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Asian Carp Population Status and Reproductive Potential in Illinois River Tributaries

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Asian Carp Population Status and Reproductive Potential in Illinois River Tributaries

(TITLE)

BY

Clinton Wesley Morgeson

THESIS

SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS
FOR THE DEGREE OF

Master of Science

IN THE GRADUATE SCHOOL, EASTERN ILLINOIS UNIVERSITY
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ASIAN CARP POPULATION STATUS AND REPRODUCTIVE POTENTIAL IN ILLINOIS RIVER TRIBUTARIES

By

Clinton Wesley Morgeson

B.S. Western Illinois University, 2013

A Thesis

Submitted for the Requirements for the Degree of

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ABSTRACT

Large floodplain rivers are among the most complex natural systems characterized by exchange between the river and its floodplain. Information regarding fishes in main channel habitats is abundant, however much less understanding extends into their tributaries. Tributaries of large rivers provide critical habitats for main channel fishes, important for foraging, spawning, and refuge. In addition to native diversity, large rivers are host to multiple exotic species invasions, threatening biodiversity, ecosystem function, and habitat quality. Two notorious invaders are Silver Carp (*Hypophthalmichthys molitrix)* and Bighead Carp (*H. nobilis*), collectively Asian Carp, presenting an imminent threat of invasion to the Great Lakes. Reproduction in lentic systems such as the Great Lakes is unlikely, however, a knowledge gap remains regarding their use of small rivers and tributaries, a large network of which is associated with the Great Lakes. This study sought to quantify invasive Asian Carp population characteristics and reproductive abilities in tributaries of the Illinois River, a highly invaded system artificially connected to Lake Michigan. To accomplish this, I sampled five Illinois River tributaries: Salt Creek, the Mackinaw, Kankakee, Sangamon, and Spoon Rivers during 2013-2014 for larval fish and adult Asian Carp, as well as the Illinois River main channel. I compared relative density, size structure, relative weight, reproductive condition, age structure, mortality, and growth of Asian Carp among the Illinois River and its tributaries. Relatively high abundances of Silver Carp were encountered in the main channel and all tributaries sampled except for the Kankakee River. Silver carp were found to temporally increase in size and age, but no differences were observed in condition. Reproductive condition of Silver Carp followed temporal hydrography, with peaks in gonad fullness and stage following peaks in discharge. In addition to adult population dynamics, I also examined spatial and temporal distributions, community assemblages, and size structure of eggs and larval fish. Peak abundances of eggs and larval fish followed peaks in discharge during May-June in each tributary, indicating that the fish in these systems are behaving like fluvial specialists in terms of spawning activity. Ten different families were sampled; clupeids and cyprinids were the most abundant taxa. Significant temporal and spatial effects ($p < 0.05$) of community assemblages were driven by secchi depth and water temperature, respectively. Post gas bladder emergent larvae of Asian Carp species were sampled in high abundance in the Spoon and Mackinaw Rivers, representing over 80% of our sample. Asian Carp were first sampled in late May with peak densities in July. My study suggests Illinois River tributaries are important habitats for early life stages of multiple fish species and contribute to the diversity of fishes in the main channel habitats. Understanding population characteristics of Asian Carps and larval fish community dynamics in small North American rivers helps bridge the knowledge gap and advance future efforts for the control of these species.

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I would like to first thank my advisor, Dr. Robert Colombo, for his support throughout my graduate career. Without his guidance and confidence, I undoubtedly would not be the successful fisheries biologist I am now. He is a dedicated mentor and friend, who invests unwavering passion in each of his students. My sincere thanks must also be extended to my co-advisor, Dr. David Wahl, without whose guidance and collaboration would have rendered this project inferior; and Dr. Anabela Maia, whose comments and direction have been invaluable.

This project would have not been possible without the assistance of the Illinois Natural History Survey and the Illinois Department of Natural Resources. I would extend a special thanks to Matt Diana and Steve Butler of the Kaskaskia Biological Station as well as Levi Solomon and Rich Pendleton of the Illinois River Biological Station for sample contributions and field assistance. Additionally, the support of the EIU Fisheries and Aquatic Research Team has made this research possible, especially Ryan Hastings and Evan Boone for field assistance, Alex Sotola for helping age every fish in the study, and Danny Flood, Kaity Knieft, and Missy Eaton for laboratory assistance.

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INTRODUCTION

Predating European settlement of North America, humans have transferred species beyond their native ranges. As geographical isolation continues to be diminished by human influence, the rate of exotic species introduction continues to increase (Courtenay 2007). Relatively few of these introductions become established, but the populations that persist have the potential to negatively influence native ecosystems. These deleterious species are known as "biological invaders" or invasive species. The introduction of non-native species to a novel environment can have detrimental effects on native populations such as competition, exclusion, displacement, hybridization, and predation (Mooney and Cleland 2001). These effects can have economic impacts as well, with invasive species costing native systems up to $120 billion in damages per year (Pimental et al. 2005). Evaluating these impacts on native systems remains of utmost importance for managers.

Successful biological invaders often possess a suite of traits which contribute to their success. Some of the attributes that contribute to the spread of successful invaders are broad environmental tolerances, wide distribution, short generation times, high growth rates, high fecundity, and high rates of dispersal (Ricciardi 2015). Humans facilitate these introductions by raising non-native species for consumption, aquaria, or biological controls (e.g. algae control in wastewater ponds), through industrial transportation (e.g. barge ballast, accidental vehicle transport), eliminating natural barriers and thus increasing connectivity of habitats (e.g. canals connecting freshwater bodies), and disturbance of habitats (Courtenay 2007). Invasive species often tend to be disturbance-driven species, rapidly colonizing disturbed areas and displacing less tolerant native species (Ricciardi 2015). The increasing levels of anthropogenic disturbance has greatly facilitated the spread of invasive species worldwide.

It has been estimated that about 50,000 non-native species have been introduced to the United States alone (Pimental et al. 2005). Fishes are no exception to this; it has been suggested more than 500 non-native fish taxa have been introduced to U.S. inland waters since the early 1800s (Nico and Fuller 1999). The world's largest freshwater resource, the Laurentian Great Lakes, has been host to multiple introductions of non-native species, some of which may have aided ecological detriment. Introductions such as the Sea Lamprey (*Petromyzon marinus*), Common Carp (*Cyprinus carpio*), and Zebra Mussels (*Dreissena polymorpha*) have been the source of major economic and ecological detriment to the Great Lakes by either competing with, or preying upon, native species (Mills et al. 1994). Asian Carp of the genus *Hypophthalmichthys* are among the most recent and notorious threats to the Great Lakes (Moy et al. 2011).

The term Asian Carp refers to two species Silver (*Hypophthalmichthys molitrix*) and Bighead Carps (*H. nobilis*), excluding other species of carp native to Asia: Grass Carp (*Ctenopharyngodon idella*), Black Carp (*Mylopharyngodon piceus*), and Common Carp (*Cyprinus carpio*) (Chick and Pegg 2001). Asian Carp are endemic to Eastern Asia and have been either cultured or introduced around the world (Kolar et al. 2005). In their native range, Asian Carp inhabit rivers, lakes, and reservoirs (Kolar et al. 2005; Jennings 1988). These carp were introduced to North America in the early 1970s for phytoplankton control in aquaculture ponds and wastewater treatment facilities in the Midwestern United States (Freeze and Henderson 1982). Subsequent flooding events allowed Asian Carp to escape into the Mississippi River basin, with reproducing populations occurring thereafter, severely impacting the newly invaded system (Williamson and Garvey 2005).

Silver and Bighead Carps are filter-feeding fishes in the family Cyprinidae, the largest family of freshwater fishes in the world (Winfield and Nelson 1991). Both species of Asian Carp are opportunistic planktivores, feeding on a variety of plankton and algae species. Bighead Carp are primarily zooplanktivores, whereas Silver Carp are primarily phytoplanktivores, exhibiting an ability to filter smaller particles than Bighead Carp (Sampson et al. 2009; Kolar et al. 2005; Jennings 1988). These two carps are voracious feeders, and have been shown to alter natural plankton communities in invaded systems (Sass et al. 2014). This impact has the potential to affect the food web at its lowest level, by affecting the recruitment of early life stages of fish that depend on plankton for nourishment (Dettmers et al. 2001). Studies suggest this feeding strategy puts Asian Carp in competition with native North American planktivores such as Gizzard Shad (*Dorosoma cepedianum*), Bigmouth Buffalo (*Ictiobus cyrinus*), and Paddlefish (*Polyodon spathula*), leading to reduced condition of native planktivores following the establishment of Asian Carp populations (Irons et al. 2007; Sampson et al. 2009). These highly efficient feeding strategies have contributed to the successful invasion in the Mississippi River system by these individuals.

Asian Carp spawn prolifically both in their native range and in introduced areas. The potential to release over half a million eggs per spawning event, in some cases multiple times per year (Abdusamadov 1987; Garvey 2006), contributes to high fecundity of these fish. Sexual maturity is reached at three to four years of age, depending on environmental conditions (Abdusamadov 1987). Peak spawning occurs in the spring and early summer associated with spring flooding events, however, Asian Carp can reproduce multiple times per year, if conditions allow (Kolar et al. 2005; Garcia et al. 2013). Spawning occurs near the surface in flows high enough to keep the semi-buoyant eggs suspended until hatch

(Abdusamadov 1987; Pflieger 1997). Prior to spawning, Asian Carp have been shown to engage in upstream migrations to spawning sites, studies have suggested that they can move over 60 km (DeGrandchamp et al. 2008), sometimes to discrete spawning sites (Kolar et al. 2005). Several specific environmental requirements have been shown to characterize Asian Carp spawning success: long stretches of unimpeded flow, water temperatures above 17°C, increased velocities, turbidity, and water levels associated with spring and summer flood events (Abdusamadov 1987). These requirements make Asian Carp unable to reproduce in lentic systems (Kolar et al. 2005; Jennings 1988).

One of the United States' greatest freshwater resources, the Laurentian Great Lakes, is at the forefront of Asian Carp invasion. These fish have been very successful invading our nation's large river systems, given their high degree of dispersal, resource consumption, and incredible fecundity; however, their potential for reproduction within the Great Lakes is limited. Therefore, it becomes important to identify areas that would satisfy the environmental requirements needed by Asian Carp to reproduce within the Great Lakes. While the Great Lakes themselves may be unsuitable for Asian Carp reproduction, the associated network of tributaries may provide appropriate spawning habitat. Similar to the tributary-targeted management efforts against Sea Lampreys within the Great Lakes (Christie and Goddard 2003), tributaries may be locations for targeted management of Asian Carp following their invasion.

Objectives

A major tributary of Lake Michigan, the Illinois River, is highly studied with regards to Asian Carp (Chick and Pegg 2001; Degrandchamp et al. 2008; Irons et al. 2007; Sampson et al. 2009; Stuck et al. 2015), but its tributaries remain unstudied. This work aims to breach this gap by studying Asian Carp and their reproductive abilities within tributaries of the Illinois River. I want to achieve the following objectives: (1) analyze population characteristics of Asian Carp within tributaries of the Illinois River, (2) determine reproductive capabilities of Asian Carp in tributaries, and (3) estimate the larval fish communities within these tributaries and the effect invasive Asian Carp have on assemblage structure. This research becomes not only important for management within the Illinois River system, but on a broader scale as a surrogate of spawning areas within the Great Lakes.

Study Area

The Illinois River is an area of high Asian Carp abundance (Baerwaldt 2013; Sass et al. 2010; Chick and Pegg 2001), formed by the confluence of the Kankakee and Des Plaines Rivers and connects the Mississippi River system to the Great Lakes via Lake Michigan. The Chicago River was redirected in 1900 and connected to the Des Plaines River by the Chicago Sanitary and Ship Canal (CSSC), effectively linking the Mississippi River to Lake Michigan for transportation and wastewater removal (Moy et al. 2011). The Illinois River is dammed in five places from its origin to its confluence with the Mississippi, forming six reaches more characteristic of slow moving lakes than a free flowing river, forming a highly disturbed habitat (Mills et al. 1966). Asian Carp were first detected in the Illinois River in 1995 (McClelland et al. 2012). Establishment of Asian Carp in Lake Michigan could have catastrophic effects, potentially impacting commercial and recreational industry worth around seven billion dollars (ASA 2008). To prevent Asian Carp introduction to Lake Michigan, two electric barriers have been implemented in the Chicago Sanitary and Ship Canal.

Five tributaries of the Illinois River were selected for this study: the Kankakee River, Mackinaw River, Spoon River, Sangamon River, and Salt Creek, a tributary of the Sangamon River. The Kankakee River originates in Indiana, where it has been channelized, and retains its natural meander for about 100 km within Illinois, where it is dammed in four places, draining 5617 km^2, supporting 89 species of fish (Page et al. 1992). The Mackinaw River flows for 209 km in Illinois, draining an area of 2942 km^2, most of which is agricultural drainage. It features a highly variable seasonal water load and supports 80 species of fish. The Mackinaw is an unimpounded principle tributary of the Illinois and joins it near Pekin, IL (Page et al. 1992). Another agricultural principle tributary is the Spoon River, 259 km long, draining 4804 km^2, joining the Illinois River adjacent to Havana, IL. This river features high turbidity, one low head dam, a fairly consistent water load, and supports approximately 66 fish species (Page et al. 1992). The Sangamon River is the largest tributary in this study, flowing 396 km and draining 14,035 km^2, much of central Illinois' agricultural drainage. The Sangamon is impounded in multiple locations, features a highly turbid water load, and supports approximately 89 fish species. The Sangamon has been diverted to join the Illinois River in Muscooten Bay near Beardstown, IL, about 14 kilometers south of its natural confluence. Salt Creek is a secondary tributary of the Illinois River and a major tributary of the Sangamon River. Salt Creek flows approximately 180 km, is dammed to form Clinton Lake, and is channelized, characterized by a sandy substrate throughout the river (Page et al. 1992). All tributaries excluding the Kankakee River have records of Asian Carp.

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POPULATION CHARACTERISTICS OF ASIAN CARP IN THE ILLINOIS RIVER AND ITS TRIBUTARIES

ABSTRACT

Invasive species introductions threaten native biodiversity, ecosystem function, and habitat quality worldwide. Two notorious invaders are Silver Carp (*Hypophthalmichthys molitrix)* and Bighead Carp (*H. nobilis*), collectively Asian Carp, threatening invasion of the Great Lakes. Reproduction in lentic systems such as the Great Lakes is unlikely, however, a knowledge gap remains regarding their use of small rivers and tributaries, a large network of which is associated with the Great Lakes. I examined population characteristics of Asian Carp in tributaries of the Illinois River, a highly invaded system artificially connected to Lake Michigan. I compared relative density, size structure, condition, reproductive condition, age structure, mortality, and growth among the Illinois River and five of its tributaries: Salt Creek, the Mackinaw, Kankakee, Sangamon, and Spoon Rivers during 2011-2014. Relatively high abundances of Silver Carp were encountered in the main channel and tributaries, with none sampled in the Kankakee River. Silver carp were found to temporally increase in size and age, but no differences were observed in condition. Mortality and growth were higher in tributary-caught carp compared to the main channel. Reproductive condition of Silver Carp followed temporal hydrography, with peaks in gonad fullness and stage following peaks in discharge. Demographic differences among the main channel and tributaries likely stems from intra-specific competition, nutrient abundance, and differences in hydrology among systems. Understanding population characteristics of Asian Carp in small North American rivers helps bridge the knowledge gap and advance future efforts for the control of these invasives.

INTRODUCTION

More than 500 non-native fish taxa have been introduced to U.S. inland waters since the early 1800s (Nico and Fuller 1999) and the world's largest freshwater resource, the Laurentian Great Lakes, has been no exception, hosting multiple introductions of non-native species, some of which may have caused ecological catastrophe. Introductions such as the Sea Lamprey (*Petromyzon marinus*), Common Carp (*Cyprinus carpio*), and Zebra Mussels (*Dreissena polymorpha*) have been the source of major economic and ecological detriment to the Great Lakes by either competing with, or preying upon, native species (Mills et al. 1994). Asian Carp of the genus *Hypophthalmichthys* are among the most recent and notorious threats to the Great Lakes (Moy et al. 2011). These carp were introduced to North America in the early 1970s for phytoplankton control in aquaculture ponds and wastewater treatment facilities in the Midwestern United States, subsequently escaping into nearby rivers during flooding events (Freeze and Henderson 1982). Both species of Asian Carp are opportunistic planktivores, feeding on a variety of plankton and algae species with potential for competition with native North American planktivores such as Gizzard Shad (*Dorosoma cepedianum*), Bigmouth Buffalo (*Ictiobus cyrinus*), and Paddlefish (*Polyodon spathula*) (Irons et al. 2007; Sampson et al. 2009). Asian Carp spawn prolifically both in their native range and in introduced areas. The potential to release over half a million eggs per spawning event, in some cases multiple times per year (Abdusamadov 1987), makes these fish highly fecund individuals. Peak spawning occurs in the spring and early summer associated with spring flooding events, however, Asian Carp can reproduce multiple times per year, as conditions allow (Kolar et al. 2005; Garcia et al. 2013). Spawning occurs near the surface in flows high enough to keep the semi-buoyant eggs suspended until hatch (Abdusamadov

1987; Pflieger 1997). Prior to spawning, Asian Carp have been shown to engage in upstream migrations to spawning sites, studies have suggested that they can move over 60 km (DeGrandchamp et al. 2008), sometimes to discrete spawning sites (Kolar et al. 2005). Several specific environmental requirements have been shown to characterize Asian Carp spawning success: long stretches of unimpeded flow, water temperatures above 17°C, increased velocities, turbidity, and water levels associated with spring and summer flood events (Abdusamadov 1987). These requirements make Asian Carp unable to reproduce in lentic systems (Kolar et al. 2005; Jennings 1988), and it is unknown if they can spawn successfully in shorter rivers typified by tributaries of major rivers.

The Illinois River has an artificial and direct link between the highly invaded Mississippi River system and Lake Michigan, putting the Great Lakes at the leading edge of the invasion by Asian Carp. These fish have been very successful invading the United States' large river systems, given their high degree of dispersal, resource consumption, and incredible fecundity; however, their potential for reproduction within the Great Lakes is limited given their specific environmental spawning requirements (Kolar et al. 2005). While Asian Carp reproduction within the Great Lakes is unlikely, an invasion means access to the network of tributaries suitable for the establishment of reproducing populations (Kocovsky et al. 2012). Therefore, it becomes important to identify areas that would satisfy the environmental requirements needed by Asian Carp to reproduce within these areas. In Lake Michigan, there are at least seven tributaries with long stretches of unimpounded flowing water, making these areas hydraulically suitable for Asian Carp reproduction (Kolar et al. 2005). It is these tributaries that targeted management should be focused, similar to the efforts for the control of invasive lotic-reproducing fish, such as Sea Lamprey, within the

Great Lakes (Christie and Goddard 2003). Since the establishment of Asian Carp in North America, multiple studies have focused on their invasion and impacts on large rivers such as the Mississippi (Williamson and Garvey 2005), Missouri (Papoulias et al. 2006), and Illinois River (Irons et al. 2011). However, relatively fewer studies focus on Asian Carp in smaller rivers and tributaries of large rivers (Hayer et al. 2014), although these rivers remain suitable for Asian Carp colonization (Kocovsky et al. 2012). The Illinois River is highly studied with regards to Asian Carp (Chick and Pegg 2001; Degrandchamp et al. 2008; Irons et al. 2007; Sampson et al. 2009; Stuck et al. 2015), but no studies focus explicitly on Asian Carp in its tributaries.

Given the specific environmental conditions required by Asian Carp for spawning and recruitment, it is important to identify reproductive abilities in small rivers. This study becomes essential to identify management potential for areas of special concern, notably the Laurentian Great Lakes, at the forefront of the Asian Carp invasion. Because of the paucity of information regarding Asian Carp in tributary systems, I sought to examine population characteristics of Asian Carp in the Illinois River and its tributaries. By comparing Asian Carp demographics between the main channel and associated tributaries, I tested the following null hypotheses: 1) there is no difference in Asian Carp population characteristics among the Illinois River and its tributaries, 2) there is no difference in population characteristics among tributaries, and 3) there are no differences in population characteristics among the years sampled. This study lays the groundwork for future work considering Asian Carp population characteristics and reproductive requirements in tributaries of large rivers and the Great Lakes.

METHODS

Study Area

Four tributaries of the Illinois River were sampled annually (2013-2014) including the Spoon, Sangamon, Mackinaw, and Kankakee Rivers, in the second year Salt Creek, a tributary of the Sangamon River, replaced the Kankakee River as no Asian Carp were sampled there in 2013 (Figure 1.1). Each river was sampled at two sites, one at a downstream location near the mouth and one at an upstream location, chosen to maximize distance between sites while considering accessibility and impoundments. Asian Carp from the Illinois River main channel were collected from the LaGrange reach annually (2011-2014) by the Illinois Natural History Survey.

Sampling

Asian Carp were sampled using boat pulsed-DC electrofishing (60Hz, 25% duty cycle) during April-October at fixed sites, both upstream in the tributaries and downstream near the confluence with the Illinois River (Figure 1.1). A single fifteen minute transect (pedal time) using a modified Long Term Resource Monitoring Program (LTRMP) protocol (Gutreuter et al. 1995) was employed to capture fish; power output was maximized according to local specific conductivity measurements with a Wisconsin ETS electrofishing box. A total of 19 hours of pulsed-DC electrofishing effort was conducted during 2013 (5.75 hrs) and 2014 (13.25 hrs) in the tributaries, no effort data was available for the Illinois River. Due to channel size differences between upstream and downstream sites and the size of watercraft necessary to accommodate these differences, one electrofishing dipper was used at upstream sites and two dippers at downstream sites, standardized to person-hour. Asian Carp were measured to the nearest millimeter (TL), weighed to the nearest gram, sexed, gonadal development stage defined via observation (Li and Mathias 1994). Gonads were weighed to the nearest gram, and a postcleithrum bone was obtained for aging estimation.

Aging

Postcleithra were returned to the lab to be dried, cleaned, and sectioned sequentially in the transverse plane using an ISOMET® low speed saw (Johal et al. 2000). Three 0.5-1.0 mm sections were mounted to glass slides for imaging using a camera-equipped stereoscopic microscope with a contrast background. Images were subsequently aged by counting annuli by two independent readers; disagreements were subsequently reconciled between the readers. Additionally, a subset of the first ten Asian Carp encountered had heads removed in the field for otolith recovery in the laboratory. Extracted sagittal otoliths were cleaned in 95% ethanol, dried, and stored in microcentrifuge tubes and then prepared for aging estimation according to Secor et. al (1991). Otolith age estimates were then subject to the same age estimation procedure as the postcleithra, and compared to determine hard structure age estimation precision.

Data Analysis

Relative abundance, size structure, and condition

Relative abundance was represented as catch-per-unit effort (CPUE), calculated as fish per person-hour to account for the difference in dip netters. Data for CPUE were $\log_{10}(x+1)$ transformed to compensate for heteroscedascity (Hubert and Fabrizio 2007) and subsequently compared among rivers and years using analysis of variance (ANOVA) with a Tukey-Kramer Honestly significant difference multiple comparison post hoc test. Differences in size structure were compared using a nonparametric Kolmogorov-Smirnov two-sample test for differences in distributions between rivers and years (Neumann and Allen 2007).

Addtionally, Bonferroni-corrected *p* values accounted for experiment-wise error of multiple comparisons for size structure (Gotelli and Ellison 2004). I calculated condition using relative weight equation (Anderson and Neumann 1996), calculated as:

$$W_r = (W/W_s) * 100,$$

where W_r is the condition factor, W is the total weight of an individual, and W_s is a length-specific standard weight predicted by a weight-length regression ($log_{10}(W_s) = a' + b(Log_{10}L)$) constructed for the specific species (Anderson and Neumann 1996). The constants a' (= -5.15765) is the intercept value and b (= 3.06842) is the slope of the *log$_{10}$ (weight)-log$_{10}$ (length)* regression equation for Silver Carp (Lamer, *unpublished data*). Condition factor of fishes among sample rivers was compared using Kruskal-Wallis nonparametric test and pairwise Wilcoxon Rank-Sum nonparametric test with a Bonferroni correction for multiple comparisons.

Sex structure and reproductive condition

I used analysis of proportions to determine if sex ratio of Asian Carp differed from 1:1. Differences in size structure between sexes were measured using a nonparametric Kolmogorov-Smirnov two-sample test for differences in distributions among rivers and years (Neumann and Allen 2007). Gonadosomatic index (GSI = gonad wt/somatic wt*100) was employed as a measure of reproductive potential. I used ANOVA with a Tukey-Kramer Honestly significant difference multiple comparison test to compare mean GSI values for each sex among sample rivers and dates.

Structure comparison, precision, and age structure

Average percent error (APE) and coefficient of variation (CV) was calculated between the two readers for each hard structure and between hard structures to determine precision, shown as:

$$APE_j = 100 * \frac{1}{R}\sum_{i=1}^{R}\frac{|x_{ij} - x_j|}{x},$$

$$CV_j = 100 * \frac{\sqrt{\sum_{i=1}^{R}\frac{\left(x_{ij} - x_j\right)^2}{R-1}}}{x_j},$$

where R is the number of readers, x_{ij} is the estimated age by reader *i*, and x_j is the mean age of all readers (Beamish and Fournier 1981, Chang 1982). I then compared the slope of each age bias plot against a slope of one using a t-test to determine age bias between structures and/or readers (Campana et al. 1995). Age structure was compared between sample rivers with a nonparametric Wilcoxon rank-sum test with a Bonferroni correction for multiple comparisons.

Growth and mortality

Length at age of Asian Carp was back-calculated using digitized images of cleithra sections. I used ImageJ® (Rasband 2015) to measure the radius length of the cross section and the length from the center to each annulus. Using these data, I back-calculated length at age using the Dahl-Lea equation:

$$L_i = \frac{S_i}{S_c} * L_c,$$

where L_i is the back-calculated length at age i, L_c is the length at capture, S_c is the radius of the cross section at capture, and S_i is the radius of the cross section at annulus i (Dahl 1907; Lea 1910; LeCren 1947).

Using these data, I modeled growth with a von Bertalanffy growth curve represented as:

$$l_t = L_\infty(1 - e^{-K(t-t_0)}),$$

where l_t is length at time t, L_∞ is the asymptotic length, K is a growth coefficient, and t_0 is a time coefficient where length would theoretically be 0 mm (Isley and Grabowski 2007) using Fisheries Analysis and Modeling Simulator 1.0 (FAMS, Slipke and Maceina 2010). Using these estimates of growth, I then compared growth differences between sampled rivers using an analysis of the residual sum of squares to account for nonlinear functions (ARSS, Chen et al. 1992).

Mortality was estimated using a linearized catch-curve regression, formed by aging all fish in the sample, and log-transforming catches at each age:

$$log_e N_t = log_e N_0 - Z(t),$$

where the absolute value of the slope of the regression line, Z, represents instantaneous mortality, N_t is frequency at time t, N_0 is frequency at time 0, and t represents time step (Miranda and Bettoli 2007). Total annual mortality (A) was then calculated from Z using the equation $A = 1 - e^{-Z}$.

Discharge

To determine the effects of discharge on reproductive development and condition of Asian Carp, daily discharge data was obtained from the United States Geologic Survey for the most downstream gauge for each sampled tributary in 2014. The Salt Creek gauge was Greenview, IL (USGS station number: 5582000); Seville, IL Spoon River (USGS station number: 5570000) for the Spoon River; Oakford, IL (USGS station number: 5583000) for the Sangamon River; and Green Valley, IL (USGS station number: 5568000) for the Mackinaw River. Pairwise correlations were used to determine relatedness between hydrographs for each sample river in 2014.

All statistics were performed using Program R© (v.3.1.1, R Core Team 2014) software and a significance level was set at $\alpha = 0.05$ unless otherwise stated.

RESULTS

A total of 1298 Asian Carp was collected from the Mackinaw, Sangamon, and Spoon Rivers and Salt Creek during 2013-2014, and a total of 447 Asian Carp was collected from the Illinois River during 2011-2014 using pulsed-DC electrofishing. No Asian Carp were sampled in the Kankakee River at any time. Of the 1745 total Asian Carp sampled, only 15 Bighead Carp were sampled, accounting for < 1% of the sample. Because of the low sample size, Bighead Carp were excluded from further analysis.

Relative abundance

Although mean CPUE was highest in the Mackinaw River and lowest in the Spoon River, there were no significant differences in CPUE among the sample rivers ($F_{3,36} =$

0.6599, $p = 0.58$, Table 1.1), however, mean CPUE was significantly higher in 2013 than in 2014 ($F_{1,38} = 4.398$, $p = 0.04$, Table 1.1).

Size structure

In the Illinois River main channel, Asian Carp sampled were between 404-888 mm, with a mean of 530.85±2.56 (mean ± SE) mm overall. In the tributaries sampled, Asian Carp were between 445-872 mm, with a mean of 562.45±1.54 mm overall. Although pairwise comparisons indicated no significant difference in length frequency distributions in the Illinois River between 2011-2012 and 2013-2014 (*KS*, $p > 0.005$, Table 1.2), all other comparisons among years differed ($p < 0.005$), with fish sampled displaying consecutively larger mean lengths each year (Figure 1.2). During 2013-2014, there were significant differences in length frequency between the Mackinaw, Sangamon, and Spoon Rivers ($p < 0.005$; Table 1.3, Figure 1.3). Pairwise comparisons between the Mackinaw and Spoon Rivers indicated no significant difference ($p > 0.005$), all remaining pairwise comparisons among tributaries were significantly different ($p < 0.005$, Table 1.3, Figure 1.3).

Condition

Mean relative weight of Asian Carp ranged from 92 to 100 in all sample rivers. Asian Carp relative weight in the tributaries was not significantly different than in the Illinois River ($\chi^2_{1691} = 1701.14$, $p = 0.43$), nor was relative weight significantly different among years ($\chi^2_{1691} = 1706.90$, $p = 0.39$) (Figure 1.4). However, pairwise Wilcoxon Rank-Sum results revealed significant differences among all interactions among sampled rivers ($p > 0.0005$, Table 1.4), except between the Illinois and Spoon Rivers; Mackinaw and Spoon Rivers; and Salt Creek and Sangamon River.

Sex structure and reproductive condition

A total of 1729 Silver Carp was sexed from each sampled river. Overall, a significant male bias was observed, however, no significant sexual bias occurred in the Sangamon River, Illinois River, or Salt Creek (Table 1.5). Length frequency distributions did not differ between the sexes ($KS = 0.06$, $p = 0.06$, Figure 1.5). Mean gonadosomatic index values reflected increased gonadal development stage in both male and female Asian Carp, with the largest values observed in stage 5 individuals and smallest values in stage 2 individuals in each tributary. Mean GSI of stages 4 and 5 females were significantly higher than stages 2, 3, and 6 ($F_{4,285} = 126$, $p < 0.001$, Figure 1.6). Similar to female Asian Carp, mean GSI values for males were significantly lower in stages 2, 3, and 6 individuals than stages 4 and 5 ($F_{4,447} = 22.39$, $p < 0.001$, Figure 1.7). Additionally, higher mean GSI values were observed during the spring (May-June) compared to summer (July-August) and fall (September-October) in both sexes (female: $F_{2,287} = 60.35$, $p < 0.001$, *Figure 1.6*, male: $F_{2,449} = 103.6$, $p < 0.001$, Figure 1.7). Peaks in GSI values for both sexes coincided with pre-spawn gonad stages (i.e. stage 4), and spent stage 6 individuals increased following decreases in mean GSI values. Predominantly overwintering and sexually immature stages were observed in early spring during April-May (Figure 1.6 and Figure 1.7).

Structure comparison

Exact agreement between otoliths and cleithra for age estimation was 32%, with 70% ± 1 yr, and 89% ± 2 yr. Precision between otoliths and cleithra showed relatively low APE (10.79) and CV (15.26) values. Illustrated by an age-bias plot, fewer age classes of cleithra were observed compared to otoliths (slope $\neq 1$, $t_{1,90} = -10.81$, $p < 0.001$; Figure 1.8).

Aging precision

Precision between readers for age using cleithra, exhibited exact agreement of 50% and ± 1 year agreement of 93%. Between-reader APE was 6.10 and the CV was 8.63. The age bias plot between readers indicates a significant reader bias (slope $\neq 1$, $t_{1,1294} = -15.56$, $p < 0.001$, Figure 1.9).

Age frequency

Pairwise Wilcoxon tests indicated no difference between age frequency between 2011 and 2012 ($W = 8060$, $p = 0.41$), all other pairwise comparisons were different ($p < 0.003$, Table 1.6, Figure 1.10). Mean age within the tributaries sampled ranged between 4.1-5.2 years during 2013-2014. During 2013, the Spoon River did not differ in age frequency from the Sangamon River ($W = 17866$, $p = 0.002$), however; during 2014, the Mackinaw River differed from Salt Creek and Sangamon River, whereas Salt Creek also differed from the Sangamon River ($p < 0.0005$, Table 1.6). Each sampled tributary displayed a significant increase in mean age between years sampled (2013 < 2014, $F_{10,1690} = 114.2$, $p < 0.0001$, Figure 1.11). Additionally, each tributary sampled differed in age from the Illinois River main channel ($p < 0.003$, Table 1.6).

Growth and mortality

Of the sampled tributaries, Asian Carp in Salt Creek were smaller at a given age compared to the Sangamon, Spoon, and Mackinaw Rivers using back-calculated mean lengths (Figure 1.12). Growth parameters of the Illinois River were calculated using length at capture, whereas back-calculated lengths were used for tributary fish (Figure 1.12). Multiple pairwise comparisons of the von Bertalanffy growth model suggest there were no significant differences in growth between rivers sampled ($p > 0.005$, Table 1.7, Figure 1.13), however there were trends in the data. Asian Carp in Salt Creek grew the fastest, opposed to Asian Carp in the Illinois River, which grew the slowest (Figure 1.13). Intermediary of these were the Sangamon and Spoon Rivers, which had comparable growth; and the Mackinaw River, similar to the Illinois River, growing relatively slower (Figure 1.13). Mortality was estimated for Asian Carp from the Illinois River using year classes 3-7 and year classes 5-7 within the tributaries. Lower age classes were determined not fully recruited to the gear. Estimated annual mortality ranged from 60-82% for Asian Carp during 2011-2014. The Illinois River displayed the lowest total annual mortality (60%), whereas the highest estimated total annual mortality occurred in the Mackinaw River (82%, Figure 15). Intermediate values ranged from 71-78% in the Sangamon River (71%), Spoon River (77%), and Salt Creek (78%, Figure 1.14).

Discharge

Mean discharge between rivers varied, with the Sangamon River showing irregular flow characteristics, with the remaining rivers exhibiting somewhat more typical spring and fall flooding events (Figure 1.15). In 2014, the Sangamon River had the highest discharge (4003.89±18.36 m^3/s) whereas the Mackinaw River (936.20±6.04 m^3/s) had the lowest with the Spoon River (1076.83±9.30 m^3/s) and Salt Creek (1276.61±8.87 m^3/s) intermediate. However, positive pairwise correlations suggest that flow patterns were similar among sampled tributaries (Table 1.8).

DISCUSSION

Silver Carp population characteristics differed between the Illinois River and associated tributaries. Asian Carp have been extensively studied in large rivers such as the Illinois River (Irons et al. 2011; Sass et al. 2010; Stuck et al. 2015; Lamer et al. 2010), and these results will help define better management strategies to deal with this invasive. Compared to the impounded Illinois River, the tributaries are either unimpounded (Mackinaw River) or have long stretches of free flowing channel. Coupled with highly variable hydraulic events, these characteristics may aid in the upstream movement of Asian Carps into these systems, especially during periods of increased discharge. Bighead Carp in the Illinois River increased movement during increased discharge (Peters et al. 2006) and both species of Asian Carp have also been shown to increase movement during elevated river stage in the Illinois River (DeGrandchamp et al. 2008).

Catch per unit effort estimates in the tributaries fall within or exceed ranges observed in other large rivers including the Illinois River: 40-70 fish per DC electrofishing hour (Stuck et al. 2015); Mississippi River: 5-20 Silver Carp per DC electrofishing hour, Ohio River: 3-41 Silver Carp per DC electrofishing hour, Wabash River: 2-18 Silver Carp per DC electrofishing hour (Tyszko et al. 2012); and South Dakota tributaries of the Upper Missouri River: 3-76 Silver Carp per DC electrofishing hour (Hayer et al. 2014). Low numbers of Bighead Carp occurred because electrofishing is more effective for silver carp and Bighead Carp tend to be pioneer species, subsequently replaced by the later-invading Silver Carp (Garvey et al. 2006; Irons et al. 2011). Asian Carp have been increasing exponentially within the Illinois River (Chick and Pegg 2001), and are likely to continue for the foreseeable future (Sass et al. 2010), probably dispersing into associated tributaries to complete portions of their life cycle.

Relatively low agreement was observed between sagittal otoliths and postcleithra for age estimation. Otoliths are commonly preferred as aging hard structures for multiple species of freshwater fishes, and are regarded as accurate and precise structures (Isley and Grabowski

2007). However, the otoliths of Asian Carp have been found to be hazy, fragile, and unusable as age structures (Johal et al. 2000). In spite of this, the asteriscii (Hayer et al. 2014, Siebert and Phelps 2013), lapilli (Siebert and Phelps 2013), and sagittal otoliths (Nuevo et al. 2004; Schrank et al. 2001) have been evaluated as age structures. Additional hard structures used for aging Asian Carp include vertebrae, scales, fin rays, and postcleithra (Nuevo et al. 2004; Johal et al. 2000; Garvey et al. 2006; Kamilov 2014; Siebert and Phelps 2013). I chose to proceed using postcleithra, given the ease of preparation, clear annuli, and support for back-calculation of growth (Johal et al. 2000; Johal et al. 2001; Stuck et al. 2015). I found sagittae to be time consuming and difficult structures for age estimation given their fragile and hazy characteristics.

Population characteristics of Asian Carp in the Illinois River followed those of previous studies in regards to size, age, growth, and mortality (Schrank and Guy 2002; Irons et al. 2011; Garvey et al. 2006; Stuck et al. 2015). I observed differences in total length and age frequency distributions between the tributaries and the Illinois River. Strong year classes dominated the populations in the Illinois River and the tributaries (2009-2010 year classes) as have been observed in previous studies of Asian Carp (Hayer et al. 2014; Irons et al. 2011; Garvey et al. 2006).

Mean back-calculated total lengths of Silver Carp in were smaller than many observed in the literature. Using postcleithra, lengths at age of Silver Carp in the Gobindsagar Reservoir, Himachal Pradesh, India were nearly 100 mm larger than our estimates for each age class (Johal et al. 2001) and in the Tudakul Reservoir, Uzbekistan from lateral scales (Kamilov 2014). Even larger lengths at age and growth rate were observed using back-calculated lengths from pectoral fin rays from Silver Carp in the Middle

Mississippi River (Williamson and Garvey 2005). Growth rates of Illinois River Silver Carp in my study were similar to those observed in other studies of the Illinois River (Stuck et al. 2015) and South Dakota tributaries of the Missouri River (Hayer et al. 2014). Silver Carp growth in Illinois River tributaries (Sangamon River, Spoon River, and Salt Creek) are comparable to those in the Middle Mississippi River (Williamson and Garvey 2005), however, the growth in the Mackinaw River is closer to that in the Illinois River and Missouri River tributaries (Hayer et al. 2014; Stuck et al. 2015). Increased growth rate in Illinois River tributaries could be attributable to increased resource availability and decreased inter- and intraspecific competition.

Our mortality estimate for the Illinois River was similar to those reported in the Illinois River (60%; Stuck et al. 2015) as well as in the Upper Mississippi River during 2004 (60%, Garvey et al. 2006). Annual mortality estimates within tributaries were substantially higher, suggesting increased mortality that may be due to dewatering events. Silver Carp in the tributaries were determined to be fully recruited to the gear at age 5, compared to ages 2-3 for the Illinois River (this study, Stuck et al. 2015). The high concentration of older age classes likely contribute to the increased mortality rates, and indicate utilization of tributaries by sexually mature adult fish, as well as an indication of the strong 5 year age class.

Significant increases in gonadosomatic indices observed during 2014 in this study coincided with elevated discharge events. Similar temporal changes were observed in the Lower Illinois River (DeGrandchamp et al. 2007) and the Missouri River (Papoulias et al. 2006), and these results are consistent with these fish in native Asia, where reproductive activity is associated with river stage (Abdusamadov 1987). GSI values in the tributaries peaked in June-July, with elevated hydrograph activity occurring simultaneously. Significant decreases and subsequent low GSI values were observed following these periods of elevated discharge, as well as the presence of spent females. Overwintering gonadal stages of Asian Carp (stage 3, both sexes; Li and Matthias 1994) were observed early in the sample during April-May, increasing to pre-spawn and spawning stages during the summer months. Female GSI values plummeted in the Spoon River during July, whereas larger decreases were observed in the Sangamon drainage during August, corresponding to high proportions of spent individuals. Similar patterns were observed for males, although more gradual declines ensued during the summer. I also observed spawning males throughout the summer, however, spent males were uncommon at any time, likely because of rapid regeneration times. Regardless, significant temporal declines were documented in male GSI values. Asian Carp spawning in Illinois River tributaries appears dependent on environmental factors such as elevated discharge associated with flooding events. Asian Carp eggs are semi-buoyant, requiring flows > 0.7 ms^{-1} to remain suspended, thus explaining the relationship between elevated discharge and gonadal fluctuations (Abdusamadov 1987).

Asian Carp inhabiting Illinois River tributaries do exhibit demographic differences from populations in the Illinois River and other large, invaded U.S. rivers. Our estimates of age, mortality, and growth for the Illinois River closely match previous studies, but our estimates within the tributaries exhibit differences. Asian Carp in the Mackinaw River, Salt Creek, Sangamon River, and Spoon River had higher growth rates, increased mortality, and higher abundances than populations of Asian Carp in other large rivers (Williamson and Garvey 2005; Hayer et al. 2014; Irons et al. 2011). Reproductive condition of tributary-residing Asian Carp indicates fluctuations consistent with spawning activity (DeGrandchamp 2007), suggesting each of these rivers supports hydrologic characteristics sufficient for Asian

Carp reproduction. These results lay the groundwork for future investigations regarding hydrologic suitability for Asian Carp reproduction in small rivers in the United States, with applicability to Great Lakes establishment.

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Table 1.1. Mean electrofishing CPUE (fish/person hour) for Asian Carp sampled using pulsed-DC electrofishing in the Kankakee, Mackinaw, Spoon, and Sangamon Rivers and Salt Creek during 2013-2014. N = number of sample sites, SE = standard error of the mean.

		Year					
		2013		2014		*Combined*	
River	N	Mean	SE	Mean	SE	*Mean*	*SE*
Kankakee	2	0	0			*0*	*0*
Sangamon	2	82.00	31.03	92.86	22.78	*88.33*	*17.71*
Spoon	2	107.00	15.61	51.14	13.60	*71.45*	*13.01*
Mackinaw	2	183.33	44.67	74.00	18.66	*106.8*	*23.97*
Salt Creek	2			74.57	25.39	*74.57*	*25.39*
Mean		*86.75*	*19.79*	*73.14*	*10.11*	*75.65*	*9.52*

Table 1.2. Kolmogorov-Smirnov test statistics for pairwise comparisons of pooled length-frequency distributions between the Illinois River, Mackinaw River, Salt Creek, Sangamon River, and Spoon River during 2011-2014. D = K-S test statistic, bolded selections indicate significance at $p < 0.005$.

	Illinois	Mackinaw	Salt Creek	Sangamon	Spoon
Illinois		**$D = 0.24$**	**$D = 0.34$**	**$D = 0.48$**	**$D = 0.29$**
Mackinaw			**$D = 0.16$**	**$D = 0.31$**	$D = 0.12$
Salt Creek				**$D = 0.22$**	**$D = 0.21$**
Sangamon					**$D = 0.40$**
Spoon					

Table 1.3. Kolmogorov-Smirnov test statistics for pairwise comparisons of length-frequency distributions between the Illinois River (IL), Mackinaw River (M), Salt Creek (SCR), Sangamon River (SG), and Spoon River (SP) during 2011 (11), 2012 (12), 2013 (13), and 2014 (14). D = K-S test statistic, bolded selections indicate significance at $p < 0.0005$.

	IL11	IL12	IL13	IL14	M13	M14	SG13	SG14	SP13	SP14	SCR14
IL11		*D* = 0.12	***D* = 0.53**	***D* = 0.66**	***D* = 0.27**	***D* = 0.60**	***D* = 0.63**	***D* = 0.77**	***D* = 0.43**	***D* = 0.67**	***D* = 0.56**
IL12			***D* = 0.54**	***D* = 0.68**	***D* = 0.29**	***D* = 0.62**	***D* = 0.65**	***D* = 0.76**	***D* = 0.50**	***D* = 0.69**	***D* = 0.58**
IL13				*D* = 0.22	***D* = 0.29**	*D* = 0.22	*D* = 0.21	***D* = 0.54**	***D* = 0.28**	*D* = 0.25	*D* = 0.22
IL14					***D* = 0.43**	*D* = 0.15	*D* = 0.08	***D* = 0.41**	***D* = 0.45**	*D* = 0.14	*D* = 0.12
M13						***D* = 0.37**	***D* = 0.38**	***D* = 0.55**	***D* = 0.23**	***D* = 0.43**	***D* = 0.33**
M14							*D* = 0.12	***D* = 0.35**	***D* = 0.35**	*D* = 0.16	*D* = 0.10
SG13								***D* = 0.37**	***D* = 0.43**	*D* = 0.10	*D* = 0.09
SG14									***D* = 0.69**	***D* = 0.30**	***D* = 0.33**
SP13										***D* = 0.46**	***D* = 0.40**
SP14											*D* = 0.13
SCR14											

Table 1.4. Wilcoxon Rank-Sum test statistics for pairwise comparisons of pooled relative weight distributions between the Illinois River, Mackinaw River, Salt Creek, Sangamon River, and Spoon River during 2011-2014. W = Wilcoxon test statistic, bolded selections indicate significance at $p < 0.005$.

	Illinois	Mackinaw	Salt Creek	Sangamon	Spoon
Illinois		**W = 66584**	**W = 18633**	**W = 37101**	W = 81329
Mackinaw			**W = 44315**	**W = 91050**	W = 60113
Salt Creek				W = 32300	**W = 49748**
Sangamon					**W = 102519**
Spoon					

Table 1.5. Sexual demographics of Asian Carp in the Illinois River, Mackinaw River, Salt Creek, Sangamon River, and Spoon River during 2011-2014. X^2 = chi-squared test statistic, p value = probability of significant differences. Bolded values indicate significance at $p < 0.05$.

	M Count	F Count	X^2	*p value*
Illinois	242	203	3.42	0.06
Mackinaw	202	163	4.17	**0.04**
Salt Creek	95	82	0.95	0.33
Sangamon	199	163	3.58	0.06
Spoon	246	134	33.01	**< 0.0001**
Mean	*984*	*745*	*30.04*	***< 0.0001***

Table 1.6. Wilcoxon Rank-Sum test statistics for pairwise comparisons of age-frequency distributions between the Illinois River (IL), Mackinaw River (M), Salt Creek (SCR), Sangamon River (SG), and Spoon River (SP) during 2011 (11), 2012 (12), 2013 (13), and 2014 (14). W = wilcoxon test statistic, bolded selections indicate significance at $p < 0.0005$.

	IL11	IL12	IL13	IL14	M13	M14	SG13	SG14	SP13	SP14	SCR14
IL11		W = 8060	**W = 2926**	**W = 711**	**W = 4135**	**W = 2319**	**W = 1890**	**W = 1720**	**W = 3607**	**W = 1098**	**W = 1677**
IL12			**W = 3403**	**W = 1054**	**W = 4677**	**W = 3140**	**W = 2557**	**W = 2451**	**W = 4695**	**W = 1620**	**W = 2391**
IL13				**W = 1484**	W = 6341	**W = 4214**	**W = 3652**	**W = 3201**	**W = 6881**	**W = 2012**	**W = 3362**
IL14					**W = 8743**	W = 9026	W = 7088	W = 7384	**W = 11643**	W = 5453	W = 7083
M13						**W = 6947**	**W = 5879**	**W = 5343**	**W = 10682**	**W = 3547**	**W = 5505**
M14							W = 18805	W = 19583	**W = 30787**	**W = 14524**	W = 18784
SG13								**W = 10277**	W = 17866	**W = 7376**	W = 10220
SG14									**W = 33209**	W = 17479	W = 21426
SP13										**W = 8116**	**W = 11890**
SP14											**W = 16849**
SCR14											

Table 1.7. Pairwise analysis of residual sums of squares (ARSS) test statistics between von Bertalanffy growth parameters for the Illinois River, Mackinaw River, Salt Creek, Sangamon River, and Spoon River. Bolded entries indicate significance at $p < 0.05$.

	Illinois	Mackinaw	Salt Creek	Sangamon	Spoon
Illinois		$F_{7,10} = 1.74$	$F_{7,10} = 0.83$	$F_{7,10} = 1.41$	$F_{8,11} = 1.69$
Mackinaw			$F_{8,11} = 2.15$	$F_{8,11} = 2.09$	$F_{9,12} = 1.82$
Salt Creek				$F_{8,11} = 2.00$	$F_{9,12} = 0.06$
Sangamon					$F_{9,12} = 1.65$
Spoon					

Table 1.8. Pairwise discharge Pearson correlation statistics between the Mackinaw River, Salt Creek, Sangamon River, and Spoon River. Bolded entries indicate significance at $p < 0.05$.

	Mackinaw	Salt Creek	Sangamon	Spoon
Mackinaw		$r_{20029} = \mathbf{0.44}$	$r_{20029} = \mathbf{0.38}$	$r_{20029} = \mathbf{0.45}$
Salt Creek			$r_{20251} = \mathbf{0.84}$	$r_{20249} = \mathbf{0.57}$
Sangamon				$r_{20249} = \mathbf{0.42}$
Spoon				

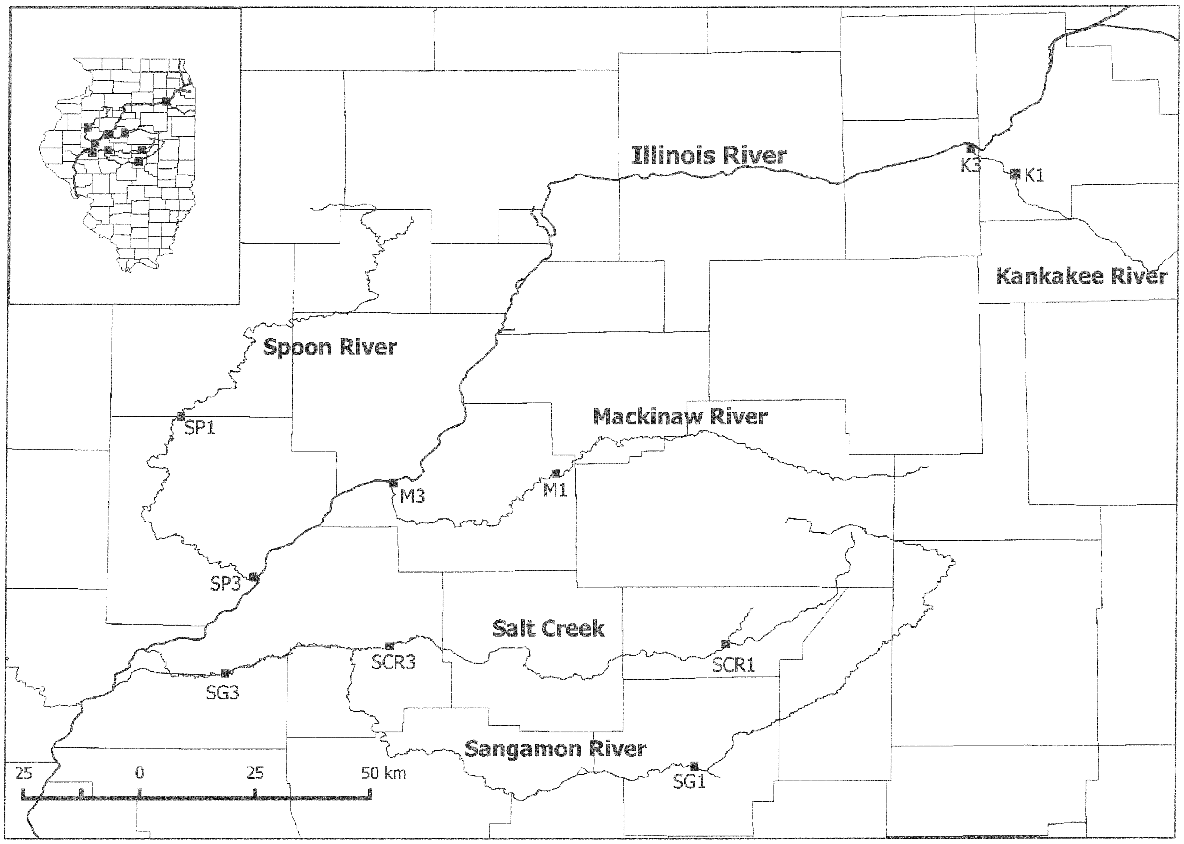

Figure 1.1. Map of five tributaries within the Illinois River system. Sites were upstream (1) and downstream (3) near the Illinois River.

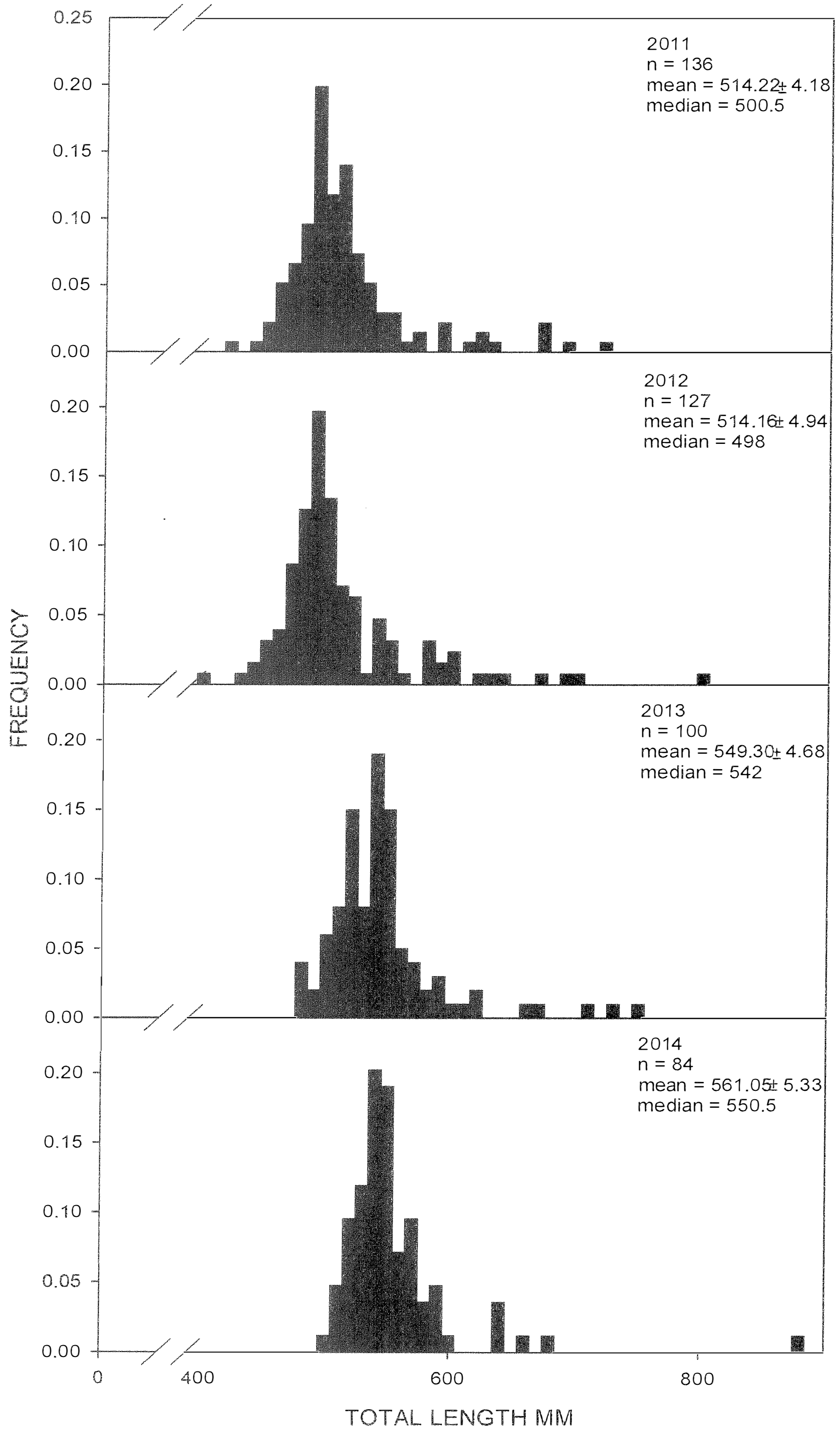

Figure 1.2. Length frequency distributions (total length, in millimeters) of Asian Carp sampled by pulsed-DC electrofishing in the Illinois River during 2011-2014.

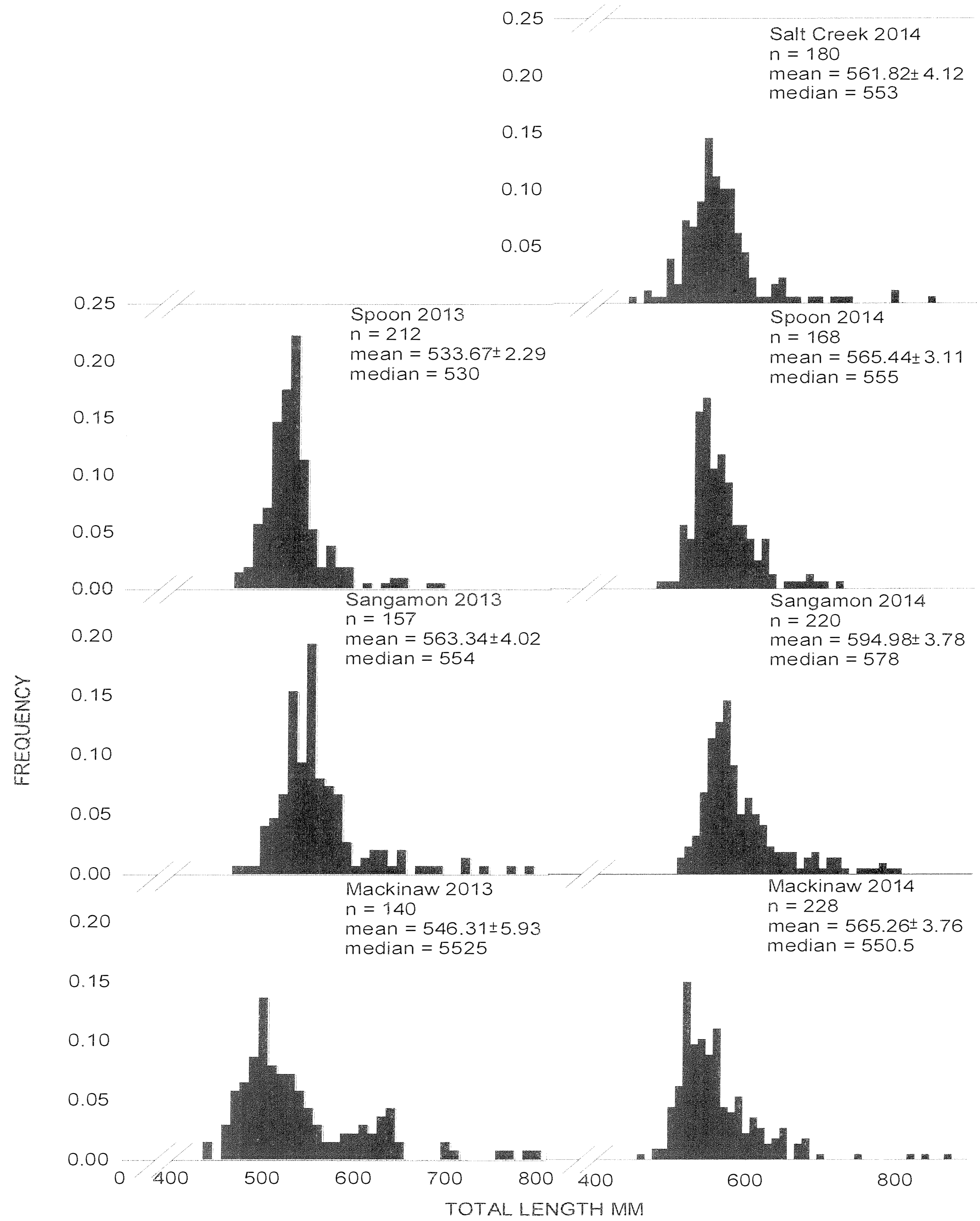

Figure 1.3. Length frequency distributions (total length, in millimeters) of Asian Carp sampled by pulsed-DC electrofishing in the Spoon, Sangamon, and Mackinaw Rivers and Salt Creek during 2013-2014.

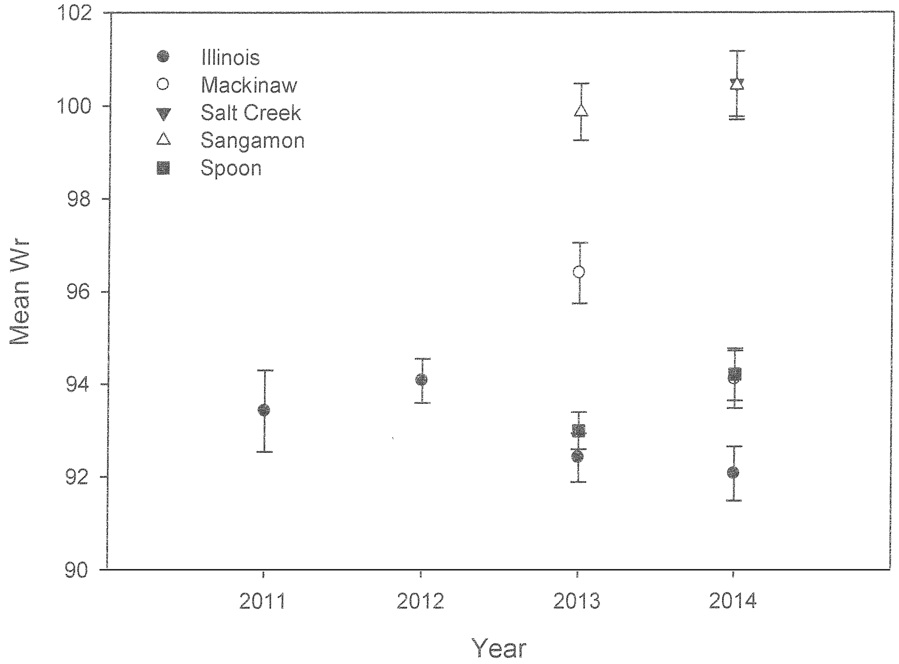

Figure 1.4. Mean relative weight ± SE (Wr) by year for Asian Carp collected from the Illinois River (filled circles; 2011-2014) and the Mackinaw (open circles), Sangamon (open triangles), Spoon Rivers (filled squares), and Salt Creek (filled triangles) (2013-2014).

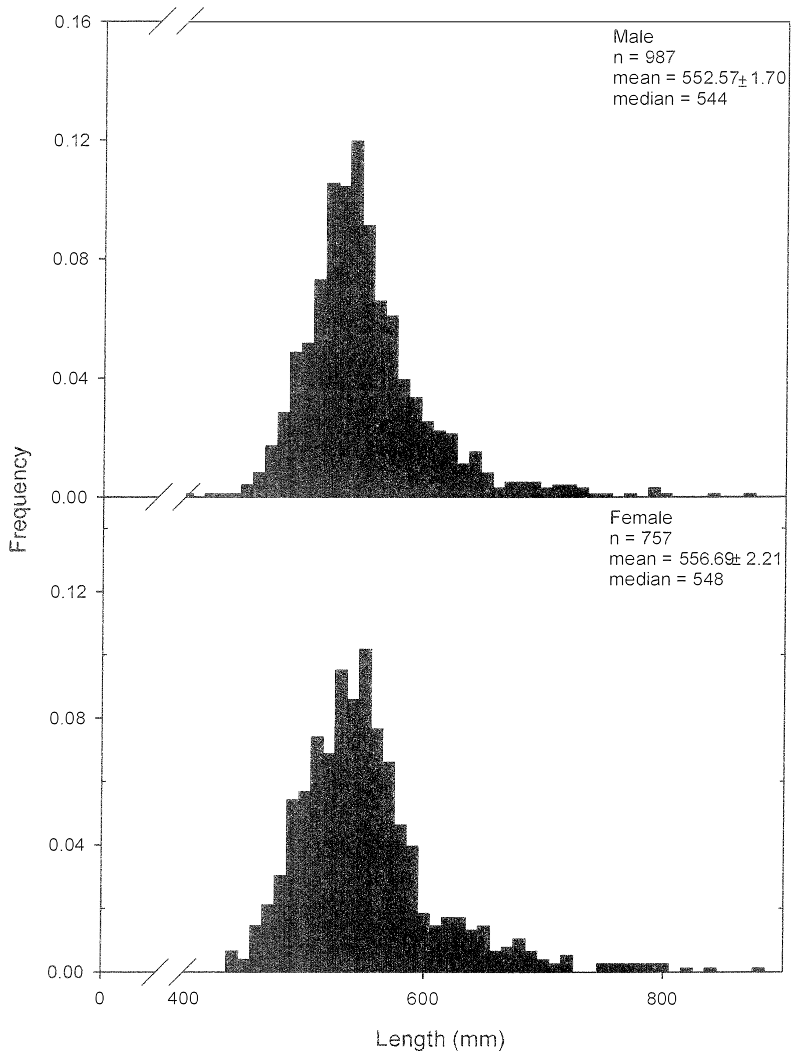

Figure 1.5. Length frequency distributions for female and male Asian Carp sampled from the Illinois River and its tributaries during 2011-2014.

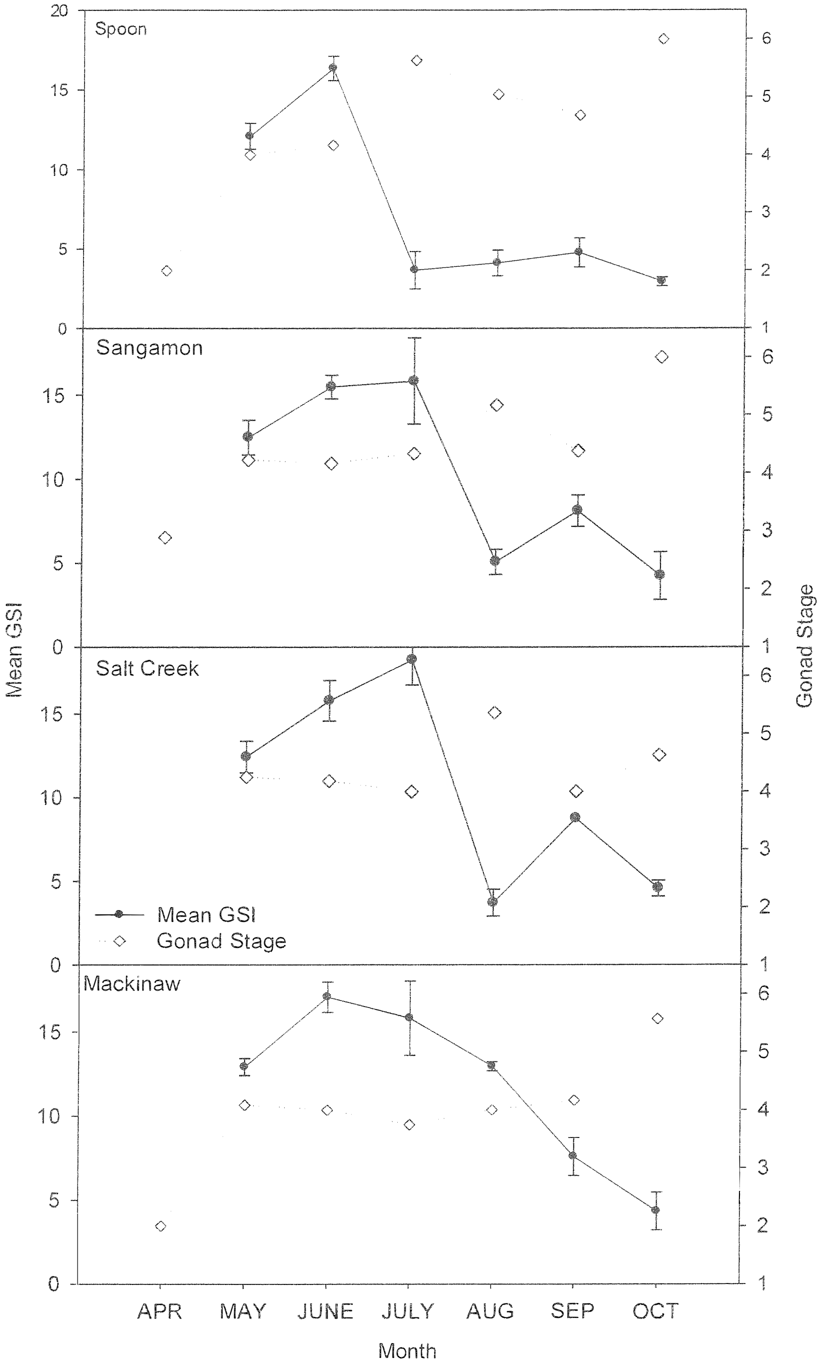

Figure 1.6. Mean GSI values (solid line, filled circles) and reproductive stage (dotted line, white diamonds) of female Asian Carp in the Spoon River, Sangamon River, Salt Creek, and Mackinaw River. Error bars represent the standard error of the mean.

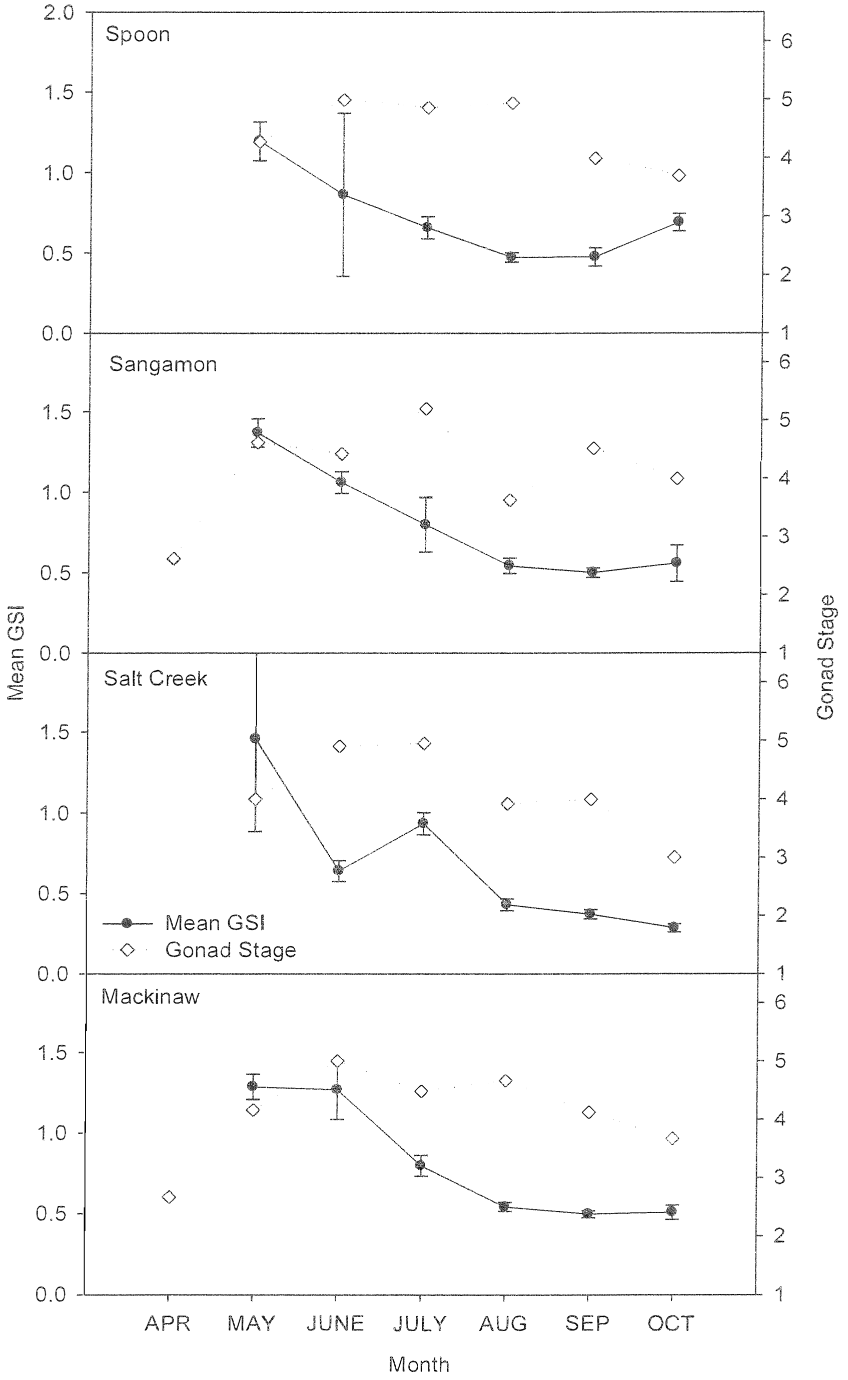

Figure 1.7. Mean GSI values (solid line, filled circles) and reproductive stage (dotted line, white diamonds) of male Asian Carp in the Spoon River, Sangamon River, Salt Creek, and Mackinaw River. Error bars represent the standard error of the mean.

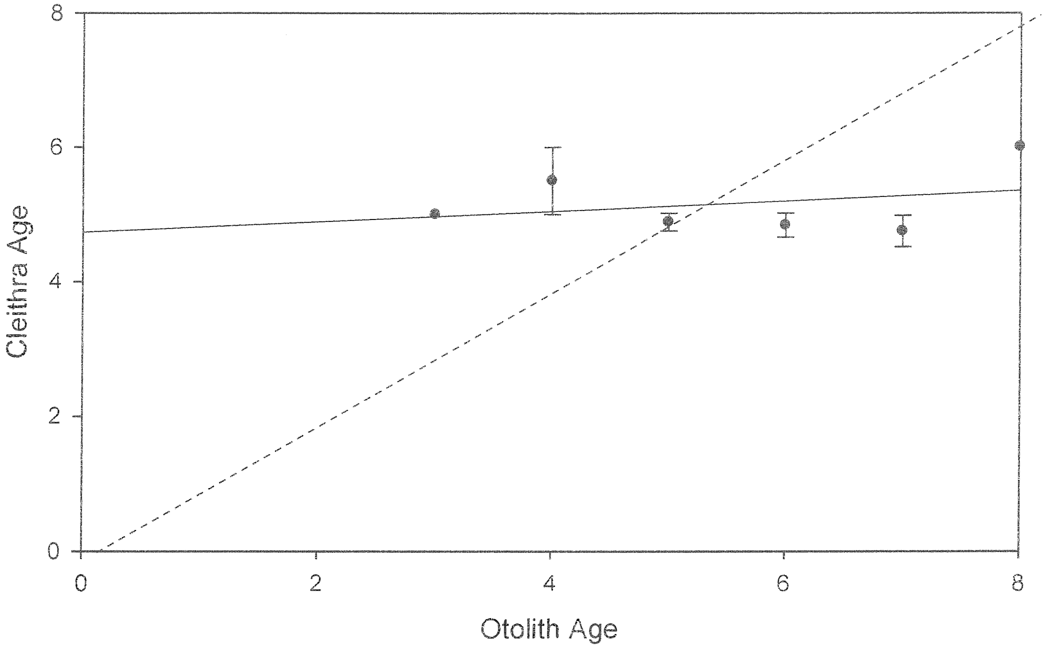

Figure 1.8. Age bias plot for otoliths compared to cleithra for Asian Carp from the Spoon, Sangamon, and Mackinaw Rivers and Salt Creek during 2013-2014 ($p > 0.05$). Solid line represents regression between otolith age and cleithra age, dashed line represents slope of one, and error bars represent standard error of the mean.

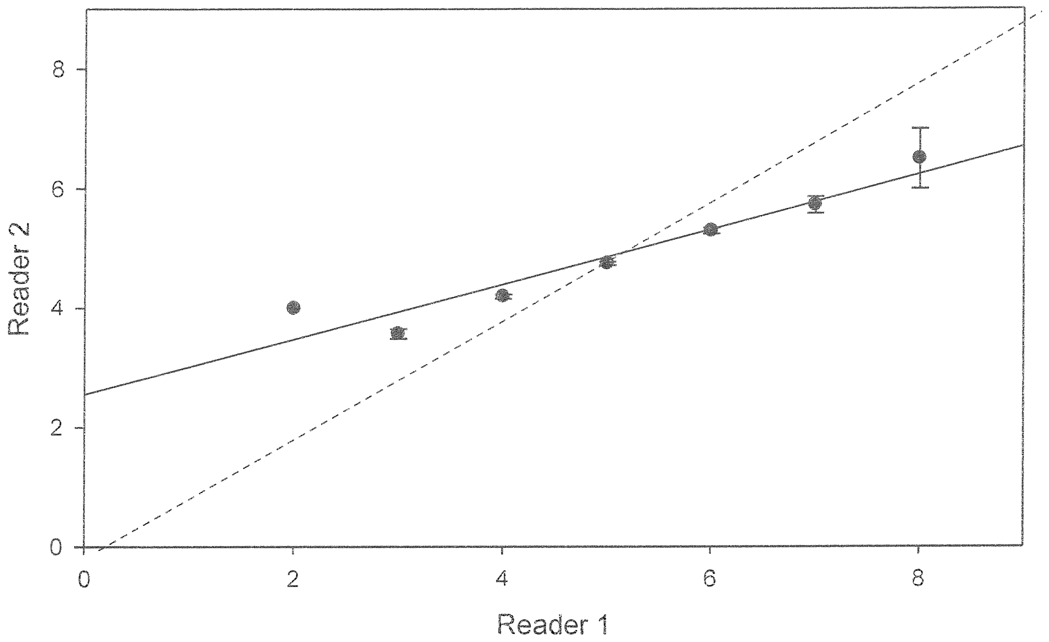

Figure 1.9. Age bias plot for Reader 1 compared to Reader 2 for Asian Carp from the Spoon, Sangamon, and Mackinaw Rivers and Salt Creek during 2013-2014 ($p > 0.05$). Solid line represents regression between Reader 1 and Reader 2 age, dashed line represents slope of one, and error bars represent standard error of the mean.

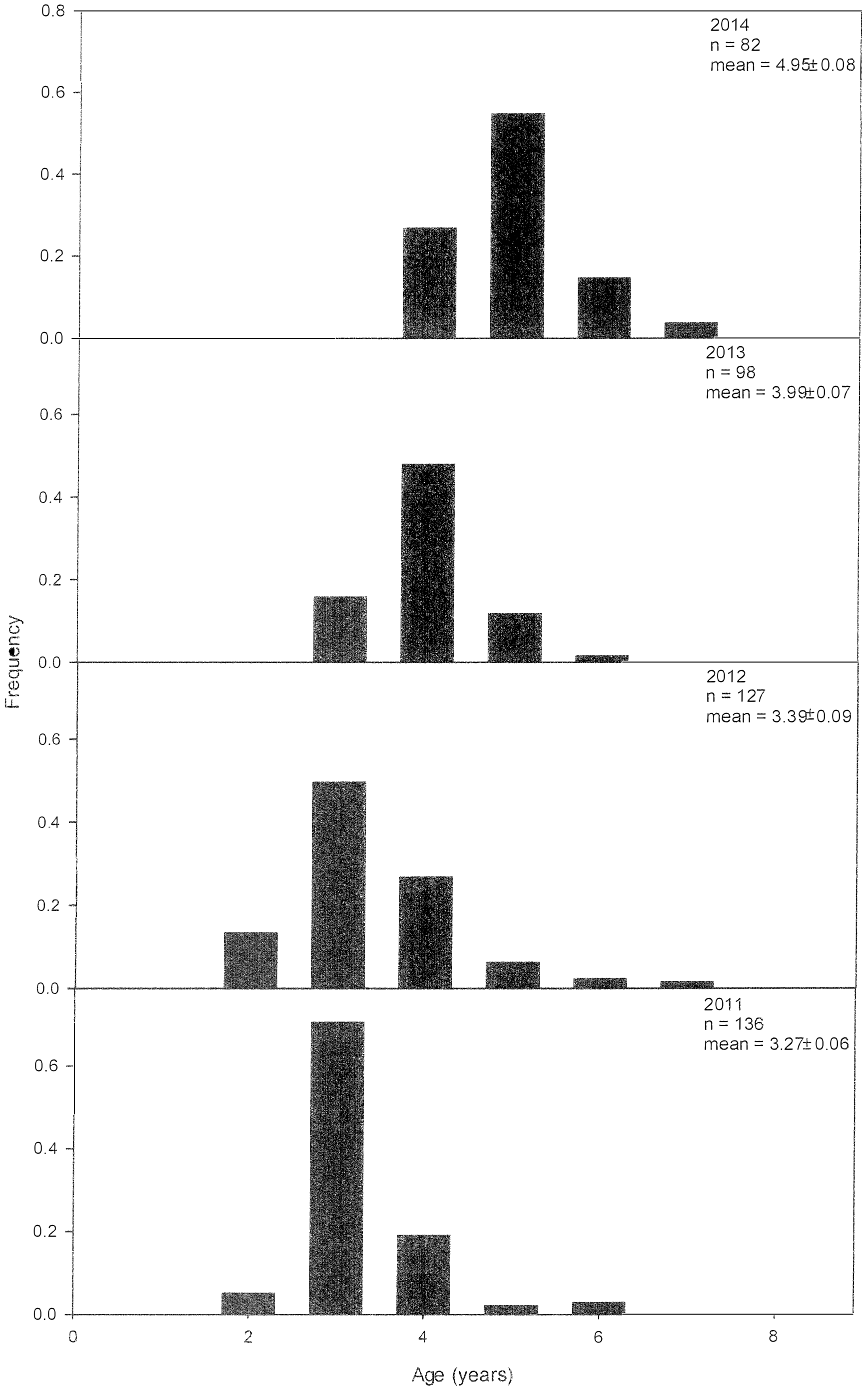

Figure 1.10. Age frequency distributions (years) of Asian Carp sampled by pulsed-DC electrofishing in the Illinois River during 2011-2014.

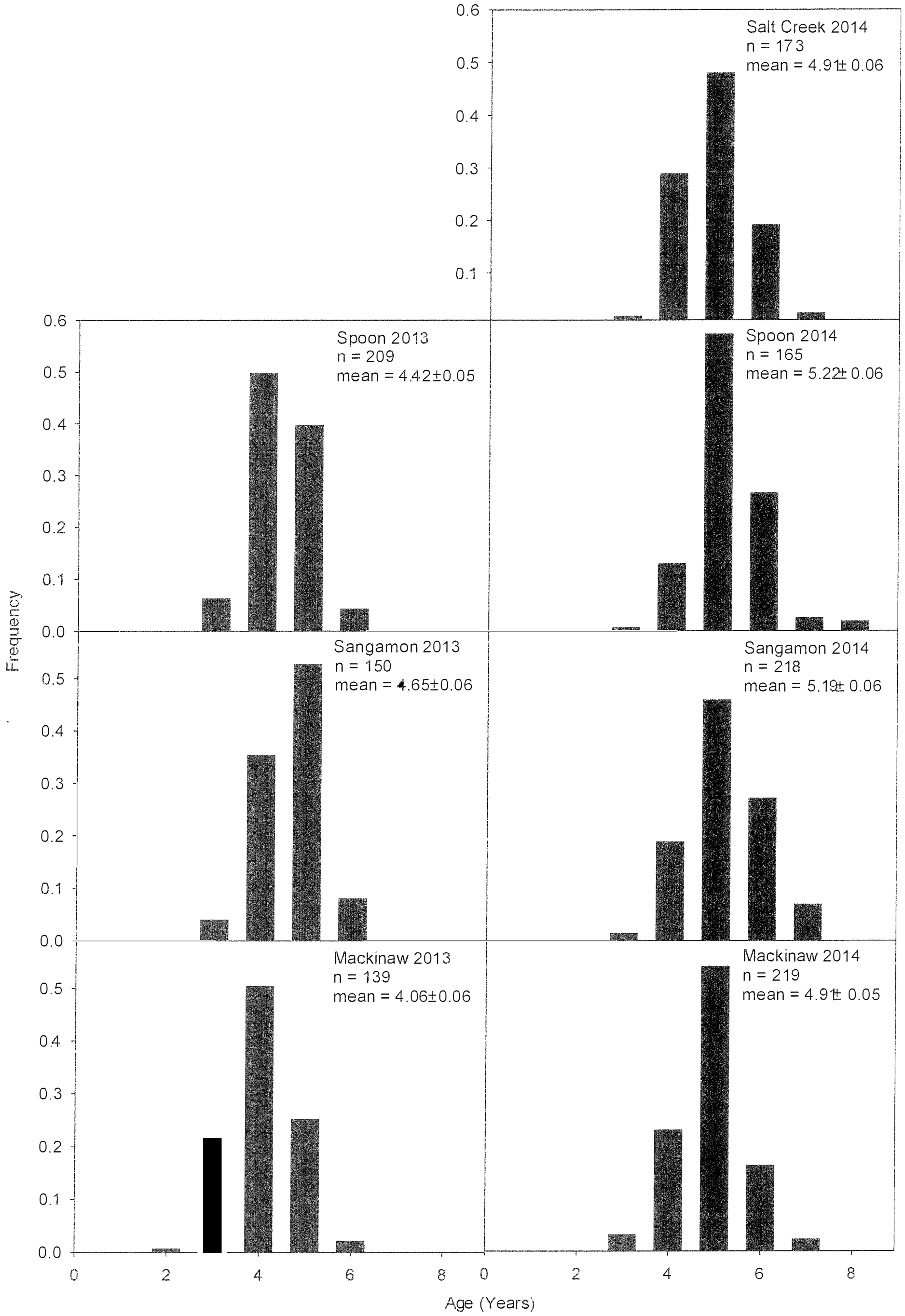

Figure 1.11. Age frequency distributions (years) of Asian Carp sampled by pulsed-DC electrofishing in the Spoon, Sangamon, and Mackinaw Rivers and Salt Creek during 2013-2014.

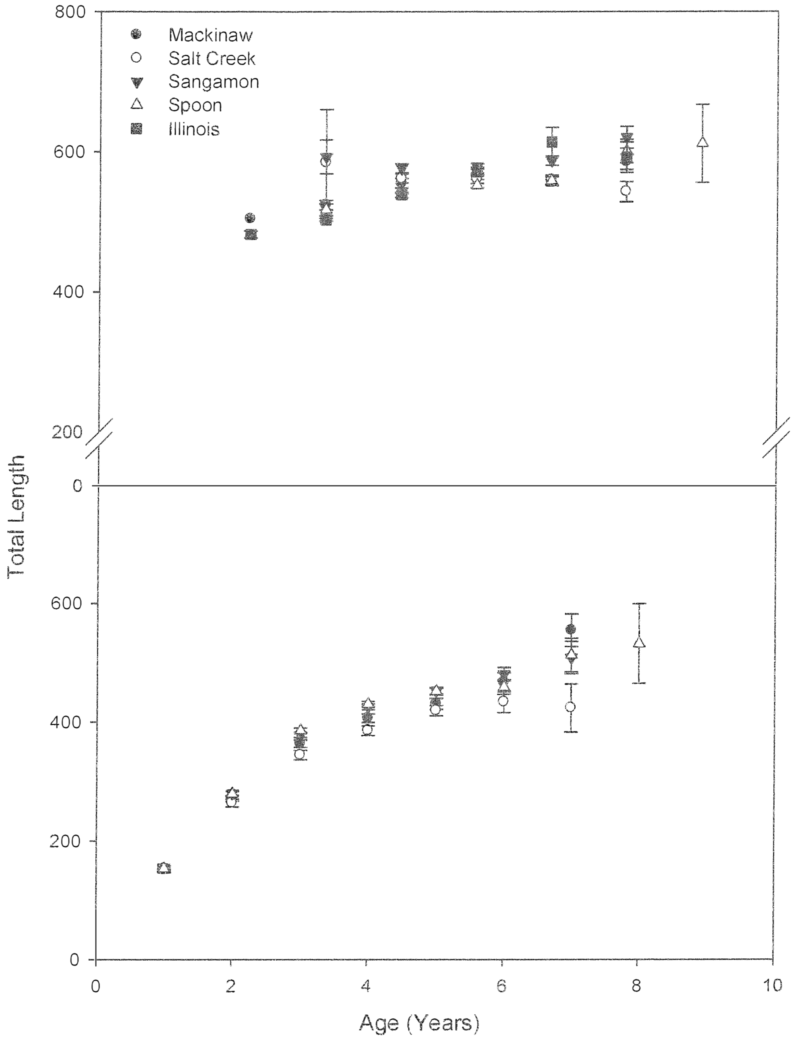

Figure 1.12. Mean length at capture ± SE (top) and mean back-calculated length at age ± SE (bottom) for Asian Carp sampled by pulsed-DC electrofishing in the Illinois (2011-2014), Mackinaw, Sangamon, and Spoon Rivers and Salt Creek during 2013-2014.

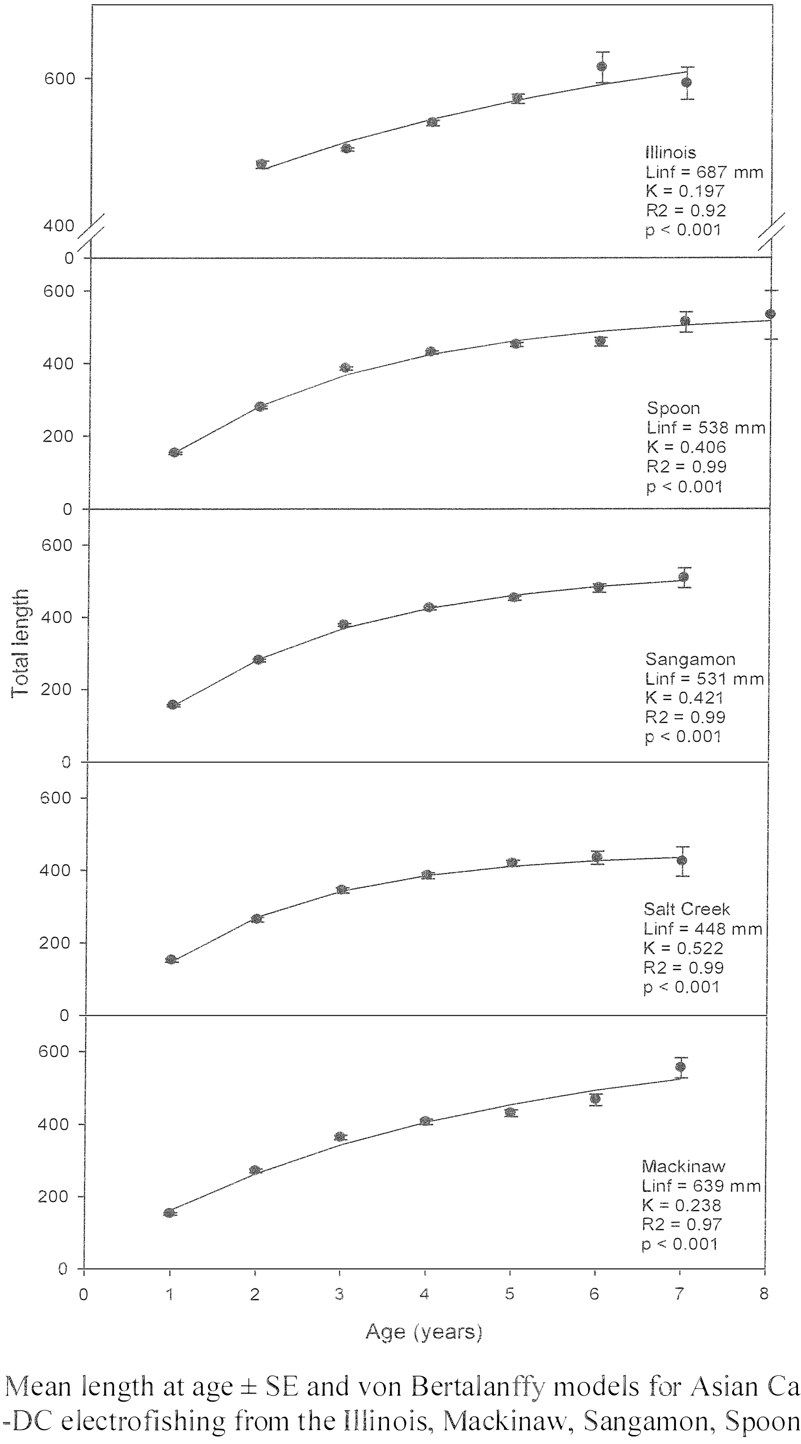

Figure 1.13. Mean length at age ± SE and von Bertalanffy models for Asian Carp sampled using pulsed-DC electrofishing from the Illinois, Mackinaw, Sangamon, Spoon Rivers and Salt Creek during 2011-2014.

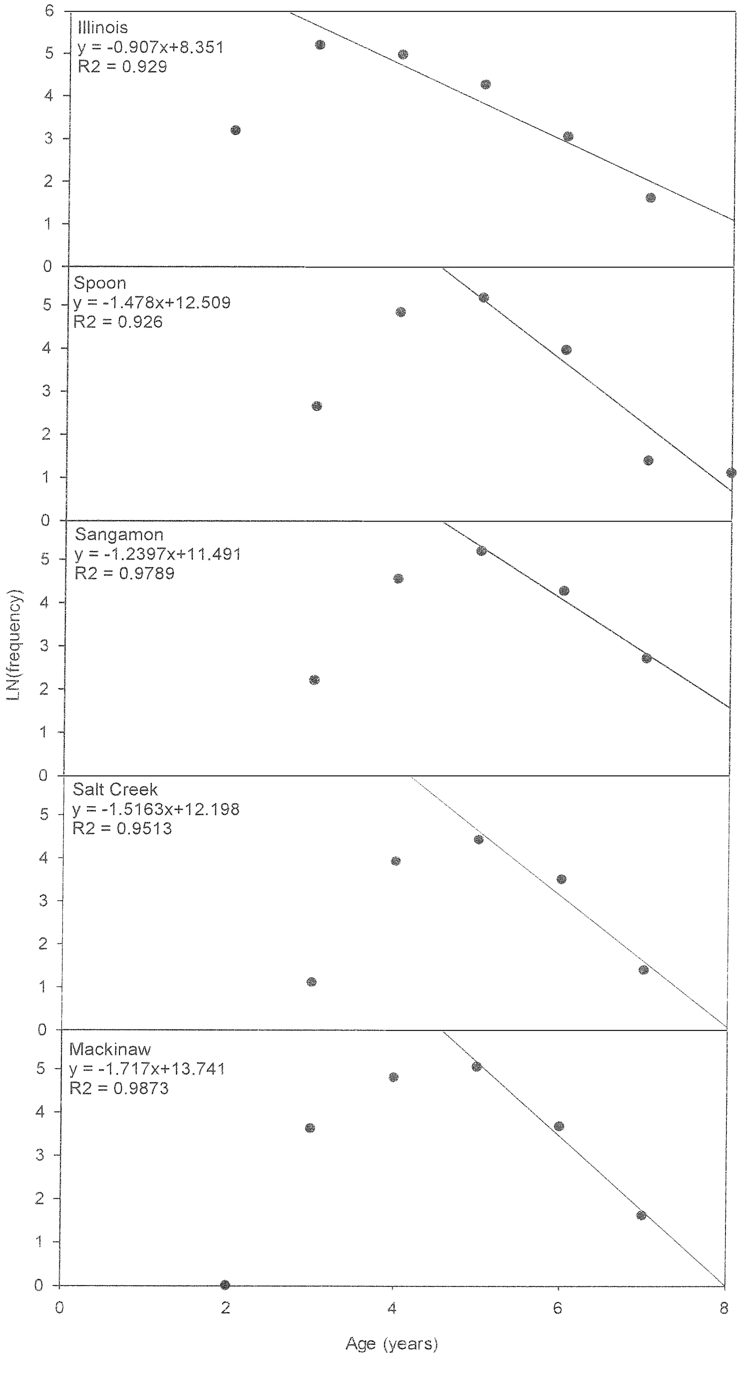

Figure 1.14. Catch-curve analysis of Asian Carp sampled by pulsed-DC electrofishing in the Illinois River (2011-2014), Spoon, Sangamon, and Mackinaw Rivers and Salt Creek during 2013-2014.

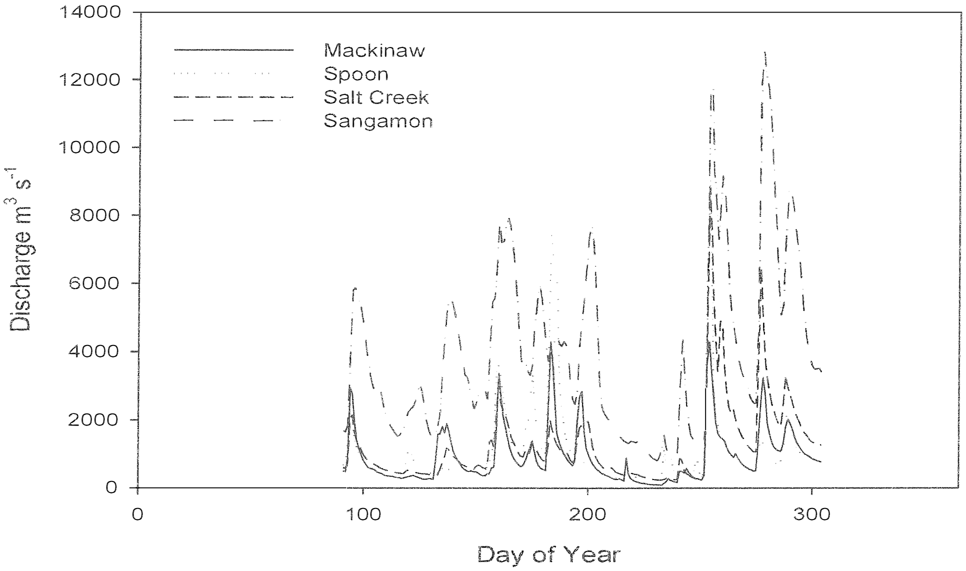

Figure 1.15. Mean daily discharge in Salt Creek (dashed line; USGS station number: 5582000), Spoon River (dotted line; USGS station number: 5570000), Sangamon River (dash-dot line; USGS station number: 5583000), and Mackinaw River (solid line; USGS station number: 5568000).

LARVAL FISH AND LARVAL ASIAN CARP COMMUNITY DYNAMICS IN ILLINOIS RIVER TRIBUTARIES

ABSTRACT

Large floodplain rivers are complex systems, driven by spatial heterogeneity and diversity. Much information is available concerning early life histories of fish in large river main channels and backwaters, however, little understanding exists about early life contributions of tributaries to large rivers. Of special concern are invasive Asian Carp, lotic specialists whose reproductive abilities in small rivers is unknown. I sampled eggs and larval fish in five tributaries of the Asian Carp-invaded Illinois River during 2013-2014: Salt Creek, the Mackinaw, Kankakee, Sangamon, and Spoon Rivers to examine abundance, spatial and temporal distributions, community assemblage structure, and size structure. Peak abundances of eggs and larval fish followed peaks in discharge in each tributary, indicating spawning activity characteristic of fluvial specialists. Ten different families were sampled; Clupeids and Cyprinids were the most abundant taxa. Temporal trends showed peak abundances of Clupeids during May, Moronids during June, and Catostomids, Centrarchids, and Cyprinids during July. Significant temporal and spatial effects ($p < 0.05$) of community assemblages were driven by secchi depth and water temperature, respectively. Post gas bladder emergent larval Asian Carp species were sampled in high abundance in the Spoon and Mackinaw Rivers, representing over 80% of larval fish sampled. Asian Carp were first present in late May with peak densities were in July. Illinois River tributaries are important habitats for fish early life stages including larval Asian Carp and contribute to the diversity of fishes in the main channel habitats. This diversity could be at risk with the high reproductive success of Asian Carp and other invasive species.

INTRODUCTION

Fish recruitment in large floodplain rivers is subject to interactions among several inter- and extra-channel habitats, making large rivers incredibly complex systems. Large floodplain rivers are characterized by a wide array of habitats, each varying in its degree of connectedness to the main channel. High habitat heterogeneity of these systems contributes to high species diversity (Taylor et al. 2006). Contributing to habitat heterogeneity are extra-channel habitats such as floodplains, backwaters, and tributaries. Through seasonal flooding and connectedness, these habitats contribute nutrients to the main river habitats, as well as provide critical habitat for many life stages of organisms (Junk et al. 1989; Winemiller 2004). Among the most influenced of these organisms are fishes, many of which rely on seasonal hydrological events and interactions between river habitats to complete various life stages.

Fluvial dependent fishes rely on flowing water, using currents to facilitate egg dispersal to adequate habitats (Schludermann et al. 2012). Although main channel habitats provide hostile conditions for larval fish, previous studies have found an abundance of larval fish in the main channel habitats of large rivers (Nannini et al. 2012; Csoboth and Garvey 2008; Brown and Coon 1994; Holland and Sylvester 1983). Early life stages of fish will often seek out refuge areas from the harsh conditions of the main channel, characterized by low flows, decreased turbidity, and high residence times (Galat et al. 2004).

Refuge areas may consist of channel border habitats, where flows are reduced, and connected backwater, side channel, or tributary habitats. Tributaries can serve as backwater areas, with downstream reaches experiencing less flow caused by backflow from the main channel, low stream gradient, and reduced depth from sedimentation (Brown and Coon 1994). Tributary habitats are important for fish communities for reproduction, foraging, and refuge; especially as anthropological disturbances, such as leveeing, channelization, and agriculture, continue to reduce and isolate critical off-channel habitats within large river systems (Koel and Sparks 2002). Tributary habitats are important for fish early life stages, from fluvial dependent species in the Great Lakes (Mansfield 1984), to recruitment sources for many fish species in the Missouri River (Brown and Coon 1994).

Like many Midwestern rivers, the Illinois River is an impounded large river heavily disturbed by anthropogenic influence including eight dams, forming pool reaches more indicative of lentic habitat than lotic (Koel and Sparks 2002, Mills et al. 1966). Fluvial dependent fishes rely on flowing water for reproductive requirements, and in disturbed habitats such as the Illinois River, these conditions are not always met. Tributaries are often less disturbed habitats because of reduced potential for transportation and recreation, and can cumulatively contribute to continuous lotic segments in disturbed systems (Koel and Sparks 2002). These segments have the potential to augment disturbed flows in impounded rivers, allowing fluvial dependent fishes to fulfill necessary spawning requirements (Brown and Coon 1994). Other taxa are tributary dependent specialists and rely on these areas for reproduction (Mansfield 1984).

Compared to studies on similar large rivers, investigations of larval fish compositions in the Illinois River have been restricted to the main channel and connected backwaters (Nannini et al. 2012; Csoboth and Garvey 2008), resulting in a significant knowledge gap regarding larval fish assemblages in associated tributaries. I examined fish early life stage use and assemblage structure of lower tributary reaches within of the Illinois River. In addition, I also wanted to determine if invasive species, such as Asian Carp, were reproducing in these areas. To accomplish this, my objectives were to 1) determine location and timing of egg and larval fish occurrences within tributaries, 2) compare community assemblages within tributaries, 3) examine the influence of abiotic factors on larval fish communities, and 4) assess larval fish size structure over time Early life history of Asian Carp has been relatively little studied in North America, especially regarding small river use by these individuals (Kocovsky et al. 2012). By studying tributary use by early life stages of fish, we may determine critical spawning habitat within tributaries of the Illinois River.

METHODS

Study Area

Four tributaries of the Illinois River were sampled each year during 2013-2014. In 2013, I sampled the Spoon (SP), Sangamon (SG), Mackinaw (M), and Kankakee (K) Rivers. In 2014, Salt Creek (SCR), a tributary of the Sangamon River, replaced the Kankakee River (Figure 2. 1). Each river was sampled at a downstream location near the mouth while taking into consideration seasonal accessibility.

Abiotic factors

Water temperature (°C), dissolved oxygen (mg/L) (YSI model 52, Yellow Springs Instruments, Yellow Springs, OH), flow (m/s) (Hach model FH950, Hach Company, Loveland, CO), secchi depth (cm), and discharge (ft^3/s) were collected during each sampling event. To determine the effects of discharge on larval fish, daily discharge data was obtained from the United States Geologic Survey for the most downstream gauge for each sampled tributary in 2014. The Salt Creek gauge was Greenview, IL (USGS station number: 5582000); Seville, IL Spoon River (USGS station number: 5570000) for the Spoon River; Oakford, IL (USGS station number: 5583000) for the Sangamon River; and Green Valley, IL (USGS station number: 5568000) for the Mackinaw River.

Larval Fish Sampling

Larval fish sampling of each tributary river occurred at least monthly from April-October at each site when accessible. In the spring and summer months during peak spawning times (Deters et al. 2012), larval fish sampling was conducted weekly or bi-weekly to increase chances of encounter. A 500µm mesh conical cylindrical ichthyoplankton net measuring 0.5m x 3m mounted to a rigid frame at the front of a boat was pushed against the current for five minutes for three replications at each site (Claramunt et al. 2005). A flow meter (model 2030R, General Oceanics, Inc., Miami, FL) attached in the center of the mouth of the net was used to estimate volume of water filtered. The net was operated just under the surface of the water and sampled the top half meter of the water column for suspended and pelagic eggs and larvae. Three five-minute samples were taken at each downstream site, one near each bank and a main channel sample during daylight hours, producing samples from main channel and channel border habitat strata.

All larval samples were fixed in 95% ethanol in the field until identification. Fixed samples were returned to the laboratory for enumeration and subsequent family identification based on meristic, morphometric, and composite characteristics (Auer 1982). Family identification was chosen to reduce risk of misidentification for most taxa; Cyprinids were further identified to include/exclude *Hypophthalmichthys sp.* (Chapman 2006). Following identification, each specimen was measured for total length to the nearest millimeter.

Data Analysis

Abiotic factors

Abiotic factors were tested using a repeated-measures analysis of variance (RMANOVA) to compare environmental factors over time in the sampled rivers, because each discrete site was sampled repeatedly. Pairwise correlations were used to determine relatedness between hydrographs for each sample river in 2014.

Larval fish abundance

Following identification, density was calculated for eggs and larval fish as number sampled per one-hundred cubic meters (number·$100m^{-3}$) of water sampled, as a measure of catch per unit effort (CPUE, count·$100m^{-3}$). Relative abundance could then be calculated. To compensate for heteroscedascity, egg and larval fish densities were log-transformed [log10(x+1)] (Hubert and Fabrizio 2007). Because *Hypophthalmichthys sp.* represented ~80% of the total sampled, separate analyses were performed with and without to examine influences of other taxa more closely. To test for differences in total egg and larval fish densities between rivers and habitat strata (main channel, channel border), I used ANOVA with a Tukey-Kramer Honestly significant difference post hoc test for multiple comparisons.

Because I sampled repeatedly at discrete sites, split-plot RMANOVA (Maceina et al. 1994) was used to compare densities among rivers through time. The main plot variable was site and the subplot variables were day sampled, using log-transformed densities of eggs and larval fish for each analysis. Low water made some sites inaccessible during August and September, therefore, densities were compared from May to July.

Larval fish assemblages

Overall assemblage structure was analyzed using nonmetric multidimensional scaling (NMDS). All data were standardized to average density and log-transformed to meet the assumptions of normality. Bray-Curtis similarity was used to explain the differences among river or sampling trip. To assess community structure over time sampled, log-transformed average density of each species was plotted against time sampled. Differences in community structure between rivers were assessed for log-transformed average density of each species for each site. Permutational multivariate analysis of variance (PERMANOVA) was chosen to explain differences in community assemblages between sampling time and sampled rivers (Anderson and Walsh 2013). Additionally, environmental variables (secchi depth, surface water temperature, dissolved oxygen, and discharge) from each site were correlated with the NMDS ordination axis using the envfit function in program R using 999 permutations (Oksanen et al. 2015). Significantly correlated variables were overlayed using biplot vectors in ordination space.

Laval fish size structure

To investigate differences in size structure of fish over time, nonparametric Kolmogorov-Smirnov cumulative distribution tests compared size structure of larval fish taxa over sampling time. Multiple pairwise comparisons determined size structure differences between sample dates with Bonferroni correction for multiple comparisons. All statistics were performed using Program R© (v.3.1.1, R Core Team 2014) software and a significance level was set at $\alpha = 0.05$ unless otherwise stated.

RESULTS

Abiotic factors

Predictably, abiotic factors reflected local water conditions, with elevated secchi depths and flows during periods of high discharge and elevated surface water temperatures during the summer months (Figure 2. 2). Of the environmental variables taken, secchi depth showed a significant temporal effect ($F_{2,22} = 3.85$, $p = 0.04$), whereas surface water temperature displayed a spatial effect, differing significantly between the rivers sampled

($F_{3,11} = 5.75$, $p = 0.01$). No significant effect of time or site was observed for flow ($F_{3,11} = 1.13$, $p = 0.38$).

Discharge between rivers varied, with the Sangamon River showing flashy characteristics, with the remaining rivers exhibiting somewhat more typical spring and fall flooding events (Figure 2. 3). In 2014, the Sangamon River had the highest discharge by mean (±SE) discharge (4003.89±18.36 m^3/s) and the lowest the Mackinaw River (936.20±6.04 m^3/s). Intermediary in mean discharge were the Spoon River (1076.83±9.30 m^3/s) and Salt Creek (1276.61±8.87 m^3/s). However, positive pairwise correlations suggest that flow patterns were similar among sampled tributaries (Table 2. 1).

Larval fish abundance

Over the sampling period, 1817 eggs and 2994 larval and juvenile fish were collected during 2013-2014 using a conical cylindrical ichthyoplankton net. We expended a total of 960 minutes of effort sampling, filtering approximately 10200 m^3 of water. Only 35 minutes of effort were expended in 2013, resulting in the collection of two larval fish, one percid and one clupeid from the Kankakee and Spoon Rivers, respectively. Given the low sample size from 2013, further analysis will include results from 2014 only. Of the 2992 larval and juvenile fish sampled during 2014, 80% were Asian Carp species (Table 2. 2). Without Asian Carp, 605 larval and juvenile fish from 10 families were sampled; Cyprinids and Clupeids dominated the remaining catch. Catostomids, Moronids, and Centrarchids made up the remainder of the sample. Atherinids, Ictalurids, Lepisosteids, Percids, and Scianids combined comprised less than 2% of the total larval and juvenile fish collected (Table 2. 3).

Total density of larval fish does not display a significant effect of river ($F_{3,179} = 0.7668$, $p = 0.514$) or habitat strata ($F_{1,181} = 3.355$, $p = 0.06863$). Similarly, removing the effects of the large number of Asian Carp reveals no significant effect of river on total density ($F_{3,179} = 0.3856$, $p = 0.7635$), however, a significant effect of habitat strata is observed with remaining taxa ($F_{1,181} = 4.446$, $p = 0.03636$) with a higher proportion of fish sampled in main channel habitats (58% of the total). Although a significant effect of strata was observed on total density, no significant effect of strata was observed when Asian Carp were removed from analyses (Table 2. 4).

Spatial and temporal distributions of eggs

Total densities of eggs did not differ between main channel and channel border habitats ($F_{1,181} = 0.1521$, $p = 0.697$), however, a significant effect of river was observed ($F_{3,179} = 3.122$, $p = 0.0273$). Tukey-Kramer HSD testing revealed significantly higher egg density in the Sangamon River than the rest of the rivers. Neither sample river ($F_{3,8} = 3.22$, $p = 0.08$) nor month sampled ($F_{11,88} = 0.13$, $p > 0.99$) had an effect on egg density, however, a significant interaction of river and month ($F_{33,88} = 3.56$, $p < 0.0001$) was observed (Figure 2. 4).

Spatial and temporal distributions of larval fishes

A significant effect of month was observed on total larval fish density ($F_{11,88} = 4.76$, $p < 0.0001$), but no effect of river was seen ($F_{3,8} = 2.89$, $p = 0.10$). Similar to total egg density, a significant interaction of river and sampling month was observed for total larval fish density ($F_{33,88} = 5.34$, $p < 0.0001$). When the large number of Asian Carp were removed for analysis, similar results were observed for sampling time ($F_{11,88} = 4.75$, $p < 0.0001$), river ($F_{3,8} = 1.02$, $p = 0.43$), and river-month interactions (Figure 2. 4; $F_{33,88} = 3.35$, $p < 0.0001$). Asian Carp displayed significant temporal ($F_{10,70} = 4.47$, $p < 0.0001$) and spatial ($F_{3,7} = 18.07$, $p = 0.001$) components, as well as a significant interaction between both factors ($F_{30,70}$

= 11.80, $p < 0.0001$). Taxa also showing temporal components were Cyprinids (without Asian Carp; ($F_{10,70} = 2.59$, $p = 0.01$), Centrarchids ($F_{10,80} = 11.26$, $p < 0.0001$), and Clupeids ($F_{11,88} = 6.38$, $p < 0.0001$). Centrarchids ($F_{3,8} = 6.74$, $p = 0.01$) and Moronids ($F_{3,8} = 9.28$, $p = 0.005$) each displayed effects of sample river, whereas Catostomids displayed no significant spatial or temporal effects ($F_{1,8} = 2.40$, $p = 0.16$) (Figure 2. 5).

Assemblage structure

Nonmetric multidimensional scaling of taxa densities sampled over time revealed grouping between months sampled (Figure 2. 6); PERMANOVA results indicate a significant effect of month sampled on taxa composition ($F_{5,16} = 1.61$, $p = 0.049$). Secchi depth had a significant correlation with the ordination axis ($r^2 = 0.45$, $p = 0.015$); no other environmental variables had a discernible effect on taxa composition during sample time (Figure 2. 6; $p > 0.05$). Taxa densities grouped by river as well (Figure 2. 7), with a significant effect of river on the model ($F_{3,11} = 2.91$, $p = 0.005$). A significant correlation of temperature was seen with the ordination axis of taxa densities by river ($r^2 = 0.62$, $p = 0.013$), however, no other significant correlations of environmental variables was observed, including habitat strata (Figure 2. 7; $p > 0.05$).

Larval fish size structure

Asian Carp varied in size between June and July ($D = 0.88$, $p < 0.0001$), with too few sampled in May for comparison. Removing Asian Carp from consideration also shows significant differences in size structure of Cyprinids, Catostomids, and Clupeids between months sampled as well ($p < 0.017$; Table 2. 5; Figure 2. 8).

DISCUSSION

Larval fish distribution and taxonomic composition

Tributaries represent critical habitats for large river fishes, whether for spawning habitat (Mansfield 1984), refugia (Junk et al. 1989), or nutrition (Thorp et al. 2006). Our data suggest that tributary habitats support early life stages of multiple taxa of fishes: fluvial dependents such as cyprinids, catostomids, and sciaenids, as well as several macrohabitat generalist taxa such as centrarchids, clupeids, lepisosteids, atherinids, percids, and ictalurids (Csoboth and Garvey 2008; Galat and Zweimuller 2001). Fluvial dependents are regarded as taxa requiring lotic environments for some portion of their life whereas macrohabitat generalists are characteristic of lentic dwelling taxa (Csoboth and Garvey 2008; Galat and Zweimuller 2001). Predictably, higher proportions of fluvial dependents were capturedcompared to macrohabitat generalists. High contributions of fluvial dependents such as cyprinids, catostomids, and moronids were likely attributable to lotic habitats (Csoboth and Garvey 2008; Nannini et al. 2012; Galat and Zweimuller 2001). Pelagic spawning sciaenids, i.e. freshwater drum (*Aplodinotus grunniens*), were sampled in low densities, although some studies report high densities in large rivers (Nannini et al. 2012). Even though conditions were appropriate for sciaenid spawning, these species have been noted to be limited by river size in their distribution (Becker 1983), possibly accounting for low densities, similar to findings in Missouri River tributaries (Brown and Coon 1994). Although considered macrohabitat generalists, clupeid larvae represented a large proportion of the catch in our sample rivers, consistent with other studies in lotic systems (Gallagher and Conner 1982; Muth and Schulbach 1984; Scheidegger and Bain 1995).

Modest presence of centrarchids, a nesting taxa, are likely a result of displacement by rapid flooding events (Floyd et al. 1984; Scheidegger and Bain 1995; Scott and Nielson 1989). Similarly low catches of macrohabitat generalists such as atherinids, ictalurids, lepisosteids, and percids occurred. This study focused on pelagic eggs and larvae, sampling the top half meter of the water column, therefore excluding taxa utilizing remaining portions of the water column. Additionally, taxa inhabiting extra-channel habitats such as connected backwaters or shallow vegetated habitats were also not sampled.

Location and timing of spawning

Temporal changes in egg and larval fish densities were apparent. Peak densities of eggs occurred during June, with peak densities of larval fish occurring during May, following spring flooding events. Early peaks of larval fish densities were driven by the high clupeid densities in May in the Mackinaw River and high densities of eggs in June in the Sangamon River. Other abundant taxa influence subsequent larval fish peaks. Clupeids spawn during April-May, however, their adhesive demersal eggs likely elude our sampling gear (Pflieger 1997; Smith 1979). Clupeids showed peaks during May-June, predominantly in the Mackinaw and Sangamon Rivers. Similar studies in large rivers report sampling larval clupeids during these times (Reeves and Galat 2010; Nannini et al. 2012; Muth and Schmulbach 1994), as well as in temperate lakes (Welker et al. 1994). Strong temporal patterns were also present in centrarchid species, sampled in highest concentrations in the Mackinaw River during June-July. Larval centrarchids were similarly sampled in rivers in the Illinois River and a Kentucky stream (Nannini et al. 2012; Floyd et al. 1984). Cyprinid species (excluding Asian Carp) additionally displayed temporal effects with peak densities sampled during July in all the sample tributaries. In the Missouri River, larval cyprinids were sampled in summer months (July-August, Reeves and Galat 2010); peak densities in a South Dakota stream were also sampled during July (Muth and Schmulbach 1984). Although no significant effects of time were observed, moronid species were exclusively sampled in the Sangamon River drainage during May and June. Moronid species typically spawn over rocky substrate during March and April, using tributaries of large rivers (Pflieger 1997). Moronids in our study are likely limited to the Sangamon River drainage because the other systems sampled lack sufficient substrate for spawning. Larval catostomid densities were among the most varied temporally of the taxa sampled in this study. Peak densities were generally observed in June, however, peak densities in Salt Creek were observed in July. These differences are likely due to species differences, but this study lacks the taxonomic refinement to make definite conclusions.

Larval fish community assemblages

Over the months sampled, larval fish assemblages were driven by secchi depth in this study. A significant effect of sample time was observed, evidenced by the groupings of samples from May, June, and July. When samples were scaled by river, temperature showed a significant correlation with larval fish assemblages. Increases in temperature led to changes in larval fish community structure. In contrast, larval fish assemblages in the Illinois River were driven by channel depth and flow (Nannini et al. 2012), whereas in the Missouri River and its tributaries, assemblages also separated with respect to depth and turbidity (Brown and Coon 1994). Overall, secchi and temperature seem to be driving factors in this study, however, effects of flow I was unable to quantify may be the result of multiple flooding events spanning sampling time, especially an unseasonable fall flood.

Size structure of larval fish

Length frequency histograms for larval fish allowed us to track spawning cohorts through time for *Hypophthalmichthys* spp., cyprinids, catastomids, and clupeids; all other taxa were not represented in large enough numbers for analyses. Significant differences were observed between months sampled, indicating differences in timing of spawning events and growth of cohorts over time. Sub-10 mm categories were observed for catostomids, cyprinids, and clupeids during each month, representing separate spawning events during each month for each taxa. Dettmers et al. (2001) provides evidence for protracted spawning for these taxa in the Lower Illinois River, whereas Floyd et al. (1984) observed species-driven differences within these taxa sampled in a Kentucky stream. Temporal progression of individuals over ~10 mm provides evidence for tracking these cohorts through time, for example, larger clupeids were seen in July compared to prior months. This remains consistent with clupeids in the Illinois River (Nannini et al. 2012) as well. Similar trends were additionally observed in cyprinids and catostomids. Low frequencies of larger bodied larval and juvenile fish may also be attributed to evasion to our gear, as individuals increase in size, motor development contributes to gear evasion (Kelso et al. 2013).

Invasive species effects on larval fish communities

Asian Carp are injurious exotic species recently established, and subsequently multiplying exponentially within the Mississippi River basin (Chick and Pegg 2001). Asian Carp represented 80% of our total larval fish sample, beginning in the Mackinaw River in May with the majority of Asian Carp larvae and juveniles were sampled in the Spoon River in July. Numbers increased dramatically into July, where growth of these individuals likely excluded Asian Carp from our gear as they began to exceed 40 mm. Post-gas bladder emergent larval Asian Carp are considered to be free swimming (Deters et al. 2012), therefore it is difficult to say whether these individuals came from the tributaries themselves or if they migrated from the main channel. However, tributary confluences have been noted to provide favorable spawning conditions for Asian Carp (Nico et al. 2005; Deters et al. 2012), further reinforcing the import of tributaries for pelagic spawning fishes. Asian Carp sampled in this study are likely using tributary mouth habitats as refuge areas from the main channel flows. Evidence of this is the presence of Asian Carp larvae in the Spoon and Mackinaw Rivers, sampled very near to their confluences, however, no larval Asian Carp are found in either the Sangamon River (sampled 33 km from its confluence), or Salt Creek, a tributary of the Sangamon River.

Many midwestern large rivers have been studied regarding fish early life stages (e.g. Csoboth and Garvey 2008; Brown and Coon 1994; Nannini et al. 2012; Gallager and Conner 1983; Reeves and Galat 2010), however, this study becomes especially relevant given the distinct lack of studies focusing on larval fish assemblages in Illinois River tributaries. The establishment of invasive planktivorous Asian Carp, shown to impact native plankton communities following establishment (Sass et al. 2014) may affect the prey base for early life stages of fish, nearly all of which rely on plankton at some life stage for nourishment (Dettmers et al. 2001; Welker et al. 1994). This study becomes important to quantify existing tributary use by larval fish and provide useful information on larval production in tributaries of the Illinois River. Further examination is necessary to elucidate changes in larval fish production over time, and compare nutritional availability and driving abiotic factors influencing early life stages of fish, however, this study documents existing larval fish assemblages in Illinois River tributaries.

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Table 2. 1. Pairwise discharge Pearson correlation statistics between the Mackinaw River, Salt Creek, Sangamon River, and Spoon River. Bolded entries indicate significance according to Bonferroni corrections for multiple comparisons (p = 0.05/6 = p = 0.008).

	Mackinaw	Salt Creek	Sangamon	Spoon
Mackinaw		$r_{20029} = \mathbf{0.44}$	$r_{20029} = \mathbf{0.38}$	$r_{20029} = \mathbf{0.45}$
Salt Creek			$r_{20251} = \mathbf{0.84}$	$r_{20249} = \mathbf{0.57}$
Sangamon				$r_{20249} = \mathbf{0.42}$
Spoon				

Table 2. 2. Larval fish total catch using boat-mounted conical cylindrical ichthyoplankton net in the Mackinaw River, Salt Creek, Sangamon River, and Spoon River during 2014.

	Mackinaw	Salt Creek	Sangamon	Spoon	Total
Atherinidae	1				1
Catostomidae	5	32	29	13	79
Centrarchidae	23	2	1	4	30
Clupeidae	109	4	54	24	191
Cyprinidae	58	33	44	2488	2623
Hypoph. spp.	25			2362	2387
Without *Hypoph.*	33	33	44	126	236
Ictaluridae			1		1
Lepisosteidae			1	1	2
Moronidae		1	52		53
Percidae	1		3		4
Scianidae	3				3
Unknown	1	2	2	1	6
Total Count	201	74	187	2531	2993

Table 2. 3. Average percent family composition (% total) of larval fish excluding Asian Carp species using boat-mounted conical cylindrical ichthyoplankton net in the Mackinaw River, Salt Creek, Sangamon River, and Spoon River during 2014.

	Mackinaw	Salt Creek	Sangamon	Spoon	Total
Atherinidae	0.17	0	0	0	0.17
Catostomidae	0.83	5.28	4.78	2.15	13.04
Centrarchidae	3.80	0.33	0.17	0.66	4.95
Clupeidae	18.00	0.66	8.91	3.96	31.52
Cyprinidae	5.45	5.45	7.26	20.79	38.94
Ictaluridae	0	0	0.17	0	0.17
Lepisosteidae	0	0	0.17	0.17	0.33
Moronidae	0	0.17	8.58	0	8.75
Percidae	0.17	0	0.50	0	0.66
Scianidae	0.50	0	0	0	0.50
Unknown	0.17	0.33	0.33	0.17	1.00
Total	29.04	12.21	30.86	27.89	100

Table 2. 4. T-test statistics for larval fish density comparisons between main channel and channel border habitats for Hypophthalmichthys, cyprinids, catostomids, moronids, centrarchids, and clupeids sampled Salt Creek, Sangamon River, Spoon River, and Mackinaw River combined during 2014.

	T	Num. DF	Den. DF	*p* value
Hypophthalmichthys	0.041	1	181	0.839
Catostomidae	0.457	1	181	0.500
Cyprinidae w/o *Hypoph.* spp.	2.277	1	181	0.133
Moronidae	1.5	1	181	0.222
Centrarchidae	0.777	1	181	0.379
Clupeidae	1.61	1	181	0.206

Table 2. 5. Pairwise Kolmogorov-Smirnov test statistics for size structure comparisons of *Hypophthalmichthys* spp., cyprinids, catostomids, and clupeids between May, June, and July sampled during 2014. Bolded entries indicate significance according to Bonferroni corrections for multiple comparisons (p = 0.05/3 = p = 0.017).

		May	June	July
Hypophthalmichthys				
	May			
	June			**$D = 0.880$**
	July			
Cyprinidae				
	May		**$D = 0.683$**	**$D = 0.670$**
	June			**$D = 0.373$**
	July			
Catostomidae				
	May		**$D = 0.769$**	**$D = 0.726$**
	June			**$D = 0.875$**
	July			
Clupeidae				
	May		**$D = 0.900$**	**$D = 0.900$**
	June			**$D = 0.675$**
	July			

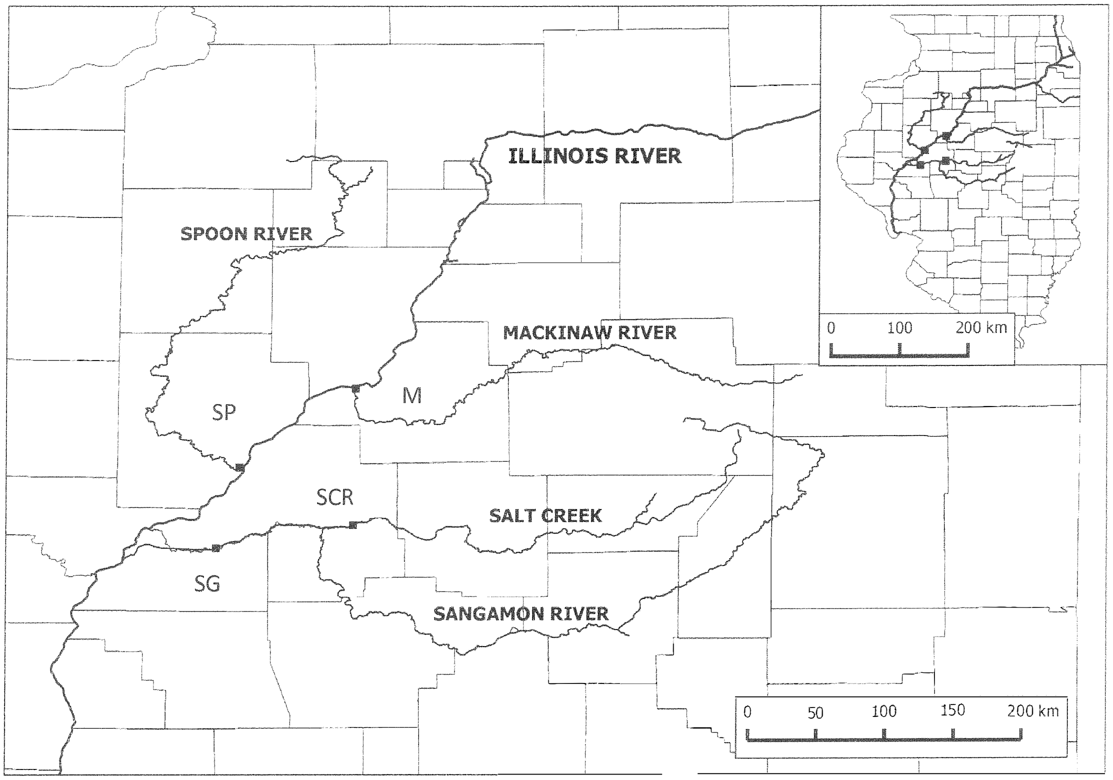

Figure 2. 1. Map of tributaries and sites within Illinois River system

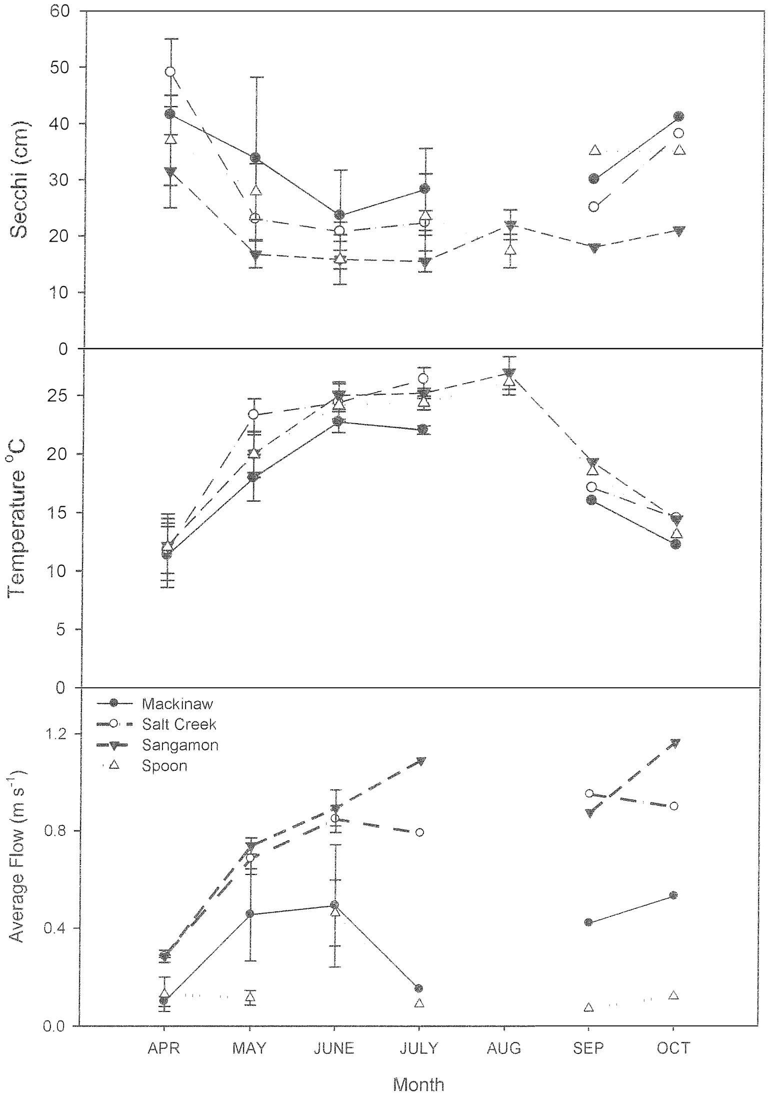

Figure 2. 2. Mean (±SE) secchi depth cm (top), surface water temperature °C (middle), and flow m·s^{-1} (bottom) from the Mackinaw River (solid lines, filled circles), Salt Creek (dash-dot lines, open circles), Sangamon River (dashed line, filled triangles), and Spoon River (dotted lines, open triangles) during 2014.

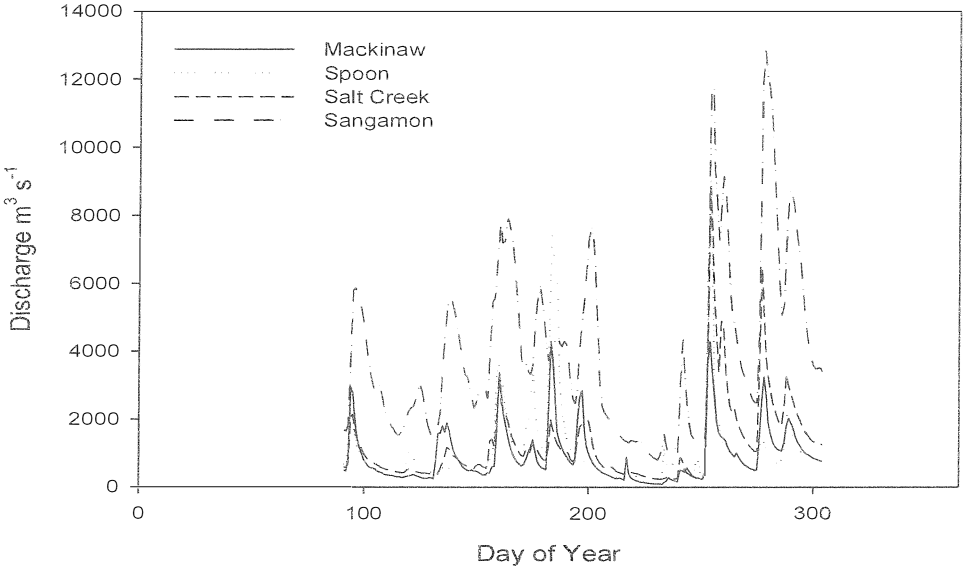

Figure 2. 3. Mean daily discharge from 1 April to 31 October in Salt Creek (short dashes, USGS station number: 5582000), Spoon River (dotted lines, USGS station number: 5570000), Sangamon River (dash-dots, USGS station number: 5583000), and Mackinaw River (solid lines, USGS station number: 5568000).

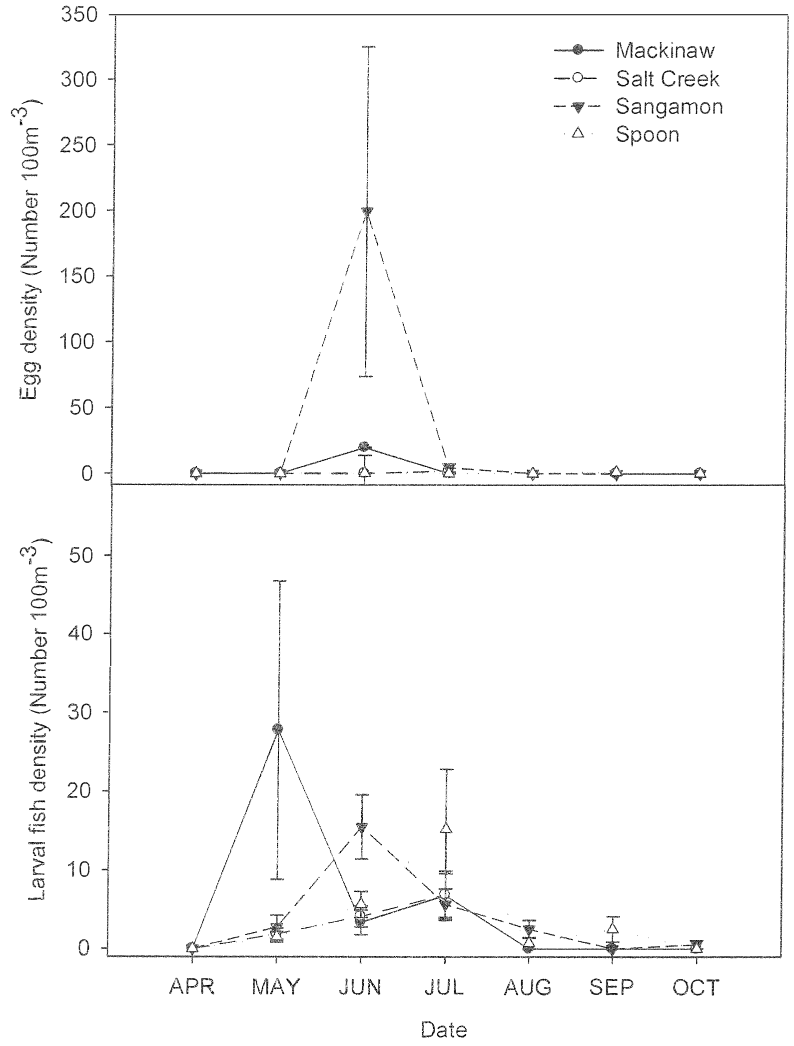

Figure 2. 4. Mean (±SE) total egg (top) and larval fish (bottom) densities excluding Asian Carp species (number·100m^{-3}) collected in from the Mackinaw River (solid lines, filled circles), Salt Creek (dash-dot lines, open circles), Sangamon River (dashed line, filled triangles), and Spoon River (dotted lines, open triangles) during 2014.

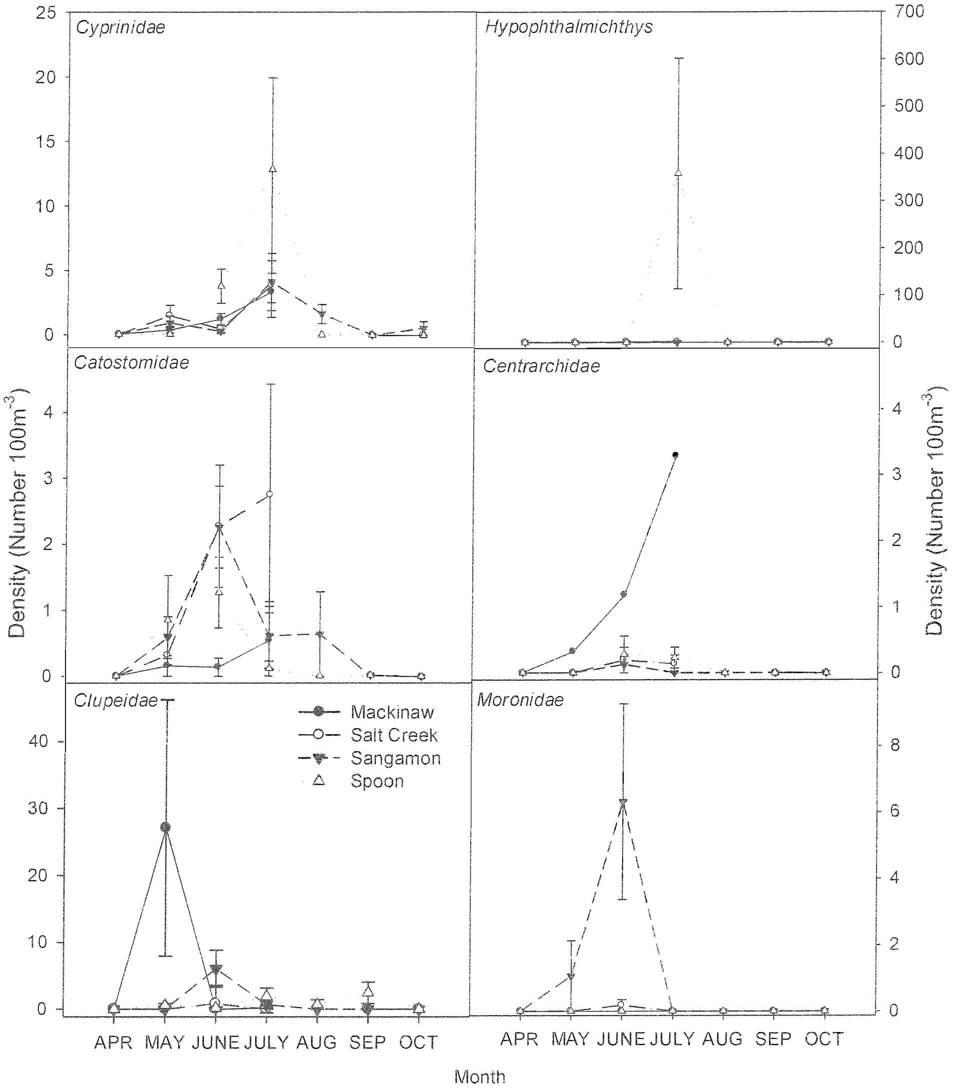

Figure 2. 5. Mean (±SE) densities (number·100 m^{-3}) cyprind spp. (excluding *Hypophthalmichthys* spp.), *Hypophthalmichthys* spp., catostomids, centrarchids, clupeids, and moronids collected in from the Mackinaw River (solid lines, filled circles), Salt Creek (dash-dot lines, open circles), Sangamon River (dashed line, filled triangles), and Spoon River (dotted lines, open triangles) during April-October, 2014.

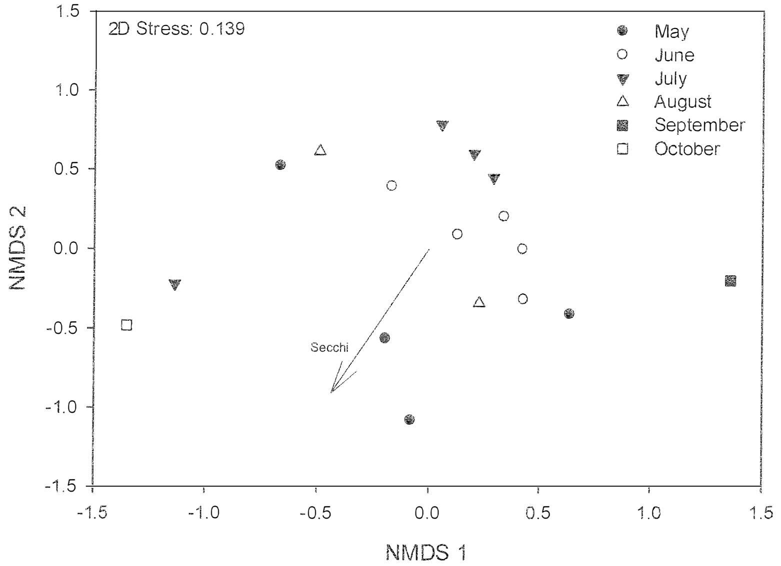

Figure 2. 6. Nonmetric dimensional scaling plot for average density of larval fish families excluding Asian Carp species sampled from the combined tributaries during May-October, 2014. Environmental vectors (arrow) plotted in ordination space to show correlation with sites.

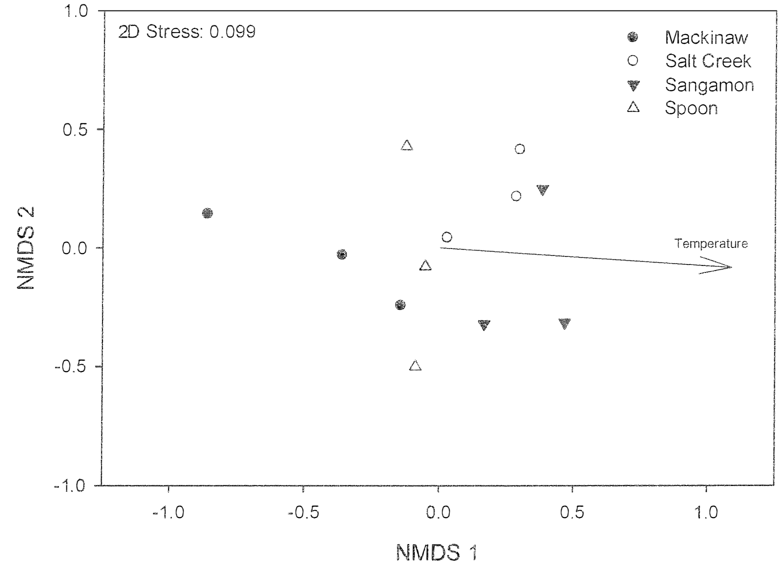

Figure 2. 7. Nonmetric dimensional scaling plot for average density of larval fish families excluding Asian Carp species sampled from the Mackinaw River (filled circles), Salt Creek (open circles), Sangamon River (filled triangles), and Spoon River (open triangles) during 2014. Environmental vectors (arrows) plotted in ordination space to show correlation with sites.

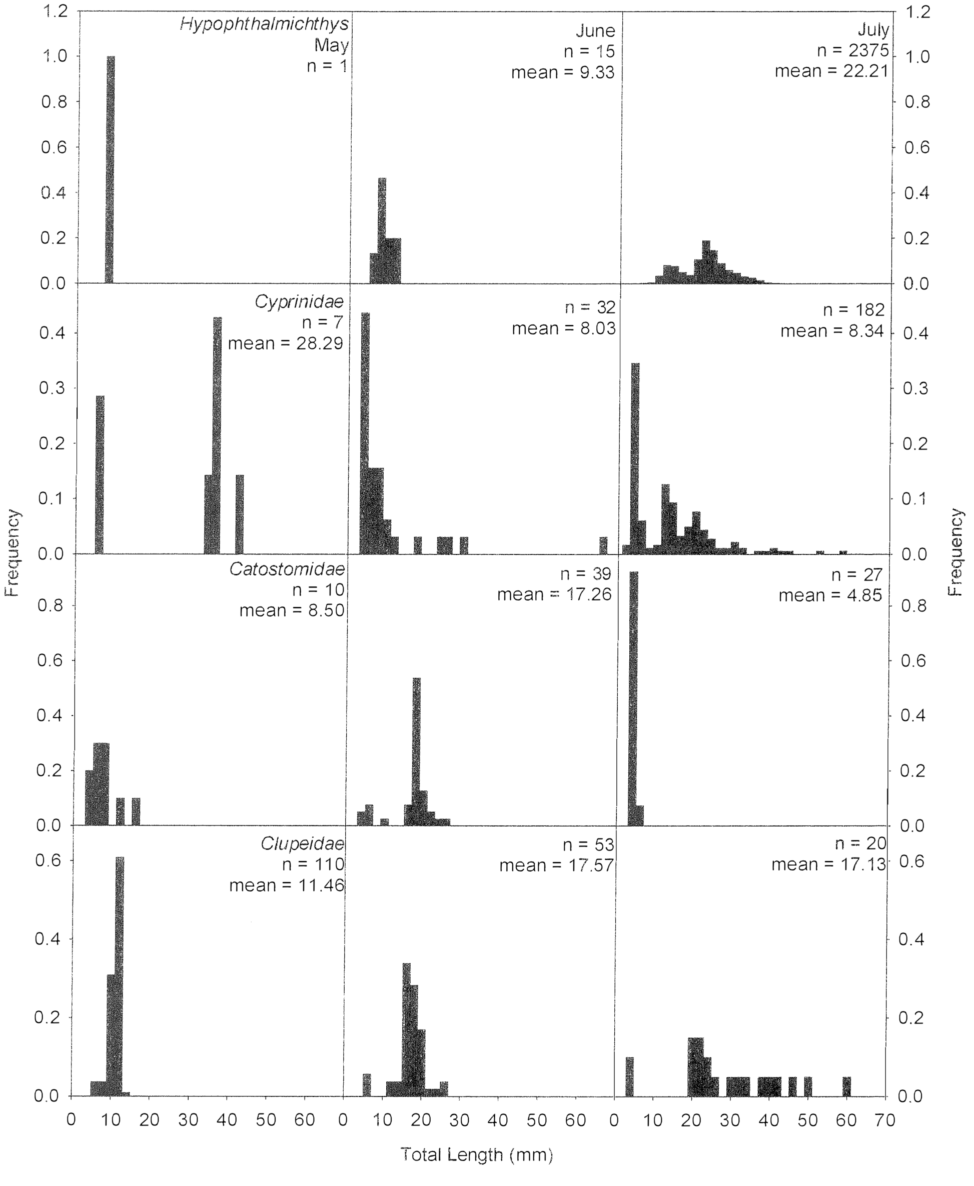

Figure 2. 8. Length-frequency histograms of total length for *Hypophthalmichthys* sp. (top), Cyprinids excluding *Hypophthalmichthys* (2nd from top), Catostomids (2nd from bottom), and Clupeids (bottom) sampled during May (left), June (middle), and July (right) during 2014.

CONCLUSIONS: PROBLEMS, IMPLICATIONS, AND FUTURE RESEARCH

By studying the tributaries of large floodplain rivers, we can expand the current knowledge base of these systems. Large rivers and the invasive species within are the focus of extensive research (Chick and Pegg 2001; Degrandchamp et al. 2008; Irons et al. 2007; Sampson et al. 2009; Stuck et al. 2015), however, associated tributaries remain largely neglected. Regulating the spread of invasive species is important, especially halting their progress into the Great Lakes, where species such as Asian Carp would cause great ecological and economic harm (Kolar et al. 2005). However, significant problems arise when studying any organism, and I will attempt to broach and remedy some issues associated with this study.

The biggest problem associated with this study is the lack of 0-2 age individuals in the sample. Many Asian Carp studies report a lack of these individuals as well, presenting gear bias, evasion, and absolute absence as the causes (Irons et al. 2011). I attempted to breach this dearth by using multiple gears (ichthyoplankton pushes and electrofishing) to sample multiple life stages, however, I was unsuccessful in sampling individuals between 40-400 mm. This size gap represents individuals age 0-2, absent from my sample. More exhaustive passive netting surveys, seines, or diel strategies may improve chances of encounter for 0-2 age fish (Irons et al. 2011), however, these strategies are often lacking in large river management. Additionally, Nico et al. (2005) reports early life stages of Asian Carps inhabiting shallow vegetated backwater habitats, areas typically not included in large river monitoring efforts. Successful recruitment of these size and age classes to gears will help ameliorate incomplete growth and mortality estimates, as well as provide more accurate demographic information, improving current management strategies. Poor recruitment to current sampling methods requires the development or improvement of gears and sampling strategies to successfully sample for these missing life stages.

Another weakness of this study was in the design of larval fish monitoring. I sampled larval fish using a boat-mounted active gear during the daytime, effective only when sites were accessible and I was actively sampling. This means diel patterns, temporal fluctuations, and shallow water stages were incompletely sampled in this study. Early life stages of Asian Carp have been noted to be nocturnally collected (Deters et al. 2012), a characteristic absent from consideration in this study. Additionally, the active nature of my design coupled with low residence times of tributaries, made it likely passively drifting larvae were incompletely sampled. For example, larval and juvenile Asian Carp sampled were all post-gas bladder emergent, meaning these individuals were free swimming larvae actively selecting habitats. Boat access also became problematic in this study, especially given the flashy nature of our sample rivers. Passive gears could address some of the issues with this study, allowing longer samples during less ideal conditions. More complete sampling design would increase sample size, further increasing the reliability of community assessment and chances of larval Asian Carp encounter.

Future studies of larval fish communities and Asian Carp populations in tributaries of large rivers should address movement, and habitat use and alteration by Asian Carp. Information on movement and habitat use of multiple life stages of Asian Carp would no doubt compliment not only this study, but the current knowledge base regarding their habits. Information regarding seasonal movements of Asian Carp exists (DeGrandchamp 2008), but seasonal habitat use, including main channel–extra channel exchanges would elucidate temporal movements. In this study we aim to quantify the abundance of larval Asian Carp in larval fish communities, however assessing impacts of high abundances of planktivorous Asian Carp in these habitats and their effects on plankton-dependent organisms would prove useful in determining their impacts on uninvaded systems. Asian Carp in the main channel of the Illinois River have been shown to impact native zooplankton communities, and this undoubtedly affects organisms feeding at these trophic levels (Sass et al. 2014). This information would reveal insight to managers regarding habitat use and influence, spawning locations, and refuge areas as locations to compliment and target management.

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APPENDIX: Supplementary Information

Table 1. Study site names and river kilometers (from confluence with main river) for study sites sampled for adult Asian Carp using pulsed-DC electrofishing on the Kankakee, Mackinaw, Spoon, and Sangamon Rivers and Salt Creek during 2013-2014.

River	Site	Latitude	Longitude	River kilometer
Kankakee	K1	41.35549	-88.20708	11.90
Kankakee	K3	41.23114	-88.15724	1.17
Sangamon	SG1	39.82933	-88.96075	236.54
Sangamon	SG3	40.064833	-90.151389	34.55
Spoon	SP1	40.713889	-90.26675	114.90
Spoon	SP3	40.307917	-90.081528	1.8
Mackinaw	M1	40.569972	-89.318778	82.17
Mackinaw	M3	40.545833	-89.729639	0.99
Salt Creek	SCR1	40.13884	-88.88793	139.94
Salt Creek	SCR3	40.13352	-89.735608	11.31

Table 2. Stages of Chinese major carps gonadal development for female and male individuals according to Li and Matthias (1994).

Description	Stage of Gonadal Development
Female	
Transparent, undeveloped blood vessels	F1
Semi-transparent, white, eggs hard to distinguish	F2
Ovaries wide, green-gray, branched blood vessels, visible eggs, overwintering stage	F3
Ovaries sac-like, green-gray, fully supplied with blood vessels, eggs plump and separable	F4
Spawning female. Eggs flowing freely in ovarian cavity, eggs flowing when abdomen gently pressed	F5
Post spawn. Blood vessels atrophy, ovaries stretched and purple-red	F6
Male	
Transparent, < 1 year of age	M1
Testes lacelike and semitransparent, age 2 individuals	M2
Testes rod-shaped and pink-yellow, hollow cavity in central lobule, reverts to this stage after exudation	M3
Testes greyish-white with wrinkled surface, blood vessels visible, male overwintering stage	M4
Spawning male. Testes plump and milky-white, milt flowing when abdomen gently pressed	M5
Post spawn. Volume of testes reduced, lobular cavity contains no or few sperms	M6