Aquatic risk assessment of pesticides in surface waters in and adjacent to the Everglades and Biscayne National Parks: II. Probabilistic analyses

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Abstract A screening-level aquatic probabilistic risk assessment was completed to determine the potential risks of organic pesticides found in surface waters of the C-111 freshwater basin (11 sites at the east boundary of the Everglades National Park) and adjacent estuarine tidal zones (two sites in northeast Florida Bay, one site in south Biscayne Bay) in south Florida. It followed the US Environmental Protection Agency (USEPA) ecological risk framework and focused only on the acute and chronic risks of endosulfan and chlorpyrifos individually and jointly with atrazine, metolachlor, and malathion by comparing distributions of surface water exposure concentrations with the distributions of species toxicity data. The highest risk of acute effects was associated with endosulfan exposure to freshwater arthropods at S-178/site C on the C-111 system, followed by endosulfan effects to estuarine arthropods at Joe Bay in northeast Florida Bay. The highest risk of acute effects from joint toxicity of pesticides was to estuarine arthropods in Joe Bay followed by freshwater arthropods in S-178/site C. For fish, the highest acute risk was for endosulfan at S-178/site C. There was low potential for acute risk of endosulfan to fish at estuarine sites. Joint probability curves indicated that the majority of potential risks to arthropods and fish were due to endosulfan concentrations and not to chlorpyrifos, at S-178/site C. In addition, the highest risk of acute effects for saltwater organisms was in Joe Bay, which receives water from the C-111. The potential risk of chronic effects from pesticide exposures was minimal at fresh- and saltwater sites except at S-178/site C, where endosulfan concentrations showed the highest exceedence of species toxicity values. In general, potential risks were higher in February than June.

Introduction

A hazard assessment (Tier 1) of five pesticides (atrazine, metolachlor, malathion, endosulfan, and chlorpyrifos) in surface waters of the C-111 freshwater basin (east boundary of the Everglades National Park), northeast Florida Bay and south Biscayne Bay, south Florida, indicated that measured concentrations of chlorpyrifos and endosulfan present potential hazard to aquatic organisms and were therefore identified as chemicals of potential ecological concern (COPECs) (Carriger and Rand, this issue). Tier 1 included problem formulation of a screening-level aquatic probabilistic risk assessment for the COPECs under the US Environmental Protection Agency (USEPA) ecological risk assessment (ERA) framework (1998), which included an overview of the physical/chemical, environmental fate, and aquatic toxicity of chlorpyrifos and endosulfan, the ecosystems at risk, endpoints (measurement and assessment) to be evaluated, and a conceptual model for risk assessment.

This paper presents a screening-level probabilistic aquatic risk assessment (Tier 2) for endosulfan and chlorpyrifos, individually and jointly with atrazine, malathion, and metolachlor, in surface waters of the C-111 freshwater

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basin, northeast Florida Bay and south Biscayne Bay. Tier 2 includes risk analyses and risk characterization of the USEPA framework and characterizes risk by comparing distributions of surface water exposure concentrations of these pesticides in these systems with species effects data from laboratory toxicity studies (Hall and Gardinali 2004). This is the first probabilistic risk analyses of these pesticides in south Florida aquatic systems.

Methods

A general description of the ecosystems of interest and the aquatic risk assessment methodology have been previously described (Carriger and Rand, this issue). Risk analyses and risk characterization are discussed below.

Risk analysis

Risk analysis contains two components: exposure and effects analysis.

Exposure: For exposure, data on the concentrations of the five pesticides in the C-111 system, south Biscayne Bay, and northeast Florida Bay were obtained by using actual measured concentrations (AMCs) in surface water from monitoring studies from 1999 to 2000 (sampling sites shown in Fig. 1; Carriger and Rand this issue). The monitoring sources included South Florida Water Management District (SFWMD), a state agency, the US Geological Survey (USGS), and the National Oceanic and Atmospheric Administration (NOAA). As discussed in Carriger and Rand (this issue) monitoring data were available for 11 freshwater sites (S-175, S-176, S-332, site A, S-177/site B, S-178/site C, S-18C/site E, site W1, site W2, site E1, site E2) on or near the C-111 and three estuarine sites (Joe Bay and Highway Creek in northeast Florida Bay and Card Sound in south Biscayne Bay). Probability distributions of exposure data for the pesticides were developed based on the AMCs to assess spatial and temporal trends.

For analytical exposure concentrations that were reported on the same day, at the same site, and by the same monitoring agency, the highest concentration was used. Some of the monitoring data from NOAA consisted of 24- or 48-h composite surface water samples. If a composite sample spanned ≥24 h and fell on the same day as a discrete sample, concentrations reported from the composite were placed on the day that did not have a discrete or composite sample available. If each of the days when a composite sample was taken fell on the day that another sample was taken, analytical concentrations were chosen in the same manner as when two discrete samples fell on the same day. Monitoring data were then assessed on an annual basis (i.e., for the 2 years, 1999 and 2000), with all sites
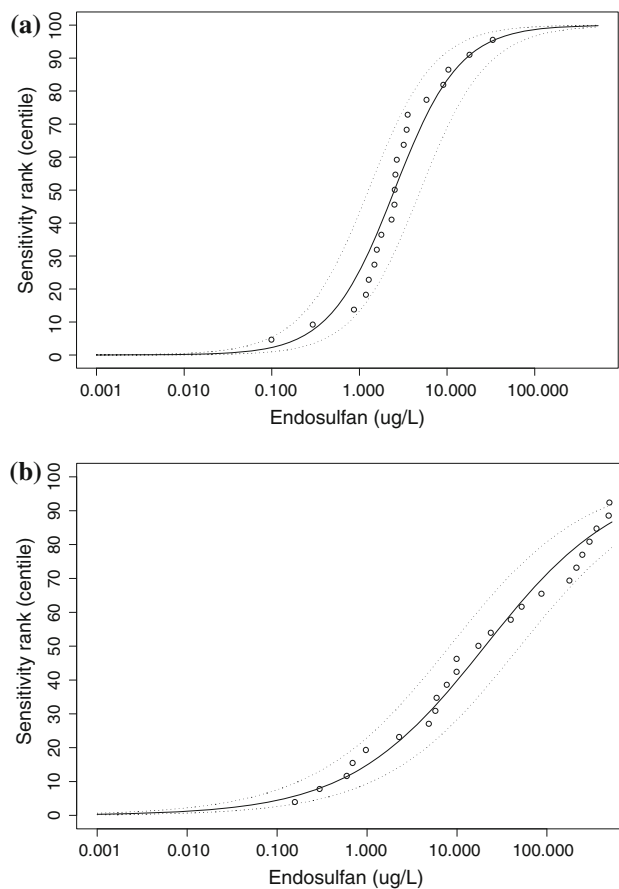
Fig. 1 Log-logistic distribution (and 95% prediction band) of endosulfan acute toxicity values for freshwater fishes (**a**) and for freshwater arthropods (**b**)

combined for each year for freshwater and saltwater, and also on a site-specific basis, with all years combined for each pesticide. In addition, to compare seasonal differences in pesticide exposure, monitoring data were combined separately for February (dry season) and June (wet season).

Measured concentration values were ranked to create cumulative distributions, and non-detects were assigned a value of zero (Giddings et al. 2000a). Concentrations were ranked from smallest to highest and assigned a centile ranking (j) at each site using the equation $j \times 100/(n + 1)$, where j is the rank and n is the total number of observations (including non-detects) (Hall and Gardinali 2004). When four concentration values were reported above the minimum detection limit (MDL), a log-logistic distribution for exposure data was created for that pesticide at a site (Hall et al. 2000).

In Tier 2, the 90th centile (exceedence of a value only 10% of the time) and 50th centile concentrations were calculated as "exposure benchmarks" (Solomon et al. 1996) for acute and chronic risks, respectively, and the corresponding concentrations were compared for each year and sampling site. The 90th centile assumes that 90% of

any samples taken over the specified duration will be below that benchmark value if it is derived from an unbiased distribution of all possible sample times (Giddings et al. 2000a). The reported centile estimates were determined using the nonparametric analyses below.

In most cases, the exposure data did not fit the log-logistic model well. Therefore a nonparametric method was utilized to calculate the centile estimates. For example, the nonparametric method for estimating the 90th centile is as follows:

Let $X_1, \ldots, X_N$ be a random sample and $X_{(1)}, X_{(2)}, \ldots, X_{(N)}$ be order statistics.

Let $np = 0.9(n + 1)$. Then, the 90th centile $= X_{(n1)} + (np - n_1)(X_{(n2)} - X_{(n1)})$, where n_1 is the integer part of np and $n_2 = n_1 + 1$. When np is greater than N, we let the 90th centile equal $X_{(N)}$.

Note that for α-endosulfan, β-endosulfan and endosulfan sulfate exposure concentrations were summed to give a value for total endosulfan (WHO 1984; You et al. 2004).

Effects: For effects, acute (LC_{50}/EC_{50}) and chronic (NOEC) laboratory toxicity data for water exposures from atrazine, chlorpyrifos, endosulfan, malathion, and metolachlor to freshwater and saltwater species were used to develop species sensitivity distribution curves (SSDs) for each pesticide. Separate acute SSDs were developed for the following taxonomic groups: aquatic plants/phytoplankton, arthropods, and fish to characterize susceptibility at the different sites. For chronic SSDs, all organism tests for a pesticide were combined because of the limited chronic toxicity data. All acute toxicity endpoints were obtained from the USEPA AQUIRE database and One Liners Database (Montague 2002). Additional data for atrazine effects, particularly on saltwater plants and algae, were taken from Giddings et al. (2000b). Due to a paucity of chronic information from AQUIRE for each pesticide, values were taken from published papers and various government monitoring reports as well.

Effects data were screened for usage in SSDs. Only toxicity endpoints that could be clearly related to changes in population structure such as growth, reproduction, and survival were used in the SSDs, and a species could only be represented once in each distribution. In order to develop an SSD with sufficient toxicity data, toxicity tests that reported nominal and/or measured concentrations were included. When a pesticide formulation in a toxicity test contained less than 80% of the active ingredient the data was excluded from the SSD (Crommentuijn et al. 1997). Endpoints with a greater-than or less-than value were not included in an SSD unless a > value exceeded the water solubility. When multiple toxicity values were available for a single-species endpoint, the geometric mean of that data was taken (Solomon et al. 2001). When data for a species with different responses (e.g., growth, biomass, survival,

LC_{50}, EC_{50}, etc.) were available, these were combined in the same manner. If life-stage information was available for a species, the most sensitive life stage, based upon the geometric means for concentration, was chosen as an endpoint for effects analysis.

For fish and arthropods acute toxicity tests with a duration of 24–96 h were considered and with plant/phytoplankton species toxicity test durations of 144–168 h were used. When plant/phytoplankton tests were not available within a 144–168 h duration, acute toxicity tests from 24 h to 14 days were used. For eelgrass (*Zostera marina*) we used several 21-day LC_{50} tests with atrazine. Plant/phytoplankton toxicity data (e.g., chlorpyrifos-freshwater species; malathion-fresh- and saltwater species; endosulfan-fresh- and saltwater species) and saltwater toxicity data were limited for several pesticides and SSDs could not be developed.

All effects (toxicity) data used for SSDs were assumed to fit a log-logistic distribution and graphical output was produced through the software program S-Plus (1999). To calculate ranks, all toxicity data were ordered by concentration and plotting positions were calculated as $(j \times 100)/(n + 1)$, where j is the rank and n is the total number of observations (Warren-Hicks et al. 2002). Values greater than the water solubility for each compound were included in the calculation of rank (n) but were not plotted (ECOFRAM 1999). Toxicity endpoints with a less than value were excluded (ECOFRAM 1999) and those with a greater-than value below the compound's solubility were also excluded and not used in the calculation of rank. For a distribution to be considered for analysis, at least four suitable species' endpoints had to be available (Aldenberg and Slob 1993).

Log-transformed concentration endpoints were then plotted against the cumulative probabilities for each species and the 10th centile for each SSD was determined. The 10th centile of the SSD has been chosen as an effects benchmark or threshold value for effects on communities of aquatic species (SETAC 1994).

The linear regression form of the model used for calculating our results from the SSDs is as follows:

$$\text{logit}(p) = \log\left(\frac{p}{1 - p}\right) = \alpha^* + \beta^* \log_{10}(x),$$

where x is the geometric mean of a species effect concentration (i.e., LC_{50}, EC_{50}) and p is the probability of an effect at a specific concentration. α and β are scale parameters derived from the sample mean of the log toxicity data and from the standard deviation of the log toxicity data multiplied by 0.55 (or the square root of 3 divided by π), respectively. For compounds with the same toxic mode of action, the β parameter, or the SSD's slope, has been found to be similar for each distribution with a

community of species tested, with a high enough n value, that share that mode of action (de Zwart 2002). The parameters from each distribution were used to estimate the 10th centiles of the SSDs and for a multiple chemical assessment to consider the potential effects of joint exposures on organisms in C-111 and estuarine sites.

Risk characterization

Individual pesticides: Risk was assessed by comparing the overlap of the pesticide distributions of the AMCs for sites and the SSDs. For acute risks to arthropods, fish, and phytoplankton/aquatic plants the 90th centile of the AMCs for exposure was compared to the 10th centile of the acute SSD (SETAC 1994). For chronic risks to aquatic organisms, the 50th centile of the AMCs for exposure was compared to the 10th centile of the NOEC chronic SSD for all species combined (Traas et al. 2002). When a centile from the exposure data was applied to a SSD, a potentially affected fraction (PAF) of species number was derived for a single pesticide. Exceedences above the 10th centile of a SSD were noted for acute and chronic risk. The PAF was calculated from the SSD at a concentration (Klepper and Van de Meent 1997) that allows one to determine that "a certain fraction of species is expected to be (potentially) affected above its acute or chronic effect level at a given environmental concentration" (Traas et al. 2002). The PAF approach allows us to assess pesticide risk both individually and in joint action (multiple substance PAF = msPAF) (Traas et al. 2002).

For individual pesticides, a PAF value was calculated in the equation for log-logistic toxicity data (Traas et al. 2002):

$$\text{PAF}_i(x) = \frac{1}{1 + e^{-(x-\alpha)/\beta}}$$

where α is the mean of log toxicity data and β is equal to $(\sigma \cdot \sqrt{3})/\pi$ with σ the standard deviation and x the log of the exposure concentration. For acute effects, x was determined to be the 90th centile of the exposure distribution for each pesticide. For chronic effects, x was the 50th centile from the exposure data.

Multiple pesticides: Methodologies used to estimate acute and chronic risks to organisms from being exposed to the joint action of pesticides were adapted from the multiple substance potentially affected fraction (msPAF) assessment approach by Traas et al. (2002). This approach is based on the toxic mode of action (TMoA) of each pesticide and toxicological mixture theories of concentration and response addition. For risk characterization of multiple chemical exposures, individual PAF values of pesticides were combined and used to calculate the risk.

Concentration addition (CA) is a concept utilized to describe the additive effects of mixtures of chemicals with the same toxic mode of action (TMoA) (Plackett and Hewlett 1952). In concentration addition, the concentrations for each pesticide at a site are expressed in hazard units (HU), representing the relative potency of the actual measured environmental concentration to an SSD:

$$\text{HU} = \frac{C_{\text{ENV}}}{10^\alpha}$$

where α represents the mean of log toxicity data and C_{ENV} is an exposure concentration of concern (e.g., 90th centile). The sum of HUs from a centile of the relevant exposure distribution was substituted into the equation below. For the msPAF values for concentration addition, HUs were summed for all pesticides with the same TMoA. The slopes of the SSDs, or β, were averaged for the pesticides with the same TMoA and the values were substituted into the following equation:

$$\text{PAF}_{\text{TMoA}} = \frac{1}{1 + e^{-\log(\Sigma \text{HU}_{\text{TMoA}})/\beta_{\text{TMoA}}}}$$

Concentration addition was used on the two organophosphate (OP) insecticides, chlorpyrifos and malathion, for arthropods and fish. Chlorpyrifos and malathion act by inhibiting the enzyme acetylcholinesterase (AChE) in these organisms. Plants and algae do not have the receptor site for chlorpyrifos and malathion and therefore response addition (RA) was used. Chlorpyrifos was the only insecticide that had enough species available ($n \geq 4$) to develop an SSD for plants and algae.

Atrazine (TMoA: inhibition of PS II in plants) and metolachlor (TMoA: inhibition of protein synthesis in plants) are herbicides from two different chemical classes with different modes of toxic action, and their joint action was modeled using RA. Chlorpyrifos and endosulfan also act by two different modes of action and therefore RA was used for these insecticides as well. RA was also used when three or more pesticides were present with different modes of action. RA assumes different modes of toxic action. Response addition theory includes the addition of a correlation coefficient, r, that accounts for the covariation of the sensitivities of organisms to the chemicals in a mixture (Könemann 1981). For SSD purposes, r refers to species sensitivities rather than individuals in a population (Traas et al. 2002). For practical purposes, r was set at 0, thus assuming no correlation. When using SSDs to estimate risks from multiple substance exposures, CA and RA models require different correlation approaches between pesticides (Traas et al. 2002). In CA, effects from two chemicals in a mixture are combined based on the toxic magnitude of each through HU scaling. While in RA, effects from two chemicals (A and B) are combined corresponding to the "probability of two nonexcluding processes" (Hewlett and Plackett, 1979, as cited in Traas et al. 2002).

The msPAF for RA was calculated with the following equation:

$$PAF_{RA} = 1 - \prod_i (1 - PAF_i)$$

for $i = 1$ to n pesticides, and PAF_{RA} calculated for all pesticides in response addition with $r = 0$.

When two pesticides with the same TMoA were detected four or more times at a similar site between 1999 and 2000 or during the same year, the distribution for the two pesticides were combined based upon CA mathematical procedures (Traas et al. 2002). For each site and each year, CA was only applied to chlorpyrifos and malathion. Concentration addition was only used if chlorpyrifos and malathion were both detected at a site. If chlorpyrifos was found by itself, an individual PAF value was determined for it alone. The PAF value for chlorpyrifos would then be combined with the PAF values for other pesticides found at the site using RA. When chlorpyrifos and malathion were both detected at a site, a msPAF value was estimated for them using CA. Additional pesticides detected at the site were then combined with the msPAF value for chlorpyrifos and malathion using response addition to obtain an overall msPAF.

Joint probability curves: The last step in the PRA used joint probability curves (JPCs, or exceedence profiles) for endosulfan and chlorpyrifos at sites which displayed the highest PAF and msPAF values. JPCs were constructed by applying the various centiles of the exposure distribution (i.e., 99th, 95th, 90th, 75th, 50th) for endosulfan and chlorpyrifos to log-logistically derived effects distributions (SSDs) for arthropods and fish following the PAF and msPAF methodology of Traas et al. (2002). The JPCs characterize "the relationship between magnitude of effect and the probability of occurrence for that effect" (ECOFRAM 1999). JPCs were used to determine the proportion of toxicity values in the SSD that are exceeded by ranges of exposure concentrations. In addition, JPCs were created with exposure concentrations from February (dry season) and June (wet season) at sites by comparing them to acute LC_{50}/EC_{50} data for fish and arthropod species to assess seasonal differences. We selected the 10% exceedence probability (10% of the exposure concentrations) as the reference to compare different JPCs. For example, a JPC at site X for endosulfan may show that there is a 10% probability that endosulfan exposures will affect 5% of the fish species (based on acute fish toxicity values) but at site Z the 10% probability of endosulfan exposures may affect 50% of the fish species. This approach provides a means of comparing relative potential risk at sites when the toxicity data base and exposure information are adequate.

Results and discussion

Risk analysis

Exposure: The 90th centiles in 1999 and 2000 combined for freshwater sites are presented in Table 1. For atrazine the 90th centiles were similar in 1999 and 2000. The highest 90th centiles occurred at S-18C/site E (0.28 µg/l), S-177/site B (0.15 µg/l), site A (0.15 µg/l), and site W1

Table 1 Freshwater sites for 1999 and 2000: 90th centiles, combined for all pesticides

Site	Pesticide	90th centile concentration (µg/l)
S-175	Atrazine	5.01E-02
S-176	Atrazine	7.77E-02
S-177/site B	Atrazine	1.54E-01
	Metolachlor	2.02E-02
	Chlorpyrifos	8.76E-03
	Malathion	2.03E-02
	Endosulfan	2.86E-02
S-178/site C	Atrazine	4.28E-02
	Metolachlor	1.66E-02
	Chlorpyrifos	3.37E-03
	Endosulfan	2.10E-01
S-18c/site E	Atrazine	2.83E-01
	Metolachlor	1.01E-02
	Chlorpyrifos	2.53E-03
	Endosulfan	1.95E-02
S-332	Atrazine	6.22E-02
Site A	Atrazine	1.51E-01
	Metolachlor	2.96E-02
	Chlorpyrifos	6.08E-03
	Endosulfan	7.61E-03
Site E1	Atrazine	2.12E-02
	Metolachlor	7.39E-03
	Chlorpyrifos	3.07E-03
	Endosulfan	1.65E-02
Site E2	Atrazine	2.01E-02
	Metolachlor	6.94E-03
	Chlorpyrifos	2.66E-03
	Endosulfan	1.23E-02
Site W1	Atrazine	1.68E-01
	Metolachlor	9.11E-03
	Chlorpyrifos	2.48E-03
	Endosulfan	1.51E-02
Site W2	Atrazine	3.90E-02
	Metolachlor	8.10E-03
	Chlorpyrifos	2.12E-03
	Endosulfan	1.02E-02

(0.17 µg/l). These values are significantly lower than the 90th centiles reported for atrazine in lakes/reservoirs in Midwestern USA (2.56–7.2 µg/l) (Solomon et al. 1996). The 90th centiles for metolachlor decreased from 1999 to 2000. The highest 90th centiles for metolachlor occurred at site A (0.03 µg/l), S-177/site B (0.02 µg/l), and S-178/site C (0.02 µg/l). The 90th centiles for chlorpyrifos also decreased from 1999 to 2000. The highest 90th centiles for chlorpyrifos were at S-177/site B (0.009 µg/l) and site A (0.006 µg/l). Chlorpyrifos monitoring data by site from the San Joaquin River (California) watershed showed that 90th centile values for chlorpyrifos ranged from 0.00009 to 0.337 µg/l (Hall and Anderson 2003). For endosulfan the 90th centiles increased from 1999 to 2000. The highest 90th centiles for endosulfan were located at S-178/site C (0.2 µg/l) and S-177/site B (0.03 µg/l) followed by S-18C/site E (0.02 µg/l) and site E1 (0.02 µg/l). The highest concentration of endosulfan (1.345 µg/l) in the C-111 was found at S-178/site C. Scott et al. (1990) found endosulfan concentrations up to 0.685 µg/l in surface water from the North Edisto River in South Carolina, and residues were attributed to surface runoff from agricultural fields.

At S-177/site B and S-178/site C, endosulfan and chlorpyrifos had higher 90th centile exposure concentrations in February, the height of the south Florida dry season, compared to June, the beginning of the wet season. Site S-18C/site E, downstream from S-177 and S-178 on the C-111, had lower 90th centile exposure concentrations for chlorpyrifos and endosulfan during June. For malathion in 1999 and 2000 there was insufficient data to calculate a 90th centile. However, at S-177/site B there was sufficient data ($n = 6$) to calculate a 90th centile (0.02 µg/l) for malathion.

The 90th centiles in 1999 and 2000 combined for saltwater sites are presented in Table 2. For atrazine, the 90th centiles slightly decreased from 1999 to 2000. In general, the 90th centiles for Card Sound, Highway Creek, and Joe Bay were similar (~ 0.02 µg/l). For chlorpyrifos, the 90th

centiles also slightly decreased from 1999 to 2000. Joe Bay had the highest number of chlorpyrifos detections and 90th centile estimates were similar in Joe Bay (0.0032 µg/l) and Highway Creek (0.0031 µg/l). The 90th centiles for endosulfan increased from 1999 to 2000. Endosulfan was the most frequently detected pesticide in Highway Creek. However, the highest 90th centile for endosulfan was at Joe Bay (0.01 µg/l). A 90th centile for metolachlor could not be calculated for Card Sound and Highway Creek because of a low percentage (1 out of 43, or 3.8%) of detections. Although a 90th centile (0.0029 µg/l) was calculated for metolachlor in Joe Bay it was based on 2 detections out of 24 samples. For malathion there was insufficient data to calculate a 90th centile at any saltwater site.

Effects: A summary of the acute and chronic toxicity data including 10th centiles for fresh- and saltwater species for all pesticides is presented in Table 3. When the term "all species" is used for acute tests it indicates that the results of all toxicity tests for fresh- and saltwater species were grouped together into one SSD to obtain a 10th centile for effects for a pesticide. The latter term is also used for chronic 10th centile SSDs of NOECs because all chronic tests for all species were combined into one SSD.

Acute toxicity: The acute freshwater 10th centile for chlorpyrifos for all species ($n = 56$) combined was 0.20 µg/l. *Ceriodaphnia dubia* was the most sensitive species with a geometric mean EC_{50} of 0.057 µg/l, and a rotifer *Brachionus calyciflorus* was the least sensitive species with a geometric mean EC_{50} of 3,713 µg/l. The acute 10th centiles for freshwater arthropods ($n = 37$) and fish ($n = 19$) were 0.11 and 2.8 µg/l, respectively. The acute saltwater 10th centile for chlorpyrifos for all species ($n = 26$) combined was 0.11 µg/l. *Americamysis bahia* was the most sensitive species with a geometric mean EC_{50} of 0.045 µg/l while *Mytilus galloprovincialis* was the least sensitive species with an LC50 of 22,500 µg/l. The acute 10th centiles for saltwater arthropods ($n = 8$) and fish ($n = 13$) were 0.026 and 0.32 µg/l, respectively. The acute 10th centile for saltwater species was lower than the acute 10th centile for freshwater species and saltwater arthropods were the most sensitive group to chlorpyrifos.

The acute 10th centile for endosulfan for all freshwater species ($n = 59$) combined was 0.37 µg/l. Acute freshwater toxicity values ranged from 0.1 µg/l in *Carassius auratus* to 39,892 µg/l in *Melanopsis dufouri*. The acute 10th centiles for freshwater arthropods ($n = 24$) and fish ($n = 21$) were 0.45 and 0.38 µg/l, respectively. The acute 10th centile for all saltwater species ($n = 27$) combined was 0.056 µg/l. Acute endosulfan toxicity values ranged from 0.04 µg/l for *Penaeus duorarum* (now called *Farfantepenaeus duorarum*) to 6,432 µg/l for *Brachionus plicatilis*. The slope of the endosulfan SSD for fish is steeper than that for arthropods (Fig. 1).

Table 2 Saltwater sites for 1999 and 2000: 90th centiles, combined for all pesticides

Site	Pesticide	90th centile concentration (µg/l)
Card Sound	Atrazine	2.00E-02
	Chlorpyrifos	2.63E-03
	Endosulfan	3.26E-03
Highway Creek	Atrazine	2.40E-02
	Chlorpyrifos	3.10E-03
	Endosulfan	6.56E-03
Joe Bay	Atrazine	2.80E-02
	Metolachlor	2.91E-03
	Chlorpyrifos	3.19E-03
	Endosulfan	1.01E-02

Table 3 Statistics for acute and chronic freshwater and saltwater toxicity data for all pesticides by test duration and trophic group

Test conditions	Trophic group	10th centile (µg/l)	Number of tests below water solubility	Total number of tests	α	β	α SE	β SE	Root MSE
Acute freshwater									
Atrazine	All species[a]	30	72	82	−4.1248	1.3041	0.1126	0.0371	0.3225
	Fish	1,700	13	17	−8.3427	1.9096	1.2927	0.3164	0.5653
	Arthropods	480	13	13	−8.7336	2.4403	0.8052	0.2224	0.4404
	Plants/algae	18	40	40	−5.2216	2.4069	0.1893	0.0836	0.3454
	Amphibians	4.0	4	5	−2.6212	0.7068	0.4350	0.1299	0.3031
Chlorpyrifos	All species	0.20	56	57	−1.1971	1.4409	0.0403	0.0303	0.2446
	Fish	2.8	19	19	−3.0119	1.8412	0.1732	0.0954	0.3278
	Arthropods	0.11	37	37	−0.5750	1.6838	0.0466	0.0468	0.2663
Endosulfan	All species	0.37	47	59	−1.6543	1.2551	0.0794	0.0625	0.4056
	Fish	0.38	21	21	−1.0765	2.6839	0.1001	0.1479	0.3695
	Arthropods	0.45	24	25	−1.7420	1.3272	0.0781	0.0486	0.2508
Malathion	All species	4.1	100	102	−2.8849	1.1182	0.0548	0.0190	0.2701
	Fish	36	35	35	−5.0919	1.8597	0.2723	0.0953	0.4622
	Arthropods	0.52	43	43	−1.8817	1.1240	0.0967	0.0444	0.4060
Metolachlor	All species	43	20	20	−4.6918	1.5282	0.1982	0.0616	0.2656
	Fish	3,200	6	6	−27.9893	7.3593	4.4333	1.1647	0.4338
	Plants/algae	11	12	12	−3.7132	1.4547	0.3293	0.1214	0.3859
Acute saltwater									
Atrazine	All species	19	25	28	−4.0277	1.4289	0.2532	0.0926	0.4037
	Fish	1,800	15	19	−9.2099	2.1517	1.1683	0.2883	0.5246
	Arthropods	130	9	11	−5.3998	1.5212	0.4351	0.1308	0.2671
	Plants/algae	19	14	14	−6.2237	3.1289	0.4305	0.2113	0.3489
Chlorpyrifos	All species	0.11	26	28	−1.0903	1.1760	0.0863	0.0644	0.3685
	Fish	0.32	13	13	−1.3026	1.8125	0.2459	0.2442	0.6211
	Arthropods	0.026	8	8	0.4629	1.6770	0.1461	0.1874	0.3863
	Plants/algae	82	4	4	−12.5184	5.3930	2.7303	1.1727	0.4245
Endosulfan	All species	0.056	27	32	−1.0082	0.9526	0.0844	0.0612	0.3976
	Fish	0.077	9	9	0.8471	2.7368	0.1917	0.3507	0.4741
	Arthropods	0.038	13	14	−0.7865	0.9921	0.0906	0.0660	0.2951
Malathion	All species	2.1	29	29	−2.5958	1.2448	0.1200	0.0496	0.3281
	Fish	17	13	13	−5.0552	2.3349	0.3792	0.1691	0.3556
	Arthropods	0.76	12	12	−2.0024	1.6157	0.1699	0.1137	0.3285
Metolachlor	All species	17	4	4	−3.6113	1.1443	0.9988	0.3060	0.5109
	Fish	3,500	7	7	−29.4459	7.7014	4.8825	1.2760	0.5037
	Plants/algae	10	13	13	−3.7134	1.4893	0.3364	0.1268	0.4137
Chronic									
Atrazine	All species[a]	7.2	28	28	−3.8119	1.8814	0.0929	0.0425	0.0341
Chlorpyrifos	All species	0.018	12	12	0.7828	1.7088	0.1169	0.1285	0.1224
Endosulfan	All species	0.12	10	10	−1.1943	1.1221	0.1819	0.1167	0.1765
Malathion	All species	0.024	15	15	−0.8085	0.8546	0.1075	0.0570	0.1296
Metolachlor	All species	2.4	7	7	−2.6403	1.1569	0.3225	0.1288	0.1227

[a] "All species" indicates that the toxicity values from all tests were included in the distributions for effects

Chlorpyrifos was the only insecticide with sufficient toxicity data for plants/algae where the minimum of four tests were available for developing a SSD for the 10th centile for saltwater. The acute saltwater 10th centile value for four tests was 82 µg/l, which was the highest acute 10th centile for any of the five pesticides. Based on low surface

water exposures, chlorpyrifos would not present any potential risk to algae/plants.

Chronic toxicity: The 10th centiles of the log-logistic distribution of NOECs are presented in Table 3. The lowest chronic 10th centile value (0.0180 µg/l) was for chlorpyrifos, followed by endosulfan (0.1227 µg/l).

Risk characterization

Acute individual pesticide assessment (PAF): For sites between 1999 and 2000 there were no acute PAF values for individual pesticides that exceeded 10% for any trophic group. However, several PAF values for pesticides were higher than other pesticides at certain sites. For example, at S-178/site C the potential risk of endosulfan was higher than at any other freshwater site (Table 4). At this site, PAF values for fish and arthropods from surface water concentrations of endosulfan were 5.2% and 6.7%, respectively. PAF values for all other pesticides and trophic groups at freshwater sites were ≤2.2%. With the exception of S-178/site C the PAF values for endosulfan in saltwater sites were the highest (3.7–5.9%) for arthropods (Table 5). The next highest PAF values were for chlorpyrifos at Joe Bay, Highway Creek, and Card Sound with a range of 2.1–2.4%.

Acute multiple pesticide assessment (msPAF): For sites between 1999 and 2000, there were no acute msPAF values for multiple pesticides that exceeded 10% for any trophic group. However, in 1999 and 2000 for arthropods in saltwater for combined sites the msPAF values for joint action of endosulfan and chlorpyrifos were 6.1% (in 1999) and 7.9% (in 2000). In both years the potential risk from joint action of both insecticides was due mainly to endosulfan. At individual saltwater sites the highest msPAF value (8.2%) was for endosulfan/chlorpyrifos at Joe Bay followed by Highway Creek (7.2%) and Card Sound (5.7%) (Table 5).

In freshwater the msPAF values for arthropods for 1999 and 2000 were 2.3% and 3.4%, respectively. At individual freshwater sites the highest msPAF for joint action was for endosulfan and chlorpyrifos and arthropods at S-178/site C (7.5%) followed by S-177/site B (6.1%) (Table 4).

The msPAF values for joint action of endosulfan and chlorpyrifos for fish for all sites combined in 1999 and 2000 in fresh- and saltwater never exceeded 1.2%. However, at individual freshwater sites the msPAF value for the joint action of endosulfan and chlorpyrifos to fish was 5.3% at S-178/site C (Table 4).

The msPAF values for the joint action of atrazine and metolachlor was ≤0.2% for 1999 and 2000. Malathion surface water exposure monitoring data were limited and therefore it only contributed to calculation for potential risk in joint action at S-177/site B.

Chronic individual (PAF) and multiple (msPAF) pesticide assessment: For fresh- and saltwater sites combined in 1999 and 2000 the chronic individual PAF values were ≤3.1% for all pesticides except endosulfan at S-178/site C, where the chronic PAF value was 6.4% (Table 6). Metolachlor chronic PAF values were only estimated at site A (0.4%) and S-177/site B (0.6%) because there was insufficient exposure data at other sites. Although atrazine was the most frequently detected pesticide, chronic PAF values in freshwater were ≤0.1% at all 11 freshwater sites and three estuarine sites. There was insufficient exposure data to estimate a chronic PAF value for malathion.

The chronic msPAF values for fresh- and saltwater sites in 1999 and 2000 were generally low (≤3.0%) for the joint action of multiple pesticides. However, at S-178/site C the chronic msPAF value was 7.0% (Table 6). At S-178/site C the magnitude of the msPAF value was a result of the joint action of predominately endosulfan followed by chlorpyrifos.

Joint probability curves: The freshwater sites S-178/site C and S-177/site B and the saltwater sites Joe Bay and Highway Creek were considered areas of concern based on the acute and chronic PAF/msPAF results for endosulfan and chlorpyrifos. Therefore, single and multiple substance JPCs were developed for these sites. Malathion was also included on the JPC distributions for fish and arthropods at S-177/site B since it appeared to contribute to the acute msPAF.

A JPC was also created for S-18C/site E since it represents downstream conditions from S-178/site C and S-177/site B, sites adjacent to agriculture. S-178/site C is on the C-111E which feeds the C-111 canal (Carriger and Rand, this issue). It is the only site in all of the monitoring programs that has no exposed headwaters and directly drains a farm area. S-178 is rarely open, therefore the waters are confined, which encourages accumulation of pesticide residues. S-177 is on the C-111 canal and has regular flushing. A JPC was also created for Card Sound for comparison purposes to Highway Creek and Joe Bay. NOAA selected Card Sound as a reference saltwater site in its monitoring program since it was outside the influence of C-111 discharges.

Figures 2–4 present the JPCs for S-177/site B, S-178/site C, and S-18C/Site E, respectively. Potential risks of endosulfan and chlorpyrifos to arthropods were highest at S-178/site C except for chlorpyrifos to arthropods, for which potential risks were higher at S-177/site B. In the multiple pesticide JPC for arthropods at S-178/site C endosulfan was contributing the largest portion of potential risk. The lowest potential risks of chlorpyrifos and endosulfan to arthropods and fish were at S-18C/site E. For fish, S-178/site C has the highest potential risk for endosulfan.

Table 4 Freshwater acute PAFs (%) and msPAFs (%)

Site	Compound	Phytoplankton/plants	Fish	Arthropods	90th centile (µg/l)
S-175	Atrazine	0.0	0.0	0.0	5.01E-02
S-176	Atrazine	0.0	0.0	0.0	7.70E-02
S-177/site B	Atrazine	0.1	0.0	0.0	1.54E-01
	Metolachlor	0.2	0.0		2.02E-02
	Chlorpyrifos		0.1	1.7	8.76E-03
	Malathion		0.0	2.2	2.03E-02
	Endosulfan		0.5	2.2	2.86E-02
	msPAF (CA)		0.1	4.0	
	msPAF (RA)	0.3	0.7	6.1	
S-178/site C	Atrazine	0.0	0.0	0.0	4.28E-02
	Metolachlor	0.2	0.0		1.66E-02
	Chlorpyrifos		0.1	0.9	3.37E-03
	Endosulfan		5.2	6.7	2.10E-01
	msPAF (RA)	0.2	5.3	7.5	
S-18C/site E	Atrazine	0.1	0.0	0.0	2.83E-01
	Metolachlor	0.1	0.0		1.01E-02
	Chlorpyrifos		0.0	0.7	2.53E-03
	Endosulfan		0.3	1.8	1.95E-02
	msPAF (RA)	0.3	0.4	2.5	
S-332	Atrazine	0.0	0.0	0.0	6.22E-02
Site A	Atrazine	0.1	0.0	0.0	1.51E-01
	Metolachlor	0.1	0.0		2.96E-03
	Chlorpyrifos		0.1	1.3	6.08E-03
	Endosulfan		0.1	1	7.61E-03
	msPAF (RA)	0.1	0.2	2.4	
Site E1	Atrazine	0.0	0.0	0.0	2.12E-02
	Metolachlor	0.1	0.0		7.39E-03
	Chlorpyrifos		0.0	0.8	3.07E-03
	Endosulfan		0.3	1.6	1.65E-02
	msPAF (RA)	0.1	0.3	2.4	
Site E2	Atrazine	0.0	0.0	0.0	2.01E-02
	Metolachlor	0.1	0.0		6.94E-03
	Chlorpyrifos		0.0	0.7	2.66E-03
	Endosulfan		0.2	1.4	1.23E-02
	msPAF (RA)	0.1	0.2	2.1	
Site W1	Atrazine	0.1	0.0	0.0	1.68E-01
	Metolachlor	0.1	0.0		9.11E-03
	Chlorpyrifos		0.0	0.7	2.48E-03
	Endosulfan		0.3	1.5	1.51E-02
	msPAF (RA)	0.2	0.3	2.2	
Site W2	Atrazine	0.0	0.0	0.0	3.90E-02
	Metolachlor	0.1	0.0		8.10E-03
	Chlorpyrifos		0.0	0.6	2.12E-03
	Endosulfan		0.2	1.2	1.02E-02
	msPAF (RA)	0.1	0.2	1.8	

Zero indicates that there was no overlap between the estimated 90th centile concentration and the SSD. Blank space indicates that there was insufficient acute toxicity data to create a SSD

Table 5 Saltwater acute PAFs (%) and msPAFs (%)

Site	Compound	Phytoplankton/plants	Fish	Arthropods	90th centile (μg/l)
Card Sound	Atrazine	0.0	0.0	0.0	2.00E-02
	Chlorpyrifos	0.0	0.3	2.1	2.63E-03
	Endosulfan		0.3	3.7	3.26E-03
	msPAF (RA)	0.0	0.5	5.7	
Highway Creek	Atrazine	0.0	0.0	0.0	2.40E-02
	Chlorpyrifos	0.0	0.3	2.3	3.10E-03
	Endosulfan		0.6	5.0	6.56E-03
	msPAF (RA)	0.0	0.9	7.2	
Joe Bay	Atrazine	0.0	0.0	0.0	2.80E-02
	Metolachlor	0.1	0.0		2.91E-03
	Chlorpyrifos	0.0	0.3	2.4	3.19E-03
	Endosulfan		1.0	5.9	1.01E-02
	msPAF (RA)	0.1	1.3	8.2	

Zero indicates that there was no overlap between the estimated 90th centile concentration and the SSD. Blank space indicates that there was insufficient acute toxicity data to create a SSD

Table 6 Chronic PAFs (%) and msPAFs (%) for freshwater and estuarine sites

Site	Compound	PAF (%)	Median (μg/l)
S-175	Atrazine	0.1	3.60E-02
S-176	Atrazine	0.2	4.10E-02
S-177/site B	Atrazine	0.1	2.11E-02
	Metolachlor	0.6	6.29E-03
	Endosulfan	1.0	1.00E-03
	msPAF (RA)	1.7	
S-178/site C	Atrazine	0.1	2.45E-02
	Chlorpyrifos	0.5	3.10E-04
	Endosulfan	6.4	4.72E-02
	msPAF (RA)	7.0	
S-18c/site E	Atrazine	0	2.29E-02
	Chlorpyrifos	0.7	4.70E-04
	Endosulfan	1.2	1.33E-03
	msPAF (RA)	2.0	
S-332	Atrazine	0.1	3.30E-02
Site A	Atrazine	0.1	2.59E-02
	Metolachlor	0.4	3.37E-03
	Chlorpyrifos	0.8	5.50E-04
	Endosulfan	1.0	8.40E-04
	msPAF (RA)	2.3	
Site E1	Atrazine	0.1	1.16E-02
	Chlorpyrifos	1.1	7.80E-04
	Endosulfan	1.4	1.86E-03
	msPAF (RA)	2.5	
Site E2	Atrazine	0.1	1.10E-02
	Chlorpyrifos	0.8	5.20E-04
	Endosulfan	1.1	1.08E-03
	msPAF (RA)	1.9	

Table 6 continued

Site	Compound	PAF (%)	Median (μg/l)
Site W1	Atrazine	0.1	1.50E-02
	Chlorpyrifos	0.7	4.10E-04
	Endosulfan	1.4	1.85E-03
	msPAF (RA)	2.1	
Site W2	Atrazine	0.1	1.18E-02
	Chlorpyrifos	0.9	6.30E-04
	Endosulfan	1.9	3.44E-03
	msPAF (RA)	2.8	
Card Sound	Atrazine	0.0	2.30E-03
	Chlorpyrifos	1.5	1.19E-03
	Endosulfan	0.7	4.20E-04
	msPAF (RA)	2.1	
Highway Creek	Atrazine	0.0	8.51E-03
	Chlorpyrifos	1.2	9.50E-04
	Endosulfan	1.0	8.60E-04
	msPAF (RA)	2.1	
Joe Bay	Atrazine	0.0	7.46E-03
	Chlorpyrifos	1.5	1.27E-03
	Endosulfan	1.1	1.27E-03
	msPAF (RA)	2.7	

Zero indicates that there was no overlap between the estimated 90th centile concentration and the SSD

The potential risk of endosulfan for fish at S-177/site B was much lower than at S-178/site C even when including malathion and chlorpyrifos in a multiple substance JPC. However, the potential risk of all three pesticides to arthropods at S-177/site B was much higher than for fish when including them in a multiple substance JPC.
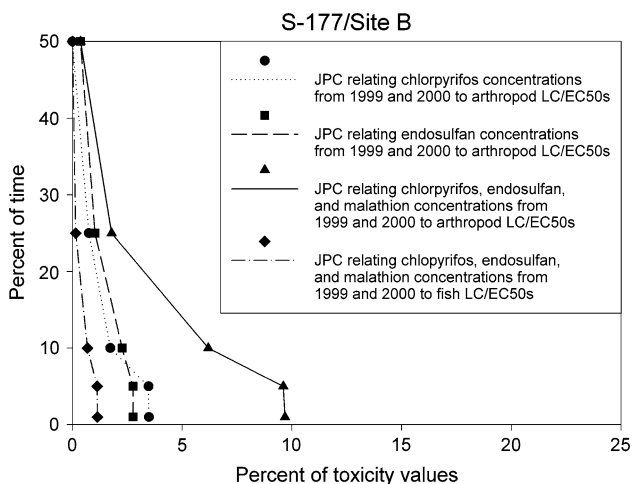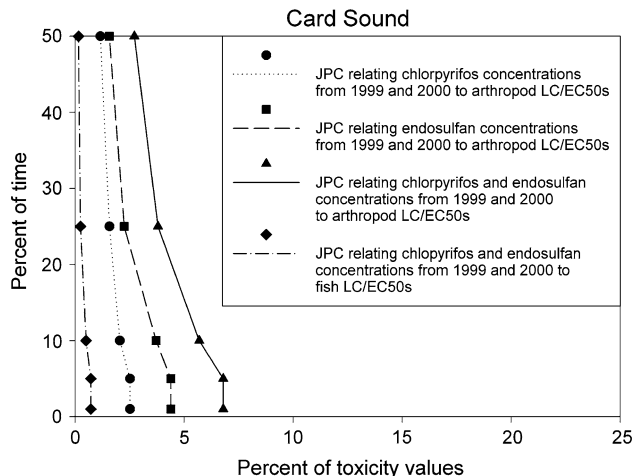
Fig. 2 Joint probability curves relating malathion, endosulfan, and chlorpyrifos concentrations in S-177/site B to arthropod and fish acute toxicity data (LC$_{50}$/EC$_{50}$s)
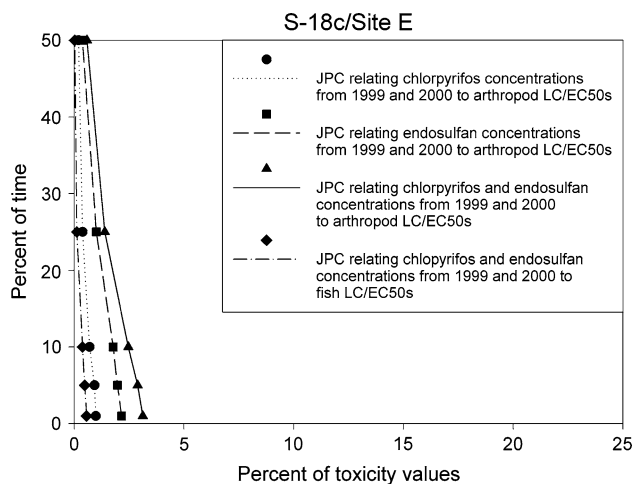
Fig. 4 Joint probability curves relating endosulfan and chlorpyrifos concentrations in S-18C/site E to arthropod and fish acute toxicity data (LC$_{50}$/EC$_{50}$s)
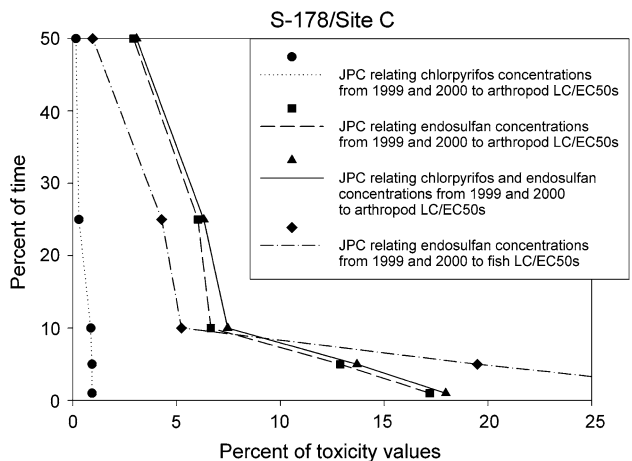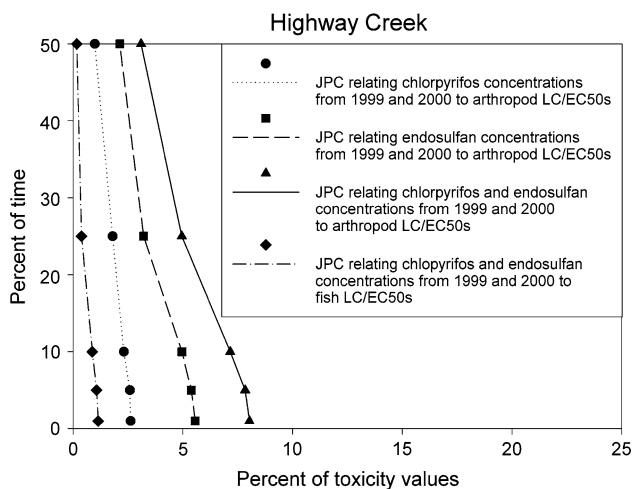

Fig. 3 Joint probability curves relating endosulfan and chlorpyrifos concentrations in S-178/site C to arthropod and fish acute toxicity data (LC$_{50}$/EC$_{50}$s)

Fig. 5 Joint probability curves relating endosulfan and chlorpyrifos concentrations in Card Sound to arthropod and fish acute toxicity data (LC$_{50}$/EC$_{50}$s)

Figures 5–7 present the JPCs for Card Sound, Highway Creek, and Joe Bay.

The highest potential risk was in Joe Bay for arthropods when exposed to chlorpyrifos and endosulfan. Conversely, the lowest potential risk at all three saltwater sites was for fish exposed to chlorpyrifos and endosulfan. Based on the results of the JPCs for Card Sound it appears that it may not be a suitable reference site, as suggested by NOAA, because of background concentrations of endosulfan.

Risks were also higher in February than in June. Miles and Pfeuffer (1997) found that the maximum total number of all pesticide detections were highest in winter to late spring. The differences in risk are evident when JPCs for February (dry season) and June (wet season) were

compared for both fresh- and saltwater sites. Joint probability curves for February and June exposure data of only S-178/site C (Fig. 8) and Joe Bay (Fig. 9) were highlighted to show seasonal differences in potential risk. At S-178/site C, 10% of the arthropod toxicity values were exceeded by endosulfan concentrations in February, 25% of the time. However, in June, the maximum percentage of arthropod toxicity values that were exceeded by endosulfan concentrations was 6% and this occurred only 1% of the time. The most dramatic difference in potential risk was to fish where, in February, 10% of the toxicity values were exceeded by endosulfan concentrations 35% of the time but in June the maximum percentage of fish toxicity values that were exceeded by endosulfan concentrations was 4% and
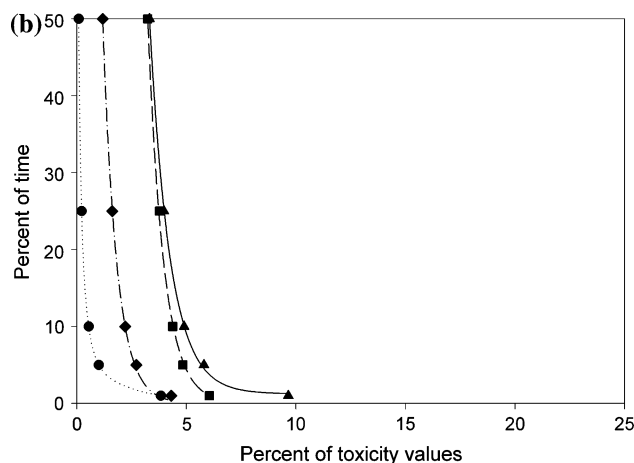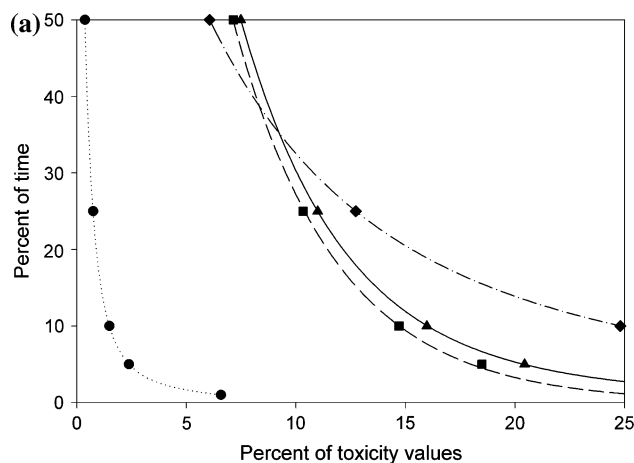
Fig. 6 Joint probability curves relating endosulfan and chlorpyrifos concentrations in Highway Creek to arthropod and fish acute toxicity data (LC$_{50}$/EC$_{50}$s)
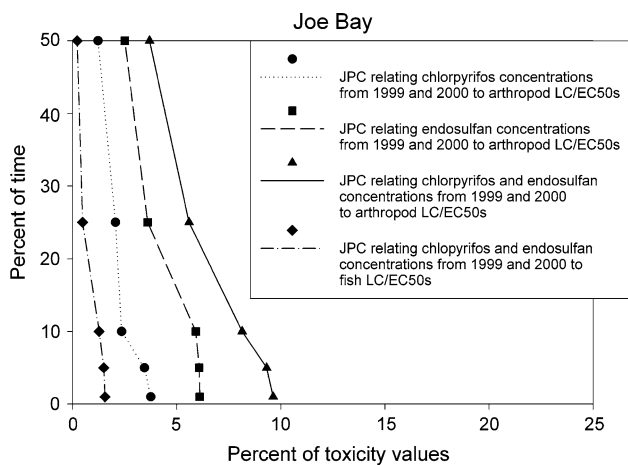
Fig. 7 Joint probability curves relating endosulfan and chlorpyrifos concentrations in Joe Bay to arthropod and fish acute toxicity data (LC$_{50}$/EC$_{50}$s)

Fig. 8 Joint probability curves relating endosulfan and chlorpyrifos concentrations in S-178/site C in February of 1999 and 2000 (**a**) and June of 1999 and 2000 (**b**) to arthropod and fish acute toxicity data (LC$_{50}$/EC$_{50}$s). *Legend*: black circles = chlorpyrifos and arthropods; black squares = endosulfan and arthropods; black triangles = endosulfan and chlorpyrifos and arthropods; black diamonds = endosulfan and fish

this occurred only 1% of the time. At S-178/site C, potential risks of endosulfan were higher than chlorpyrifos to arthropods in both February and June. The potential risk of joint action of chlorpyrifos and endosulfan to arthropods was also higher in February than June but there was no change in potential risk of chlorpyrifos to arthropods from February to June. At Joe Bay, 10% of the arthropod toxicity values were exceeded by endosulfan concentrations in February, 3% of the time. However, in June, the maximum percentage of arthropod toxicity values that were exceeded by endosulfan concentrations was 5% and this occurred only 1% of the time. At Joe Bay, potential risks of endosulfan are higher than chlorpyrifos to arthropods in February but this is reversed in June. The potential risk of joint action of chlorpyrifos and endosulfan to arthropods was also higher in February than June but there was little

change in potential risk of chlorpyrifos to arthropods from February to June.

Figure 10 shows the rest of the monthly JPCs for sites that exceeded hazard criteria and S-18C/site E and Card Sound.

Although Card Sound was selected as a reference site for comparisons, potential risks of pesticides in surface waters did exist at this site. Risks extrapolated from JPCs, however, were usually lower at Card Sound than they were at Highway Creek and Joe Bay.

Assessment endpoints

Analysis of the predicted adverse ecological effects related to the assessment endpoints indicates the following:

Primary producers: There was no potential risk of the herbicides or insecticides, either singly or jointly, to
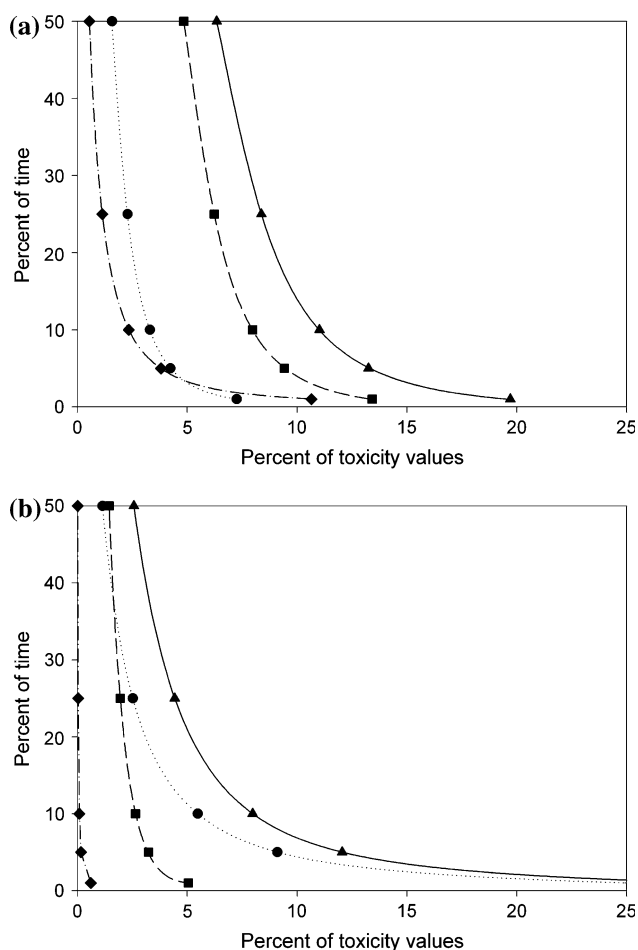
Fig. 9 Joint probability curves relating endosulfan and chlorpyrifos concentrations in Joe Bay in February of 1999 and 2000 (**a**) and June of 1999 and 2000 (**b**) to arthropod and fish acute toxicity data (LC_{50}/EC_{50}s). *Legend*: black circles = chlorpyrifos and arthropods; black squares = endosulfan and arthropods; black triangles = endosulfan and chlorpyrifos and arthropods; black diamonds = endosulfan and fish

primary producers. However, atrazine is ubiquitous in surface waters of south Florida canals at low concentrations (Pfeuffer and Rand 2004). The significance of exposure to low concentrations of atrazine, either singly or jointly with other herbicides, on primary producers requires further investigation in south Florida surface waters. However, a recent aquatic probabilistic risk assessment indicates that atrazine, neither singly nor jointly with other herbicides, produces any potential acute risks to primary producers (Schuler and Rand 2008).

Arthropods: Arthropods may be at low risk (directly) from single and multiple exposures to endosulfan in freshwater and saltwater. For example, pink shrimp may especially be at low risk from endosulfan exposures in Joe Bay and Highway Creek since the 96-h LC_{50} for pink shrimp and endosulfan is 0.04 μg/l.

Fish: Fish may be at risk (directly) from pulsed exposures to endosulfan. The maximum concentration of endosulfan measured at S-178/site C (1.345 μg/l) is close to the 96-h LC_{50} for channel catfish (1.5 μg/l), a native species within C-111. This concentration also exceeds the 96-h LC_{50} for bluegill (1.2 μg/l), a native species of Taylor Slough. Fish may also be at low risk (indirectly) from low food resources impacted by endosulfan at fresh- and salt-water sites. For example, at S-178/site C, Highway Creek, and Joe Bay the potential acute risk of endosulfan to arthropods is >5% at the 90th centile for exposure. Direct effects of endosulfan on arthropods (on percentage survival) will present less available food and indirect effects on fish that rely on this resource.

Uncertainties

Exposure analysis: The characterization of exposure in the C-111 system and in Florida and Biscayne Bays had severe limits in both spatial and temporal dimensions. For example, although the C-111 canal consists of over 100 square miles of basin and several ecologically distinct discharge sites, only a total of 11 sites were sampled for pesticides in areas related to C-111. There were only two sampling sites in northeast Florida Bay and one in Biscayne Bay.

One of the largest sources of uncertainty in the analysis of exposure arose from analytical data for pesticides that were detected frequently or at high concentrations by some monitoring programs but were not analyzed in other monitoring programs. The SFWMD maintains an ongoing pesticide monitoring program at several sites on the C-111 system and related tributaries (C-111E). Water sampling occurs quarterly throughout the year. However, chlorpyrifos was not analyzed by the SFWMD.

Spatial data for various sites in C-111 came from a project by NOAA. Sampling was initiated to measure changes in wet, dry, and transition seasons. However, only a few dates were sampled for each season. Although endosulfan was a pesticide of importance in all tiers of the C-111 risk assessment, only α-endosulfan was analyzed by the USGS in surface waters in 1998 and none of the isomers or degradates of endosulfan were measured by the USGS at S-177 in following years. In addition, NOAA did not analyze for malathion or its metabolite, malaoxon, in any of its sampling sites.

The measured surface water concentrations of α-, β-endosulfan, and endosulfan sulfate were added at each site to create exposure distributions for endosulfan (total). These exposure distributions for endosulfan (total) allowed a comparison to a large toxicity effects database for technical-grade endosulfan, which is a mixture of the α- and β-isomers of endosulfan. It would have been appropriate to

Fig. 10 Joint probability curves relating endosulfan and chlorpyrifos concentrations in freshwater sites (S-177/site B, S-18c/site E) and saltwater sites (Card Sound, Highway Creek) in February and June to arthropod and fish acute toxicity data (LC_{50}/EC_{50}s). *Legend*: black circles = chlorpyrifos and arthropods; black squares = endosulfan and arthropods; black triangles = endosulfan and chlorpyrifos and arthropods; black diamonds = endosulfan and fish; black diamonds in (**f**) represent chlorpyrifos and fish
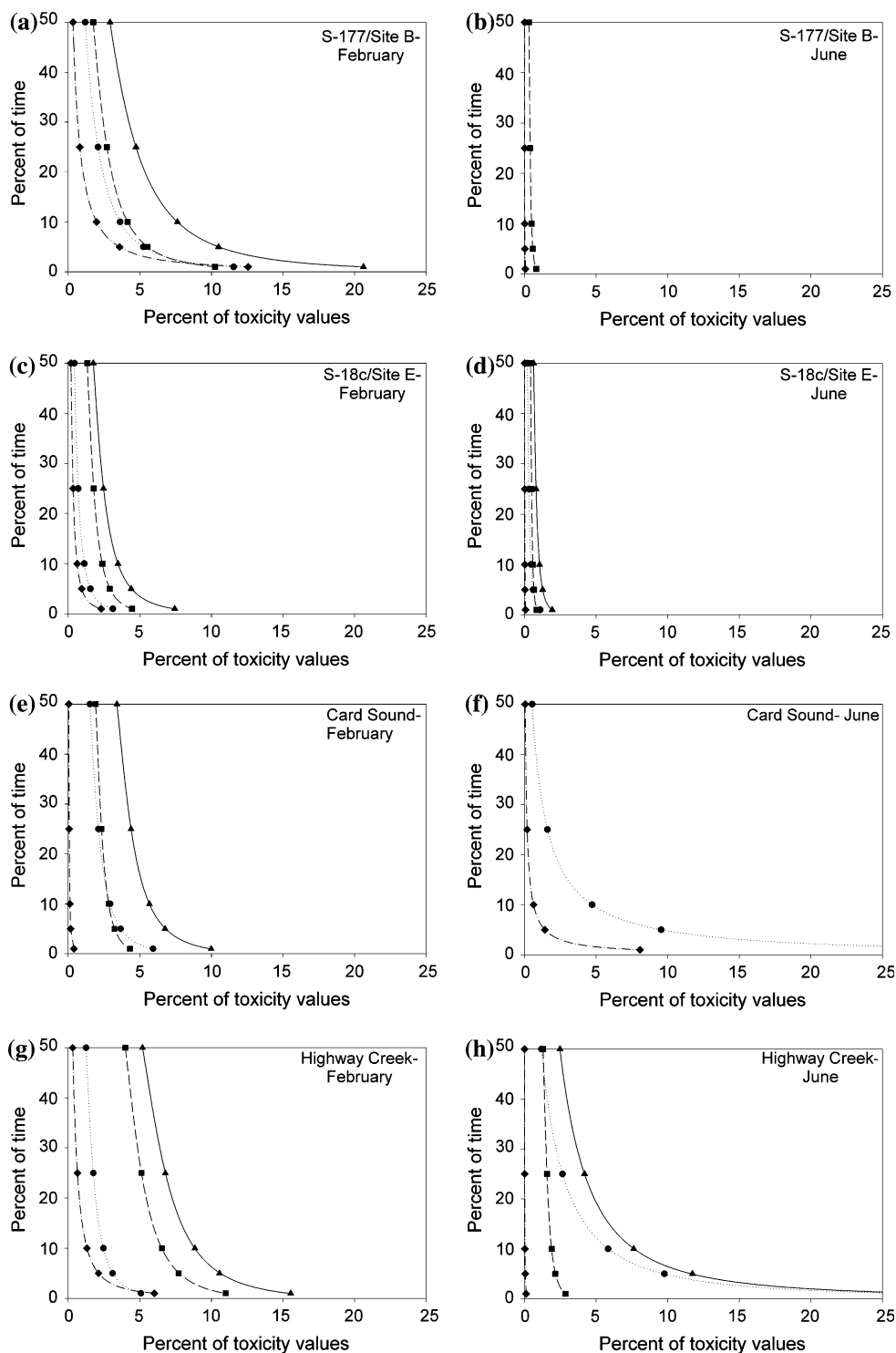
compare individual exposure distributions for α-endosulfan, β-endosulfan, and endosulfan sulfate with their respective toxicity distributions but the data is limited. SSDs for each chemical therefore could not be developed. However, in the preparation of agricultural formulations technical-grade endosulfan is used. In a sediment-water system organisms are potentially exposed to all three

chemicals. Therefore, it is a very valid and conservative approach to use total endosulfan based on the actual exposure that the aquatic organisms are experiencing.

Effects analysis: Pesticide registration testing under the USEPA Federal Insecticide Fungicide and Rodenticide Act (FIFRA) requires single-species testing on a limited number of aquatic species with an ultimate goal of predicting

effects on many nontarget biota. Relying on a limited database of single-species toxicity tests to predict effects on higher levels of organization can lead to problems, especially when the standard test species do not include native species in the area undergoing an ecological risk assessment. It was evident from the screening level ecological risk assessment (SERA) that more ecotoxicity testing has to be completed for native species in the C-111 basin and for northeast Florida Bay and south Biscayne Bay.

Another source of effects uncertainty is the exposure duration used in the laboratory versus the field. Most standard acute tests have durations that range from 24 to 96 h and most standard chronic tests are 30 days or less with invertebrates. Full chronic tests are not routinely conducted with pesticides. In the field, pesticide exposures are often pulses and vary with application frequency. Dissipation of pesticide residues in the water column commonly takes hours to days but pesticide applications can be repeated multiple times within a growing season. In south Florida, some pesticides are applied frequently (up to ten or more times) with a year-round growing season. Early life stages of fish and zooplankton with short life cycles may thus be adversely affected by pulsed exposures. Pulsed exposures are not considered in the ecotoxicity testing of pesticides by chemical manufacturers because they are not required under FIFRA ecotoxicity testing guidelines.

Although the toxicity of α-endosulfan, β-endosulfan, and endosulfan sulfate have been designated as being similar (WHO 1984), recent available toxicity data from the literature indicates that α-endosulfan may be more toxic than β-endosulfan and endosulfan sulfate to *H. azteca* and *C. tentans* (You et al. 2004) and *Daphnia magna* and rainbow trout (*Oncorhynchus mykiss*) (Wan et al. 2005). However, the toxicity database for each isomer and the metabolite of endosulfan are still inadequate for developing separate acute and chronic SSDs.

Risk characterization: Concentration and response addition may not always account for the joint effects of mixture toxicity. The application of laboratory toxicity tests to these models has validated the use of these theories, in many situations, for predicting the joint effects from mixtures (Altenburger et al. 2000; Backhaus et al. 2000; Deneer et al. 1988; De Wolf et al. 1988; Faust et al. 2000). However, the development of methods for a probabilistic assessment of joint effects from mixtures (i.e., multiple chemical stressors) is not fully developed (Giesy et al. 1999; De March 1987; Suter et al. 2002). The effects of a mixture of atrazine and chlorpyrifos, and atrazine and malathion in water on laboratory-reared *C. tentans* found effects greater than additivity in both studies (Pape-Lindstrom and Lydy 1997). However, atrazine concentrations in the tests were one to two orders of magnitude higher than typical environmental concentrations and synergistic interactions from chemical stressors is still the exception, particularly in pesticide mixtures exceeding binary and tertiary combinations (Pape-Lindstrom and Lydy 1997). Although they may not always be representative, concentration addition and response addition are still the accepted models for predicting the joint toxicity of mixtures.

Conclusions

A screening-level aquatic probabilistic risk assessment was completed to determine the potential risks of organic pesticides found in surface waters of the C-111 basin and adjacent estuarine tidal zones (northeast Florida Bay, south Biscayne Bay) in south Florida. It followed the USEPA ecological risk framework (USEPA 1998) and focused only on the potential acute and chronic risks of endosulfan and chlorpyrifos, individually and jointly and with atrazine, metolachlor, and malathion. From the hazard assessment (Tier 1), endosulfan and chlorpyrifos were determined to be COPECs and potential hazards in surface waters but atrazine, metolachlor, and malathion did not exceed numerical criteria and therefore were not considered potential hazards (Carriger and Rand, this issue).

From the results of this probabilistic aquatic risk assessment on 11 freshwater sites in or around the C-111 basin and three estuarine sites several conclusions were reached. The highest risk of acute effects was associated with endosulfan exposure on freshwater arthropods at S-178/site C (PAF 6.7%), followed by endosulfan effects on estuarine arthropods at Joe Bay (PAF 5.9%). The highest risk of acute effects from joint toxicity of pesticides was to estuarine arthropods in Joe Bay (msPAF 8.2%) followed by freshwater arthropods in S-178/site C (msPAF 7.5%). For fish, the highest acute risk was for endosulfan at S-178/site C (PAF 5.3%). There was low potential for acute risk of endosulfan to fish at estuarine sites. The highest potential risk of acute effects in fresh water was associated at sites near water control structure S-178 on C-111E, a branch of the C-111. Also, in the JPCs it is apparent that the majority of potential risks to arthropods and fish were due to endosulfan concentrations and not to chlorpyrifos, at S-178/site C. The highest risk of acute effects for saltwater organisms was in Joe Bay, which receives water from the C-111. The potential risk of chronic effects from pesticide exposures was minimal in fresh- and saltwater except at S-178/site C, where the endosulfan PAF was 6.4% and the msPAF was 7.0%. In general, potential risks were higher in February than June.

S-178 has no exposed headwaters and it drains an agricultural area. The water control structure, near the sampling site, is rarely open and therefore persistence of

endosulfan and its metabolites would be enhanced in this area, especially in sediment since it has low water solubility and a high K_{oc} (Carriger and Rand, this issue). Carriger et al. (2006) showed that endosulfan had the highest potential chronic risk to arthropods at S-178 when bulk sediment concentrations were considered in a probabilistic risk assessment.

The study demonstrated that there is low potential risk of endosulfan to arthropods and fish at localized freshwater (S-178/site C) and estuarine (Joe Bay, Highway Creek) sites. Therefore, future contaminant monitoring programs in south Florida should address more intensive sampling regimes in these locations with periodic preparation of quantitative aquatic risk assessments.

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