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Mark-Recapture and Fish Monitoring Activities in the Little Colorado River in Grand Canyon from 2000 to 2013

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U.S. Fish and Wildlife Service volunteer (David K. McDermitt-Van Haverbeke) checking a hoop net in Little Colorado River, October 2011. Photograph by David Van Haverbeke.

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Mark-Recapture and Fish Monitoring Studies in the Little Colorado River in Grand Canyon from 2000 to 2013

By David R. Van Haverbeke, Kirk Young, Dennis M. Stone, and Michael J. Pillow

Abstract

Since 2000, monitoring using hoop nets has been conducted in the lower 13.57 river kilometers of the Little Colorado River (LCR) to track abundance and trend of the endangered humpback chub (*Gila cypha*), and to monitor other native and nonnative fishes. These monitoring activities have occurred during the spring and fall seasons. Closed mark-recapture methods and catch per unit effort data have been used to estimate absolute or relative abundances of humpback chub of various size classes. Between 2000 and 2006, adult humpback chub (≥ 200 mm) maintained stable but low abundances of $< 3,000$ individuals during the spring and fall seasons. After 2006, abundances of adult humpback chub significantly increased. Since 2008 adult humpback chub have remained at higher abundance levels ($\sim 4,000$ to $7,000$ individuals). Abundances of sub-adult humpback chub (150-199 mm) during the spring, although more variable than adults, have also generally displayed higher abundance levels in the post-2006 timeframe and have maintained levels $\geq 1,000$ fish since 2007. Smaller size classes of humpback chub (< 150 mm) have displayed significant annual variation, thought to be related to the LCR hydrograph.

Bluehead sucker (*Catostomus discobolus*) and flannelmouth sucker (*C. latipinnis*) have been monitored using catch per unit effort. Relative catches of both native sucker species significantly increased during the post-2006 timeframe, but have since declined.

Reasons for the post-2006 increases of humpback chub, bluehead sucker, and flannelmouth sucker are thought to be related to several factors, including warmer mainstem Colorado River water temperatures since 2003, and mechanical removal of nonnative fish in the mainstem Colorado River during the 2003-2006 timeframe. Additionally, it is thought that translocations of humpback chub within the LCR watershed (upstream of Chute Falls) have contributed to increasing abundances. Translocated humpback chub have demonstrated an ability to grow and persist upstream of Chute Falls, with annual adult abundance estimates ranging from approximately 1 to 143 individuals. Many translocated humpback chub eventually vacate the area upstream of Chute Falls and return to

the lower portions of the LCR. Finally, the LCR continues to be dominated by native fish species that have comprised 91 percent of fish captures since 2000.

Introduction

With completion of the EIS on Operations of Glen Canyon Dam (USBR 1996), the Glen Canyon Dam Adaptive Management Program was initiated. The Adaptive Management Work Group (AMWG) within the program is responsible for defining management objectives associated with resources downstream from Glen Canyon Dam, and provides recommendations about development of long-term monitoring programs to assess those resources. The U.S. Geological Survey Grand Canyon Monitoring and Research Center (GCMRC) is responsible for developing and implementing long-term monitoring programs and assuring that they are fulfilling the needs of the AMWG. Assessing the status of the humpback chub (*Gila cypha*) is particularly important because of its status as a federally endangered species (U.S. Office of the Federal Register 32:48 [1967]:4001).

Because of the above needs, GCMRC and U.S. Fish and Wildlife Service (USFWS) initiated a program in fall 2000 to conduct long-term monitoring of humpback chub in the Little Colorado River (LCR). Between 2000 and 2013, USFWS obtained closed mark-recapture population abundance estimates of humpback chub ≥ 150 mm in the lower 13.57 river kilometers (rkm) of the LCR during the spring spawning season and during the fall season (Van Haverbeke 2010, Van Haverbeke et al. 2013). The spring mark-recapture effort is aimed to coincide with the peak of humpback chub spawning in the LCR and provides GCMRC with an annual estimate of the spring spawning abundance. The fall mark-recapture efforts provide data more representative of humpback chub overwintering in the LCR, particularly those in the juvenile and sub-adult life stages. Additionally, the fall mark-recapture efforts temporally expand marks and recaptures of humpback chub in the LCR, thereby strengthening the open Age-Structured Mark-Recapture (ASMR) models (Coggins et al. 2006, Coggins and Walters 2009).

In addition to tracking abundance and trend of sub-adult and adult humpback chub ≥ 150 mm, there is interest in tracking abundance and trend in smaller size classes of humpback chub < 150 mm. This interest largely stems from a desire to further understand recruitment dynamics of humpback chub (e.g., Coggins and Walters 2009), and to track abundances of smaller size classes of humpback chub in the LCR; a portion of which are annually removed for translocations (NPS 2013, Trammel et al. 2012) and for establishing a refuge of humpback chub at the Southwest Native Aquatic Research and Recovery Center (SNARRC).

Finally, in addition to the ongoing monitoring in the lower 13.57 rkm of the LCR, between 2006 and 2013 humpback chub monitoring has occurred upstream of rkm 13.57 in the LCR (Van Haverbeke 2010, Van Haverbeke et al. 2013, Stone et al. *in prep*). The purpose for this monitoring has been two-fold. First, this portion of the LCR

(rkm 13.57 to 17.7) became of interest after a series of translocations initiated in 2003 moved juvenile humpback chub from the lower reaches of the LCR (rkm 1.15 to 9.85) to upstream of a natural travertine structure called Chute Falls. Chute Falls is located at rkm 14.1 and translocated chub were released at rkm 16.2. Mark-recapture efforts upstream of Chute Falls provide annual population estimates of these translocated chub as they grow into adulthood. Second, there is a small reach of river (rkm 13.57 to 14.1) that is not included during our mark-recapture efforts downstream of rkm 13.57. This is a section of river in which chub naturally occur, and also a section of river that many of the translocated chub eventually appear to occupy as they disperse downstream.

Previous Investigations

Early studies on humpback chub in Grand Canyon began in the 1970s and focused on morphology, life history, and ecology (Suttkus and Clemmer 1977, Kaeding and Zimmerman 1983, Minckley 1996). In Grand Canyon, humpback chub are potadromous, with adults typically migrating from nearby areas in the Colorado River to the LCR to spawn during early spring (Douglas and Marsh 1996, Gorman and Stone 1999). Young humpback chub rear in the LCR and many remain until early adulthood (Douglas and Marsh 1996, Gorman and Stone 1999) unless they emigrate or are transported out of the LCR by seasonal flood events (Valdez and Ryel 1995). In addition to the LCR population, there are several smaller “aggregations” of humpback chub inhabiting the mainstem Colorado River in Grand Canyon (Valdez and Ryel 1995). With few exceptions, post-dam mainstem reproduction in other aggregations is absent (but see Valdez and Masslich 1999, Anderson et al. 2010), and exchange of individuals from these aggregations to the LCR is limited (Paukert et al. 2006).

Since emplacement of Glen Canyon Dam, the Colorado River throughout Grand Canyon has been predominately characterized by cold hypolimnetic release waters (Wright et al. 2009) that negatively affect incubation period, egg and larval survival, larval to juvenile transition time, and growth of humpback chub (Hamman 1982, Clarkson and Childs 2000, Robinson and Childs 2001). These factors are considered to have caused range contraction and decreases in abundances of humpback chub in Grand Canyon (Kaeding and Zimmerman 1983, Douglas and Marsh 1996, USFWS 2002). Because of the need to conserve and recover this relict species, several studies have focused on population abundance of the LCR population (Douglas and Marsh 1996, Coggins et al. 2006, Coggins and Walters 2009, Van Haverbeke et al. 2013).

Purpose and Objectives

The purpose of this report is to summarize humpback chub mark-recapture studies in the LCR from 2000 through 2013, evaluating the status and trends of humpback chub during those years, particularly those humpback chub in the sub-adult (150-199 mm) and adult (≥ 200 mm) size classes. Population variables evaluated for the study include closed population estimates of humpback chub during the spring and fall seasons, length frequency distributions, and external parasites. In addition, data is presented on relative abundance (catch per unit effort) of smaller size classes of humpback chub (< 150 mm), and flannelmouth sucker (*Catostomus latipinnis*) and bluehead sucker (*C. discobolus*) ≥ 150 mm. The specific objectives of this report are:

1. Present closed Chapman modified Peterson (Chapman Petersen) abundance estimates of humpback chub ≥ 150 mm and ≥ 200 mm in the lower 13.57 rkm of the LCR during the spring and fall seasons.
2. Present Chapman Petersen abundance estimates of age 0 humpback chub (40-99 mm) in the lower 13.57 rkm of the LCR during the fall season.
3. Present Chapman Petersen abundance estimates of humpback chub in the LCR between rkm 13.57 and 14.1 and between rkm 14.1 to 17.7 (Chute Falls reaches).
4. Collect data on Passive Integrated Transponder (PIT) tagged fish in support of the open ASMR models (e.g., Coggins et al. 2006, Coggins and Walters 2009).
5. Present additional information related to physical parameters of the LCR, length frequencies, species composition, and parasites.
6. Present 2013 annual data alongside previous data to provide data congruency.

For additional detailed information about monitoring activities that may or may not be included in this report, readers should consult Pillow (2013) concerning the spring 2013 mark-recapture trips; Stone (2013) concerning the 2013 Chute Falls monitoring trip; and Stone and Pillow (2013) concerning the fall 2013 mark-recapture trips. Stone and Pillow (2013) also cover details on the 2013 translocation of humpback chub to Chute Falls and to SNARRC.

Methods

Trips and Participating Personnel

Between September 2000 and October 2013, 54 field trips were conducted to perform 27 mark-recapture efforts to estimate abundance of humpback chub ≥ 150 mm and ≥ 200 mm in the lower 13.57 rkm of the LCR. A mark-recapture event occurred each spring (generally during April and May), and each fall (generally during September and October). In addition, between 2006 and 2013, twelve trips were conducted to estimate abundances of humpback chub upstream of rkm 13.57 in the LCR (Chute Falls reaches). These trips occurred during May and June. Personnel on the above mentioned trips included USFWS staff from the Arizona Fish and Wildlife Conservation Office, USFWS volunteers, and collaborative staff from GCMRC, Arizona Game and Fish Department, and Grand Canyon National Park.

During 2013, four trips were conducted: 17-26 April, 14-24 May, 13-20 September, and 29 October to 8 November. The 14-24 May trip included monitoring in the Chute Falls reaches, and the 29 October to 8 November trip included capture and transport of juvenile humpback chub to upstream of Chute Falls and to SNARRC.

Study Area

Work during the spring and fall mark-recapture trips in the lower 13.57 rkm of the LCR was conducted downstream of a large travertine structure called Lower Atomizer Falls, with the confluence of the LCR and the mainstem Colorado River designated as rkm 0 (Figure 1). During these trips, the LCR was divided into three contiguous ~ 4.5 km reaches (Boulders, Coyote, and Salt reaches). Each reach was divided into three subreaches (Table 1).

During the Chute Falls trips, the LCR upstream of rkm 13.57 was divided into a lower reach from the top of Lower Atomizer Falls (rkm 13.57) to the base of Chute Falls (rkm 14.1), and an upper reach from the top of Chute Falls (rkm 14.1) to rkm 17.7 (Figure 1, Table 1).

Gear and Effort

During spring and fall trips in the lower 13.57 rkm of the LCR, unbaited hoop nets (0.5 - 0.6 m diameter, 1.0 m length, 6 mm [1/4"] mesh, with a single 0.1 m throat) were used to sample fishes. During 2001 and 2002, nets were baited with Aquamax Grower 600 for Carnivorous Species (Purina Mills, Inc., Brentwood, Missouri). With few exceptions, ~ 540 net sets were deployed during each monitoring trip. Each net set consisted of a hoop net being deployed in the river for ~ 24 hours, after which it was checked for fish. This resulted in ~ 180 net sets being deployed per reach (i.e., Boulders, Coyote, and Salt reaches), or ~ 60 net sets being deployed per subreach. More

specifically, 20 nets were set for three 24-hour periods in each subreach, resulting in netting effort being uniform among sampling trips across years (Figure 2). Exceptions to the above were five trips during 2001 and 2002, when nets were set for four 24-hour periods rather than three. Minor variation also occurred because helicopter logistics occasionally resulted in slightly shortened trips (e.g., spring 2006, and fall 2011, 2013 mark trips). Between fall 2000 and fall 2013, a total of 29,458 hoop net sets were deployed during spring and fall trips combined yielding 691,371 hours of fishing effort. With exclusion of the five trips in 2001 and 2002, average hoop netting effort among spring trips was 12,620 hrs per trip (SE = 463), and among fall trips was 12,292 hrs per trip (SE = 1,118). During spring 2013 trips, 1,080 hoop net sets were deployed, yielding 25,332 hours of effort. During fall 2013 trips, 883 hoop net sets were deployed, yielding 20,377 hours of effort.

During the Chute Falls monitoring trips, hoop nets as described above were baited near their cod ends by attaching nylon mesh bags (30 x 30 cm, 6 mm mesh) filled with ~160 g AquaMax Grower 600 for Carnivorous Species (Purina Mills Inc., Brentwood, MO). This was done to increase fish captures in order to more closely track translocated fish. Typically, the reach between Lower Atomizer Falls and the base of Chute Falls was sampled with 17 hoop nets, and the reach upstream of Chute Falls to rkm 17.7 fished with 33 hoop nets, all of which were run for three 24-hr periods. During 2006-2009, the Chute Falls monitoring trips entailed both an annual marking and recapture trips, but thereafter (2010-2013) included only annual marking trips. Sampling effort (hours of hoop netting) was uniform across trips, both in the lower reach (Lower Atomizer to Chute Falls, rkm 13.57-14.1) and in the upper reach (Chute Falls to rkm 17.7, Figure 3). Average trip effort in the lower reach was 1,143 hours (SE = 62), and average trip effort in the upper reach was 2,357 hours (SE = 42). During 2013, 51 net sets were deployed in the lower reach yielding 1,096 hours of effort, and 99 net sets were deployed in the upper reach yielding 2,320 hours of effort.

On all LCR monitoring trips, nets were positioned in habitat suspected to catch humpback chub, and were frequently repositioned or moved if the catch was poor and if an alternative site was available. Nets were spread throughout each subreach uniformly within constraints of river hydrology and depth. Most nets were set near shore, but some were set further midstream if access allowed. Each net was checked and emptied of fish daily. All net locations on all sampling trips above were recorded as distance (rkm) upstream of the confluence.

Fish Handling and Data Collection

Data collected from fish included species, total and fork length (mm), and location (rkm). Speckled dace and non-native fish were generally measured only for total length. Humpback chub and other fish were visually

checked for the presence of the external copepod parasite (*Lernaea cyprinacea*), but the internal Asian fish tapeworm (*Bothriocephalus acheilognathi*) was not monitored. All fish lengths reported refer to total length.

Humpback chub, flannelmouth sucker, and bluehead sucker ≥ 150 mm were scanned for a Passive Integrated Transponder (PIT) tag (Biomark Inc., Boise, Idaho), and if lacking a tag were PIT tagged. From 2000 through 2002, 400 khz PIT tags were used. Thereafter, 134 khz PIT tags were used and fish containing a 400 khz tags were retagged with a 134 khz tag. From 2000 to 2002 and from 2009 onward humpback chub ≥ 100 mm were PIT tagged. From 2000 through spring 2003, most fish were weighed (g). Methods for collection of fish data followed the Standardized Methods for Grand Canyon Fisheries Research 2012 (Persons et al. 2013), or earlier draft versions of this document with the following additions:

1. Humpback chub ≥ 100 mm in the Chute Falls monitoring efforts were PIT tagged from 2006 onward.
2. From 2010 onward during September marking trips in the lower 13.57 rkm of the LCR, humpback chub 40-99 mm received a red Visible Implant Elastomer (VIE) tag, and humpback chub 40-99 mm during the October recapture trips received a blue VIE tag. Tagging humpback chub with blue VIE tags during October trips was performed in order to account for duplicate recaptures.

Water Quality

Measured water quality parameters for the spring and fall trips included a single point daily afternoon temperature reading ($^{\circ}\text{C}$, between 1200 to 1800 hrs) and turbidity readings (nephelometric turbidity units, NTUs) collected near Salt Camp (rkm 10.4). Generally, three or more turbidity readings were taken with a Hach 2100P Turbidimeter (Loveland, CO) and averaged. During Chute Falls trips, these measurements were taken at Translocation Camp (rkm 16.2). Provisional data (mean daily discharges in cubic feet per second; cfs) were downloaded (<http://nwis.waterdata.usgs.gov>) for USGS gage station 0940200 located on the LCR near Cameron, Arizona.

Closed Mark-Recapture

Between 2000 and 2013, closed Chapman Petersen mark-recapture efforts were conducted biannually (during the spring and fall) to estimate abundance of humpback chub ≥ 150 mm in the lower 13.57 rkm of the LCR. During 2000, no spring mark-recapture effort occurred. Like Douglas and Marsh (1996), our approach was to obtain closed abundance estimates of humpback chub ≥ 150 mm via fishing the lower 13.57 rkm of the LCR with hoop nets. However, our efforts only provide abundance estimates during the spring and fall seasons of each year rather than on a monthly basis as in Douglas and Marsh (1996). This is because Douglas and Marsh (1996)

conducted a monthly as opposed to a seasonal sampling regime. Nevertheless, within a given set of months, and within a given size class of fish (≥ 150 mm), our abundance estimates are comparable to Douglas and Marsh (1996).

The target population for determining abundance estimates was all humpback chub ≥ 150 mm, with the exceptions mentioned above (e.g., humpback chub ≥ 100 mm from 2010 onward, and humpback chub ≥ 40 mm during fall trips 2010 onward). We first examined our data to define the sampled population. Bernard and Hansen (1992) suggest setting the lower boundary of the sampled population equal to the length of the smallest fish recaptured. However, we allowed for growth and measurement error that could have occurred between the marking and recapture events (generally ~ 10 mm). We did not truncate the upper length end of our abundance estimates, because the types of hoop nets used in our study have been shown to capture large humpback chub.

The closed Chapman modified Petersen two-sample mark-recapture model (Seber 2002) was used to estimate abundance. Using the Chapman Peterson model requires only two trips (i.e., a marking trip and a recapture trip) to generate abundance estimates rather than three trips as open models require, and therefore required fewer expenditures and less handling of fish. In addition, data gathered to produce the Chapman Petersen estimates can be incorporated into open Age Structured Mark Recapture models (Coggins et al 2006, Coggins and Walters 2009). Assumptions necessary for unbiased estimates of abundance using the Chapman Petersen estimator are:

1. The population is closed with no additions or losses between marking and recapture events; either through recruitment, immigration, mortality, or emigration.
2. Marking does not affect capture probability during the recapture event.
3. All individuals in the target population have an equal probability of capture during the marking event or the recapture event; or marked individuals mix completely with unmarked individuals prior to the recapture event.
4. Marks (tags) are not lost between the mark and recapture events.
5. All marked individuals captured can be recognized from unmarked individuals.

The first assumption, addressing population closure, could potentially be violated in this system because humpback chub in the LCR have access to the mainstem Colorado River. This assumption has a higher probability of being violated during the spring than during the fall mark-recapture events. Humpback chub movement and migration is known to occur during the springtime (Kaeding and Zimmerman 1983, Douglas and Marsh 1996), but is thought to be much lower during the fall and winter months (Douglas and Marsh 1996). We minimized potential

for violating this assumption by allowing less than a month to elapse between mark and recapture events. It was also assumed that growth-related recruitment was minimal because of the short time span between the mark and recapture events. Finally, all fish captured during the mark-recapture efforts were handled with care and according to the standardized protocols (Persons et al. 2013) to avoid injury or stress related mortality.

If humpback chub emigrate from the LCR or die between sampling events, and both marked and unmarked fish are lost at a rate that is proportional to their abundance in the population, the Chapman Petersen estimator can still be used, but the population estimate will be germane for the population during the marking event. If humpback chub immigrate into the LCR between the two events, the population estimate will only be germane for the population during the recapture event. If both immigration and emigration occur between the events, there is no possible correction and the estimator will overestimate abundance. For further explanation about population estimation, and measures taken to minimize assumption violation during these studies, see Van Haverbeke et al. (2013).

Population estimation using capture probability

In certain instances, capture probability (p) data rather than mark-recapture was used to estimate abundances of fish. For example, from 2010 onward p data was used to estimate abundance of adult humpback chub for the Chute Falls monitoring efforts. This was because there were few humpback chub in the Chute Falls reaches after 2009 and numbers of expected recaptured fish may have been too low for mark-recapture to function properly. Population estimation was performed by dividing trip catch of adult humpback chub by the p values for humpback chub ≥ 200 mm derived from four mark-recapture events conducted between 2006-2009. The p values were derived by dividing the number of recaptured humpback chub by the total number of humpback chub captured in the recapture event, providing p values for the marking trips. Confidence intervals were constructed from the variance in the abundance estimates derived with the distribution of p values.

Catch per Unit Effort

Catch per unit effort (CPUE) was used to monitor relative trends in smaller size classes of humpback chub (<150 mm) and for flannelmouth and bluehead suckers ≥ 150 mm. This was because humpback chub <150 mm were not PIT tagged during all years of this study, and because in several years numbers of recaptured suckers were insufficient for population estimation. In the LCR, catch rates of these fish in hoop nets can be significantly affected by high turbidities resulting from flood events (Stone 2010). To minimize for this effect, we calculated CPUEs using only data gathered during the second spring trips (May or June) of each year, and only from October trips during the fall. During these trips, LCR conditions were generally at base flow and low turbidities. Because

nets were set very close to 24 hours each, all CPUEs in this report are expressed as number of fish captured per net set (Stone 2010).

Results –rkm 0-13.57

Physical Parameters

Temperature

Average afternoon water temperatures during monitoring trips in the lower 13.57 rkm generally ranged between 16 and 22 °C (Figure 4). Average trip water temperature during the spring mark-recapture trips (2001-2013) was 20.5 °C (SE = 2.5). Excluding the unusually cold fall of 2000, average trip water temperature during the fall mark-recapture trips (2001-2013) was 19.8 °C (SE = 1.6). For the spring 2013 trips, average water temperature was 21.1 °C (SE = 1.8), and for the fall 2013 trips was 20 °C (SE = 0.3).

Turbidity and Flow

Turbidities encountered during 2000-2013 fall mark-recapture events trips were generally much higher than those of 2001-2013 spring mark-recapture events in the lower 13.57 rkm (Figure 5). This is because the dramatic monsoonal flooding events during September generally cause far higher turbidities than the spring runoff flows of April. Average trip turbidities encountered during the spring ranged from 4 to 6,343 NTUs (mean = 589 NTUs, N = 26 trips). With the exception of 2005, turbidity was low on all spring recapture trips (4 to 45 NTUs, mean = 18 NTUs). Turbidity tended to be higher during spring mark trips because of snow run-off, and tended to be low during the spring recapture trips when the LCR generally returned to base flows (Figure 6). Average trip turbidities during the fall ranged from 5 to 75,956 NTUs (mean = 12,460 NTUs, N = 28 trips). Turbidity tended to be high during the fall mark trips because of monsoonal flooding, and low during the fall recapture trips when the LCR generally returned to base flows (Figure 6). For 2013, average daily turbidity during the spring trips was 200 NTUs (SE = 240), and for the fall trips was 31,302 NTUs (SE = 42,216).

Species Composition

Humpback chub have generally dominated hoop net catches in the lower 13.57 rkm of the LCR since fall 2000 (Figure 7). Bluehead sucker display a high variance in percent species composition because of highly variable annual catches. Native fish have comprised 91% of all fish captured since fall 2000. Fathead minnow

Pimephales promelas) were generally the dominant nonnative fish captured, comprising 7.2% of catch since 2000, but catches were variable and thought to be a result of seasonal flooding in the LCR and transport of these fish from upriver (Stone et al. 2007), or from extended periods of base flows during which local populations may expand. Other nonnative fish captured since 2000, listed in order of decreasing catch, included common carp (*Cyprinus carpio*), black bullhead (*Ameiurus melas*), red shiner (*Cyprinella lutrensis*), channel catfish (*Ictalurus punctatus*), plains killifish (*Fundulus zebrinus*), rainbow trout (*Oncorhynchus mykiss*), green sunfish (*Lepomis cyanellus*), and brown trout (*Salmo trutta*). A few fish thought to be razorback sucker (*Xyrauchen texanus*) and flannelmouth sucker hybrids have been captured since 2000, but are treated as flannelmouth sucker for this report. Such hybrids were infrequently captured during monitoring efforts in the early 1990s (Douglas and Marsh 1998). Presumably under-represented in hoop net catches were adult channel catfish and adult common carp, which seldom enter our nets but are seen by field crews and captured by angling. In 2013, humpback chub again dominated trip catches (62% of captures), followed by bluehead sucker (24%), speckled dace (9%) and flannelmouth sucker (4%). Native fish comprised 98.7% of fish captures.

Length Frequency Distributions

Humpback Chub

Humpback chub in the lower 13.57 rkm of the LCR show a widely distributed population structure, with all size classes represented and recruitment of young fish apparent (Figure 8-A). Cumulatively since 2001, the spring season shows sizeable production of age 1 fish in the roughly 80-130 mm size class. Some smaller humpback chub can also be seen during the spring, representing the oncoming age 0 cohorts born each spring. This smaller group of humpback chub may be under-represented since fish <40-50 mm escape the hoop nets. Humpback chub in the 130-300 mm size class also show good representation, indicating multiple but blurred age classes. Most of these fish are likely in the 2-15 year age classes (see Figure 4 in Coggins and Pine 2010). Finally, above 300 mm representation noticeably declines, with some individuals reaching maximum lengths of ~470-480 mm. Most humpback chub above 300 mm are likely at least 15 years of age, but some of the largest ones could extend to 30+ years of age. Since 2001, 3,204 humpback chub >300 mm have been captured in the LCR during the spring spawning season.

During the fall in the lower 13.57 rkm of the LCR, there is also indication of a healthy population structure for humpback chub, with multiple size class representation (Figure 8-B). Cumulatively since 2000, there has been sizeable production of age 0 humpback chub. These fish hatch during the spring seasons, and most are still <100

mm by fall (Figure 8-B). There is another sizeable mode of fish centered at ~145-150 mm. This mode would largely represent the age 1 cohort that were in the roughly 80-130 mm size class the previous spring (Figure 8-A), although the mode blends into older fish. Noticeably fewer fish are captured in the ~200-300 mm size class during the fall compared to spring, since many of these fish presumably vacate the LCR after the spring spawning season. Finally, captures of the largest of humpback chub >300 mm are noticeably fewer during the fall season, with 677 captured since 2000.

Bluehead sucker

Bluehead sucker also show signs of a healthy, long-term population structure in the lower 13.57 rkm of the LCR, with adult fish present as well as recruiting young fish (Figure 9-A). Cumulatively from 2001-2012, the spring season displays a sizeable group of presumed age 0 fish (mode at 65-70 mm), and another group of sub-adult/adult fish (mode ~230 mm). By fall, both size groups remain, but the mode for small fish is somewhat larger (100 mm; Figure 9-B).

Flannelmouth Sucker

Cumulatively (2001-2012), flannelmouth sucker show distinct size class patterns during the spring (Figure 10-A), with representatives from the age 0 (<80 mm) and 1 cohorts (~80-150 mm) being most distinguishable (i.e., older cohorts tend to blur). Like bluehead suckers, the spring age 0 size class of flannelmouth suckers is more pronounced compared to humpback chub, possibly because they spawn earlier than chub. By fall, the age 0 cohorts of both sucker species (Figures 9-B and 10-B) tend to be very diminished compared to humpback chub (Figure 8-B).

Parasites

Compared to fall trips, percent occurrence of the external parasite (*Lernaea cyprinacea*) on humpback chub during spring trips was generally lower, with infestation rate representing the percent of chub captured observed carrying the parasite (Figure 11). Infestation rates during spring trips averaged 2.2% (range 0.1 to 8%), while those on fall trips averaged 9.2% (range 0.1% to 35%). The highest infestation rate was fall 2009 at 35%. Typically, only one or two parasites are seen on any individual humpback chub in the LCR, although numbers are sometimes higher. Very infrequently, this parasite is seen on other species of native and nonnative fish (e.g., flannelmouth, bluehead, dace, fathead minnows), again typically at low frequencies with only one or two parasites per fish.

Population Abundance Estimation (rkm 0-13.57)

Adult and sub-adult humpback chub

Following the decline in sub-adult (150-199) and adult (≥ 200 mm) humpback chub abundance documented in the early-mid 90s (Coggins et al. 2006), abundance in the LCR remained at reduced but stable levels during the early-mid 2000s (Figure 12). Following this was a post-2006 period in which the abundances of humpback chub in the LCR during the spring and fall seasons significantly increased. Since this increase, the abundances of humpback chub ≥ 150 mm and ≥ 200 mm have remained at elevated levels (Figure 12). For spring 2013, the abundance estimate of fish ≥ 150 mm was 8,549 (SE = 757), of which an estimated 5,734 (SE = 512) were ≥ 200 mm. For fall 2013, the abundance estimate of humpback chub ≥ 150 mm was 4,946 (SE = 1,141), of which an estimated 3,022 (SE = 1,240) were ≥ 200 mm. The 95% confidence intervals around the fall estimate of fish ≥ 200 mm were quite large in 2013 because of abnormally few recaptures (6 fish) in this size class.

Since 2007, the spring abundance estimates for humpback chub in the 150-199 mm size class have all been $>1,000$ fish (Figure 13). The mean spring point estimate for humpback chub in the sub-adult category (150-199 mm) from 2007-2013 is 1,829 fish. The spring abundance estimates for fish in this size class are important for reasons mentioned in the Discussion. For spring 2013, the abundance estimate for humpback chub in the 150-199 mm size class was 1,583 (SE = 178).

Juvenile (age 1 and age 0) humpback chub

Mark-recapture estimates of humpback chub in the age 1 size class (100-149 mm) during the spring season were conducted in 2001, 2002 and 2009-2013, and have ranged between 762 and 11,619 fish (Figure 14-A). For spring 2013, the abundance estimate for humpback chub in this size class was 4,714 (SE = 714). CPUE is used in all years (2001-2013) to provide trend, and ranged between 0.14 and 3.91 fish per 24-hour net set (mean = 1.82) during the spring recapture trips (Figure 14-A). For spring 2013, CPUE for age 1 humpback chub was 2.41 fish per net set during the recapture trip. Several years (2001, 2003, 2005, 2007 and 2010) can be characterized as low relative abundance years. Four of these years (2001, 2003, 2007 and 2010) correspond to the low age 0 CPUEs of the previous fall (Figure 14-B). The age 1 mark-recapture estimates show good correlation with the age 1 CPUE calculations ($r^2 = 0.85$, $N = 7$ years).

Annual mark-recapture efforts of humpback chub in the age 0 size class (40-99 mm) in the LCR began in fall 2010 and have continued through fall 2013. As mentioned in the Methods, VIE tagging has been utilized to estimate the size of the age 0 humpback chub cohort. Between fall 2010 and fall 2013, abundance estimates of

the fall age 0 cohort ranged between 3,479 and 10,511 fish (Figure 14-B). For 2013, we obtained an estimate of 3,479 (SE = 997) humpback chub in this size class. Relative abundance (catch per unit effort, CPUE) has also been used to show patterns of the fall age 0 humpback chub cohort, and has ranged between 0.05 and 2.93 fish per 24-hour net set (mean = 1.46) during fall October trips (Figure 14-B). For fall 2013, CPUE for age 0 humpback chub was 1.09 fish per net set during the recapture trip. Unfortunately, the age 0 mark-recapture estimates show poor correlation with the age 0 CPUE calculations ($r^2 = 0.004$).

Bluehead and flannelmouth sucker

Bluehead sucker and flannelmouth sucker spring trend was gaged by use of CPUE data (Figure 15). Similar to humpback chub, bluehead and flannelmouth suckers both underwent a post-2006 period of significant increase, although this increase was noticeably less for flannelmouth sucker. From 2009-2011, bluehead sucker then significantly declined, and increased again in 2012 and 2013. Flannelmouth sucker have also been trending downward somewhat since 2010 (Figure 15). For spring 2013, CPUE for bluehead sucker was 2.93 (SE = 0.55), and for flannelmouth sucker was 0.28 (SE = 0.03).

Results Chute Falls (rkm 13.57 to 17.7)

Physical Parameters

Temperature

Average water temperatures during the 2006-2013 Chute Falls monitoring trips were higher and less variable (22.1 °C, SE = 1.0) than downstream of 13.57 km (Figure 4). This was likely because the Chute Falls trips were conducted during the warmer months of May and June, and because the Chute Falls reaches are closer to Blue Springs with a discharge temperature of ~20.5 °C (pers. com., D. Stone). For the 2013 trip, average water temperature in the Chute Falls reach was 20.7 °C (SE = 0.3).

Turbidity and Flow

Average trip turbidities during the 2006-2013 June Chute Falls monitoring trips were consistently low, ranging from 0.9-6.9 NTUs (mean = 2.9 NTUs, N = 12 trips), and were accompanied by base flows. For the 2013 trip, average turbidity in the Chute Falls reach was 2.2 NTUs (SE = 0.8).

Length Frequency Distributions and Catch

Length frequencies demonstrate that humpback chub translocated to upstream of Chute Falls can remain and grow to adulthood (Figure 16-A). Most fish were ~80-130 mm at the time of their translocation. Immediately downstream of Chute Falls, a sizeable number of humpback chub fall into the approximately 110-160 mm size class (Figure 16-B), however the origin of these fish remains uncertain. It is hypothesized that these fish represent small humpback chub that vacated habitat upstream of Chute Falls shortly after translocation, or that there is spawning of humpback chub in the reach of river immediately downstream of Chute Falls. Interestingly, the long term mode (2006-2012) of small humpback chub during May/June immediately downstream of Chute Falls is ~140 mm (Figure 16-B), while the long term mode (2001-2012) of small fish downstream of rkm 13.57 during May/June is ~110 mm (Figure 8-A), suggesting that growth of small humpback chub is higher upstream of lower Atomizer Falls (above rkm 13.57).

Parasites

Similar to spring trips in the lower 13.57 rkm, infestation rates of *Lernaea cyprinacea* on humpback chub in the Chute Falls reaches tended to be low, ranging from 0% to 6% (mean = 1%). The highest infestation rate was 6% in 2009. During 2013, infestation rate was 0.13%, with usually only 1 parasite per infected fish.

Population Abundance Estimation

Recall that mark-recapture was only performed in the Chute Falls reaches from 2006-2009, and that from 2010-2013 capture probability (p) data derived from these mark-recapture efforts was used to estimate abundance (Figures 17-A and 17-B). Upstream of Chute Falls was a period of decline in adult humpback chub ≥ 200 mm (2006-2008) that was accompanied by no translocations occurring in 2006 and 2007 (Figure 17-A). Translocations resumed again in 2008 and this was followed by a higher abundance of adults by 2009. A prolonged spring flood in 2010 (Figure 6) is thought to have resulted in a near absence of humpback chub upstream of rkm 13.57 during summer of 2010 (Figures 17-A and 17-B). Translocations continued during 2010-2012, again resulting in an increase in abundance of adult humpback chub upstream of Chute Falls through 2013 (Figure 17-A). In November 2013, another 303 humpback chub were translocated to upstream of Chute Falls (Stone and Pillow 2013). Importantly, the abundance estimates calculated with mark-recapture (2006-2009) and those calculated using p data for 2006-2009 were highly correlated ($r^2 = 0.93$), and did not differ significantly during any given year (see Figures 17-A and 17-B), lending credibility to the post 2010 abundance estimates derived using p data. For 2013, it

was estimated that there were 99 humpback chub ≥ 154 mm (SE = 22) in the upper reach, and 280 ≥ 143 mm (SE = 52) in the lower reach. Of these, it was estimated that there were 62 humpback chub ≥ 200 mm (SE = 12) in the upper reach, and 218 ≥ 200 mm in the lower reach (SE = 38).

Discussion

Lower 13.57 rkm of the LCR

Population estimates of humpback chub indicate that sometime between the early 1990s and 2000, the abundance of adult humpback chub (≥ 200 mm) underwent a decline in the LCR (Coggins et al. 2006). This was followed by a period of relatively low but stable abundance between 2000 and 2006, and by a post-2006 period of significant increasing trend (Van Haverbeke et al. 2013). This trend is also reflected in humpback chub ≥ 150 mm, largely because fish ≥ 200 mm generally comprise a large portion of the population ≥ 150 mm (mean = 68% during 26 spring trips since 2001, SE = 12). The post-2006 increases in humpback chub ≥ 150 mm are visible during both the spring and fall seasons, but are more apparent during spring months (Figures 12-A and 12-B). The significant decline in the 2011 spring abundance of humpback chub ≥ 150 mm (Figure 12-A) may be a result of relative low production of age 0 fish during 2009 as suggested by Figure 14-B.

Regarding the spring annual abundance estimates of humpback chub strictly within the 150-199 mm size class (Figure 13), there is more annual variation than in viewing the more inclusive set of fish ≥ 150 mm (Figure 12-A). This should be expected, since the former is a narrower and smaller size class. Since 2008 there have been only two years (2008 and 2011) in which the lower 95% confidence interval of the spring abundance estimate dropped below 910 fish (Figure 13). This is important for two reasons. First, the Biological Opinion associated with mechanical removal of trout (USFWS 2011) implemented a set of triggers under which mechanical removal of trout would resume. One of the measures listed that could help trigger mechanical removal was if the lower 95% confidence interval of humpback chub in the 150-199 mm size category drops below 910 fish in any 3 of 5 years extending retrospectively since 2008. Clearly, our results indicate this has not been the case. Second, to help meet the recovery criteria for humpback chub, the Recovery Goals (USFWS 2002) called for the mean estimated recruitment of age 3 (150–199 mm TL) naturally produced fish to equal or exceed mean annual adult mortality. Mean adult mortality rate has been estimated at 0.13 and the adult population size has been estimated at $\sim 7,650$ individuals (Coggins and Walters 2009). Using these numbers as a base guideline, this translates into 995 sub-adult (150-199 mm) humpback chub needed annually to equal or exceed adult mortality. Our results indicate that this guideline is being met, with all spring point population estimates for humpback chub in the sub-adult category

(150-199 mm) since 2007 being $\geq 1,019$ fish and the average of these spring point population estimates since 2007 being 1,829 fish (SE = 663).

Concerning juvenile humpback chub <150 mm, there is high annual variation in size of cohorts. Both mark-recapture and CPUE indices suggest high annual variability in abundance of the fall 40-99 mm size class (age 0) and the spring 100-149 mm (largely age 1) size class of humpback chub (Figures 14-B and 14-A, respectively). Since 2010, sizes of the fall age 0 cohort (40-99 mm) have ranged from 3,479 to 10,511 fish. As mentioned in the Introduction, one reason for estimating this parameter is to gain insight into what percentage of the age 0 cohort is being harvested (cropped) each year for the Shinumo and Havasu creek translocations and for the establishment of a refuge at SNARRC. Humpback chub cropped for Chute Falls translocations are generally between 80-130 mm, and those for other translocations outside of the LCR and for SNARRC <100 mm. Humpback chub are cropped from the Boulders and Coyote reaches, since these reaches typically have much higher catches for these size classes of fish than the Salt reach (e.g., Van Haverbeke et al. 2011). Since 2010, when mark-recapture efforts began to track abundance of humpback chub <100 mm during the fall, it is estimated that on average we have been cropping ~10% of the age 0 cohort for translocations outside of the LCR and for SNARRC. This would seem to be an acceptable level according to the population viability modeling of Pine et al. (2013), however, we caution that population estimation using VIE methodology may not be as reliable or accurate as we would prefer, as suggested by the poor correlation between mark-recapture estimates and CPUEs (Figure 14-B). Alternatively, this lack of good correlation could be an effect of disparate turbidities on age 0 catch during the October trips of 2010-2013 (range of mean trip turbidities = 24 to 913 NTUs).

In addition to sub-adult and adult humpback chub abundance increases, CPUE of bluehead sucker and flannelmouth sucker ≥ 150 mm significantly increased in the post-2006 timeframe during the spring spawning season (Figure 15). Unlike humpback chub, the two sucker species since about 2009 have undergone significant declines in abundances. Interestingly, bluehead sucker catches have been about 10-fold lower during the fall season, suggesting mass emigration for this species to the mainstem. Alternatively, they could be seeking refuge in shallow fast water habitat during the off spawning season where hoop nets are not usually deployed. Finally, because few age 0 suckers are captured in the fall season, this may indicate that the age 0 cohorts for both sucker species tend to drift as larvae and emigrate from the LCR before the onset of the fall season (see Robinson et al. 1998).

Chute Falls (rkm 13.57-17.7)

Since 2003, 2,361 juvenile humpback chub have been translocated from the lower reaches of the LCR to upstream of Chute Falls. Since translocation activities commenced in 2003, humpback chub have consistently been found to grow to adulthood in the upper reach, although not all translocated fish have stayed upstream of Chute Falls. Many have either moved into the lower reach, or have moved further downriver (downstream of Lower Atomizer Falls). Some have even been detected in the mainstem. Sometime between summer of 2009 and summer 2010, nearly all humpback chub vacated both the upper and lower Chute Falls reaches (Figure 17), as well as the upper portion of Salt reach (12.3 to 13.57 km). It is thought that this was a result of a prolonged spring flood during 2010 that deposited a heavy sand load in the upper reaches of the LCR, filling in much of the available deep pool habitat. This phenomenon may well explain why there were not humpback chub upstream of Chute Falls prior to translocations. With the commencement of translocation activities again in 2010, humpback chub have again been found to remain and grow to adulthood in the upper and lower reaches of Chute Falls (Figure 17). The post 2010 increase in adult abundance immediately downstream of Chute Falls (Figure 17-B) may be largely a result of translocation efforts, plus natural immigration from downriver. It is beyond the scope of this document to discuss all aspects of the Chute Falls translocation and monitoring activities, which will be presented in Stone et al. (*in prep*).

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Table 1. List of reaches, subreaches, and river kilometers within each subreach; Little Colorado River.

Sub-reach	Reach	River km (range)
Boulders		
Confluence-Jump Off Rock		0.0-1.8
Jump Off Rock-Powell Pool		1.8-3.0
Powell Pool-5.0 rkm		3.0-5.0
Coyote		
5.0 rkm - White Spot (Kachina Falls)		5.0-6.5
White Spot-Redbud Canyon		6.5-8.0
Redbud Canyon-House Rock		8.0-9.6
Salt		
House Rock-Hell Hole		9.6-11.2
Hell Hole-Triple Drop		11.2-12.3
Triple Drop-Lower Atomizer Falls		12.3-13.57
Chute Falls		
Lower Atomizer Falls-Chute Falls		13.57-14.1
Chute Falls-18 rkm		14.1-17.7

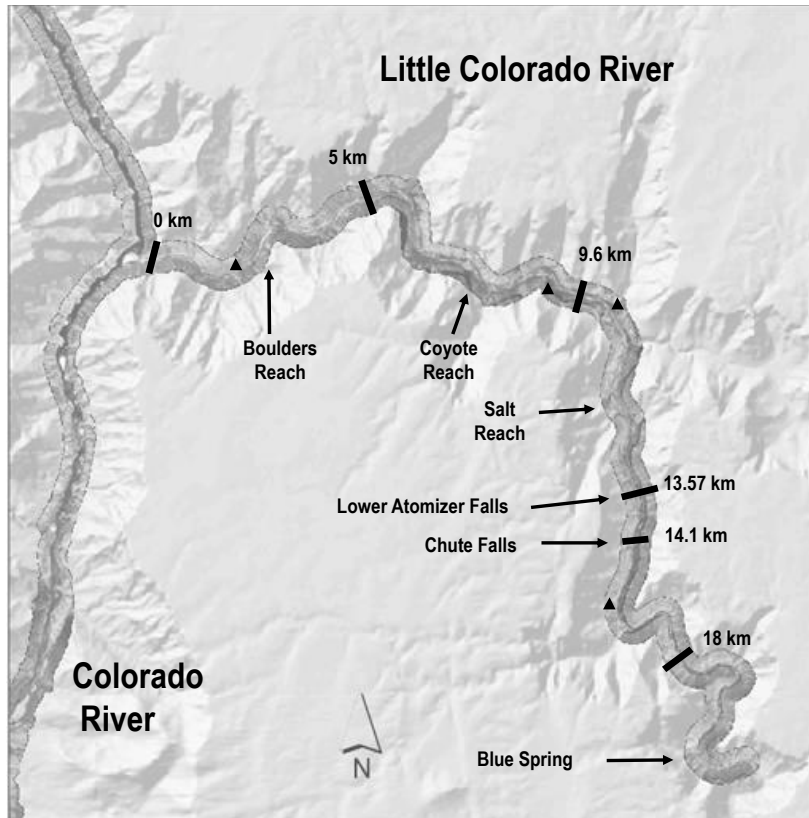


Figure 1. Map of study areas, showing Boulders (river km [rkm] 0 to 5), Coyote (rkm 5 to 9.6) and Salt (rkm 9.6 to 13.57) reaches, and lower Chute Falls (rkm 13.57 to 14.1) and upper Chute Falls (rkm 14.1 to 18) reaches; Little Colorado River. Camps are designated by triangles.

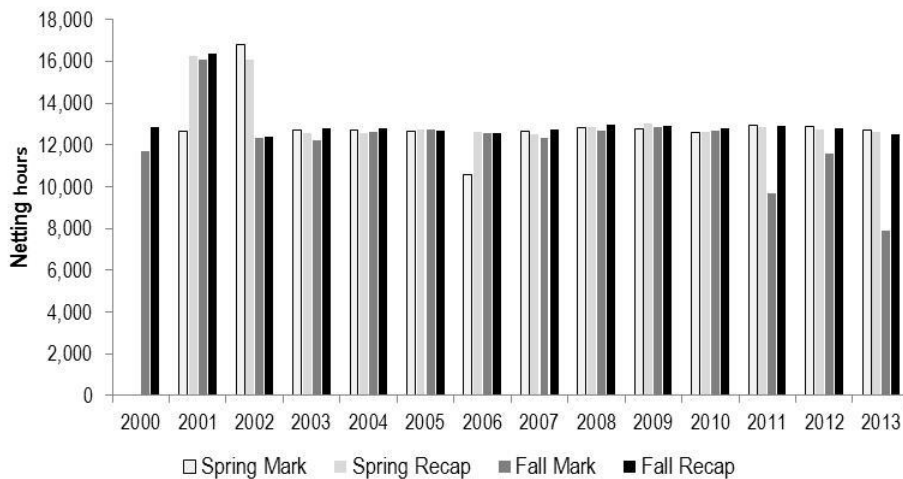


Figure 2. Hoop net sampling effort (hours) across all spring and fall monitoring trips 2000 to 2013, Little Colorado River.

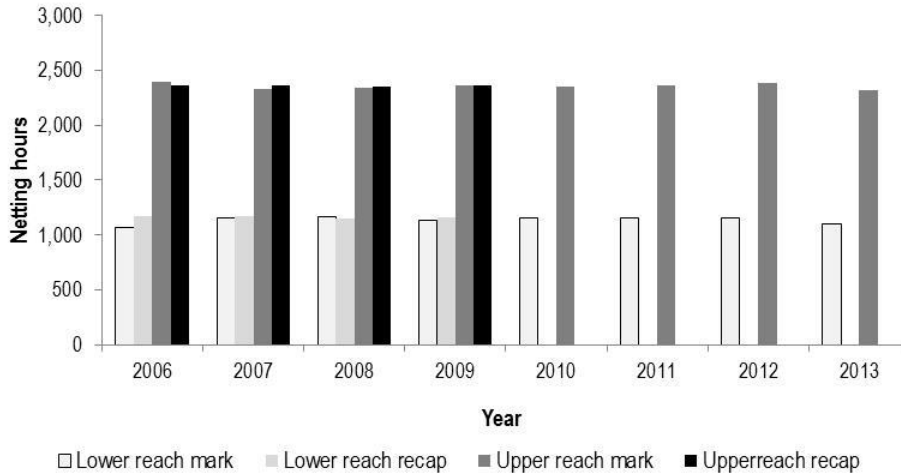


Figure 3. Hoop netting effort (hours) in the upper and lower Chute Falls reaches during monitoring efforts between 2006 and 2013, Little Colorado River. Note: Lower reach extended from Lower Atomizer Falls (river km [rkm] 13.57) to Chute Falls (rkm 14.1). Upper reach extended from top of Chute Falls (rkm 14.1) to rkm 17.7. Both reaches included a mark and recapture event from 2006-2009, but thereafter were inclusive of only one marking trip per year.

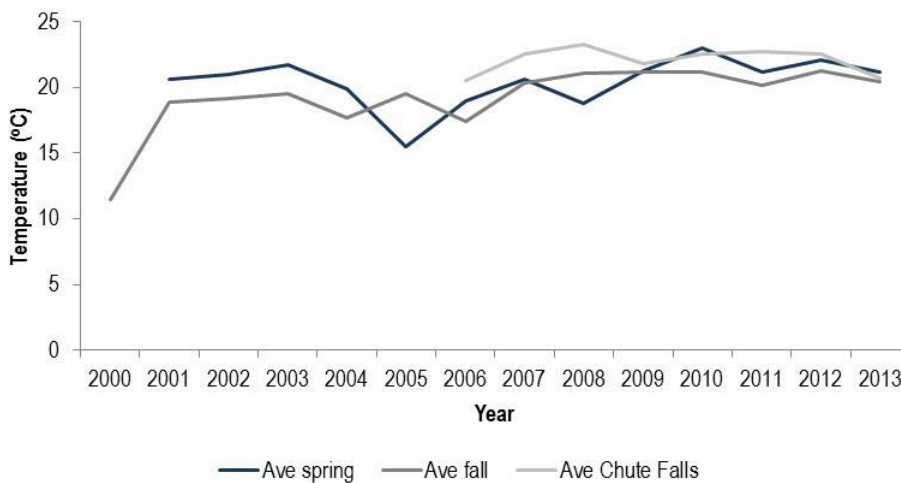


Figure 4. Average trip water temperatures collected in the Little Colorado River between 2000 and 2013.

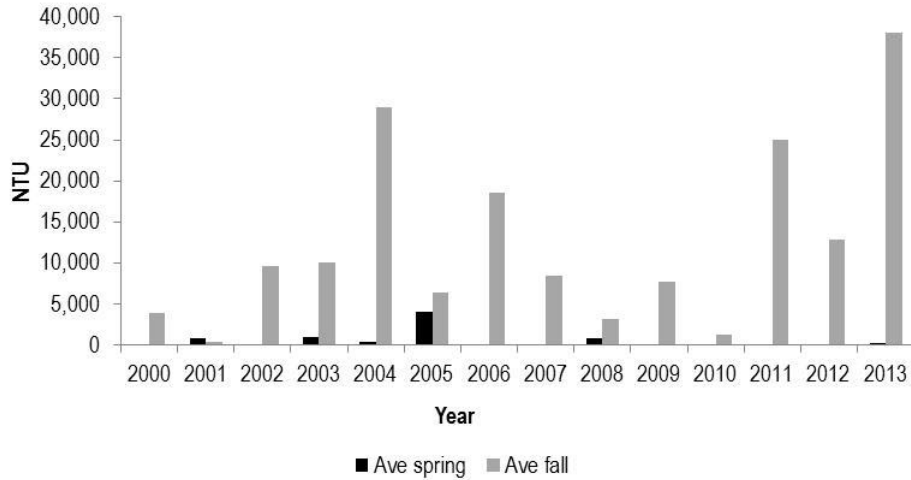


Figure 5. Average turbidities (Nephelometric Turbidity Units, NTU) during spring and fall mark-recapture events in the Little Colorado River, 2000-2013.

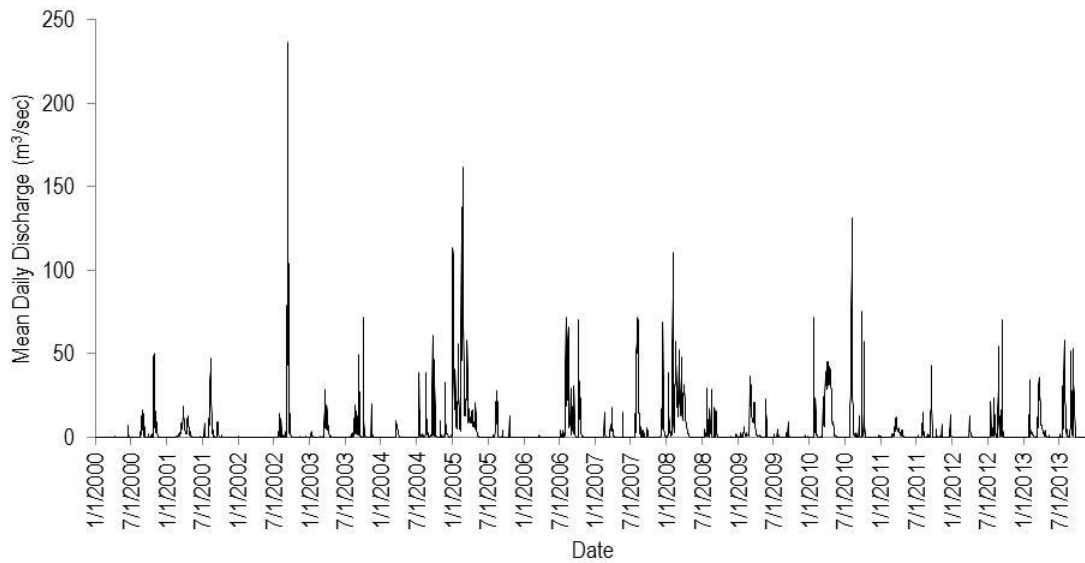


Figure 6. Provisional mean daily discharge (cubic feet/second; cfs) from USGS gage station 0940200; Little Colorado River (LCR), Arizona, 2002-2013.

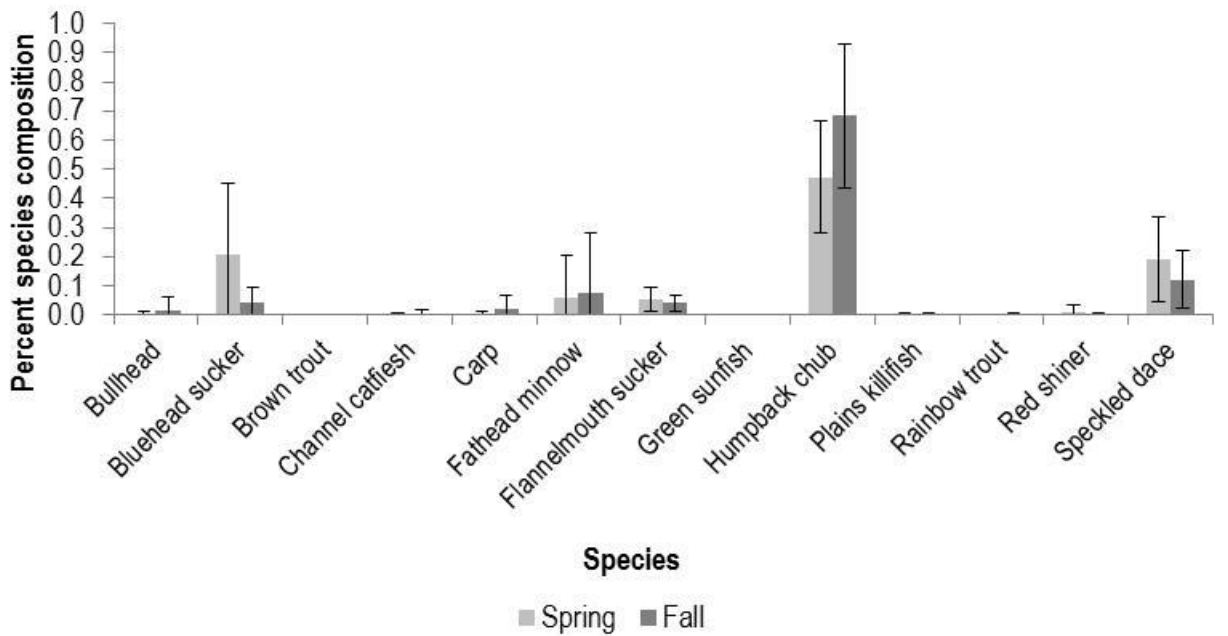
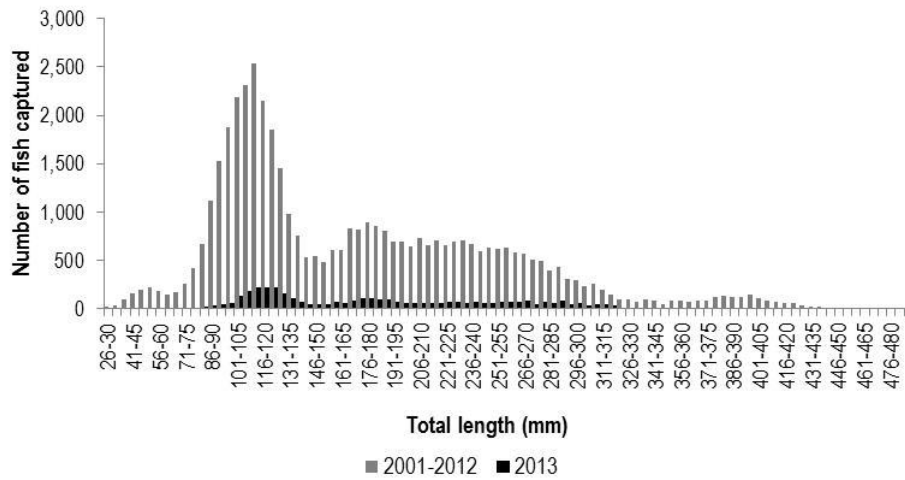


Figure 7. Spring and fall species composition of fish captured in hoop nets between fall 2000 and fall 2013; Little Colorado River (N = 160,556 fish).

A.



B.

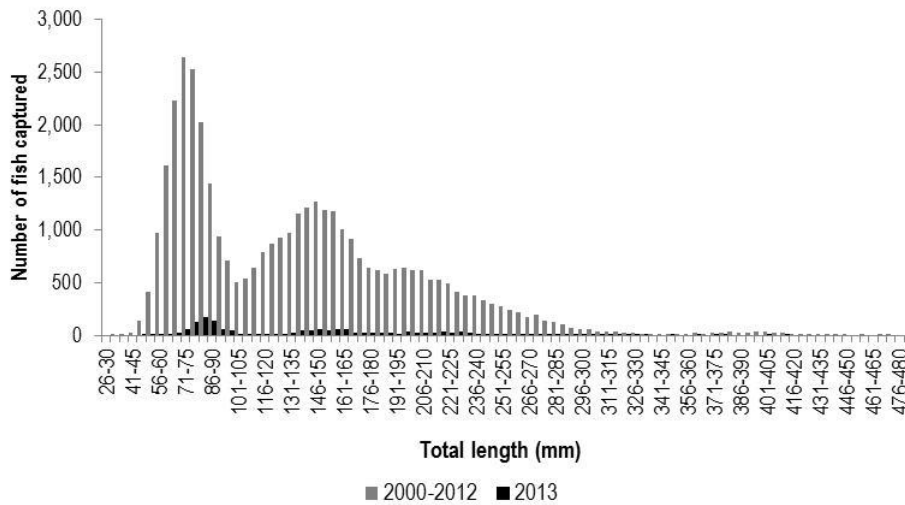
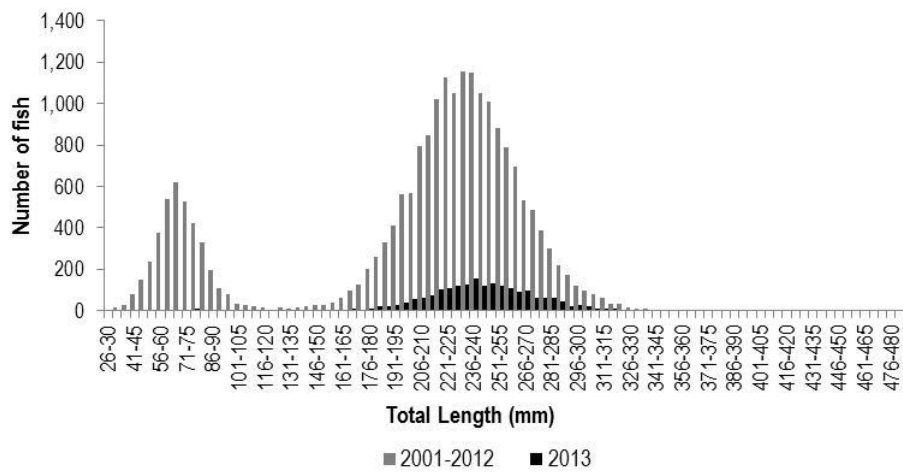


Figure 8. Length frequency distributions of humpback chub captured in the Little Colorado River (river km 0-13.57) during (A) spring 2001-2012 and 2013 and (B) fall 2000-2012 and 2013.

A.



B.

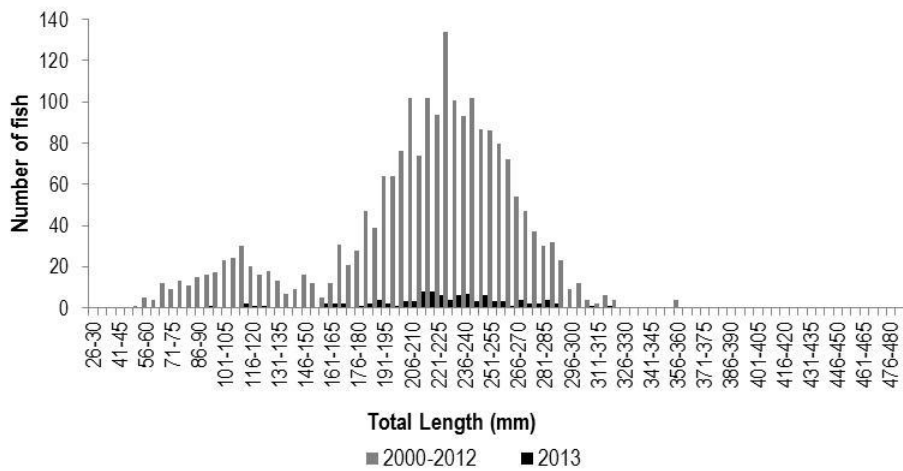
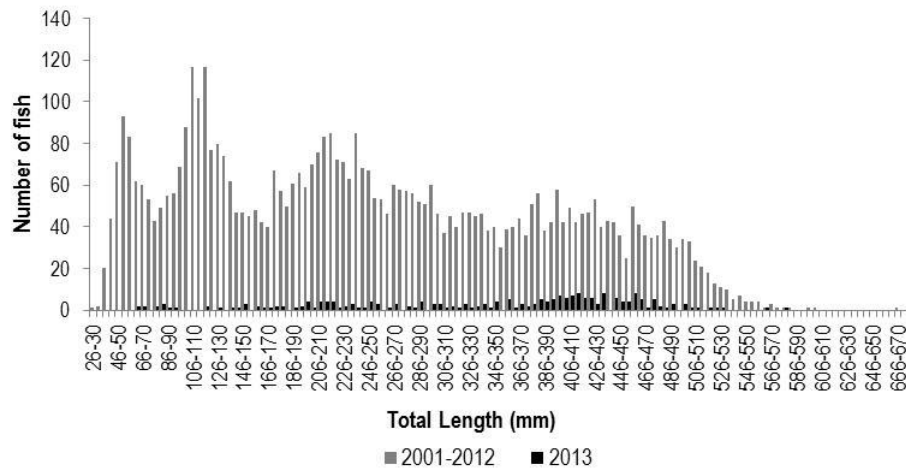


Figure 9. Length frequency distributions of bluehead suckers captured in the Little Colorado River (river km 0-13.57) during (A) spring 2001-2012 and 2013 and (B) fall 2000-2012 and 2013.

A.



B.

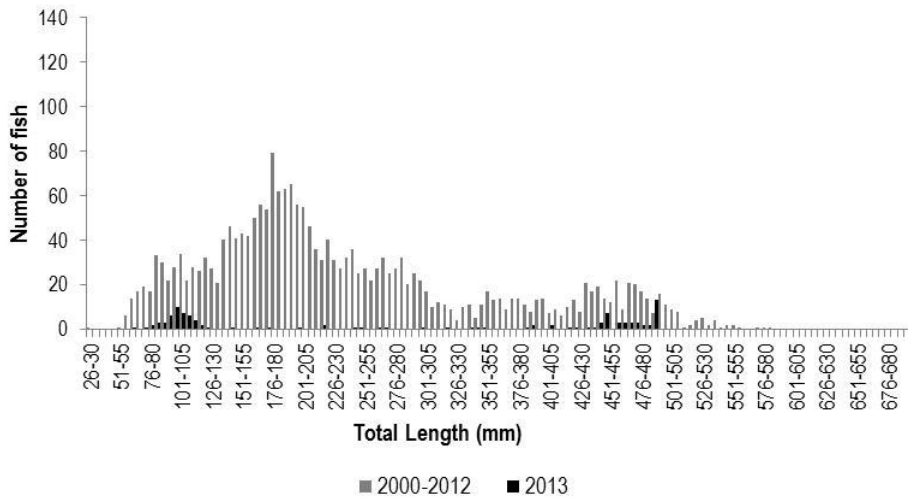


Figure 10. Length frequency distributions of flannelmouth suckers captured in the Little Colorado River (river km 0-13.57) during (A) spring 2001-2012 and 2013 and (B) fall 2000-2012 and 2013.

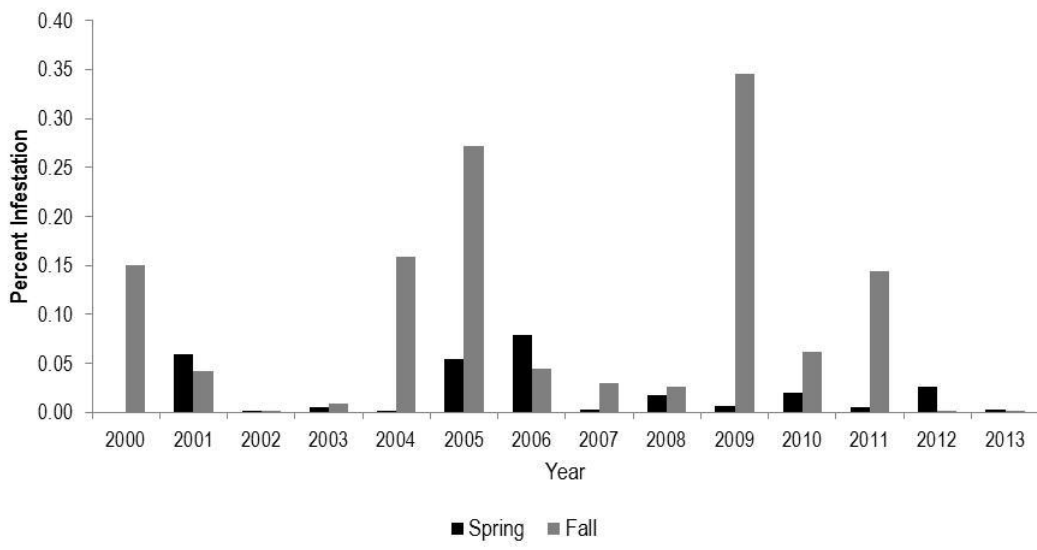
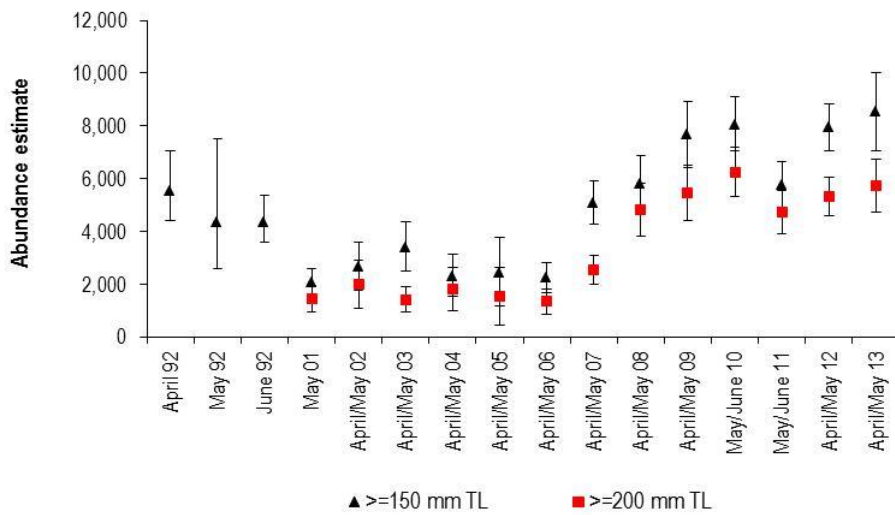


Figure 11. Percent occurrence of the external copepod parasite (*Lernaea cyprinacea*) on humpback chub in the Little Colorado River (river km 0-13.57), 2000-2013.

A.



B.

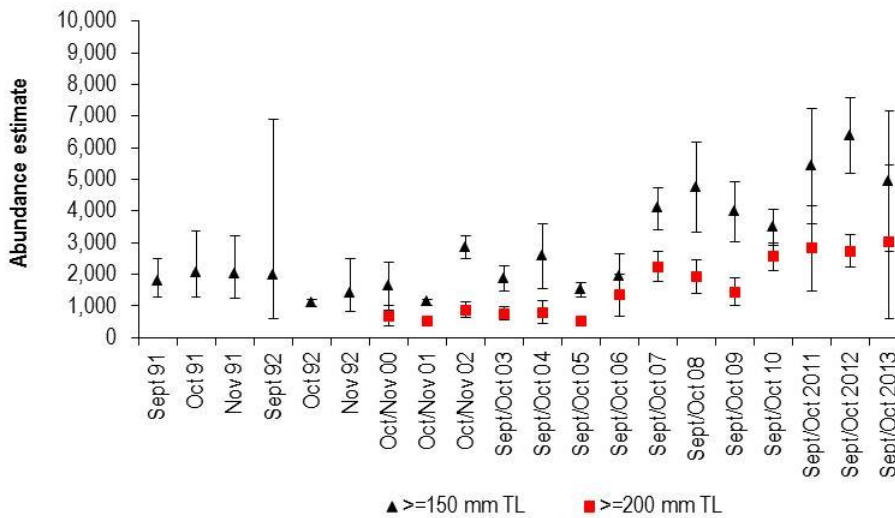


Figure 12. Abundance estimates ($\pm 95\%$ CI) of humpback chub ≥ 150 mm and ≥ 200 mm in the Little Colorado River during (A) spring (2001-2013) and (B) fall seasons (2000-2013). Closed spring and fall abundance estimates of humpback chub > 150 mm in the Little Colorado River during 1991 and 1992 are from Douglas and Marsh (1996).

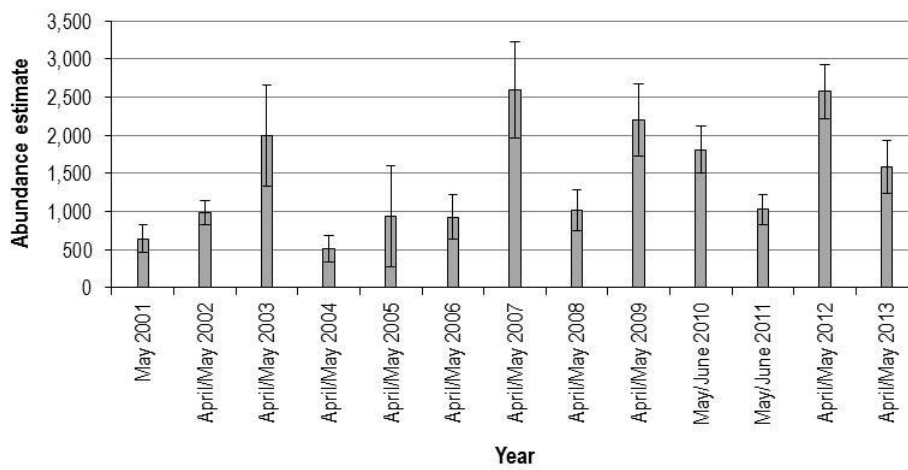
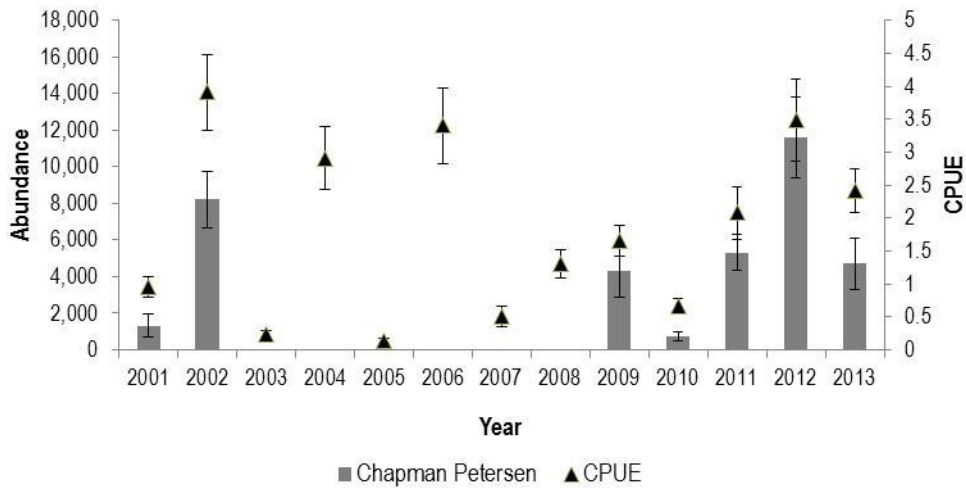


Figure 13. Spring abundances 2001-2013 of humpback chub in the 150-199 mm size category; Little Colorado River.

A.



B.

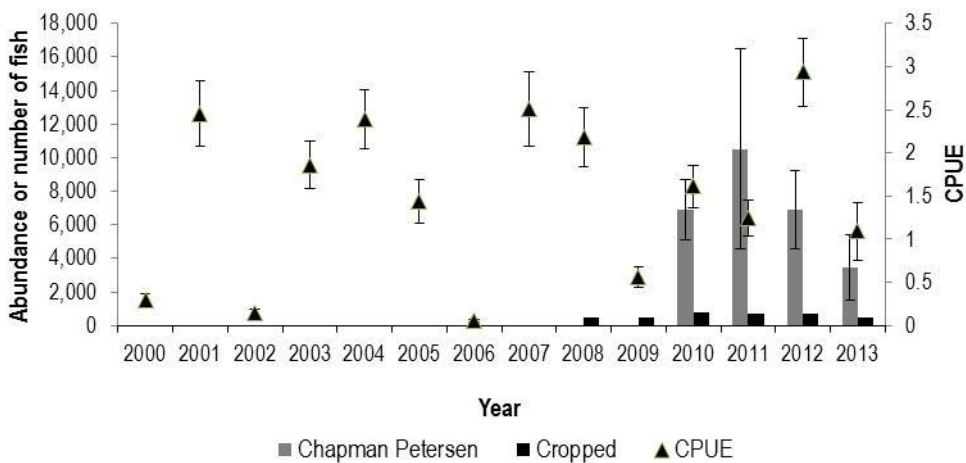


Figure 14. (A) Chapman Petersen abundance estimates of humpback chub in the 100-149 mm size class during spring seasons (2001,2002 and 2009-2013) and spring season catch per unit effort (CPUE) of humpback chub in the 100-149 mm size class (2001-2013). (B) Chapman Petersen abundance estimates of age 0 humpback chub (40-99 mm) during fall seasons (2010-2013) and fall season CPUEs of age 0 humpback chub (2000-2013). Also shown in (B) are the number age 0 humpback chub that have been cropped from the lower ~1.5 to 10 river km of the LCR since 2008 for Shinumo and Havasu creek translocation purposes or for establishment of a refuge at Southwestern Native Aquatic Research and Recovery Center.

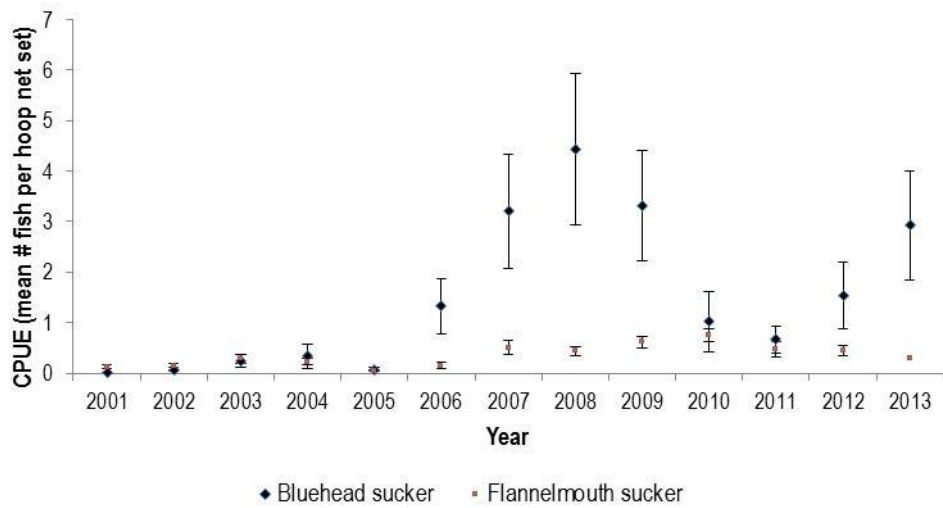
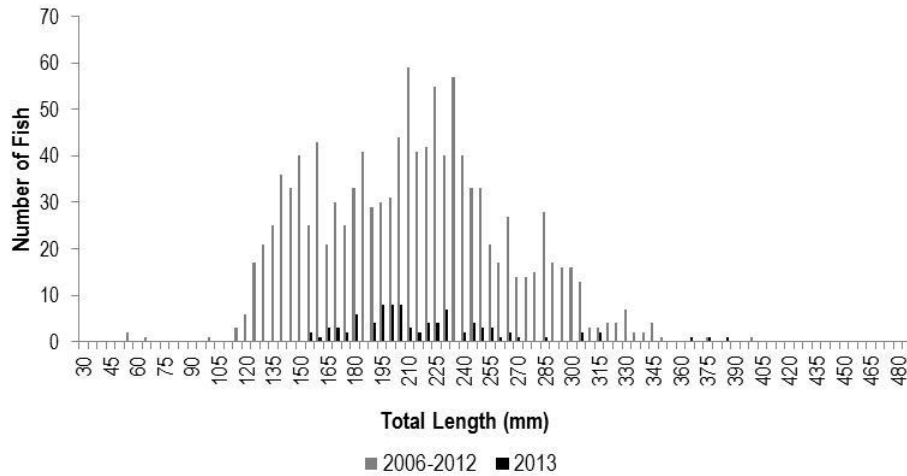


Figure 15. Relative abundance (catch per unit effort, CPUE) of bluehead and flannelmouth sucker ≥ 150 mm in the Little Colorado River (river km 0-13.57), 2001-2013.

A.



B.

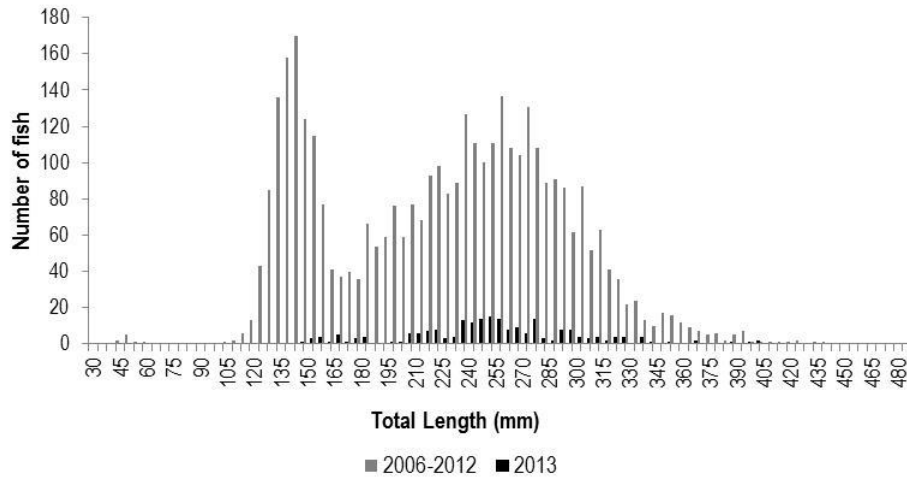
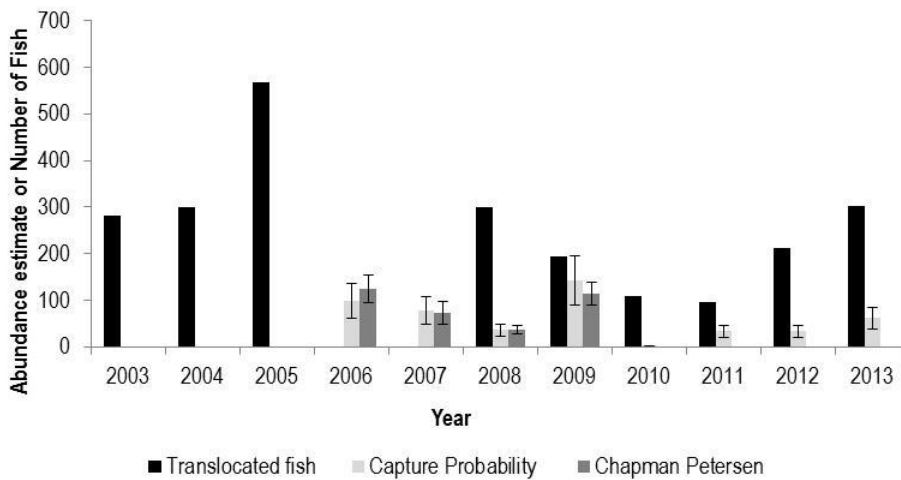


Figure 16. Length frequency distributions of unique humpback chub captured A) upstream of Chute Falls (river km [rkm] 14.13 to 17.68), and B) downstream of Chute Falls (rkm 13.58-14.11) during Chute Falls monitoring trips 2006-2012 and 2013; Little Colorado River.

A.



B.

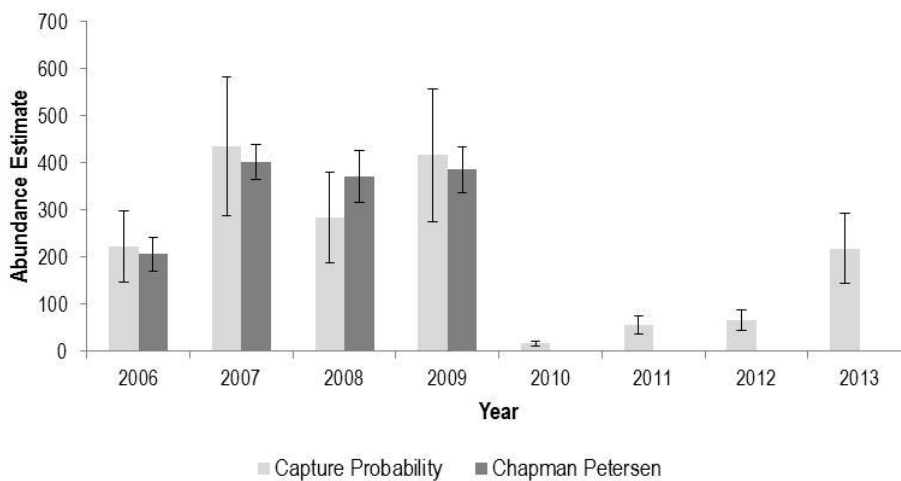


Figure 17. (A) Numbers of humpback chub that have been translocated to upstream of Chute Falls since 2003 (black bars) and abundances of adult humpback chub (≥ 200 mm) in upper reach upstream of Chute Falls (river km [rkm] 14.1 to 17.7) since summer 2006, and (B) abundances of adult humpback chub in lower reach downstream of Chute Falls since summer 2006. Note, abundances for both upper and lower reaches are shown as those estimated with Chapman Petersen mark-recapture (dark grey bars) and those estimated using capture probability data (light grey bars) derived from the Chapman Petersen mark-recapture efforts.