**(6): 1653-1655.

Caro, A. U., et al. (2010). "Ecological convergence in a rocky intertidal shore metacommunity despite high spatial variability in recruitment regimes." Proceedings of the National Academy of Sciences **107**(43): 18528-18532.

Chase, J. M. and M. A. Leibold (2003). Ecological niches: linking classical and contemporary approaches, Chicago, IL, University of Chicago Press.

Chave, J. (2004). "Neutral theory and community ecology." Ecology Letters **7**(3): 241-253.

Chesson, P. (2000). "Mechanisms of maintenance of species diversity." Annual Review of Ecology and Systematics **31**: 343-366.

Colwell, R. K. and D. J. Futuyma (1971). "On the measurement of niche breadth and overlap." Ecology **52**(4): 567-576.

Connell, J. H. (1961). "Effects of competition, predation by Thais lapillus, and other factors on natural populations of the barnacle Balanus balanoides." Ecological Monographs **31**(1): 61-104.

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

Connell, J. H. (1985). "The consequences of variation in initial settlement vs. post-settlement mortality in rocky intertidal communities." Journal of Experimental Marine Biology and Ecology **93**(1): 11-45.

Connell, J. H. and W. P. Sousa (1983). "On the evidence needed to judge ecological stability or persistence." American Naturalist **121**(6): 789-824.

Connolly, S. R. and J. Roughgarden (1998). "A latitudinal gradient in northeast Pacific intertidal community structure: evidence for an oceanographically based synthesis of marine community theory." The American Naturalist **151**(4): 311-326.

Connolly, S. R. and J. Roughgarden (1999). "Theory of marine communities: competition, predation, and recruitment-dependent interaction strength." Ecological Monographs **69**(3): 277-296.

Coulatti Robert I, M. H. J. (2004). "A neutral terminology to define 'invasive' species." Diversity and Distributions **10**: 135-141.

Cowen, R. K., et al. (2000). "Connectivity of marine populations: open or closed?" Science **287**(5454): 857-859.

Crisp, D. and P. Meadows (1962). "The chemical basis of gregariousness in cirripedes." Proceedings of the Royal Society of London. Series B. Biological Sciences **156**(965): 500-520.

Cunha, R. L., et al. (2014) Wider sampling reveals a non-sister relationship for geographically contiguous lineages of a marine mussel. Ecology and Evolution. **4**(11):2070-2081.

Davenport, J. and X. Chen (1987). "A comparison of methods for the assessment of condition in the mussel (Mytilus edulis L.)." Journal of Molluscan Studies **53**(3): 293-297.

Davis, M. A., et al. (2011). "Don't judge species on their origins." Nature **474**(7350): 153-154.

Dayton, P. K. (1971). "Competition, disturbance, and community organization: the provision and subsequent utilization of space in a rocky intertidal community." Ecological Monographs **41**(4): 351-389.

Diamond, J. M. (1978). "Niche shifts and the rediscovery of interspecific competition: Why did field biologists so long overlook the widespread evidence for interspecific competition that had already impressed Darwin?" American Scientist **66**(3): 322-331.

Díaz, E. R. and C. D. McQuaid (2011). "A spatially explicit approach to trophic interactions and landscape formation: patchiness in small‐scale variability of grazing effects along an intertidal stress gradient." Journal of Ecology **99**(2): 416-430.

Elkin, C. and D. J. Marshall (2007). "Desperate larvae: influence of deferred costs and habitat requirements on habitat selection." Marine Ecology Progress Series **335**: 143-153.

Elton, C. S. (1958). The Ecology of Invasions by Animals and Plants. Chicago, IL, University of Chicago Press.

Elton, C. S. and R. S. Miller (1954). "The ecological survey of animal communities: with a practical system of classifying habitats by structural characters." Journal of Ecology **42**(2): 460-496.

Emanuel, B., et al. (1992). "A zoogeographic and functional approach to the selection of marine reserves on the west coast of South Africa." South African Journal of Marine Science **12**(1): 341-354.

Eränen, J. K. and M. V. Kozlov (2008). "Increasing intraspecific facilitation in exposed environments: consistent results from mountain birch populations in two subarctic stress gradients." Oikos **117**(10): 1569-1577.

Fahrig, L. (1992). "Relative importance of spatial and temporal scales in a patchy environment." Theoretical Population Biology **41**(3): 300-314.

Folmer, O., et al. (1994). "DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. Molecular Marine Biology and Biotechnology **3**: 294-299.

Freckleton, R. P. and O. T. Lewis (2006). "Pathogens, density dependence and the coexistence of tropical trees." Proceedings of the Royal Society B: Biological Sciences **273**(1604): 2909-2916.

Fridley, J., et al. (2007). "The invasion paradox: reconciling pattern and process in species invasions." Ecology **88**(1): 3-17.

Fridley, J. D., et al. (2004). "Null models of exotic invasion and scale-dependent patterns of native and exotic species richness." Ecology **85**(12): 3215-3222.

García-Ramos, G. and M. Kirkpatrick (1997). "Genetic models of adaptation and gene flow in peripheral populations." Evolution **51**(1): 21-28.

Gause, G. F. (1934). "Experimental analysis of Vito Volterra's mathematical theory of the struggle for existence." Science **79**(2036): 16-17.

Gaylord, B. and S. D. Gaines (2000). "Temperature or transport? Range limits in marine species mediated solely by flow." The American Naturalist **155**(6): 769-789.

Grace, J. (1991). "A clarification of the debate between Grime and Tilman." Functional Ecology **5**(5): 583-587.

Grant, B. R. and P. R. Grant (1982). "Niche shifts and competition in Darwin's finches: Geospiza conirostris and congeners." Evolution **36**: 637-657.

Grant, P. R. and B. R. Grant (2006). "Evolution of character displacement in Darwin's finches." Science **313**(5784): 224-226.

Grantham, B. A., et al. (2003). "Dispersal potential of marine invertebrates in diverse habitats." Ecological Applications **13**(1): S108-S116.

Gravel, D., et al. (2006). "Reconciling niche and neutrality: the continuum hypothesis." Ecology Letters **9**(4): 399-409.

Griffiths, R. J. (1977). "Reproductive cycles in littoral populations of Choromytilus meridionalis (Kr) and Aulacomya ater (Molina) with a quantitative assessment of gamete production in the former." Journal of Experimental Marine Biology and Ecology **30**: 53-71.

Griffiths, C. L. and J. A. King (1979) "Energy expended on growth and gonad output in the ribbed mussel Aulacomya ater." Marine Biology **53:** 217-222.

Griffiths, C., et al. (1992). "Marine invasive aliens on South African shores: implications for community structure and trophic functioning." South African Journal of Marine Science **12**(1): 713-722.

Grosberg, R. K. and D. R. Levitan (1992). "For adults only? Supply-side ecology and the history of larval biology." Trends in Ecology & Evolution **7**(4): 130-133.

Guichard, F., et al. (2003). "Mussel disturbance dynamics: signatures of oceanographic forcing from local interactions." The American Naturalist **161**(6): 889-904.

Gurevitch, J. and D. K. Padilla (2004). "Are invasive species a major cause of extinctions?" Trends in Ecology & Evolution **19**(9): 470-474.

Harris, J. M., et al. (1998) "Spatial and temporal variability in recruitment of intertidal mussels around the coast of southern Africa. South African Journal of Zoology. 33:1-11.

Hairston, N. G. (1980). "Evolution under interspecific competition: field experiments on terrestrial salamanders." Evolution **34**(3): 409-420.

Hairston, N. G., et al. (1960). "Community structure, population control, and competition." The American Naturalist **94**(879): 421-425.

Hilbish, T. J., et al. (2000). "Origin of the antitropical distribution pattern in marine mussels (Mytilus spp.): routes and timing of transequatorial migration." Marine Biology **136**(1): 69-77.

Hixon, M. A. and G. P. Jones (2005). "Competition, predation, and density-dependent mortality in demersal marine fishes." Ecology **86**(11): 2847-2859.

Hubbell, S. P. (2001). The unified neutral theory of biodiversity and biogeography (MPB-32), Princeton, New Jersey., Princeton University Press.

Hubbell, S. P. (2005). "Neutral theory in community ecology and the hypothesis of functional equivalence." Functional Ecology **19**(1): 166-172.

Hubbell, S. P. (2006). "Neutral theory and the evolution of ecological equivalence." Ecology **87**(6): 1387-1398.

Hunt, C. E. and S. B. Yamada (2003). "Biotic resistance experienced by an invasive crustacean in a temperate estuary." Biological Invasions **5**(1-2): 33-43.

Hunt, H. L. and R. E. Scheibling (1997). "Role of early post-settlement mortality in recruitment of benthic marine invertebrates." Marine Ecology Progress Series **155**: 269-301.

Hutchinson, G. E. (1959). "Homage to Santa Rosalia or why are there so many kinds of animals?" The American Naturalist **93**(870): 145-159.

Jensen, G. C., et al. (2007). "Biotic resistance to green crab, Carcinus maenas, in California bays." Marine Biology **151**(6): 2231-2243.

King, J. R. and W. R. Tschinkel (2006). "Experimental evidence that the introduced fire ant, Solenopsis invicta, does not competitively suppress co‐occurring ants in a disturbed habitat." Journal of Animal Ecology **75**(6): 1370-1378.

Knight-Jones, E. (1951). "Gregariousness and some other aspects of the settling behaviour of Spirorbis." Journal of the Marine Biological Association of the United Kingdom **30**: 201-222.

Knight-Jones, E. (1953). "Laboratory experiments on gregariousness during setting in Balanus balanoides and other barnacles." Journal of Experimental Biology **30**(4): 584-598.

Kourtev, P. S., et al. (2002). "Exotic plant species alter the microbial community structure and function in the soil." Ecology **83**(11): 3152-3166.

Krebs, J. R. (1978). "Optimal foraging: decision rules for predators." Behavioural Ecology: an Evolutionary Approach: 23-63.

Lee, J. and S. Chown (2007). "Mytilus on the move: transport of an invasive bivalve to the Antarctic." Marine Ecology, Progress Series **339**: 307-310.

Leibold, M. A. and M. A. McPeek (2006). "Coexistence of the niche and neutral perspectives in community ecology." Ecology **87**(6): 1399-1410.

Leigh Jr, E. G., et al. (1993). "The decline of tree diversity on newly isolated tropical islands: a test of a null hypothesis and some implications." Evolutionary Ecology **7**(1): 76-102.

Levin, L. A. (2006). "Recent progress in understanding larval dispersal: new directions and digressions." Integrative and Comparative Biology **46**(3): 282-297.

Levin, S. A. (1992). "The problem of pattern and scale in ecology: the Robert H. MacArthur award lecture." Ecology **73**(6): 1943-1967.

Levine, J. M., et al. (2004). "A meta‐analysis of biotic resistance to exotic plant invasions." Ecology Letters **7**(10): 975-989.

Levine, J. M. and C. M. D'Antonio (1999). "Elton revisited: a review of evidence linking diversity and invasibility." Oikos **87**: 15-26.

Lotka, A. J. (1920). "Analytical note on certain rhythmic relations in organic systems." Proceedings of the National Academy of Sciences of the United States of America **6**(7): 410.

Lövei, G. L. (1997). "Biodiversity: global change through invasion." Nature **388**(6643): 627-628.

Lubchenco, J. (1980). "Algal zonation in the New England rocky intertidal community: an experimental analysis." Ecology **61**(2): 333-344.

MacArthur, R. and R. Levins (1967). "The limiting similarity, convergence, and divergence of coexisting species." American Naturalist **101**(921): 377-385.

MacDougall, A. S. and R. Turkington (2005). "Are invasive species the drivers or passengers of change in degraded ecosystems?" Ecology **86**(1): 42-55.

Mack, M. C. and C. M. D'Antonio (1998). "Impacts of biological invasions on disturbance regimes." Trends in Ecology & Evolution **13**(5): 195-198.

May, R. and R. Macarthur (1972). "Niche overlap as a function of environmental variability." Proceedings of the National Academy of Sciences of the United States of America **69**: 1109-1113.

Mayr, E. (1983). "How to carry out the adaptationist program?" American Naturalist **121**(3): 324-334.

McCann, K., et al. (1998). "Weak trophic interactions and the balance of nature." Nature **395**(6704): 794-798.

McGill, B. J. (2003). "A test of the unified neutral theory of biodiversity." Nature **422**(6934): 881-885.

McGill, B. J., et al. (2006). "Empirical evaluation of neutral theory." Ecology **87**(6): 1411-1423.

McQuaid, C. and T. Phillips (2000). "Limited wind-driven dispersal of intertidal mussel larvae: in situ evidence from the plankton and the spread of the invasive species Mytilus galloprovincialis in South Africa." Marine Ecology Progress Series **201**: 211-220.

McQuaid, C. D. and F. Arenas (2009). Biological Invasions: Insights from Marine benthic communities, Springer Berlin Heidelberg**:** 309-320.

Mead, A., et al. (2011). "Revealing the scale of marine bioinvasions in developing regions: a South African re-assessment." Biological Invasions **13**(9): 1991-2008.

Menge, B. A. (1976). "Organization of the New England rocky intertidal community: role of predation, competition, and environmental heterogeneity." Ecological Monographs **46**(4): 355-393.

Menge, B. A., et al. (1994). "The keystone species concept: variation in interaction strength in a rocky intertidal habitat." Ecological Monographs **64**(3): 249-286.

Michalek-Wagner, K. and B. Willis (2001). "Impacts of bleaching on the soft coral Lobophytum compactum. I. Fecundity, fertilization and offspring viability." Coral Reefs **19**(3): 231-239.

Minchinton, T. E. and R. E. Scheibling (1991). "The influence of larval supply and settlement on the population structure of barnacles." Ecology **72**(5): 1867-1879.

Morgan, S. G., et al. (2009). "Nearshore larval retention in a region of strong upwelling and recruitment limitation." Ecology **90**(12): 3489-3502.

Munday, P. L., et al. (2001). "Interspecific competition and coexistence in a guild of coral-dwelling fishes." Ecology **82**(8): 2177-2189.

Nicastro, K., et al. (2010). "Differential reproductive investment, attachment strength and mortality of invasive and indigenous mussels across heterogeneous environments." Biological Invasions **12**(7): 2165-2177.

Nichols, F. H., et al. (1990). "Remarkable invasion of San Francisco Bay (California, USA) by the Asian clam Potamocorbula amurensis. II: Displacement of a former community. Marine Ecology Progress Series **66**: 95-101.

Nottale, L. (1996). "Scale relativity and fractal space-time: applications to quantum physics, cosmology and chaotic systems." Chaos, Solitons & Fractals **7**(6): 877-938.

Paine, R. (1984). "Ecological determinism in the competition for space." Ecology **65**(5): 1339-1348.

Paine, R. T. (1966). "Food web complexity and species diversity." American Naturalist **103**(929): 65-75.

Palmer, M. A., et al. (1996). "Dispersal as a regional process affecting the local dynamics of marine and stream benthic invertebrates." Trends in Ecology & Evolution **11**(8): 322-326.

Pechenik, J. A., et al. (1998). "Metamorphosis is not a new beginning." BioScience **48**(11): 901-910.

Petes, L. E., et al. (2007). "Environmental stress decreases survival, growth, and reproduction in New Zealand mussels." Journal of Experimental Marine Biology and Ecology **351**(1): 83-91.

Petraitis, P. S. (1995). "The role of growth in maintaining spatial dominance by mussels (Mytilus edulis)." Ecology **76**(4): 1337-1346.

Phillips, T. (1994). Dispersal, settlement and recruitment: their influence on the dynamics of intertidal mussels, Ph. D. thesis, Department of Zoology and Entomology, Rhodes University: 246 pp.

Pimentel, D., et al. (2005). "Update on the environmental and economic costs associated with alien-invasive species in the United States." Ecological Economics **52**(3): 273-288.

Pineda, J., et al. (2010). "Causes of decoupling between larval supply and settlement and consequences for understanding recruitment and population connectivity." Journal of Experimental Marine Biology and Ecology **392**(1): 9-21.

Pitcher, G. and D. Calder (1998). "Shellfish mariculture in the Benguela system: phytoplankton and the availability of food for commercial mussel farms in Saldanha Bay, South Africa." Journal of Shellfish Research **17**(1): 15-24.

Porri, F. et al. (2006). "Spatio-temporal variability of larval abundance and setllement of Perna perna: differential delivery of mussels." Marine Ecology Progress Series 315: 141-150.

Porri, F. et al. (2007). "Tidal height, rather than habitat selection for conspecifics, controls settlement in mussels." Marine Biology **152**(3): 631-637.

Porri F, et al. (2008.) "Fine-scale spatial and temporal variation in settlement of the intertidal mussel Perna perna indicates differential hydrodynamic delivery of larvae to the shore." Journal of Experimental Marine Biology and Ecology **367**: 213-218.

Porri, F. et al (2014). "The effect of mesoscale oceanographic features on the distribution of mussel larvae along the south coast of South Africa." Journal of Marine Systems 132, 162-173.

Porter, S. D. and D. A. Savignano (1990). "Invasion of polygyne fire ants decimates native ants and disrupts arthropod community." Ecology **71**(6): 2095-2106.

Quinn, J. F. and A. E. Dunham (1983). "On hypothesis testing in ecology and evolution." The American Naturalist **122**(5): 602-617.

Reaugh, K. E., et al. (2007). "Further refutation of the primary-secondary settlement hypothesis for the brown mussel Perna perna. " African Journal of Marine Science **29**(3): 545-549.

Richmond, C. and S. Woodin (1996). "Short-term fluctuations in salinity: effects on planktonic invertebrate larvae." Marine Ecology Progress Series **133**(1): 167-177.

Ricklefs, R. E. (2003). "A comment on Hubbell's zero‐sum ecological drift model." Oikos **100**(1): 185-192.

Rius, M. and C. D. McQuaid (2006). "Wave action and competitive interaction between the invasive mussel Mytilus galloprovincialis and the indigenous Perna perna in South Africa." Marine Biology **150**(1): 69-78.

Rius, M. and C. D. McQuaid (2009). "Facilitation and competition between invasive and indigenous mussels over a gradient of physical stress." Basic and Applied Ecology **10**(7): 607-613.

Rius, M. (2004). The effect of the invasive mussel Mytilus galloprovincialis and human exploitation on the indigenous mussel Perna perna on the South Coast of South Africa, M. Sc. thesis, Department of Zoology and Entomology, Rhodes University, South Africa.

Roberts, M., et al. (2010). "Shelf currents, lee-trapped and transient eddies on the inshore boundary of the Agulhas Current, South Africa: their relevance to the KwaZulu-Natal sardine run." African Journal of Marine Science **32**(2): 423-447.

Robinson, T., et al. (2005). "Marine alien species of South Africa—status and impacts." African Journal of Marine Science **27**(1): 297-306.

Romanuk, T. N. and J. Kolasa (2005). "Resource limitation, biodiversity, and competitive effects interact to determine the invasibility of rock pool microcosms." Biological Invasions **7**(4): 711-722.

Rose, G. A. and W. C. Leggett (1990). "The importance of scale to predator-prey spatial correlations: an example of Atlantic fishes." Ecology **71**(1): 33-43.

Rosenzweig, M. L. (2001). "The four questions: what does the introduction of exotic species do to diversity?" Evolutionary Ecology Research **3**(3): 361-367.

Roughgarden, J. (1983). "Competition and theory in community ecology." The American Naturalist **122**(5): 583-601.

Roughgarden, J. et al. (1985). "Demographic theory for an open marine population with space-limited recruitment." Ecology **66**(1): 54-67.

Ruiz, G. M., et al. (1997). "Global invasions of marine and estuarine habitats by non-indigenous species: mechanisms, extent, and consequences." American Zoologist **37**(6): 621-632.

Ruiz, G. M., et al. (2000). "Invasion of coastal marine communities in North America: apparent patterns, processes, and biases." Annual Review of Ecology and Systematics **31**: 481-531.

Russell, R., et al. (2006). "Scale, environment, and trophic status: the context dependency of community saturation in rocky intertidal communities." The American Naturalist **167**(6): E158-E170.

Sale, P. F. (1974). "Overlap in resource use, and interspecific competition." Oecologia **17**(3): 245-256.

Sax, D. F., et al. (2007). "Ecological and evolutionary insights from species invasions." Trends in Ecology & Evolution **22**(9): 465-471.

Schoener, T. W. (1982). "The controversy over interspecific competition: despite spirited criticism, competition continues to occupy a major domain in ecological thought." American Scientist **70**(6): 586-595.

Sebens, K. P. (1982). "Competition for space: growth rate, reproductive output, and escape in size." The American Naturalist **120**(2): 189-197.

Semenikhina, O. Y., et al. (2008). "Morphology of larvae of the family Mytilidae (Bivalvia) from the north-western part of the Sea of Japan." Journal of the Marine Biological Association of the United Kingdom **88**(2): 331-339.

Shanks, A. L. and L. Brink (2005). "Upwelling, downwelling, and cross-shelf transport of bivalve larvae: test of a hypothesis." Marine Ecology Progress Series **302**: 1-12.

Shanks, A. L., et al. (2003). "Propagule dispersal distance and the size and spacing of marine reserves." Ecological Applications **13**(sp1): 159-169.

Shea, K. and P. Chesson (2002). "Community ecology theory as a framework for biological invasions." Trends in Ecology & Evolution **17**(4): 170-176.

Siddall, S. E. (1980). "A clarification of the genus Perna (Mytilidae)." Bulletin of Marine Science **30**(4): 858-870.

Southwood, T. (1977). "Habitat, the templet for ecological strategies?" Journal of Animal Ecology **46**(2): 337-365.

Stachowicz, J. J., et al. (1999). "Species diversity and invasion resistance in a marine ecosystem." Science **286**(5444): 1577-1579.

Stohlgren, T. J., et al. (1999). "Exotic plant species invade hot spots of native plant diversity." Ecological Monographs **69**(1): 25-46.

Strathmann, R. (1974). "The spread of sibling larvae of sedentary marine invertebrates." American Naturalist **108**(959): 29-44.

Strayer, D. L., et al. (2006). "Understanding the long-term effects of species invasions." Trends in Ecology & Evolution **21**(11): 645-651.

Swearer, S. E., et al. (1999). "Larval retention and recruitment in an island population of a coral-reef fish." Nature **402**(6763): 799-802.

Thompson, R. J. (1979). "Fecundity and reproductive effort in the blue mussel (Mytilus edulis), the sea urchin (Strongylocentrotus droebachiensis) and the snow crab (Chionoecetes opilio) from populations in Nova Scotia and Newfoundland." Journal of the Fisheries Board of Canada **36**: 955-964.

Tilman, D., et al. (1997). "The influence of functional diversity and composition on ecosystem processes." Science **277**(5330): 1300-1302.

Tilman, D. (2004). "Niche tradeoffs, neutrality, and community structure: a stochastic theory of resource competition, invasion, and community assembly." Proceedings of the National Academy of Sciences of the United States of America **101**(30): 10854-10861.

Travaset A, and D. Richardson (2006). "Biological invasions as disruptors of plant rprodcutive mutualisms." Trends in Ecology & Evolution **21**(208-216).

Underwood, A. (1986). The analysis of competition by field experiments. Melbourne, Blackwells.

Underwood, A. and P. Fairweather (1989). "Supply-side ecology and benthic marine assemblages." Trends in Ecology & Evolution **4**(1): 16-20.

United Nations system-wide earthwatch <www.un.org/earthwatch>

Van den Brink, F., et al. (1993). "Ecological aspects, explosive range extension and impact of a mass invader, Corophium curvispinum Sars, 1895 (Crustacea: Amphipoda), in the Lower Rhine (The Netherlands)." Oecologia **93**(2): 224-232.

Van Erkom Schurink, C. and C. Griffiths (1990). "Marine mussels of southern Africa–their distribution patterns, standing stocks, exploitation and culture." Journal of Shellfish Research **9**(1): 75-85.

Van Erkom Schurink, C. and C. Griffiths (1991). "A comparative study of reproductive cycles and reproductive effort in four southern African mussel species." Marine Ecology Progress Series **76**: 123-134.

Van Erkom Schurink, C. and C. Griffiths (1992). "Physiological energetics of four South African mussel species in relation to body size, ration and temperature." Comparative Biochemical Physiology **101**(4): 779-789.

Van Erkom Schurink , C. and C. Griffiths (1993). "Factors affecting the relative growth in four South African mussel species". Aquaculture **109**: 257-273.

Vance, R. R. (1973). "More on reproductive strategies in marine benthic invertebrates." The American Naturalist **107**(955): 353-361.

Volterra, V. (1928). "Variations and fluctuations of the number of individuals in animal species living together." Journal du Conseil International por l'Exploration de la Mer. **3**(1): 3-51.

Von der Meden, C., et al. (2008). "Coastline topography affects the distribution of indigenous and invasive mussels." Marine Ecology Progress Series **372**: 135-145.

Von der Meden, C. E., et al. (2010). "Fine-scale ontogenetic shifts in settlement behaviour of mussels: changing responses to biofilm and conspecific settler presence in Mytilus galloprovincialis and Perna perna." Marine Ecology Progress Series **411**: 161-171.

Wiens, J. A. (1989). "Spatial scaling in ecology." Functional Ecology **3**(4): 385-397.

Wiens, J. J. (2004). "Speciation and ecology revisited: phylogenetic niche conservatism and the origin of species." Evolution **58**(1): 193-197.

Wiens, J. J. and C. H. Graham (2005). "Niche conservatism: integrating evolution, ecology, and conservation biology." Annual Review of Ecology, Evolution, and Systematics: 519-539.

Wilcove, D. S., et al. (1998). "Quantifying threats to imperiled species in the United States." BioScience **48**(8): 607-615.

Wilson, E. O. (1992). "The diversity of life." Belknap, Cambridge, Massachusetts.

Wonham, M., et al. (2000). "Fish and ships: relating dispersal frequency to success in biological invasions." Marine Biology **136**(6): 1111-1121.

Wootton, J. T. (2005). "Field parameterization and experimental test of the neutral theory of biodiversity." Nature **433**(7023): 309-312.

Zardi, G., et al. (2006). "Hydrodynamic stress and habitat partitioning between indigenous (Perna perna) and invasive (Mytilus galloprovincialis) mussels: constraints of an evolutionary strategy." Marine Biology **150**(1): 79-88.

Zardi, G. et al. (2007). "Balancing survival and reproduction:seasonality of wave action, attachment strength and reproductive output in indigenous Perna perna and invasive Mytilus galloprovincialis mussels." Marine Ecology Progress Series **334**: 155-163.

Zardi, G., et al. (2007). "Unexpected genetic structure of mussel populations in South Africa: indigenous Perna perna and invasive Mytilus galloprovincialis." Marine Ecology, Progress Series **337**: 135-144.

Zardi, G., et al. (2011). "The combination of selection and dispersal helps explain genetic structure in intertidal mussels." Oecologia **165**(4): 947-958.