# RESULTS OF FISH PASSAGE MONITORING AT THE VERN FREEMAN DIVERSION FACILITY, SANTA CLARA RIVER, 1996

Prepared for:

UNITED WATER CONSERVATION DISTRICT Santa Paula, CA

Prepared by:

**ENTRIX, Inc.** Walnut Creek, CA

Project No. 324402

December 26, 1996

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Prepared for:

UNITED WATER CONSERVATION DISTRICT 725 E. Main Street, Suite 30 Santa Paula, California 93061

Prepared by:

ENTRIX, Inc. 590 Ygnacio Valley Road, Suite 200 Walnut Creek, California 94596

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cutting of the river bed also contributed to repeated failures of the previous sand dike diversion structure. The permanent concrete structure, completed in 1991, has since halted the headcutting, stabilized the river bed both upstream and downstream of the project, and improved the ability of UWCD to divert streamflow to groundwater recharge basins.

The project was permitted through U.S. Army Corps of Engineers (COE) 404 Permit No. 86-116-T5. The Freeman Diversion also includes a two-entrance denil fish ladder, a fish screen, and by-pass facilities as described in Special Condition A of the COE 404 permit. Special Condition B of the 404 permit focuses on the fisheries mitigation features of the project, and states:

"B. The District shall institute a plan for evaluation of the mitigation features of the project to determine their effectiveness at accomplishing their designated purpose. This evaluation process may include studies on fish movement, flows and timing and will be conducted for a period of 5 years after the project is completed. The plan is to be developed by the District within 18 months of permit issuance and is to be approved by the COE in consultation with the involved resource agencies. The implementation of the plan shall include the installation of some functional, mutually agreeable device for counting fish passage through the ladder."

An appropriate study plan (ENTRIX 1991) to monitor steelhead utilization of the fish ladder was developed and approved by the California Department of Fish and Game (CDFG), United States Fish and Wildlife Service (USFWS) and the COE. The plan centered on the installation of a semi-permanent fish trap and counting device that became operational in February of 1993. The counting tubes (described in ENTRIX (1994 and 1995) were installed with the hope that adult steelhead migrating through the fish ladder would be automatically counted, thus eliminating the need for the trap. However, the counting tubes, operated during the 1994 and 1995 sampling seasons, did not operate as planned. The Santa Clara River transports a high debris load during periods of relatively high streamflow. The passage of debris through the tubes apparently resulted in several erroneous counts (i.e., a count registered on the counting device that was not related to the passage of an upstream migrating steelhead as evidenced by the absence of trapped fish). In addition, adult lamprey migrating upstream apparently pass through the tubes, also resulting in counts. These factors resulted in a high number of "non-steelhead" counts. Thus, the number of adult steelhead passing upstream could not be assessed by use of the counting device. As a result, the tubes were removed at the beginning of the 1996 sampling season. Prior to the installation of the semi-permanent fish trap and counter, upstream fish migration was monitored with a temporary trap (described in ENTRIX 1991) in 1991 and 1992. A number of details pertaining to the operation of the semi-permanent fish trap and counting device required refinement after installation. As a result, an agreement was reached between UWCD and CDFG to view the 1993 trapping season as a preliminary year to work out the details of the operating

The 1996 monitoring season marks the third year of a five year study monitoring the upstream movement of adult steelhead and the downstream movement of juvenile steelhead past the Vern Freeman Diversion Facility. The 5-year monitoring study began with the 1994 trapping season (winter 1993-1994), and will continue through the 1998 trapping season. This report presents the results of the 1996 trapping season, and discusses the 1996 results in relation to the previous data collected. Limited observations of fish in the upstream fish ladder were also made between 1991 and 1993. Pertinent data collected prior to 1996 will be presented in tables to facilitate this discussion. The results of the 1991 through 1995 trapping seasons are presented in previous reports (ENTRIX 1993, 1994 and 1995).

#### 1.1 BACKGROUND

The Santa Clara River is intermittent and drains portions of Los Angeles and Ventura counties in southern California. The mainstem Santa Clara River flows through a narrow alluvial valley onto a large coastal plain, and is fed by several tributaries that flow out of local mountains. The major tributaries are Santa Paula, Sespe and Piru creeks. Streamflow is typical of most southern California rivers; extremely low (often zero) during the dry summer and fall months, but can experience relatively high peak flows during winter storms. During the low flow period, a sand bar forms at the mouth of the Santa Clara River estuary, forming an intermittent barrier to fish migration to or from the ocean. Fish also are prevented from migrating through the lower Santa Clara River until sufficient rainfall in the basin provides adequate streamflow for passage.

The Santa Clara River supplies water primarily through groundwater recharge for domestic, municipal and agricultural uses in the basin. Water is diverted at Saticoy, approximately 16.8 kilometers upstream from the ocean, by the United Water Conservation District (UWCD) into a series of percolation ponds to recharge the Oxnard Coastal Plain aquifers. Prior to 1989, the diversion consisted of an earthen dike. In 1991, the UWCD constructed the Freeman Diversion Improvement Project to improve the existing diversion works on the Santa Clara River. This action was taken at the direction of the State Water Resources Control Board to combat sea water intrusion in the Oxnard Coastal Plain aquifers. This intrusion results from overdraft of groundwater to supply water for irrigation, industry, and municipal uses. The improvement consists primarily of a permanent concrete riverbed stabilization structure and diversion. These were necessary for the UWCD to maintain its ability to divert water to groundwater recharge basins in the Oxnard Plain Forebay Basin. Historical in-river aggregate mining destabilized and degraded the Santa Clara River bed, which had lowered approximately 22 feet opposite the diversion headworks since 1928, when diversions began. This downprocedures. The five year monitoring phase began with the 1994 water year. Accordingly, this report documents the third year of the required five year study.

#### 1.2 SPECIES COMPOSITION

The fish assemblage in the Santa Clara River is comprised of five native species (including two subspecies of stickleback), four species native to southern California streams that have been introduced into the system, and several species that are not native to California, but have been introduced into the system. Fish native to the Santa Clara River Basin include steelhead (rainbow) trout (Oncorhynchus mykiss), Pacific lamprey (Lampetra tridentata), partially armored threespine stickleback (Gasterosteus aculeatus microcephalus) and the unarmored threespine stickleback (G. a. williamsoni), a federally and state listed endangered species. The tidewater goby (Eucyclogobius newberryi) (also a federally listed endangered species) and Pacific staghorn sculpin (Leptocottus armatus) are primarily estuarine species which are seldom found upstream in freshwater habitats.

Fish found in the Santa Clara river that are native to southern California, but not to the Santa Clara River system include Santa Ana sucker (Catostomus santaanae) and the arroyo chub (Gila orcutti). The native range of the arroyo chub and the Santa Ana sucker is the Los Angeles basin. Two additional species native to California have established populations in the Santa Clara River, the Owens sucker (C. fumeiventris) and the prickly sculpin (Cottus asper). The Owens sucker, native to the Owens River, was apparently introduced through the transfer of water (through the Los Angeles Aqueduct) into the basin. The prickly sculpin is native to coastal streams as far south as the Ventura River (the next drainage to the north), but the population in the Santa Clara River is thought to have entered the basin through transfer of water (through the State Water Project into Pyramid Lake from the Sacramento River system) (Bell 1978). Several additional species have been stocked into reservoirs throughout the basin and are occasionally found in the river, including threadfin shad (Dorosoma peteneses), bullhead (Ameiurus spp.) and green sunfish (Lepomis cyanellus), as well as hatchery-reared rainbow trout. Although the mainstem Santa Clara River provides habitat for several of the fish species listed above, the lower Santa Clara River (in the vicinity of the Vern Freeman Diversion Facility) serves primarily as a migration corridor for steelhead and lamprey. Tributaries provide most of the spawning and rearing habitat (Puckett and Villa 1985). No federally or state protected species of fish inhabit the Santa Clara River in the vicinity of the Vern Freeman Diversion.

#### 1.2.1 STEELHEAD

Maintaining the steelhead trout population was the impetus for the construction of the fish ladder past the Vern Freeman Diversion. The Santa Clara River historically supported steelhead, although the size of the population was never quantified. Steelhead (and lamprey) use the lower Santa Clara River as a migration corridor and do not spawn or rear in this portion of the river (Puckett and Villa 1985). Spawning and rearing of the young takes place in upstream tributaries. Historically, Santa Paula, Sespe and Piru

creeks were utilized as the primary spawning and rearing habitat, with several smaller streams also providing habitat.

The decline in the steelhead population in the Santa Clara River is probably related to a number of factors, including, development, loss of habitat, planting of hatchery fish, drought, and ocean conditions. The old Vern Freeman Diversion may have impeded upstream migration and entrained emigrating smolts. In Santa Paula Creek, the Santa Paula Diversion blocks upstream access and reduces or eliminates flow downstream of the diversion during the dry season. Santa Felicia Dam blocks upstream access on Piru Creek approximately 10 kilometers upstream of its confluence with the Santa Clara River. A minimum release of five cfs is maintained at the dam which may provide spawning and rearing habitat for steelhead in the lower creek. Sespe Creek has historically been heavily stocked with hatchery-reared rainbow trout, but remains substantially undeveloped and probably provides the best steelhead spawning and rearing habitat in the basin. Severe droughts can reduce the quantity and quality of spawning and rearing habitat in upstream tributaries. In addition, streamflow necessary to breach the sand bar at the mouth of the river and to provide the streamflow necessary to allow for passage to and from the ocean may be reduced or absent during dry years. The steelhead population in the Santa Ynez River experienced a large scale decline after the drought of 1947-1951 (the second driest five year period since 1890) (Bedwell 1952, Lantis 1967, ENTRIX 1995). In comparison, the drought of 1987-1991 was the third driest five year period since 1890. Oceanic conditions, most notably El Niño events, may also affect survival of juvenile and adult steelhead (McEwan and Jackson 1996). During El Niño events, coastal water temperature increases and productivity decreases. Thus, juvenile steelhead may need to migrate further offshore to find suitable conditions. In addition, adult steelhead would have to migrate back through the relatively warm, unproductive waters to reach their natal rivers.

2.0 Methods

The study design for this program was presented in ENTRIX (1991). The study was designed to monitor the upstream (adult) and downstream (juvenile) migrations of steelhead trout through the fish ladder. In 1993, a semi-permanent fish trap was installed in the fish ladder. Prior to 1993, a temporary trap and fyke net were placed in the ladder during high flow events to monitor fish movement upstream through the diversion facility. Under agreement with the CDFG, USFWS and COE, the fish ladder is to be operated throughout the upstream migration period in the Santa Clara River at the Vern Freeman Diversion provided certain headwater elevation criteria are met (i.e., the headwater elevation is between 160-164 feet per the COE 404 permit). The upstream ladder is closed on occasions when streamflows exceed 20,000 cfs, and when turbidity readings exceed 2,000 NTU's (in 1997, the turbidity level for restarting the fish ladder will be increased to 3,000 NTU's). Closing the ladder during these periods reduces build up of sand and debris at the fish trap and allows for more consistent and efficient ladder operation. The ladder may also be closed 48 hours after streamflow above the diversion declines below 415 cfs, and at the end of the 48 hour period, streamflow has further declined below 375 cfs.

#### 2.1 UPSTREAM MIGRATION

A denil type fish ladder provides access for upstream migrating fish around the diversion structure. During periods of high streamflow, a relatively high velocity current is required to attract upstream migrating fish into the fish ladder. The water surface elevation inside the fish ladder (at the downstream fish entrance) is maintained 1.5 feet higher than the river outside the fish ladder. The head created by this elevation difference results in a water velocity flowing out of the fish ladder at a calculated eight feet per second.

Steelhead migrating upstream through the fish ladder are directed by a series of guide bars which directs the fish into the trap. A screen "funnel" (300 mm high by 100 mm wide) was placed at the upstream end of the guide bars. The funnel allows steelhead to easily move into the trap, while making it difficult for fish to move back out of the trap. The guide bars are spaced sufficiently far apart to allow lamprey (but not adult steelhead) to migrate past the trap unhindered. The trap is formed by the upstream trash rack and downstream guide bars (metal slats) and their supports. This forms a holding compartment measuring approximately 10' by 15'.

The trap is serviced once per day during the sampling season. During servicing, the fish ladder is drained, and the debris and sand that has collected around the trap are removed.

The fish trap is checked during this time. In addition, the rest of the fish ladder is surveyed for fish stranded as a result of dewatering the ladder.

Fish collected in the trap (or captured in the ladder during dewatering for trap maintenance) are identified to species and measured (fork length (FL) or total length (TL) depending upon caudal configuration of the individual species) to the nearest millimeter (mm). Photographs are taken of representative individuals. Scale samples are taken from rainbow trout for age determination.

Tissue samples were taken from steelhead trout for mitochrondrial DNA (mtDNA) and/or nuclear microsatellite analysis in 1995 and 1996 (results for tissue samples collected in 1995 are presented in Appendix A, tissue samples collected in 1996 have not been analyzed at this time). Steelhead and lamprey were then released upstream of the trap to continue their upstream migration.

#### 2.1.1 STREAMFLOW AND TURBIDITY MEASUREMENTS

Streamflow measurements are recorded from a staff gage reading by UWCD every morning at approximately the same time (0730). Turbidity measurements are recorded twice weekly by UWCD when the diversion is in operation. Water samples are collected at the diversion intake and measured using a Hach portable turbidity meter (model 2100 P).

#### 2.2 DOWNSTREAM MIGRATION

Downstream-migrating steelhead smolts entering the diversion facility are prevented from entering the diversion canal by a fish screen which directs the fish to the downstream fish by-pass facility. The fish by-pass allows the smolts to exit the diversion and return back to the river downstream of the diversion structure to continue their migration to the ocean. During periods when streamflow between the diversion and the ocean is not contiguous, fish are collected in a trap. Smolts trapped during these conditions are transported by truck to the lagoon downstream. The trap consists of a mesh cage that can be lowered into the chamber adjacent to the weir, through which all by-pass water (and fish) flows. The downstream trap was used throughout the study (1994-1996) to verify the existence of steelhead smolts in the system. Smolts collected in the by-pass trap are counted, measured (FL) and scale samples are collected for age determination.

Tissue samples were taken from steelhead/rainbow trout for mtDNA and/or nuclear micro satellite analysis in 1995 and 1996 from steelhead/rainbow trout and (results for tissue samples collected in 1995 are presented in Appendix A, tissue samples collected in 1996 have not been analyzed at this time). The smolts were then either released into the river through the by-pass, or, if flow was not contiguous to the lagoon, smolts were transported by truck to the lagoon. Juvenile and spawned-out adult Pacific lamprey also are collected in the downstream trap. Lamprey collected in the downstream trap are counted, measured, and released into the fish by-pass facility.

#### 3.0 1996 Results of Fish Passage Monitoring at the Vern Freeman Diversion Facility

Rainfall data has been collected at Santa Paula in the Santa Clara River Basin since 1890 (107 year period of record). Since 1890, annual rainfall has ranged from 6.42 to 38.11 inches, averaging 17.28 inches (Figure 3-1). Annual rainfall in the basin is skewed towards drier years. The median rainfall at Santa Paula is 15.01 inches and the Mode (combining the yearly rainfall data into 0.5 inch increments) falls between 11.00 and 11.49 inches. On a percentile basis, annual rainfall in at Santa Paula exceeds the "average rainfall" in only 37 percent of the years since 1890. Thus, the mean annual rainfall total provides a more descriptive means of assessing wet and dry years.

Two of the three years during the current study have been moderately dry compared to the median annual rainfall. Rainfall totaled 13.39 inches in 1994 (64 percentile) and 13.90 inches in 1996 (59 percentile). 1995 was the fourth wettest year (35.34 inches) recorded at Santa Paula since 1890. The differences in yearly rainfall, and the timing and intensity of rainfall events, are evident in the streamflow measured at the Vern Freeman Diversion and the number of days each year that the fish ladder and fish by-pass facility were operated. In 1996, the fish ladder was operated for 23 days, compared to 1995, when the ladder was operated for 116 days. The fish ladder was operated for 74 days in 1994. The downstream migrant traps was operated for 91 and 83 days in 1994 and 1996, respectively, and for 145 days in 1995.

#### 3.1 UPSTREAM MIGRATION

The sand bar at the mouth of the lagoon breached on or about 19 February, 1996 permitting upstream migrants to enter the river. The upstream fish ladder and trap were operated for nine days in February, and for 14 days in March (Figure 3-2).

#### 3.1.1 STEELHEAD

Two steelhead, measuring 263 and 436 mm FL were captured in the upstream fish trap on 5 and 17 March, respectively. The 263 mm specimen was aged as a two-year-old. The scales from the 436 mm specimen were unreadable. The steelhead were released upstream of the diversion to continue their spawning migration. Streamflow below the diversion was 40 cfs (fish ladder releases) on both days that steelhead were captured.

#### 3.1.2 PACIFIC LAMPREY

A total of 307 adult Pacific lamprey were counted in the fish ladder in 1996 (Table 3-1). Lamprey were observed in the ladder from 25 February, approximately six days after the lagoon breached. The upstream migration continued through 25 March when the ladder









Figure 3-2. Dates the Upstream Migrant Trap was in Operation, Vern Freeman Diversion, Santa Clara River, 1991-1996.

Month	Week Ending	1994 <sup>2</sup>	1995 <sup>3</sup>	19964
January	7	~~	0	
	14		0	
	21		I	—
	28		2	
February	4	0	95	_
	11	0	74	_
	18	1	35	
	25	19	16	14
March	4	11	24	85 <sup>5</sup>
	11	7	2	17
	18	283	4	36
	25	240	20	155
April	1	201	20	_
	8	133	34	
	15	13	9	
	22		9	_
	29		3	
May	6		13	
	13		8	
	20	_	2	_
	27			<u></u>
lune	4			<b>-</b>
	11	—	_	_

#### Weekly Summary of Adult Pacific Lamprey Caught in the Upstream Table 3-1. Migrant Trap, Vern Freeman Diversion, Santa Clara River, 1994<sup>1</sup>

<sup>1</sup>1994 and 1995 data from ENTRIX (1994, 1995) <sup>2</sup>Upstream migrant trap was operated from 4 February through 9 April, 1994. <sup>3</sup>Upstream migrant trap was operated from 6 January through 20 May, 1995.

<sup>4</sup>Upstream migrant trap was operated on 2-3 February, from 22 February through 5 March, and from 14 through 25 March, 1996

<sup>5</sup>Includes leap year day (8 day period)

was closed per the 404 permit. Approximately 43 percent of the Pacific lamprey migrated past the fish ladder during a three day period (19-21 March). Although upstream migration of lamprey peaked during the last week that the ladder was in operation, the daily counts decreased sharply during the final four days, from 12 to 1, 5, and 2, respectively. The upstream migrating lamprey ranged in total length from 490 to 675 mm TL, averaging 604 mm (Figure 3-3).

Rainfall in 1996 was below the median (Figure 3-1). Consequently, streamflow was relatively low. Streamflow below the diversion was generally less than 100 cfs during the majority of the 1996 season (compared to 1995 when streamflow exceeded 1,000 cfs during the majority of the season). Adult Pacific lamprey were observed in the fish ladder at streamflows ranging from approximately 15 to 950 cfs (Figure 3-4). The majority of the lamprey were observed in the ladder when streamflows below the diversion was 40 cfs. In comparison, adult lamprey located and ascended the fish ladder when the in-river flows exceeded approximately 4,000 cfs in 1995 (Figure 3-5 - Note the differences in the Y axis (flow) between the 1995 and 1996 graphs; also note that the axis' depicting streamflow were observed in the fish ladder).

#### 3.2 DOWNSTREAM MIGRATION

The downstream migrant trap was operated 23 days in February, 27 days in March, 30 days in April, and three days in May (the downstream migrant trap was closed on 3 May). When the diversion was not in operation, the river spills over the diversion structure. Any fish migrating downstream during this time would pass over diversion unhindered. Rainbow trout collected in the downstream trap and the diversion canal were separated into four categories. These categories were; wild smolts, wild resident rainbow trout, hatchery-reared rainbow trout, and hatchery-reared smolts. The descriptions of the four types are as follows:

- Wild smolts were typically bright silver in color, their fins were fully formed and did not show signs of unusual wear, and their scales were easily removed. Smolts also have larger heads, slimmer bodies, and a longer caudal peduncle than resident rainbow trout (Beeman et al. 1995).
- Wild rainbow trout had perfectly formed fins, but had more color than smolts. In addition, "resident rainbow trout" did not possess the typical body configuration characteristic of smolts (described above). It was assumed that these were not outmigrating juvenile steelhead.
- Hatchery-reared rainbow trout had fins that showed excessive wear as a result of rubbing against concrete raceways. The hatchery-reared rainbow trout tended to be longer, "fatter," and darker in color than the wild fish.
- Hatchery-reared smolts had fins showing excessive wear similar to hatchery rainbow trout, but were noted as being bright silver in color.











#### 3.2.1 STEELHEAD SMOLTS

A total of 82 wild smolts were captured in the fish trap between 18 March and 23 April (37 days). The weekly catch of smolts in the downstream trap was fairly consistent between 20 March and 14 April, with a peak in migration occurring in early April (Table 3-2). The majority of the smolts (91 percent) were counted over a 27 day period (20 March to 15 April), with 50 percent of the run counted by 5 April (Figure 3-6).

Readable scales were collected from 55 smolts, 26 of which were aged as being-one-year old (1+) (1995 cohort). The remaining 29 smolts were aged as two-years-old (2+) (1994 cohort). The scales from a few individuals were unreadable (i.e., the fish could not be aged from scale samples collected), and scales were not collected from all specimens captured in the downstream migrant trap. The remaining 29 smolts were assigned to an age class based on their length compared to smolts of a known age. For example, all smolts ranging in length between 150 and 159 mm FL were aged as 1+. Therefore, the four smolts in the 150 to 159 mm ranged for which scales were not available were assigned to the 1+ age group. There was some overlap between 1+ and 2+ age groups in the size classes greater than 160 mm length. Fish of unknown age were assigned an age class based on the percentage of known ages for each size class. For example, four smolts measuring between 180 and 189 mm FL were aged as 2+, two were aged as 1+ and five were unaged. Since approximately 70 percent of the smolts of a known age were aged as 2+, three (rounded to the whole fish) of the remaining five fish were assigned to 2+ age group, and two fish were assigned to the 1+ age group. On the basis of this analysis, 51.2 percent (42) of the smolts captured were age 1+ and 46.3 percent (38) were age 2+ (Figure 3-7). The two largest smolts (measuring 260 and 261 mm FL, respectively) could not be aged from the scale samples collected, and could not be assigned to an age group based on the above analysis (the largest fish aged in 1996 measured 235 mm FL).

Overall, smolts ranged in size from 134 to 261 mm (Figure 3-7). The one-year-old smolts ranged in length from 134 to 184 mm, averaging 164.6 mm. The two-year-old smolts ranged in length from 168 to 235 mm, averaging 201.6 mm.

Nine wild resident rainbow trout were captured in the downstream migrant trap between 16 March and 5 April. Resident rainbow trout ranged in length from 177 to 525 mm FL. Six resident fish were aged as two years old (177 to 225 mm FL), two were aged as three years old (326-345 mm FL), and one fish was at least four years old (525 mm FL). The growth pattern on the scales of the 525 mm specimen was consistent, and suggest that the fish had reared in a stable environment such as a reservoir.

Twenty-seven hatchery rainbow trout were collected in the downstream trap between 1 March and 5 April. Seventy percent (19) of the hatchery fish were captured on two days, March 1 and 18. The hatchery trout ranged in length from 222 to 293 mm, averaging 245.9 mm FL. Six of the 27 hatchery rainbow trout appeared to have been smolting.

Month	Week Ending	1994 <sup>2</sup>	1995 <sup>3</sup>	1996 <sup>4</sup>
January	7		0	
	14	—	0	_
	21	—	0	
	28	—	1	_
February	4	0	1	0
-	11	0	0	0
	18	0	1	0
	25	0	0	0
March	4	0	0	0
	11	0	0	0
	18	2	0	1
	25	9	2	16
April	1	16	0	14
-	8	25	0	28
	15	0	0	17
	22	15	2	5
	29	9	11	1
May	6	2	27	0
•	13	3	25	_
	20	0	16	_
	27	0	10	
June	3	_	5	
	10	_	6	_
	17	—	I	_
	24		0	
July	1		0	
-	8	—	0	<u> </u>
	15	_	0	_
	22	—	0	
	29		0	_

#### Weekly Summary of Steelhead Smolt Caught in the Downstream Table 3-2. Migrant Trap, Vern Freeman Diversion, Santa Clara River, 1994<sup>1</sup>

<sup>1</sup>Data From ENTRIX (1994) <sup>2</sup>Downstream migrant trap closed on 25 May, 1994. <sup>3</sup>Downstream migrant trap closed on 27 July, 1995.

<sup>4</sup>Downstream migrant trap closed on 3 May, 1996







These fish were bright silver in color (similar to wild smolts) and their scales were easily removed. These fish ranged from 215 to 270 mm FL, averaging 233.8 mm.

#### 3.2.2 PACIFIC LAMPREY

A total of 56 Pacific lamprey were captured in the downstream migrant trap in 1996 (Table 3-3). Lamprey were captured in the trap from 17 February through 29 April, with 55.6 percent of the captures recorded during the last two weeks in March. Pacific lamprey ranged in length from 152 to 680 mm TL (Figure 3-3). The 152 mm lamprey did not possess eyes and was thus categorized as an ammocoete. The 680 mm TL specimen was an apparent upstream migrating adult that had wandered into the diversion canal and into the downstream trap. The remaining 54 lamprey captured in the downstream trap appeared to spent spawners. These lamprey were covered with soars and lesions possibly as the result of constructing nests and spawning. The spawned-out lamprey ranged between 355 and 615 mm TL, averaging 438 mm TL. Since upstream migrating lamprey average greater than 600 mm TL in 1995 and 1996, the spawned-out lamprey collected in the downstream trap likely migrated upstream in 1995, held over in freshwater for one year, and spawned in 1996.

On average, Pacific lamprey that migrated upstream in 1995 decreased 173 mm in length (28 percent) by the time they returned downstream after spawning in 1996 (Table 3-4). Pacific lamprey that migrated upstream in 1994 decreased on average 183 mm (30 percent) by the time they were collected moving downstream.

#### 3.2.3 NON-ANADROMOUS FISH

Six additional species were caught in the downstream trap (and occasionally in the fish ladder) in 1996, including arroyo chub, prickly sculpin, Santa Ana sucker, partially armored threespine stickleback, bullhead and green sunfish. The number of fish caught varied by month and species. General observations of the abundance of the non-anadromous species indicated that arroyo chub, prickly sculpin, and Santa Ana suckers were the most abundant species present.

Month	Week Ending	1994 <sup>2</sup>	1995 <sup>3</sup>	1996 <sup>4</sup>
January	7		0	
•	14	_	0	_
	21	_	0	_
	28		0	
February	4	0	2	_
•	11	0	3	0
	18	0	1	1
	25	0	0	5
March	4	0	2	2
	11	2	1	0
	18	18	1	2
	25	20	0	18
April	1	13	10	12
	8	10	6	5
	15	5	4	2
	22	0	8	3
	29	0	18	4
May	6	1	32	0
	13	1	5	—
	20	0	3	
	27	_	5	
June	3		9	—-
	10	—	3	-
	17		0	<u>_</u>
	24	—	0	—-
July	1		0	—
	8	—	0	
	15	<u> </u>	0	
	22	_	0	

Weekly Summary of Pacific lamprey Caught in the Downstream Table 3-3. Migrant Trap, Vern Freeman Diversion, Santa Clara River, 1994<sup>1</sup>

<sup>1</sup>1994 data from ENTRIX (1994) <sup>2</sup>Downstream migrant trap closed on 25 May, 1994. <sup>3</sup>Downstream migrant trap closed on 27 July, 1995.

<sup>4</sup>Downstream migrant trap closed on 3 May, 1996.

# Table 3-4.A Comparison of Lengths of Adult Pacific Lamprey at the Time of<br/>Upstream Migration and After Spawning the Following Year.

	Average Length (mm)	Range (mm)	Difference (mm)
Length in 1994	610	485 - 750	
Length in 1995	427	355 - 510	183
Length in 1995	610	490 - 675	
Length in 1996	437	345 - 615	173

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4.0 DISCUSSION

#### 4.1 UPSTREAM MIGRATION

Steelhead trout were captured in the upstream and downstream migrant traps in each of the first three years of this study. The capture of the upstream migrants indicates that steelhead trout are attracted to, and are able to negotiate, the fish ladder over the Vern Freeman Diversion structure. In addition, the presence of juveniles in the downstream trap indicates that the fish screen and by-pass system allowed smolts diverted from the river to return downstream of the diversion to continue their migration to the ocean. Adult Pacific lamprey were also captured in the fish ladder. Most of the lamprey capture in the downstream trap appear to be spawned-out adults from the previous years run. Therefore, the results of the upstream and downstream trapping for adult Pacific lamprey will be discussed together.

#### 4.1.1 ADULT STEELHEAD

One adult steelhead was captured in the fish ladder in both 1994 and 1995, and two adults were collected in 1996. All four adult fish have been captured in the upstream trap during the month of March. The four steelhead captured in the upstream trap ranged in length from 263 to 635 mm FL. Based on scale analysis, the adult captured in 1994 (475 mm FL) had spent one year in freshwater and one year in the ocean (age 2+) before returning to spawn. The adult captured in 1995 (635 mm FL) was aged as a 4+, having spent two years in freshwater and two years in the ocean. The two adults captured in 1996 measured 263 and 436 mm FL, respectively. The smaller fish was aged as a 2+, having spent one year in freshwater and one year in the ocean. The larger fish could not be aged from the scale sample collected.

Adult steelhead have migrated through the lower Santa Clara River and ascended the fish ladder when streamflows below the diversion ranged from approximately 40 to 1,175 cfs. In 1996, streamflow below the diversion was approximately 40 cfs (fish ladder release) on both days when steelhead were collected in the fish ladder. Streamflow below the diversion was also 40 cfs on the day preceding the capture of the steelhead as well. This suggest that adult steelhead (ranging in size from 263 to 437 mm FL) are able to migrate upstream and ascend the fish ladder when flow in the river is limited to fish ladder releases. Streamflow in the river was approximately 1,000 cfs in the days prior to the capture of the adult in 1995.

#### 4.2 DOWNSTREAM MIGRATION

#### 4.2.1 JUVENILE STEELHEAD

When streamflow is continuous to the ocean, fish diverted into the facility are shunted through the fish by-pass and returned via a pipe back into the river downstream of the diversion structure. When streamflow is discontinuous, the smolts are collected in the downstream migrant trap and transported downstream to the lagoon where they can continue their migration to the ocean. However, the downstream trap has been monitored continuously during the study (1994-1996) to document the presence of smolts in the river. Only the diverted portion of the river was sampled. Therefore, the results of the downstream migrant trapping are only indicative of the minimum number of smolts that migrated downstream. There is no way to count smolt that may pass over the diversion structure when it spills to the river downstream.

The numbers of rainbow trout/steelhead counted in the downstream trap provide information on the seasonal timing, size ranges and age classes of outmigrating smolts. The number of smolts captured in the downstream migrant trap in 1994, 1995, and 1996 were 81, 111, and 82 respectively. The mean daily streamflow in 1995 was considerably higher compared to 1994 and 1996, and a greater number of smolts were captured even though the percentage of water sampled (diverted) was considerably less.

The total duration of outmigration (from the date of the first smolt captured in the downstream trap to the date of the last smolt captured) has ranged from 37 days in 1996 to 152 days in 1995 (56 days in 1994). However, the majority of the smolts migrate past the diversion over a five week period. Between 1994 and 1996, 91.4, 82.9, and 98.7 percent of the smolts outmigrated past the diversion over a 35 day period, respectively.

Shapovalov and Taft (1954) reported that downstream migration of smolts began earlier in low flow years compared to high flow years. During relatively low flow years (1994 and 1996), smolt outmigration began in mid-March, peaked in early-April, and was essentially completed by the second week of May. In 1995, a high flow year, the outmigration period essentially began in late-April, peaked in early-May, and continued into early-June. The date at which 50 percent of the run was counted at the diversion occurred on 6 April in 1994, 9 May in 1995, and 5 April in 1996.

Smolts outmigrated primarily as one-year-old fish in 1994 and 1995. Age 1+ smolts comprised 68.0 and 82.2 percent of the outmigrants in 1994 and 1995, respectively. In 1996, age 1+ smolts comprised only 51.2 percent of the outmigrants. Age 1+ smolts were slightly larger, on average, in 1994 compared to 1995 and 1996 (175 mm in 1995 compared to approximately 165 mm FL in 1995 and 1996). Age 2+ smolts comprised 30.6, 17.8, and 46.3 percent of the out migrants in 1994, 1995, and 1996, respectively. The average length of age 2+ smolts was approximately 220 mm in 1994 and 1995 compared to approximately 202 mm in 1996. Only one smolt has been aged as 3+ in three years of trapping (1994).

Hatchery-reared rainbow trout have been identified in the downstream migrant trap in 1995 and 1996. CDFG stocks hatchery trout into a tributary of the Santa Clara River during the winter and spring to provide for a put-and-take fishery (Adams pers. comm. 1995). Hatchery rainbow trout that have survived for at least one year after release into the wild have been identified at the Vern Freeman Diversion. In addition, six apparently smolting hatchery rainbow trout have also been captured.

#### 4.3 PACIFIC LAMPREY

Pacific lamprey have a life history pattern similar to steelhead in that the young are spawned and reared in freshwater, then migrate to the ocean were they grow and mature before returning to freshwater to complete their life cycle. Beamish (1980) reported that Pacific lamprey typically migrate into rivers in British Columbia from April through June, and remain in freshwater for one year prior to spawning. In California, Pacific lamprey have been reported to migrate upstream between April and late July (Moyle 1976), although in Trinity River in northern California, Moffett and Smith (1950, cited by Moyle 1976) reported some adult lamprey migrating upstream in August and September. Pacific lamprey have been reported to spawn between April and July (Beamish 1980, Moyle 1976, and Wang 1986). The spawning migration in the Santa Clara River is regulated by streamflow, and has begun as early as late January in 1995 and as a late as early March in 1991, depending on the onset of winter rains. Adult lamprey are not thought to range far from their natal streams during the oceanic phase of their life history (Moyle 1976). The 1994 through 1996 data suggest that the lamprey are in close proximity to the mouth of Santa Clara River just prior to the spawning season based on their presence in the fish ladder (approximately 16.8 kilometers inland) within two weeks of the sand bar breaching. Pacific lamprey were first observed in the fish ladder 10 days after the probable breaching date of the lagoon mouth in 1994, 13 days after the lagoon breached in 1995, and six days after the lagoon breached in 1996. However, the number of adult lamprey captured in the ladder have typically remained low during the first two weeks of the run.

The adult Pacific lamprey run spanned a period of about three months (late January though early May) in 1995, the only year to date that the entire lamprey spawning migration was sampled. Although the start of the lamprey run was missed in 1993, the run also continued into the first week of May. In 1996, the daily number of lamprey counted in the fish ladder had declined to very low numbers prior to the close of the fish ladder on 25 March. However, it is not known if additional lamprey would have entered the river with additional streamflow.

Adult Pacific lamprey have been observed in the fish ladder over a range of in-river streamflows from approximately 15 to over 4,000 cfs in 1995 and 1996. In 1995, a relatively wet year, adult Pacific lamprey were able to locate and ascend the fish ladder when in-river streamflows ranged between 1,000 and over 4,000 cfs. In 1996, a relatively dry year, streamflow seldom exceeded 100 cfs below the diversion, however, adult Pacific lamprey utilized the fish ladder in numbers similar to the 1995 run. This suggest

that streamflows within this range (approximately 15 to 4,000 cfs) do not appear to affect the ability of lamprey to locate and ascend the fish ladder over the Vern Freeman Diversion.

The number of Pacific lamprey counted in the fish ladder has ranged from 74 in 1991 to 908 in 1994. However, an unknowable number of adult lamprey potentially migrate through the fish ladder without being detected. In addition, the number of lamprey counted was dependent upon the length of time that the fish ladder was in operation. The fish ladder was in operation but not monitored during the beginning of the run in 1993. The upstream migration of lamprey was still in progress when the fish ladder was closed in 1994. In 1995, essentially the entire lamprey run (371) was monitored prior to the fish ladder being closed. In 1991, 74 adult lamprey were counted in the fish ladder over a four day period, and streamflow sufficient for lamprey to migrate upstream continued for only four or five days after the cessation of monitoring. In 1996, 308 adult lamprey were counted in the ladder. Although the weekly counts of adult lamprey peaked the last week that the ladder was in operation, relatively few lamprey were counted during the last four days of operation.

Pacific lamprey tend to migrate upstream in surges (Moyle 1976). In 1994, the daily catch of lamprey ranged between 0 and 11 in the fish ladder between 17 February and 15 March (65 total). On March 16 and 18, 142 and 118 lamprey were collected in the fish ladder, and released upstream of the diversion, respectively. The fish ladder was closed on 16 March in accordance with the 404 permit. The fish ladder was reopened on the morning of 17 March to allow the lamprey spawning migration to continue. In total, 332 lamprey were collected in the fish ladder between March 13 and 19. The fish ladder was closed on 9 April in accordance with the 404 permit. At the time the ladder was closed, adult lamprey were still being observed in the fish ladder. In 1995, the lagoon mouth breached approximately one month earlier and the upstream run of adult lamprev also peaked one month earlier compared to 1994. By 3 February, only three lamprey had been collected in the fish ladder. The lamprey run then peaked over the next two weeks when approximately 46 percent of the run was counted in the fish ladder. After this initial pulse of upstream migrants, the weekly number of lamprey observed in the ladder declined to a total of six lamprey captured between March 5 and 18. A smaller, secondary pulse of 74 upstream migrants occurred between 19 March and 8 April. In 1996, a total of 135 lamprey (43 percent) were counted between March 19 and 21, while only eight were counted between March 23 and 25. The upstream run was interrupted for 10 days in mid-March by low streamflow and the closure of the fish ladder. During this time period, rising groundwater resulted in streamflow in the lower reaches of the river, and flow was maintained to the ocean. Approximately 63 percent of the run migrated past the ladder after it was reopened following rains in mid-to-late March.

Adult lamprey collected in the fish ladder ranged in size from 535 to 700 mm TL in 1994, and from 485 to 750 mm TL in 1995 (although only one individual less than 535 mm was captured) and from 490 to 675 mm TL in 1996. Pacific lamprey averaged 610 mm TL in 1994 and 1995, and 604 mm TL in 1996.

Adult lamprey do not feed after returning to freshwater to spawn (Hardistry and Potter 1971). As a result, they decrease in length prior to spawning. In British Columbia, adult Pacific lamprey decreased by an average of 20 percent by the time that spawning actually took place (Beamish 1980). A similar pattern of holding over in freshwater for one year prior to spawning appears to be occurring in the Santa Clara River Population. In 1995 and 1996 several apparently spawned out lamprey averaging 427 mm TL (range 355 to 510 mm TL) and 437 mm TL (345 to 615 mm TL) were captured in the downstream trap. Since the adult lamprey in the upstream ladder during both years were considerably larger than those captured in the downstream trap, the spawned out lamprey in the downstream trap could not have been part of that years upstream migration. The 1994 adult lamprey decreased, on average, 183 mm or 30 percent of their original body length, and the 1995 run decreased, on average, 173 mm, or 28 percent of their original body length, prior to

Pacific lamprey die after spawning. However, the spawned-out lamprey captured in the downstream trap were still alive at the time of capture. This would suggest that spawning had recently taken place (i.e., after spawning, the lamprey were drifting downstream in a weakened condition). Based on the capture of spawn-out lamprey in the downstream trap in 1995 between 4 February and 11 June, it appears that spawning may begin in January and continue into May during years with ample rainfall. In 1996, spawned out lamprey were captured in the downstream trap between 17 February and 28 April.

Adult Pacific lamprey apparently have little difficulty negotiating the fish ladder. Marking studies conducted in 1994 and 1995 found that approximately 92 percent of the lamprey were able to negotiate the ladder in less than 24 hours.

Young adult Pacific lamprey have been reported to average 130 mm (Beamish 1980) and 140 to 160 mm (Moyle 1976) in length when they metamorphose and migrate to the ocean. In the Santa Clara River, young adult Pacific lamprey migrating to the ocean average approximately 150 mm in length (Swift pers. comm. 1996). Although no young adult lamprey in the process of metamorphosing have been collected during the study, young lamprey in the 130 to 160 mm size class are probably small enough to pass though the screens of the downstream migrant trap. Ammocoetes of several species of lamprey out-migrate during periods of high streamflow (Hardistry and Potter 1971). Beamish and Levings (1991) reported that young adult Pacific lamprey migration was initiated by abrupt increases in stream flow. Thus, if young adult lamprey in the Santa Clara River outmigrate during high streamflow events, they will be less likely to be enter the diversion canal and be observed.

#### 4.4 NON-ANADROMOUS SPECIES

Eight non-anadromous species have been collected in the Vern Freeman fish ladder and downstream migrant traps during the study. The most abundant species collected during the study was the prickly sculpin. The sculpin were reported in Piru and Castaic creeks during a basin wide fisheries investigation (Bell 1978). The presence of prickly sculpin

at the diversion site suggest that they expanded their range considerably since 1978. However, the Santa Clara River typically becomes dry over much of its lower reach (above and below the diversion), and unless suitable habitat exist in the lagoon for freshwater fish, the non-anadromous fish moving downstream in the vicinity of the diversion will be lost as the river dries. Arroyo chub and Santa Ana suckers were the only other species collected in relatively large numbers to date.

# 5.0 CONCLUSIONS AND RECOMMENDATIONS

The fish monitoring results indicate that the fish passage and fish monitoring facilities performed satisfactorily in 1994, 1995 and 1996. Although only one adult steelhead was captured in the upstream migrant trap in 1994 and 1995, and two in 1996, there were no indications that steelhead were impeded or prevented from negotiating the fish ladder during the period of flow. In addition, the downstream fish by-pass facility functioned as designed. Smolts entering the diversion facility successfully passed from the diversion canal into the fish by-pass pipe where they would be returned to the river downstream of the diversion structure (as evidenced by fish caught in the downstream migrant trap). The 1996 trapping season marks the third year of the five year monitoring study. This study will continue through the 1998 migration period.

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APPENDIX A

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EVALUATION OF STEELHEAD/RAINBOW TROUT STOCKS

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#### APPENDIX A EVALUATION OF STEELHEAD/RAINBOW TROUT STOCKS

#### A.1 INTRODUCTION

There are four objectives for this study:

- to determine whether steelhead and rainbow trout collected at the Vern Freeman Diversion (VFD) included steelhead of native southern California heritage
- to determine whether steelhead and rainbow trout collected at the VFD included fish of hatchery or northern California heritage
- to determine the heritage of recently released hatchery rainbow trout collected at the VFD, and
- to review the available information regarding the heritage of steelhead and rainbow trout inhabiting Santa Clara River Basin tributaries.

We used mitochondrial DNA (mtDNA) sequencing to identify the heritage of steelhead and rainbow trout collected at the VFD. MtDNA sequencing requires a very small fish tissue sample, and is thus non-destructive. Our technique followed that of Neilsen (1994), who sequenced tissue samples from steelhead/rainbow trout from a number of California rivers, including Sespe, Santa Paula, and Piru creeks in the Santa Clara River Basin.

#### A.1.1 BACKGROUND: STEELHEAD/RAINBOW TROUT LIFE HISTORY

Coastal populations of rainbow trout in freshwater are typically comprised of resident "rainbow trout" and anadromous "steelhead" (Shapovalov 1954). Although rainbow trout and steelhead are the same species (*Oncorhynchus mykiss*), resident rainbow trout remain in freshwater through-out their life. Steelhead migrate to the ocean as juveniles to mature, then return to freshwater to spawn. Coastal rainbow trout and steelhead have a common ancestry (Benke 1992) and are thought to be flexible in their life history strategy (e.g., resident parents may produce offspring that become anadromous). This plasticity in life history strategy may allow rainbow trout, including steelhead, to persist in uncertain environments like those found in southern California. During periods of prolonged drought, migration opportunities to and from the ocean are restricted or non-existent. During droughts, populations of coastal rainbow trout may be restricted to the resident fish life history strategy, surviving in perennial headwater streams. With the return of years with abundant rainfall, a portion of the population can revert to an anadromous life history strategy and take advantage of the relatively abundant food supply in the ocean. Thus, the numbers of rainbow trout expressing the anadromous life history strategy would expand and contract with the natural variation in rainfall. For example, the Santa Ynez River steelhead runs apparently increased and decreased with wet and dry cycles between 1930 and 1950 (ENTRIX 1995a).

Prior to ocean migration, the juveniles of the two forms cannot be differentiated (and are referred to as "steelhead/rainbow trout" in this report). However, juvenile steelhead migrating to the sea go through a process called smoltification. This process is a change in the physiology of the fish which must adjust from living in freshwater to a saltwater environment. The juvenile steelhead going through these changes are called smolts. Smolts typically become bright silver in color, and their body shape changes (compared to their freshwater or "parr" stage). Smolts have larger heads, slimmer bodies, and a longer caudal peduncle than parr (Beeman et al. 1995). In addition to the native rainbow trout and steelhead, hatchery rainbow trout are stocked into basin streams such as Sespe, Santa Paula and Piru creeks. Hatchery rainbow are typically characterized by having eroded fins (from rubbing on the concrete raceways that they are reared in at the hatchery), are generally darker in color, and are usually larger and heavier than smolts.

Although survival of hatchery rainbow trout in the wild is generally low, a small percentage of planted rainbow trout may survive after release into basin streams. For example, in 1996 a number of hatchery reared rainbow trout captured at the VFD were identified by J. Adams, fish hatchery manager for CDFG's Fillmore Hatchery, as having been stocked in 1995 (the fish were larger than the rainbow trout stocked in 1996). Rainbow trout of hatchery origin have the potential to spawn in the wild. The offspring of hatchery reared parents that spawn after being stocked are considered to be "wild" but "non-native". These are spawned and reared in the wild as opposed to a hatchery, however, their genetic heritage are from rivers outside of southern California.

#### A.1.2 BIOGEOGRAPHIC DISTRIBUTION OF THE MTDNA TYPES FOUND IN SANTA CLARA RIVER STEELHEAD/RAINBOW TROUT

Tissue samples for mtDNA analysis were collected from upstream and downstream migrating steelhead and hatchery rainbow trout captured at the VFD. The mtDNA sequencing was conducted by K. Thomas and J. Vida at the University of Kansas City, Missouri (J. T. Vida pers. comm. 1996), using techniques described in Nielsen (1994). MtDNA data are also available for rainbow trout collected in three Santa Clara River Basin tributaries, Sespe, Santa Paula, and Piru creeks (Nielsen pers. comm. 1994).

Nielsen (1994) used sequencing of mitochondrial DNA on 547 coastal steelhead from 33 streams and five hatcheries throughout California, and identified 13 "mtDNA types" within California populations. Nielsen identified steelhead/rainbow trout with seven of the thirteen mtDNA types in Santa Clara River stocks, referred as Types 1, 3, 5, 8, 9, 10, and 14 (Nielsen pers. comm. 1994 and this report). Types 5, 9, 10 and 14 were represented by only one or two individuals in Nielsen's study.

Overall, four of thirteen types appear to dominate in coastal steelhead populations, Type 1, Type 3, Type 5 and Type 8 (Nielsen 1994). Although the four main types (1, 3, 5, 8)

were found throughout coastal California, the distribution of these types suggests a distinct cline in frequency distribution along the coast from north to south. For example, steelhead with the Type 1 mtDNA marker were far more common in northern California streams (Humboldt Bay to Gualala Point) than they were in streams to the south, while steelhead with the Type 3 mtDNA marker were the most common variety between the Russian River and Point Sur (Nielsen 1994). The frequency of occurrence of steelhead with the Type 5 mtDNA marker increased in streams south of San Francisco, and steelhead with the Type 8 mtDNA marker were found predominantly in southern California rivers (from San Simeon Point to Santa Monica Bay) (Nielsen et al. 1994). In addition, Type 1 and Type 3 steelhead were used to develop several hatchery strains, and have been widely introduced throughout California. The Type 9 steelhead/rainbow trout were found in a few tributaries to the upper Sacramento River and possibly in some hatchery stocks (Nielsen pers. comm. 1996), however, their overall distribution was The Type 10 genetic marker has been found in resident rainbow trout unclear. (Kamloops strain) in British Columbia. The Kamloops and Whitney hatchery strains have bee crossed and have been stocked into basin streams in the past (Adams pers. comm. 1996). Steelhead/rainbow trout with the Type 14 mtDNA marker are fairly rare, and therefore the true distribution of this type is unclear (Nielsen pers. comm. 1994).

The geographic distributions of the different mtDNA types listed above are not absolute boundaries, but the frequency of their occurrence will differ based on geographic location. For example, native steelhead with the Type 1 genetic marker have been found in trout all along the coast to Baja California, and Type 8 steelhead have been found in the Van Duzen River, a tributary to the Eel River in northern California. In addition, past stocking practices have led to the introduction of several strains throughout the state. However, the prevalence of these markers is low outside their normal range.

At present, there are at least four streams that could be the source of the smolts collected at the Vern Freeman Diversion facility (VFD). These are Sespe, Santa Paula, Piru, and Hopper creeks. MtDNA data has been collected from steelhead/rainbow trout inhabiting all of the streams except for Hopper Creek. Hopper Creek has been reported to support steelhead/rainbow trout and has not been stocked with hatchery fish in the past 20 years by CDFG (Adams pers. comm. 1996). However, it is likely that all streams in the Santa Clara River Basin were stocked at some point in the past, including Hopper Creek (Adams pers. comm. 1996). Steelhead and hatchery rainbow trout in Sespe and Hopper creeks have access to and from the ocean. The rainbow trout sampled in Santa Paula and Piru creeks were above barriers to upstream migration and represent a combination of self-sustaining wild and hatchery reared stocks. It is possible for wild and hatchery reared rainbow trout to migrate, or be transported by high streamflow, downstream into the mainstem Santa Clara River, and from there to the ocean.

#### A.2 RESULTS AND DISCUSSION

#### A.2.1 ADULT STEELHEAD

MtDNA sequencing analysis was conducted for the one adult steelhead captured in the upstream migrant trap. Following the classification scheme presented in Nielsen (1994), the adult steelhead was sequenced as a Type 3 (Table A-1).

Steelhead/rainbow trout stocks with the Type 3 genetic marker are common in hatchery stocks and from rivers that enter the ocean between the Russian River and Point Sur, although the historic range of Type 3 steelhead may extend into southern California. It is likely that the adult steelhead was a stray from another river system or of hatchery heritage.

#### A.2.2 Smolts

MtDNA sequencing analysis was conducted on tissue samples from a total of 33 wild smolts collected in the downstream migrant trap. The 33 smolts were sequenced into four of Nielsen's 13 "mtDNA Types"; Type 1 (9), Type 3 (10), Type 5 (1), and Type 8 (13) (Table A-1).

Based on mtDNA sequencing of apparently wild smolts collected at the VFD, approximately 43 percent of the smolts have the "southern California" mtDNA genetic marker (predominantly Type 8). The remaining 57 percent of the apparent wild smolts were sequenced as Type 1 or Type 3. These are also the same mtDNA types found in hatchery rainbow trout collected at the VFD. The Type 1 and 3 genetic markers are also common in steelhead that have a more northern distribution. Thus, the origin of the smolts captured at the VFD appear to be a mixture of native southern California stocks and hatchery origin.

#### A.2.3 HATCHERY RAINBOW TROUT

MtDNA sequencing analysis was conducted on tissue samples from a total of 19 hatchery-reared rainbow trout that were stocked into basin streams during January and February, 1995. Hatchery rainbow trout were sequenced into two mtDNA types, Type 1 (4) and Type 3 (15) (Table A-1). Since a portion of the hatchery rainbow trout stocked into basin tributaries are known to survive at least one year after being released into the wild, they provide a potential source for the Type 1 and Type 3 steelhead and rainbow trout collected in basin streams and at the VFD.

#### A.2.4 SESPE, SANTA PAULA, AND PIRU CREEKS

Nielsen (pers. comm. 1994) sequenced mtDNA samples from steelhead/rainbow trout from three basin streams, Sespe, Piru, and Santa Paula. Thirty-five steelhead/rainbow trout collected in Sespe Creek were classified into three mtDNA types; Type 3 (24), Type 8 (10), and Type 10 (1) (Table A-1). Forty-one steelhead/rainbow trout were collected in

Santa ]	Paula, an	d Piru C	reeks <sup>2</sup> .					-
River/Life History Strategy	Type 1	Type 3	Type 5	Type 8	Тур <del>с</del> 9	Type 10	Type 14	Total
VFD (Adult Steelhead) <sup>1</sup>	0	1	0	0	0	0	0	1
VFD (Smolt) <sup>1</sup>	9	10	1	13	0	0	0	33
VFD (Hatchery)	4	15	0	0	0	0	0	19
Sespe Creek <sup>2</sup>	0	24	0	10	0	1	1	36
Santa Paula Creek <sup>2</sup>	36	3	1	0	l	0	0	41
Piru Creek <sup>2</sup>	15	5	0	3	0	0	0	23
Total	64	57	2	26	1	I	1	152

## Rainbow Trout MtDNA Types Collected at the VFD<sup>1</sup>, and Sespe, Table A-1.

<sup>1</sup>Data from present study <sup>2</sup>Data from Nielsen (pers. comm. 1994)

Santa Paula Creek were classified into four types, Type 1 (36), Type 3 (3), Type 5 (1) and Type 9 (1). Twenty-three steelhead/rainbow trout collected in Piru Creek were classified into three types; Type 1 (15), Type 3 (5) and Type 8 (3).

Type 8 steelhead/rainbow trout were relatively abundant only in the Sespe Creek samples (10 of 36 fish sampled). No Type 8 steelhead/rainbow trout were found in the Santa Paula Creek sample, and three (13 percent) were found in the Piru Creek sample. Only one rainbow trout was sequenced with the Type 5 genetic marker (from Santa Paula Creek). Overall, Type 1 and Type 3 genetic markers were sequenced in approximately 67 percent of the Sespe Creek sample. All of these creeks are planted with hatchery rainbow trout.

#### A.3 CONCLUSIONS

Tissue samples were successfully sequenced from steelhead and recently released hatchery rainbow trout collected at the VFD in 1995. The sample included one adult steelhead, 33 outmigrating juvenile steelhead (smolts), and 19 recently released hatchery rainbow trout. In addition, tissue samples have been analyzed from steelhead/rainbow trout collected in three basin tributaries (Nielsen 1994 pers. comm.). The major findings of this study are:

- The one adult steelhead collected at the VFD in 1995 was sequenced with the Type 3 genetic marker. Thus, this fish was likely either a stray from a more northern river, or of a hatchery heritage.
- The smolts collected at the VFD are a combination of native southern California stocks and likely hatchery origins. The Type 8 (southern California) genetic marker comprised 40 percent of the smolts sequenced. Type 1 and Type 3 genetic markers (hatchery and northern California stocks) were sequenced in 57 percent of the sample.
- Recently released hatchery rainbow trout collected at the Vern Freeman Diversion were sequenced with the Type 1 and Type 3 genetic marker. Thus, the hatchery rainbow trout provide a potential source for the Type 1 and Type 3 genetic marker found in the majority of the wild fish sequenced.
- Including the results of Nielsen (1994 pers. comm.), trout with the Type 1 and Type 3 mtDNA markers comprise the majority (77.4 percent, excluding recently released hatchery fish) of the Santa Clara River Basin steelhead/rainbow trout sequenced. Steelhead/rainbow trout with the "southern California" mtDNA marker (Type 8) were relatively abundant only in Sespe Creek and VFD samples (28 and 42 percent of the samples, respectively). Overall, wild steelhead/rainbow trout with the Type 8 genetic marker accounted for 19.5 percent of the fish sequenced. Based on the mtDNA data collected to date for Santa Clara River Basin stocks, it appears that hatchery rainbow trout are influencing the steelhead/rainbow trout gene pool.

#### A.4 REFERENCES

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