Bull. Southern California Acad. Sci.
108(1), 2009, pp. 1–15

Die Off and Current Status of Southern Steelhead Trout (*Oncorhynchus mykiss*) in Malibu Creek, Los Angeles County, USA

Rosi Dagit,[1] Stevie Adams,[2] and Sabrina Drill[3]

[1,2]*Resource Conservation District of the Santa Monica Mountains, P.O. Box 638, Agoura Hills, CA 91376-0638*
[3]*University of California Cooperative Extension, Los Angeles and Ventura Counties, 4800 Cesar Chavez Blvd., Los Angeles, CA 90022*

Abstract.—A die-off of native and exotic fish and invertebrate species, including the endangered southern steelhead trout (*Oncorhynchus mykiss*) was observed in Malibu Creek, Los Angeles County, during the summer and fall of 2006. Death was preceded by a period of illness during which trout in particular exhibited a noticeable yellow coloration. Physical, chemical and biological variables, including temperature, dissolved oxygen, a variety of chemical contaminants, presence of toxin producing algae, and direct pathology were examined but results remain inconclusive. The first day of a 12-day high temperature event occurred on the same date yellow trout were first observed. This sustained event is different from shorter term temperature spikes recorded in other years. Recovery monitoring documented re-colonization by all exotic fish species and crayfish, but limited numbers of southern steelhead trout in 2007. Surveys in summer 2008 documented a record number of anadromous adults (five silvery fish over 50 cm total length) and young of the year (over 2,200 under 10 cm).

Malibu Creek in Los Angeles County is home to one of the southernmost reproducing populations of the endangered southern steelhead trout (*Oncorhynchus mykiss*). Located in arid southern California, this stream offers access to the ocean only during the rainy winter season when high flows breach a sand berm across the mouth of the estuary. Adults entering the stream from the ocean reach an impassible barrier presented by the 30 meter high Rindge Dam, restricting them to the lowest 3.2 km of more than 112 km of historic steelhead habitat. The dam, originally built for water storage and flood control in 1926, no longer functions as the reservoir is completely filled with sediment. Over the past decade several government agencies and non-profit organizations have been trying to remove the dam to restore access to upstream spawning habitat. In 1997, the southern Evolutionarily Significant Unit (ESU) of steelhead trout was added to the federal list of endangered species, with Malibu Creek as the southernmost boundary. Since 1997, the protected range of this ESU has been extended to the U.S./Mexican border. The National Marine Fisheries Service (NMFS) estimates that only 500 anadromous adults remain within this ESU (NMFS 2007).

Located within the 283sq kilometer Malibu Creek watershed (Figure 1), Malibu Creek and its tributaries are impacted by source and non-point source pollutants emerging from the residential, commercial, animal husbandry and infrastructure facilities covering at least 22% of the total area (Dagit et al 2005). Malibu Creek has been placed on the federal list of impaired water bodies under section 303 of the Clean Water Act due to water

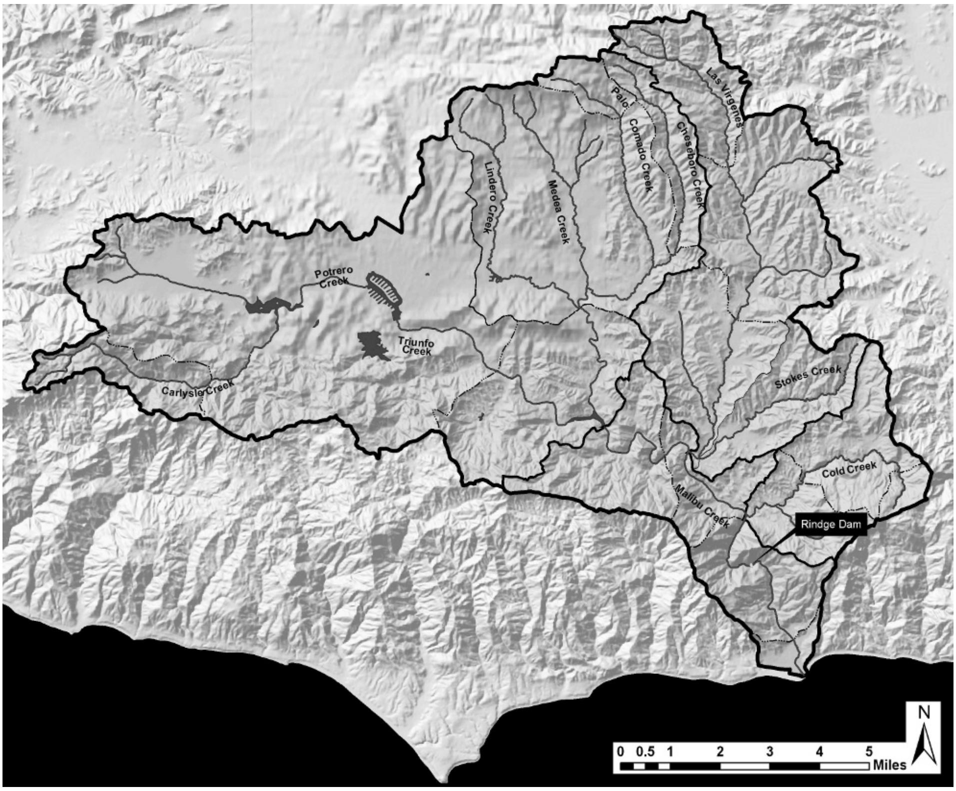

Fig. 1. Malibu Creek Watershed map, Study reach is located downstream of Rindge Dam (Map provided by M. Grimmer, Heal the Bay).

quality degradation related to excess sedimentation, nutrients, coliform bacteria and pathogens, trash, scum and foam (LARWQCB 2007). The creek is also host to numerous invasive exotic species, including red swamp crayfish (*Procambarus clarkii*), common carp (*Cyprinus carpio*), and recently introduced New Zealand mud snails (*Potamopyrgus antipodarum*) (Dagit and Abramson 2007). In recent years we have noticed a sulfurous unpleasant odor in portions of the creek, and the substrate in shallow pools is often covered with a slimy black muck that may itself be covered with a white/grey powdery looking substance.

The Resource Conservation District (RCD) of the Santa Monica Mountains has conducted monthly snorkel surveys to monitor the *O. mykiss* population in Malibu Creek since June 2005. In May 2006, we recorded the highest numbers of *O. mykiss* in Malibu Creek seen since the surveys began (245 individuals including more than 70 young-of-the year). In July 2006 we observed a yellow coloration in a few otherwise healthy small (< 15 cm) juvenile trout. The next month, yellow trout of all sizes were observed, and appeared stressed or sick, swimming sluggishly with mouths agape. In September, only nine *O. mykiss* were observed in Malibu Creek. Other species including crayfish, carp, and catfish, while not exhibiting the yellow coloration, had declined in number and showed similar sluggishness. By November 2006, no fishes at all were found in Malibu Creek.

In December 2006, a Technical Advisory Committee was convened to frame an investigation into the possible causes of the yellow coloration observed in *O. mykiss*, and the morbidity and mortality observed among these fish and other aquatic species. This group included aquatic biologists, chemists, and other experts from the RCD of the Santa Monica Mountains, University of California Cooperative Extension, California Department of Parks and Recreation, the National Park Service – Santa Monica Mountains National Recreation Area, California Department of Fish and Game (CDFG), NMFS, Las Virgenes Municipal Water District (LVMWD), Malibu Creek Watershed Monitoring Program/City of Calabasas, UC Santa Barbara, UC Los Angeles, California State University-San Marcos, Heal the Bay, Los Angeles County Vector Control (LACVC), and the Los Angeles Regional Water Quality Control Board (LARWQCB).

This group collected and reviewed existing data and developed the following list of questions for further study:

1. What caused the yellow coloration?
2. What caused the morbidity and mortality of the trout and other aquatic species?
3. Was there an unusual water quality problem such as low dissolved oxygen, extreme nutrient loading, or extreme high temperatures?
4. Did the recent invasion by New Zealand mud snails (*P. antipodarum)* play a role?
5. Did the thick layer of muck covering the channel substrate contribute to the morbidity and mortality, and what was the composition of this material?

These questions guided investigation into the die-off. All of these questions were not conclusively answered, but several variables were identified that should be helpful in developing a quick response research program if this phenomenon is observed again.

Materials and Methods

Snorkel surveys were conducted monthly from June 2005 to June 2007 between the upper extent of Malibu Lagoon (upstream of Cross Creek Road) and Rindge Dam, covering approximately 3.2 km of stream reach. An additional reach (500 m upstream of the dam) was added to the survey from February–June 2007 so that we could observe the status of exotic fish species density where no die-off was documented. Additional monthly surveys were done between June and October 2008.

Surveys were conducted by a trained team of 3–4 divers and one observer/data recorder. Divers enter the water on the downstream end of the area and slowly work their way upstream searching under every boulder undercut, bubble curtain or other instream feature where fish might hide. The observer is positioned with a clear view into the water in order to see fish, and determine if any fish swim away from the divers. Each habitat unit where fish are located is characterized by type (pool, run, riffle), depth, width, dominant substrate, canopy cover, instream cover, and shelter value, according to the standards of the *California Salmonid Stream Habitat Restoration Manual* (Flosi and Reynolds 1998). Large, deep pools were often surveyed twice by divers swimming in a transect formation, first from the surface, and then again after about 30 minutes, with divers inspecting every undercut as well. Counts were verified by the observer, who discussed the number and condition of fish observed with each diver to avoid recording duplicate fish observations from different divers.

Water temperature was measured every 15–30 minutes by use of in-stream recording thermometers (Stowaway Tidbits, HOBO, Inc.). Loggers were placed in two locations each year where trout were consistently observed (Malibu dam pool-2005, 2006, 2008, lower twin pool-2008, and just downstream of the start pool- 2005, 2006) within the study reach (Figure 2). These two locations were selected since each represented conditions in

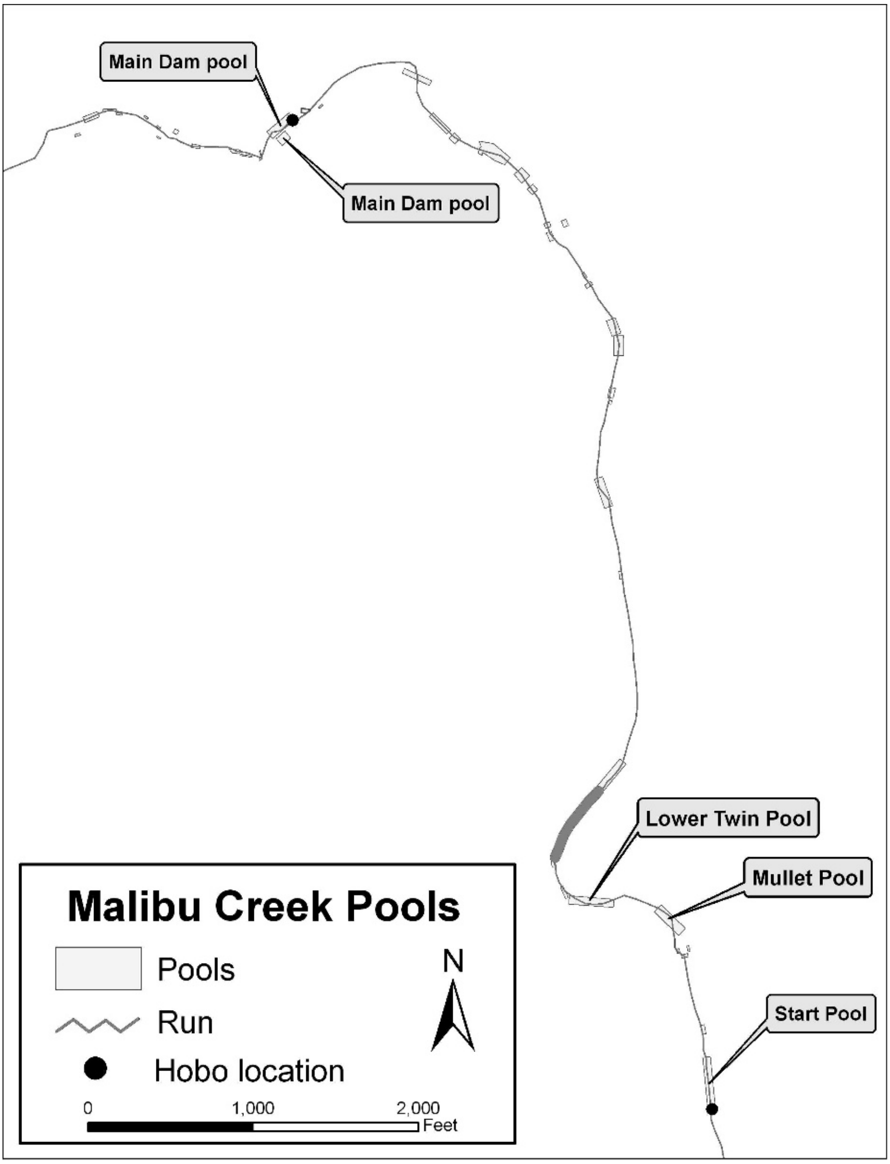

Fig. 2. Snorkel Survey reach showing pool locations, Malibu Creek. The start pool is approximately 500 m upstream of the upper end of Malibu Lagoon. (Map provided by M. Grimmer, Heal the Bay).

the upper and lower reach of the study area. The dam pool has continuous flow under even extreme drought conditions when portions of the lower reach go dry or are shallow and experience greater temperature fluxes.

Data were collected in the start pool from July 29–October 21, 2005. Breaks in the data occurred when the logger was exposed to air. The logger in the dam pool was stolen prior to downloading in 2005. In 2006, a logger in the dam pool collected data from May 18–July 19 (it was then stolen), and in the start pool from July 18–September 12. In 2008, the logger was installed in lower twin pool (start pool was too shallow) from 13 June–July 24 (when the logger was stolen) and in the dam pool from June 13–September 10.

All temperature data were examined and outliers were removed if we had evidence (based on either field observations or data irregularities) that the logger had been recording while out of the water. Daily maximum and mean temperatures were calculated. Data were also examined to determine proportion of time that water temperatures exceeded 27.5 degrees C at each location.

Grab samples were collected monthly in 2005–2006 by the Heal The Bay Malibu Stream Team just below the start pool (HtB- 1) and above the influence of the Tapia Wastewater Treatment Plant located approximately 1.6 km upstream of Rindge Dam, and tested for nitrate-N, ammonia- N, orthophosphate, pH, turbidity, and conductivity. The LVMWD also collects water quality data regularly from the outfall of the Tapia Wastewater Treatment Plant. In September 2006, samples were specifically collected and tested for a suite of toxic pollutants and heavy metals. A chronic bioassay test on *Ceriodaphnia* and Fathead minnow with copper chloride control was also conducted (Aquatic Bioassay Report 2006).

Application records for the mosquito larvicide *Bacillus thuringienses isrealensis* (Bti) were obtained from the LACVC for the four locations within or affecting the study reach where the fish die-off occurred.

In December 2006, sediment samples were collected and tested for toxicity using the freshwater amphipod *Hyalella azteca*, by the Nautilus Environmental Bioassay Laboratory, San Diego, CA.

Grab samples of the muck were collected in three locations (start pool, mullet pool and lunch pool) in August, September and December 2006. Each sample included both the surface scum of white, and the more dominant black, sulfurous smelling bulk. The muck felt gelatinous, and dispersed into a cloud when touched. Samples were kept cool and transported for examination by microscope by Dr. Robert Sheath at CSU San Marcos, Robert Gilbert at UCLA, and Dr. Tom Dudley at UC Santa Barbara.

In January 2007, over 100 New Zealand mud snails and 10 native snails were collected alive from the impacted study reach. Additional live samples were collected upstream in Malibu Creek State Park, where no fish die-off was observed. These snails were examined for trematode infestations by Dr. Brian Fredensborg at UC Santa Barbara.

A total of three *O. mykiss* were caught by hook and line in September 2006 under the supervision of the National Oceanographic and Atmospheric Administration (NOAA) Office of Law Enforcement. Gross pathology, parasitology, bacteriology and histology tests were performed by CDFG Fish Health Laboratory veterinarian Dr. Joe Maret, both in the field and in subsequent laboratory efforts. Dr. Ron Hedrick (UC Davis) reviewed and commented on the histology.

Results

Snorkel Surveys

In May 2006, a total of 245 trout were observed, along with the usual suite of exotic species dominated by crayfish, carp, bluegill (*Lepomis macrochirus*), green sunfish (*L. cyanellus*), black bullhead (*Ameiurus melas*), mosquitofish (*Gambusia affinis*), fathead minnow (*Pimephales promelas*), and largemouth bass (*Micropterus salmoides*). That same month, New Zealand Mud Snails were identified in a macroinvertebrate sample from Malibu Creek that had been collected in 2005 by Heal the Bay. Researchers from a variety of agencies working in streams in the Santa Monica Mountains voluntarily suspended field work until decontamination procedures for field equipment and clothing were established to prevent spread of the snails. Hence, no snorkel survey was conducted

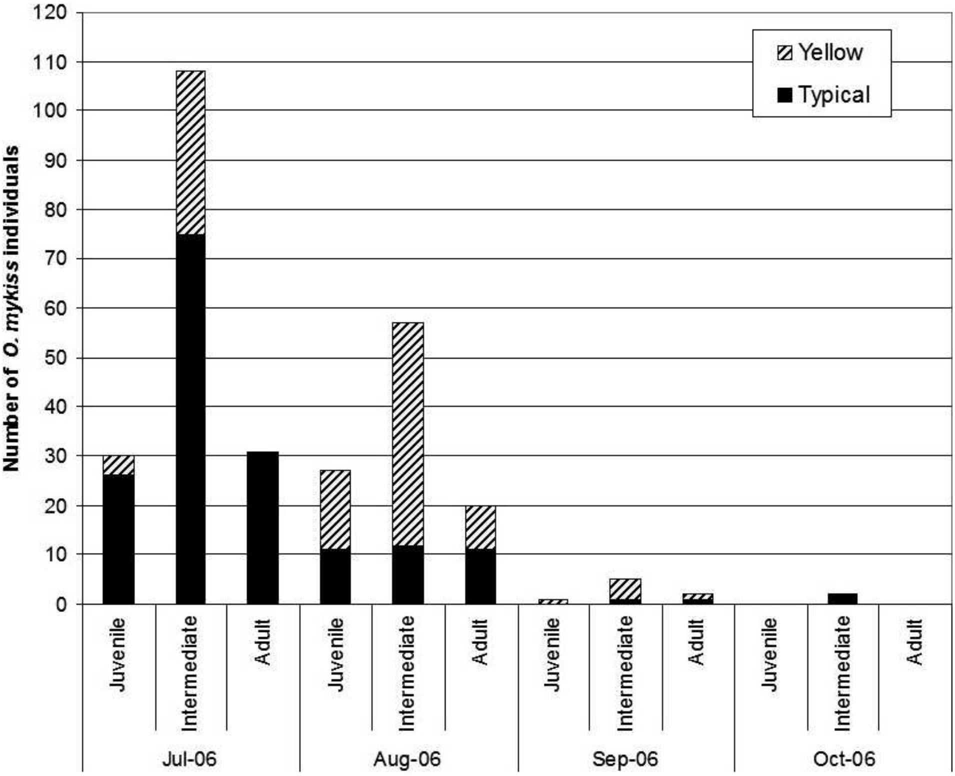

Fig. 3. Yellow coloration progressed through size classes of *O. mykiss* in July and August 2006. In September 2006, abundance decreased dramatically and by November 2006 no fish of any species were observed in the study area of Malibu Creek. Juvenile = 0–10 cm, Intermediate = 11–25 cm, Adult > 26 cm total length.

in Malibu Creek in June 2006. During the July 18–19 2006 survey, we observed 37 trout with yellow coloration, all under 15 cm, as well as another 145 trout with normal coloration in all size classes. This corresponded to the start date of a 12-day high temperature episode (described below).

In August 2006, the number of trout observed exhibiting normal coloration was 36, and the yellow trout of all size classes increased to 75. Dead and dying crayfish were noted throughout the study area and a 75 cm carp that appeared ill was caught by hand. In September 2006 few fish of any species were observed. A total of nine trout were observed, of which seven exhibited the yellow coloration. After collecting three trout for pathology and histological testing in September, only two normal trout were observed in October (Figure 3). From November 2006 until March 2007, no trout and only a few carp were seen (Figure 4). The lagoon was open when the berm was breached for much of this time, allowing both in and out migration.

In April 2007, 11 trout between 35–65 cm were observed, still retaining the steel gray color indicating recent movement from the ocean. In June 2007, we counted a total of 32 trout, including eight young of the year, indicating that some spawning had occurred. Unfortunately, the rebound of exotic fishes and crayfish populations were explosive, with crayfish literally covering the bottom in some pools. Due to funding constraints, surveys were suspended until September 2007, when a total of 30 trout were recorded. Of these 30

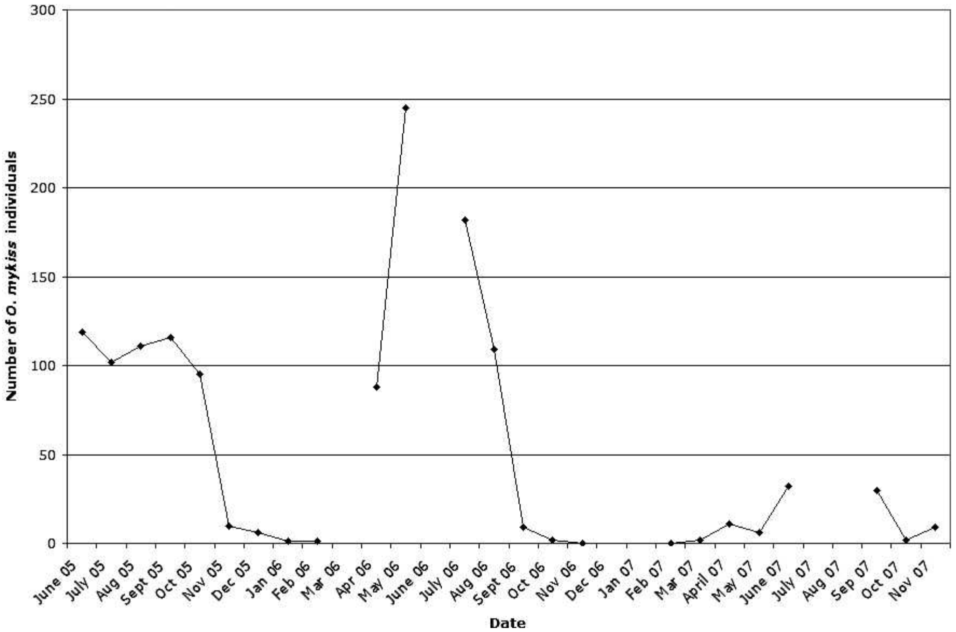

Fig. 4. Abundance of *O. mykiss* in Malibu Creek June 2005–November 2007.

trout, three adults (over 30 cm total length) were quite pale in color and one adult exhibited the same yellow color and sluggish behavior observed the previous year. By November 2007, the number of trout observed had dropped to nine adult fish, possibly due to reduced visibility during the survey. A subsequent stream walk survey done in January 2008 by CDFG biologists observed a total of five adult trout and a recently excavated redd (McKibbin, 2008).

Snorkel surveys conducted in June and July 2008 counted over 2,500 trout, with fish under 10 cm most abundant (2,293 in June and 2,327 in July). In June surveys, 24 adults over 50 cm long were observed, compared to five in July. The remainder of the fish ranged from 10–25 cm in length. Invasive non-native species have also rebounded, attaining abundances similar to those observed before the die-off event.

Flow

Flow was continuous throughout the study area and to the ocean during the entire die-off event, with levels ranging between 0.141–0.297 cubic meters/second (cms), with mean flows of 0.2175 cms in July 2006, 0.1662 cms in August and 0.1494 cms in September 2006.

Water temperature

In 2006, water temperatures ranged between 15.68–29.15 degrees C (Figure 5). Peak temperatures were observed from July 18–30, when water temperatures exceeded 27.5 degrees C, a temperature commonly considered to be near the upper end of the range of *O. mykiss*, for much of this 12-day period (Table 1). The proportion of the time in which temperatures exceeded 27.5 degrees C was much greater in 2006 than were observed in either 2005 or 2008 (Figure 6). Table 2 compares the percent time at each 2-degree temperature interval recorded in 2005, 2006, 2008. No temperature data were collected in 2007. The 2006 data set consists of information generated from two different reaches of

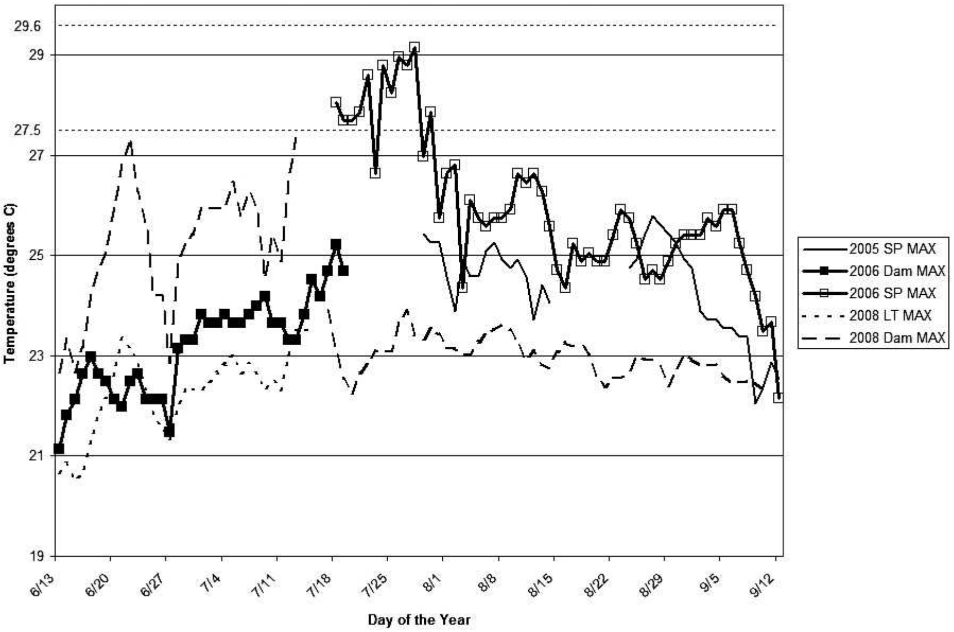

Fig. 5. Daily maximum and mean of Water Temperatures recorded in Malibu Creek from June–September 2005, 2006, 2008.

the creek because the logger in the dam was stolen following the July 19 download. It is not clear if the lower maximum temperatures observed in the dam pool prior to July 18 also reflect conditions downstream at the start pool.

Water Quality

Nutrient levels documented by HTB during the months of the fish die off did not differ significantly from levels regularly documented in recent years. Using its site HTB-1, which is located at the downstream end of the study reach just below the start pool (Figure 2), nitrate-nitrites ranged from a high of 13.5 ppm with a mean value of 2.76 ppm

Table 1. Duration of time that logged water temperatures equaled or exceeded 27.5°C, 2006.

Date	Hours >27.5°C	Hours >28°C	Hours >29°C
7/18/06	4	1.5	0
7/19/06	2	0	0
7/20/16	2.5	0	0
7/21/06	3	0	0
7/22/06	6	4	0
7/23/06	0	0	0
7/24/06	7	5.5	0
7/25/06	7	3	0
7/26/06	10	6.5	0
7/27/06	8	6	0
7/28/06	10	8	1.5
7/29/06	0	0	0
7/30/06	4	0	0

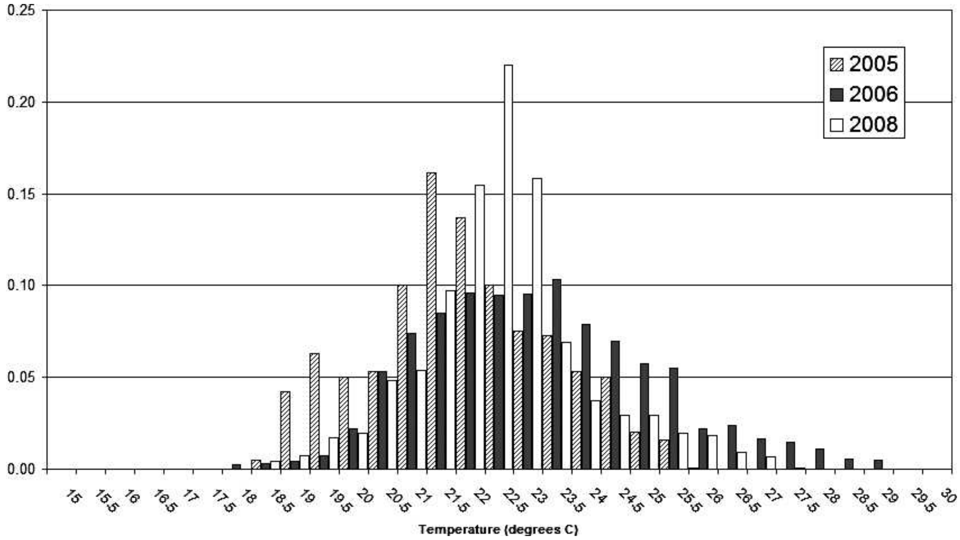

Fig. 6. Frequency of occurrence of water temperatures measured at 15–30 minute intervals in steelhead occupied pools in Malibu Creek, 2005, 2006, 2008 (2005 N=1,791; 2006 N=4,445; 2008 N=8,750).

during the fish die-off time frame. Ammonia-nitrogen levels were 0.35 ppm during August 2006, and orthophosphate levels were 0.56 ppm. A summary of all data is found at www.healthebay.org/streamteam/data/chem/query.

Dissolved oxygen levels were only directly measured mid-day at the surface, and were found to be 10 mg/l. No continually recorded DO data sets are available for Malibu Creek.

No evidence could be found for any toxic spills or acute pollution event. Toxicity testing of the grab sample collected on September 21, 2006 by the LVMWD was negative.

Bti application by the LACVC followed the same procedures as previous years, with a volume of 0.4841–0.9682 liters per event released at each of four locations within the study reach. Two of these locations received applications at the normal rate between June and August 2006 (M6 and M11)(LACVC 2006).

Sediment Toxicity

At the request of the Technical Advisory Committee, a sample of sediment was collected on 14 December 2006, refrigerated and shipped to Nautilus Environmental Bioassay Laboratory, San Diego for analysis. Mean survival of the amphipod *Hyallela*

Table 2. Percentage of logged time that water temperatures occurred in Malibu Creek between June 13–September 12 2005, 2006, 2008. (2005 N=1,791; 2006 N=4,445; 2008 N=8,750).

Degrees C	2005	2006	2008
<16.75	0%	0%	0%
16.75–18.75	1%	1%	0%
18.75–20.75	21%	9%	9%
20.75–22.75	50%	35%	53%
22.75–24.75	25%	35%	29%
24.75–26.75	4%	16%	8%
>26.75	0%	5%	1%

Table 3. Taxa found in Malibu muck samples collected in 2006.

1.	*Enteromorpha clathrata* (green algae)
2.	*Pleurosira laevis* (marine diatom)
3.	*Phormidium retzii* (cyanobacteria)
4.	*Oscillatoria tenuis* (cyanobacteria)
5.	*Scenedesmus quadricauda* (green algae)
6.	*Closterium sp.* (freshwater green algae)
7.	*Amphora sp.*(diatom)
8.	*Entonomeis sp.*
9.	*Cymbella sp.* (diatom)
10.	*Synedra sp.* (diatom)
11.	*Frustulia sp.* (diatom)
12.	*Spirogyra sp. 2* (green algae)
13.	*Cladophora cf. glomerata* (green algae)
14.	*Epithemia sp.* (diatom)
15.	*Cocconeis sp.*(diatom)

azteca in the sediment control was 92%, and mean survival of the test sediment was 98%. No toxicity was evident. (Nautilus Environmental 2007).

Muck Composition

Examination revealed a diverse community of both high and low ionic condition periphyton species, which are common to nutrient enriched environments such as that found in Malibu Creek (Table 3). No unusual or toxic species were noted (T. Dudley; R. Gilbert, R. Nhemh, unpublished data).

Pathology and Histology Results

Three adult trout ranging in size from 15–30 cm total length were captured by hook and line. All fish were examined alive and then killed for further examination. All fish appeared normal. The stomach of the 30 cm female was full of green algae. Anchor worm, a common skin parasite, was found on two of the three fish. Two of the three fish exhibited unusual inflammation of the olfactory rosette, which was not explainable. Digenetic trematode metacercariae were encysted in the dermis sections of their heads. Unfortunately, it was not possible to identify the species of metacercariae to determine if it was associated with a vector of native or non-native snails (Maret 2006). Otoliths were destroyed in the preparation of the histologic samples, so age was undetermined. Fin clips were sent to the NMFS Genetic Tissue Laboratory in Santa Cruz, CA for analysis.

Snail Examination

In order to address the question of whether the metacercariae were associated with native or non-native snail vectors, over 100 New Zealand mud snails and 10 native snails were collected from two locations in Malibu Creek in February 2007. No trematodes were found (Lund Fredensborg, pers. comm.)

Discussion

Despite concerted effort, it has not been possible to assign a single causal agent to the total loss of all species in lower Malibu Creek during the die-off period of July–December 2006. It seems unlikely that high temperatures alone explain the die-off. Infectious agents are an unlikely causative agent due to the wide taxonomic range of organisms (from vertebrates to arthropods) affected. Based on die-offs observed in other locations, dissolved oxygen remains notable as a potential stressor, and low DO levels, combined with seasonally high water temperatures and consistently high levels of nutrients found in the water, could have led to the die-off. However, stream flow was continuous from July–December 2006, indicating that mixing was occurring and all habitat units were hydrologically connected. Of all possible contributing factors identified, the 12-day high temperature episode in July 2006 and the presence of the muck covering all wetted substrate, appear to be the two variables that are different from conditions observed in other years.

While we could find no accounts of the yellow coloration previously occurring in trout from Malibu Creek or other streams in Southern California, pale yellowish coloration has been noted in *O. mykiss* from other watersheds. Yellow trout were observed in the Salinas River during snorkel surveys in 2006–2007, and were associated with high temperature conditions (L. Thompson, pers. comm.). A study of summer water temperature conditions in the Eel River system also contained observations of both live and deceased yellow trout correlated with temperatures of between 28 and 30+ degrees C (Kubicek 1977), and this same study discussed previous sightings of yellow trout (Wales 1938, as described in Kubicek 1977). In 1938, Wales observed yellow trout in the Eel River associated with a heat wave. Wales attributed the yellow coloration to a loss of melanophore control due to disease or parasitism, but was unable to identify any causative agent.

Other observations of yellow coloration in trout have been associated with genetic mutation (Dobosz et al. 2000), though given the progression of this phenotype through the population, and the correlation with illness, we do not think this played a role in Malibu Creek.

Fish physiologists recognize that extreme heat stress could cause a loss of coloration in salmonids (Cech, pers. comm., Cech et al. 1990). Unlike the compromised water quality found in Malibu Creek, the Eel River was thought to have high water quality when yellow trout were observed. In situations where non-lethal levels of toxicants or pathogens are present along with high temperature conditions, it is possible that the cumulative impacts could result in a loss of melanophore control, stress related illness, and subsequent death even at temperatures below 28.0 degrees C.

Parasite infection such as the trematode infection observed in two of the three trout autopsied could add to the cumulative stress impacts, though it would appear that parasitism alone was not sufficient to cause death (J. Maret; J. Cech, pers. comm.). Parasitism could however be a factor contributing to a reduced ability to withstand environmental stresses, as has been observed in a related species (Keefer et al, 2007).

Thermal stress is a chronic concern in small southern California coastal streams that support these endangered fishes. In 2006, temperatures in Malibu Creek were observed to remain at the upper end of the documented thermal limits of *O. mykiss* for a 12-day period in July, with spikes of over 27.5 degrees C for up to 10 hours at a time. However, it remained below 29.6 degrees C, the critical thermal maximum observed for steelhead acclimated to high temperatures (19 degrees C) (Myrick and Cech, 2005; for a detailed discussion of the temperature, tolerances of southern steelhead see Spina 2007).

The 2006 high temperature event differs from the temperature patterns observed in 2005 and 2008 when fish mortality was not observed. In 2005, the temperature never exceeded 26.75 degrees C, and was over 22.75 degrees C for less than 30% of the time between July 29 and October 21. In 2008 short spikes of higher temperatures were observed less than 1% of the time between July and September, concurrent with the presence of thousands of young of the year and larger trout. This contrasts with 2006, when temperatures remained over 27.5 degrees C for between 2–10 hours daily, starting on the same day that the first yellow trout were observed. Unfortunately, we do not have temperature data from 2006 from this reach prior to July 18, so it is not possible to determine if the yellow trout were responding to a previous or perhaps more prolonged high temperature event, but the data from the dam pool suggests that water temperatures in the creek were lower. It is important to note that the percentage and size class of yellow trout increased significantly in the months that followed this prolonged high temperature event, as did mortality.

O. mykiss in other southern California streams have been found to withstand similar temperatures to those we observed. Data from nearby Topanga Creek (2004–2006) indicates that resident fish routinely survive daily spikes of up to 26 degrees C (observed in late afternoon), although during the months of May through October the daily averages are closer to 18–20 degrees C (Dagit et al 2007). Carapanzano, (1996) recorded maximum temperatures up to 28 degrees C in the Ventura River. Researchers working in Sespe Creek in Ventura County also documented temperatures reaching 28 degrees C for short time periods in refugia pools where *O. mykiss* remained for most of the summer (Matthews and Berg 1997). Spina (2007) determined that juvenile steelhead were able to forage and remain active with an elevated body temperature, supporting the idea that *O. mykiss* found in the southern California region may be able to withstand, and possibly even thrive, in temperatures that would stress steelhead from cooler areas.

Despite their higher temperature tolerance, exotic species observed in Malibu Creek were found to die off along with *O. mykiss* in 2006. Temperature ranges for these species vary as described in Moyle (2002), but are consistently in line with or higher than the maxima we observed in 2006, and higher than those reported in the literature for *O. mykiss*. Common carp prefer temperatures from 4–24 degrees C, but can withstand temperatures between 31–36 degrees. Bluegill can survive temperatures as high as 40–41 degrees C but prefer temperatures between 27–32 degrees C and green sunfish prefer temperatures between 26–30 degrees C, but can survive high temperatures $>$38 degrees C. Black bullhead can survive temperatures up to 38 degrees C under laboratory conditions. Mosquitofish can occur at temperatures between 0.5 and 42 degrees C, but persist at temperatures between 10–35 degrees C. Fathead minnow prefer temperatures between 22–23 degrees C, but can withstand temps up to 33 degrees C. Optimal temps for largemouth bass are 25–30 degrees C, but they can handle temperatures ranging from 10–35 degrees C. Red swamp crayfish can withstand temperatures from 5–38 degrees C (Wizen, et al, 2008).

O. mykiss in Southern California and other areas are routinely observed living in warm streams, and laboratory tests confirmed their ability to acclimate to higher temperatures. However, the temperatures observed in Malibu Creek during most of the 2006 summer season were at the high end of the temperature tolerance of this species, and exceeded 27.5 degrees C for 12 days. Malibu Creek trout did not exhibit visible signs of stress, yellow coloration, or mortality in 2005 or 2008 when temperatures were slightly cooler, but still above 24 degrees C for 4% and 9% of the summer. Exotic species with significantly higher temperature tolerances also died only in 2006. Therefore, we do not think that heat stress alone can explain the die-off, but likely contributed to sensitivity to other factors.

While Southern California trout may be able to tolerate exposure to high temperatures, it still appears that they will select for lower temperatures and higher oxygen content when these conditions are available (Santa Ynez River Technical Advisory Committee

2000). Dissolved oxygen levels were not measured during the die-off, although flow was continuous, potentially indicating that oxygen levels might have been maintained through physical mixing. In Sespe Creek, pools, or sections of pools having low dissolved oxygen levels were avoided by *O. mykiss*, who selected for pool areas with higher dissolved oxygen, even if the temperatures were a bit higher as well (Matthews and Berg 1997). It could be that the combination of biological oxygen demand related to the decomposition of the muck, in addition to the higher water temperatures, reached a stress threshold that proved to be too great for not only the *O. mykiss*, but also the other fish species and crayfish.

In 2007 and 2008 "Malibu muck" re-appeared and remains an element in the benthic zone. The possible influx of a transient toxin or pollutant cannot be totally discounted, and the continued presence of the muck indicates that if eutrophication contributed to the 2006 decline, the threat still remains. There is also concern that a pathogen associated with the diatom community could be a factor in the die-off, as was noted in Sycamore Creek, Arizona, where invasive bacteria caused a massive die-off of the benthic diatom community (Peterson and Dudley 1995).

The die-off was a sobering reminder of how quickly a small population can be extinguished and reinforces the need to monitor and preserve all geographic populations of southern steelhead. It also points to the need to have a rapid response plan in hand, in case such an event should occur again. To that end, the following protocol has been developed and permits are being obtained to facilitate a more coordinated and rapid response to observation of discolored or sick fish in the future.

Should trout be observed with the yellow coloration, sluggish swimming, gaping and/or other evidence of disease or illness, the following steps will be taken:

1. Notify NMFS and CDFG immediately and invite agency staff to confirm the problem.
2. Document conditions with video if possible (visibility can be a limiting factor) and send images to NMFS, CDFG pathologists for review.
3. Convene the ad-hoc Technical Advisory Committee (TAC) described above, and add additional expertise if needed.
4. Initiate continuous year round water quality monitoring to include, but not be limited to: temperature, dissolved oxygen, pH, nutrients, heavy metals, pesticides and toxicity. (A Troll 9500 probe is being installed near lower twin pool in 2008 to collect temperature, dissolved oxygen, conductivity, pH and pressure)
5. Check for acute pollution event, in collaboration with Las Virgenes Municipal Water District, Heal the Bay, Malibu Creek State Park, Los Angeles County Vector Control and the Los Angeles Regional Water Quality Control Board.
6. Evaluate in-stream conditions regarding muck and algal decomposition. Collect samples for examination of species composition, bacteria and toxicity in collaboration with UC Santa Barbara, Cal State San Marcos, UCLA.
7. Collect specimens of other fish species and crayfish that exhibit signs of stress or illness for examination by the CDFG pathologists.
8. Continue examination of other accounts of yellow morphology in trout.
9. Collect specimens of both native and New Zealand mud snails for examination for trematodes.
10. If conditions warrant, collect one – three affected *O. mykiss*, under the direct supervision of NMFS and CDFG pathologists, for both live examination and post-mortem histology and pathology testing.

11. If conditions warrant, install secured live car cages in the creek containing triploid, sterile, disease free hatchery rainbow trout to test survival and further infection potential, under the direct supervision of CDFG pathologists.

It is hoped that this die-off was an isolated incident and the recovery of both *O. mykiss* and other fish species in 2008 has demonstrated that an area can re-populate quickly following a traumatic event. Given the precarious status of *O. mykiss* in Southern California, we need to develop regional strategies for rapid response should this kind of event occur again.

Acknowledgments

This work was funded by grants from the California Department of Fish and Game Contract No. P0450012, Sea Grants Program Project No. R/FISH-128PD and Pacific States Marine Fisheries Commission No A06-S3. We would like to recognize the expertise and enthusiasm of the snorkel survey and fishing crew (Nat Cox, Ian Cuthbertson, Kevin Gautrey, Delmar Lathers, Lisa Openshaw, Jayni Shuman, Clark Stevens, Ken Wheeland, Ken Widen, Josh Widen, Steve Williams). Experts who contributed their time to examining samples and conducting tests included Dr. Tom Dudley, Robert Gilbert and Dr. Robert Sheath who examined the muck; Drs. Joe Maret, Mark Chifford and Ron Hickman of the CDFG Fish Pathology Lab, Rancho Cordoba; Drs. Armand Kuris and Brian Lund Fredensborg for their work with the snails, Dr. Randal Orton and the Las Virgenes Municipal Water District for their help with water quality analysis, Robert Saviskas of Los Angeles County Vector Control for his help with the Bti information, Mark Abramson, Mike Grimmer and Heal the Bay Stream Team for their assistance with water quality data, and Suzanne Goode, California Department of Parks and Recreation, for her help in obtaining the sediment toxicity data. Drs. Lisa Thompson, Paul Kubicek, and Joseph Cech contributed insights about yellow coloration and temperature stress around California. Dr. Camm Swift was ever generous with his thoughts. NMFS staff that contributed their insights included Anthony Spina and Stan Glowacki, along with Special Enforcement Officer Monica Hamm. CDFG Region 5 biologists Mary Larson, John O'Brien and Valerie Taylor also lent their expertise. Finally, the entire Technical Advisory Committee provided great guidance and networking related to this problem. We also thank our reviewers whose comments assisted us.

Literature Cited

Aquatic Bioassay and Consulting Laboratories. 2006. Chronic Fathead Minnow Survival and Growth Bioassay, 20 September 2006, Malibu Creek. Provided to Las Virgenes Municipal Water District, October 2006.

Carpanzano, C.M. 1996. Distribution and habitat associations of different age classes and mitochondrial genotypes of *Oncorhynchus mykiss* in stream in southern California. Masters Thesis UC Santa Barbara.

Cech, J.J., S.J. Mitchell, D.T. Castleberry, and M. McEnroe. 1990. Distribution of California stream fishes: influence of environmental temperature and hypoxia. Environmental Biology of Fishes, 29: 85–105.

Dagit, R. and M. Abramson. 2007. Malibu and Arroyo Sequit Creeks Southern Steelhead Monitoring. Prepared for Contract No. P4050012 California Department of Fish and Game. Resource Conservation District of the Santa Monica Mountains, Agoura Hills, CA.

———, B. Meyer, and S. Drill. 2005. Historical Distribution of Southern Steelhead Trout in the Santa Monica Bay. Prepared for NOAA Fisheries and CA Department of Fish and Game. Resource Conservation District of the Santa Monica Mountains, Topanga, CA.

———, K. Reagan, and V. Tobias. 2007. Topanga Creek Southern Steelhead Monitoring: Habitat Suitability and Monitoring Summary. Prepared for California Department of Fish and Game Contract No. P0450011. Resource Conservation District of the Santa Monica Mountains, Agoura Hills, CA.

Dobosz, S., K. Kohlmann, K. Goryczko, and H. Kuzminski. 2000. Growth and vitality in yellow forms of rainbow trout. J. of Applied Ichthyology, 16(3): 117–120.

Dudley, Tom. UCSB. Unpublished data regarding results of muck examination.

Flosi, G. and F.L. Reynolds. 1998. California Salmonid Stream Habitat Restoration Manual, Third Edition. Inland Fisheries Division, California Department of Fish and Game, The Resources Agency, Sacramento, CA.

Fredensborg, Brian Lund. 2007. email results of snail examination.

Gilbert, Robert. UCLA, 2006. Unpublished data regarding muck examination.

Heal the Bay. 2007. Stream Team Water Quality Monitoring Results (B1). www.healthebay.org/streamteam/data/chem/query

Keefer, M.L., C.A. Peery, and M.J. Heinrich. 2007. Temperature-mediated en route migration mortality and travel rates of endangered Snake River sockeye salmon. Ecology of Freshwater Fish, 17: 136–145.

Kubicek, Paul F. 1977. Summer Water Temperature Conditions in the Eel River System, with Reference to Trout and Salmon. Master's Thesis, Humboldt State University.

Las Virgenes Municipal Water District. 2007. Malibu Creek Flow measurements.

Lee, R.M. and J.N. Rinne. 1980. Critical thermal maxima of five trout species in the southwestern United States. Trans. of the American Fisheries Society, 109:632–635.

Los Angeles Regional Water Quality Control Board. 2007. 303(d) List of Impaired water bodies.

Los Angleles County Vector Control. 2006. phone conversation and maps providing locations, amount and timing of Bti releases.

Maret, J. 2006. Fish Pathologist Report, Malibu Creek below Rindge Dam, September 26, 2006. Calif. Dept. of Fish and Game, Fish Health Laboratory, Rancho Cordova, CA.

Matthews, K.R. and N.H. Berg. 1997. Rainbow trout responses to water temperature and dissolved oxygen in two Southern California stream pools. J of Fish Biology, 50:50–67.

McKibbin, C. 2008. CDFG report of Malibu Creek Stream Walk on 13 January 2008.

Myrick, C.A. and J.J. Cech, Jr. 2005. Effects of Temperature on Growth, Food Consumption, and Thermal Tolerance of Age-0 Nimbus-Strain Steelhead. North American Journal of Aquaculture, 63:324–330.

National Marine Fisheries Service. 2007. 2007 Federal Recovery Outline for the Distinct Population Segment of Southern California Coast Steelhead. NMFS Southwest Regional Office, September 2007.

Nautilus Environmental. 2007. Sediment Toxicity Evaluation for Malibu Creek December 2006. Prepared for Suzanne Goode, California Department of Parks and Recreation, Calabasas, CA. February 2007.

Peterson, C.G. and T.L. Dudley. 1995. Infection, Growth and Community-Level Consequences of a Diatom Pathogen in a Sonoran Desert Stream. J of Phycology, 29(4): 442–452.

Santa Ynez River Technical Advisory Committee. 2000. A Review of Effects of Warm Water Temperature on Steelhead/Rainbow Trout. Appendix G. Prepared for Santa Ynez River Consensus Committee. October 2, 2000.

Sheath, R. CSU San Marcos, 2006. Unpublished data regarding muck examination.

Spina, A. 2007. Thermal ecology of juvenile steelhead in a warm-water environment. Environmental Biology of Fishes. Vol. 80(1): 23–34.

Thompson, Lisa. 2006. emails and photos of yellow trout from Salinas River.

Wales, J.H. 1938. Mortality in young Eel River steelhead. California Department of Fish and Game. Inland Fisheries Branch Administrative Report No. 38-10.

Wizen, G., Bella S. Galil, Alex Shlagman, and Avital Gasith. 2008. First record of red swamp crayfish *Procambarus clarkia* in Israel – too late to eradicate? Aquatic Invasions, 3:181–185.