

1-1-2013

Assessing The Habitat Use, Diet, And Sex Ratios Of Atlantic (*Acipenser Oxyrinchus*) And Shortnose Sturgeon (*Acipenser Brevirostrum*) In The Saco River, ME

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MOVEMENT AND HABITAT USE OF ATLANTIC (ACIPENSER OXYRINCHUS
OXYRINCHUS) AND SHORTNOSE STURGEON (ACIPENSER BREVIROSTRUM) IN THE
SACO RIVER ESTUARY SYSTEM

By

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B.S. Cedar Crest College, 2008

THESIS

Submitted to the University of New England
in Partial Fulfillment of the
Requirements for the Degree of

Master of Science

in

Marine Sciences

January, 2013

This thesis has been examined and approved.

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ACKNOWLEDGEMENT

I would like to thank the individuals without who this research would not have been possible. I want to thank the members of Dr. Sulikowski's research lab and other graduate students at the University of New England. All of your assistance, dedication and support were invaluable to me during my time at UNE. Without it, I would not have been able to successfully complete this project. You each made working in the lab enjoyable for me every day. For that, I am forever grateful. To James Sulikowski, I am thankful for all of your helpful efforts in my research. The opportunities that you provided have helped me to achieve so much since I entered your lab. I also thank my committee members, Eric Hoffmayer and Gayle Zydlewski, for their thoughtful comments on my work. Thank you. This work would also not have been possible without the help of Marian Reagan, Tim Arienti and Shaun Gill, who were always available to help with any and all problems. Finally, I would like to thank my friends and family. You stayed with me through the good and the bad over the last three years. Without your encouragement and support, I would not have completed this research. This thesis work was supported by the NOAA Species of Concern Research Program and the NOAA Section 6 Research Program. I would also like to thank the National Science Foundation GK-12 program for funding and those involved with the program at UNE for their never ending encouragement and support.

TABLE OF CONTENTS

TITLE PAGE i

THESIS APPROVAL ii

ACKNOWLEDGEMENTS iii

TABLE OF CONTENTS iv

ABSTRACT vii

GENERAL INTRODUCTION 1

CHAPTER 1: FIRST DOCUMENTED OCCURRENCES OF THE SHORTNOSE STURGEON
(ACIPENSER BREVIROSTRUM) IN THE SACO RIVER, MAINE, USA 4

 Abstract 5

 1. Introduction 5

 2. Materials and Methods 7

 2.1 Study Area 7

 2.2 Acoustic Receiver Array 7

 2.3 Field Sampling Methods 8

 3. Results 9

 3.1 Field Sampling 9

 3.2 Acoustic Telemetry 10

 3.2.1 Fish Tagged in Saco River 10

 3.2.2 Fish Tagged in Other Rivers 10

 4. Discussion 13

References 16

Tables 19

 Table 1 19

 Table 2 20

Figure Captions 21

Figures 22

 Figure 1 22

CHAPTER 2: HABITAT USE AND MOVEMENT PATTERNS OF ATLANTIC STURGEON (ACIPENSER OXYRINCHUS) IN THE SACO RIVER, ME WITH OBSERVATION ON SEX RATIO AND DIET 23

Abstract	24
1. Introduction	25
2. Materials and Methods	27
2.1 Study Site	27
2.2 Fish Capture and Sampling	28
2.3 Acoustic Telemetry	29
2.3.1 Acoustic Array	29
2.3.2 Transmitter Implantation	30
2.3.3 Data Processing.....	31
2.4 Steroid Hormone Analysis	33
2.5 Diet Analysis	36
3. Results	37
3.1 Capture and Tagging	37
3.2 Telemetry	37
3.3 Steroid Hormones	40
3.4 Diet	42
4. Discussion	43
4.1 Movement	43
4.2 Size Classes	48
4.3 Reproduction	49
4.4 Diet	54
References	56
Tables	61
Table 1	61
Table 2	62
Table 3	63
Table 4	64

Table 5	65
Table 6	66
Figure Captions	67
Figures	68
Fig. 1	68
Fig. 2	69
Fig. 3	70
Fig. 4	71
Fig. 5	72
Fig. 6	73
GENERAL CONCLUSION	74

ABSTRACT

ASSESSING THE HABITAT USE, DIET AND SEX RATIOS OF ATLANTIC (ACIPENSER OXYRINCHUS) AND SHORTNOSE STURGEON (ACIPENSER BREVIROSTRUM) IN THE SACO RIVER, ME

By

Caitlyn Little

University of New England, August 2012

Due to the questionable status of sturgeon populations along the east coast of the United States, there has been considerable research focus on these species in recent years. Previous studies have overlooked the Saco River as an important habitat for sturgeon in the Gulf of Maine. However, the incidental capture of a sturgeon in the Saco River in the fall of 2007 raised questions about the presence of this species and the role that the Saco River might play in the life cycle of these fish. The goals of this thesis were to evaluate the utilization of this habitat by sturgeon species and determine the sex ratio of the population inhabiting the river. Chapter one documents the first reported occurrences of the shortnose sturgeon (*Acipenser brevirostrum*) in the Saco River. Two shortnose sturgeon were captured in the Saco River between August of 2010 and June of 2011. In addition to these two individuals, six other shortnose sturgeon that were tagged in the Merrimack River, MA were also detected in the Kennebec and Saco Rivers in Maine. This indicates that coastal movements in the Gulf of Maine may be more important than previously

thought. Smaller rivers, such as the Saco, may play an important role as foraging habitat on these longer migrations. Chapter two examines the habitat use and sex ratio of Atlantic sturgeon in the Saco River using acoustic telemetry, stomach content analysis and quantification of circulating sex steroids. During this study a total of 196 individual Atlantic sturgeon (*Acipenser oxyrinchus oxyrinchus*) were captured in gill nets in the Saco River. Internal acoustic transmitters were implanted in 43 of these sturgeon to track movements within the Saco River system. Of these tagged sturgeon 42 were subsequently detected on the acoustic receiver array in the Saco River and 40 were detected in the Saco River during more than one sampling season. Lattice-based density analysis of all detections from the Saco River indicated that sturgeon in this system primarily utilize the area at the mouth of the river. Stomach content analysis revealed that American sand lance (*Ammodytes americanus*) was the primary food item in the diet of Atlantic sturgeon in the Saco River. Steroid hormone analysis indicated that the sex ratio of Atlantic sturgeon in the river is biased toward females. However, there was some degree of uncertainty in assignment of sex, and further investigation into steroid hormones is warranted to fine-tune the methodology.

GENERAL INTRODUCTION

In recent decades overexploitation of fish stocks has become a major threat for many large marine species (Casey and Myers, 1998; Baum et al. 2003, Myers et al. 2007). This overfishing has led to dramatic reductions in some fish stocks worldwide, including Canadian cod and several groundfish species off the coast of Canada and New England (Botsford, 1997; Jacquet and Pauly, 2007). It has also been estimated that most large shark species in the northwest Atlantic, with the exception of the mako shark have declined by more than 50% in the past 8 to 10 years (Baum et al. 2003). Mullan et al. (2005) suggests that overfishing has led to the complete collapse of one in every four commercial fisheries over the last 50 years. These patterns of decline in fish stocks have highlighted the importance of proper management for extant fish species.

Impacts of increased fishing activities are especially serious when the targeted species are those that live to advanced ages and mature late in life (Botsford, 1997, Casey and Myers, 1998). Species that display these life history characteristics wait many years before being capable of spawning and may be removed from the population before they are able to reproduce. Because fisheries focus their efforts on the largest individuals in a population, mature and maturing individuals are most commonly removed from the stock (Botsford, 1997). Removal of these individuals may reduce the reproductive input of a population and lead to faster declines in biomass and slower recovery rates for populations that are already depleted (Botsford, 1997).

There are currently 25 extant species of sturgeon worldwide (Pikitch et al. 2005). In recent years, concern over the conservation of this taxonomic group has become a focus for researchers (Birstein, 1993; Boreman, 1997; Secor et al. 2000, Pikitch et al. 2005). Fishing pressures, pollution and habitat degradation have caused dramatic population declines in many sturgeon species (Beamesderfer and Farr, 1997; Bemis and Kynard, 1997, Pikitch et al. 2005). Failure to implement proper conservation measures may lead to the continued decline and possible extinction of several sturgeon species worldwide (Pikitch et al. 2005).

Some Rivers along the northeastern coast of the United States play host to two sturgeon species, the Atlantic (*Acipenser oxyrinchus oxyrinchus*) and the shortnose sturgeon (*Acipenser brevirostrum*). A thriving sturgeon fishery along the east coast operated from colonial times through the mid 20th century. This led to drastic declines in the populations of both species. In 1967 the shortnose sturgeon was officially listed as endangered under the ESA, effectively ending commercial harvest of this fish. However, fishing for Atlantic sturgeon continued into the 1990's. In 1997, the National Marine Fisheries Service listed the Atlantic sturgeon as a federal species of concern and as of April 6, 2012 it was listed under the ESA. Listing of this species is currently based on distinct population segments (DPSs). The south Atlantic, Carolina, Chesapeake Bay and New York Bight DPSs have all been listed as endangered, while the Gulf of Maine DPS is currently listed as threatened.

Until recently the Saco River has been overlooked as potentially important habitat for sturgeon species in the northeastern United States due to a general lack

of documentation of sturgeon in this river. However, incidental capture of an Atlantic sturgeon by researchers at the University of New England in 2007 brought focus to the presence of these creatures in the Saco River. During subsequent fishing efforts to gain insight into the presence of Atlantic sturgeon, a shortnose sturgeon was captured just inside the mouth of the river. This master's thesis reports current research into the presence of these two fish in the river. The first chapter focuses on the first documented occurrences of shortnose sturgeon in the Saco River and some of their coastal movements in the Gulf of Maine. The second chapter examines the home range, diet and sex ratio of Atlantic sturgeon captured in the Saco River.

CHAPTER 1

FIRST DOCUMENTED OCCURRENCES OF THE SHORTNOSE STURGEON (ACIPENSER BREVIROSTRUM) IN THE SACO RIVER, MAINE, USA

Abstract

During sampling efforts to study the more abundant Atlantic sturgeon, *Acipenser oxyrinchus oxyrinchus*, between May of 2009 and November of 2011, four shortnose sturgeon were captured in gill nets near the mouth of the Saco River. Two of these individuals were tagged with acoustic transmitters to monitor their movement within the Saco River. Additionally, six shortnose sturgeon that had been tagged with acoustic transmitters in the Merrimack River, MA were detected on the acoustic array deployed within the Saco River and its estuary over this time period. These incidences represent the first verified documentation of shortnose sturgeon within this estuary.

1. Introduction

The shortnose sturgeon, *Acipenser brevirostrum*, was commercially harvested in the United States from the late nineteenth to the early twentieth century (NMFS 1998). This directed harvest, coupled with deteriorating water quality in riverine habitats and construction of dams restricted passage to spawning grounds, leading to drastic declines in shortnose sturgeon abundance (NMFS 1998). Due to these circumstances, the shortnose sturgeon was designated an endangered species in 1967, a status it retains today.

Currently, the geographic distribution of *Acipenser brevirostrum* encompasses large rivers along the majority of the East Coast of the United States (U.S.), extending to the St. Johns River in New Brunswick, Canada (NMFS 1998).

Historically, populations of shortnose sturgeon were reputed to display the life history characteristics of a freshwater amphidromous species (Bain 1997; Kieffer and Kynard 1996; Buckley and Kynard 1985; Taubert 1980). These fish were thought to spend the majority of their lifecycle in discrete areas of large natal rivers with limited movement into estuarine or marine waters (Kieffer and Kynard 1993; Buckley and Kynard 1985). Specimens are rarely captured in coastal marine waters and were not typically considered coastal migrants (Bain et al. 2007; Dadswell et al. 1984). However, more recent studies have found that movements between major river systems in the northeastern U.S. are more common than previously thought and that habitat connectivity may be an important consideration for the recovery of this species throughout its range (Zydlewski et al. 2011; Fernandes et al. 2010).

In the northeastern U.S., populations of shortnose sturgeon have been studied in several rivers. These include the Penobscot River (Zydlewski et al. 2011; Fernandes et al. 2010) and Kennebec-Androscoggin-Sheepscoot complex in Maine (Squiers et al. 1982), as well as the Merrimack (Kieffer and Kynard 1996; Kieffer and Kynard 1993) and Connecticut Rivers (Buckley and Kynard 1985; Taubert 1980) in Massachusetts. The Saco River is the sixth largest river system entering the coastal waters of the Gulf of Maine (Tilburg et al. 2011) and it represents a midpoint between these southern and northern rivers of New England. It is also known to host a population of Atlantic sturgeon (*Acipenser oxyrinchus oxyrinchus*) during the summer months (Sulikowski, unpublished data), a species that frequently inhabits many of the same major rivers as shortnose sturgeon. Despite this location and potentially suitable habitat, shortnose sturgeon have never been documented in this

river system. Herein, we present the first description of the endangered shortnose sturgeon within this river system and possible implications for the life history and management of this species.

2. Materials and Methods

2.1 Study Area.

Sampling for this study focused on the estuarine portion of the Saco River as it enters its associated bay (Fig 1). The Cataract Dam joins the towns of Saco and Biddeford at a distance of about 10 river kilometers (rkm) from the mouth of the river (Brothers et al. 2008). This dam separates the tidally influenced estuary from upstream portions of the river and presents an impassable boundary for sturgeon. As a result, the study area is limited to the stretch of river downstream of this barrier. At the mouth of the river, two jetties have been constructed that extend about 1.5 km from the natural mouth of the river into Saco Bay.

2.2 Acoustic Receiver Array.

An array of seven VEMCO VR2W receivers was deployed in this lower reach of the Saco River (Fig. 1) to monitor movement of fish tagged with acoustic transmitters. The theoretical maximum detection range of a VR2W receiver in a turbid river in fresh water is between 900 and 1000 m (VEMCO). Given that the greatest river

width at a receiver location is 330 m, it was assumed that the receivers covered the width of the river at each location. Each year from 2009-2011, receivers were deployed in the Saco River in early April or May and remained in the water throughout the summer and fall until their removal in late November or early December. Two acoustic receivers were positioned near the mouth of the river from November through May each year to monitor passage of fish into and out of the river over the winter months.

2.3 Field Sampling Methods.

Net sampling was conducted twice a month between May and November from 2009-2011. Bottom-set monofilament gill nets measuring 100 m long x 2 m deep and with stretched mesh sizes of 15.2 cm and 30.5 cm were used to capture sturgeon (Atlantic and shortnose) of varying sizes. Fishing was conducted at low tide and nets were set perpendicular to the jetties at the mouth of the river. Gear was allowed to soak for a maximum of 30 minutes to minimize stress to captured individuals. Captured sturgeon were held in a floating net pen (2.1 m x 0.9 m x 0.9 m) attached to the side of the boat until processing.

Each fish was individually brought onboard and placed in a holding tank measuring 2.1 m x 0.5 m x 0.4 m. External measurements, including total length (TL), fork length (FL), head length (HL), interorbital width (IOW), and mouth width (MW) were obtained. Three fish were implanted with passive integrated transponder (PIT) tags for long-term identification. PIT tags were injected in the

fleshy base of the dorsal fin using a Biomark MK10 implanter with a 6-gauge stainless steel needle. External T-bar tags were implanted in the opposite side of the base of the dorsal fin using an Avery Dennison Mark III tagging gun. Two shortnose sturgeon captured in the Saco River were surgically implanted with coded VEMCO V16 acoustic transmitters with an estimated battery life of 2993 days. Transmitters were inserted through a 5 cm c-shaped incision on the ventral surface approximately 10 cm anterior of the vent. A single polydioxanone (absorbable) suture was used to close the incision. Individuals were returned to the net pen for recovery and observation before release.

3. Results

3.1 Field Sampling.

A total of four shortnose sturgeon, ranging in total length from 81.0-92.5 cm, were captured in gill nets set in the Saco River between June of 2009 and June of 2011. The first capture occurred on June 16, 2009 (81.0 cm TL), the second occurred on August 30, 2010 (86.0 cm TL), the third on May 25, 2011 (92.5 cm TL) and the fourth on June 9, 2011 (83.0 cm TL). All shortnose were returned to the Saco River at the location of capture with no outward signs of stress. VEMCO V16 acoustic tags were surgically implanted in the sturgeon captured on August 30, 2010 (SNS A) and June 9, 2011 (SNS B) to monitor movement within the Saco River.

3.2 Acoustic Telemetry

3.2.1 Fish Tagged in Saco River

The first shortnose sturgeon (SNS A) surgically implanted with an acoustic tag was released into the Saco River on August 30, 2010 (Table 1). This individual remained in the river for approximately 18 d, departing on September 17. Over this period, SNS A utilized the entire study area (up to rkm 9.5). SNS A returned to the Saco River on July 3, 2011 and remained in the river for nine days, exiting the system on July 12. In 2011, this sturgeon used a smaller portion of the study area, venturing to rkm 6.

The second shortnose sturgeon (SNS B) was implanted with an acoustic tag and released in the Saco River on June 9, 2011. This individual remained in the study area for 146 d before departing on November 2, 2011. The farthest upriver this sturgeon ventured over this time period was to rkm 6. Neither of the two shortnose sturgeon tagged in the Saco River have been detected on receiver arrays in other river systems.

3.2.2 Fish Tagged in Other Rivers

The Saco River acoustic receiver array detected six individual shortnose sturgeon that had been implanted with transmitters upon prior capture in the Merrimack River, MA. These immigrant tag detections occurred between April 6, 2010 and

November 1, 2011 (Table 2). No shortnose tagged in other systems were detected in the Saco, but some of the Merrimack (tagged) fish that entered the Saco were previously or subsequently detected in additional river systems in the Gulf of Maine.

The first Merrimack-tagged sturgeon (SNS #1) to arrive in the Saco River was a female with late stage eggs that was last detected in the Merrimack on April 3, 2010. This fish was detected just inside the mouth of the Saco River 53 times on April 6 before departing the study site the same day. This fish was then detected in the Kennebec River, ME from April 12 to May 5, 2010. It subsequently returned to Saco Bay on May 9, 2010, almost one month after it was last detected in the system. Upon returning to the study site, this fish was detected in Saco Bay 12 times over an hour and a half before exiting the study area again on the same day.

The second immigrant shortnose (SNS #2) was last detected in the Merrimack on April 6, 2010 and arrived in the Saco River three days later. Receivers logged 1280 detections between April 9 and 11, 2010. After departing the Saco River, this individual moved north and was detected in the Kennebec system on April 18. It remained in this system until May 2, 2010. It then returned south to the Saco River on May 8. It subsequently remained in the system until May 9 and was detected on receivers up to rkm 5.5.

The third shortnose (SNS #3) detected in the Saco River was last detected in the Merrimack on March 28, 2010. This individual initially traveled north past the Saco River and was detected in the Kennebec River from 6-30 April, 2010. This fish was subsequently detected on receivers at the mouth of the Saco River 39 times on May 3, 2010 before departing the study area.

The fourth Merrimack-tagged shortnose (SNS #4) to be detected in the study area was last detected in the Merrimack on October 18, 2010. It remained in the estuary for only a brief period of time. This fish was detected on a receiver in Saco Bay five times on October 20 before moving on. It was then detected on a single receiver in the Casco Bay from 25-27 October 2010. This fish was detected in the Kennebec River the following spring and remained in that system from April 24, 2011 to December 12, 2011

A fifth Merrimack-tagged shortnose sturgeon (SNS #5) was last detected in that river on April 15, 2011 and was detected in the Saco River on April 20, 2011. This individual remained in the river for approximately four days, departing the Saco River on April 24. During its residency in the river, this animal was detected on all receivers up to the Cataract Dam (rkm 9.5). This animal then continued north and was detected in the Kennebec River from 4-24 May, 2011.

The sixth and final sturgeon (SNS #6) from the Merrimack to be detected in the Saco was last detected in the Merrimack on April 13, 2011. This animal then migrated north and entered the Kennebec River on April 22, 2011. It remained in this system for 85 d before exiting on July 16, 2011. SNS #6 then traveled south and arrived at the mouth of the Saco River on July 21, 2011. This individual remained in the system for several months, utilizing the entire study area and exiting the system on November 1, 2011.

4. Discussion

Few studies have examined the fish community in the Saco River estuary. One long-term study by Reynolds and Casterlin (1985) found that the Saco River estuary and Saco Bay hosts at least 18 different species of fish as well as a variety of crustaceans, echinoderms and mollusks. Additionally, a two-year study by Furey and Sulikowski (2011) from 2007 to 2008 documented 24 fish species inhabiting the estuarine reaches of this system, including two Atlantic sturgeon (*Acipenser oxyrinchus oxyrinchus*). Although those studies suggest that the Saco River estuary is an important habitat for other species of fish, *Acipenser brevirostrum* had not previously been reported in the system prior to the current study. Use of the Saco estuary by this endangered species suggests this watershed might serve an important habitat function.

Brief movements into and out of the estuary by six shortnose sturgeon tagged in the Merrimack River indicates that the Saco River may serve as a stopover site on a larger migration among rivers in the Gulf of Maine. The timing of these stopovers suggests a potential link to spawning. This is most clearly illustrated by SNS #1 and SNS #2, both of which entered the Saco River while travelling to and returning from the Kennebec River. Several other shortnose sturgeon either briefly entered the Saco River estuary at the beginning of April before departing and entering the Kennebec River system or were detected in the Kennebec River before returning south to the Saco River. This time window of absence from the Saco River or entry into the Kennebec River (April-May) is consistent with the known period of

spawning in the Kennebec/Androscoggin system (Squiers et al. 1982). Moreover, all shortnose sturgeon tagged in the Merrimack, with the exception of SNS #5, were known to be females with late stage eggs that should have been approaching spawning condition as described by Kieffer and Kynard (2012). None of these fish were detected in freshwater reaches of the Saco, suggesting that spawning did not occur in this system.

One individual tagged in the Merrimack River (SNS #6) and both individuals tagged in the Saco River demonstrated extended residence times and movements throughout the Saco system. The period of residence for two of these fish (SNS A during its return year and SNS #6 in its tagging year) occurred too late in the year to likely be tied to a spawning movement (Dadswell et al. 1984; Buckley and Kynard 1985; Kieffer and Kynard 1996). The remaining Saco River fish (SNS B) was tagged too late in 2011 to know whether it was resident in the system during the spring of its capture year (as is true for SNS A), and it is not known at the time of writing if and when it might reenter the system (unlike SNS A). Extended summer residence in the Saco estuary is likely more consistent with use of the system for foraging.

On several occasions, shortnose sturgeon that entered the Saco River only ventured as far as rkm 6. This location represents one of the widest portions of the river accessible to sturgeon and would be expected to have a lower flow velocity than other portions of the study area. It is not currently known why several of the fish ended their upriver movements at this point and additional study is necessary to determine if this area serves a specific function or limitation for some individuals.

Historically it was thought that shortnose sturgeon do not typically make coastal migrations (Dadswell et al. 1984). However, Fernandes et al. (2010) and Zydlewski et al. (2011) found that this species undertakes regular, seasonal migrations between the Kennebec River complex and the Penobscot River with short ventures into smaller coastal rivers in Maine. This discovery, coupled with our findings, indicates that the movements and population ecology of *Acipenser brevirostrum* in the northeastern United States is more complex than previously thought. Furthermore, the repeated occurrence of shortnose sturgeon in the Saco River during consecutive years suggests that this estuary may be an important habitat for this species on both short (days) and long (months) time scales. Indeed, intermediate rivers like the Saco may require careful consideration under the current management scenario, since it is possible that they are in part responsible for enabling the unique migratory ecology of this species in the Gulf of Maine.

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Table 1. Tagging and observation dates (mm/dd/yy) for shortnose sturgeon acoustically tagged in the Saco River, ME. 'NYD' indicates that return of the fish was 'not yet documented'. Fish were assigned IDs of SNS A (shortnose sturgeon A) and SNS B (shortnose sturgeon B) to differentiate between individuals. 'Furthest Upriver' indicates the position of the acoustic receiver furthest inland at which the individual was detected.

Fish ID	Date Tagged	Exited Saco	Date Returned to Saco	Furthest Upriver (rkm)
SNS A	08/30/10	09/17/10	07/03/11	9.5
SNS B	06/09/11	11/02/11	NYD	6

Table 2. Observation dates (mm/dd/yy) for shortnose sturgeon acoustically tagged in the Merrimack River, MA and recorded in other river systems in the Gulf of Maine, April 2010 to November 2011. Fish were assigned IDs of SNS #1 - #6 (shortnose sturgeon #1 - #6) in order to differentiate between individuals.

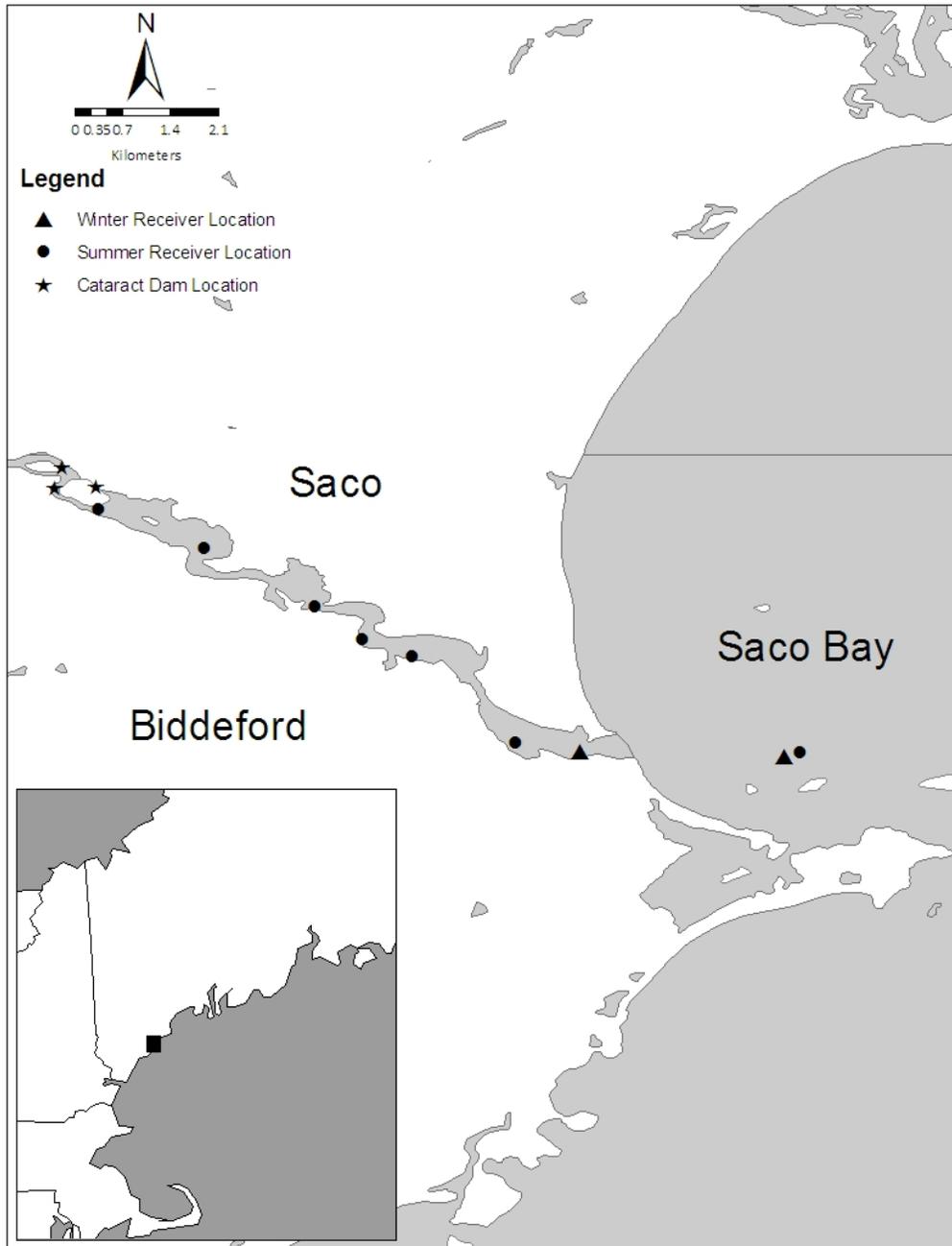
Fish ID	Depart Merrimack	Arrive Saco	Depart Saco	Arrive Kennebec	Depart Kennebec	Arrive Saco	Depart Saco
SNS #1	04/03/10	04/06/10	04/06/10	04/12/10	05/05/10	05/09/10	05/09/10
SNS #2	04/06/10	04/09/10	04/11/10	04/18/10	05/02/10	05/08/10	05/09/10
SNS #3	03/28/10			04/06/10	04/30/10	05/03/10	05/03/10
SNS #4	10/18/10	10/20/10	10/20/10	04/24/11	12/12/11		
SNS #5	04/15/11	04/20/11	04/24/11	05/04/11	05/24/11		
SNS #6	04/13/11			04/22/11	07/16/11	07/21/11	11/01/11

Figure Captions

Fig 1.

Location of acoustic receiver array in the Saco River estuary during summer (May - November) and winter (December - April) months of 2009 - 2011. Area of the array is denoted by the small, black rectangle on the inset map. Approximate river kilometers are denoted with numbers along the southern edge of the river.

Fig. 1



CHAPTER 2

HABITAT USE AND MOVEMENT PATTERNS OF ATLANTIC STURGEON (ACIPENSER OXYRINCHUS) IN THE SACO RIVER, ME WITH OBSERVATIONS ON SEX RATIO AND DIET

Abstract

Little research has been done regarding Atlantic sturgeon in the Gulf of Maine. More specifically, this study represents the first study to examine movement patterns, reproduction, and diet information gathered on the population of Atlantic sturgeon utilizing the Saco River. A total of 196 Atlantic sturgeon ranging from 77.0 to 180.0 cm FL were captured between 2008 and 2011. Forty-two sturgeon were implanted with acoustic transmitters to monitor movement within the Saco River system and the Gulf of Maine. Atlantic sturgeon appear to inhabit the Saco River seasonally and prefer the first few river kilometers (1-5) near the mouth of the system. Reproductive hormones were utilized to assess the reproductive structure of the population and indicated that the majority of sturgeon using this system are female. The diet of Atlantic sturgeon in the river was composed primarily of American sand lance, which are known to school near the mouth of the river where sturgeon appear to spend the majority of their time in the system, and could indicate that the Saco River provides an important seasonal foraging ground for Atlantic sturgeon. This study represents the first information regarding the population of Atlantic sturgeon in this region of Maine and is important for developing a better understanding of the life history of Atlantic sturgeon in the immediate area and throughout the Gulf of Maine.

1. Introduction

The Atlantic sturgeon (*Acipenser oxyrinchus oxyrinchus*) is a large (average 6 to 9 feet), long-lived (60 years or more), late maturing (up to 20 years), anadromous fish species that ranges from St. Croix, ME to the Saint Johns River, FL, in US waters (Smith and Clugston 1997). This species was historically abundant throughout its range along the east coast of North America, and fisheries for Atlantic sturgeon existed in most major river systems this species inhabited (Smith and Clugston 1997). However, the aforementioned life history characteristics make Atlantic sturgeon highly susceptible to fishing pressures and other anthropogenic impacts (Beamesderfer and Farr 1997; Boreman 1997). Overharvest, pollution and construction of dams, that limited passage to historic spawning grounds, resulted in drastic declines in Atlantic sturgeon populations along the coast of the United States (Smith and Clugston 1997). By the early 20th century, all Atlantic sturgeon fisheries had suffered drastic catch reductions and most major fisheries had collapsed by 1905 (Smith 1985).

Due to the population declines along the east coast, the National Marine Fisheries Service (NMFS) listed the Atlantic sturgeon as a federal species of concern throughout its range in the United States in 1997 (ASSRT 2007). A comprehensive status review was completed by NMFS in 2007 to evaluate the status and current threats to the species. This review identified five distinct population segments (DPSs) of Atlantic sturgeon along the east coast of the United States (ASSRT, 2007). The northernmost DPS encompasses the Gulf of Maine, including the entire coasts of Maine and New Hampshire, as well as the coast of Massachusetts north of Cape Cod. As of 6 February

2012 the Gulf of Maine DPS has been listed as threatened under the Endangered Species Act (ESA) due to decreased population numbers from historical levels, limited spawning and threats that will presumably continue to hinder population recovery in the foreseeable future (NMFS 2012). The other DPSs of Atlantic sturgeon include the New York Bight, Chesapeake Bay, Carolina, and South Atlantic and have all been listed as endangered under the ESA.

Despite efforts to study the Atlantic sturgeon throughout its range in recent decades, information on this species remains patchy. Atlantic sturgeon spend several years of their lives in the marine environment and are known to make large-scale coastal migrations. However, habitat use in the marine environment remains largely unknown (Bain 1997; Collins et al. 2000). Similarly, information regarding habitat use in riverine environments is incomplete for many river basins this species inhabits. The population in the Hudson River has been most thoroughly documented, but other rivers are lacking critical life history information, potentially because sturgeon population levels in these basins are too low to establish habitat use with certainty (Bain 1997; Gross et al. 2002). This is especially true for smaller coastal rivers where spawning may not be occurring and studies on habitat use and life history have not been conducted (ASSRT 2007).

Although this species has been known to occur in several New England Rivers, including the Penobscot and Kennebec complexes in Maine (ME; Squiers and Smith 1987; Smith and Clugston 1997; Fernandes 2008), their presence had not been reported in the Saco River, ME within the last 50 years until the incidental capture of a specimen by researchers from the University of New England in 2007. With the use of acoustic telemetry, circulating levels of reproductive hormone concentrations, and stomach

contents analysis we present the first examination of the movement patterns, habitat use and population structure of Atlantic sturgeon in the Saco River watershed. This information is critical for the proper assessment of the status of this species in the northern portion of its range and for implementation of proper management strategies to ensure the persistence of this species in the Gulf of Maine.

2. Materials and Methods

2.1 Study Site

The Saco River is a rock bound, mid-latitude estuary (Brothers et al. 2008). It is one of the largest rivers in the region and has been recognized as the primary source of sand to the surrounding beach systems (Kelley et al. 2005). Sampling for this study focused on the estuarine portion of the Saco River as it enters its associated bay (Fig 1). The Cataract Dam joins the towns of Saco and Biddeford a distance of approximately 10 km from the river mouth (Brothers et al. 2008). This dam separates the estuary from upstream portions of the river and presents an impassable boundary for sturgeon, preventing utilization of any greater portion of the Saco River. As a result, the study area is limited to the stretch of river downstream of this barrier. At the mouth of the Saco River, two jetties have been constructed that extend about 2.3 km from the natural mouth of the river into Saco Bay (Brothers et al. 2008).

2.2 Fish Capture and Sampling

Sampling for Atlantic sturgeon was conducted twice a month between May and November from 2009-2011. Fish were captured using bottom-set monofilament gill nets measuring 100 m long x 2 m deep and with stretched mesh sizes of 5.08 cm, 15.2 cm and 30.5 cm. Fishing was conducted at slack, low tide and nets were set perpendicular to the jetties at the mouth of the river. Gear was allowed to soak for a maximum of 30 minutes to minimize stress to captured individuals. Captured sturgeon were held in a floating net pen (2.1 m x 0.9 m x 0.9 m) attached to the side of the boat until ready for processing.

Sturgeon were processed one at a time upon retrieval of the gill nets. Each fish was brought onboard and placed in a holding tank (2.1 m x 0.5 m x 0.4 m) supplied with fresh ambient water. External measurements including total length (TL), fork length (FL), head length (HL), interorbital width (IOW), and mouth width (MW) were obtained. Passive integrated transponder (PIT) tags were implanted for long-term identification (model HPT12, Biomark Inc., Boise, Idaho, USA). Passive integrated transponder tags were injected in the fleshy base of the dorsal fin using a Biomark MK10 implanter with a 6-gauge stainless steel needle. External T-bar tags (model TBA, Hallprint Pty Ltd, Hindmarsh Valley, South Australia) were also implanted in the opposite side of the base of the dorsal fin using an Avery Dennison Mark III tagging gun.

Blood samples were collected from 168 of the 196 sturgeon captured during this study for quantification of circulating steroid hormone concentrations. Samples were collected via caudal venipuncture using a double-sided 22-gauge needle and a 3ml lithium heparinized vacutainer. Collected blood was stored on ice while in the field. In

the laboratory, plasma was separated from whole blood in a centrifuge (model VWR Clinical 50; 3,500 RPM; 5 min) and stored at -20°C until hormone extraction.

Stomach contents were obtained from 31 sturgeon via gastric lavage methods modified from Haley (1998). For this technique, a length of PVC pipe with rounded edges (1.2 cm diameter x 15.4 cm length) was inserted into the mouth of the animal and past the gills to provide a guide for the lavage tube. A length of aquarium tubing (120 cm long, 6.25 mm inside diameter, 9.5 mm outside diameter) attached to a 3.1 L garden sprayer was inserted into the PVC tube and slowly advanced down the esophagus of the sturgeon into the stomach. The sturgeon was manually lifted into a vertical position and the food bolus was flushed from the stomach using the garden sprayer. Stomach contents were collected on a mesh screen and transferred to an individually labeled plastic bag. Contents were stored in a freezer until further analysis could take place.

2.3 Acoustic Telemetry

2.3.1 Acoustic Array

An array of ten acoustic receivers (model VR2W; VEMCO, Halifax, Nova Scotia, Canada) was deployed in the lower reach of the Saco River to monitor movement of acoustically tagged sturgeon (Fig. 1). Each year from 2009-2011, receivers were placed in the river in early April or May and remained in the water throughout the summer and fall until their removal in late November or early December due to ice cover. Two acoustic receivers were positioned near the mouth of the river where ice cover is minimal

from November through May each year to monitor passage of fish into and out of the river over the winter months. Receivers were deployed using an anchor, line and float system. This consisted of an anchor with 2 m of anchor chain attached to a line with a buoy on the opposite end. The receiver was attached to the line approximately 1m below the surface of the water. The theoretical maximum detection range of a VR2W receiver in a turbid river in fresh water is between 900 and 1000 m (VEMCO). Given that the greatest river width at a receiver location is 330 m, it was assumed that the receivers covered the width of the river at each location.

2.3.2 Transmitter Implantation

A total of 43 individually coded acoustic transmitters (model V16, 69KHz, VEMCO, Halifax, Nova Scotia, Canada) were surgically implanted into the body cavities of Atlantic sturgeon. Estimated battery life for V16 tags was approximately 2500 days. Nominal delay on transmitters was 240 seconds with a minimum of 170 seconds and a maximum of 310 seconds. During transmitter implantation, fish were removed from the holding tank and placed in a hooded stretcher to minimize movement of the animal and cover the eyes to minimize stress. Transmitters were coated with antibiotic ointment and inserted into the abdominal cavity through a 5 cm c-shaped incision on the ventral surface approximately 10 cm anterior of the vent. One to two polydioxanone sutures (PDO II violet monofilament absorbable suture, Oasis, Mettawa, IL, USA) were used to close the incision. The wound was coated with a layer of antibiotic ointment for protection. After transmitter implantation, individuals were returned to the net pen for recovery and

observation. Individuals were released once they were able to maintain an upright position, indicating that the swim bladder had been deflated, and could actively swim upon removal from the net pen.

2.3.3 Data Processing

Detection data from the acoustic receivers was filtered to ensure all detections utilized in the analyses were valid. Simultaneous detections from the same transmitter at geographically separate locations were eliminated from the analyses. Observations were considered valid if a transmitter was detected on the same hydrophone two or more times within a 60-minute time frame (Wrege et al. 2011). Residence times were calculated for each individual sturgeon. Groups of detections that were separated from other detection events by 24 hours or more were considered a separate residence event.

Data were analyzed for the presence of diel movement patterns using methodology from Wrege et al. (2011). Initially, occurrence data (% total) was plotted against hour of the day. Sturgeon occurrences were then grouped by time of day; night (18:00 – 6:00) and day (6:00 – 18:00), and season; winter (December-February), spring (March-May), summer (June-August) and fall (September – November). Chi-square analyses were performed to identify differences in occurrence based on season and time of day. Statistical tests values were considered significant at an α level of 0.05.

Lattice-based density estimation was conducted to evaluate the home range of individual sturgeon using procedures modified from Barry and McIntyre (2010). A polygon representing the study area was created using the statistical program R (version

13.2). A grid of nodes was laid in the polygon and neighbor relationships were used to define a lattice structure for the study area. Detection data were read into the program and parsed out for analysis. Lattice based home range analysis was run based on several variables, including time of day, month, season, tidal cycle and lunar cycle to elucidate the influence of these parameters on sturgeon movement.

In order to examine habitat use in relation to time of day, data was separated based on the hour of the day (0-23). The data for each hour was plugged into the lattice based density-estimator model to determine the areas of the river most commonly used during each hour. Similarly, to examine monthly and seasonal habitat use, data were parsed out based on the month (1-12) or season (1-4) during which detections occurred. Lunar illumination data (the fraction of the moon illuminated on a given date) was obtained (<http://aa.usno.navy.mil/data/docs/MoonFraction.php>) to examine possible effects of the lunar cycle on sturgeon habitat use. A value of 0 was assigned to detections on dates when the fraction of the moon illuminated was less than 0.33. These dates were grouped together as being “new moon”. A value of 0.5 was assigned to all dates with luminance values of between 0.33 and 0.66. These dates were grouped together as “half moon”. Dates when the fraction of the moon illuminated was greater than 0.66 were considered to be “full moon.” The lattice based home range analysis was run for each of these groups to determine which areas of the study area are most commonly used during different phases of the lunar cycle.

A similar methodology was used for the tidal cycle where detections were divided into 4 categories based on the part of the tidal cycle during which they occurred. A value of 1-4 was assigned to each detection to classify it as low incoming (1), high incoming

(2), high outgoing (3) or low outgoing (4). Detections occurring between low and mid-tide on an incoming tide were assigned a value of 1. Those between mid and high tide on an incoming tide were assigned a value of 2. Detections between high and mid-tide on an outgoing tide were assigned a value of 3. Those between mid-tide and low tide on an outgoing tide were assigned a value of 4.

Each set of detections for each of these variables was analyzed using detection data pooled from all transmitters, as well as for all individual tags. A series of random walks was run in R for each set of detections for each parameter to determine the portions of the study area most frequently utilized by sturgeon. Images that displayed home range as an area of 95% utilization (general activity space) and an area of 50% utilization (core area of utilization) were generated for all data sets. Results were compared to examine changes in habitat use based on season, month, lunar cycle and tidal cycle. Additionally, the total number of detections at each receiver was examined by month to see if utilization of the river changes throughout the course of the year.

2.4 Steroid Hormone Analysis

Plasma samples were extracted twice with 10 volumes of diethyl ether and snap frozen. The remaining ether was evaporated under a stream of nitrogen. Dried extracts were reconstituted in phosphate-buffered saline with 0.1% gelatin (PBSG). All extracted samples were stored at -20°C until assay. To account for loss during the extraction procedure, 1000 counts min⁻¹ (cpm) of the appropriate radio labeled steroid hormone was added into each plasma sample. The mean recoveries for estradiol (E₂) and testosterone

(T) were 68% and 80% respectively. Recovery values were used to correct for the appropriate steroid concentration in each sample.

Radioimmunoassay procedures modified from Sulikowski *et al.* (2004, 2005, 2006) were used to determine plasma concentrations of E₂ and T. Non-radiolabeled steroids were obtained from Steraloids, Inc. (http://www.bioportfolio.com/search/steraloids_Inc.html). The specifics of the radiolabeled steroids, the antibody characteristics, and titers are found in Sulikowski *et al.* (2004). Radioactivity was determined in a Perkin Elmer Tri-Carb 2900 PR liquid scintillation counter (<http://las.perkinelmer.com>). Intra-assay coefficients of variance were 17% for T and 24% for E₂. Inter-assay coefficients of variance were 40% for T and 55% for E₂.

Hormone concentrations obtained from the assays were used to assign sex to each sturgeon. Sex assignments were generated using two methods. The first assignment was made by comparing values obtained in this study with those obtained by Van Eenennaam *et al.* (1996), which examined reproductive hormones of mature Atlantic sturgeon in the Hudson River. The mean androgen and E₂ concentrations for male sturgeon in Van Eenennaam *et al.* (1996) were 126.8 ± 62.5 and 0.33 ± 0.15 ng ml⁻¹, respectively. This mean testosterone value was greater than for that of female sturgeon and the E₂ value was lower than for that of female sturgeon. As a result, similar concentrations observed in sturgeon in this study were used as conservative estimators for sex. Individuals with T concentrations at or above 130 ng ml⁻¹ and sturgeon with E₂ concentrations at or below 0.50 ng ml⁻¹ were assigned a sex of male. All other sturgeon in the study were assigned a sex of female.

The second method of sex assignment was based on equations derived by Webb et al. (2002), which utilized stepwise discriminant function analysis (DFA) to determine the best predictors for sex of wild white sturgeon. Plasma concentrations of E₂ and T were the best predictors for sex and classification functions were derived for both female

$$-1.6727 + 2.3678(\log_{10}T) - 3.5783(\log_{10}E_2)$$

and male sturgeon

$$-5.2972 + 5.2524(\log_{10}T) - 7.5539(\log_{10}E_2).$$

Hormone values were entered into these equations. If the value obtained from the male equation was greater, the sturgeon was assigned a sex of male. If the value obtained from the female equation was greater, the sturgeon was assigned a sex of female.

Sex assignments from these two methods were combined to produce a final sex ratio estimate for the Saco River estuary. Sex ratios were compared for each year and for each season to determine if the sex ratio in the estuary changed over time or if different sexes utilized the estuary more or less at different times of the year.

Finally, the ratio of T to E₂ was examined for all of the individuals in this study. The final concentration of T was divided by the final concentration of E₂. These values were compared between males and females to examine how the ratio differed between sex based on the assigned individuals and to provide information about potential error in sex assignments.

2.5 Diet Analysis

The diet composition of 31 Atlantic sturgeon captured in the Saco River was evaluated using a variety of indices (Pinkas et al. 1971; George and Hadley 1979) comprising the numerical index (%N), the gravimetric index based on wet weight (%W), the frequency of occurrence (%F), and the index of relative importance (IRI), which incorporates the three previous indices and was expressed as a percentage (%IRI). Prey items recovered from each stomach were separated and identified to the lowest possible taxa. The total number and weight of each species was recorded for each individual sturgeon. IRI was calculated as:

$$\text{IRI} = \%F * (\%N + \%W)$$

where %F is the percentage of stomachs containing a particular prey species, %N is the percentage of the total number of prey items in stomachs and %W is the percentage of total weight of prey in the stomachs. The %IRI was used as a final indicator of the importance of each prey species in the diet of Atlantic sturgeon in the Saco River. In addition, a cumulative prey curve was created as an indicator for how completely this study describes the diet of Atlantic sturgeon in the Saco River. The order of the stomachs in the study was randomized 10 times to minimize any bias.

3. Results

3.1 Capture and Tagging

A total of 196 Atlantic sturgeon were captured in gill nets at the mouth of the Saco River between September 10, 2008 and November 15, 2011. Four of these individuals were captured twice over the course of the study, resulting in 200 total capture events (Table 1). Sampled sturgeon ranged in size from 77.0 to 180.0 cm FL, with an average FL of 124.3 ± 1.3 cm. During the course of the study, 184 T-bar tags and 159 PIT tags were deployed.

3.2 Telemetry

Acoustic transmitters were surgically implanted in 43 Atlantic sturgeon between 2009 and 2011. Forty-two of these individuals were subsequently detected on the acoustic array deployed in the Saco River. The only tagged individual that was not detected in the river was tagged on 15 November, 2011 near the mouth of the Saco River. It is possible that this individual exited the river shortly after the tagging event and was not detected by the receiver in Saco Bay. All 21 sturgeon tagged in the Saco River during the 2009 season were redetected in the river in 2010. Forty of the acoustically tagged sturgeon were detected in the Saco River across two seasons, and 17 were detected across all three seasons of acoustic monitoring in the Saco River. All sturgeon with transmitters exited the Saco by late

November or early December during sampling seasons. In 2010 and 2011, tagged sturgeon began to arrive back at the Saco River in mid-April.

A total of 558,593 detections were logged during the study with an average of $12,695 \pm 2,106$ (mean \pm SE) detections per fish. The total number of detections per location ranged from 0 to 200,390. Receivers located at or below rkm 5 accounted for the majority (87%) of the total detections. The length of residence times for Atlantic sturgeon in the Saco River ranged from 0.02 to 3,517.17 hours (110.9 hours ± 6.9 ; mean \pm SE). Four Atlantic sturgeon were detected on acoustic receivers throughout the entire study area (to rkm 10). These four sturgeon ranged in size from 77.0 – 134.9 cm FL. Across all sampling seasons, sturgeon were detected in the river between the months of April and December, with the number of detections increasing through the summer months and peaking in October before sturgeon exited the river in late November or early December (g).

All detection data were pooled and entered into the lattice based home range model to obtain areas of 95% (general) and 50% (core) utilization for all Atlantic sturgeon that had been surgically implanted with transmitters in the Saco River (Fig 3). The results of this analysis indicate that 95% of sturgeon activity in the Saco River occurs between the natural river mouth and rkm 7. The core area of use (50% of activity) includes the area just inside the natural mouth of the river and extends to rkm 3.5. Analysis for each individual sturgeon, based on month, season, lunar cycle and tidal cycle revealed only small changes in the region of the study site being utilized based on these parameters. When color values were assigned to each receiver location based on the total number of detections at each site, it was

revealed that the two receivers just inside the natural mouth of the river had the most detections in each month. The receivers up river did display some variation in the number of pings detected. These receivers tended to log a greater proportion of the pings in April and December than other months (Fig 4)

When detections (% total) within the array were plotted against the time of day, it was evident that Atlantic sturgeon utilized the Saco River estuary more frequently at night during spring, summer, and fall (all $p < 0.001$; Fig 5). However, during winter, Atlantic sturgeon were observed more frequently during daytime ($\chi^2 = 4.9, p=0.027$).

Atlantic sturgeon tagged in this study were detected on acoustic receivers in several other locations in the Northeast region of the United States in addition to the array in the Saco River. A single individual that was last detected on October 12, 2009 was later detected about 220km away along the coast in Plymouth Bay, MA (41°59.22'N, -70°39.50'W) between May 25 and June 1, 2010. It returned to Plymouth Bay again on 3 June 2010 and remained in the area until 5 June 2010. This individual returned the Saco River on 9 June 2010 and remained until 13 October 2010. In addition to this individual, another Atlantic sturgeon tagged in the Saco River moved southward and was captured off the coast of Delaware by researchers from Delaware State University about 850km from the mouth of the Saco River. It was identified as a Saco River sturgeon by the PIT tag implanted at the base of the dorsal fin.

Five Atlantic sturgeon tagged in the Saco River were also detected on the DeepCwind test site buoy located south of Monhegan Island (43° 42.38'N, 69° 19.18'

W) about 95km northeast of the Saco River. These individuals were last detected in the Saco River during fall of 2010 and were recorded on the buoy between mid-January and mid-February of 2011 (Table 2). All of these fish subsequently returned to the Saco River in the spring or summer of 2011.

Four Atlantic sturgeon tagged in the Saco River between 4 April 2009 and 21 October 2011, were detected on a single acoustic receiver located at the mouth of the Presumpscot River in Casco Bay (43° 41.22'N, 70° 14.33'W), about 40 km from the mouth of the Saco River. One sturgeon tagged in September of 2010 moved north and was detected in the Casco Bay on 27 September 2010. Three other Atlantic sturgeon tagged in the Saco River in 2010 were detected in the Casco bay at various times between May and October of 2011.

3.3 Steroid Hormones

Blood samples were collected from a total of 168 sturgeon between October 2008 and September 2011. Of these samples, E₂ and T values were obtained for 164 individuals. Other samples did not yield enough plasma to obtain values for both T and E₂. Testosterone and estradiol values ranged from 2.8 to 177.3 ng ml⁻¹ and from 0.1 to 15.9 ng ml⁻¹, respectively. Initial sex assignment for each sturgeon was based on a comparison of obtained hormone values to those from Van Eenennaam et al. (1996). Based on these values, 138 individuals were assigned as female and 28 were assigned as male. Based on equations developed by Webb et al. (2004), 104 individuals were assigned as females and 60 were assigned as male. A comparison

between these two methods revealed a 78.1 % agreement with regard to the sex of sturgeon (Table 4).

The combination of the two sex determination methods revealed an overall sex ratio of 1:3 (M:F). Observed sex ratios were also examined for each sampling season during the study (Table 5). During each of the first two sampling seasons (2008 and 2009), the sex ratio was evenly spread between males and females. During the last two sampling seasons (2010 and 2011) the number of females per male increased to more than four females for every male sampled in the river. Dividing the sex ratio in the Saco River by season revealed that at all times of the year more female sturgeon were captured in the river than male sturgeon (Table 5). The sex ratio of captured sturgeon was nearly 1:1 during spring; however, increased significantly to almost three females for every male caught in the Saco River during summer and fall.

Values for (T/E_2) ranged from 0.56 to 48.94 for the 101 individuals identified as female by both assignment methods used in this study. Of these individuals, 95 had values below 25.00. Values for the 22 individuals identified as male by both methods ranged from 13.74 to 376.42. Of these, 19 had values above 30.00. Values for the 41 individuals for which agreement on sex was not achieved between the two methods ranged from 6.03 to 191.37.

3.4 Diet

Stomach contents were successfully obtained from a total of 31 Atlantic sturgeon between September of 2010 and November of 2011. Total weight of prey items in stomachs ranged from 0.4 to 40.6 g (6.1 ± 1.9 ; mean \pm SE). The total number of prey items identified in stomachs ranged from 2 to 155 (29.5 ± 5.8 ; mean \pm SE).

Sand lance (*Ammodytes americanus*) was the most dominant prey item (by weight) in 30 of the 31 stomachs examined (2.3-100.0%, mean = 84.8%) (Table 3), accounted for 48.7% of all prey items collected, and made up 83.4 % of the %IRI. Amphipod species were the second most important prey item consumed by Atlantic sturgeon in the Saco River (%IRI = 14.0), and accounted for 25.7% of the total number of prey items recovered from sturgeon stomachs. The %IRI for all other taxonomic groups encountered ranged from 1.3% to 0.01%, indicating that these prey items are of potentially of minor importance in the diet of Atlantic sturgeon in the Saco River. The cumulative prey curve approached an asymptote, indicating that the results of this study provide a good representation of the diet of Atlantic sturgeon in the Saco River estuary (Fig 6).

4. Discussion

While Atlantic sturgeon have been studied to some extent throughout the majority of its geographical range, much of the research on this species has been focused on the Hudson River estuary. Little work has been conducted in the Gulf of Maine and this study represents the first research conducted on this species in the Saco River. As such, this study provides new insights into habitat use, diet and movement of the Gulf of Maine population of Atlantic sturgeon and can be utilized for more complete future assessments of Atlantic sturgeon in this region.

4.1 Movement

Atlantic sturgeon were detected in the Saco River more frequently during nighttime than daytime. Overall, these results are generally consistent with those found by Wrege et al. (2011) for the Gulf of Mexico sturgeon. Wrege et al. (2011) found that the percentage of total detections began to decrease around 06:00 and began to increase again at about 15:00. However, differences were observed between these two studies when data were broken down by season. Saco River Atlantic sturgeon were detected more frequently in daytime during the winter months, while daytime detections were more common during summer months for Gulf of Mexico sturgeon (Wrege et al. 2011). These differences indicate that diel movement habits may change based on latitude.

Of the 42 Atlantic sturgeon tagged over the course of this study, only four individuals ranging in size from 77.0 – 134.9 cm FL used the entirety of the study area to

rkm 10. The sizes of these sturgeon indicate that they were juveniles or potentially maturing adults. Therefore, it was unlikely that these individuals were utilizing the river as a spawning habitat. Additionally, the majority of these upriver movements in the Saco River occurred outside the spawning season for this region. Water temperature is the primary cue for spring spawning migrations in Atlantic sturgeon and as a result, the timing of these movements varies across latitudes throughout their geographical range. For example, the presence of spent male sturgeon in rivers in the southern portion of their range indicate that spawning could take place as early as February or early March (Collins et al. 2000). Several studies in the Hudson River indicate that spawning occurs several months later in April or May (Dovel and Berggren 1983). In the Canadian portion of their range, Atlantic sturgeon are documented to spawn between May and late July (Collette and Klein-MacPhee 2002; Hatin and Caron 2002). Three individuals in the Saco River reached the most upriver portions of the study during August and September, placing these movements outside the observed spawning period for this region. Additionally, lattice-based home range analysis of these upriver movements indicated that the environmental parameters examined do not influence these upriver movements, further supporting that these are exploratory or foraging movements into the river.

A single individual was detected at the dam (rkm 10) in the Saco River during the spawning season, between 25 and 31 May, 2010. This sturgeon was the largest of the four that utilized the entire portion of the river available (134.9 cm FL). Based on the aforementioned size classes, this individual is likely still immature but may be maturing within the next few years. While the majority of sturgeon in the river are likely juveniles that are potentially looking for foraging habitat, the presence of this larger individual at

the dam during this time of year may indicate that individuals are exploring the river in search of other resources or habitats.

In addition to spawning migrations during spring and summer, studies have documented a secondary Atlantic sturgeon migration that occurs during fall where the sturgeon move into estuarine waters to presumably overwinter (Smith et al. 1984; Smith et al. 1985; Collins et al. 2000; Laney et al. 2007). This seaward movement appears to occur throughout the range of Atlantic sturgeon and has been documented in various months from October to December along different portions of the east coast. This same seaward movement was exhibited by sturgeon that were acoustically tagged in the current study. Detection data indicate that Atlantic sturgeon begin to arrive in the Saco River in April or May of each year. The number of detections in the river increases steadily across summer months, peaking in October and then beginning to decrease. It was noticed that a greater number of detections came from upstream receivers very early in the season (April) or very late in the season (December). This apparent shift in regions that sturgeon utilize most during different times of the year could indicate that sturgeon utilize different portions of the river more or less based on other factors. The full array of acoustic receivers was replaced with two overwinter receivers near the mouth of the river in November. Detections on these receivers ceased in late November or early December each year, indicating that all sturgeon had exited the Saco River for the winter.

The habitat use and movement patterns of Atlantic sturgeon in the marine environment are relatively unknown. However, several tagging studies have been conducted to better understand habitat use and movement of this species in the marine environment. A tag and recapture study by the Delaware Department of Fish and Wildlife

(n=120) in the Delaware River suggested that juvenile Atlantic sturgeon make extensive journeys along the eastern coast of the United States (Greene et al. 2009), since several individuals were recaptured as far south as Cape Hatteras, North Carolina during the winter and early spring months and as far north as the coastal waters of Maine during the summer months (Greene et al. 2009).

Stein et al. (2004) also examined the distribution of Atlantic sturgeon in the marine environment based on bycatch data from monitored fishing trips from 1989-2000. Atlantic sturgeon were typically caught in shallow (≤ 50 m depth) coastal waters from Massachusetts to Cape Hatteras, NC, and were found to utilize any habitat that supported benthic organisms for foraging.

A satellite tag study was conducted by Erickson (2011) to document their long-term movements and examined the movements of 23 adult Atlantic sturgeon from the Hudson River. Movement data was obtained from 15 of 23 individuals tagged during the course of this study. Thirteen of these sturgeon remained in the mid-Atlantic region between Cape Cod, MA and Cape Hatteras, NC. Kernel analysis of the movements of tagged individuals revealed that sturgeon from the Hudson River primarily aggregated off Long Island, Delaware Bay and the Chesapeake Bay. However, one sturgeon ventured as far south as Georgia and another as far north as Cobequid Bay, Nova Scotia, indicating that while most sturgeon remain in the presumed region of origin, others make longer migrations along the east coast of the United States.

Several Atlantic sturgeon tagged in the Saco River displayed long or short term coastal migrations. Both sturgeon that displayed larger scale migratory behavior moved south along the coast. One individual was detected at the mouth of Plymouth Bay, MA

and the other was recaptured off the coast of Delaware. The individual that was detected at the mouth of Plymouth Bay made the same coastal movement in more than one year. These movements are greater in scale than other coastal movements made by other sturgeon in this study, but remain within the Gulf of Maine region, similar to the individuals from the Hudson River that remained within the mid-Atlantic region (Erickson et al. 2011). Conversely, the other Atlantic sturgeon that made a long distance migration ventured outside of the Gulf of Maine and was captured off the coast of Delaware. This movement was similar in scope to the single individual from the Hudson River that moved north into Canada (Erickson et al. 2011) and the juvenile sturgeon that was tagged in Delaware and relocated off the coast of Maine (Greene et al. 2009). The river of origin for this individual is not currently known. As a result, it is not possible to determine if this southerly movement was away from a natal river in the Gulf of Maine or a movement to return to a natal river in the mid-Atlantic region.

In addition to the long-range movements that two individuals made to more southerly locations, several Atlantic sturgeon made shorter range movements within the Gulf of Maine at different times of the year. Nine Atlantic sturgeon were detected on receivers located to the north of the Saco River. Four of these individuals were detected in the Casco Bay (about 20 km northeast of the Saco River). Additionally, five of these individuals were detected on the buoy at the DeepCwind Test Site (43.70633N, 69.31967W). The fact that five of these sturgeon were detected in the same location within a short time span indicates that sturgeon may travel in groups in the marine environment or that there are other resources in the area that sturgeon are utilizing. It is also interesting to note that these individuals moved north during January and February at

the same time that the previously mentioned individual moved south to Massachusetts. The fact that several sturgeon moved north while others moved south over the winter months may be an indication that individuals from more than one natal river are utilizing the Saco River during the summer months. Given the uncertainty in the seaward migration patterns, greater tracking efforts are necessary to elucidate the movement patterns of Atlantic sturgeon in the marine environment and to determine how much populations may overlap at different times of the year.

4.2 Size Classes

It has been established that Atlantic sturgeon display clinal variation in growth based on latitude, with individuals from southern DPSs growing more quickly than those in the northern portion of their range. However, despite these differences in growth rates, Atlantic sturgeon reach sexual maturity at approximately the same size throughout their range. This results in differing age at maturity based on latitude. For example, Collins et al. (2000) reported mature male Atlantic sturgeon as young as age seven in South Carolina rivers. In contrast, Van Eenennaam and Doroshov (1998) found that the youngest mature male captured in the Hudson River over the course of their study was 12 years of age. Due to the consistency of size at maturity throughout their range, life stages of sturgeon are often generalized based on size. Life stages have previously been defined as juvenile (2-134.0 cm FL), non-spawning adults (≥ 135.0 cm FL), female spawners (≥ 180.0 cm FL) and male spawners ($\geq 135.0 - 190.0$ cm FL) (Bain 1997). The average size for Atlantic sturgeon captured in the Saco River was 124.30 (± 1.28 cm; sem) and

these fish were designated as juveniles. The lack of large adult females captured in the river indicates that the sturgeon inhabiting this system are likely not utilizing the estuary as an active spawning ground. However, utilization of the river in this manner is consistent with observations by Kieffer and Kynard (1993) in the Merrimack River, MA and Murawski and Pacheco (1977) in the Saint Lawrence River, Quebec, Canada, where late juvenile sturgeon will often inhabit riverine reaches during the summer months. Bain (1997) hypothesized that juveniles from larger rivers may annually use smaller rivers before returning to their natal rivers to spawn. If this is the case, the Saco River may function as an important seasonal habitat for juvenile sturgeon produced in a larger river along the east coast of the United States.

4.3 Reproduction

Reproductive hormones have been examined as an indicator of sex and reproductive stage in a variety of sturgeon species (Van Eenennaam et al. 1996; Webb and Erickson 2007; Webb et al. 2002; Wildhaber et al. 2006; Feist et al. 2004; Barannikova et al. 2004; Semenkova et al. 2002; Mojazi Amiriet et al. 1996a, 1996b). This method of sex determination is of particular interest in sturgeon species due to their threatened status. Blood collection is a simple, quick, inexpensive, minimally invasive procedure that can provide valuable information about the reproductive structure of a population while causing minimal stress to sampled individuals (Craig et al. 2009).

Results from previous studies indicate that although hormone concentrations vary between species, the pattern of hormone production throughout the reproductive cycle

remains consistent across species. Levels of both E₂ and T are low in young sturgeon until the differentiation of gonads (Craig et al. 2009). In male sturgeon, levels of testosterone increase and remain high throughout development of the testes (Mojazi Amiri et al. 1996a; Van Eenennaam et al. 1996; Webb et al. 2002; Erickson and Webb 2004) and decrease after spermiation (Webb et al. 2001). Similarly, T levels in female sturgeon increase during development of the ovaries (Mojazi Amiri et al. 1996b; Van Eenennaam et al. 1996; Webb et al. 1999; Webb and Erickson 2007) but decrease following ovulation (Webb et al. 1999). Levels of E₂ generally remain low in male individuals, but increase in females during vitellogenesis and subsequently decrease after spawning (Webb and Erickson 2007; Van Eenennaam et al. 1996).

Reproductive hormones have been used extensively in aquaculture as an early indicator of sex in several sturgeon species (Petoichi et al. 2011; Barannikova et al. 2004; Feist et al. 2004; Semenikova et al. 2002) and have recently been explored by several studies as a potential method for determining sex in wild individuals (Van Eenennaam et al. 1996; Webb et al. 2002; Craig et al. 2009). Determination of the sex ratio of a population of wild sturgeon is important for proper management (McLeod et al. 1999). This information is useful in determining the potential spawning stock biomass of a population because the number of females is crucial for these calculations (Craig et al. 2009). Additionally, this information is important when considering species that display differences in growth rate, reproductive periodicity or mortality between sexes (Craig et al. 2009).

Webb et al. (2002) examined the potential for using plasma indicators for sexing wild white sturgeon. They collected gonad samples from 257 individuals and examined a

suite of blood plasma indicators, and concluded that E₂ and T were the most reliable blood parameters for indicating sex and reproductive stage of white sturgeon. More recently, Webb and Erickson (2007) also utilized steroid hormone concentrations to describe the reproductive structure of the green sturgeon population in the Rogue River, Oregon. This study found that concentrations of E₂ and T in green sturgeon differed from those of other species, but that the pattern of hormone production throughout the reproductive cycle remains the same, with E₂ levels peaking near 5 ng/mL in vitellogenic females and T peaking near 125 ng/mL in spermiating males.

Van Eenennaam et al. (1996) conducted a study on Atlantic sturgeon in the Hudson River to determine the reproductive conditions of the population. This study focused on mature individuals (> 270.0 cm FL) and involved collection of blood and gonad tissue to verify the sexes of individual sturgeon. Van Eenennaam et al. (1996) found that the plasma concentrations of sex steroids in Atlantic sturgeon were similar to those observed in brood stock of white and Siberian sturgeon. Levels were also similar to those obtained by Webb and Erickson (2007) for the green sturgeon. Mature male Atlantic sturgeon demonstrated a mean T level of 126.83 ng/mL. Female sturgeon displayed higher levels of E₂, with gravid females averaging 5.10 ng/mL.

Finally, Craig et al. (2009) utilized reproductive hormones as an indicator of sex in lake sturgeon. This study was unique in that researchers were unable to collect gonad tissue due to the protected status of the lake sturgeon. This study also utilized the previously described model for white sturgeon of Webb et al. (2002) to aid in identification of sex. Craig et al. (2009) found that this model correctly predicted the sex of 75% of females and 88% of the males that could be visually identified in the field

based on the release of eggs or milt. These values were comparable to the accuracy of the model itself, and indicate that statistical models developed for sturgeon species may be able to effectively assist in identification of sex for species that cannot be sampled as extensively due to protected status.

The sex ratio of the Atlantic sturgeon population sampled in the Saco River (M:F) ranged from 1:1.2 (2009) to 1:4.4 (2010) across years. When all samples from all seasons were pooled, the total sex ratio for the Saco River was 1:2.7. These sex ratios are markedly different from previously mentioned studies on wild sturgeon populations. Craig et al. (2009) found that male lake sturgeon outnumbered females by a ratio of 1:2.7. Van Eenennaam et al. (1996) found a similar situation in Hudson River Atlantic sturgeon with an overall observed sex ratio of 1:2.4 (F:M). The only other study that recorded a population with larger numbers of females than males (M:F) was Webb et al. (2002) (1:1.4). The greater number of females in the Saco River population indicates that the population would be more resilient to fishing pressures and other causes of population declines if these were mature females spawning in the river. However, although the population is comprised of a greater percentage of females (73.1%) only a single female was large enough to be classified as a female spawner, indicating that the Atlantic sturgeon inhabiting the Saco River are not likely to be a spawning population. It is also important to note that when the T/E_2 ratios were examined, there was some uncertainty about the assignment of sexes. There was overlap in the ratios between those assigned as male and female. The 95 individuals assigned a sex of female and having a T/E_2 ratio less than 25.0 were considered to be positively female. The 19 that were assigned a sex of male and had a T/E_2 ratio greater than 30 were considered to be positively male. This

leaves a total of 50 sturgeon for which an agreement of sex could not be agreed upon by the two methods used or for which the T/E₂ ratio could not definitively separate them as one sex or the other. Additional research is necessary on circulating hormones in Atlantic sturgeon in order to minimize some of this uncertainty and promote definitive identification of sex using this methodology.

In New England and Canada spawning of Atlantic sturgeon is typically documented in tidal freshwater reaches of rivers above the salt wedge (Dovel 1978, 1979; Smith 1985; Van Eenennaam et al. 1996; Bain et al. 2000). The majority of research on spawning Atlantic sturgeon in this region has taken place in the Hudson River. Spawning in this system reportedly occurs at great distances from the coast, with ovulating female sturgeon being found 136 km upriver (Van Eenennaam et al. 1996). During spawning, eggs are released into flowing water and are dispersed downstream for a period of about 20 minutes before the surface of the eggs becomes adhesive and they settle onto solid substrate (Van den Avyle 1984). Van Eenennaam et al. (1996) suggested that Atlantic sturgeon spawning requires significant clearance in front of the salt water interface to accommodate this dispersal of embryos and larvae as even small quantities of salt are lethal to early life stages of Atlantic sturgeon.

Reynolds and Casterlin (1985) evaluated the hydrographic environment in the Saco River estuary at different times of the year and different parts of the tidal cycle. At low tide during the spring, the estuary was filled top to bottom with fresh water to a point just inside the natural mouth of the river. However, at high tide during the fall, near bed salinity was found to reach levels of approximately 12ppt at rkm 10. Based on this information, it is not currently known if there is sufficient

clearance in front of the salt wedge for an extended period of time to accommodate sturgeon spawning activity in the future.

4.4 Diet

Home range analysis of acoustic data from the Saco River indicates that the majority of Atlantic sturgeon activity occurs very close to the natural mouth of the river. Previous studies on the fish community of the Saco River and its associated bay documented the presence of American sand lance in the sandy areas at the mouth of the river (Furey and Sulikowski 2011; Reynolds and Casterlin 1985). Reynolds and Casterlin (1985) conducted a comprehensive survey of the vagile macrofauna in the Saco River. Over the course of this study, American sand lance were captured over sand substrates near the mouth of the river. Similarly, Furey and Sulikowski (2011) used beach seines, beam trawls and otter trawls to survey the fish assemblage structure of the Saco River estuary. This study recorded a total of twenty-four species of fish in the Saco River estuary. American sand lance was one of the five most commonly captured species throughout the course of the study. In addition, unpublished data from beach seines conducted from June through September by Sulikowski confirms that large schools of American sand lance (*Ammodytes americanus*) are still present in the sandy areas near the mouth of the river during the summer months, with greatest numbers of sand lance being captured in June and July.

Scott and Crossman (1973) reported that the diet of juvenile Atlantic sturgeon in Canadian waters consists primarily of sludge worms, mayfly larvae, isopods, amphipods

and small bivalve mollusks. The diet of adult Atlantic sturgeon inhabiting riverine stretches was reported to include gastropods and benthic organisms, while the diets of adults in salt water are reported to consist primarily of amphipods, isopods, shrimp, polychaete worms, mollusks and small fishes. Sand lances were the most commonly reported fish species (Scott and Crossman 1973). A more recent study by Johnson et al. (1997) examined the diet of Atlantic sturgeon off the Central New Jersey coast. Based on 275 stomachs donated by local commercial fishermen polychaete worms and isopods were determined to be the most important prey items in the diets of sturgeon in this region. These disparities between the diet of Atlantic sturgeon in the two regions may be a function of time or of regional prey availability. Although the current study has a limited sample size, the major components of the diet are similar to those found by Scott and Crossman (1973) and the cumulative prey curve began to approach an asymptote, indicating that the diet is adequately described, despite the small sample size. The extensive amount of time spent at the mouth of the Saco River by Atlantic sturgeon and the continued presence of large schools of sand lance throughout the summer months indicates that this estuary provides a summer foraging habitat for Atlantic sturgeon that several individuals return to annually.

This study provides the first comprehensive examination of the Atlantic sturgeon population in the Saco River, ME. While this river is not likely an active spawning site, it still serves a potentially important function as a foraging ground for late juvenile sturgeon during the summer and fall months and the importance of intermediate rivers in the range of the Atlantic sturgeon should not be overlooked when examining matters of policy as the status of this species changes. Indications that the sex ratios in the juvenile

population may be skewed towards females are promising for the future of this species in the Saco River and beyond. Further examination of the movements of this species in the marine environment is also necessary to further refine the movement patterns and behavior of Atlantic sturgeon to ensure that the best possible measures are taken to protect this species and facilitate a full recovery of the population.

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Table 1. Summary of capture and tagging events for Atlantic sturgeon between October 2008 and November 2011 in the Saco River estuary. Table presents total number of capture events per season as well as average total length (TL), fork length (FL), number of PIT tags, T-bar tags and acoustic transmitters deployed each season. CPUE is calculated as number of sturgeon caught per hour of soak time during the season. Observed sex ratios are based on hormone values obtained from blood samples.

Year	Capture Events	Avg TL (cm)	Avg FL (cm)	PIT Tags	T-Bar Tags	Acoustic Transmitters	CPUE	Observed Sex Ratio (M:F)
2008	16	128.7	112.7	0	0	0	12.47	1:1.7
2009	33	143.3	127.5	33	33	21	2.80	1:1.2
2010	55	135.1	119.2	56	55	20	8.89	1:2.0
2011	96	142.6	125.5	70	96	4	6.40	1:4.4
Totals	200	137.4	121.2	159	184	45	5.70	1:2.7

Table 2. Percentage of detections during night (18:00-06:00) and day (06:00-18:00) hours, total number of observations (n), chi-square statistic, and the probability of rejecting the null hypothesis of Atlantic sturgeon recorded on receivers in the Saco River from June, 2009 through November, 2011.

Season	Night	Day	n	χ^2	p
Fall	60.1	39.9	284,297	14968.63	<0.001
Winter	67.5	32.5	1,037	127.07	<0.001
Spring	48.6	51.4	46,517	36.61	<0.001
Summer	48.2	51.8	226,742	296.69	<0.001
Combined	54.3	45.7	558,593	4144.44	<0.001

Table 3. Transmitter numbers for sturgeon detected on the DeepC Wind buoy and dates these individuals exited the river in 2010, were detected on the buoy, and returned to the Saco River in 2011.

Transmitter Number	Exited Saco River	Detected on DeepC Wind Buoy	Returned to Saco River
46817	11/29/2010	2/10/2011	4/25/2011
46829	12/6/2010	1/11/2011	6/6/2011
54186	11/5/2010	1/11/2011	7/2/2011
54188	10/31/2010	2/16/2011	4/14/2011
54189	11/25/2010	2/14/2011	4/14/2001

Table 4. Results of the sex assignment for both methods based on steroid hormone values obtained using radioimmunoassay.

Sex Assignment Method	Total Males	Total Females	Approximate Sex Ratio (M:F)
Van Eenannam et al. (1996)	28	136	1:5
Webb et al. (2004)	60	104	1:2
Combined	88	240	1:3

Table 5. Seasonal sex ratios observed. Ratios were generated by combining results from both methods used to determine sex of sturgeon captured in the Saco River.

Season	Males	Females	Observed Sex Ratio
Spring	5	7	1:1.4
Summer	40	120	1:3
Fall	43	113	1:2.6

Table 6. Identified taxon in stomach contents of sturgeon in the Saco River (n=31), the frequency (%F), percent of total stomachs (%N), and calculated %IRI for each taxon.

Taxon	Common Name	%F	%N	%W	%IRI
Ammodytidae	Sand Lance	36.1	48.7	87.8	83.4
Gammaridae	Amphipod spp.	27.9	25.7	4.0	14.0
Nemertea	Tube worms, ribbon worms	1.2	16.4	3.9	0.4
Crangonidae	Sand Shrimp	12.8	4.1	1.9	1.3
Portunidae	European Green Crab	2.3	0.2	0.1	<0.1
Nereididae	Clam Worm	2.3	0.4	1.8	0.1
Myidae	Surf Clam	4.7	0.8	0.1	0.1
Mytilidae	Blue Mussel	11.6	3.5	0.2	0.7
	Unidentified Egg Mass	1.2	0.1	0.1	<0.178573

Figure Captions

Fig 1.

Location of acoustic receiver array in the Saco River estuary. Area of the array is denoted by the small, black rectangle on the inset map. Approximate river kilometers are denoted with numbers along the southern edge of the river.

Fig 2.

Observations (% total) in relation to month for Atlantic sturgeon in the Saco River estuary from June 2009 through November 2011.

Fig 3.

Lattice based structure of the lower portion of the Saco River and Saco Bay. Darker shading represents areas of a) 95% (general activity space) and b) 50% (core area) utilization by Atlantic sturgeon in the Saco River estuary between June, 2009 and November, 2011.

Fig. 4.

Maps of the Saco River displaying receiver locations. Total number of detections by month at each location is indicated by the color of the receiver. More detections are indicated by darker shades of red. Fewer detections are indicated by lighter shades of yellow

Fig 5.

Observations (% total) in relation to time of day (h) for Atlantic sturgeon in the Saco River on acoustic receivers from June, 2009 through November, 2011.

Fig 6.

Cumulative prey curve for stomachs evaluated during this study (n=31).

Fig. 1

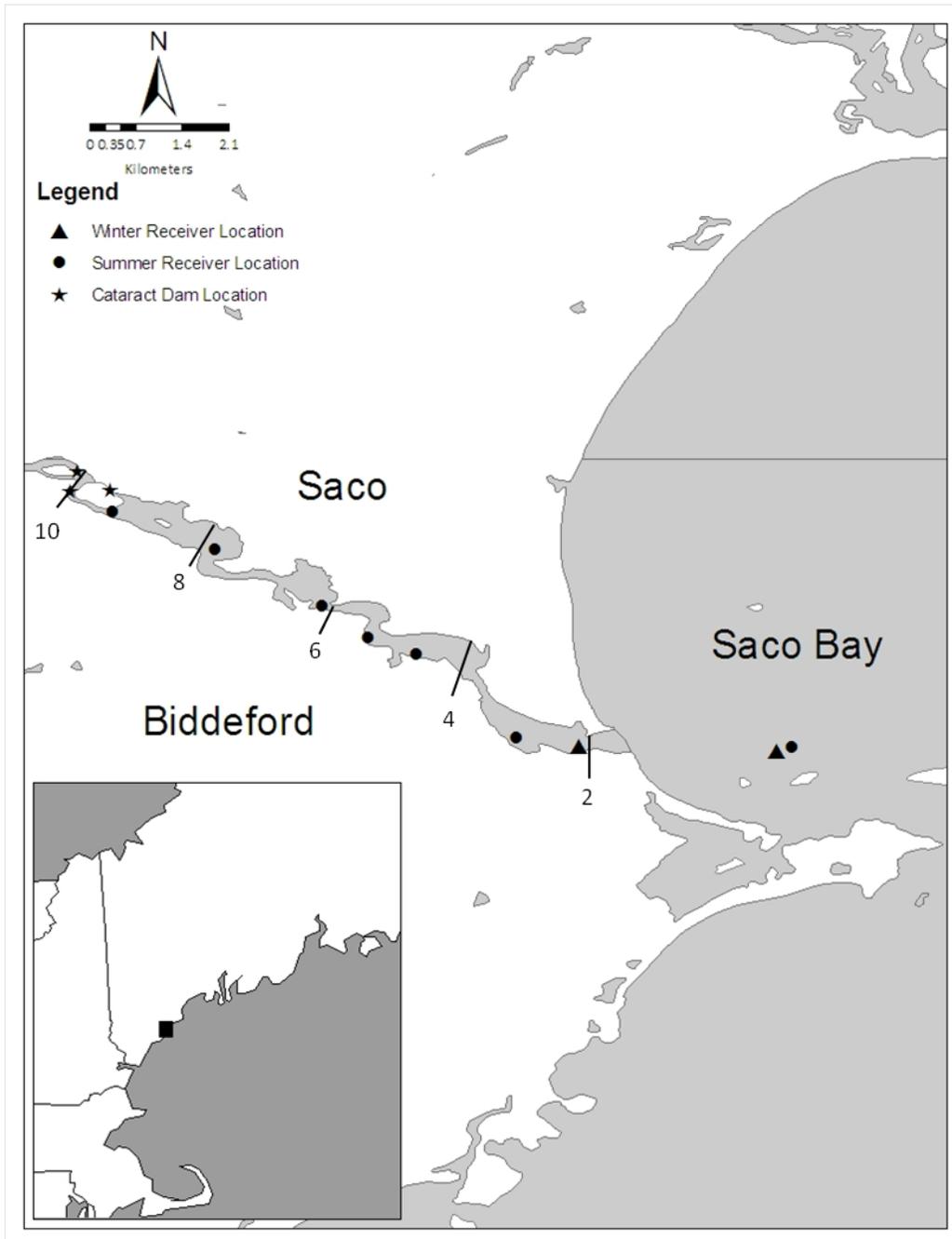


Fig. 2

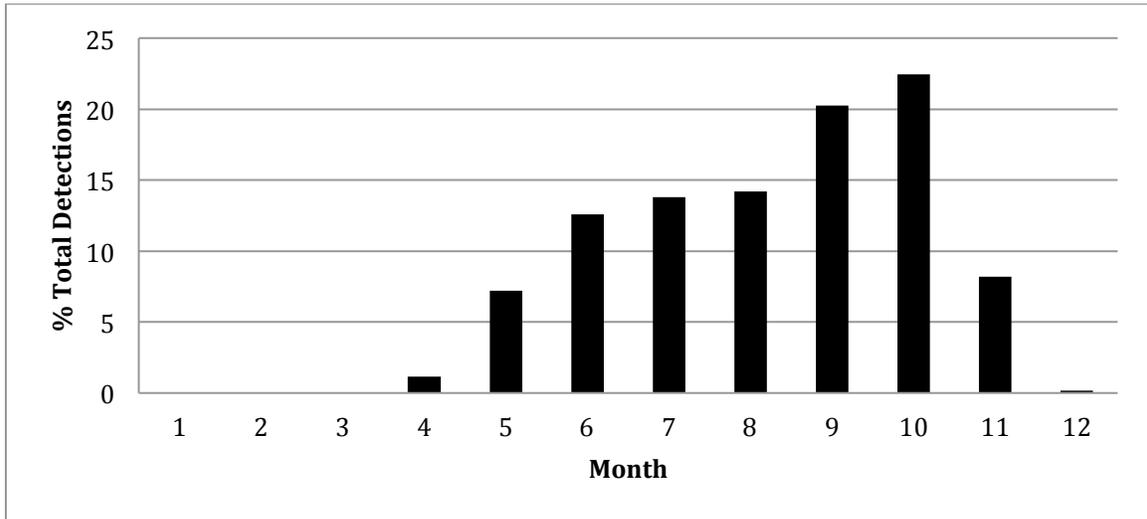


Fig. 3

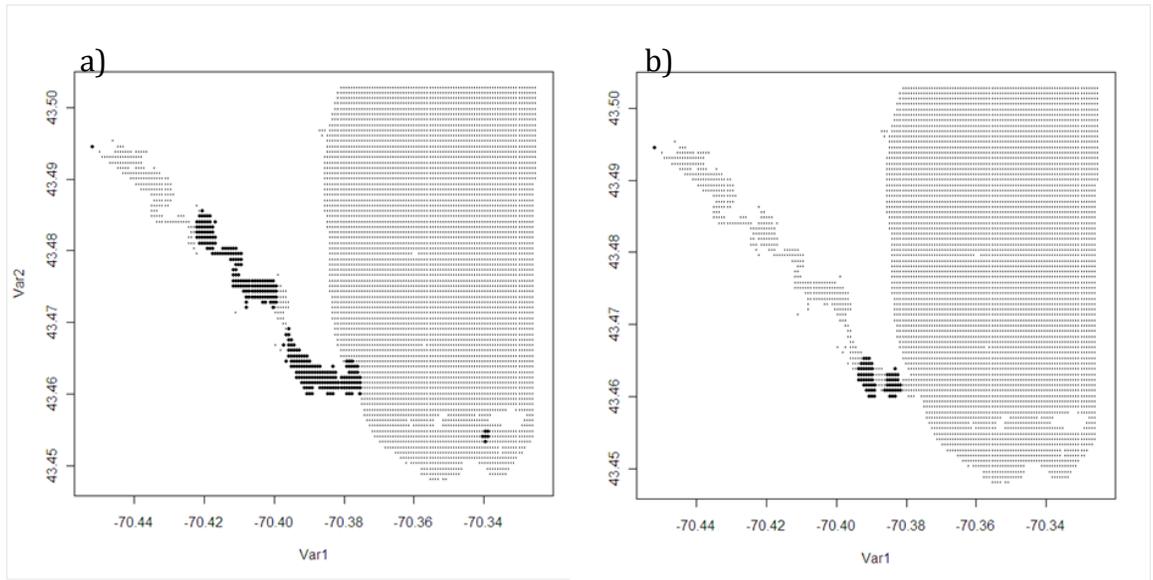


Fig. 4

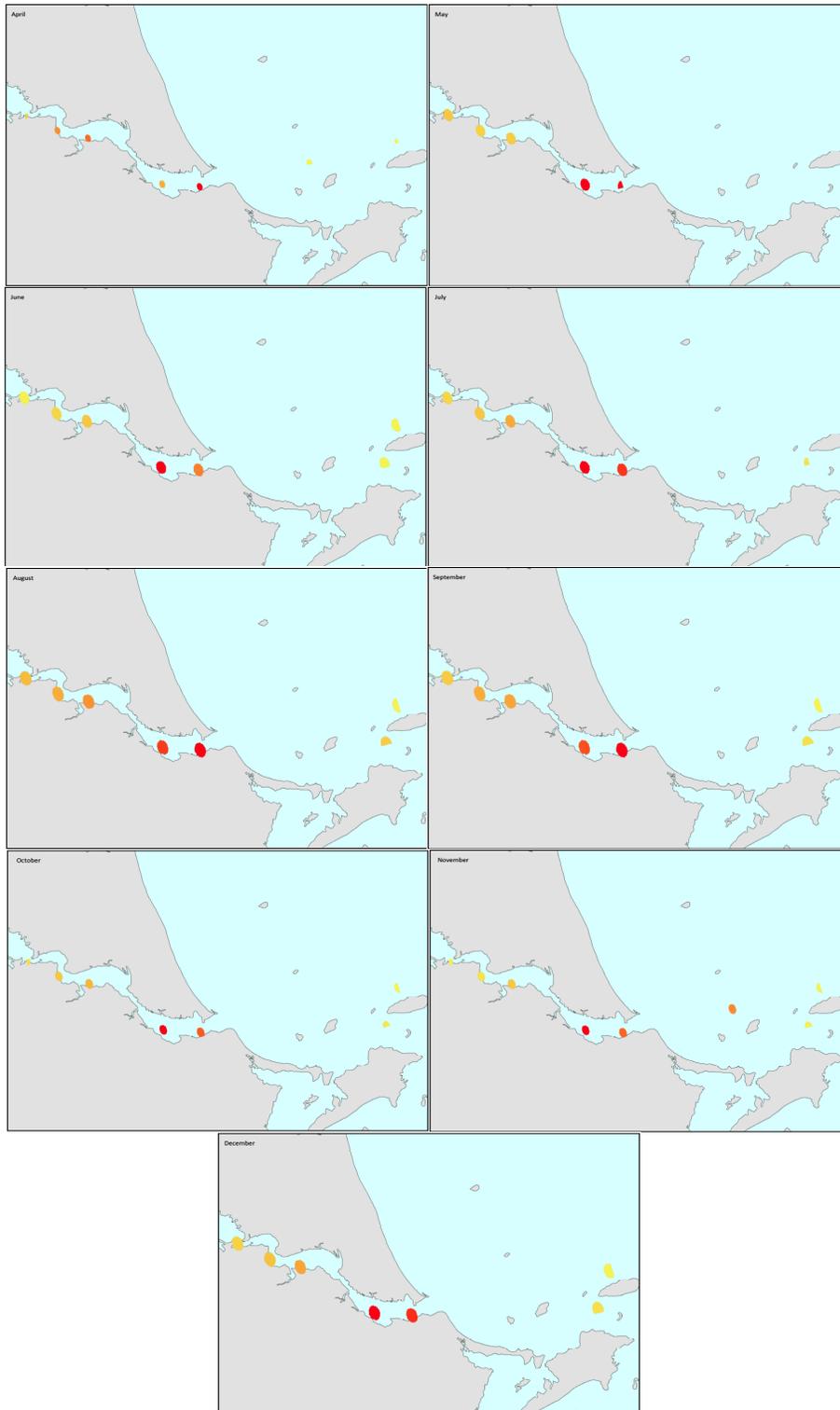


Fig. 5

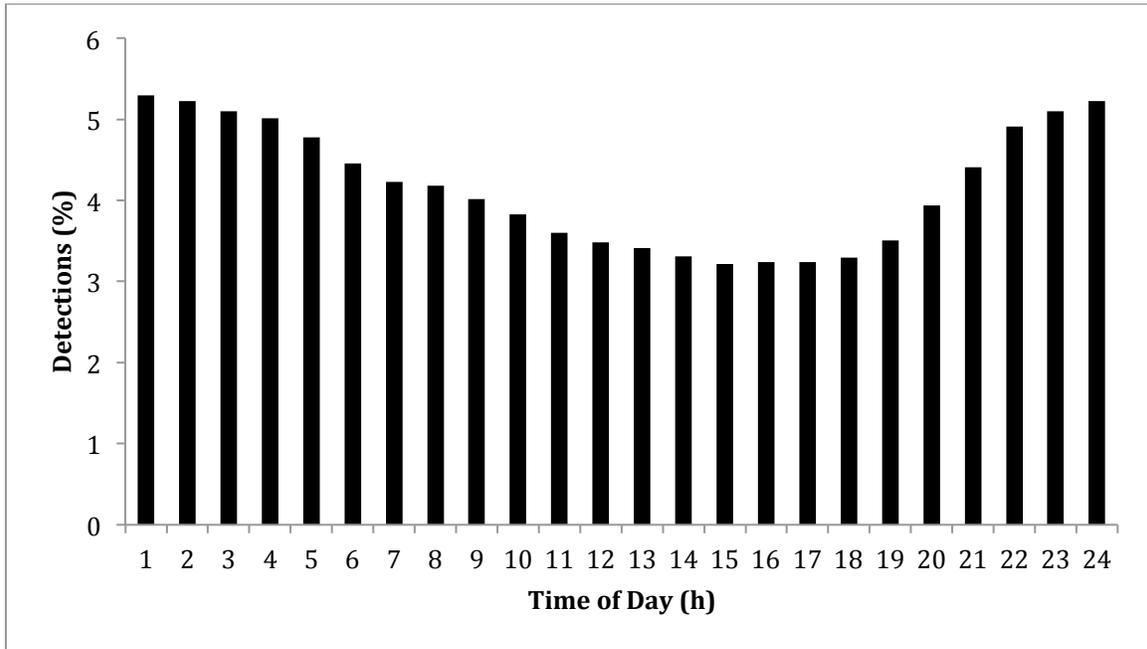
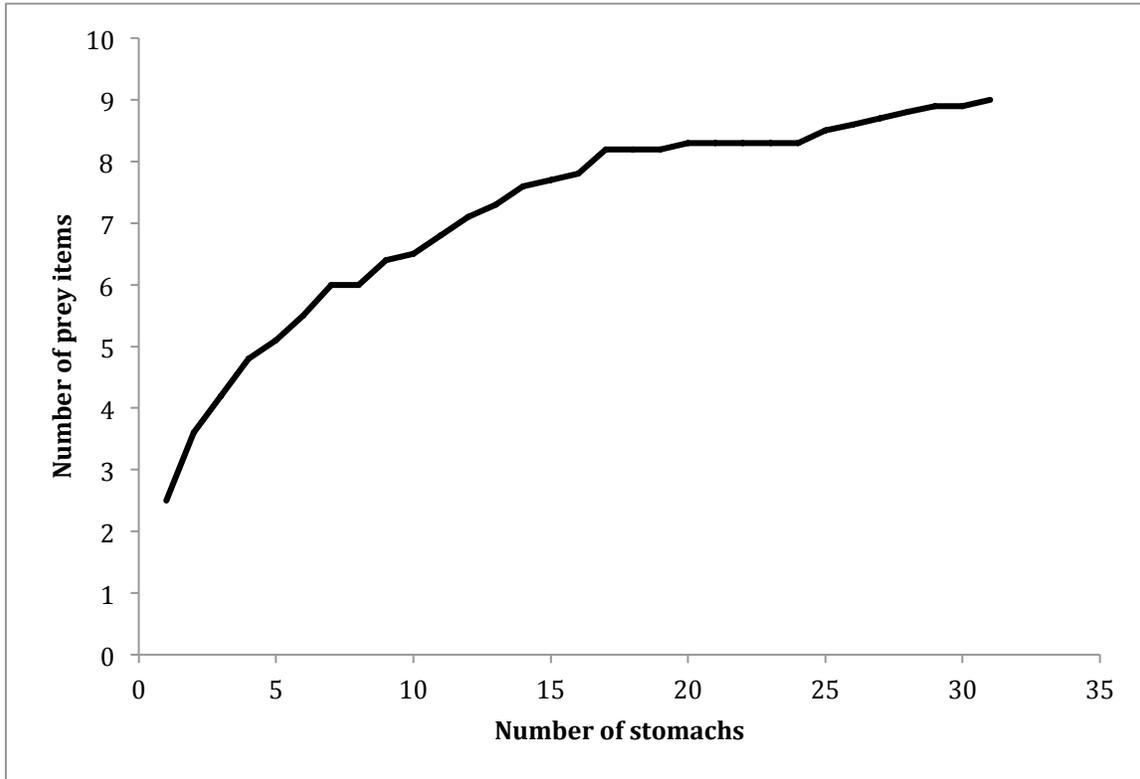


Fig. 6



GENERAL CONCLUSION

The data presented in this thesis represents the first study of the population of Atlantic sturgeon in the Saco River estuary and bay to date. The results indicate that the Saco River may be more important to the life cycle of these species that was previously thought. Published studies on the fish community of the Saco River either failed to document these species or documented only few sturgeon in the system (Reynolds and Casterlin 1985; Furey and Sulikowski 2011).

This study provides the first documentation of the shortnose sturgeon, an endangered species, in this system. Movements of individuals between several river systems in the Gulf of Maine are in agreement with current studies suggesting that coastal migrations for this species may be more common than previously thought (Fernandes et al. 2010; Zylewski et al. 2011). Further investigation is warranted to determine how many sturgeon are using the river, how much time they are spending in the river and if the river is functioning as a foraging ground for this species.

The number of individual Atlantic sturgeon captured in the Saco River indicates that it is an important habitat for this species between the spring and fall months. Movement data collected within the system shows that the majority of activity for this species in the river is focused near the mouth. Furey and Sulikowski 2011 found that American sand lance was the predominant fish species in this part of the river. This indicates that the Saco River may function as an important spawning ground for the Atlantic sturgeon from the spring to the late fall. Sizes of sturgeon in the river indicate that the population is composed primarily of juveniles

and subadults. This indicates that this river does not currently serve as a spawning ground. However, the presence of a few sturgeon of adult size could indicate that the river may provide spawning habitat in the future for individuals looking for a new river to inhabit. In general, more research is necessary to determine the full extent of sturgeon utilization of the Saco River.

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