

Toxicity of Stormwater from Ballona and Malibu Creeks

By Steven Bay, Darrin Greenstein, Andrew Jirik, and Ann Zellers

ABSTRACT

Stormwater samples from Ballona Creek and Malibu Creek were collected during four storms in January and February, 1996, and tested to determine the magnitude and characteristics of toxicity. Toxic effects were measured using sea urchin fertilization and abalone larval development tests. Every sample of Ballona Creek stormwater collected during the four storm events was toxic to sea urchin sperm. Toxicity to red abalone larvae was also detected, but this test had lower sensitivity. Malibu Creek samples were also often toxic, but the magnitude of toxicity was usually lower than comparable samples from Ballona Creek. Phase I toxicity identification evaluations (TIEs) were conducted on selected samples from the two creeks. The TIE results were similar between samples and indicated that stormwater toxicants had characteristics similar to trace metals. Dissolved copper and zinc concentrations were near toxic levels in many of the Ballona Creek samples, but a significant correlation with toxicity could not be established with the limited number of samples tested. The TIE response patterns indicated that little (if any) of the toxicity was associated with nonpolar organics (e.g., pesticides and petroleum hydrocarbons), particulates, or oxidants.

INTRODUCTION

Surface runoff (stormwater and dry weather flow) has become an increasingly important source of contaminants to the Southern California Bight (SCB). Although municipal wastewater discharge was a major contributor of contaminants in the 1970s (SCCWRP 1973), improvements in source control and treatment methods over the past two decades have dramatically reduced contaminant inputs from this source. During this period, runoff inputs changed little. Consequently, recent estimates of SCB contaminant inputs indicate that runoff now contributes comparable or greater amounts of contaminants to the SCB than does municipal wastewater (SCCWRP 1996).

Municipal wastewater discharges have been intensively monitored since the early 1970s, providing a record of variations in effluent toxicity, chemical composition, and marine community responses. In contrast, little information has been collected on the composition, toxicity, and ecological effects of runoff discharges to the marine environment. An assessment of the environmental effects of contaminants in storm drains is necessary to evaluate management options for source control or treatment of storm drain discharges. In addition, knowledge of the characteristics of the contaminants and their toxicity is needed to assist environmental managers in determining the best management practices to reduce biological impacts.

Urban runoff consists of two broad types of discharges, dry weather flow and stormwater. Dry weather flow is a relatively consistent source of runoff discharge, resulting from daily residential activities (e.g., landscape irrigation) and permitted discharges of effluents from events such as groundwater pumping, swimming pool maintenance, and air conditioning condensate. Stormwater discharges are markedly different, consisting of highly variable flows that provide 50 to 80% of the annual runoff discharge during relatively few days (approximately 10%) of the year. Approximately 70 to 97% of the annual mass emission of runoff contaminants result from stormwater (SCCWRP 1992).

Previous studies of southern California stormwater have detected toxicity in samples from most locations investigated (SCCWRP 1990, Schiff and Stevenson 1996, Skinner *et al.* 1994, Kinnetic Laboratories Inc. 1995). Use of these data to evaluate effects on marine animals is difficult, however, because toxicity tests were often conducted using microorganisms or freshwater animals. Since the ultimate fate of river runoff is the marine environment, tests using marine organisms are essential for determining potential impacts.

The value of toxicity tests has been greatly enhanced in recent years by the refinement of methods for conducting TIEs. The initial phase of most TIEs includes various physical/chemical manipulations to separate a toxic sample into well-characterized fractions. Once characterized, subsequent TIE procedures are applied to identify specific chemicals contributing to the toxicity. The TIEs are usually conducted with freshwater species and have been success-

fully applied to municipal and industrial effluents, receiving water, and sediment interstitial water. A few stormwater samples also have been evaluated with TIEs (Cooke and Lee 1993). Some TIE procedures are compatible with marine toxicity test species (Burgess *et al.* 1995) and have been used in preliminary investigations of dry weather urban runoff (Bay *et al.* 1996). Application of TIEs to stormwater has the potential to provide substantial benefits to agencies responsible for stormwater management, as treatment or source control actions can be focused to reduce those chemicals contributing to the toxic input.

The objectives of this study were (1) to compare the magnitude of toxicity from two subwatersheds representing different land uses, and (2) to characterize the toxicants present.

MATERIALS AND METHODS

Sample Collection

Samples were collected from Ballona Creek and Malibu Creek during four storm events that occurred in January and February 1996. These creeks were selected because they represent two of the largest sources of stormwater discharge to Santa Monica Bay and have different land use patterns (Figure 1). Ballona Creek carries runoff from highly urbanized areas, and previous studies have identified toxicity in dry and wet weather discharges for this area (SCCWRP 1990, Bay *et al.* 1996). The Malibu Creek site, which was selected as a reference, drains predominantly undeveloped land in the Santa Monica Mountains.

Samples from Ballona Creek were obtained from the bridge at Sawtelle Boulevard, approximately 6 km upstream from the mouth. Samples from Malibu Creek were obtained from a bridge located just downstream of the Tapia Water Reclamation Facility (TWRP), approximately 6 km upstream of the creek's discharge into Malibu Lagoon. Both sites were located near flow gauging stations oper-

ated by the Los Angeles County Department of Public Works (LACDPW).

Surface water samples were collected from each creek using a stainless steel bucket (samples for metals analysis were collected with a plastic bucket). The samples were transferred to 3.8 L glass jars and chilled with ice until they were delivered to the laboratory. At the laboratory, samples were stored at 5 °C until they were used for toxicity tests. Toxicity tests were initiated within 48 h of the end of each storm sampling event. A summary of the sampling dates is shown in Table 1. The first two storms in January were relatively small, and only one sample was collected from the middle of each event. Four to five samples were collected from each site during the last two storms. Sampling was distributed over the course of the storm in order to obtain runoff samples representative of initial, peak, and ending flows. Most of the samples were tested individually, except for three samples from the final Malibu Creek sampling that were composited (equal volumes per sample) into a single sample for toxicity measurement.

Toxicity Measurement

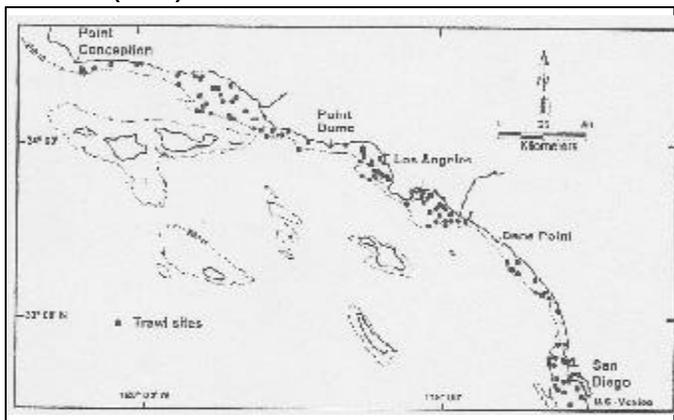
Two toxicity tests were used: the 48-h red abalone (*Haliotis rufescens*) larval development test, and the 20-min purple sea urchin (*Strongylocentrotus purpuratus*) fertilization test. These tests are among the methods recommended by the State of California's Water Resources Control Board (SWRCB 1996) for evaluating the toxicity of discharges to the marine environment.

Larvae of the red abalone (*H. rufescens*) were exposed to selected stormwater samples (Table 1) for 48 h according to the method described by Chapman *et al.* (1995). Newly fertilized eggs were added to 10 mL of test solution in glass vials and allowed to develop at 15 °C for 48h. The resulting larvae were preserved and examined with a microscope to assess the percentage of normal development. Toxic effects are expressed as a reduction in the percentage of normally developed veliger larvae.

All samples were tested using the purple sea urchin (*S. purpuratus*) fertilization test (Chapman *et al.* 1995). The test consists of a 20-min exposure of sperm to 10 mL samples at 15 °C. Eggs are then added and given 20 min for fertilization to occur. The eggs are preserved and examined later with a microscope to assess the percent fertilized. Toxic effects are expressed as a reduction in fertilization percentage.

Stormwater samples were centrifuged (3,000 x g for 30 min) in order to remove most of the particulates before testing. Particulates were removed to minimize potential test interferences; prior studies have shown that particulate removal has little effect on runoff toxicity. The salinity of

FIGURE 1. Location of Ballona Creek and Malibu Creek watersheds. Land use data are from Stenstrom and Strecker (1993).



the sample was then adjusted to 34 g/kg (typical seawater value) by the addition of brine produced by freezing seawater. Laboratory seawater was added to produce concentrations of 50, 25, 12, 6, and 3% stormwater. Toxicity test organisms were added to each sample within three h of dilution. Three replicates of each concentration were tested.

The salinity, dissolved oxygen, pH, and total ammonia content of each test sample was measured at the beginning of the toxicity tests. Measurements were made using electrodes that were calibrated daily. Water quality measurements were also made at the termination of each 48-h abalone development test. Electronic thermometers were used to measure water temperature continuously throughout each experiment.

Selected samples from Ballona and Malibu Creeks were treated using modifications of EPA Phase I TIE methods (Norberg-King *et al.* 1992) in order to characterize the toxicants present. The TIEs were conducted on a field duplicate sample stored at 5 °C. The Phase I manipulations consisted of filtration through a 1.0 µm glass fiber filter followed by three treatments (Figure 2): solid phase extraction using a C18 column, addition of 3 to 30 mg/L ethylenediamine tetraacetic acid (EDTA), or addition of 10 to 25 mg/L sodium thiosulfate. The TIE manipulations were conducted on stormwater samples prior to salinity adjustment. Sea urchin fertilization tests were used to evaluate the effectiveness of the TIE treatments. Each treatment was adjusted to normal seawater salinity and diluted to produce concentrations of 50, 25, or 12%.

Chemical Analysis

Each stormwater sample was analyzed to determine the concentration of dissolved trace metals using procedures described in EPA Method 200.7 (EPA 1983).

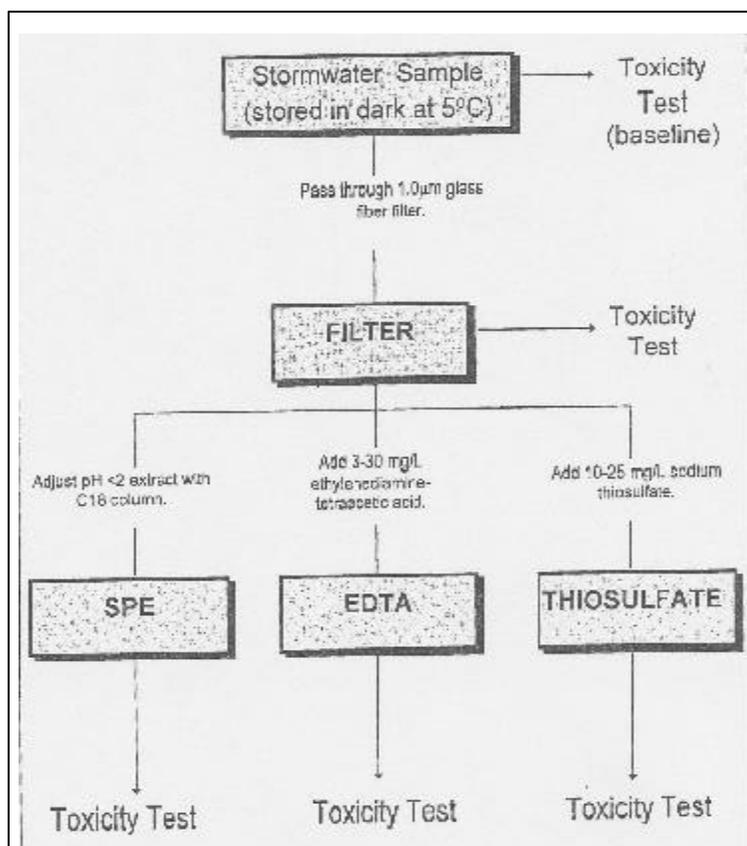
Water samples were filtered (1 µm glass fiber filter) within 24 h of collection, acidified with nitric acid, and stored at 4 °C until digested. Hydrogen peroxide was added to the filtrates in order to digest particles that may have passed through the filter. Samples were analyzed using an inductively coupled plasma atomic emission spectrometer (ICP-AES).

Data Analysis

The highest concentration that did not produce significant toxicity (NOEC) was calculated for each sample. This value was determined by analysis of variance followed by a multiple comparison test to determine which sample concentrations were significantly different from the seawater control ($p \leq 0.05$). Median response values (EC50, the concentration producing a 50% toxic effect) were calculated by probit analysis in most cases. Some data could not be analyzed by probit analysis; EC50s for these samples were estimated by nonlinear interpolation. Toxicity data were converted to toxic units (100/EC50). This conversion facilitates comparisons between samples by scaling the data so that it is directly proportional with toxicity magnitude. Pearson product moment correlation coefficients

were calculated to identify potential relationships between toxicity and trace metal concentrations.

FIGURE 2. Flow chart of toxicity identification evaluation (TIE) manipulations applied to stormwater samples. Toxicity changes were measured with the sea urchin fertilization test.



RESULTS

Relative Toxicity

Only a single sample from each creek was obtained for testing during the first two storm events studied (January 16 and 21, 1996). No toxicity was detected in either Malibu Creek sample at the highest concentration tested (50%), resulting in NOEC and EC50 values of >50% (Table 1). No evidence of increased flow was found in Malibu Creek during sampling, indicating that the water samples represented base flow conditions instead of stormwater. These two storms were relatively small (less than 0.5 inch of precipitation), and it is

assumed the rain was absorbed by soil in the Malibu watershed.

Elevated flow was present in Ballona Creek during collection of the January 16 and 21 samples. Both the sea urchin and abalone tests detected toxicity in these samples (Table 1), with greater toxicity being present in the January 16 sample.

Four water samples from each creek were evaluated for toxicity during the third sampling event on January 31 and February 1, 1996. The sea urchin fertilization test was consistently more sensitive than the abalone development test, detecting a greater magnitude of toxicity in every sample (Table 1). Comparison of the dose-response curves produced by each test method (Figure 3) illustrates the greater responsiveness of the sea urchin fertilization test to stormwater. The abalone development test detected toxicity in only one Malibu Creek sample (initial low flow)

and three of the four Ballona Creek samples (Table 1). The initial low flow sample from Malibu Creek also produced the greatest toxicity to sea urchin sperm.

Toxicity of stormwater collected during the final sampling period (February 19 to 22, 1996) was evaluated using only the sea urchin fertilization test. The four Ballona Creek samples from this set contained a similar magnitude of toxicity as previous samples, while Malibu Creek toxicity was detected in only the initial sample, apparently representing base flow (Table 1). Of the three Malibu Creek samples tested from the final collection event, only the initial sample (base flow) was toxic to sea urchin sperm.

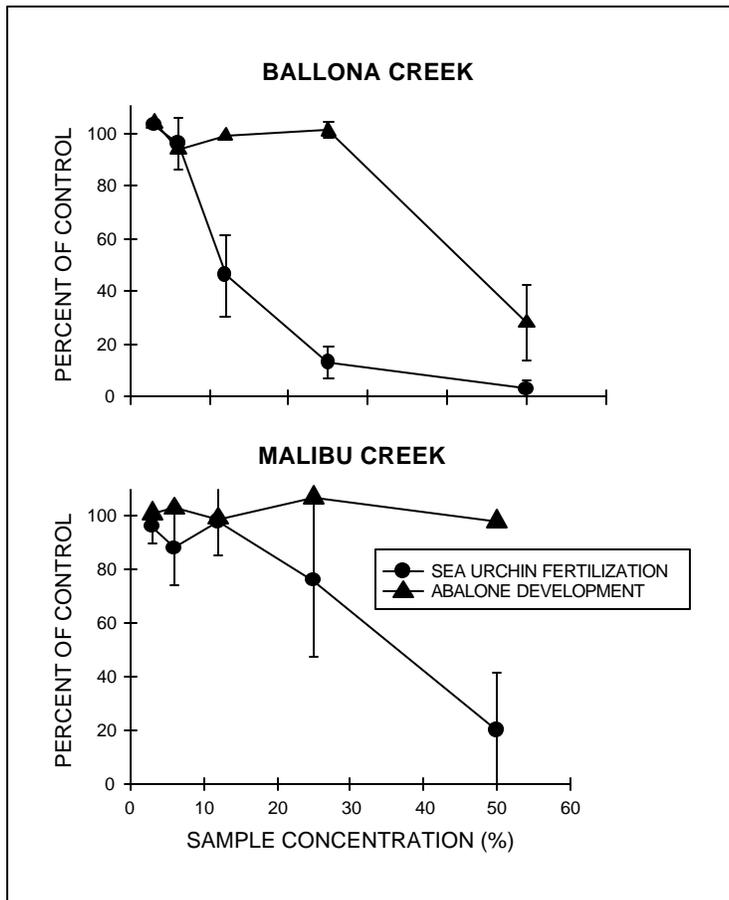
No consistent relationship was found between toxicity magnitude and variations in Ballona Creek flow during a storm (Figure 4). Samples obtained at the beginning of a storm had similar toxicity to samples obtained during much higher flow rates. No relationship was evident between

TABLE 1. Summary of toxicity test results for stormwater samples from Ballona and Malibu Creeks. Samples from the same creek are listed in chronological order. Creek flow rate above nonstorm value and the peak flow for each storm event were obtained from unpublished monitoring data from the Los Angeles County Department of Public Works. The abalone embryo development test was not conducted on stormwater collected 2/19-22/96.

Date	Flow (cfs)		Sea urchin fertilization			Abalone development		
	Storm	Peak	NOEC	EC50	95% CI	NOEC	EC50	95% CI
Malibu Creek								
1/16/96	0		≥50	>50		≥50	>50	
1/21/96	0	0	≥50	>50		≥50	>50	
1/31/96	1	3	3	5 ^d	NC	25	44	NC
1/31/96	1		25	35	32-39	≥50	>50	
2/01/96 ^a	2		12	27	23-30	≥50	>50	
2/01/96	1		25	28 ^d	NC	≥50	>50	
2/19/96	0	476	≥50	32	24-41			
2/20-21/96 ^e	4-77		≥50	>50				
2/22/96	0		≥50	>50				
Ballona Creek								
1/16/96 ^a	NA ^b	NA	6	11	10-13	12	22	NC ^c
1/21/96 ^a	2,600	4,700	25	>50		25	>50	
1/31/96	325	9,500	6	13 ^d	8-17	25	39	NC
1/31/96 ^a	4,200		6	14	12-15	25	49	NC
1/31/96	7,875		6	20	17-22	≥50	>50	
1/31/96	275		6	14 ^d	6-24	25	>50	
2/19/96 ^a	400	9,750	3	12	10-14			
2/19/96	3,250		12	12	8-15			
2/20/96	1,900		12	16	12-19			
2/21/96	250		25	30 ^d	NC			

^aTIE sample.
^b NA = Data not available.
^c NC = Not calculated; confidence limits could not be calculated due to poor fit to probit model or use of linear interpolation to estimate EC50.
^d Poor fit of data to probit model (heterogeneity or slope not significantly different from zero).
^e Composite of three samples.

FIGURE 3. Dose-response plots for purple sea urchin fertilization and red abalone larval development tests of stormwater samples collected on January 31, 1996. Data are mean and standard deviation, normalized to the control response.



toxicity and variations in water quality parameters, including ammonia (data not shown).

One sample from Ballona Creek was selected from each of the four storm events for TIE analysis. A range of flow conditions (400 to 4,200 cfs) was represented by these samples. In addition, one sample from Malibu Creek (collected on February 1) was also prepared for the TIE. Each of the Ballona Creek samples showed a remarkably similar pattern of response to the TIE treatments (Figure 5). Treatment with EDTA (3 mg/L final concentration) completely removed the toxicity in each case. Filtration and sodium thiosulfate addition were not effective on any of the samples. Solid phase extraction (SPE) partially removed toxicity on only one sample from Ballona Creek. Baseline toxicity (prior to filtration) was usually similar to initial toxicity values measured 6 to 10 d earlier, indicating that the toxicants were relatively stable in solution.

A different response pattern was obtained for the Malibu Creek sample TIE. The EDTA completely removed the toxicity (similar to the Ballona Creek samples), but partial toxicity reductions were also

produced by filtration and sodium thiosulfate (Figure 5).

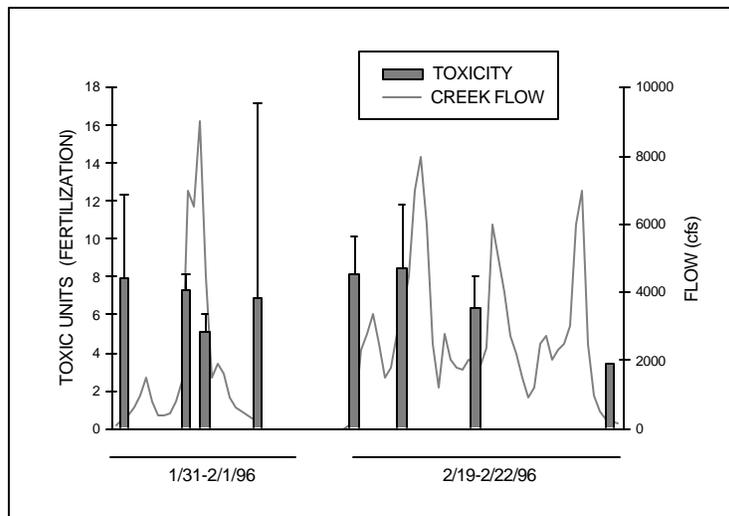
Chemistry-Toxicity Relationships

Since the TIE results suggested that metals were a likely source of toxicity, dissolved metal concentrations were examined for cadmium, copper, manganese, lead, zinc, and nickel. These trace elements were most frequently detected in the samples and/or are associated with toxicity to aquatic life. Cadmium, lead, and nickel were detected sporadically (30-60% of samples) at Ballona Creek and were present in similar concentrations at both sites (Table 2). Dissolved copper, manganese, and zinc were detected in all Ballona Creek samples. Manganese and zinc were frequently detected in Malibu Creek samples also, but usually at reduced concentrations. Copper was not detected in any sample from Malibu Creek.

Correlation coefficients were calculated among the nine Ballona Creek samples having calculable EC50s and the corresponding concentration of each metal. Negative correlation coefficients (indicating greater toxicity as concentration increased) were obtained for cadmium, copper, manganese, nickel, and zinc (Table 2). Correlation coefficients were relatively low (-0.268 to -0.435) and not statistically significant ($r_{0.05,1,7}=0.582$).

Comparison of the metals data to California Ocean Plan chronic toxicity values indicates that copper and zinc were occasionally present at concentrations above the estimated toxicity threshold (Table 2). A more specific assessment of the potential for metal toxicity was made by comparing stormwater concentration with metal toxicity estimates (EC50) specific for the sea urchin fertilization test. Fertilization EC50 data were available for

FIGURE 4. Variation in Ballona Creek toxicity and flow for two storm events. Toxicity is expressed as toxic units (100/fertilization EC50).



cadmium, copper, zinc, and manganese only. Of these metals, zinc was the only element present at concentrations higher than the EC50.

DISCUSSION

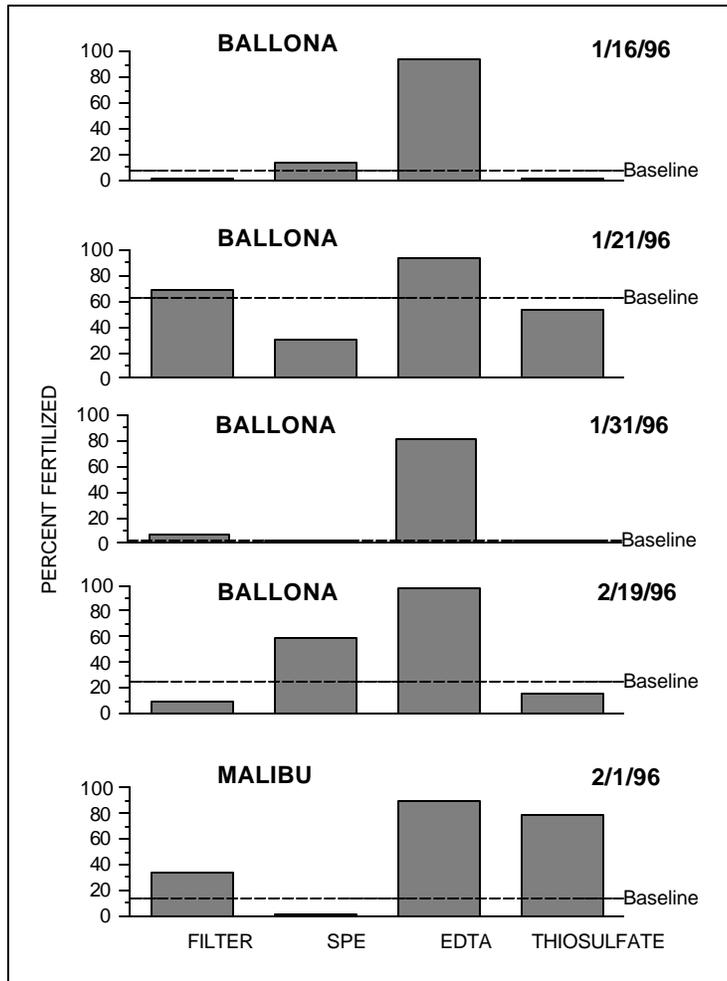
Relative Toxicity

The sea urchin fertilization test results show that Ballona Creek stormwater was usually more toxic than comparable samples from Malibu Creek. The mean sea urchin fertilization NOEC for Ballona Creek samples was 16% stormwater, while the NOEC for Malibu Creek samples was nearly three times higher (31% stormwater). A similar pattern is illustrated by the fertilization EC50 data or abalone development test results.

The presence of greater toxicity at Ballona Creek compared to Malibu Creek is consistent with differences in land use between the two watersheds (Figure 1). The Ballona Creek watershed has a much higher degree of development, and its runoff would be expected to contain more toxic materials. This assumption is supported by the greater chemical contamination of Ballona Creek stormwater, as indicated by dissolved metal measurements (Table 2) and previous data showing elevated metal and organic contaminants (SCCWRP 1992).

The presence of measurable toxicity in some Malibu Creek samples is unexpected, given the low degree of urbanization within its watershed. It is not known whether this toxicity reflects anthropogenic inputs (e.g., agricultural runoff) or natural constituents. Some of the toxicity may have been produced by municipal wastewater effluent discharged from the TWRP, located upstream of the sampling site. The influence of TWRP effluent on the toxicity results cannot be evaluated at this time, as the proportion of effluent in the sample is not known and the

FIGURE 5. Summary of toxicity identification evaluation (TIE) results for samples of 50% stormwater. The baseline value indicates the toxicity of the stored sample before TIE manipulations.



data difficult. Stormwater toxicity, however, is frequently present at most of the locations studied. Accurate comparisons of stormwater toxicity from different watersheds will not be possible until more extensive studies are conducted using consistent test methods. We recommend that the sea urchin (*S. purpuratus*) fertilization test be used for such a testing program.

Toxicant Characterization

The consistent effectiveness of EDTA in the TIEs suggests that cationic metals are an important factor in Ballona Creek stormwater toxicity. The EDTA complexes with many divalent trace metals and renders them nontoxic. Thiosulfate also has a similar effect on some divalent metals.

A difference in the effectiveness of EDTA and thiosulfate was observed, which may provide a more specific indication of the toxicants in Ballona Creek stormwater.

toxicity of the effluent has not been measured using the sea urchin fertilization test.

Limited southern California stormwater toxicity data are available for comparison. A survey of stormwater toxicity at multiple locations was conducted by SCCWRP in 1986 and 1988 (SCCWRP 1989 and 1990). This study utilized the Microtox™ bacterial luminescence test to measure toxicity and identified Ballona Creek as one of the locations with the greatest frequencies of toxic stormwater samples (Table 3). No previous tests of Malibu Creek toxicity have been conducted.

More recent tests have reported a high frequency of stormwater toxicity for several creeks and rivers in San Diego County (Table 3). These data were obtained using different test methods (primarily freshwater fish and an invertebrate), which makes comparisons with the Ballona and Malibu Creek

TABLE 2. Dissolved metals concentration in stormwater samples. Pearson correlation coefficients with corresponding Ballona Creek sea urchin fertilization EC50 data are also shown, along with comparative toxicity data for selected metals.

Sample Date	Concentration (mg/L)					
	Cadmium	Copper	Zinc	Manganese	Lead	Nickel
Malibu Creek						
1/16/96 Base	NT ^a	NT	NT	NT	NT	NT
1/21/96 Base	ND ^b	ND	0.050	0.019	ND	ND
1/31/96 Base	ND	ND	0.058	0.009	ND	ND
1/31/96 Initial	ND	ND	0.079	ND	0.004	ND
2/01/96 Peak	ND	ND	0.015	0.012	ND	ND
2/01/96 End	ND	ND	0.042	0.010	ND	ND
2/19/96 Base	ND	ND	0.058	0.009	ND	ND
2/20/96 Initial	0.002	ND	0.084	0.019	0.014	ND
2/20/96 Peak	ND	ND	0.015	0.012	ND	ND
2/21/96 Peak	0.002	ND	0.006	0.016	0.013	0.002
2/22/96 End	0.001	ND	0.010	0.021	0.006	0.002
Ballona Creek						
1/16/96 Peak	ND	0.021	0.171	0.084	ND	0.005
1/21/96 Peak	0.001	0.017	0.183	0.068	0.016	0.003
1/31/96 Initial	ND	0.003	0.045	0.015	ND	ND
1/31/96 middle	0.001	0.006	0.036	0.012	0.003	0.002
1/31/96 Peak	ND	0.006	0.047	0.006	ND	ND
1/31/96 End	ND	0.022	0.077	0.013	ND	ND
2/19/96 Initial	0.001	0.028	0.107	0.114	0.001	0.003
2/19/96 Peak 1	ND	0.005	0.044	0.001	0.001	0.001
2/20/96 Peak 2	ND	0.009	0.076	0.005	ND	0.001
2/21/96 End	ND	0.005	0.045	0.015	ND	ND
Correlation with						
Ballona Creek toxicity	-0.268	-0.372	-0.379	-0.330	0.299	-0.435
EC50 (mg/L) ^c	12.4	0.031	0.022	>40		
Chronic toxicity value (mg/L) ^d	0.008	0.005	0.051		0.022	0.048

^a NT = Sample not tested for this parameter.
^b ND = Nondetectable.
^c SCCWRP sea urchin fertilization test data.
^d California Ocean Plan (SWRCB 1990).

The EDTA and thiosulfate have different affinities for complexing cationic metals. In experiments using freshwater tests, both EDTA and thiosulfate were highly effective in removing toxicity from copper, cadmium, and mercury (Hockett and Mount 1996). The pattern observed for Ballona Creek samples was different; EDTA was highly effective and thiosulfate was not effective in removing the toxicity. Hockett and Mount (1996) identified three metals (zinc, lead, and nickel) that showed a similar pattern of toxicity removal (EDTA > thiosulfate). The limited chemistry and toxicity information available for these three metals identifies zinc as the metal most likely to be present at toxic concentrations.

The weak correlations between metal concentration and toxicity (Table 2) could have resulted from many factors and do not contradict a conclusion that trace metals are important toxicants in Ballona Creek. The low correlation coefficients obtained for Ballona Creek stormwater may

reflect the use of a small sample size (n=9); differences in metal concentration between samples used for chemistry and those measured for toxicity; or the effects of complex interactions among multiple chemicals. Additional research is needed before the cause of Ballona Creek stormwater toxicity can be described in greater detail.

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TABLE 3. Summary of southern California stormwater toxicity data from various sources and years.

Location	Bacteria ^a		Invertebrates			Fish		
	# Samples	% Toxic	# Samples	% Toxic	Test ^b	# Samples	% Toxic	Test
Ventura County								
Santa Clara River	6	33						
Calleguas Creek	2	0						
Los Angeles Co.								
Ballona Creek	9	89	16	100	SF			
Ballona Creek			6	83	AD			
Malibu Creek			6	67	SF			
Malibu Creek			3	0	AD			
L.A. River	46	76						
Dominguez Channel	2	0						
San Gabriel River	9	89						
Orange County								
Santa Ana River	16	25						
San Diego County								
San Diego River	30	17				2	100	JMD
Chollas Creek	29	55	4	100	CS	4	100	FMG
Chollas Creek						2	100	JMD
Rose Creek	26	23				3	100	JMD
Tecolote Creek			4	100	CS	4	100	FMG
Switzer Creek						1	100	JMD
Encinas Creek						4	75	JMD
Los Penasquitos Ck.						1	100	JMD
Sweetwater River						3	67	JMD
Tijuana River	28	100						

^aMicrotox™ luminescence (SCCWRP 1990).
^bSF = Sea urchin fertilization (this study), AD = Abalone larval development (this study), CS = Daphnid (*Ceriodaphnia*) survival (Kinnetic Laboratories Inc. 1995), FMG = Fathead minnow growth/survival (Kinnetic Laboratories Inc. 1995), and JMD = Japanese medaka embryo development (Skinner *et al.* 1994).

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