Attachment C

SANTA ANA RIVER SUCKER

STUDY

FY-99 PROGRESS REPORT FOR PROJECT #98-304

Prepared for:

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Introduction

The Santa Ana sucker, *Catostomus santaanae*, is endemic to the Los Angeles, San Gabriel, and Santa Ana river drainages of southern California (Moyle 1976). Although commonly found in these drainages during the 1970s, Moyle and Yoshiyama (1992) indicated that only the San Gabriel River population is now believed to be viable and self-sustaining over multiple generations. However, recent fish surveys conducted by various government agencies or their consultants have captured this species in portions of the Santa Ana River and Big Tujunga Creek, a tributary of the Los Angeles River (Barrett 1999). An introduced population occurs in the Santa Clara River, but it has apparently hybridized with another introduced sucker (Swift et al. 1993).

In response to evidence of declining populations, FWS was petitioned by the Sierra Club Legal Defense Fund, Inc., to list the Santa Ana sucker as an endangered species (Barrett 1999). The petition and supporting documents were reviewed and, on July 9, 1996, determined to be sufficient to warrant further consideration for listing. On April 3, 1997, FWS determined that listing the Santa Ana sucker was warranted but precluded by higher listing priorities. On January 26, 1999, FWS formally proposed the Santa Ana sucker for listing as a threatened species. The ruling is scheduled to become effective on January 26, 2000, unless FWS determines that additional time is needed to complete ongoing studies of this species (G. Knowles, FWS, Carlsbad, CA, personal communication).

The purpose of this project is to develop a better understanding of environmental factors responsible for the decline of the Santa Ana sucker. Specifically, this project will attempt to identify the physicochemical variables that are most strongly associated with variations in population densities of this sucker. The general approach is to compare population densities in selected streams (or stream reaches) within the Los Angeles basin where suckers were historically present but are now absent or very rare, and where suckers are still common or abundant. Physicochemical variables such as stream discharge, water quality, and environmental contaminants will also be assessed. Key associations between sucker abundance and physicochemical variables will then be identified by using multivariate statistical procedures.

Results from this project will enable FWS biologists to better establish a scientifically defensible link between the decline of sucker populations and deteriorating environmental conditions. Data collected during this project will also provide a realistic baseline for designing future laboratory tests that estimate the tolerance thresholds of the Santa Ana sucker to selected variables. Knowledge of environmental conditions and tolerance thresholds are necessary for developing scientifically sound conservation agreements, action plans, management alternatives, etc., that protect the Santa Ana sucker and its remaining habitat.

Study Area and Methods

Duplicate sampling sites separated by a distance of at least 0.2 km were established on three stream reaches in the Los Angeles basin that historically supported or currently support endemic populations of the Santa Ana sucker (Figure 1). The stream reaches include (i) the Santa Ana River near the Metropolitan Water District's pipeline in Riverside County (about 1.8 km upstream from the Van Buren Boulevard bridge; henceforth referred to as MWD), (ii) the Santa Ana River at the Imperial Highway (California State Highway 90) bridge in Orange County (henceforth referred to as IMP), and (iii) the East Fork of the San Gabriel River adjacent to the terminus of East Fork Road in Los Angeles County (within the Angeles National Forest; henceforth referred to as SGR). According to available information, the Santa Ana sucker is expected to be very rare or absent at IMP, common at MWD, and abundant at SGR (P.J. Barrett, personal communication).

Fish measurements. Fish collections were initiated at all study sites during December 12-17, 1998, by using a battery-operated backpack electroshocker (Model 12-B electrofisher, Smith-Root, Inc., Vancouver, WA). Subsequent collections occurred at three-month intervals (March 6-12, June 17-22, and September 14-18, 1999) to comply with permit restrictions imposed by the California Department of Fish and Game. At each site, five passes (each lasting 10-15 min as determined from a voltage-activated timer on the variable voltage pulsator unit) were made with the electroshocker at the lowest output voltage setting needed to stun fish. Care

was taken not to re-fish a given area within the sampling site. Stunned fish were retrieved with wood-handled dipnets (3.2-mm mesh) and placed into 18.9-L plastic buckets filled with site water.

Immediately after completing a pass with the electroshocker, Santa Ana suckers were hand-sorted from the catch for additional processing. All other fish were identified, counted, and promptly released. Santa Ana suckers were measured (standard length, SL) and weighed (with Pesola scales); superficially examined for presence of skeletal deformities, external parasites and infectious conditions (e.g., parasitic copepods, ulcers, fin erosion, lesions), physical trauma (e.g., puncture wounds from bird pecks), and the presence of breeding tubercles (reproductive males develop tubercles over most of the body but are most dense on the caudal and anal fins and the caudal peduncle, whereas reproductive females possess tubercles only on the caudal fin and the peduncle; Moyle 1976); then returned unharmed to the water. When unidentifiable fish species were encountered, voucher specimens were preserved in 10% buffered formalin for later identification with taxonomic keys (e.g., Moyle 1976).

Length and weight data of Santa Ana suckers from the combined sites were used to determine a generalized length-weight equation by using the formula, Log W=a+b(Log SL), where "a" is a constant, "b" is the slope, WT is fish weight, and SL is standard length (Bagenal and Tesch 1978). In addition, the data were used to compute measurements of relative weight (K"), an index of fish body condition (Bagenal and Tesch 1978). Relative weight is computed from the formula, K" = WT/WT\, where WT is the actual weight and WT\ is estimated from the

generalized length-weight equation. In general, K'' >>> 1.00 indicates a fish in good body condition, K=1.00 indicates a fish in average condition, and K'' <<< 1.00 indicates a fish in poor body condition.

Water quality and other environmental measurements. The following water quality variables (also referred to as "routine variables") were measured continuously with Hydrolab DataSonde 4 multiprobe loggers (Hydrolab Corporation, Austin, TX) at 15-min intervals from near the bottom at a fixed station located at the upstream boundary of each sampling reach: water temperature, dissolved oxygen concentration, pH, specific conductance, turbidity, and ammonia concentration. These continuous measurements were taken only during daylight hours when field personnel were at a given sampling site. (Note: Although 24-hr sampling is necessary to describe diurnal variations in water quality variables, the risk of vandalism is so high that DataSondes could not be left unattended during nighttime hours.) Calibrated readings from the DataSonde were verified (for quality assurance purposes) by taking occasional readings with alternative instruments (for temperature and dissolved oxygen concentration, a YSI Model 57 temperature-oxygen meter; for pH, a Cole-Parmer Digisense pH meter; for specific conductance, a Cole-Parmer Model 9100-00 conductivity meter; for turbidity, a portable LaMotte Model 2008 turbidimeter; and for ammonia, an Orion portable ion/pH meter equipped with an Orion Model 95-12 ammonia electrode. Although not determined for this progress report, stream discharge will be roughly estimated from measurements of current velocity (taken with a Price AA current meter; Scientific Instruments, Inc., Milwaukee, WI), stream width (taken with a 30.5-

m Arizona fiberglass tape; Peco Sales, Inc., Jackson, MI), and water depth (taken with a meter stick or calibrated sounding line). Current velocity and water depth were measured at five equally spaced intervals along a cross-sectional profile of the stream channel. Color photographs were also taken at each sampling reach to supplement written descriptions of salient habitat features.

Fish-tissue contaminant measurements. In September 1999, a total of 60 Santa Ana suckers were sampled from MWD and SGR for inorganic and organic determinations. Although fishes were sampled from IMP, Santa Ana suckers were not captured. The late-summer fish collections coincided with the dry season when flows in the Santa Ana River consist almost entirely of municipal/industrial wastewater. This time period also coincided with several previous contaminant surveys (e.g., the NAWQA program of USGS).

The 60 Santa Ana suckers were rinsed in their respective site waters, then separated into 12 composites of five fish each. Six of the composites targeted for inorganic determinations were wrapped in plastic sheets (Saran Wrap), then double bagged in plastic ziplock bags. The remaining six composite samples were wrapped in acetone-rinsed aluminum foil (shiny side out), then double bagged in plastic ziplock bags. All samples were then either held on wet ice or dry ice until they could be frozen in a freezer (-10°C). The composite samples are currently stored in a freezer at the USGS-BRD facility in Dixon, CA, while awaiting shipment to an analytical laboratory.

The composite samples will be used in an exploratory attempt to determine if selected environmental contaminants are present in inordinately high concentrations. The specific list of chemical determinations will include trace elements and organic compounds (organochlorine pesticides and polychlorinated biphenyls) measured by the now defunct National Contaminant Biomonitoring Program (NCBP), which provided a nationwide baseline for concentrations that typically occur in freshwater fish (Schmitt and Brumbaugh 1990; Schmitt et al. 1990). Moisture content will also be measured in samples destined for inorganic determinations whereas lipid content will be measured in samples destined for organic determinations.

Statistical analysis. Raw data were stored as Lotus 1-2-3 files (Lotus Development Corporation, Cambridge, MA) and summarized with SAS (SAS Institute Inc., Cary, NC) or other statistical and graphical software. For this progress report, correlation analysis and comparison of 95% confidence intervals were used to identify patterns in the data and detect environmental variables that best explain variations in population densities of the Santa Ana sucker. Final selection of statistical procedures will be made after consulting with statisticians at the University of California-Davis Statistical Laboratory. Unless indicated otherwise, the probability of type 1 error for all statistical tests was specified as *P*=0.05.

Preliminary Results and Discussion

A total of 4,079 fish representing 14 species were captured during sampling trips in December 1998, March 1999, and June 1999 (Table 1). Data from September 1999 are still awaiting entry into the computerized database. Native fishes captured during the first three trips included Santa Ana sucker, arroyo chub (*Gila orcutti*), rainbow trout (*Oncorhynchus mykiss*), and speckled dace (*Rhinichthys osculus*). Only native fishes occurred in catches from SGR, whereas a mixture of native and nonnative fishes occurred at MWD and IMP.

Overall, the abundance of all fishes combined was highest at SGR and MWD (about 30 fish/10-min of electrofishing; Figure 2), whereas fish abundance was relatively low at IMP (about 10 fish/10-min of electrofishing). Speckled dace was most numerous at SGR, followed by rainbow trout, Sacramento sucker, and arroyo chub (Figure 3). By comparison, nonnative fishes were most numerous at both MWD and IMP, followed by arroyo chub. Sacramento sucker was relatively numerous at MWD but not captured at IMP.

Length frequencies of Santa Ana suckers. A total of 248 Santa Ana suckers varying in SL from 22 mm to 168 mm was captured from SGR and MWD during the three sampling trips. In general, fish were longer at SGR (mean SL, 96 mm; 95% CI, 88-106 mm) than at MWD (mean SL, 66 mm; 95% CI, 62-71 mm). In addition, at least two distinct size classes were present at SGR whereas more than one size class was not always distinguishable at MWD

(Figure 4). Nevertheless, the occurrence of small suckers at MWD indicates that the Santa Ana River population is reproducing successfully.

Chadwick Ecological Consultants (1996) suggested that reproduction by the Santa Ana sucker occurs primarily in perennial tributaries such as Anza Park Drain, Tequesquite Arroyo, and Sunnyslope Channel (located upstream from MWD) because these localities contain the types of spawning substrate (boulder/cobble/large gravel) that this species presumably requires. If Chadwick Ecological Consultants are correct, then downstream drift from tributaries could serve as a recruitment source for juvenile suckers at MWD.

Length-weight relationship and relative weight of Santa Ana suckers. The length-weight relationship of 248 Santa Ana suckers is described by the formula, Log WT= -4.447895+2.866104*Log SL (Figure 5). According to Carlander (1969), a length-weight relationship yielding a slope <3.0 indicates that small fish are plumper than larger fish (i.e., the weight of a fish decreases relative to its length as the fish grows).

On average, Santa Ana suckers were in higher body condition at SGR (mean K", 1.04; 95% CI, 1.00-1.08; N, 132) than at MWD (mean K", 0.95; 95% CI, 0.93-0.98; N, 116).

Although a statistical comparison was not attempted due to small sample size, the low body condition of fish from MWD was especially apparent among longer individuals (Figure 6).

Although corroborating evidence is not available for Santa Ana suckers, low body conditions in other fishes have been associated with poor growth (Bennett 1971; Carlander 1969), overpopulation or disease (Bennett 1971), or a temporarily low food supply (Bennett 1971). At

MWD, the seemingly poorer body condition of large suckers could be related to a shortage of animal forage. Casual observations by USGS field personnel suggested that larval and adult aquatic insects were more numerous at SGR than at MWD and IMP. According to Greenfield et al. (1970), juvenile Santa Ana suckers in the Santa Clara River fed primarily on algae, diatoms, and detritus, whereas older suckers included a significant amount of larval aquatic insects in their diets. However, Greenfield et al. (1970) also mentioned that hybridization between the Santa Ana sucker and the Owens sucker (*Catostomus fumeiventris*) makes it possible that life history information gathered on suckers from the Santa Clara River may differ from that of Santa Ana sucker populations in other drainages.

Water quality and other environmental characteristics. Physicochemical conditions varied considerably among the three sampling sites. At SGR, the habitat consists of a relatively high gradient stream flowing over a mostly coarse gravel-cobble-boulder substrate with numerous pools, riffles, and runs in a narrow steep-sided canyon containing little riparian vegetation. However, even the deepest pools rarely exceed 1 m in depth. In general, this site is characterized by cool, clear, well-oxygenated waters. Specific conductance (a measure of the dissolved mineral content) of water from this site is relatively low (Table 2). By comparison, MWD consists of a relatively wide and shallow low-gradient run (no pools or riffles) flowing over a shifting-sand substrate, with extensive riparian vegetation (mostly giant reed, cattails, and some willow) growing along the shores. Water depth is very shallow, averaging about 0.3 m deep or less. The water at this site is warmer than at SGR and IMP, but it is still well oxygenated

(Table 2). In addition, turbidity and specific conductance at MWD are intermediate between conditions occurring at SGR and IMP. At IMP, the reach consists of a channelized (rip-rapped) low-gradient stream flowing over a sandy or gravelly substrate with some riffles, pools, and shallow backwaters that lack riparian vegetation. Water temperature is warmer at IMP than at SGR but cooler than at MWD (Table 2). Although water at IMP is well oxygenated, it is more turbid and has a higher specific conductance than water from the other two sites.

Relation of Santa Ana sucker abundance to selected environmental variables.

According to results from Pearson product-moment correlation analysis, the relative abundance of Santa Ana suckers is not associated with water temperature, dissolved oxygen concentration, pH, electrical conductance, and turbidity (Table 3). Moreover, when compared with the abundance of other fish species, significant correlations occurred only with nonnative species such as bluegill (*Lepomis macrochirus*), largemouth bass (*Micropterus salmoides*), and common carp (*Cyprinus carpio*). In such instances, inverse associations were detected, indicating that the Santa Ana sucker was most numerous when nonnative species were least abundant. These findings agree with the concern expressed by some fishery managers (e.g., Moyle and Yoshiyama 1992; Barrett 1999) that predation and competition from nonnative fishes are likely to "...threaten the continued existence of Santa Ana suckers throughout most of the species' range." According to results from stepwise multiple regression analysis, common carp and bluegill (the only independent variables retained by the statistical model; Table 4) collectively accounted for nearly 88% of the variation in abundance of Santa Ana suckers.

Recommendations

The preliminary indications that Santa Ana suckers at IMP are either very rare or absent (no individuals have been captured) and that Santa Ana suckers from MWD are generally smaller and in poorer body condition than those from SGR strongly suggest that the Santa Ana River study sites represent substandard habitat for this species. However, future work should focus on verifying these results and determining if suckers from the Santa Ana River exhibit evidence of poor health or reduced survivability as determined by an appropriate combination of physiological, behavioral, and biochemical measures of fitness (e.g., swimming performance, predator avoidance, lipid content [a measure of nutritional status]). If suckers from the Santa Ana River are indeed suffering from poor health/survivability, the next step would be to identify and conduct cause-effect studies linked to specific environmental variables or combinations of the variables. Concurrently, other studies should be conducted to identify and quantify the geographic locations within the watershed used by suckers for spawning, rearing, and other necessary life history requirements. Collectively, these studies should yield the information needed to design restoration plans and designate critical habitats.

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Table 1. Occurrence of fish species from the East Fork of the San Gabriel River (SGR), the Santa Ana River at the Metropolitan Water District Pipeline Crossing (MWD), and the Santa Ana River at Imperial Highway (IMP). N=4,079 fish.

Family	Species	SGR	MWD	IMP
Catostomidae	Santa Ana sucker, Catostomus santaanae ¹	X	X	
Centrarchidae	Green sunfish, Lepomis cyanellus		X	
	Bluegill, Lepomis macrochirus			X
	Largemouth bass, Micropterus salmoides		X	X
Cichlidae	Redbelly tilapia, <i>Tilapia zilli</i>		X	
Cyprinidae	Common carp, Cyprinus carpio			X
	Arroyo chub, Gila orcutti ¹	X	X	X
	Fathead minnow, Pimephales promelas		X	X
	Speckled dace, Rhinichthys osculus ¹	X		
Ictaluridae	Yellow bullhead, Ameiurus natalis		X	X
	Brown bullhead, Ameiurus nebulosus		X	X
	Channel catfish, Ictalurus punctatus			X
Poeciliidae	Western mosquitofish, Gambusia affinis		X	X
Salmonidae	Rainbow trout, Oncorhynchus mykiss ¹	X		

¹Native species.

Table 2. Comparison of selected water quality variables from the East Fork of the San Gabriel River (SGR), the Santa Ana River at the Metropolitan Water District Pipeline Crossing (MWD), and the Santa Ana River at Imperial Highway (IMP). Values are geometric means (95% confidence intervals in parentheses).

Variable	SGR	MWD	IMP
Water temperature (°C)	15.0 (14.2-15.8)	19.2 (18.6-19.8)	16.4 (15.7-17.1)
Dissolved oxygen (mg/L)	9.7 (9.6-9.9)	8.6 (8.5-8.7)	9.3 (9.1-9.4)
pН	8.35 (8.21-8.49)	8.18 (8.15-8.21)	8.20 (8.18-8.21)
Electrical conductance (mmhos/cm @ 25°C)	0.32 (0.31-0.34)	0.93 (0.92-0.95)	1.01 (0.99-1.03)
Turbidity (NTUs)	1.4 (1.1-1.9)	6.2 (4.4-8.8)	79.7 (70.1-90.7)

Table 3. Pearson's product-moment correlations between the relative abundance of Santa Ana suckers and selected physicochemical and biological variables. Codes: $*, P \le 0.05$; $**, P \le 0.01$.

Variable		Correlation coefficient, r
Physicochemical:	Water temperarature	0.01
	Dissolved oxygen	-0.02
	рН	0.15
	Electrical conductance	-0.53
	Turbidity	-0.66
Biological:	Green sunfish	0.52
	Bluegill	-0.84**
	Largemouth bass	-0.79*
	Redbelly tilapia	0.39
	Common carp	-0.86**
	Arroyo chub	0.38
	Fathead minnow	-0.49
	Speckled dace	0.47
	Yellow bullhead	0.19
	Brown bullhead	-0.30
	Channel catfish	-0.47
	Western mosquitofish	0.17
	Rainbow trout	0.48

Table 4. Relation of relative abundance of Santa Ana suckers to physicochemical and biological variables retained by a stepwise multiple regression model. (Note: See Table 3 for the complete list of variables.)

Variable	Partial r ²	Model r^2	F	P
Common carp	0.7372	0.7372	19.64	0.0030
Bluegill	0.1423	0.8795	7.08	0.0375

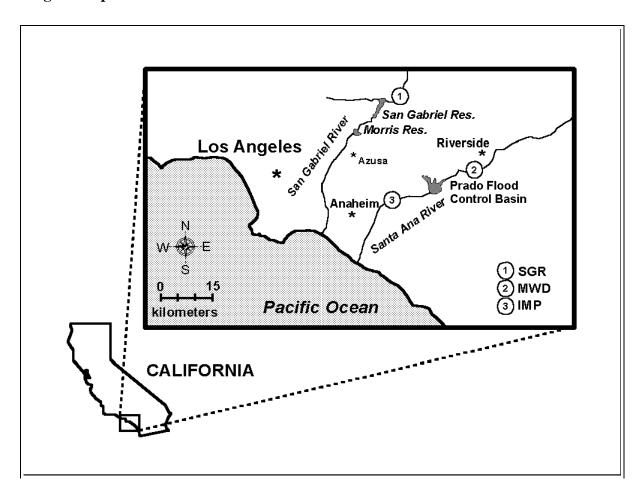


Figure 1. Map of the study area showing locations of sampling sites on the East Fork of the San Gabriel River (SGR), the Santa Ana River at the Metropolitan Water District Pipeline crossing (MWD), and the Santa Ana River at Imperial Highway (IMP).

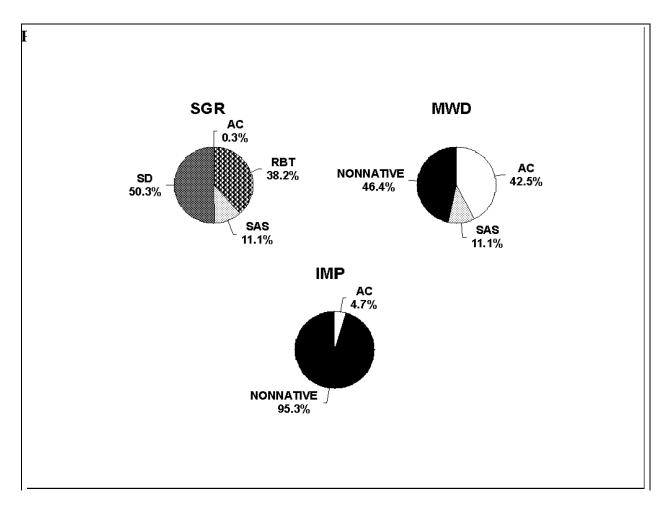


Figure 2. Composition of fish species assemblages as determined by electrofishing surveys from sampling sites on the East Fork of the San Gabriel River (SGR), the Santa Ana River at the Metropolitan Water District Pipeline crossing (MWD), and the Santa Ana River at Imperial Highway (IMP). Codes: nonnative, all nonnative species combined; SD, speckled dace; SAS, Santa Ana sucker; RBT, rainbow trout; and AC, arroyo chub.

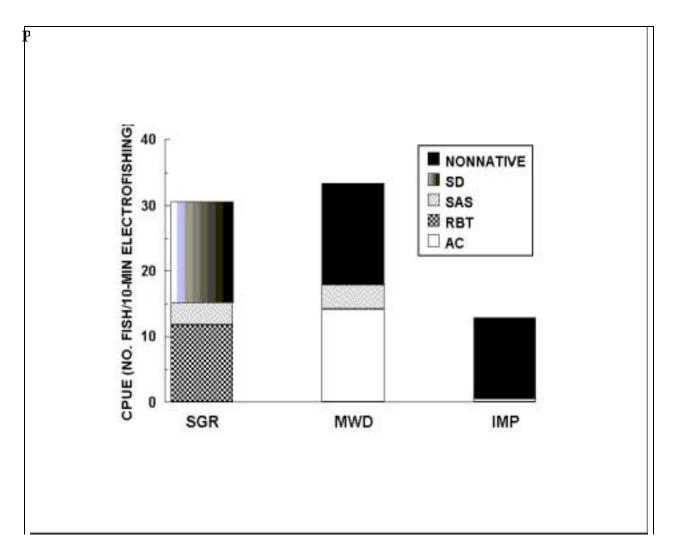


Figure 3. Relative abundance of native and nonnative fishes to the total catch at sampling sites on the East Fork of the San Gabriel River (SGR), the Santa Ana River at the Metropolitan Water District Pipeline crossing (MWD), and the Santa Ana River at Imperial Highway (IMP). Codes: nonnative, all nonnative fish species combined; SD, speckled dace; SAS, Santa Ana sucker; RBT, rainbow trout; and AC, arroyo chub.

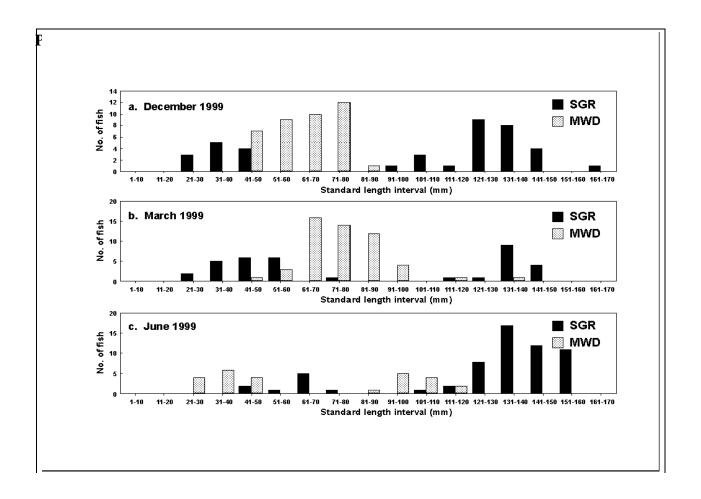


Figure 4. Length frequencies of Santa Ana suckers captured by electrofishing from sampling sites on the East Fork of the San Gabriel River (SGR) and the Santa Ana River at the Metropolitan Water District Pipeline crossing (MWD). Suckers were not captured from the Santa Ana River at Imperial Highway.

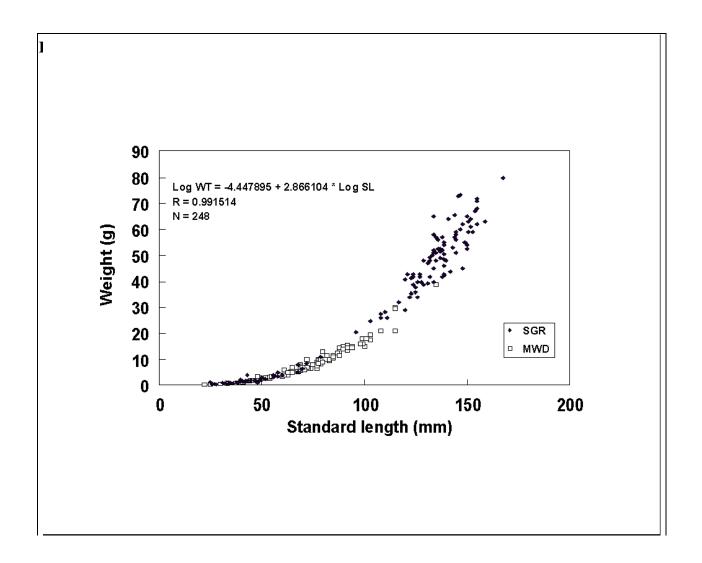


Figure 5. Length-weight relationship of Santa Ana suckers captured by electrofishing from sampling sites on the East Fork of the San Gabriel River (SGR) and the Santa Ana River at the Metropolitan Water District Pipeline crossing (MWD).

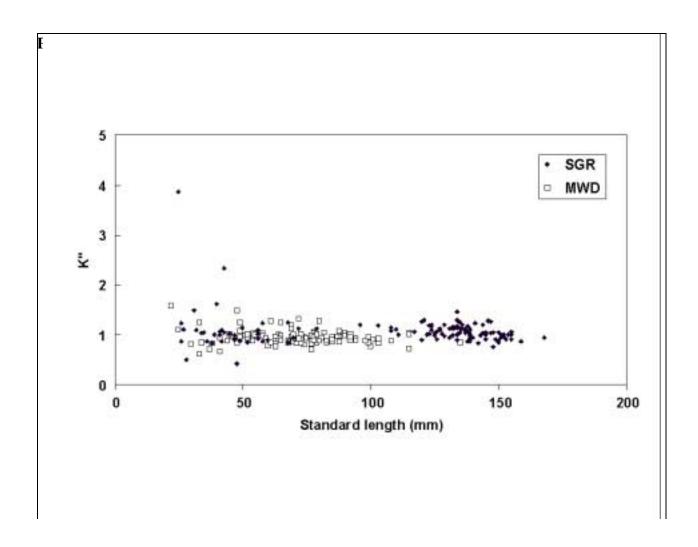


Figure 6. Relation of relative weight (K") to standard length of Santa Ana suckers captured by electrofishing from sampling sites on the East Fork of the San Gabriel River (SGR) and the Santa Ana River at the Metropolitan Water District Pipeline crossing (MWD).