

Potential Exposure of Larval and Juvenile Delta Smelt to Dissolved Pesticides in the Sacramento–San Joaquin Delta, California

KATHRYN M. KUIVILA* AND G. EDWARD MOON

U.S. Geological Survey, Placer Hall, 6000 J Street, Sacramento, California 95819-6129, USA

Abstract.—The San Francisco Estuary is critical habitat for delta smelt *Hypomesus transpacificus*, a fish whose abundance has declined greatly since 1983 and is now listed as threatened. In addition, the estuary receives drainage from the Central Valley, an urban and agricultural region with intense and diverse pesticide usage. One possible factor of the delta smelt population decline is pesticide toxicity during vulnerable larval and juvenile stages, but pesticide concentrations are not well characterized in delta smelt spawning and nursery habitat. The objective of this study was to estimate the potential exposure of delta smelt during their early life stages to dissolved pesticides. For 3 years (1998–2000), water samples from the Sacramento–San Joaquin Delta were collected during April–June in coordination with the California Department of Fish and Game’s delta smelt early life stage monitoring program. Samples were analyzed for pesticides using solid-phase extraction and gas chromatography/mass spectrometry. Water samples contained multiple pesticides, ranging from 2 to 14 pesticides in each sample. In both 1999 and 2000, elevated concentrations of pesticides overlapped in time and space with peak densities of larval and juvenile delta smelt. In contrast, high spring outflows in 1998 transported delta smelt away from the pesticide sampling sites so that exposure could not be estimated. During 2 years, larval and juvenile delta smelt were potentially exposed to a complex mixture of pesticides for a minimum of 2–3 weeks. Although the measured concentrations were well below short-term (96-h) LC50 values for individual pesticides, the combination of multiple pesticides and lengthy exposure duration could potentially have lethal or sublethal effects on delta smelt, especially during early larval development.

Introduction

Delta smelt *Hypomesus transpacificus* is a small, typically annual fish endemic to the San Francisco Estuary (Moyle 2002). The abundance of delta smelt has declined greatly since the early 1980s, leading to its listing as a threatened species in 1993 (Sweetnam 1999). The combination of a 1-year life cycle and a weak stock–recruitment relationship suggests that environmental conditions may highly influence its population success (Moyle et al. 1992).

Many factors have been suggested as contributing to the declining population of delta

smelt. One of these factors is pesticide toxicity during vulnerable larval and juvenile stages (Bennett and Moyle 1996). During early life stages, the primary geographic range of delta smelt is the Sacramento–San Joaquin Delta. Although pesticides have been detected in the delta (Kuivila and Foe 1995; MacCoy et al. 1995; Kuivila et al. 1999), there are minimal pesticide data in delta smelt habitat. The data are particularly sparse in spring and summer months when larval and juvenile life stages of delta smelt are present.

The objectives of this study were to identify and characterize dissolved pesticide concentrations in the delta during 1998–2000 and to assess the potential exposure of larval and

*Corresponding author: kkuivila@usgs.gov

juvenile delta smelt to these dissolved pesticides. Sampling of water for dissolved pesticides was coordinated with delta smelt surveys by the California Department of Fish and Game (CDFG). Results show co-occurrence of multiple pesticides and delta smelt and are discussed within the context of possible toxic effects to delta smelt.

Study Area

The delta has two major freshwater inputs: Sacramento River from the north and San Joaquin River from the southeast (Figure 1). During spring and summer, Sacramento River discharge is typically three to eight times that of the San Joaquin River. Net seaward flow of the delta is westward through Suisun Bay, while water is exported for agricultural and municipal consumption via two water diversions, the State Water Project (SWP) and Central Valley Project (CVP), located in the south delta. During most years, combined export pumping is approximately equal to San Joaquin River discharge during spring and exceeds San Joaquin River discharge in summer. The result is that Sacramento River water is drawn through the central delta towards the pumps.

Delta smelt primarily spawn during early April through mid-May (Moyle et al. 1992; Moyle 2002). Spawning locations vary from year to year, depending on environmental conditions; however, a consistent spawning area is the northwestern delta, including Cache and Lindsey sloughs (Figure 1). Newly hatched delta smelt are semibuoyant, float near the bottom, and remain in the general area where they hatched. After several weeks, larval delta smelt develop swim bladders and fins, move up in the water column, and are transported downstream (Moyle 2002). Juvenile delta smelt tend to congregate near the confluence of the Sacramento and San Joaquin rivers (confluence; Moyle et al. 1992; Figure 1).

The delta receives drainage from the Central Valley, an urban and agricultural region with intense and diverse pesticide usage. Generally, pesticide sources can be attributed to specific uses (Kuivila and Foe 1995; Kuivila et al. 1999). The combination of use and a mechanism to transport pesticides off-site to surface

waters can explain most pesticide occurrences. There are several sources of pesticides to the delta in spring and summer when delta smelt larvae and juveniles are present (Kuivila 1999). These include both external or riverine inputs and local, or within-delta inputs. One major riverine input is rice pesticides, which occurs every year when rice field water is released in May and June (Crepeau and Kuivila 2000). Other inputs include runoff of alfalfa pesticides following late rainfall in spring and irrigation return-flow transport of a variety of herbicides in summer. In addition, pesticides applied on delta islands on a wide variety of crops can cause elevated pesticide concentrations in localized areas (Kuivila et al. 1999).

Methods

Study design

Most water samples for pesticide analysis were collected in conjunction with delta smelt surveys by CDFG in spring and summer of 1998–2000. Concurrent sampling allowed for direct comparison of pesticide concentrations and fish abundances. The start of pesticide sampling was triggered by detection of adult delta smelt in spawning areas.

Pesticide sampling sites (Figure 1) varied from year to year as the sampling design was modified. In 1998, five delta sites were selected to cover known spawning areas. Five new sites were added in 1999 to further characterize the spatial variability within the delta. In 2000, sampling was modified to focus on source inputs. Three new sites were selected to characterize the two major riverine inputs (Sacramento and San Joaquin rivers) and a potential within-delta source of one pesticide, eptam. None of these sites were delta smelt sampling sites. Two sites (Cache Slough and Middle River at Bacon Island), sampled in 1998 and 1999, were also sampled in 2000. Delta sites were sampled biweekly with the exception of Sacramento River at Sacramento, which was sampled weekly in 2000.

Exposure of juvenile delta smelt at the confluence was characterized by sampling at one site, Suisun Bay at Mallard Island. In 1998 and 1999, sampling was based on the hypothesis that juvenile delta smelt generally con-

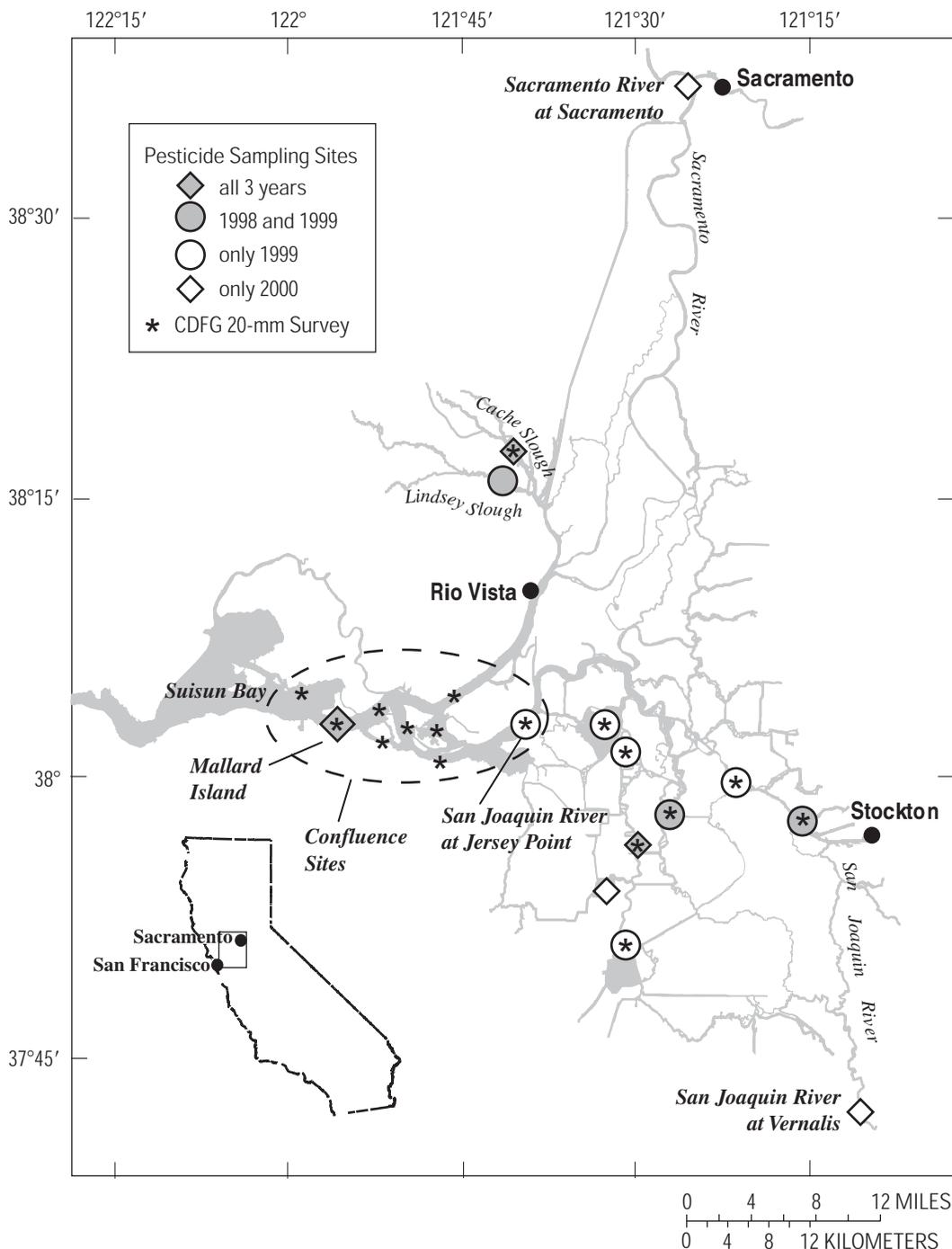


FIGURE 1. Map of sampling locations in the Sacramento-San Joaquin Delta.

gregate at 2‰ salinity (Moyle 2002). Auto-samplers were used to collect samples and were programmed to collect no more than one sample per day on the ebb tide as 2‰ water

passed the site. Because of equipment difficulties in 1998, samples were not collected successfully at this site. In 1999, samples were collected daily from mid-June to mid-August.

Pesticide sampling was changed from salinity-based to weekly from May to August 2000 to be consistent with the timing of delta smelt sampling.

Pesticide sampling and analysis

Water samples for pesticide analyses were collected using a weighted sampler holding either two 1-L amber glass bottles or a 3-L Teflon bottle. Samples were collected at 1 m below the surface at mid-channel from a bridge or from the fish monitoring vessel. Samples were preserved on ice until extraction in the laboratory.

One liter of water was filtered through a 0.7-mm baked glass fiber filter. Terbutylazine was added as a surrogate to verify cartridge efficiency. The water sample was extracted by pumping through a 500-mg C-8 solid-phase extraction cartridge at a flow rate of 20 mL/min. The cartridges were dried by applying a positive pressure of carbon dioxide and stored frozen until analysis. Thawed cartridges were eluted with 9 mL of ethyl acetate, internal standards were added, and eluant was evaporated to approximately 200 mL. Samples were analyzed for 28 current-use pesticides by gas chromatography/mass spectrometry with ion-trap detection. Details of the method, including accuracy and precision of data, are described in Crepeau et al. (2000).

Quality control data included field blanks, replicate samples, matrix spikes, inter-laboratory samples, and surrogate recovery. No pesticides were detected in equipment blanks. Replicate and interlaboratory comparison samples agreed within method limits (Crepeau et al. 2000). Recovery of all compounds was verified using matrix spike samples, and recovery of the surrogate, terbutylazine, was recorded to assess the efficiency of each extraction. More information about the quality-assurance practices are detailed in Crepeau et al. (2000).

Sampling of larval and juvenile delta smelt

Delta smelt density data from 17 sites (Figure 1) were obtained from the CDFG 20-mm Survey (Dege and Brown 2004, this volume, provide extensive details on the sampling pro-

gram). Samples were collected with a 5.1-m-long, 1,600-mm stretched mesh plankton net mounted on a townet frame. The volume of water sampled was estimated with a flowmeter mounted at the net mouth. Abundance of delta smelt is reported as number per unit volume of water sampled.

Data analysis

Exposure of delta smelt to dissolved pesticides was estimated by examining overlap of fish densities and detected pesticide concentrations. This analysis was only done for 1999 and 2000 data, since fish densities in 1998 were very low at delta sites. Total pesticide concentrations were calculated as the sum of all pesticides measured in a single sample. Examination of data for San Joaquin River at Jersey Point (Figure 1) revealed that fish densities and pesticide concentrations were very similar to the confluence site, Suisun Bay at Mallard Island. Therefore, data from Jersey Point were excluded from the delta data and grouped with the confluence data for purposes of interpretation.

Total pesticide concentrations increased and decreased over time in a similar pattern at all delta sites; therefore, data were averaged to simplify presentation. For the delta, fish densities were averaged at eight sites for both years, while total pesticide concentrations were averages of nine sites in 1999 and four sites in 2000. For the confluence, fish densities were averaged at nine sites for both years, while total pesticide concentrations were averages of concentrations at Jersey Point and Suisun Bay at Mallard Island in 1999, but were measured only at Suisun Bay at Mallard Island in 2000.

Results

Multiple pesticides in delta smelt habitat

Water samples collected in the delta and the confluence contained a variety of pesticides (Table 1). Throughout the 3 years of study, 202 water samples were analyzed for 28 pesticides. Twenty-three pesticides were detected, while five pesticides were not detected: fonofos, malathion, methidation, methyl parathion, and phosmet. Metolachlor, the most

TABLE 1. Pesticides detected in the Sacramento-San Joaquin Delta, 1998-2000. Frequency of detection and concentrations of pesticides shown as maximum and 90th percentile concentration for each year. Fonofos, malathion, methidathion, methyl parathion, and phosmet were not detected. Concentrations are in ng/L. * = not analyzed; nd = not detected.

Pesticide	All Years (N = 202)			1998 (N = 41)			1999 (N = 107)			2000 (N = 54)		
	Percent samples detected	Percent samples detected	Maximum	90th percentile	Maximum	Percent samples detected	90th percentile	Maximum	Percent samples detected	90th percentile	Maximum	
Alachlor	8	28	21	11	21	2	nd	56	6	nd	17	
Atrazine	35	78	43	16	43	10	1	24	54	21	32	
Butylate	3	0	nd	nd	nd	0	nd	nd	11	11	22	
Carbaryl	7	0	nd	nd	nd	5	nd	210	17	15	72	
Carbofuran	42	58	29	23	29	29	39	82	57	25	50	
Chlorpyrifos	12	47	12	4	12	0	nd	nd	11	10	32	
Cyanazine	15	61	54	14	54	0	nd	nd	*	*	*	
Cycloate	11	*	*	*	*	*	*	*	11	11	38	
Dacthal	6	22	1	1	1	0	nd	nd	6	nd	11	
Diazinon	24	97	46	17	46	0	nd	nd	24	15	30	
Diethaty/ethyl	2	11	3	1	3	0	nd	nd	0	nd	nd	
EPTC	43	50	1,500	33	1,500	36	27	7,700	52	88	550	
Ethafuralin	1	0	nd	nd	nd	0	nd	nd	4	nd	30	
Metolachlor	95	100	210	130	210	96	120	210	91	85	210	
Molinate	74	53	2,500	320	2,500	77	290	440	83	690	3,000	
Napropamide	14	75	15	11	15	0	nd	nd	0	nd	nd	
Oxyfluorfen	14	67	10	6	10	0	nd	nd	7	nd	86	
Pebulate	4	17	140	7	140	1	nd	84	0	nd	nd	
Pendimethalin	13	44	9	3	9	0	nd	nd	19	26	31	
Simazine	57	100	68	37	68	42	51	64	57	42	49	
Sulfotep	20	0	nd	nd	nd	37	66	120	0	nd	nd	
Thiobencarb	61	14	330	130	330	70	100	150	76	130	1,300	
Trifluralin	42	69	8	4	8	25	10	65	59	40	44	

frequently detected pesticide, occurred in 95% of samples. Molinate and thiobencarb were detected in 74% and 61% of samples, respectively. Other frequently detected pesticides (35–57%) included atrazine, carbofuran, eptam, simazine, and trifluralin.

Each water sample contained multiple pesticides. All samples contained detectable concentrations of at least 2 pesticides, median number of pesticides detected per sample was 5, and maximum number of pesticides detected in a sample was 14 (Figure 2).

Annual variability of pesticide distribution

Although multiple pesticides were always detected in samples collected each year, there was high variability in actual pesticide occurrence from year to year. This included both temporal and spatial variation in detection and concentration.

Frequency of detection of individual pesticides can be compared from year to year (Table 1). In all years, metolachlor had the highest frequency of detection (91–100%), and simazine was frequently detected (100%, 42%, and 57%). In contrast, frequency of detection of other pesticides varied considerably from year to year (Table 1). Frequency of detection of molinate and thiobencarb was low in 1998, but substantially higher in 1999 and 2000. In contrast, atrazine, carbofuran, eptam, and trifluralin were frequently detected in 1998 and 2000, but not in 1999.

There was considerable variability in pesticides that were not detected in an individual year (Table 1). Carbaryl was not detected in 1998, but was detected in 1999 and 2000. Chlorpyrifos, dacthal, diazinon, oxyfluorfen, and pendimethalin were not detected in 1999 (but were detected the other 2 years), while sulfotep was only detected in 1999.

Pesticide concentrations varied from year

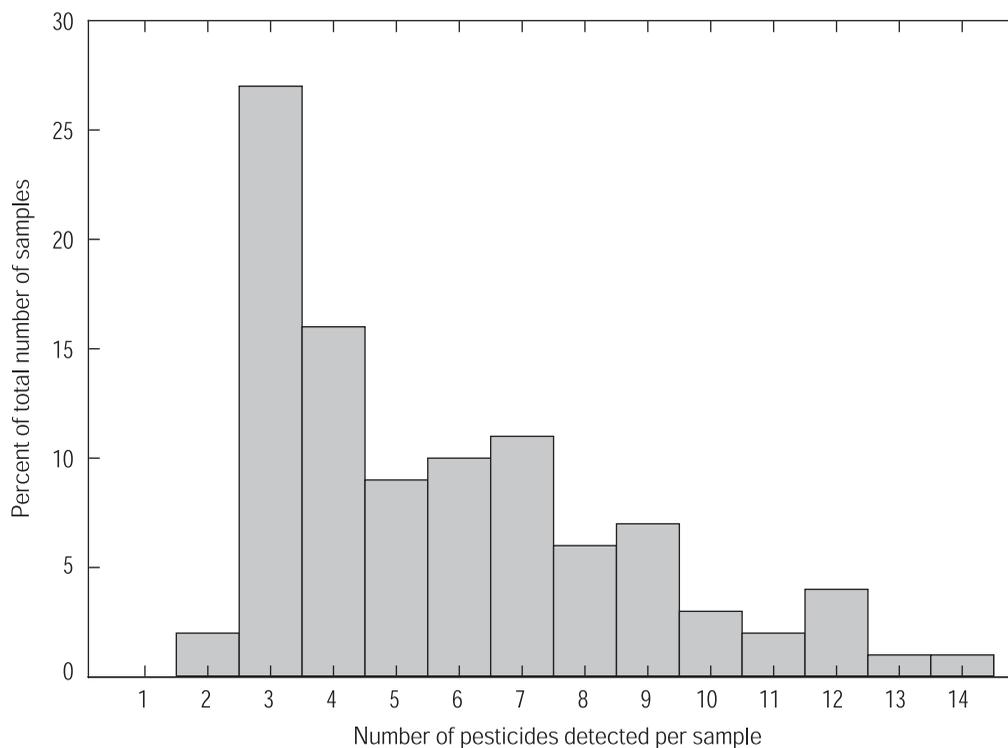


FIGURE 2. Number of pesticides detected per sample, 1998–2000.

to year (Table 1). Maximum concentrations of eptam, molinate, pebulate, sulfotep, and thiobencarb varied considerably from year to year; however, 90th percentiles of their concentrations were very similar. This suggests that only a few samples had unusually high concentrations. For example, highest concentrations of eptam were measured in one or two samples every year and were at least five times higher than any other concentrations. This was likely due to localized "hot spots" from within-delta inputs that were inadequately characterized by our sampling design.

Another approach to examine annual variability is by comparing concentrations at one site over all 3 years. The concentrations of molinate and thiobencarb at Cache Slough varied considerably with 1998 greater than 2000 and 2000 greater than 1999 (Figure 3). The two pesticides are applied to rice, and observed differences in concentration have been explained previously by variations in actual holding time of water on rice fields before release (Crepeau and Kuivila 2000).

Overlap of larval and juvenile delta smelt with dissolved pesticides

In 1998, few delta smelt were caught at sites with concurrent pesticide sampling. High spring outflows resulted in the delta smelt population being centered downstream in Suisun Bay. Since delta smelt were not captured (but could have still been present in low numbers) at the pesticide sampling sites in 1998, no estimate was made of their potential exposure to pesticides.

Delta smelt and pesticide sampling sites overlapped in 1999 and 2000 (Figures 4 and 5). In 1999, highest densities of delta smelt occurred at delta sites from 10 May to 7 June (Figure 4A). During this time, pesticides co-occurred with fish. The highest pesticide concentrations were detected on 10 May. Extremely high concentrations of eptam (7,700 and 4,300 ng/L) detected at two sites strongly influenced total pesticide concentration on 10 May. Even without eptam, however, pesticide concentrations were elevated throughout the period that delta smelt were present.

Peak fish abundance at the confluence lagged delta sites by 2 weeks in 1999. The highest density of delta smelt was found on 21 June (Figure 4B). As at the delta sites, highest densities of delta smelt co-occurred with highest concentrations of dissolved pesticides. But this time, peak pesticide concentrations were composed primarily of rice pesticides molinate and thiobencarb. These results suggest that a significant fraction of the delta smelt population was being exposed to these pesticides.

For 2000, total pesticide concentration for Sacramento River at Sacramento is shown separately (Figure 5A) because pesticide concentrations were much higher than other delta sites. This difference occurred because of input of rice field water, which contained elevated concentrations of molinate and thiobencarb, into the Sacramento River (Crepeau and Kuivila 2000). Delta smelt were present in the delta from mid-April to the end of the CDFG 20-mm Survey in late June, during which densities remained relatively constant (Figure 5A). Concentrations of pesticides were also elevated throughout this period.

At the confluence, a distinct peak in fish density in 2000 occurred on 14 June (Figure 5B), in contrast to the relatively constant abundance in the delta. Highest densities of delta smelt co-occurred with highest total concentrations of pesticides. Elevated concentrations of molinate and thiobencarb, similar to 1999, strongly influenced the observed total pesticide concentration peak.

Discussion

Variability in overlap of fish and pesticides

The spatial distribution of pesticides and delta smelt densities varied from year to year. One factor that probably influenced both pesticide and fish transport is hydrodynamics. The importance of flow can be illustrated by comparing the 3 years. In 1998, concentrations of molinate and thiobencarb in Cache Slough were very high (Figure 3); however, these pesticides were not even detected at any other delta sites. In contrast, in 1999 and 2000, concentrations of molinate and thiobencarb in Cache Slough were much lower (Figure 3), yet

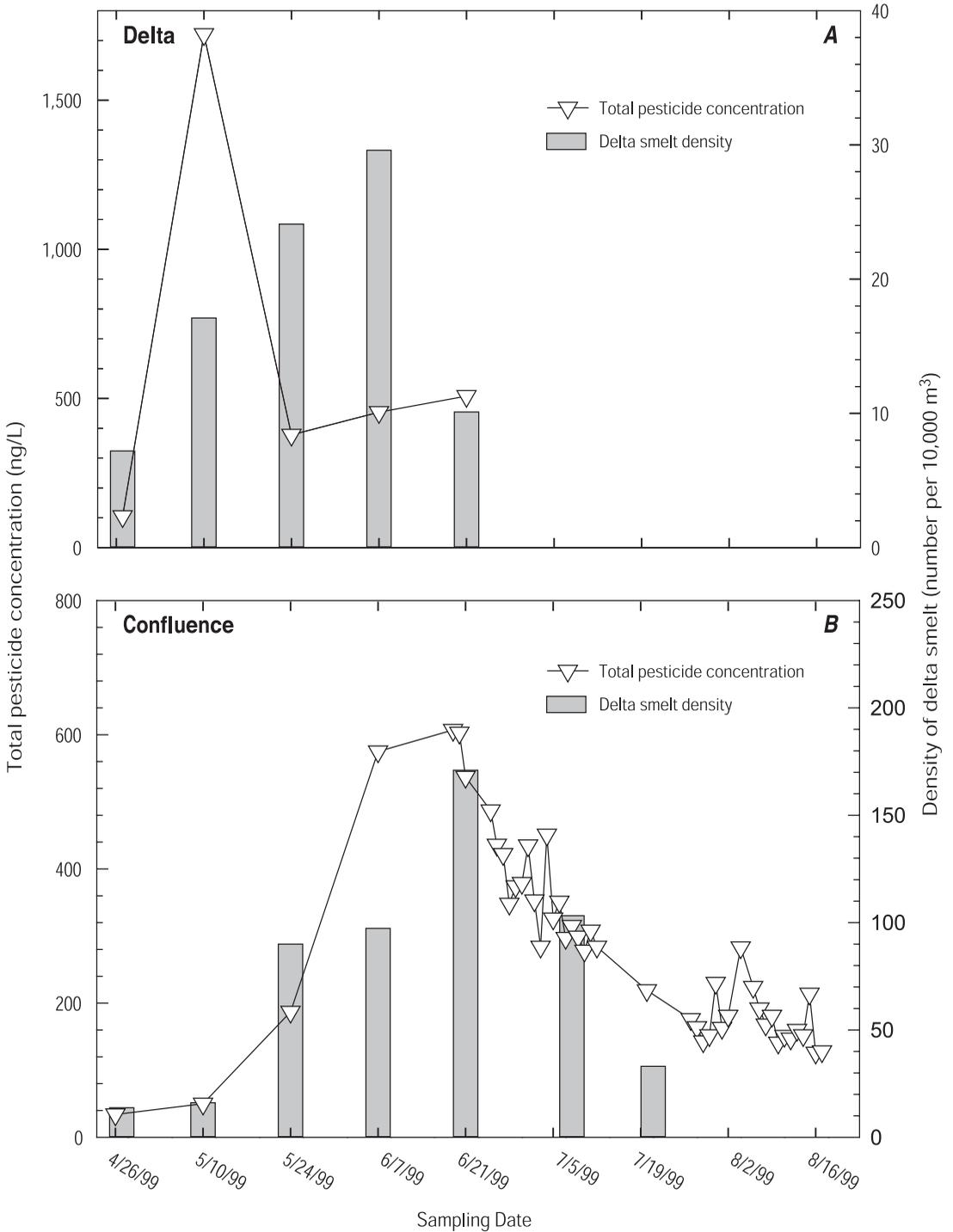


FIGURE 4. Co-occurrence of pesticides and larval and juvenile delta smelt in the delta (A) and at the confluence (B) in 1999.

concentrations at central delta sites were very similar to Cache Slough. This difference in

pesticide distribution between years can be explained by the effect of delta hydrodynam-

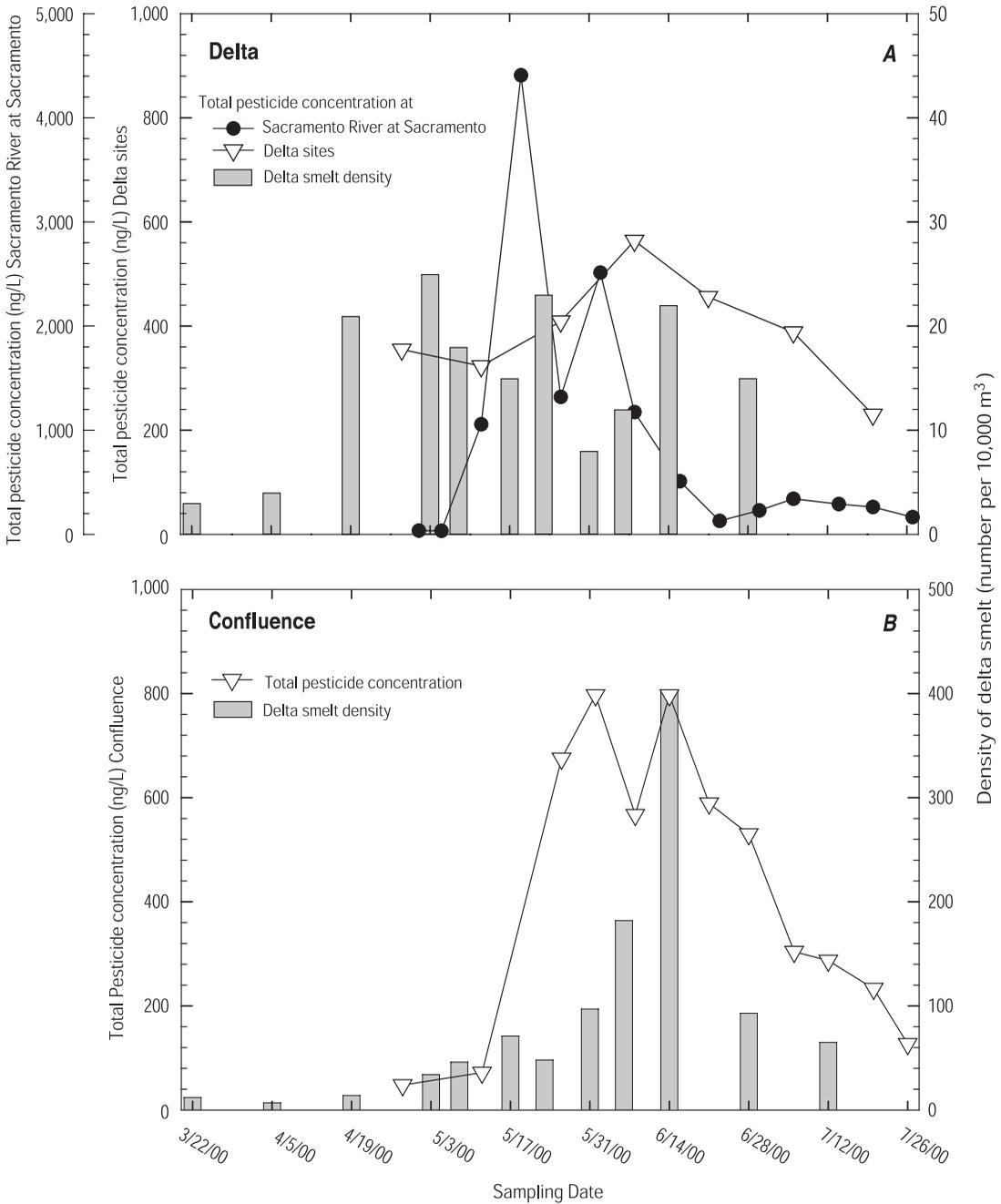


FIGURE 5. Co-occurrence of pesticides and larval and juvenile delta smelt in the delta (A) and at the confluence (B) in 2000.

ics on pesticides originating from the Sacramento River watershed. In 1998, San Joaquin River flow was high, relative to export pumping by the SWP and CVP; therefore, flow from the San Joaquin River was sufficient to sup-

ply export pumps, and the amount of Sacramento River water drawn into the central delta was minimal. Conversely, during spring 1999 and 2000, export pumping rates, at times, equaled or exceeded discharge from the San

Joaquin River. This resulted in Sacramento River water with its associated pesticide load being drawn into the central delta in 1999 and 2000. The high flows in 1998 also shifted the distribution of delta smelt into Suisun Bay; whereas, in 1999 and 2000, delta smelt distribution was more typical (i.e., centered in the delta).

Previous studies (Bennett 1996) have suggested that timing of pesticide pulses is offset from fish abundance and that exposure to pesticides is not important. Bennett (1996) found that maximum densities of striped bass larvae and concentrations of molinate did not occur concurrently; however, he noted that his two data sets were not collected concurrently and suggested that future studies should be designed accordingly. Our results, conducted with concurrent sampling, found that delta smelt were exposed to a complex mixture of dissolved pesticides in both 1999 and 2000. But variability in fish abundance and pesticide concentrations from year to year is evidence that caution is needed when extrapolating and estimating exposure of delta smelt to pesticides.

Estimating potential exposure

Actual exposure of delta smelt to pesticides was not measured because of the impossibility of tracking individual delta smelt; however, potential exposure can be estimated by making assumptions about delta smelt behavior. The time interval from hatching to swim bladder and fin development is several weeks, during which time larvae are thought to remain near the bottom to avoid being washed downstream (Moyle 2002; Mager et al. 2004, this volume). Therefore, it seems reasonable to assume that an individual larval delta smelt spawned in the delta remained in the same general geographic area for this 2–3-week period.

Within a given year, the rise and fall of total pesticide concentrations over time at all delta sites followed a similar pattern. The graph of average pesticide concentrations represents the pattern of concentrations over time throughout the entire delta while underestimating local "hot spots." In 2 of 3 years, peaks in density of delta smelt and pesticide con-

centrations overlapped at the delta sites. These results suggest that large numbers of individual larval delta smelt were potentially exposed to these elevated pesticide concentrations for 2–3 weeks.

A similar analysis can be done for the potential exposure of juvenile delta smelt at the confluence. Juvenile delta smelt are thought to maintain their position in the mixing zone by moving up and down in the water column for weeks to months (Moyle 2002). At the confluence sites, distinct peaks in fish density and total pesticide concentrations coincided in both 1999 and 2000. So it is likely that a large number of juvenile delta smelt were exposed to a complex mixture of pesticides for a period of weeks.

Potential biological effects of pesticides

The maximum concentration for each pesticide was two to four orders of magnitude below 96-h LC50 values for many fish species (Tomlin 1997); therefore, it is unlikely that an individual pesticide caused short-term, acute toxicity to delta smelt. But two other important factors must be considered: (1) delta smelt were likely exposed to multiple pesticides for a minimum of 2–3 weeks, which suggests that chronic effects may be important; and (2) exposure occurred during larval and juvenile life stages, when organisms are particularly sensitive to toxic effects (Heath 1987; Kristensen 1994; Rand 1995).

Chronic exposure to individual and multiple pesticides may hinder growth rate, reproduction, and swimming performance of fish (Rand 1995). No chronic toxicity studies have been conducted with delta smelt. The effects of pesticide exposure have been tested with other larval fish. Heath et al. (1993) found that high concentration of molinate (3,125 mg/L) caused decreased swimming performance of larval striped bass, but low concentration (69 mg/L) did not cause any measurable effects. Although these concentrations are one to three orders of magnitude higher than any values measured in this study, the laboratory exposure time was only 4 d, in contrast to estimated potential exposure times of several weeks in the field.

This study showed that larval and juve-

nile delta smelt were potentially exposed to a complex mixture of pesticides for weeks at a time during both 1999 and 2000. The toxicological effects of this exposure on delta smelt population cannot be evaluated fully until more data are available on lethal and sublethal effects of chronic exposure of delta smelt larvae and juveniles to complex mixtures of pesticides.

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