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Nearshore Salinity and Juvenile Pink Shrimp (Farfantepenaeus duorarum): Integrating Field Observations, Laboratory Trials, and Habitat Suitability Simulations

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UNIVERSITY OF MIAMI

NEARSHORE SALINITY AND JUVENILE PINK SHRIMP (*FARFANTEPENAEUS DUORARUM*): INTEGRATING FIELD OBSERVATIONS, LABORATORY TRIALS, AND HABITAT SUITABILITY SIMULATIONS

By

Ian C. Zink

A DISSERTATION

Submitted to the Faculty
of the University of Miami
in partial fulfillment of the requirements for
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A dissertation submitted in partial fulfillment of
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NEARSHORE SALINITY AND JUVENILE PINK SHRIMP (*FARFANTEPENAEUS DUORARUM*): INTEGRATING FIELD OBSERVATIONS, LABORATORY TRIALS, AND HABITAT SUITABILITY SIMULATIONS

Ian C. Zink

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Nearshore Salinity and Juvenile Pink Shrimp (*Farfantepenaeus duorarum*): Integrating Field Observations, Laboratory Trials, and Habitat Suitability Simulations (December 2017)

Abstract of a dissertation at the University of Miami.

Dissertation supervised by Professors Joseph Serafy and Diego Lirman.
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The pink shrimp *Farfantepenaeus duorarum* has been selected as one of many ecological indicators to assess implementation of the Comprehensive Everglades Restoration Plan (CERP). One element of CERP, the Biscayne Bay Coastal Wetlands (BBCW) project, intends to reduce salinity conditions along southwestern Biscayne Bay nearshore areas while also redistributing freshwater inflows to expand the spatial extent of reduced salinity (mesohaline [5-18 ppt] and oligohaline [0.5-5 ppt]) conditions. CERP restoration goals for Biscayne Bay juvenile pink shrimp nearshore populations presumed these salinity regimes would benefit pink shrimp abundances. Using this hypothesis as a backdrop, my dissertation was designed to test whether increased area of mesohaline and oligohaline conditions within a 50-m strip of shallow-water shoreline area likely would increase pink shrimp densities. I conducted four studies consisting of a literature review, both field and laboratory studies, and a habitat suitability modeling studies to test this hypothesis. First, I carried out a comprehensive literature review to gather, synthesize, and, where possible, conduct meta-analysis to provide a context for salinity effects on pink shrimp “Essential Fish Habitat” (EFH) metrics (i.e., distribution/occurrence, density/relative abundance, growth, survival, and productivity) and behavior. Polyhaline and/or euhaline salinity regimes generally maximized pink shrimp occurrence, abundance, growth, and survival. However, exceptions to this generalization were noted, such as interaction with benthic aquatic vegetation conditions or limitations with respect to observed salinity ranges. Meta-analysis of survival data yielded a parabolic relationship that maximized at ~30 ppt. Next, I conducted an analysis of field data representing 10 years of pink shrimp density and habitat observations from a monitoring project on southwestern Biscayne Bay nearshore epifaunal communities. Quantile regression detected density limitation by temperature, salinity, total submerged aquatic vegetation percent cover, and water depth. Cluster analysis of pink shrimp spatial density patterns revealed density reductions in a sub-region of the shoreline marked by low mean salinity and higher salinity variation. Temporal clustering indicated low inter-annual and seasonal variability in density patterns. However, wet season densities were more often lower than those of dry season, especially during the 2013 wet season microalgal bloom event. Third, I completed a laboratory study to investigate salinity effects (5, 10, 15, 25, 35, 45, and 55 ppt treatment levels) on pink shrimp growth, in terms of molt increment and intermolt period, and survival. Linear growth in weight and length exhibited parabolic responses to salinity with maximal growth at 34 and 33 ppt, respectively. No statistical relationship between salinity and molt increment nor intermolt period was observed. Pink shrimp survival was reduced at salinities < 15 ppt. This study may have been compromised by an as yet unidentified disease displayed by the shrimp. Lastly, I used quantile regression of 35 d antecedent mean salinity as the basis for a habitat suitability model to simulate salinity effects on pink shrimp habitat suitability area under two restoration scenarios. The resulting dome-shaped functional response yielded maximal potential pink shrimp density at 29.25 ppt. This habitat suitability model was applied to Base Flow and High Flow (~2x Base Flow freshwater discharges) 35 d antecedent mean salinities simulated by the Biscayne Bay Simulation Model v4. Under the Base Flow scenario, the areal extent of wet season good and optimal habitat suitability totaled 91.5% of the simulated spatial domain (i.e., 50 m nearshore area from Shoal Point to Turkey Point). Wet season good and optimal habitat suitability decreased to 74.3% of the spatial domain in the High Flow scenario. Considering the results of these four studies, reduction of nearshore salinities to oligohaline (0-5 ppt) and mesohaline (5-18 ppt) regimes would negatively impact pink shrimp density, growth, survival, and habitat suitability. Furthermore, analysis of field data revealed that pink shrimp densities were generally higher in the dry season (Jan.-Mar.), whereas the CERP pink shrimp restoration goal focused on Aug.-Oct. abundances when densities were presumed to be the highest. These results should prove useful to freshwater and resource managers, scientists, and policy advocates involved in CERP implementation as well as to those with a general interest in pink shrimp, penaeid, or estuarine ecology.

DEDICATION

I dedicate this work to my family. My parents have been my best cheerleaders since this little boy told them of his dream. My sister commiserated with me through the trials and tribulations of higher education. My brother inspired me not sweat the little things and to see a bigger picture. My wife came into my life and motivated me to achieve my best while showering me with patience and understanding. My son re-energized my work as I recognized an example - very dear to me – as to why I had embarked on this quest: to improve our present world and leave it a little better for the next generation. All of you have preserved with me, sacrificed for me, and encouraged me to be my best.

Thank you for the love that envelops me.

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Without Joan Browder, the opportunity to pursue this degree would not have been possible. I began work under the umbrella of the Browder Lab a little over seven years prior to completion of this document. I often joked that you were my "financial advisor" during the pursuit of this degree. You provided the majority of the funding that allowed for this work to proceed, an arrangement that provided much freedom for me to think long and deep during formulation this dissertation. Your institutional knowledge of all things Everglades and pink shrimp provided a vast store of knowledge for me to build upon and acted as a spring-board for my ideas. You too have dedicated much of your time to help me improve both technically – especially with my writing – as well as personally.

Forces external of my committee caused Diego Lirman to elevate to the level of co-advisor. While it the title was forced, in retrospect it was only natural that you ended up with it. You too have devoted much time and energy in shaping my analyses, writing, and thinking. I thank you for being a moderating force on my committee. You also kept my thinking relevant to the big picture, and helped me move forward when my wheels were spinning in the details. Your knowledge of subtropical benthic ecosystems contributed greatly to the interpretation of these results.

Jiangang Luo and David Die completed my dissertation committee. Your application of personal expertise and unique talents to this work often forced me to see things from a different perspective. You carefully guided me in the precision of language, and corrected my transgressions of over-simplification or generalizations of my results. Jiangang provided much assistance with my foray in modeling and simulation. David's expertise of penaeids provided an excellent sounding board as I considered my results within a broader ecological context. Although not a committee member, I also would like to thank Erik Stabenau, who provided BBSM model outputs. Without your prior work, as well as discussions and motivation, Chapter 5 would not exist.

Two of the following chapters relied heavily on the long-term, southwestern Biscayne Bay nearshore epifaunal monitoring dataset which was initiated in 2005. The existence of this dataset was possible only because of the efforts of many individuals over the past 12 years. While many more individuals contributed to collection of this data, I would especially like to thank those that I had the pleasure of spending field days with: Eric Buck, Tom Jackson, Gladys Liehr, Ana Griefen, Laura Petteway, Michelle Harangody, Simeon Yurek, Robin Casciolli, Tim Creed, David Bouck, Michelle La Martina, Katarzyna Kulpa, Nicole Besemer, Josh Goldston, Diego Lirman, Travis Thyberg, Stephanie Schopmeyer, Rolando Santos, Crawford Drury, Kelley Peebles, Dalton Hesley, Jane Carrick, Ryan Sirota, Rich Karp, Herve Jobert, and Jordi Robles - I hope I got you all. Over the years we have spent many long hot, cold, rainy, exhausting days on the water dodging lightening, sharks, hurricanes, cold fronts, algal blooms, angry fishermen, and manatees. Some you have also spent hours sorting through the half rotten seagrass, identifying organisms at scopes, and compiling the databases from samples we collected. Thank you all for making the fun times more fun and banding together in the bad times. Here's to many more dozens of Dunkin' Donuts!

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I came to Miami – and somehow haven't left after 17 years! During the intervening time I developed a network of friends that have become like family: James Herlan; Mark Fitchett; Lyza Johnston; Rolando Santos; Sascha Cushner; Stephanie Schopmeyer; and Dave, Dana, Drew, and Celine Viggiano (D^3 + C!). There are many more great friends I have made through my years in the RSMAS community and in greater Miami. Whether waxing on science or just enjoying the spoils of our location, you have all helped achieve this goal. ¡Muchas gracias y un gran abrazo a todos!

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1.1 The Focal Species: Pink Shrimp

The pink shrimp *Farfantepenaeus duorarum* was first recognized as a species during Burkenroad's (1939) specific differentiation of the *Penaeus* (later *Farfantepenaeus*) *brasiliensis* complex. After populations capable of supporting targeted commercial fishing were identified (Idyll, 1950a, 1950b), interest in the *F. duorarum* life history and ecology grew substantially. The bulk of United States pink shrimp commercial landings originate from the Tortugas Grounds, northwest of Key West on the southwestern Florida Gulf shelf (Fig. 1.1, Table 1.1; Iversen et al. 1960, Hart et al. 2012). Between offshore food fisheries and nearshore bait and food fisheries, pink shrimp fisheries generate millions of dollars in economic yields (Table 1.1, 1.2). In an ecological context, pink shrimp are noted to be an important trophodynamic linkage between primary and tertiary consumers (Browder and Robblee 2009).

Similar to other penaeid shrimps (Dall et al., 1990), the pink shrimp life history (Fig. 1.1) involves migration between nearshore juvenile nursery habitats and offshore adult spawning grounds, which may be spatially separated by hundreds of kilometers (Tabb et al., 1962a; Iversen and Idyll, 1960; Costello and Allen, 1966; McCoy, 1968; Cody and Fuls, 1981; Klima et al., 1987). South Florida nearshore areas, bays, and estuaries have long been recognized as important nursery grounds for pink shrimp. Postlarval pink shrimp immigrant abundance in nearshore nursery areas has been correlated to subsequent commercial landings of newly recruited, small adults (Roessler and Rehrer 1971). Juvenile pink shrimp emigration from nurseries has also been linked to fishery catch of newly recruited adults (Yokel et al., 1969; Ehrhardt and Legault, 1999). Density-dependence in adult recruitment has been associated with juvenile productivity (Ehrhardt et al., 2001). Mark-recapture studies of juvenile pink shrimp released in southern Florida bays and estuaries directly linked inshore populations to Tortugas and Sanibel adult spawning grounds (Iversen and Idyll, 1960; Tabb et al., 1962a; Costello and Allen, 1966).

Postlarval, juvenile, and subadult pink shrimp reliance on nearshore nursery areas makes these ontogenetic stages vulnerable to coastal ecological degradation and habitat perturbations. One habitat alteration affecting pink shrimp nearshore ontogenetic stages in south Florida coastal areas is modification of the timing, quantity, quality, and distribution of freshwater inflows and ground water discharges by freshwater management (Browder et al., 2005; Rudnick et al., 2005; Browder and Robblee, 2009). South Florida freshwater management has developed into a vast network of canals, levees, and pumping stations that are used to control freshwater water tables, movements, and inflows to coastal areas. Tabb (1962a) was among the first to express concern that upland freshwater management would alter nearshore habitat to the detriment of juvenile pink shrimp productivity. Not only had freshwater management severely affected terrestrial ecosystems, but altered freshwater flows also directly impacted nearshore salinity conditions, and thus juvenile, and consequently later adult, pink shrimp productivity (Browder 1985, Sheridan 1996, Ehrhardt and Legault 1999, Ehrhardt et al. 2001). A deeper understanding of salinity influences on juvenile pink shrimp productivity - conditional on distribution, abundance, growth, and survival - would not only advance pink shrimp ecology, but also help scientists and resource managers make informed freshwater management decisions.

1.2 The Problem: A Brief History of South Florida Freshwater Management

Water is the basis for the unique features of southern Florida that make it of national park caliber.
-A.E. Demaray, Acting Director of the National Park Service, 1939

Estuaries are semi-enclosed areas where river inflows or other terrestrial drainages join and mix with salt water originating from a free connection to the open ocean (Pritchard 1967). The quantity and timing of freshwater deliveries to estuaries have been considered the most fundamental characteristics determining the functioning of an estuary (Montagna et al. 2002). Although other physical conditions may also be important, freshwater inflow regimes that determine estuarine salinity cycles are dominant estuarine attributes that shape spatial and temporal biological communities (Hedgepeth 1966). Humans have long depended on estuaries as sources of food, for commerce, as transportation corridors, for disposal of waste, and as bases for further exploration (Cronin 1967). Explosive industrial development and human population growth during the mid-19th century led to increased pressure on estuarine stability and functionality (Cronin 1967). Alteration of freshwater inflows - whether for agricultural, navigational, energy production, land reclamation, flood control, recreation - affects estuarine salinity regimes with sometimes dire ecological consequences (Smith 1966, Cronin 1967).

Across the geographic range of pink shrimp, various human activities affecting freshwater inflows have influenced nearshore areas that successful completion of their life history cycle depends upon. For example, drought in the 1950's led to realization of a need to intensively manage Texas freshwater resources, including the desire to reserve water for future urban and agricultural growth and development. This vision was realized with the Texas Water Plan of 1961 (Vandertulip 1961). Perhaps no greater example of the complicated nature of allocating fresh water for human consumption and retaining sufficient quantities to maintain ecosystem functionality has been realized in south Florida. There, the largest ecosystem restoration project ever attempted is currently underway: the Comprehensive Everglades Restoration Plan (CERP).

Beginning in the mid-1800s, the newly created Florida state legislature expressed interest in drainage and development of the vast swamplands in the southern reaches of the state (Godfry and Catton 2011). Despite construction of drainage canals, flooding still occurred in the Everglades region, a point made evident after the 1926 and 1928 hurricanes caused substantial loss of life and property damage (Godfry and Catton 2011). In 1930, Congress passed a general river and harbor bill that authorized direct federal financing and involvement of U.S. Army Corps of Engineers (USACE) in south Florida flood control projects. However, as freshwater management efforts increased, their negative consequences became realized. A growing south Florida environmental movement began advocating for protection of remaining unspoiled Everglades areas at the southern extreme of the Florida peninsula resulting in Congressional approval for Everglades National Park in 1934. In 1939, meetings were held in south Florida regarding saltwater intrusion effects on municipal water supplies; wildfire (“muck fire”) consumption of soil organic matter as well as soil subsidence caused by oxidation of soil organic content were other negative impacts of over-drainage (Godfry and Catton 2011).

Recognizing these concerns, USACE developed a new flood control plan requiring enlargement of existing canals as well as construction of new canals, impoundments, levees, water pumping stations, to balance flood control, municipal and agricultural water needs, and provisions for reducing negative impacts (Godfry and Catton 2011). The Central and South Florida Flood Control Project (CSFFCP) was federally authorized by the Flood Control Act of 1948, which established water conservation areas (WCAs). Although it was noted that WCAs were anticipated to benefit fish and wildlife, the CSFFCP effects on south Florida ecosystems were considered inconsequential and seemed peripheral to anthropocentric goals and benefits. Meanwhile, the south Florida environmental movement grew in step with urban development throughout the latter half of the 20th century. Biscayne National Monument (later National Park) received federal protection in 1968, and Big Cypress National Preserve was created in 1974. However, by the 1980's, concern for water resource allocation to maintain ecosystem functioning was at an all-time high as extreme manifestations of environmental degradation became apparent. One example was the mass mortality of Florida Bay seagrass beds (Robblee et al. 1991, Zieman et al. 1999), which altered Florida Bay fish and invertebrate communities (Robblee and DiDomenico 1991, Matheson et al. 1999, Thayer et al. 1999).

By the end of the 1980's, the "Marshall Plan" was being discussed by politicians and resource managers as a means to "repair" the Everglades, although this plan initially focused on Kissimmee River basin restoration north of Lake Okeechobee(Godfrey and Catton 2011). Support for a broader Everglades restoration continued to build, and in the Water Resources Development Act (WRDA) of 2000, Congress authorized USACE to implement the Comprehensive Everglades Restoration Plan (CERP: Fig. 1.2; Godrey and Catton 2011). Among other goals, CERP seeks to improve the quality, quantity, timing, and distribution of terrestrial freshwaters as well as their subsequent discharge to south Florida nearshore areas.

1.3 The Study Site: Biscayne Bay

Biscayne Bay is a large (1,110 km^2), shallow (depths generally < 3 m), subtropical lagoon system located along the southeast coast of Florida, USA (Fig. 1.1). Its western shoreline extends approximately 56 km north to south. Virtually the entire bay falls under a federal (Biscayne Bay National Park) or state (Biscayne Bay Aquatic Preserve) ecologically-protected status. Where coastal urban development is low, its shorelines consist of a mangrove-seagrass ecotone punctuated by natural tidal creeks, artificial channels, and freshwater canals (Serafy et al. 2007). Overland freshwater discharges, along with groundwater seepage, create a salinity gradient perpendicular to the shoreline with three salinity zones: (1) western nearshore areas usually affording the lowest salinities; (2) the bay central axis marked by near oceanic salinities; and (3) oceanic salinities near the eastern passes to the open ocean (Lirman and Cropper 2003, Caccia and Boyer 2005, Lirman et al. 2008a). Tidal ranges are generally on the order of 0.5 to 1 m (Schneider 1969, Wang et al. 2003). Biscayne Bay is just one target area among those considered as the Southern Coastal Systems in reference to CERP ecological restoration.

Historically, most freshwater delivery to Biscayne Bay was through transverse glades: low topography channels through the Miami Coastal Ridge that allowed

Everglades Basin surface water drainage (Davis 1943, Halley et al. 1977) and groundwater seepage (Shaler 1895, Smith 1896, Kohout 1967, Kohout and Kolipinski 1967, Langevin 2003, Stalker et al. 2009). On the eastern side of the Coastal Ridge, transverse glades fed fresh water from the Everglades into creek networks that spread surface water flows along the shoreline. However, canalization converted the freshwater delivery system to one dominated by pulsed point-source (i.e., canal mouth) discharges that altered benthic submerged aquatic vegetation (SAV), infaunal, epifaunal, and nekton communities (Brook 1982; Irlandi et al. 1997; Serafy et al. 1997; Lirman et al. 2008a, b, 2014; Santos et al. 2011). Canalization also lowered the water table, which reduced nearshore groundwater seepage (Langevin 2003, Stalker et al. 2009).

The Biscayne Bay Coastal Wetland (BBCW) project is a CERP component designed to distribute freshwater inflows to Biscayne Bay via restoration of overland sheet flows as well as increased overall freshwater volume to restore historic nearshore salinity regimes. Post-BBCW salinity goals for western central and southern Biscayne Bay (Shoal Point to Turkey Point: Fig. 1.1) include nearshore areas with oligohaline (0.5-5 ppt) and mesohaline (5-18 ppt) regimes at the shoreline trending towards ~20-28 ppt 600 m from the coast and farther (RECOVER 2008). These salinity conditions are anticipated to enrich estuarine faunal assemblages as well as increase estuarine species distributions and abundances (Browder et al. 2005a, SFNRC 2006, RECOVER 2008, McManus et al. 2014). Expansion of continuous submerged aquatic vegetation (SAV) habitat dominated by *Halodule wrightii*, a species commonly associated with low and variable salinity, is also foreseen (Lirman and Cropper 2003; RECOVER 2004; SFNRC 2006; Lirman et al. 2008a, b, 2014; Santos et al. 2011). BBCW implementation goals for benthic habitat include increased spatial extent of nearshore seagrass beds, especially expansion of *H. wrightii* toward the bay's interior (RECOVER 2004, SFNRC 2006). Increased overlap of optimal salinity conditions with preferred benthic SAV habitats would yield indirect, synergistic benefits to estuarine fauna, presumably including pink shrimp (Sklar and Browder 1998, Browder et al. 2005a, RECOVER 2008, Browder and Robblee 2009).

1.4 Dissertation Objectives

The focus of this dissertation was to investigate effects of nearshore salinity regimes and their changes on the juvenile pink shrimp productivity. These investigations were intended to be broadly applicable to pink shrimp ecology throughout its geographic range. However, the immediate motivation was to contribute to the understanding of how CERP, and in particular BBCW, implementation may impact juvenile pink shrimp populations inhabiting the southwestern Biscayne Bay shoreline. More specifically, I used CERP restoration goals and targets for Biscayne Bay juvenile pink shrimp populations as a general working hypothesis for these investigations. According to RECOVER (2005), restoration of nearshore salinity regimes is anticipated to increase wet season juvenile pink shrimp abundances by shifting salinity habitats to conditions more favorable for pink shrimp abundances. Salinity regime shifts are also anticipated to create greater spatial extent of Submerged Aquatic Vegetation (SAV) habitat favorable to pink shrimp abundance (RECOVER 2005). I considered results of my investigations in the context of stated restoration salinity targets and goals: restoration of oligohaline and mesohaline salinity regimes in nearshore areas of western Biscayne Bay (RECOVER 2005).

The studies constituting this dissertation were organized into four chapters that are flanked by an introductory Chapter 1 and a concluding Chapter 6. Chapter 2 reviewed the pink shrimp literature and summarized information linking pink shrimp Essential Fish Habitat (US CFR Title 50 CH VI §600.815) metrics directly to salinity. The purpose was to integrate, summarize, and synthesize information from field, laboratory, and modeling studies. Chapter 3 investigated spatiotemporal trends, especially with respect to salinity conditions, in empirical pink shrimp density observations spanning 10 years of ecological monitoring. Assumptions that nearshore mesohaline salinity goals would yield increased pink shrimp abundance and that pink shrimp peak abundance occurs during the wet season were addressed. In Chapter 4, a controlled, laboratory study investigated salinity effects on pink shrimp molt-process growth. The study was designed to investigate pink shrimp intermolt period and molt increment, as well as other growth metrics, and survival across a salinity gradient representing the range of conditions pink shrimp may face in south Florida nearshore areas. In Chapter 5, I developed a pink shrimp habitat suitability index model, which was then applied to assess the impact of increased freshwater discharge on pink shrimp habitat along Biscayne Bay's southwestern shoreline. Quantification of predicted habitat suitability spatial extents from modeled (BBSMv4) salinity conditions simulated for a "Base Flow" and "High Flow" scenario revealed areal changes in pink shrimp habitat suitability under one of many means of achieving altered western Biscayne Bay nearshore salinity conditions.

This dissertation investigates direct responses of pink shrimp, a selected ecological indicator species for CERP, to salinity conditions. My results should be of immediate use to resource managers, scientists, and policy advocates involved with CERP. However, an important limitation of this work is its focus is on a narrow (i.e., 50 m strip of shallow-water area along the shoreline. This is due to the spatial domain of the empirical data supporting the analyses. The specific language of the stated salinity goals for southwestern Biscayne Bay refer to the area within 500m of the shoreline (RECOVER 2008), a 10 times greater area. The outcomes of this research provide insight into future impacts of restoration and freshwater management activities on southwestern Biscayne Bay coastal pink shrimp abundances. Results will also be useful to those studying juvenile pink shrimp and penaeid ecology and productivity more generally.

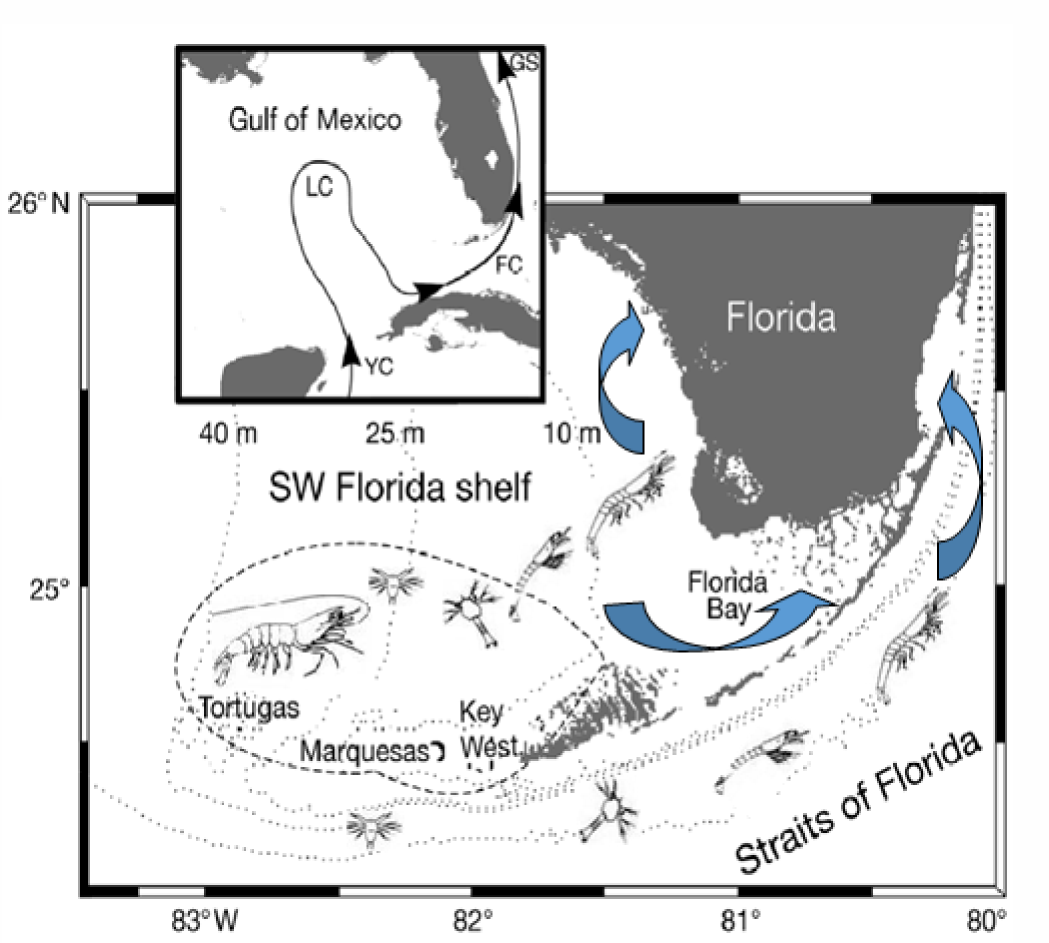


Fig. 1.1: Map of south Florida depicting Tortugas adult pink shrimp spawning grounds, larval development, and postlarval recruitment to nearshore areas, bays, and estuaries (blue arrows). Dotted lines contours depict water depth while the dashed contour encircles the area primarily recognized as adult pink shrimp habitat. The map inset depict the south Florida region within the greater Gulf of Mexico and along the Yucatan Current (YC), Loop Current (LC), and Florida Current (FC) that dominate oceanic surface flows in the region. Figure modified from Criales et al. 2006.

Table 1.1: Pink shrimp landings (lbs), number of trips, and estimated value ($) for differing Florida fisheries areas from 2016.

Year	Area Description	Landings (lbs)	Trips	Estimated Value
2016	APALACHEE BAY	323,075	140	656,715
2016	CAPE CANAVERAL	1,792	9	6,821
2016	CRYSTAL RIVER-TARPON SPRINGS	353,728	388	1,381,193
2016	DESTIN	1,898	9	3,558
2016	EVERGLADES	17,087	5	67,718
2016	FORT MYERS	44,312	132	194,825
2016	KEY WEST	918,309	96	2,163,155
2016	MIAMI	176,858	292	182,075
2016	PANAMA CITY	28,568	55	78,828
2016	PENSACOLA	1,689	12	6,191
2016	TAMPA	65,216	9	156,668
2016	TORTUGAS	5,104,868	488	11,546,681

Table 1.2: State of Florida pink shrimp fisheries landings, trips, and estimated value of landings as tabulated Florida Fish and Wildlife Conservation Commission.

Year	Landings (lbs)	Trips	Estimated Value ($)
1984	2,376,385	691	4,512,456
1985	16,018,418	6,794	30,855,155
1986	14,036,879	6,468	34,043,540
1987	12,452,296	5,517	28,969,173
1988	9,298,439	4,694	26,399,748
1989	9,254,671	4,672	23,492,747
1990	8,422,151	4,680	21,560,655
1991	8,558,833	5,078	25,404,618
1992	8,078,206	4,660	19,868,110
1993	11,420,772	4,884	27,928,880
1994	9,721,358	4,793	24,526,096
1995	14,823,997	5,379	33,264,421
1996	19,532,275	7,425	38,535,743
1997	14,273,445	9,038	36,326,292
1998	19,548,151	9,571	44,338,884
1999	10,605,264	6,603	28,274,335
2000	9,313,208	5,713	26,380,235
2001	11,312,953	5,775	29,975,006
2002	11,697,458	5,542	24,080,110
2003	11,498,126	3,947	21,552,355
2004	12,620,857	3,937	23,492,155
2005	10,902,403	3,652	20,801,071
2006	9,991,713	3,194	20,725,956
2007	5,238,355	1,578	10,234,540
2008	7,183,840	1,565	13,221,087
2009	5,833,265	1,666	10,826,854
2010	8,900,398	2,544	15,374,705
2011	6,753,641	1,367	13,924,317
2012	5,527,668	1,957	12,182,901
2013	5,473,604	1,069	13,410,593
2014	8,371,682	1,451	24,142,278
2015	7,408,972	1,632	17,286,874
2016	7,037,399	1,635	16,444,427
2017*	4,763,945	834	12,514,830

* Data incomplete for 2017

Fig. 1.2: Map of south Florida depicting CERP projects. Reproduced from USACE materials.

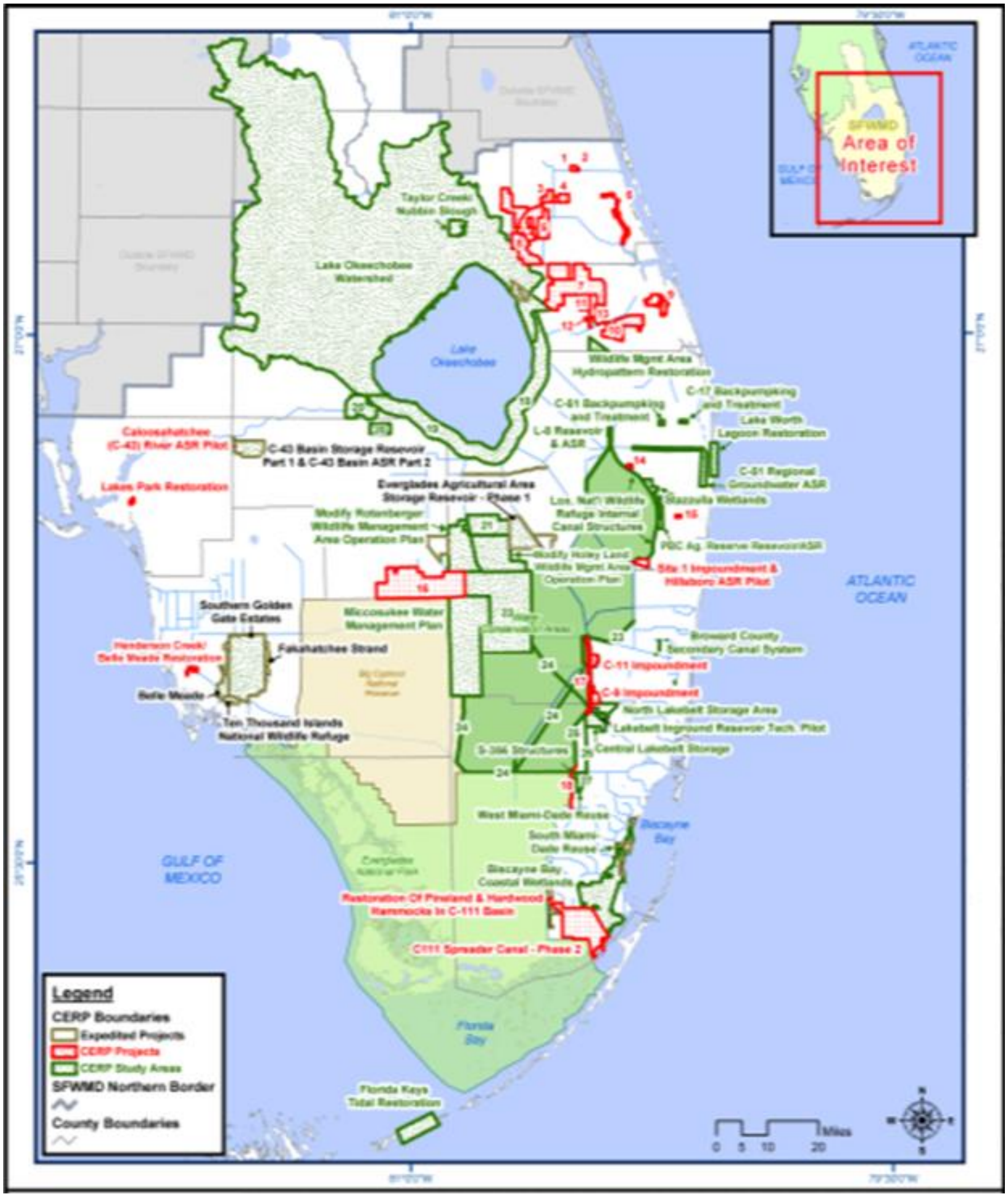
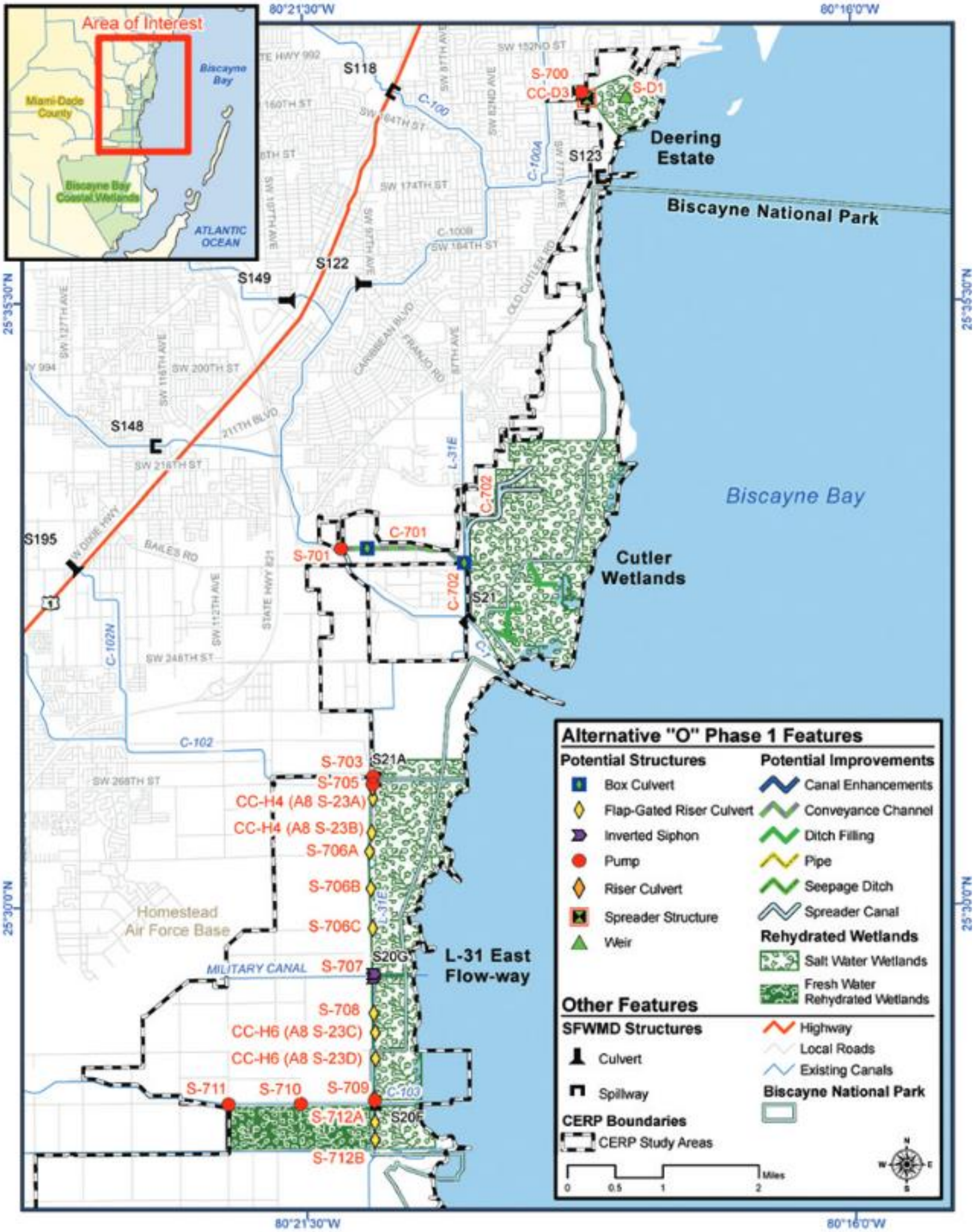


Fig. 1.3: Map of southwestern Biscayne Bay depicting BBCW features. Reproduced from USACE materials.



Chapter 2: Review of Salinity Effects on Abundance, Growth, and Survival of Nearshore Life Stages of Pink Shrimp (*Farfantepenaeus duorarum*)

2.1 Summary

The pink shrimp (*Farfantepenaeus duorarum*) has been selected as an ecological indicator to assess ecological effects on estuaries of implementation of the Comprehensive Everglades Restoration Plan that seeks to restore historical freshwater flows and nearshore salinity regimes in southern Florida. Concern over altered freshwater delivery impacts on pink shrimp productivity was expressed as early as the 1960s. The present review assessed pink shrimp scientific literature of the past 75+ years (>500 publications) to glean information relevant to understanding potential influence of freshwater management on pink shrimp productivity. The review was organized around "Essential Fish Habitat" metrics concerning abundance, growth, survival, distribution, productivity, and behavior. It summarizes previous pink shrimp field, laboratory, and modeling studies. Where possible, statistical analyses and meta-analyses of previously published data were performed to investigate consistency among independent findings. Pink shrimp occur in a wide range of salinities (0.5 to 67 ppt). A majority of studies (53.3%) reported maximal abundance between ~20 to 35 ppt salinities. One laboratory study reported maximal growth at 30 ppt. Meta-analysis of reported growth rates did not yield results due to non-convergence of regression models. Reported survival was maximal at ~30 ppt and remained high (>80% survival) across salinities of ~15 to 40 ppt. A regression model that combined survival data across studies confirmed a previously reported parabolic relationship between salinity and survival; in this regression, 35 ppt

maximized survival. Productivity, conditional upon survival and growth, was maximized at polyhaline (18-30 ppt) conditions. Inshore hypersalinity (>40 ppt) may elicit young pink shrimp behavioral cues counterproductive to settlement in nearshore areas. Virtually no information exists regarding postlarval pink shrimp movement or preference relative to salinity gradients. Realization and preservation of nearshore polyhaline conditions and elimination of hypersalinity should maximize growth, survival, and density, thus improving pink shrimp productivity. New and updated statistical models predicting pink shrimp distribution, abundance, growth, survival, and productivity relative to salinity conditions are needed to better guide freshwater management decisions.

2.2 Background

Freshwater management and restoration actions in central and southern Florida are expected to alter future nearshore salinity regimes in the region (Barnes et al. 2005, Browder et al. 2005, Crigger et al. 2005, Davis et al. 2005, Rudnick et al. 2005, Sime 2005, VanArman et al. 2005). The Comprehensive Everglades Restoration Plan (CERP) seeks to restore the quality, quantity, timing, and distribution of freshwater flows through terrestrial ecosystems and ultimately to downstream bays and nearshore areas that historically received these flows. It is expected that CERP implementation will restore nearshore salinity gradients extending perpendicular to the shoreline by expansion of nearshore mesohaline (5-18 ppt) and polyhaline (18-30 ppt) salinity habitat (Barnes et al. 2005, Browder et al. 2005, Davis et al. 2005, Rudnick et al. 2005, Sime 2005, VanArman et al. 2005). In addition, reductions in the intensity, duration, and spatial extent of hypersaline (> 40 ppt) conditions are anticipated (Browder et al. 2005, Rudnick et al. 2005, RECOVER 2008, Browder and Robblee 2009). Increased overlap of favorable salinity conditions and structured benthic habitats is expected to enrich estuarine species assemblages as well as increase abundance and distribution of individual species (Browder et al. 2005, Crigger et al. 2005, Rudnick et al. 2005, RECOVER 2008, Browder and Robblee 2009).

The pink shrimp (*Farfantepenaeus duorarum*) is one of many indicators selected to monitor ecological changes associated with CERP implementation (RECOVER 2005, Browder and Robblee 2009). Selection was by the REstoration COordination & VERification (RECOVER) multi-agency team of scientists, modelers, planners, and resource specialists. Pink shrimp were selected because: (1) they are commercially and ecologically important members of southern Florida estuarine epibenthic communities; and (2) freshwater management and restoration actions are anticipated to directly impact their distribution, abundance, growth, survival, and productivity (Browder and Robblee 2009). Both inshore bait and food fisheries (Higman 1952, Salomon 1965, Berkeley et al. 1985, Gandy et al. 2001, Johnson et al. 2012) and offshore food fisheries (Idyll 1950a, 1950b; Hart et al. 2012) have long relied on pink shrimp as an economic resource. Pink shrimp are an important trophodynamic link due to predation upon them by organisms of ecological or economic significance (Palmer 1962; Odum and Heald 1972; Johnson 1976; Overstreet and Heard 1978; Laughlin 1979; Rutherford et al. 1982, 1983). Although their reported abundance is likely influenced by sampling gear (Diaz 2001), pink shrimp are often reported to dominate epibenthic crustacean communities by number and/or biomass (Tabb et al. 1962a; Hooks et al. 1976; Livingston et al. 1977;

Subrahmanyam and Coultas 1980; Gore et al. 1981; Yokel 1983; Holmquist et al. 1989a, 1989b; Sheridan 1992; Sheridan et al. 1997; Glancy et al. 2003; King and Sheridan 2006; Rozas et al. 2012).

Like other penaeid shrimps (Dall et al. 1990), pink shrimp exhibit a life history pattern involving migration between nearshore juvenile nursery habitats and offshore adult spawning grounds, which may be spatially separated by hundreds of kilometers (Tabb et al. 1962a, Iversen and Idyll 1960, Costello and Allen 1966, McCoy 1968, Cody and Fuls 1981, Klima et al. 1987). The estuaries and bays of southern Florida are recognized as important nursery grounds for pink shrimp. Roessler and Rehrer (1971) correlated relative abundance of postlarval pink shrimp immigrating to nursery areas and subsequent commercial landings of newly recruited, small adults. Abundance of pink shrimp juveniles emigrating from nursery areas has also been linked to fishery catch of newly recruited adults (Yokel et al. 1969, Ehrhardt and Legault 1999). Similarly, it has been suggested that density-dependent limitation of adult recruitment is highly influenced by juvenile productivity (Ehrhardt et al. 2001). Marked juvenile pink shrimp released in southern Florida bays and estuaries were recaptured on the Tortugas and Sanibel shrimping grounds (Iversen and Idyll 1960, Tabb et al. 1962a, Costello and Allen 1966).

Previous studies have suggested how pink shrimp ontogenetic stages residing in estuarine and other nearshore areas can be affected by climate, land use changes, and freshwater management (Fig. 2.1) (Browder et al. 2005, Rudnick et al. 2005, Browder and Robblee 2009). However, the observation of Costello et al. (1986) that "…factors other than salinity *per se* control abundance of the euryhaline juveniles…" counters the notion of pink shrimp as a salinity-habitat indicator. For pink shrimp to effectively act as an indicator (Carignan and Villard 2002), these conflicting views need to be reconciled.

The purpose of this review was to survey the pink shrimp literature and summarize information linking pink shrimp Essential Fish Habitat (EFH) (US CFR Title 50 CH VI §600.815) metrics directly to salinity, an environmental parameter heavily influenced by freshwater management (Fig 2.1). This review integrates, summarizes, and synthesizes information from field and laboratory studies, as well as modeling results, and has immediate utility for scientists and managers involved in CERP planning and implementation. Priority was given to postlarval, juvenile, and subadult ontogenetic stages whose distribution, abundance, growth, survival, and ultimate productivity are likely to be directly influenced by alterations in freshwater management due to their occupancy of nearshore areas (Fig. 2.1). Indirect impacts on these life stages, such as alteration of predator-prey dynamics, potential exacerbation of parasitic or disease states, or salinity influences on benthic habitat conditions, were generally not considered in this review (Fig. 2.1). Once relevant articles were assembled, tabulation of study characteristics (date, location, purpose, type, and methods), observations, and results relevant to pink shrimp production (distribution, abundance, growth, mortality, productivity, and behavior) revealed the chronology and development of relevant pink shrimp research over the past ~75 years.

2.3 Methods

2.3.1 Literature Collection and Data Tabulation

Systematic literature searches were conducted of publicly-accessible literature databases: 1) Aquatic Sciences and Fisheries Abstracts (ProQuest LLC, http://search.proquest.com/asfa), 2) Web of Science (Thompson Reuters, webofscience.com), 3) ProQuest Electronic Dissertations (ProQuest LLC, search.proquest.com/dissertations), and 4) Google Scholar (Google Inc, scholar.google.com). Search terms including "*Penaeus duorarum,*" "*Farfantepenaeus duorarum*," and "pink shrimp" reflected nomenclature changes over time (Burkenroad 1939; Costello and Allen 1970; Burukovsky 1972, 1997). The resulting collection was augmented by investigation of literature cited within the studies. Other collections (National Oceanographic and Atmospheric Administration online library, Florida Fish and Wildlife Conservation Commission online library) were also searched for relevant documents. Further information came from the authors' and colleagues' personal collections. Following Faunce and Serafy (2006), "Essential Fish Habitat" (EFH) information and other relevant information were tabulated from the accumulated studies, and these were categorized by ontogenetic stage(s), shrimp size range studied, study type, and study region (North Carolina, Southeastern United States, South Florida, Gulf of Mexico East, Gulf of Mexico West, Gulf of Mexico South).

EFH habitat delimitation metrics and relevant information were reviewed by the following topics: 1) distribution/occurrence, 2) abundance, 3) growth, 4) survival, 5) productivity, and 6) behavior. Productivity was considered in the traditional ecological context as the cumulative biomass of an ecosystem; it is conditional on cumulative growth and survival rates. To facilitate inter-study comparisons, study type was characterized as field, laboratory, aquaculture, review, or modeling. Aquaculture studies generally did not provide information relevant to the present study and thus were generally excluded. Field studies were categorized by sampling design, duration, gear, diel sampling nature, and geographic location. Seasons were defined as 1) spring: March, April, May; 2) summer: June, July, August; 3) fall: September, October, November; and 4) winter: December, January, February. The Venice salinity scale was used to define salinity regimes as 1) hypersaline: >40 ppt, 2) euhaline: 30-40 ppt, 3) polyhaline: 30-18 ppt, 4) mesohaline: 18-5 ppt, and 5) oligohaline: 5-0.5 ppt (Anonymous 1958).

Pink shrimp ontogenetic stages were defined as: 1) egg, 2) larval, 3) postlarval, 4) juvenile, 5) subadult, and 6) adult. In keeping with the trend in the literature, some ontogenetic stage classifications were delimited by shrimp carapace length (CL): 1) postlarval and early juvenile: 1.5 to 7 mm CL, 2) juvenile shrimp: 7 to 17.5 mm CL, 3) subadult shrimp: 17.5 to 24 mm CL, 4) adults: >24 mm CL. These CL size bins correspond to reported size of postlarval estuarine immigration and settlement (Bearden 1961, Tabb et al. 1962a, Allen et al. 1980, Costello et al. 1986, Wenner and Beatty 1993, Diaz 2001, Criales et al. 2003, Flores-Coto et al. 2010), age/size effects on salinity range tolerance (Criales et al. 2011a, Zink et al. Unpub. Data), changes in somatic growth rate between juvenile and subadult (Diaz et al. 2001), and size at sexual maturity (Cummings 1961, Kennedy and Barber 1981). Pinks shrimp postlarvae are considered “settled” in nearshore nursery areas at 3 mm CL (Eldred et al. 1961, Costello et al. 1986). Some studies only provided total length (TL) measurements; these were converted to CL using the TL to CL relationships of Diaz et al. (2001). When neither TL nor CL were reported, ontogenetic classification relied on the authors' qualitative descriptions. Where possible, linear growth rates were calculated as final size minus initial size divided by the duration of study, time at large, or inter-sampling duration, depending upon the context. Where appropriate, linear growth rates were converted to CL growth.

2.3.2 EFH Metric Meta-Analysis

Data on pink shrimp abundance, growth, and survival from the reviewed studies were analyzed to glean broader understanding of their relationships to salinity conditions. Bayesian hierarchical probabilistic models were coded in OpenBUGS (www.openbugs.net) and executed using two Markov Chain Monte Carlo (MCMC) simulations and uninformative priors. MCMC chains were run until model convergence was observed. Unless otherwise explicitly mentioned, model fitting diagnostics and selection values were computed by the software. Vagueness of priors was affirmed by graphical investigation of posterior parameter value density plots. Posterior parameter estimate convergence was checked graphically via Brooks-Gelman-Rubin diagnostic, quantile, acceptance rate, autocorrelation, and jump rate plots (Lunn et al. 2013, Spiegelhalter et al. 2014a). Ratios of Monte Carlo error to parameter estimate standard deviation less than 1% were used to confirm convergence (Lunn et al. 2013).

Model fit was verified by investigation of residual vs. fitted observations plots, standardized residuals plots, and normality and variance statistical properties of residuals (Kéry 2010). Bayesian p values and χ^2 discrepancy plots were calculated to assess model fit (Kéry 2010). Ratio of variance explained (i.e., R^2) was also computed. Deviance Information Criterion (DIC) was used to select "best" models from a suite of similar formulations. DIC penalizes model "fit" (i.e., posterior mean deviance: $\overline{D}$) by its "complexity" formulated as an "effective number of parameters" (*pD*) (Spiegelhalter et al. 2002, 2014b). Posterior parameter estimates were considered significant if their 95% credible intervals (CI) did not include zero (McCarthy 2007, Kéry 2010, Lunn et al. 2013).

Summary abundance data presented by Minello (1999) relative to both salinity regime (n = 3: polyhaline, mesohaline, and oligohaline) and habitat (n = 4: seagrass, *Spartina alterniflora* marsh edge, mixed spp. vegetation marsh edge, and non-vegetated) was deemed sufficient for further analysis. A mixed effects ANOVA model was formulated to treat salinity regime as a fixed effect and habitat conditions as "nuisance" random effects (Bolker et al. 2008). To accommodate the three salinity treatment ontrasts, an "effects" ANOVA (sensu Kéry 2010) was constructed with oligohaline condition as the "base" treatment and both mesohaline and polyhaline conditions as "indicator" parameters. Meta-analysis was achieved by incorporating each reported mean density's respective standard error estimate (N = 12, one for each habitat and salinity regime combination) as input error (i.e., fixed precisions) of estimated posterior mean values. Models were considered convergent after 100,000 MCMC iterations; simulations were carried out for another 100,000 iterations to estimate parameter posterior probability densities.

Accumulated growth and/or survival rate data either appearing in the original publication (Zein-Eldin 1963, Zein-Eldin and Aldrich 1964, Browder et al. 1999, Rand and Bachman 2007, Zink et al. 2013) or provided by the author (Browder et al. 2002) were assimilated across studies and various salinity treatment conditions via Bayesian mixed effects linear models. Model formulations included fixed linear and quadratic salinity terms as well as a random categorical "nuisance" term to account for inter-study variability (Bolker et al. 2008). Models were considered convergent after 200,000 MCMC iterations; simulations were carried out for another 100,000 iterations to estimate parameter posterior probability densities.

To obtain exponential growth-in-length coefficients from Browder et al. (2002), CL size data were natural-log transformed and regressed against day of study. Per the methods used by Browder et al. (2002), only growth or survival data associated with "extreme" salinity conditions (i.e., ≤ 10 ppt or ≥ 45 ppt) were used in this analysis if acclimation to the test salinity had occurred. After removal of negative growth coefficients and treating primary sampling units as independent observations, the process yielded 1084 growth coefficients. To obtain an average survival per salinity treatment from Browder et al. (2002) data, survival was averaged across primary sampling units within each salinity treatment of a specific study trial (n = 10), yielding 139 survival observations (see Browder et al. 2002). Similarly, wet weight (WWT) growth data from Zein-Eldin (1963: Table 3) also required natural log transformation and regression against day of study, which yielded 5 growth rates. Due to slight differences in study conditions (e.g., feeds, stocking density, and initial size), data from the separate months (n = 2) of the Zink et al. (2013) study were treated as independent, thus yielding 6 growth coefficients per month (N = 12). Other survival and growth data were used directly as reported (e.g., Table 2: Browder et al. 1999; Figure 3: Zein-Eldin 1963; Table 4: Rand and Bachman 2007).

2.4 Results and Discussion

2.4.1 Overview

The literature search revealed over 500 studies, including peer-reviewed papers, dissertations, theses, reports, abstracts, conference proceedings, and other documents that provided pink shrimp biological and ecological information. The accumulated literature was reduced to 255 works by screening out studies not relevant to understanding potential impacts of freshwater change on nearshore ontogenetic stages (see Supplementary Materials for complete list of considered literature). Within this count, 68.6% were peer-reviewed journal articles and other literary sources (e.g., technical memorandums, dissertations, and theses). With further scrutiny, a subset of these studies (i.e., those cited throughout the remainder of this text) was found to provide the most relevant information.

The 255 selected studies spanned the entire pink shrimp geographic range but were unequally distributed among regions ($\chi^2 = 285.46$, df = 7, $P < 0.0001$) (Table 2.1, Fig. 2.2). The SF region has been the most productive pink shrimp fishing area (Hart et al. 2012) and is the most highly represented region (n = 122, 47.4%) (Table 2.1, Fig. 2.2). An increase in studies during the 1960s and 1970s was likely prompted by CPUE declines observed (Iversen and Jones 1961) a decade after the discovery and rapid expansion of the Tortugas and Sanibel pink shrimp fisheries (Idyll 1950a, 1950b). Tabb (1962a) was among the first to express concern that alteration of freshwater delivery to SF estuaries and coastal bays likely contributed to those declines.

At least one type of EFH information was provided in 65.6% (n = 165) of the studies (Table 2.1). Density information was the most often reported EFH metric (n =

87, 34.1%). Distribution/occurrence, growth, survival, or productivity metrics were each supported by less than 25% of reviewed studies, and 29% contained behavior information (Table 2.1). Field studies dominated study type (n = 169, 66.3%), followed by laboratory studies (n = 66, 25.9%), computer simulation or modeling studies (n = 34, 13.3 %), and descriptive or review studies, classified as "other" (n = 12, 4.7%: Table 2.1). Here, focus is on field and laboratory studies that provided primary empirical evidence of salinity effects on pink shrimp EFH metrics.

Most field studies (n = 96, 56.8%) relied on fixed sampling designs (Table 2.2). More than half of field studies (n = 96, 56.2%) were less than 2 yr duration, while fewer were of intermediate (≥ 2 and 5 ≤ yr) duration (n = 26, 15.4%) or longer (>5 yr) duration (n = 17, 10.1%). Longer duration studies (>5 yr) were generally related to fisheries CPUE and landings analyses. The most commonly used sampling gears were trawl nets (45.6%) and throw traps (11.8%) (Table 2.2). Size selectively and efficiency differences between these two gear types complicates direct comparison between them (Browder et al. 2003, Campos and Berkeley 2003). Almost half of the studies (48.5%) did not clearly report whether sampling was diurnal, nocturnal, or during both times (Table 2.2). Pink shrimp exhibit nocturnal activity, as observed by fishing and field studies (Woodburn et al. 1957, Ingle et al. 1959, Eldred et al. 1961, Idyll et al. 1964) and later confirmed via controlled field and laboratory observational studies (Fuss 1964; Fuss and Ogren 1966; Wickham 1967; Hughes 1968, 1969a; Wickham and Minkler 1975; Subrahmanyam 1976; Reynolds and Casterlin 1979). Diel sampling could affect study results and reduce the clarity of inter-study comparisons (Brusher and Ogren 1976, Trent et al. 1976, Ault et al. 1999a, Diaz 2001, Sánchez 1997, Ogburn et al. 2013).

2.4.2 Distribution / Occurrence

This review identified 25 studies that contained occurrence or distribution information of postlarval, juvenile, or subadult ontogenetic stages. These included 23 field and 2 laboratory studies. The geographic range of pink shrimp extends south from Chesapeake Bay, USA, along the eastern coastline of the United States and throughout the Gulf of Mexico to Isla Mujeres, Quintana Roo, Mexico, including nearby insular areas such as Bermuda, Bimini, and Cay Sal Bank (Saloman et al. 1968, Perez-Farfante 1969, Perez-Farfante and Kensley 1997) (Fig. 2.3). Thus, pink shrimp are potentially exposed to a wide range of temperatures. They have been caught from 5 to 38 °C (Williams 1955a, 1955b; Copeland and Bechtel 1974). Adult pink shrimp are distributed across wide areas of the continental shelf (Idyll 1950c, Bur. Comm. Fish. 1961, Costello and Allen 1966, Salomon et al. 1968, Kennedy and Barber 1981, Darcy and Gutherz 1984, Roberts 1986, Ramírez-Rodríguez 2002) to depths down to 300m (Bur. Comm. Fish. 1961, Salomon et al. 1968, Huff and Cobb 1979), but are most abundant between 9 and 65 m (Kutkuhn 1962, Huff and Cobb 1979).

While many studies reported pink shrimp occurrence at various salinities, few (n = 9) reported occurrence information in terms of salinity regime and ranges. Pink shrimp can occur in a wide range of salinities; they have been collected from waters described as "…without measurable salt content…" (Tabb et al. 1962a) and as low as 0.5 ppt (Gunter and Hall 1965, Williams and Deubler 1968) and 0.65 ppt (Joyce 1965) or 0.7 ppt (Loesch 1976). At the other extreme, they have been collected from waters up to 60 ppt (Hedgpeth 1967), 65 ppt (Gunter et al. 1964), and even 67 ppt (Simmons 1957). Statistical analysis of pink shrimp occurrence has resulted in maxima at ~25 ppt across wet and dry seasons (Lirman et al. 2014a) and ~17 ppt during the summer (Rubec et al. 2016). Costello et al. (1986) concluded that, within an estuary, salinity regime alone does not necessarily limit pink shrimp distribution. Rapid and extreme salinity fluctuations due to high riverine (or canal) discharge events have been postulated to elicit migrations to and from a coastal system, resulting in sporadic and transitory pink shrimp occurrence in places with highly variable salinities (Ingle 1957).

Pink shrimp move away from lower, variable salinities towards higher, less variable salinities as they develop into subadults (Burkenroad 1934, Gunter 1950, Williams 1955b, Tabb et al. 1962a, Gunter et al. 1964, McCoy and Brown 1967, Brusher and Ogren 1976, Diaz 2001). For example, Tabb et al. (1962a) reported collecting pink shrimp ranging from 3 to 28 mm CL from 5 to 47 ppt and only larger shrimp (28-32 mm CL) from 25 to 45 ppt waters. Similarly, Gunter et al. (1964) noted catch composition mean size ($\bar{X}$) increased at higher salinities (0-29.9 ppt: $\bar{X} \approx 13$ mm CL, ≥30.0 ppt: $\bar{X} \approx 18.4$ mm CL). I analyzed salinity and mean size information presented by Brusher and Ogren (1976) and found a significant, positive correlation with mean salinity ($r_{n=5} = 0.9828$, $p = 0.0027$) and a significant, negative correlation with salinity range ($r_{n=5} = -0.9184$, $p = 0.0276$).

Pink shrimp habitat selection depends on interacting factors such as depth, salinity regime, shrimp abundance, and ontogenetic stage and/or size. Diaz (2001) reported a high nocturnal proportion of occurrence (≥ 0.78) of postlarval and juvenile pink shrimp in intertidal (<1 m depth) and subtidal (1-2 m depth) locations in variable and lower salinity regime seagrass beds during summer and fall seasons. Subadults were virtually absent from these locations (Diaz 2001). Diaz (2001) did not observe any postlarvae in near-oceanic, stable salinity regime intertidal seagrass areas (annual $\bar{X}$ = 34 ppt), although others suggested postlarvae prefer any shallow, low-energy seagrass beds (Eldred et al. 1961, Costello et al. 1986; Murphey and Fonseca 1995) where current regimes are capable of delivering them (Hovel et al. 2002). Subadults were generally collected from deeper *T. testudinum* habitats (Eldred et al. 1961, Pitre, 2001) and deeper inshore waters (Eldred et al. 1961, Costello et al. 1986, Ault et al. 1999a, Diaz 2001) at depths <4 m (Rubec et al. 2016).

2.4.3 Density / Relative Abundance

A total of 59 studies contained density or relative abundance information on postlarval, juvenile, or subadult ontogenetic stages. Most (n = 56) were field studies, although 5 studies investigated habitat preference and relative abundance in laboratory settings. Fewer (n = 18) reported salinity-density statistical relationships of relevance to this review. Some investigators considered salinity to have a minimal influence on pink shrimp abundance in estuaries. Hoese (1960) noted that habitat conditions other than salinity regime can drive juvenile pink shrimp abundance, reducing the influence of salinity on habitat selection. Costello et al. (1986) suggested that salinity regime was less important than other environmental factors.

Early pink shrimp studies reported that salinity regime influenced pink shrimp abundance (Hildebrand 1955, Gunter 1961, Gunter et al. 1964). Most studies summarized in Table 2.3 showed significant negative (n = 5), positive (n = 8), parabolic (n = 1), and spline (n = 1) associations to observed salinity ranges. A few studies (n = 3) reported no significant relationship (Table 2.3). A plot of the significant abundance trends against each trend's corresponding observed salinity range revealed that 8 of the 15 (53.3%) were maximized between salinities of ~20 to 35 ppt (Fig. 2.4). Trends in the remaining 7 studies indicated maxima at either lower (n = 3) or higher (n = 4) salinities (Fig. 2.4). In two studies that reported pink shrimp maximal abundance at low salinity (Howe and Wallace 2000, Robblee et al. 2008), the shrimp were associated with *Ruppia maritima*, a seagrass species usually confined to oligohaline and mesohaline conditions, which provided the only seagrass habitat available in the area (Tabb et al. 1962b, Fourqurean et al. 2002, Merino et al. 2009). These two studies support the conclusions of Hoese (1960) and Costello et al. (1986) that salinity makes little or no difference to pink shrimp abundance. In contrast, Rubec et al. (2016) studied salinities ranging from 0 to 35 ppt, finding a maximal concentration from ~0 to 15 ppt and a distinct density maxima at ~17 ppt in an ecological system where seagrasses occurred under all salinity condition and thus may reflect true selection for high mesohaline and lower polyhaline salinity conditions. These relationships were developed for summer pink shrimp abundances, when a wider range of salinity habitat was present (Rubec et al. 2016).

For those reporting a maximal abundance trend at higher salinities (i.e., > ~35 ppt), two investigated wide salinity ranges >30 ppt (Perez-Castañeda and Defeo 2001, Robblee et al. 2008), which might have been better investigated by fitting parabolic rather than linear assumptions of abundance relative to salinity (Table 2.3, Fig. 2.4). Two studies (Wakida-Kusunoki et al. 2008, Blanco-Martínez and Pérez-Castañeda 2016) were conducted in the Laguna Madre ecosystem, a location dominated by euhaline and hypersaline conditions and virtual nonexistence of polyhaline or lower salinities (Hildebrand 1969). Laguna Madre pink shrimp salinity relationships were likely complicated by late fall and early winter recruitment maxima coincident to the early dry season, when salinity was increasing (Wakida-Kusunoki et al. 2008, Blanco-Martínez and Pérez-Castañeda 2016).

Four field studies provided sufficient density and salinity data to allow categorization by salinity regime (Sheridan 1992, Rozas and Minello 1998, Minello 1999, Sheridan and Minello 2003). However, experimental control of salinity condition in three of these studies eliminated the confounding influence of salinity on the studies' focus of habitat type (Sheridan 1992, Rozas and Minello 1998, Sheridan and Minello 2003). Therefore, I included only the data reported by Minello (1999) in my analysis of pink shrimp mean densities in relation to salinity. The resulting model (Table 2.4) revealed significant higher density in polyhaline and mesohaline regimes relative to oligohaline regimes but no statistical difference between polyhaline and mesohaline effects (Table 2.5). Substantial ΔDIC reductions revealed the importance of including both the standard error of mean density estimates and the random habitat effect (Table 2.4). The final selected model exhibited adequate fit (Bayesian $p = 0.588$), but only explained approximately 16% of the data variability (Table 2.4). Model residuals were non-normally distributed (Shapiro-Wilk test: $W = 0.7681$, p value = 0.00416 with heterogeneous variances between random habitat groups (Bartlett's Test: $K^2 = 15.18$, d.f. = 3, p value = 0.00167). However, the balanced design of this analysis may tolerate moderate departures from ideal conditions (Zar 2010). Counter to the previous graphical analysis of abundance trends (Fig. 2.4), these results extended maximal pink shrimp density into mesohaline conditions.

Qualitative observations confirm the preceding statistical analysis of Minello (1999) data. While they did not provide data that allowed graphical presentation or inclusion in meta-analyses, many studies reported peak pink shrimp abundance at polyhaline (Gunter et al. 1964, Gunter and Hall 1965, Copeland and Bechtel 1974, Weinstein et al. 1980, Gracia and Soto 1990, Zimmerman et al. 1990, Minello 1999, Perez-Castañeda and Defeo 2003a) or mesohaline conditions (Gunter 1950, Loesh 1976, Flores et al. 1996, Rubec et al. 2016). Browder and Robblee (2009) discussed prior findings of predicted lower pink shrimp abundances at salinities <20 ppt (Browder et al. 2002). Pink shrimp also occur in oligohaline areas, although not very abundantly (Tabb et al. 1962a; Darnell and Williams 1956; Odum 1970; Loesch 1976; Gracia and Soto 1990; Minello 1999; Pérez-Castañeda and Defeo 2001, 2004; Robblee et al. 2008).

Amplitude and frequency of salinity fluctuation can also affect pink shrimp abundance. Higher abundance has been qualitatively attributed to areas exhibiting stable, higher salinity areas relative to nearby lower mean, variable salinity regimes (Berkeley and Campos 1984, Ault et al. 1999a, Diaz 2001, Campos and Berkeley 2003). Salinity variability can be negatively related to pink shrimp abundance (Berkeley and Campos 1984). Salinity regimes within estuaries and bays are dependent on terrestrial freshwater inflows, including seasonally-varying localized precipitation runoff, riverine discharge, and aquifer (groundwater) seepage (Copeland 1966, Browder and Moore 1981, Sklar and Browder 1998, Browder et al. 2005). Intra- and inter-annual pink shrimp abundance has been positively associated with localized precipitation (Browder 1985, Gracia and Soto 1990, Rice 1997, Sheridan 1996, Browder et al. 1999).

2.4.4. Growth

Fifty-five publications contained growth information for postlarval, juvenile, or subadult ontogenetic stages. A total of 1 review, 26 laboratory, and 28 field studies reported relevant growth information Only four studies, all conducted in the laboratory, directly investigated growth rates relative to salinity (Table 2.6), even though) salinity is a paramount factor affecting estuarine organismal growth (Kinne 1964, 1967). Only a few of the 17 field studies reporting relevant growth rates provided accurate salinity conditions. For those that did so (n = 7), interpretation was complicated by the generally broad salinity ranges reported (> ~10 ppt: Table 2.7) and inclusion of multiple ontogenetic stages.

Comparison of growth rates was complicated by differences in the reported functional growth trajectories: 3 laboratory studies reported exponential growth rates while 1 reported linear rates (Table 2.6). A total of 17 field studies reported comparable growth rates, all of which were linear (Table 2.7). Although many von Bertalanfy-type growth rates were identified during the literature review, only a three included relevant ontogenetic stages (Kutkuhn 1965, Klima et al. 1987, Campos and Berkeley 2003). These three studies reported maximal size parameter estimates (L_∞ or W_∞) that were at odds with empirically-observed maximum sizes (50.0 to 55.0 mm CL) reported elsewhere (Iversen and Idyll 1960; Iversen and Jones 1961; Costello and Allen 1966, 1968; Berry 1970; Fontaine and Neal 1971). Further complicating comparisons, some laboratory studies reported growth rates as changes of either TL or CL, whereas others reported changes in weight (Table 2.6). Field studies only presented growth in terms of changes in length (Table 2.7). Differing culture conditions affected comparison of reported laboratory growth rates (Table 2.6).

The mechanistic growth of crustaceans follows a stair-step trajectory consisting of ecdysis (i.e., molting, shedding of the exoskeleton) and longer duration inter-molt periods of tissue deposition and physiological preparation for the act of molting (Drach 1939, Hiatt 1948, Scheer 1960, Dall et al. 1990). Three studies investigated molt-process growth in pink shrimp; none considered differing salinity conditions. While salinity influences on pink shrimp inter-molt period and/or growth-per-molting event have not been studied, salinities resulting in maximal growth of penaeid shrimps have been observed to minimize inter-molt periods (Staples and Heales 1991, O'Brien 1994, Vijayan and Diwan 1995) and maximize growth-per-molt increments (Staples and Heales 1991, O'Brien 1994, Vijayan and Diwan 1995).

The most extensive pink shrimp growth study I identified reported a parabolic relationship with salinity (Browder et al. 2002). Across an experimental range of 2 to 55 ppt, maximal growth was observed at 30 ppt (Browder et al. 2002). Zink et al. (2013) found reduced growth at 55 ppt treatment conditions, but similar growth under 15 and 35 ppt conditions. Rand and Bachman (2007) studied growth in salinity treatments of 2 to 50 ppt and revealed significant influences on WWT, but post-hoc testing did not reveal significant comparisons. Unfortunately, two studies (Zein-Eldin 1963, Zein-Eldin and Aldrich 1964) were hindered by difficulty with species identifications, resulting in admixtures of *F. duorarum* and *F. aztecus*, thus complicating reported results. However, maximal growth was observed at 25 ppt (Zein-Eldin 1963) although "…growth occurred at a wide salinity range" (Zein-Eldin and Aldrich 1964).

Coefficients of exponential growth in length (Fig. 2.5) or weight (Fig. 2.6), plotted against their treatment salinities, revealed substantial differences among studies. Use of hierarchical mixed effects models to develop growth regressions that integrated across studies was unsuccessful. Inter-study differences may have been too great to allow determination of a collective growth rate relationship relative to salinity condition. Dissimilar experimental conditions, including mean initial size (CL mm), stocking density (shrimp L^{-1}), diet, shrimp source, and temperature treatment levels, likely contributed to inter-study variability (Table 2.6, Fig. 2.5, 2.6).

Laboratory growth study results may have been confounded by crowding too many individuals, or even a single one of too large a size, into an unsuitably small space. Several reviewed growth studies acknowledged crowding issues (Eldred et al. 1961, Zein-Eldin 1963, Teinsongrusmee 1965, Caillout et al. 1972, Browder et al. 2002, Zink et al. 2013). Eldred et al. (1961) noted "…there are indications that crustaceans may molt with little or no growth when confined to small containers." Alternatively, growth variation within a study, at least in part, could result from differential mortality and subsequently altered biomass density (Zein-Eldin 1963, Teinsongrusmee 1965, Gullian et al. 2010, Zink et al. 2013).

Only one study has investigated stocking density influences upon pink shrimp growth; the lowest density treatment resulted in maximal growth (Gullian et al. 2010). Comparison of exponential growth of WWT coefficient values from Gullian et al. (2010: 0.036 at 5 ppt and 23.5 - 26.6 °C) to similar, but lower, values reported by Zink et al. (2013: 0.0316 at 32 ˚C and 15 ppt), was complicated by inter-study differences in stocking density, temperature, salinity, and other potentially influencing conditions.

Field studies have also reported both intra- and inter-specific negative population density effects on growth (Álvarez et al. 1987; Pérez-Castañeda and Defeo 2002, 2005).

Field studies contributed a more general understanding of potential freshwater management effects, but precision in their results was confounded by lack of controlled study conditions and concurrent consideration of multiple ontogenetic stages. Only one field study of pink shrimp growth rates directly investigated salinity effects on pink shrimp growth and did not find a significant influence (Pérez-Castañeda and Defeo 2005). However, a previous study from the same ecosystem found that condition factor (i.e., the exponent of length-weight power functions) was higher in the higher salinity regime studied (≥30 ppt) (Pérez-Castañeda and Defeo 2002). Williams (1969) postulated that mesohaline waters were suitable for postlarval pink shrimp growth and concluded "…oligohaline upstream stations [were] apparently too fresh for regular utilization."

Plotting of linear field growth rates across their associated salinity ranges allowed for direct comparison to similar rates from laboratory studies (Fig. 2.7). The overlap of higher laboratory growth rates with most field rates (81.8%) suggests a congruence between them (Fig. 2.7). However, two field growth rates were double the value of others or of typical laboratory growth rates (Williams 1955b, Álvarez et al. 1987). Given the previously mentioned agreement in values, these two growth rates were either erroneous or else they provide an estimate of maximum attainable growth capacity.

2.4.5 Survival / Mortality

A total of 32 studies provided survival and/or mortality information for the postlarval, juvenile, and subadult ontogenetic stages. Of those, 17 were conducted in laboratory settings and 12 were obtained from field studies. Within laboratory studies, 6 investigated longer-term (10 d ≥ and ≤ 30 d; Table 2.8) and 3 tested short-term (i.e., abrupt: ≤ 4 d; Table 2.9) salinity exposure. Of the reviewed field studies, 3 investigated *in situ* mortality rates (Álvarez et al. 1987; Pérez-Castañeda and Defeo, 2003b, 2005). Although these field studies do not report salinity conditions, they do provide natural mortality rates that could be adjusted by laboratory-reported salinity effects on survival for incorporation into production modeling. Eight studies included modelling of survival, either alone or in combination with field and laboratory studies. However, modelling studies were of little utility given their basis on laboratory survival experimentation or use of more generalized survival rates that did not consider salinity influences. While many fishery studies reported natural and fishing mortality estimates, these were not further considered due to lack of salinity or relevant ontogenetic information.

Kinne (1964, 1967) considered salinity effects of paramount importance to understanding natural mortality of estuarine invertebrates. Longer-term laboratory mortality studies reported that survival/mortality was affected by salinity condition. Two laboratory studies (Browder et al. 1999, 2002) and one field study (Pérez-Castañeda and Defeo 2005) included sufficient salinity information to suggest a parabolic response.

High survival over a wide salinity range was reported from several laboratory studies (Zein-Elden 1963; Zein-Elden and Aldrich 1964; Browder et al. 1999, 2002; Rand and Bachman 2007; Zink et al. 2013). For example, Browder et al. (2002) reported survival >80% in salinities ranging from 15 to 40 ppt; similarly, Zink et al. (2013) reported highest survival in 15 and 35 ppt treatments. Browder et al. (2002) reported maximum survival at 30 ppt, which was consistent with reported isosmotic values of ~ 26 to 28.9 ppt (Williams 1960, Bursey and Lane 1971, Castille and Lawrence 1981, Bonilla-Gómex et al. 2012). Hypersaline (>40 ppt) conditions can cause drastic survival reductions (~40 – 90%) relative to polyhaline/euhaline conditions (Browder et al. 1999, 2002; Zink et al. 2013). Although one field study (Perez-Castaneda and Defeo 2005) reported a parabolic survival response to survival, equation coefficients were not reported, precluding computation of a salinity maxima.

I used laboratory survival data presented directly by authors or calculated from their data (Zein-Eldin 1963; Browder et al. 1999, 2002; Rand and Bachman 2007; Zink et al. 2013) in a mixed-effects hierarchical Bayesian regression model relating survival to treatment. The selected final model included a random effects factor for study (Table 2.10). Only one study (Zein-Eldin 1963) significantly differed from the others (Table 2.11). This difference may be due to use of *F. aztecus* and *F. duorarum* during experimentation due to species identification complications. Inclusion of a "nuisance" parameter random study effect substantially lowered DIC and increased R^2 values (Table 2.10). A significant parabolic survival-salinity condition relationship was detected (Table 2.10, Fig. 2.8), which reinforces similar previous independent observations (Browder et al. 1999, 2002). The model predicted a maximal survival at 35 ppt and high survival (>80%) from ~20 ppt to ~45 ppt (Fig. 2.8). This maximum and range were both greater than previously reported (Browder et al. 1999, 2002). Differing conditions, such as diet, stocking density, shrimp size and source, and study duration, likely contributed to intra-study variability (Table 2.8).

Studies of effects of abrupt (≤ 4 d) salinity change on pink shrimp survival (Table 2.9) generally agreed with the salinity ranges reported in longer-exposure (10 d ≥ and ≤

30 d) studies (Williams 1960, Zein-Eldin and Aldrich, 1964, Criales et al., 2011a). This is interesting given that longer-exposure studies generally include acclimation to extreme salinities (Browder et al. 2002, Zink et al. 2013). Pink shrimp exhibited high tolerance (i.e., survival) to abrupt changes of salinity conditions ~15-20 ppt different from ambient (Williams 1960, Criales et al. 2011a). More extreme changes in salinity can lead to total mortality (Zein-Eldin and Aldrich 1964, Criales et al. 2011a). Mortality related to abrupt salinity changes generally ceased after 24 hr (Criales et al. 2011a). This observation agrees with earlier osmoregulatory studies, which found restoration of homeostasis ~24hr after exposure to a salinity different from ambient (Bursey and Lane 1971, Castille and Lawrence 1981). Evidence of shifts in salinity tolerance range related to postlarval ontogeny and acclimation salinity (Criales et al. 2011a) suggest the importance of the salinity condition to which postlarvae are exposed during settlement, which may act as a physiological imprinting at an especially receptive stage of development.

Interaction of temperature and salinity on mortality has been reported (Zein-Elden and Aldrich 1964; Browder et al. 1999, 2002). Specifically, combinations of low salinity and high temperature or, conversely high salinity and low temperature, were especially detrimental to pink shrimp survival (Browder et al. 2002). Reduced osmoregulatory capabilities have been experimentally observed after rapid (48 and 96 hr) reductions in temperature (Williams 1960). “Broken-back syndrome”, resulting from the cumulative stress of handling, low temperature, and low salinity conditions, was presumed to be caused by muscular swelling during osmotic stress and extensive abdominal flexures when displaying escape behaviors (Couch 1978). Complete mortality was reported for captive shrimp exposed to rapid salinity reductions (from 15-18 down to 3 ppt) in conjunction with relatively cold temperatures (8 °C) (Couch 1978).

2.4.6 Productivity

A total of 23 studies contained information on pink shrimp productivity at relevant ontogenetic stages. Ten of these provided information relevant to freshwater management. Four (Browder 1985, Sheridan 1996, Ehrhardt and Legault 1999, Ehrhardt et al. 2001) reported field-measured productivity, five (Ault et al. 1999b; Browder et al. 1999, 2002; Ramírez-Rogríquez et al. 2003, 2006) modeled productivity, and one (Zink et al. 2013) investigated potential productivity in laboratory settings relative to salinity or freshwater flow conditions.

Relationships between productivity of nearshore ontogenetic stages and later fisheries landings have been observed (Yokel et al. 1969, Roessler and Rehrer 1971, Ehrhardt and Legault 1999, Ehrhardt et al. 2001, Johnson et al. 2012). Early investigations of pink shrimp ecology postulated linkages between upland freshwater conditions, related coastal discharges, and pink shrimp productivity (Tabb et al. 1962a). Ecological conditions experienced during early life stages, such as salinity in nearshore pink shrimp nursery habitats, have been linked to pink shrimp production. Browder (1985) was the first to directly link offshore adult fishery landings to freshwater management. Regression analysis that included terrestrial water levels explained substantial amounts (i.e., 30 to 68%) of quarterly and annual landings variability not explained by fishery effort (Browder 1985). These relationships were later substantiated via linkage of both rainfall patterns and terrestrial water levels to pink shrimp annual landings (Sheridan 1996) and fishery recruitment of subadults (Ehrhardt and Legault 1999, Ehrhardt et al. 2001). In Biscayne Bay, Florida, fishers have reported that unseasonably low freshwater discharges were detrimental to inshore pink shrimp fisheries (Berkeley et al. 1985).

Laboratory study of the effect of salinity regime on pink shrimp productivity has been limited. The only study found statistically lower potential productivity under hypersaline conditions (55 ppt) (Zink et al. 2013); whereas, euhaline and mesohaline (35 and 15 ppt, respectively) treatments provided similar higher potential productivity (Zink et al. 2013).

Model simulations suggested substantial influence of salinity regime on pink shrimp productivity (Browder et al. 1999, 2002). The productivity scenarios incorporated empirical salinity conditions with laboratory-based salinity relationships on growth and survival (Browder et al. 1999, 2002). Extreme salinities, particularly hypersalinity, reduced productivity potential, whereas, higher freshwater inflows associated with higher rainfall increased the overlap of favorable salinity and seagrass conditions and maximized potential production (Browder et al. 1999, 2002). To the advantage of overall production, detrimental extreme low salinity conditions coincided with areas where pink shrimp postlarval recruitment was minimal (Browder et al. 2002). Counter to these results, virtual population modeling of pink shrimp offshore recruitment was positively correlated both with salinities (range of study: 22 – 38.5 ppt) experienced three months prior to recruitment (Ramírez-Rogríquez et al. 2003) and with annual average salinity (range of study: 29 – 38 ppt) (Ramírez-Rogríquez et al. 2006). Ault et al. (1999b) modeled pink shrimp productivity, which incorporated salinity into natural mortality rates; however, close examination of the relationship revealed a single, coarse scaling metric that included broad salinity conditions (i.e., < or > 34 ppt) as a modifier of natural mortality.

2.4.7 Behavior

Although 55 studies were identified that contained behavioral information for relevant ontogenetic stages, most were not applicable to freshwater management alterations. However, one highly pertinent behavioral topic was selective tidal stream transport (STST), used by both pink shrimp postlarvae for immigration into nearshore areas and by subadults for emigration to offshore waters (Criales et al. 2006, 2007, 2010, 2011b; Flores-Coto et al. 2010). Three laboratory studies were identified that investigated habitat influences on STST.

Pink shrimp have long been thought to selectively utilize tidal currents to facilitate migration transport as well as to maintain their position within tidally influenced estuaries (Hughes 1969b, 1969c). Laboratory study has demonstrated that STST activity is mediated by alterations of salinity (Hughes 1969b, 1969c) and turbulence (Criales et al. 2013) regimes. Specifically, higher salinity (> 32.5 ppt) currents elicit positive rheotactic movement; conversely, exposure to salinity reductions as little as 2 ppt will elicit juvenile drift or active swimming with the current (Hughes 1969b, 1969c). Exposure of active postlarvae to declines in salinity of as little as 2 to 3 ppt usually caused them to settle to the substrate (Hughes 1969b), although not consistently (Hughes 1969c). Degree of behavioral response was dependent on rate of salinity change; while low rates (-3 ppt hr^{-1}) did not elicit a response, larger reductions in salinity (10-15% from previous conditions) did so (Hughes 1969b). Hughes (1969c) found minor postlarval size differences altered behavioral patterns: smaller postlarvae (1.7 - 2.1 mm CL) were less likely to penetrate a 34 to 30 ppt salinity discontinuity barrier than were larger postlarvae (2.2 - 2.6 mm CL). Pink shrimp postlarvae may be able to detect salinity differences as low as 1 ppt (Hughes 1969c).

Alterations in salinity regime could influence swimming behaviors and effective immigration, settlement, and maintenance within nursery areas. Indeed, Hughes (1969b, 1969c) expressed concern that altered salinity regimes (i.e., negative estuarine salinity gradients with interior waters of higher salinity) could provide misleading behavioral stimuli to both juvenile and postlarval pink shrimp. Criales et al. (2010) speculated that observed positive salinity gradients from oceanic to nearshore waters (i.e., inshore hypersaline conditions) could expose immigrating postlarvae to a salinity regime that discouraged further progress into the estuary.

2.5 Conclusions

Maintenance of adequate freshwater inflows to estuaries and nearshore areas is necessary to sustain high productivity in these areas (Moore et al. 1970, Browder and Moore 1981, Deegan et al. 1986, Browder 2000). However, large volume freshwater inflows within short time periods can be detrimental to estuarine species assemblages (Serafy et al. 1997) and their productivity (Fraser 1981, Van Os et al. 1981). CERP intends to reduce abrupt point-source freshwater inputs and resultant salinity fluctuations and the duration, frequency, severity, and extent of deleterious hypersaline events (Browder et al. 2005, Crigger et al. 2005, Rudnick et al. 2005, RECOVER 2008). CERP-driven creation, expansion, and maintenance of nearshore mesohaline and polyhaline salinity habitats is also anticipated (Barnes et al. 2005, Browder et al. 2005, Davis et al. 2005, Rudnick et al. 2005, Sime 2005, VanArman et al. 2005).

I conclude that restoration of freshwater inflow by CERP implementation would, for the most part, improve pink shrimp habitat. Reduction of intensity, duration, and spatial extent of hypersaline events should directly improve pink shrimp occurrence, abundance, growth, survival, and productivity. Maintenance of nearshore polyhaline conditions should maximize growth, survival, and density, thus improving pink shrimp productivity. Elimination or reduction of rapid salinity fluctuations (Irlandi et al. 1997, Lirman et al. 2008) should reduce osmoregulatory energy expenditures, avoidance behavior, and, possibly, mortality. Indirectly, altered predator-prey relationships (Ault et al. 1999b), and/or overlap of favorable salinity regimes with favorable benthic habitats (Browder and Robblee 2009) may yield survival benefits. Despite some evidence of mesohaline habitat usage by pink shrimp (Tabb et al. 1962a, Howe and Wallace 2000, Robblee et al. 2008), my results suggest that expansion of the area and duration of nearshore mesohaline salinities could negatively impact abundance, growth, and/or survival.

2.6 Research Recommendations

Despite the availability of pink shrimp information, major gaps in knowledge about pink shrimp productivity exist, and new targeted studies could fill them. Laboratory study of salinity and temperature influences on pink shrimp molt-processes would yield more realistic growth representations (Staples and Heales 1991, O’Brien

1994). No information exists regarding the influence of salinity fluctuation frequency, magnitude, and duration on pink shrimp growth and survival. Other penaeid species attained maximal growth at moderate salinity fluctuations (± 4 – 7 ppt: Mu et al. 2005, ± 5-10 ppt: Su et al. 2010), whereas greater magnitude fluctuations (±10 – 15 ppt) reduce growth and survival (Su et al. 2010). The potentially confounding effect of density-dependence on laboratory studies of the effect of salinity on growth and mortality should be given more consideration. Given laboratory (Eldred et al. 1961, Zein-Eldin 1963, Teinsongrusamee 1965, Caillout et al. 1972, Browder et al. 2002, Zink et al. 2013) and field studies' (Álvarez et al. 1987; Pérez-Castañeda and Defeo, 2002, 2005) concern of density-dependent growth, directed study of growth at a range of laboratory and ecologically relevant densities should be conducted. No information is presently available regarding pink shrimp bioenergetics (Ault et al. 1999b). Such information, including osmoregulatory relationships studies, should be combined with molt-process growth studies to better define growth mechanics relative to salinity conditions for use in ecological production models. Salinity preference studies like those conducted for other estuarine organisms (McManus et al. 2014, Doerr et al. 2015) currently do not exist for pink shrimp but could provide further insight into altered freshwater management impacts.

Both short (< 1 yr) and longer (> 1 yr) duration field investigations could confirm salinity effects on growth and survival obtained from laboratory studies. For example, short term i*n-situ* growth studies have been used to study brown shrimp (*F. aztecus*) growth between natural and modified salinity regimes and salinity gradients (Rozas and Minello 2011), while tethering studies have yielded direct evidence of habitat-moderated predation pressure (Minello 1993, Minello et al. 2003). Longer duration cohort analyses could yield *in situ* growth and mortality rates across different habitat or temporal conditions (Álvarez et al. 1987, Minello et al. 1989, Perez-Casteñada and Defeo 2005). Cohort analysis comparing inter-annual or spatial differences in salinity habitats could either validate or refute laboratory results and further contribute to pink shrimp production modeling. Few longer duration (> 1 yr) nearshore or estuarine pink shrimp occurrence and abundance field studies exist, limiting knowledge of inter-annual variability. No information on pink shrimp daily home range or movement patterns on the nursery grounds was identified; this information could lead to better spatially explicit ecosystem models (Ault et al. 1999b).

These studies should lead to updated, improved, and/or new distribution, abundance, and production modeling exercises that provide deeper insight into freshwater management impacts on pink shrimp EFH. Similar modeling exercises have provided such insight on habitat suitability for other penaeids and other estuarine species (Clark et al. 2004, Haas et al. 2004, Adamack et al. 2012, McManus et al. 2014, Lirman et al. 2014b). Although a habitat suitability index model of pink shrimp occurrence has been published (Mulholland 1984), its approach did not use statistically validated, continuous predictive functions (Boyce and McDonald 1999, Boyce et al. 2002). Statistically rigorous prediction models could simulate pink shrimp distribution, abundance, growth, survival, and productivity altered by differing freshwater management scenarios.

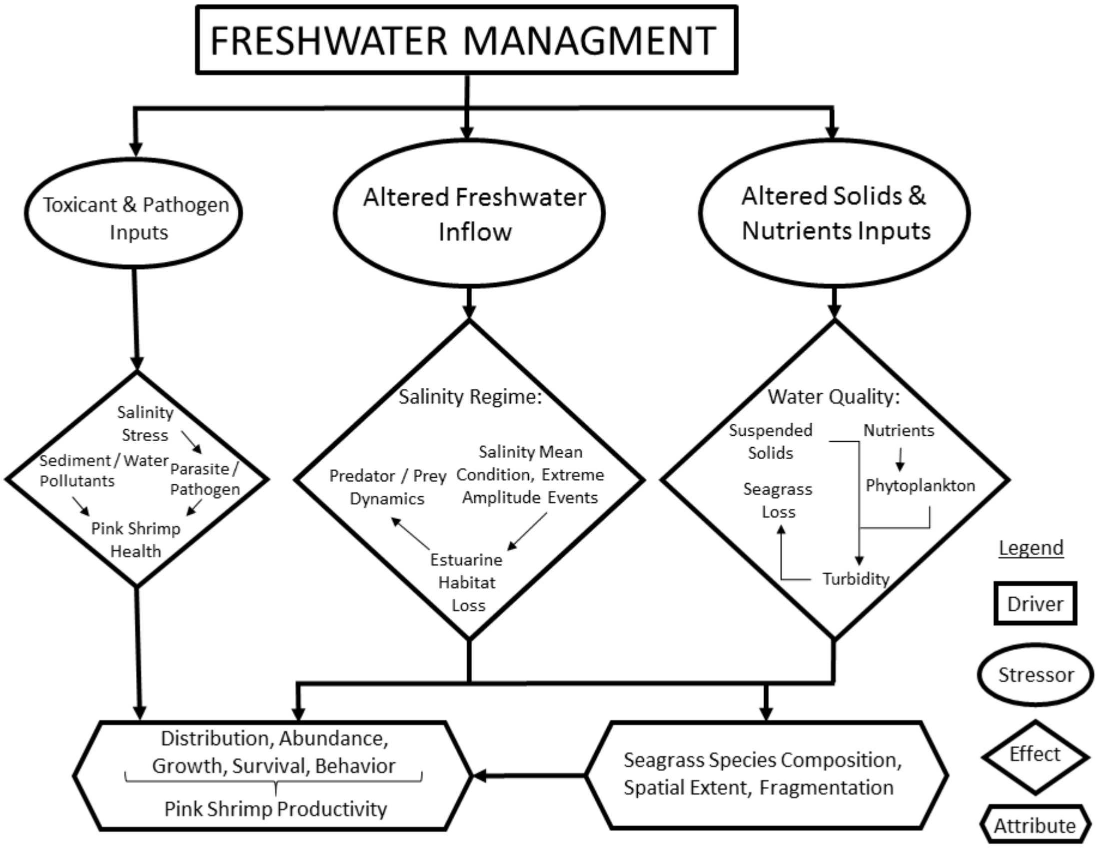


Fig. 2.1: Conceptual model depicting direct and indirect linkages between freshwater management and pink shrimp Essential Fish Habitat (EFH) Metrics following style of Harwell et al. (1999). Relative sizes of stressor and effect symbols depict degree of understanding and suspected degree of impact. The present review focuses on the middle pathway: “Altered Freshwater Inflow” in relation to direct salinity effects upon pink shrimp EFH Metrics.

Table 2.1: Summary of geographic subregions, EFH informational content, and ontogenetic stages studied in the reviewed studies (N = 255).

Classification Category	Condition of Category	Number of Studies	Proportion of Total
Geographic Subregions	North Carolina	25	0.098
	South Atlantic	14	0.055
	South Florida	122	0.474
	Gulf of Mexico East	45	0.177
	Gulf of Mexico West	30	0.118
	Gulf of Mexico South	39	0.153
	Not Applicable	8	0.031
	Not Reported	1	0.004
EFH Information	Distribution/Occurrence	46	0.180
	Density	87	0.341
	Survival	37	0.145
	Growth	58	0.227
	Productivity	29	0.114
Behavior	-	74	0.290
Study Type	Field	169	0.663
	Laboratory	66	0.259
	Simulation/Modeling	34	0.133
	Other	12	0.047
Ontogenetic Stage	Egg	16	0.063
	Larval	24	0.094
	Postlarval/Early Juvenile	93	0.365
	Juvenile	125	0.490
	Subadult	106	0.416
	Adult	84	0.329
	NA	1	0.004
	NR	73	0.287

Table 2.2: Summary of sampling design, duration, sampling gear (those used in >5% of reviewed studies), and diel nature of methods used in reviewed field studies (N = 169).

Classification Category	Condition of Category	Number of Studies	Proportion of Total
Sampling Design	Fixed (FX)	96	0.568
	Haphazard (HZ)	12	0.071
	Stratified Random (StR)	11	0.065
	Various (VA)	7	0.041
	Not Reported (NR)	32	0.189
Duration (time [t] = yr)	$t \leq 0.5$	19	0.112
	$0.5 < t \leq 1$	44	0.260
	$1.0 < t \leq 2.0$	32	0.190
	$2.0 < t \leq 5.0$	26	0.154
	$5.0 < t \leq 10.0$	10	0.059
	$10.0 < t \leq 15.0$	4	0.024
	$t > 15.0$	3	0.018
	Not Reported (NR)	26	0.154
Sampling Gear	Channel Net	14	0.082
	Plankton Net	17	0.101
	Seine	16	0.095
	Trawl	77	0.456
	Drop Net/Samplers	13	0.077
	Throw Trap	20	0.118
	Not Applicable (NA)	2	0.012
	Not Reported (NR)	21	0.124
Diel Nature of Study	Diurnal	17	0.172
	Nocturnal	29	0.101
	Both	40	0.237
	Not Applicable (NA)	1	0.006
	Not Reported (NR)	82	0.485

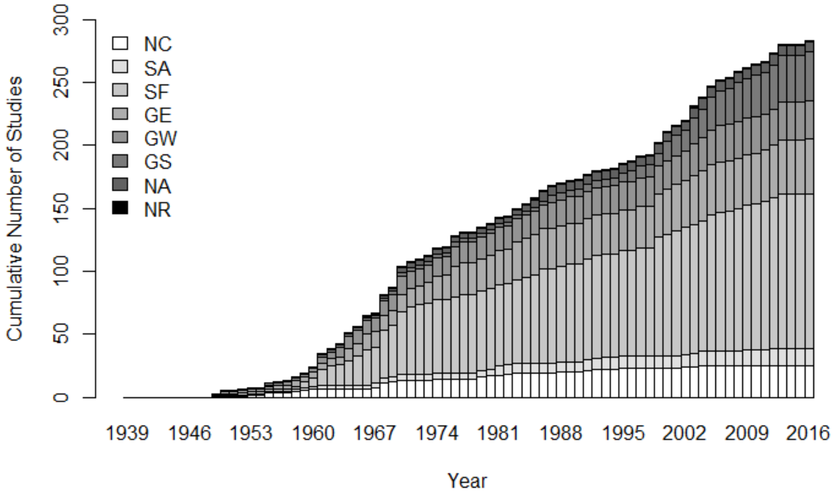


Fig. 2.2: Cumulative number of reviewed pink shrimp studies (N = 255) by geographic subregion beginning with delineation as a distinct species (1939) until the present (2016). NC = North Carolina, SA = South Atlantic, SF = South Florida, GE = East Gulf of Mexico East, GW = Gulf Mexico West, GS = Gulf of Mexico South, NA = not applicable, NR = not reported.

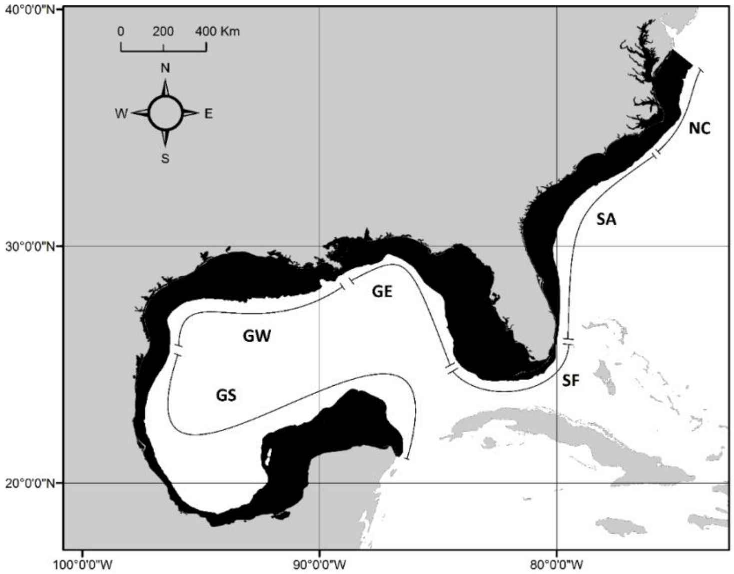


Fig. 2.3: Map of southeastern North American Atlantic Coast, Caribbean, and Gulf of Mexico regions depicting the geographic range of pink shrimp (black shading) to a depth of 200 m. Not depicted on this map are reported observations from Bimini, Bermuda, and Cay Sal Bank. NC = North Carolina, SA = South Atlantic, SF = South Florida, GE = East Gulf of Mexico East, GW = Gulf Mexico West, GS = Gulf of Mexico South.

Table 2.3: Summary of studies that investigated statistical trends between pink shrimp relative abundance and salinity. NR = not reported, NS = not significant, PL = postlarvae, JV = juvenile, SB = subadult, N = number of sampling events, CL = carapace length.

Study #	Source	N	Ontogenetic Stage	Size Range (mm CL)	Salinity Range (ppt)	Statistical Relationship	Shape	Abundance Trend (+ / -)
1	Wakida-Kusunoki et al. 2008	136	NR	NR	28 – 42	Correlation	Linear	+
2	Berkeley and Campos 1984	630	NR	6 – 42	21.2 – 34.7	Correlation	Linear	-
3	Howe and Wallace 2000	108	PL, JV	NR	3.3 – 12.4	Correlation	Linear	-
4	Howe et al. 1999	59	PL, JV	NR	0.6 – 28.0	Correlation	Linear	+
5	Howe et al. 1999	351	PL, JV	NR	0.0 – 29.1	Correlation	Linear	+
6	Sheridan and Minello 2003	61	NR	NR	28 – 33	Regression	Linear	+
7	Browder and Robblee 2009	20	NR	NR	28.3 – 45.3	Regression	Linear	-
8	Perez-Castañeda and Defeo 2001	72	JV, SB	8 – 24	9 – 42	Correlation	Linear	+
9	Johnson et al. 2002	NR	NR	NR	5 – 33	Regression	Linear	+
10	Johnson et al. 2002	NR	NR	NR	28 – 60	Regression	Linear	-
11	Robblee et al. 2008	NR	NR	NR	2 – 28	Regression	Log-linear	-
12	Robblee et al. 2008	NR	NR	NR	2 – 43	Regression	Log-linear	+
13	Blanco-Martínez and Pérez-Castañeda 2016	33	JV	NR	32 – 45	Regression	Linear	+
14	Rubec et al. 2016	NR	JV	NR	2 – 37	Regression	Spline	NA
15	Lirman et al. 2016	NR	NR	NR	2 – 48	Regression	Parabolic	NA
16	Flores et al. 1996	240	NR	NR	8 – 28	Correlation	Linear	NS
17	Rice 1997	NR	PL, JV	< 12	2 – 33	Regression	Linear	NS
18	Browder and Robblee 2009	17	NR	NR	22 – 48	Regression	Parabolic	NS

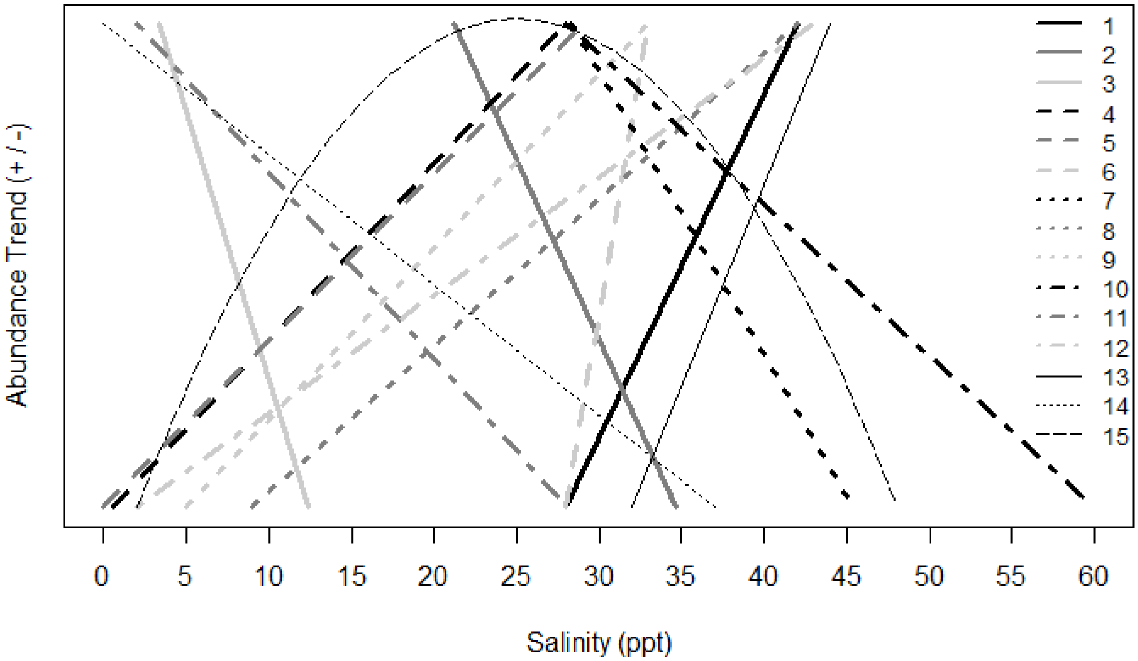


Fig. 2.4: Plot of reported statistically significant positive, negative, or parabolic trends in abundance over investigated salinity range. Legend study numbers coincide with those presented in Table 2.3. Difficulty in graphical recreation of the one spline regression trend (14) led to representation as a linear trend. Of the 13 studies depicted, 8 reported trend maxima between ~20 and ~35 ppt salinities.

Table 2.4: Results of hierarchical Bayesian model selection including fixed categorical salinity (FSal); a single, fixed intercept (FIntercept); multiple, random factor intercepts (RIntercept); a single, fixed precision of residual error (FPrec); and a known (i.e., meta-analytic) standard error (i.e., $\sqrt{(1/\text{precision})}$) input for each density estimate (MPrec) as reported by Minello (Table 9: 1999). The selected final model is highlighted by boldface type. DIC = Deviance Information Criterion, ΔDIC = change in DIC among models, Bayesian *p* = model fit diagnostic (0.5 = perfect fit, $0 \leq P \leq 1$), R^2 = fraction of variance explained, pD = effective number of parameters.

	Model Terms	DIC	ΔDIC	Bayesian *p*	R^2	pD
Model 1	FSal+FIntercept+FPrec	38.22	27.95	0.4997	0.26	3.555
Model 2	FSal+RIntercept+FPrec	41.35	31.08	0.4974	0.092	5.843
Model 3	FSal+FIntercept+MPrec	22.15	11.88	0.7397	0.12	3
Model 4	**Fsal+RIntercept+MPrec**	**10.27**	**0**	**0.588**	**0.16**	**5.992**

Table 2.5: Mean coefficient values and 95% Credible Interval (2.5% and 97.5% quantiles of parameter estimates) calculated for Model 4 of Table 2.4.

Parameter	Q2.5%	Mean	Q97.5%
RBase1	-0.07834	0.101	0.2817
RBase2	0.5109	1.035	1.562
RBase3	-0.0409	0.1729	0.3873
RBase4	-0.0101	0.0095	0.0290
FSal1 (Polyhaline)	0.1149	0.2522	0.3896
FSal2 (Mesohaline)	0.0132	0.1627	0.3109

Table 2.6. Pink shrimp linear and exponential growth parameters of length and/or weight derived and study characteristics reported in laboratory studies. When available, associated salinity (ppt) levels and treatment, diet, stocking density (shrimp L^{-1}), duration (d), shrimp source, and size ranges studied (mm CL) are also provided. NR = Not Reported, NA = Not Applicable, V= varies, LN = length (mm CL), WT = weight (g), DWT = dry weight (g), PEL = pellet, ART = *Artemia*, WLD = wild sourced, CUL = cultured shrimp (from broodstock held in captivity),

Study #	Source	Linear Growth Rate	Exponential Growth Coefficient	# Salinity Levels	Salinity Treatments (ppt)	Diet	Stocking Density (shrimp L^{-1})	Duration (d)	Shrimp Source	Size Range (mm CL)
1	Browder et al. 2002*	NA	0.0 – 0.0279 LN	12	2, 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55	PEL	1	28	WLD	3.2 – 14.5
2A	Zink et al. 2013[§]	0.033 – 0.16 mm d^{-1}[‡]	0.0114 – 0.0316 LN	3	15, 35, 55	PEL	4	56	CUL	2.3 – 5.6
2A	Zink et al. 2013[§]	0.0005 – 0.0035 g d^{-1}[‡]	0.0388 – 0.100 WT	**3**	15, 35, 55	PEL	4	56	CUL	2.3 – 5.6
2B	Zink et al. 2013[§]	0.0 – 0.16 mm d^{-1}[‡]	0.0112 – 0.0252 LN	3	15, 35, 55	PEL	0.81	56	CUL	4.6 – 9.7
2B	Zink et al. 2013[§]	0.001 – 0.017 g d^{-1}[‡]	0.0254 – 0.0817 WT	3	15, 35, 55	PEL	0.81	56	CUL	4.6 – 9.7
3	Zein-Eldin 1963	0.0032 – 0.0063 g d^{-1}[‡]	0.121 – 0.139 WT[‡]	5	2, 5, 10, 25, 40	ART	NR	28-29	WLD	1.9 – 7.8
4	Zein-Eldin and Aldrich 1964	0 – 0.22 mm d^{-1}	NR	5	2, 5, 15, 25, 35	NR	NR	NR	NR	NR

* Calculated from original study data which included a 2 ppt salinity treatment but growth coefficients could not be computed due to no survival at that treatment level.

[‡] Calculated from data provided therein

[§] Study repeated for growth rates provided from differing trials conducted therein

Table 2.7. Reported pink shrimp field linear growth rates. When available, study method, associated salinity (ppt) levels and ranges, sampling frequency (mo^{-1}), duration (d), and size ranges studied (mm CL) are also provided. NR = Not Reported, V = Varies, S-F = Size-Frequency (cohort) analysis, M-R = Mark-Recapture, SP = Spring, SU = Summer, AU = Autumn, W = Winter.

Study #	Source	Study Method	Linear Growth Rate (mm d^{-1})	# Salinity Levels	Salinity Ranges (ppt)	Sampling Frequency (mo^{-1})	Duration (yr)	Size Range (mm CL)
1	Murphey and Fonseca 1995	S-F	0.051	1	28 – 37	1	1	0.75 – 23.5
2	Hoese and Jones 1965[#]	S-F	SP: 0.14; SU: 0.19	2	SP: 22 – 25; SU: 39 – 41	1-2	1	3.4 – 20.9
3 (A+B)	Álvarez et al. 1987[*]	S-F	0.13; 0.25	2	13.4 – 36.4; 10 – 28.5	4	0.92	1 – 17.2
4 (A+B+C)	Pérez-Castañeda and Defeo 2005[#]	S-F	AU-W 0.086; SP: 0.13, SU-AU: 0.14	3	AU-W 7-35; SP: 25-35, SU-AU: 10-25	1	2	8 – 24.3
5	Tabb et al. 1962a[#]	S-F	0.033 – 0.133	2	12-37	NR	NR	2 – 32
6 (A+B)	Williams 1955b[#]	S-F	W: 0.040 – 0.062; SU: 0.30 – 0.42	2	W: 5.6 – 33.0; SU: 6.1 – 37.2	~ 1	2.2	2.3 – 34.7
7	Eldred et al. 1961	S-F	0.14	V	24.8 – 27.7	1	0.58	2.9 – 14.1
8	Pérez-Castañeda and Defeo 2003b	S-F	0.14	NR	NR	1	2	8 – 24.3
9	Campos and Berkeley 2003	S-F	0.13 – 0.17	NR	NR	V	0.83	8 – 31
10	Idyll et al. 1970[#]	S-F	0.082; 0.066; 0.14	3	W – SP; W; SU	1	~ 1	1.8 – 21.4
11	Pérez-Castañeda and Defeo 2003b	S-F	0.09 – 0.21	NR	NR	~1	2	8 – 24.3
12	Eldred 1962	NR	0.14	NR	NR	NR	NR	NR
13	McCoy and Brown 1967	M-R	0.099[‡]	NR	NR	V	0.75	14.1 – 36
14	Costello and Allen 1959	M-R	0.067 – 0.10	NR	NR	~1	NR	13.2 – 18.7
15	Cody and Fuls 1981	M-R	-0.067 – 0.752	NR	NR	V	2	NR
16	Costello and Allen 1961	M-R	0.19	NR	NR	NA	0.18	15.3 – 21.7
17	Monsreal-Vela et al. 2016	S-F	0.007 – 0.30	NR	NR	1	0.917	8 – 38.4

[#] Treatment levels are based upon different study seasons

[*] Treatment levels are based upon different study locations

[‡] Linear equation provided: TL (mm) = 2.152 + 0.414*Time-at-Large (d)

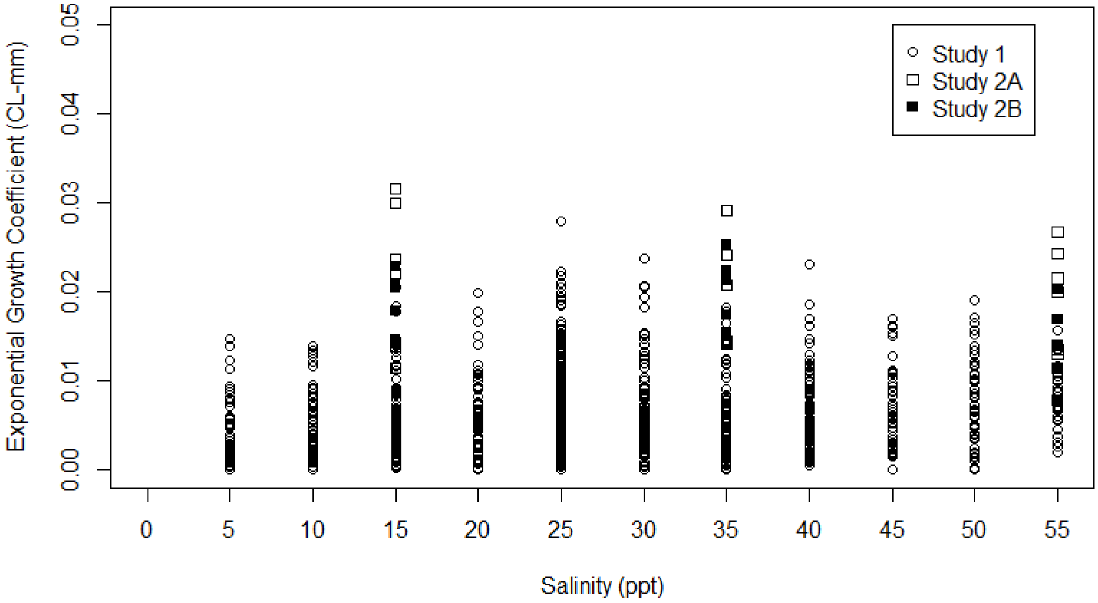


Fig. 2.5: Pink shrimp exponential growth coefficients of CL (mm) plotted against treatment salinity (ppt). Legend study numbers coincide with those presented in Table 2.6.

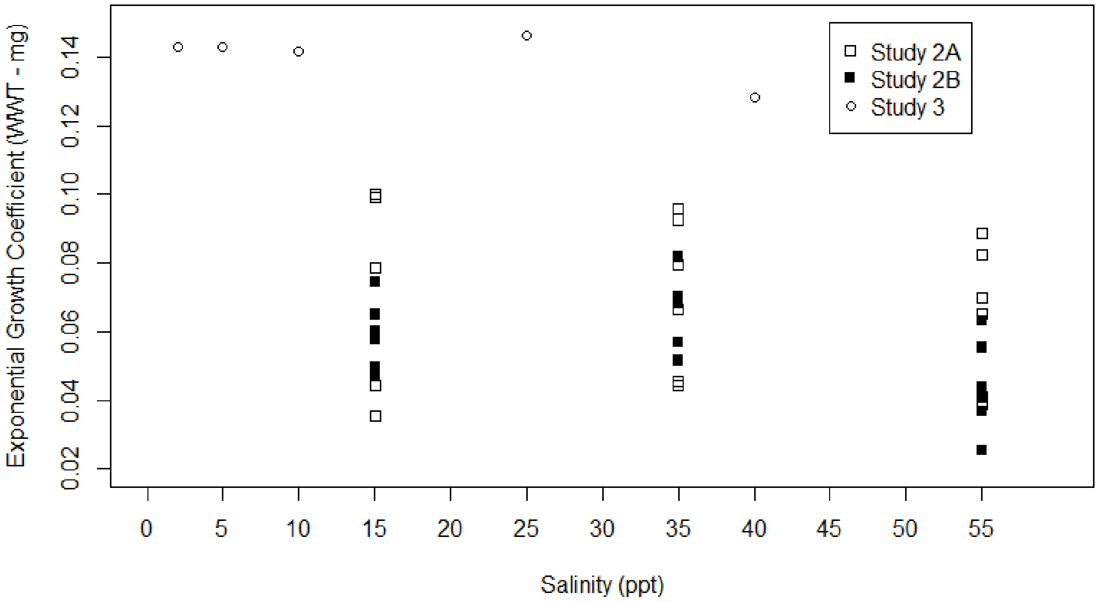


Fig. 2.6: Pink shrimp exponential growth coefficients of WWT (mg) plotted against treatment salinity (ppt). Legend study numbers coincide with those presented in Table 2.6.

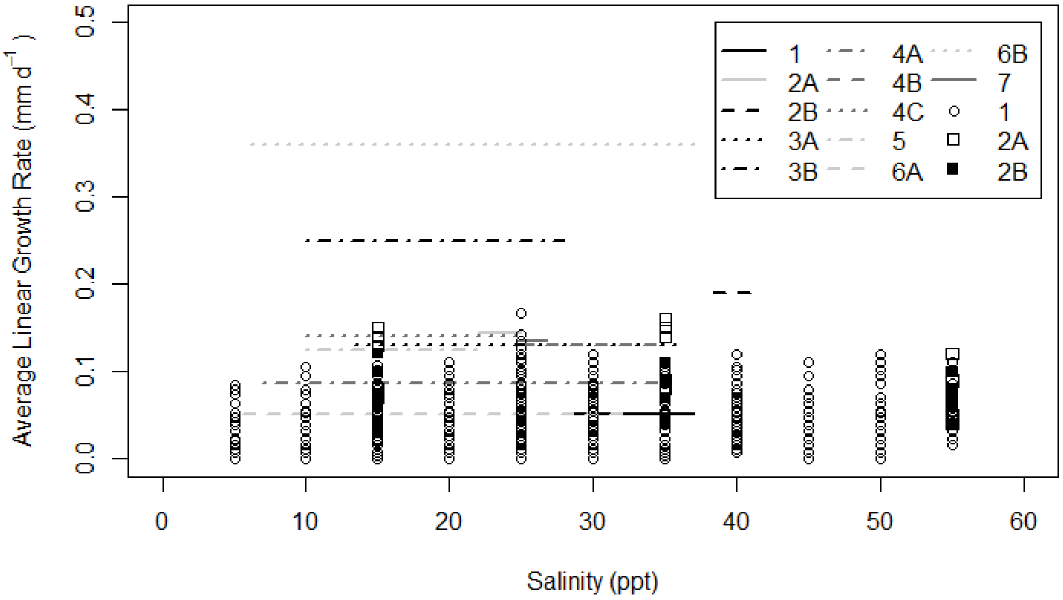


Fig. 2.7: Field linear growth rates (Table 2.7) depicted by lines and laboratory linear growth rates (Table 2.6) depicted by points. The overlap of the upper range of laboratory linear growth rates and the majority (81.8%) of field linear growth rates suggests general congruence. Legend study numbers coincide with those presented in Table 2.6 (points) and 2.7 (lines).

Table 2.8: Reported pink shrimp survival rates observed during longer duration (≥10 to ≤ 30 d) laboratory study. When available, associated salinity (ppt) levels and treatments, diet, stocking density (L^{-1}), duration (d), shrimp source, and size ranges studied (mm CL) are also provided. CL = carapace length, CUL = cultured, WLD = wild, NR = not reported, ART = *Artemia*, PEL = pellets.

Study #	Source	Survival Rate (% population d^{-1})	# Salinity Levels	Salinity Treatments (ppt)	Diet	Stocking Density (L^{-1})	Duration (d)	Shrimp Source	Size Range (mm CL)
1	Browder et al. 1999	0.957 – 0.061	5	30,35, 40, 45, 50	NR	NR	10	NR	NR
2	Browder et al. 2002	1.0 – 0.0	12	2, 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55	PL	1	28	WLD	3.2 – 14.5
3A	Zink et al. 2013	0.840 – 0.273	3	15, 35, 55	PEL	4	28	CUL	~2.3 – ~5.6
3B	Zink et al. 2013	0.933 – 0.533	3	15, 35, 55	PEL	0.81	28	CUL	~4.6 – ~9.7
4	Zein-Elden 1963*	0.84 – 0.0	5	2, 5, 10, 25, 40	ART	NR	30	WLD*	NR
5	Rand and Bachman 2007	1.0 – 0.5	11	2, 5, 10, 15, 20, 25, 30, 35, 40, 45, 50	PEL	2	30	CUL	NR
6	Zein-Elden and Aldrich 1964*	1.0 – 0.0	5	2, 5, 15, 25, 35	NR	NR	30	WLD*	NR

* Mixed group of *F. aztecus* and *F. duorarum* due to difficulties with species differentiation

Table 2.9: Reported pink shrimp mortality rates observed during short-duration ($\leq$ 4 d) laboratory studies investigating abrupt salinity change. When available, associated salinity (ppt) levels and treatments, diet, stocking density (L^{-1}), duration (d), shrimp source, and size ranges studied (mm CL) are also provided. NR = not reported, ART = *Artemia*, PEL = pellets, CUL = cultured, WLD = wild, CL = carapace length.

Study #	Source	Mortality Rate (% population d^{-1})	# Salinity Levels	Salinity Treatments (ppt)	Diet	Stocking Density (L^{-1})	Duration (d)	Shrimp Source	Size Range (mm CL)
1	Williams 1960	0.0 – 0.167 d^{-1}	5	10, 15, 20, 25, 30	NR	NR	2, 4	NR	7.4 – 22.2
2	Criales et al. 2011a	0.0 – 0.995 d^{-1}	9	2, 5, 10, 15, 25, 35, 45, 55, 60	ART, PEL	5	4	CUL	2.1 – 7.3
3	Zein-Elden and Aldrich 1964	0.0 – 1.0 d^{-1}	NR	2 to 40	NR	NR	1	WLD*	NR

* Mixed group of *F. aztecus* and *F. duorarum* due to difficulties with species differentiation

Table 2.10: Results of Bayesian hierarchical model selection incorporating fixed effect linear salinity (FSal) and quadratic salinity (FSal_2) terms and/or a random study effect (RStudy) term as predictors of reported or calculated survival rates (Table 2.8). The selected final model is highlighted by boldface type. DIC = Deviance Information Criterion, ΔDIC = change in DIC among models, Bayesian *p* = model fit diagnostic (0.5 = perfect fit, $0 \leq P \leq 1$), R^2 = fraction of variance explained, pD = effective number of parameters.

	Model Terms	DIC	ΔDIC	Bayesian *p*	R^2	pD
Model 1	FSal	101	37.93	0.4881	0.0	3.0
Model 2	FSal+FSal_2	88.79	25.72	0.4875	0.04	3.957
Model 3	FSal+RStudy	81.08	18.01	0.4893	0.094	7.792
Model 4	**FSal+FSal_2+Rstudy**	**63.07**	**0**	**0.485**	**0.17**	**8.895**

Table 2.11: Mean coefficient values and 95% Credible Interval (CI: 2.5% and 97.5% quantiles of parameter estimates) calculated for Model 4 of Table 2.9.

	Q2.5%	Mean	Q97.5%
Intercept	0.3883	0.7179	1.047
FSal	0.01586	0.0278	0.03929
FSal2	-5.93E-04	-4.09E-04	-2.20E-04
RStudy1	-0.5337	-0.2064	0.09641
RStudy2	-0.4034	-0.08164	0.2256
RStudy3A	-0.3577	-0.05351	0.245
RStudy3B	-0.5345	-0.2087	0.09342
RStudy4	0.0259	0.3532	0.7383
RStudy5	-0.113	0.2016	0.5317

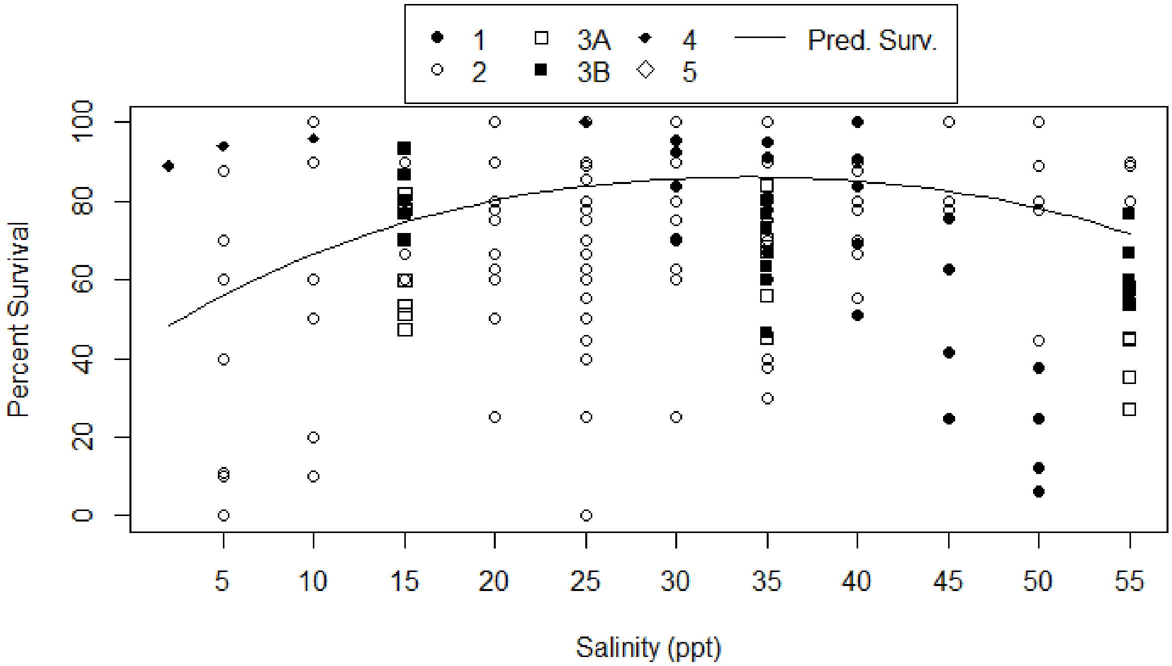


Fig. 2.8: Pink shrimp laboratory study percent survival observations (points; N = 211) (Table 2.8) and the predicted regression trend (Pred. Surv.) of Model 4 (Table 2.10) plotted against salinity (ppt). Legend study numbers coincide with those presented in Table 2.8 (points).

Chapter 3: Pink Shrimp *Farfantepenaeus duorarum* Spatiotemporal Abundance Trends Along an Urban, Subtropical Shoreline Slated for Restoration

3.1 Summary

The Biscayne Bay Coastal Wetlands (BBCW) project of the Comprehensive Everglades Restoration Plan (CERP) aims to reduce point-source freshwater discharges and redirect freshwater flow along the mainland shoreline of southern Biscayne Bay. An increase in pink shrimp (*Farfantepenaeus duorarum*) density to ≥ 2 individuals m^{-2} during the wet season (i.e., August-October) along the mainland shoreline is proposed as an indicator of BBCW success. This study examined pre-BBCW baseline densities and compared them with the above (≥ 2 individuals m^{-2}) target. Densities were monitored via seasonal (wet, dry) throw-trapping at 47 sites along ~ 22 km of southwestern Biscayne Bay coastline over 10 years (2007-2016). Substantial inter-annual density variability was observed; densities were most often higher in dry seasons. Quantile regression revealed potential density limitation by four habitat attributes: water temperature (°C), depth (m), salinity (ppt), and submerged aquatic vegetation (SAV: % cover). Procrustean analyses tested significance of habitat attribute observation congruence to shrimp density observations. Water temperature, water depth, and salinity explained ~ 28%, 28%, and 22% of density variability, respectively, but no significant relationship with SAV was observed. Hierarchical clustering was used to identify spatially and temporally similar groupings of pink shrimp densities that could be later investigated with respect to potentially limiting habitat attributes. Six site and four year-season clusters were identified. Although habitat attributes significantly differed among spatial clusters, median densities within the clusters did not correlate to habitat attributes' within-cluster

minimum, maximum, median, or standard deviation. Pink shrimp densities corresponded significantly with, and appeared limited by, salinity conditions, an environmental attribute that will be directly influenced by CERP implementation. However, the lack of difference in salinities among spatiotemporal clusters indicated either that salinity did not have a major influence on shrimp density patterns or that other habitat attributes complicated the spatiotemporal analyses.

3.2 Background

The watershed of Biscayne Bay was heavily modified during the 20th century and is currently highly managed to prevent urban, suburban, and agricultural flooding while also meeting agricultural, commercial, and residential freshwater demands. Implementation of the Comprehensive Everglades Restoration Plan (CERP) seeks to restore the quality, quantity, timing, and distribution of freshwater deliveries to southern Florida nearshore areas (Water Resources Development Act of 2000: Public Law 106-541, 114 Stat. 2680; RECOVER 2005), including Biscayne Bay. The Biscayne Bay Coastal Wetlands (BBCW) project, a CERP component, aims to restore a more natural hydrology and salinity regime along the bay's southwestern shoreline (USACE and SFWMD 2012, USACE 2014). Three actions are needed to make this improvement: (1) increasing the total volume of freshwater deliveries; (2) diverting part of the volume of point-source freshwater discharge (i.e., canal discharges) to reestablish water delivery as overland sheet flow; and (3) altering the present timing of deliveries by means of lengthening discharges through the wet season (May-October) and into the dry season (November-April) (RECOVER 2008, USACE 2014). The REstoration COordination and

VERification (RECOVER) team established Interim Goals (IGs) to link ecological indicator metrics to CERP activities and thus evaluate restoration performance and realization of post-implementation ecological benefits at 5-yr intervals (RECOVER 2005).

The pink shrimp *Farfantepenaeus duorarum* is one of many ecological indicators selected to assess ecological impacts of CERP implementation (RECOVER 2005, Browder and Robblee 2009). Pink shrimp was selected to assess estuarine ecosystems due to previously suggested abundance linkages to salinity condition (RECOVER 2005, Browder and Robblee 2009). As reviewed by Zink et al. (2017: see Chapter 2), salinities within polyhaline (18 – 30 ppt: Anonymous 1958) and euhaline (30 - 40 ppt) ranges would directly improve pink shrimp productivity. Expansion of southwestern Biscayne Bay estuarine habitat was anticipated to benefit pink shrimp residing there (Browder et al. 2005). Indirectly, reduced salinity variability stress on seagrass communities would yield increased pink shrimp abundance due to increased seagrass cover and spatial extent (Browder et al. 2005a, Browder and Robblee 2009). Higher abundance of pink shrimp has been reported in areas exhibiting higher and more stable salinities (Diaz 2001, Browder et al. 2005b, Santos 2014); these same stretches of coast exhibit more continuous seagrass cover (Santos et al. 2011). Expansion of the areal cover of shoal grass (*Halodule wrightii*) could further amplify pink shrimp abundance due to an apparent affinity for this seagrass species (Costello et al. 1986). The stated pink shrimp IG for southwestern Biscayne Bay is “2 shrimp m^{-2} in nearshore optimal habitat (i.e., seagrasses)” during August-October peak abundance periods (RECOVER 2005). This IG was based upon a peak density of ~1.8 shrimp m^{-2} observed in September during a relatively short duration (2 yr) pilot study (Browder et al. 2005b).

Historically, most freshwater delivery to Biscayne Bay was through transverse glades: low topography channels through the Miami Coastal Ridge that allowed Everglades Basin surface water drainage (Davis 1943, Halley et al. 1977) and groundwater seepage (Shaler 1895, Smith 1896, Kohout 1967, Kohout and Kolipinski 1967, Langevin 2003, Stalker et al. 2009). On the eastern side of the Coastal Ridge, transverse glades fed fresh water from the Everglades into creek networks that spread surface water flows along the shoreline. However, canalization converted the freshwater delivery system to one dominated by pulsed point-source (i.e., canal mouth) discharges that altered benthic submerged aquatic vegetation (SAV), infaunal, epifaunal, and nekton communities (Brook 1982; Irlandi et al. 1997; Serafy et al. 1997; Lirman et al. 2008a, b, 2014; Santos et al. 2011). Canalization also lowered the water table, which reduced nearshore groundwater seepage (Langevin 2003, Stalker et al. 2009).

Post-BBCW salinity goals for western central and southern Biscayne Bay (Shoal Point to Turkey Point: Fig. 3.1) include nearshore areas with oligohaline (0.5-5 ppt) and mesohaline (5-18 ppt) regimes at the shoreline trending towards 20 ppt 500 m from the coast (RECOVER 2008). These salinity conditions are anticipated to enrich estuarine faunal assemblages as well as increase estuarine species distributions and abundances (Browder et al. 2005a, RECOVER 2008, McManus et al. 2014). Expansion of continuous submerged aquatic vegetation (SAV) habitats dominated by *Halodule wrightii*, a species commonly associated with low and variable salinity, is also foreseen (Lirman and Cropper 2003; RECOVER 2004; Lirman et al. 2008a, b, 2014; Santos et al.

2011). Indeed, BBCW implementation goals for benthic habitat include increased spatial extent of nearshore seagrass beds, especially expansion of *H. wrightii* seaward into Biscayne Bay (RECOVER 2004). Increased overlap of optimal salinity conditions with preferred benthic SAV habitats would yield indirect, synergistic benefits to estuarine fauna such as pink shrimp (Sklar and Browder 1998, Browder et al. 2005a, RECOVER 2008, Browder and Robblee 2009).

The purpose of this study was to investigate spatiotemporal trends in pink shrimp density along the southwestern Biscayne Bay shoreline using CERP IG as working reference threshold. I investigate the plausibility of the post-implementation ≥2 shrimp m^{-2} IG. Further, I address presumptions that: (1) pink shrimp peak abundance occurs during the wet season; and (2) nearshore mesohaline salinity goals would yield increased pink shrimp abundance (within the sampled spatial domain). Pink shrimp density relationships to species-specific and total benthic SAV % cover, as well as SAV canopy height, were also investigated. My focus was to evaluate temporal (i.e., seasonal and inter-annual) and spatial pink shrimp density trends relative to spatiotemporal patterns in habitat attributes. This was achieved by: (1) using quantile regression to identify habitat attributes that potentially limit pink shrimp density; (2) organizing pink shrimp density and habitat observations via heatmaps to visually assess spatiotemporal variability and trends; (3) using Procrustean analysis to measure concordance between density and habitat attribute matrices; (4) employing hierarchical clustering analysis to identify spatiotemporal density groupings; and (5) investigating distributional aspects (median, minimum, maximum, and standard deviation) of habitat attributes (temperature, salinity, water depth, and SAV % cover) within density clusters to link density patterns to the environment. These analyses employ data from two seasons (wet and dry) of 10 years of epifaunal community monitoring data collected from 47 sites lying within 50 m of shore and spanning ~ 22 km of shoreline.

3.3 Methods

3.3.1 Study Area

Biscayne Bay is a large (1,110 km^2), shallow (depths generally < 3 m), subtropical lagoon system located along the southeast coast of Florida, USA (Fig. 3.1). Its western shoreline extends approximately 56 km north to south. Where coastal urban development is low, the shorelines consist of mangrove-seagrass ecotone punctuated by natural tidal creeks, artificial channels, and freshwater canals (Serafy et al. 2007). Overland freshwater discharges, along with groundwater seepage, create a salinity gradient perpendicular to the shoreline with three salinity zones: (1) western nearshore areas usually affording the lowest salinities; (2) the bay central axis marked by near oceanic salinities; and (3) oceanic salinities near the eastern passes to the open ocean (Lirman and Cropper 2003, Caccia and Boyer 2005, Lirman et al. 2008a). Tidal ranges are generally on the order of 0.5 to 1 m (Schneider 1969, Wang et al. 2003).

3.3.2 Field Surveys

Epifaunal communities and SAV habitats were surveyed seasonally at fixed sampling sites (n = 47) located along the southwestern Biscayne Bay nearshore zone (0-50 m) from Shoal Point to Turkey Point (Fig. 3.1). Surveys were stratified by the dry (January-March sampling) and wet (July-September sampling) seasons that dominate south Florida’s climate. The primary sampling unit was defined as the 20 m buffer around GPS coordinates that identified permanent sampling sites. These sites were located in the shallow, open water along the western shoreline mangrove-seagrass ecotone, an area likely to be directly affected by CERP implementation. During each survey, the 47 fixed sampling sites were visited within 3 hr of high tide over 4 to 6 days within a relatively short timeframe (~ 2 weeks or less). Water quality and habitat parameters, including water temperature (˚C), salinity (ppt), pH, dissolved oxygen saturation (%), dissolved oxygen concentration (mg/L), water depth (m), and sediment depth (m), were recorded at each site. Benthic habitats were assessed for species-specific SAV % cover by visual assessment of 10 replicate 0.5 m^2 quadrats per site (Lirman et al. 2008a, b). In addition, canopy height (maximum seagrass blade length) was measured to provide a topography metric. Species-specific and total SAV % cover data following the methods of Lirman et al. (2008b) available from 2008 to 2016 were used.

Epifaunal communities were sampled in triplicate (n = 3) using an open-ended, rigid-sided aluminum box (i.e., throw trap) measuring 45 cm by 1 m^2 (Kushlan 1981, as modified by Robblee et al. 1991). Two, 3 mm stretch mesh cover nets affixed to opposite sides of the throw-trap upper surface prevented epifauna escape during deployment. Once deployed, the throw-trap was cleared of trapped epifauna by sweeping the trap (n = 4) from alternating directions with a metal-framed seine fitted with 3 mm stretch mesh, while gently tapping the substrate with the seine frame. The collected organisms from the four sweeps were stored together. Organisms collected from each replicate throw-trap deployment were bagged and numbered separately for storing and processing. Samples were frozen for storage until processing.

3.3.3 Epifuana Identification and Measurement

Taxonomic identification and size measurement were conducted in the laboratory. Organisms collected from each replicate throw-trap deployment at a site were maintained and processed independent of each other. Where possible, carapace length (CL, mm), total length (TL, mm), and sex were recorded for each farfantepenaeid shrimp. Shrimps >8.0 mm CL were identified to species primarily using petasma and thelycum morphology, although other characteristics were also used (Perez-Farfante 1969, Perez-Farfante 1970, Perez-Farfante 1988, Perez-Farfante and Kensley 1997). Shrimps <8.0 mm CL were identified to genus due to low degree of sexual morphological development (Perez-Farfante 1970).

3.3.4 Statistical Analysis

All statistical analyses were performed using the R statistical package (The R Foundation, https://www.r-project.org/). Statistical analyses were performed with a Type 1 error criterion of $\alpha = 0.10$ to reduce potential Type 2 errors. Distributions of sizes for all shrimp collected were investigated using Kolmogorov-Smirnov tests to examine potential spatial and seasonal influences. Spatially, potential influences of recruitment limitation to more southern sites was investigated by comparing size distributions for all sampling sites north of Black Point (sites 1 -17) to that of all sites south of Black Point (sites 18 – 47). All further analyses included only farfantepenaeid shrimps >5 mm CL. Combining the data for all three trap samples taken at each site, density was calculated as the sum of observed shrimps per 3 m^2 per site. Density data were natural logarithm (x + 1) transformed before analysis to reduce influence of outlying observations.

3.3.4.1 Potential Habitat Limitations on Pink Shrimp Density

As a statistical interpretation of the ecological concept of Leibig's Law of the Minimum (Taylor 1934, Thompson et al. 1996), quantile regression (QR) has been presented as a method to identify limitation of species distribution or abundance by specific habitat attributes by focusing on specifically on the upper bound of the abundance vs. habitat attribute relationships (Cade et al. 1999, Cade and Noon 2003, Lancaster and Belyea 2006). Use of traditional linear regression models to predict abundance and habitat attribute relationships often yields non-significance; these models focus on the interior of the data distribution in attempts to predict a "mean" response while assuming variability about the response is due to the influence of other factors (Scharf et al. 1998, Cade et al. 1999, Cade and Noon 2003, Lancaster and Belyea 2006). However, examination of functional relationships along the edges of distributions, particularly at the upper bound where the highest abundances may appear limited by the habitat factor of interest, may provide clearer statistical relationships between abundance and this factor (Scharf et al. 1998, Cade et al. 1999, Cade and Noon 2003, Lancaster and Beylea 2006). Here, QR was used to examine whether shrimp density was limited by the various habitat variables measured. Specifically, focus was placed on the magnitude and sign of the 90th regression quantile coefficients emerging from each model developed for each habitat attribute.

Pink shrimp density was first plotted against individual habitat factors to graphically assess potential limiting factors. QRs (function "rq" of package "quantreg") fit to the 0.5 and 0.9 density percentiles were used to statistically identify a subset of habitat attributes that suggested limitation at the median and upper edge of the density distribution. Analyses considered water temperature (˚C), salinity (ppt), pH, dissolved oxygen saturation (%), dissolved oxygen concentration (mg/L), water depth (m), sediment depth (m), and the following SAV metrics: *Thalassia testudinum* % cover, *H. wrightii* % cover, total seagrass % cover, total SAV % cover, and total SAV canopy height. As in previous studies in the same region (Lirman et al. 2008a, b; 2014), S*yringodium filiforme* was rarely encountered (n = 10, 1.6% of total samples) and thus was not further considered.

Multiple QR functional response shapes were investigated including linear, quadratic, cubic, log-linear, natural cubic splines (function "ns" of package "splines") (Wood 2006), and additive quantile smoothing spline (AQSS) response curves (functions "rqss" and "qss" of package "quantreg": Koenker et al. 1994, Koenker 2010). Natural cubic splines were constructed with 3 (0.25, 0.50, and 0.75 quantiles of the predictor), 2 (0.33 and 0.66 quantiles of the predictor) and 1 (0.5 quantile of the predictor) internal knots (Wood 2006). Resultant QR coefficient confidence intervals were constructed and tested for significance by *xy*-pair bootstrapping (function "summary.rq" of package "quantreg"). Polynomial QRs were considered significant if all terms of the first order and higher were significant; significance of the intercept was not considered. Natural cubic spline QRs were modeled without intercepts; these QRs were considered significant if all splines described specified data ranges were individually significant. For those QRs found to be statistically significant, they were only further considered as candidate models if the resulting shape of the QR was “ecologically plausible” (i.e., if their trends predicted increasing density at the either extreme of the observed salinity range).

3.3.4.2 Spatiotemporal Relationships

Heatmaps were generated to visualize spatiotemporal trends in pink shrimp density and the habitat attributes found by QR to potentially limit pink shrimp density. Observation data were converted to 47 row by 20 column matrices to display their spatial (47 sampling sites) and temporal (10 yr by 2 seasons) patterns, and color gradients were used to represent the scale of density observation values.

Procrustean analyses allowed direct testing of statistical concordance between shrimp density and habitat attribute matrices that were used to develop the heatmaps (Jackson 1995, Peres-Neto and Jackson 2001, Lisboa et al. 2014). Procrustean analysis minimizes the residual sum of the squares between a target matrix (**X**: here, shrimp density) and the second, fitted matrix (**Y**: here, habitat attributes) by superimposing the fitted matrix to the target by scaling, rotating, and dilating the fitted matrix (Jackson 1995, Peres-Neto and Jackson 2001). The Procrustean Sum of Squares (PSS, also known as Gower’s Statistic: $m^2_{X,Y}$), the minimized residual sum of squares from the fitting procedures, is used to assess Procrustean fit ranging from 0 to 1 with higher values presenting poorer fit (Jackson 1995, Peres-Neto and Jackson 2001, Lisboa et al. 2014). The PSS metric is equivalent to $1 - r^2$, where r is a Pearson correlation coefficient (Peres-Neto and Jackson 2001). Because the method hinges on one-to-one relationships between the matrices being compared, missing values cannot be handled by Procrustean analysis (Adams et al. 2004, Arbour and Brown 2014). Following Adams et al. (2004) and Arbour and Brown (2014), missing habitat attribute values were imputed with linear regressions that included site, season, and year as potential factors. PROTEST (function "protest," package "vegan", permutation n = 9999) provided statistical significance of Procrustean fits between density and habitat attribute matrices (Jackson 1995).

Hierarchical clustering procedures were used to identify similar density groups among sites and year-seasons (Legendre and Legendre 1998, Clarke et al. 2014). Bray-Curtis dissimilarity matrices were constructed ("vegdist" function, "vegan" package) with respect to site (i.e., spatial) and year-season (i.e., temporal) density observations. Hierarchical agglomerative clustering (function "hclust") using the "Ward.D2" agglomeration method identified spatially and temporally similar density groupings. *A prioiri* statistical significance of clusters was tested via similarity profiling (function "simprof" of package "clustsig") (permutations = 999, number of expected groups = 1000) of identified density cluster memberships (Clarke et al. 2014). Permutational multivariate ANOVA (PERMANOVA: function "adonis2" of package "vegan") testing provided *a posteriori* cluster significance (Anderson 2001, McArdle and Anderson 2001). PERMANOVA was also used to investigate inter-annual and seasonal density differences using year-season cluster membership as a categorical nesting factor. To investigate potential dispersion influences on PERMANOVA significance, multivariate homogeneity of dispersions analysis (function "betadisper" of package "vegan") was used to test for inter-cluster differences in dispersion (i.e., distance to centroid) (Anderson 2006). The density heat map was rearranged to reflect site and year-season cluster memberships.

3.3.4.3 Pink Shrimp Density and Habitat Attributes Among Density Clusters

Pink shrimp density and habitat attributes (previously detected as potentially limiting to pink shrimp density via QR were investigated among site and year-season clusters. First, medians (± CI) of density and habitat attributes were computed for each site and year-season cluster. Confidence intervals (CIs) about median values were computed as:

$$1.58*\mathrm{IQR}/\mathrm{sqrt}(n)$$

where IQR = interquantile ranges and n = sample size, as described in McGill et al. (1978) and Chambers et al. (1983). Plots of density and habitat attributes' median, CIs, minimum, and maximum values were used to visualize their distributions within site and year-season clusters. Density and habitat attributes were analyzed with respect to site or year-season clusters. Nonparametric tests were used because parametric normality and equality of variance assumptions were usually violated. Kruskal-Wallis tests were used to investigate differences in distribution shape and range (i.e., location: Sokal and Rohlf 1995) of density and habitat attributes among site or year-season clusters. Post-hoc Tukey-type nonparametric Conover multiple comparison tests (function "posthoc.tukey.conover.test" of package "PMCMR") were used to test for significant pairwise differences. These tests were implemented as χ^2 distributions to correct for data ties, and p-values were Bonferroni-corrected (Sokal and Rohlf 1995, Conover 1999). A series of correlation analyses was used to identify habitat attribute distribution characteristics that associated with site or year-season cluster median densities. Pearson correlation analyses were applied to median, minimum, maximum, and standard deviation of habitat attributes within site or year-season clusters.

3.4 Results

Of a total of 3,179 penaeid shrimp specimens collected, 9 individuals were identified as species other than *F. duorarum* and were removed from further analysis. Carapace length was measurable for 2,619 specimens. Shrimp size frequency distributions were found to significantly differ both spatially ($D_{2\text{-tailed}} = 0.230$, $p < 0.0001$) and seasonally ($D_{2\text{-tailed}} = 0.092$, $p < 0.0001$: Fig. A1). For the seasonal size distribution comparison, the maximal difference was found at 5.53 mm CL; the maximal size difference for the spatial comparison was associated with 7.54 mm CL. The distribution of shrimp sizes suggested a gear capture inefficiency for individuals <5mm CL; therefore, data only for shrimps ≥5 mm CL (2,419 shrimps) were retained for further analysis (Fig. A1). Of the retained shrimp, 1,573 were identified as *F. duorarum* and the remaining 844 were identified as farfantepenaeids due to difficulties with species identification of individuals <8 mm CL. Of the 1,937 individuals with measured CL, 1,931 individuals (79.9%) were considered juveniles (≤17.5 mm CL) and the remaining 36 individuals were subadults.

Pink shrimp density observations ranged from 0 to 13.0 shrimp m^{-2}; 105 instances (11.2%, N = 940 samples) of densities ≥2 shrimp m^{-2} were observed, while no penaeid shrimps were observed in 377 samples (40.1%). Overall, shrimp density averaged 0.86 (SD = 1.32) shrimp m^{-2}, was significantly lower than the 2 shrimp m^{-2} threshold ($t_{(\alpha=0.10,2),939} = -26.53$, $P < 0.0001$). Average density in any year-season was always < 2.0 shrimp m^{-2} (Table 3.1), although the highest year-season density (2014 Dry: 1.62 ± 2.02 shrimp m^{-2}; Table 3.1) was the only case that did not significantly differ from 2 shrimp m^{-2} ($t_{(\alpha=0.10,2),46} = -1.30$, $p > 0.10$). Averaged over all sites, mean dry season shrimp densities were higher than those of those of the subsequent wet season 50% of the time. Averaged over year-seasons, the highest mean site density was 2.15 (±1.95) shrimp m^{-2} at site 33 (Table 3.2). Seven sites (7, 10, 12, 33, 34, 43, and 44: 14.9%) exhibited temporally averaged densities that did not significantly differ from 2.0 shrimp m^{-2} ($t_{(\alpha=0.10,2),19}$ = -0.080, -1.69, -0.30, 0.34, -1.63, -1.57, -0.32, respectively; $p > 0.10$). Most sites (n =33, 70.2%) exhibited average densities below 1 shrimp m^{-2} (Table 3.2).

Temperatures ranged from 12.49 to 36.06 °C. Across year-seasons, average temperatures demonstrated a clear pattern of cooler (22.73 ± 2.65 °C) and warmer (30.65 ± 1.53 °C) values for dry and wet seasons, respectively (Table 3.1, Fig. 3.3C). This difference was anticipated given the late-winter/early-spring and late-summer/fall sampling regime. The dry season record was punctuated by an extreme cold front event that occurred during the 2010 field sampling. No pattern of variation in average temperatures among sites was readily discernable (Table 3.2, Fig. 3.3C). Salinities ranged from 2.48 to 39.71 ppt; overall average salinity was 23.64 (± 7.05) ppt (Table 3.2). Spatially averaged wet season salinities were generally lower than those of dry seasons although 2011, 2014, and 2015 wet seasons were notable exceptions with higher average salinity than both the preceding and following dry seasons (Table 3.1). Across year-seasons, sampling sites' mean salinity and standard deviation of salinity negatively correlated with each other (Pearson $r = -0.63$, $t = -5.49$, d.f. = 45, $p < 0.0001$; Fig. A2). Wet 2011 and 2015 were deemed "hypersaline" due to duration (average = 16.91 and 11.54 d, maximum = 54.50 and 32.65 d for wet 2011 and 2015, respectively) and frequency (average = 0.18 and 0.12, maximum = 0.36 and 0.34 of observations for wet 2011 and 2015, respectively) of hypersaline (>40 ppt) observations recorded by in-situ continuous salinity loggers (Lirman et al. 2016). Only four temporally averaged site salinities were mesohaline, most (n = 42) were polyhaline, and one was euhaline (Table 3.2). Water depths ranged from 0.19 to 1.5 m and averaged 0.74 (± 0.20) overall (Table 3.2) with no appreciable trends among year-seasons or among sites (Fig. 3.3E). Total SAV % cover ranged from 4.57 to 100% and averaged 66.57% (± 21.97) with no clear year-season variation patterns (Table 3.1, 3.2; Fig. 3.3F). A planktonic microalgal bloom event was observed in parts of the Biscayne Bay coastal area during the 2013 wet season (Lirman et al. 2016, Wachnicka and Browder 2016).

3.4.1 Habitat Limitations on Pink Shrimp Density

Of the multiple habitat attributes investigated, logical and significant QR analyses revealed that temperature (°C), salinity (ppt), water depth (m), and SAV (% cover) potentially limited pink shrimp density (Table 3.3, Fig. 3.2). QR of density vs. temperature yielded a single-knot natural cubic spline relationship. This relationship was roughly dome-shaped that maximized at 26.6 °C, with tails that tapered off at higher and lower temperatures (Table 3.3, Fig. 3.2A). Temperatures between 21.08 and 31.33 °C did not appear to limit pink shrimp densities to <2 shrimp m^{-2} (Fig. 3.2A). Although a series of functional shapes was considered for the QR density vs. salinity response curve, only the linear and log-linear responses were found to be both significant and ecologically plausible (Fig. A3, Table A1). It was decided the log-linear response was more plausible than the linear response as the former implied a saturation of response that likely represented habitat saturation. The log-linear response suggested severe density limitation below10 ppt and a near-linear increase at salinities above 10 ppt (Fig. 3.2B).

Salinities less than ~18 ppt limited shrimp density to <2 shrimp m^{-2} (Fig. 3.2B). QR of pink shrimp density against water depth (m) yielded a 3 knot (0.25, 0.5, 0.75 quantile) splined relationship with rapid increases in limitation below ~0.6 m and above ~ 1.0 m and a bimodal midsection (Fig. 3.2C). Apparent limitation of density to <2 shrimp m^{-2} occurred at water depths less than 0.43 m and greater than 1.05 m (Fig. 3.2C). Shrimp density had a logarithmic linear relationship with SAV (% cover: Fig. 3.2D). SAV cover less than 45% limited density to <2 shrimp m^{-2} (Fig. 3.2D). For the four habitat attributes, significant QRs were observed only at the 0.9 percentile (Table 3.3). Given that salinity was a major focus of this study, the suite of functional responses considered for QR between density and salinity are presented in the Appendix (Fig. A2, Table A1).

3.4.2 Spatiotemporal Relationships

Heatmap visualization of pink shrimp spatiotemporal density trends revealed a general absence from sites 13 to 28 (approximately Black Point to Fender Point, Fig. 3.1) and sites 45 to 47 (near Turkey Point, Fig. 3.1) across all year-seasons (Fig. 3A). Within these groups of sites, only 16 (4.4%, N = 360) and 4 (6.7%, N=60) instances, respectively, of pink shrimp densities >2 shrimp m^{-2} were observed. Generally higher densities were observed in sites 31 through 44 and sites 1 to 12, where 42 (15%, N=280) and 40 (16.7%, N = 240) instances, respectively, of densities >2 shrimp m^{-2} were observed across all year-seasons. Densities were particularly low during 2007, 2009, 2013, and 2014 wet seasons, when shrimp were absent from a high proportion of samples (55.3, 51.1, 70.2, and 76.6%, respectively: Fig 3.3A). These collections exhibited among the lowest average densities (Table 3.1). Other year-seasons (2008 dry, 2009 dry, 2012 dry, 2012 wet, 2014 dry, and 2015 wet: Fig. 3.3A) exhibited high average density because of a preponderance of higher density observations, which offset low and zero-catch observations from Black Point to Fender Point (Fig. 3.1). Average densities in these year-seasons yielded the highest year-season average densities, which were all >1 shrimp m^{-2} (Table 3.1).

Heatmaps were also developed to visualize spatiotemporal trends in temperature, salinity, water depth, and SAV (Fig. 3.3C, D, E, F). Procrustean analyses revealed significant concordance of the shrimp density matrix to water depth, temperature, and salinity habitat attribute matrices but not to the SAV matrix (Table 3.4). Water depth and temperature exhibited the highest correlations, followed by salinity (Table 3.4). High $m^2_{X,Y}$ values indicated high residual sum of squares for each comparison and relatively weak explanatory power of individual habitat attributes (Table 3.4). Procrustean fitting procedures explained 28.3, 27.1, and 22.1% of the variability in density (r^2) for water depth, temperature, and salinity, respectively.

SIMPROF testing identified six significant site clusters and four significant year-season clusters (Fig, 3.3B). Three site clusters included only four or fewer sites. Two large site clusters, together, included most of the sampling stations; one contained 16 sites (cluster 2) exhibiting sporadic, intermediate densities and the other had 15 sites (cluster 6) that generally exhibited intermediate to higher densities (Fig. 3.3B). Two year-season clusters consisted of only single sampling events, one cluster included three year-seasons, and one large cluster contained the rest of the year-seasons (n = 15) (Fig. 3.3B). The smaller clusters, which contained mainly wet seasons (80%), generally exhibited lower pink shrimp densities. PERMANOVA testing of cluster membership confirmed SIMPROF results for both site ($F_{5,41} = 4.765$, $p = 0.001$, $R^2 = 0.368$) and year-season ($F_{7316} = 3.727$, $p = 0.001$, $R^2 = 0.411$) clustering. However, testing of multivariate homogeneity of dispersions identified significant differences among cluster dispersions of both sites ($F_{5,41} = 14.886$, $p < 0.0001$) and year-seasons ($F_{3,16} = 22.987$, $p = <0.0001$). Differences in dispersion were likely influenced by the large variation in clusters' membership sizes (i.e., number of sites or year-seasons). Within the year-season distance matrix, a 2-factor test with PERMANOVA showed significant season ($F_{1,9} = 1.912$, $p = 0.0063$), but not year ($F_{9,9} = 1.020$, $p = 0.4279$), categorical temporal effects. Multivariate dispersions also differed significantly between seasons ($F_{1,18} = 7.047$, $p = 0.0161$) and years ($F_{9,10} = 5.56*10^{29}$, $p < 0.0001$). When greater multivariate dispersion is observed in larger sample sized groups, PERMANOVA p values tend to be conservative. Therefore, although differences in dispersion were noted, this presumably did not hinder detection of differences given significance levels observed with the PERMANOVA tests. Although not presented here, plots of multivariate dispersion test results on principle coordinate axes revealed greater variability in the wet season than in the dry season.

3.4.3 Pink Shrimp Density and Habitat Attributes Among Density Clusters

Significant differences in density distributions were detected among both site and year-season clusters (Table 3.5, Fig. A3A, Fig. A4A). Site clusters represented three relative median density levels: high (~0.7 shrimp m^{-2}: site cluster 6); intermediate (~0.3 shrimp m^{-2}: site clusters 2, 3, and 5); and low density (0.0 and ~0.14 shrimp m^{-2}: site clusters 1 and 4, respectively: Table 3.5A, Fig. A3A). Year-season clusters also exhibited three relative density levels: high (0.51 shrimp m^{-2}: year-season cluster 2), intermediate (0.29 shrimp m^{-2}: year-season cluster 4) and low density (0.0 shrimp m^{-2}: year-season clusters 1 and 3) (Table 3.5B, Fig. A2A). Significant differences were also observed for salinity, water depth, and SAV distributions among site clusters (Table 3.5A; Fig. A3B, C, D), while temperature distributions did not differ among site clusters. All four habitat attributes exhibited significant differences among year-season clusters (Table 3.5B, Fig. A2B, C, D, E). No significant correlations were found between site cluster median density and clusters' median, minimum, maximum, or standard deviation of habitat attributes. Low sample size prevented investigation of correlations of year-season clusters' median density or maximum density with clusters' habitat attributes' distribution characteristics.

3.5 Discussion

CERP implementation was expected to result in more favorable salinity conditions for pink shrimp, leading to higher shrimp densities (Browder et al. 2005a, RECOVER 2008, Browder and Robblee 2009). The analysis of 10 years of monitoring data revealed few instances (11.2 %) of pink shrimp densities > 2 shrimp m^{-2}, the IG for Biscayne Bay pink shrimp populations (RECOVER 2008). All but one spatially averaged year-season density and all but a few temporally averaged site densities in the 2007-2016 database were significantly below 2 shrimp m^{-2}. Furthermore, only one average site density and no average year-season densities were above the target. When CERP is implemented and the salinity environment of the southern half of the study area (i.e., Black Point to Turkey Point: Fig. 3.1) becomes less volatile, then perhaps average densities > 2 shrimp m^{-2} across the entire system could be achieved following periods of high postlarval recruitment.

However, my results suggest that salinity conditions of ~ 8ppt (i.e., low mesohaline to oligohaline: Fig. 3.2B) potentially represented a threshold below which pink shrimp densities were more severely limited. Above this threshold, the log-linear response continued to increase in a more linear fashion. Re-analyses of data presented by Brusher and Ogren (1976) and Minello (1999) by Zink et al. (2017; Chapter 2) found increasing abundance with increasing salinity and no statistical difference between polyhaline and mesohaline pink shrimp abundances. Limitation of pink shrimp densities at salinities <8 ppt does not support coexistence of CERP post-restoration IGs of >2 shrimp m^{-2} and reduction of Biscayne Bay nearshore salinity regimes to oligohaline and low mesohaline conditions. It was unexpected to not find a limitation of pink shrimp density at higher salinities, especially at conditions >35 ppt (e.g., a significant, negative quadratic term or significance the cubic splines functions in Fig. A2, Table A1). The range of salinity values observed in the study did not include hypersaline values (>40 ppt). Perhaps this range of salinity observations was not broad enough to characterize suspected reduction of densities in extreme hypersaline conditions (Browder and Robblee 2009).

Due the field sampling design, the present results apply only to extreme nearshore areas (≤50 m). Application of the present results to further offshore areas should proceed with caution, if done at all, due to potential interaction with other habitat attributes that influence trends in pink shrimp density. Indeed, I found that SAV and water depth both exhibit limitations on pink shrimp densities. These two habitat attributes can change rapidly in nearshore areas >50 m from the coast, and therefore the present results may not apply to those areas.

Procrustean analysis confirmed direct relationships between salinity and density observations, but suggested that salinity was less influential than temperature (i.e., seasonal) or water depth on spatiotemporal density patterns. This finding is consistent with Rubec et al. (2016), who found multiple habitat attributes, including salinity, temperature, dissolved oxygen concentration, water depth, and benthic habitat characteristics, influenced summer-season pink shrimp occurrence and concentration in Tampa Bay, Florida. Similarly, Berkeley and Campos (1984) found in multiple regression modeling that sampling station standard deviation of turbidity, median sediment size, density of seagrass, and density of hard bottom assemblages all explained greater abundance variability in Biscayne Bay pink shrimp abundance than station salinity standard deviation, the only significant salinity metric within the model. As re-iterated by Zink et al. (2017; Chapter 2), Costello et al. (1986) stated that "…factors other than salinity per se control abundance of the euryhaline juveniles…"

The stretch of coastline from Black Point to Convoy Point (Fig. 3.1) was dominated by membership within one low (cluster 1) and one intermediate (cluster 2) density site clusters. This zone is well recognized as an area strongly influenced by canal discharges (Serafy et al. 1997; Caccia and Boyer 2005; Lirman et al. 2008a, 2008b, 2014; Santos et al. 2011; Stabenau et al. 2015). Salinity reductions that can be both rapid (<60 min to 2 d) and extreme (~25 ppt) occur along this stretch of coastline (Wang and Cofer-Shabica 1988; Irlandi et al. 1997; Lirman and Cropper 2003; Lirman et al. 2003, 2008a, b). These rapid salinity fluctuations alter fish community assemblages (Serafy et al.

1997) and can cause mortality and altered foraging behavior (Irlandi et al. 1997, Serafy et al. 1997). Pink shrimp may avoid these deleterious conditions just as they have been reported to migrate to avoid large-volume riverine inflows (Ingle 1957). Rapid salinity reductions of greater than 20 ppt cause near complete mortality in laboratory settings (Williams 1960, Zein-Eldin and Aldrich 1964, Criales et al. 2011a). BBCW implementation is focused on the same stretch of coastline covered by the monitoring data of this study; perhaps moderation in salinity fluctuations in this area could enable the CERP pink shrimp IG to be attained across this system.

Interestingly, site clusters 1 and 2 included sites 45, 46, and 47, which are located at Turkey Point and exhibited moderate minimum salinities (≥11.06 ppt) and the highest average salinity across all sites (≥29.57 ppt: Table 3.2). These sites also generally have high SAV cover (Table 3.2, Fig. 3.3F). Perhaps limited recruitment of pink shrimp to southern reaches of the bay did not allow for higher abundances (Ault et al. 199b, Wang et al. 2003). However, the presence of nearby sites (40, 41, 43, 44) that clustered with other "high" density sites (cluster 6: Fig. 3.1) does not support this notion. The cause of low pink shrimp density in these sites remains undetermined but does not seem to be related to limitation due to salinity or SAV cover conditions. Site cluster 4, with site 42 as its only member, also exhibited an intermediate to low median density (0.14 ± 0.20 shrimp m^{-2}: Table 3.5A). The minimum salinity at site 42 was also moderate (13.46 ppt) and the site's average salinity was also among the highest of the study (28.47 ± 5.85 ppt: Table 3.1). No reason(s) can presently be provided to explain this observation, but perhaps the density pattern observed at site 42 is unique due its close proximity to what was previously the Florida City Canal mouth, the only canal in this region of Biscayne Bay to be plugged from discharging directly to the bay.

Site cluster 6 (Fig. 3.1) exhibited the highest median density and was comprised of sites not located within the stretch of shoreline highly influenced by canal discharges (i.e., Black Point to Convoy Point). This site cluster exhibited the second highest minimum salinity (9.62 ppt), which was the only other cluster higher than the apparent ~8 ppt limitation. The sites comprising site cluster 6 were generally further from canal mouths and exhibited higher minimum salinities (>10 ppt). Previous field (Ault et al. 1999a, Diaz 2001) and modeling (Ault et al. 1999b, Wang et al. 2003) studies describe the area corresponding to the more northern sites of this cluster as an area of relatively high shrimp abundance. The proximity of these northern sites to a large ocean inlet known as “the safety valve”, considered a primary postlarval immigration pathway (Ault et al. 1999b, Wang et al. 2003), may have contributed to their high densities (Hovel et al. 2002, Blanco-Martínez and Pérez-Castañeda 2016). Indeed, shrimp size frequencies were found to differ between these northern (sites 1 - 17) and southern (sites 18 – 47) sampling sites. The shrimp size (7.54 mm CL) associated with the maximal size distribution difference suggested a higher number of juvenile sized shrimps occurred in the north, which could be caused by differences in recruitment, growth and/or mortality, or both factors.

However, several other cluster 6 sites also exhibited high density despite being located towards the southern part of the sampling domain, known for deleterious salinity fluctuations (Lirman et al. 2003, Lirman et al. 2008a, Appendix A). These sites are located near mangrove creeks that drain more natural watersheds, although their watersheds are likely reduced due to inland canalization. Similarly, several sites, both northern and southern, are located near, but not immediately adjacent to, canals that discharge relatively small volumes of freshwater (Military Canal: 1994-2003 annual mean canal output = 21.9 cfs; Cutler Drain C-100: 1994-2003 annual mean output = 46.1 cfs; Caccia and Boyer 2005). One might expect that segments of the shoreline that experience lower-volume freshwater discharges have consistently higher shrimp densities. However, sites that were located closest to low-volume discharge canals (i.e., site 6 and site 32) clustered with intermediate and low density site clusters (cluster 1 and 2, respectively: Fig 3.1, 3.3B, 3.4A).

Most year-seasons (75%) were aggregated within one large cluster, indicating a general lack of inter-annual and inter-season variability in Biscayne Bay juvenile pink shrimp populations. However, the majority (60%) of the year-seasons comprising year-season cluster 2 were dry seasons. Sampled densities were higher in the dry season than the wet season. However, the pink shrimp IG presently focuses on improvement of "peak" fall (wet) season abundances (RECOVER 2008). A low between-season difference in shrimp size distributions agreed with the notion that shrimp abundances did not differ greatly on a temporal scale. However, the shrimp size associated with this significant difference (5.53 mm CL) pointed out that abundance of small juveniles – presumably due to recent postlarval recruitment - caused this seasonal difference. Temperature QR revealed a peak in density at 26.6 °C, which is lower than late summer/early fall water temperatures recorded during this study. A strong seasonal pattern was also suggested by Procrustean analysis, with temperature yielding one of the higher concordances of the habitat attributes tested. Although this pattern was not consistent, multiple lines of evidence suggested generally higher abundances, and more recent recruitment, of shrimp in the Dry Season.

The pink shrimp IG was based upon observation of a summer/fall (i.e., wet season) peak in abundance (Browder et al. 2005b). This summer/fall peak abundance agreed with Diaz (2001), who reported June and September peak abundances of ~ 2 shrimp m^{-2} from the same spatial domain. However, Diaz's (2001) sampling was limited to four months: June, July, August, and September. Unlike these studies, Ault et al. (1999a) and Berkeley and Campos (1984) reported peak juvenile abundances in late fall/early winter (i.e., November/December). Further, Diaz (2001) estimated maximal Biscayne Bay juvenile pink shrimp population level occurred in November (i.e., late fall). However, differences in sampling gear and spatial extent of sampling, both of which could lead to targeting of different sized and ontogenetic stage shrimps, complicate comparisons between these studies and the present study. Further, the relatively short durations (≤2 yr) of these four reference studies prevented examination of longer temporal abundance patterns (Browder et al. 2005b, Diaz 2001, Ault et al. 1999a, Berkeley and Campos 1984). Although of greater duration, the present study's bi-seasonal sampling effort may be insufficient to identify the period of the year of peak pink shrimp density and especially if that changes from year to year.

A lack of understanding of Biscayne Bay pink shrimp recruitment complicates study of their abundance patterns: perhaps year-season cluster 4 is the result of poor recruitment as there is currently no other explanation for the low densities observed within this cluster. The only study available on Biscayne Bay postlarval pink shrimp recruitment reported a late fall through early winter peak (i.e., October through March), although the study's short duration (1 yr) limited consideration of inter-annual trends (Criales et al. 2000). This peak agreed with juvenile abundance studies reporting a late fall/early winter peak (Ault et al. 1999a, Berkeley and Campos 1984). Pink shrimp postlarval recruitment modeling found that oceanographic processes favored Florida Keys potential recruitment during late wet season and early dry season months (Criales et al. 2015) - presumably these conditions also favor Biscayne Bay recruitment. Modeling of larval permit *Trachinotus falcatus* originating from spawning grounds near those of pink shrimp also found similar recruitment patterns for the Florida Keys and Biscayne Bay, although no seasonal (i.e. spring and fall) difference in magnitude was observed (Bryan et al. 2015). Oceanographic, coastal, and climatic processes and conditions affect pink shrimp adult reproductive activity (Cummings 1961, Kennedy and Barber 1981) and larval abundances (Jones et al. 1970). These processes interact with behavior to influence recruitment of early life stages to nearshore areas from offshore spawning sites such as the Tortugas (Tabb et al. 1962; Allen et al. 1980; Criales et al. 2003, 2005, 2006, 2007, 2010, 2011b, 2015). Use of ecological indicators is complicated by life cycles affected by unrelated, prior, external and stochastic conditions (Serafy et al. 2007, Browder and Robblee 2009).

Several environmental perturbations occurred during this study. Variability in climatic conditions led to both wetter and drier than normal wet seasons (Fig. 3.3D). Lower salinities (3.34 to 22.08 ppt) across most of the spatial domain (44 sites: 93.6%) accompanied record rainfall during the 2012 wet season and coincided with the second highest wet season pink shrimp average density (1.29 ± 1.65 shrimp m^{-2}) observed during the 10 yr record (Table 3.1). Conversely, the highest wet season pink shrimp average density (1.45 ± 2.25 shrimp m^{-2}: Table 3.1) observed during the study occurred during the 2015 wet season, which was denoted as a “hypersaline” period (Fig. 3.3A). Record dry season rainfall occurred during the 2016 dry season, yielding the lowest average dry season salinity (Table 3.1), and low salinity observations across most of the study’s spatial domain (Fig. 3.3D). The 2016 wet season also saw the lowest average salinity observed in the 10-yr record (Table 3.1) and extended lower salinity conditions across the entire study spatial domain (Fig. 3.3D). Mean shrimp densities in 2016 dry and wet seasons were 0.84 (1.18) and 0.96 (0.93), respectively, and were moderate compared to the other year-seasons examined (Table 3.1). Despite the range of salinity conditions observed in these four differing year-seasons, they all were assigned to the same density cluster, suggesting a general lack of salinity influence on the density trends (Fig. 3.3B). The only pattern possibly suggesting an environmental disturbance negatively impacting pink shrimp abundance is the microalgal bloom that occurred during the 2013 wet season (Lirman et al. 2016, Wachnicka and Browder 2016). This year-season clustered separate from all other year-seasons (Fig. 3.3B) and exhibited a median density of 0.00 (±0.051 shrimp m^{-2}: Table 3.5B). Although an extreme cold front event occurred during the 2010 dry season, no impact on pink shrimp densities was observed.

The Biscayne Bay pink shrimp IG suggested a pink shrimp preference for seagrasses, and presumed that increased % cover of seagrasses would increase pink shrimp abundance (RECOVER 2004, RECOVER 2008, Browder and Robblee 2009). Besides increased % seagrass cover, CERP restoration is also expected to increase the seaward spatial extent of *H. wrightii* (RECOVER 2004). Total SAV QRs yielded the most plausible relationship of the benthic habitat metrics investigated. Although 21.2%

of the variability between SAV % cover and pink shrimp density matrices was accounted for in Procrustean analysis, test results were non-significant. The seemingly weak statistical relationships with either total or species-specific SAV metrics was unexpected. Pink shrimp associations with *H. wrightii* have been previously reported (Eldred et al. 1961, Eldred 1962, Costello et al. 1986, Johnson et al. 2002). Although most studies reporting pink shrimp benthic habitat associations focused on seagrasses, three reported maximal pink shrimp densities relative to total SAV biomass or % cover of multiple SAV taxa, including both seagrass and macroalgae (Gore et al. 1981, Yokel 1983, Santos 2014). However, Pérez-Castañeda et al. (2010) found negative impacts of drift and attached algal biomass on farfantepenaeid recruit (< 8.0 mm CL) abundance. Conflicting results between the present study and relationships reported in the literature indicate further study of pink shrimp associations with SAV is needed. However, indirect influences of salinity on pink shrimp density because of increased SAV % cover and reduced fragmentation due to reduced salinity variability (Lirman et al. 2008a, Santos et al. 2011) would likely increase pink shrimp abundance.

Water depth explained ~28% of pink shrimp density variability, the most of any habitat attribute presently investigated. The strong influence of water depth was unexpected given the narrow spatial sampling domain adjacent to the mangrove-seagrass ecotone. Associations between nearshore pink shrimp abundance and depth have been previously reported, although these studies included a greater depth range than the present study (Berkeley and Campos 1984, Ault et al. 1999a, Sheridan and Minello 2003, Rubec et al. 2016). Other studies that focused on very nearshore areas (<100 m) also found higher abundances there (Costello et al. 1986, Diaz 2001, Johnson et al. 2002, Browder et al. 2005b, Santos 2014). Recruiting postlarval pink shrimp often concentrate in SAV near the low-tide mark along shorelines (Williams 1955, Eldred 1962, Costello and Allen 1966, Allen and Hudson 1970, Costello et al. 1986, Murphy and Fonseca 1995, Diaz 2001, Pitre 2001). Decreased throw trapping detection probability of pink shrimp with increasing water depths may complicate the abundance-water depth relationships (Robblee et al. 2008). Both limitations of tide on distribution and reduced abundance and/or detection probability at greater depths likely contributed to the domed shape of the QR relationship. Perhaps movements concurrent to flooding and ebbing tides resulted in a concentration of shrimps along the tidal front, causing the bimodal QR distribution.

Results of this study (QR) identified four habitat attributes exhibiting limitation of pink shrimp densities, two of which (i.e., salinity regime and SAV % cover) can be influenced by freshwater management. It should be noted that these habitat attributes vary at differing time scales; for example, water depth can differ by as much as 1.3 m within 12 hr during extreme tidal cycles while SAV % cover may be integrative of salinity, nutrient, water clarity, and other influential factors on the order of 6 mo. to 1 yr. Procrustean analysis revealed a significant one-to-one correspondence between shrimp density observations and water depth, temperature, and salinity observations, but not with SAV % cover. The lack of significance despite a correspondence correlation between SAV and density similar in magnitude to the other habitat attributes may have been due the smaller sample size that reduced power in this analysis (SAV was only available for 9 yr instead of 10 yr). SIMPROF analysis clustered shrimp density observations into groups exhibiting statistically similar density patterns in space (sites) or time (year-seasons). However, median pink shrimp density within spatial clusters did not correlate with either the median, maximum, minimum, or standard deviation of the four habitat attribute groups corresponding to the shrimp density clusters. The lack of significance for spatial density clusters may have been due to low testing power or another environmental factor not considered in these analyses. Further investigation is warranted. For temporal density clusters, correlation analysis could not be applied because too few of clusters were formed, suggesting a general similarity of inter-year-season density patterns.

The study was limited by apparent low catchability of recently settled pink shrimp by the throw trap gear. Pink shrimp postlarvae are generally considered settled in their nursery habitat by 3 mm CL (Chapter 2). Pink shrimp postlarvae settle in the shallow (≤1 m), calm water areas along shorelines (Eldred et al. 1961, Costello et al. 1986) which would suggest they should be readily available to the present field sampling program that samples nearshore waters generally < 1 m deep. While it would be expected that recently settled pink shrimp would be more numerous than larger shrimps given mortality processes, frequency histograms revealed lower numbers of pink shrimp less than 5 mm CL (Fig. A1). This was attributed to a size-related catchability for the throw-trap sampling gear, and further analyses were appropriately adjusted. However, this situation prevented inclusion of recently settled postlarval shrimp in spatio-temporal or density limitation investigations.

The RECOVER Biscayne Bay IG anticipates >2 shrimp m^{-2} as a target wet season pink shrimp density to be achieved with CERP BBCW implementation. But achievement of BBCW and CERP salinity IGs, which include low mesohaline (<10 ppt) and even oligohaline conditions (<5 ppt), may negatively impact pink shrimp density. On the other hand, alteration of salinity regimes is anticipated to increase seagrass % cover, the major component of total SAV cover, which would lessen limitation of pink shrimp abundances due to low SAV % cover. Dry season densities were higher than wet season densities in all but one year covered: 2016. The Biscayne Bay pink shrimp IG may need modification to clarify whether the ≥ 2 shrimp m^{-2} target refers to all monitoring observations, or a seasonal or annual average density across the entire shoreline and to further consider spatial and seasonal abundance patterns.

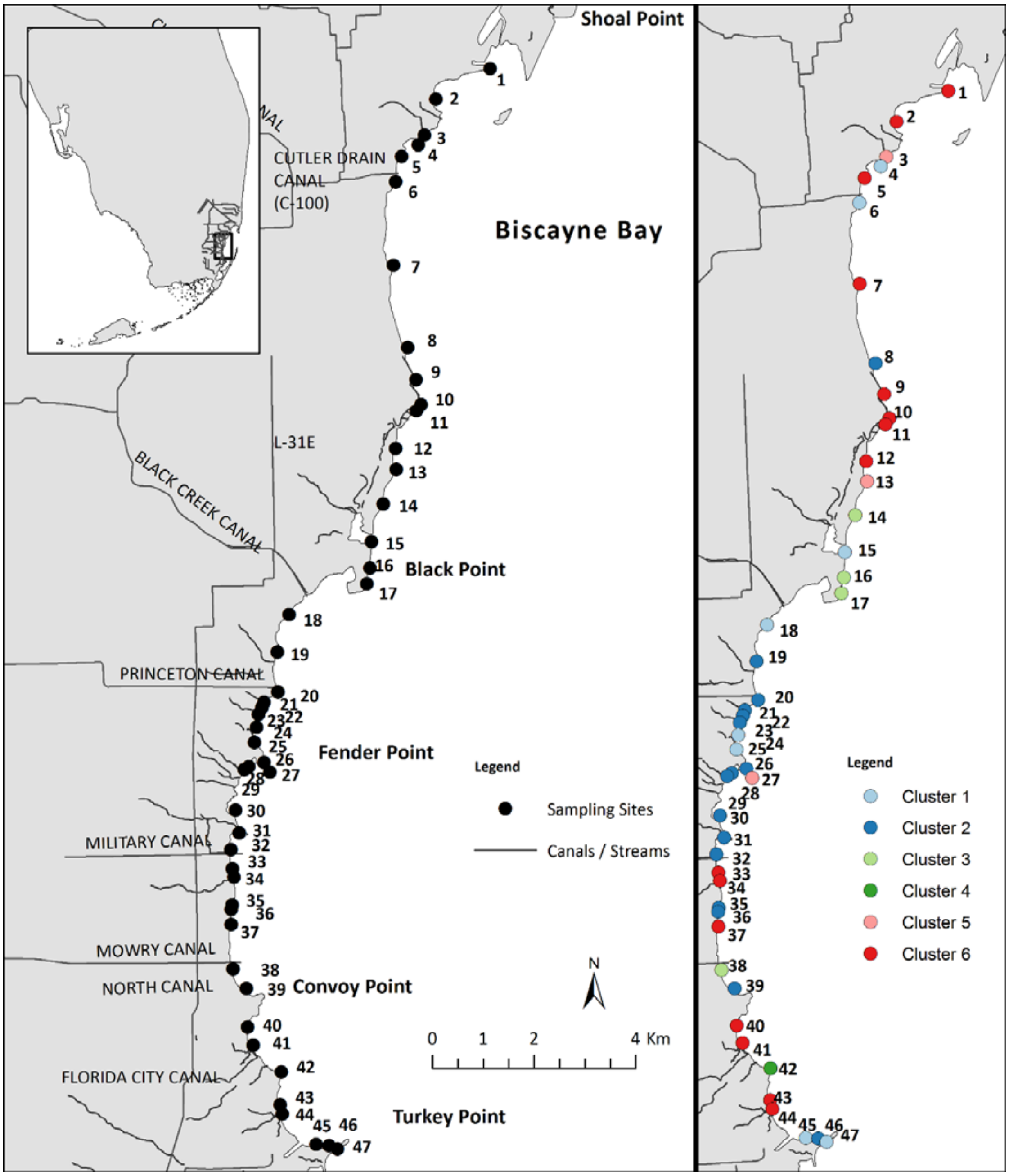


Fig. 3.1: Map of study area, including referenced geographical features, and location of survey sites. The second panel depicts the same sites color-coded to match significant site clusters (Fig. 3.3B and 3.4).

Table 3.1: Number of pink shrimp collected, average pink shrimp density (± SD), and average (± SD) of water quality and habitat attributes for survey year-seasons.

Year	Season	# Shrimp	Density (# m^{-2})	Temp (°C)	Sal (ppt)	Depth (m)	SAV (% Cover)
2007	Dry	131	0.93 ± 1.11	23.90 ± 1.99	26.11 ± 5.62	0.67 ± 0.18	NA
	Wet	63	0.45 ± 0.9	30.81 ± 1.07	20.36 ± 5.98	0.72 ± 0.17	NA
2008	Dry	165	1.17 ± 1.95	22.29 ± 1.44	25.28 ± 4.12	0.73 ± 0.17	52.45 ± 26.52
	Wet	104	0.74 ± 1.09	29.84 ± 0.99	23.57 ± 5.32	0.73 ± 0.17	70.38 ± 15.97
2009	Dry	198	1.4 ± 1.4	21.61 ± 1.53	25.95 ± 5.61	0.63 ± 0.15	78.36 ± 14.13
	Wet	56	0.4 ± 0.58	31.25 ± 1.85	22.93 ± 6.47	0.65 ± 0.15	75.25 ± 20.76
2010	Dry	98	0.7 ± 0.94	19.28 ± 2.89	25.50 ± 2.90	0.59 ± 0.14	62.28 ± 23.08
	Wet	123	0.87 ± 1.34	31.62 ± 1.17	24.18 ± 6.92	0.71 ± 0.20	63.46 ± 31.63
2011	Dry	98	0.7 ± 0.9	21.31 ± 1.83	27.09 ± 3.02	0.60 ± 0.18	72.35 ± 19.22
	Wet	112	0.79 ± 0.99	31.72 ± 1.56	31.86 ± 3.68	0.73 ± 0.22	69.96 ± 22.34
2012	Dry	182	1.29 ± 1.73	22.52 ± 1.00	24.47 ± 3.45	0.71 ± 0.16	83.74 ± 12.07
	Wet	182	1.29 ± 1.65	31.10 ± 1.85	15.30 ± 5.64	0.73 ± 0.15	65.44 ± 16.61
2013	Dry	82	0.58 ± 0.68	21.45 ± 1.07	28.85 ± 3.41	0.86 ± 0.18	57.98 ± 19.80
	Wet	29	0.21 ± 0.43	29.30 ± 0.87	15.98 ± 7.24	0.79 ± 0.18	60.07 ± 17.74
2014	Dry	228	1.62 ± 2.02	22.89 ± 1.39	23.33 ± 5.71	0.69 ± 0.15	61.41 ± 20.49
	Wet	15	0.11 ± 0.22	29.44 ± 1.12	29.30 ± 4.84	1.02 ± 0.20	53.20 ± 20.96
2015	Dry	92	0.65 ± 0.81	26.06 ± 1.48	28.85 ± 3.42	0.74 ± 0.19	68.51 ± 19.71
	Wet	204	1.45 ± 2.25	30.87 ± 1.45	23.11 ± 6.19	0.80 ± 0.17	64.23 ± 19.75
2016	Dry	119	0.84 ± 1.18	26.03 ± 2.06	18.60 ± 6.08	0.77 ± 0.20	60.63 ± 21.48
	Wet	136	0.96 ± 0.93	30.28 ± 1.22	12.22 ± 2.94	0.83 ± 0.17	59.73 ± 21.19
Overall	Dry	1393	0.99 ± 1.38	22.73 ± 2.65	25.40 ± 5.27	0.70 ± 0.19	66.54 ± 22.00
	Wet	1024	0.73± 1.25	30.65 ± 1.53	21.87 ± 8.10	0.77 ± 0.20	64.64 ± 21.93

Table 3.2: Number of pink shrimp collected, average pink shrimp density (± SD), and average (± SD) of water quality and habitat attributes for survey sites.

Site	# Shrimp	Density (# m^{-2})	Temp (°C)	Sal (ppt)	Depth (m)	SAV (% Cover)
1	73	1.22 ± 1.65	27.42 ± 4.04	29.03 ± 5.09	0.84 ± 0.14	71.44 ± 20.10
2	57	0.95 ± 1.54	26.90 ± 4.23	26.75 ± 4.92	0.88 ± 0.17	70.37 ± 19.51
4	35	0.58 ± 1.49	26.68 ± 4.06	27.78 ± 4.94	0.79 ± 0.2	74.40 ± 16.07
5	66	1.10 ± 1.93	26.78 ± 4.06	25.67 ± 5.09	0.76 ± 0.19	67.94 ± 21.58
6	19	0.32 ± 0.69	26.58 ± 3.89	25.10 ± 5.55	0.65 ± 0.14	79.31 ± 14.20
7	117	1.95 ± 2.80	26.72 ± 4.20	27.05 ± 5.10	0.63 ± 0.15	72.07 ± 15.29
8	27	0.45 ± 0.64	26.68 ± 4.23	26.05 ± 5.63	0.5 ± 0.12	67.45 ± 18.53
9	85	1.42 ± 1.10	26.58 ± 4.00	25.23 ± 5.82	0.59 ± 0.15	51.46 ± 19.84
10	94	1.57 ± 1.15	26.14 ± 3.87	24.97 ± 5.91	0.73 ± 0.18	77.71 ± 18.04
11	80	1.33 ± 1.36	26.31 ± 3.65	24.91 ± 5.99	0.66 ± 0.15	68.94 ± 17.80
12	111	1.85 ± 2.27	26.34 ± 3.64	24.46 ± 6.42	0.86 ± 0.17	73.90 ± 19.70
13	51	0.85 ± 1.11	26.41 ± 3.77	24.53 ± 6.17	0.8 ± 0.15	80.85 ± 10.49
14	45	0.75 ± 1.65	26.70 ± 3.75	23.75 ± 6.17	0.8 ± 0.15	82.69 ± 14.27
15	13	0.22 ± 0.36	26.36 ± 4.10	23.10 ± 5.46	0.75 ± 0.18	71.29 ± 18.34
16	36	0.60 ± 0.88	26.67 ± 3.96	22.78 ± 5.20	0.77 ± 0.21	72.08 ± 22.62
17	57	0.95 ± 1.77	26.58 ± 4.00	23.07 ± 5.26	0.86 ± 0.15	75.96 ± 17.11
18	9	0.15 ± 0.23	26.75 ± 4.13	17.10 ± 9.03	0.64 ± 0.17	70.33 ± 17.95
19	36	0.60 ± 0.65	26.77 ± 4.05	17.71 ± 7.82	0.7 ± 0.17	66.30 ± 21.90
20	38	0.63 ± 0.71	26.76 ± 3.99	17.72 ± 7.59	0.73 ± 0.22	54.80 ± 22.54
21	29	0.48 ± 0.64	26.68 ± 4.37	18.35 ± 6.88	0.68 ± 0.22	59.85 ± 26.64
22	33	0.55 ± 0.60	26.82 ± 4.64	18.93 ± 6.82	0.7 ± 0.2	57.64 ± 23.73
23	30	0.50 ± 0.72	26.84 ± 4.77	17.98 ± 7.25	0.67 ± 0.2	62.61 ± 19.56
24	12	0.20 ± 0.23	27.25 ± 5.18	18.29 ± 7.34	0.64 ± 0.19	66.70 ± 17.74
25	12	0.20 ± 0.35	27.05 ± 4.83	18.20 ± 6.45	0.65 ± 0.15	54.74 ± 23.68
26	27	0.45 ± 0.74	27.08 ± 5.07	20.53 ± 5.51	0.7 ± 0.14	53.51 ± 20.39
27	41	0.68 ± 0.96	26.73 ± 4.74	20.60 ± 5.79	0.67 ± 0.17	66.51 ± 19.81
28	29	0.48 ± 0.51	26.82 ± 4.38	20.51 ± 5.86	0.85 ± 0.19	38.07 ± 21.13
29	49	0.82 ± 1.02	26.82 ± 4.43	20.69 ± 5.78	0.79 ± 0.2	49.53 ± 23.86
30	33	0.55 ± 0.55	26.76 ± 4.09	22.13 ± 5.32	0.74 ± 0.23	53.08 ± 19.99
31	38	0.63 ± 0.69	27.27 ± 4.59	21.92 ± 6.26	0.77 ± 0.19	55.20 ± 23.45
32	43	0.72 ± 0.78	27.49 ± 4.91	22.06 ± 5.89	0.78 ± 0.23	55.56 ± 18.24
33	129	2.15 ± 1.95	26.80 ± 4.61	22.00 ± 6.02	0.82 ± 0.2	64.78 ± 15.60
34	88	1.47 ± 1.46	26.79 ± 4.79	22.50 ± 5.80	0.76 ± 0.22	63.24 ± 18.32
35	52	0.87 ± 0.84	26.90 ± 4.79	22.43 ± 6.96	0.74 ± 0.22	56.12 ± 23.22
36	52	0.87 ± 0.98	26.64 ± 5.11	22.04 ± 6.57	0.73 ± 0.18	53.76 ± 21.81
37	52	0.87 ± 1.13	26.43 ± 4.97	22.18 ± 6.88	0.81 ± 0.2	54.10 ± 22.31
38	33	0.55 ± 0.60	27.00 ± 5.28	21.01 ± 7.61	0.77 ± 0.18	51.91 ± 22.86
39	63	1.05 ± 2.89	27.24 ± 5.93	24.51 ± 6.65	0.84 ± 0.17	41.68 ± 22.78
40	76	1.27 ± 1.28	26.48 ± 5.57	27.16 ± 5.94	0.82 ± 0.17	79.73 ± 12.79
41	74	1.23 ± 1.24	26.31 ± 5.42	27.53 ± 5.76	0.69 ± 0.17	73.72 ± 18.74
42	42	0.70 ± 1.27	26.51 ± 5.34	28.47 ± 5.85	0.85 ± 0.22	66.68 ± 21.59
43	92	1.53 ± 1.33	26.17 ± 5.17	28.51 ± 6.09	0.68 ± 0.17	72.63 ± 17.57
44	113	1.88 ± 1.65	25.98 ± 5.06	28.77 ± 6.18	0.54 ± 0.16	59.35 ± 15.19
45	12	0.20 ± 0.31	26.15 ± 5.71	29.83 ± 5.78	0.72 ± 0.17	89.86 ± 4.83
46	45	0.75 ± 1.00	25.87 ± 5.74	29.57 ± 6.03	0.58 ± 0.19	63.17 ± 17.5
47	10	0.17 ± 0.35	25.82 ± 5.69	30.02 ± 5.87	0.81 ± 0.18	89.86 ± 6.12
Overall	2417	0.86 ± 1.32	26.68 ± 4.52	23.64 ± 7.05	0.74 ± 0.20	65.57 ± 21.97

Table 3.3: Statistical significance of 0.5 and 0.9 quantile regressions of pink shrimp density (shrimp m^{-2}: LN([x+1]) against temperature (°C), salinity (ppt), water depth (m), and submerged aquatic vegetation (SAV: % cover). LN = natural logarithm.

Quantile	Predictors	Coefficients (± SE)	t value	p value
0.5	Spline1(Temperature)	-0.59 ± 0.057	10.39	0.5551
	Spline2(Temperature)	-0.33 ± 0.16	-2.08	0.0375
0.9	Spline1(Temperature)	2.24 ± 0.094	23.80	< 0.0001
	Spline2(Temperature)	-0.49 ± 0.22	-2.26	0.0242
0.5	LN(Salinity)	0.00 ± 0.044	0.00	1.0000
	Intercept	0.29 ± 0.12	2.44	0.0147
0.9	LN(Salinity)	0.26 ± 0.091	3.17	0.0016
	Intercept	0.34 ± 0.28	1.22	0.2213
0.5	Spline1(Water Depth)	0.27 ± 0.053	5.04	< 0.0001
	Spline2(Water Depth)	0.15 ± 0.068	2.26	0.0243
	Spline3(Water Depth)	0.57 ± 0.14	4.07	0.0005
	Spline4(Water Depth)	-0.22 ± 0.16	-1.43	0.1544
0.9	Spline1(Water Depth)	1.07 ± 0.11	10.04	< 0.0001
	Spline2(Water Depth)	0.75 ± 0.23	3.29	0.0011
	Spline3(Water Depth)	2.15 ± 0.18	11.70	< 0.0001
	Spline4(Water Depth)	-0.83 ± 0.32	-2.62	0.0089
0.5	LN(SAV)	0.00 ± 0.036	0.00	1.0000
	Intercept	0.29 ± 0.13	2.18	0.0298
0.9	LN(SAV)	0.21 ± 0.075	2.75	0.0061
	Intercept	0.33 ± 0.30	1.10	0.2733

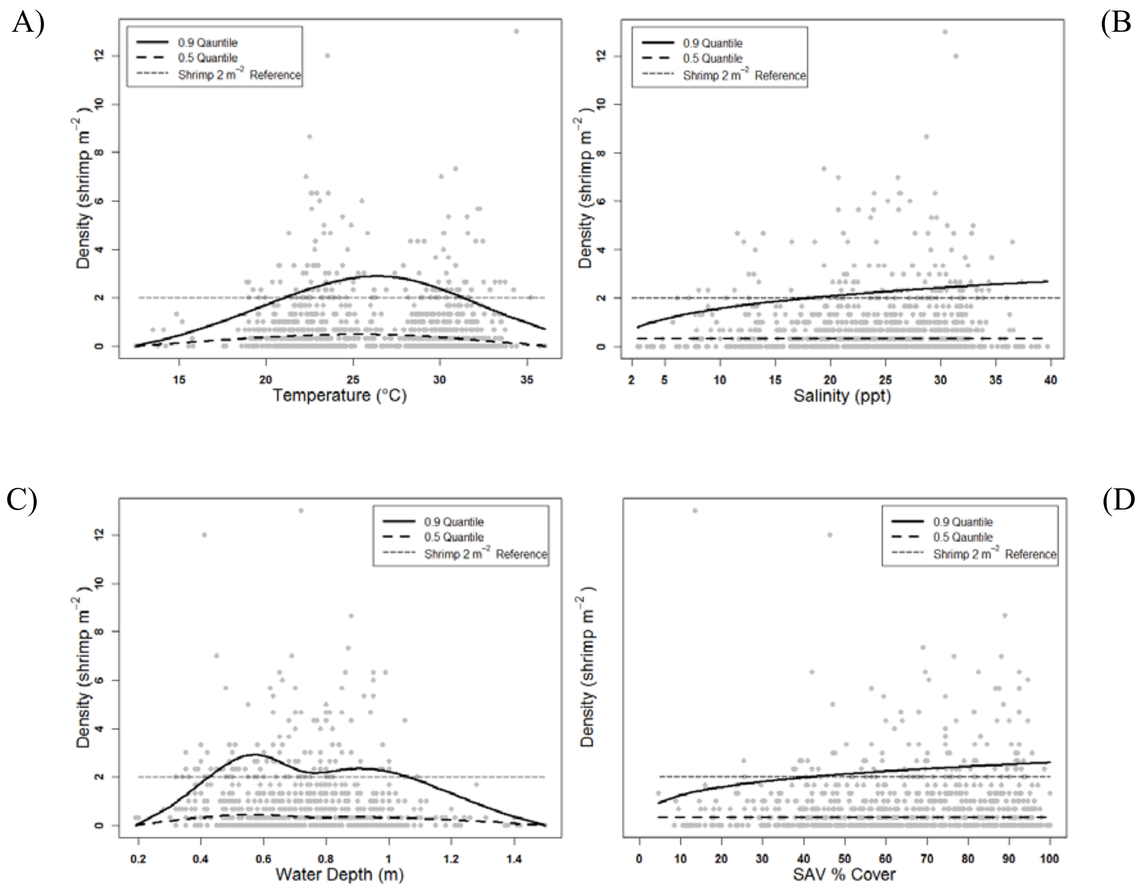


Fig. 3.2: Pink shrimp density (shrimp m^{-2}) and back-transformed 0.50 and 0.90 quantile regressions lines of predicted density (LN x+1) plotted against A) temperature (°C), B) salinity (ppt), C) water depth (m), and D) submerged aquatic vegetation (SAV: % cover). Predicted regression lines depict relationships reported in Table 2.

Fig 3.3: Heatmaps depicting spatial (i.e., site) and temporal (i.e., year-season) trends in A) pink shrimp density (shrimp m^{-2}: LN[x+1]), C) temperature (°C), D) salinity (ppt), E) depth (m), and F) SAV (% cover). Shrimp densities are also depicted as organized (B) by site and year-season clusters. Color bars along the left and top margins of B) reflect significant sites and year-season clusters as denoted in the legend. Black cells in A) and B) highlight 0 shrimp m^{-2} observations while in C) through F) black bars represent missing values. Year-season label colors depict ecological perturbations: red = hypersalinity event, blue = cold snap, green = algal bloom. Labels on the left margin of (A) refer to canal outlets (blue) and coastline features (black) depicted in Fig. 1.

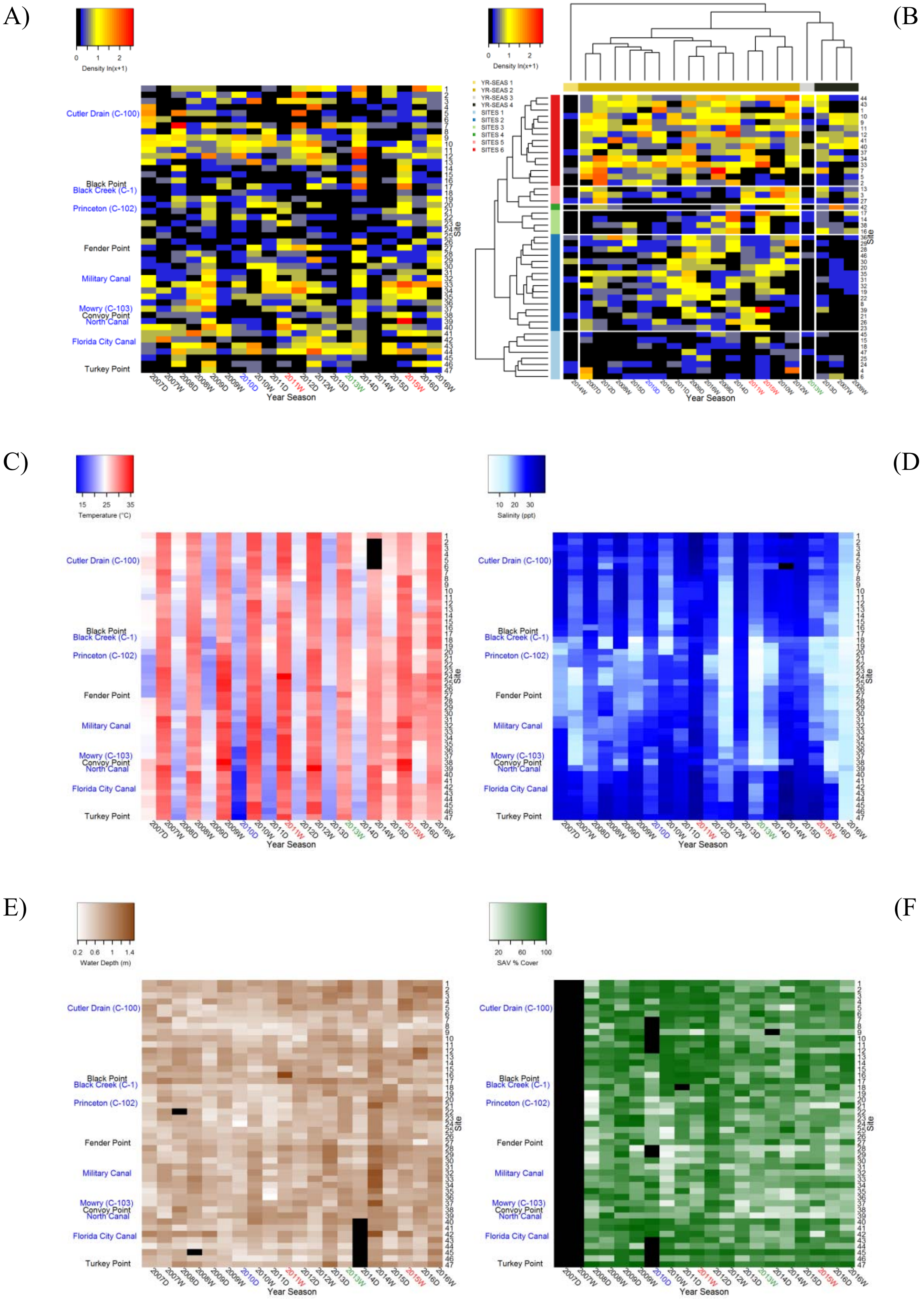

A)
(B
C)
(D
E)
(F
Density ln(x+1)
0 1 2
YR-SEAS 1
YR-SEAS 2
YR-SEAS 3
YR-SEAS 4
SITES 1
SITES 2
SITES 3
SITES 4
SITES 5
SITES 6
Temperature (°C)
15 25 35
Salinity (ppt)
10 20 30
Water Depth (m)
0.2 0.6 1 1.4
SAV % Cover
20 60 100
Cutler Drain (C-100)
Black Point
Black Creek (C-1)
Princeton (C-102)
Fender Point
Military Canal
Mowry (C-103)
Convoy Point
North Canal
Florida City Canal
Turkey Point
Site
Year Season
2007D
2007W
2008D
2008W
2009D
2009W
2010D
2010W
2011D
2011W
2012D
2012W
2013D
2013W
2014D
2014W
2015D
2015W
2016D
2016W

Table 3.4: Results of Procrustean analysis of density (shrimp m^{-2}: LN([x+1]) relative to temperature (°C), salinity (ppt), water depth (m), and SAV (% cover) including goodness-of-fit measure (m^{2}), correlation of the Procrustean rotation (r), and p value of the fit.

	m^2	r	p value
Temperature	0.7287	0.5209	<0.0001
Salinity	0.779	0.4701	<0.0001
Water Depth	0.7169	0.5321	<0.0001
SAV	0.7777	0.4715	0.1162

Table 3.5: Median and ~95% CI of density (shrimp m^{-2}: LN([x+1]) , temperature (°C), salinity (ppt), water depth (m), and submerged aquatic vegetation (SAV: % cover) and the χ^2, d.f., and p values associated with Kruskal-Wallis testing of density clusters relative to A) site and B) year-season. Median CI computed as described in the text.

A)

Site Cluster	n	Density	Temperature	Salinity	Water Depth	SAV
1	160	0.00 ± 0.036	27.73 ± 1.04	21.15 ± 1.29	0.68 ± 0.032	79.00 ± 4.00
2	320	0.29 ± 0.061	28.50 ± 0.71	22.38 ± 0.87	0.72 ± 0.024	56.50 ± 2.97
3	80	0.29 ± 0.090	28.05 ± 1.32	23.79 ± 1.54	0.80 ± 0.039	78.00 ± 5.65
4	20	0.14 ± 0.20	28.70 ± 3.32	29.67 ± 2.24	0.80 ± 0.113	69.75 ± 10.05
5	60	0.29 ± 0.15	28.60 ± 1.58	25.37 ± 1.69	0.76 ± 0.047	77.00 ± 4.13
6	300	0.69 ± 0.074	27.80 ± 0.72	26.57 ± 0.79	0.75 ± 0.025	70.25 ± 2.86
χ^2	NA	148.27	0.64	71.07	21.41	98.24
d.f.	NA	5	5	5	5	5
p value	NA	<0.0001	0.9861	<0.0001	0.0007	<0.0001

B)

Year-Season Cluster	n	Density	Temperature	Salinity	Water Depth	SAV
1	47	0.00 ± 0.00	29.40 ± 0.25	28.12 ± 0.96	1.00 ± 0.041	53.00 ± 5.94
2	705	0.51 ± 0.075	25.30 ± 0.48	24.55 ± 0.59	0.71 ± 0.015	70.00 ± 2.10
3	47	0.00 ± 0.051	29.20 ± 0.28	18.26 ± 2.59	0.83 ± 0.60	59.00 ± 6.51
4	141	0.29 ± 0.18	30.01 ± 1.20	24.58 ± 1.25	0.72 ± 0.40	66.25 ± 5.36
χ^2	NA	89.77	32.28	70.07	85.68	22.03
d.f.	NA	3	3	3	3	3
p value	NA	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001

Chapter 4: Salinity Influences on Juvenile Pink Shrimp (*Farfantepenaeus duorarum*) Growth and Survival with Notes on Potential Disease Interactions

4.1 Summary

Pink shrimp (*Farfantepenaeus duorarum*) has been selected as an indicator of salinity regime alterations by Comprehensive Everglades Restoration Plan implementation. Though previous studies have investigated salinity effects on pink shrimp growth, none have used the mechanistically realistic intermolt period (IMP) and molt increment (MI) growth process approach. Crustacean growth consists of stanzas of no growth (i.e., IMPs) punctuated by ecdysis events and subsequent rapid growth bursts (i.e., MIs). Here, I studied the IMP/MI process as well as survival of juvenile pink shrimp on a salinity gradient (5, 10, 15, 25, 35, 45, and 55 ppt treatment levels). IMP and MI were observed until all shrimps molted twice, yielding at least one IMP and two MI observations each. No statistical relationship between salinity and the MI and IMP was observed. Average daily increase in weight (ADW: g d^{-1}) and average daily increase in length (ADL: mm carapace length d^{-1}) followed parabolic responses relative to salinity with maximal growth at 34 and 33 ppt, respectively. MI significantly decreased with increasing shrimp size. Females exhibited significantly higher MI, ADW, and ADL growth metrics. Logistic curves were fit to survival vs. salinity for both acclimation and IMP/MI study phases, resulting in >80% survival at ≥25 ppt and ≥15 ppt, respectively. Salinity influences on ADW, ADL, and survival generally agreed with previous studies of pink shrimp growth.

4.2 Background

Pink shrimp (*Farfantepenaeus duorarum*) has been selected as an indicator of altered salinity regimes and other ecological conditions altered by Comprehensive Everglades Restoration Plan (CERP) implementation (Browder et al. 2005a, Browder and Robblee 2009). Among its goals, CERP seeks to restore estuarine salinity regimes, while minimizing rapid salinity fluctuations and/or hypersaline conditions associated with altered freshwater deliveries (Barnes et al. 2005, Browder et al. 2005, Davis et al. 2005, Rudnick et al. 2005, Sime 2005, VanArman et al. 2005, RECOVER 2008, Browder and Robblee 2009). Thus, a thorough understanding of salinity effects on pink shrimp growth is of critical interest (Chapter 2).

Crustacean, including penaeid shrimp (Dall et al. 1990), molt-process growth follows a discontinuous, cyclical pattern of intermolt periods (IMPs) punctuated by ecdysis (i.e., exuviation), which is immediately followed by rapid growth bursts (i.e., molt increments: MIs) (Drach 1939, Passano 1960, Scheer 1960, Botsford 1985). Investigation of crustacean growth in terms of a stair-step, discontinuous trajectory of MIs and IMPs most realistically portrays the molt-growth process (Gray and Newcombe 1938, Hiatt 1948). However, researchers often approximate growth as a linearized, continuous process due to difficultly in molt-growth data collection (Dall et al. 1990). Penaeid growth trajectories exhibit rapid postlarval growth that decreases with increasing size (i.e., age), resulting in a sigmoid-shaped curve (Dall et al. 1990). Reduction in growth rate is manifested by increases in IMP and decreases in MI (Dall et al. 1990, Chang et al. 2012). Thus, shrimp size (i.e., age) may be the most important determinant of both IMP and MI (Kurata 1962, Mauchline 1977, Hartnoll 1983, Dall et al. 1990).

However, a number of external environmental factors, sex, and health also influence penaeid growth trajectories (Kurata 1962, Mauchline 1977, Dall et al. 1990, Chang et al. 2012).

Salinity can exhibit considerable influence on penaeid growth (Dall et al. 1990). Most investigations report a significant parabolic relationship between penaeid IMP and salinity (*Penaeus. merguiensis*: Staples and Heales 1991; *P. chinensis*: Chen et al. 1992; *P. esculentus*: O'Brien 1994; *P. monodon*: Ye et al. 2009). One study reported a linear relationship (*P. chinensis*: Chen et al. 1996), likely because the study did not include extreme (<10 and >40 ppt) salinity treatments. Another study reported significant salinity influences on IMP, but did not present a functional relationship (*P. indicus*: Vijayan and Diwan 1995). Study of molting frequency (i.e., average number of molts per group per given time period) has also revealed significant salinity influences (*P. monodon*: Allan and Maguire 1992, *P. chinensis*: Chen et al. 1992). A few studies report no significant salinity influence on IMP (*P. latisulcatus*: Sang and Fotedar 2004) or molt frequency (*P. vannamei*: Pante 1990; *P. chinensis*: Chen et al. 1996). Conflicting parabolic relationships have been reported, with either IMP maximization (Chen et al. 1992, Ye et al. 2009) or minimization (Staples and Heales 1991, O'Brien 1994) at the salinity value yielding maximal growth.

Only three studies have investigated penaeid MI in relation to salinity conditions. Two reported parabolic relationships with maximal MI and growth occurring at intermediate salinity conditions (20 to 35 ppt for *P. merguiensis*: Staples and Heales 1991; 25 to 30 ppt for *P. esculentus*: O'Brien 1994). The third reported no significant salinity effect on MI (*P. latisulcatus*: Sang and Fotedar 2004).

The present study was designed to investigate pink shrimp IMP and MI and other growth metrics as well as survival across a salinity range representative of natural conditions pink shrimp may face in south Florida nearshore areas. The results of this investigation are directly relevant to understanding impacts of CERP implementation on growth and survival of juvenile pink shrimp and could lead to improved growth and survival models for investigating potential effects of CERP on pink shrimp productivity. Observations on sex influences on *F. duorarum* growth and notes on observed disease conditions are also provided.

4.3 Methods

4.3.1 Experimental Design

The study was designed to investigate salinity effects on juvenile pink shrimp molt-process growth represented by IMP duration and post ecdysis MI. Individuals were maintained in isolation at one of seven experimental salinities (5, 10, 15, 25, 35, 45, 55 ppt). Detection of exuvae during morning hours indicated occurrence of ecdysis during the previous scotophase (Eldred 1958, Bishop and Herrnkind 1976, Diaz 2001). When exuvae were observed, shrimp size (MI) and duration between subsequent ecdysis events (IMP) were recorded. Individuals were maintained until at least two consecutive ecdysis events or mortality occurred. Photoperiods were maintained at approximately 12:12 hr light:dark with photophase lasting from ~ 08:00 hr to ~20:00 hr and scotophase from ~20:00 hr to ~08:00 hr of the following day.

4.3.2 Experimental Organisms

Juvenile pink shrimp were obtained from a live bait retailer who reported the capture location as the vicinity of Black Point, Biscayne Bay (Florida, USA). Salinity trials were conducted in the laboratory to determine the effect of salinity on IMP and MI. Individuals of similar size (range: 12.4 to 18.6 mm CL) were selected from bait shop holding tanks and transported to the laboratory (Rosenstiel School of Marine and Atmospheric Science, University of Miami, Miami, FL, USA) in aerated buckets. Upon termination of the IMP/MI study phase, sex was determined by examination of external morphology (Perez-Farfante and Kensley 1997).

Individuals were held in a 209 L glass aquarium filled with filtered seawater (~33.5 ppt) for at least 24 hr prior to experimental use. Sand and shell hash substrate was provided (SuperReef and Caribbean Crushed Coral substrates, CaribSea, Fort Pierce, Florida, USA) at a depth of ~ 3.0 cm. Filtration consisted of filtration floss and biofiltration media. Aeration was provided via airstones. Submersible titanium heating elements were used to control temperature (~28 °C). Shrimps were offered feed at a rate of 15% of their mass each evening. Feed consisted of a juvenile-specific commercial shrimp grow-out diet (40 % protein, 9% lipid: 40-9 V-Pak 2.0 mm pellet, Ziegler Bros., Inc., Gardners, PA, USA). Shrimps were not fed on the day of transfer to salinity acclimation tanks.

4.3.3 Acclimation to Experimental Salinities

Acclimation to experimental salinities (5, 10, 15, 25, 35, 45, and 55 ppt) was achieved by subjecting groups of shrimps maintained in 13.3 L polyethylene tanks to regular 3 hr interval) water exchanges. Replacement water was 5 ppt closer to the target experimental salinity condition. Thus, salinity acclimation proceeded in a stepped pattern of 1.7 ppt hr^{-1} salinity change. Water exchanges consisted of freshly prepared mixtures of deionized (DI) water and Instant Ocean artificial sea salts (Instant Ocean Spectrum Brands, Blacksburg, VA, USA).

Seven acclimation tanks, one for each final salinity treatment, were arranged within a larger fiberglass tank. Submersible titanium heaters were used in conjunction with a freshwater bath within the fiberglass tank to control water temperature (~28 °C). Acclimation tank lids prevented escape. Aeration as previously described was provided to each acclimation tank. During this stage, observed cannibalistic antagonism among shrimps led to offering of small amounts of feed during acclimation.

Immediately before each water exchange, temperature (°C), salinity (ppt), pH, and dissolved oxygen concentration (mg L^{-1}) were measured using an YSI Model 556 or YSI Pro Plus (YSI, Inc., Yellow Springs, OH, USA). Mean temperature (°C), pH, and dissolved oxygen concentration (mg L^{-1}) did not substantially differ among salinity treatments (Table 4.1) or, where differences were observed, their degree of dissimilarity was not considered physiologically meaningful. Therefore, no further statistical analysis of experimental conditions was conducted.

4.3.4 Experimental Procedures and System

The experimental system consisted of three fiberglass water trough, each of which held seven ~4.25 L glass experimental aquaria (Fig. 4.1). Aquaria were bisected across their shorter axis with affixed plexi-glass panels into two experimental chambers. The panels achieved physical separation between adjacent experimental chamber waters although visual detection among test subjects was not hindered. Each aquarium was randomly assigned one salinity treatment (5, 10, 15, 25, 35, 45, or 55 ppt); both chambers of each aquaria received the same salinity treatment. The three water baths drained to a common sump tank in which a submersible pump continuously circulated municipal fresh water to inlet pipes near the head of the baths. Bath drain stand pipes maintained water levels so that aquaria were approximately 1/3 submerged. Two submersible titanium heaters, placed near the head and middle of each bath, as well as one heater in the sump, maintained constant water temperatures (~28 °C). The two experimental chambers per aquaria replicated in the three baths were considered independent primary sampling units, resulting in six (n = 6) replicates per salinity treatment. A total of 42 experimental chambers were used to investigate the 7 salinity treatments.

Within each experimental chamber, benthic substrates covered under-gravel filter plates. Aeration applied to the air lifts of the under-gravel filter plate drew water through the sand substrate, up the air-lifts, and through activated charcoal media pods. Thus, mechanical, chemical, and biological filtration and substrates for burrowing behavior (Fuss 1964) were provided. Test waters of differing salinities were prepared from DI water and artificial sea salts. Glass lids fitted to experimental aquaria prevented escape from the test chambers and limited both evaporation and cross contamination of waters between test chambers.

Daily maintenance of the experimental system included assessment of water quality, siphoning of debris, feeding, mortality and growth assessment, and water salinity adjustments, as needed. Feed was offered each evening at the previously described rate.

A bulb pipette was used to remove debris, uneaten feeds, and feces. Salinity adjustments generally consisted of addition of small amounts (~100 to 250 mL) of DI water to account for evaporation.

Water quality measurements (temperature (°C), salinity (ppt), pH, and dissolved oxygen concentration (mg L^{-1}) were made in each experimental chamber each morning. Total ammonia nitrogen (TAN) concentration was tested once (study day 18). Temperature (°C), pH, and dissolved oxygen concentration (mg L^{-1}) water quality conditions did not substantially differ among salinity treatments (Table 4.2), or, where they did so, the differences were not considered physiologically relevant. Targeted experimental temperature (28 °C) and salinities (5, 10, 15, 25, 35, 45, 55 ppt) were generally achieved; means of observed values were ≤ 0.49 units from target conditions (Table 4.2). Therefore, no further statistical analysis was conducted.

Post-acclimation to salinity treatments, each experimental chamber was stocked with one shrimp acclimated to the appropriate salinity. Immediately prior to stocking, an initial carapace length (CL: mm) and wet weight (WWT: g) of each acclimated shrimp was obtained using digital calipers (Mitutoyo Model CH-6"CSX, Mitutoyo Corp., Kawasaki, Japan) and a digital precision balance (Ohaus Adventurer Pro, Ohaus Corporation, Parsippany, NJ, USA), respectively. Once initial size measurements were taken, acclimated shrimp were haphazardly assigned to appropriate salinity treatment test chambers. More shrimps were stocked in lower target salinity treatments because I expected reduced survival at those salinities (Table 4.2). Exuvae observations were made each morning. MI was assessed the evening following an ecdysis event (~ 12 hr later). Individuals were removed from test chambers with a small dip net, and CL (mm) was measured with a digital caliber and recorded along with the date. Notation of an ecdysis event in the morning may have inflated IMP by ~2-10 hr since ecdysis has previously been observed to occur during late evening/early morning scotophase periods (Eldred 1958, Bishop and Herrnkind 1976). The experiment was carried out until individuals molted at least twice yielding two MI events and one IMP event each, given survival to study termination. However, observation of multiple 3rd and a few 4th molting events allowed for division of MI measurements into 1st, 2nd, 3rd, and 4th MI events and similarly 1st, 2nd, and 3rd IMP events.

4.3.5 Disease Observations

During the course of the study, qualitative observation of shrimp behavior, poor growth/molting performance, and gross pathological signs led to the conjecture that a disease was affecting the experiment. Upon completion of the study, gross pathological observations were recorded for each surviving individual at the end of the experiment. A subjective scale of "disease acuity" ranging from 0 to 3 with 4 outcome levels (e.g., 0, 1, 2, 3 in order of increasing acuity) were applied to the pathological notations.

4.3.6 Statistical Analysis

All statistical analysis was carried out using the R statistical package (The R Foundation, https://www.r-project.org/). Sex ratio was analyzed with a χ^2 test. Linear regressions and logistic generalized linear model (GLM) analyses were used to investigate the significance of salinity condition and other potential predictors of growth and survival, respectively. Where relevant, normality and equality of variance assumptions of residuals were tested via Anderson-Darling (AD) and Breusch-Pagan (BP) Studentized Residual statistics and graphically investigated with quantile plots and residual vs. fitted values plots, respectively (Kutner et al. 2005). Likelihood ratio tests were used to assess successive inclusion of predictors (Kutner et al. 2005). IMP, MI, average daily increase in weight (ADW), and average daily increase in length (ADL) data were analyzed against salinity, salinity^2, and initial CL (mm) or prior CL (mm) as continuous predictors and molting event, sex, and disease acuity as categorical predictors. Acclimation survival was assessed relative to acclimation group and salinity, whereas IMP/MI study phase survival was assessed relative to salinity and disease condition. Parabolic relationships with respect to salinity were anticipated over the range of test salinities (see Chapter 2, Browder et al. 2002). Thus, inclusion of both linear and quadratic terms was generally considered relative to a null model even if linear term inclusion was not initially supported. Prior CL (mm) was defined as the size measured immediately prior to each ecdysis event while initial CL (mm) was defined as the first CL observed during experiment stocking.

4.4 Results

4.4.1 Experimental Organisms

Two groups of test subjects stocked the experimental system (Table 4.3). Group 1 (n = 75) experienced substantial mortalities during the pre-experiment holding period: 9.3% mortality had occurred by 5.5 hr, 22.3% by 16.5 hr, and 37.3% by 24 hr after stocking. A combination of these losses and further losses to mortality during salinity acclimations and post-acclimation experimental chamber stocking resulted in incomplete stocking of the experimental system. A second group of shrimps (Group 2: n = 36) was introduced to make up for these losses. The second group was held for 48 hr before stocking into acclimation tanks and experienced 61.1% mortality in the interim. Forty-six remained in Group 1 and 18 remained in Group 2 at the beginning of the salinity acclimation study phase.

Mean ± SD CL was 15.74 ± 1.29 mm and CL ranged from 12.4 to 18.6 mm inclusive of both acclimation groups at the onset of the study. Mean ± SD WWT was 3.02 ± 0.76 g and ranged from 1.54 to 4.69 g. Although a higher number of females than males was identified at the study termination (Table 4.3), the observed sex ratio was not statistically different from 1:1 ($\chi^2 = 1.32$, df = 1, p = 0.2498).

4.4.2 Survival during Acclimation to Experimental Salinities

Both groups were successfully acclimated to treatment salinity conditions within ~ 15 hr. Although mortality occurred at most salinities, lower salinity treatments (< 25 ppt) exhibited lower survival (Table 4.3, Fig. 4.2). A significant logistic regression was fit to acclimation survival that included a continuous predictor salinity treatment term (Z = 3.026, p = 0.00248). Likelihood-ratio tests confirmed inclusion of the salinity predictor ($\chi^2 = 14.222$, df = 1, p < 0.001). Inclusion of categorical acclimation group and continuous quadratic salinity terms was not supported. Shrimps were not sexed during the acclimation phase and thus the influence of this variable could not be assessed. Residuals distribution plots suggested an adequate model fit with no major outliers observed. Survival was predicted to increase relative to increasing salinity with survival ≥ 80% at ≥ 25 ppt salinity conditions (Fig. 4.2).

4.4.3 Intermolt Period

During the 37 d duration of the IMP/MI study phase, 58 IMP observations were made, 34 of which were 1st IMP events, 22 were 2nd IMP events, and 2 were 3rd IMP events (Table 4.4). Mean ± SD of all IMP observations was 12.26 ± 4.00 d; minimum and maximum values were 4 d (5 ppt) and 23 d (35 ppt). Linear and quadratic salinity predictors were not found to significantly affect IMP. Investigation of prior CL (mm), sex, and disease acuity predictors also did not yield statistical significance. However, IMP event yielded a significant linear regression that accounted for a modest amount of IMP variability ($R^2_a = 0.169$). Mean duration of the 2nd IMP event significantly differed from that of the 1st IMP ($t = 3.682$, $p = 0.0005$) (Fig. 4.3). Residual normality ($AD = 1.576$, $p = 0.5824$) and homogeneity of variance ($BP = 0.817$, $df = 2$, $p = 0.6644$) assumptions were satisfied.

4.4.4 Molt Increment

A total of 94 MI observations was made during the 37 d of the IMP/MI study phase; 37 were 1st MI events, 33 were 2nd MI events, 22 were 3rd MI events, and 2 were 4th MI events (Table 4.5). The mean ± SD of all MIs was 0.65 ± 0.42 mm; minimum and maximum values were -0.19 (55 ppt) and 1.83 mm (35 ppt). Initial model building detected significant linear and quadratic salinity effects on MI. However, other predictors were found to explain higher amounts of MI variability leading to a final model that did not include either salinity term. A significant linear regression of MI included shrimp sex and initial CL predictors (Table 4.6, Fig. 4.4). The homogeneity of variance assumption was not violated ($BP = 3.737$, $df = 2$, $p = 0.1544$), although the normality assumption was (AD = 10.17, p = 0.0256). Likelihood ratio testing supported successive inclusion of the sex (χ^2 = 6.0055, df = 1, p = 0.0143) and prior CL (mm) (χ^2 = 5.3911, df = 1, p = 0.0202) terms. However, explanatory power of the model was low (R^2 = 0.1202, R^2_a = 0.0997). The model predicted a decrease in MI in males relative to females and decreasing MI with increasing prior CL (mm) (Table 4.6, Fig. 4.4).

4.4.5 Average Daily Increase in Weight

Using initial and final WWT observations, a total of 36 ADW estimates were made. Mean ± SD ADW ranged from -0.00833 ± 0.0035 g d^{-1} in 5 ppt salinity to 0.0317 ± 0.0052 g d^{-1} in 45 ppt salinity, and the overall mean ± SD was 0.0209 ± 0.0177 g d^{-1} (Table 4.7). Some weight increments were negative. A significant regression of ADW included both a quadratic salinity term as well as shrimp sex as predictors (Table 4.8, Fig. 4.5A). Likelihood ratios substantiated successive inclusion of linear salinity (χ^2 = 4.25, df = 1, p = 0.03916), quadratic salinity (χ^2 = 11.75, df = 1, p = 0.0006), and sex (χ^2 = 8.22, df = 1, p = 0.004) predictors relative to reduced model versions. Residuals' homogeneity of variance (BP = 3.162, df = 3, p = 0.3674) assumption was not violated, although the normality assumption was (AD = 9.547, p = 0.0327). Inclusion of disease acuity or initial CL (mm) did not yield statistically significant results. The model predicted a maximum ADW at 34 ppt (Table 4.8, Fig. 4.5A) and explained almost half of the observed variability (R^2_a = 0.4289).

4.4.6 Average Daily Increase of Length

A total of 57 measurements of ADL (mm CL d^{-1}) were recorded. Mean ± SD ADL ranged from 0.0073 ± 0.0161 in 5 ppt to 0.0711 ± 0.0289 mm d^{-1} in the 45 ppt salinity treatment and the overall mean ± SD was 0.0533 ± 0.0366 (Table 4.7). A significant regression of ADL included both a quadratic salinity and sex predictors (Table 4.8, Fig. 4.5B). Likelihood ratio tests confirmed inclusion of quadratic salinity (χ^2 = 15.444, df = 2, p = 0.0004) although initially the initial linear salinity term alone was not supported (χ^2 = 1.298, df = 2, p = 0.2545). Shrimp sex was also supported as a predictor (χ^2 = 4.7016, df = 1, p = 0.0301). Inclusion of disease acuity did not reveal statistically significant influences. Homogeneity of variances (BP = 3.314, df = 3, p = 0.3456) and normality of residuals (AD = 8.484, p = 0.0864) were confirmed. The model predicted a maximum ADL at 33 ppt (Table 4.8, Fig. 4.5B) and explained a moderate amount of the observed variability (R^2_a = 0.2343).

4.4.7 Survival during IMP/MI Experiment

Survival during the IMP/MI portion of study tended to be lower at lower salinity treatments (i.e., 5 and 10 ppt: Table 4.3, Fig. 4.6). A significant logistic GLM of percent mortality included a continuous predictor salinity treatment term (Z = 1.992, p = 0.0464). Likelihood-ratio tests confirmed the significance of this model relative to the intercept-only model (χ^2 = 12.039, df = 1, p < 0.0005). Predicted survival from the model reflected a rapid increase in survival with increasing salinity condition: predicted survival was ~80% at 15 ppt and was ~100% for 25 through 55 ppt salinity treatments (Fig. 4.6). Inclusion of a categorical predictor of relative disease condition was not significant.

4.4.8 Observations of Suspected Disease Conditions

At study termination, a relative disease acuity assessment revealed all shrimps exhibited some signs of gross pathological disease (Table 4.9). Approximately midway through the study, white spots reminiscent of White Spot Syndrome Virus (Fig. 4.7) were noted on test subjects. Other gross pathological signs included a pale yellowish hue and flaccid tissues. A black gill condition was also observed during the course of this study (Table 4.9, Fig. 4.8). A few shrimps (n = 5) exhibited a pale, opaque “milk-like” white appearance to their flesh (Table 4.9). Behavioral indications of poor health included poor appetite and general lethargy or lack of escape response. No effort was made to determine the etiologic agents responsible for these signs. Inclusion of a disease acuity predictor in growth analyses did not yield statistically significant results. After completion of the experiment, all equipment was disinfected with hypochlorite to prevent further spread of any potential diseases present.

4.5 Discussion

The primary objective of this investigation was to examine the effect of a salinity on pink shrimp IMP and MI. However, study results did not clearly elucidate salinity effects on either IMP or MI. Given previous observations of a parabolic response of MI to a salinity gradient in other penaeid shrimps (*P. merguiensis*: Staples and Heales 1991, *P. esculentus*: O’Brien 1994) as well as a salinity influence on IMP (Staples and Heales 1991, Chen et al. 1992, O’Brien 1994, Vijayan and Diwan 1995, Chen et al. 1996, Ye et al. 2009), the lack of response of MI and IMP to salinity in the present study was unexpected. Exploratory analysis of MI did detect a significant parabolic influence of salinity. However, model selection procedures opted for a formulation that included prior CL (mm) and sex, and further likelihood ratio testing did not support adding either linear or quadratic salinity terms. Similar exploratory analysis of IMP did not reveal any salinity influence. Previous studies have reported a minor salinity influence on IMP in other penaeids (Dall et al. 1990, Staples and Heales 1991, O'Brien 1994). Perhaps the present lack of a statistically significant response of either IMP or MI to salinity was due to low sample size. Disease presence in the test animals may also have affected these outcomes.

Finding significant prior CL (mm) and sex effects on MI was not surprising. MI generally changes with shrimp size (Dall et al. 1990). Diaz (2001) observed increasing MI with increasing size for juvenile pink shrimp< 17.6 and 17.2 mm CL, females and males, respectively, and decreasing MI with increasing size for subadults. MI only decreased with increasing size in the present study (Fig. 4.4); no change in trajectory was apparent, counter to Diaz's (2001) observations. Perhaps the lack of inflection in the present study was due to use of a smaller size range (12.41 to 20.16 mm CL) that included sizes intermediate to the range of sizes used by Diaz (2001: 7.6 to 28.0 mm CL). Diaz (2001) noted that, for a given CL, MI appeared larger for female pink shrimp. My results confirmed this observation as females exhibited molt increments ~0.24 mm CL greater than males (Table 4.6, Fig. 4.4).

The MI range observed here (-0.19 to 1.83 mm CL) **w**as broader than has been previously reported for similarly sized pink shrimp (0.1 to 1.2 mm CL: Diaz (2001). This may be related to MI measurement error due to flexibility in the carapace and could possibly explain observed negative MI values. The ~12 hr duration between measurements that I used may not have been sufficient to ensure exoskeleton hardening and accurate measurement. Eldred et al. (1961) attributed observations of negative pink shrimp growth over multiple molting events to small culture vessel size. However, maximal growth rates observed at intermediate salinity conditions agreed with previously reported rates, suggesting limitation by vessel size was not presently a concern or that it affected this and previous studies similarly. Since instances of negative MI were observed at both extremes of the salinity distribution as well as at one intermediate salinity (one each in 5, 15, and 55 ppt), one should be cautious in interpreting their presence in relation to salinity conditions.

The only significant predictor of IMP was the IMP event. Sex was not a significant factor explaining IMP, in agreement with Diaz (2001) for pink shrimp and Hansford and Hewitt (1994) for *P. monodon.* The overall mean IMP (12.3 ± 4.0 d) and range (4 d to 23 d) observed in the present study were of similar magnitude to pink shrimp values reported elsewhere (13 d for 31 mm CL: Eldred 1958, 14 d for 11.8 mm to 15.8 mm CL: Bishop and Herrnkind 1976, ~ 6 d to ~32 d for 7.6 mm to 28.0 mm CL: Diaz 2001, 5 d to 18 d: Bonilla-Gomez et al. 2013).

IMP may have changed with successive events because of the stress associated with capture, holding, and acclimation of wild-caught bait shrimp to study conditions. Bonilla-Gomez et al. (2013); who studied growth, molt frequency, and physiological differences between wild and F2 cultured subadult pink shrimp, discussed how wild shrimp did not “perform” as well immediately after introduction to laboratory conditions. However, they later mention that final growth conditions between wild and cultured shrimp were similar (Bonilla-Gomez et al. 2013). Their longer study duration (55 d) and observed shorter IMP (wild: 10.1 d; cultured: 11.39 d) (Bonilla-Gomez et al. 2013) allowed for more IMP observations compared to the present study (duration: 37 d, mean IMP: 12.26 d).

Although the present study failed to detect a salinity effect on pink shrimp MI or IMP, parabolic responses to salinity were observed for both ADW and ADL (Fig. 4.5). These results agreed with previously reported parabolic-shaped responses of growth to salinity (Browder et al. 2002) and low growth at hyperhaline conditions relative to mesohaline, polyhaline, and euhaline conditions (Browder et al. 2002, Zink et al. 2013). ADL rates concurred with values reported from previous studies (Fig. 2.7: Chapter 2, Zink et al. 2017). Similarly, the maximum observed ADL (0.175 mm d^{-1} at 25 ppt), agreed with previously observed maximal growth rates relative to salinity condition (Fig. 2.7: Chapter 2, Zink et al. 2017). Maximal values of ADW (0.0257 - 0.0317 g d^{-1} in 15 – 45 ppt salinities) concurred with that previously reported for wild shrimps in captivity (0.024 g d^{-1} in 22 ppt: Bonilla-Gomez et al. 2013). Predicted maximal ADW and ADL occurred at 34 and 33 ppt, respectively. These values were similar to a maximum growth rate at 30 ppt as reported by Browder et al. (2002). A significant sex difference in ADW and ADL was also evident (Table 4.8, Fig. 4.5). Along with the MI sexual dimorphism, these observations agreed with previously reported sex differences in pink shrimp growth (Eldred et al. 1961, Iversen and Jones 1961, Kutkuhn 1965, Berry 1967, McCoy 1972, Cohen and Fishman 1980, Diaz 2001, Diaz et al. 2001) and other penaeids (Dall et al. 1990).

High survival at moderate salinities (i.e., 15 – 40 ppt) and low survival at extreme, low salinities (i.e., ≤10 ppt) was consistent with previous studies (Browder et al. 1999, 2002; Criales et al. 2011; Zink et al. 2013). However, high survival at hyperhaline conditions was contradictory to previous studies (Browder et al. 1999, 2002; Criales et al. 2011; Zink et al. 2013). Consequently, survival did not respond to the salinity gradient in the parabolic fashion previously reported (Browder et al. 1999, 2002; Criales et al. 2011) and that was supported by meta-analysis (Fig. 4.8: Chapter 2, Zink et al. 2017). Perhaps the larger shrimp size of the present study relative to those of previous studies (3.2-14.5 mm CL: Browder et al. 2002, 2.3 - ~9.6 mm CL: Zink et al. 2013) revealed a previously unexplored hypersalinity tolerance at larger sizes. Ontogenetic shifts in salinity tolerance range of pink shrimp were reported by Criales et al. (2011), although the phenomenon has not been studied across all relevant pink shrimp sizes. Another explanation could be that an interaction with disease conditions and salinity altered survival rates.

Other than the study target salinity parameter, water quality should not have negatively influenced growth or survival. Although decreasing dissolved oxygen concentration with increasing salinity was not unexpected (Millero 2002); the lowest observed dissolved oxygen concentrations were greater than reported levels inhibiting penaeid growth (Seidman and Lawrence 1985, Rosas et al. 1998, Wei et al. 2009) or molting (Clark 1986). Similarly, pH conditions were also acceptable for penaeid culture (Allan and Maguire 1992). Low TAN concentrations were well within long term penaeid exposure limits (Allan et al. 1990, Chen et al. 1990, Chen and Kou 1992), which suggested effective experimental chamber biological filtration.

The present results should be considered with reservation. Observation of gross pathological disease signs cause one to question the validity of the study results, especially those counter to previously reported results or my own observations. For example, instances of negative MI values could have been caused by the shrimps' diseased state. In retrospect, high mortality during introduction of test subjects to the laboratory and salinity acclimation did not agree with my previous experience with pink shrimp transport and rearing. These mortalities at the study onset could have selected for hardier shrimps, thus affecting subsequent results such as high survival in hypersaline conditions. Although it was reassuring that the disease acuity factor was eliminated during model selection procedures, this may also reflect a lack "control" (i.e., non-diseased) shrimps to compare with. But ADL and ADW growth rates were comparable to, or even higher than, previously reported values (Browder et al. 2002, Bonilla-Gomez et al. 2013, Zink et al. 2013). Why would disease conditions affect IMP and MI but not ADL or ADW?

Disease signs included "white spots" (Fig. 4.7), possibly related to White Spot Syndrome Virus, exhibited by all shrimps; "black gill disease" (Fig. 4.8) by one individual; and possible "cotton shrimp" microsporidian infections (Table 4.9). Without further investigation, present assessment of suspected diseases amount to conjecture based upon gross pathology, incidence rates, and logical routes of transmission.

In summary, the present study was unable to detect salinity influences on pink shrimp IMP and MI. Sex and shrimp size effects were observed for MI, and IMP duration significantly differed between the 1st and 2nd IMP events. In contrast, trends in ADW and ADL were each parabolic with respect to salinity condition, and growth in these terms was comparable to that reported in other studies. Sexual dimorphism affected measures of growth.

Survival under lower salinity conditions generally agreed with previous studies, but high survival at hypersaline conditions was unexpected. Future study of the pink shrimp molt-growth process should include larger sample sizes to ensure detection of trends in IMP and MI. Providing a longer duration between observation of ecdysis and measurement of post ecdysis size would allow the new skeleton to harden and reduce measurement error and its influence on molt increment estimation. Extending this study to a greater size range of shrimps would improve understanding of ontogenetic shifts in pink shrimp salinity tolerance.

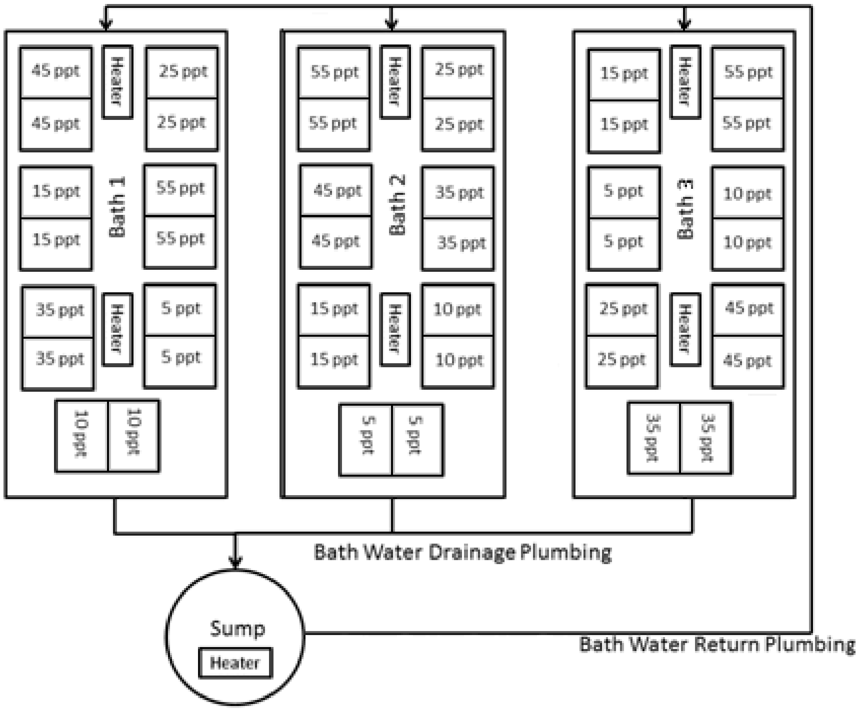


Fig. 4.1: Diagram of the IMP/MI experimental system depicting bath tanks, experimental aquaria and their chambers, sump, water heaters, and plumbing connections. Experimental chamber salinity labels reflect random assignment of salinity treatments to experimental aquaria.

Table 4.1: Sample size and mean ± SE of temperature (°C), pH, and dissolved oxygen (D.O.) concentration (mg L^{-1}) observations per salinity treatment and acclimation group.

Acclimation Group	Treatment Salinity	Sample Size	Water Quality Parameter		
			Temperature (°C)	pH	D.O. (mg L^{-1})
			$\bar{X}$ ± SE	$\bar{X}$ ± SE	$\bar{X}$ ± SE
Group 1	5	6	26.56 ± 0.19	7.87 ± 0.08	5.85 ± 0.13
	10	6	26.62 ± 0.11	7.94 ± 0.07	6.21 ± 0.09
	15	6	26.78 ± 0.21	8.00 ± 0.05	5.82 ± 0.12
	25	6	26.72 ± 0.19	8.01 ± 0.05	6.16 ± 0.09
	35	5	26.74 ± 0.29	8.00 ± 0.06	5.72 ± 0.13
	45	6	26.74 ± 0.20	7.99 ± 0.06	6.13 ± 0.10
	55	6	26.54 ± 0.20	8.02 ± 0.02	5.61 ± 0.09
Group 2	5	6	26.19 ± 0.19	8.01 ± 0.05	5.94 ± 0.09
	10	6	26.22 ± 0.13	7.96 ± 0.06	5.39 ± 0.09
	15	6	26.22 ± 0.18	8.01 ± 0.03	5.59 ± 0.07
	25	6	26.28 ± 0.24	7.91 ± 0.07	5.21 ± 0.11
	35	6	26.41 ± 0.27	7.99 ± 0.03	5.40 ± 0.20
	45	6	26.62 ± 0.19	7.90 ± 0.07	5.20 ± 0.13
	55	6	26.62 ± 0.23	8.03 ± 0.03	5.30 ± 0.25

Table 4.2: Sample size and mean ± SE of temperature (°C), salinity (ppt), pH, dissolved oxygen (D.O. mg L^{-1}), and total ammonia nitrogen (TAN: mg L^{-1}) observations during IMP/MI study phase. Witihin a column (i.e., within each water quality parameter), superscript letters denote statistically similar groupings as detected by post hoc means comparisons.

Treatment Salinity (ppt)	Temperature (°C)		Salinity (ppt)		pH		D. O. (mg L^{-1})		TAN (mg L^{-1})	
	n	$\bar{X}$ ± SE	n	$\bar{X}$ ± SE	n	$\bar{X}$ ± SE	n	$\bar{X}$ ± SE	n	$\bar{X}$ ± SE
5	172	27.51 ± 0.03	172	5.10 ± 0.01	166	8.25 ± 0.01	172	6.96 ± 0.07	6	0.04 ± 0.01
10	172	27.59 ± 0.04	172	10.09 ± 0.01	166	8.16 ± 0.01	172	6.81 ± 0.07	6	0.03 ± 0.01
15	184	27.65 ± 0.04	184	15.14 ± 0.02	180	8.09 ± 0.01	184	6.66 ± 0.06	6	0.02 ± 0.01
25	186	27.72 ± 0.03	186	24.99 ± 0.02	182	7.97 ± 0.01	186	6.29 ± 0.06	6	0.02 ± 0.01
35	186	27.60 ± 0.03	184	34.95 ± 0.03	182	7.85 ± 0.01	186	5.96 ± 0.07	6	0.00 ± 0.00
45	188	27.57 ± 0.03	186	45.12 ± 0.05	184	7.83 ± 0.01	188	5.63 ± 0.06	6	0.01 ± 0.00
55	186	27.70 ± 0.04	186	54.77 ± 0.06	182	7.79 ± 0.01	186	5.26 ± 0.06	6	0.01 ± 0.00

Table 4.3: Numbers of shrimp stocked and their survival in each salinity treatment during salinity acclimation and IMP/MI study phases. Sex, assessed at the termination of the IMP/MI study phase, is also included.

Salinity Treatment	Acclimation to Salinity Treatments				IMP/MI Study				
	Number Stocked		Survival		Number Stocked	Survival		Sex*	
	Group 1	Group 2	Group 1	Group 2		n	Percent	Male	Female
5	8	5	37.5%	40.0%	5	3	60%	1	3
10	7	3	57.1%	66.7%	6	3	50%	2	3
15	7	4	28.6%	100.0%	6	5	83%	2	3
25	6	1	83.3%	100.0%	6	6	100%	3	3
35	6	1	100.0%	100.0%	6	6	100%	2	3
45	6	2	83.3%	100.0%	6	6	100%	3	3
55	6	2	100.0%	100.0%	6	6	100%	2	4
Total	46	18	67.4%	81.8%	41	35	85%	15	22

* Sex was assessed only on those shrimps surviving until study termination

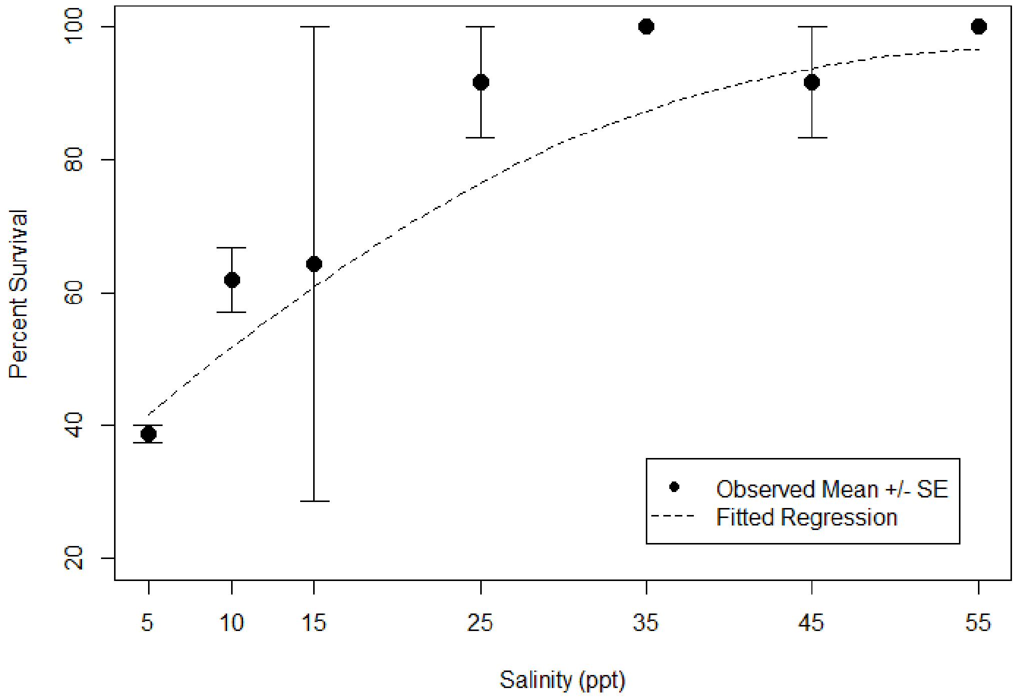


Figure 4.2: Acclimation study phase mean percent survival ± SE and fitted logistic survival regression plotted against treatment salinties (ppt).

Table 4.4: Sample size and mean ± SE by salinity treatment for each IMP event. Superscript letters in Overall row denote statistically similar groupings.

Salinity Treatment	1st IMP Event		2nd IMP Event		3rd IMP Event		Overall	
	n	$\bar{X}$ ± SE	n	$\bar{X}$ ± SE	n	$\bar{X}$ ± SE	n	$\bar{X}$ ± SE
5	3	10.7 ± 4.8	2	13.5 ± 5.5	0	-	5	11.8 ± 3.2
10	4	10.8 ± 1.1	2	14.5 ± 2.5	0	-	6	12.0 ± 1.2
15	5	13.4 ± 2.0	2	15.0 ± 1.0	0	-	7	13.9 ± 1.5
25	5	10.0 ± 1.4	5	15.2 ± 1.9	2	12 ± 0.0	10	12.5 ± 1.2
35	5	9.0 ± 0.4	3	16.0 ± 3.5	0	-	8	11.6 ± 1.7
45	6	11.3 ± 1.7	6	13.8 ± 1.4	0	-	12	12.6 ± 1.1
55	6	10.5 ± 1.7	2	13.0 ± 0.0	0	-	8	11.1 ± 0.6
Overall	34	10.8 ± 0.6^{A}	22	14.5 ± 0.8^{B}	2	$12 \pm 0.0^{A,B}$	58	12.3 ± 0.5

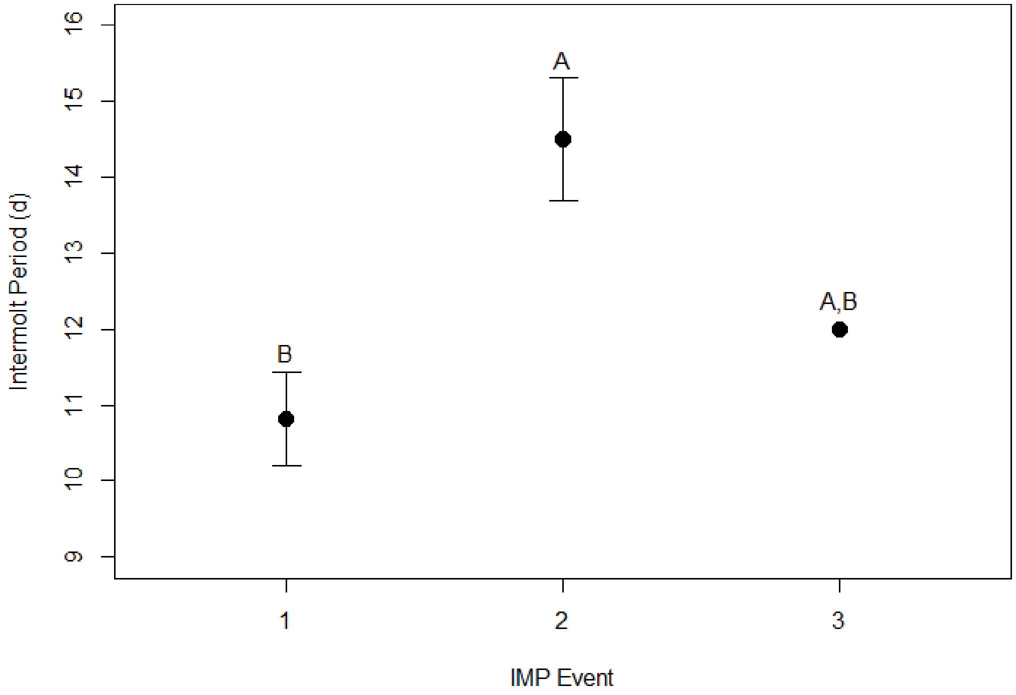


Figure 4.3: Mean ± SE of IMP (d) for each molting event. Letters denote statistical similarities.

Table 4.5: Sample size and mean ± SE molt increment (MI) by salinity treatment and MI event.

Salinity Treatment	1st MI Event		2nd MI Event		3rd MI Event		4th MI Event		Overall	
	n	$\bar{X}$ ± SE	n	$\bar{X}$ ± SE	n	$\bar{X}$ ± SE	n	$\bar{X}$ ± SE	n	$\bar{X}$ ± SE
5	2	0.59 ± 0.19	2	0.21 ± 0.17	2	0.09 ± 0.22	0	-	6	0.30 ± 0.13
10	6	0.55 ± 0.10	4	0.52 ± 0.11	2	0.37 ± 0.17	0	-	12	0.51 ± 0.07
15	6	0.78 ± 0.24	5	0.65 ± 0.24	2	0.70 ± 0.25	0	-	13	0.72 ± 0.14
25	5	0.39 ± 0.11	5	0.76 ± 0.17	5	0.64 ± 0.09	2	0.79 ± 0.15	17	0.62 ± 0.07
35	6	0.73 ± 0.06	5	0.50 ± 0.09	3	1.22 ± 0.54	0	-	14	0.75 ± 0.13
45	6	0.67 ± 0.19	6	0.84 ± 0.22	6	0.88 ± 0.12	0	-	18	0.80 ± 0.10
55	6	0.95 ± 0.21	6	0.23 ± 0.11	2	0.84 ± 0.84	0	-	14	0.62 ± 0.14
Overall	37	0.68 ± 0.07	33	0.56 ± 0.07	22	0.73 ± 0.10	2	0.79 ± 0.15	94	0.65 ± 0.04

Table 4.6: Coefficient estimates, degrees of freedom (df), and relevant statistics from molt increment ($R^2_a = 0.0997$) regression.

Source of Error	Coefficient Estimate ± SE	df	F value	p value
Intercept	1.869 ± 0.4933	-	14.349	0.0003
Sex (= Male)	-0.2397 ± 0.0854	1	7.885	0.0062
Prior CL (mm)	-0.0723 ± 0.0312	1	5.368	0.0229
Residuals	-	86	-	-
Model	-	2	5.874	0.0041

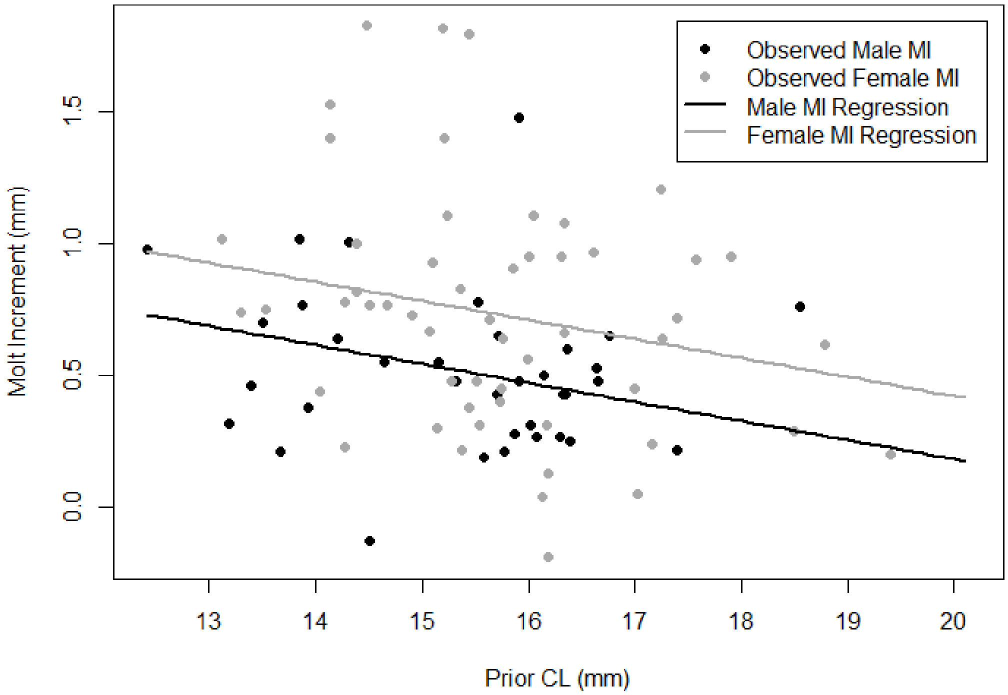


Figure 4.4: Molt increment (MI: mm) plotted against prior CL (mm). Significant prediction of MI ($R^2_a = 0.0997$) by both sex and prior CL (mm) depicted by regression lines.

Table 4.7: Sample size and mean ± SE of average daily increase in weight ($g\ d^{-1}$) and average daily increase in length ($mm\ d^{-1}$).

Salinity Treatment (ppt)	Weight		Length	
	n	$\bar{X} \pm SE$	n	$\bar{X} \pm SE$
5	4	-0.00833 ± 0.0018	4	0.0073 ± 0.0081
10	5	0.0163 ± 0.0045	6	0.0411 ± 0.0080
15	5	0.0257 ± 0.0044	7	0.0494 ± 0.0107
25	6	0.027 ± 0.0069	12	0.0667 ± 0.0127
35	5	0.0278 ± 0.0111	8	0.0641 ± 0.0140
45	6	0.0317 ± 0.0052	12	0.0711 ± 0.0083
55	5	0.0172 ± 0.0053	8	0.0316 ± 0.0103
Overall	36	0.0209 ± 0.0177	57	0.0533 ± 0.0366

Table 4.8: Coefficient estimates, degrees of freedom (df), and relevant statistics from average daily incrase in wieght ($R^2_a = 0.4289$) and length ($R^2_a = 0.2343$) regressions.

Average Daily Increase	Predictor	Coefficient Estimate ± SE	df	F value	p value
Weight	Intercept	-6.576e-3 ± 6.684e-3	1	0.968	0.3328
	Salinity	2.496e-3 ± 5.530e-4	1	20.367	<0.0001
	Salinity^2	-3.591e-5 ± 8.985e-6	1	15.976	0.0004
	Sex (=Male)	-1.190e-2 ± 4.153e-3	1	8.208	0.0074
	Residuals	-	31	-	-
	Model	-	3	9.513	0.0001
Length	Intercept	-1.555e-4 ± 1.568e-2	1	0.0001	0.9921
	Salinity	4.750e-3 ± 1.184e-3	1	16.088	0.0002
	Salinity^2	-7.230e-5 ± 1.877e-5	1	14.838	0.0003
	Sex (=Male)	-1.824e-2 ± 8.5513-3	1	4.550	0.0377
	Residuals	-	51	-	-
	Model	-	3	6.509	0.0008

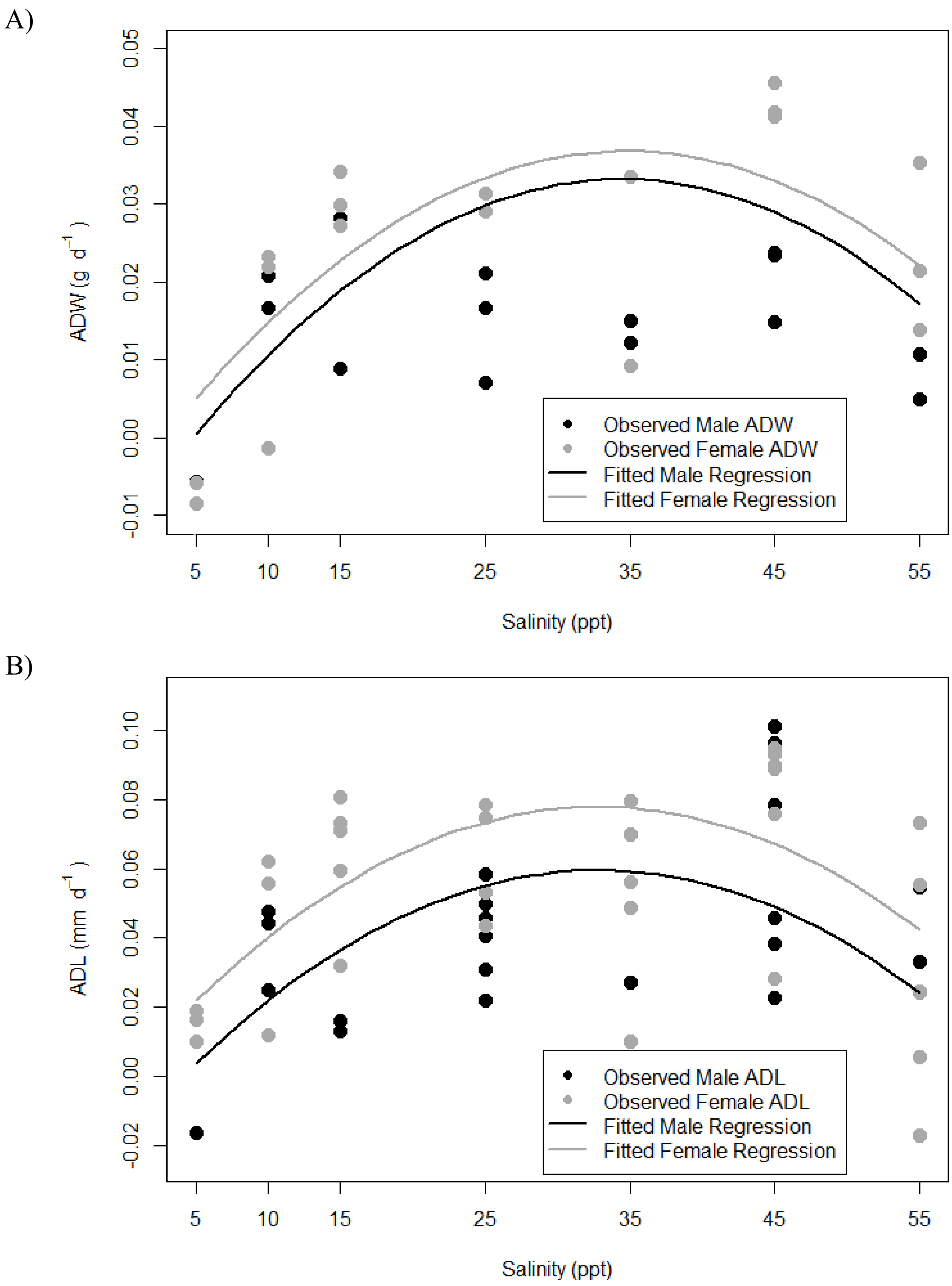


Figure 4.5: Plot of A) average daily in weight (ADW: g d^{-1}) and B) average daily increase in length (ADL: mm CL d^{-1}) plotted against treatment salinities (ppt). Significant prediction ADW and ADL by both sex and salinity condition (ppt) depicted by regression lines.

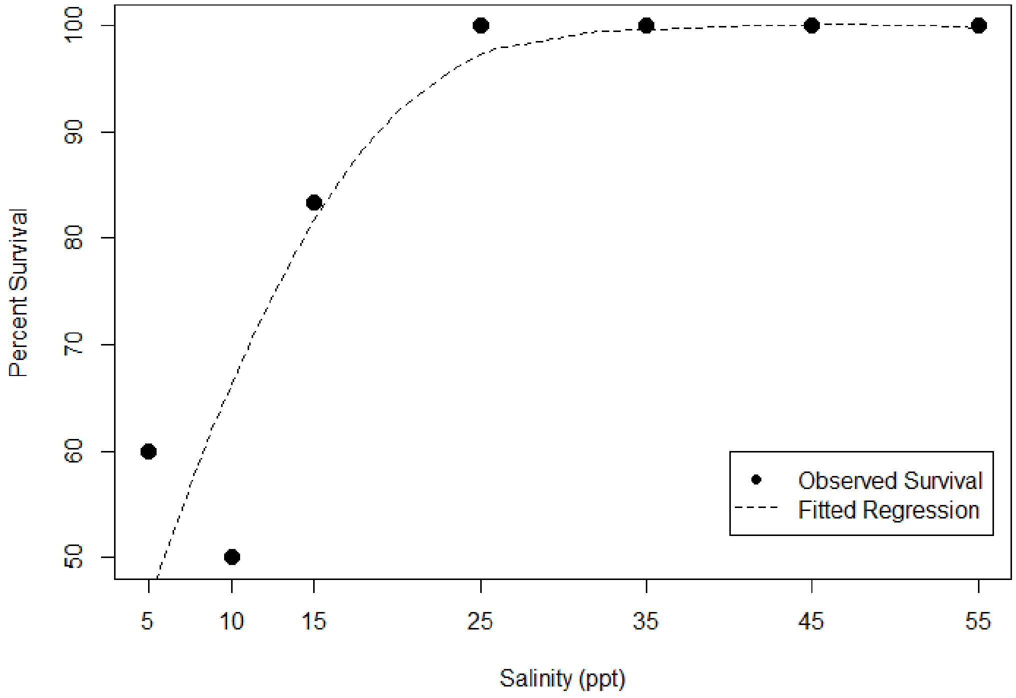


Figure 4.6: Percent suvival during IMP/MI study phase and fitted logistic regression plotted against salinity condition (ppt).

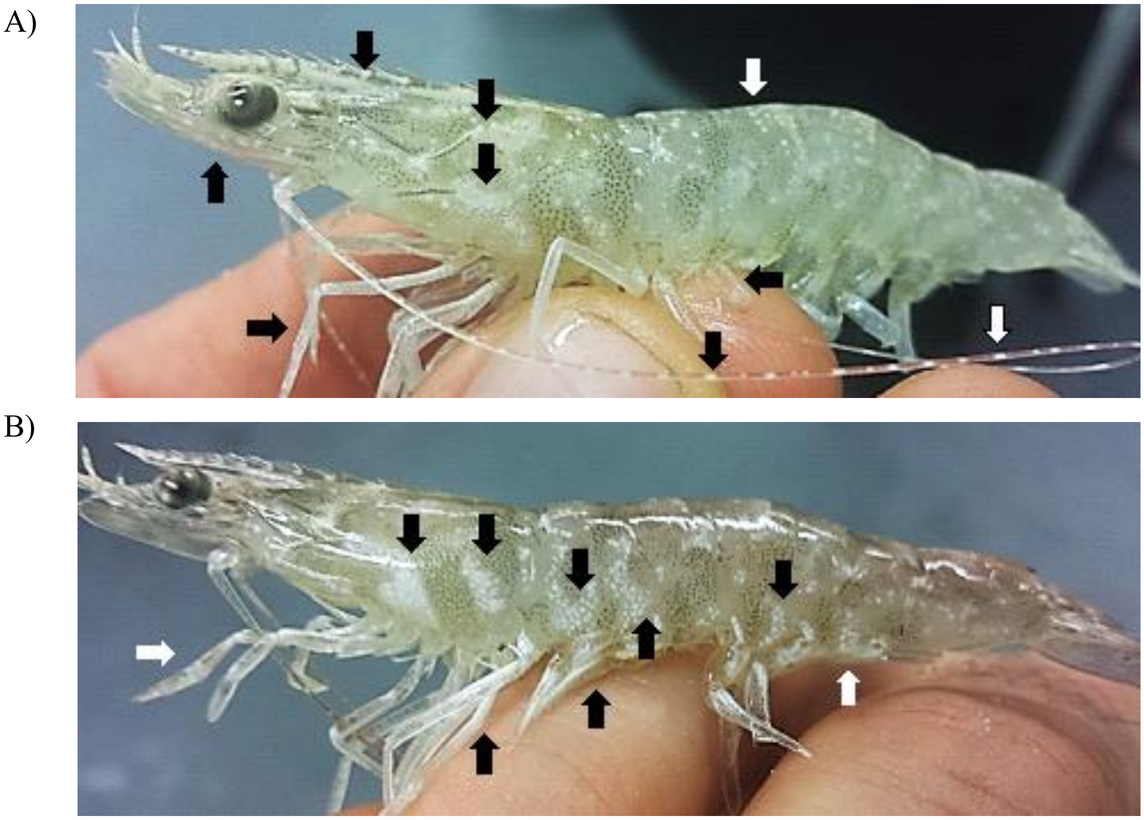


Figure 4.7: Photos of A) heavy (acuity scale = 3) and B) moderate (acuity scale = 2) white spot infestations of experimental subjects taken at the conclusion of the study (37 d). Occurrence of white spots gross pathological signs (arrows) can be observed on virtually every body surface, including rostrum, scaphocerite, antenna, carapace, pereiopods, pleopods, abdominal segments, and uropods as highlighted by arrows. “Heavier” infestations demonstrated dense clusters of white spots which made localized areas appear nearly solid white (photos digitally sharpened and contrast increased to aid appearance).

Table 4.9: Acuity of white spots scores assigned to individual test subjects and other potential disease observations.

Bath	Tank	Treatment Salinity (ppt)	Acuity of White Spots	
			Replicate 1	Replicate 2
1	1	45	2	1[‡]
1	2	25	3	1
1	3	15	2	2
1	4	55	3	3[‡]
1	5	35	2	3
1	6	5	2	N/A[†]
1	7	10	N/A[†]	N/A[†]
2	1	55	1	N/A[†]
2	2	25	1	3
2	3	45	1[‡]	3
2	4	35	1[‡]	3*
2	5	15	2	2
2	6	10	2	1
2	7	5	3	2
3	1	15	1	N/A[†]
3	2	55	2	1[‡]
3	3	5	N/A[†]	N/A[†]
3	4	10	3	2
3	5	25	3	2
3	6	45	3	2
3	7	35	1	1

*Individual also displayed apparent "black gill disease"

[‡] Individual also displayed apparent "milky flesh" condition

[†] Acuity was not assessed as individual died before study termination

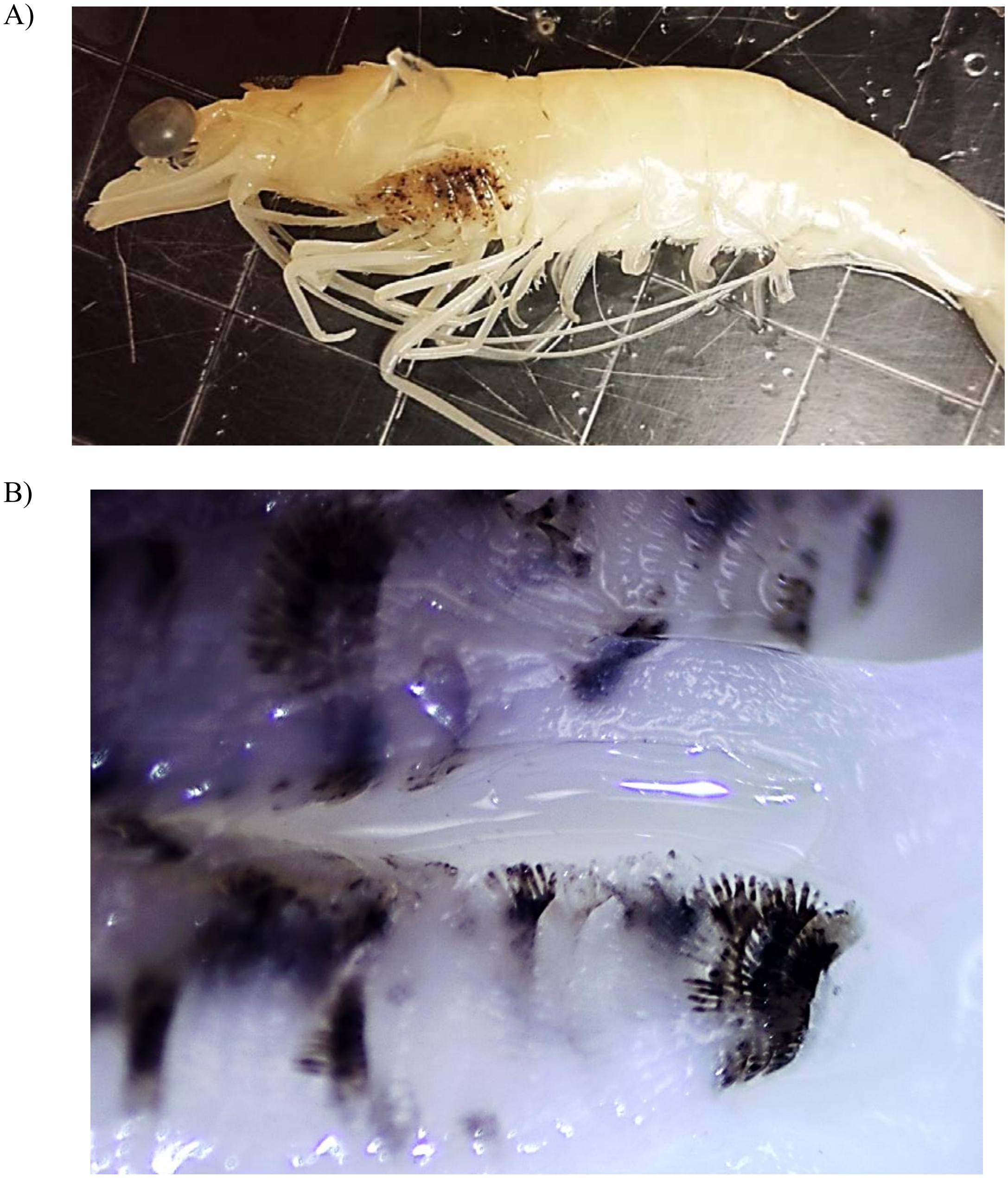


Figure 4.8: Photos A) whole body and B) magnified view of gill lamellae depicting discoloration of black gill condition (photos digitally sharpened and contrast increased to aid appearance).

Chapter 5: The Potential Impact of Doubling Biscayne Bay Freshwater Canal Discharges on Nearshore Juvenile Pink Shrimp *Farfantepenaeus duorarum* Habitat Suitability

5.1 Summary

Habitat suitability indices (HSIs) have proven useful to environmental managers involved with the Comprehensive Everglades Restoration Plan (CERP) for gaining insight into the potential impacts of salinity modifications and other habitat changes on nearshore biota. Here, I developed a juvenile pink shrimp, *Farfantepenaeus duorarum*, HSI for southwestern Biscayne Bay, FL, nearshore waters (0-50 m) to explore the potential effects of doubling the volume of freshwater discharges through the existing canal system. While other habitat attributes were initially considered, the developed HSI model focused on a significant 3-knot natural cubic spline function 0.9 quantile regression relationship between salinity and juvenile pink shrimp density. This regression yielded maximal potential pink shrimp density at 29.25 ppt. This density-salinity relationship was partitioned into five habitat suitability levels designated as optimal, good, fair, poor, and critical suitability. Using the Biscayne Bay Simulation Model (v4), wet (May-Oct.) and dry (Nov.-April) seasonally averaged 35 d antecedent mean salinities were calculated for “Base Flow” and “High Flow” (a two-fold increase in canal discharge volume) scenarios over 15 Water-years (WYR: 1997-2011). Tabulation of these HSI predictions at the 47 sampling sites for each WYR and season (N = 705 estimates per season) revealed High Flow scenario reductions in good and optimal habitat suitability during the wet (High Flow = 64.6%, Base Flow = 84.4%) and dry seasons (High Flow = 74.4%, Base Flow = 90.7%).

To quantify areal changes in habitat suitability, seasonally averaged 35 d antecedent mean salinities were simulated every 50 m along~22 km of the shoreline; considering a 50-m wide buffer, habitat suitability was classified for 158.75 ha of habitat area. In 2004 WYR, a wet season marked by substantially higher freshwater discharge, High Flow and Base Flow percentage of area classified as good and optimal habitat suitability were similar (67.2% and 60.6%, respectively); however, areal extent of High Flow critical suitability (23.8%) was just over 3x greater than that of the Base Flow scenario (7.7%). Average WYR wet season areal extent of good and optimal suitability was 91.8% in the Base Flow scenario; no areas within the study domain were classified as critical. In the High Flow scenario, good and optimal area was reduced to 74.3%, and 6.5% of the area was classified as critical. These results indicate creation of mesohaline and oligohaline salinity conditions in southwestern Biscayne Bay nearshore areas (0-50 m) by doubling freshwater discharges would negatively impact pink shrimp habitat suitability within the simulated study domain.

5.2 Background

Freshwater management has altered salinity regimes along Biscayne Bay's western shoreline due to the operation of a vast system of canals, locks, levees, and pumping stations. This system has resulted in flood control, surface water retention, and lowering of the water table to allow agricultural, urban, and suburban development of southeast Florida. Over the course of the 20th century, salinity gradients perpendicular to the western shoreline were greatly compressed or eliminated due to alteration of the timing, distribution, and volume of surface and groundwater discharges (Kohout 1967, Kohout and Kolipinski 1967, Lindall 1973, Parker 1974, Meeder 2001, Browder et al. 2005, Renken et al. 2005). Today, western Biscayne Bay nearshore salinities are generally polyhaline (18-30 ppt) and euhaline (30-40ppt) (Caccia and Boyer 2005). These conditions are punctuated by rapid salinity reductions due to surface freshwater discharges from point sources (i.e., canal mouths: Caccia and Boyer 2005, 2007), where salinities may rapidly decrease 15 ppt or more in only a few hours (Wang and Cofer-Shabica 1988, Lirman et al. 2003). Western Biscayne Bay nearshore wet season salinities are generally lower than those of the dry season (Lirman et al. 2003, Caccia and Boyer 2005), although reversal of this seasonality can occur (Chapter 3). Hypersalinity (>40 ppt) can occur along the shoreline as well (Roessler and Tabb 1974, Brook, 1977; Lirman et al. 2016).

The Comprehensive Everglades Restoration Plan (CERP) was authorized as a multi-agency initiative to restore the quality, quantity, timing, and distribution of freshwater flows through the greater Everglades terrestrial system and subsequent discharge to south Florida nearshore areas. A component of CERP, the Biscayne Bay Coastal Wetlands (BBCW) project, is anticipated to help achieve these goals in Biscayne Bay and improve its estuarine functionality. BBCW salinity goals for western central and southern Biscayne Bay (Shoal Point to Turkey Point: Fig. 5.1) include nearshore areas with oligohaline (0.5-5 ppt) and mesohaline (5-18 ppt) regimes at the shoreline trending towards ~20-28 ppt 600 m from the coast and beyond (RECOVER 2008).

To evaluate restoration performance and monitor ecological changes associated with CERP implementation, The REstoration COordination and VERification (RECOVER) team established a number of ecological indicators as well as Interim Goals that link indicator metrics to CERP activities (RECOVER 2005). One such indicator is pink shrimp *Farfantepenaeus duorarum*, which was selected to assess estuarine ecosystems due its purported direct and indirect responses to salinity conditions (Browder and Robblee 2009). Pink shrimp is ecologically important as a trophic link between primary and tertiary consumers (Browder and Robblee 2009). It is also economically important as multiple commercial fisheries target pink shrimp (Idyll 1950, Higman 1952, Saloman 1965, Berkeley et al. 1985, Hart et al. 2012, Johnson et al. 2012). CERP pink shrimp restoration goals for southwestern Biscayne Bay predicted increases in pink shrimp density during their peak abundance (i.e., Aug. to Oct., which was determined to be their time of peak abundance in western Florida Bay) :RECOVER 2005) with expansion of estuarine conditions (Browder et al. 2005a).

Browder and Robblee (2009) also proposed that reduction of salinity stress, including extreme variability, would lead to increased seagrass cover and spatial extent, resulting in increased pink shrimp densities. An apparent pink shrimp preference for shoal grass *Halodule wrightii* was reported by Costello et al. (1986). CERP is anticipated to increase total seagrass cover, especially of *H. wrightii*, along the southwestern shore of Biscayne Bay due to restoration of historical discharge patterns (RECOVER 2004). The CERP Pink Shrimp Interim Goal for southwestern Biscayne Bay predicts increased density averaging 2 shrimp m^{-2} in optimal nearshore benthic habitat (i.e., seagrasses) during August-October peak abundance periods (RECOVER 2005). To evaluate these predictions, I investigated potential changes in juvenile pink shrimp habitat suitability under a simplified scenario of doubling freshwater inflow from the present canal system. This differs from potential CERP scenarios, which will divert a portion of canal flow through many smaller distribution points rather than just increasing discharge, not necessarily doubling. Salinity simulations that better match the CERP scenarios are presently under development (J. Luo, Pers. Comm.).

In Chapter 2, I noted a recognized need for tools enabling prediction of pink shrimp habitat quality changes with respect to CERP implementation (Zink et al. 2017). Habitat suitability index (HSI) models represent one type of tool. HSI models provide quantitative estimates of habitat preference and/or utilization based on application of simple mathematical expressions relating species-specific relationships to habitat attributes (USGS 1981, Brooks 1997). Habitat suitability scores are assigned to specific times/locations based on a standardized score ranging from 0 to 1, indicating a gradient of suitability conditions, such as critical, poor, fair, good, and optimal, for the habitat attribute in question. Where appropriate, an HSI designation might reflect a composite model integrating multiple habitat attributes into one standardized score (USGS 1981, Brooks 1997). Recently, HSI models have been developed to provide insight into habitat quality for goldspotted killifish (*Floridichthys carpio*) in Biscayne Bay (McManus et al. 2014) and changes in habitat suitability with respect to climate change for selected fishes and invertebrates in Florida Bay (Kearney et al. 2015). While a pink shrimp HSI model exists (Mulholland 1984), its functional relationships relating habitat suitability to habitat attributes are not supported statistically.

Historically, HSI models developed from abundance data relied on index values derived from “index graphs” and assigned location-specific suitability values based on relative abundance given habitat attribute value(s) at that location (USGS 1981). More recently, statistically supported multivariate HSI models based on either probability of use (e.g., logistic regression) or mean abundance (e.g., generalized linear model) relationships have been preferred (Brooks 1997, Boyce and McDonald 1999, Boyce et al. 2002).

Logistical constraints or imperfect knowledge of species' requirements may constrain HSI model development to consider only a subset of biophysical factors that potentially limit the presence or abundance of an organism (Terrell et al. 1996, Cade et al. 1999, Cade and Noon 2003, Cade et al. 2005, Lancaster and Beylea 2006). Quantile regression overcomes these restrictions when investigating ecological constraints on organismal presence, or abundance, relative to a habitat attribute (predictor), by focusing on the distribution's upper edge, where other measured or unmeasured limiting habitat attributes are not limiting (Thompson et al. 1996, Cade et al. 1999, Cade and Noon 2003, Lancaster and Beylea 2006). Previous studies have successfully incorporated quantile regression in habitat suitability modeling exercises (Eastwood et al. 2003, Vaz et al. 2008, VanDerWal et al. 2009). This statistical approach aligns conceptually with niche theory, potential vs. realized habitat, and Liebig's law of the minimum (Taylor 1934, Thompson et al. 1996, Cade et al. 1999, Hiddink and Kaiser 2005).

The Biscayne Bay Simulation Model v4 (BBSMv4) is the most recent iteration of a series of hydrodynamic models designed to simulate salinity regimes and other circulation features within Biscayne Bay (Stabenau et al. 2015). Presently, two model scenarios are available for the BBSMv4: High Flow and Base Flow (Stabenau et al. 2015). In designing the High Flow model scenario, various mechanisms of delivering additional freshwater to the bay were considered (E. Stabenau, Pers. Comm.). Presently, much of the high-volume point source freshwater discharges via canals are "lost to tide."

This loss is caused by high canal discharge rates and freshwater buoyancy resulting in much of the inflow volume being advected and mixed further from the nearshore environment into higher tidal exchange regions. The transient influence on nearshore salinity regimes leads to increased variability in salinity conditions along the shoreline. BBCW features already in place or under construction would divert sizeable fractions of coastal freshwater outputs away from canal mouths and disburse it to multi-point flow-ways that lead into coastal wetlands. These redistributions will both reduce the direct high flow rate loss of freshwater to tide while discharging it more uniformly and at lower flow rates across wide-spread coastal wetlands and mangrove fringe. More efficient production of temporally stable nearshore mesohaline habitats is anticipated since lower freshwater discharge volume would be more effective at producing low salinity conditions along the shoreline relative to higher volume canal discharges. Conceivably, under full CERP implementation, larger volumes of freshwater may be available to discharge to southwestern Biscayne Bay.

In this study, I develop a pink shrimp HSI model and then use it to assess the impact of doubling freshwater canal discharges on pink shrimp habitat along Biscayne Bay's southwestern shoreline. The HSI is based on quantile regression results in which 10 years (2007-2016) of shrimp density observations are regressed against a suite of potentially limiting habitat attributes. Analyses focused on habitat attributes that were previously identified as limiting to pink shrimp densities (Chapter 3) and are anticipated to be affected by CERP implementation. The HSI model was applied to 15 calendar years (CYR: 1996-2011) of BBSMv4 modeled salinity conditions to predict changes in habitat suitability between Base Flow and High Flow BBSMv4 freshwater discharge scenarios. Quantification of predicted HSI values was used to reveal areal changes in pink shrimp habitat suitability under potential western Biscayne Bay nearshore salinity conditions (i.e., 50 m buffer along the coastline) should salinity Interim Goals (i.e., mesohaline and oligohaline regimes) be achieved. The results of this work will be of direct relevance to environmental managers and scientists wishing to glean insight of potential future impacts of CERP implementation on Biscayne Bay nearshore juvenile pink shrimp habitat suitability. It should be recognized, however, that juvenile pink shrimp habitat in Biscayne Bay extends beyond the 50-m strip of shallow open water that is the focus of the simulation presented in this section. More generally, the following study contributes to better understanding of the habitat needs of pink shrimp, a species that occurs across a broad geographical area (Zink et al. 2017, Chapter 2).

5.3 Methods

5.3.1 Study Site

Biscayne Bay is one of many south Florida bays and estuaries where salinity regimes were drastically altered by the Central and South Florida Flood Control Project (CSFFCP). Although mainland shoreline salinity conditions are presently higher than historical conditions, rapid and large, but ephemeral salinity reductions occur due to freshwater discharge from canals and water control structures. These unnatural salinity conditions have altered Biscayne Bay's benthic submerged aquatic vegetation (SAV), infaunal, epifaunal, and nektonic communities (Brook 1982; Irlandi et al. 1997; Serafy et al. 1997; Lirman et al. 2008a, b, 2014; Santos et al. 2011). Total annual surface and groundwater discharges are lower than those prior to CSFFCP development (Langevin

2003, Stalker et al. 2009). These changes have greatly diminished the estuarine nature of Biscayne Bay's western mainland shoreline, resulting in more polyhaline and euhaline conditions as well as loss of estuarine species and communities (Smith 1896, Kohout and Kolipinski 1967, Serafy et al. 2003, Browder et al. 2005). The present study focused on habitat suitability predictions matching the spatial domain of the field sampling data used to construct the HSI model. This area consisted of 158.75 ha of shallow-water habitat within a 50 m buffer of the shoreline.

5.3.2 Field Sampling and Sample Processing

Pink shrimp density information was obtained from seasonal (wet: May to Oct.; dry: Nov. to April) sampling at 47 fixed sampling sites situated along ~22 km of Biscayne Bay's southwestern shoreline from Shoal Point to Turkey Point (Fig. 5.1) during calendar years (CYR) 2005 through 2016. All sites lay within 50 m of the shoreline. Input data and results are sometimes discussed by Water year (WYR). In this application, each water year starts in May of the previous calendar year (CYR). For example, WYR 2010 started in May of CYR 2009 and ended in April of CYR 2010.

Sampling procedures previously described in Chapter 3 will be summarized here. Upon each site visit, water temperature (˚C), salinity (ppt), pH, dissolved oxygen saturation (%), dissolved oxygen concentration (mg/L), and water depth (m) were recorded. Species-specific seagrass and macroalgal % cover and total SAV canopy height were assessed within 10 replicate 0.5 m^2 quadrats per site (Lirman et al. 2008a, b). An open-ended, rigid-sided aluminum throw trap measuring 0.45 m (height) x 1 m (width) x 1 m (length) (Kushlan 1981, as modified by Robblee et al. 1991) was used to sample epifaunal communities with 3 throws at each site (summing to a $3m^2$ sample).

Samples were frozen for storage until sample processing. Taxonomic identification and size measurements of captured penaeid shrimp, including carapace length (CL, mm), total length (TL, mm), and sex, were recorded. Shrimps >8.0 mm CL were identified to species, where possible, while specimens <8.0 mm CL were identified only to genus because species identification depended on sexual morphological characteristics lacking sufficient development in smaller individuals (Perez-Farfante 1969, Perez-Farfante 1970, Perez-Farfante 1988, Perez-Farfante and Kensley 1997).

5.3.3 HSI Development and Validation

All statistical analyses and model prediction calculations were conducted using the R statistical package (The R Foundation, https://www.r-project.org/). Type 1 error criterion was set to $\alpha = 0.10$ to reduce probability of Type 2 errors. Only farfantepenaeids and *F. duorarum* ≥5 mm CL were included due to apparent sampling inefficiency in capturing smaller sized shrimps (based on size-frequency distribution, in which smallest sizes should be most numerous if sampled as efficiently as the larger sizes: Chapter 3). Shrimp counts from replicate throw-trap deployments were summed and divided by 3 m^2 to obtain site density values. Shrimp densities were log-transformed before further analysis.

Consistent with analyses presented in Chapter 3, quantile regression (QR) was used to identify the functional shape and respective coefficients of equations describing pink shrimp density versus habitat relationships. While four identified habitat attributes (temperature, salinity, water depth, and total SAV % cover) were identified in Chapter 3 as potentially limiting to pink shrimp density, analyses focused on those attributes likely to be affected by CERP implementation (i.e., direct salinity and indirect effects of salinity via SAV % cover responding to salinity: Robblee and Browder 2009). Other habitat attributes likely limit potential maximal densities; however, in single-predictor quantile regression, focusing on the upper edge of the density-related habitat attribute being addressed allows the practitioner to examine potential limitation on a case-by-case basis with relative isolation from other potentially interacting factors (Cade et al. 1999, Cade and Noon 2000, Lancaster and Belyea 2006). The potential limitation of SAV % cover by salinity was also investigated. Relationships emerging along the upper edge (i.e., the 0.9 quantile) of the examined density-habitat distributions, if statistically significant, were considered as indicative of a density-limiting effect. Training data for revealing these relationships included the 10 years from CYR 2007-2016.

Instead of observed salinity at the time of sampling, the present analyses used salinity metrics 35 days antecedent to the time of sampling. The use of antecedent salinity metrics was presumed to yield improved density-salinity limitation relationships by providing integrative metrics (average, maximum or other) of salinity conditions experienced by pink shrimp prior to the time of sampling. Salinity observations were collected at 15 min intervals via a network of 17 water quality sondes (YSI 6600) located at fixed water quality monitoring stations throughout nearshore Biscayne Bay within 50 m of the shoreline (Lirman et al. 2017). These observations were used to calculate antecedent salinity metrics (e.g., maximum and mean salinity). Several gaps in the continuous salinity data due to undetected instrument failure were filled with multiple linear regression from five nearby salinity stations. The continuous salinity records were assigned to epifauna sampling sites based on their proximity to the water quality sonde locations.

Antecedent salinity metrics were computed for a 35 d period prior to the time of sampling based on a post-settlement estimate of linear growth rate of 0.15 mm CL per day. This is an average linear field growth rate for median size shrimp, 8.14 mm CL pink shrimp, developed from previously published rates representing a variety of field conditions (Table 2.7: Chapter 2, Zink et al. 2017). The calculated linear growth rate of 0.15 mm CL per day and resulting 35 d post-settlement period of residence seem reasonable when assuming a settlement size of 3 mm CL (Chapter 2).

To investigate potential functional relationships between pink shrimp density and habitat attributes, a series of linear, quadratic, cubic, log-linear, and natural cubic spline (function “ns” of R package “splines”) functional response QR models were tested using the “qr” function (R package “quantreg”). Multiple knot-structure natural cubic spline shapes were considered; these were constructed with one, two, or three internal knots located at the 0.5; 0.33 and 0.66; or 0.25, 0.5, and 0.75 percentiles of the predictor distribution, respectively, both with and without estimation of intercepts. Although QR theoretically accommodates environmental variability not associated with the specific predictor under investigation by focusing on the upper edge of the predictor-dependent variable relationship, the effect of removal of a number of potentially confounding habitat conditions was investigated. Specifically, once the functional form to be used in further analysis was selected, the influence of extreme temperatures (<18 °C and >35 °C) as well as water depths (<20 cm and >120 cm) on the coefficient values of this QR were investigated. Concern about limitations in recruitment of settling pink shrimp postlarvae south of Black Point (Ault et al. 1999, Chapter 3) led to investigation of QRs focusing solely on density observations to the north of Black Point.

The final selected QR model was used to classify predictor habitat attributes by HSI level. First, shrimp densities were calculated for the respective upper and lower HSI level limits. The maximum HSI value attainable (i.e., 1) was assigned to the maximum density value predicted by the QR model, while the minimum predicted density (i.e., 0 shrimp m^{-2}) was set as the lowest HSI value (i.e., 0). Between these values, 80, 60, 40, and 20% of the maximum predicted shrimp density were established as thresholds between five levels of habitat suitability: optimal, good, fair, poor, and critical, respectively (McManus et al. 2014). Once the density limits were calculated, habitat attribute values corresponding to the HSI thresholds were predicted. These habitat attribute predictions were used to categorize modeled habitat attribute conditions with respect to the five habitat suitability levels. Where necessary, composite HSI values were computed as the geometric mean of multiple habitat attribute-specific HSI values. The range of predicted values spanned the range of HSI values from 0 to 1, and, therefore, calibration was unnecessary.

Validation of the HSI was tested against an independent validation data set consisting of pink shrimp density and habitat attribute observations collected during 2005-2006 CYR sampling events (i.e., 2 yr). Validation procedures recognized the reliance of the HSI development on a QR statistical framework and used QRs, again focusing on the 0.9 quantile, to investigate consistency of the relationships between observed pink shrimp densities and habitat attributes, predicted densities, and predicted

HSI levels (e.g., Eastwood et al. 2003, Vaz et al. 2008). Three approaches were used to check for consistency in the developed HSI model: (1) similarity between QR coefficients resulting from the training and validation datasets; (2) significance and validity of QR coefficients from observed shrimp densities versus densities predicted by the HSI model; and (3) significance and validity of QR coefficients of observed shrimp density versus predicted HSI value predictions. Nonparametric rank-correlation (i.e., Kendall's tau) identified whether a positive, significant correlation existed between observed densities and predicted HSI levels (Vaz et al. 2008).

5.3.4 Hydrodynamic Model Framework and HSI Application

The Biscayne Bay Simulation Model v4 (BBSMv4) is the most recent version of a Biscayne Bay hydrodynamic model (Wang 1980, Wang et al. 1988, Wang et al. 2003, Stabenau et al. 2015). Briefly, the model uses nonlinear hydrodynamic equations to resolve a finite element, vertically integrated (two-dimensional) array of 6,857 nodes and 13,075 elements to determine salinity fields affected by water motion, advection, and diffusion across the model grid. For a full description of BBSMv4, see Stabenau et al. (2015). Two model scenarios were developed to investigate Biscayne Bay salinity conditions under a *status quo* and a possible CERP restoration scenario: Base Flow and High Flow (Stabenau et al. 2015). The High Flow Model scenario aimed to produce, on average, at least 4,406 ha (10,000 acres) of mesohaline habitat (Stabenau et al. 2015), a target for Biscayne Bay nearshore salinity regime restoration advanced by the National Park Service (SFNRC 2006). This was achieved within the model scenario by drastic increases, generally representing a near doubling, of coastal freshwater point-source (i.e., canal) discharges (Table 5.1).

Investigation of salinity and HSI trends in the pink shrimp study focused on the temporal resolution of the empirical observations (i.e., field sampling events used to develop the quantile regression HSI model) following the South Florida Water Management District Water-year (WYR) convention, with wet season (May-Oct.) and the following dry season (Nov.-April), which spans the end and beginning of a CYR. First, daily 35 d antecedent salinity metrics were calculated by averaging the 20 min time step salinities simulated by the model for the 35 d prior to each day of the simulation (i.e., 00:20 hr time step of 35th d prior to 24:00 hr time step of the present day). Seasonally averaged 35 d antecedent salinity values were then calculated as the mean daily values for each wet and dry season. Heatmaps were generated to depict spatial (i.e., across sites) and temporal (i.e., across 15 consecutive wet-dry seasons) seasonal 35 d antecedent salinity value trends between model scenarios as well as the difference in seasonal 35 d antecedent salinity value (i.e., ΔSalinity) between scenarios.

Heatmaps depicted spatial and temporal HSI level trends classified from seasonally averaged 35 d antecedent salinity values calculated from the simulated BBSMv4 outputs for both scenarios. Base Flow, High Flow, and change in HSI level (ΔHSI) classifications between the model scenarios were depicted for each WYR and season at the 47 field sampling sites. Time series of daily 35 d antecedent salinity values and predicted HSI levels were visualized at three representative sampling sites (sites 10, 18, and 33: Fig 1) for WYR 2004 and both model scenarios. These sites were previously noted as exhibiting high, intermediate, and low shrimp density temporal trends (Chapter 3), while WYR 2004 was an example of extreme wet season low salinity conditions.

Distances between adjacent field sampling sites (N = 47) ranged from ~114 to 1,213 m. A finer spatial resolution of simulated salinities was desired to better quantify changes in areal extent among the five habitat suitability levels. A series of interpolated location latitude and longitude coordinates spaced ~50 m apart was established from along the shoreline from sampling site 1 in the north to sampling site 47 in the south (Fig. 5.1; ESRI, Redlands, CA, USA). This interpolation of shoreline sites yielded 635 average salinity estimates. A series of daily 35 d antecedent salinity values were calculated from 20 min time step simulated salinities extracted from the BBSMv4 at the 635 interpolated sites. Seasonally averaged simulated salinities for each WYR were then averaged to generate a series of Average WYR values. Predicted HSI levels were assigned to the 35 d antecedent salinity values for each interpolated location. To visually compare seasonal and model scenario average salinity and HSI value predictions, a series of maps was created depicting WYR 2004 and Average WYR values along the shoreline. Differences in areal extent of each level of HSI were quantified for WYR 2004 and Average WYRs as the number of data points, each representing 50 m of shoreline length multiplied by 50 m of offshore extent, which reflected the nearshore sampling domain of the original pink shrimp density observations. A total habitat area of 158.75 ha was assessed (635 cells of 2,500 m^2 each).

5.4 Results

A total of 2,419 shrimps from 2,820 throw trap samples covering wet and dry seasons of 2007-2016 were used for statistical analysis and HSI development. Pink shrimp density observations ranged from 0 to 13.0 shrimp m^{-2}. Continuous salinity observations could be reliably calculated for sites 1 to 44 of the 47 field sampling sites because sites 45-47 were too far distant from salinity sonde sites for reliable assignment of salinity values. Observed 35 d antecedent mean salinities ranged from 6.46 to 42.37 ppt; hypersaline (>40 ppt) conditions were observed in 12 instances (1.4%, N =880). All instances of hypersalinity were observed during the WYR 2012 wet season (July 2011). Observed 35 d antecedent maximum salinities ranged from 16.40 to 46.09 ppt and hypersaline conditions were slightly more prevalent (n = 52, 5.9%, N = 880). Most instances of 35 d antecedent maximum salinity occurred in the WYR 2012 wet season (n = 42, 95.5% of observations for that year-season), with the remaining values occurring in wet seasons of WYR 2009 (n = 5) and WYR 2016 (n = 5). Benthic SAV information was available only from 2008 through 2016, when SAV % cover observations ranged from 4.57 to 100%, 0 to 86% for seagrasses, 0 to 72.5% for *T. testudinium*, and 0 to 86% for *H. wrightii*.

5.4.1 HSI Development and Validation

Quantile regression revealed several significant functional shapes that could have been used to quantify limitation of pink shrimp density as a function of 35d antecedent mean or maximum salinity (ppt) (Fig. B1 and B2; Table B1 and B2). The 3-knot natural cubic splining function representing limitation of shrimp density by either 35 d antecedent mean or maximum salinity (ppt) represented the most ecologically plausible functional response that also yielded a statistically significant quantile regression. A graphical examination revealed that density observations associated with extreme temperature and water depth observations were located generally within the main spread of the data and thus exclusion of them was unlikely to influence QR results (Fig. B3B, Fig. B4B). Indeed, comparison of QR coefficients calculated with the full dataset or with these extreme values removed revealed no significant change in their values (Table B3, Fig. B3D, Table B4, Fig. B4D). Focus only on observations north of Black Point prevented inclusion of a number of relatively high density observations at higher salinities (Fig. B3C, Fig. B4C). These observations proved valuable to the resultant QR, as the shape of the predicted curve using only the northern observations drastically differed from the curve based on the full dataset (Fig. B3D, Fig. B4D) and lacked a statistical significance for the natural cubic spline fit between the median and maximum 35 d antecedent mean salinity values (i.e., knots 2 and 3) (Table B3, B4). In comparison to alternatives of data inclusion and model structure, the 3-knot natural cubic spline using 35 d antecedent mean salinity at all possible sites produced a better fit and was more inclusive of salinity conditions experienced by pink shrimp prior to observation of their density.

Lack of significance or ecologically implausible functional responses were observed with QR of pink shrimp density with either *T. testudinium*, *H. wrightii*, or total seagrass % cover and so these predictors were not further considered (Chapter 3). Furthermore, SAV % cover was not significantly limited by salinity (ppt) (Table 5.2). Therefore, potential effects on shrimp abundance via salinity effects on SAV were not pursued. Computed habitat suitability values, therefore, focused solely on salinity (ppt) at sampling site locations (Fig. 5.2).

Application of habitat suitability thresholds to the 3-knot natural cubic spline density-35 d antecedent mean salinity QR relationship yielded a dome-shaped response that maximized density at 29.25 ppt. Classification of this QR into the five habitat suitability levels yielded optimal suitability ranging from 38.68 down to 21.77 ppt, good ranging from 42.37 ppt (i.e., the maximum 35 d antecedent mean salinity observed) down to 38.68 ppt and from 21.77 down to 18.31 ppt, fair ranging from 18.31 down to 15.15 ppt, and poor ranging from 15.15 down to 11.44 ppt; 35 d antecedent mean salinities <11.44 ppt were classified as critical habitat suitability (Fig. 5.2).

All three HSI model validation methods yielded significant QRs (Table B5). Although quantile regression trends of the HSI model and validation of shrimp density observations to salinity appeared different visually (Fig. B5A), the validation salinity coefficients did not statistically differ from that of the training model (Table B5). Validation QRs relating both predicted densities and computed HSI values to observed densities yielded significant, positive slopes, suggesting the HSI model was valid (Fig. B5B, C). Although significant ($z = 5.97$, $p < 0.0001$), a low Kendall tau (0.36) was evident between predicted HSI levels and observed shrimp densities, but the case for validation was supported by a lack of high observed densities in plot regions corresponding to both low QR density (Fig. B5B) and low habitat suitability classification (Fig. B5C).

5.4.2 Base and High Flow Scenario Salinities

The 20-min step resolution of the recording sondes resulted in 13,428 and 13,032 (leap year = 13,104) time steps for wet and dry season salinity analyses, respectively. A total of 2,820 sampling site seasonally averaged 35 d antecedent mean salinity observations were simulated for wet and dry seasons and Base Flow and High Flow model scenarios across the 15 WYR simulations. Increased water management structure discharges in the High Flow scenario (Fig. 5.1, Table 5.1) decreased seasonally averaged 35 d antecedent mean salinity (Fig. 5.3, A5). The most drastic reductions in 35 d antecedent mean salinity occurred along the stretch of coastline bound by sampling sites 18 and 39 (Fig. 5.3, B6). These northern and southern bounds coincide with the Black Creek and Mowry Canals, respectively, which exhibit the highest canal discharges to the Shoal Point to Turkey Point stretch, as reflected by modeled discharge volumes (Table 5.1). Average WYR (i.e., averaged across years and seasons) 35 d antecedent mean salinities were lower in the High Flow model scenario for both wet ($\Delta = 2.61$ ppt) and dry ($\Delta = 2.47$ ppt) seasons (Table 5.3). The distribution of High Flow 35 d antecedent mean salinities was significantly lower than those of the Base Flow scenario for both wet ($D = 0.213$, $p < 0.0001$) and dry ($D = 0.220$, $p < 0.0001$) seasons. Between-scenario Average WYR minimum and maximum salinities did not substantially differ, indicating similar ranges in simulated site-averaged salinity conditions (Table 5.3). However, 100% of annual WYR High Flow minimum salinities were lower, or equal to, those of the Base Flow model scenario for both wet and dry seasons. Similarly, 96.9% of High Flow maximum salinities were lower than those of the Base Flow model scenario for both wet and dry seasons.

Among sampling sites and years (N = 705), 99.1% (n = 699) of dry season and 100.0% (n = 705) of wet season average modeled 35 d antecedent mean salinity observations were lower in the High Flow relative to the Base Flow model scenario (Fig. 5.3). For the dry season, the maximum increase in 35 d antecedent mean salinity was 0.90 ppt (site 38, WYR 2010: Fig. 5.4B). For those seasonally averaged 35 d antecedent mean salinities that were lower in the High Flow model scenario, the average 35 d antecedent mean salinity decrease was 2.62 and 2.50 ppt in the wet and dry seasons, respectively, while the maximum decreases were 8.04 and 10.23 ppt, respectively. Reflecting the modeled approach for achieving increased mesohaline habitat via increased freshwater discharges from water management structures, the most drastic salinity reductions occurred from site 18 to 37 for both seasons (Fig. 5.3, B6). The sum of 35 d antecedent salinity reductions due to increased water discharges was greater in the wet season (Fig. 5.3A) than in the dry season (Fig. 5.3B).

Across all sampling sites, dry season 35 d antecedent mean salinities averaged by season were greater than wet season averaged mean salinities for just over half the observations in both the Base Flow (n = 400, 56.7%) and High Flow (n = 411, 58.3%) model scenarios (Fig A5). Similarly, 60.0% (n = 9) of Base Flow and 53.3% of High Flow scenario annual WYR salinities (i.e., averaged across sites) were lower in the wet season (Table 5.3). Generally, minimum 35 d antecedent mean salinities across sampling sites were lower for the wet season in both model scenarios (Table 5.3). However, the WYR 1999 dry season minimum salinity was lower than that of the wet season in the High Flow scenario. Maximum 35 d antecedent mean salinities were more often observed in the wet season for both Base Flow (n = 10, 66.7%) and High Flow (n = 10, 66.7%) model scenarios (Table 5.3). No instances of hypersalinity were observed for seasonally averaged 35 d antecedent mean salinity (Table 5.3). Further, hypersalinity was not observed in daily 35 d antecedent mean salinity observations.

5.4.3 Spatial and Temporal HSI Trends

Application of the HSI regression model to the simulated salinity outputs across wet and dry seasons for the 15 WYRs and the Base Flow and High Flow model scenarios resulted in 2,820 HSI estimates at the original field sampling sites. Sampling sites' classified HSI values were generally high throughout both seasons over both model scenarios (Table 5.4, Fig. 5.4). Summarizing across all WYRs and sampling sites and both seasons, the majority (n = 2,215, 78.5%) of HSI predictions were categorized as either “optimal” or “good” habitat suitability. Few instances were identified as “fair,” “poor,” or “critical” habitat suitability (Table 5.4, Fig. 5.4), although collectively these habitat suitability levels represented 21.4% of the total simulated HSI estimates.

Only two instances (0.14%, N = 1410) of increased habitat suitability were realized between Base to High Flow model scenarios: these occurred during the dry season at sites 38 and 39 (Fig. 5.4F). The majority of habitat suitability values (n = 850, 60.3%) did not change between Base Flow and High Flow scenarios for both wet and dry seasons (Fig. 5.4C, F). Habitat suitability was reduced in 558 cases (39.6%: Fig. 5.4C, F). Most decreases in habitat suitability were by one HSI level (n = 418, 74.9%). Fewer instances of reduction by two HSI levels were observed (n = 139, 24.9%) and only one instance of reduction by three HSI levels was observed (0.17%).

Spatially, trends in HSI predictions indicated a general reduction within the central region of the sampling domain (sites 18-39) for both Base and High Flow model scenarios (Fig. 5.4). Indeed, 95.7% (n = 534) of all habitat suitability reductions between model scenarios occurred in this central region of the sampling domain (Fig. 5.4C, F). Northern (sites 1-17) and southern (sites 40-47) regions of the sampling domain exhibited "optimal" and a few instances of "good" suitability conditions (Fig. 5.4). The northern region (n = 13, 2.3%) and southern region (n = 11, 2.0%) accounted for few of the total changes in habitat suitability level between model scenarios (Fig. 5.4C, F).

Seasonal and inter-annual patterns of reductions in HSI levels were less consistent than spatial trends (Fig. 5.4). Between model scenarios, similar amounts of habitat suitability reduction were observed in both seasons (~40%). Reductions in predicted HSI levels in the central region of the sampling domain (sites 18-39) for both model scenarios (Fig. 5.4A, B) in the wet season. Habitat suitability levels were not as drastically reduced in the central region relative to the north and the south in the dry season, nor were reductions as consistent on an inter-annual basis (Fig. 5.4D, E).

Time series of daily 35 d antecedent mean salinity simulated for the Base and High Flow scenarios were plotted for WYR 2004 at three sampling sites (Fig. 5.5), selected as representative of high (site 10), intermediate (site 33), and low (site 18) pink shrimp densities (Chapter 3). WYR 2004 was selected because wet season 35 d antecedent salinities were the than in any other WYR (Table 5.3, Fig. B6). Simulated 35 d antecedent mean salinity values remained higher at site 10 (minima = 18.25 and 16.18 ppt for Base and High Flow model scenarios, respectively) than at site 18 (minima = 4.56 and 1.77 ppt for Base and High Flow model scenarios, respectively) and site 33 (minima

= 12.34 and 6.47 ppt for Base and High Flow model scenarios, respectively; Fig. 5.5). The investigation of daily salinity values, as opposed to seasonal averages, allowed consideration of more extreme salinity and HSI designation changes and transient behavior. Color coding of habitat suitability levels for each scenario revealed that the best conditions under either scenario occurred at sampling site 10 and the worst at sampling site 18. Based on color-coded HSI's, shrimp are favored by the Base condition, over the High Flow condition, at all three sampling sites, but especially at sites 18 and 33 (Fig 5C).

Areal changes in predicted HSI levels were calculated by expansion of simulated 35 d antecedent mean salinities and habitat suitability level predictions to points located every 50 m along the shoreline of the study domain. This resulted in 635 estimates of 35d antecedent mean salinity and corresponding HSI level along the focal Biscayne Bay shoreline for each model scenario and season. To depict these estimates, simulated 35 d antecedent mean salinity conditions and predicted habitat suitability for WYR 2004 and Average WYR for the Base Flow and High Flow scenarios were mapped as one-dimensional strips along shoreline study domain (Fig. 5.6). Spatial and seasonal patterns in simulated 35 d antecedent mean salinity (Fig. 5.6A, C) and corresponding HSI level (Fig. 5.6B, D) were readily apparent. Lower 35 d antecedent mean salinities during the wet season were apparent from the centrally located Black Point Canal mouth south to Convoy Point, whereas, in the dry season, the region of lower salinity was more spatially limited with its southern limit at about Fender Point. These trends apparent in the Base Flow model scenario, were accentuated in the High Flow scenario, which expanded regions of lower salinity further north and south, while further reducing salinity within the core lower salinity region. HSI values followed these spatial patterns, with fair, poor, and/or critical designations extending from the Black Creek Canal mouth south to at least Fender Point, and good habitat designations stretching further north and south. In summary, the High Flow scenario highlighted spatial trends, with areas of lower habitat suitability extending further north and south from their Base Flow scenario cores.

Summation of areal HSI estimates between High Flow and Base Flow scenarios allowed for visualization of changes between model scenarios and seasons (Fig. 5.7). Average WYR wet season areal extent of good and optimal suitability was 91.8% in the Base Flow scenario; no areas were classified as critical. For the High Flow scenario, Average WYR good and optimal suitability was reduced to 74.3%, and 6.5% of the area was classified as critical. No Average WYR dry season critical habitat suitability was observed in either flow scenario (Fig. 5.7). However, the area of good and optimal suitability was substantially reduced in the High Flow scenario (76.2%) relative to that of the Base Flow (93.7%). These trends were exacerbated in the lower salinity conditions of WYR 2004. For example, wet season critical habitat increased 3-fold from 7.7% in the Base Flow scenario to 23.8% in the High Flow scenario. Despite this substantial increase in critical habitat, good and optimal habitat totals were similar between Base Flow (67.2%) and High Flow (60.6%) scenarios of WYR 2004.

5.5 Discussion

The present study coupled a pink shrimp HSI model, developed from southwestern Biscayne Bay shrimp density observations and antecedent salinity conditions, to Base and High Flow model scenario outputs computed by the BBSMv4 hydrodynamic model. The High Flow scenario was developed to simulate a doubling of freshwater input into the focal system to reduce salinities and maintain them longer in the nearshore zone. My present HSI modeling results suggest that achievement of the anticipated salinity reductions would reduce pink shrimp habitat suitability in the presently simulated spatial domain (i.e., 0 – 50 m nearshore area from Shoal Point to Turkey Point). Nevertheless, ~75% of the study domain area remained of good or optimal habitat suitability in both wet and dry seasons of the High Flow scenario when considering average conditions over the entire simulation time period.

Wet season pink shrimp abundances were anticipated to increase in response to expansion of nearshore estuarine salinity habitat conditions (RECOVER 2005, Browder et al. 2005a). However, given the apparent limitation on juvenile pink shrimp densities by low salinity conditions, it is not surprising that the southwestern Biscayne Bay nearshore (0-50 m) area between Black Point and Fender Point exhibited lower habitat suitability. These results were not previously excluded (RECOVER 2005); indeed, as discussed by Browder and Robblee (2009), prior Florida Bay pink shrimp juvenile modeling exercises predicted reductions in abundance at salinities <20 ppt (Browder et al. 2002). This spatial pattern was more prominent in the wet season and under the High Flow model scenario, although it was also evident under the Base Flow scenario. Pink shrimp habitat suitability was generally higher in the dry season when 35 d antecedent mean salinity conditions were also higher. High habitat suitability along the coastline north of Black Point is consistent with previously reported pink shrimp abundance patterns in this region (Diaz 2001, Browder et al. 2005, Santos 2014, Chapter 3), as well as with suspected recruitment pathways and settlement patterns (Ault et al. 1999a, Wang et al. 2003). Pink shrimp distribution within coastal lagoons has elsewhere been observed limited to regions with salinities > ~15 ppt (Pérez-Casteñeda and Defeo 2001, 2004; Blanco-Martínez and Pérez-Casteñeda 2016). Juvenile distributions may also be affected by recruitment limitation (Criales et al. 2010).

The present analyses do not support previous predictions that pink shrimp densities and, therefore, habitat suitability, may be affected by seagrass species composition (Browder and Robblee 2009). Exploratory quantile regressions did not reveal seagrass species-specific limitation of pink shrimp densities. Although a significant limitation was noted in relation to total SAV % cover, the present failure to detect limitation of total SAV % cover by salinity precluded consideration of this habitat attribute as it relates to alteration of nearshore salinity regimes. However, the present analyses are limited by inability to account for SAV fragmentation, which would be influenced by salinity variability (Lirman et al. 2008a, Santos et al. 2011). Increased SAV cover, even if patchy, as well as increased heterogeneity in SAV cover, may also influence pink shrimp densities (Browder and Robblee 2009).

The High Flow model scenario achieved nearshore salinity reduction goals by effectively doubling water management control structure discharges (i.e., canal discharges: Stabenau et al. 2015). However, this scenario does not accurately recreate the intended post-BBCW implementation spatial redistribution and dispersion of freshwater inputs. From a modeling perspective, simulating this future scenario presents a challenge. The BBSMv4 model was calibrated against the existing conditions, and, while it has been validated against a range of flow rates, it has not been calibrated against new, yet unimplemented, CERP freshwater discharge features. Still, since the model is vertically integrated and, other than the additional volume of freshwater lost to tide, treats all freshwater input along the boundary as equivalent, it was appropriate to alter the input flow volumes to reveal expected results. Given these practical and logical considerations, the High Flow scenario was modeled by simply increasing the coastal freshwater point-source discharge until the mesohaline habitat area reached, on average, the NPS target 4,406 ha. Achieving this target under the less efficient point-source scenario required a near doubling of canal flow volumes, due to loss to tide. Full CERP restoration may achieve this goal by more efficiently utilizing freshwater deliveries.

Seasonal trends in 35 d antecedent mean salinities predicted by the BBSMv4 generally followed empirical observations except that its estimates did not reach empirically observed wet season hypersaline conditions. Poor representation of evaporation, a major pathway for freshwater loss, may play a role in this poor performance. Presently, the BBSMv4 models evaporation as an "average year" calculated from daily evaporation observations spanning 1996 to 2000 (Stabenau et al. 2015). Monthly mean values are likely unable to accurately recreate the high evaporation rates that, when paired with little or no freshwater outflow, result in hypersaline conditions (J. Luo, Pers. Comm.) Hypersaline conditions in Biscayne Bay have been observed in previous field studies (Roessler and Tabb 1974, Brooks 1977, RECOVER 2008, Lirman et al. 2014). Observed (i.e., calculated from water quality sonde) 35 d antecedent mean hypersaline conditions were not extreme (maximum = 42.37 ppt) although instantaneous values up to 47 ppt have been recorded in the region (RECOVER 2008). Often, outflows are withheld by water management operations until approximately June of each year, even when wet season rains start in May (Caccia and

Boyer 2007). Discharges generally remain high into the early dry (i.e., non-rainy) season despite the tapering of wet season precipitation in November, leaving little freshwater in the substrate and shallow aquifer to discharge into the late dry season (Caccia and Boyer 2007). This water management strategy often results in gradually increasing salinities during the spring (Caccia and Boyer 2007), leading to early wet season high-euhaline or even hypersaline conditions (Roessler and Tabb 1974, Brooks 1977, Lirman et al. 2003, Lirman et al. 2014). These conditions are exacerbated by increasing temperatures and evaporation of surface waters.

Given the limited realism of the High Flow scenario relative to BBCW plans, a future model scenario should be developed that would emulate the spatially and temporally redistributed freshwater inflows expected with CERP and BBCW restoration actions, as well as hypersaline conditions when and where interacting flow and evaporation rates would dictate them. While BBSMv4 refinement could address the present limitation of spatial redistribution of freshwater discharges, more realistic modeling scenarios would not alter the nearshore mesohaline goal driving both this High Flow scenario and development of the BBCW. In regard to salinity reductions, the general results of these simulations probably would not be drastically altered with redistribution of freshwater inflows. The predicted upper bound for the "critical" habitat suitability level was 11.44 ppt, which includes all future oligohaline and some mesohaline habitat that might be created. However, if hypersalinity were better emulated by the BBSMv4, then benefits to pink shrimp habitat suitability would likely include reduction in the spatial extent, duration, and severity of deleterious hypersaline conditions. This is suggested by the 38.68 ppt upper bound for optimal habitat suitability. Reduction of hypersalinity could also benefit pink shrimp abundances by reducing negative impacts on postlarval pink shrimp recruitment to nearshore areas (Criales et al. 2010). However, benefits of increased recruitment cannot be separated from post-settlement growth and survival processes with the current sampling design.

The association of lower salinity conditions (e.g., <15.15 ppt) with poor habitat suitability generally agrees with previously published studies of pink shrimp abundance (Gunter et al. 1964, Pérez-Castañeda and Defeo 2004, Zink et al. 2017: Chapter 2), habitat suitability modeling (Mulholland 1984, Rubec et al. 2016), and population simulations (Browder et al. 2002, as discussed in Browder and Robblee 2009). The present HSI model structure also recreated suspected limitation of pink shrimp densities at the highest observed 35 d antecedent salinities (≥35 ppt). The present HSI quantile regression predicted a maximal potential shrimp density at 29.25 ppt, which agreed with maximal growth (Browder et al. 2002; Zink et al. 2013, 2017; Chapter 2; Chapter 4) and survival (Browder et al. 1999, 2002; Zink et al. 2013; Chapter 2) relative to salinity condition. In their review of pink shrimp abundance relationships with salinity, Zink et al. (2017, Chapter 2) reported most (53.3%) pink shrimp density-salinity relationships maximized between ~20 and 35 ppt. Conversely, Rubec et al. (2016) predicted maximal pink shrimp summer concentrations at ~17 ppt with rapid declines below 10 ppt and above 20 ppt.

The habitat suitability model peaked at ~ 30 ppt. However, this dome-shaped response was non-symmetric with the descent in suitability to the left of the optimum (i.e., toward lower salinity) being steeper than the one to its right (toward higher salinity). This suggested that pink shrimp are not as adversely associated with euhaline conditions as with mesohaline and lower salinities. A similar non-symmetric response was presented by Mulholland's (1984) pink shrimp habitat suitability model: the descending slope to right of the optima was lower between 35 and 45 ppt; after this point, suitability decreased more rapidly. The present HSI model is limited in that it does not consider salinity conditions higher than the maximum 42.37 ppt 35 d antecedent mean salinity in the model input data. Other regions of south Florida exhibit higher pink shrimp densities than occur in our data from the southwestern Biscayne Bay nearshore area: in western Florida Bay they can average ~5 shrimp m^{-2} (Costello et al. 1986, Browder and Robblee 2009) and in the middle and lower Florida Keys 2.3 shrimp m^{-2} (Costello et al. 1986). Salinities at these locations are more stable and near-oceanic (Costello et al. 1986, Browder and Robblee 2009). Pink shrimp also were found at extremely high densities (annual average: 11.9 m-^{2}) in higher, more stable salinities (~36 ppt: Sheridan 1992) in the Ten Thousand Islands area of southwest Florida. These average densities are greater than those of Biscayne Bay (<2 shrimp m^{-2}: Chapter 3). However, study duration (1 yr: Costello et al. 1986, Sheridan 1992) and differential larval supply (Criales et al. 2015) likely influence these comparisons. At other locations throughout their range, pink shrimp occur in bays and lagoons known for high, and at times extreme, salinity conditions (Simmons 1957, Hedgpeth 1967, Hildebrand 1969, Wakida-Kusunoki et al. 2008, Ocana-Luna et al 2008, Pérez-Castañeda et al. 2012). Simmons (1957) reported pink shrimp abundance was only reduced above 45 ppt (Simmons 1957) and Ocana-Luna et al. (2008) reported maximal pink shrimp abundances in high euhaline and low hypersaline conditions (between ~ 38 and 44 ppt).

Consideration of daily HSI predictions allowed cursory investigation of effects of 35 d antecedent mean salinity variability on pink shrimp habitat suitability. Low wet season habitat suitability was predicted for sampling sites 18 to 26 under the Base Flow model scenario and for sites 18 to 34 under the High Flow model scenario. This "canal zone" region of Biscayne Bay (McManus et al. 2014), is known for low and highly variable salinity conditions (Irlandi et al. 1997; Serafy et al. 1997; Lirman et al. 2003, 2008a, b). Spatially, negative correlation between mean salinity and salinity variability has been observed in Biscayne Bay in both modeled (Lirman et al. 2008a) and empirical observations (Chapter 3). Daily 35 d antecedent mean salinities and HSI predictions illustrated the impact salinity fluctuations and low salinity conditions on nearshore pink shrimp habitat; during much of the wet season, site 18 salinity and HSI predictions were low throughout the WYR 2004 wet season. At site 33, fluctuating 35 d antecedent mean salinity conditions yielded mostly "critical" daily habitat conditions with interspersed higher habitat suitability conditions during the wet season of WYR 2004. Should simulation of hypersalinity within this spatial domain be improved, similar plots could be used to also demonstrate the ephemeral improvement in habitat suitability via reduction of these deleterious conditions.

Only one previous study has considered salinity variability effects on pink shrimp abundance, reporting on results of multiple regression models that salinity variability, and not mean salinity condition, was significant in predicting abundance (Berkeley and Campos 1984). While relatively little research is available relating salinity variability to penaeid shrimp abundance, salinity variability is known to affect estuarine organisms' bioenergetics (Wheatly 1988). Moderate salinity variability (abrupt changes of 4 to 10 ppt every 4 to 16 d) may maximize growth of at least some penaeids when fluctuations are extreme and/or rapid (Mu et al. 2005, Su et al. 2010). Further study to partition salinity mean versus variability effects on pink shrimp is warranted.

The present pink shrimp HSI model contributes to the presently available HSI models available for CERP restoration indicator species (McManus et al. 2014, Kearney et al. 2015). Further development of this pink shrimp HSI model may reveal other influences of salinity restoration on pink shrimp habitat suitability. With further refinement, the model may be adapted for other predictive applications, such as assessing potential changes in pink shrimp habitat suitability under various climate change scenarios (Obeysekera et al. 2011, Kearney et al. 2015). While the present single-species HSI predicts reductions in habitat suitability for pink shrimp due to low salinity conditions within the spatial domain, some other species are likely to benefit from restoration of mesohaline and/or oligohaline conditions. For example, goldspotted killifish *Flordichthys carpio* and *Palaemonetes* spp. shrimps might benefit from these salinity conditions (Lirman et al. 2016). The present study at least partially addresses an important line of inquiry for Biscayne Bay: “How is juvenile pink shrimp abundance affected by changes in the quantity, timing, and distribution of freshwater inflow?” (Browder et al. 2005). While some insight is provided, expansion of the present HSI’s development and its application to other south Florida estuaries might expand its applicability and better address this question. Further refinement of the BBSMv4 model conditions could also lead to habitat suitability predictions that better reflect intended BBCW freshwater delivery and distribution as well as better recreate empirically observed hypersaline conditions.

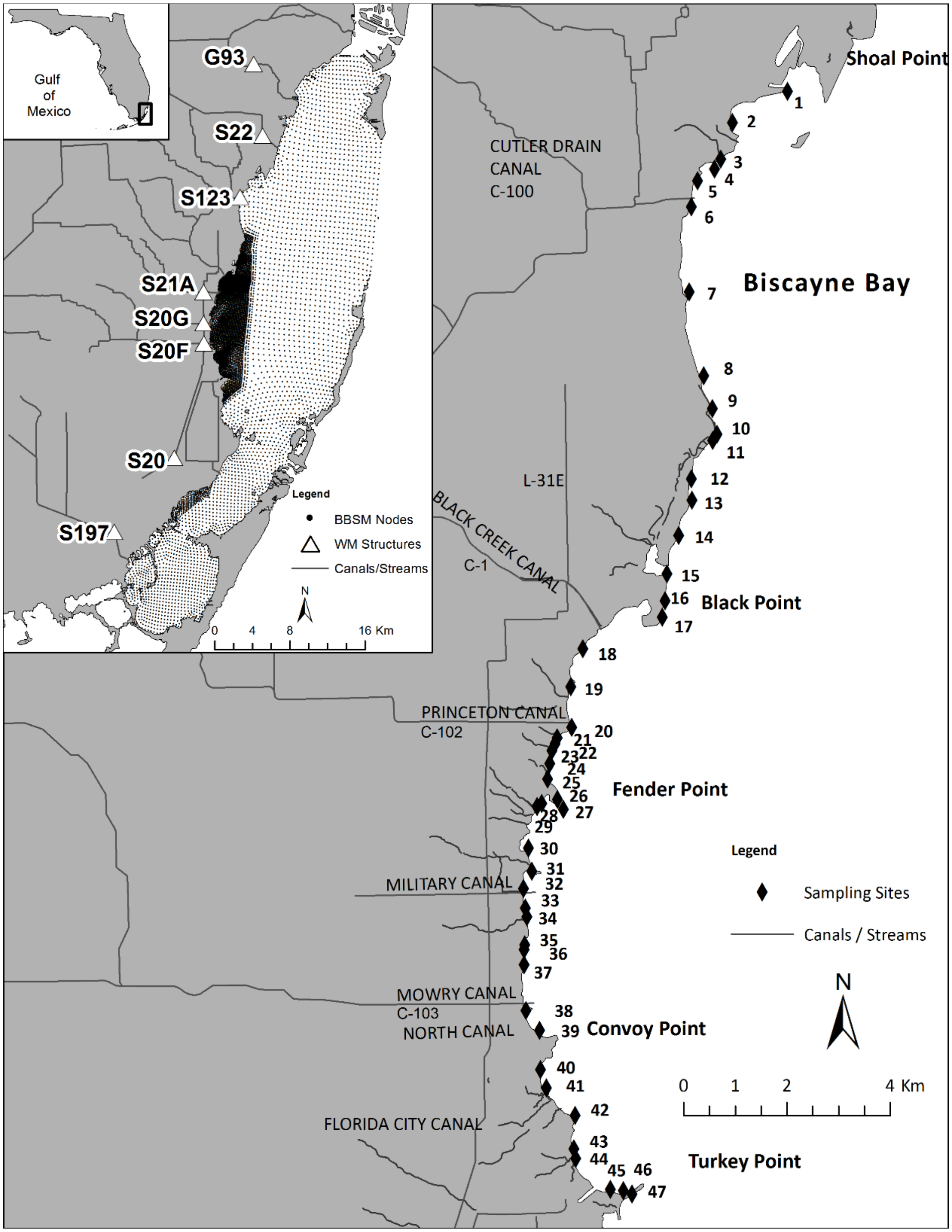


Fig. 5.1: Main map depicts the southwestern Biscayne Bay shoreline and the 47 epifaunal monitoring sites used to develop and investigate the pink shrimp HSI model. Map insert depicts the spatial domain and tessellation of the Biscayne Bay Simulation Model v4 as well as water control structures discharging to Biscayne Bay and the third insert orients the study region within Florida.

Table 5.1: Base (~0.57 million acre-ft) and High Flow (~1.1 million acre-ft annually) model scenarios' mean annual discharge, as computed from the Biscayne Bay Simulaition Model v4, from freshwater control structures discharging to Biscayne Bay (see Fig. 1).

Canal	Freshwater Control Structure	Base Flow – Mean Annual Discharge (acre-ft)	High Flow – Mean Annual Discharge (acre-ft)	Difference – Mean Annual Discharge (acre-ft)
Coral Gables Canal	G93	9,453	18,244	8,791
Snapper Creek Canal	S22	106,334	205,224	98,890
C-100 (Cutler Drain Canal)	S123	42,777	82,560	39,783
C-1 (Black Creek Canal)	S21	130,490	251,846	121,356
C-102 (Princeton Canal)	S21A	81,691	157,665	75,974
Military Canal	S20G	14,281	27,563	13,282
C-103 (Mowry Canal)	S20F	149,088	287,740	138,652
C-107	S20	14,032	27,082	13,050
C-111	S197	20,793	40,131	19,338
Total		568,940	1,098,055	529,115

Table 5.2: Statistical significance of 0.9 quantile regressions of pink shrimp density (shrimp m^{-2}) against salinity (ppt) and submerged aquatic vegetation (SAV: % cover) as well as for SAV % cover against salinity. LN = natural logarithm.

Dependent Variable	Predictors	Coefficients (± SE)	t value	p value
Density	Spline 1	2.47 ± 0.089	27.62	<0.0001
	Spline 2	0.37 ± 0.16	2.38	0.0177
Density	LN(SAV)	0.62 ± 0.19	2.59	0.0098
	Intercept	-0.28 ± 0.71	-0.38	0.7068
SAV	LN(Salinity)	2.27 ± 1.47	1.55	0.1216
	Intercept	85.53 ± 4.77	17.93	<0.0001

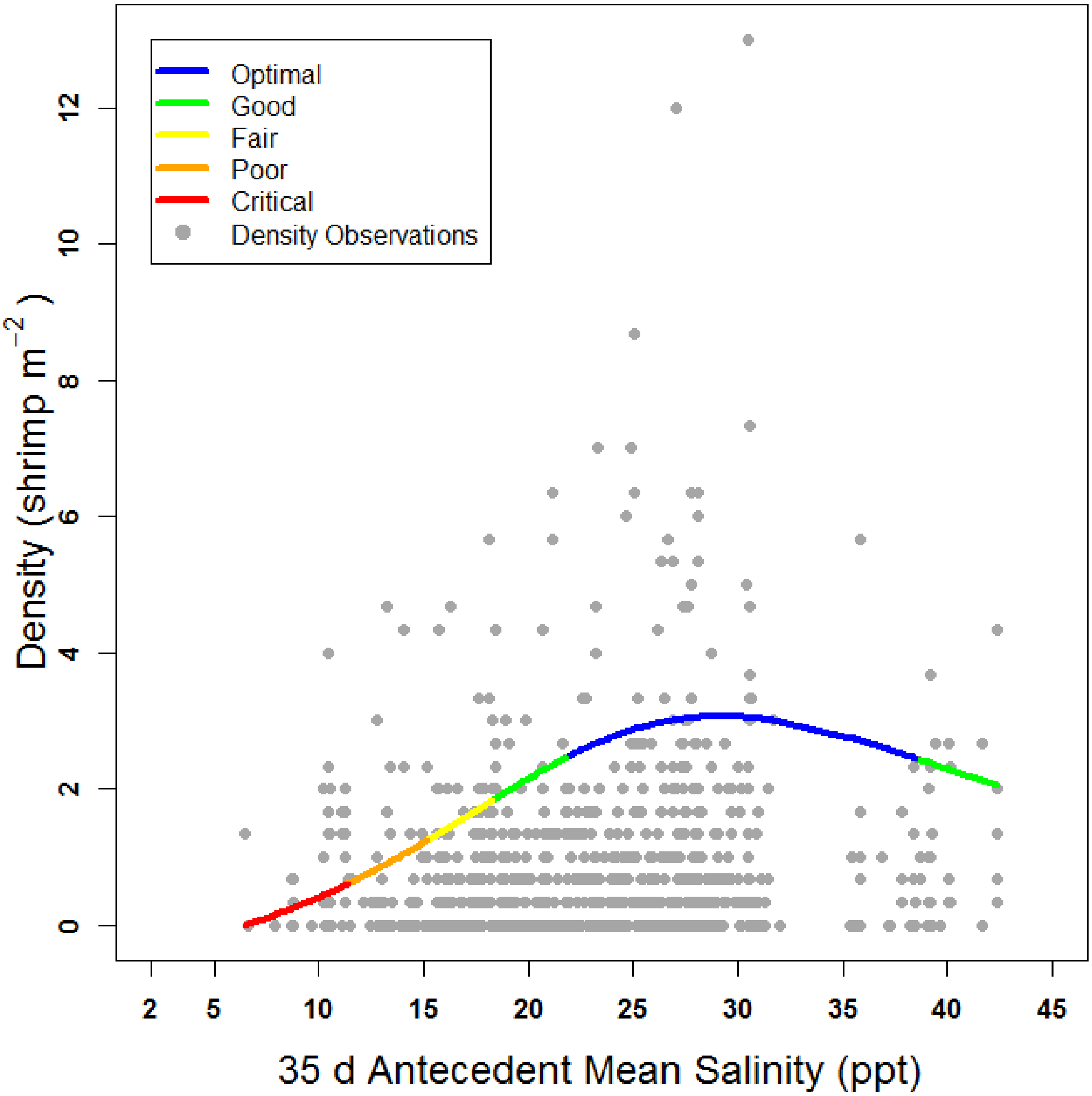


Fig. 5.2: Plot of final habitat suitability equation depicting limitation of pink shrimp density (shrimp m^{-2}) over the range of 35 d antecedent mean salinity (ppt) calculated from the *in-situ* salinity loggers. Points represent individual pink shrimp density observations used to develop the quantile regression. Colors represent five habitat suitability levels: optimal (0.8-1.0), good (0.6-0.8), fair (0.4-0.6), poor (0.2-0.4), and critical (0.0-0.2).

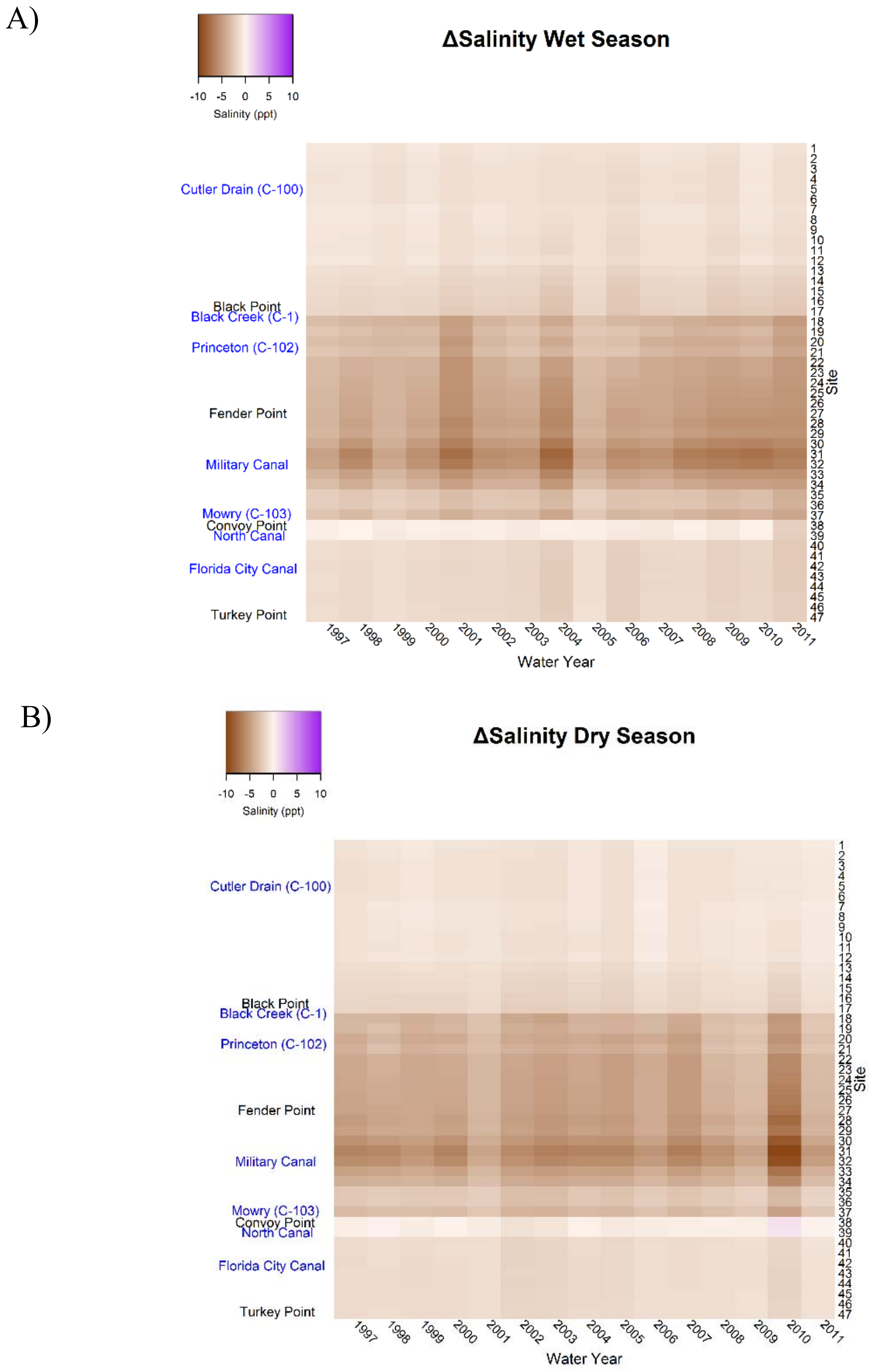


Fig. 5.3: Difference between Base and High Flow model scenarios' (A) wet- and (B) dry-seasonally averaged 35 d antecedent mean salinities for each of the 47 sampling sites and 15 water-years.

Table 5.3: Mean ± SD, minimum, and maximum 35 d antecedent mean salinity values of the 47 sampling sites simulated by Base and High Flow model scenarios for each water-year (WYR) and Average WYR. Mean ± SD were computed as mean of seasonally averaged values across sites while minimum and maximum represent 20 min time step values.

WYR	Season	Base Flow			High Flow		
		Mean ± SD	Min	Max	Mean ± SD	Min	Max
1997	Wet	22.46 ± 8.07	2.54	36.92	20.42 ± 9.14	1.06	36.79
	Dry	27.40 ± 4.93	9.97	36.13	24.84 ± 6.52	4.70	35.84
1998	Wet	20.67 ± 6.88	4.21	36.13	18.29 ± 7.93	1.61	35.86
	Dry	18.10 ± 4.70	5.47	28.84	15.79 ± 5.62	2.89	27.55
1999	Wet	23.49 ± 4.65	6.76	32.15	21.33 ± 5.91	3.38	31.42
	Dry	23.58 ± 6.31	6.87	35.68	21.20 ± 7.46	3.36	35.44
2000	Wet	22.90 ± 7.41	6.48	36.40	20.57 ± 8.93	3.30	36.34
	Dry	22.88 ± 4.59	7.41	33.34	20.39 ± 5.88	3.61	32.37
2001	Wet	26.43 ± 6.19	8.86	36.53	23.32 ± 8.02	4.43	36.13
	Dry	24.90 ± 5.01	9.27	34.51	22.83 ± 6.17	5.36	34.29
2002	Wet	22.90 ± 7.40	4.18	34.60	20.45 ± 8.78	1.74	34.39
	Dry	21.54 ± 5.16	7.48	31.70	18.80 ± 6.34	3.69	30.47
2003	Wet	21.96 ± 8.68	1.74	35.54	19.50 ± 9.92	0.56	35.4
	Dry	23.41 ± 4.66	8.80	31.99	20.52 ± 6.23	4.44	31.12
2004	Wet	19.68 ± 6.22	2.64	30.77	16.42 ± 7.46	0.77	28.92
	Dry	22.54 ± 5.70	7.65	34.01	20.02 ± 7.05	3.18	33.58
2005	Wet	28.74 ± 7.08	6.10	37.46	26.68 ± 9.17	1.96	37.48
	Dry	26.45 ± 5.15	7.60	34.46	23.68 ± 6.87	2.98	33.87
2006	Wet	21.21 ± 7.87	3.07	34.80	18.46 ± 9.35	0.94	34.36
	Dry	23.70 ± 5.65	7.55	34.99	21.50 ± 6.52	3.93	34.4
2007	Wet	24.79 ± 7.08	5.37	36.21	22.30 ± 8.50	2.16	35.82
	Dry	28.22 ± 3.88	14.15	34.81	25.49 ± 5.89	8.47	34.43
2008	Wet	22.83 ± 6.65	4.32	33.79	20.15 ± 7.69	2.22	33.23
	Dry	25.68 ± 4.48	8.10	32.98	23.47 ± 5.85	3.77	32.32
2009	Wet	25.43 ± 6.56	6.38	35.89	22.39 ± 8.44	2.28	35.63
	Dry	25.51 ± 6.22	6.74	36.33	23.60 ± 7.33	2.79	35.9
2010	Wet	24.81 ± 7.91	4.61	38.21	22.04 ± 9.60	1.66	38.12
	Dry	23.17 ± 4.61	11.87	32.17	19.73 ± 6.48	5.70	31.19
2011	Wet	22.00 ± 5.11	5.93	30.60	18.75 ± 6.54	2.22	28.71
	Dry	25.49 ± 5.53	11.09	35.80	23.62 ± 6.92	7.46	35.73
Over All	Wet	23.35 ± 7.35	1.74	38.21	20.74 ± 8.76	0.56	38.12
	Dry	24.17 ± 5.69	5.47	36.33	21.70 ± 6.96	2.79	35.9

Table 5.4: Counts and percentage of sampling sites classified among the five habitat suitability index (HSI) levels: [optimal (1.0-0.8 = 5), good (0.8-0.6 = 4), fair (0.6-0.4 = 3), poor (0.4-0.2 = 2), and critical (0.2-0.0 = 1)] across all water-years and sampling sites.

Season	Model Scenario	HSI Level									
		1		2		3		4		5	
		n	%	n	%	n	%	n	%	n	%
Both	Both Base and High (N = 2,820)	85	3.0	221	7.8	299	10.6	486	17.2	1,729	61.3
Both	Base Flow (N = 1,410)	11	0.8	47	3.3	117	8.3	276	19.6	959	68.0
Both	High Flow (N = 1,410)	74	5.2	174	12.3	182	12.9	210	14.9	770	54.6
Wet	Base Flow (N = 705)	8	1.1	32	4.5	70	9.9	139	19.7	456	64.7
	High Flow (N = 705)	45	6.4	106	15.0	99	14.0	73	10.4	382	54.2
Dry	Base Flow (N = 705)	3	0.4	15	2.1	47	6.7	137	19.4	503	71.3
	High Flow (N = 705)	29	4.1	68	9.6	83	11.8	137	19.4	388	55.0

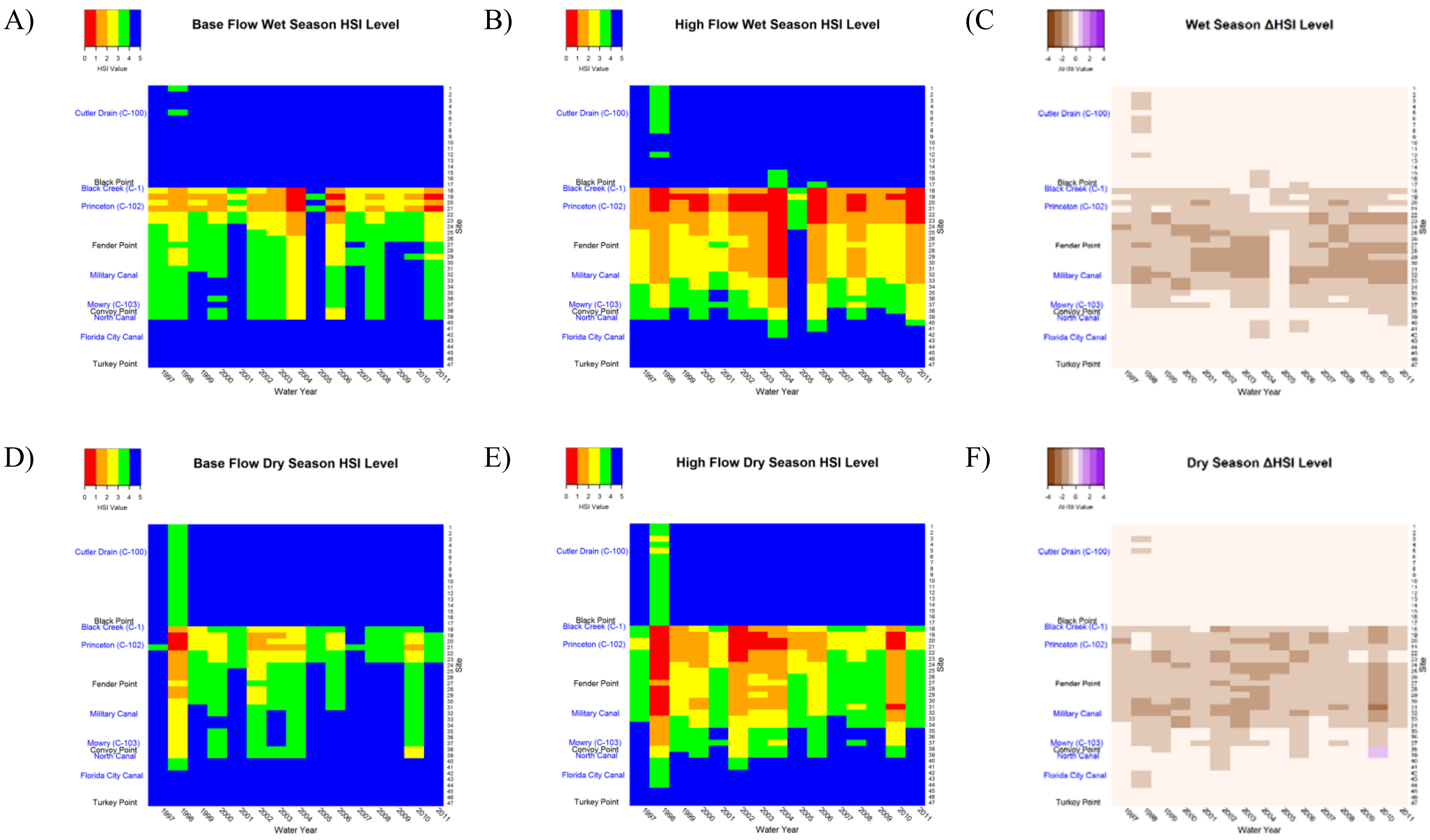


Fig. 5.4: Predicted habitat suitability index (HSI) levels for wet and dry season Base Flow (A, C), High Flow (B, D), and the change between the two model scenarios (C, F), respectively, for water-year and sampling site. Colors in panels A, B, D, and F) represent five habitat suitability levels: optimal (1.0-0.8 = 5), good (0.8-0.6 = 4), fair (0.6-0.4 = 3), poor (0.4-0.2 = 2), and critical (0.2-0.0 = 1) while in panels C) and F) darker shading of brown and purple represent increasingly negative and positive changes in HSI value, respectively.

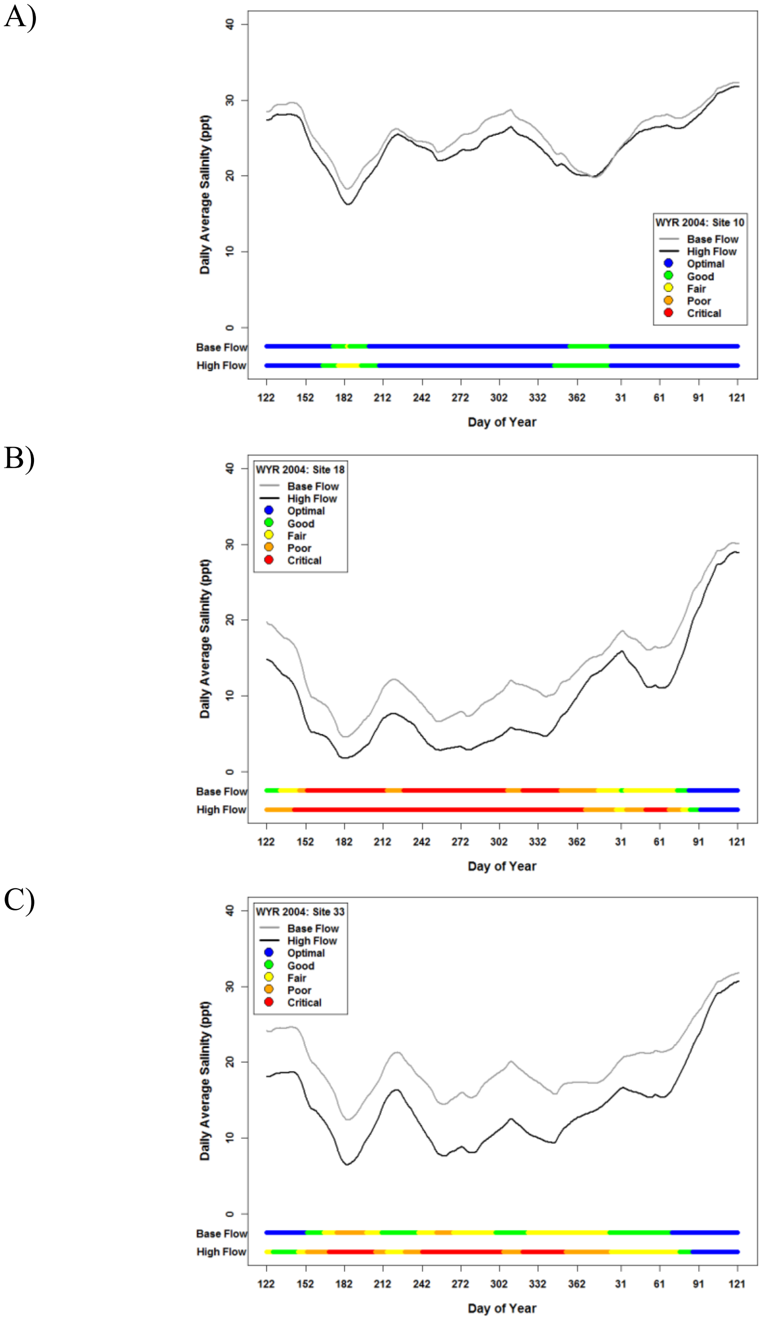


Fig. 5.5: Daily time series of simulated 35 d antecedent mean salinity (ppt) values from Base and High Flow model scenarios for water-year (WYR) 2004 at selected sampling sites: A) Site 10, B) Site 18, and C) Site 33. Points just above the x axis depict daily habitat suitability index (HSI) values for Base and High Flow model scenarios, respectively, predicted for respective salinity conditions. Colors represent five habitat suitability levels: optimal (1.0-0.8), good (0.8-0.6), fair (0.6-0.4), poor (0.4-0.2), and critical (0.2-0.0).

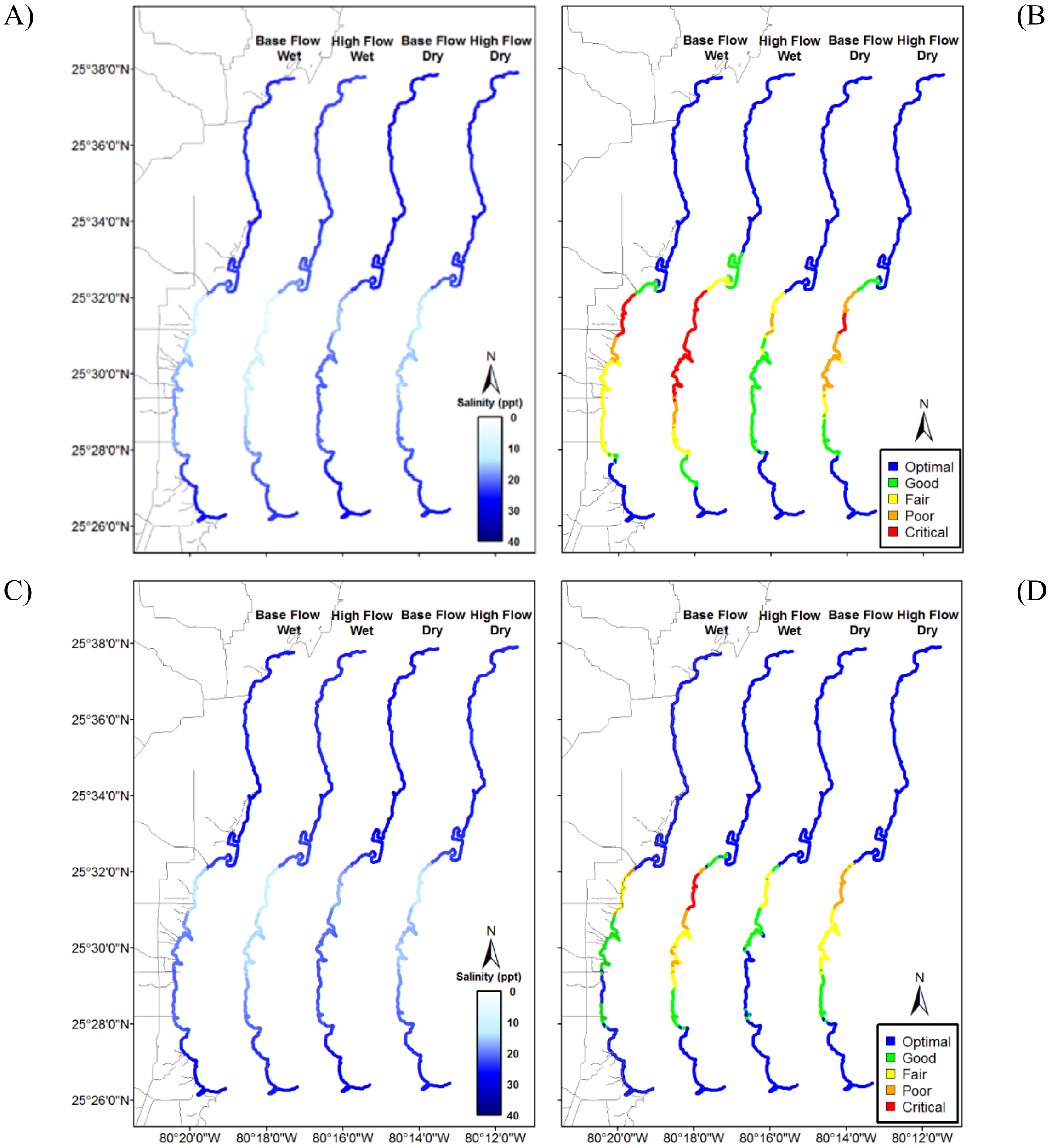
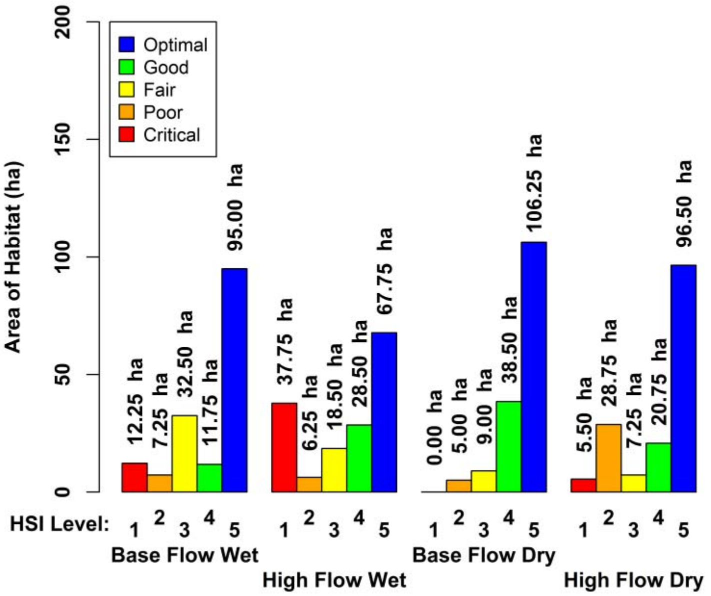


Fig. 5.6: Maps depicting simulated 35 d antecedent mean salinity and habitat suitability index levels (HSI) along the southwestern Biscayne Bay shoreline for each season and freshwater flow model scenario at the 635 interpolated points for A, B) water-year (WYR) 2004 and C, D) Average WYR. Colors in C and D represent five habitat suitability levels: optimal (1.0-0.8), good (0.8-0.6), fair (0.6-0.4), poor (0.4-0.2), and critical (0.2-0.0). Canals and creeks are represented by grey lines interior of the shoreline. Color ramp of A) and C) are the same as those used in Fig. A2.

A)



B)

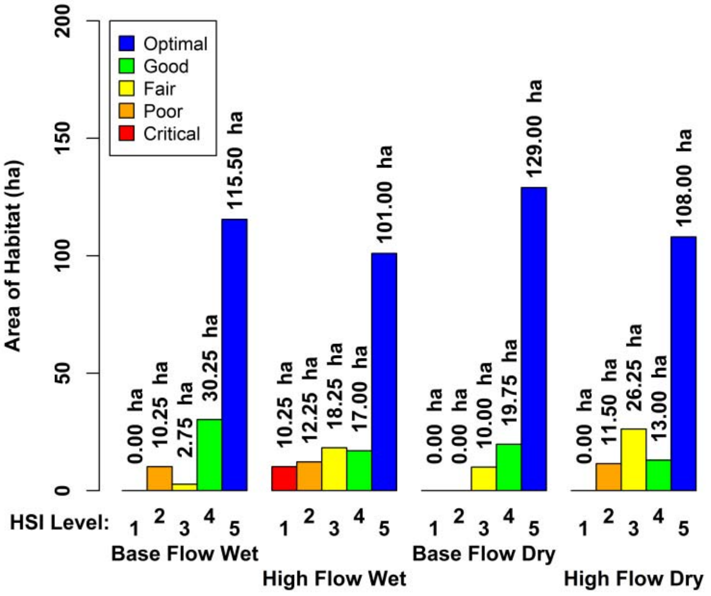


Fig. 5.7: Histograms depicting distribution of areal extent (ha) of each of the five habitat suitability levels optimal (1.0-0.8), good (0.8-0.6), fair (0.6-0.4), poor (0.4-0.2), and critical (0.2-0.0) between Base and High Flow model scenarios and seasons for A) water-year (WYR) 2004 and B) Average WYR available from the hydrodynamic model (1997-2011). Values above bars indicate areal values (ha) for each bar.

Chapter 6: Dissertation Key Findings, Conclusions, Future Research, and Management Implications

6.1 Background

This dissertation was initiated to investigate salinity influences on nearshore (≤ 50 m) juvenile pink shrimp productivity. These investigations were motivated by the need to assess impacts of salinity regime alteration on the productivity of pink shrimp, which has been identified as an ecological indicator for Comprehensive Everglades Restoration Plan (CERP) implementation. These studies have immediate management utility by informing resource managers, scientists, and policy advocates involved with CERP implementation. Beyond this immediate use, the findings in these chapters contribute to the broader ecological knowledge of juvenile pink shrimp productivity in nearshore and estuarine areas and how it may be influenced by freshwater management.

6.2 Chapter 2: Review of Nearshore Life Stage Pink Shrimp Essential Fish Habitat Metrics in Relation to Salinity

6.2.1 Key Findings

A thorough review of the scientific literature revealed that nearshore life stage pink shrimp distribution/occurrence, abundance, and survival were generally maximized under polyhaline (18 – 30 ppt) or euhaline (30 – 40 ppt) salinity conditions. A majority of studies (53.3%) agreed that nearshore pink shrimp abundance was highest between ~20 to 35 ppt; however, a number of studies reported abundance-salinity relationships that maximiza at either higher or lower salinity conditions. The only study to measure growth within a sufficient number and broad enough range of salinity treatments found a maximum at 30 ppt; meta-analysis of growth could not be achieved due to inconsistent growth patterns across the few laboratory studies available. Field growth rates relative to salinity condition were not available. Meta-analysis of survival from laboratory studies yielded a maximum at 30 ppt with high survival (>80%) across salinities of ~ 15 to 40 ppt. Few salinity-productivity investigations have been undertaken.

6.2.2 Discussion

Polyhaline and/or euhaline salinities generally maximized nearshore life stage pink shrimp EFH metrics. However, relationships of pink shrimp abundance to salinity regime were not consistent across all studies. While most studies (53.3%) reported maximum abundance between ~20 to 35 ppt, an almost equal number did not corroborate this generalization. The conflicting studies were likely influenced by interaction with benthic habitat conditions (Howe and Wallace 2000, Robblee et al. 2008), ranges of observed salinities during the study (Wakida-Kusunoki et al. 2008, Blanco-Martínez and Pérez-Castañeda 2016), or perhaps lack of thorough investigation of potential functional responses, beyond linear regression (Perez-Castañeda and Defeo 2001, Robblee et al. 2008). Meta-analytic regression of growth versus salinity did not yield a statistically significant result; however, several studies agreed that extreme hypersalinity (>45 ppt) decreased juvenile pink shrimp growth (Browder et al. 1999, 2002; Zink et al. 2013). A more complete understanding of pink shrimp EFH metric-salinity relationships was also limited by a general lack of studies investigating productivity-salinity relationships. Preservation of nearshore polyhaline conditions and elimination of hypersalinity would maximize density, growth, and survival, and thus improve pink shrimp productivity.

6.2.3 Management Implications

Specific Interim Goals for CERP have assumed that hydrologic restoration will increase pink shrimp abundance in both Florida Bay and Biscayne Bay (RECOVER 2005, Browder et al. 2005a). Conditions faced by pink shrimp differ between these two systems. Extreme hypersalinity occurs in Florida Bay, whereas hypersaline conditions develop less frequently in Biscayne Bay. This review supported prior conclusion that reduction in occurrence, severity, and persistence of hypersalinity would benefit juvenile pink shrimp populations (RECOVER 2005, Rudnick et al. 2005, Browder and Robblee 2009). However, the present findings contradicted the presumption that expansion and persistence of estuarine conditions (salinities <20 ppt) in Biscayne Bay would increase juvenile pink shrimp abundance (RECOVER 2005, Browder et al. 2005a).

6.2.4 Future Research

Relationships of juvenile pink shrimp EFH metrics with other habitat attributes, such as species-specific and/or total submerged aquatic vegetation (SAV) cover, have also been postulated as indirect routes by which alteration of salinity regime may influence pink shrimp productivity (RECOVER 2005, Browder et al. 2005a, Browder and Robblee 2009). A thorough review of pink shrimp-benthic habitat relationships could provide information to either confirm or refute this hypothesis. This knowledge may be especially beneficial given anticipated SAV responses to CERP-related salinity regime alteration (RECOVER 2005).

Review of the salinity habitat requirements of the congeneric pink spotted shrimp *F. brasiliensis* could be helpful. This species has also been reported to occur within Biscayne Bay (Eldred 1960, Browder et al. 2005a, 2005b), where at times it may comprise a substantial proportion (~40%) of the bay's penaeid population (Saloman et al. 1968). Direct comparison of the habitat requirements for *F. duorarum* and *F. brasiliensis* may provide insight as to whether alteration of salinity regimes could produce a species composition shift in Biscayne Bay. Should a species shift occur, it may be possible that the ecological function of penaeids within Biscayne Bay is maintained despite potential negative impacts to *F. duorarum* habitat. However, similar to *F. duorarum* (Bursey and Lane 1971, Criales et al. 2011), survival of euhaline-acclimated early postlarval *F. brasiliensis* was substantially reduced when they were abruptly exposed to salinities ≤10 ppt (Brito et al. 2000). Maximal growth appears to occur at salinities greater than the isosmotic points for both *F. duorarum* (isosmotic point ~28.9 ppt: Bursey and Lane 1971; maximal growth ~30 ppt: Browder et al. 2002) and *F. brasiliensis* (isosmotic point ~ 25 ppt; maximal growth 35 ppt: Brito et al. 2000). With an optimal growth at salinities greater than even *F. duorarum*, reduced salinities are unlikely to benefit *F. brasiliensis*.

6.3 Chapter 3: Spatial and Temporal Patterns in Biscayne Bay Juvenile Pink Shrimp Density

6.3.1 Key Findings

Nearshore juvenile pink shrimp density was limited by four benthic and water column habitat attributes: temperature, water depth, salinity, and total SAV % cover. Cluster analysis of spatial density patterns yielded high-, moderate-, and low-density groupings: moderate densities were generally observed within the "canal zone" between

Black Point and Convoy Point, and sampling sites exhibiting “low” density patterns were even closer to canal or natural creek mouths. “Higher” density sampling sites were located further from canals in the extreme north and south of the sampling domain. On average, pink shrimp densities were higher during the dry-season (i.e., late winter and spring). However, clustering of inter-annual density patterns identified one large group comprised of most year-seasons, indicating low inter-seasonal or inter-annual pink shrimp density variability. Temporal clusters that differed from the main group mostly consisted of wet season sampling events; one in particular was marked by a microalgal bloom within Biscayne Bay.

6.3.2 Discussion

Quantile regression of southwestern Biscayne Bay nearshore juvenile pink shrimp densities from 10 years of monitoring observations detected limitation of potential maximum densities by temperature, salinity, total SAV % cover, and water depth. The salinity-density relationship was best described by a log-linear functional response with salinities ≤~10 ppt most limiting to pink shrimp densities. Inability to detect the anticipated dome-shaped functional response seen in other studies was likely related to few high salinity (>35 ppt) and no hypersaline-pink shrimp density observations.

Spatial clustering of nearshore juvenile pink shrimp densities revealed low and moderate density patterns within the “canal zone” of the southwestern Biscayne Bay coast, an area marked by lower and more variable salinities. Conversely, consistently higher densities were observed along the shoreline north of Black Point and in the southern sampling domain, areas that exhibited higher and more stable salinity conditions. Similar Biscayne Bay pink shrimp spatial density patterns were previously reported by shorter duration studies in similar (i.e., likely overlapping) spatial domains (Diaz 2001, Santos 2014). However, a spatial difference in shrimp size distributions suggested higher recruitment, or other factors, may influence pink shrimp abundance in the northern most stations.

Despite these broad spatial patterns, within-spatial-cluster median density was not significantly correlated with cluster minimum, standard deviation, and median salinity. Perhaps temporally averaged salinity within sampling sites may have been an insufficient predictor for spatial cluster-salinity correlation testing since Procrustean analysis found a significant one-to-one relationship between salinity at the time of sampling and pink shrimp density. Alternatively, interaction with finer scale habitat conditions, such as limitation by SAV cover, may have complicated the spatial trend and prevented correlation from being detected.

Temporal clustering identified three low shrimp density groupings comprised of a few year-seasons, most of which were wet seasons. The 4th temporal cluster exhibited higher densities and was mostly comprised of dry seasons. The shrimp size (5.53 mm CL) associated with the maximal shrimp size distribution difference revealed higher abundance of small juveniles in the dry season, which suggested more recent recruitment in that season. A microalgal bloom during the wet season of 2013 corresponded with one single year-season temporal cluster, which may be indicative of poor habitat conditions that caused increased mortality or avoidance of the sampling domain by pink shrimp.

6.3.3 Management Implications

Pre-CERP-implementation juvenile pink shrimp spatial density patterns along the southwestern Biscayne Bay nearshore (≤ 50 m) between Shoal Point and Turkey Point indicated an association with the higher, more stable salinities presently encountered in the area. These conditions currently exist mainly north and south of a stretch of coastline becoming known as "the canal-zone" because it is bracketed by two major water management canals, C-1 (Black Creek) and C-103 (Mowry). Conversely, lower (<10 ppt) and more variable salinities occurring in the canal-zone limited juvenile pink shrimp density. These results indicate that further salinity reductions in these areas would negatively impact pink shrimp densities. This conclusion is counter to current predictions that salinity reductions via hydrologic restoration would benefit Biscayne Bay pink shrimp densities (RECOVER 2005, Browder et al. 2005a). However, this analysis was not able to address how reduction or elimination of hypersaline conditions in Biscayne Bay by salinity regime restoration could benefit pink shrimp density. Further, these results may not be applicable to pink shrimp density-salinity relationships in nearshore areas further from the coast (>50 m). Observation of generally higher pink shrimp densities in the dry season, as well as apparent more recent recruitment in the dry season, suggest that salinity conditions in the late wet season and dry season would exert greater influence on pink shrimp productivity in the study domain.

6.3.4 Further Research

Continued monitoring of pink shrimp densities is necessary to verify reduction in nearshore salinities to mesohaline and/or oligohaline conditions will indeed negatively impact southwestern Biscayne Bay nearshore pink shrimp density. However, another facet of the salinity regime alteration is the reduction in extreme salinity variability events by at least partially redistributing freshwater discharges from point-sources (i.e., canal mouths) to alongshore inflows via rehydration of coastal wetlands. Using linear regression models, Berkeley and Campos (1984) found that salinity variability, rather than mean salinity condition, was retained as a better predicted Biscayne Bay pink shrimp abundance during multivariate regression analysis. However, they also reported a higher correlation coefficient with mean salinity rather than standard deviation of salinity (Berkeley and Campos 1984). Future studies could investigate whether mean salinity, salinity variability, or both factors sufficiently explain Biscayne Bay pink shrimp abundance patterns. However, these investigations would also have to carefully consider the significant correlation between salinity variability and salinity condition, and it is difficult to separate the two in the environment (Lirman et al. 2008a, Appendix A). Continuously collected data from salinity data-loggers could be used to investigate mean and standard deviation of salinity values computed over similar or differing durations of time to determine best predictors of shrimp density. Related manipulative field studies, such as mesocosm experiments, could be used in to investigate extreme salinity reduction event effects on localized pink shrimp growth and survival.

Laboratory experiments in which the two factors, value and variability, are separately controlled, may be necessary to truly investigate the impact of each and both salinity value and variability simultaneously. Indeed, the direct effects of salinity fluctuation on juvenile pink shrimp growth and survival should also be investigated. Moderate amounts of salinity variability (e.g., abrupt changes of 4 to 10 ppt with a periodicity of 4 to 16 d) were found to maximize growth in other penaeids (Mu et al. 2005, Su et al. 2010). The applicability of present pink shrimp density-water depth relationships, as well as density-salinity relationships, should be investigated in areas further than the 50 m nearshore domain examined here.

6.4 Chapter 4: Salinity Effects on Pink Shrimp Molt Process Growth

6.4.1 Key Findings

Observed growth patterns confirmed previously reported sex and salinity effects on juvenile pink shrimp growth in length and weight. Females grew faster than males. Growth exhibited a parabolic relationship with respect to salinity with average daily growth in length and weight peaking at 33 and 34 ppt, respectively. However, the investigation of salinity effects on juvenile pink shrimp molt process growth were compromised by an unknown disease. Small sample sizes and high measurement error also likely affected statistical results.

6.4.2 Discussion

Parabolic relationships between salinity and growth in both length and weight were consistent with the functional response reported by Browder et al. (2002). Suppression of survival at low salinities (≤ 15 ppt) was also consistent with Browder et al. (2002) but contrasted with the high (~80%) survival at 15 ppt reported by Zink et al. (2013), who acclimated test subjects to these salinity conditions during postlarval development. High survival (100%) in hypersaline conditions (45 and 55 ppt salinity treatments) contrasted with previous studies (Browder et al. 1999, 2002; Zink et al. 2013).

Despite the above findings of salinity impacts on growth, the investigation of salinity impacts on intermolt period and growth-per-molt did not yield statistically significant results. The present molt-process growth study likely suffered from low sample size, which resulted in low statistical power and difficulty in detecting salinity effects that have been demonstrated in other penaeid species (Staples and Heales 1991, O'Brien 1994). Measurement error could be reduced by waiting 48 hr after a molting event to measure the molt increment to allow for hardening of the exoskeleton. However, the results of this laboratory study of salinity effects on the juvenile pink shrimp molt growth process was compromised by probable disease conditions that were not investigated.

6.4.3 Management Implications

Growth in juvenile pink shrimp was reduced in salinities ≤15 ppt and ≥45 ppt, while survival was decreased at salinities ≤15 ppt. If southwestern Biscayne Bay nearshore mesohaline and oligohaline salinity targets are achieved, then density of juvenile pink shrimp will be negatively impacted. Presently, the etiologic agent(s) of the disease conditions observed in this study are unknown, although their identity should be found in order to better interpret the experiment's results and investigate potential salinity relationships.

6.4.4 Further Research

Identification of the etiologic agent(s) that affected the test subjects in my study may provide valuable information relevant to freshwater management. Experimental infection of pink shrimp with the etiologic agent(s), combined with short term salinity tolerance trials and longer term growth and survival studies, might identify interactions relevant to freshwater management.

Not all salinities are equal, and ionic composition of south Florida fresh waters (especially groundwater) and receiving waters may influence their effect on pink shrimp growth, survival, and distributions. Inputs of groundwater rich in certain ions may facilitate use by pink shrimp. For example, some marine fishes enter south Florida oligohaline and fresh waters, which is likely due to their ionic composition and, more specifically, calcium concentrations (Odum 1953, Hulet et al. 1967). In salinities ≤30 ppt, penaeids regulate hemolymph calcium and potassium levels higher than ambient concentrations (Dall and Smith 1981, Dall et al. 1990). Variable success of inland penaeid aquaculture has been attributed partly to differing ionic composition of groundwater sources (Saoud et al. 2003; Davis et al. 2005; Prangnell and Fotedar 2005, 2006; Zhu et al. 2006; Roy et al. 2007). Browder and Robblee (2009) suggested that water hardness of freshwaters entering Whitewater Bay from the Everglades system possibly buffer pink shrimp from negative physiological effects of low salinity. Restoration of groundwater discharges along with surface inflows may be needed to best recreate pre-drainage ionic composition in nearshore waters that allow organisms to thrive at lower salinities.

6.5 Chapter 5: Simulated Pink Shrimp Habitat Suitability Under Base and High Freshwater Discharge Scenarios

6.5.1 Key Findings

Nearshore juvenile pink shrimp habitat suitability Base Flow and High Flow simulations incorporated simulated 35 d antecedent mean salinity conditions hindcast by the hydrodynamic Biscayne Bay Simulation Model v4 (BBSMv4) model. Under the Base Flow scenario, the areal extent of wet season good and optimal habitat suitability totaled 91.5% of the simulated spatial domain (i.e., 50 m nearshore area from Shoal Point to Turkey Point). Good and optimal habitat suitability decreased to 74.3% of the wet season spatial domain in the High Flow scenario. These reductions were not so extreme as to make restoration of Biscayne Bay nearshore salinity conditions incompatible with continued use of the area as pink shrimp nursery habitat. However, reduction in habitat suitability was counter to original predictions that salinity regime restoration in the Biscayne Bay's southwestern nearshore areas would benefit pink shrimp abundances there. Salinity conditions along the southwestern mainland Biscayne Bay shoreline (≤ 50 m) during the dry season generally provided better habitat suitability than those of the wet season.

6.5.2 Discussion

The 3-knot spline natural cubic spline quantile regression ultimately selected for application in the habitat suitability simulations yielded a dome-shaped functional response between potential maximal pink shrimp densities and 35 d antecedent mean salinity conditions. This relationship revealed a 35 d antecedent mean salinity optimum at ~ 30 ppt. Therefore, it was not surprising that habitat suitability reductions were predicted under the High Flow model scenario, which was designed to reduce salinity to oligohaline to mesohaline conditions in the nearshore zone. The spatial patterns of habitat suitability--namely the reduced suitability within the canal-zone--also were not surprising given Chapter 3's spatial analysis of pink shrimp densities. While the present analyses were more rigorous due to use of quantitative, statistical relationships to partition habitat suitability, the declines in habitat suitability at salinities <~15 ppt were consistent with previously reported qualitative declines in habitat suitability at salinities <15 ppt (Mulholland 1984). However, these simulations were limited by their spatial extent (50 m nearshore area) and should not necessarily be applied to nearshore areas further from the coast (i.e, >50 m). These simulations were further limited by an inability of the BBSMv4 to recreate hypersaline conditions, and therefore this potential benefit to pink shrimp habitat suitability was not detected. Reduction of hypersalinity could benefit pink shrimp abundances by reducing negative impacts on postlarval pink shrimp recruitment to nearshore areas (Criales et al. 2010). However, benefits of increased recruitment cannot presently be separated from post-settlement growth and survival processes given the current sampling design. Stronger positive salinity gradients engendered by more freshwater inflow might even attract greater recruitment of available postlarval shrimp from coastal waters into the bay, but this possibility has not been investigated and is outside the realm of this study.

Browder and Robblee (2009) predicted pink shrimp abundance would benefit from stabilization of seagrass communities that reduced patchiness and spatial heterogeneity in cover. Consistent with this prediction, investigations reported in Chapter 3 found limitation of pink shrimp densities at low levels of total SAV % cover.

However, analyses reported on in Chapter 5 did not detect limitation of total SAV % cover by salinity. Therefore, total SAV % cover was not considered in the present simulations. The lack of a salinity response in SAV was likely driven by a transition in dominance between *Halodule wrightii* and *Thalassia testudinum* across salinity conditions; these species often occur in mixed stands at intermediate salinities (Lirman et al. 2014).

6.5.3 Management Implications

The present relationship predicts low habitat suitability for low mesohaline (<~15 ppt) and oligohaline estuarine habitats in nearshore areas (≤50 m) of southwestern Biscayne Bay. Future freshwater management actions that reduce nearshore salinity to these levels would reduce juvenile pink shrimp nearshore habitat suitability in this area.

6.5.4 Further Research

The present BBSMv4 High Flow scenario achieved salinity targets (SFRNC 2006) by increasing freshwater discharges through existing water control structures (Stabenau et al. 2015). These conditions do not reflect spatial redistribution of point-source discharges as restored sheet-flows along the shoreline as intended in Biscayne Bay Coastal Wetlands (BBCW) project construction plans. Improvements to the BBSMv4 that recreate this spatial redistribution of freshwater discharges would improve the realism of habitat suitability and other ecological predictions made from the salinity model. Subtleties in reduction of extreme salinity fluctuations may result in higher predicted habitat suitability values. However, if the redistributions yield oligohaline and/or mesohaline salinity conditions along longer stretches of coastline, then nearshore juvenile pink shrimp habitat suitability may further decline.

Evaporation rates were modeled as an "average" annual trend across a wide time frame (1965-2000) in the BBSMv4 (Stabenau et al. 2015). This generalization did not allow inter-annual variability in evaporation rates, which likely contributed to an inability to recreate empirically observed hypersaline conditions (J. Luo, Pers. Comm.). Improving the implementation of evaporation rates in the BBSMv4 scenarios would add further realism to the model simulations. This could ultimately lead to observation of benefits from increased freshwater discharge to southwestern Biscayne Bay nearshore areas in that reductions of simulated hypersaline conditions would benefit pink shrimp habitat suitability there.

The quantile regression used as the foundation for the habitat suitability predictions relied on antecedent mean salinities calculated for the 35 d period prior to the density observation sampling event. While this time frame seems reasonable given average *in situ* juvenile pink shrimp growth rates, other antecedent durations should be considered. For example, tagging studies could be used to determine shrimp home ranges (Taylor and Ko 2011) and possible avoidance of deleterious salinity conditions. These studies, coupled with assessment of pink shrimp densities before, during, and after an extreme salinity fluctuation event, could indicate whether shorter or longer antecedent durations may better describe patterns in pink shrimp distribution and abundance.

While it was decided not to include SAV influences on pink shrimp density in the present simulations, these investigations were limited by an inability to account for influence of SAV fragmentation on pink shrimp abundance nor salinity influences on

SAV fragmentation. Browder and Robblee (2009) suggested that salinity may indirectly benefit shrimp density via influence of SAV characteristics. Positive influences of increased SAV heterogeneity, which is influenced by salinity variability (Lirman et al. 2008a, Santos et al. 2011), on pink shrimp densities may yet be realized (Santos 2014).

6.6 Conclusions: Pink Shrimp as an Ecological Indicator

The summation of my findings led me to conclude that juvenile pink shrimp abundances will be negatively impacted by restoration of oligohaline and/or mesohaline salinity conditions in southwestern Biscayne Bay nearshore areas. Spatial patterns of pink shrimp density along the southwestern shore of Biscayne Bay indicate areas with lower mean salinity and higher salinity variability are not conducive for maintaining relatively higher shrimp densities. My habitat suitability simulations confirmed this trend by predicting increased spatial extent and degree of deleterious salinity conditions under a model scenario that increased freshwater discharges and decreased nearshore salinity conditions. These dissertation results suggest that Biscayne Bay Interim Goals for salinity and juvenile pink shrimp density, as they are presented stated, and at least when applied to the immediate nearshore area (≤ 50 m), are at odds with each other. There will always be winners and losers when ecosystems are altered, and restoration effects on ecosystems should be considered more broadly than by effects on one or two species.

This “discovery” really represents a circular logic, as my final conclusions echo a conceptual model that depicted selected penaeid species whose placement along a salinity gradient represented their relative reliance on estuarine or oceanic salinity conditions during ontogenetic development (Kutkuhn 1966). Pink shrimp was placed closer to the marine end of this spectrum (Kutkuhn 1966). Overall, my literature review, field, laboratory, and habitat suitability studies of pink shrimp-salinity relationships generally agree with this observation. However, interaction with other habitat conditions could confound this generalization.

The findings of this dissertation were inconsistent with pink shrimp density-salinity regime restoration hypotheses. However, pink shrimp may yet serve as an important ecological indicator of changes in south Florida nearshore salinity conditions. Pink shrimp satisfy many qualities required of “good” ecological indicators: ease in measurement, sensitivity to the anticipated ecological stressors, predicable response to the stressor, and low variability in response (Dale and Beyeler 2001, Carignan and Villard 2002).

Juvenile pink shrimp respond to the stressor (i.e., salinity) in a predicable manner. The presently observed density limitation by salinity quantile regression yielded a dome-shaped (i.e., 3-knot natural spline function) response curve that clearly indicated density changes across the range of stressor conditions (i.e., 35 d antecedent mean salinities ranging from mesohaline to hypersaline conditions for Biscayne Bay). Optimal habitat suitability was predicted for salinities ranging from 21.77 to 38.68 ppt, a difference of 16.91 ppt that represented 47.1% of the total range of 35 d antecedent salinities observed (6.46 to 42.37 ppt). This suggests a low sensitivity response to the stressor. However, since CERP salinity goals include reduction of hypersaline conditions and increase of mesohaline conditions, depending upon the specific location in question, this range of optimal suitability would be sensitive to reduction or increase in temporal and spatial extent, respectively, of these salinity regimes. Consideration of other south Florida estuaries could lead to inclusion of more extreme salinity conditions which further strengthen this understanding. For example, analysis of pink shrimp density limitation should consider locations such as Whitewater Bay, an ecosystem marked by low-salinity conditions that are often ≤20 ppt (Tabb et al. 1962b), and Florida Bay, where extreme hypersaline conditions often develop (Kelble et al. 2007).

From a practicality point of view, monitoring pink shrimp densities has not proven difficult: the present sampling program may require as little as 2 weeks of field effort annually to collect the epifaunal samples at 47 regularly monitored sites over the course of the year (wet and dry seasons). These can be sorted, identified and catalogued by diligent efforts of one full time employee, although in some instances more effort may be needed to process the samples. Expansion of sampling efforts in areas further offshore (i.e., up to 500m from the coast) of the immediate sampling domain would substantially increase field and laboratory efforts. Field sampling may also be complicated by a necessity to use compressed air by throw-trappers in order to clear throwtraps deployed in deeper waters.

Ecological indicator population variations caused by unrelated phenomena should, at a minimum, be distinguishable from those induced by the focal stressor of interest (Carignan and Villard 2002). Juvenile pink shrimp densities are influenced by postlarval supply and recruitment, which depends on stochastic events external to juvenile habitats. However, temporal clustering analyses revealed low inter-annual differences in patterns of pink shrimp density within Biscayne Bay nearshore areas (≤50 m), suggesting that recruitment variability is not a major factor influencing results. Inter-annual consistency in pink shrimp density in other south Florida estuaries should be investigated

This is not to say that pink shrimp would be a good indicator of hydrologic restoration across all south Florida nearshore systems likely to be impacted by CERP implementation (RECOVER 2008). Pink shrimp recruitment may be limited to western Florida Bay (Criales et al. 2010) and thus could not serve as a useful indicator in interior areas of Florida Bay such as Joe Bay and Little Madeira Bay. Pink shrimp would likely not be a good ecological indicator for upper reaches of Shark, Harney, Broad, Lostman's, or Chatham River mangrove systems that are located deeper within the freshwater-estuarine Everglades system ecotone. On the other hand, the Fish and Invertebrate Network Monitoring study results (Robblee and Browder 2010) suggested that pink shrimp were relatively abundant in the Oyster Bay and Whitewater Bay mangrove systems of lower southwest Florida, as well as at the mouth of Lostman's River. Therefore, the use of pink shrimp as an ecological indicator for southwest coast mangrove areas should not be precluded.

Juvenile pink shrimp densities may also not be the best ecological indicator for CERP-related nearshore salinity regime changes. Ideally, multiple indicators should be selected representing various taxa, life histories, and ontogenetic stages in order to ensure a complete view of an ecological system (Dale and Beyeler 2001, Carignan and Villard 2002). Reliance on one or few indicator species can hinder assessment of ecosystem conditions, (Dale and Beyeler 2001, Carignan and Villard 2002), and in the present situation one species cannot reasonably be assumed to indicate changes in salinity regime in all target areas and conditions. Indeed, other nearshore transient and resident species are already being investigated as ecological indicators of nearshore salinity regimes (e.g., Kearney et al. 2015; Lirman et al. 2008a,b, 2011, 2014, 2016; McManus et al. 2014; Serafy et al. 1997, 2003, 2007). Beyond this, the search for community-based ecological indicators within the epifuanal community dataset, which was the basis for these investigations should be continued. Given the ecological and economic importance of pink shrimp (Chapter 1, 2), monitoring of its juvenile populations should be continued.

These results also contributed to a deeper understanding of pink shrimp ecology and will inform scientists and fisheries managers throughout the pink shrimp geographic range of North Carolina to Mexico (Chapter 2). However, and more immediately, it is prudent to continue monitoring juvenile populations of this valuable resource in the face of likely ecological changes caused by freshwater management and climate change. Ultimately, the investigations conducted within this dissertation were limited by the capabilities of one student who did not have the ability to alter previously collected epifauna community monitoring data or hydrologic modeling scenarios, much less acquire the resources needed to consider pink shrimp density patterns in nearshore areas beyond that which was already being sampled. The limitations of the present studies should be recognized as opportunities where further investigation of effects of freshwater management changes on nearshore pink shrimp densities could promptly yield greater understanding.

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Appendix A: Chapter 3 Supplementary Material

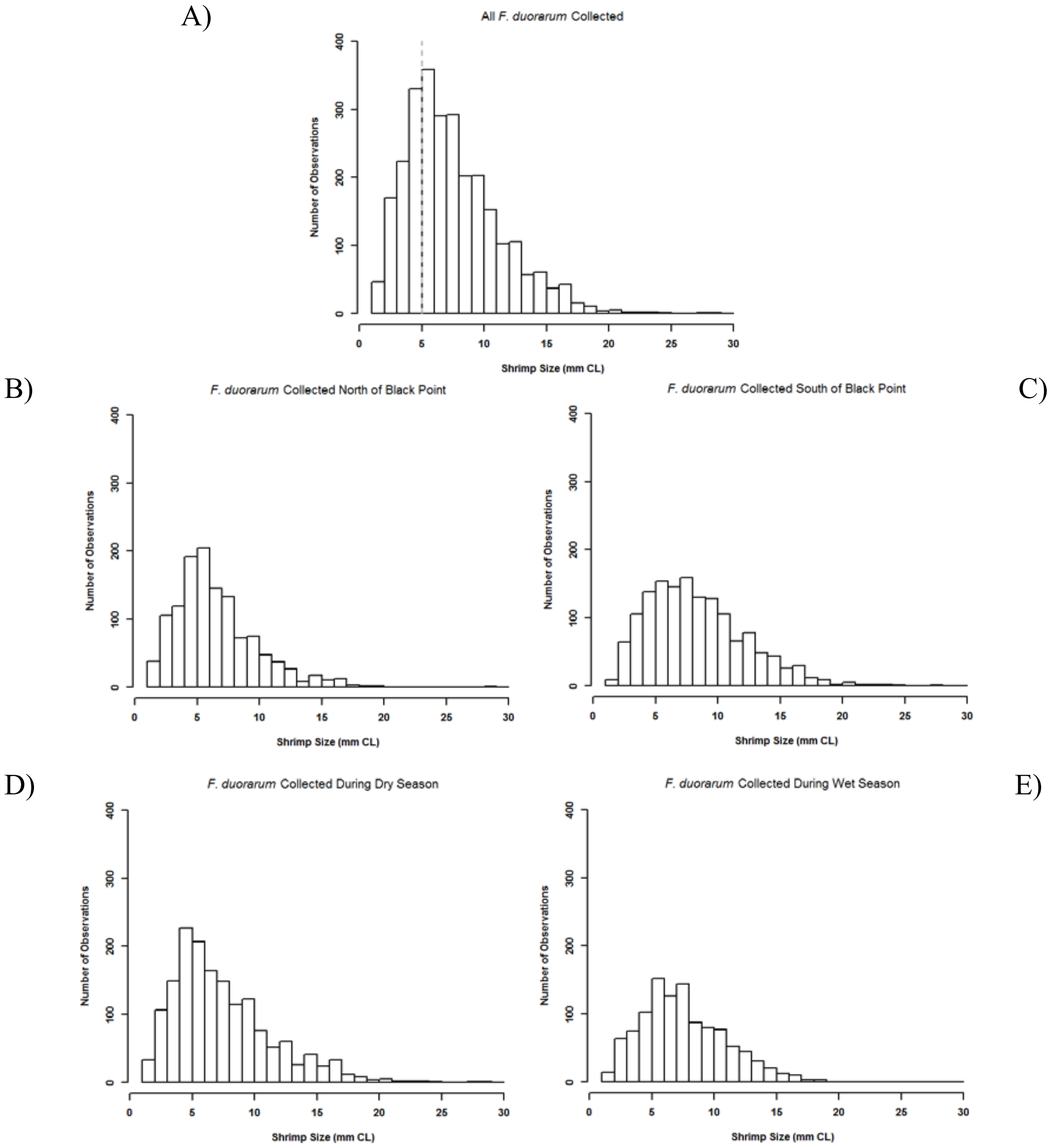

Fig. A1: Histograms depicting size frequencies (mm CL) of A) all *F. duorarum* collected, B) those collected north of Black Point, and C), those collected south of Black Point, D) those collected in the dry season, and E) those collected in the wet season. Vertical dashed line in A) represents removal of smaller shrimps from analysis due to suspected catchability concerns. Shrimp size frequency differences were detected between the two regions ($D_{2\text{-tailed}} = 0.230$, $p < 0.0001$) and between the wet and dry seasons ($D_{2\text{-tailed}} = 0.092$, $p < 0.0001$).

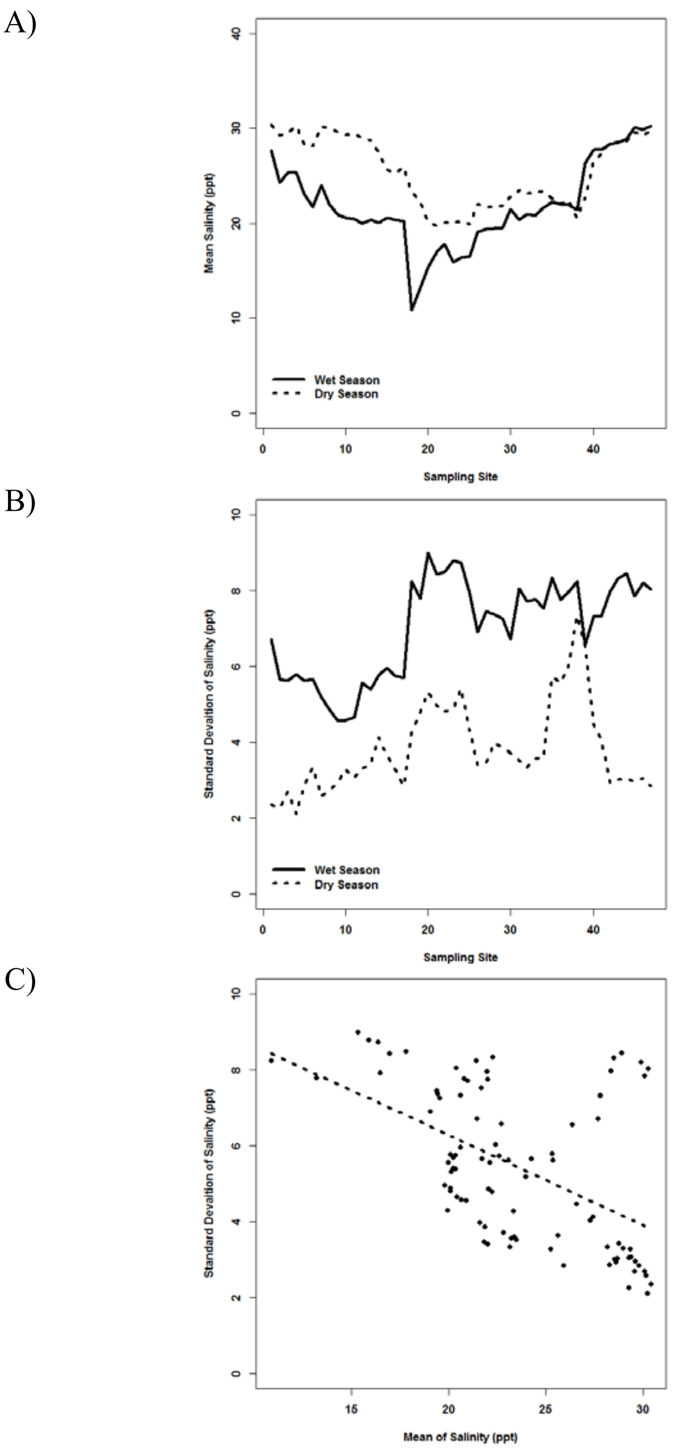

Fig A2: Salinity distributional trends of A) mean salinity (ppt) and B) standard deviation of salinity (ppt) across sampling sites across all year-seasons sampled while C) depicts the scatter of salinity mean and standard deviation values as well as the significant correlation trend line between them.

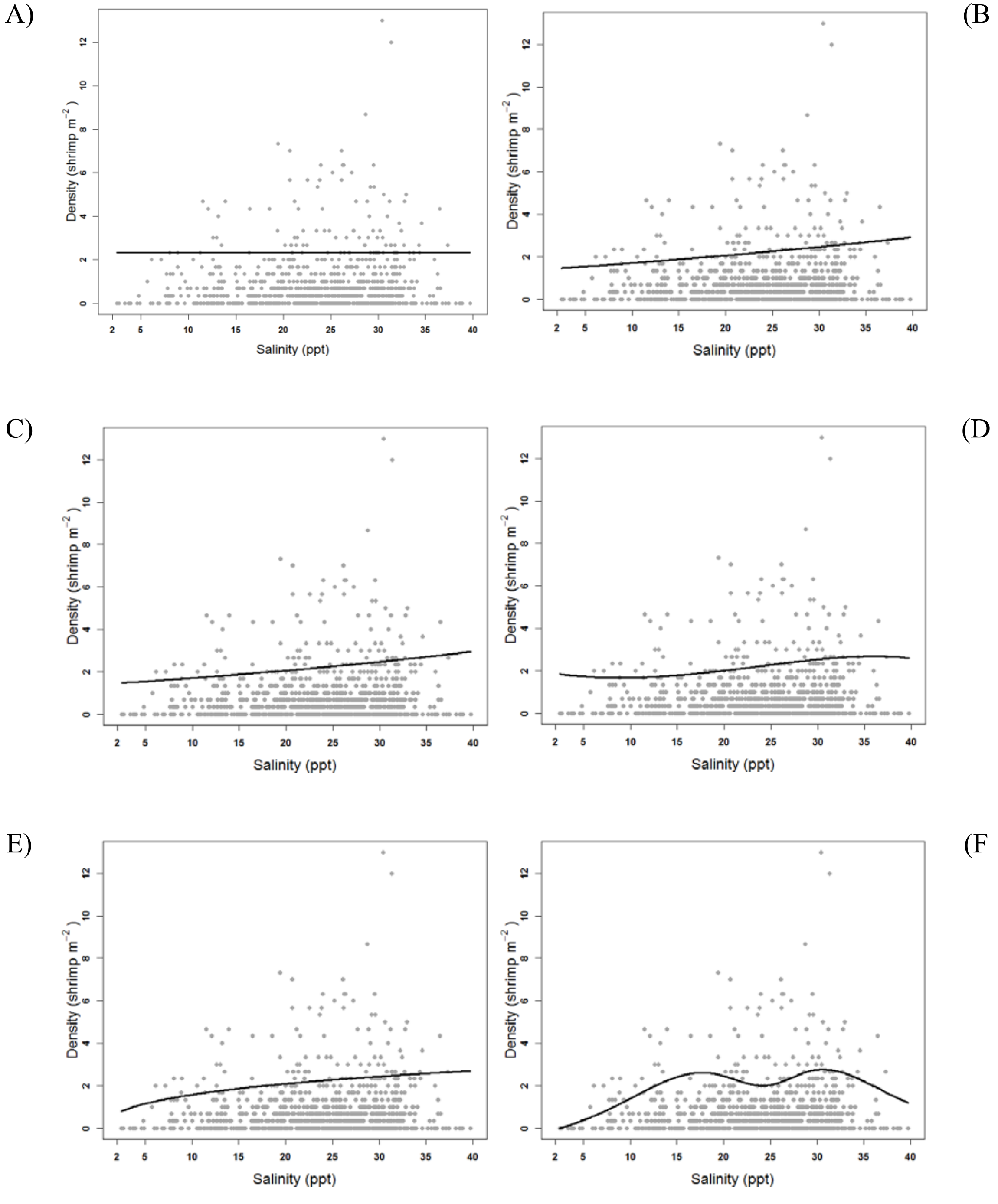

Fig. A3: Plots of functional shape fitting attempted during quantile regression of pink shrimp density (# m^{-2}) against salinity (ppt) for A) null (i.e., intercept only), B) linear, C) quadratic, D) cubic, E) log-linear response curves, and F) 5-knot cubic splines.

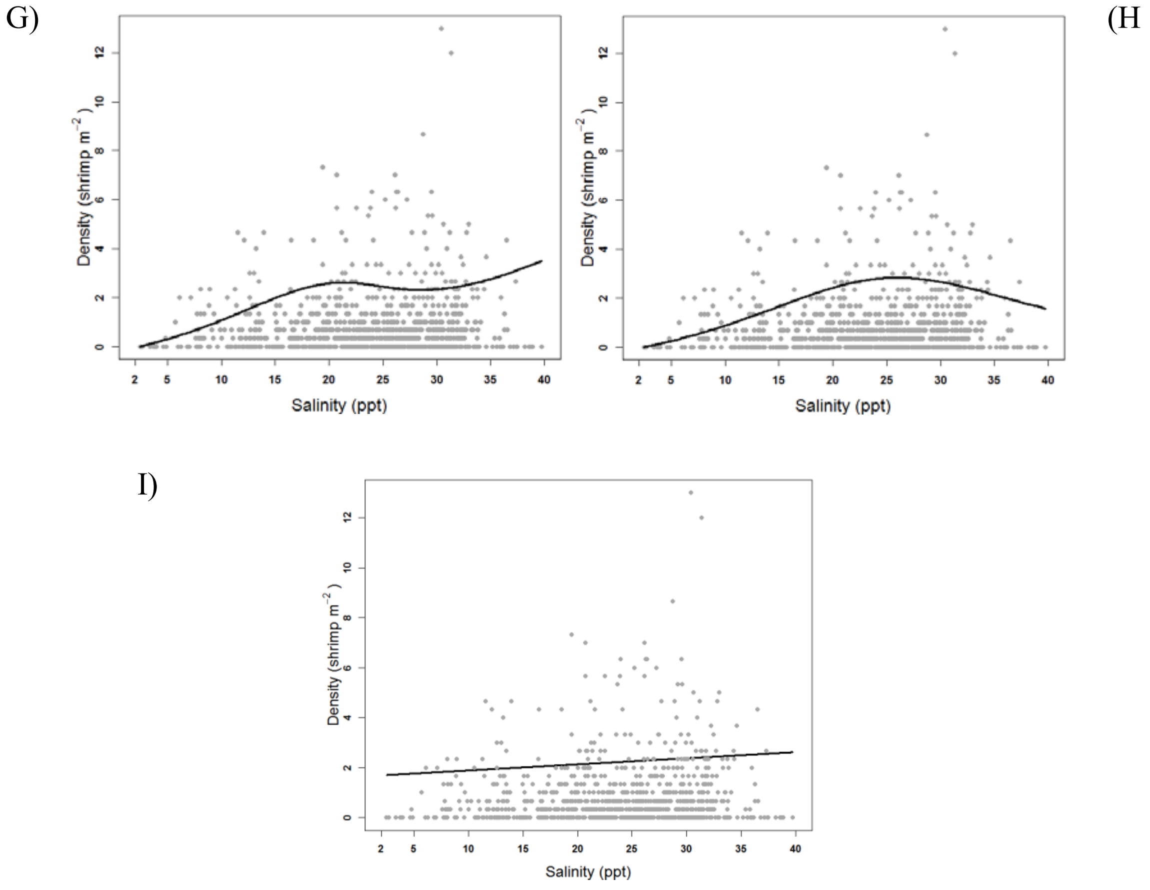

Fig. A3 (cont’d): Plots of functional shape fitting attempted during quantile regression of pink shrimp density (# m^{-2}) against salinity (ppt) for G) 4 knot cubic splines, H) 3-knot cubic splines, and I) additive quantile smoothing spline (AQSS) response curves.

Table A1: Siginficance of parameters used to fit differing functional shapes for quantile regression of pink shrimp density (# m^{-2}) against salinity (ppt) as presented in Fig. A2. Ecological plausibility indicates whether resultant functional shapes (Fig. A2) are reasonable response shapes. AQSS = Additive Quantile Smoothing Spline.

Functional Shape	Parameter	Parameter Values	t value	p value	Ecological Plausibility
Linear	Coefficient 1	0.012 ± 0.11	2.70	0.0071	Yes
	Intercept	0.87 ± 0.11	7.89	0.0000	
Quadratic	Coefficient 2	0.00003 ± 0.00058	0.05	0.9600	No
	Coefficient 1	0.011 ± 0.026	0.44	0.6635	
	Intercept	0.88 ± 0.27	3.24	0.0012	
Cubic	Coefficient 3	-0.00003 ± 0.00008	-0.42	0.6750	No
	Coefficient 2	0.0022 ± 0.0048	0.45	0.6550	
	Coefficient 1	-0.031 ± 0.094	-0.33	0.7424	
	Intercept	1.11 ± 0.55	2.02	0.0436	
Log-Linear	Coefficient 1	0.26 ± 0.091	3.17	0.0016	Yes
	Intercept	0.34 ± 0.28	1.22	0.2213	
Cubic Splines: 5 knots	Coefficient 1	0.97 ± 0.13	7.19	0.0000	Yes
	Coefficient 2	0.95 ± 0.19	5.1	0.0000	
	Coefficient 3	2.55 ± 0.26	9.7	0.0000	
	Coefficient 4	-0.070 ± 0.49	-0.14	0.8876	
Cubic Splines: 4 knots	Coefficient 1	0.55 ± 0.13	2.59	0.0098	No
	Coefficient 2	2.65 ± 0.18	14.88	0.0000	
	Coefficient 3	0.72 ± 0.28	2.59	0.0098	
Cubic Splines: 3 knots	Coefficient 1	2.35 ± 0.081	29.22	0.0000	Yes
	Coefficient 2	0.10 ± 0.17	0.57	0.5711	
AQSS	Intercept	1.70 ± 0.71	2.39	0.0170	Yes
	Lambda	72.93	∞	NA	

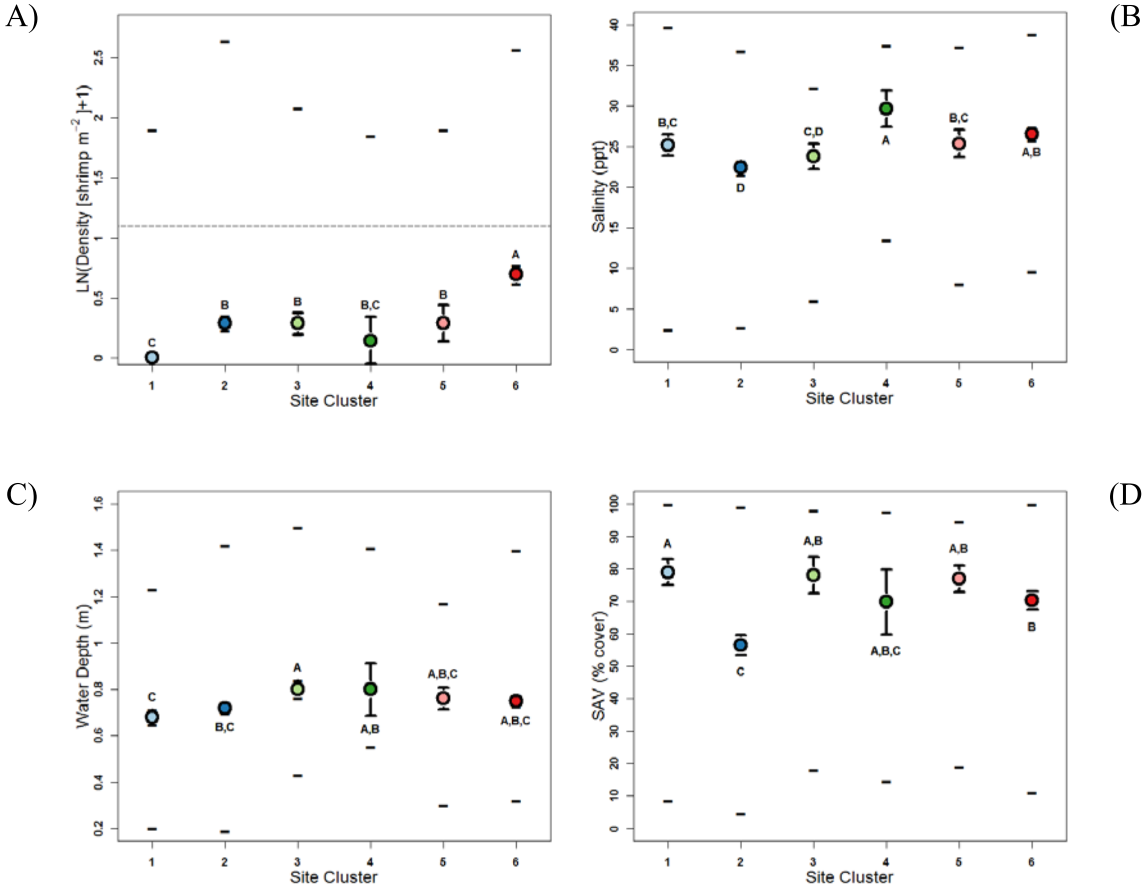

Fig. A3: Median (± CI) and maximum, and minimum values of A) density (shrimp m^{-2}: LN([x+1]), B) salinity (ppt), C) water depth (m), and D) submerged aquatic vegetation (SAV: % cover) relative to shrimp density site clusters. Point colors coincide with Fig. 1 and 3B. Letters denote statistically similar groups. Horizontal line of A) depicts the 2 shrimp m^{-2} CERP Interim Goal.

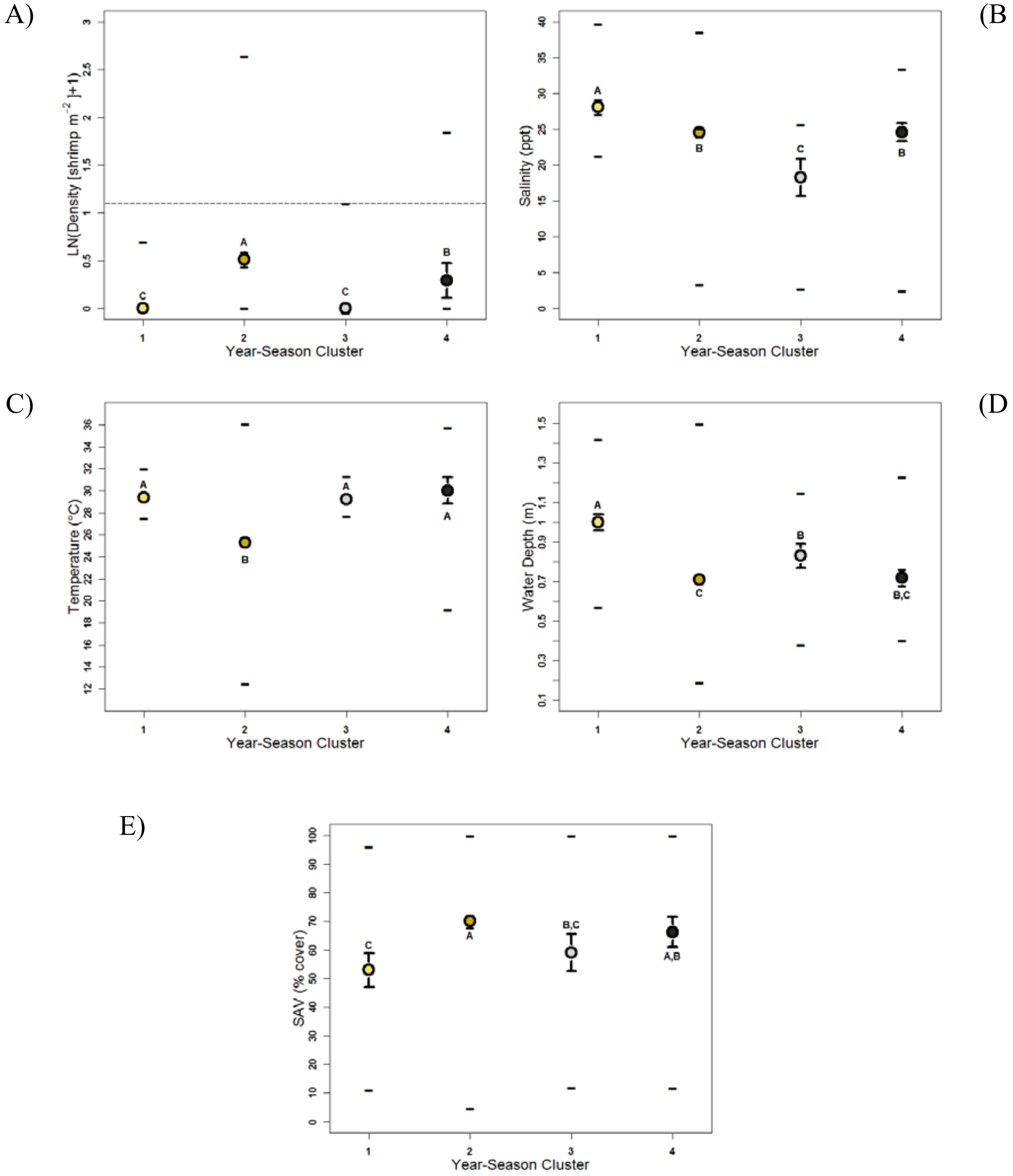

Fig. A4: Median (± CI) and maximum, and minimum values of A) density (shrimp m^{-2}: LN([x+1]), B) temperature (°C), C) salinity (ppt), D) water depth (m), and E) submerged aquatic vegetation (SAV: % cover) relative to shrimp density year-season clusters. Point colors coincide with Fig. 3B. Letters denote statistically similar groups.

Appendix B: Chapter 5 Supplementary Materials

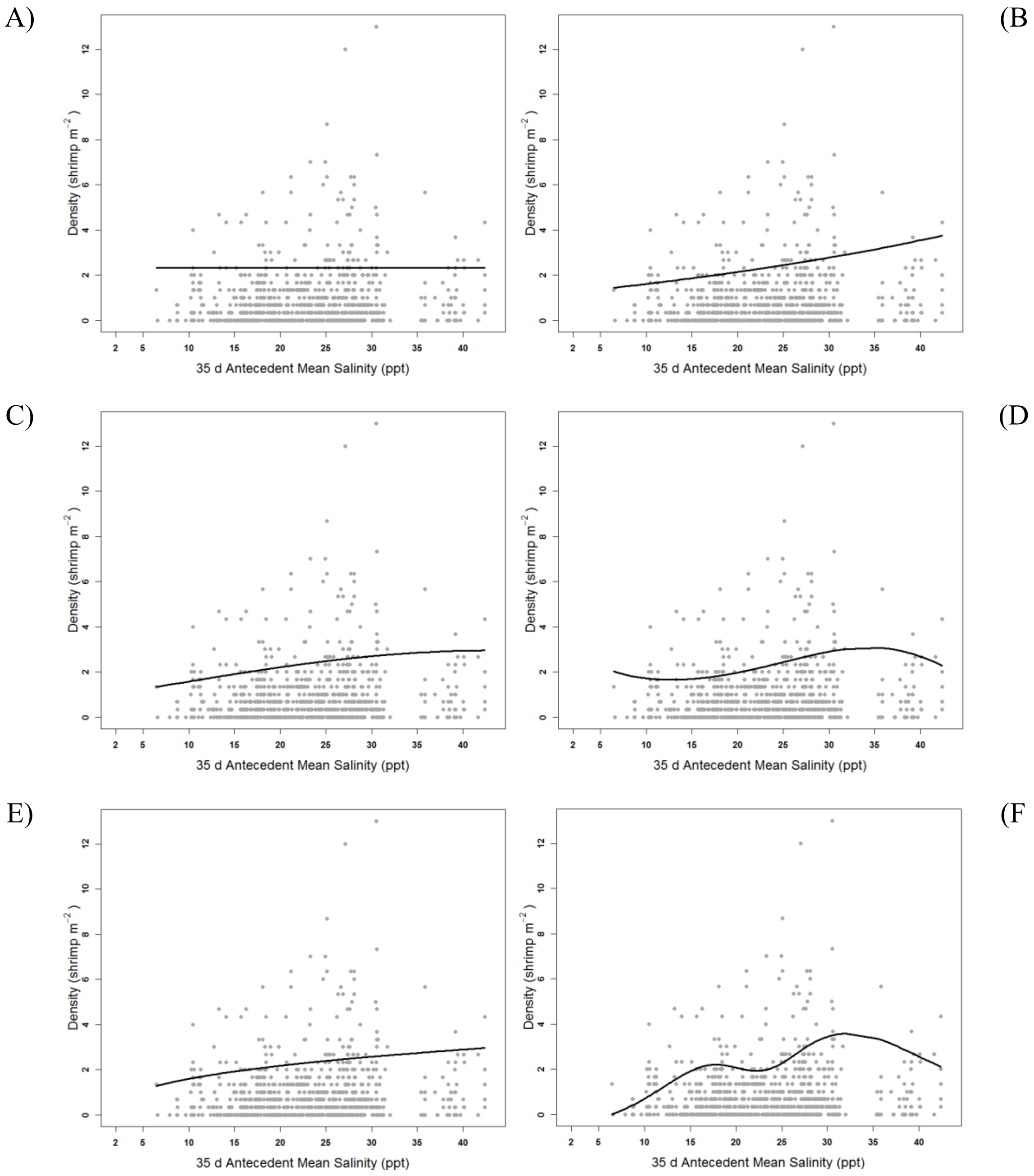

Fig. B1: Plots of functional shape fitting attempted during quantile regression of pink shrimp density (# m^{-2}) against 35 d antecedent mean salinity (ppt) for A) null (i.e., intercept only), B) linear, C) quadratic, D) cubic, E) log-linear response curves, and F) 5-knot cubic splines.

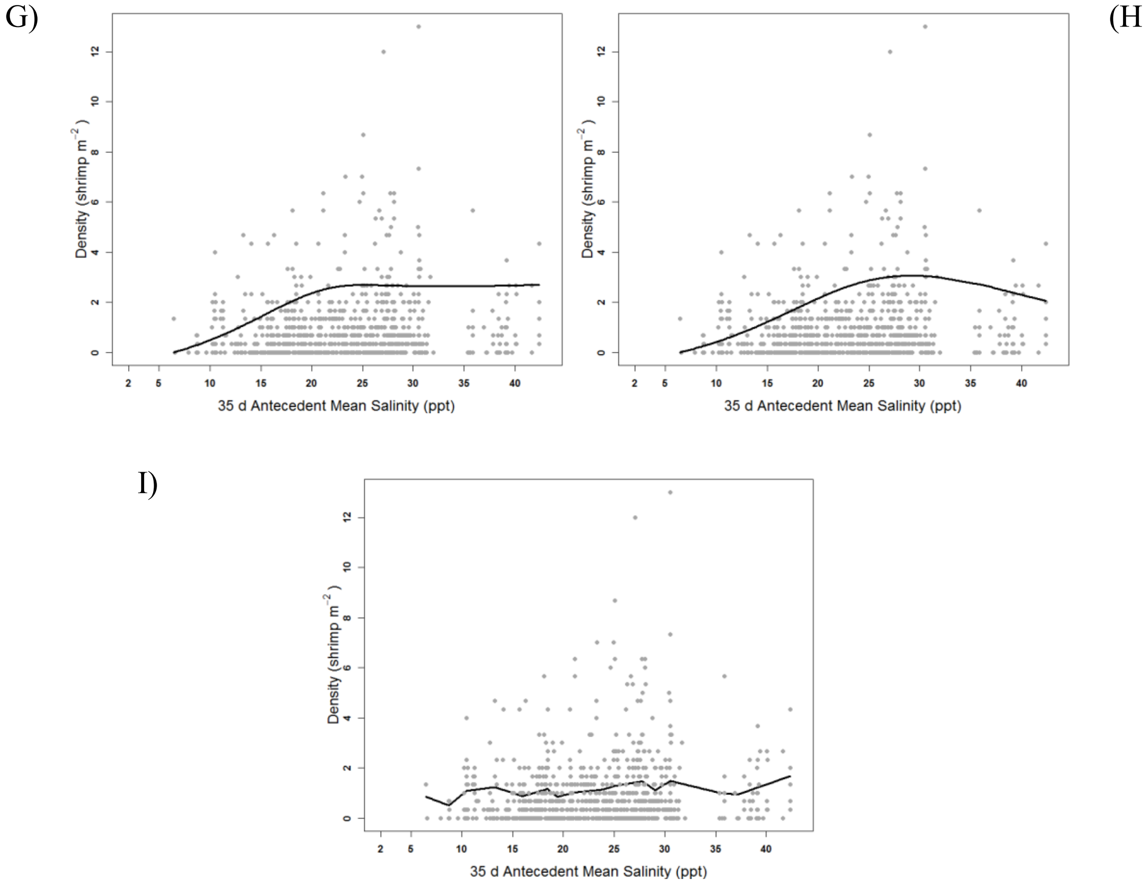

Fig. B1 (cont'd): Plots of functional shape fitting attempted during quantile regression of pink shrimp density (# m^{-2}) against 35 d antecedent mean salinity (ppt) for G) 4 knot cubic splines, H) 3-knot cubic splines, and I) additive quantile smoothing spline (AQSS) response curves.

Table B1: Siginficance of models as well as model fit diagnostics for differing functional shapes of quantile regression (τ = 0.9) of pink shrimp density (# m^{-2}) against 35 d antecedent mean salinity (ppt) as presented in figure above. Ecological plausibility indicates whether resultant functional shapes are reasonable response shapes. AQSS = Additive Quantile Smoothing Spline.

Functional Shape	AIC	ΔAIC	Parameter	Parameter Values	t value	p value	Significant	Ecologically Plausible	Fig. B#
Null	2081.64	84.80	Intercept	1.20 ± 0.045	26.8	<0.0001	Yes	No	2A
Linear	2050.20	53.36	Coefficient 1	0.018 ± 0.0056	3.33	0.0009	Yes	Yes	2B
			Intercept	0.77 ± 0.12	6.30	<0.0001			
Quadratic	2051.34	54.50	Coefficient 2	-0.00039 ± 0.00047	-0.84	0.4011	No	Yes	2C
			Coefficient 1	0.034 ± 0.022	1.53	0.1257			
			Intercept	0.65 ± 0.25	2.58	0.0101			
Cubic	2044.19	47.35	Coefficient 3	-0.00008 ± 0.00006	-1.48	0.1401	No	No	2D
			Coefficient 2	0.0060 ± 0.0043	1.38	0.1669			
			Coefficient 1	-0.11 ± 0.10	-1.11	0.2674			
			Intercept	1.60 ± 0.71	2.24	0.0253			
Log-Linear	2052.11	55.27	Coefficient 1	0.29 ± 0.11	2.79	0.0054	Yes	Yes	2E
			Intercept	0.27 ± 0.32	0.87	0.3818			
Splining: 5 knots	2105.73	138.89	Coefficient 1	0.93 ± 0.11	8.40	<0.0001	Yes	Yes	2F
			Coefficient 2	1.25 ± 0.18	6.95	<0.0001			
			Coefficient 3	2.67 ± 0.22	12.13	<0.0001			
			Coefficient 4	0.42 ± 0.19	2.22	0.0266			

Table B1: Siginficance of models as well as model fit diagnostics for differing functional shapes of quantile regression ($\tau = 0.9$) of pink shrimp density (# m^{-2}) against 35 d antecedent mean salinity (ppt) as presented in figure above. Ecological plausibility indicates whether resultant functional shapes are reasonable response shapes. AQSS = Additive Quantile Smoothing Spline.

Functional Shape	AIC	ΔAIC	Parameter	Parameter Values	t value	p value	Significant	Ecologically Plausible	Fig. B#
Splining: 4 knots	2144.08	177.24	Coefficient 1	0.83 ± 0.14	5.89	<0.0001	Yes	Yes	2G
			Coefficient 2	2.44 ± 0.13	19.08	<0.0001			
			Coefficient 3	0.64 ± 0.17	3.75	0.0002			
Splining: 3 knots	2167.26	200.42	Coefficient 1	2.47 ± 0.087	28.43	<0.0001	Yes	Yes	2H
			Coefficient 2	0.37 ± 0.14	2.58	0.0101			
AQSS	1966.84	0	Intercept	0.85 ± 0.27	3.10	0.0020	Yes	No	2I
			Lambda	1.35	6.5	<0.0001			

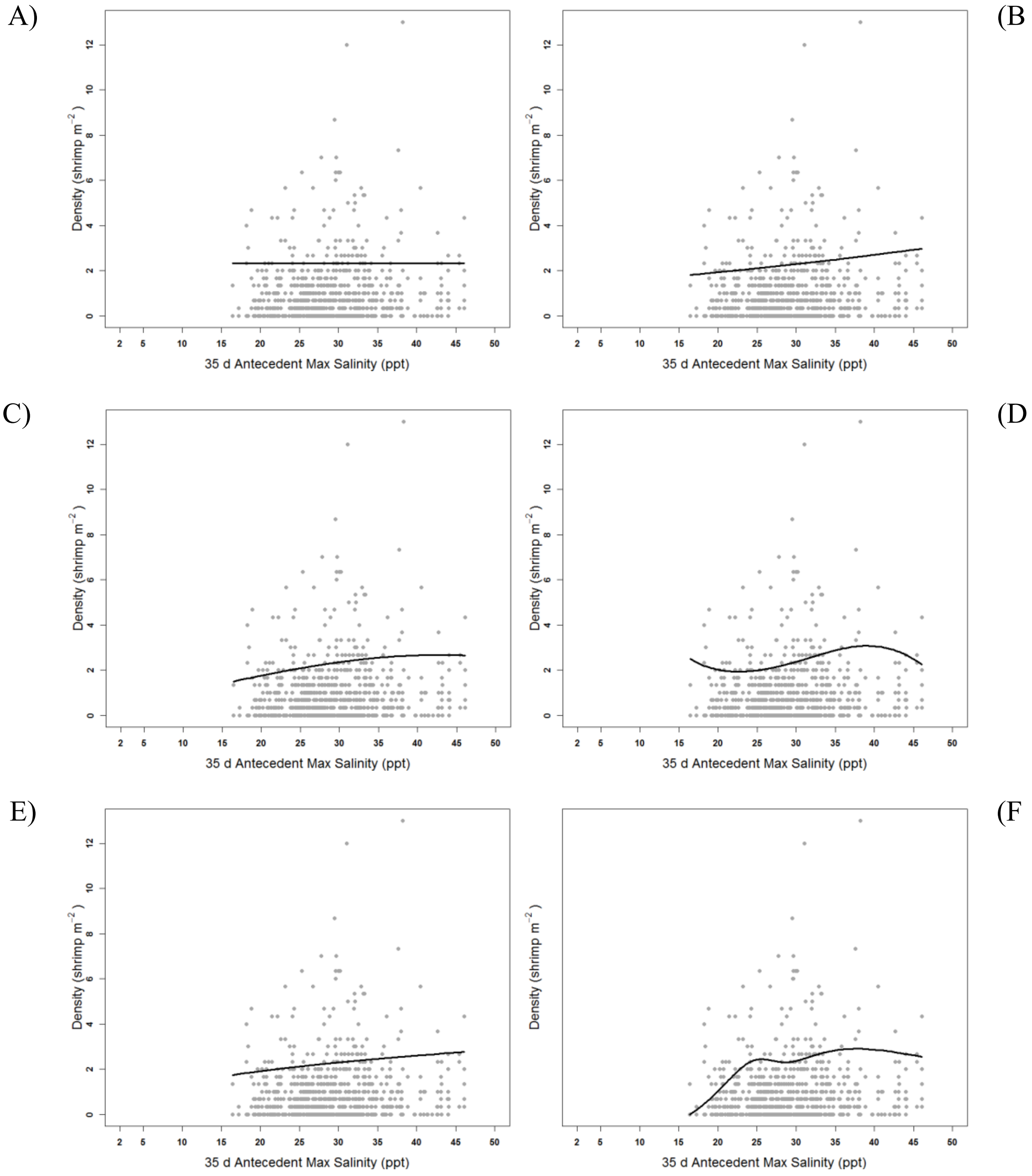

Fig B2: Plots of functional shape fitting attempted during quantile regression of pink shrimp density (# m^{-2}) against 35 d antecedent maximum salinity (ppt) for A) null (i.e., intercept only), B) linear, C) quadratic, D) cubic, E) log-linear response curves, and F) 5-knot cubic splines.

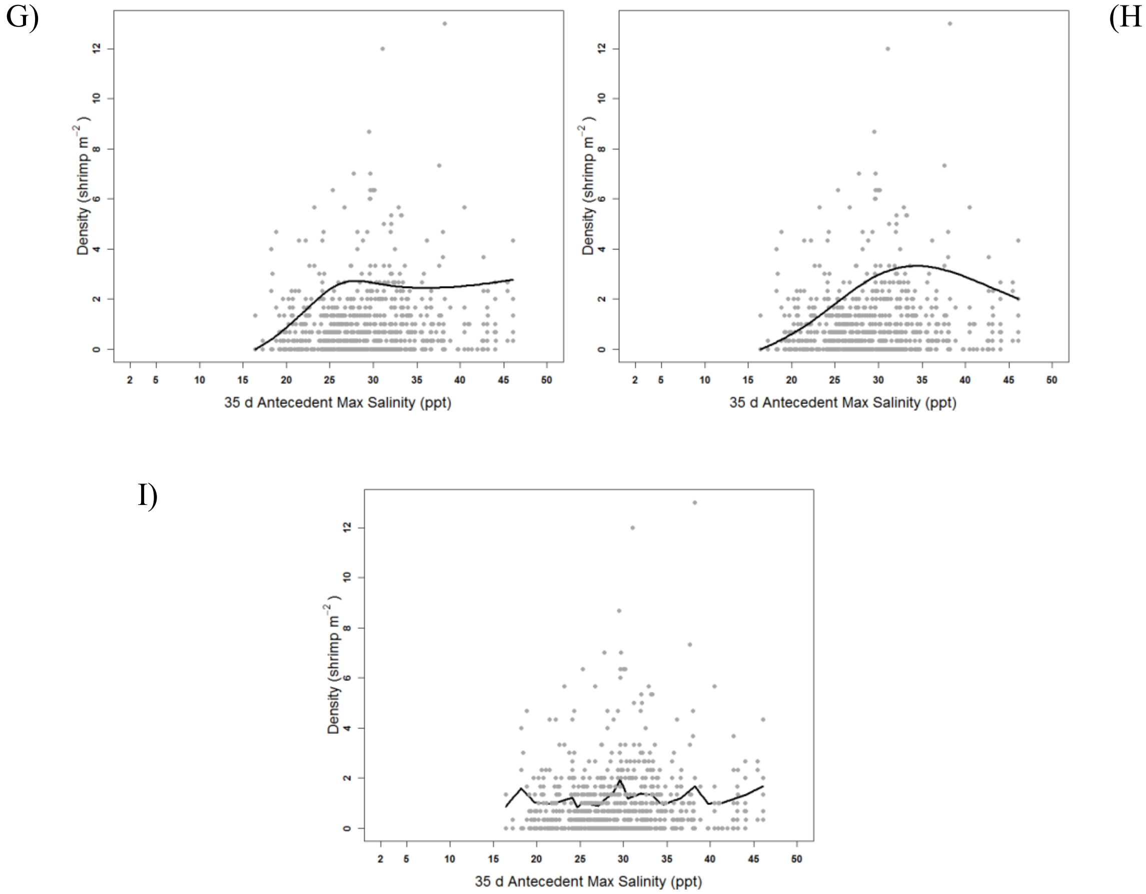

Fig. B2 (cont'd): Plots of functional shape fitting attempted during quantile regression of pink shrimp density (# m^{-2}) against 35 d antecedent maximum salinity (ppt) for G) 4 knot cubic splines, H) 3-knot cubic splines, and I) additive quantile smoothing spline (AQSS) response curves.

Table B2: Siginficance of models as well as model fit diagnostics for differing functional shapes of quantile regression ($\tau = 0.9$) of pink shrimp density (# m^{-2}) against 35 d antecedent maximum salinity (ppt) as presented in figure above. Ecological plausibility indicates whether resultant functional shapes are reasonable response shapes. AQSS = Additive Quantile Smoothing Spline.

Functional Shape	AIC	ΔAIC	Parameter	Parameter Values	t value	p value	Significant	Ecologically Plausible	Fig. B#
Null	2081.64	129.93	Intercept	1.20 ± 0.048	24.9	<0.0001	Yes	No	3A
Linear	2071.71	120.00	Coefficient 1	0.012 ± 0.0070	1.66	0.0970	Yes	Yes	3B
			Intercept	0.84 ±0.21	4.09	0.0001			
Quadratic	2071.06	119.35	Coefficient 2	-0.00055 ± 0.00072	-0.76	0.4460	No	Yes	3C
			Coefficient 1	0.047 ± 0.045	1.04	0.2968			
			Intercept	0.29 ± 0.69	0.42	0.6748			
Cubic	2067.12	115.41	Coefficient 3	-0.00015 ± 0.00011	-1.35	0.1766	No	No	3D
			Coefficient 2	0.014 ± 0.011	1.30	0.19			
			Coefficient 1	-0.39 ± 0.33	-1.20	0.23			
			Intercept	4.64 ± 3.27	1.42	0.16			
Log-Linear	2070.90	1198.19	Coefficient 1	0.31 ± 0.19	1.59	0.11	No	Yes	3E
			Intercept	0.14 ± 0.65	0.22	0.83			
Splining: 5 knots	2155.27	203.56	Coefficient 1	1.13 ± 0.13	8.92	<0.0001	Yes	No	3F
			Coefficient 2	0.91 ± 0.19	4.71	<0.0001			
			Coefficient 3	2.57 ± 0.17	14.89	<0.0001			
			Coefficient 4	0.57 ± 0.8	3.24	0.0006			

Table B2 (cont'd): Siginficance of models as well as model fit diagnostics for differing functional shapes of quantile regression (τ = 0.9) of pink shrimp density (# m^{-2}) against 35 d antecedent maximum salinity (ppt) as presented in figure above. Ecological plausibility indicates whether resultant functional shapes are reasonable response shapes. AQSS = Additive Quantile Smoothing Spline.

Functional Shape	AIC	ΔAIC	Parameter	Parameter Values	t value	p value	Significant	Ecologically Plausible	Fig. B#
Splining: 4 knots	2169.14	217.43	Coefficient 1	0.63 ± 0.13	4.92	<0.0001	Yes	Yes	3G
			Coefficient 2	2.51 ± 0.12	21.58	<0.0001			
			Coefficient 3	0.61 ± 0.18	3.47	<0.0001			
Splining: 3 knots	2231.56	279.85	Coefficient 1	2.57 ± 0.095	27.21	<0.0001	Yes	Yes	3H
			Coefficient 2	0.35 ± 0.18	1.97	0.0494			
AQSS	1951.71	0	Intercept	0.85 ± 0.30	2.82	0.0049	Yes	No	3I
			Lambda	0.65					

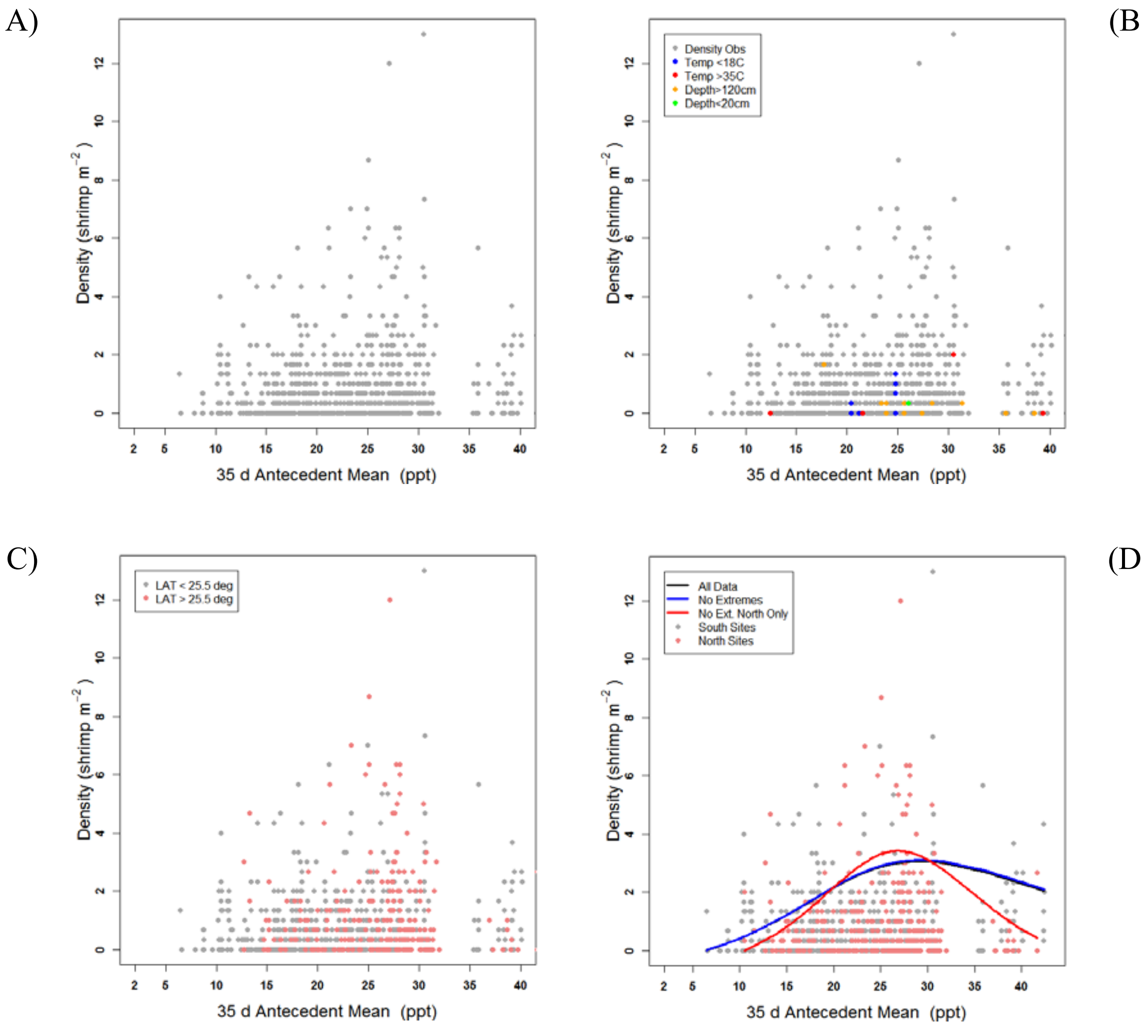

Fig. B3: Plots of pink shrimp density against 35 d antecedent mean salinity (ppt) depicting A) the raw data, B) the raw observations with extreme temperature (°C) and water depth (cm) at the time of sampling highlighted, C) the raw observations with sampling sites north of Black Point highlighted, and D) the raw observations highlighted as in C) but also depicting 3-knot natural cubic spine function quantile regression fitted lines resulting from regression of all data (black line), the removal of the extreme habitat conditions (blue line), and the removal of the southern sampling sites and the extreme habitat conditions (red line).

Table B3: Quantile regression parameter values and statistical significance for 3-knot natural cubic spline functions presented in Fig. A1D.

Data Set	Parameter	Parameter Values	t value	p value
All Data	Coefficient 1	2.47 ± 0.089	27.62	<0.0001
	Coefficient 2	0.37 ± 0.16	2.38	0.0177
Extreme Habitat Conditions Removed	Coefficient 1	2.57 ± 0.10	25.33	<0.0001
	Coefficient 2	0.26 ± 0.15	1.76	0.0795
Northern Sites and Extreme Habitat Conditions Removed	Coefficient 1	2.39 ± 0.20	11.74	<0.0001
	Coefficient 2	-0.60 ± 0.49	-1.24	0.2151

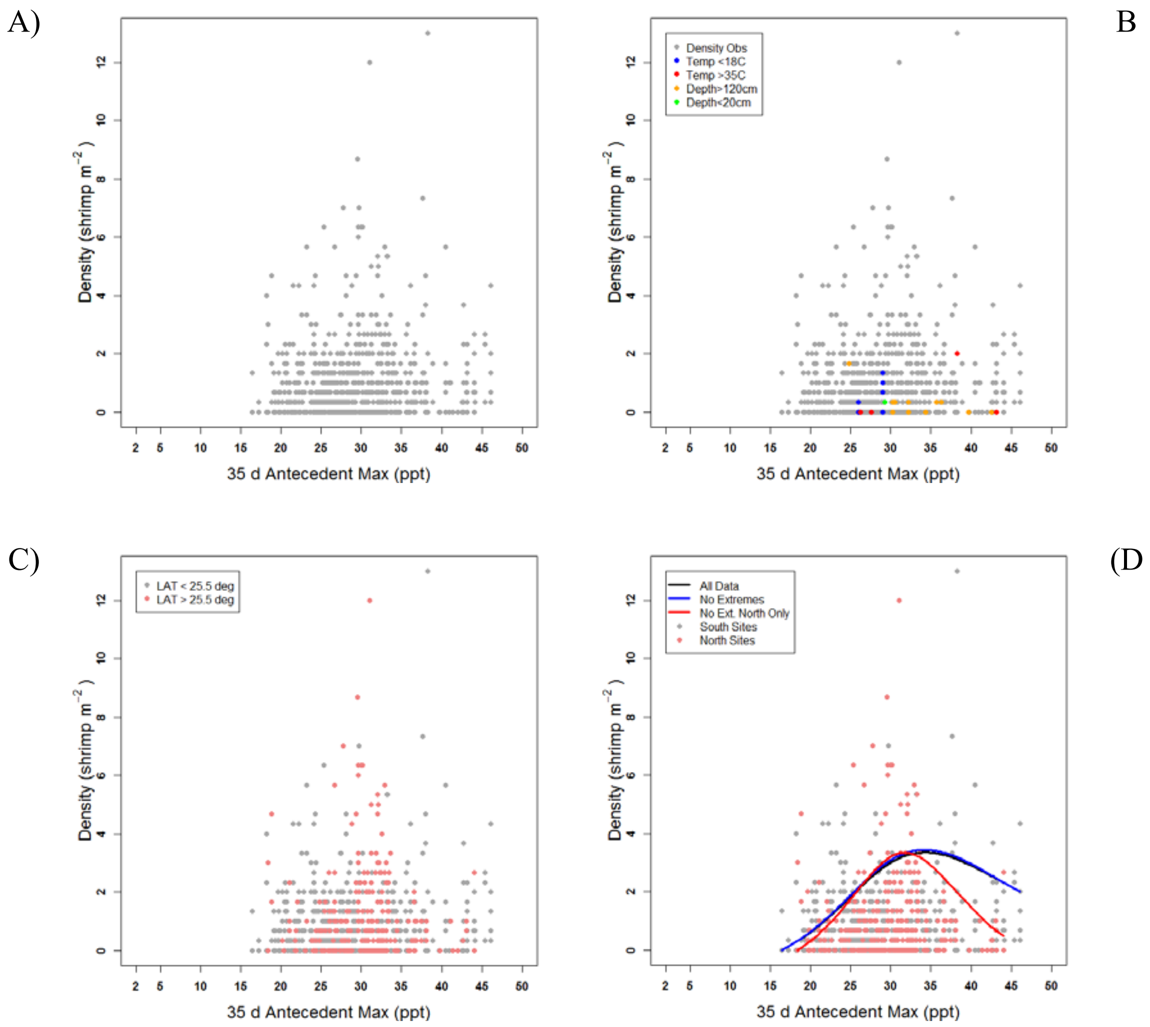

Fig. B4: Plots of pink shrimp density against 35 d antecedent maximum salinity (ppt) depicting A) the raw data, B) the raw observations with extreme temperature (°C) and water depth (cm) at the time of sampling highlighted, C) the raw observations with sampling sites north of Black Point highlighted, and D) the raw observations highlighted as in C) but also depicting 3-knot natural cubic spine function quantile regression fitted lines resulting from regression of all data (black line), the removal of the extreme habitat conditions (blue line), and the removal of the southern sampling sites and the extreme habitat conditions (red line).

Table B4: Quantile regression parameter values and statistical significance for 3-knot natural cubic spline function of 35 d antecedent maximum salinity presented in Fig. A1D.

Data Set	Parameter	Parameter Values	t value	p value
All Data	Coefficient 1	2.57 ± 0.095	27.21	<0.0001
	Coefficient 2	0.35 ± 0.8	1.96	0.0093
Extreme Habitat Conditions Removed	Coefficient 1	2.65 ± 0.11	25.05	<0.0001
	Coefficient 2	0.33 ± 0.17	1.75	0.0080
Northern Sites and Extreme Habitat Conditions Removed	Coefficient 1	2.38 ± 0.20	11.86	<0.0001
	Coefficient 2	-0.53 ± 0.33	-1.61	0.1088

Table B5: Results of HSI model validation via prediction of pink shrimp density (shrimp m^{-2}) via quantile regression (tau = 0.9) against 35 d antecedent mean salinity, density predicted from the HSI model, and predicted HSI value.

Dependent Variable	Predictors	Coefficients (± SE)	t value	p value
Observed Density	Spline 1	2.49 ± 0.11	23.53	<0.0001
	Spline 2	0.57 ± 0.11	4.99	<0.0001
Observed Density	Predicted Density	0.73 ± 0.15	4.88	<0.0001
	Intercept	0.67 ± 0.38	1.76	0.0796
Observed Density	Predicted HSI Value	0.44 ± 0.12	3.83	0.0002
	Intercept	0.56 ± 0.47	1.17	0.2419

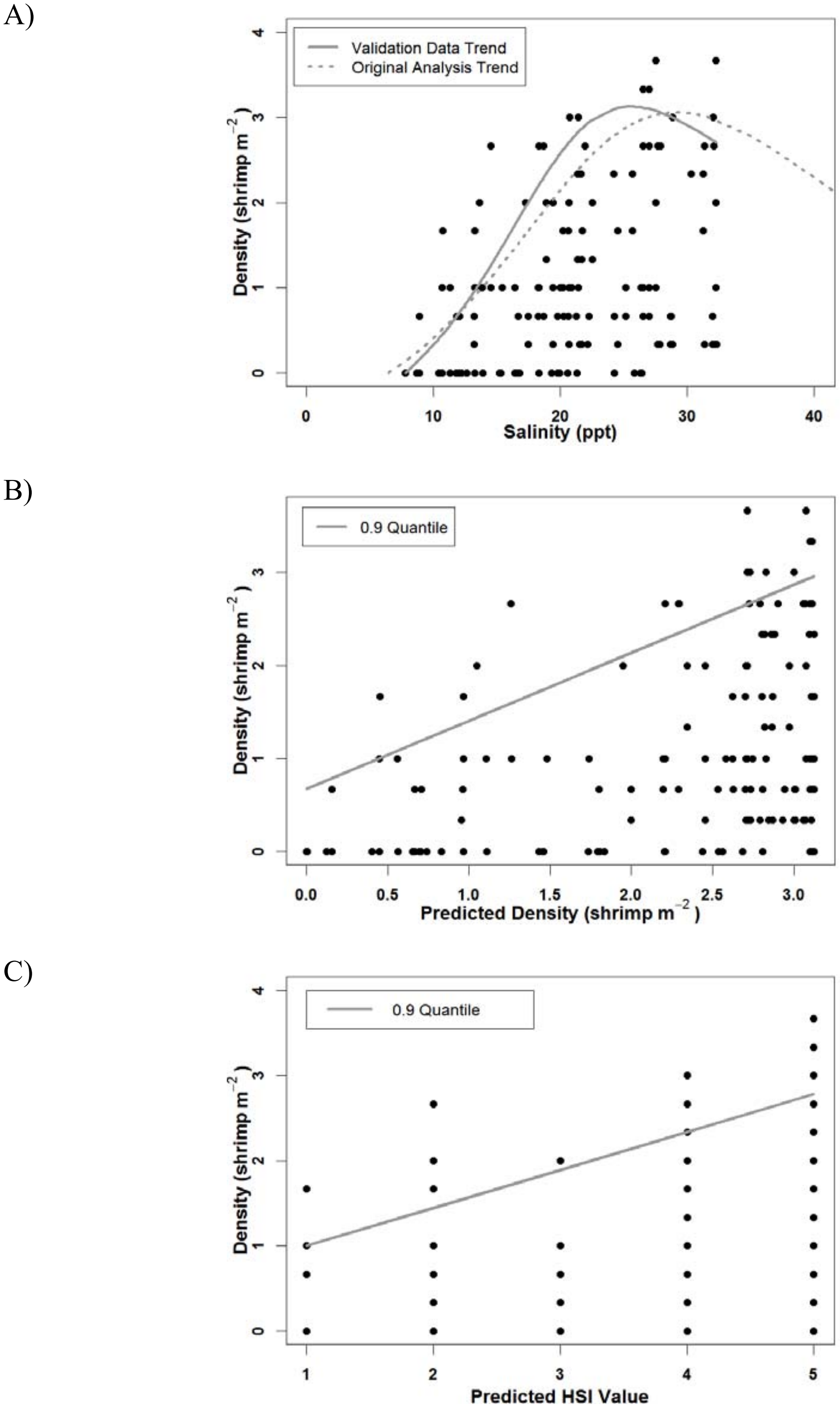

Fig. B5: Calendar year 2005-2006 observed pink shrimp densities (shrimp m^{-2}) plotted against A) observed salinity (ppt), B) density predicted from the log-linear HSI salinity-density, and C) predicted HSI values. Curves in figure A) represent log-linear responses of density to salinity while lines in B) and C) represent 0.9 quantile regression relationships. See text for details.

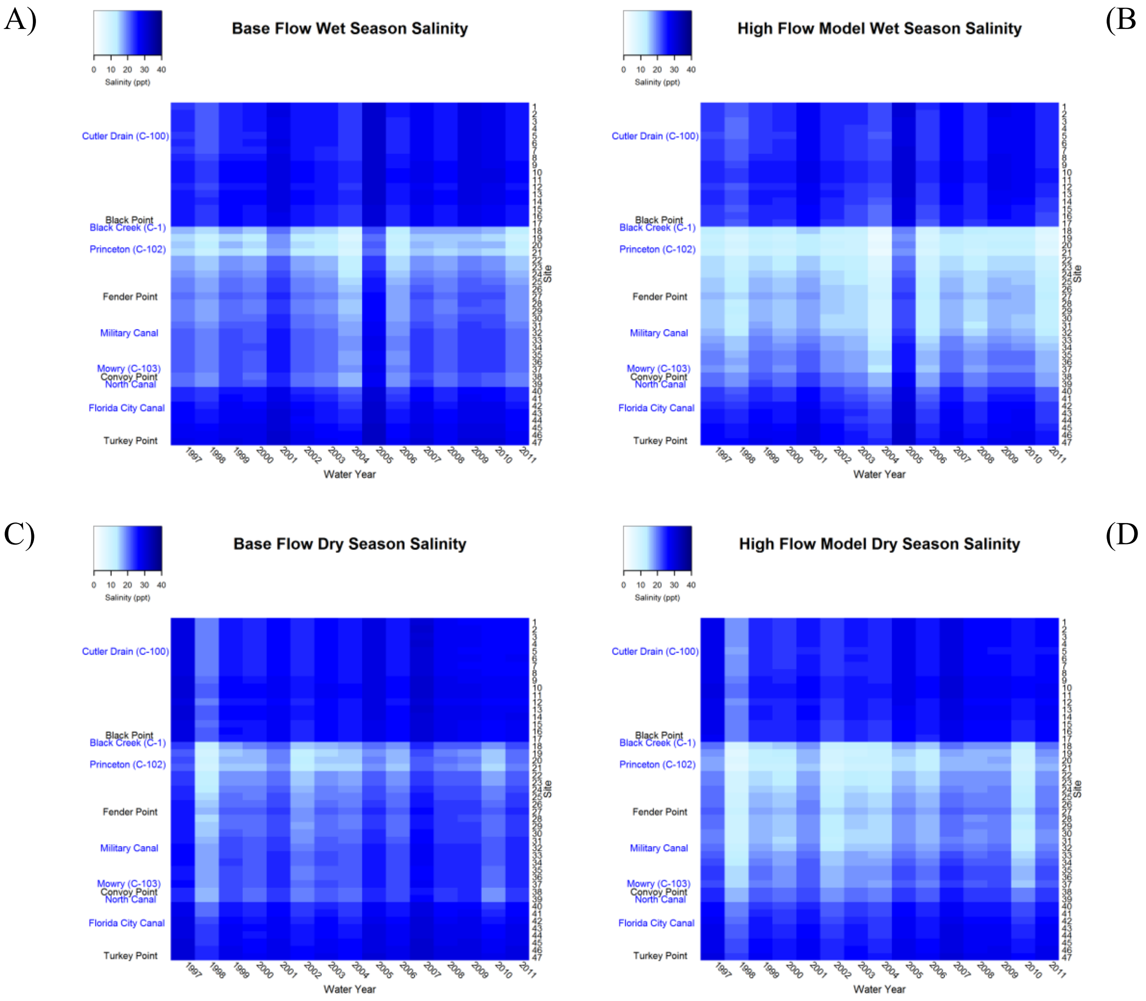

Fig. B6: Temporal and spatial trends in simulated, seasonally averaged 35 d antecedent mean salinity (ppt) from A) Base and B) High Flow model scenario wet seasons and C) Base and D) High Flow model scenario dry seasons across WYRs and sampling sites. Color ramps are the same as those used in Fig. 6A, C.