ABSTRACT

Title of Document:	DEPTH PREFERENCES OF OVERWINTERTING JUVENILE BLUE CRABS (*CALLINECTES SAPIDUS*) IN THE MARYLAND WATERS OF THE CHESAPEAKE BAY: A LOCAL SEASONAL STUDY AND PRELIMINARY SHALLOW WATER SURVEY
	Anthony Gartner Kaufman, Master of Science, 2014
Directed By:	Professor Thomas Miller Research Associate Professor Eric Johnson Marine, Estuarine, and Environmental Science

A series of experimental studies designed to quantify the spatial and temporal patterns of juvenile blue crabs abundance in upper Chesapeake Bay, with an emphasis on the depth distribution during the winter, were conducted. A seasonal sampling of the Rhode and West Rivers was conducted. Concurrently, a pilot-scale shallow water survey (SWS) in four river systems throughout Maryland allowed the abundance of juvenile blue crabs to be estimated and compared to abundance estimates from the Winter Dredge Survey (WDS). The seasonal study results indicated that depth, sediment type and the time of year sampling took place were a significant predictor of juvenile blue crabs distribution and abundance. The SWS indicated that the abundance of juvenile blue crabs is underestimated by the WDS. This work highlights the potential need for additional surveys to index the abundance of juvenile blue crabs as an aid to ongoing management and conservation efforts.

DEPTH PREFERENCES OF OVERWINTERTING JUVENILE BLUE CRABS (*CALLINECTES SAPIDUS*) IN THE MARYLAND WATERS OF THE CHESAPEAKE BAY: A LOCAL SEASONAL SURVEY AND PRELIMINARY SHALLOW WATER SURVEY

By

Anthony Gartner Kaufman

Thesis submitted to the Faculty of the Graduate School of the
University of Maryland, College Park in partial fulfillment
of the requirements for the degree of
Master of Science
2014

Advisory Committee:

Professor Thomas Miller, Co-Chair
Research Associate Professor Eric Johnson, Co-Chair
Assistant Professor Hongsheng Bi

Acknowledgements

I would like to thank my advisors; Dr. Tom Miller, Dr. Eric Johnson, and Dr. Hongsheng Bi for their guidance, support, and patience; their encouragement and enthusiasm made the pursuit of my Master's an adventure I will never forget. I would like to also thank the Smithsonian Environmental Research Center, specifically the Fish and Invertebrate Ecology Lab, for hosting me and for providing the necessary resources while I conducted my sampling. A special thanks goes to Mike Goodison, for captaining a variety of research vessels needed to complete the data collection, and to Rob Aguilar, for his help collecting data as well as all of his help with GIS. I would also like to thank Paige Roberts, Midge Kramer and the many "Crab Lab" interns for their help in collecting the data used in this thesis; without their assistance and dedication I would not have been able to complete my sampling. I would like to Glenn Davis for providing me with the raw data collected from the Winter Dredge Survey.

I would like to thank my family for all of their support. Uncle Tim- thank you for taking me on my first SCUBA dive and sparking my passion for the marine environment. Ron- thank you for introducing me to fisheries research and management during my internship senior year of high school. Amanda, Ben, and Christina- thank you for being such great siblings. Dad- even though you are no longer here, thank you for your friendship and for instilling in me a desire to explore the world around me. Last, but most importantly; Mom- thank you for always being there for me and providing endless encouragement and love.

This research, and my fellowship, was funded by Maryland Sea Grant. Thank you for the funding necessary to complete my research as well as my academic requirements.

TABLE OF CONTENTS

List of Tables

List of Figures

Figure 4.6 Plot of the proportional distribution of the raw juvenile blue crabs caught per tow in both the Shallow Water Survey (SWS) and Winter Dredge Survey (WDS) for the Patuxent.

CHAPTER 1:

INTRODUCTION

The blue crab, *Callinectes sapidus*, is a species of swimming crab native to the Western Atlantic, from Maine to Argentina, and the Gulf of Mexico (Rathbun 1896; Williams 1974). One of the more defined populations of blue crab is in found in the Chesapeake Bay (Rugolo et al. 1997), where this species is a key component of the estuarine foodweb (Baird and Ulanowicz 1989), and supports both important recreational fishing opportunities and the Chesapeake Bay's most lucrative commercial fishery (Kennedy et al. 2007).

Like many estuarine-dependent species, the blue crab exhibits a complex life history which involves movement between marine and estuarine habitats (Figure 1.1). Mating occurs in shallow water tributaries throughout Chesapeake Bay when females molt to maturity; on the molt to maturity, males cradle females and transfer sperm during her soft stage (Jivoff et al. 2007). In general, mating occurs between May and October (Hines et al. 2003). After mating, inseminated females undergo a long-distance migration from subestuaries throughout the Bay to the spawning areas in the lower Bay, with peak movement occurring in the fall (Aguilar et al. 2005, 2008; Hines et al. 2008). Most females then overwinter and will begin producing broods of eggs in the following year. A female will produce several broods in her lifetime potentially over multiple years (Hines et al. 2003; Darnell et al. 2009). The resulting larvae (zoea) are then carried into the waters off the coastal shelf (Johnson and Hess 1990; Roman and Boicourt 1999; Natunewicz and Epifanio 2001).

After undergoing a series of molts, zoea will metamorphose into postlarvae, called megalopae, and return to the Chesapeake Bay (Epifanio 2007). Re-entering the Chesapeake Bay from the Atlantic Ocean in late summer and fall

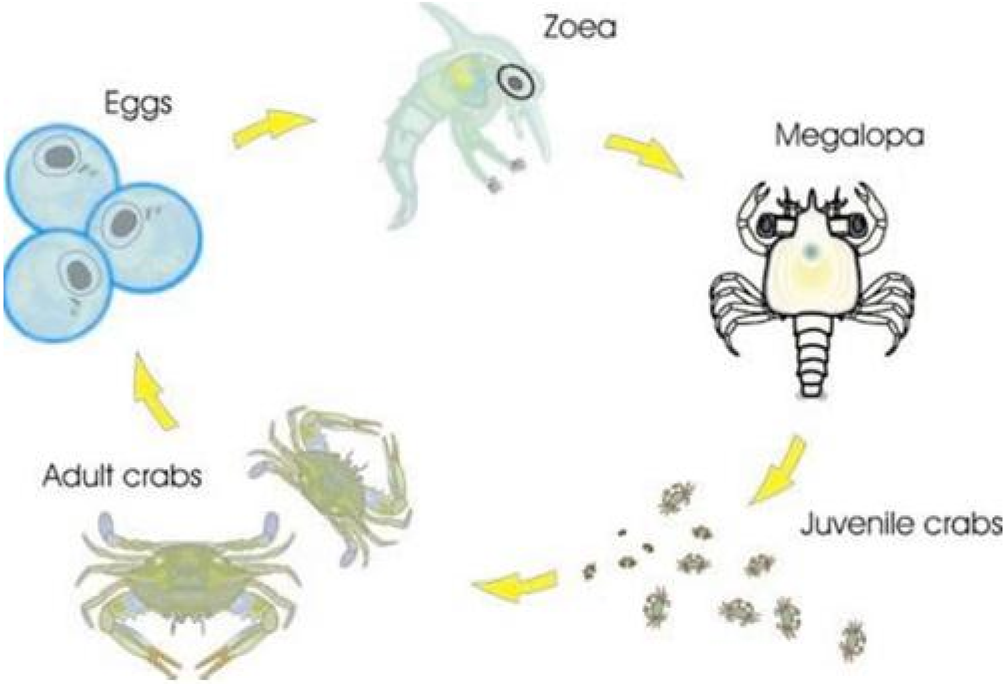

Figure 1.1 Life history cycle of the blue crab, *Callinectes sapidus*. (Diagram provided by the Smithsonian Environmental Research Center)

(van Montfrans et al.1990, 1995), megalopae then undergo metamorphosis to the first juvenile instar (C1) and settle in primarily in seagrass beds or other structured habitats of the lower Bay (Orth and van Montfrans 1987; Lipcius et al. 2005, 2007). Juvenile blue crabs continue to feed and grow within initial lower Bay settlement habitats until a size of approximately 20 mm carapace width (CW; 5th to 7th crab instar, C5-C7 stage). At this stage, juveniles commence the secondary dispersal phase (Etherington and Eggleston 2000), whereby they redistribute into a wide array of shallow water habitats throughout Chesapeake Bay (Pile et al. 1996; Hines et al. 1987; Hines 2007) which serve as secondary nurseries. Secondary dispersal may also occur in early instars (C1-C2; Reyns and Eggleston 2004; Reyns et al. 2006) and appears to be mediated by density-dependent competition and cannibalism among conspecifics (Gunther 1992; Moksnes et al. 1997; Hines and Ruiz 1995; Etherington and Eggleston 2003). Secondary dispersal in this species plays a large role in structuring population dynamics by redistributing juveniles from initial settlement habitats that harbor high densities of juveniles to low density habitats that may receive little or no postlarval recruitment (Etherington and Eggleston 2003).

The juvenile stage plays a vital role in the population dynamics of the Chesapeake Bay blue crab. Juvenile blue crabs grow rapidly in their first year, some will even reach a marketable size. Until recently the stock assessment models assumed 100% of the age 0 crabs would reach marketable size before the season was over. However, recent changes have reduced this percentage to 60% (Miller et al. 2011). This is very important as any changes in the abundance of juveniles susceptible to the fishery will impact the historical exploitation rates leading to changes in the assessment of the population’s health.

Therefore it is imperative we have an accurate estimate of the abundance of juvenile blue crabs.

In Chesapeake Bay, secondary dispersal occurs during fall with juvenile blue crabs typically arriving in upper Bay subestuaries in October-November (Hines et al. 1987). With the onset of winter juveniles will then bury in the sediment and become inactive and as water temperatures decline. Although the timing of secondary dispersal to upper Bay subestuaries in Chesapeake Bay is relatively well documented (Hines et al. 1995; Hines 2007), the fine-scale patterns of seasonal depth and habitat selection by juvenile blue crabs remain poorly documented in Chesapeake Bay. The primary objectives of this study were to quantify spatio-temporal patterns of juvenile crab abundance in the shallow water tributaries of upper Chesapeake Bay.

Objectives

For this study I had three objectives.

Objective 1: Calculate gear efficiencies for active juvenile blue crabs using two gear types, and one gear type for overwintering juvenile blue crabs.

To address this objective, a series of gear efficiency experiments were conducted for two gear types: (1) a commercial grass scrape and (2) a modified commercial grass scrape fitted with a tooth bar that allowed it to act in the field as a small scale dredge. A gear efficiency study for both types of gear was conducted on active juvenile blue crabs during summer using mark-recapture methodologies. Secondly, a gear efficiency study was conducted for only the small-scale dredge on overwintering juvenile blue crabs using a series of depletion experiments; this component of the study did not include the commercial grass scrape because gear efficiency was assumed to be near zero based on previous observations. This objective is addressed in Chapter 2.

Objective 2: Quantify the spatiotemporal patterns of juvenile blue crabs densities in two rivers within the upper Chesapeake Bay with an emphasis on the spatial distribution during winter.

To address this objective the depth distribution and of juvenile blue crabs was tracked between October 2010 and July 2011 in the Rhode and West rivers using a fishery-independent survey involving a stratified sampling design with depth and bottom sediment type as strata. A generalized additive model was used to predict the density and distribution of juvenile blue crabs in the area sampled. This study is presented in Chapter 3 of this thesis.

Objective 3: Conduct a study of the feasibility of a fishery-independent winter survey by sampling the shallow waters of the Maryland section of the Chesapeake Bay for juvenile blue crabs and comparing the density of juvenile blue crabs within the shallow waters to the densities observed in the Winter Dredge Survey.

To address this objective, two creeks were sampled in each of four river systems selected to be broadly representative of four regions of the upper Chesapeake Bay. The average density of juvenile blue crabs found in each river system was compared to the average density of juvenile blue crabs in all winter dredge survey sites within nearby sites (<16 km). An estimate of absolute juvenile abundance was then calculated using combined Shallow Water Survey and Winter Dredge Survey(WDS) data and compared to estimates generated for the WDS alone to determine if the juvenile crab population is underestimated by the current WDS. This objective is addressed in Chapter 4 of this thesis.

CHAPTER 2:

EFFICIENCY OF GEAR FOR SAMPLING ACTIVE AND OVERWINTERING JUVENILE BLUE CRABS

Abstract

An accurate estimate of sampling gear efficiency is critical when estimating the absolute abundance of a species from fisheries independent surveys. For this thesis, three gear efficiencies were calculated using alternative gear that could be used to sample juvenile blue crabs: (1) commercial grass scrape sampling active juvenile blue crabs, (2) small-scale dredge sampling active juvenile blue crabs, and (3) small-scale dredge sampling overwintering juvenile blue crabs. Two estimate methods were used to calculate the three gear efficiencies: a mark recapture method estimated the efficiencies of gears sampling active juvenile blue crabs and a depletion method estimated the efficiency of the gear that sampled overwintering blue crabs. There was no significant difference in the efficiencies of each gear efficiency calculated. The individual efficiencies, each approximately 25%, are similar to the weighted average gear efficiency for all boats participating in the 2011 winter dredge survey as well as previous summer surveys.

Introduction

Searches rely upon the relative movement of target and sensor (Koopman 1956). In passive searches, the sensor is fixed in time and the search relies on the motion of the target toward the sensor, e.g. RADAR or a sit and wait predator (Gerritsen and Strickler 1977). Active searching requires movement of the sensor relative to the target, e.g., search and rescue or a cruise predator (Gerritsen and Strickler 1977). In many instances of active searching both the sensor and the target move – because often one party wants to find the other which is in turn seeking to evade being found (e.g., predator – prey interactions). In both active and passive search strategies the sensitivity and efficiency of the sensor are important determinants of the overall effectiveness of the search. A sensor with a wide encounter radius is generally better than one with a smaller search radius, and a more sensitive sensor better than a less sensitive one. The encounter radius of a sensor is generally easy to determine, but the efficiency of the sensor is often challenging to quantify. Yet both are important determinants of the effectiveness of the search process.

In natural resource management, searches are often conducted to determine the number of individuals of a target species in an area. The resultant abundance estimates are used to derive estimates of abundance which when sequentially combined can provide estimates of population growth and survival. Often the search is conducted within a rigorous statistical framework, (e.g., random and stratified random surveys), which then provide estimates of the variance in the abundances estimated in each area surveyed. Subsequently, the abundance per area can be multiplied by the total survey area to provide an estimate of the total abundance. However, this approach assumes that the sensor used in the survey has an absolute encounter radius and a known efficiency. Thus, knowledge of the encounter radius and efficiency of sensors used in surveys is central to the reliability of the abundances and vital rates, if estimated, of any survey.

Fishery management use a variety of sensors to conduct surveys. Increasingly managers use electronic sensors (both passive and active hydroacoustics and lidar) to estimate abundance. However, nets are still commonly used to assess abundance. These have the advantage of having a known and binary encounter radius – indexed by the overall size of the net. However, the efficiency of nets can be difficult to estimate and is often poorly known. In many cases, managers of fishery resources rely on indices of relative abundance and use a statistical model to expand these relative abundances to yield an absolute abundance. But even in this case, it is necessary to assume that the sampling gear is equally efficient in all habitats.

The blue crab, *Callinectes sapidus*, is a species of swimming crab native to the Western Atlantic, from Maine to Argentina, and the Gulf of Mexico (Rathbun 1896; Williams 1974). One of the more defined populations of blue crab is in found in the Chesapeake Bay (Rugolo et al. 1997), where this species is a key component of the estuarine foodweb (Baird and Ulanowicz 1989), and supports both important recreational fishing opportunities and the Chesapeake Bay’s most lucrative commercial fishery (Kennedy et al. 2007). Management of blue crab in the Chesapeake Bay relies upon data from fishery independent surveys and from the fisheries which are combined in a stock assessment model to establish management reference points (Miller et al. 2011). Central to the reliability of the assessment is a winter dredge survey which estimates the abundance of blue crabs during winter months when crabs are quiescent in the sediments (Sharov et al. 2003). Substantial effort is invested to estimate the efficiency of the winter dredge gear so that the fishery managers can provide stakeholders with estimates of absolute abundance. However, the estimate of the efficiency of the survey for juvenile crabs is known to be highly uncertain because juvenile crabs are distributed outside of the survey area (shallow water), thereby allowing availability to the survey to potentially vary, because juveniles may not be fully retained by the mesh on the dredge itself (efficiency), and because the vagility of crabs changes seasonally with distinct active and quiescent periods.

Estimating efficiency of active fishery survey gear is challenging. One common method of estimating gear efficiency appropriate for use with the chosen gear, on active crabs, is mark recapture. Mark recapture relies on release and resampling of a known number of marked individuals. In simple terms, the known density of marked individuals released can be compared to the density of marked individuals caught in the sample. The proportional difference of sampled density and known density is the estimate of gear efficiency. The most common method of estimating the gear efficiency of inactive crabs is the depletion method (Sharov et al. 2003; Volstad et al. 2000). Depletion studies require that you sample the same area multiple times and record the decline in catch per sample. While this method is commonly used, it is dependent on being able to sample the exact same area multiple times. There are methods that can be used if you deviate from the original sampled path (Rago et al. 2006); however, the calculation of the gear efficiency is more complicated. An alternative method for use on inactive crabs, is one in which you take an initial sample within a defined area larger than the sampled area to get a density estimate. The defined area can then be sampled till no crabs are left. The total crabs caught can then be used to calculate a known true density. This method requires that no immigration or emigration occurs while the defined area is being depleted. This method also requires that the gear remains in the defined area while depleting it of juvenile blue crabs. Like the mark recapture estimate, the proportional difference of initial sample density and known density is the estimate of gear efficiency.

Here, I evaluate the gear efficiencies of two alternative gears that could be used to target juvenile crabs in both the active and quiescent phase. Commercial grass scrapes are a familiar gear type used in the sampling of juvenile blue crabs. The grass scrape is similar to a bottom trawl but provides a rigid opening that feeds to a mesh net. This is beneficial because it provides a fixed gear width which can then be easily multiplied by the distance the gear is dragged to accurately calculate the total area sampled. The grass scrape is the ideal gear for active juvenile blue crabs; however it is ineffective when sampling for stationary crabs that are buried into the sediment. A dredge is commonly used to sample overwintering blue crab, but this gear type requires larger vessels that cannot access the shallow water. A commercial grass scrape can be modified with a tooth bar that allows the gear to act as a dredge while being small enough to be pulled by smaller boats with less draft (Figure 2.1).

Gear efficiencies were predicted to differ between gear types, and among winter when crabs are inactive and buried and other months when crabs are active and mobile. These differences necessitated estimation of three separate three gear efficiencies: (1) efficiency of the commercial grass scrape on active blue crabs, (2) efficiency of the

Figure 2.1 Picture of the modified commercial grass scrape fitted with a tooth bar that allowing it to act as a small scale dredge. The commercial grass scrape was identical except with a solid bar rather than a tooth bar.

small-scale dredge on active juvenile blue crabs, and (3) efficiency of the small-scale dredge for overwintering blue crabs. Gear efficiency for the grass scrape in winter was not estimated because it is known that this gear type is not effective for sampling inactive blue crabs.

Materials and Methods

Gear efficiency for active juvenile blue crabs

A small-scale mark-recapture experiment was designed and conducted during June 2010 to estimate the gear efficiency for the commercial crab scrape and the small dredge sampling active juvenile blue crabs. A sample of crabs were collected from around the Chesapeake Bay using the commercial grass. A subsample of 512 crabs, 408 of which were less than 60mm (mean size 44 mm carapace width (cw), range 11-167 mm cw), were marked using coded microwire tags (CWTs; Northwest Marine Technology, Inc. NMT), Shaw Island, WA 98286) prior to release. CWTs have negligible rates of short-term tag loss and have been successfully used in a variety of mark-recapture experiments with this species (Eggleston and Johnson 2008; Johnson and Eggleston 2010; Johnson et al. 2011). Tagged crabs were released in July of 2010 into a small cove (0.32 ha) within the Rhode River resulting in an approximately uniform density of 0.16 crabs m^{-2} for all crabs and 0.13 crabs m^{-2} for crabs less than 60mm. Emigration from the study area was not likely to occur given the spatial configuration of the cove which had a narrow opening and because sampling commenced immediately following release.

Each gear was then towed randomly within the study area for a distance of 75m and all captured blue crabs were scanned with a magnetic moment detector (NMT) to detect the presence of a CWT. The carapace width and sex of each recaptured crab was then recorded, and crabs were returned to the study site along the approximate path of the tow prior to beginning each subsequent tow. In total, 22 tows were completed with the commercial grass scrape and 18 tows with the small scale dredge over the course of a sampling period of four hours. The gear efficiency was calculated by dividing the average observed density of tagged crabs by the expected density of juvenile blue crabs, those less than 60mm.

Gear efficiency of overwintering juvenile blue crabs

Gear efficiency for juvenile blue crabs during winter may differ from that in summer because crabs are known to bury into the sediment at low temperatures which may make them less vulnerable to capture by the survey gear. Winter gear efficiency experiments were conducted in the Patuxent River given that in 2010 it had the highest summer juvenile blue crabs densities of any river on the western Maryland section of the Chesapeake Bay. Three independent locations in shallow tributaries of the lower Patuxent River were randomly chosen to conduct the gear efficiency experimental trials. To begin a trial, a 5m × 75m experimental area was delineated by marking the four corners with PVC pipe driven into the sediment. At least one tow was performed inside the marked area and a minimum of two additional tows in the area immediately adjacent to the marked area using a small scale winter dredge (Figure 2.2). The density of juvenile blue crabs in the marked area was assumed to be identical to the density just outside the

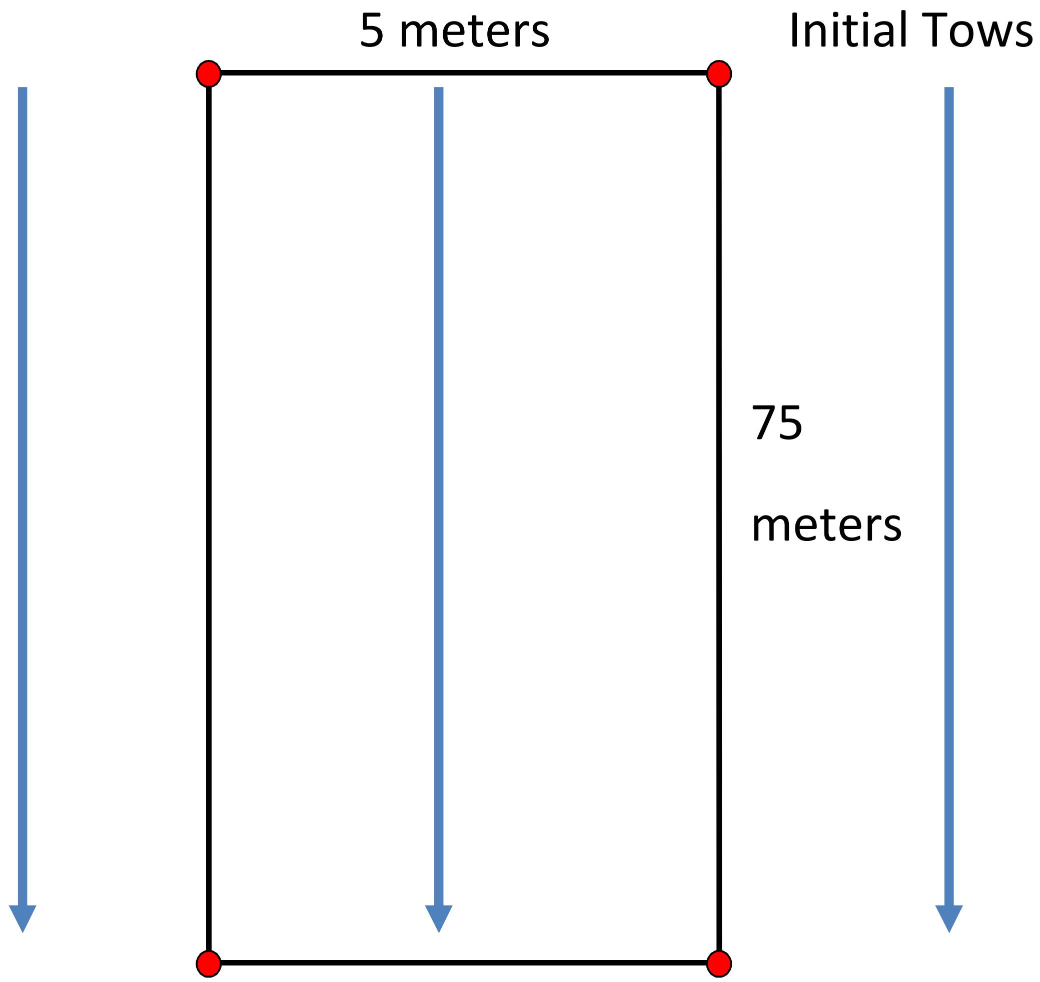

Figure 2.2. Schematic diagram of the overwintering juvenile blue crabs gear efficiency study sites. The red circles represent PVC pipes that mark the study area. The black lines show the border lengths. The blue lines represent the initial tows performed.

marked area. Additional tows were performed within the marked area until the study area was assumed to be completely depleted of crabs. Care was taken not to have the small-scale dredge leave the defined area. If the gear appeared to be drifting out of the area the tow was stopped and reset. To be reasonably sure that the area was completely depleted, sampling continued until the following two conditions were met: (1) a minimum of 20 tows had been completed within the study area and (2) no blue crabs were captured on three successive tows. This process was repeated in three experimental trials to generate three independent estimates of gear efficiency. However, due to gear failure on the third attempt only two trials were used in the calculation of the gear efficiency.

For each winter tow the carapace widths of all blue crabs caught were measured. To calculate the gear efficiency for juvenile blue crabs, first the sum total of all blue crabs less than 60mm collected in all tows inside the study area was calculated. This value was an estimate of the actual number of juvenile blue crabs within the marked area. This value was then divided by ($375 m^2$), which was the size of the study area (5 × 75m) to calculate the absolute density (no. m^{-2}) of juvenile blue crabs (CW < 60mm) in the study area. A density estimate of blue crabs less than 60mm was calculated for each of the initial tows. The gear efficiency was calculated by dividing this estimate of relative density by the estimated absolute density. For each trial, the gear efficiency estimates calculated from the initial tows were averaged. The trials were averaged to get the final gear efficiency estimate. During the sampling of the third site the boats davit broke and sampling could not be completed. Thus, the marked area could not be completely depleted. Therefore, only the first two sites were used to calculate the gear efficiency.

Results

In summer for active juvenile blue crabs, an area with a known density of 9.56 juvenile crabs less than 60 mm per $75m^2$ was sampled. The grass scrape caught an average of 2.4 ± 2.9 crabs per $75m^2$ (range 0 – 12 crabs per 75 m^2, n=22, Table 2.1) The distribution of catches in the grass scape is not normal and appears to follow the negative binomial distribution given the standard deviation is larger than the mean, this is common in count data. For the small scale dredge the average tow contained 2.8 ± 2.6 crabs per $75m^2$ (range 0 – 7 crabs per 75 m^2, n=18, Table 2.1). Like the distribution of grass scrape catches, the distribution of catches for the small scale dredge most closely resembles that of the negative binomial distribution. The resultant gear efficiencies were 24.7% for the commercial grass scrape and 25.3% for the small scale dredge.

In sampling of inactive juvenile crabs in the Patuxent River 27 and 32 tows were completed to meet the depletion requirement established in the sampling design. The third trail could not be completed due to a gear failure. The resultant average abundance among the two completed areas was 33.8 ± 22.4 crabs per 375 m^2 (Table 2.2). The estimated abundance in the adjacent unmarked areas was 7.2 ± 8.6 crabs per $375m^2$ (Table 2.2). The estimated gear efficiency for overwintering blue crabs, less than 60mm, for the small scale dredge was 24.2 %. A summary of the site specific gear efficiency estimates for overwintering juvenile blue crabs can be found in Table 2.2.

Table 2.1 A Summary of the gear efficiency for the small scale dredge and commercial grass scrape on active juvenile blue crabs.

	Average Crabs per 75m Tow	Released Density in crabs / 75m^2	Gear Efficiency Estimate	Standard Error
Grass Scrape	2.364	9.556	0.247	0.06
Dredge	2.778	9.556	0.253	0.05

Table 2.2. A Summary of the gear efficiency estimates and the overall average gear efficiency for the small scale dredge on overwintering juvenile blue crabs. The standard error is calculated from the two trial estimates.

	Gear Efficiency Estimate	Standard Error
Inside Left	0.1128	
Outside Left	0	
Outside Right	0.5039	
Trial 1	0.2056	0.15267
Inside Left	0.2778	
Inside Right	0.5556	
Outside Left	0	
Outside Right	0.27778	
Trial 2	0.27778	0.1134
Average	0.24167	

Discussion

The purpose of this study was to accurately estimate the gear efficiencies for the commercial grass scrape on active blue crabs, the efficiency of the small-scale dredge on active juvenile blue crabs, and the efficiency of the small-scale dredge for overwintering blue crabs. The efficiency estimates ranged from 24.2% to 25.3% and the confidence intervals for all three estimates overlapped with one another. Therefore there was no significant difference among the three gear efficiencies calculated. I expected to find a lower gear efficiency estimate on active crabs than inactive crabs, using the small scale dredge, because I assumed they might avoid the gear. The fact that the gear efficiency estimate for the small scale dredge on active and buried crabs is so similar suggests that active crabs are not attempting or are not able to avoid the gear when they are active.

The use of mark recapture studies in calculating gear efficiency is well documented in many fisheries. In this study the estimated gear efficiency for the commercial grass scrape is consistent with other similar studies. However, this study included a modified commercial grass scrape that would act as a small scale dredge. It was not known whether the modifications to the commercial grass scrape would have an impact on the gear efficiency. It was determined that the gear modifications did not have an appreciable effect on the gear efficiency given the small difference between the two estimates and significant overlap between the two confidence intervals. The study requires the following assumptions: (1) No tagged crabs left the sampling area and (2) the crabs were homogeneously distributed within the sampling area. Given the mobility of blue crabs it is likely both assumptions were violated. However, it is possible the impacts of the violations are not very large. Given the relatively short duration of sampling it is unlikely that many crabs left the small cove. Even if the crabs did not leave the cove they must be evenly distributed within the cove. When the crabs were first released great care was taken to ensure the crabs were scattered evenly within the cove. It is possible that once released the crabs would move towards the shallow waters and away from other crabs. However, as the tows were conducted within the cove the collected crabs were again scattered around the entire cove. Some improvements could be made if this mark recapture study was going to be conducted again. Firstly, the cove should be blocked off by a net that would prevent any crabs from leaving. Secondly, a defined sampling pattern could be implemented that reduces the impacts of the crabs migrating within the cove. Given the known limitations and issues with these gear efficiency studies, a degree of caution should be taken in any subsequent results that rely on converting raw catch to absolute density/abundance.

An emphasis was put on ensuring the gear efficiency estimates were accurate, a comparison can be made between the estimates generated from this study and to those of other similar depletion style efficiency estimates for dredge gear. In the study conducted by Volstad et al. (2000) they estimated changes in gear efficiency on overwintering blue crabs of all sizes using different dredge linings. They found gear efficiencies ranging from 15% to 22% with standard errors ranging from 2% to 3%. These gear efficiencies are slightly lower than those observed in this study. The study conducted by Sharov et al. (2003) is most comparable to this winter gear efficiency study, here they found the gear efficiencies ranged from 17% to 42% with standard errors ranging between 1% and 9%. The wide range of gear efficiencies can be explained by the design, in this study they ran the gear over a defined path and used the rate of depletion to estimate the gear efficiency. Therefore, they must assume that the gear never leaves the original path. A problem with this assumption is that it is incredibly difficult to follow the exact same path every time. Any deviation from the original path will result in a slower apparent depletion rate which will imply a lower gear efficiency estimate. Each year the returning captains would be better able to keep the gear on the original path. This resulted in an apparent increase in gear efficiency over time.

This winter gear efficiency study was designed in such a way that the gear was only required to remain within a defined area and not to a defined path. The Sharov et al. (2003) study looked at the rate of depletion, while this study compared the density of crabs within an initial tow to the density of crabs observed by fully depleting the defined area. In this study two assumptions must be made: (1) the gear does not leave the defined area while depleting it of crabs, and (2) the density of crabs within the defined area is the same as the area directly adjacent. While every attempt was made to ensure the gear never left the defined area it was possible. If the gear left the area, any crabs collected outside the area would be included in the final observed density and result in an artificially lowered gear efficiency rate. If the second assumption is violated then the tows that were taken outside the defined area could not be compared to the density observed in the depleted area. If the winter gear efficiency were to be repeated, none of the initial tows would occur outside of the marked area and I would increase the number of trials conducted. This would eliminate the need for the assumption of homogeneous density within and adjacent to the depleted area. Additionally I would attach a floating marker to a line tied to the mouth of the gear that would allow the boat operator to know if the gear was going to leave the defined area.

CHAPTER 3:

SPATIOTEMPORAL PATTERNS IN JUVENILE BLUE CRABS DENSITIES IN THE RHODE AND WEST RIVERS, MD

Abstract

The life history of the Chesapeake Bay blue crab population is well described. However, the habits of juvenile blue crabs after they leave the lower bay habitats are not as well described as other life stages. In particular, little is known about the depth distribution of juvenile blue crabs during winter. The spatial and temporal patterns of juvenile blue crabs densities in the Rhode and West Rivers, two adjacent tributaries of upper western Chesapeake Bay, was examined in a seasonal study. To quantify the distribution of juvenile blue crabs in these systems, a stratified survey was conducted using depth and bottom sediment type as strata. The spatiotemporal patterns of juvenile blue crabs density was described. Most relevant to this investigation was the finding that depth, sediment and sampling time were significant predictors of juvenile blue crabs density. During the winter sampling period juvenile blue crabs were found mostly in the shallow waters on the edge of the rivers sampled. Consistent with earlier work in these systems, it was observed that in the summer month's juvenile blue crabs were predominantly found in shallow waters less than 1.5m.

Introduction

Following a period of larval development in oceanic waters, juvenile blue crabs settle in high salinity estuarine waters. Structured habitats, such as seagrass, appear to be preferred habitats, for early stage juveniles, as a protection against high mortality rates (van Montfrans eta al. 1995; Stockhausen and Licpius 2003; see Lipcius et al. 2007 for review). As the juveniles grow, competition for valuable food resources in seagrass beds increases (Etherington et al. 2003) and the value of seagrass as a refuge from predation diminishes (Pile et al. 1996; Lipcius et al. 2007). At this stage, many juveniles undergo secondary dispersal. This process appears to be driven by density-dependent competition among conspecifics (Günther 1992; Etherington and Eggleston 2003; Reyns and Eggleston 2004), which is not surprising given the cannibalistic nature of the species (Moksnes et al. 1997; Hines and Ruiz 1995). Secondary dispersal results in a wide distribution of juvenile crabs away from the primary settlement sites (Etherington and Eggleston 2003), such that juveniles become distributed to lower density juvenile habitats that receive little or no postlarval settlement.

Secondary juvenile habitats are often shallow tributary or littoral habitats. Juveniles tend to remain in these shallow secondary habitats until they mature, (Hines et al. 1995; Davis et al. 2005). However, juveniles remain active as they grow, moving within and between secondary habitats reflecting both ontogenetic changes in habitat use and patterns of food availability. For example, juvenile crabs tend to move into shallow waters in the warmer months to escape predation and cannibalistic interactions with larger crabs in deeper waters (Dittel et al. 1995; Hines et al. 1995). In contrast, the depth preference of juvenile blue crabs during winter is poorly understood; although several studies have documented the impact of winter mortality at low temperatures (Bauer and Miller 2010a, Bauer and Miller 2010b, Rome et al. 2005). Therefore it is reasonable to hypothesize that juvenile crabs may leave the shallows and move to deeper waters which may provide a thermal refuge and reduced potential for ice scour. Although this has been reported anecdotally (Hines 2007), this hypothesis has not been rigorously tested.

Habitat selection remains an important unanswered question in juvenile blue crabs for both ecological and management reasons. An accurate understanding of the distribution patterns of juvenile blue crabs during winter has important management implications. For example, a bay-wide winter dredge survey is conducted in the Chesapeake Bay to provide estimates of absolute abundance for management purposes (Volstad et al. 2000; Sharov et al. 2003; Miller et al. 2005; Miller et al. 2011). This survey is conducted annually at approximately 1,500 randomly assigned stations, but is restricted to water depths > 1.6m. If juvenile blue crabs routinely occur in waters < 1.6 m then not all of the population of juvenile blue crabs are available to the survey gear. This would not be a concern if a constant fraction of the juvenile population occurred in waters < 1.6m in winter, as the bias would be constant from year to year. However, if differing proportions of the juvenile population occur at depths that are not available to the survey, then survey estimates become less reliable. If, as suggested above winter depth preference is temperature dependent, then the depth distribution of juvenile blue crabs likely does change from year to year. Understanding the absolute number of juvenile blue crabs is important because stock assessments for this population assume a pattern of recruitment of juvenile blue crabs to the fishery over the course of the subsequent fishing year (Rugolo et al. 1997; Miller and Houde 1999; Miller et al. 2005; Miller et al. 2011).

Uncertainties regarding interannual variability in the availability of the juvenile blue crabs to the winter dredge survey can be evaluated by implementing a survey of juvenile blue crabs in water depths < 1.6m. Accordingly, the spatiotemporal patterns of juvenile blue crabs density in the Rhode and West rivers, two representative shallow tributaries of upper Chesapeake Bay, were quantitatively assessed. Numerous studies have described the distribution of juvenile blue crabs in these rivers from spring through fall (Hines et al. 1987), however the winter distribution of juvenile blue crabs is not known. The main objective was to quantify the spatial and temporal patterns of juvenile blue crabs in the Rhode and West rivers, with an emphasis on the overwintering depth and spatial distribution. To accomplish this task, a stratified survey was conducted from October 2010 to July 2011. The strata were determined by unique combinations of depth and bottom sediment type. Using observed density data, a generalized additive model incorporating depth, bottom sediment type and sampling month was used to predict the density of juvenile blue crabs at each potential sampling cell. These predictions were mapped so that the distribution of juvenile blue crabs could be observed for each sampling period.

Materials and Methods

The density and depth distribution of juvenile blue crabs was estimated during eight individual sampling periods, between early October 2010 and mid-July 2011 using two gear types, a commercial grass scrape and a modified grass scrape, in the Rhode and

West Rivers, two tributaries on the western shore of the Chesapeake Bay (Figure 3.1). The two rivers have a combined surface area of approximately 14.27 square kilometers. A majority of the Rhode River shoreline is undeveloped, however much of the shoreline in the West River is developed. Both rivers are commercially and recreationally fished, predominantly by commercial watermen using trot-lines in the summer months. These rivers were ideal systems for conducting this work because (1) they are broadly representative of upper western shore tributaries in Chesapeake Bay, (2) their long history of ecological research, particularly for blue crabs, and (3) the close proximity to the Smithsonian Environmental Research Center.

The survey employed a stratified sampling design involving depth and bottom sediment type. To determine sampling locations, a GIS shape-file (obtained from rimmer.ngdc.noaa.gov) was used to get the shoreline of the Rhode and West Rivers, the boundary separating the sampling area from the Chesapeake Bay is the no crab pot line. A 100m × 100m grid was then overlaid on top of the sampling area to get unique cells that would then become potential sample sties (Figure 3.1). This grid size was chosen to allow a tow length of 75m to be completed entirely within a sample site. If a cell had an area of less than $3000m^2$ it was omitted from the list of potential sample sites. This was done to reduce the risk of leaving the sample site when sampling.

Bottom sediment type information was obtained from bottom-penetrating sonar data provided by NOAA (http://chesapeakebay.noaa.gov/acoustic-seafloor-mapping/rhode-and-west-rivers-md-benthic-habitat-characterization; S. Giordano, *pers. comm.*). The information was provided in a JPEG that was scaled, georeferenced and

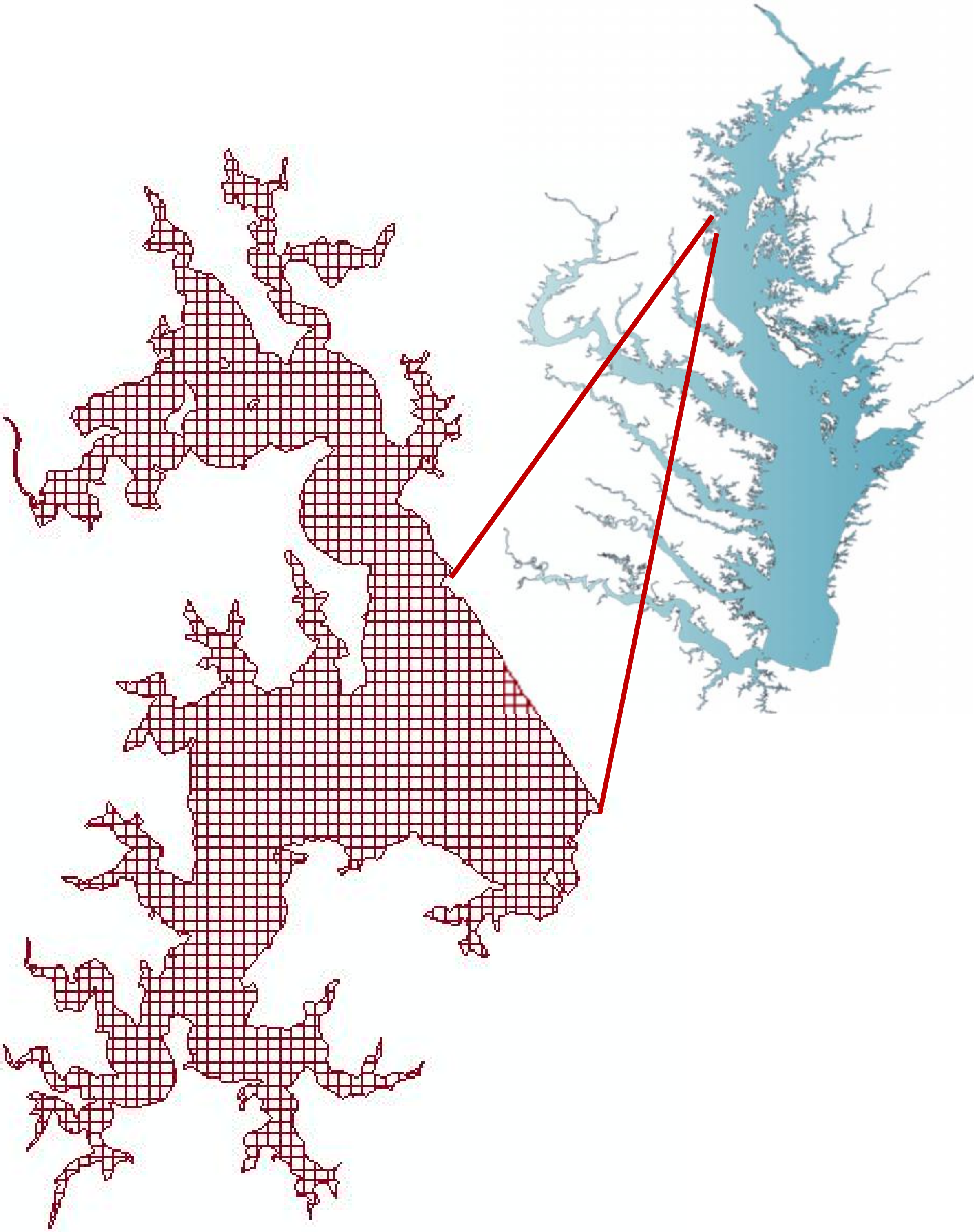

Figure 3.1. A map of the shoreline of the Rhode and West Rivers. The bay boundary is the no crab potting line. The sampling area has a 100mX100m grid overlaid to assist in site selection.

superimposed on the Rhode and West River Grid (Figure 3.2). Because not all of the area in the rivers was mapped by the side scan sonar, the bottom sediment type of unmapped areas was extrapolated using observed nearby sediment types. This assumption was validated by groundtruthing all unmapped cells and a subset of mapped cells using a standard box-core (0.04 m^2) to obtain the samples from which the actual sediment type was determined. The sediment in only 2.3% of cells sampled differed from that predicted by the sonar maps. Bottom type was classified into four major categories: (1) Clay/Silt, (2) Silt/Sand, (3) Sand and (4) hard-bottom/oyster. Because cells were not homogenous with respect to sediment type, if a cell did not have at least 60% coverage of one sediment type it was omitted as a potential sampling location to reduce confounding effects (Figure 3.2); however, few cells fell into this category, and only 9.4% of the 1933 potential sites were excluded. All of the cells that were defined as having hard-bottom/oyster as the dominant sediment type were excluded to reduce the potential impact of the survey on the local remnant oyster populations; only 8 potential sites were excluded using this criterion. The depth for each sampling site cell was determined by using a GIS point file of mean low tide depth information. The average of the points in each sampling site cell (Figure 3.3) was used as the cell depth. Potential sampling cells were classified into four strata according to depth: (1) less than 1m (2) 1-2m, (3) 2-3m, and (4) 3-4m; no cells in the sampling area had an average depth greater than 4m.

The survey utilized two types of gear, a commercial grass scrape and a modified grass scrape to act as a small scale dredge (Figure 2.1). Both gears were 1m in width, but the modified scrape was fitted with a tooth bar with teeth approximately 100 mm in length at a 45 degree angle spaced about 20mm apart. The tooth bar was affixed to the

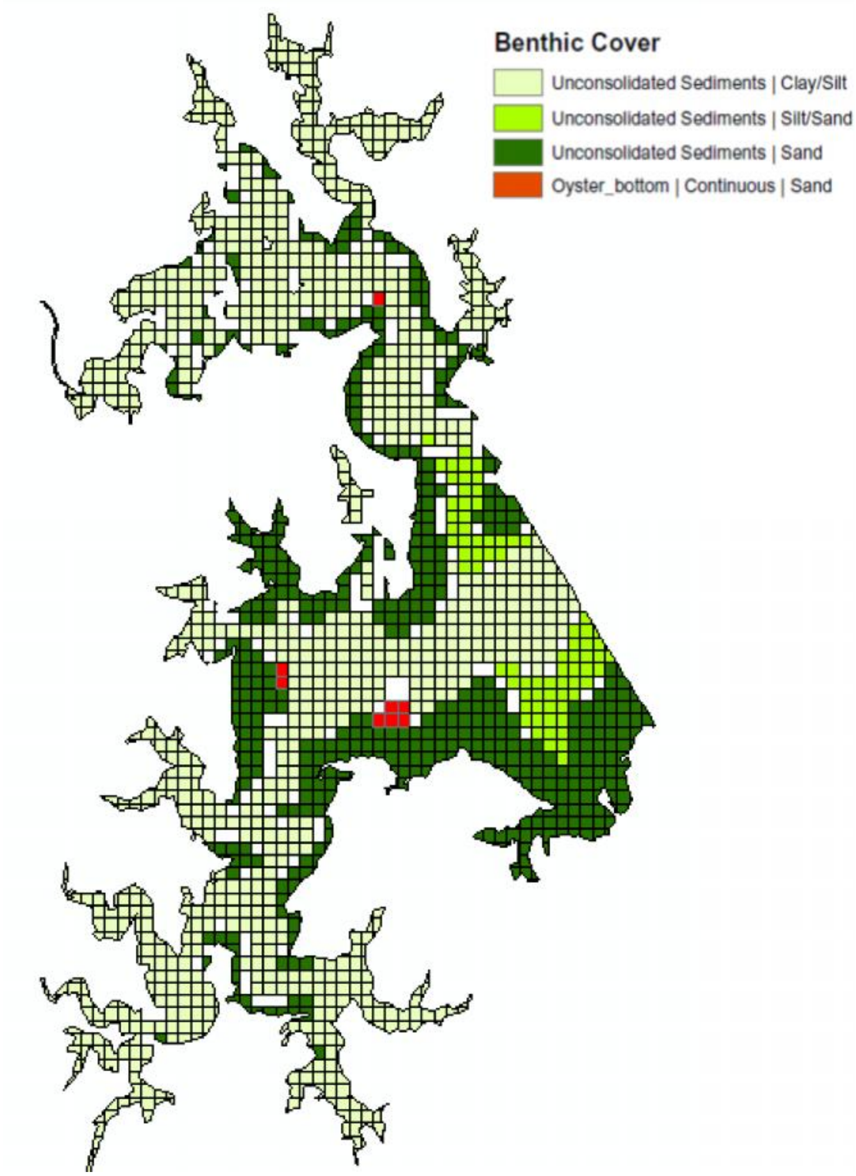

Figure 3.2. Sediment identification for each of the potential sampling sites. The cells that are blank represent sites that did not have at least 60% coverage of one sediment type.

Figure 3.3. Resulting depth identities for each of the potential sampling site cells. Lighter cells represent shallower depths while the darkest areas represent the deepest areas.

bottom of the scrape and allowed the gear to dig into the sediment and effectively sample buried fauna. The commercial grass scrape was towed by a 13ft johnboat and the small scale dredge was towed by the R/V Macoma, an 18ft Boston Whaler. The grass scrape was used during the winter; however, this gear type was ineffective due to low capture efficiency when crabs are buried (see Chapter 2). Optimal effort allocation between the two gear types was determined by using data obtained during field testing of the gears prior to beginning the seasonal sampling. On average, it took 50% longer to perform a 75m tow with the small scale dredge as it did to perform the same tow using the commercial grass scrape. With the available personnel, it was determined approximately 100 sites could be sampled within a week. Therefore, 60 grass scrape samples and 40 dredge samples could be collected for each sampling period during fall, spring and summer when water temperatures are warmer and crabs are active. Due to the ineffectiveness of the commercial grass scrape and to better characterize distribution patterns in during overwintering, an additional 21 sample sites were added for the small scale dredge during winter sampling to increase the sample size and spatial coverage of the data.

Selected sites were sampled from October of 2010 to July of 2011. In total, eight semi-monthly surveys were conducted and subdivided into three sampling periods defined by distinct biological transition events. The first period encompassed juvenile blue crabs recruitment into the Rhode and West Rivers, which typically occurs in late fall during the months of October and November (Hines et al. 1987). The second period was defined as the period of overwintering, or the months of December through March when water temperature declined below 10 °C and crabs bury and become inactive (Brylawski and Miller 2006). The final period was defined as post-winter and encompassed the period from April through July when water temperatures warm and crabs resume activity. Table 3.1 contains the dates of the sampling periods as well as a detailed summary of the crabs caught in each sampling period

All combinations of depth and sediment strata present in the rivers were identified. The sample was then stratified by depth and sediment to ensure all combinations were adequately represented in the data, providing a basis for model-based estimates. Half of the samples in each combination of depth and sediment were fixed (sampled in all periods) and the other half were random. If a combination of depth and sediment resulted in a sample size of two both sites were fixed. An additional site for each gear type was added to the silt/sand sediment in depths between 3m to 4m to increase the number of cells sampled for that particular combination since it appeared it was underrepresented based on the early October sampling.

The fixed sites were selected using a random number generator whose output corresponded to an individual cell with a specific combination of depth and bottom sediment type. If random selection resulted in two sites that shared a border both sites were re-selected. This same method was used to select the random sites for each sampling period. For each sampling location, a random tow direction was chosen; however, in some cases this direction was not followed depending on wind and tidal currents. The latitude and longitude of the center of the cell was used to identify the sample site coordinates that could be uploaded to a handheld GPS unit.

Table 3.1. Summary of the raw data collected from the seasonal study sampling. *After the first sampling period, early October, two additional sites were included for each gear type to increase the coverage of the silt/sand sites in water between 3m and 4m deep. **The winter sampling period contains fewer sampled sites because we did not include the grass scrape data since the gear was not able to effectively sample the buried crabs.

Sampling period	**Gear**	**Total Sampled sites**	**Minimum crabs per $75m^2$**	**Average crabs per $75m^2$**	**Maximum crabs per $75m^2$**	**% of tows with 1 or more crabs**
Early October* (10/05/2010-10/15/2010)	Grass Scrape	60	0	0.21	2	15%
	Dredge	40	0	0.30	4	20%
	Total	**100**	**0**	**0.25**	**4**	**17%**
Late October (10/26/2010-10/28/2010)	Grass Scrape	61	0	0.72	9	31%
	Dredge	41	0	0.85	6	44%
	Total	**102**	**0**	**0.77**	**9**	**37%**
November (11/17/2010-11/19/2010)	Grass Scrape	61	0	0.61	6	28%
	Dredge	41	0	0.99	10	39%
	Total	**102**	**0**	**0.76**	**10**	**33%**
Winter** (02/16/2011-03/17/2011)	Grass Scrape		0	.	.	.
	Dredge	62	0	0.31	3	26%
	Total	**62**	**0**	**0.31**	**3**	**26%**
April (04/19/2011-05/02/2011)	Grass Scrape	61	0	0.26	4	16%
	Dredge	41	0	0.15	1	15%
	Total	**102**	**0**	**0.22**	**4**	**16%**
May (05/16/2011-05/24/2011)	Grass Scrape	61	0	0.70	10	31%
	Dredge	41	0	0.63	10	32%
	Total	**102**	**0**	**0.68**	**10**	**32%**
June (06/07/2011-06/14/2011)	Grass Scrape	61	0	0.74	11	26%
	Dredge	41	0	1.26	6	53%
	Total	**102**	**0**	**0.95**	**11**	**38%**
July (07/05/2011-07/13/2011)	Grass Scrape	61	0	1.18	12	43%
	Dredge	41	0	3.50	25	51%
	Total	**102**	**0**	**2.12**	**25**	**47%**

Standard Field Sampling Methods

Identical sampling methodologies were used for both gear types. Since the coordinates defined the center of the sample cell, the gear was deployed approximately 35m from the coordinates opposite of the tow direction so that entire length of each tow would be within the appropriate sample cell. A handheld recreational GPS (Garmin GPSMAP76) was used to measure the tow distance. A standard tow was 75m in length; however, any deviations were recorded in the data. If a sample could not be completed after three attempts it was discarded and a new site was randomly selected.

At the completion of each tow, bottom temperature, salinity and dissolved oxygen were recorded using a standard handheld YSI Professional Plus (YSI, Inc. Yellowspring, OH). The collection bag was then rinsed to remove mud and sediment before being opened and sorted. For each tow, any species of fish present were recorded; if the fish was a known predator of blue crabs the total length was measured for the first ten individuals of that species. For each crab the sex, carapace width, molt stage and limb loss were recorded. If a dead crab was collected, only the carapace width and sex were recorded.

Identifying the Age 0 Cohort

Because juvenile crabs are growing throughout the survey duration, size cutoffs were estimated for each sampling period to identify juvenile blue crabs (Age-0 cohort) from larger, older individuals (Age 1+). To determine the appropriate size cutoffs, the observed carapace width distributions for each sampling period were used to identify potential cutoffs (Figure 3.4.). The following size cutoffs were used to identify juvenile blue crabs for sample periods 1 thru 7, respectively; less than 40mm, less than 45mm, less than 50mm, less than 60mm, less than 70mm less than 80mm and less than 90mm respectively (Figure 3.4). For sample period 8 (July), juvenile blue crabs had grown sufficiently that two unique cohorts could no longer be clearly identified so all crabs were included in the analysis. With these cutoffs the numbers of juvenile (Age-0) crabs caught in a 75m tow for each sample site was identified for each sampling period.

Statistical Analysis

The overall goal was to observe changes in the spatial distribution of juvenile blue crabs between the recruitment phase and the early summer within the Rhode and West Rivers. Therefore, model based estimates are required to gain a finer level of detail than can be provided by design based estimates. There are two prominent model frameworks that could be used. The most simple is a single process model which assumes a single statistical distribution can describe presence/absence and abundance. A second approach is a two-stage model which separate distributions for presence/absence and abundance (Jensen et al 2005). For this analysis a single stage Negative Binomial Generalized Additive Model (NBGAM) was used to describe the distribution of juvenile blue crabs over a series of months. While environmental data was collected it was determined that the range of observed value would have no effect on the presence of juvenile blue crabs within a given sample period, therefore a two-staged model is unnecessary.

Previous analyses indicate that spatial and temporal autocorrelation were not present in the data (See Appendix I). This suggests a mixed model is not necessary and a

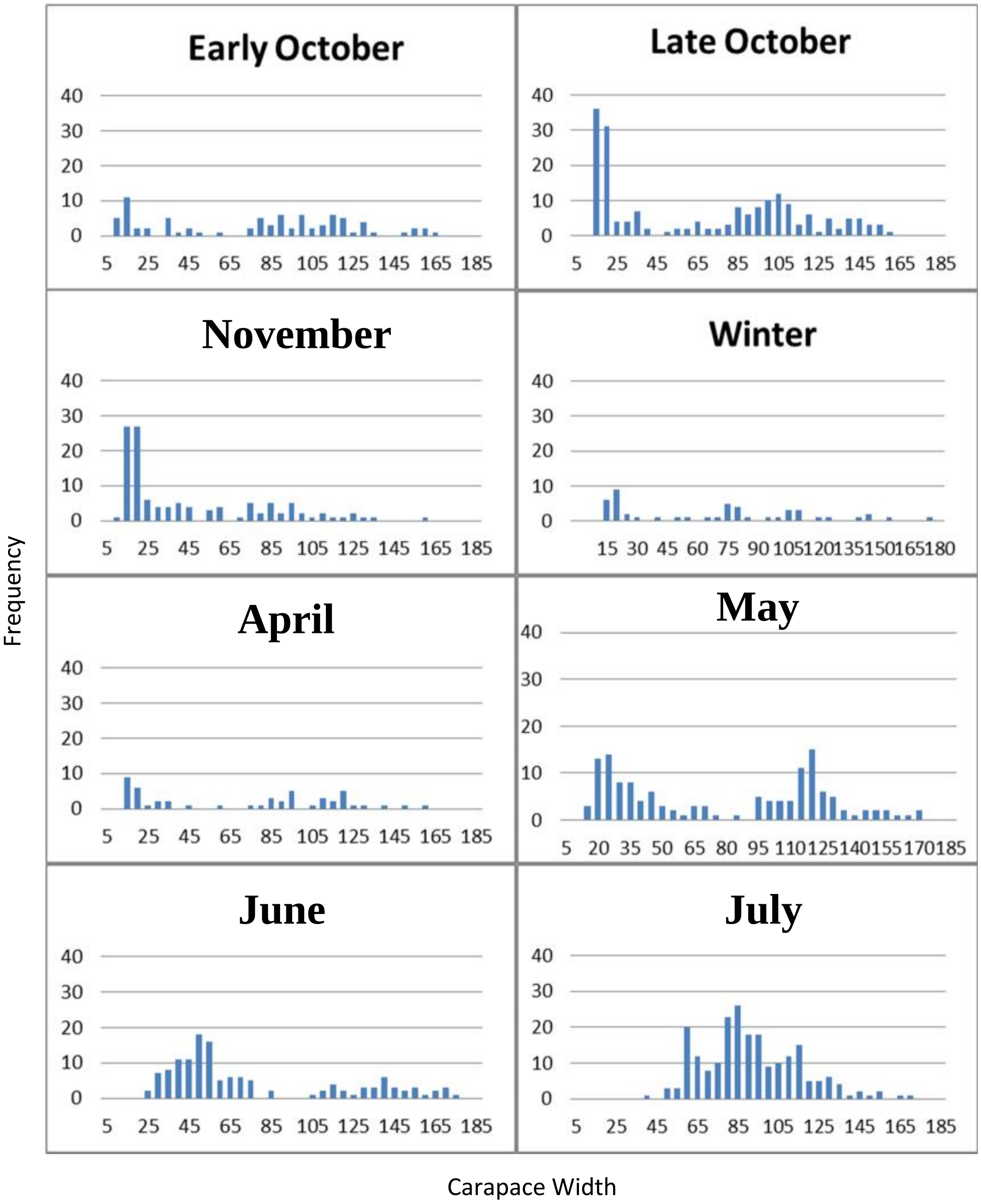

Figure 3.4. Size frequency data for each sampling period used to identify juvenile size cutoffs. For periods 1-7 I used less than 40mm, less than 45mm, less than 50mm, less than 60mm, less than 70mm less than 80mm and less than 90mm, respectively, as the cutoffs. No cutoff was set for period 8.

NBGAM could be used. The density of crabs per tow was modelled as a negative binomial process (Zuur et al. 2009) where

$$g(Crabs) = NB(\mu_{ij}, \theta) \qquad (1)$$

Where g() is some function of the standardized number of crabs in a 75 m tow, NB is the negative binomial distribution, $\mu_{i,j}$ is the mean of the negative binomial process, and θ is the overdispersion process. $\mu_{i,j}$ can be given by

$$\mu_{i,j,k,l} = e^{\beta_0 + \beta_1(\text{Sediment}_i) + f_1(\text{Depth}_j, k=0.4) + f_2(\text{Sample Period}_l, k=0.4) + f_3(\text{Depth}_j\ \text{Sample Period}_l, k=0.4)} \qquad (2)$$

Where β's are fitted parameters for each model factor or interaction of factors. A smoothing factor of k=4 was used for each function within the model. Models defined by equation 1 and 2 were fit using a range of ϴ (Eq. 1). Theta values of 0.1 to 10 by 0.1 were input into the model and the value of ϴ was chosen that minimized the Akaike Information Criterion (AIC) for the model. All models were fit using R and the package mgcv (Wood 2000; 2006)

The best fitting model was then used to predict the number of juvenile blue crabs per 75m tow in each of the 1743 potential sampling sites for each of the eight sampling periods. The density for a given site within a sample period was adjusted by dividing by the maximum density estimate for that sample period; this allows for the observation of relative trends that are not influenced by seasonal changes in the absolute abundance of juvenile blue crabs. These values were then used to create a density heat map (ArcGIS 10.2.1, ESRI Corporation).

Results

The survey was completed in all months. In general there is an increase in juvenile blue crabs from early October through November (Table 3.1). Juvenile blue crabs densities decreased from December – May and then increased from May through June. While samples were taken using the commercial grass scrape in the winter months the data was not included in the models since the gear was not able to effectively sample the buried crabs.

Negative Binomial Generalized Additive Model (NBGAM).

AIC values indicated that the best fitting model was achieved with $\theta = 0.4$. Depth, sediment, sample period and the tensor interaction between depth and sample period were all significant predictors of juvenile blue crabs density in the Rhode and West Rivers at the $p=0.05$ significance level (Table 3.2).

Smoothing functions were estimated from the model for the main effects of sample period and depth. The predicted smoother for sample period is nonlinear and multimodal (Figure 3.5), indicating the pulsed pattern of recruitment. The predicted smoother for depth was generally a declining linear function, indicating that crabs were approximately twice as likely at shallow depths (<0.5m) than at 4m (Figure 3.6).

Figure 3.7 shows the coefficient estimates generated from the model for each sediment classification, solid lines, and +/- two standard errors, dashed lines. The Clay/Silt sediment has a coefficient estimate of zero since the parameter was not

Table 3.2. The model summary for the Negative Binomial Generalized Additive Model.

Family: Negative Binomial(0.4) Generalize Additive Model
Link Function: Log
Formula: CrabsAdj ~ ti(Sample Period, k = 4) + ti(Depth, k = 4) +
ti(Sample Period, Depth, k = 4) + Sediment

Coefficient	Estimate	Standard Error	T Value	P Value
Intercept	-0.863	0.106	-8.146	<0.0001
Sediment-Sand	0.682	0.171	3.983	<0.0001
Sediment- Silt/Sand	0.689	0.262	2.635	0.0084
Tensor Interaction Smoother Terms		Estimated Degrees of Freedom	Chi Square value	P Value
Sample period		2.864	46.88	<0.0001
Depth		1.000	13.41	0.0003
Sample Period x Depth		2.708	16.66	0.0008
R-Square(adjusted)	0.187			

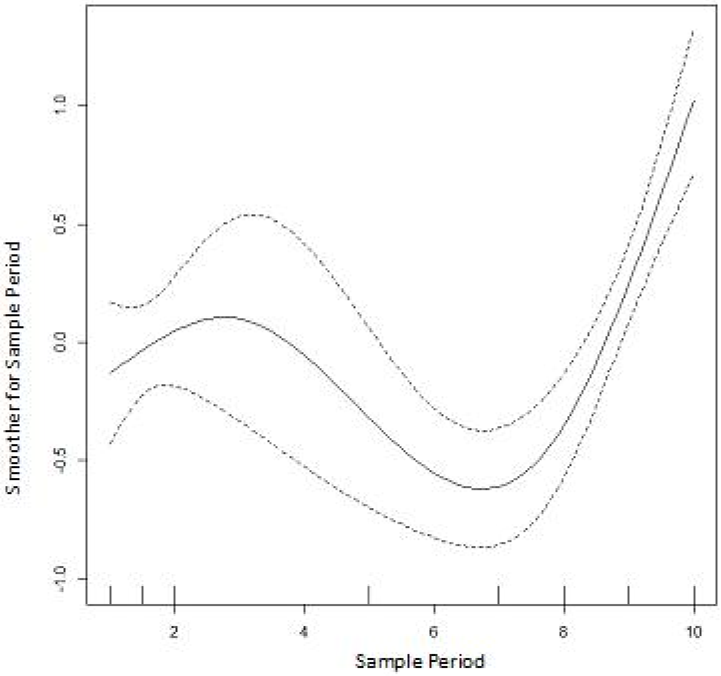

Figure 3.5. Smoothing function (solid line) for sample period estimated from the best fitting negative binomial general additive model. The x-axis shows the sampling period, with the tick marks indicating the values used in model fitting. The y-axis indicates the relative impact of sample period on the standardized density of blue crab relative to the mean (0-value). The dashed lines are ± 1 SE of the mean.

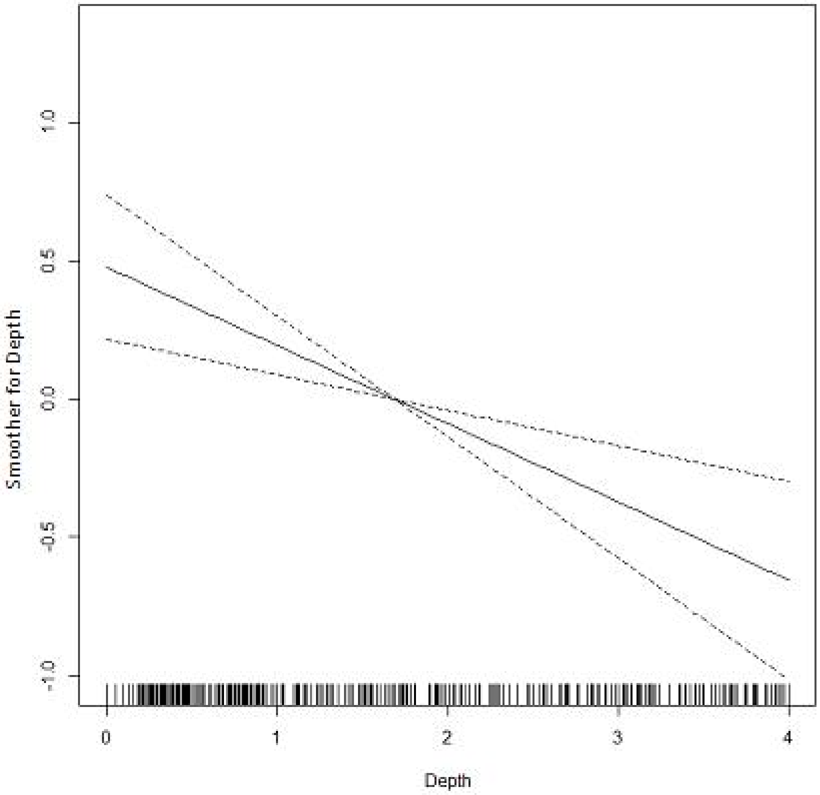

Figure 3.6. Smoothing function (solid line) for depth estimated from the best fitting negative binomial general additive model. The x-axis shows the sample depth, with the tick marks indicating the values used in model fitting. The y-axis indicates the relative impact of depth on the standardized density of blue crab relative to the mean (0-value). The dashed lines are ± 1 SE of the mean.

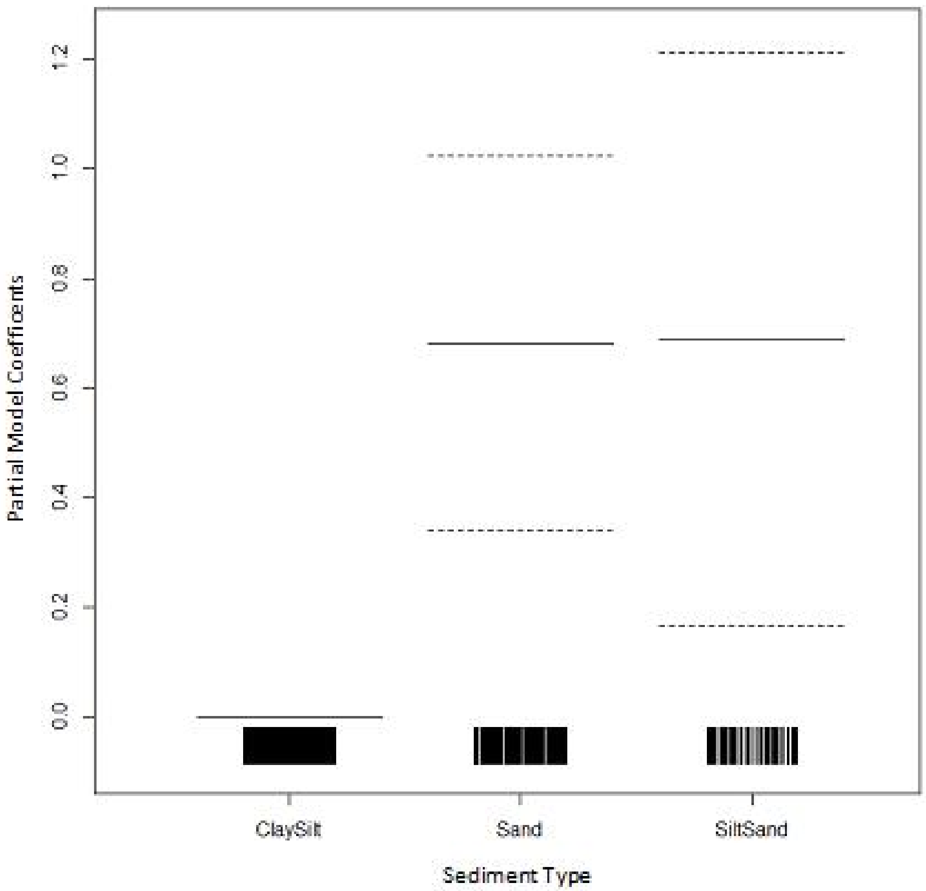

Figure 3.7. Plot of the sediment coefficient estimates and 95% confidence intervals generated from the Negative Binomial Generalized Additive Model.

estimated. The dashed vertical lines on the x-axis represent the sediment type of each site sampled across all sample periods.

The best fitting model was used to predict the density of juvenile crab in all potential grid cells in all sampling periods (Table 3.3). Relative density estimates were plotted as heat maps for each sampling period (Figures 3.8 – 3.15). These maps indicated that juvenile crabs are relatively evenly distributed in the Rhode / West River system in the first three sampling periods (October – November). Thereafter, the distribution maps indicate higher densities of blue crab in shallower water, and in more up river sites further from the mouth of the West River.

Discussion

The main objective of the seasonal study was to quantify seasonal patterns of juvenile blue crabs distribution and abundance with particular interest in depth utilization during winter. This was accomplished using model based estimates generated from data collected during a stratified survey taking place from October 2010 to July 2011. The key findings of this study were (1) Depth, bottom sediment type, sample period and the interaction between depth and sample period were significant predictor of juvenile blue crabs density. (2). It appears that as depth increases the density of juvenile blue crabs decreases; more than 50% of all juvenile blue crabs were predicted to be in waters less than 1.6m, currently the minimum depth sampled by the Winter Dredge Survey (WDS), across all sample periods (Figure 3.16).

Table 3.3. Minimum and maximum predicted crabs per 75m^2 for each sample period, generated by the Negative Binomial Generalized Additive Model. *In the winter sampling period only data collected by the small scale dredge were used in the analysis.

Sampling period	Minimum predicted crabs per 75m^2	Average predicted crabs per 75m^2	Maximum predicted crabs per 75m^2
Early October	0.29	0.53	1.21
Late October	0.33	0.59	1.33
November	0.36	0.64	1.44
Winter*	0.17	0.47	0.98
April	0.10	0.38	0.73
May	0.11	0.52	1.00
June	0.18	0.98	1.97
July	0.34	2.24	4.70

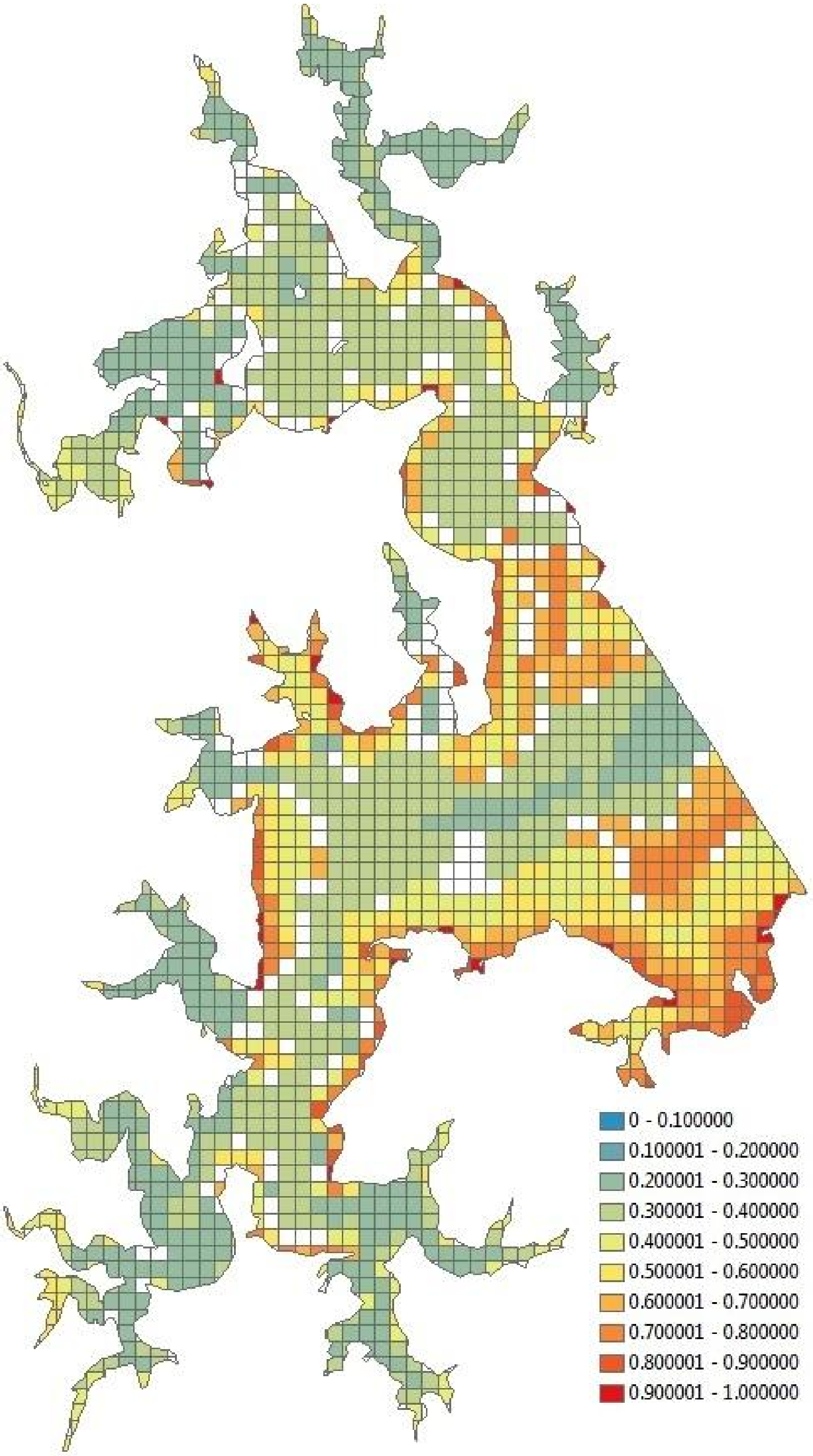

Figure 3.8. Heat map of the predicted relative density of juvenile blue crabs for early October.

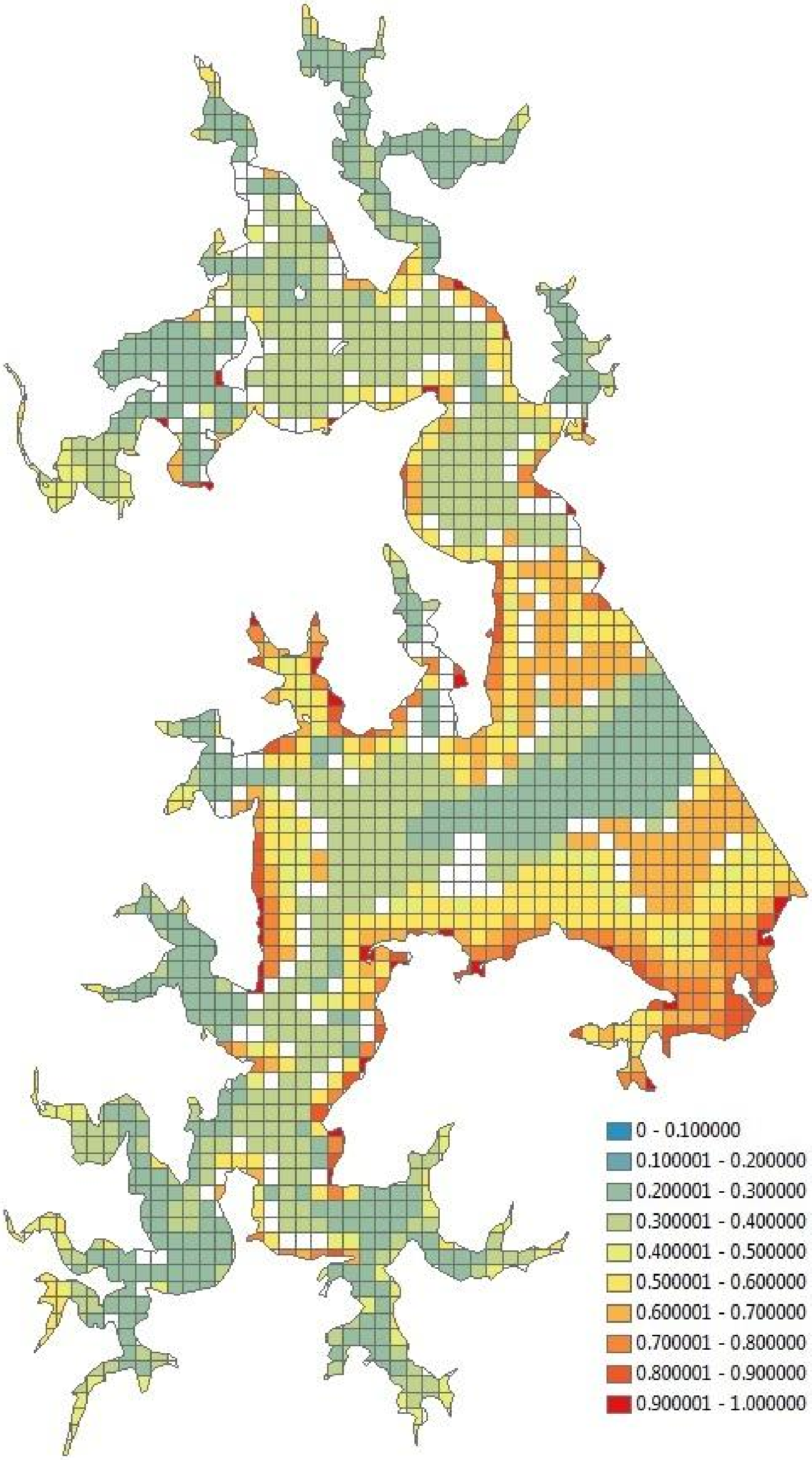

Figure 3.9. Heat map of the predicted relative density of juvenile blue crabs for late October.

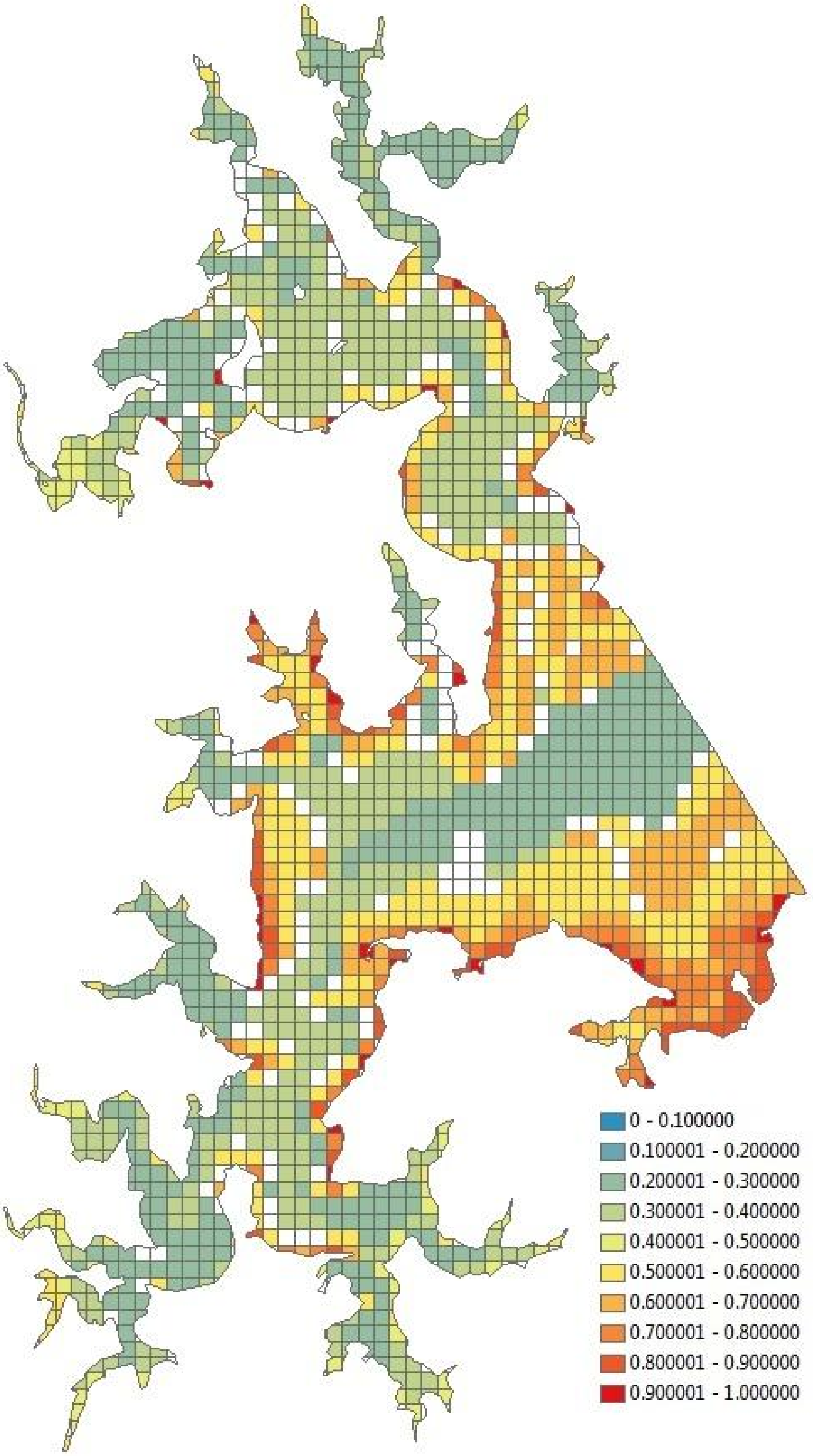

Figure 3.10. Heat map of the predicted relative density of juvenile blue crabs for November.

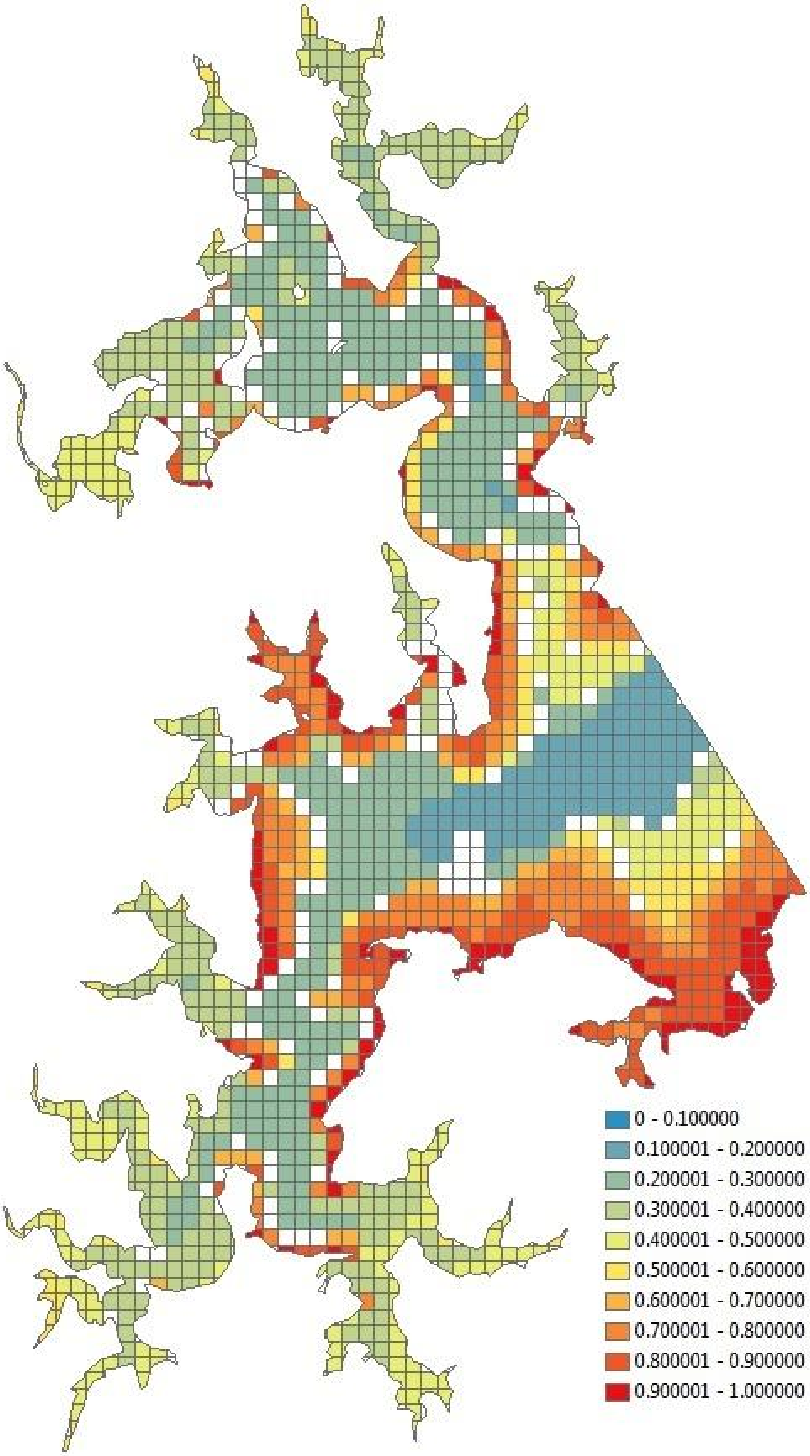

Figure 3.11. Heat map of the predicted relative density of juvenile blue crabs for the winter sampling period.

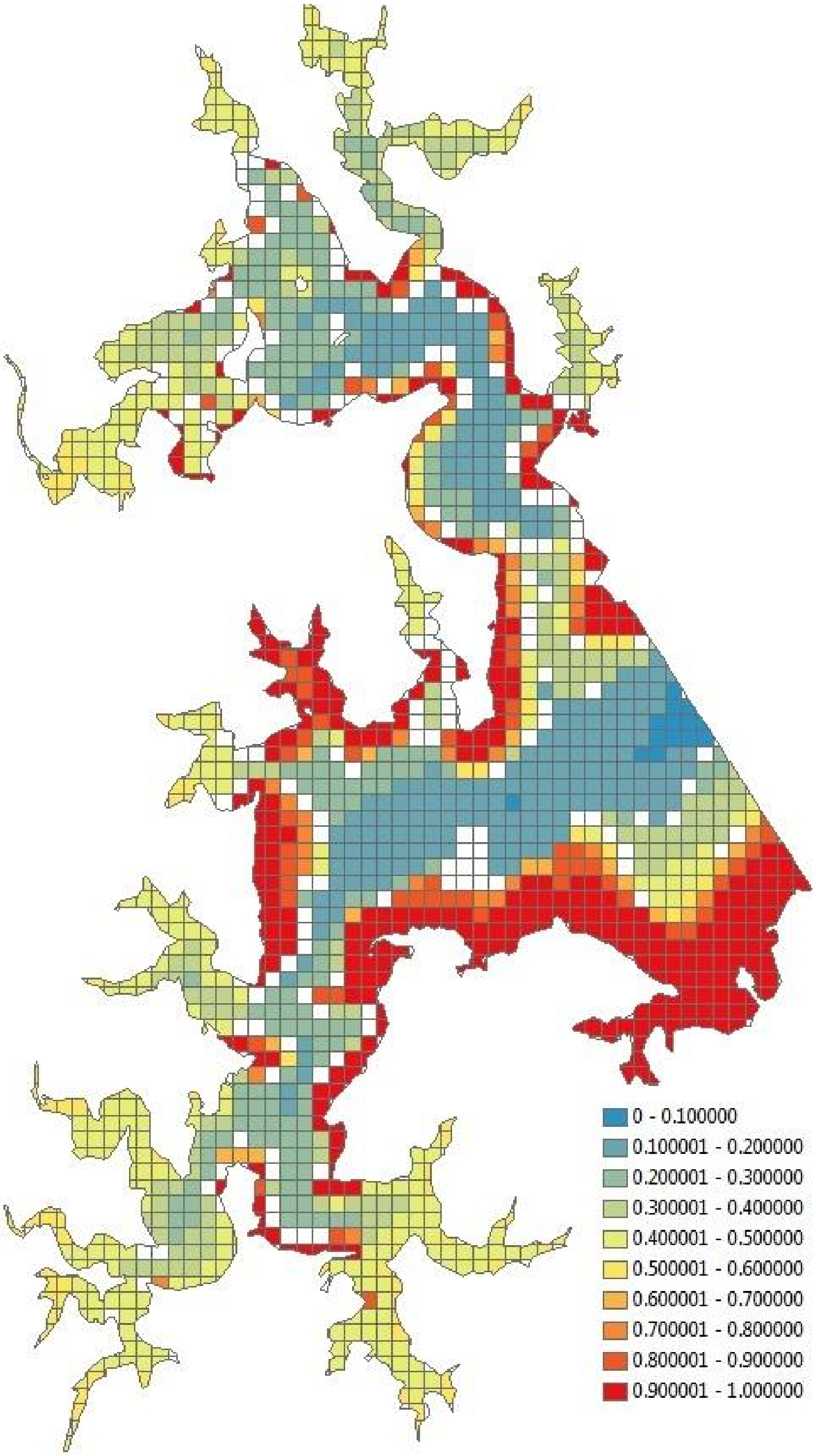

Figure 3.12. Heat map of the predicted relative density of juvenile blue crabs for April.

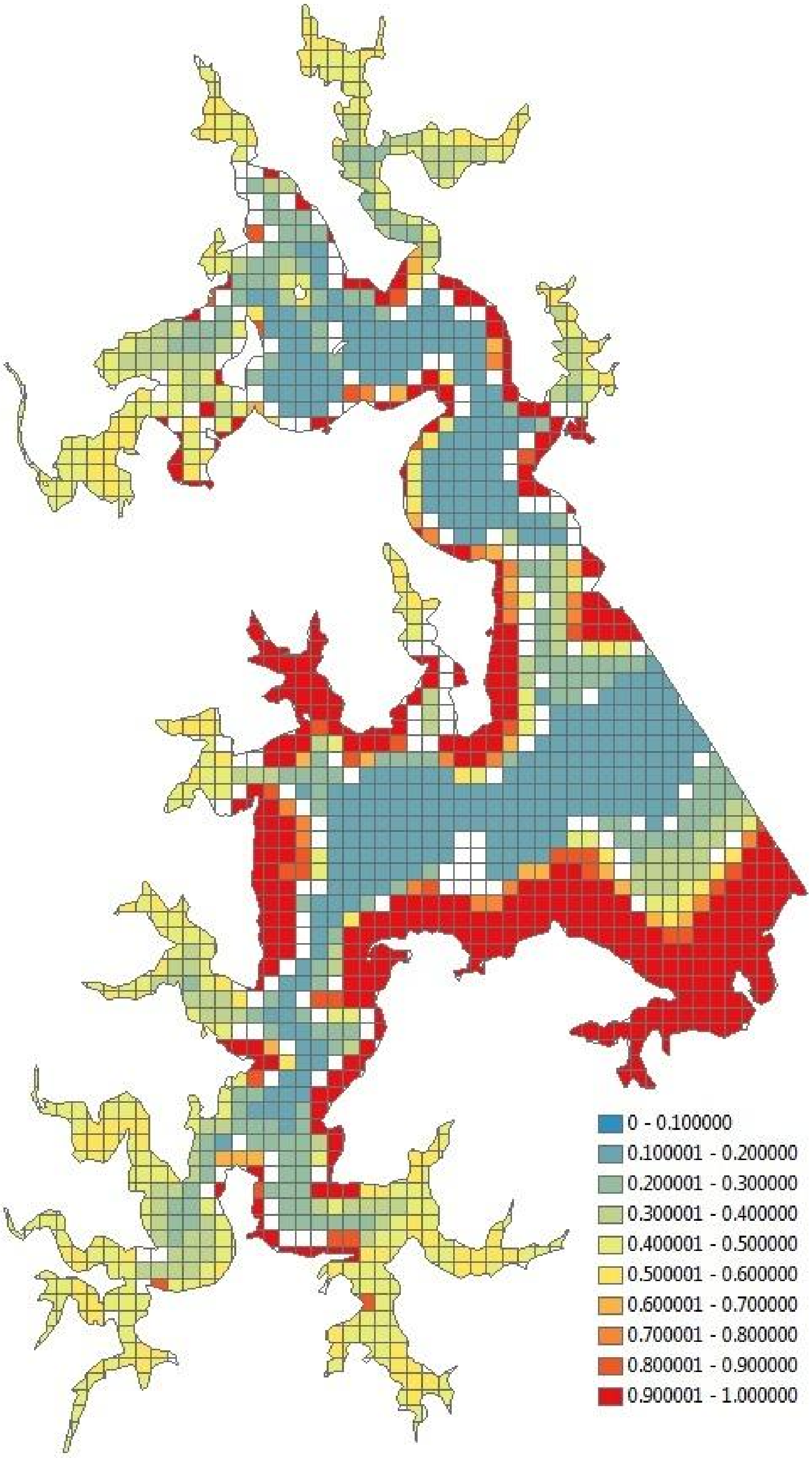

Figure 3.13. Heat map of the predicted relative density of juvenile blue crabs for May.

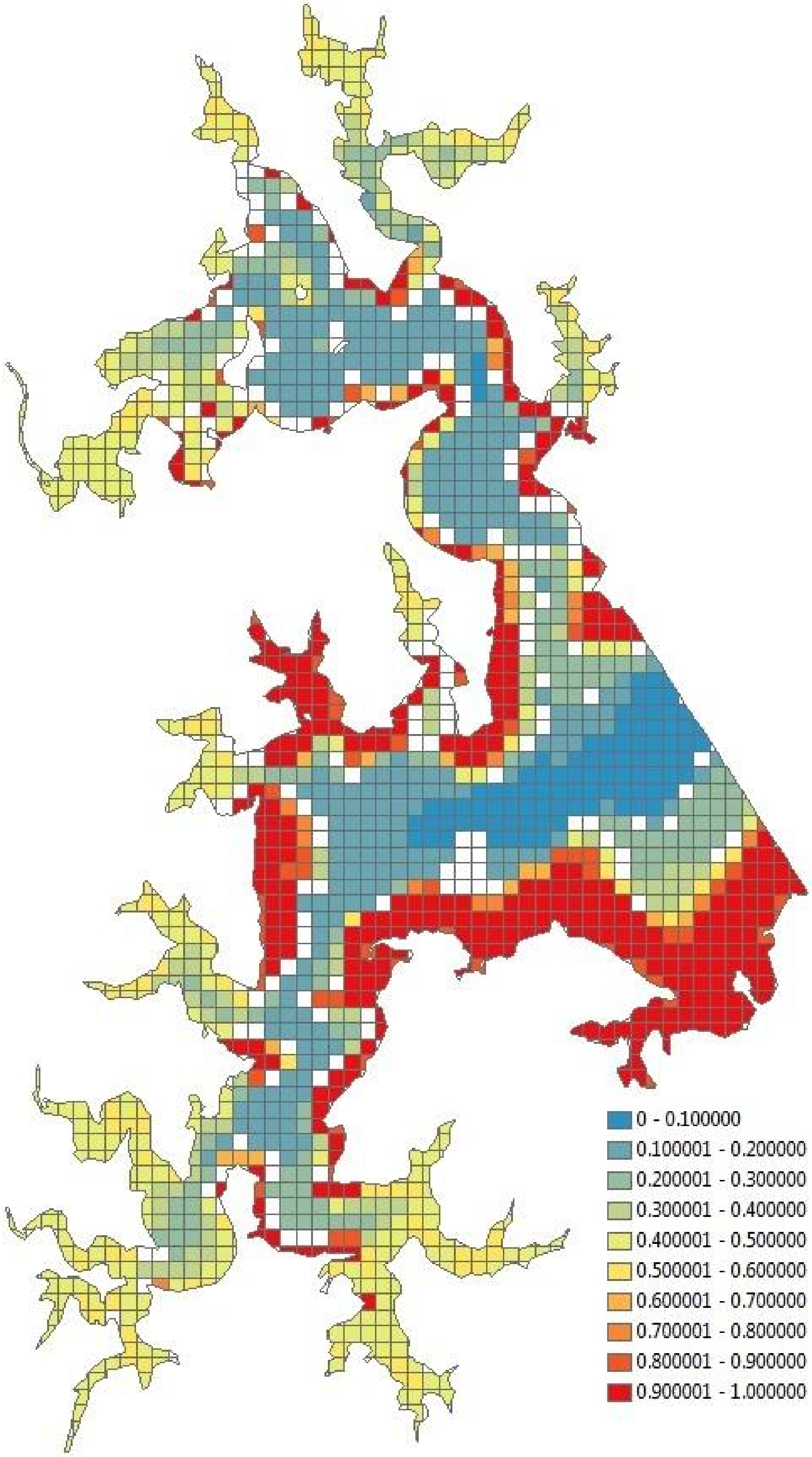

Figure 3.14. Heat map of the predicted relative density of juvenile blue crabs for June.

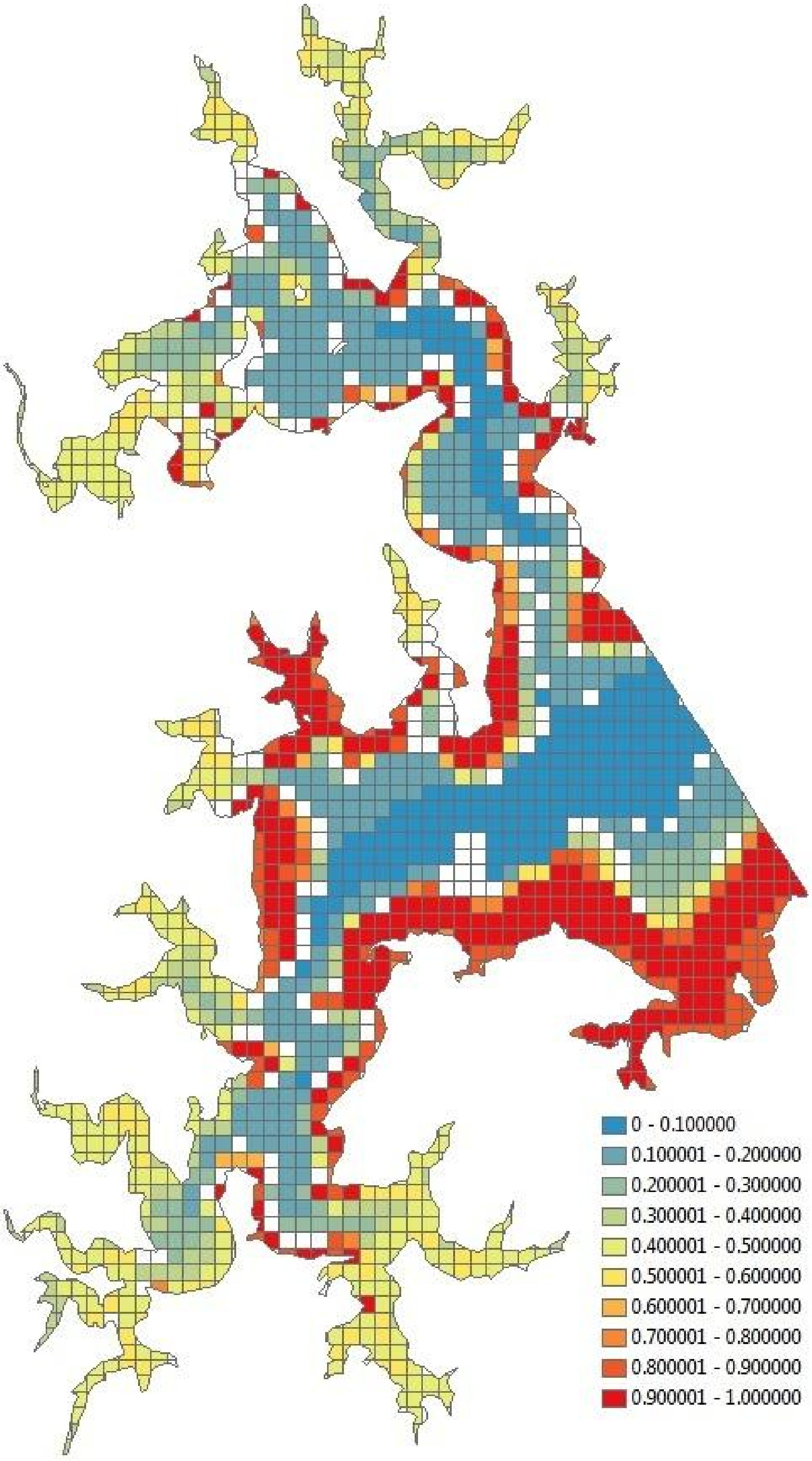

Figure 3.15. Heat map of the predicted relative density of juvenile blue crabs for July.

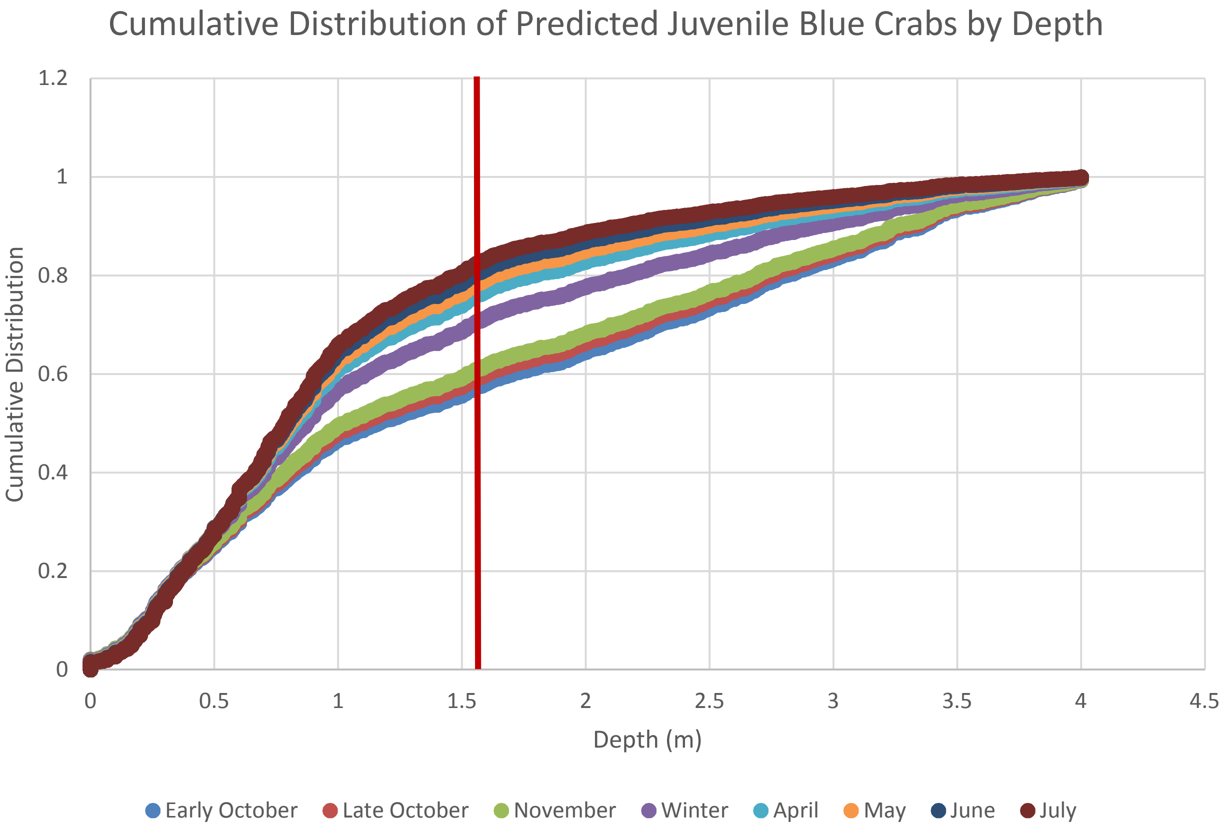

Figure 3.16. Cumulative distribution of predicted juvenile blue crabs by depth in meters. The area to the left of the red vertical line represents depths unsampled by the Winter Dredge Survey.

The need for detailed density estimates dictated the use of model based estimators. It is common in fisheries surveys whose data contains many zeros; this is especially true when encountering the target species is a rare occurrence. Sometimes the data is zero-inflated, meaning there are certain underlying factors that exclude or deter the target species from occupying the sampled area. In an aquatic environment temperature, salinity, and dissolved oxygen can be important factors that determine the habitability. During the seasonal study the temperature, salinity, and dissolved oxygen were measured; however, variations within a sampling period were not enough to suggest these factors were contributing to zero-inflated data. It is most likely that the zeros in the data are explained by relatively low abundances and small area sampled during each tow. It is likely that if the tow distance was significantly increased there would be fewer zeros in the data. This suggests that a single stage model is most suitable for observing spatiotemporal patterns in seasonal density estimates.

Count data is typically non-normally distributed, usually displaying properties of a poisson or negative binomial distribution. Data whose mean and standard deviation are identical generally best described by the poisson distribution. Sometimes the standard deviation is greater than the mean, indicating over dispersion of the data. In that case the data might be best described by the negative binomial distribution. For this study the data is best described by the negative binomial distribution given the relatively small means, large maximum observed counts per tow and large standard deviations for each sample period.

Generalized models can be used when analyzing non-normal data; two common generalized models are the Generalized Linear Model (GLM) and the Generalized Additive Model (GAM). As the name suggests the GLM uses a linear combination of variables to predict a response, whereas the GAM incorporates the use of smoothing functions to describe nonlinear processes. For this study a GAM is most appropriate given the relationship between juvenile blue crabs density and depth and/or time of year the sampling took place is most likely not linear. Since it is likely changes in the depth preferences of juvenile blue crabs is not uniform across all months sampled a tensor interaction was included in the model. Ideally a tensor interaction would be included to examine the interaction between sediment type and sample period, unfortunately the bottom sediment type was not diverse enough to support an additional tensor interaction. Therefore, we must assume that, for a given depth, the relative density of juvenile blue crab for each sediment type is consistent across all sample periods. This poses a significant problem if the juvenile blue crabs seek specific substrate depending of the time of year. It is possible that juvenile blue crabs prefer one substrate in the summer and move to a softer substrate just before overwintering. Without the ability to better classify specific sediment types a tensor interaction cannot be included in the model; therefore, no statements can be made about changes in the juvenile blue crabs sediment type preferences across the sampling periods.

A more complex Negative Binomial Generalized Additive Mixed Model (NBGAMM) can be used to incorporate spatial and temporal autocorrelation. A NBGAMM was used to plot a semivariogram and autocorrelation function. Neither plot suggested any autocorrelation. Therefore the density of juvenile blue crabs can be described by the NBGAM model alone. This implies some interesting behavioral interactions. At least for this study, it appears that the density of crabs within a specific site is uncorrelated to the density of crabs in nearby sites. That is the density can be predicted by the parameters within the model alone. This could be explained by a lack of competition between juveniles. If resources were limited then you might expect to see a degree of spatial autocorrelation. Based on this research alone, the level of resource completion between individuals cannot be surmised.

Even with these limitations the Negative Binomial Generalized Additive Model (NBGAM) is the best option for predicting the spatiotemporal patterns in the density of juvenile blue crabs. The heat maps generated from the model predictions yield some very interesting results that can provide insight into the spatiotemporal patterns of juvenile blue crabs densities within the Rhode and West Rivers. In the first sampling period, early October, the distribution of juveniles appears to be fairly uniform. The sampling for this period took place during the first stages of juvenile recruitment into the Rhode and West Rivers. At this point the crabs have not fully recruited into the river systems and are still very mobile. This pattern is also seen in the late October and November sampling periods.

The winter sampling period was of greatest interest because it was during this sample period the depth distribution of juvenile blue crabs that were overwintering could be observed. For this sampling period juvenile blue crabs were seen in higher densities in the shallow waters near the shore, having moved from the deeper waters where they had been located in previous months. This migration appears to continue through the spring and summer months. This data, broadly supports previous findings that juvenile blue crabs tend to move towards shallow water in the warmer summer months to escape cannibalism from larger adult blue crabs (Dittel et al. 1995; Hines et al. 1995).

One possibility why higher relative densities were seen in the shallow waters after the winter might have been because of a rapid drop in water temperature; perhaps encouraging burial before the preferred depth could be reached. Another explanation might be that the juveniles avoided the extremely shallow waters as a form of thermal refuge. Sampling would need to be repeated over many years to make any conclusive statement about juvenile blue crabs overwintering habits in the Rhode and West Rivers. The most important finding is that in the winter sampling period approximately 70% of all juvenile blue crabs in the Rhode and West Rivers were in waters less than 1.6m, the current minimum sampling depth for the current WDS. In all sampling periods more than 50% of all juvenile blue crabs were located in waters less than 1.6, with the proportion ranging from about 58% to 83% of all juvenile blue crabs in the shallow unsampled waters.

In addition to the trends in depth preferences across months, an interesting trend in the average density estimates was observed. From early October through November the average predicted densities showed a gradual increase. A decrease in the density estimates was observed in the winter and April sampling periods followed by an increase from May through July. While there are many other possible explanations for this observation the most likely reason is we are observing winter mortality followed by continued recruitment from the lower bay into the Rhode and West Rivers. The sudden drop in temperature could have resulted in the juvenile crabs overwintering before they could fully disperse throughout the Chesapeake Bay. If the study were to be repeated I would need to devise a method to observe emigration and immigration to better explain trends in the average density estimates seen within the Rhode and West Rivers.

Even though the model provides interesting results care must be taken when making statements about the results. Firstly the model is limited, not allowing for a tensor interaction between the sediment type and sample period. Another topic that requires further investigation is the choice of size cutoffs for identifying juvenile blue crabs throughout the sampling. When the size cutoffs for classifying juvenile blue crabs were identified the WDS was the base reference. Since the WDS uses a size cutoff of 60mm that was the size cut off used for the winter sampling period. The carapace width frequency data did not provide enough evidence to change the WDS size cutoff for the winter sampling period. Logical size cutoffs were selected for the previous months based on the frequency data, meaning no previous month could have a size cutoff greater than 60mm. This was also done for the sample periods after the winter sample period, making sure no size cutoff was less than 60mm. For the July sample period a clear cutoff could not be identified; because of this any trends observed for this sample period were for all blue crabs not just juveniles. While these cutoffs are fairly arbitrary, they are appropriate given the data collected. There are many reasons why a time varying size cutoff of would be more appropriate. Lower than normal temperatures leading up to winter might reduce the growth of juvenile blue crabs resulting in a smaller cutoff being necessary. However, it is extremely unlikely that an age 1+ crab is smaller than 60mm after a full year of growth. Conversely if the period leading up to the winter was warmer than normal it might facilitate faster growth, necessitating larger size cutoff. Proper size cutoffs are important so that the age 0 and age1+ crabs can be properly identified; improper age class identification will could result in an inaccurate estimate of the proportion of juvenile blue crabs in the areas unsampled by the WDS.

This study yields interesting results that demand further study. A single year of observations is not enough to make broad statements about the spatiotemporal patterns of juvenile blue crabs abundance. A multi-year study is required to make a more generalized statement about the depth and sediment preferences of juvenile blue crabs throughout the year, with an emphasis on the distribution during the overwintering period. If this study was to be repeated it could benefit from a few changes to the design. Firstly there should be no fixed sites sampled, the addition of fixed sites needlessly complicates the analysis. Secondly, given the similarities in gear efficiencies between both gear on active crabs, and the inability for the commercial grass scrape to effectively sample the overwintering blue crabs, only the small scale dredge should be used. Because encountering a juvenile blue crabs while sampling a site was relatively low the area sampled should be increased. This can be accomplished by increasing the sample cell size and increasing the tow distance or by increasing the number of sites. Increasing the tow distance would reduce the number of zero counts in the data, while increasing the number of sites sampled would decrease the standard error of the density estimates. This study could also be completed in other river systems to see if the trends seen in this study are also seen in other rivers around the Chesapeake Bay. With these changes more generalized statements can be made about the spatiotemporal patterns of juvenile blue crabs densities within the shallow waters of the entire bay. Additional information on the spatiotemporal patterns of juvenile blue crabs might lead to a shallow water survey that can be used to generate more accurate indices of abundance for juvenile blue crabs. These indices might be included in future stock assessments to improve management targets.

CHAPTER 4:

PRELIMINARY SHALLOW WATER SURVEY AND COMPARISON TO THE CURRENT WINTER DREDGE SURVEY

Abstract

Annual estimates of blue crab abundance from the Bay-wide winter dredge survey (WDS) provide precise estimates of large blue crabs that will be exploitable during the coming fishery. However, the WDS targets deep water areas (>1.5m) and may underestimate the abundance of very young blue crabs overwintering in shallow waters that are not accessible to the WDS. A coordinated preliminary shallow water survey (SWS) was conducted in four shallow water tributaries in Maryland and compared the abundance of juvenile blue crabs with estimates from nearby (<16 km) sampling locations in the WDS as well as compare estimates of total juvenile abundance in the Maryland waters of the Chesapeake Bay generated by the WDS alone and the SWS and WDS combined. The estimates of juvenile abundance in shallow water tributaries were much greater than those predicted by the WDS data alone. In fact, the actual abundance of juvenile blue crabs in Maryland portions of Chesapeake Bay may be underestimated by nearly 70%. These results have important implications for both stock assessment models and their associated biological reference points and fisheries management for blue crabs in Chesapeake Bay.

Introduction

The blue crab is both economically and ecologically important in Chesapeake Bay where it provides valuable ecosystem services and supports the Bay's most lucrative fishery. The Chesapeake Bay fishery is complex with commercial and recreational sectors, regional variation in fishing gear, multi-jurisdictional management, and a variety of markets including "live hard crab", "soft and peeler" and "processed crab meat" industries (Kennedy et al. 2007). Despite efforts to reduce fishing pressure and improve habitat quality in Chesapeake Bay, blue crab populations declined since the early 1990s and persisted at low levels of abundance with little sign of recovery until 2009 (Lipcius and Stockhausen 2002; Sharov et al. 2003; Chesapeake Bay Stock Assessment Committee (CBSAC) 2010). Most troubling was the precipitous 84% decline in spawning stock abundance in the lower Bay spawning sanctuary (Lipcius and Stockhausen 2002), with concurrent evidence of recruitment limitation in local populations in many areas in the Bay (Jensen et al. 2006; Johnson et al. unpublished data). In response to stock declines and the designation of the stock as "fishery disaster" by NOAA, recent conservation efforts enacted since 2008 have focused on conserving mature females. Mature female crab abundance significantly increased in 2009 and again in 2010 as a result of coordinated management actions to reduce exploitation and favorable environmental conditions. In 2011 there was a slight decrease in the density of spawning females, and in 2012 the decline continued more precipitously, followed by a slight increase in 2013 and in 2014 the density fell to its lowest value since 2002. For the most recent winter dredge survey the density of spawning females increased to a level above what was seen in 1994-2008, but below the high densities reported in 2009, 2010 and 2011.

Due to its economic importance in Chesapeake Bay, the blue crab has been the focus of intense scientific study, fishery-independent monitoring programs, and coordinated regional management. A great deal is known about the ecology and life history of the blue crab (Kennedy and Cronin 2007), providing assessment biologists with valuable life history information necessary for conducting ever more advanced stock assessments (Rugolo et al. 1997; Miller 2001; Miller et al. 2005; Miller et al. 2011). The intensive winter dredge survey (WDS) conducted annually by MDNR and VIMS provides fishery-independent indices of abundance that form the backbone of these assessments (Sharov et al. 2003; Miller et al. 2005; Miller et al. 2011). Fishery-dependent harvest data is collected from the commercial fishery; accurate data on fishery landings is an essential component in assessing the impact of fishing on population dynamics (Hilborn and Walters 1992). In the Chesapeake, blue crab fishery exploitation targets and thresholds are a direct function of removals making an accurate estimate of total landings critical for effective management. As a result, considerable effort has been expended to collect and rigorously analyze harvest data for the blue crab in Chesapeake Bay to ensure reported landings accurately reflect actual removals from the system (Miller and Houde 1999; Fogarty and Miller 2004; Colton 2011; Miller et al. 2011).

To help guide management in maintaining a sustainable fishery, a stock assessment of the Chesapeake Bay blue crab population is conducted, on average, every five years. At the heart of the most recent assessment (Miller et al. 2011) assessment is a sex-specific catch multiple survey model (SSCMSA) that utilizes both fisheries-dependent and -independent data to estimate population abundance and generate biological reference points. Fisheries-dependent data comes from three management agencies; the Virginia Marine Resource Commission, the Potomac River Fisheries Commission and the Maryland Department of Natural Resources. The fisheries-independent data comes from three major surveys; the Virginia juvenile finfish and blue crab trawl, the Maryland DNR trawl survey and the Winter Dredge Survey (WDS) (Miller et al. 2011); however the only WDS occurs Bay-wide and is considered to most accurately reflect actual population abundance (Sharov et al. 2003; Jensen et al. 2006). The WDS started in 1989 as a stratified random sample with the strata being region, bottom sediment type and depth. In 1991, the survey design was changed so that there are only three regional strata which correspond roughly to low, mid and high salinity areas: (1) all tributaries of the Chesapeake and northern bay, (2) middle Chesapeake, and (3) lower Chesapeake Bay. However, the entire bay is not sampled during the winter dredge survey since the survey is only conducted in waters greater than five feet leaving shallow water habitats unsampled. Approximately 1200 stations are sampled every winter with the sample size in each stratum proportional to the area they cover. A 1.83m wide Virginia crab dredge is used to sample each station. All crabs collected are sexed and sorted into two age classes, age-0 and age-1+. To identify the age class all crabs less than 60mm are classified as age-0 and anything greater is age-1+. Using the WDS catch and gear efficiency data an absolute abundance estimate is then calculated for age-1+ crabs. However, the stock assessment model calculates; which will be referred in this document as the susceptibility coefficient, for age-0 crabs, across all years and adjusts the WDS abundance estimates accordingly (Miller et al. 2005; Miller et al. 2011). The most recent stock assessment estimated that the susceptibility was 0.4, indicating only 40% of all juvenile crabs are susceptible to the WDS, the remaining 60% are not susceptible because they are outside the sampled area or are not captured by the gear within the sampling area since the gear efficiency is less than 100% (Miller et al. 2011). Rothschild et al. 1992 found no significant difference in the densities of juvenile blue crabs in waters not sampled by the WDS.

Unlike previous assessment models, the current SSCMSA model assumes that a fraction of age-0 crabs will recruit to the fishery before the years end (Miller et al. 2011). Thus, there are two potential problems with the current model. The first is the model uses a single mortality value for both age-0 and age-1+ blue crabs. Tethering studies have shown that relative mortality decreases as the crabs grow in size (Hines and Ruiz 1995). Therefore it is possible the natural mortality of juveniles is higher than it is for adults. However, related studies have suggested that smaller juveniles will seek shallow waters as a refuge from predation and cannibalism (Dittel et al. 1995). This makes estimating the natural mortality of juveniles very difficult.

The second potential problem arises if the portion juvenile blue crabs in shallow, unsampled, waters changes year to year. Any juveniles that stay in waters below 1.5m will not be susceptible to the WDS. The current model can cope with this to a certain degree by adjusting using the susceptibility coefficient (q) to scale up the abundances of juvenile blue crabs. However, this is coefficient does not change year to year; this is also confounded by the model using one value of natural mortality for adults and juveniles. If the portion of juvenile blue crabs in the shallow waters varies year to year it becomes difficult to accurately estimate the juvenile blue crabs abundance. If the natural mortality of juveniles is greater than what is assumed then the susceptibility coefficient will be overestimated and as a result the abundance of age-0 blue crabs will be underestimated. Since a portion of juvenile blue crab will be exposed to the fishery in their first year, approximately 60%, it is important to have an accurate estimate of age-0 natural mortality and abundance to allow for a more accurate calculation of the exploitation fraction, of age-0+ crabs, at maximum sustainable yield (MSY).

My research lab coordinated with researchers (Dr. Romuald Lipcius) at the Virginia Institute of Marine Science (VIMS) to conduct a pilot-scale shallow water survey to assess the potential need for an additional shallow water compliment to the WDS and to provide preliminary estimates of sampling variance to aid in survey design if a juvenile survey were to be recommended by management. Four river systems were selected to be sampled in the Maryland waters of the Chesapeake Bay, and VIMS selected four rivers in the Virginia waters of the Chesapeake Bay. In this chapter, two comparisons are made between juvenile blue crabs densities in the four Maryland river systems and the WDS densities.

For the first comparison, a series of contrasts were used to evaluate the differences in juvenile blue crabs densities between the four selected river system in the SWS and the average density of juvenile blue crabs in all WDS sites within 16 km of each river. A Negative Binomial Generalized Linear Model was used to calculate the contrasts. The second comparison involved calculating absolute abundance estimates for the Maryland waters of the Chesapeake Bay using just the WDS data and comparing this to an abundance estimate for the Maryland waters using the combined shallow water survey data and the WDS data. An estimate of the susceptibility coefficient of juvenile blue crabs in the Maryland waters of the Chesapeake Bay was then calculated. I expect that the abundance of juvenile blue crabs in the shallow, un-sampled, Maryland waters of the Chesapeake Bay to be much greater than the waters susceptible to the WDS. Therefore I expect that the susceptibility coefficient I calculate will be less than the value estimated by the current stock assessment model.

Materials and Method

The following methods are for one part of a joint preliminary study whose goal was to assess the potential need for a shallow water compliment to the Winter Dredge Survey (WDS). This survey was conducted along with a group from the Virginia Institute of Marine Science (VIMS). My group was responsible for conducting the survey in Maryland, and VIMS conducted the complementary survey in Virginia. Four rivers were chosen to be sampled and the same small-scale dredge was used from the previous seasonal study to perform the sampling. The rivers selected for this study were all sampled in February and early March; all WDS sites used in the comparisons were sampled between late December and late February.

Selection of rivers

To select the rivers for this study the Maryland area of the Chesapeake Bay was separated into four sections using existing information from the spatial coverage of the WDS (G. Davis, MDNR, Figure 4.1). The lower boundary of the upper bay region was set by connecting the northern points of the mouth of the Magothy and Chester Rivers.

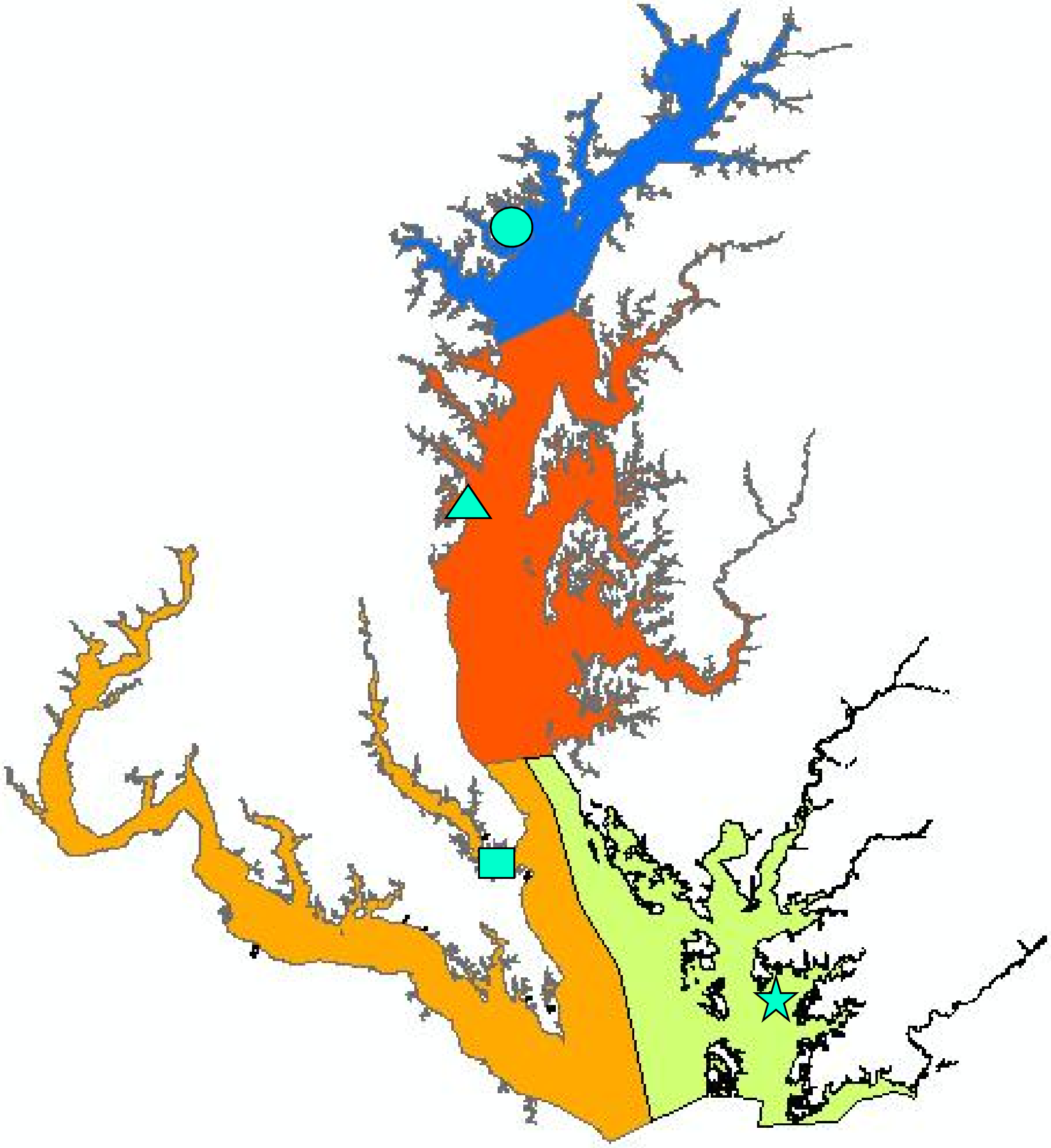

Figure 4.1. The four regions of the Maryland section of the Chesapeake Bay. The blue area is the northern bay represented by the Middle River (Circle), the red area is the middle bay represented by Rhode and West Rivers (Triangle), the orange area is the lower west bay represented by the Patuxent River (Square) and last the green area is the lower eastern bay represented by the Manokin River (Star).

The upper boundary of the lower bay section was the same as the upper boundary of the second stratum of the WDS. The lower section was then split into two; a lower western and a lower eastern section. The lower boundary of the upper bay section and the upper boundary of the lower bay section now provide the boundaries for the middle Bay. One river system was selected in each section from a list of rivers previously sampled during a bay-wide summer juvenile blue crabs survey. This was done because they were familiar locations, and to allow for future comparisons of winter and summer juvenile blue crabs densities in these selected areas. The Middle River was selected to represent the upper Chesapeake Bay, the Rhode and West Rivers were selected to represent the middle Bay, the Patuxent River was selected to represent the lower western Bay and the Manokin River to represent the lower eastern Bay. In each river two replicate creeks were sampled and each creek was divided into the main-stem and tributaries of the creek.

Site Selection and Sampling Methods

Within each creek, the main-stem and adjacent tributary were sampled in 12 random locations using a small-scale dredge (75 m tows). Earlier work has shown that sampling variance was asymptotic at this number of replicate tows in similar shallow cove systems in Maryland. The shoreline of the selected creeks and tributaries was digitized and a random number generator was used to select a point on the length of the shoreline. Six of the twelve sites were near-shore while the other six were in the in the middle of the creek/tributary. A tow direction of up or downstream was also randomly assigned. I used Google Earth to place the sites and capture the latitude and longitude data, and uploaded those coordinates to a handheld recreational GPS unit (Garmin GPSMAP76). These coordinates represent the starting location for each sample tow.

Like the previous study I used a modified commercial grass scrape, which acts as a small scale dredge, and was towed by an 18ft Boston Whaler center console. A standard tow was 75m in length; however, any deviations were recorded in the data. The tow length was measured using a tracking feature on a handheld Garmin GPS unit. Once the tow was complete YSI (YSI Professional Plus, YSI, Inc. Yellowspring, OH) readings of the bottom temperature, salinity and dissolved oxygen were taken. The collection bag was then rinsed before being opened and sorted. For each sampling site any species of fish present were recorded, if the fish was a predator to the blue crab the total length was measured for the first ten individuals. For each crab the sex, carapace width, molt-stage and limb loss was recorded.

Comparative Analysis

A key goal for this work was to facilitate a comparison between densities of juvenile blue crabs in the SWS and the densities found in the WDS. Any blue crab with a carapace width of less than 60mm was identified as a juvenile, this matches the size cutoff for the WDS. Two comparative methods were chosen, the first method simply took the average density of blue crab in each of the four river systems sampled and compare it to the average density of juvenile blue crabs in any WDS sites within a 16 km radius of the river. This range was chosen to allow for an adequate sample size of WDS sites and ensure no WDS site was used in more than one comparison, ensuring we were not extending our range too far and maintain an independence of each rivers comparison. The second method of comparison involved estimating the total juvenile blue crabs abundance in Maryland waters using just the WDS data and comparing that to an abundance estimate from a combination of the shallow water survey and the Maryland waters of the WDS. The current blue crab stock assessment model calculates that the WDS yields an abundance estimate that is only 40% of the true abundance for the entire Chesapeake Bay.

For the first comparison I started by calculating an average density of juvenile blue crabs for each of the four rivers. I used the raw catch per tow data and calculated a density per $1000m^2$ for each sample site, secondly I adjusted the density to account for gear efficiency to get an estimate of catchability corrected juvenile blue crabs density, using the gear efficiency estimates from chapter 2. Following that I identified a point that would become the center of the circle that would be used to identify nearby WDS sites for comparison. The latitude and longitude of the mouth of both creeks sampled in a given river system were identified and the latitude and longitude of the midpoint between the two mouths were used as the identification point for each river system. This midpoint was used to calculate the geographical distance to each of the WDS sampling sites surveyed during the winter of 2011. All sites within 16 km of the river identification point were selected for inclusion in this comparison. The catch per tow of juvenile blue crabs was the converted to juvenile blue crabs per $1000m^2$ for each site. I again corrected for gear efficiency (gear and vessel specific efficiencies were provided by G. Davis, MDNR) to get a catchability corrected estimate of juvenile blue crabs density.

The density of juvenile blue crabs was modelled as a negative binomial process (Zuur et. al 2009) where

$$g(Crabs) = NB(\mu_{ij}, \theta) \quad (3)$$

Where g() is some function of the density of crabs in a 1000m2 area, NB is the negative binomial distribution, $\mu_{i,j}$ is the mean of the negative binomial process, and θ is the overdispersion process. $\mu_{i,j}$ can be given by

$$\mu_{i,j} = e^{\beta_0 + \beta_1(\text{River}_i) + \beta_2(\text{Survey}_j) + \beta_3(\text{River}_i\ x\ \text{Survey}_j)} \quad (4)$$

Where β's are fitted parameters for each fitted model or interaction. The average catchability corrected density of juvenile blue crabs in sites within 16 km was compared to the average catchability corrected density of juvenile blue crabs within the sampled river systems using a series of contrasts. All catchability corrected density values were rounded to the nearest whole crab.

For the second comparison, the absolute abundance of juvenile blue crabs in Maryland waters was calculated using, (1) the WDS alone, and (2) the estimate from the SWS and WDS combined. To begin, all WDS sites in Maryland waters were selected and a density estimate was calculated for each site. Using information obtained from Maryland Department of Natural Resources the WDS spatial coverage for Maryland waters using was charted using ArcGIS (Figure 4.2). The area of three unique sections was then calculated; the first area is the first strata of the WDS, the second area is the second strata of the WDS and the final area is the portion not sampled by the WDS. The area not sampled by the WDS was then split into four subsections based on the four

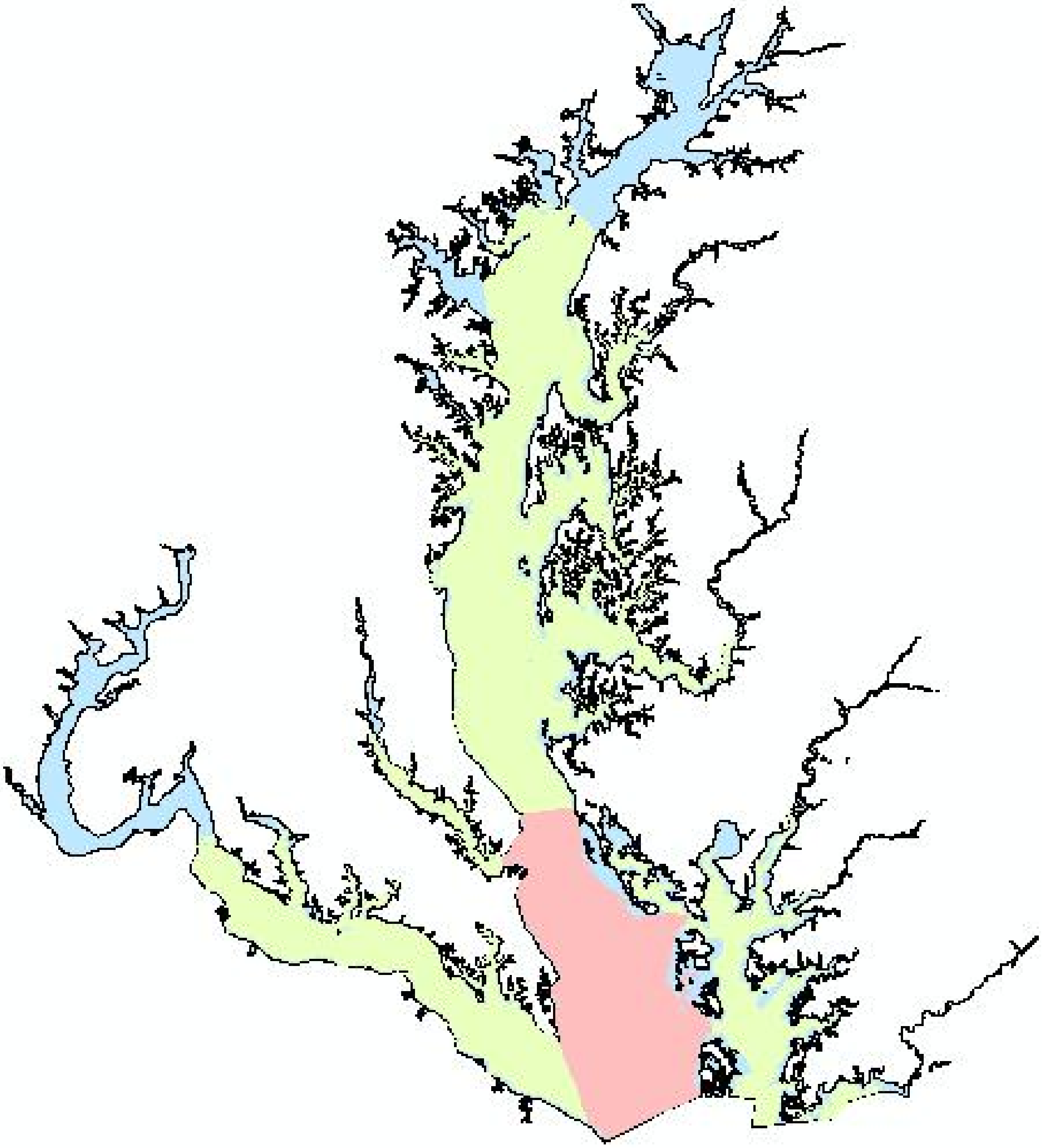

Figure 4.2. The figure above shows the spatial coverage of the Winter Dredge Survey (WDS) in the Maryland area of the Chesapeake Bay. The green area represents the first stratum of the WDS, the red area represents the second stratum of the WDS and the blue area represents area not sampled by the WDS.

quadrants previously defined; each quadrant had a corresponding river system that was sampled to estimate the shallow water density of juvenile blue crabs (Figure 4.1).

The weighted catchability corrected average density of juvenile blue crabs from the WDS data was then calculated, weighting by the WDS strata area coverage. An estimate of total juvenile blue crabs abundance based on the WDS data alone was calculated by multiplying the average catchability corrected density of juvenile blue crabs and the total area of the Maryland waters of the Chesapeake Bay and tributaries. ArcGIS was used to calculate area not sampled by the WDS for each of the four quadrants. The catchability corrected density of juvenile blue crabs estimated for each of the four river systems sampled was multiplied that by their corresponding area of water not sampled by the WDS to get an abundance of juvenile blue crabs in the area not sampled by the WDS. The abundance of juvenile blue crabs in the first and second stratum of the WDS was calculated and the three abundance estimates were then summed to get a total abundance estimate of juvenile blue crabs in Maryland waters. The two estimates of total abundance were then compared.

Results

A total of 192 samples were taken for the Shallow Water Survey (SWS), 48 in each of the 4 rivers sampled. A total of 170 sites in the Winter Dredge Survey (WDS) were within 16km of the rivers I sampled in the SWS. We saw an increase in the observed juvenile blue crabs density moving down the Chesapeake Bay, North to South, in both the SWS and nearby WDS sites. A summary of the raw and catchability corrected densities for both surveys can be seen in Table 4.1. Figures 4.3 to 4.6 show the

Table 4.1 Summary of the raw and catchability corrected crabs per 1000m^2 for each river sampled by the Shallow Water Survey (SWS) all Winter Dredge Survey (WDS) sites within 16km. The minimum and maximum values are rounded to the nearest whole crab.

River	Survey	Sample Size	Raw Mean	Standard Error	Corrected Mean	Standard Error	Minimum	Raw Maximum	Corrected Maximum
Middle	SWS	48	1.39	0.59	5.52	2.38	0	13	53
	WDS	31	0.56	0.27	1.35	0.64	0	5	11
Rhode /	SWS	48	1.67	0.85	6.67	3.42	0	27	107
West	WDS	39	3.89	1.29	9.34	3.10	0	41	99
Patuxent	SWS	48	13.61	4.06	54.35	16.23	0	147	587
	WDS	58	9.57	1.74	22.93	4.17	0	77	185
Manokin	SWS	48	181.10	31.49	724.44	125.95	0	1373	5493
	WDS	42	33.04	5.10	79.26	12.23	0	148	355

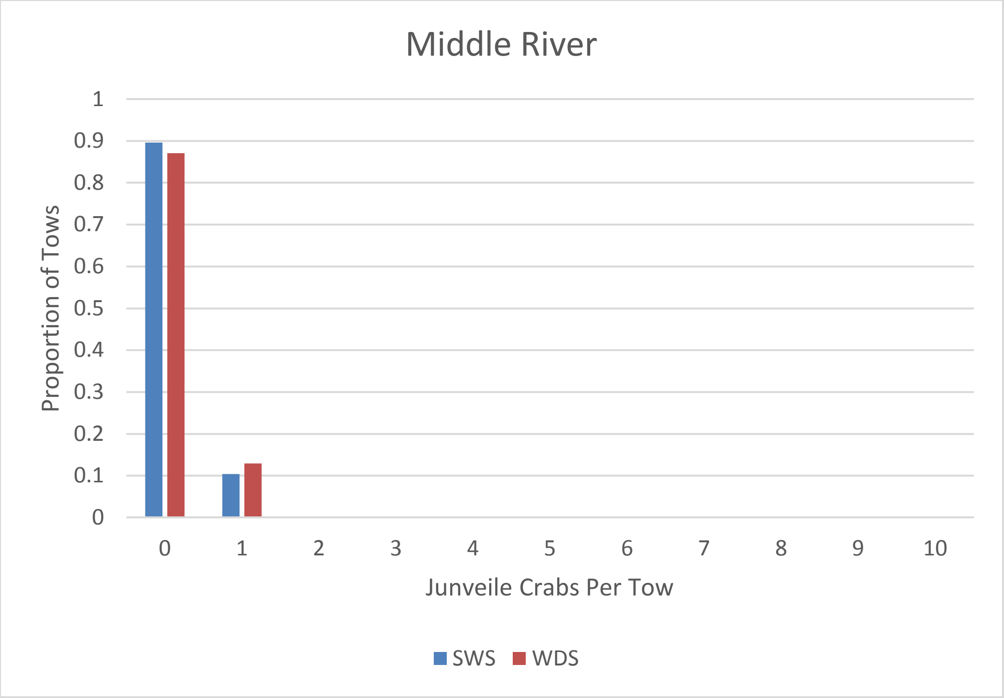

Figure 4.3 Plot of the proportional distribution of the raw juvenile blue crabs caught per tow in both the Shallow Water Survey (SWS) and Winter Dredge Survey (WDS) for the Middle River.

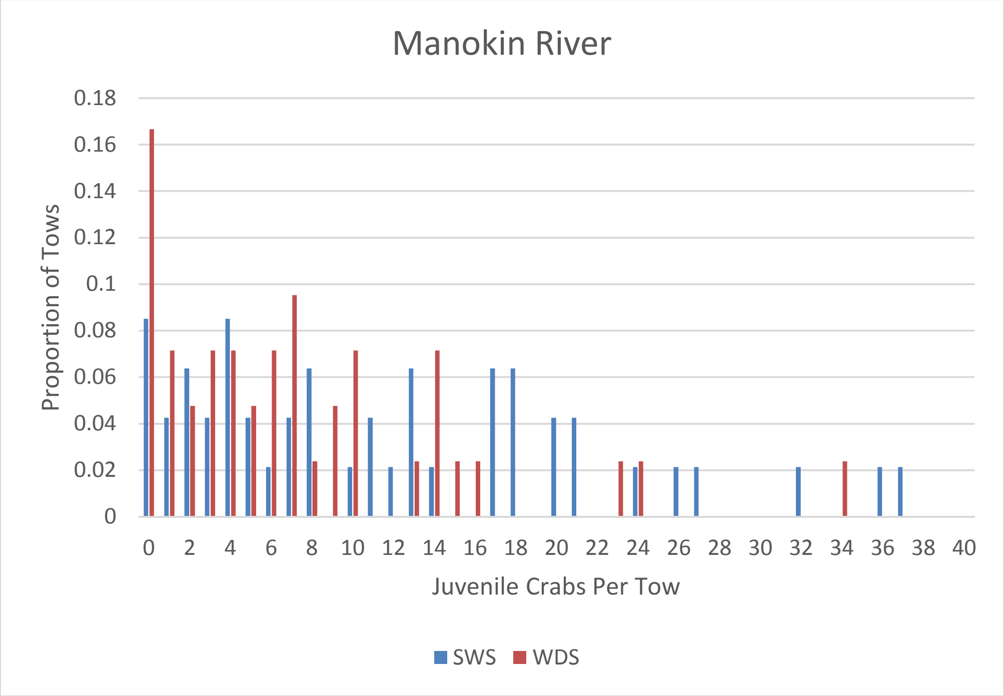

Figure 4.4 Plot of the proportional distribution of the raw juvenile blue crabs caught per tow in both the Shallow Water Survey (SWS) and Winter Dredge Survey (WDS) for the Manokin River. A single tow from the SWS contained 103 juvenile blue crabs and is not included in the proportional distribution above.

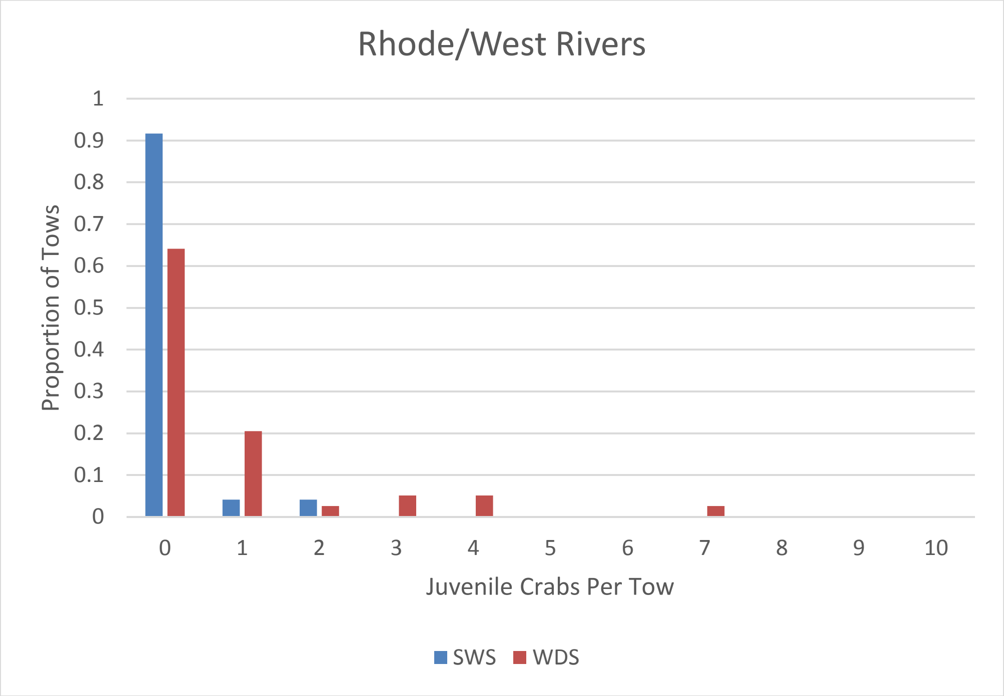

Figure 4.5 Plot of the proportional distribution of the raw juvenile blue crabs caught per tow in both the Shallow Water Survey (SWS) and Winter Dredge Survey (WDS) for the Rhode and West Rivers.

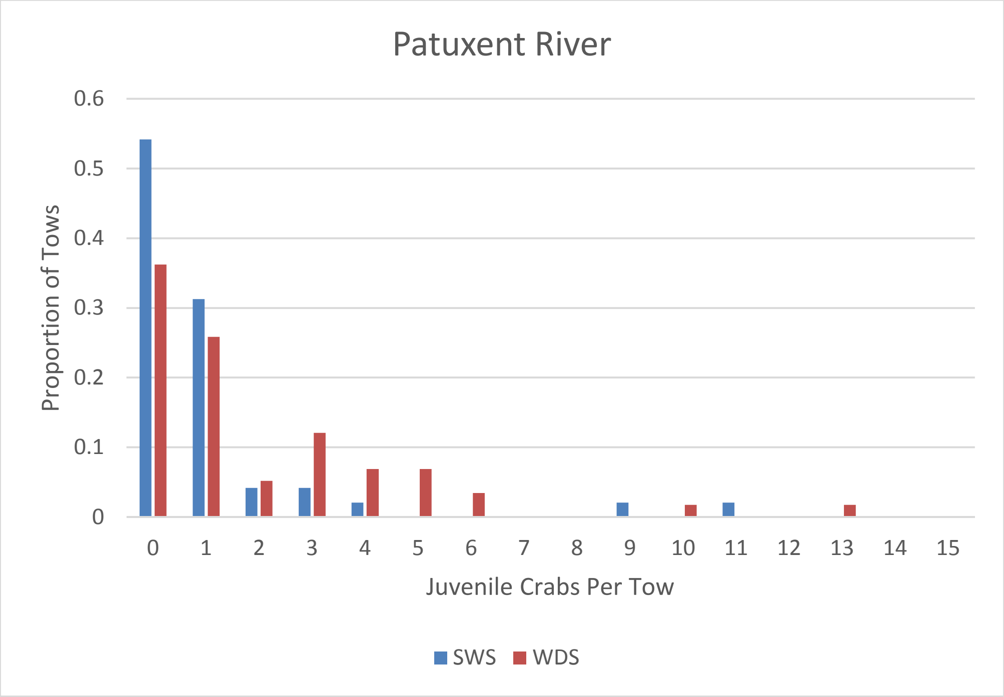

Figure 4.6 Plot of the proportional distribution of the raw juvenile blue crabs caught per tow in both the Shallow Water Survey (SWS) and Winter Dredge Survey (WDS) for the Patuxent.

proportional distribution of the raw catch per tow of juvenile blue crabs in both the SWS and WDS. In all of the rivers sampled, except for the Manokin, a majority of tows contained zero crabs. In general the distributions widened the further south the river is located. One of the samples in the Manokin River contained 103 juvenile blue crabs and is not included in the frequency distribution but is used in the two comparative methods.

In the Middle River both the SWS and nearby WDS sites contained no more than one crab per tow. In the Rhode/West and the Patuxent Rivers the WDS had a slightly wider distribution than the SWS but in general the distributions are very similar. In the Manokin River the distribution of juvenile blue crabs catch per tow for both the SWS and the WDS were more uniform than in the other rivers. The SWS appeared to be more dispersed than the WDS and contain relatively fewer 0 catch tows. Overall the proportional distributions appear to show the properties of a negative binomial distribution.

Comparison of Shallow Water Survey to nearby Winter Dredge Survey sites

For the first method of comparison a series of contrasts generated from a Negative Binomial Generalized Linear Model (NBGLM) (Eq.3, 4) were used. The interaction between river sampled and survey was found to be significant at the 0.05 level, a summary of the model can be seen in Table 4.2. This indicates at least one combination of river and survey differed from another combination. This interaction produces 28 different comparisons between unique combinations of river sampled and survey method. However, only four combinations that compare the two surveys within each river are of importance (Table 4.3). Of the four contrasts only the Manokin and Middle rivers shows

Table 4.2. The model summary for the Negative Binomial Generalized Linear Model used to generate the contrasts necessary to compare the rivers sampled by the Shallow Water Survey (SWS) and nearby Winter Dredge Survey (WDS) sites.

Family: Negative Binomial(0.6) Generalize Linear Model
Formula: CrabsAdj ~ River + Survey + River x Survey

Parameter	Estimate	Standard Error	Wald Chi-Square	P Value
Intercept	2.231	0.404	30.51	<.0001
River- Manokin	2.142	0.559	14.69	0.0001
River- Middle	-1.927	0.623	9.56	0.0020
River- Patuxent	0.902	0.521	2.99	0.0837
SWS	-0.334	0.545	0.38	0.5400
Manokin x SWS	2.546	0.759	11.26	0.0008
Middle x SWS	1.739	0.810	4.61	0.0318
Patuxent x SWS	1.197	0.732	2.67	0.1021
Dispersion	6.254	0.586		

Table 4.3. Results of the selected contrasts comparing the two survey results for each river. * Indicates significance at the p=0.5 level.

Contrast	Chi-Square Value	P Value
Middle	4.74	0.0294*
Rhode/West	0.38	0.5388
Patuxent	3.08	0.0792
Manokin	13.82	0.0002*

significantly different juvenile blue crabs densities between the two surveys. For both significant contrasts the densities in the SWS are greater than the nearby WDS sites.

No significant difference was seen between the average density of juvenile blue crabs in the SWS for the Rhode/West and Patuxent Rivers and the nearby WDS sites. For the Rhode/West River location the SWS had an average catchability corrected density of 6.67 crabs per 1000 m^2 whereas nearby WDS sites had an average catchability corrected density of 9.34 crabs per 1000 m^2. In the Patuxent River the average catchability corrected density was 54.35 crabs per 1000 m^2 for the SWS and 22.93 crabs per 1000 m^2 for the WDS (Table 4.1, 4.3).

A significant difference was seen between the average catchability corrected density of juvenile blue crabs in the SWS for the Middle and Manokin River and the nearby WDS sites. For the Middle River the SWS had an average density of 5.52 crabs per 1000 m^2 whereas nearby WDS sites had an average density of just 1.35 crabs per 1000 m^2. In the Manokin River the average density was 724.44 crabs per 1000 m^2 for the SWS and 79.26 crabs per 1000 m^2 for the WDS (Table 4.1, 4.3).

Comparison of Juvenile blue crabs Abundance Estimates

A weighted average density of juvenile blue crabs in Maryland Chesapeake Bay waters was calculated to be approximately 24.48 crabs per 1000 m^2 based on only the WDS data (Table 4.4). This density results in a juvenile blue crabs abundance estimate of approximately 149,073,775 individuals in the Maryland waters of the Chesapeake Bay. The average density of juvenile blue crabs in unsampled Maryland waters, weighted by

Table 4.4. A summary of the areas and density estimates used to calculate the discrepancy between the Winter Dredge Survey (WDS) juvenile blue crabs abundance estimates and the abundance estimates generated from both the WDS and the Shallow Water Survey.

	Area m2	**Abundance**	**Density Crabs per 1000m2**
WDS Stratum 1	2,983,414,269	911,772,601	30.56
WDS Stratum 2	1,000,371,090	6,344,631	6.34
Total Sampled	**3,983,785,359**	**97521891**	**24.48**
Upper Not Sampled	606,737,366	3,349,190	5.75
Middle Not Sampled	477,108,597	3,182,314	6.89
Lower West Not Sampled	545,881,530	29,668,661	56.32
Lower East Not Sampled	476,175,466	344,960,554	749.36
Total Not Sampled	**2,105,902,958**	**381,160,720**	**180.99**
Total MD Bay Area	**6,089,688,317**	**478,682,611**	**78.61**

regional area coverage, was estimated to be approximately 180.99 crabs per 1000 m^2. The weighted average density of juvenile blue crabs, in the Maryland waters of the Chesapeake Bay using the SWS densities and the WDS sites together, is approximately 78.61 crabs per 1000m^2. This then translates to an abundance estimate of approximately 478,682,611 juvenile blue crabs in the Maryland waters of the Chesapeake Bay. A summary of these findings can be found in Table 4.4. The pilot study yielded an abundance estimate approximately 3.21 times higher than the abundance estimate generated by the WDS data alone. In other words only 31% of the juvenile blue crabs are susceptible to the WDS within the Maryland waters of the Chesapeake Bay; this translates to a susceptibility value of 0.31.

Discussion

This goals of this study were: (1) Compare the densities of juvenile blue crabs in the shallow, unsampled, waters with estimates from nearby (<16 km) sampling locations in the Winter Dredge Survey (WDS) and (2) Compare estimates of total juvenile abundance in the Maryland waters of the Chesapeake Bay generated by the WDS alone and the Shallow Water Survey (SWS) and WDS combined. They key findings were (1) two of the four rivers sampled, the Middle and Manokin Rivers, showed significantly higher densities than nearby WDS sites, and (2) Using both the SWS and the WDS data to estimate the abundance of juvenile blue crabs in the Maryland waters of the Chesapeake Bay resulted in an abundance estimate approximately 3.21 time greater than using the WDS data alone.

However, there are some restrictions to the implications of the results of this study. Future stock assessments for the Chesapeake Bay Blue Crab will rely less on absolute abundances and more on relative indices of abundance. Relying less on estimates of absolute abundance is advantageous given the limitations in calculating reliable gear efficiency estimates, as seen in Chapter 2. A multi-year study would need to be conducted in order to determine if the proportion of juvenile blue crabs in the shallow unsampled waters relative to the juvenile blue crabs in the sampled waters changes from year to year. If the proportion changes annually a separate shallow water survey would need to be conducted to provide an additional index of relative abundance that can be used along with the WDS. Many things learned during this study can be applied to any future shallow water surveys. Firstly, a greater number of regions would need to be sampled within the entire Chesapeake Bay. One major limitation of this study is only 4 river systems were sampled and these rivers might not truly represent of the regions they are located within. Also, a greater number of sites would need to be sampled. A total of 192 SWS sites represented over 2,100 km^2 of unsampled water within the Maryland section of the Chesapeake Bay. Whereas, 826 WDS sites represented just under 4,000 km^2 of sampled water within the Maryland section of the Chesapeake Bay

Even with the limitations this study yielded interesting results. Ideally the Negative Binomial Generalized Linear Model (NBGLM) would have shown an insignificant interaction between the survey methods and river, thus indicating that the proportion of juvenile blue crabs in the unsampled waters relative to the waters sampled by the WDS remains constant across all regions of the Maryland Waters of the Chesapeake Bay. However, because the interaction term was significant the ad hoc contrasts were needed to see which rivers contained significantly different densities between the SWS and WDS. Interestingly the two rivers furthest apart are also the only two rivers where the SWS had a significantly higher catchability corrected density compared to nearby WDS sites.

The Middle River catchability corrected density estimate for the SWS was approximately 4 times higher than that of the nearby WDS sites. The Middle River represented the uppermost region of the Chesapeake Bay and had the lowest density estimates compared to the other rivers within the same survey. Figure 4.3 shows the proportional distribution of the raw count of juvenile blue crabs caught per tow for the SWS sites sampled within the Middle River and nearby WDS sites. Neither survey had a single tow with more than 1 crab caught; this, combined with the relatively low sample size for both surveys, n=48 for the SWS and n=31 for the WDS, suggests that the results might not be truly representative of the region as sampling such a rare event would require a substantially larger sample size.

The catchability corrected density estimate of juvenile blue crabs within the shallow waters of the Manokin River is approximately 9 times greater than the density estimate of nearby WDS sites. The Manokin River represented the south eastern region of the Maryland waters of the Chesapeake Bay and had the highest density estimates compared to the other rivers within the same survey. Figure 4.4 shows the proportional distribution of the raw count of juvenile blue crabs caught per tow. Both surveys show similar patters with the WDS containing proportionally more zero hauls and the SWS containing proportionally more tows containing 15 or more juvenile blue crabs in a single tow. Not represented in the figure is the single SWS tow that contained 103 juvenile blue crabs in a 75m tow; this data is included when creating the model and contrasts

Both the Rhode/West and Patuxent Rivers did not have significantly different catchability corrected densities between the SWS and nearby WDS sites. The Rhode and West Rivers represented the middle of the Maryland waters within the Chesapeake Bay and the Patuxent River represents the south western section of the Maryland waters within the Chesapeake Bay. Figure 4.5 shows the proportional distribution of the raw count of juvenile blue crabs caught per tow within the Rhode and West Rivers. Both surveys show similar patters with the SWS containing proportionally more zero hauls and the WDS containing proportionally more tows containing 3 or more juvenile blue crabs in a single tow. Figure 4.6 shows the proportional distribution of the raw count of juvenile blue crabs caught per tow within the Patuxent River. Both surveys show similar patterns with the SWS containing proportionally more zero hauls and the WDS containing proportionally more tows containing 3 or more juvenile blue crabs in a single tow.

However, comparing absolute densities should be done with caution. Firstly, the SWS sites were sampled up to a month and a half after the WDS sites were sampled. The natural mortality that occurs between the time the WDS sites are sampled and the SWS sites were sampled would reduce the density observed in the SWS. Therefore the differences in abundances between the two surveys would be greater, but without an accurate estimate of winter mortality it is impossible to determine the magnitude of the impact and if the increase in the SWS densities would result in significant contrasts for the Rhode/West and Patuxent Rivers. Additionally, as stated in Chapter 2, the difficulty of calculating an accurate gear efficiency estimate means care should be taken when making inferences based on comparisons of absolute densities.

When comparing the absolute abundance of juvenile blue crabs in Maryland waters a greater abundance was calculated using both the SWS and WDS data than the abundance estimated from the WDS data alone. It could be expected that this trend would also be seen in Virginia waters and potentially with a greater difference in abundance estimates given juvenile blue crabs must pass through this region on their way from the ocean to the upper sections of the bay. This is supported by a long term summer juvenile blue crabs survey that takes place in several rivers around the Chesapeake Bay; in general juveniles are in much greater densities in the Virginia Rivers than in the Maryland Rivers (Johnson et al. unpublished data). This would suggest that the susceptibility coefficient value is lower than predicted by the model and have a greater impact on reference point calculations, specifically the exploitation fraction at maximum sustainable yield (MSY).

The exploitation fraction is calculated by dividing the estimated catch by the sum of age-0 and age-1+ crabs. The age-0 crabs are divided by the susceptibility coefficient to adjust the WDS abundance estimates. To estimate the susceptibility coefficient the current multiple catch-survey model uses a sex specific Ricker stock-recruitment model to predict the combined number of age 0 crabs at time *t* that survived to time *t*+1and the number of age 1+ crabs at time *t* that survived to time *t*+1 to predict the total number of age 1+ crabs at time *t*+1. This abundance estimate is then compared to the abundance estimate generated from the WDS at time *t*+1 and the discrepancy is the susceptibility coefficient for age 0 crabs at time *t*. The model predicts that the WDS generates a juvenile abundance estimate that is about 40% of the true abundance (Miller et al. 2011). However, based on the SWS and the WDS, that in Maryland the WDS abundance estimate is roughly 30% of the true population. This value may be overestimated because of the timing of the SWS versus the WDS. SWS sites were sampled up to a month and a half after the WDS sites. The natural mortality that occurred between the two surveys would result in lower abundance estimates; thus resulting in a larger susceptibility coefficient. Like the previous comparison, without an accurate estimate of winter mortality it is impossible to determine the magnitude of the impact it has on the susceptibility coefficient.

If this trend is also seen in Virginia, some of the discrepancy between this susceptibility coefficient estimate and the stock assessment models susceptibility coefficient estimate could be due to an incorrect natural mortality value for juvenile blue crabs. If juvenile blue crabs experience higher natural mortality the susceptibility coefficient is being underestimated since the proportionally fewer age 0 crabs will survive to the age 1+ size class. If a shallow water survey was added to the WDS there would be no need to estimate a susceptibility coefficient, and therefore any discrepancy would be due to random/observation errors and an incorrect natural mortality rate. Without the need to calculate a susceptibility coefficient the natural mortality for juvenile blue crabs could be estimated, as long as the natural mortality of adults is known or assumed.

CHAPTER 5:

SUMMARY

The blue crab, *Callinectes sapidus,* is an estuarine-dependent species supports both important recreational and commercial fisheries (Kennedy et al. 2007). Because of its economical, biological, and cultural importance the blue crab has been the object of many scientific studies, fishery-independent monitoring programs, and coordinated regional management projects. One of the main goals in the management of blue crabs is the development of advanced stock assessment methodologies. The newest stock assessment, completed in 2011, introduced a new sex-specific catch multiple survey assessment model (Miller et al. 2011). There were two major changes to the previous stock assessment, the first is the separation of age 1+ males and females in the assessment model. The second major change was a partial recruitment of juvenile blue crabs to the fishery.

The Winter Dredge Survey (WDS) is a Bay-wide stratified random sample of waters greater than 1.6 meter. Of all large scale surveys conducted in the Chesapeake Bay, the WDS is considered to provide the most accurate reflection population abundance (Sharov et al. 2003; Jensen and Miller 2006). However, studies have suggested that smaller juveniles will seek shallow waters as a refuge from predation and cannibalism (Dittel et al. 1995). Any juveniles that stay in waters below 1.5m will not be susceptible to the WDS. The current model calculates a susceptibility coefficient (q) to scale up the abundances of juvenile blue crabs not susceptible to the WDS. However, this is confounded by the model using one value of natural mortality. If the natural mortality of juveniles is greater than what is assumed then the susceptibility coefficient will be overestimated and as a result the abundance of age-0 blue crabs will be underestimated. Since a portion of juvenile blue crabs will be exposed to the fishery in their first year it is important to have an accurate estimate of age-0 natural mortality and abundance to allow for a more accurate calculation of the exploitation rate, of age-0 crabs, at maximum sustainable yield.

Three objectives were identified to help assess the need for an additional shallow water survey to compliment the current WDS. Those three objectives are; (1) calculate gear efficiencies for active juvenile blue crabs using two gear types, and one gear type for overwintering juvenile blue crabs, (2) quantify the spatiotemporal patterns of juvenile blue crabs densities in two rivers within the upper Chesapeake Bay with an emphasis on the spatial distribution during winter and (3) conduct a study of the feasibility of a fishery-independent winter survey by sampling the shallow waters of the Maryland section of the Chesapeake Bay for juvenile blue crabs and comparing the density of juvenile blue crabs within the shallow waters to the densities observed in the Winter Dredge Survey.

The first objective is addressed in Chapter 2; a series of gear efficiency experiments were conducted for two gear types: (1) a commercial grass scrape and (2) a modified commercial grass scrape fitted with a tooth bar that allowed it to act in the field as a small scale dredge. To accomplish this a gear efficiency study for both types of gear was conducted on active juvenile blue crabs during the summer using mark-recapture methodologies. Secondly, a gear efficiency study was conducted for the small-scale dredge on overwintering juvenile blue crabs using a series of modified depletion experiments; this component of the study did not include the commercial grass scrape because gear efficiency was assumed to be near zero based on previous observations.

Gear efficiency estimates were consistent across gear type, season, and estimation methodology; my estimates were about 25%. An emphasis was put on ensuring the gear efficiency estimates were accurate; similar studies have been conducted on overwintering blue crabs that have assumptions that may not be valid (Volstad et al. 2000; Sharov et al. 2003). Design changes were made for this gear efficiency study to reduce the risk of violating our assumptions. The most important difference between the methodology presented in this thesis and the methods used by Sharov el al. (2003) is in the role of depletion. The Sharov el al. (2003) methodology uses the rate of depletion to estimate the gear efficiency, however this requires the assumption that all passes happen over exactly the same area every time. The methodology used in this thesis only requires that the sampling gear does not leaved a defined area while the area is depleted. Once depleted, the first pass within the area can be compared to the known density. The assumptions made in this thesis are less likely to have been validated than those made by Sharov et al. (2003). When estimating the gear efficiency of both gear types on active blue crabs a series of assumptions were made that were likely violated. It was assumed that no tagged crabs left the cove during sampling and that the tagged crabs were homogenously distributed within the cove.

Improvements can be made to the methodology used in this thesis to further reduce the likely hood that any of our assumptions will been violated in the future. For estimating the gear efficiency of active juvenile blue crabs the cove where sampling is occurring should be blocked off to prevent emigration and changes should be made to how the cove is sampled to maintain a homogeneous distribution of juvenile blue crabs. For estimating the gear efficiency of overwintering juvenile blue crabs the gear should be better marked so that it can be seen if it is leaving the defined area. Also, the number of trials should be increased.

The second objective is addressed in Chapter 3, the depth distribution of juvenile blue crabs was tracked between October 2010 and July 2011 in the Rhode and West rivers based on a stratified sampling design with depth and bottom sediment type as strata. Size cutoffs for each sample period to categorize the crabs caught into juvenile blue crabs and adult blue crab size classes. A Negative Binomial Generalized Additive Model (NBGAM) was used to predict the density of juvenile blue crabs for each potential sampling site within the Rhode and West Rivers. Heat maps were generated of the relative densities of juvenile blue crabs to facilitate the observation of spatiotemporal patterns in juvenile blue crabs densities, focusing on the depth distribution of overwintering juveniles.

The most important finding was that in all sampling periods over 50% of all juvenile blue crabs in the Rhode and West Rivers were located in waters less than 1.6m. In the winter nearly 70% of all juveniles are located in waters unsampled by the WDS. However, this study was only conducted over a single year and should be replicated over several years and within different river systems in order to make general statements about the depth preferences of overwintering juvenile blue crabs. If the proportion of juvenile blue crabs with the shallow, unsampled, waters changes from year to year this would be strong support for the creation of a new shallow water survey to obtain a better index of abundance for juvenile blue crabs. If this study were to be replicated a few hurdles must be overcome, mainly the lack of a tensor interaction between sampling period and sediment type may be problematic if juveniles change their sediment preference depending on the time of year. Additionally, the relatively small area sampled within the river system resulted in a high number of zeros. This can be overcome by increasing the area covered by each sample and increasing the number of sites sampled.

The final objective is addressed in Chapter 4; for this study a preliminary Shallow Water Survey (SWS) was conducted where two creeks in each of four river systems, selected to be broadly representative of four regions of the upper Chesapeake Bay, were sampled. The average density of juvenile blue crabs found in each river system was compared to the average density of juvenile blue crabs within all nearby winter dredge survey sites (<16 km). An estimate of absolute juvenile abundance was then calculated using combined SWS and WDS data and compared to estimates generated for the WDS alone to determine if the juvenile crab population is underestimated by the current WDS.

Only two of the four rivers sampled showed significantly higher juvenile blue crabs in the shallow waters than nearby WDS sites. Higher density of juvenile blue crabs were found in the unsampled waters of the Middle and Manokin rivers when compared to nearby WDS sites. The juvenile blue crabs density in the unsampled waters of the Rhode/West and Patuxent Rivers was not significantly different than nearby WDS sites. The results of this analysis are mixed and suggest that future studies would benefit from an increase in sample size for each region sampled. In addition to increasing the sample size the number of regions sampled should be increased to better represent the entire bay not just a few small portions.

When comparing juvenile blue crabs abundances in Maryland waters a greater abundance was calculated using both the SWS and WDS data than the abundance estimated from the WDS data alone. The current stock assessment model predicts that the WDS generates a juvenile abundance estimate that is about 40% of the true abundance (Miller et al. 2011). In the stock assessment this value is called the susceptibility and is expressed as q0=0.4. According to the abundances calculated in this chapter, within the Maryland waters of the Chesapeake Bay the WDS generates a juvenile abundance estimate that is 30% of the population abundance calculated from both the WDS and SWS, therefore the q0 value is approximately 0.3. Currently the stock assessment model uses one value for natural mortality for both juvenile and adult blue crabs, however some studies have suggested juvenile blue crabs mortality is higher than that of adults (Hines and Ruiz 1995; Lipcius et al. 2005; Johnson et al. 2011). Some of the discrepancy seen between the estimate of susceptibility calculate here and the susceptibility predicted by the model could be explained by juvenile blue crabs having a higher natural mortality than adults. Care should be taken when interpreting these results, converting from catch to density requires an accurate estimate of gear efficiency. Given the concerns about the accuracies of the gear efficiency estimates and the move away from the use of absolute abundances in future stock assessments, the shallow water survey should be redesigned to better meet the future needs of the stock assessment scientists.

Overall, the results of the seasonal and shallow water survey may indicate a potential need for a bay-wide shallow water survey to compliment the current Winter Dredge Survey. It might be worthwhile to further explore spatiotemporal patterns of juvenile blue crabs densities in the shallow waters of other rivers to see if similar patterns as those in the Rhode and West Rivers are seen elsewhere. These studies should be repeated for several years to determine if the proportion of juvenile blue crabs in the unsampled waters remains constant or if it varies from year to year. If this proportion is time varying it would be strong evidence for a separate shallow water survey to generate an additional index of abundance for juvenile blue crabs. The addition of an additional shallow water survey could improve current stock assessment models and allow for better management of the blue crab stock in the Chesapeake Bay.

Appendix I: Results of Negative Binomial Generalized Additive Mixed Model for the Rhode and West River Seasonal Study

Methods:

The autocorrelation function and vairogram of a Negative Binomial Generalized Additive Mixed Model (NBGAMM), seen in equations 5 and 6, were examined to determine if there was any spatial and/or temporal autocorrelation in the data collected as part of the season study conducted in the Rhode and West Rivers. Crabs represents the number of juvenile blue crabs in a 75m tow. In the event the tow length was not 75m the crab count was corrected and rounded to the nearest whole juvenile crab. A smoothing factor, of k=4, was applied to the sample period term of the equation. Theta values of 0.1 to 10 by 0.1 were input into the model and the most appropriate theta was chosen by minimizing the Akaike Information Criterion (AIC). For the variogram, the distance between two samples was determine from the latitude and longitude of the center of the sampled cells; paired distances were calculated for each sample period. A maximum distance of 2.9km was used for the variogram; this represents half the maximum distance between any two cells sampled in this study.

The abundance of crabs per tow was modelled as a negative binomial process (Zuur et al. 2009) where

$$g(Crabs) = NB(\mu_{ij}, \theta) \tag{5}$$

Where g() is some function of the standardized number of crabs in a 75 m tow, NB is the negative binomial distribution, $\mu_{i,j}$ is the mean of the negative binomial process, and θ is the overdispersion process. $\mu_{i,j}$ can be given by

$$\mu_{i,j,k,l} = e^{\beta_0 + \beta_1(\text{Sediment}_i) + \beta_2(\text{Depth}_j) + f_2(\text{Sample Period}_l, k=0.4)} \tag{6}$$

Where β's are fitted parameters for each model factor or interaction of factors. A smoothing factor of k=4 was used for each function within the model. Models defined by eq 1 and 2 were fit using a range of ϴ (Eq. 5). Theta values of 0.1 to 10 by 0.1 were input into the model and the value of ϴ was chosen that minimized the Akaike Information Criterion (AIC) for the model. All models were fit using R and the package mgvc (Wood 2000, 2006)

Results:

The AIC for the NBGAMM, was minimized at theta= 0.6. The model summary can be found in Table AI.1). The best fitting model indicated that depth, sediment and sample periods were significant predictors of juvenile blue crabs density in the Rhode and West Rivers. The autocorrelation function plot indicated a correlation of 1 at time lag 0, and small but significant correlations at temporal lags 2, 5, and 8 (Figure AI.1). However, the pattern in the autocorrelation function did not indicate the presence of temporal autocorrelation in the data. Similarly, the spatial variogram did not indicate presence of substantial spatial correlation (Figure AI.2).

Table AI.1 The model summary for the Negative Binomial Generalized Additive Mixed Model.

Family: Negative Binomial(0.6) Generalize Additive Mixed Model
Link Function: Log
Formula: CrabsAdj ~ Depth + Sediment + s(Sample Period, k = 4)

Coefficent	Estimate	Standard Error	T Value	P Value
Intercept	-0.331	0.179	-1.850	0.0647
Depth	-0.286	0.085	-3.375	0.0008
Sediment-Sand	0.678	0.184	3.678	0.0003
Sediment- Silt/Sand	0.659	0.288	2.288	0.0224
Smoother term		Estimated Degrees of Freedom	F value	P Value
Sample period		2.695	16.67	<0.0001
R-Square(adjusted)	0.152			

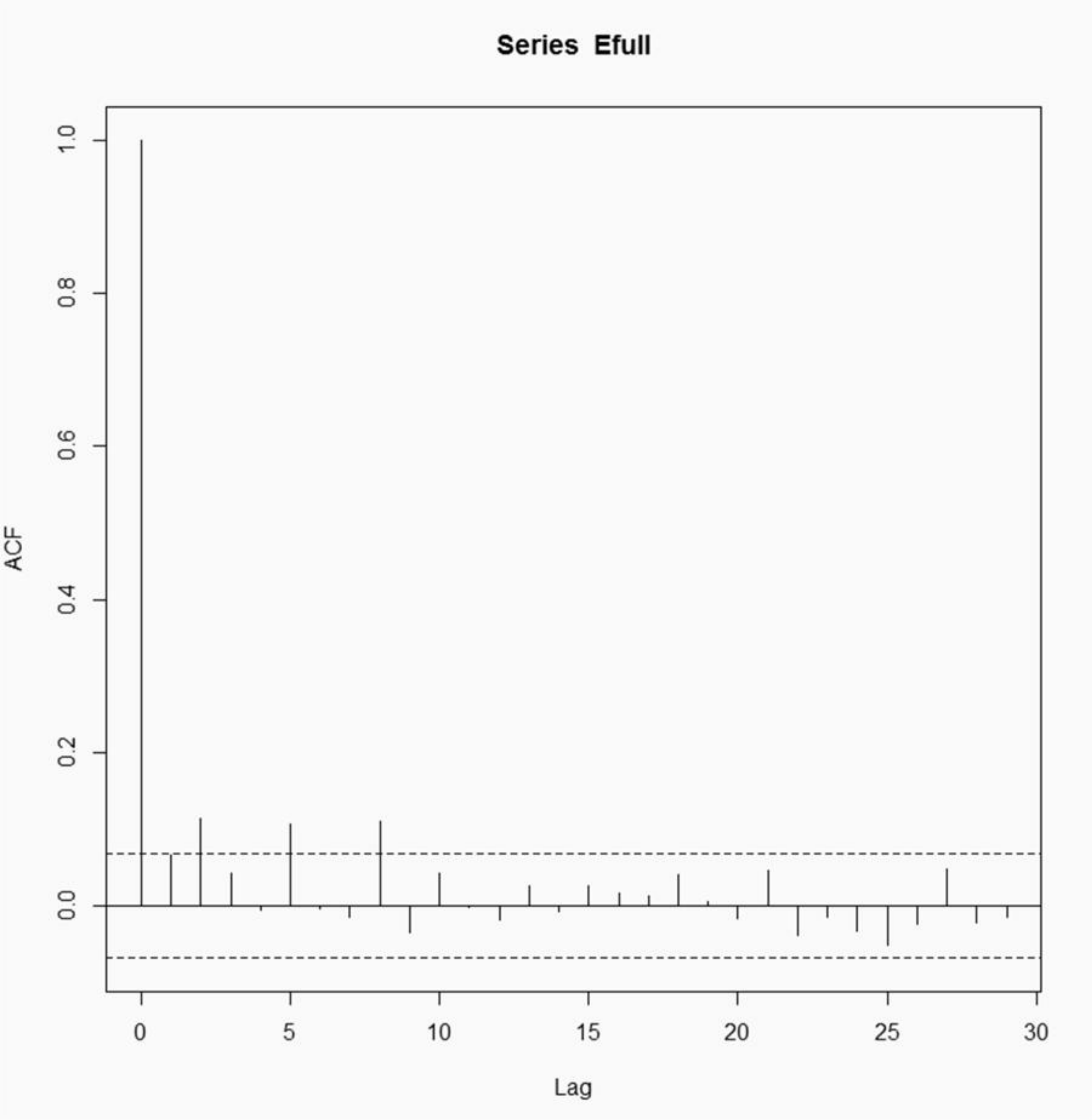

Figure AI.1 The Autocorrelation Function plot generated from the results of the Negative Binomial Generalized Additive Mixed Model. The plot suggests there is no temporal autocorrelation.

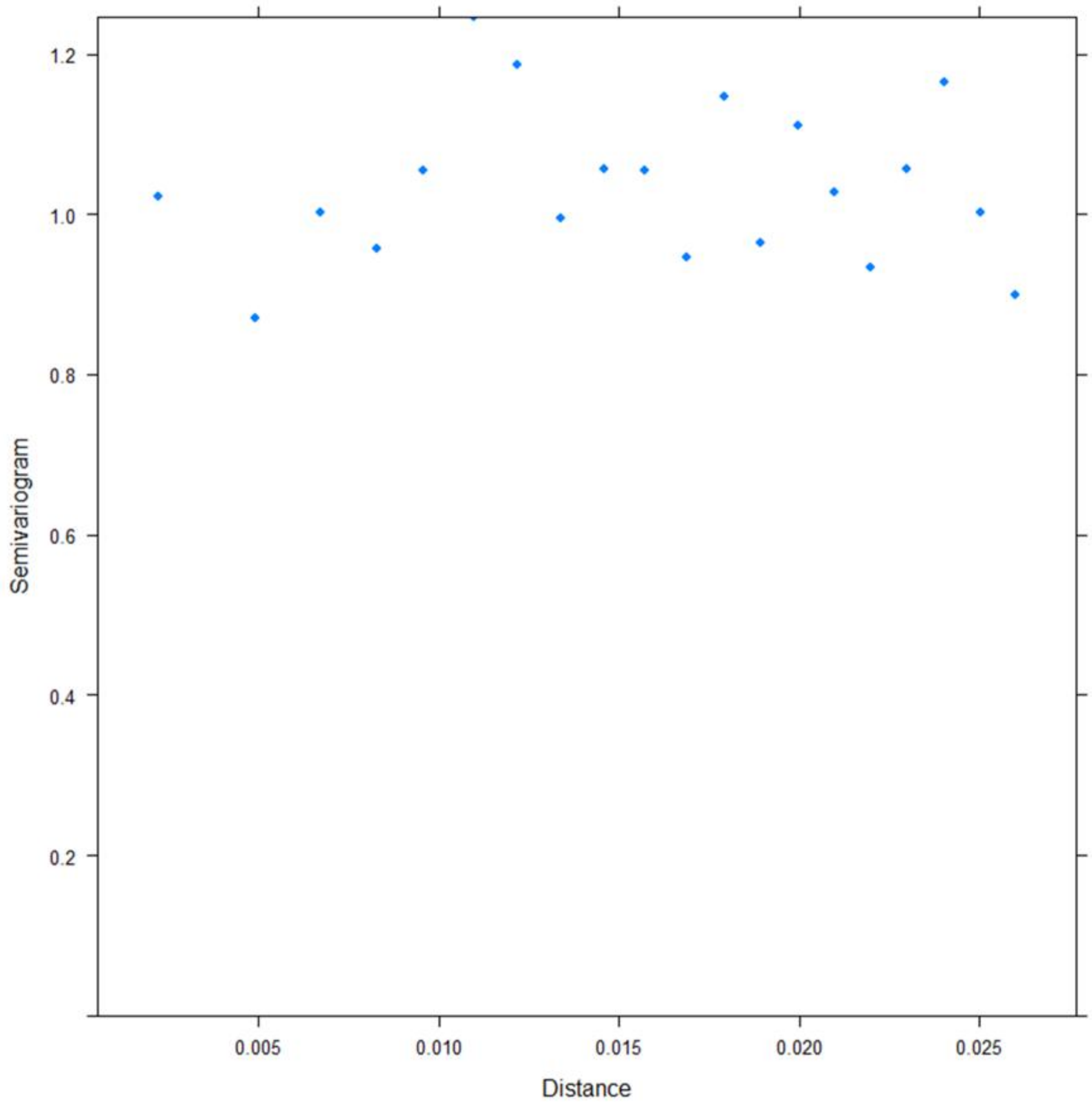

Figure AI.2 Variogram generated from the results of the Negative Binomial Generalized Additive Mixed Model and used to determine the presence or absence of spatial autocorrelation. The plot suggests there is no spatial autocorrelation.

Appendix II: R code for Rhode and West River Seasonal Study

Below is the Code used to observe any spatial or temporal autocorrelation and predict juvenile blue crabs densities for each potential sample site in the Rhode and West Rivers.

```
##### Need packages nlme gstat mgcv MASS
setwd("f:/Masters")

SPmissingdata=read.csv("mastersdata_missing_sp.csv")

######################################################

#### Data with Sample Period by 0.5 month intervals Where October is SP=1 and July is
SP=10 SP=1.5 is for Late OCT Sampling
#### Sled data for winter sampling period = NA since gear efficiency is near 0 and not
estimated

#### Changing negbin by 0.1 to get lowest AIC
AICtest<-NULL

for (i in (1:100)) {

CrabModel1<-gamm(CrabsAdj ~ Depth + Sed + s(SP,k=4),
data = SPmissingdata,
family = negbin((i/10))
)

AICtest[i]<-AIC(CrabModel1$lme)

}

cat(AICtest,sep="\n")

##### AIC minimized at theta=0.6

CrabModel2<-gamm(CrabsAdj ~ Depth + Sed + s(SP,k=4),
data = SPmissingdata,
family = negbin(0.6)
)

#### Look at residuals of model And ACF

M3Resid<-residuals(CrabModel2$gam, type = c("pearson"))
EAll<-vector(length=length(SPmissingdata$CrabsAdj))
```

```
EAll[]<-NA
I1<- !is.na(SPmissingdata$CrabsAdj)
EAll[I1]<- M3Resid
plot(SPmissingdata$SP,EAll)

#### Create ACF to examine temporal autocorrelation

M3Resid2<-residuals(CrabModel2$gam, type = c("pearson"))
I1<- !is.na(SPmissingdata$CrabsAdj)
Efull<-vector(length=length(SPmissingdata$CrabsAdj))
Efull<-NA
Efull[I1]<- M3Resid2
acf(Efull, na.action =na.pass)

#### ACF shows no temporal autocorrelation

#### Create Variogram to determine if there is spatial autocorrelation

CrabModel2Var2<-Variogram(CrabModel2$lme, form=~ Long+Lat|SP, data=
SPmissingdata, maxDist=0.0265)

plot(CrabModel2Var2, smooth=FALSE)

#### since there is no spatial or temporal correlation a GAM is more appropriate than
GAMM

#### Rerun as GAM
#### Add tensor to create"Interaction" between depth and SP

AICtest<-NULL

for (i in (1:100)) {

CrabModel3<-gam(CrabsAdj ~ ti(SP,k=4) + ti(Depth,k=4) + ti(SP,Depth, k=4) + Sed,
data = SPmissingdata,
family = negbin(i/10)
)

AICtest[i]<-AIC(CrabModel3)

}

cat(AICtest,sep="\n")

#### AIC minimized at theta=0.4
```

```
#### Final Model selected

CrabModel4<-gam(CrabsAdj ~ ti(SP,k=4) + ti(Depth,k=4) + ti(SP,Depth, k=4) + Sed,
data = SPmissingdata,
family = negbin(0.4)
)

FinalCrabModelPredCell<-predict(CrabModel4, newdata=SPcelldata, se = T,
type="response")

write.csv(FinalCrabModelPredCell, "FinalCrabModelPredCell.csv")

M3Resid<-residuals(CrabModel4, type = c("pearson"))
EAll<-vector(length=length(SPmissingdata$CrabsAdj))
EAll[]<-NA
I1<- !is.na(SPmissingdata$CrabsAdj)
EAll[I1]<- M3Resid
plot(SPmissingdata$SP,EAll)

M3Resid2<-residuals(CrabModel4, type = c("pearson"))
I1<- !is.na(SPmissingdata$CrabsAdj)
Efull<-vector(length=length(SPmissingdata$CrabsAdj))
Efull<-NA
Efull[I1]<- M3Resid2
acf(Efull, na.action =na.pass)

plot(CrabModel4)
```

Appendix III: SAS Code for Shallow Water Survey

Below is the SAS Code used to create the contrasts used for comparing the Shallow Water Survey juvenile blue crabs densities to nearby Winter Dredge Survey samples.

```
proc import datafile= "E:\SWS.csv"
out=sws
dbms=csv;
getnames=yes;
run;

data sws;
set sws;
crabsadj= round(crabsadj);
run;

proc genmod data=SWS;
class River Sample;
model crabsadj = river|sample/ dist=negbin type3;
contrast 'Manokin' sample 1 -1 river*sample 1 -1 0 0 0 0 0 0 ;
contrast 'Middle' sample 1 -1 river*sample  0 0 1 -1 0 0 0 0 ;
contrast 'Patuxent' sample 1 -1 river*sample  0 0 0 0 1 -1 0 0 ;
contrast 'RhodeWest' sample 1 -1 river*sample  0 0 0 0 0 0 1 -1 ;
run;

proc means data=sws mean median stderr min max;
class river sample;
var crabsadj;
run;

proc import datafile= "E:\SWSRW.csv"
out=swsRW
dbms=csv;
getnames=yes;
run;

data swsRW2;
set swsRW;
crabsadj= round(crabsadj);
run;

proc genmod data=SWSRW2;
class River Sample;
model crabsadj = river|sample/ dist=negbin type3;
contrast 'Manokin' sample 1 -1 river*sample 1 -1 0 0 0 0 0 0 ;
```

```
contrast 'Middle' sample 1 -1 river*sample  0 0 1 -1 0 0 0 0 ;
contrast 'Patuxent' sample 1 -1 river*sample  0 0 0 0 1 -1 0 0 ;
contrast 'RhodeWest' sample 1 -1 river*sample  0 0 0 0 0 0 1 -1 ;
run;

proc means data=swsRW2 mean median stderr min max;
class river sample;
var crabsadj;
run;
```

Literature Cited

Aguilar, R., A. H. Hines, T. G. Wolcott, D. L. Wolcott, M. A. Kramer, and R. N. Lipcius. 2005. The timing and route of movement and migration of post-copulatory female blue crabs, *Callinectes sapidus Rathbun*, from the upper Chesapeake Bay. *Journal of Experimental Marine Biology and Ecology* 319:117-128.

Aguilar, R., E.G. Johnson, A.H. Hines, M.A. Kramer, and M.R. Goodison. 2008. Importance of blue crab life history for stock enhancement and spatial management of the fishery in Chesapeake Bay. *Reviews in Fisheries Science* 16:117-124.

Baird, D., R.E Ulanowicz. 1989. The seasonal dynamics of the Chesapeake Bay ecosystem. *Ecological Monographs*. 59: 329-364.

Bauer, L. J. and T. J. Miller. 2010a. Spatial and interannual variability in winter mortality of the blue crab (*Callinectes sapidus*) in the Chesapeake Bay. Estuaries and Coasts.

Bauer, L. J. and T. J. Miller. 2010b. Temperature, salinity and size-dependent winter mortality of juvenile blue crabs (*Callinectes sapidus*). Estuaries and Coasts.

Brylawski, B. J. and T. J. Miller. 2006. Temperature-dependent growth of the blue crab *(Callinectes sapidus)*: a molt process approach. *Canadian Journal of Fisheries and Aquatic Sciences* 63:1298-1308

Colton, A. R. 2011. *An evaluation of the synchronization in the dynamics of blue crab (Callinectes sapidus) populations in the western Atlantic* (Masters thesis).

Darnell, Z. M., D. Rittschof, K. M. Darnell, and R. E. McDowell. 2009. Lifetime reproductive potential of female blue crabs *Callinectes sapidus* in North Carolina, USA. *Marine Ecology Progress Series*, 394:153-163.

Davis, J. L. D., A. C. Young-Williams, A. H. Hines, and Y. Zohar. 2005. Assessing the potential for stock enhancement in the case of the Chesapeake Bay blue crab (Callinectes sapidus). *Canadian Journal of Fisheries and Aquatic Sciences* 62:109-122.

Dittel, A. I., A. H., Hines, G. M., Ruiz, & K. K. Ruffin. 1995. Effects of shallow water refuge on behavior and density-dependent mortality of juvenile blue crabs in Chesapeake Bay. *Bulletin of Marine Science*, 57: 902–916.

Eggleston, D. B., Johnson, E. G., Kellison, G. T., Plaia, G. R., & Huggett, C. L. 2008. Pilot evaluation of early juvenile blue crabs stock enhancement using a replicated BACI design. *Reviews in Fisheries Science, 16*(1-3), 91-100.

Epifanio, C. E. 2007. Biology of larvae. Pages 513-533 *in* V. S. Kennedy and L. E. Cronin, editors. The Blue Crab, *Callinectes sapidus*. Maryland Sea Grant, College Park, MD.

Etherington, L. L. and D. B. Eggleston. 2000. Large-scale blue crab recruitment: linking postlarval transport, post-settlement planktonic dispersal, and multiple nursery habitats. *Marine Ecology Progress Series,* 204:179-198.

Etherington, L. L., D. B. Eggleston, and W. T. Stockhausen. 2003. Partitioning loss rates of early juvenile blue crabs from seagrass habitats into mortality and emigration. *Bulletin of Marine Science* **72**:371-391.

Fogarty, M. J., & Miller, T. J. 2004. Impact of a change in reporting systems in the Maryland blue crab fishery. *Fisheries research*, *68*(1), 37-43.

Günther C.P. 1992. Dispersal of intertidal invertebrates: a strategy to react to disturbances of different scales? *Netherlands Journal of Sea Research* 30:45 - 56

Hilborn, R., & Walters, C. J. 1992. Quantitative fisheries stock assessment: choice, dynamics and uncertainty. *Reviews in Fish Biology and Fisheries*, *2*(2), 177-178.

Hines, A. H., Lipcius, R. N., & Haddon, A. M. 1987. Population dynamics and habitat partitioning by size, sex, and molt stage of blue crabs Callinectes sapidus in a subestuary of central Chesapeake Bay. *Marine Ecology Progress Series*, *36*, 55-64.

Hines, A. H., & Ruiz, G. M. 1995. Temporal variation in juvenile blue crabs mortality: nearshore shallows and cannibalism in Chesapeake Bay. *Bulletin of Marine Science,* **57**, 884–901.

Hines, A. H., P. R. Jivoff, P. J. Bushmann, J. van Montfrans, S. A. Reed, D. L. Wolcott, and T. G. Wolcott. 2003. Evidence for sperm limitation in the blue crab, Callinectes sapidus. *Bulletin of Marine Science* 72:287-310.

Hines, A. H. 2007. Ecology of juvenile and adult blue crabs. 565 - 654 in V. S. Kennedy and L. E. Cronin, editors. The Blue Crab Callinectes sapidus. Maryland Sea Grant College, College Park, MD.

Hines, A.H., E.G. Johnson, A.C. Young, R. Aguilar, M.A. Kramer, M. Goodison, O. Zmora, and Y. Zohar. 2008. Release strategies for estuarine species with complex migratory life cycles: Stock enhancement of Chesapeake blue crabs, Callinectes sapidus. *Reviews in Fisheries Science* 16(1):175-185.

Jensen, O. P. and T. J. Miller. 2005. Geostatistical analysis of blue crab *(Callinectes sapidus)* abundance and winter distribution patterns in Chesapeake Bay. *Transactions of the American Fisheries Society* 134:1582-1598.

Jensen, O. P., Christman, M. C., & Miller, T. J. 2006. Landscape-based geostatistics: a case study of the distribution of blue crab in Chesapeake Bay. *Environmetrics, 17*(6), 605-621.

Jivoff, P. R., A. H. Hines, and L. S. Quackenbush. 2007. Reproductive biology and embryonic development. Pages 255-298 in V. S. Kennedy and L. E. Cronin, editors. The Blue Crab, *Callinectes sapidus*. Maryland Sea Grant, College Park, MD.

Johnson, D. F. and K. W. Hess. 1990. Numerical simulations of blue crab larval dispersal and recruitment. *Bulletin of Marine Science* 46:195-213.

Johnson, E.G., and D.B. Eggleston. 2010. Population density, survival and movement of blue crabs in estuarine salt marsh nurseries. Marine Ecology Progress Series 407:135-147.

Johnson, E.G., A.H Hines, A.C. Young, M.A. Kramer, M. Goodison, R. Aguilar, and M. Bademan. 2011. Field comparison of survival and growth of hatchery-reared versus wild blue crabs, Callinectes sapidus Rathbun. *Journal of Experimental Marine Biology and Ecology* 402:35-42.

Kennedy, V. S., & Cronin, L. E. (Eds.). 2007. *The blue crab: Callinectes sapidus*. Maryland Sea Grant College University of Maryland.

Kennedy, V. S., M. Oesterling, and W. A. Van Engel. 2007. History of blue crab fisheries on the U.S. Atlantic and Gulf Coasts. Pages 655 - 710 in V. S. Kennedy and L. E. Cronin, editors. *The Blue Crab, Callinectes sapidus*. Maryland Sea Grant College, College Park, MD.

Koopman, D. O. 1956. The Theory of Search. I Kinematic Bases. *Operations Research* 4:324-345

Lipcius, R. N., & Stockhausen, W. T. 2002. Concurrent decline of the spawning stock, recruitment, larval abundance, and size of the blue crab Callinectes sapidus in Chesapeake Bay. *Marine ecology. Progress series*, *226*, 45-61.

Lipcius, R. N., D. B. Eggleston, K. L. Heck Jr, R. D. Seitz, and J. van Montfrans. 2007. Ecology of postlarval and young juvenile blue crabs. 535 - 564 in V. S. Kennedy and L. E. Cronin, editors. The Blue Crab, Callinectes sapidus. Maryland Sea Grant, College Park, MD.

Miller, T.J. and E. D. Houde. 1999. Blue Crab Target Setting. Technical Report [UMCES]CBL TS-177–99. Chesapeake Biological Laboratory, Solomons, Maryland

Miller, T. J. 2001. Matrix-based modeling of blue crab population dynamics with applications to the Chesapeake Bay. *Estuaries*, *24*(4), 535-544.

Miller, T. J., S. J. D. Martell, D. B. Bunnell, G. Davis, L. A. Fegley, A. F. Sharov, C. F. Bonzek, D. A. Hewitt, J. M. Hoenig, and R. N. Lipcius. 2005. Stock Assessment of Blue Crab in Chesapeake Bay: 2005. Final Report Ref: [UMCES]CBL 05-077. UMCES Tech. Ser. No. TS- 487-05-CBL., University of Maryland Center for Environmental Science Chesapeake Biological Laboratory, Solomons, MD.

Miller, T.J., M.J. Wilberg, A.R. Colton, G.R. Davis, A. Sharov, R.N. Lipcius, G.M. Ralph, E.G. Johnson, A.G. Kaufman. 2011. Stock Assessment for the Blue Crab in Chesapeake Bay 2011. Technical Report Series No. TS-614-11 of the University of Maryland Center for Environmental Science.

Moksnes, P. O., Lipcius, R. N., Pihl, L., & van Montfrans, J. 1997. Cannibal-prey dynamics in young juveniles and postlarvae of the blue crab. *Journal of Experimental Marine Biology and Ecology*, *215*(2), 157-187.

Natunewicz, C. C. and C. E. Epifanio. 2001. Spatial and temporal scales of patches of crab larvae in coastal waters. *Marine Ecology Progress Series*, 212:217-222.

Orth, R. J. and J. van Montfrans. 1987. Utilization of a seagrass meadow and tidal marsh creek by blue crabs Callinectes sapidus. I. Seasonal and annual variations in abundance with emphasis on post-settlement juveniles. *Marine Ecololgical Progress Series* 41: 283-294.

Pile, A. J., Lipcius, R. N., van Montfrans, J., & Orth, R. J. 1996. Density-dependent settler-recruit-juvenile relationships in blue crabs. *Ecological Monographs*, 277-300.

Rago, P. J., Weinberg, J. R., & Weidman, C. 2006. A spatial model to estimate gear efficiency and animal density from depletion experiments. *Canadian Journal of Fisheries and Aquatic Sciences*, *63*(10), 2377-2388.

Rathbun, M.J. 1896. The genus *Callinectes*. Proceedings of the U.S. National Museum, 18(1070): 349-375

Reyns, N. B., & Eggleston, D. B. 2004. Environmentally-controlled, density-dependent secondary dispersal in a local estuarine crab population. *Oecologia*, *140*(2), 280-288.

Reyns, N. B., Eggleston, D. B., & Luettich Jr, R. A. 2006. Secondary dispersal of early juvenile blue crabs within a wind-driven estuary. *Limnology and oceanography*, *51*(5), 1982-1995.

Roman, M. R. and W. C. Boicourt. 1999. Dispersion and recruitment of crab larvae in the Chesapeake Bay plume: Physical and biological controls. *Estuaries* 22:563-574.

Rome, M. S., A. C. Young-Williams, G. R. Davis, and A. H. Hines. 2005. Linking temperature and salinity tolerance to winter mortality of of Chesapeake Bay blue crabs (Callinectes sapidus). *Journal of Experimental Marine Biology and Ecology* 319:129-145.

Rothschild, B. J., J. S. Ault, E. V. Patrick, S. J. Smith, H. Li, T. Maurer, B. Daugherty, G. R. Davis, C. Zhang, and R. McGarvey. 1992. Assessment of the Chesapeake Bay blue crab stock. CBL 92 - 003 - 036, Chesapeake Biological Laboratory, Solomons, MD.

Rugolo, L., A. Lange, K. Knotts, C. Stagg, R. O'Reilly, M. Terceiro, V. Crecco, and D. Vaughan. 1997. Chesapeake Bay blue crab stock assessment. National Oceanic and Atmospheric Administration, National Marine Fisheries Service, Chesapeake Bay Stock Assessment Committee, Annapolis, Maryland.

Sharov, A.F., J.H. Vølstad, G.R. Davis, B.K. Davis, R.N. Lipcius, and M.M. Montane. 2003. Abundance and exploitation rate of the blue crab (Callinectes sapidus) in Chesapeake Bay. *Bulletin of Marine Science* 72 (2): 543–565.

Stockhausen, W. T. and R. N. Lipcius. 2003. Simulated effects of seagrass loss and restoration on settlement and recruitment of blue crab postlarvae and juveniles in the York River, Chesapeake Bay. *Bulletin of Marine Science* **72**:409-422.

Van Montfrans, J., C. A. Peery and R. J. Orth. 1990. Daily, monthly and annual settlement patterns by Callinectes sapidus and Neopanope sayi on artificial collectors deployed in the York River, Virginia. *Bulletin of Marine Science*. 46: 214-228.

Van Montfrans, J., C. E. Epifanio, D. M. Knott, R. N. Lipcius, D. J. Mense, K. S Metcalf, E. J. Olmi III, R. J. Orth, M. H. Posey, E. L. Wenner, T. L. West. 1995. Settlement of blue crab postlarvae in western North Atlantic estuaries. *Bulletin of Marine Science*. 57: 834-854.

Vølstad, J. H., Sharov, A. F., Davis, G., & Davis, B. 2000. A method for estimating dredge catching efficiency for blue crabs, Callinectes sapidus, in Chesapeake Bay. *Fishery Bulletin*, *98*(2).

Williams, A.B. 1974 The swimming crabs of genus Callinectes (Decapoda: Portunidae). *Fishery Bulletin*, 72:685-798

Wood, S.N. 2000 Modelling and smoothing parameter estimation with multiple quadratic penalties. *Journal of the Royal Statistical Society*, 63(2):413-428.

Wood, S.N. 2006 Generalized Additive Models: An Introduction with R. *Chapman and Hall/CRC*.

Zuur, Alain, Elena N. Ieno, Neil Walker, Anatoly A. Saveliev, and Graham M. Smith. *Mixed effects models and extensions in ecology with R*. Springer, 2009.