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STORMWATER INPUT OF PYRETHROID INSECTICIDES TO AN URBAN RIVER

DONALD P. WESTON*† and MICHAEL J. LYDY‡
†Department of Integrative Biology, University of California, Berkeley, California, USA
‡Fisheries and Illinois Aquaculture Center and Department of Zoology, Southern Illinois University, Carbondale, Illinois, USA

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Abstract—The American River flows for nearly 50 km through highly urbanized lands surrounding Sacramento, California, USA. Twenty-three streams, drainage canals, or pumping stations discharge urban runoff to the river, with the cumulative effect of nearly doubling the river's flow during rain events. During winter storms, the water column in the most downstream 13-km reach of the river exhibited toxicity to the standard testing species, *Hyalella azteca*, in 52% of samples, likely because of the pyrethroid insecticide bifenthrin. The compound is heavily used by professional pest controllers, either as a liquid perimeter treatment around homes or as granules broadcast over landscaped areas. It was found in 11 of 12 runoff sources examined, at concentrations averaging five times the *H. azteca* 96-h EC50. Quantified inputs of bifenthrin should have been sufficient to attain peak concentrations in the river twice those actually observed, suggesting loss by sedimentation of particulates and pesticide adsorption to the substrate and/or vegetation. Nevertheless, observed bifenthrin concentrations in the river were sufficient to cause water column toxicity, demonstrated during six storms studied over three successive winters. Toxicity and bifenthrin concentrations were greatest when river flow was low ($<23\ m^3/s$) but persisted even at atypically high flows ($585\ m^3/s$). Environ. Toxicol. Chem. © 2012 SETAC

Keywords—Pyrethroids Bifenthrin *Hyalella azteca* Urban runoff

INTRODUCTION

Pyrethroid insecticides are widely used in urban environments and have frequently been found in sediments of urban waterways in California [1,2], Texas [3], Illinois [4], and Washington [5] (USA), often exceeding concentrations acutely toxic to standard sediment toxicity testing species. Past studies generally have not focused on the water column, given the extreme hydrophobicity of pyrethroids (log K_{OC} typically 5–6 [6]). However, pyrethroids have recently been found to be responsible for water column toxicity to the amphipod *Hyalella azteca* in several California creeks and rivers [7].

Because of the hydrophobicity of pyrethroids, water column concentrations are very low, yet the compounds are extraordinarily toxic in the dissolved phase. Acute toxicity at less than 5 ng/L has been reported for the amphipod *H. azteca* [8], the grass shrimp *Palaemonetes pugio* [9], and the phantom midge *Chaoborus obscuripes* [10]. Monitoring to detect concentrations at these low levels is challenging, given that detection limits for dissolved pyrethroids have been variously reported as 1 to 7 ng/L [11], 3.1 to 6.3 ng/L [12], and 2.1 to 11.7 ng/L [13]. Given that detection limits are essentially at acute LC50s for sensitive species, it is likely that chronic effects occur at concentrations below those currently measurable.

Among the water bodies in which water column toxicity from pyrethroids has been reported is the American River, located in northern California [7]. Although the results of our previous study [7] were intriguing and suggested that the traditional focus solely on the sediments might be misdirected, the work left several critical questions unanswered. No data were available on the specific inputs contributing pyrethroids to the river; thus it could not be established whether they were due to widespread urban sources that would be of broader regional or national concern or were due to one or a few sources unique to the American River. The river flow was also at unusually low levels, so it was unclear whether toxicity was a common phenomenon potentially applicable to other urban areas or was limited to drought conditions prevailing at the time.

The present study expanded upon the earlier observations to resolve questions of their general applicability. In particular, we assessed pyrethroid concentrations and toxicity over variable flows, at times exceeding flows during the previous work by nearly 30-fold. The study also included measurement of pyrethroids in various inputs to the river (e.g., tributaries, drains, pump stations), to identify the principal sources, establish which pyrethroids were of greatest concern in urban runoff, and quantify pyrethroid inputs from urban environments to aquatic systems.

MATERIALS AND METHODS

Description of study area

The American River begins on the western slopes of the Sierra Nevada Mountains in northern California. Three forks of the river converge in a reservoir formed by the Folsom Dam. Water levels in the reservoir are manipulated for flood control and other purposes, so the amount of water released from the dam is carefully managed. Water that is released flows for 50 km to the river's confluence with the Sacramento River.

Upstream of the reservoir, most of the watershed is rural or forested. Downstream of the dam, the watershed is heavily urbanized as it passes through a succession of cities comprising the Sacramento metropolitan area. The river receives stormwater runoff from these urban areas, although sanitary sewage is diverted to the Sacramento River and does not affect American River water quality. Agricultural inputs downstream of the dam are negligible.

All Supplemental Data may be found in the online version of this article.
* To whom correspondence may be addressed (dweston@berkeley.edu).
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The extensive urbanization of the lower watershed has earned the American River the title of "California's largest urban stream" [14], although the term "stream" belies the fact that flow is considerable, often 30 to 120 m^3/s. The river provides habitat to several protected salmonids, most notably fall-run chinook salmon (*Oncorhynchus tshawytscha*) and steelhead (*Oncorhynchus mykiss*), both of which spawn in the reach below the dam.

Sample collection

Water samples were collected from four river locations, ranging from station 1, just below Folsom Dam, to station 4, just prior to the confluence with the Sacramento River (Fig. 1). Samples were collected within a few meters of the river's banks. Glass bottles certified clean for pesticide sampling were immersed in the river and filled approximately 10 cm below the surface.

Stormwater runoff enters the American River through seven natural creeks or sloughs, several earthen or concrete-lined drainage canals, and 14 pump stations, which raise runoff over the levees that flank the river channel as it passes through Sacramento. The creeks and the two main drainage canals were sampled in a manner comparable to the river. Among the 14 pump stations that discharge to the American River, three of the four with the largest watersheds were sampled. The pump stations were sampled by lowering a stainless-steel bailer into the concrete sumps from which the pumps draw.

Sampling was structured around three storm events. The first was 2 d in length (October 13–14, 2009) and produced 6.4 cm of precipitation (Chicago weather station, east of Sacramento; http://www.cdec.water.ca.gov). The second storm was 6 d in length (January 17–22, 2010) and produced 9.2 cm. The third yielded 6.9 cm over 3 d (December 17–19, 2010). One or two days before each storm event, water samples were collected from the river at stations 2 and 4. During the storms, all four river sites were sampled two to four times, depending on storm duration. The various creeks, drains, and pump stations were sampled twice during each storm event. To increase temporal coverage, some data presented incorporate previous observations from rains during February to May 2009 [7].

The present study provided an opportunity to study the American River over an extremely broad range of flows. Dam releases are commonly in the range of 30 to 120 m^3/s, but, in early 2009, northern California was in the midst of a lengthy drought and releases declined to $<23\,m^3$/s for several weeks, an event that had not occurred since 2001. Conversely, in December 2010, releases exceeded 800 m^3/s as dam operators sought to create reservoir capacity for flood management in anticipation of heavy rains. Flows of this magnitude had not occurred since April 2006. Samples were collected over a wide range of flows, from 21 m^3/s (March 3, 2009) to 585 m^3/s (December 19, 2010).

Flow rates were established by a variety of methods, depending on the site. Flows released from the Folsom Dam are available on the Internet (http://cdec.water.ca.gov/cgi-progs/queryDaily?FOL). Flows in the creeks and open drainage canals were determined by using a Swoffer 2100-C140 current meter (Swoffer Instruments), taking multiple measurements across the cross-sectional area of flow [15]. Flows from the pump stations were determined by the pump run times on the days of interest, as provided by the operators, multiplied by the nominal capacity of each pump. Flow data were obtained from five larger pump stations, and the smaller ones were estimated. The five pump stations with data serve 60% of the 29 km^2 of the watershed served by all 14 Sacramento pump stations. The flow per square kilometer of land area for each of the five pump stations was determined for any given day, and an average was calculated

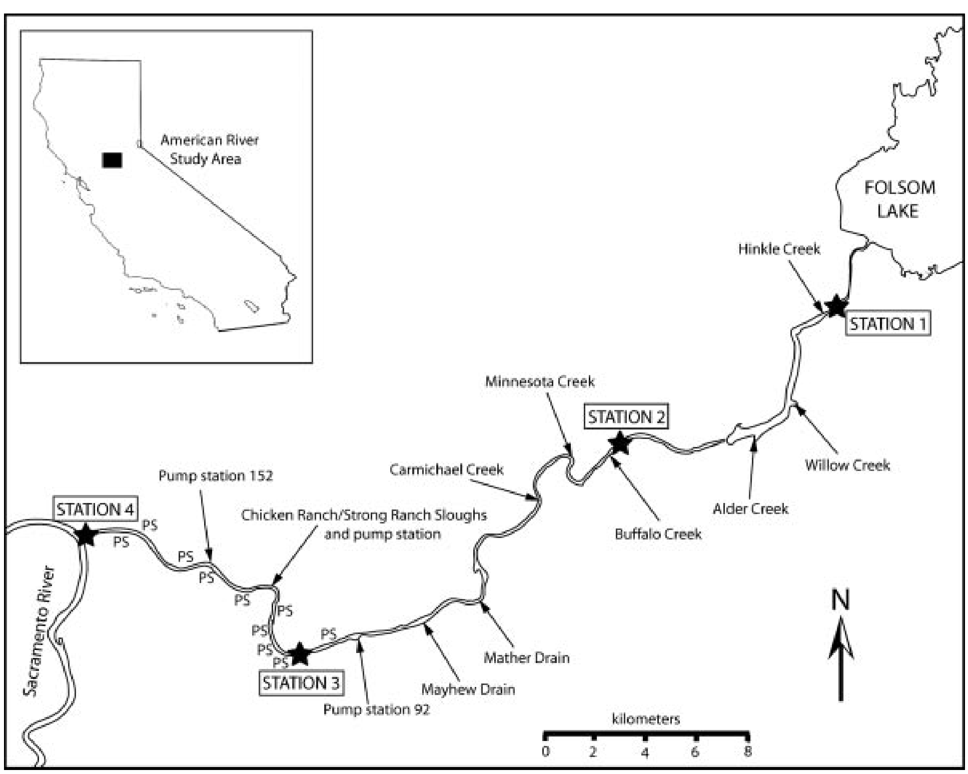

Fig. 1. Map of the lower American River, downstream of Folsom Dam. Stations 1 to 4 in the main river channel are shown as well as the various streams, drainage channels, and pump stations (PS) sampled. The locations of unsampled PS are indicated.

(relative standard deviation was typically ~30%). That average value was then applied to the remaining 40% of the land area served by the remaining nine pump stations to estimate their flow during the same day.

Analytical chemistry

Analytical methods have been previously described in Wang et al. [16]. Water was liquid:liquid extracted following U.S. Environmental Protection Agency (U.S. EPA) method 3510C, using three 60-ml additions of dichloromethane. The extract was reduced to 1 ml, exchanged to hexane, and cleaned by a dual-layer graphitized black carbon and primary/secondary amine column (Supelclean ENVI-Carb II/Supelclean primary/secondary amine column, 3.0 mg/600 mg, 6.0 ml; ResPrep). Extracts were analyzed on an Agilent 6890 gas chromatograph with a micro-electron capture detector (Agilent Technologies), using two columns, an HP-5ms and a DB-608. Qualitative identity was established using a retention window of 0.5%, with confirmation on a second column. Calibration used the external standard method. Analytes included eight pyrethroids (bifenthrin, cyfluthrin, cypermethrin, deltamethrin, esfenvalerate, fenpropathrin, lambda-cyhalothrin, permethrin). Quality assurance procedures included surrogate spikes with every sample, blanks, laboratory control spikes, matrix spikes, matrix spike duplicates, and field duplicates. Detection limits using these methods have previously been shown to vary from 0.75 to 1.65 ng/L depending on the matrix and the specific pyrethroid [16]. Because pyrethroid data were used to help interpret toxicity findings, and as little as 1 ng/L is near the threshold of acute toxicity to the test organisms [8], data are reported to 1 ng/L for all analytes when the analyst believed the quantification to be reliable.

The organophosphate pesticide chlorpyrifos was also included among the analytes, although the data are not shown. Concentrations were <10 ng/L in all river samples and in nearly all (85%) of the runoff samples, well below the reported EC50 for *H. azteca* of 96 ng/L [17]. At the highest observed concentrations (39 ng/L in Chicken/Strong Ranch Sloughs, 27 ng/L in Mather Drain), chlorpyrifos might have had some contribution to toxicity secondary to the pyrethroids.

Toxicity testing

Testing generally followed U.S. EPA protocols for freshwater acute tests [18], with the substitution of *H. azteca*, a species used for water testing in the past [7,17,19,20]. Water samples were tested within 48 h of collection and were distributed to five replicate 80-ml beakers, each with a 1-cm^2 nylon screen to which the *H. azteca* often cling. Amphipods were obtained from cultures maintained at the University of California, Berkeley, and 10 organisms, 7 to 14 d in age, were added to each beaker. The beakers were held at 23°C, under a 16:8 h light:dark photocycle and without aeration. After 48 h, 1 ml yeast/cerophyll/trout food was added. After a 6-h feeding period, approximately 80% of the water was removed and replaced with fresh sample. Replacement water had been maintained at 4°C since collection, and its temperature was raised to 23°C just prior to water exchange. Conductivity, alkalinity, hardness, and pH were measured at the beginning and end of the test; temperature and dissolved oxygen were measured throughout the test. After 96 h, the test was terminated. Pyrethroids are neurotoxins, and we often observe paralysis, with these organisms usually lying on the bottom of the exposure container unable to move except for occasional twitching. Tests were scored by recording the number of organisms able to swim normally, the remainder being dead or paralyzed. All tests included a concurrent control using deionized water made moderately hard by addition of salts [18].

Several pyrethroid-focused toxicity identification evaluation (TIE) procedures [17] were used to help identify the cause of toxicity. Piperonyl butoxide (PBO) is a synergist expected to cause greater toxicity if a pyrethroid is present. It was added to test and control waters at 50 μg/L in a methanol carrier, with methanol concentration kept below 12.5 μl/L. The PBO was replaced at the 48-h water change.

Enzymes engineered to break down specific pesticides can be a useful TIE tool [8] and would be expected to decrease toxicity if their target substrate were responsible. The enzymes are not yet commercially available but were obtained through a research collaboration with Orica Limited. An enzyme, E3-013, is capable of catalyzing the hydrolysis of several pyrethroids, but has been optimized for the degradation of the pyrethroid bifenthrin. It was available as a freeze-dried powder of lysed bacterial cells, of which the enzyme is a small fraction of the mass. The powder was added to the water at a concentration of 5 mg/L, and 4 h was provided for pesticide degradation before exposing *H. azteca* to the water. Fresh enzyme was added with the water replacement at 48 h. A concurrent TIE trial was performed with an OpdA enzyme designed to break down organophosphate insecticides. These compounds were not a concern in the American River, given that diazinon and chlorpyrifos have not been sold for urban uses for at least six years and, in those samples in which OpdA was used, chlorpyrifos never exceeded 3% of its reported *H. azteca* EC50 [17]. Therefore, OpdA was used as a dissolved organic matter control for the E3-013 enzyme. This control treatment ensures that any reduction in toxicity is due to the catalytic action of the enzyme and not a reduction in bioavailability resulting simply from addition of the dissolved organic matter introduced with the freeze-dried enzyme preparation. Bovine serum albumin has previously been used [17,21], but the OpdA freeze-dried preparation is preferable, because the majority of the mass, excluding the specific enzyme, is identical in composition to the E3-013 product. Treatment controls for PBO, E3-013, and OpdA were included whenever performing the TIE manipulations, and none showed toxicity.

Statistical analyses were carried out in CETIS (Tidepool Scientific Software). Comparisons between the field samples and the controls were made by using *t* tests, with the additional criterion that a sample is denoted as "toxic" only if the effect is greater than or equal to 20% relative to the control [22]. Comparisons between TIE treatments and the concurrent controls were done by *t* test for samples tested with only one TIE treatment or by Dunnett's multiple comparisons for samples tested with multiple treatments.

RESULTS AND DISCUSSION

Characterization of runoff

Urban runoff to the American River was characterized in seven creeks, two stormwater drainage canals, and three of the larger pump station sumps along the river. In addition, another 11 pump stations were not sampled for pyrethroids. The combination of all these inputs can represent a substantial volume of the river's flow by the time it reaches the confluence with the Sacramento River. For example, during the October 2009 rain event, 58.5 m^3/s was released from Folsom Dam. The combined inputs of the 12 sampled and 11 unsampled inputs totaled 39.5 m^3/s, resulting in the river's flow at the Sacramento River confluence consisting of 40% urban runoff. Comparable results were seen in the January 2010 rain event, when the river was 44% runoff. At the other extreme, during the December 2010 rain event, releases from the dam were the largest in nearly five years (585 m^3/s), and runoff contributions were only 2.5% of total flow.

The various inputs were sampled for pyrethroids during three rain events, with a sample from any given input usually collected during 2 d of each event (Supplemental Data, Table S1). There was no consistent difference in concentrations between the first and second samples during a single event. The pyrethroid bifenthrin was detected in 88% of the samples (Table 1). Concentrations were commonly 10 to 30 ng/L and reached a maximum of 106 ng/L in a Carmichael Creek sample. Other detected pyrethroids were permethrin (31% detection, 111 ng/L maximum), cyfluthrin (25% detection, 20.5 ng/L maximum), and cypermethrin (15% detection, 9.4 ng/L maximum). Lambda-cyhalothrin was detected in two samples at <5 ng/L, and deltamethrin, esfenvalerate, and fenpropathrin were not detected.

The various inputs could be grouped into four categories based on their relative loadings of bifenthrin to the American River, derived from the median flow and bifenthrin concentrations in Table 1. First, Alder and Willow Creeks contained low or unmeasurable concentrations of bifenthrin and were not significant sources. Second, although samples for the present study were collected at some of the larger pump stations, the bifenthrin inputs from the 11 small stations were estimated based on a median bifenthrin concentration of 15.5 ng/L (range, <1–34 ng/L) derived from sampled pump stations. Estimated bifenthrin inputs from these 11 unsampled sources range from 0.2 to 12 μg/s each, or 56 μg/s in aggregate. Third, most of the other inputs listed in Table 1 are moderate sources, releasing 10 to 50 μg/s bifenthrin during rain events. Carmichael Creek is at the high end of this range, owing to higher than typical bifenthrin concentrations. The fourth group, and largest of the sources, is Chicken Ranch and Strong Ranch Sloughs. These creeks have a larger urban watershed than the other inputs studied. The flows from the two creeks combine just prior to reaching the American River and then continue to the river either in the stream channel or, at high flows, through a diversion to a pump station. Their combined flow is the largest contributor of bifenthrin to the river, owing to both high bifenthrin concentrations (median 35.8 ng/L) and a flow 5 to 20 times that of other inputs. Chicken Ranch and Strong Ranch Sloughs contributed approximately 350 μg/s bifenthrin to the river, averaged over all storm events.

Total suspended solids (TSS) concentrations in the runoff from creeks and pump stations were below 100 mg/L in 79% of the samples but reached a maximum of 662 mg/L. However, TSS concentration was not the primary determinant of pyrethroid concentration. Among the five waterways for which Table 1 shows five to six samples taken, only one waterway showed a significant correlation between bifenthrin and TSS concentrations (Buffalo Creek; $r = 0.89$; $p < 0.05$). Some of the pyrethroids carried in the runoff are likely to be particle-associated given pyrethroid hydrophobicity, but, at least based on the limited data available, it appears that factors such as timing of pesticide application or rainfall intensity were more important determinants of pyrethroids in runoff. In addition, only about one-third of the bifenthrin in Sacramento urban runoff is likely to be particle associated if equilibrium partitioning is assumed, given

$$K_{OC} = \frac{C_{sed}}{C_{wat} \times OC}$$

and

$$(C_{sed} \times TSS) + C_{wat} = C_{tot}$$

where the K_{OC} of bifenthrin is 237,000 [6], C_{sed} is the concentration of bifenthrin on particulates, C_{wat} is the dissolved concentration of bifenthrin, OC is the organic carbon content of the particulate matter (5% based on our two direct measurements of particulate matter from Sacramento runoff; 5.1 and 5.8%; unpublished data), TSS is the median suspended sediment concentration in runoff from the present study (50 mg/L), and C_{tot} is the median bifenthrin in whole-water runoff samples from the present study (18 ng/L). Based on these calculations, only approximately 8 ng of the 21 ng/L bifenthrin in a typical Sacramento urban runoff sample is likely to be particle-associated, further explaining the lack of correlation between TSS and pyrethroid content of runoff. However, these estimates would imply a typical bifenthrin concentration on suspended sediment of 160 ng/g, a value lower than that reported for winter urban runoff from Sacramento suburbs (473–1,211 ng/g [23]). Particle-bound pyrethroids could be more significant than these estimates indicate if the equilibrium

Table 1. Suspended sediment, flow, and pyrethroid concentrations in creeks, drainage channels, and pump stations discharging to the American River (median and range)[a]

Source	No. of samples	Total suspended solids (mg/L)	Flow (m^3/s)	Bifenthrin (ng/L)	Cyfluthrin (ng/L)	Cypermethrin (ng/L)	Permethrin (ng/L)
Hinkle Creek	4	41.7 (2.2–141)	0.36 (0.11–0.63)	26.8 (1.0–43.7)	6.4 (U–13.0)	U (U–U)	U (U–11.3)
Willow Creek	2	60.2 (21.3–99.0)	0.59 (0.49–0.68)	4.7 (U–9.3)	U (U–U)	U (U–U)	U (U–U)
Alder Creek	2	37.4 (28.3–46.4)	2.8 (1.8–3.7)	U (U–U)	U (U–U)	U (U–U)	U (U–U)
Buffalo Creek	6	29.7 (14.4–104)	1.5 (0.89–2.2)	9.0 (U–28.9)	U (U–U)	U (U–U)	U (U–U)
Minnesota Creek	5	158 (15.5–258)	0.45 (0.09–1.2)	20.7 (1.3–34.0)	U (U–U)	U (U–U)	U (U–U)
Carmichael Creek	5	193 (22.0–662)	1.4 (0.17–2.3)	37.3 (6.2–106.4)	8.7 (2.0–20.5)	U (U–9.4)	8.1 (U–21.1)
Mather Drain	6	38.4 (12.3–89.1)	2.6 (0.18–8.0)	13.3 (U–31.7)	U (U–26.6)	U (U–U)	U (U–U)
Mayhew Drain	4	38.1 (9.3–54.7)	1.3 (0.50–2.0)	19.0 (10.1–34.6)	U (U–U)	U (U–U)	U (U–U)
Sump 92	4	38.6 (21.1–55.2)	0.67 (0.35–1.7)	21.1 (12.4–34.0)	5.0 (U–11.4)	U (U–3.9)	13.3 (8.4–29.8)
Chicken/Strong[b]	6	74.0 (51.3–286)	9.9 (4.7–10.8)	35.8 (17.4–83.4)	U (U–3.2)	U (U–4.3)	13.6 (U–111)
Sump 152	4	32.3 (14.8–55.7)	1.8 (0.95–3.5)	11.9 (4.5–21.0)	U (U–14.6)	U (U–U)	3.5 (0–15.2)

[a] When six samples were taken, they represent 2 d in each of three rain events (October 13–14, 2010; January 18–20, 2010; December 18–19, 2010), though not all sites could be sampled on every occasion.
[b] Chicken Ranch Slough and Strong Ranch Slough converge just prior to discharge to the American River, and their combined flow can enter the river through both the stream channel and a pump station. The given flow combines these two inputs.
U = undetected (<1 ng/L).

partitioning assumptions on which they are based are not applicable to urban runoff.

Water quality in the American River

Water clarity was extremely high in the American River, even during runoff events. Median TSS during storm sampling was 5.5 mg/L and ranged from 0.7 to 47.2 mg/L. Dissolved organic carbon was not measured in the present study but has previously been found to be approximately 2 mg/L during rain events [7].

Ten samples were collected from the river (primarily from stations 2 and 4) during winter periods without rain (six samples from present study, four samples from Weston and Lydy [7]). None contained detectable pyrethroids. In total, 30 river samples were collected for pesticide analyses during rain events in the present study, with an additional seven samples available from Weston and Lydy [7] (Supplemental Data, Table S2). Station 1, located below Folsom Dam, was analyzed six times, and never showed measurable pyrethroids. Station 2 contained no pyrethroids on eight of nine occasions, although one sample contained 1.2 ng/L bifenthrin.

The two downstream sites frequently contained pyrethroids at times of rain, with bifenthrin commonly found and permethrin present in two samples. Station 3 contained bifenthrin 50% of the time (among 10 samples) with a range of detected concentrations of 1.5 to 3.8 ng/L. Station 4 contained bifenthrin 45% of the time (among 11 samples), with a range of detected concentrations of 1.1 to 5.6 ng/L. The relationship between the concentration of bifenthrin and the amount of water released from Folsom Dam was not significant ($r^2 = 0.029$; $p > 0.05$), although the higher concentrations tended to occur at times of lower flow. Bifenthrin remained measurable at station 4, even when the river was at flood stage with flows $>500\,m^3$/sec, although concentrations were just barely above the detection limit (1.1–1.6 ng/L).

It was possible to compare the observed concentrations of bifenthrin with those expected based on known inputs to the river (Fig. 2). To generate this figure, the median concentration from each sampled input (Table 1) was applied to its median

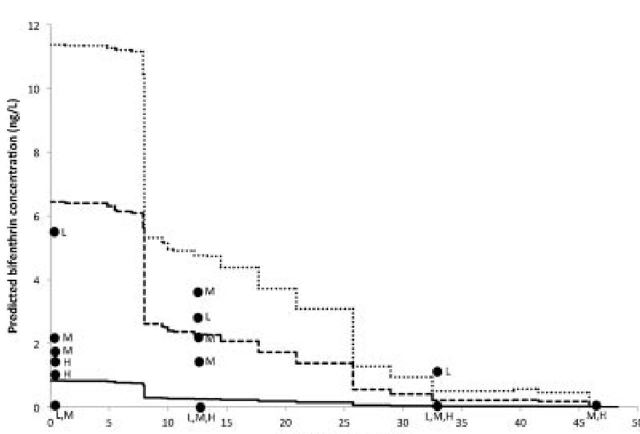

Fig. 2. Lines indicate predicted bifenthrin concentrations in the American River. Concentrations increase as the river passes each known discharge point of urban runoff in accordance with median flows and bifenthrin concentrations measured or estimated for that specific input. Data are shown for occasions when river flow was unusually low (dotted line; $21\,m^3$/s on March 3, 2009), moderate (dashed line; $55\,m^3$/s on October 13, 2009), or unusually high (solid line; $585\,m^3$/s on December 19, 2010). Points indicate measured concentrations in the river at stations 1 to 4 and the flow regime prevailing at the time of collection. Undetected concentrations (<1 ng/L) are plotted as zero. L = low; M = moderate; H = high.

flow (usually a median of five measurements over three rain events) to estimate the amount of bifenthrin released to the American River from that input during an average rain event. For unsampled pump stations, flow was estimated (see *Materials and Methods* section), and a median bifenthrin concentration from sampled pump stations of 15.5 ng/L was applied. This value represents a median of 14 Sacramento pump station samples, eight from the present study and six wet-season pump-station samples from Weston and Lydy [7]. Using the calculated bifenthrin loading from each input, predicted bifenthrin concentrations in the river were derived for occasions when river flow was unusually low ($21\,m^3$/s; March 3, 2009), moderate and typical for winter months ($55\,m^3$/s; October 13, 2009), and unusually high ($585\,m^3$/s; December 19, 2010). Bifenthrin concentrations in the river as it left the dam were assumed to be zero, as supported by the lack of detections at station 1.

Predicted bifenthrin concentrations in the river, based on known dam releases and median storm-driven inputs from all known major inputs, were expected to reach 6.4 ng/L during moderate flows and 11.4 ng/L at minimum flows, assuming no loss of bifenthrin once it enters the river (Fig. 2). The largest increases were expected as the river receives input from Chicken Ranch and Strong Ranch Sloughs (river km 8 in Fig. 2), and, secondarily, Carmichael Creek (river km 26). During December 2010, when dam releases were at their highest point in nearly five years, dilution of stormwater inputs should have been sufficient to permit bifenthrin concentrations in the river to reach only 0.8 ng/L, a concentration on the threshold of toxicity but slightly below the threshold of detection.

In general, observed concentrations of bifenthrin in the river tend to be lower than the values predicted based on river flow and known bifenthrin inputs, with the discrepancy greatest at low flows. This difference suggests some mechanism of bifenthrin loss from the water column, once the pesticide enters the river. Deposition of particle-associated bifenthrin is one possible loss, a hypothesis supported by the fact that median TSS in the river is about one-tenth that of median TSS in the inputs, when dilution of the inputs with river water would account for only about a factor of two difference. Adsorption of dissolved bifenthrin onto the substrate and plants along the riverbanks may also represent significant losses [24]. Both routes of loss would be expected to be greatest at times of lowest flow.

Water column toxicity

Bifenthrin is toxic to sensitive aquatic species at low parts per trillion concentrations [25]. The 96-h EC50 for paralysis of *H. azteca* is 3.3 ng/L, and the 96-h LC50 for mortality is 7.7 ng/L [8]. The study incorporated toxicity testing with *H. azteca* because bifenthrin concentrations in the river were near these benchmarks, and they were routinely exceeded in urban runoff.

Toxicity of the creeks and sloughs prior to their discharge to the river was tested only during the October 13, 2009, rain event (Supplemental Data, Table S1). Water samples from five of the seven waterways tested caused toxicity to *H. azteca* (Carmichael = 96% dead/paralyzed, Hinkle = 96%, Chicken/Strong = 92%, Minnesota = 92%, Buffalo = 74%; Alder and Willow nontoxic; control = 2% dead/paralyzed).

River samples were tested throughout the study, and control performance (0–10% dead/paralyzed) met test acceptability criteria. At the most upstream site, station 1, all seven samples collected during rain events showed no toxicity. At station 2, all but one of 11 samples were nontoxic.

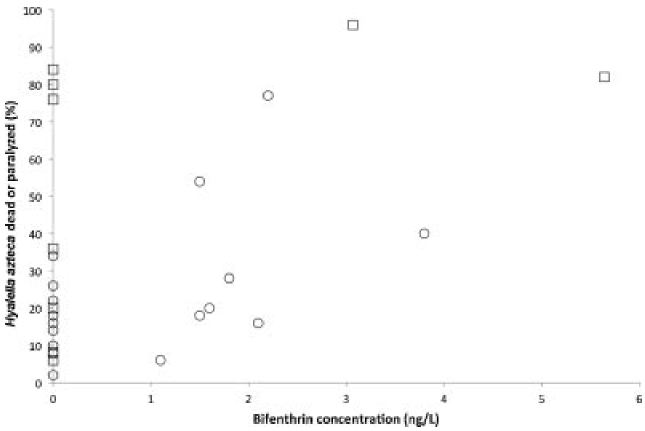

Fig. 3. Relationship between bifenthrin concentration in American River water at stations 3 and 4 and the proportion of affected *H. azteca* observed in toxicity tests. Circles are data from the present study; squares show additional data from Weston and Lydy [7]. Undetected concentrations (<1 ng/L) are plotted as zero.

Most of the toxicity was seen at stations 3 and 4, in the most heavily urbanized reaches of the river (Supplemental Data, Table S2). One of five prestorm samples showed toxicity (station 4; March 18, 2009; 36% dead/paralyzed). In the 23 samples collected at stations 3 and 4 during storm events, 35% caused mortality to *H. azteca*, and 52% were toxic based on the swimming endpoint. Toxicity was present even during the high-flow event of December 2010, when river flow reached 585 m^3/s (station 4; 20% dead/paralyzed), although toxicity was diminished relative to the previous times of lower river flow. There was a significant relationship between bifenthrin concentration and toxicity to *H. azteca* (Fig. 3; $r^2 = 0.211$; $p < 0.05$). The few samples with high toxicity but undetectable bifenthrin may reflect unmeasured toxicants. However, this might also be a consequence of the fact that the 1 ng/L detection limit for pyrethroids is essentially at the threshold of toxicity, and in fact, for one of these samples (the point at 80% on y axis), addition of piperonyl butoxide (PBO) caused a tripling of toxicity, consistent with pyrethroids as the cause [7]. Although samples with undetected pyrethroids are plotted as zero in Figure 3, actual concentrations could be any value up to 1 ng/L.

Of particular note was the duration of elevated bifenthrin concentrations and toxicity during the January 2010 storm event in which rain fell for 6 d. Bifenthrin was above detection limits in the lower reaches of the river for 3 d (January 18–20), and toxicity was apparent over a 5-d period (January 18–22; 28–77% dead/paralyzed). Although laboratory toxicity tests employed a standardized 4-d exposure, this duration appears environmentally relevant given the persistence of bifenthrin and toxicity in the river during the January storm.

Testing was performed at the standard *H. azteca* test temperature of 23°C [26]. American River winter temperatures can reach approximately 8°C. Had the tests been conducted at in situ temperatures, pyrethroid toxicity would have at least tripled [27], and it is likely that even more samples would have proved toxic [5].

Four samples were further tested with TIE treatments developed for use with potential pyrethroid toxicity (Table 2). Adding PBO substantially increased toxicity in all four cases, consistent with pyrethroids as the cause. Adding the E3-013 enzyme, engineered to hydrolyze bifenthrin, mitigated toxicity in one instance and had no effect in another. An organophosphate-degrading enzyme (OpdA), used as a control for dissolved organic matter influence on toxicity, independent of catalytic activity, failed to mitigate toxicity on either occasion when it was used, although it increased toxicity in one instance for unknown reasons. With the exception of the enzyme treatments of station 4, TIE results are consistent with pyrethroids as the cause of toxicity and are consistent with historical data from the river one year previously, in which PBO increased toxicity and E3-013 reduced it [7]. All but one of the four samples used for the TIE tests contained bifenthrin at concentrations of one-half to two-thirds of the reported *H. azteca* EC50 (3.3 ng/L; [8]), indicating that its role in toxicity is plausible. Its nondetection in the one sample, despite PBO data supporting its presence, is reflective of the challenges of working with a compound having a detection limit (1 ng/L) at the threshold of toxicity. No pyrethroids other than bifenthrin were found in any of the TIE samples.

Management considerations

It is apparent that current pesticide use practices allow bifenthrin applied to urban properties to be carried offsite by stormwater runoff. The concentrations in runoff are, on average, approximately five times those that cause paralysis and/or death to sensitive species, typified by *H. azteca*. The amount of pyrethroid-contaminated runoff from a metropolitan area, such as Sacramento, is sufficient to cause toxicity in a large river system. At least a 13-km reach of the lower American River routinely exhibited toxicity after winter rains, now documented in six storm events over three successive winters. The present study did not examine the effects of the bifenthrin on resident invertebrates, such as chironomids, mayflies, and caddisflies, that provide a food source for the chinook salmon and steelhead that spawn in the American River [28], although at least some members of these broad taxonomic groups are less sensitive to bifenthrin than is *H. azteca* [29,30].

Bifenthrin is commonly used in northern California by professional pest controllers to treat outside areas for pests, particularly ants. A recently completed survey of applicators (http://www.cdpr.ca.gov/docs/registration/reevaluation/

Table 2. Percentage of *Hyalella azteca* dead or paralyzed in the initial test and in various subsequent toxicity identification evaluation (TIE) treatments (mean and standard deviation)

Station and sample date	Bifenthin (ng/L)	% affected in initial test	TIE: unamended	TIE: PBO treatment	TIE: OpdA treatment	TIE: E3-013 treatment
Station 4, Jan. 19, 2010	1.8	28 ± 22	4 ± 5	82 ± 4^b	24 ± 18^b	22 ± 33
Station 4, Jan. 22, 2010	0	34 ± 26	20 ± 14	96 ± 9^b		
Station 3, Jan. 18, 2010	2.2	77 ± 18	52 ± 28	96 ± 9^b	46 ± 17	22 ± 4^b
Station 3, Jan. 19, 2010	1.5	18 ± 8	16 ± 13	98 ± 4^b		

[a] All control treatments, including those with piperonylbutoxide (PBO) and E3-013 or OpdA enzymes, had <8% dead or paralyzed.
[b] Values are significantly different ($p < 0.05$) than the unamended treatment.

chemicals/pyrethroids.htm, [31]) provides insights on the treatments from which the bifenthrin may originate. Pest control applicators reported that most applications are done on a contract basis, either monthly or every other month; 80% of bifenthrin is used in residential applications, with only 20% applied to commercial properties; liquid bifenthrin formulations are used as a barrier treatment around the perimeter of homes, whereas granular formulations are broadcast over lawns or similar large areas of the property; and there is little seasonality in use, with comparable quantities applied in all four seasons. In 2009 and 2010, 22,200 and 26,300 kg, of bifenthrin was used, respectively, for nonagricultural purposes by professional applicators in California, of which approximately 15% was used in Sacramento County, the location of the study area (http://www.cdpr.ca.gov/docs/pur/purmain.htm). Retail sales are not included in these figures, but professional use of bifenthrin in California exceeds retail sales by a factor of approximately four [23].

Bifenthrin's prominence in urban runoff can be explained by several factors. First, its nonagricultural use in Sacramento County is greater than any other pyrethroid, exceeding cypermethrin by a factor of three, permethrin by a factor of four, and cyfluthrin by a factor of 13. Second, bifenthrin has a greater persistence in soils and aquatic sediments than other pyrethroids [6,32,33]. Finally, its dominance in runoff could reflect mobility differences resulting from the manner in which the various products are applied or their formulations (e.g., granular formulations are commonly applied for bifenthrin but are few or lacking for the other pyrethroids).

Given the ubiquity of bifenthrin in urban runoff of the present study, as well as elsewhere in California [1,34], it is clear that the problem is not confined to an isolated geographic area or a very specific urban land use. The volume of runoff probably precludes any treatment prior to discharge in most localities. Chicken Ranch and Strong Ranch Sloughs alone discharge approximately 30 g/d bifenthrin during a storm event, or about half the quantified inputs to the American River, yet this bifenthrin is dispersed in 400 to 800 million liters per day of runoff, originating over a 33-km^2 watershed. Urban stormwater management practices, such as infiltration of runoff or the use of detention basins [35,36] may provide some benefit, but opportunity for adoption of these measures is limited, particularly in established neighborhoods, and it is likely that control will have to focus on the application practices by pest control professionals.

SUPPLEMENTAL DATA

Tables S1–S2. Toxicity testing and analytical chemistry results for inputs to the American River and in the river itself. (171 KB PDF).

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REFERENCES

1. Amweg EL, Weston DP, You J, Lydy MJ. 2006. Pyrethroid insecticides and sediment toxicity in urban creeks from California and Tennessee. *Environ Sci Technol* 40:1700–1706.
2. Brown JS, Sutula M, Stransky C, Rudolph J, Byron E. 2010. Sediment contaminant chemistry and toxicity of freshwater urban wetlands in Southern California. *J Am Water Res Assoc* 46:367–384.
3. Hintzen EP, Lydy MJ, Belden JB. 2009. Occurrence and potential toxicity of pyrethroids and other insecticides in bed sediments of urban streams in central Texas. *Environ Poll* 157:110–116.
4. Ding Y, Harwood AD, Foslund HM, Lydy MJ. 2010. Distribution and toxicity of sediment-associated pesticides in urban and agricultural waterways from Illinois, USA. *Environ Toxicol Chem* 29:149–157.
5. Weston DP, Asbell AM, Hecht SA, Scholz NL, Lydy MJ. 2011. Pyrethroid insecticides in urban salmon streams of the Pacific Northwest. *Environ Poll* 159:3051–3056.
6. Laskowski DA. 2002. Physical and chemical properties of pyrethroids. *Rev Environ Contam Toxicol* 174:49–170.
7. Weston DP, Lydy MJ. 2010. Urban and agricultural sources of pyrethroid insecticides to the Sacramento–San Joaquin Delta of California. *Environ Sci Technol* 44:1833–1840.
8. Weston DP, Jackson CJ. 2009. Use of engineered enzymes to identify organophosphate and pyrethroid-related toxicity in toxicity identification evaluations. *Environ Sci Technol* 43:5514–5520.
9. McKenney CL, Hamaker DB. 1984. Effects of fenvalerate on larval development of >*Palaemonetes* >*pugio* (Holthuis) and on larval metabolism during osmotic stress. *Aquat Toxicol* 5:343–355.
10. Van Wijngaarden RPA, Barber I, Brock TCM. 2009. Effects of the pyrethroid insecticide gamma-cyhalothrin on aquatic invertebrates in laboratory and outdoor microcosm tests. *Ecotoxicology* 18:211–224.
11. Bondarenko S, Spurlock F, Gan J. 2007. Analysis of pyrethroids in sediment porewater by solid phase microextraction. *Environ Toxicol Chem* 26:2587–2593.
12. Heines RL, Halpin PW. 2008. Analysis of pyrethroid pesticides in sediment and waters by EPA Method 8270 gas chromatography/mass spectrometer (GC/MS) narrow-range scan selected ion monitoring. In Gan J, Spurlock F, Hendley P, Weston D, eds, *Synthetic Pyrethroids: Occurrence and Behavior in Aquatic Environments*. ACS Symposium Series 991. American Chemical Society, Washington, DC, USA, pp 114–129.
13. Hladik ML, Smalling KL, Kuivila KM. 2008. A multi-residue method for the analysis of pesticides and pesticide degradates in water using HLB solid-phase extraction and gas chromatography–ion trap mass spectrometry. *Bull Environ Contam Toxicol* 80:139–144.
14. Williams JG. 2001. Chinook salmon in the lower American River, California's largest urban stream. *Fish Bull* 174:1–38.
15. Rantz SZ. 1982. *Measurement and Computation of Streamflow. Volume 1: Measurement of Stage and Discharge*. Water-Supply Paper 2175. U.S. Geological Survey, Washington, DC, USA.
16. Wang D, Weston DP, Lydy MJ. 2009. Method development for the analysis of organophosphate and pyrethroid insecticides at low parts per trillion levels in water. *Talanta* 78:1345–1351.
17. Weston DP, Lydy MJ. 2010. Focused toxicity identification evaluations to rapidly identify the cause of toxicity in environmental samples. *Chemosphere* 78:368–374.
18. U.S. Environmental Protection Agency. 2002. Methods for measuring the acute toxicity of effluents and receiving waters to freshwater and marine organisms. Fifth edition. EPA 821/R-02/012. Washington, DC.
19. Ingersoll CG, Ivey CD, Brunson EL, Hardesty DK, Kemble NE. 2000. Evaluation of toxicity: Whole-sediment versus overlying water exposures with amphipod *Hyalella azteca*. *Environ Toxicol Chem* 19:2906–2910.
20. Werner I, Deanovic LA, Markiewicz D, Khamphanh M, Reece CK, Stillway M, Reece C. 2010. Monitoring acute and chronic water column toxicity in the northern Sacramento–San Joaquin Estuary, California, USA, using the euryhaline amphipod, Hyalella azteca: 2006 to 2007. *Environ Toxicol Chem* 29:2190–2199.
21. Wheelock CE, Miller JL, Miller MJ, Phillips BM, Huntley SA, Gee SJ, Tjeerdema RS, Hammock BD. 2006. Use of carboxylesterase activity to remove pyrethroid-associated toxicity to *Ceriodaphnia dubia* and *Hyalella azteca* in toxicity identification evaluations. *Environ Toxicol Chem* 25:973–984.
22. U.S. Environmental Protection Agency. 2010. National Pollutant Discharge Elimination System test for significant toxicity technical document. EPA 833/R-10/004. Office of Wastewater Management. Washington, DC.
23. Weston DP, Holmes RW, Lydy MJ. 2009. Residential runoff as a source of pyrethroid pesticides to urban creeks. *Environ Poll* 157:287–294.
24. Bennett ER, Moore MT, Cooper CM, Smith S Jr, Shields FD Jr, Drouillard KG, Schulz R. 2005. Vegetated agricultural drainage ditches for the mitigation of pyrethroid-associated runoff. *Environ Toxicol Chem* 24:2121–1127.

25. Solomon KR, Giddings JM, Maund SJ. 2001. Probabilistic risk assessment of cotton pyrethroids: I. Distributional analyses of laboratory aquatic toxicity data. *Environ Toxicol Chem* 20:652–659.
26. U.S. Environmental Protection Agency. 2000. Methods for measuring the toxicity and bioaccumulation of sediment-associated contaminants with freshwater invertebrates. Second edition. EPA 600/R-99/064. Office of Research and Development. Duluth, MN.
27. Weston DP, You J, Harwood AD, Lydy MJ. 2009. Whole sediment toxicity identification evaluation tools for pyrethroid insecticides: III. Temperature manipulation. *Environ Toxicol Chem* 28:173–180.
28. Merz JE, Vanicek CD. 1996. Comparative feeding habits of juvenile chinook salmon, steelhead, and Sacramento squawfish in the lower American River, California. *Cal Fish Game* 82:149–159.
29. Siegfried BD. 1993. Comparative toxicity of pyrethroid insecticides to terrestrial and aquatic insects. *Environ Toxicol Chem* 12:1683–1689.
30. Anderson BS, Phillips BM, Hunt JW, Connor V, Richard N, Tjeerdema RS. 2006. Identifying primary stressors impacting macroinvertebrates in the Salinas River (California, USA): Relative effects of pesticides and suspended particles. *Environ Pollut* 141:402–408.
31. Pyrethroid Working Group. 2010. California 2009 urban pesticide use pattern study. Prepared by the Pyrethroid Working Group and Meta Research. Submitted to the California Department of Pesticide Regulation, Sacramento, CA, USA.
32. Gan J, Lee SJ, Liu WP, Haver DL, Kabashima JN. 2005. Distribution and persistence of pyrethroids in runoff sediments. *J Environ Qual* 34:836–841.
33. Budd R, O'Geen A, Goh KS, Bondarenko S, Gan J. 2011. Removal mechanisms and fate of insecticides in constructed wetlands. *Chemosphere* 83:1581–1587.
34. Holmes RW, Anderson BS, Phillips BM, Hunt JW, Crane DB, Mekebri A, Connor V. 2008. Statewide investigation of the role of pyrethroid pesticides in sediment toxicity in California's urban waterways. *Environ Sci Technol* 42:7003–7009.
35. Tsihrintzis VA, Hamid R. 1997. Modeling and management of urban stormwater runoff quality: a review. *Water Resource Manag* 11:137–164.
36. Birch GF, Fazeli MS, Matthai C. 2005. Efficiency of an infiltration basin in removing contaminants from urban stormwater. *Environ Monit Assess* 101:23–38.