ENANTIOMER-SPECIFIC MEASUREMENTS OF CURRENT-USE PESTICIDES IN AQUATIC SYSTEMS

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Table S1. EF values, sample description, and sampling date for river-, storm-, and waste- water samples contributed by Caltest

Sample	Description	Date (2013)	Fipronil EF	Bifenthrin EF	*cis*-Permethrin EF
	Racemic standards		0.453–0.513	0.479–0.504	0.498–0.518
1.	American River, Sacramento CA	5/1	NM[a]	NF[b]	NF
2.	American River, Sacramento CA	5/1	0.478	NF	NF

Sample	Description	Date (2013)	Fipronil EF	Bifenthrin EF	*cis*-Permethrin EF
3.	American River, Sacramento CA	5/1	0.501	NF	NF
4.	American River, Sacramento CA	5/1	0.490	NF	NF
5.	American River, Sacramento CA	5/7	0.511	NF	NM
6.	Freshwater, S California	5/21	0.495	NF	NF
7.	Freshwater, S California	5/21	NM	0.489	NF
8.	Freshwater, S California	5/21	0.493	0.494	NM
9.	Matrix Spike		NM	0.500	NM
10.	Matrix Spike		0.480	0.492	NM
11.	Matrix Spike		0.489	0.492	NM
12.	Matrix Spike		NM	0.491	**0.494**[c]
13.	Matrix Spike		0.480	0.502	NF
14.	Matrix Spike		0.487	0.496	NM
15.	Matrix Spike		0.464	**0.513**	NF
16.	Matrix Spike		**0.452**	**0.521**	NF
17.	POTW Effluent, N California	5/20	**0.523**	NF	NF
18.	POTW Effluent, N California	5/20	0.481	NF	NF
19.	POTW Effluent, N California	5/20	NM	NF	NF
20.	Seawater	5/21	NM	NF	NF
21.	Seawater	5/21	NM	0.494	**0.494**
22.	Seawater	5/21	NM	NF	NF
23.	Seawater	5/21	NM	0.489	NF
24.	Seawater	5/21	0.495	0.486	NF
25.	Seawater	5/21	NM	0.494	**0.486**
26.	Seawater	5/21	NM	0.494	**0.496**

Sample	Description	Date (2013)	Fipronil EF	Bifenthrin EF	*cis*-Permethrin EF
27.	Seawater	5/21	NM	0.493	NM
28.	Seawater	5/21	0.512	0.491	**0.490**
29.	Seawater	5/21	NM	0.492	0.500
30.	Seawater	5/21	NM	NF	0.499
31.	Seawater	5/21	NM	0.491	**0.484**
32.	Seawater	5/21	0.490	0.489	0.500
33.	Stormwater S California[d]	5/1	0.482	NF	NF
34.	Stormwater S California	5/1	0.493	NF	NF
35.	Stormwater S California	5/1	0.497	NF	NF
36.	Stormwater S California	5/1	NM	NF	NF
37.	Stormwater/Urban N California	5/1	NM	NF	NF
38.	Stormwater/Urban N California	5/1	0.486	NF	NF
39.	Stormwater/Urban N California	5/1	NM	NF	NF
40.	Stormwater/Urban N California	5/1	0.470	NF	NF
41.	Stormwater/Urban N California	5/1	NM	NF	NF
42.	Stormwater/Urban N California	5/1	0.498	NF	NF
43.	Stormwater/Urban N California	5/1	0.498	NF	NF
44.	Unknown	5/1	0.492	NF	NF
45.	Unknown	5/1	0.496	NF	NF

[a]NM = EF not measured; peaks detected but QC checks did not pass so data cannot be used.

[b]NF = Not found; no peaks detected during enantioselective analysis.

[c]Values in bold are non-racemic based on the range of EFs measured for racemic standards.

[d]All Stormwater and Stormwater/Urban were collected under dry weather conditions.

Table S2. Sampling date, sampling station ID, pesticide concentration, pesticide EF (italicized), and toxicity results for urban estuary sediment contributed by Southern California Coastal Water Research Project

Sample	Date	Station	Fipronil Conc (ng/g dry wt) EF	Bifenthrin Conc (ng/g dry wt) EF	*cis*-Permethrin Conc (ng/g dry wt) EF	*E. estuarius* % survival
Racemic standards			0.450–0.592	0.481–0.506	0.498–0.525	
1.	Oct 2007	1	ND[a] *NM*[b]	3.05 *NM*	3.94 *NF*[c]	89
2.	Oct 2007	2	ND *NM*	26.6 *NF*	31.8 *NM*	3
3.	Oct 2007	3	ND *NF*	79.6 *NF*	100 *NM*	0
4.	Oct 2007	4	ND *NM*	3.64 *NF*	3.92 *NF*	16
5.	Oct 2007	4	ND *0.537*	3.64 *NF*	3.92 *NF*	
6.	Oct 2007	4	ND *NM*	3.64 *NF*	3.92 *NF*	
7.	Oct 2007	5	ND *0.507*	4.57 *NF*	5.33 ***0.562***[d]	18
8.	Oct 2007	6	ND *0.506*	3.16 ***0.508***	2.86 *0.500*	8
9.	June 2008	Blank	–[e] *0.495*	– *NF*	– *NF*	

Sample	Date	Station	Fipronil Conc (ng/g dry wt) EF	Bifenthrin Conc (ng/g dry wt) EF	*cis*-Permethrin Conc (ng/g dry wt) EF	*E. estuarius* % survival
10.	June 2008	1	ND *NM*	8.3 *NF*	16.7 *NF*	72
11.	June 2008	1	ND *0.492*	8.3 *NM*	16.7 *NF*	
12.	June 2008	2	0.7 *0.495*	24.6 *NM*	50.6 *NF*	41
13.	June 2008	3	ND *NM*	5.1 *NF*	17.1 *NM*	57
14.	June 2008	4	0.1 *NM*	2.8 *NF*	6.6 *NF*	89
15.	June 2008	5	1.1 *0.504*	67.6 *NF*	92.1 *0.516*	0
16.	June 2008	6	0.2 *0.490*	6.6 *0.503*	11.5 ***0.532***	49
17.	Oct 2008	Blank	– *NM*	- *NF*	- *NF*	
18.	Oct 2008	1	ND *NM*	17.9 *NF*	43.6 *NF*	
19.	Oct 2008	1	ND *NM*	17.9 *NF*	43.6 *NF*	
20.	Oct 2008	2	ND *0.500*	6.99 *NF*	15.4 *NF*	78

Sample	Date	Station	Fipronil Conc (ng/g dry wt) EF	Bifenthrin Conc (ng/g dry wt) EF	*cis*-Permethrin Conc (ng/g dry wt) EF	*E. estuarius* % survival
21.	Oct 2008	2	ND *0.471*	6.99 *NF*	15.4 *NF*	
22.	Oct 2008	3	ND *0.490*	34 *NF*	99.4 *NM*	
23.	Oct 2008	4	ND *NM*	4.37 *NF*	7.05 *NF*	
24.	Oct 2008	5	ND *NM*	13.5 *NM*	23 *NF*	88
25.	Oct 2008	5	ND *0.514*	15.5 *NF*	23 *NM*	
26.	Oct 2008	6	ND *0.485*	3.08 ***0.526***	6.12 *NF*	
27.	Aug 2009	Blank	– *NM*	– *NF*	– *NF*	
28.	Aug 2009	1	ND *NM*	1.79 *NF*	ND *NF*	
29.	Aug 2009	2	ND *0.500*	15.3 *NF*	21.4 *NM*	
30.	Aug 2009	3	0.118 *0.529*	19.4 *NF*	23.2 *0.506*	
31.	Aug 2009	4	0.069 *0.516*	3.84 *NF*	6.24 *NM*	

Sample	Date	Station	Fipronil Conc (ng/g dry wt) EF	Bifenthrin Conc (ng/g dry wt) EF	*cis*-Permethrin Conc (ng/g dry wt) EF	*E. estuarius* % survival
32.	Aug 2009	5	0.144 *NM*	2 *NF*	2.56 *NM*	
33.	Aug 2009	6	0.434 *0.463*	5.83 ***0.526***	13.7 *NF*	
34.	Nov 2009	Blank	– *0.507*	– *NF*	– *NF*	
35.	Nov 2009	1	0.798 *NF*	25.3 *NM*	61.5 *NF*	5
36.	Nov 2009	2	0.422 *0.499*	14.3 *NF*	24 *NF*	12
37.	Nov 2009	2	0.422 *0.513*	14.3 *NF*	24 *NM*	
38.	Nov 2009	3	0.082 *0.484*	4.16 *NF*	5.27 ***0.460***	23
39.	Nov 2009	5	0.036 *0.474*	0.684 *0.499*	1.52 *NF*	37
40.	Dec 2009	Blank	- *0.495*	- *NF*	- *NF*	
41.	Dec 2009	1	0.43 *0.475*	8.36 *NM*	17.6 *NF*	15
42.	Dec 2009	1	0.43 *0.472*	8.36 *NF*	17.6 *NF*	

Sample	Date	Station	Fipronil Conc (ng/g dry wt) EF	Bifenthrin Conc (ng/g dry wt) EF	*cis*-Permethrin Conc (ng/g dry wt) EF	*E. estuarius* % survival
43.	Dec 2009	2	0.613 *0.500*	11.6 *NF*	16.8 *NM*	20
44.	Dec 2009	3	0.349 *0.560*	3.41 *NF*	6.12 *NF*	24
45.	Dec 2009	5	0.227 *NM*	0.592 *NF*	0.665 *NM*	8

[a]ND = concentration below detection limit (varies).

[b]NM = EF not measured; peaks detected but QC checks did not pass so data cannot be used.

[c]NF = Not found; no peaks detected during enantioselective analysis.

[d]Values in bold are non-racemic based on the range of EFs measured for racemic standards.

[e]Concentrations were not reported for blanks.

Table S3. Time of sample collection, formulation, and pesticide EF for runoff samples from concrete treated with bifenthrin and permethrin contributed by UC Riverside

Sample	Day	Formulation[a]	Fipronil EF	Bifenthrin EF	*cis*-Permethrin EF
Racemic standards			0.351 -0.540[b]	0.457-0.500	0.477-0.518
1.	1	S	0.484	0.499	NF[c]
2.	1	S	NF	0.478	0.490
3.	1	S	NM[d]	0.496	NF
4.	1	S	NF	0.483	0.496
5.	7	L	0.480	0.478	**0.094**[e]
6.	7	L	NF	NF	**0.122**

Sample	Day	Formulation[a]	Fipronil EF	Bifenthrin EF	*cis*-Permethrin EF
7.	7	L	NF	0.482	**0.175**
8.	7	L	NF	NF	**0.205**
9.	7	P	0.399	**0.452**	**0.454**
10.	7	P	0.435	0.459	**0.445**
11.	7	P	NF	0.487	0.487
12.	7	P	NF	NF	0.496
13.	7	P	0.421	NM	0.494
14.	7	P	NF	NF	0.492
15.	7	P	0.424	**0.353**	0.498
16.	7	P	NF	NF	0.495
17.	7	S	0.477	0.476	**0.431**
18.	7	S	NF	**0.507**	**0.425**
19.	7	S	NF	NF	NM
20.	7	S	NF	NM	0.493
21.	20	L	NF	NF	NM
22.	20	L	NF	NF	**0.391**
23.	20	P	NF	NF	0.502
24.	20	P	NF	NF	NM
25.	20	P	0.480	NF	NM
26.	20	P	0.473	0.473	0.502
27.	20	P	NF	NF	0.501
28.	20	P	NF	NF	0.491

[a]L = ready-to-use liquid, P = professional concentrate, S = ready-to-use solid.

[b]Some racemic standard EFs for fipronil are lower than what is typically measured. The EF of the 10 ng/µL standard was measured three times at 0.399, 0.383, and 0.415 at the beginning, middle, and end of the sequence of samples, respectively. Additionally, the EF of the 100 ng/µL

standard was measured at 0.351 at the end of the sequence (the other two measurements were typical). Samples and standards measured just before or after these unusually low EFs were not remarkable, nor did they indicate carryover. While surprising and inexplicable, these low EFs for racemic standards met all QA/QC criteria, and thus were not removed from the data set as we feel they correctly reflect variability in our measurements.

[c]NF = Not found; no peaks detected during enantioselective analysis.

[d]NM = EF not measured; peaks detected but QC checks did not pass so data cannot be used.

[e]Values in bold are non-racemic based on the range of EFs measured for racemic standards.

Table S4. Time of sample collection, health status, length, weight, tissue concentration, and EF of bifenthrin for salmon uptake samples dosed with 0.2 µg/L bifenthrin in water contributed by U.S. Geological Survey

Sample	Day since dose	Status[a]	Length (mm)	Weight (g)	Tissue Conc (ng/g)	Bifenthrin EF
Racemic standards						0.465-0.521
1.	1	A	70	3.949	428	**0.451**[b]
2.	1	A	66	3.227	478	**0.449**
3.	1	A	65	3.203	552	**0.440**
4.	1	A	65	2.862	449	**0.401**
5.	1	A	68	3.626	465	**0.460**
6.	1	A	64	2.770	408	**0.458**
7.	2	D	70	3.235	2062	**0.526**
8.	2	D	69	3.283	1850	**0.447**
9.	2	D	63	2.626	1684	**0.399**

Sample	Day since dose	Status[a]	Length (mm)	Weight (g)	Tissue Conc (ng/g)	Bifenthrin EF
10.	2	D	70	3.394	1341	**0.417**
11.	2	A	72	3.286	1216	**0.441**
12.	2	A	74	3.832	1474	0.466
13.	2	A	69	2.981	1922	**0.428**
14.	3	A	-[c]	3.863	1377	**0.411**
15.	3	A	65	3.413	1452	**0.409**
16.	3	D	74	4.896	1115	**0.392**
17.	3	D	62	2.293	1731	**0.454**
18.	3	D	60	2.654	1614	**0.399**
19.	3	D	62	3.208	1373	**0.441**
20.	3	D	62	2.914	1921	**0.378**
21.	3	D	70	4.530	1109	**0.440**
22.	3	D	62	3.379	1186	0.466
23.	3	D	61	2.953	1455	**0.384**

[a]A = Alive; D = Dead.

[b]Values in bold are non-racemic based on the range of EFs measured for racemic standards.

[c]Reported fish length believed to be in error, therefore removed.

Additional samples

No additional sample information was available for the California Department of Pesticide Regulation (CDPR) surface water samples (Figure SI S2A and Table S5), however it is interesting to note that of 12 bifenthrin EFs, 6 were less than and 3 were greater than the EF for racemic standards, making 9 of 12 samples non-racemic. It would have been interesting to investigate if the EFs above/below racemic standards came from similar or different sample locations, sampling dates, or other water conditions, but this was not possible without further information.

No additional sample information was available for the California Department of Fish and Wildlife (CDFW) water and sediment samples (Figure SI S2B and Table S6). Half (5 of 10) of the bifenthrin EFs and both of the permethrin EFs (2 of 2) were non-racemic. It would have been interesting to explore whether EF values in the sediment and water were similar at a given location, or if enantioselective processes were occurring solely in one of the matrices, but this was not possible without further information. Overall, the CDRP and CDFW samples showed similar trends as other environmental samples reported here.

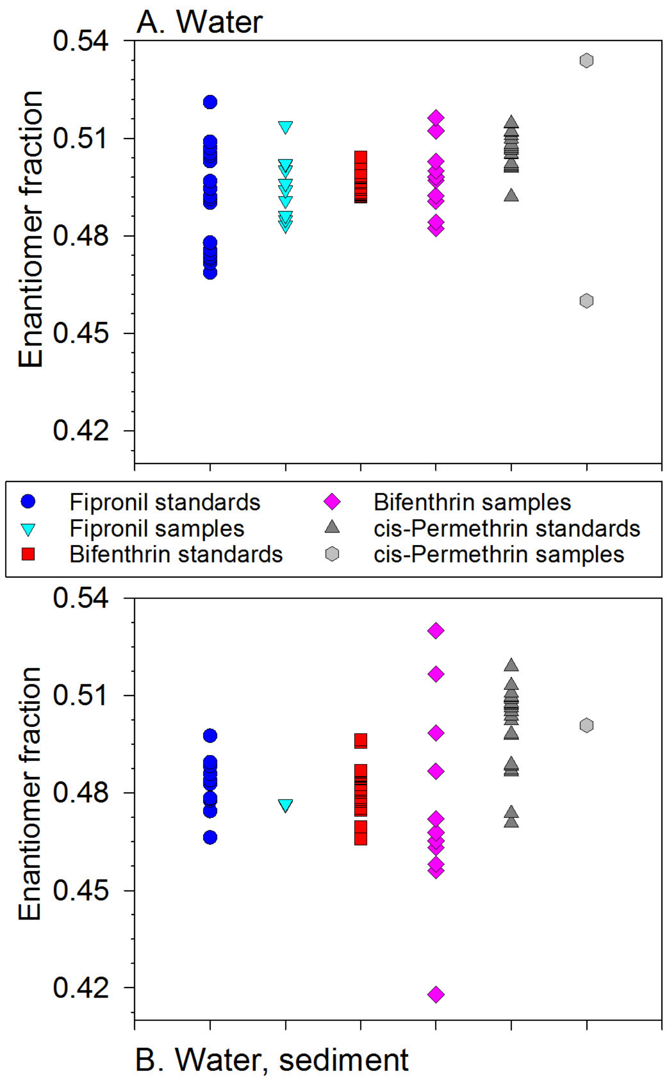

Figure S2. Enantiomer fractions measured in racemic standards and samples donated from two organizations. Panel A shows surface water samples donated by California Department of Pesticide Regulation; Panel B shows surface water and urban sediment samples donated by the California Department of Fish and Wildlife. Details for these standards and samples are listed in Supplemental Tables 5-6.

Table S5. EF values for surface water samples contributed by the California Department of Pesticide Regulation. No additional sample details were available.

Sample	Fipronil EF	Bifenthrin EF	*cis*-Permethrin EF
Racemic standards	0.466–0.498	0.466–0.496	0.471–0.519
1.	NF[a]	NF	NM[b]
2.	NF	**0.499**[c]	NF
3.	0.477	**0.465**	NF
4.	NF	0.468	NF
5.	NF	NM	NF
6.	NF	NM	NF
7.	0.476	0.487	NF
8.	NF	**0.463**	NF
9.	NF	NM	NF
10.	NF	NM	NF
11.	NF	NM	NM
12.	NF	NM	NM
13.	NF	NF	NF
14.	NF	NF	NF
15.	NF	NM	NF
16.	NF	NF	NF
17.	NF	NF	NF
18.	NF	NF	0.501
19.	NF	NM	NF
20.	NF	NM	NF
21.	NF	**0.465**	NF

Sample	Fipronil EF	Bifenthrin EF	*cis*-Permethrin EF
22.	NF	NM	NF
23.	NF	0.472	NM
24.	NF	NF	NF
25.	NF	**0.456**	NM
26.	NF	**0.458**	NF
27.	NF	NM	NM
28.	NF	**0.530**	NF
29.	NF	NM	NF
30.	NF	NM	NF
31.	NF	NM	NF
32.	NF	**0.418**	NF
33.	NF	NM	NF
34.	NF	NM	NF
35.	NF	NF	NF
36.	NF	**0.517**	NF

[a]NF = Not found; no peaks detected during enantioselective analysis.

[b]NM = EF not measured; peaks detected but QC checks did not pass so data cannot be used.

[c]Values in bold are non-racemic based on the range of EFs measured for racemic standards.

Table S6. EF values for surface water and urban sediment samples contributed by the California Department of Fish and Wildlife. No additional sample details were available

Sample	Fipronil EF	Bifenthrin EF	*cis*-Permethrin EF
Racemic standards	0.469–0.521	0.492–0.504	0.492–0.515
1.	0.502	**0.491**[a]	**0.534**

Sample	Fipronil EF	Bifenthrin EF	*cis*-Permethrin EF
2.	NM[a]	NM	NF[b]
3.	NM	NM	NF
4.	0.486	**0.484**	NF
5.	0.514	0.492	NF
6.	NM	NM	NF
7.	0.491	**0.482**	NF
8.	NM	**0.516**	NF
9.	0.485	0.498	NF
10.	0.494	0.497	**0.460**
11.	0.483	0.500	NF
12.	NM	NM	NF
13.	0.500	**0.512**	NF
14.	0.502	NM	NF
15.	0.496	0.503	NF

[a]Values in bold are non-racemic based on the range of EFs measured for racemic standards.

[b]NM = EF not measured; peaks detected but QC checks did not pass so data cannot be used.

[c]NF = Not found; no peaks detected during enantioselective analysis.

Discussion of enantiomer assignment reversal from Table 1.

The assignment of absolute structure and light rotation is an important but difficult task for chiral compounds. For a number of reasons, it appears that the assignment of (+) and (-) bifenthrin has been reversed in reference [1]. The first indication that there might be an issue was that the more toxic enantiomer for *Daphnia pulex* reported in reference [1] is opposite that reported for similar species by other researchers [2–4]. One study determined that the toxicity of an enantiopure 1R-*cis* bifenthrin obtained from FMC Corporation was the only enantiomer contributing toxicity to a racemic mixture for *Ceriodaphnia dubia* [2]. The 1R-*cis* bifenthrin is identified as the (+) enantiomer in reference [3].

Further investigation revealed the elution order for bifenthrin on a Sumichiral OA-2500I HPLC column with a mobile phase of > 99% hexane with small amounts of various modifiers is reversed between references [1] and [3]. It is expected that the elution order with such similar conditions would be the same. The elution order reported in reference [3] on a BGB-172 GC column determined using an enantiopure 1R-*cis*-bifenthrin standard provided by FMC Corporation matches the elution order of this study. The method for determining the assignment of (+) and (-) in reference [3] was by laser polarimeter at 675 nm, in full agreement with polarimetry (wavelength unknown) for single enantiomers prepared by Chirosolve used in this study. Reference [1] used specific rotation at 365 nm to assign the enantiomers, which lead to the disagreement of enantiomer assignment.

The authors were unable to locate errata for the article in question, but determined it was more scientifically justified given the evidence to not perpetuate the error. Therefore we have reversed the assignment of (+) and (-) bifenthrin from reference [1] for Table 1 of this manuscript.

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