California Environmental Protection Agency
Department of Pesticide Regulation
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STUDY 167: PROTOCOL FOR MONITORING ACUTE AND CHRONIC TOXICITY IN THE SAN JOAQUIN RIVER WATERSHED, WINTER 1997-98

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I. INTRODUCTION

In the San Joaquin Valley, the organophosphorus insecticides diazinon, chlorpyrifos, or methidathion are generally applied with a dormant oil on nut and stone fruit trees to control peach twig borer, San Jose scale, European red mite, and brown mite pests. The best time to achieve control of these pests is December through February, when trees are dormant and better pesticide coverage is possible (Zalom *et al.*, 1995). This dormant orchard spray application period, however, coincides with seasonal rainfall. Thus, these pesticides have the potential to wash off target areas and migrate with runoff waters to the San Joaquin River. This study is designed to monitor toxicity of pesticides in the San Joaquin River watershed during winter months when dormant insecticidal sprays are being applied to orchards.

From 1988 to 1990, the Central Valley Regional Water Quality Control Board conducted an aquatic toxicity survey in the San Joaquin Valley. Surface water samples collected from certain reaches of the San Joaquin River watershed during this survey were acutely toxic to the water flea, *Ceriodaphnia dubia* (Foe and Connor, 199 1). The cause of toxicity was not determined but was attributed to pesticides in general. Further study was conducted in the Valley during the winter of 1991-92, and the resultant toxicity was attributed to the presence of chlorpyrifos and diazinon (Foe and Sheipline, 1993; Foe, 1995; Kuivila and Foe, 1995). The toxicity found in these studies was in violation of the Central Valley Regional Water Quality Control Board's narrative water quality objective (Foe, 1995). The toxicity objective states that, "All waters shall be maintained free of toxic substances in concentrations that produce detrimental physiological responses in human, plant, animal, or aquatic life" (CVRWQCB, 1994).

The Department of Pesticide Regulation (DPR) monitored the San Joaquin River watershed during the winters of 1991-92 and 1992-93 and reported the detection of chlorpyrifos, diazinon, and methidathion in 10, 72, and 18 percent of the 108 water samples collected, respectively (Ross, 1997). Of these positive samples, 2, 13, and 1 percent exceeded the LC_{50} for *C. dubia,* respectively, indicating potential acute toxicity. In addition, diazinon concentrations in the San Joaquin River at Vernalis ranged from 0.148 to 1.07 µg/L, on 12 consecutive days in 1993, and the authors concluded that chronic toxicity due to diazinon might be problematic at this site (Kuivila and Foe, 1995). During the 12 days, diazinon exceeded the 96 hour LC_{50} for *C. Dubia* four times (Table 1). Dormant spray insecticides, at levels acutely toxic to test organisms, were also reported in Orestimba Creek, a tributary to the San Joaquin River, during the 1992-93

dormant spray period (Domagalski, 1995). Consequently, programs to reduce the mass of dormant orchard spray insecticides leaving target areas have been under investigation by DPR and growers (Ross, 1997; Ando, 1996; Anonymous, 1996; Biermann, 1996).

During the winter of 1996-97, DPR conducted toxicity monitoring at sites along the San Joaquin River and Orestimba Creek (Bennett, 1997). During the first half of winter, there was unusually wet weather and flooding followed by unseasonably dry weather during the second half of winter. Water samples from Orestimba Creek contained residues of diazinon, carbofuran, and dimethoate in 20, 13, and 7 percent of the samples collected, respectively. The maximum diazinon, carbofuran, and dimethoate concentrations detected were 0.092, 0.238, and 0.082 ug/L, respectivley. Twelve percent of water samples from the San Joaquin River near Vernalis were found to have diazinon residues with a maximum concentration of 0.070 ug/L. *Ceriodaphnia dubia* survival ranged from 45 to 100 percent for acute toxicity from Orestimba Creek with only one toxic sample coinciding with a pesticide detection. Chronic toxicity tests conducted on water samples from the San Joaquin River near Vernalis displayed either 90 or 100 percent survival. The concentrations detected did not exceed LC_{50}s listed for these insecticides (Table 1). Complete analysis of last winter's study data will appear in a report later this year.

In this study we will continue to look at acute toxicity to *C. dubia* in a small watershed where the discharging waters do not contain municipal or industrial contaminants. We will also investigate the potential for chronic toxicity in a reach of the San Joaquin River downstream from major orchard and tributary inputs in the watershed. Due to the lack of water quality criteria with which to compare detected concentrations with, long-term monitoring of acute and chronic toxicity will provide an assessment of the effects on aquatic life and will help scientists at DPR to evaluate the effectiveness of programs designed to decrease the runoff of dormant spray insecticides.

During the winter season, herbicides are also applied in the San Joaquin River watershed and are known to migrate in surface water to the San Joaquin River. During the period December through March, U.S. Geological Survey has detected the herbicides simazine, cyanazine and dacthal in the San Joaquin River at Vernalis (MacCoy et al., 1995). As part of an ongoing effort to gain information about pesticide residues in state surface waters as recommended in the memo "Catagory and recommendation of currently registered pesticides for surface water monitoring during FY 97-98" (Goh, 1997), additional samples will be collected during this study and analyzed for selected herbicides.

II. OBJECTIVE

The objective of this study is to monitor the occurrence of acute and chronic toxicity in the San Joaquin River watershed during the dormant spray season. Additionally, levels of specific organophosphate and carbamate insecticides and selected herbicides that have potential to enter the San Joaquin River with surface runoff will also be monitored. A companion study will be conducted to monitor toxicity and pesticide levels in the Sacramento River.

III. PERSONNEL

This project will be conducted by the Environmental Hazards Assessment Program (EHAP) under the general direction of Don Weaver, Ph.D., Senior Environmental Research Scientist (Supervisor). Key personnel are listed below:

Project Leader: Carissa Ganapathy
Field Coordinator: Jeff Schuette and Bridget Hansen
Senior Scientist: John Troiano, Ph.D.
Statistician: Terre11 Barry, Ph.D.
Contractor (toxicity tests): Charlie Huang, Ph.D., California Department of Fish & Game
Chemists: Jorge Hernandez, Jean Hsu, and Hsiao Feng, California Department of Food & Agriculture
Agency and Public Contact: Pat Dunn

Questions concerning this monitoring project should be directed to Pat Dunn at (916) 324-4100. Fax: (916) 324-4088.

IV. STUDY PLAN

The San Joaquin River extends approximately 2 15 km from Friant Dam to Stevinson where flows are intermittent. The flow from Stevinson to Vernalis (about 97 km), is perennial. Acute toxicity sampling will be conducted at Orestimba Creek, a western tributary to the San Joaquin River, as this site receives runoff that is predominantly agricultural (Figure 1). Sampling for chronic toxicity will be conducted on the San Joaquin River at Vernalis, as this site receives discharges from all of the River's major agricultural tributaries, including the Merced, Tuolumne, and Stanislaus Rivers. Discharge records for both the Orestimba Creek and Vernalis sampling sites are available from collocated gauging stations. This information will be used to correlate any changes in chemical concentrations to fluctuations in flow and may be useful for modeling efforts, should they be undertaken.

Monitoring will commence prior to the onset of the dormant spray season (December 1997) and continue through the middle of March 1998. Background samples will be collected for one week, beginning prior to dormant spray applications, then monitoring will resume once applications have begun in the watershed. Monitoring will occur weekly and continue until no later than March 20, 1998.

V. SAMPLING METHODS

Acute toxicity sampling will be conducted on Monday and Wednesday of each week. Sampling for chronic toxicity will be conducted weekly. One chronic sample constitutes the collection of a sample on days zero, two, and four of each week (e.g. Mon., Wed., and Fri.). Water collected on those days will be delivered the following day to the laboratory for testing and sample renewal. Chemical analyses will also be performed on each sample collected for both acute and chronic tests. Selected organophosphate and carbamate pesticides will be analyzed using three chemical analytical screens (Table 2). The herbicides are not expected to reach levels that would

contribute to C. *dubia* toxicity (Table 1) but will be monitored to look for their possible effects on other aquatic life.

A center channel water sample will be collected from bridges at each site. This will be done using a depth-integrated sampler (D-77) with a 3-liter Teflon' bottle and nozzle. Surface water subsamples will be composited temporarily in a stainless steel container until the appropriate volume of water has been collected. This composited sample will then be stored on wet ice until delivered to the processing facility in West Sacramento. Immediately upon arrival at West Sacramento, the samples will be split into amber glass bottles using a Geotech® 10-port splitter then sealed with Teflon@-lined lids. Samples to be analyzed for organophosphate and carbamate pesticides will be acidified with 3N hydrochloric acid to a pH between 3.0 and 3.5. At this pH, most of the organophosphate and carbamate pesticides are sufficiently preserved with the exception of diazinon. Diazinon degrades more rapidly at an acidic pH and therefore will be analyzed from a separate, unacidified, split sample. Samples submitted for herbicide analysis and toxicity tests will not be acidified. Sufficient water will be collected during each sampling event to provide approximately four liters for chemical analysis, two liters each for both acute and chronic toxicity tests, and any additional water required for quality control samples. All samples will be stored at 4°C until delivered to the laboratories for toxicity testing and chemical analyses. In addition, samples submitted for toxicity testing will be delivered within 24 hours of collection.

Dissolved oxygen, pH, specific conductance, and water temperature will be measured *in situ,* at each site, during each sampling period. As part of the toxicity testing, the California Department of Fish and Game's Aquatic Toxicology Laboratory will measure and record other parameters of the delivered toxicity samples, including totals of alkalinity, hardness, and ammonia, and specific conductivity. These measurements will be made in the laboratory, thus they will not directly reflect on-site conditions.

VI. TOXICITY TESTING AND CHEMICAL ANALYSIS

Toxicity testing, conducted by the California Department of Fish and Game's Aquatic Toxicology Laboratory, will follow current U.S. Environmental Protection Agency procedures using the cladoceran *Ceriodaphnia dubia* (U.S. EPA, 1993). The Aquatic Toxicology Laboratory has been accredited by the California Department of Health Services' Environmental Laboratory Accreditation Program. Acute toxicity will be determined using a 96-hour, static-renewal bioassay in undiluted sample water. Chronic toxicity will be determined using a seven-day bioassay with *C. dubia* in undiluted sample water, and will follow current U.S. Environmental Protection Agency guidelines (U.S. EPA, 1994). For example, test organisms used in chronic testing will be subjected to sample water from day zero on the following day (day 1). Sample water collected on days two and four will then replace test water on days three and five, respectively. All bioassays will commence within 36 hours of sample collection. In addition, data will be reported to the project leader as percent survival on each day for the duration of the tests.

Chemical analysis will be performed by the California Department of Food and Agriculture Center for Analytical Chemistry. The reporting limit will be used to record the lowest concentration of analyte that the method can detect reliably in a matrix blank. The method titles and reporting limits for the study are listed in Table 2. Comprehensive chemical analytical methods will be provided in the final report.

Number of Toxicity Tests		
2 acute/week x 11 weeks of study		22
1 chronic sample per week x 11 weeks of study		11
	Total	33
Number of Chemical Analyses		
4 (OP, CB, diazinon and herbicides) per acute toxicity sample: 4 analyses x 2 acute toxicity sampling events/week x 11 weeks		88
4 (OP, CB, diazinon and herbicides) per chronic toxicity sampling event: 4 analyses x 3 chronic sampling events (=1 chronic sample)/week x 11weeks		132
	Subtotal	220
Quality Control		
Continuing QC (approx. 10% of total chemical analyses)		22
	Total number of chemical analysis samples	242

VII. QUALITY ASSURANCE/QUALITY CONTROL

Chemical Analysis

Quality control will be conducted in accordance with Standard Operating Procedure QAQC001 .00. Ten percent of the total number of primary analyses will be submitted with field samples as blind matrix spikes and rinse blanks. The total number of samples is presented above.

VIII. DATA ANALYSIS

Toxicity data will be used to establish baseline information on the occurrence of acute or chronic events at the Orestimba and Vemalis sites. A correlation matrix will be generated to investigate potential relationships between measured environmental parameters, discharge, toxic events, and chemical concentrations. Measured concentrations will be compared to various established water quality criteria including California Quantitative Response Limits, U.S. EPA objectives, and California Department of Fish and Game suggested criteria. Pesticide concentrations will also be compared with acute and chronic LC_{50}s for *C. dubia* to aid in interpreting toxicity test results.

With a fixed monitoring schedule, sample collection during storm events, when pesticide levels are typically highest, may not occur. Therefore, records of storm events will be kept, and an analysis of discharge, chemical data, and toxicity will be discussed in relation to sampling periods. Depending on the results of the preliminary analysis, further analysis may include multivariate analysis.

IX. TIME TABLE

Field Sampling - December 1997 and application onset through March 1998
Toxicity Testing and Chemical Analysis - December 1997 through April 1998
Preliminary Memorandum - August 1998
Final Report - January 1999

X. REFERENCES

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