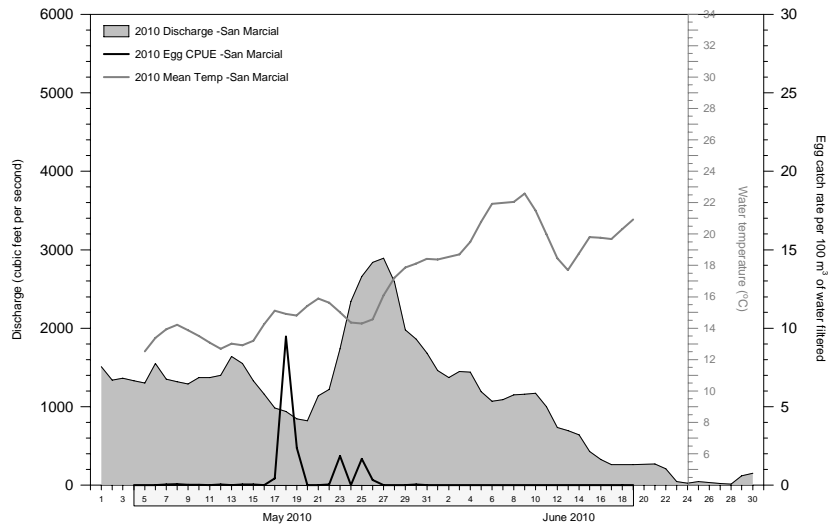
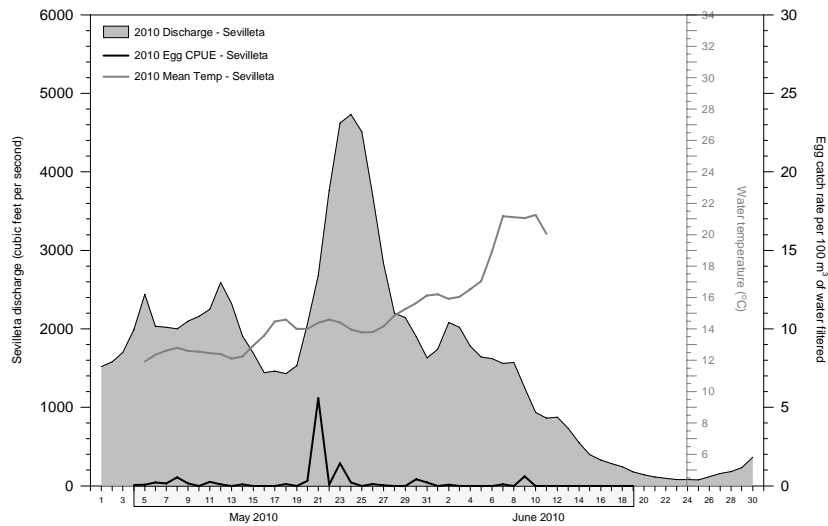


**SPATIAL SPAWNING PERIODICITY OF RIO GRANDE SILVERY MINNOW
DURING 2010**

**A MIDDLE RIO GRANDE ENDANGERED SPECIES ACT
COLLABORATIVE PROGRAM FUNDED RESEARCH PROJECT**



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30 September 2010

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EXECUTIVE SUMMARY

Systematic monitoring of the reproductive output of Rio Grande silvery minnow at multiple sites in the Middle Rio Grande was first conducted in 1999 and has continued annually (except 2005) since 2001. Previous studies demonstrated May and June as the primary period of spawning activity. The 2009-2010 studies were structured to monitor the spatial and temporal (May-June) reproductive output of Rio Grande silvery minnow in the two downstream-most river reaches (Isleta and San Acacia), where the majority of the population currently persists.

Sampling at both 2010 Rio Grande silvery minnow spawning periodicity study sites was conducted from 5 May through 19 June 2010 (47 days). The cumulative volume of water sampled at the two Rio Grande sites in 2010 was 171,335.8 m³ (138.9 acre-feet). The total volume of water filtered was higher at the San Marical Site (105,421 m³) as compared with the Sevilleta Site (65,915 m³). A cumulative total of 586 Rio Grande silvery minnow eggs were collected at the two sites during 2010. The majority ($n = 364$; 62.1%) of the catch was taken at the San Marcial Site while the number and cumulative percent of Rio Grande silvery minnow eggs collected at the Sevilleta site ($n = 222$; 37.9%) was slightly lower. Daily egg catch rates at the Sevilleta Site ranged between 0.04 and 5.57 eggs per 100 m³ of water sampled ($n = 1$ and $n = 125$, respectively) while daily egg catch rates at the San Marcial Site ranged between 0.03 and 9.47 eggs per 100 m³ of water sampled ($n = 1$ and $n = 223$, respectively). During the study, mean daily egg catch rates at the Sevilleta and San Marcial sites were 0.34 and 0.35 eggs per 100 m³ of water sampled, respectively. The number of eggs estimated to be transported downstream of the Sevilleta Site over the duration of the study was 727,010 with a daily maximum of 365,187. The number of eggs estimated to be transported downstream of the San Marcial Site over the duration of the study was 500,755 with a daily maximum of 217,576.

Analysis of reproductive output revealed a significant difference ($F = 6.36$; $p < 0.0001$) among mean values of catch rate (#/100m³) at the San Marcial Site for the eight years of the study (2002-2004, 2006-2010). The natural log-transformed mean egg catch rate (standardized for discharge) was highest in 2002 (5.86 ± 0.87), followed by 2007 (4.77 ± 0.34), 2008 (3.22 ± 1.22), 2003 (2.89 ± 0.50), 2009 (2.57 ± 0.59), 2010 (2.24 ± 0.55), 2006 (1.44 ± 0.67), and 2004 (0.96 ± 1.22). Additional statistical analyses for the Isleta Reach (2006-2010) revealed a significant difference ($F = 9.45$; $p < 0.0001$) among mean values of catch rate (#/100m³) among years. The natural log-transformed mean egg catch rate (standardized for discharge) was highest in 2009 (4.68 ± 0.45), followed by 2008 (4.28 ± 0.40), 2007 (4.19 ± 0.36), 2010 (2.60 ± 0.36), and 2006 (1.97 ± 0.40) in the Isleta Reach.

Additional comparisons of reproductive output (2006 to 2010) revealed significant differences ($p < 0.05$) in mean values of catch rate (#/100m³) among reaches in 2006, 2007, and 2009. In 2006, the natural log-transformed mean egg catch rate (standardized for discharge) was higher ($p < 0.05$) in the Angostura Reach than in either the Isleta or San Acacia reaches. This pattern was reversed in 2007 when the natural log-transformed mean egg catch rate (standardized for discharge) was higher ($p < 0.05$) in the San Acacia Reach than in Angostura Reach. The natural log-transformed mean egg catch rate (standardized for discharge) was higher ($p < 0.05$) in the Isleta Reach than in the San Acacia Reach during 2009.

Regression analyses of reproductive effort and key river parameters (i.e., discharge and water temperature) revealed several significant, albeit variable, relationships. A comparison between log-transformed egg catch rate (standardized for discharge) and discharge at the Sevilleta and San Marcial sites yielded significant ($F = 11.71$; $p < 0.001$; $r^2 = 0.11$ and $F = 10.68$; $p < 0.01$; $r^2 = 0.09$, respectively) positive relationships. Similarly, a comparison between log-transformed egg catch rate (standardized for discharge) and the percentage increase in discharge (over two days) at the Sevilleta and San Marcial sites yielded significant ($F = 9.33$; $p < 0.01$; $r^2 = 0.09$ and $F = 15.42$; $p < 0.001$; $r^2 = 0.13$, respectively) positive relationships. Comparisons between log-transformed egg catch rate

(standardized for discharge) and water temperature yielded a significant ($F = 10.77$; $p < 0.01$; $r^2 = 0.10$) negative relationship at the Sevilleta Site but a non-significant negative relationship at the San Marcial Site ($F = 1.20$; $p = 0.28$; $r^2 = 0.01$).

While some spawning events occurred at cool water temperatures ($<17^{\circ}\text{C}$) at either site, most spawning occurred between water temperatures of about 17 to 22°C. At warmer water temperatures (ca. 20 to 26°C), there was a gradual decline in the flow at which spawning was observed (i.e., generally coinciding with low June discharge). The highest densities of eggs were collected from the Sevilleta Site between about 16.5 to 19.5°C, coinciding with flows between about 750 to 3,250 cfs. In contrast, the highest densities of eggs were collected at the San Marcial Site between about 19 to 22°C, coinciding with either low flows (ca. 200 to 600 cfs) or high flows (ca. 1,750 to 2,250 cfs).

These findings suggest that the number of eggs produced in the river appears to be related to some combination of discharge and water temperature conditions. Large increases in flow over a relatively short period (one to two days) seemed particularly related to increased spawning activity in Rio Grande silvery minnow. Increased flow leads to a series of changes to the physical dynamics of the river (e.g., increased water velocities/depths in some areas and augmented or new “flooded” low velocity habitats in other areas) and changing water chemistry conditions (e.g., increased turbidity and salinity levels) that could be important spawning cues associated with rising flows. While increases in discharge appear to be the primary cue for spawning, water temperature seems to be an important factor as well. Spawning appears to occur over a wide range of mean daily water temperatures (ca. 15 to 26°C) but with the majority occurring over a narrower range temperatures (ca. 17 to 22°C). While the complex interactions among abiotic and biotic variables in the early survival, growth, and recruitment of Rio Grande silvery minnow give some insight to the patterns of their spatial spawning periodicity, there are still many unanswered questions.

Population trends lend support to the observation that substantial numbers of eggs (i.e., several million during 2009 and 2010), and presumably larvae, are being transported downstream every year. In support of these observations, the highest densities of juvenile Rio Grande silvery minnow are most frequently found in the southern reaches of the Middle Rio Grande. Recent population monitoring surveys (June 2010) show the majority (59.5%) of the population is located in the San Acacia Reach. This trend was first noted over two decades ago and, with few exceptions, it has persisted to the present time.

Despite the seemingly large number of Rio Grande silvery minnow propagules transported downstream every year, some portion do remain upstream. It is likely that the proportion of individuals retained and successfully recruited upstream is related to the complexity of instream habitat conditions and the long-term availability of nursery habitat. Years with elevated and extended spring runoff conditions appear to create the favorable habitat conditions required for the successful recruitment of early life stages of Rio Grande silvery minnow.

The high snow pack runoff and elevated discharge that stimulates spawning by Rio Grande silvery minnow resulted in a modest spawning effort during the 2010 study period. Rio Grande silvery minnow appear to have had a good year for spawning and recruitment in 2010 (as compared with drought years), which could translate into increased numbers of reproductively capable females available to spawn in the spring of 2011. While the population of Rio Grande silvery minnow appears to have stabilized since 2007, the lack of an adequately high spring runoff (high magnitude over an extended duration) combined with summer drying could result in a rapid decline to 2002/2003 population levels. The loss of individuals from downstream reaches during river drying events is particularly problematic as these are the areas that most frequently and consistently support the highest densities of Rio Grande silvery minnow (except in years immediately following downstream drying). The future conservation status of Rio Grande silvery minnow appears dependent on ensuring adequate flow and habitat conditions during the spawning and early recruitment phases of this species while also allowing upstream recolonization by individuals transported downstream.

INTRODUCTION

The reach of the Rio Grande between Cochiti Dam and Elephant Butte Reservoir (Middle Rio Grande) has been greatly modified over the last 50 years; this has alternatively led to aggradation, degradation, armoring, and narrowing of the river channel in different portions of the reach (Lagasse, 1985). This section of the river flows through the massive Rio Grande rift and historically resulted in a wide floodplain within the sparsely vegetated Rio Grande valley. Extensive braiding of the river through the relatively linear Rio Grande rift valley was common as it flowed over shifting sand and alluvium substrata; flow in the Middle Rio Grande was generally perennial except during times of severe or extended drought (Scurlock, 1998).

The Middle Rio Grande was relatively shallow throughout most of the year because of regionally low precipitation levels (Gold and Dennis, 1985) but was subjected to periods of high discharge. Flow was generally greatest during the annual spring snow melt runoff (April-June), however intense localized rainstorms (monsoonal events that generally occur in July and August) often caused severe flooding and were important in maintaining perennial flow through the summer. The cyclic pattern of drought and flooding over mobile substrata likely helped to promote the active interaction between the river and its floodplain. Historically, the Middle Rio Grande in many ways possessed all of the characteristics distinctive of a dynamic semi-arid river ecosystem.

The reduced species diversity typical of semi-arid ecosystems was also reflected in the depauperate ichthyofaunal composition of the Middle Rio Grande. Despite the reduced species richness of the Rio Grande, the river supported many native cyprinids that were endemic to this drainage (Platania and Altenbach, 1998). However, of the few native cyprinids that historically occupied the Rio Grande basin (i.e., speckled chub, *Macrhybopsis aestivalis*, Rio Grande shiner, *Notropis jemezianus*, and Rio Grande bluntnose shiner, *Notropis simus simus*) many have been extirpated from the Middle Rio Grande over the past century. A fourth species, phantom shiner, *Notropis orca*, is extinct (Bestgen and Platania, 1990). Rio Grande silvery minnow, *Hybognathus amarus*, is the only extant member of the native cyprinid fish fauna (Bestgen and Platania, 1991; Platania, 1991) and is found only in the Middle Rio Grande.

This group of native cyprinids shared several life-history characteristics. All were small (generally <100 mm TL), short-lived (2-5 years), fishes that occupied mainstem habitats. In addition to these shared traits, all five species were members of a reproductive guild of fishes that are pelagic spawners of semibuoyant eggs (Platania and Altenbach, 1998).

Reproduction in fish in this guild is characterized by the production of non-adhesive eggs that, upon expulsion from the female, swell rapidly with water and become nearly neutrally buoyant. Spawning is generally associated with increases in discharge, such as spring run-off or summer rainstorms. The eggs are about 1.6 mm in diameter shortly after spawning but quickly expand (ca. 3.0 mm) and remain suspended in the water column during development. Egg hatching time is temperature dependent, but rapid, occurring in 24-48 hours. Recently hatched larval fish remain a component of the drift until development of the gas bladder. This physiological development corresponds with a shift in swimming behavior as larvae actively seek low-velocity habitats.

The 3-5 days necessary for propagules to attain the developmental stage necessary to control horizontal movements and freely disperse allows for considerable downstream displacement of eggs and larvae in the Middle Rio Grande. As has been well documented for other aquatic organisms, it is necessary for at least some portion of the drifting propagules to settle in appropriate nearby low-velocity habitats or move upstream to maintain viable populations (Speirs and Gurney, 2001). Downstream transport distance of the progeny of Rio Grande silvery minnow is dependent on a variety of factors including flow magnitude and duration, water temperature, and channel morphology (Dudley and Platania, 2007b). Historically, there were no permanent barriers to upstream dispersal of fishes in the Middle Rio Grande. There are currently three instream diversion

structures between Cochiti Dam and Elephant Butte Reservoir that act as barriers to upstream movement of fishes and fragment the once continuous range of the Rio Grande silvery minnow.

Systematic monitoring of the reproductive output of Rio Grande silvery minnow at several sites in the Middle Rio Grande was first conducted in 1999 (Platania and Dudley, 2000). The 1999 monitoring effort involved collecting and quantifying catch rate of Rio Grande silvery minnow eggs at several Middle Rio Grande sites during the relatively short spawning period of this species. Limited Rio Grande silvery minnow egg collecting efforts were also conducted at selected sites in the Middle Rio Grande (Platania and Hoagstrom, 1996) and in the Low Flow Conveyance Channel (Smith, 1998, 1999) from 1996 to 1999. These latter samples provide information on the magnitude of reproduction during certain times and for specific sites. However, consistent monitoring throughout the spawning season produces the most reliable measure of the duration and magnitude of Rio Grande silvery minnow reproductive output. The first site specific sampling effort to document the magnitude of the reproductive effort of Rio Grande silvery minnow occurred daily throughout May and June 2001 (Platania and Dudley, 2002) at a location near the southern end of the San Acacia Reach of the Middle Rio Grande (River Mile 58.8). Monitoring of the reproductive effort of Rio Grande silvery minnow also occurred daily at this site in May and June 2002 (Platania and Dudley, 2003), 2003 (Platania and Dudley, 2004), and 2004 (Platania and Dudley, 2005). More intensive monitoring efforts were conducted from 2006 to 2008 (Platania and Dudley, 2006, 2007, 2008) and resulted in the sampling of the Angostura, Isleta, and San Acacia reaches of the Middle Rio Grande. Since 2009, monitoring of the Rio Grande silvery minnow reproductive effort has occurred annually in the Isleta and San Acacia reaches of the Middle Rio Grande.

Population monitoring efforts of the Middle Rio Grande fish community over the past decade have documented vast changes (i.e., order of magnitude increases and decreases) in the abundance of Rio Grande silvery minnow (Dudley and Platania, 1999, 2000, 2001, 2002; Dudley et. al., 2003, 2004, 2005; Dudley and Platania, 2007a, 2008, 2009). Recent monitoring efforts (Dudley and Platania, 2009) show that the October density of Rio Grande silvery minnow was significantly lower ($p < 0.05$) in 2008 compared to 2005. However, the October density of this species was higher ($p < 0.05$) in 2008 than in 1996, 2000-2004, and 2006. The San Acacia Reach yielded most of the Rio Grande silvery minnow in October of 2008, followed by the Angostura Reach and Isleta Reach. This was in contrast to population monitoring in October of 2007, when the largest catch rates were recorded in the Angostura and Isleta reaches.

The marked 2002-2003 decline in wild Rio Grande silvery minnow and increased stocking efforts in the upper reaches of its range (Angostura and Isleta reaches) apparently resulted in a temporary reapportioning of this species' relative abundance. Between June 2002 and November 2004, over 301,000 silvery minnow were released in the Angostura and Isleta reaches of the river. While over 90% of the total Rio Grande silvery minnow catch had been recorded in the San Acacia Reach during 2000-2002, by the end of calendar year 2003, the largest percentage (58%) of individuals collected (albeit extremely reduced numbers; $n=224$) were taken in the Angostura Reach. This trend continued into 2004 and by October of that year, approximately 78% of the cumulative 2004 Rio Grande silvery minnow catch had been taken in the Angostura Reach, 13% in the Isleta Reach, and only 9% in the San Acacia Reach. However, the relative abundance of Rio Grande silvery minnow among reaches has fluctuated from 2005-2009 (e.g., highest densities in the Angostura Reach during 2006 and 2007, in the Isleta Reach during 2005, and in the San Acacia Reach during 2008 and 2009). This reapportionment of the Rio Grande silvery minnow population, in combination with the meager 2004 catch of reproductive propagules in the San Acacia Reach necessitated a modification of subsequent reproductive monitoring protocols beginning in 2006. Sampling sites were established in each of the three downstream reaches of the Middle Rio Grande (Angostura, Isleta, and San Acacia) to provide a more complete data set from 2006 to 2008. The Middle Rio Grande Endangered Species Act Collaborative Program (MRGESACP) changed the

scope of the project in 2009 to focus sampling efforts in the Isleta and San Acacia reaches, where the majority of the spawning effort occurs.

The study conducted herein is at its core a continuation of the systematic Rio Grande silvery minnow reproductive monitoring research activity, fulfills multiple recovery goals, and will provide detailed catch-per-unit-effort (CPUE) values for Rio Grande silvery minnow eggs. The primary objective of this study is to provide data that will enable characterization of the timing, duration, and magnitude of Rio Grande silvery minnow reproduction in two reaches of the Middle Rio Grande to assess temporal and spatial differences in spawning effort. This document presents the results of the 2010 spawn of Rio Grande silvery minnow and compares data collected under the auspices of this study from 2001-2004 and 2006-2010. Long-term monitoring of the reproductive effort of Rio Grande silvery minnow provides insight to potential factors affecting annual reproductive output, remains necessary for ongoing recovery efforts, and helps facilitate effective management decisions in the Middle Rio Grande.

Institutional background and considerations

Monitoring the reproductive effort of Rio Grande silvery minnow was identified as a requirement of the 29 June 2001 Programmatic Biological Opinion of the Effects of Actions Associated with the U. S. Bureau of Reclamation's, U. S. Army Corps of Engineers', and Non-Federal Entities Discretionary Actions related to Water Management on the Middle Rio Grande, New Mexico as authored by the U. S. Fish and Wildlife Service. This work was part of an ongoing effort to document changes in the distribution and abundance of the federally endangered Rio Grande silvery minnow. This research effort provided an assessment of the reproductive output (eggs) for Rio Grande silvery minnow within the Middle Rio Grande and specifically addressed the task: "Evaluate the status and trend of the Rio Grande silvery minnow" as identified by the MRGESACP.

The Rio Grande silvery minnow Recovery Plan (U. S. Fish and Wildlife Service, 1999) also outlined research objectives (2.2. Determine spawning periodicity of silvery minnow under multiple flow regimes; 2.2.1. Determine environmental factors that cue spawning in silvery minnow) that were addressed through this research. This investigation provided an assessment of the relative magnitude of the Rio Grande silvery minnow spawning effort. This project was also a central component of the Rio Grande silvery minnow propagation and genetics research efforts, both requirements of the 29 June 2001 Programmatic Biological Opinion (see "Project Objectives" 2 and 3).

In 2002-2003, MRGESACP members met and discussed Rio Grande flow issues and impacts of the hydrological conditions on Rio Grande silvery minnow. The dismal 2002-2003 snow pack in the Rio Grande headwaters meant there would not be a natural spring flow spike in 2003 in the Middle Rio Grande and, therefore, it was unlikely that there would be a spring spawn by Rio Grande silvery minnow. Personnel from the MRGESACP decided to create an artificial flow spike during mid-May 2002, using reservoir storage, to initiate a spawn by Rio Grande silvery minnow. As 2003 climatic conditions were similar to those experienced in 2002, an artificial flow spike was also created in the Rio Grande in 2003, using reservoir storage, to initiate spawning of Rio Grande silvery minnow. Snow pack runoff and ambient flow conditions in 2004 were sufficient enough that, for the first time in two years, an artificial flow spike was not necessary. Artificial flow spikes have not been generated since 2004 for the purpose of inducing spawning.

STUDY AREA

The principal area of interest in the Middle Rio Grande is the reach between the outflow of Cochiti Reservoir and inflow to Elephant Butte Reservoir as it encompasses the known range of Rio Grande silvery minnow (Figure 1). Five upstream reservoirs and numerous irrigation diversion dams regulate flow in the Middle Rio Grande. Cochiti Reservoir has been operational since 1973, is located 76 km upstream of Albuquerque, and is the primary flood control reservoir that regulates flow in the Middle Rio Grande. Reach names are taken from the diversion structure at the upstream boundary of that reach of river. In the Cochiti Reach (between Cochiti Dam and Angostura Diversion Dam), the Rio Grande flows through Cochiti, Santo Domingo, and San Felipe pueblos, respectively.

The reproductive effort of Rio Grande silvery minnow has, in the past, been sporadically determined at selected collecting localities in the Angostura and Isleta reaches. In 2003 and 2004, our sampling efforts were restricted to the single San Acacia Reach collection location. The San Acacia Reach of the Middle Rio Grande is about 56 miles (91 km) long extending from the apron of San Acacia Diversion Dam to the head of Elephant Butte Reservoir. Sections of this reach are characterized by a wide and braided river channel, sand substrate, high suspended sediment load, and a broad variety of aquatic mesohabitats. Conversely, some segments in this reach are relatively narrow and result in increased water velocity and decreased habitat heterogeneity. The 12 mile (19 km) reach of the Rio Grande downstream of San Marcial Railroad bridge crossing is confined to a channel that is about 50 m wide. Substrate in this segment of the river is predominately sand and braiding of the channel is uncommon except under conditions of relatively low flow.

Given the downstream drift of the eggs, the original location of the collecting activities was selected so as to maximize the number of eggs collected and potentially rescue eggs destined to drift into Elephant Butte Reservoir where, if hatched, larvae would be subjected to a wide array of nonnative predators. This site was located near in the downstream-most portion of the San Acacia Reach. The sampling site is downstream of U. S. Geological Survey stream gauging station (# 08358400), which is the nearest upstream San Acacia Reach gauge. In addition to easy accessibility and favorable river conditions (i.e., wide river channel, current being carried through a single river channel, gently sloped banks, moderate gradient), the only means of vehicle access to this site was gated and could be secured. This area has been sampled annually from 2001-2004 and from 2006-2010.

The Isleta Reach site is on the Sevilleta National Wildlife Refuge (near confluence of the Rio Grande and Canada Ancha) and about 4.8 river miles upstream of San Acacia Diversion Dam. The Sevilleta site is downstream of U. S. Geological Survey stream gauging station (# 08331510), which is the nearest upstream Isleta Reach gauge (Figure 2). The Sevilleta sampling site has been sampled annually from 2006-2010.

MATERIALS AND METHODS

The egg collecting device, developed specifically for the collection of large numbers of live and undamaged semibuoyant fish eggs (Moore Egg Collector; MEC), was the only sampling apparatus used in this project (Altenbach et al., 2000). Numerous modifications have been made to the collecting gear, since the original publication detailing the construction and operation of the MEC (Altenbach et al., 2000), that have resulted in increased effectiveness and efficiency of the MEC (i.e., greater catch rate per sampling period). Catch rate of Rio Grande silvery minnow eggs in the Middle Rio Grande was determined following the sampling protocol described in Altenbach et al. (2000). A mechanical flow-meter was attached to the MEC so that volume of water filtered could be calculated and catch rate per unit of water determined. The catch-per-unit-effort (CPUE) of drifting eggs was

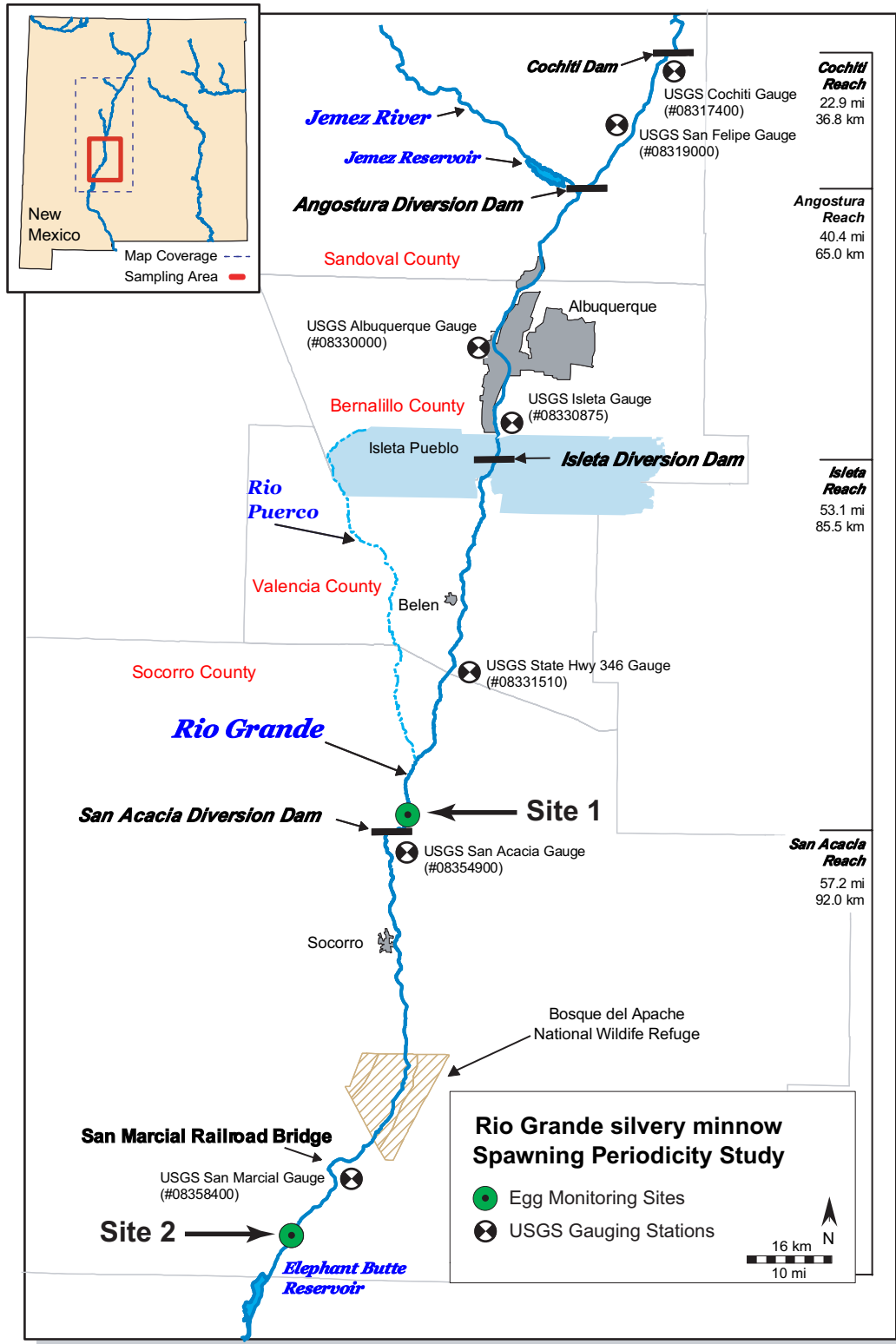


Figure 1. Map of the Middle Rio Grande, New Mexico, and the 2010 study site locations.

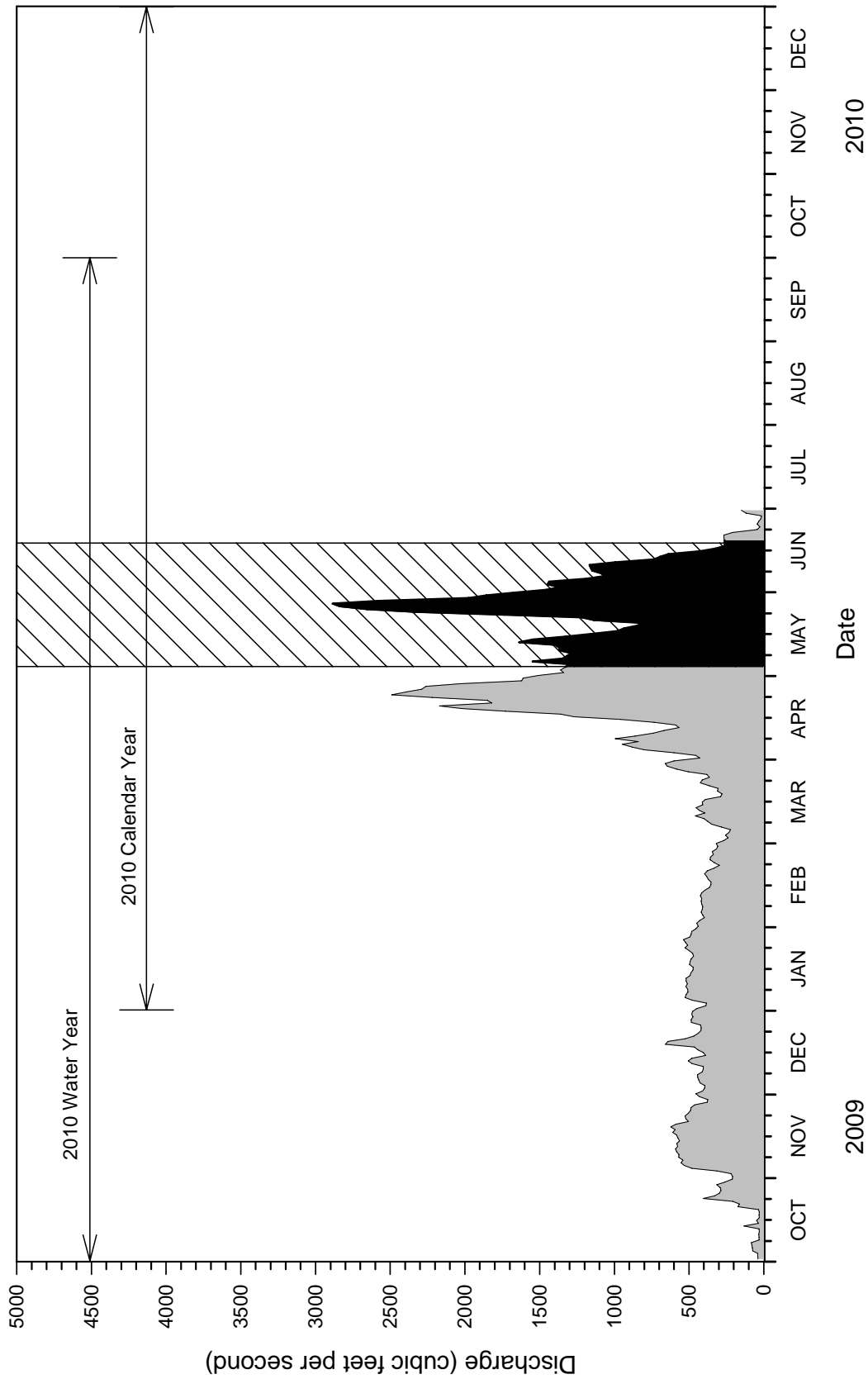


Figure 2. Hydrograph of the Rio Grande, New Mexico, at the San Marcial Gauging Station before, during, and after the 2010 study period. Cross-hatching indicates annual study periods.

calculated as the total number of eggs collected · volume of water sampled⁻¹ · 100 (i.e., N [eggs] · 100 m³ water⁻¹). The total number of eggs passing a sampling site in a 24 hour period was estimated by using egg sampling data from the site (over a 6 hour period) and flow data from the nearest upstream USGS gauging station (i.e., (number of eggs collected / (volume of water sampled / volume of water available)) · 4).

Previous studies (Platania and Dudley 2000, 2002-2009) demonstrated May and June as the primary period of Rio Grande silvery minnow reproductive activity. The normal sampling regime in 2009 was comprised of three daily efforts (morning, noon, and afternoon), each of two-hour duration. Eggs were not staged (i.e., determining approximate time from spawning) as this would require substantial laboratory work outside of the current core objectives of this study. Staging eggs would give a general idea of drift time but extrapolating to drift distance would be subject to a series of simplifying assumptions. Also, determining drift distance is a complex modeling exercise of which eggs are only a component (i.e., eggs can be present in the drift for about one day but larvae can be present in the drift for about another three days post-hatching). Two MEC's were operated so as to increase the volume of water sampled per unit of time. Research personnel worked daily at both sampling sites from 4 May through 19 June 2010.

Volumetric determination of the number of Rio Grande silvery minnow eggs collected, as employed in 2001, lacked the rigor necessary for effective evaluation of the relative level of spawning by this species. Minor changes initiated in the 2002 sampling protocol were instituted to increase the amount and utility of the information acquired from this research activity. The result was that the two principal 2002 project objectives, determining the reproductive output of Rio Grande silvery minnow and obtaining eggs for use in Rio Grande silvery minnow propagation activities, were accomplished through slightly different sampling protocols. The aforementioned differences in egg catch rate determination between 2001 and post-2001 studies preclude use of 2001 data for quantitative or statistical comparison with data from subsequent years. There have not been changes in the methodology for quantitative determination of egg catch rates since 2002.

Rio Grande silvery minnow egg CPUE values are (in part) dependent on flow conditions thereby precluding unadjusted comparison of inter-annual catch rates (e.g., higher flow volume will result in lower CPUE assuming number of eggs in water column remains constant). To account for these differences, catch rate was standardized (CPUE_S) to CPUE (N [eggs]/100 m³) based on mean daily discharge (Q) using the formula: $CPUE_S = CPUE \cdot Q$.

Assumptions of normality in annual Rio Grande silvery minnow egg catch rates were evaluated using the Shapiro-Wilk test. This statistical procedure has been shown to be excellent when testing for departures from a normal distribution. Critical values of *W* were calculated and significant differences assessed using a goodness-of-fit procedure. The 2002-2004 and 2006-2010 egg catch rate time-series distributions were compared with a normal distribution using the Shapiro-Wilk test. To meet normality assumptions, all data were log-transformed ($X' = \ln(X+1)$). Normal quantile plots of empirical data were also examined in reference to Lilliefors's confidence bounds.

The log-transformed CPUE_S values were compared among years and sites to determine general differences in spawning magnitude. Differences among independent samples were tested using ANOVA. This statistical procedure was used to detect differences among years (at a single site) and among sites (during a single year). Differences between sample means were evaluated based on the critical value of *F* based on sample size. Multiple pair-wise comparisons were made using the Tukey-Kramer HSD procedure.

Linear regression modeling was used to determine the strength of the relationships among log-transformed CPUE_S values, discharge, percentage increase in discharge (over two days), and water temperature. Regression models were developed for both the Sevilleta Site and the San Marcial Site. A negative or positive trend in population abundance was defined as occurring when the slope of the regression was significantly different ($p < 0.05$) from zero.

A new filtering screen to separate drifting debris from Rio Grande silvery minnow eggs was developed and tested for the MEC in 2009. The new screen was designed to allow the passage of much of the very fine particulate debris while preventing the passage of drifting eggs. Experimental tests revealed that the new screen was consistently more efficient at sampling a larger volume of water than was the old screen over the same time period (Platania and Dudley, 2009). All MEC's were fitted with the new screen for sampling conducted in 2010.

Water temperature was recorded by temperature logging devices deployed at the study site and programmed to record hourly water temperature. Hourly water temperature data from the primary temperature logger are presented in this report as mean, minimum, and maximum daily water temperatures. Data from past spawning periodicity studies are also included for comparative purposes.

RESULTS

Hydrology

Flows in the Middle Rio Grande have fluctuated dramatically among years since the beginning of this study in 2001 (Figures 3 to 7). The drought that enveloped the study region since 2000 was somewhat interrupted in 2004 by a moderate snow pack and wetter than normal April. These precipitation events helped but did not replenish the already diminished water reserves in upstream reservoirs. Despite the presence of a more normal spring runoff in 2004, an artificial flow spike was released (as was done in 2002 and 2003) to stimulate spawning in Rio Grande silvery minnow. Snow pack runoff in 2005 was larger (greater magnitude and duration) than any of the previous four study years (egg sampling was not conducted in 2005). Conversely, flow in the Rio Grande during 2006 (prior to 27 June) was extremely low because of minimal spring snowmelt runoff. Spring flow conditions improved markedly from 2007 to 2010, as compared with 2006, and there was an extended period of time during May and June when flows exceeded 1,500 cfs at the State Hwy 346 Near Bosque Gauge (USGS Gauge 08331510).

Base flow in the Rio Grande at the State Hwy 346 Near Bosque Gauge (USGS Gauge 08331510) during May and early June 2009 was generally between 1,500 and 2,500 cfs (Figure 8). Mean daily discharge at the Isleta Reach Site (Sevilleta) for the period 1 April through 30 June 2009 was 1,597.4 cfs (SD = 1,040.6). From 19 May to 24 May, there was a substantial spike in flow as mean daily discharge rose from 1,530 to 4,730 cfs; flows then declined rapidly until 31 May (1,630 cfs) when a second smaller peak began. There was a final steady decline in flow for the month (2,080 cfs [2 June] to <500 cfs [after 14 June]).

Discharge in the Rio Grande at the San Marcial Railroad Bridge Crossing (USGS Gauge 08358400) during the 2009 water year closely mirrored that of the State Hwy 346 Near Bosque Gauge (except at a reduced magnitude). From 1 April to 30 June 2009, daily discharge in the Rio Grande at the San Marcial Gauge ranged from 413 to 3,330 cfs (mean = 1,178.5 cfs, SD = 712.8). Flows peaked on 27 May (2,890 cfs) and then declined steadily throughout June, reaching lows of <200 cfs from 22-30 June.

Water temperature

There was little difference (ca. 1 to 2°C) in mean daily water temperatures at the Sevilleta and San Marcial sites during the beginning of this study (Figure 9). There was a steady increase in mean daily water temperature from about 16°C in early May to about 21°C in late May. The San Marcial Site consistently generated mean daily water temperatures of 20°C or more by late May and

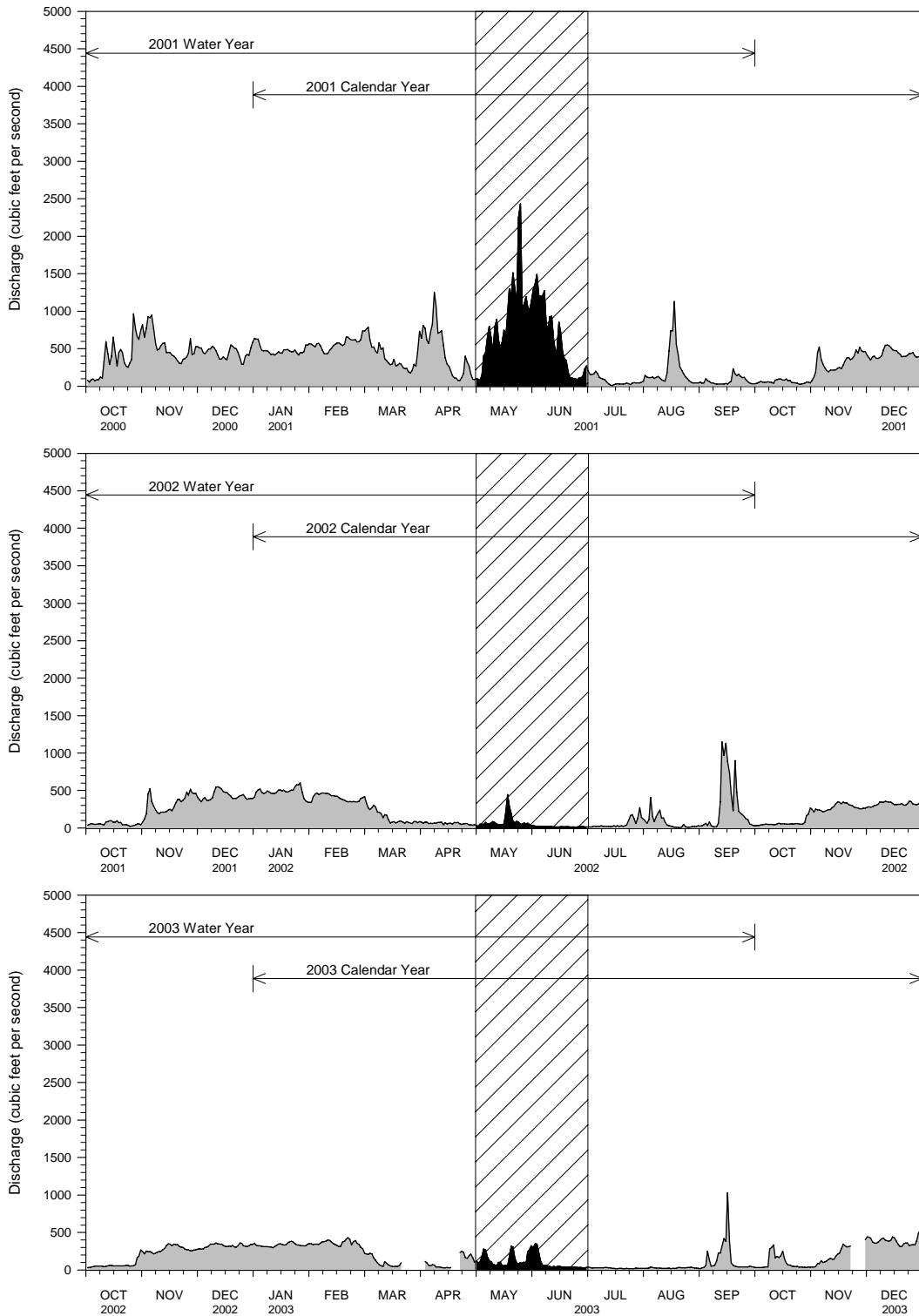


Figure 3. Annual hydrographs of the Rio Grande, New Mexico, at San Marcial before, during, and after the 2001-2003 Rio Grande silvery minnow spawning periodicity study periods. Cross-hatching indicates annual study periods.

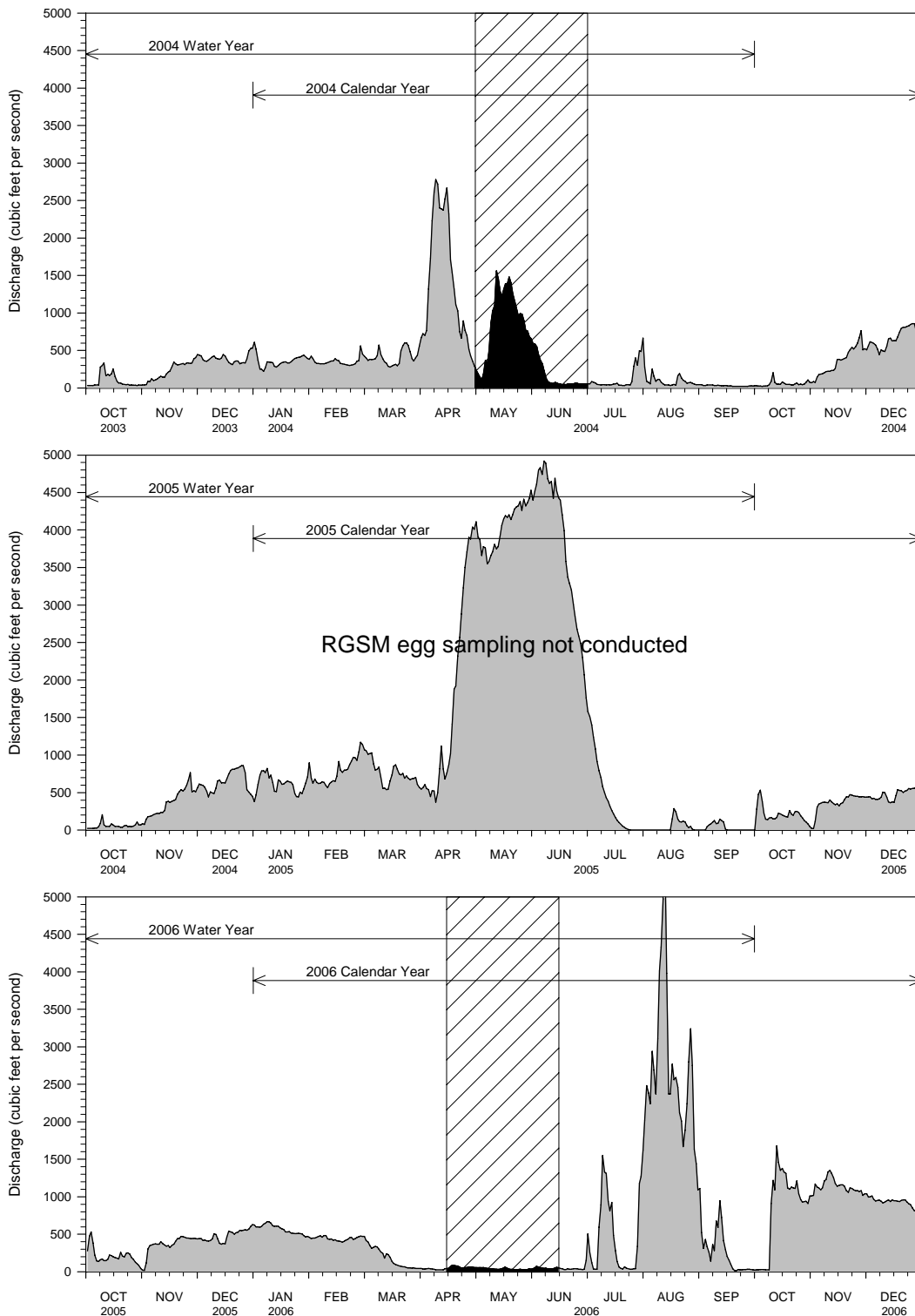


Figure 4. Annual hydrographs of the Rio Grande, New Mexico, at San Marcial before, during, and after the 2004 and 2006 Rio Grande silvery minnow reproductive monitoring study periods. Cross-hatching indicates annual study periods. Sampling was not conducted in 2005.

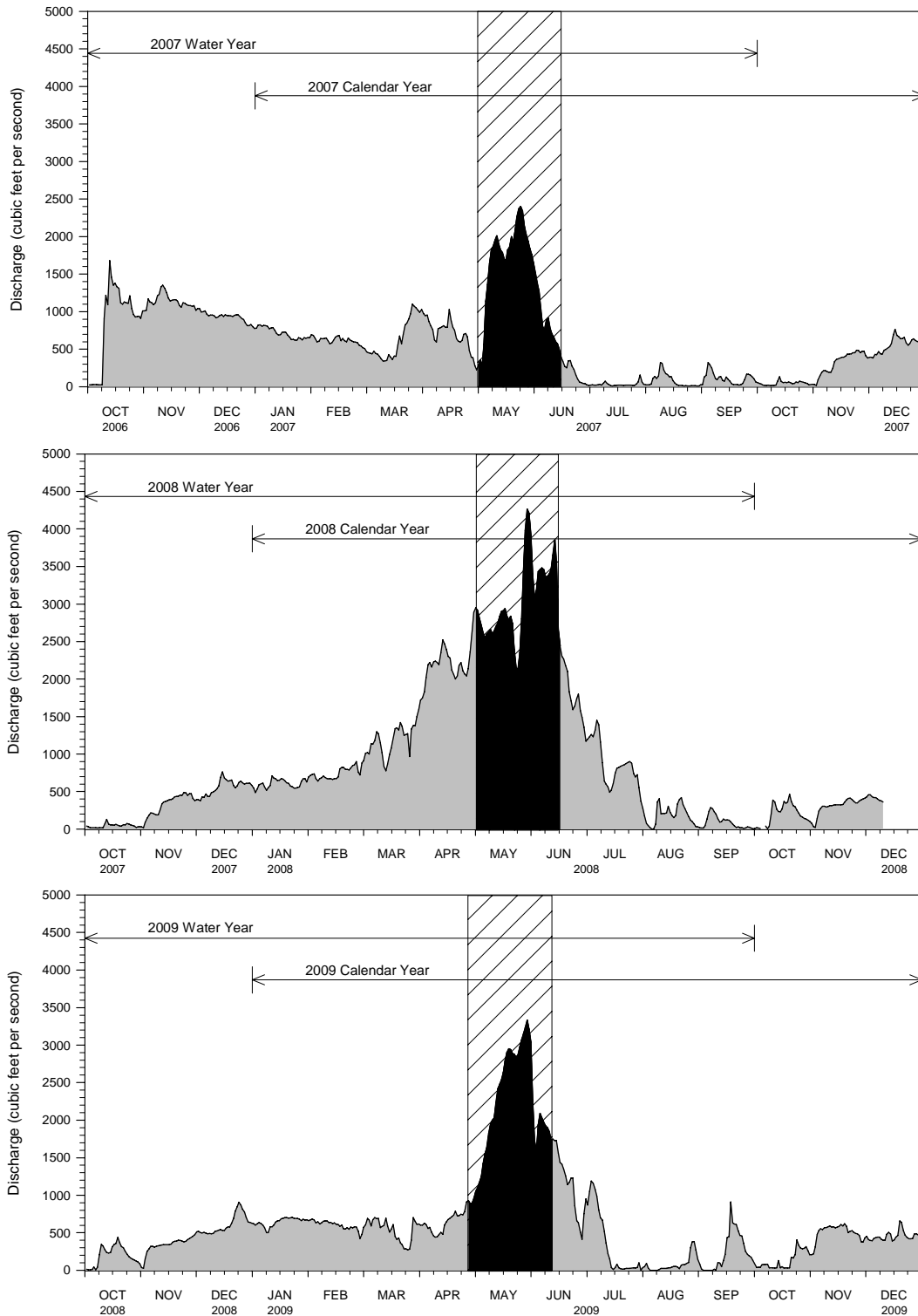


Figure 5. Annual hydrographs of the Rio Grande, New Mexico, at San Marcial before, during, and after the 2007-2009 Rio Grande silvery minnow reproductive monitoring study periods. Cross-hatching indicates annual study periods.

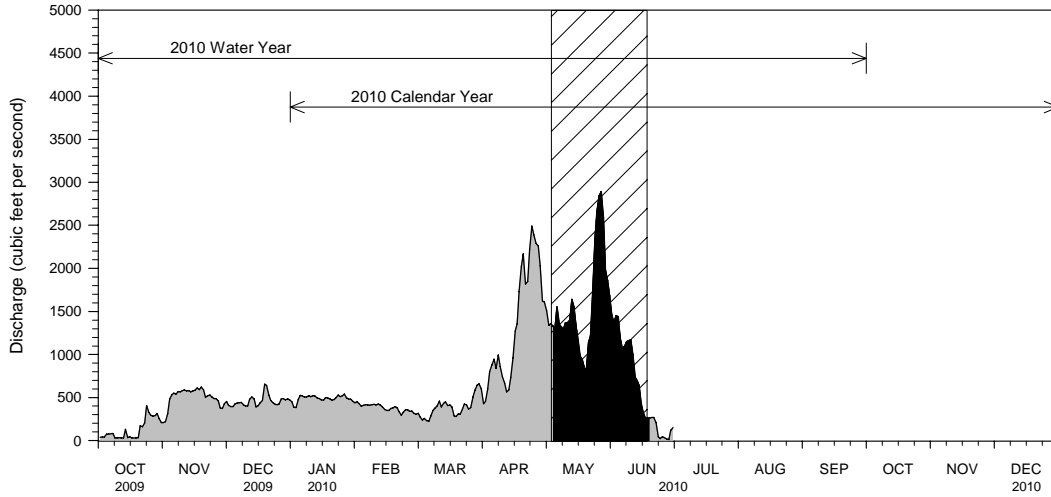


Figure 6. Annual hydrograph of the Rio Grande, New Mexico, at San Marcial before, during, and after the 2010 Rio Grande silvery minnow reproductive monitoring study period. Cross-hatching indicates annual study period.

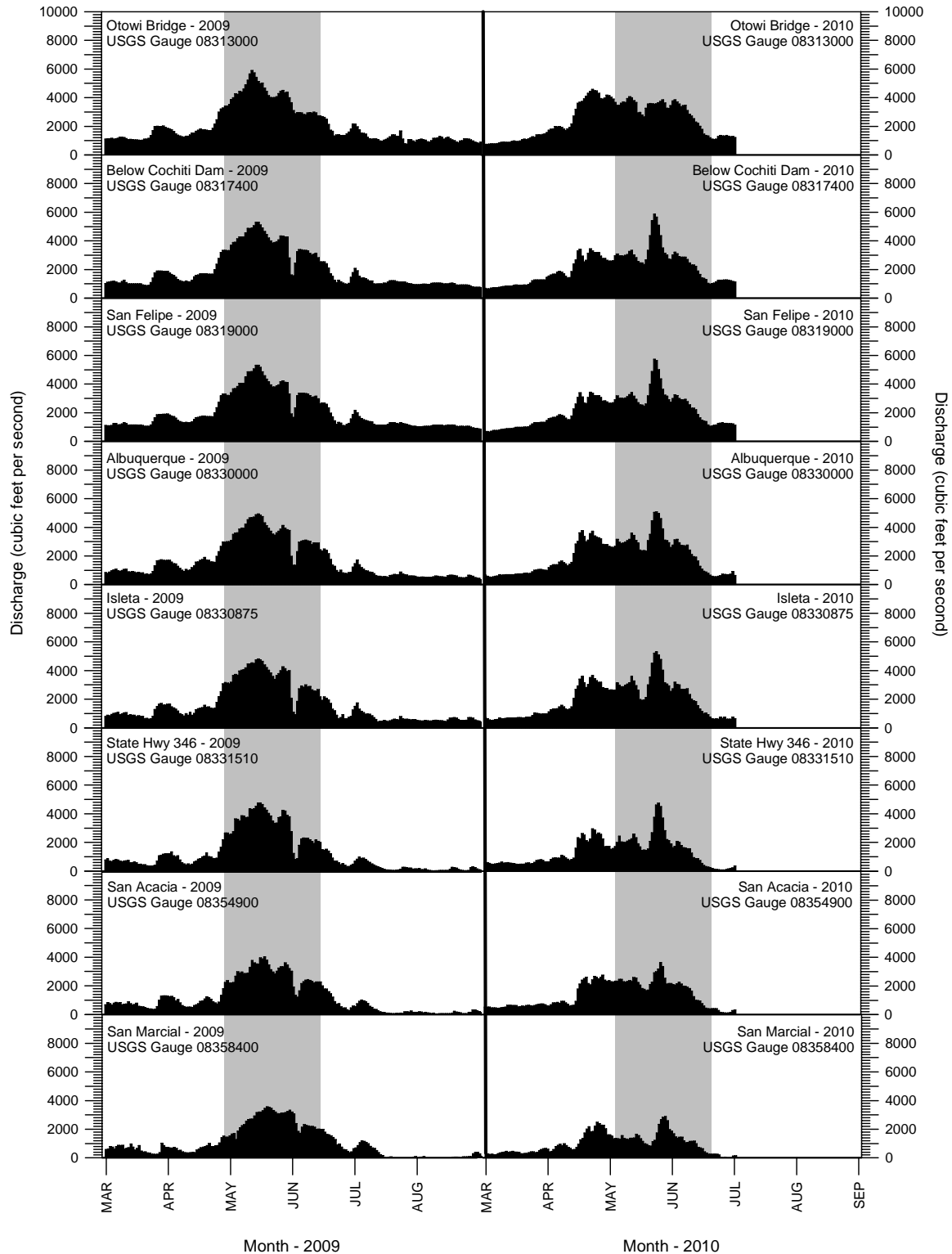


Figure 7. Rio Grande discharge from March through August 2009 and March through June 2010 at seven U. S. Geological Survey Gauge Stations (see Figure 1). The Otowi Bridge gauge is not shown in Figure 1 but it provided for reference. Gray rectangles indicate study periods in 2009 and 2010. Discharge data are provisional and may change.

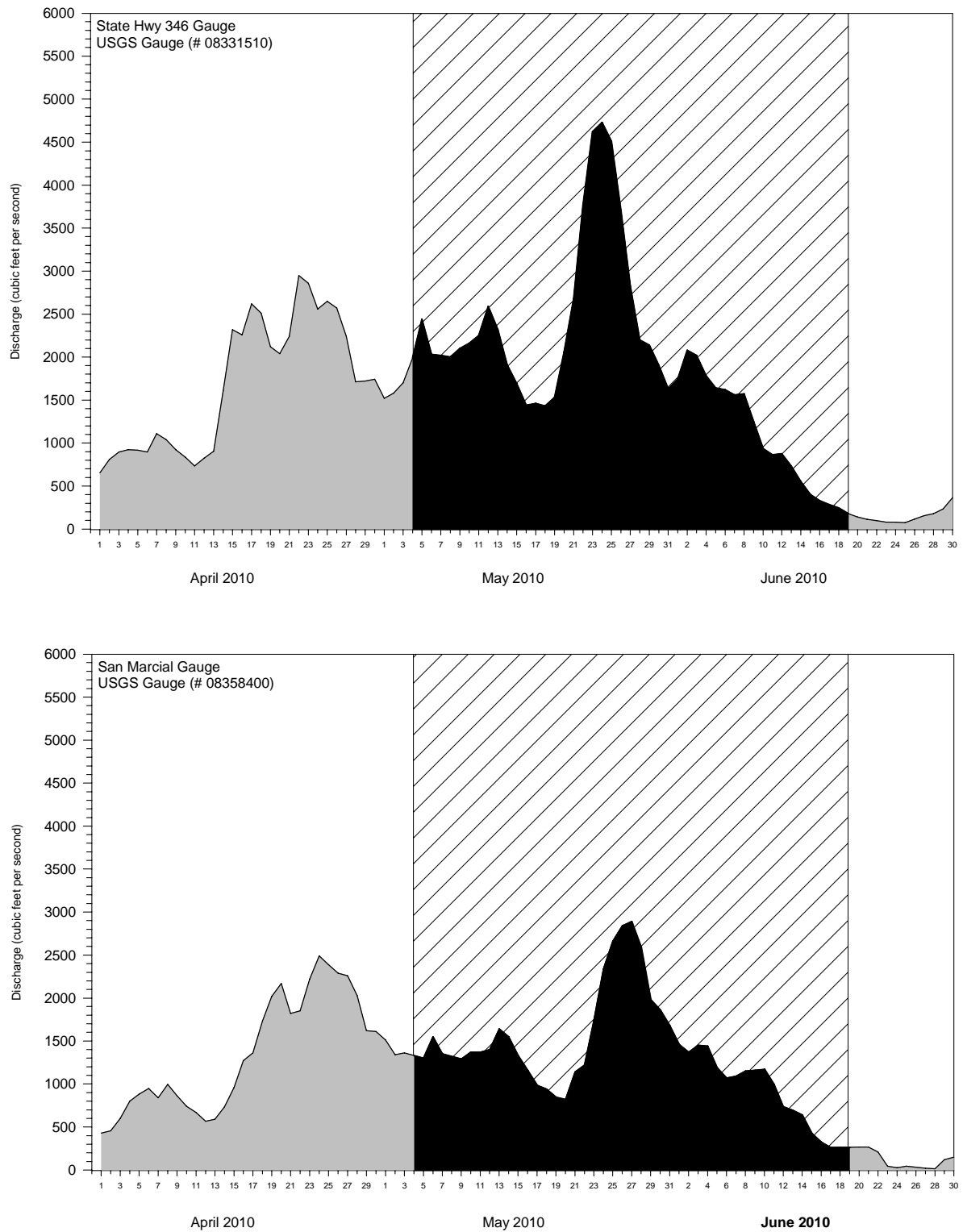


Figure 8. April-June 2010 hydrographs (dark gray) of the Rio Grande, New Mexico, from State Hwy. 346 and San Marcial gauges. Cross-hatching indicates annual study period.

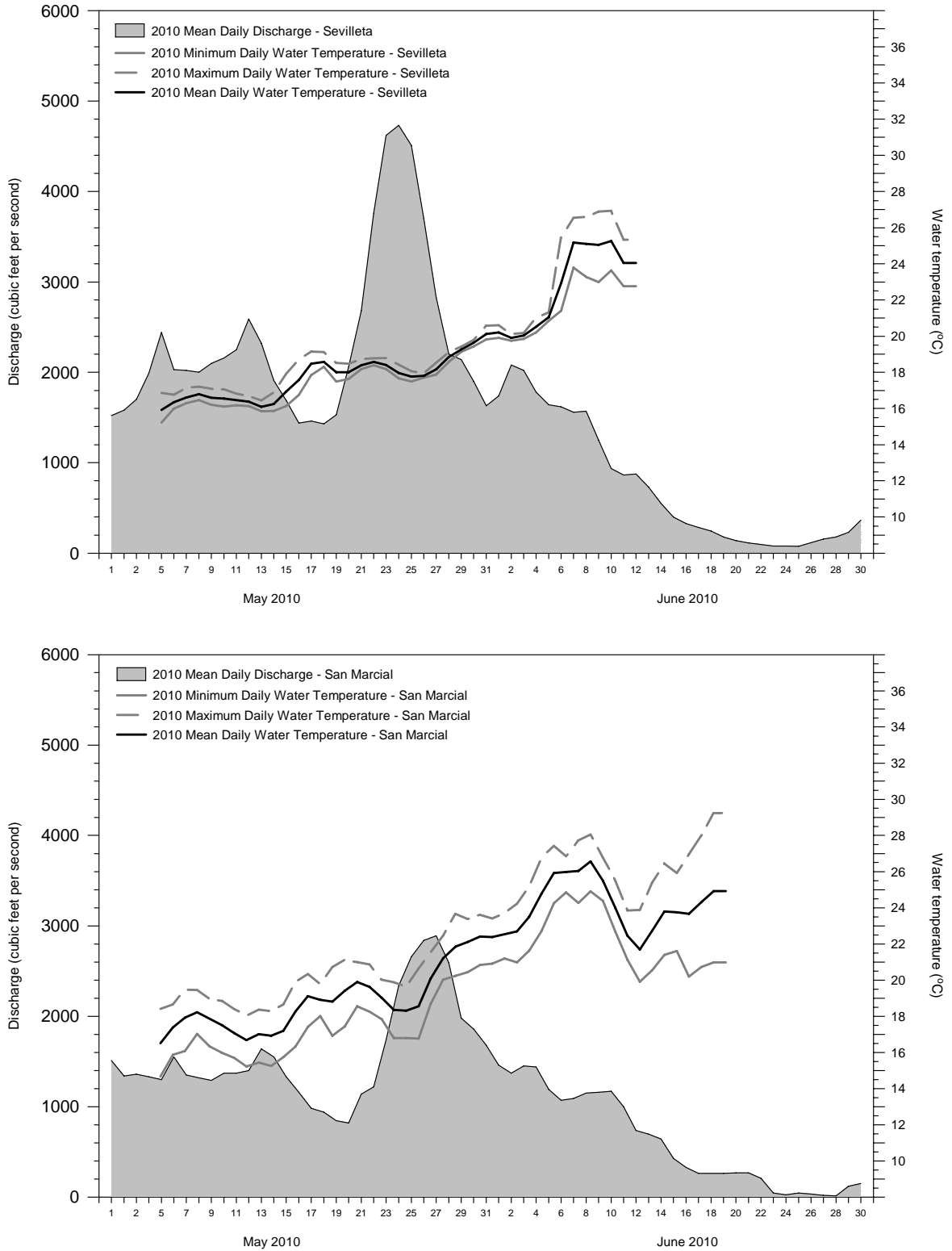


Figure 9. Daily water temperatures (mean, minimum, and maximum) at the 2010 Rio Grande silvery minnow spawning periodicity sampling sites. Approximate mean daily discharge in the Rio Grande at each sampling site is highlighted in light gray.

early June. A similar pattern was observed at the Sevilleta Site but with delayed onset (i.e., early June). By the second week of June, mean daily water temperatures were $>25^{\circ}\text{C}$ at both the Sevilleta and San Marcial sampling sites.

Temperature extremes (minimums and maximums) at the two sampling sites followed different trajectories over the course of the study. In general, there was more diurnal fluctuation in water temperatures at the San Marcial Site as compared with the Sevilleta Site. This was particularly true in late May when flows were elevated. From 5 May to 11 June, maximum water temperatures were 26.9°C at the Sevilleta Site and 28.0°C at the San Marcial Site. Maximum daily water temperature exceeded 20°C at the Sevilleta Site from 31 May until the end of temperature monitoring (11 June) and exceeded 20°C at the San Marcial Site from 16 May until the end of temperature monitoring (22 June). Minimum water temperatures followed a similar pattern as the maximum water temperatures with the coldest temperatures recorded from early to mid-May at both sampling sites. From 5 May to 11 June, minimum water temperatures were 15.2°C at the Sevilleta Site and 14.7°C at the San Marcial Site.

Comparison of 2001 to 2010 water temperatures at the San Marcial Site during May and June demonstrated only modest differences in mean daily water temperature trends across this period (Figures 10 to 12). However, there were notable day-to-day differences in mean water temperatures among years (e.g., 19.8°C [28 May 2004] vs. 25.7°C [28 May 2003]). The minimum daily water temperatures were consistently the lowest in 2002 and 2006, both years with very low flows. Maximum water temperatures were also consistently the highest during these same two years (2002 and 2006) as compared with other years of the study. The general trend for minimum temperatures was a slow steady increase through May and June. In contrast, maximum temperatures generally rose slowly in May and then rose rapidly at some point during June (i.e., coinciding with the reduction in flow following spring runoff). There were sometimes notable differences in the day-to-day maximum water temperatures among years (e.g., 18.2°C [19 May 2008] vs. 30.5°C [19 May 2006]) but the difference in day-to-day minimum water temperatures were not as pronounced. The principal reason for these patterns, besides ambient temperature, was the volume of water in the river channel during the respective study periods. The relatively high flows present in 2001, 2004, and 2007 to 2010 (May to mid-June) served to ameliorate water temperatures and minimize diel variation. Conversely, the very low flow conditions present throughout most of the spawning period in 2002, 2003, and 2006 typically resulted in large daily temperature fluctuations.

2010 Rio Grande silvery minnow spatial spawning periodicity studies

Sampling at both 2010 Rio Grande silvery minnow spawning periodicity study sites was conducted from 5 May through 19 June 2010 (47 days). The cumulative volume of water sampled at the two Rio Grande sites in 2010 was $171,335.8\text{ m}^3$ (138.9 acre-feet). The total volume of water filtered was higher at the San Marcial Site as compared with the Sevilleta Site (Table 1). The cumulative volume of water sampled at the Sevilleta Site was $65,915\text{ m}^3$ and the total amount of water sampled at the San Marcial Site was $105,421\text{ m}^3$.

A cumulative total of 586 Rio Grande silvery minnow eggs were collected at the two sites during 2010 (Table 2). The majority ($n = 364$; 62.1%) of the catch was taken at the San Marcial Site while the number and cumulative percent of Rio Grande silvery minnow eggs collected at the Sevilleta site ($n = 222$; 37.9%) was slightly lower. Rio Grande silvery minnow eggs were collected on 22 days at the Sevilleta Site and 15 days at the San Marcial Site. On 10 days, Rio Grande silvery minnow eggs were taken concurrently at both the Sevilleta and San Marcial sites.

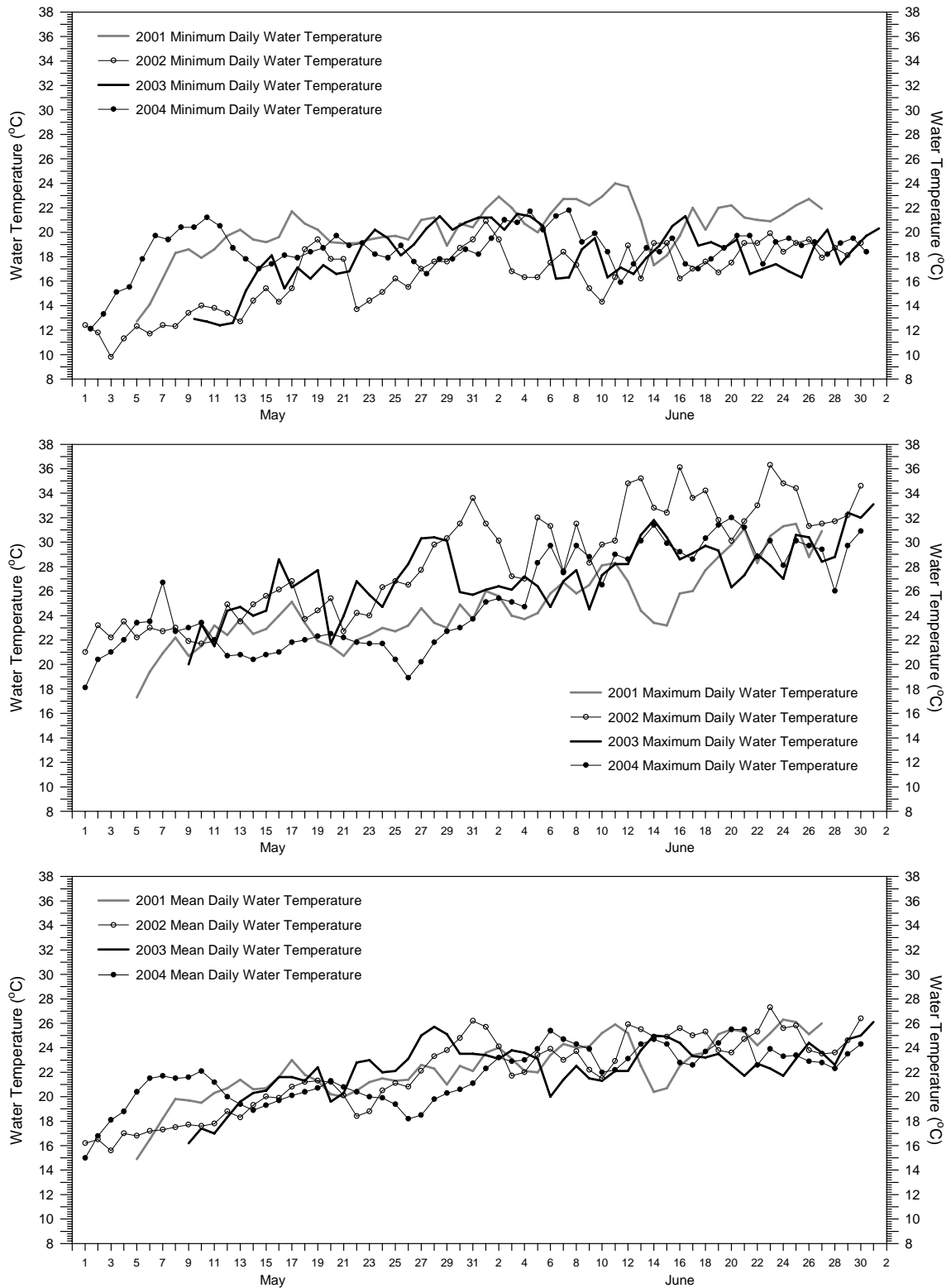


Figure 10. Annual minimum, maximum, and mean daily water temperatures at the San Marcial site during the 2001-2004 Rio Grande silvery minnow spawning periodicity study periods.

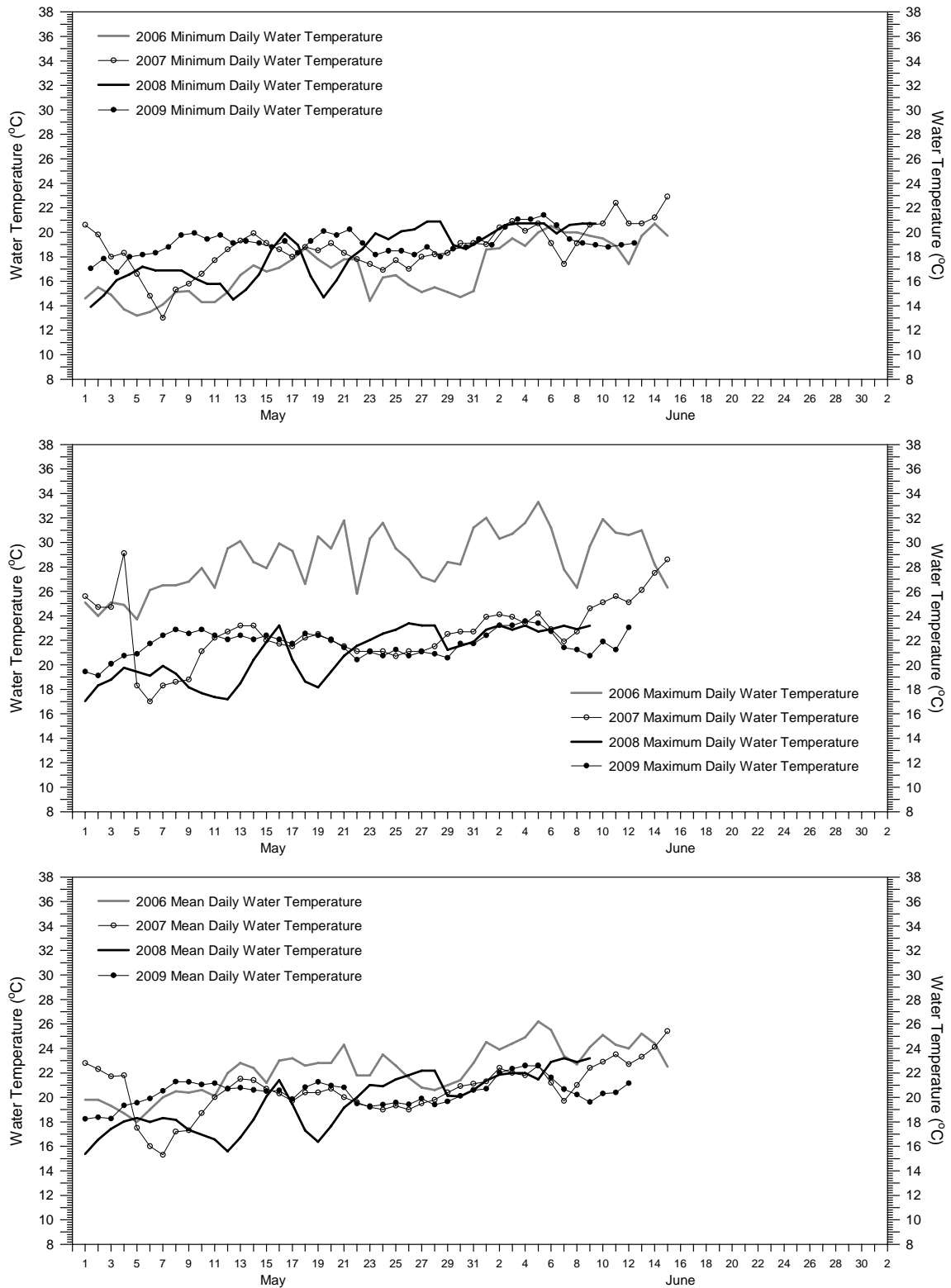


Figure 11. Annual minimum, maximum, and mean daily water temperatures at the San Marcial site during the 2006-2009 Rio Grande silvery minnow spawning periodicity study periods.

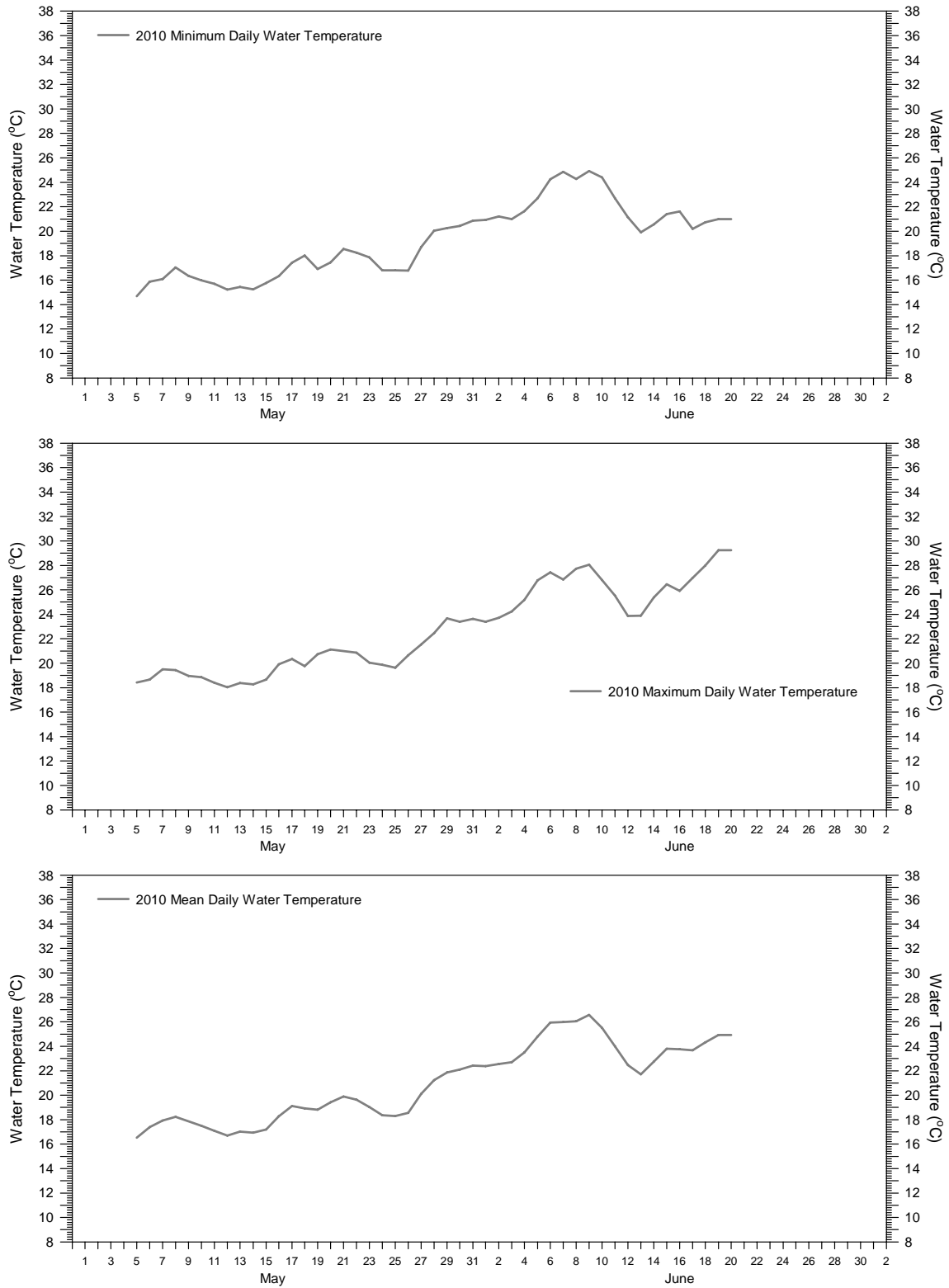


Figure 12. Annual minimum, maximum, and mean daily water temperatures at the San Marcial site during the 2009-20109 Rio Grande silvery minnow spawning periodicity study periods.

Table 1. Summary of 2010 mainstem Rio Grande sampling effort for Rio Grande silvery minnow eggs by site.

SAMPLING INFORMATION		NUMBER OF EGGS	EGG CATCH RATE ¹	VOLUME OF WATER SAMPLED (M ³)	NUMBER OF DAYS SAMPLED	DATES SAMPLED	
SYSTEM:	SITE					START	STOP
RIO GRANDE:	SEVILLETA	222	0.34	65,915	47	4 May 2010	19 JUNE 2010
RIO GRANDE:	SAN MARCIAL	364	0.35	105,421	47	4 May 2010	19 JUNE 2010

¹ Value based on number of Rio Grande silvery minnow eggs collected per 100 m³ of water sampled for all sampling days.

Table 2. Number of Rio Grande silvery minnow eggs collected per day by site. Table does not include dates that eggs *were not* collected at either site.

LOCATION : RIVER MILE :	SEVILLETA RM 121.0	SAN MARCIAL RM 58.8
4-May-10	1	0
5-May-10	2	0
6-May-10	5	0
7-May-10	3	1
8-May-10	9	2
9-May-10	3	1
10-May-10	0	1
11-May-10	7	0
12-May-10	2	1
14-May-10	1	1
15-May-10	0	1
17-May-10	0	12
18-May-10	1	223
19-May-10	0	60
20-May-10	6	0
21-May-10	125	0
22-May-10	1	1
23-May-10	33	40
24-May-10	4	0
25-May-10	0	11
26-May-10	3	8
27-May-10	1	0
30-May-10	4	1
31-May-10	2	0
2-Jun-10	1	0
7-Jun-10	1	0
9-Jun-10	7	0
TOTALS	222	364
%	37.9	62.1

Dates of egg collection ranged from 4 May to 9 June at the Sevilleta Site and 7 to 30 May at the San Marcial Site. Daily egg catch rates at the Sevilleta Site ranged between 0.04 and 5.57 eggs per 100 m³ of water sampled ($n = 1$ and $n = 125$, respectively) while daily egg catch rates at the San Marcial Site ranged between 0.03 and 9.47 eggs per 100 m³ of water sampled ($n = 1$ and $n = 223$, respectively). During the study, mean daily egg catch rates at the Sevilleta and San Marcial sites were 0.34 and 0.35 eggs per 100 m³ of water sampled, respectively (Figure 13).

The volume of water sampled at both the Sevilleta and Corral sampling sites constituted only a small fraction of the total volume available (i.e., nearly three orders of magnitude lower or about 0.1%) over each daily six hour sampling period (Table 3). The number of eggs estimated to be transported downstream of the Sevilleta Site over the duration of the study was 727,010 with a daily maximum of 365,187. The number of eggs estimated to be transported downstream of the San Marcial Site over the duration of the study was 500,755 with a daily maximum of 217,576.

Comparison of 2002-2004 and 2006-2010 studies

There were several similarities apparent regarding Rio Grande silvery minnow reproduction during 2002-2004 and 2006-2010 (Table 4). Based on the results of data taken from all years of the project, there was an apparently extended duration of spawning (April-July). However, most spawning consistently occurred during the early to middle portion of May over the period of record. The start of the spawning season varied across years and generally occurred later in upstream reaches. Spawning at the San Marcial Site was first documented during the early part of May from 2001-2004 (8 May 2001, 7 May 2002, 5 May 2003, and 6 May 2004). In 2006, the study was expanded to include upstream reaches and eggs were collected at the Albuquerque Site starting on 23 May, at the Sevilleta Site on 2 May, and at the San Marcial Site on 26 April 2006. Spawning started on 8 May 2007 at the Albuquerque Site but was documented on 4 May at the Sevilleta Site and 1 May at the San Marcial Site. A similar early spawning pattern was observed in 2008 when eggs were first collected on 10 May at the Albuquerque Site, 1 May at the Sevilleta Site, and 7 May at the San Marcial Site. In 2009, spawning was first observed on 27 April (first day of project) at the Sevilleta Site and at the San Marcial Site. In 2010, spawning was first observed on May 4 at the Sevilleta Site (first day of the project) and on May 7 at the San Marcial Site.

Mean daily water temperatures during the initial and peak spawning events were relatively similar among years (Figures 14-17). In 2001, maximum spawning of Rio Grande silvery minnow occurred when water temperatures ranged between 19-20°C while 2002 mean daily water temperatures during maximum spawning were 18-20°C. The extended Rio Grande silvery minnow spawning period in 2003 occurred at higher temperatures (20-24°C) than recorded during previous years. During the 2004 spawn (7-12 May), mean daily water temperatures were 20-22°C with maximum daily water temperatures of 21-27°C. In 2006, mean daily water temperatures during the late-May/early-June 2006 Albuquerque spawn were 22-24°C. At the Sevilleta Site, during that same period, mean daily water temperatures were 20-26°C. Mean daily water temperatures during spawning at the San Marcial Site were 18°C in late April, 23°C in mid-May, and 24°C in early June 2006. Mean daily water temperatures ranged between 20.0 and 21.3°C at the San Marcial Site during peak spawning [13-15 May] in 2007. The range of mean daily water temperatures over the full period of spawning (4 May to 10 June) was between 14.9 and 22.2°C. Mean daily water temperatures during spawning in 2008 ranged between 14.3 and 19.3°C at the Albuquerque Site, between 14.1 and 21.6°C at the Sevilleta Site, and between 18.0 and 18.3°C at the San Marcial Site. In 2009, mean daily water temperatures during spawning ranged between 16.1 and 18.9°C at the Sevilleta Site and between 17.0 and 21.3°C at the San Marcial Site. Mean daily water temperatures ranged between 16.6 and 22.1 during spawning in 2010 at the San Marcial Site.

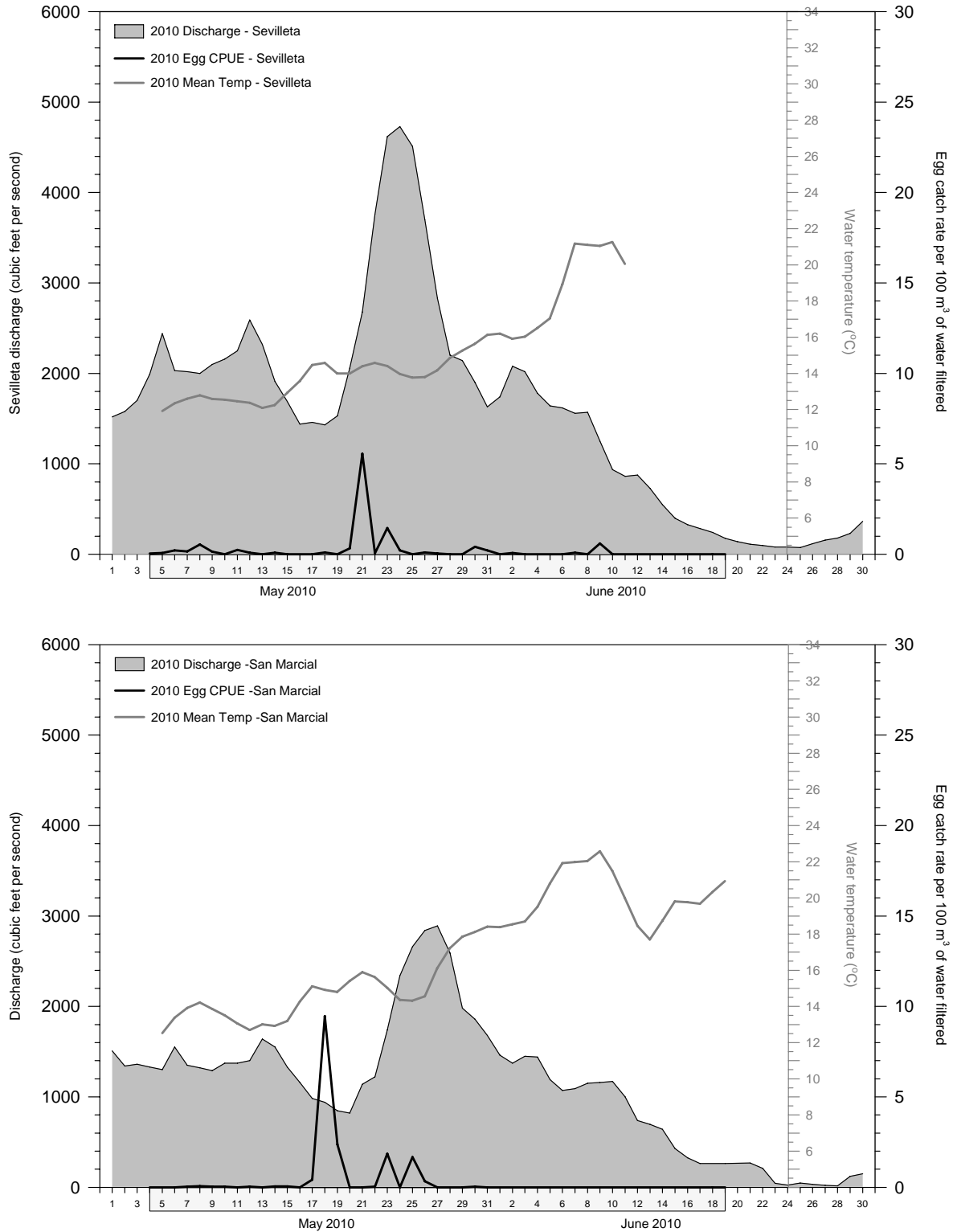


Figure 13. Mean daily discharge, mean daily egg catch rate, and mean daily water temperature during the 2010 Rio Grande silvery minnow spawning periodicity study period (sampling period is highlighted in gray along the abscissa axis).

Table 3. Volume of water sampled (6 hours), volume of water available (6 hours), and number of Rio Grande silvery minnow eggs estimated to be transported downstream of sampling location (24 hours) by date and site. Table does not include dates that eggs *were not* collected at either site.

DATA TYPE: LOCATION:	VOLUME SAMPLED (ft ³) SEVILLETA	VOLUME SAMPLED (ft ³) SAN MARCIAL	VOLUME AVAILABLE (ft ³) SEVILLETA	VOLUME AVAILABLE (ft ³) SAN MARCIAL	EGGS TRANSPORTED SEVILLETA	EGGS TRANSPORTED SAN MARCIAL
4-May-10	81,187	54,125	42,984,000	28,728,000	2,118	0
5-May-10	93,894	88,285	52,704,000	28,080,000	4,491	0
6-May-10	84,418	87,133	43,848,000	33,480,000	10,388	0
7-May-10	67,615	82,548	43,632,000	29,160,000	7,744	1,413
8-May-10	59,015	114,828	43,200,000	28,512,000	26,352	1,986
9-May-10	75,878	104,683	45,360,000	27,864,000	7,174	1,065
10-May-10	88,524	105,909	46,656,000	29,592,000	0	1,118
11-May-10	100,401	61,572	48,600,000	29,592,000	13,554	0
12-May-10	81,771	100,685	55,944,000	30,240,000	5,473	1,201
14-May-10	39,764	76,016	41,256,000	33,480,000	4,150	1,762
15-May-10	40,388	70,105	36,504,000	28,728,000	0	1,639
17-May-10	31,066	99,428	31,536,000	21,232,800	0	10,250
18-May-10	32,475	83,152	30,888,000	20,282,400	3,805	217,576
19-May-10	33,726	89,393	33,048,000	18,252,000	0	49,002
20-May-10	66,720	65,083	44,496,000	17,712,000	16,006	0
21-May-10	79,258	78,871	57,888,000	24,624,000	365,187	0
22-May-10	54,621	83,159	81,216,000	26,352,000	5,948	1,268
23-May-10	80,473	76,268	99,792,000	37,584,000	163,689	78,846
24-May-10	63,771	0	102,168,000	50,544,000	25,634	0
25-May-10	32,076	23,231	97,416,000	57,456,000	0	108,824
26-May-10	102,270	85,851	79,920,000	61,344,000	9,378	22,865
27-May-10	85,596	13,970	61,128,000	62,424,000	2,857	0
30-May-10	33,796	82,820	41,040,000	40,176,000	19,429	1,940
31-May-10	32,459	129,101	35,208,000	36,288,000	8,677	0
2-Jun-10	47,962	96,011	44,928,000	29,592,000	3,747	0
7-Jun-10	43,628	114,662	33,696,000	23,544,000	3,089	0
9-Jun-10	41,717	110,885	27,000,000	25,056,000	18,122	0
TOTALS	1,674,469	2,177,775	1,402,056,000	879,919,200	727,010	500,755
%	43.5	56.5	61.4	38.6	59.2	40.8

Table 4. Catch rates of Rio Grande silvery minnow eggs by year, site, and category (mean daily, maximum daily, and maximum sample).

SAMPLING INFORMATION			MEAN DAILY CATCH RATE ¹ (# / 100 M ³)	MAXIMUM DAILY CATCH RATE (# / 100 M ³)	MAXIMUM SAMPLE CATCH RATE (# / 100 M ³)
YEAR:	SITE				
2002	RIO GRANDE:	SAN MARCIAL	1,622.04	14,222.00	96,558.00
2003	RIO GRANDE:	SAN MARCIAL	34.85	476.00	1,027.00
2004	RIO GRANDE:	SAN MARCIAL	0.07	0.09	0.22
2005			Not Sampled	Not Sampled	Not Sampled
2006	RIO GRANDE:	ALBUQUERQUE	3.47	19.15	22.64
2006	RIO GRANDE:	SEVILLETA	4.50	44.88	53.82
2006	RIO GRANDE:	SAN MARCIAL	30.81	289.33	621.97
2007	RIO GRANDE:	ALBUQUERQUE	0.34	0.94	1.43
2007	RIO GRANDE:	SEVILLETA	13.00	147.13	158.56
2007	RIO GRANDE:	SAN MARCIAL	11.55	106.12	201.55
2008	RIO GRANDE:	ALBUQUERQUE	0.32	1.33	2.65
2008	RIO GRANDE:	SEVILLETA	5.70	76.87	136.31
2008	RIO GRANDE:	SAN MARCIAL	1.76	5.10	7.67
2009	RIO GRANDE:	SEVILLETA	3.86	24.42	38.72
2009	RIO GRANDE:	SAN MARCIAL	1.33	8.05	8.57
2010	RIO GRANDE:	SEVILLETA	0.49	5.57	10.8
2010	RIO GRANDE:	SAN MARCIAL	1.10	9.47	13.5

¹ Catch rate determinations in this table are not corrected for discharge and only incorporate daily sample totals that contained Rio Grande silvery minnow eggs.

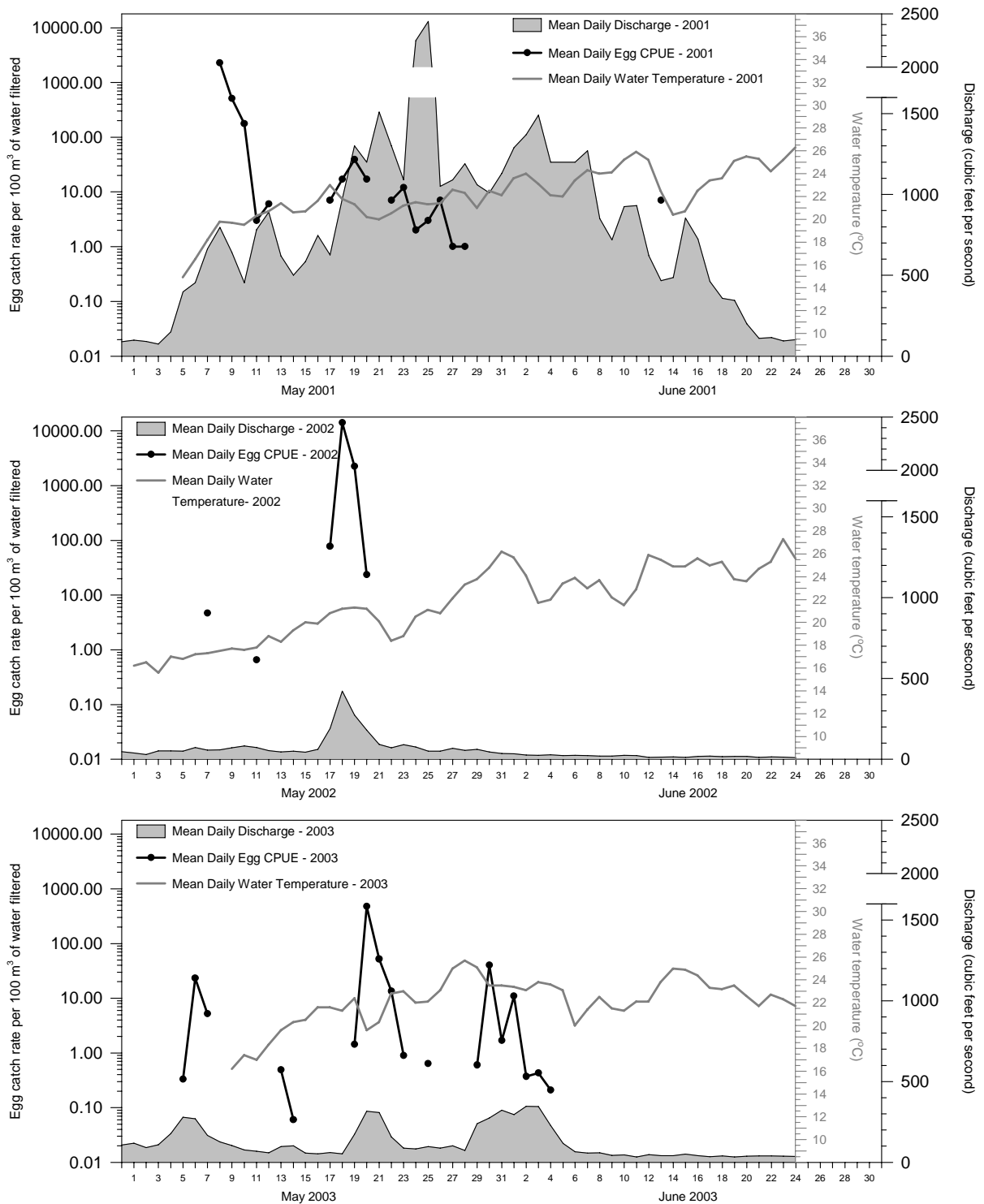


Figure 14. Mean daily discharge, mean daily egg catch rate, and mean daily water temperature during the 2001-2003 Rio Grande silvery minnow spawning periodicity study periods at San Marcial. Note that the Y-axis for egg catch rate is a log-scale.

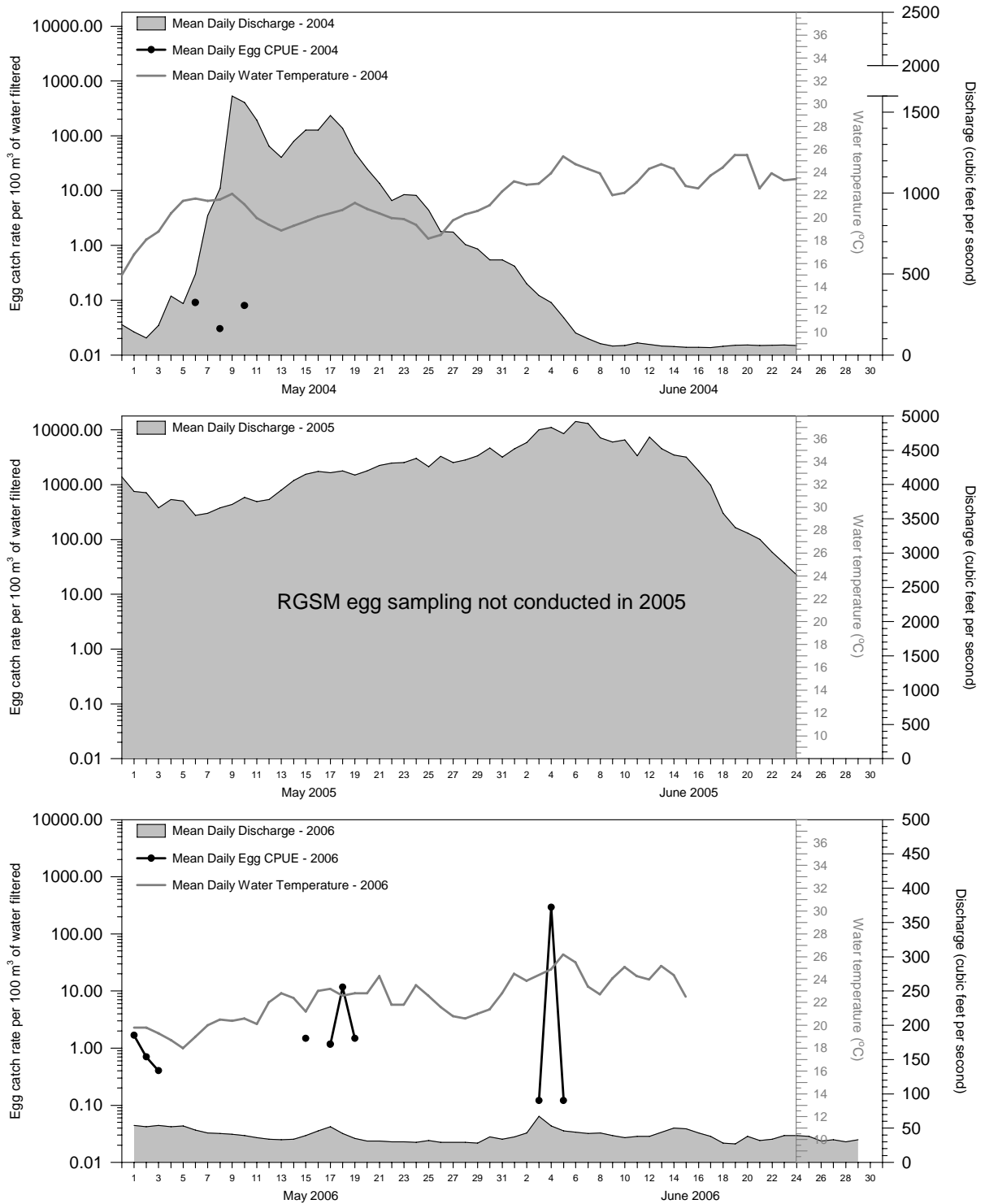


Figure 15. Mean daily discharge, mean daily egg catch rate, and mean daily water temperature during the 2004 and 2006 Rio Grande silvery minnow spawning periodicity study periods at San Marcial. Note that the Y-axis for egg catch rate is a log-scale. Sampling was not conducted in 2005.

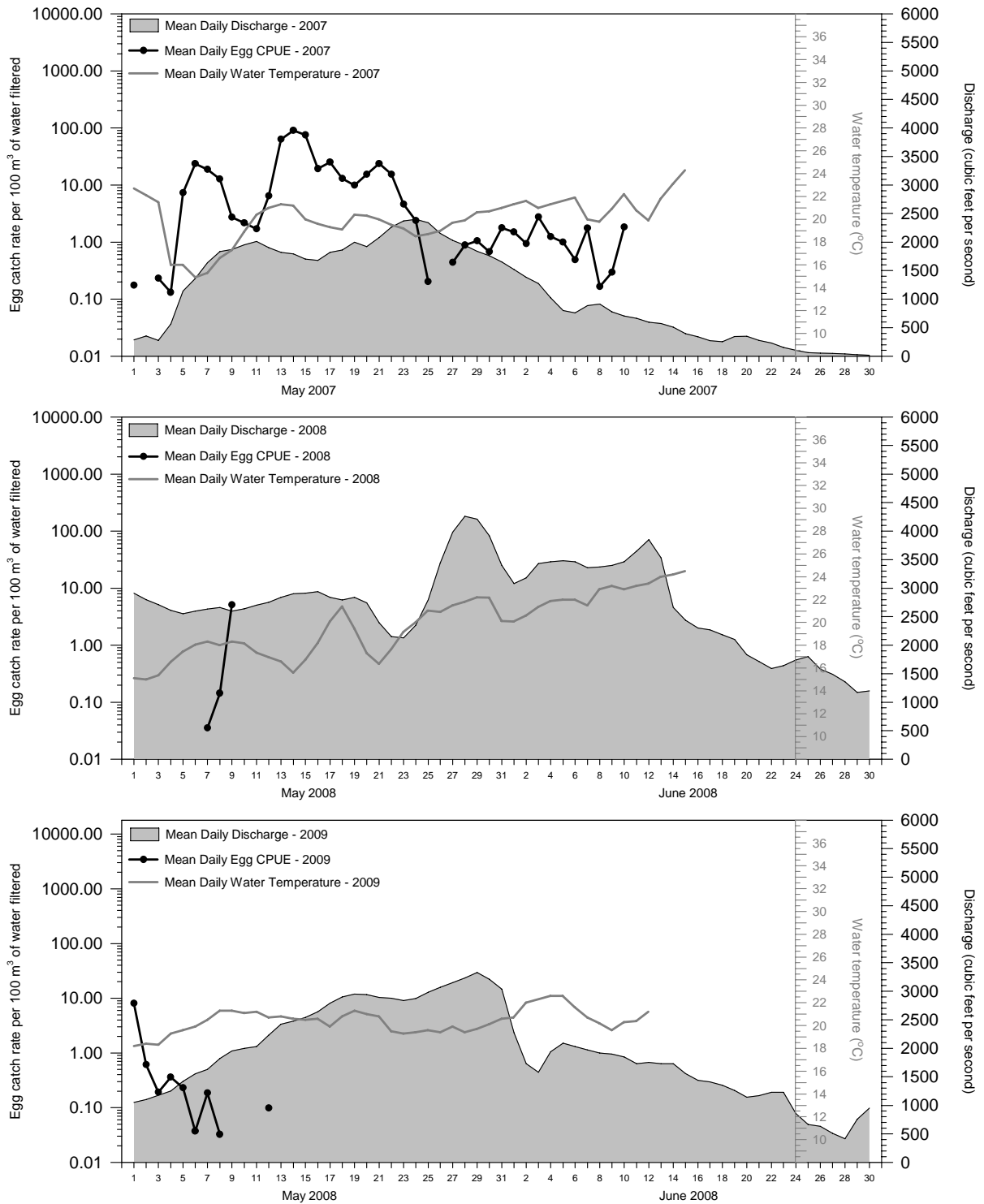


Figure 16. Mean daily discharge, mean daily egg catch rate, and mean daily water temperature during the 2007-2009 Rio Grande silvery minnow spawning periodicity study periods at San Marcial. Note that the Y-axis for egg catch rate is a log-scale.

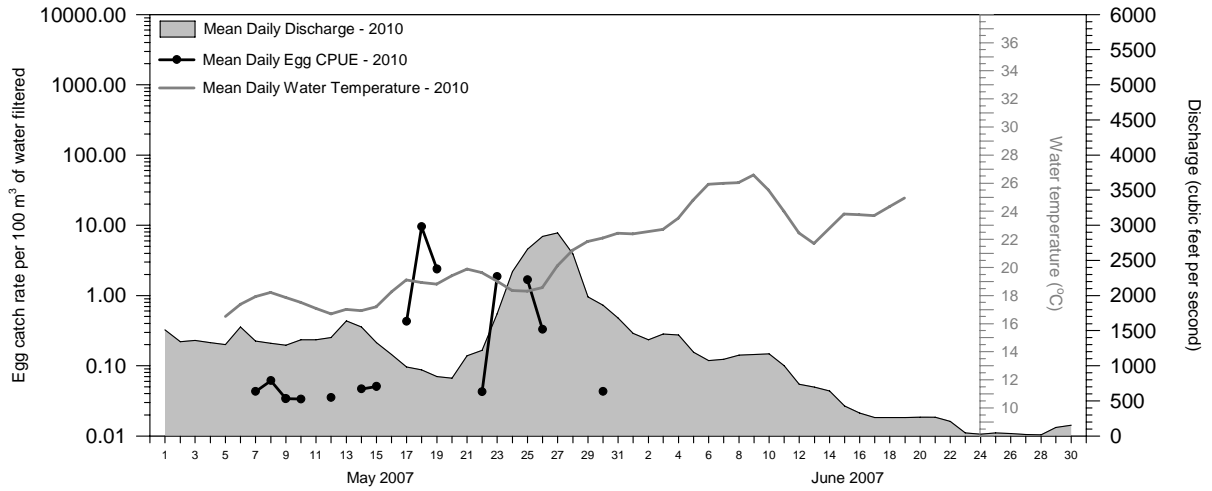


Figure 17. Mean daily discharge, mean daily egg catch rate, and mean daily water temperature during the 2010 Rio Grande silvery minnow spawning periodicity study period at San Marcial. Note that the Y-axis for egg catch rate is a log-scale.

Statistical analyses among all years were made using data from the San Marcial sampling locality since that site was the only common one for all years. Analysis of reproductive output revealed a significant difference ($F = 6.36$; $p < 0.0001$) among mean values of catch rate ($\#/100\text{m}^3$) for the eight years of the study (2002 to 2004 and 2006 to 2010). The following pair-wise comparisons were significant ($p < 0.05$) over the period of record (2002 vs. 2004, 2006, 2009, 2010; 2003 vs. 2007; 2006 vs. 2007; 2007 vs. 2009, 2010). The natural log-transformed mean egg catch rate (standardized for discharge) was highest in 2002 (5.86 ± 0.87), followed by 2007 (4.77 ± 0.34), 2008 (3.22 ± 1.22), 2003 (2.89 ± 0.50), 2009 (2.57 ± 0.59), 2010 (2.24 ± 0.55), 2006 (1.44 ± 0.67), and 2004 (0.96 ± 1.22) (Figure 18).

Additional statistical analyses for the Isleta Reach were made over the period of time that data were available (2006 to 2010). Analysis of reproductive output revealed a significant difference ($F = 9.45$; $p < 0.0001$) among mean values of catch rate ($\#/100\text{m}^3$) for the five years of the study in the Isleta Reach. The following pair-wise comparisons were significant ($p < 0.05$) over the period of record (2006 vs. 2007, 2008, 2009; 2007 vs. 2010; 2008 vs. 2010; 2009 vs. 2010) in the Isleta Reach. The natural log-transformed mean egg catch rate (standardized for discharge) was highest in 2009 (4.68 ± 0.45), followed by 2008 (4.28 ± 0.40), 2007 (4.19 ± 0.36), 2010 (2.60 ± 0.36), and 2006 (1.97 ± 0.40) in the Isleta Reach.

Comparisons of catch rates among reaches were also made for years for which those data were available (i.e., 2006 to 2010). Analysis of reproductive output revealed significant differences ($p < 0.05$) in mean values of catch rate ($\#/100\text{m}^3$) among reaches in 2006, 2007, and 2009. In 2006, the natural log-transformed mean egg catch rate (standardized for discharge) was higher ($p < 0.05$) in the Angostura Reach than in either the Isleta or San Acacia reaches. This pattern was reversed in 2007 when the natural log-transformed mean egg catch rate (standardized for discharge) was higher ($p < 0.05$) in the San Acacia Reach than in Angostura Reach. The natural log-transformed mean egg catch rate (standardized for discharge) was higher ($p < 0.05$) in the Isleta Reach than in the San Acacia Reach during 2009.

A comparison between log-transformed egg catch rate (standardized for discharge) and discharge at the Sevilleta and San Marcial sites yielded significant ($F = 11.71$; $p < 0.001$ and $F = 10.68$; $p < 0.01$, respectively) positive relationships (Figure 19). The Sevilleta Site model was derived from 97 observations while the San Acacia Site model was derived from 107 observations. The predictive relationships for the Sevilleta Site ($y = 2.43 + (4.78 \times 10^{-4})(x)$) and the San Marcial Site ($y = 2.40 + (9.41 \times 10^{-4})(x)$) had low values for the coefficient of determination ($r^2 = 0.11$ and $r^2 = 0.09$, respectively).

Similarly, a comparison between log-transformed egg catch rate (standardized for discharge) and the percentage increase in discharge (over two days) at the Sevilleta and San Marcial sites yielded significant ($F = 9.33$; $p < 0.01$ and $F = 15.42$; $p < 0.001$, respectively) positive relationships (Figure 20). The Sevilleta Site model was derived from 97 observations while the San Acacia Site model was derived from 107 observations. The predictive relationships for the Sevilleta Site ($y = 3.27 + (1.51 \times 10^{-2})(x)$) and the San Marcial Site ($y = 3.14 + (8.80 \times 10^{-3})(x)$) had low values for the coefficient of determination ($r^2 = 0.09$ and $r^2 = 0.13$, respectively).

Comparisons between log-transformed egg catch rate (standardized for discharge) and water temperature yielded a significant ($F = 10.77$; $p < 0.01$) negative relationship at the Sevilleta Site but a non-significant negative relationship at the San Marcial Site ($F = 1.20$; $p = 0.28$) (Figure 21). The Sevilleta Site model was derived from 97 observations while the San Acacia Site model was derived from 107 observations. The predictive relationships for the Sevilleta Site ($y = 8.07 - (0.24)(x)$) and the San Marcial Site ($y = 5.83 - (0.12)(x)$) had low values for the coefficient of determination ($r^2 = 0.10$ and $r^2 = 0.01$, respectively).

The complex relationships among discharge, water temperature, and log-transformed egg densities at the Sevilleta and San Marcial sites are presented as contour plots (Figure 22). The

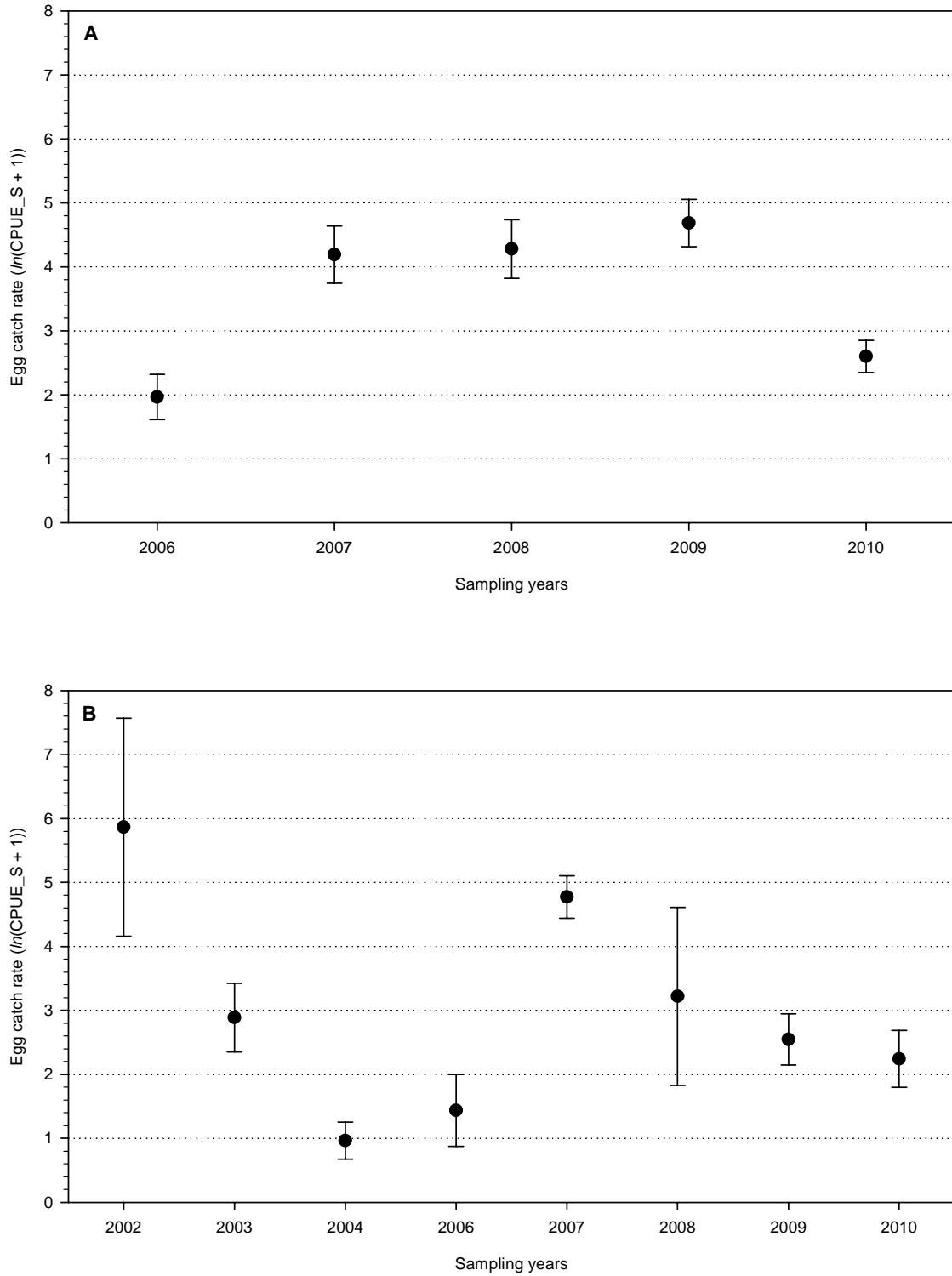


Figure 18. Log-transformed egg densities (standardized for discharge, Q) at the Sevilleta Site (A) and the San Acacia Site (B) over the period of record. Solid circles indicate means and capped-bars represent the standard error. Dotted horizontal lines represent different orders of magnitude.

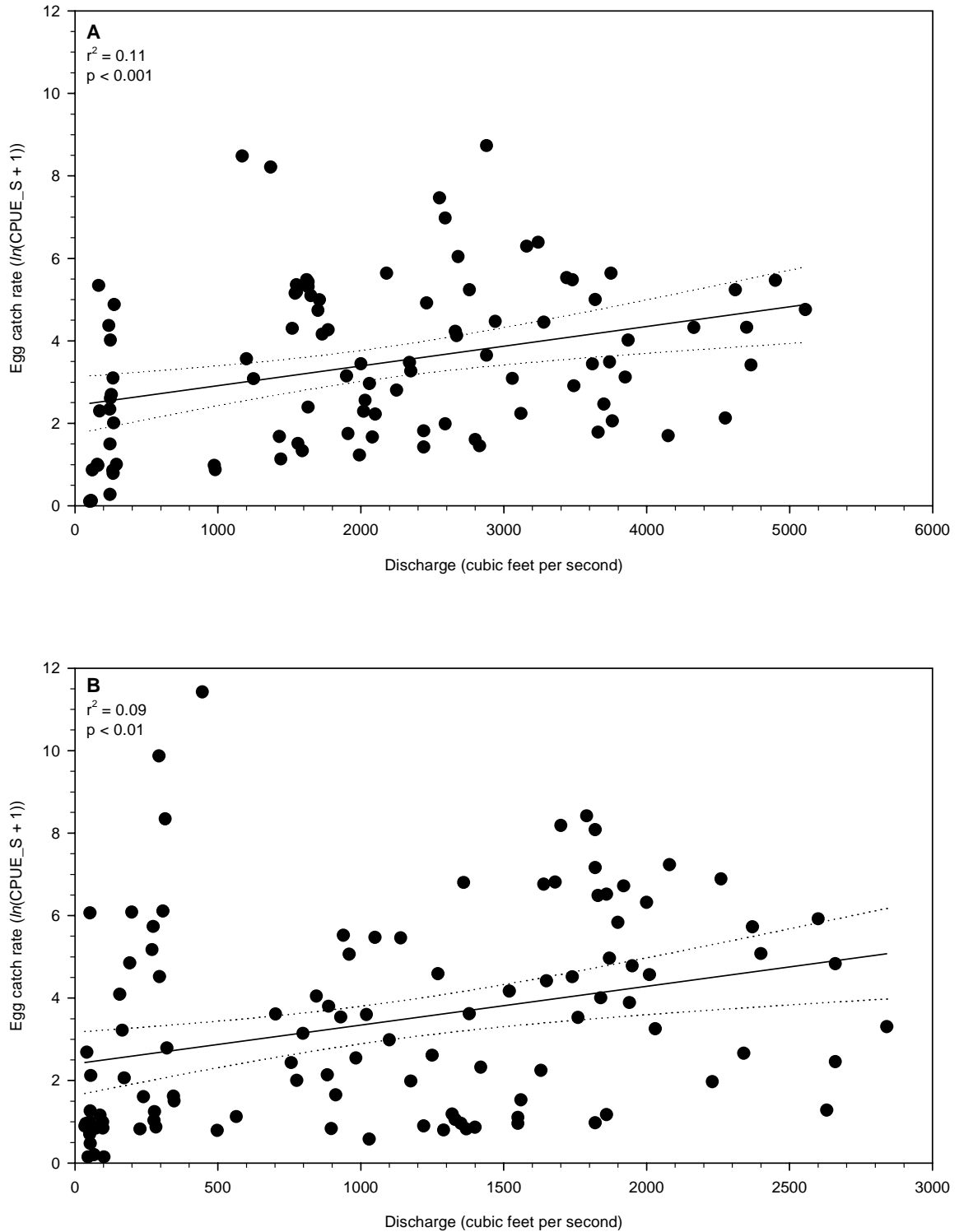


Figure 19. Regression analysis of log-transformed egg densities (standardized for discharge) and discharge at the Sevilleta Site (A), using USGS Gauge # 08331510, and the San Marcial Site (B), using USGS Gauge # 08358400, over the period of record. Graph shows regression line (solid) and 95% confidence intervals (dotted).

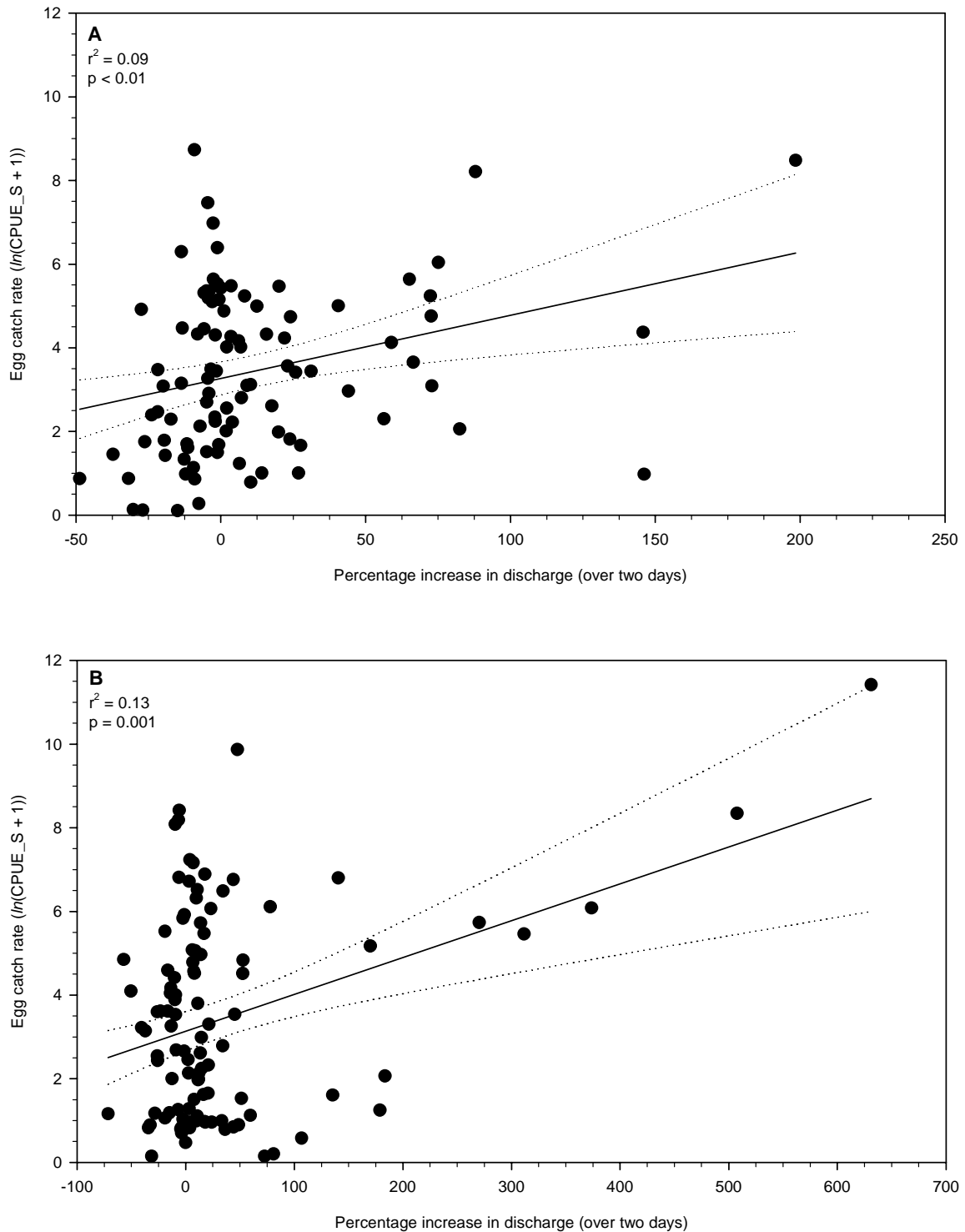


Figure 20. Regression analysis of log-transformed egg densities (standardized for discharge) and percentage increase in discharge (over two days) at the Sevilleta Site (A), using USGS Gauge # 08331510, and the San Marcial Site (B), using USGS Gauge # 08358400, over the period of record. Graph shows regression line (solid) and 95% confidence intervals (dotted).

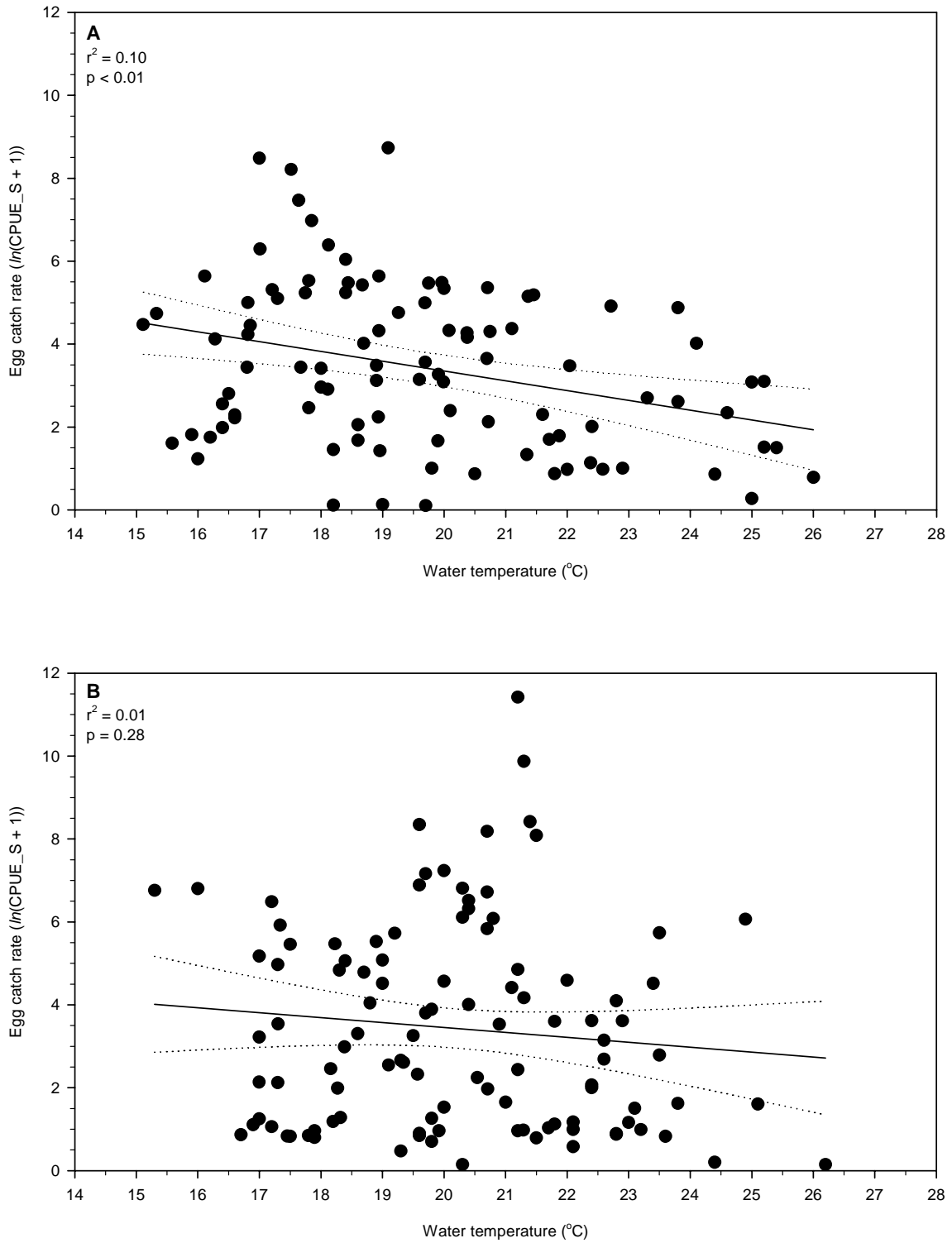


Figure 21. Regression analysis of log-transformed egg densities (standardized for discharge) and water temperature at the Sevilleta Site (A) and the San Marcial Site (B) over the period of record. Graph shows regression line (solid) and 95% confidence intervals (dotted).

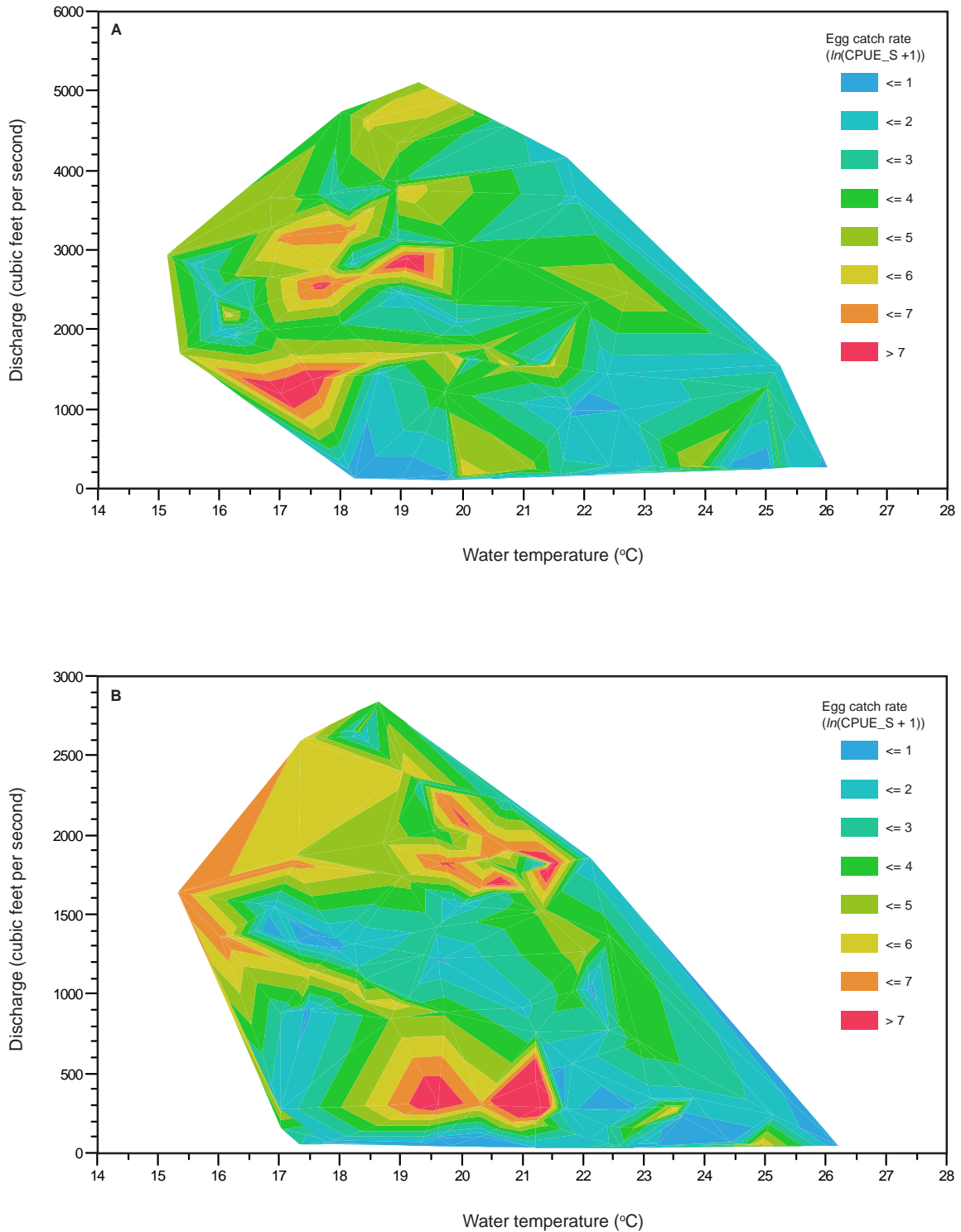


Figure 22. Contour plot of discharge, water temperature, and log-transformed egg densities (standardized for discharge) at the Sevilleta Site (A) and the San Marcial Site (B) over the period of record.

colored space shown on each graph illustrates the boundary of all possible discharge/temperature combinations, resulting in an observed spawning event, over the period of record for each site. There were some spawning events at cool water temperatures (<17°C) at either site, but only at moderate flows. At warmer water temperatures (ca. 20 to 26°C), there was a gradual decline in the flow at which spawning was observed (i.e., generally coinciding with low June discharge). There was a mottled pattern of spawning intensity across the range of discharge-temperature combinations for both sampling sites. The highest densities of eggs were collected from the Sevilleta Site between about 16.5 to 19.5°C, coinciding with flows between about 750 to 3,250 cfs. In contrast, the highest densities of eggs were collected at the San Marcial Site between about 19 to 22°C, coinciding with either low flows (ca. 200 to 600 cfs) or high flows (ca. 1,750 to 2,250 cfs).

DISCUSSION

As rivers have become increasingly fragmented, an important factor limiting the recolonization of upstream reaches and imperiling pelagic spawning cyprinids is the downstream transport of reproductive products below barriers or displacement into highly degraded downstream riverine habitats and reservoirs (Dudley and Platania, 2007b). The negative impacts of dam-related modifications of flow and habitat on Great Plains stream cyprinids that employ drifting eggs and larvae as an early life history strategy have been well documented (Stanford and Ward, 1979; Cross et al., 1983; Cross et al. 1985, Cross and Moss, 1987; Winston et al., 1991; Luttrell et al., 1999; Dudley and Platania, 2007b). In the Middle Rio Grande, large numbers of eggs of the federally endangered Rio Grande silvery minnow have been estimated to be transported past downstream collecting sites (Platania and Dudley, 2009). The downstream transport of this reproductive effort from upstream sources is potentially one factor that led to the apparent loss of Rio Grande silvery minnow from the Cochiti Reach and to its decline in the Angostura Reach in the Middle Rio Grande (Platania and Altenbach, 1998).

In addition to the problems created by river fragmentation, habitat simplification (caused by flow regulation, bank armoring etc.) also appears to contribute to the downstream displacement of the Rio Grande silvery minnow reproductive effort. The closure of Cochiti Dam resulted in a greatly reduced passage of fine sediments through the Middle Rio Grande which has, in turn, contributed to channel degradation, armoring, and narrowing (Lagasse, 1985). Arroyos, backwaters, and other "nursery habitats" may result in increased upstream retention of eggs and larvae because their off-channel location results in low or no water velocities (Porter and Massong, 2004; Pease et al., 2006). The reduction in the number and size of low velocity mesohabitats has likely reduced egg retention in upper reaches of the Middle Rio Grande.

Since Rio Grande silvery minnow is the only extant species of the previously discussed reproductive guild in the Middle Rio Grande, the species-specific identification of any semibuoyant egg collected during this study was unambiguous. The only other fish eggs that we have captured in the Middle Rio Grande during this and previous investigations were those of the common carp, *Cyprinus carpio*. Fortunately, there are numerous differences between eggs of these species that aid in identification. As the eggs of common carp are adhesive, there are usually small pieces of particulate matter attached to the chorion. Additionally, common carp eggs are smaller and more opaque than Rio Grande silvery minnow eggs, and the eyes of carp embryos become pigmented very early in development. Conversely, the eggs of Rio Grande silvery minnow are clear, nonadhesive, smooth, large, and the embryos lack discernible pigment.

Spawning of Rio Grande silvery minnow and other members in its reproductive guild (Platania and Altenbach, 1998) appears to be triggered by specific environmental cues. These fishes exhibited notably increased spawning efforts shortly following increases in flow during spring and

summer months. The peak spawning event by Rio Grande silvery minnow generally occurs soon after the initiation of runoff (often during the first two weeks of May). Egg catch rates in the Pecos River and Rio Grande appear most closely related to flow increases as opposed to absolute water volume. This relationship has been observed throughout the Middle Pecos River from early-May until late-September. Spawning was closely correlated to sharp increases in flow from local rainstorms and egg catch rates would drop as soon as flows began to drop. This sequential pattern (increased flow, increased spawning, decreased flow, decreased spawning) occurred throughout the summer in the Pecos River, NM. By late-September, the association between spawning and flow was minimal, indicating the end of the reproductive season for the five members of the reproductive guild that occupy the Pecos River.

Sampling efforts to document spawning in 2010 began on 4 May at the Sevilleta and San Marcial sampling sites. However, flows were elevated from mid to late April in both the Isleta and San Acacia reaches. It is possible that some level of spawning occurred during this period that preceded the commencement of this study. While eggs were collected on the first day of the study at the Sevilleta Site (and on 7 May at the San Marcial Site), peak spawning didn't occur until mid-May. Also, only a single larval Rio Grande silvery minnow (12 mm SL) and no early stage juveniles were collected during the May 2010 Population Monitoring Program effort (17 to 21 May). This observation suggests that recruitment was minimal if spawning did indeed occur during mid to late April.

The results of this study suggest that the number of eggs produced in the river appears to be related to some combination of discharge and water temperature conditions. Large increases in flow over a relatively short period (one to two days) seemed particularly related to increased spawning activity in Rio Grande silvery minnow. Increased flow leads to a series of changes to the physical dynamics of the river (e.g., increased water velocities/depths in some areas and augmented or new "flooded" low velocity habitats in other areas) that could be important spawning cues associated with rising flows. Additionally, there are frequently changes in water chemistry that accompany flow increases, particularly when large amounts of sediment are carried into the river from formerly dry shoreline areas, eroding banks, or flowing arroyos. The increased sediment load results in increased turbidity levels (decreased water clarity), slightly decreased water temperatures, and can lead to substantial increases in salinity levels. For example, three-fold increases/decreases in salinity levels (at the same sampling site) are regularly documented as a result of different flow conditions during spring/summer population monitoring surveys. It is possible the Rio Grande silvery minnow are spawning as a result of some combination of changing habitat and water chemistry conditions that result from these increased flow events.

While increases in discharge appear to be the primary cue for spawning, water temperature seems to be an important factor as well. Spawning appears to occur over a wide range of mean daily water temperatures (ca. 15 to 26°C) but with the majority occurring over a narrower range of temperatures (ca. 17 to 22°C). However, this interaction is quite complex and varies among years and reaches. Also, sampling has focused on May through mid-June as part of this project but spawning has been documented from late March into late June during several early years of this project (Platania and Dudley, 2000, 2002, 2003, 2004, 2005). The mean daily water temperatures during these extended periods were at the limits of the range at which spawning has been documented (i.e., 15 to 26°C).

It is possible that this range of spawning temperatures is even broader, particularly at warmer water temperatures, but there have been no longer-duration systematic studies conducted to document this possibility. Despite the lack of field spawning studies earlier or later in the year, experimental water temperature treatments on eggs and larvae of Rio Grande silvery minnow revealed that mortality was notably higher at 15°C or 30°C as compared with 20°C or 25°C (Platania, 2000). It is likely that individuals spawned earlier in the year (e.g., April) or later in the year (e.g., July), when water temperatures are at the presumed limits of their spawning range, have an

increased rate of mortality. Those individuals spawned earlier in the year might have an increased chance of early survival as compared to those spawned later in the year since there would presumably be reduced competitive pressure from other early stage larval fishes, which are generally less abundant in May as compared with June or July (Pease et al., 2006).

There are, however, multiple factors that affect survival of Rio Grande silvery minnow throughout the spring and summer, including numerous abiotic (e.g., flow, temperature, water quality) and biotic (e.g., temporal/spatial resource availability, competitive interactions, predation pressure) factors. Genetic analyses of wild Rio Grande silvery minnow eggs and adults suggested that survival was highly variable, leading to large differences in reproductive success among individuals (Osborne et al., 2005). In fact, the broad range of conditions that result in Rio Grande silvery minnow reproduction could indicate that there is no "perfect" spawning cue (i.e., combination of abiotic/biotic conditions) that would result in a consistently higher chance of early survival and recruitment into the population. The closest combination of favorable conditions, based on the last decade of spawning studies, appears to be increased flows that occur with appropriately warm water temperatures. While increased flows can, and often do, lead to the creation of new or the expansion of existing larval fish nursery habitats and presumably higher recruitment success, there is no guarantee that flows will continue to rise or remain stable after spawning. Sometimes flow in the river will briefly increase and then return to low levels either as a result of changes in ambient temperature (affecting the rate of snowmelt) or as a result of short-term precipitation events. The spawned eggs and subsequent larvae that are produced as a result of these flow events are often subjected to biotic and physical conditions that may preclude their successful growth and survivorship, particularly during summer months. Excessively elevated water temperatures (>30°C) in the Rio Grande, caused by warm ambient conditions and low flows, may reduce the hatching success of newly spawned eggs and survival of larvae. In addition to high water temperatures and possibly poor water quality, the likelihood of intra- and inter-specific interactions (e.g., predator-prey or competition) would be expected to increase during low flows as available aquatic habitat decreases. While the complex interactions among abiotic and biotic variables in the early survival, growth, and recruitment of Rio Grande silvery minnow give some insight to the patterns of their spatial spawning periodicity, there are still many unanswered questions. Future studies may illuminate some of these mysteries as additional data over a wider range of conditions and over a longer time period can fill in the gaps of our understanding.

Efforts were made in 2009 and 2010 to estimate the number of eggs transported downstream of each sampling site based on the total number of eggs collected, volume of water sampled, volume of water available to be sampled, and duration of sampling. This approach required several simplifying assumptions including 1) eggs were approximately evenly distributed within the volume of water passing the sampling site, 2) eggs collected during a six hour period of sampling in a given day approximately represented the rate at which eggs were transported downstream of the site during that day, and 3) the volume of water at the nearest upstream USGS station approximately represented the volume of water passing the sampling site. Even with modest violations of these assumptions, the number of eggs estimated to be transported downstream of sampling sites would still likely be quite high (i.e., several million during 2009 and 2010). These results indicate a substantial downstream transport of drifting eggs at the two sampling sites in the Middle Rio Grande despite the seemingly low numbers collected in individual MECs.

Population trends lend support to the observation that substantial numbers of eggs (and presumably larvae) are being transported downstream every year. In 2009, large numbers of larval Rio Grande silvery minnow were collected along the shoreline in MECs several weeks following the peak spawning events (unpubl. data). In support of these observations, the highest densities of juvenile Rio Grande silvery minnow are most frequently found in the southern reaches of the Middle Rio Grande (Dudley and Platania, 2009). The most recent population monitoring surveys (June

2010) show the majority (59.5%) of the population is located in the San Acacia Reach. This trend was first noted over two decades ago (Bestgen and Platania, 1991), before Rio Grande silvery minnow was listed as a federally endangered species, and it persists to the present time. The few exceptions to this trend (i.e., higher densities found in upstream reaches) have almost always occurred following years when flows were very low in the San Acacia Reach, resulting in moderate to substantial river drying and loss of fish in that area.

Despite the seemingly large number of Rio Grande silvery minnow propagules transported downstream every year, some portion do remain upstream. It is likely that the proportion of individuals retained and successfully recruited upstream is related to the complexity of instream habitat conditions and the long-term availability of nursery habitat. Since successful growth and survival of Rio Grande silvery minnow from the egg to the juvenile stage requires about six weeks (Platania, 1995), the persistence of these nursery habitats throughout this critical phase of development could lead to improved recruitment success (Pease et al., 2006). Years with elevated and extended spring runoff conditions appear to create the favorable habitat conditions required for the successful recruitment of early life stages of Rio Grande silvery minnow (Dudley and Platania, 2009).

The high snow pack runoff and elevated discharge that stimulates spawning by Rio Grande silvery minnow resulted in a modest spawning effort during the 2010 study period. Summer monsoonal rainstorms that began in late June and early July contributed considerable water to the Middle Rio Grande and helped maintain flow throughout the study area. Rio Grande silvery minnow appear to have had a good year for spawning and recruitment in 2010 (as compared with drought years), which could translate into increased numbers of reproductively capable females available to spawn in the spring of 2011.

While the population of Rio Grande silvery minnow appears to have stabilized since 2007, the lack of an adequately high spring runoff (high magnitude over an extended duration) combined with summer drying could result in a rapid decline to 2002/2003 population levels. The loss of individuals from downstream reaches during river drying events is particularly problematic as these are the areas that most frequently and consistently support the highest densities of Rio Grande silvery minnow (except in years immediately following downstream drying). The future conservation status of Rio Grande silvery minnow appears dependent on ensuring adequate flow and habitat conditions during the spawning and early recruitment phases of this species while also allowing upstream recolonization by individuals transported downstream.

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