EVALUATION OF AMBIENT TOXICITY TESTS FOR DETECTING GROUNDWATER POLLUTION ENTERING STREAMS AND RIVERS: FINAL REPORT

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ABSTRACT

Groundwater pollution is an emerging environmental concern in the Rocky Mountain region. In this two-year study, we evaluated the utility of two sublethal toxicity tests for detecting migration of contaminated ground water into streams and rivers.

During Year 1, we tested groundwater or surface-water samples from five locations at each of two study sites: 1) the Laramie River as it flowed past a former railroad tie treating plant south of Laramie, Wyoming, from June 1985 to October 1985; and 2) Crow Creek as it flowed past an oil refinery in Cheyenne, Wyoming, from June 1985 to April 1986. During Year 2, we tested groundwater and surface-water samples only at the Crow Creek site, from June to September 1986. Each water sample was tested for its effects on survival and reproduction of *Ceriodaphnia dubia* (an aquatic invertebrate) and survival and growth of fathead minnow (*Pimephales promelas*) larvae. Chemical analyses of water samples included routine water chemistry parameters, major inorganic ions, 11 trace elements, dissolved organic carbon, reverse-phase HPLC gradients, and GC-MS analyses of organics.

At the Laramie River, toxic ground water underlaid sediments adjacent to the tie treating plant. However, migration of ground water into the Laramie River did not adversely affect fathead minnows and *Ceriodaphnia*. Some groundwater and surface-water samples from Crow Creek also were toxic. The oil refinery's effluent appeared to cause much of the adverse effects in surface water downstream from that discharge. Adverse effects upstream from the refinery discharge may have been caused by contaminated ground water or storm sewer runoff.

Results of this study indicate that (1) ambient toxicity tests can be used in alkaline surface waters of the western U.S.; (2) they are sensitive enough to detect migration of contaminated ground water into surface waters; (3) they may be more sensitive in some cases than routine, inexpensive chemical analyses for detecting the presence of contaminants; (4) toxicity of contaminated ground water and an industrial discharge varied considerably during the two-year study; and (5) toxicity of interstitial ground water did not always correspond with toxicity of the overlying surface water or downstream surface water.

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INTRODUCTION

Groundwater pollution is an emerging environmental problem in the Rocky Mountain region of the United States. Contamination of near-surface aquifers is an especially important concern because many potential pollutants are applied directly to the soil surface (e.g., herbicides, pesticides, solid and liquid wastes) or are buried at relatively shallow depths (e.g., landfills, hazardous chemical storage ponds, burial pits), where they can migrate into or be leached by ground water. Subsequently, contaminated water flowing through near-surface aquifers can enter surface waters and degrade water quality downstream. Chemical analyses of water from monitoring wells can sometimes be used to trace the transport of groundwater pollutants. However, identifying and quantifying all of the potential inorganic and organic pollutants can be time-consuming and expensive. And a major question still arises: Do contaminant concentrations identified in these analyses pose environmental hazards in receiving waters?

Therefore, it would be desirable to have sensitive biological tests to complement the chemical analyses that are routinely used to detect groundwater pollutants. The U.S. Environmental Protection Agency (EPA) recently developed relatively quick, sublethal toxicity tests using fish and invertebrates for monitoring the effects of industrial and municipal effluents that are discharged into surface waters (Mount and Norberg 1984, Norberg and Mount 1985). Those tests are designed to detect adverse effects on fish growth and invertebrate reproduction and to be more sensitive than traditional acute lethality tests. Because of this improved sensitivity, they have been used successfully (1) for testing instream (ambient) toxicity of effluents after they are discharged into receiving waters and (2) for more traditional serial-dilution testing of effluents before they enter receiving waters (e.g., Mount et al. 1984, Mount and Norberg 1985).

Unfortunately, groundwater contamination has not yet been addressed in the EPA ambient toxicity testing programs. Therefore, we conducted a two-year study to evaluate the utility of the EPA fish and invertebrate toxicity tests to detect chemical pollution from groundwater and effluent sources along the Laramie River and Crow Creek in southeastern Wyoming. In this report, we (1) present results of these investigations, (2) evaluate the toxicity test methods as they are currently being used by EPA and contract laboratories, and (3) compare costs and sensitivity for detecting pollutants at our study sites among several levels of chemical analyses and toxicity tests.

OBJECTIVES

The objectives of this two-year study were as follows.

1. Evaluate EPA ambient toxicity tests as monitors of biological effects of groundwater pollutants.
2. Compare the sensitivity of those biological tests to the sensitivity of chemical analyses for detecting the presence of groundwater contaminants.
3. Assess temporal variability of groundwater and surface-water contamination in two Wyoming streams.

By sampling surface waters adjacent to suspected groundwater contamination sources, we anticipated that the emergence of contaminated ground water into streams and rivers could be detected using biological tests. Then the near-surface ground water could be sampled to determine its toxicity, identify its chemical constituents, and ascertain whether it could have caused the observed instream toxicity. Using this sequential testing procedure, we hoped that a cost-effective approach could be developed to evaluate the potential environmental hazards of contaminated ground water entering streams and rivers.

RELATED RESEARCH

Crossey and Bergman (1985) reported initial investigations of organic contaminant transport in ground water, surface water and sediments at the Union Pacific Tie Treatment Plant adjacent to the Laramie River in Laramie, Wyoming. Although toxicity tests were not conducted in that study, Crossey and Bergman (1985) demonstrated that (1) creosote oil underlaid Laramie River sediments and occasionally entered the Laramie River directly by way of oil seeps along the river bank, and (2) surficial sediments downstream from the tie treating plant were contaminated with chemical constituents identified in creosote oil.

Steadman (1986) reported preliminary studies of contamination along Crow Creek in Cheyenne, Wyoming. In that study, in situ biomonitoring and subsequent biochemical analyses of exposed fish at two sites adjacent to Frontier Oil Refinery property (formerly Husky Oil Refinery and Husky/RMT Properties, Inc.) demonstrated that (1) Crow Creek water downstream from the refinery's NPDES discharge was toxic to rainbow trout, and (2) groundwater or surface-water contaminants might be entering Crow Creek along the refinery's property upstream from the NPDES discharge pipe. Unfortunately, fathead minnow survival and growth and Ceriodaphnia survival and reproduction tests were not conducted in that study. Therefore, results of Steadman's (1986) biochemical analyses on rainbow trout cannot be interpreted directly with respect to the current study of ambient toxicity in Crow Creek.

Fathead minnow and Ceriodaphnia ambient toxicity tests are rapidly becoming accepted by regulatory agencies as sensitive indicators of instream biological effects of industrial and municipal effluent discharges. But because these two toxicity tests are relatively new, they are continually being revised and tested in inter-laboratory comparisons. For example, we recently participated in a round-robin evaluation of the fathead minnow test that was coordinated by Dr. G. Michael DeGraeve of Battelle Columbus Laboratories in Columbus, Ohio (DeGraeve et al. 1987). Dr. DeGraeve is currently investigating improved culture techniques for the *Ceriodaphnia* test and coordinated a round-robin evaluation of that test protocol in 1987 and 1988. Additionally, Dr. Donald Mount and Ms. Teresa Norberg-King of the U.S. EPA Environmental Research Lab in Duluth, Minnesota, continue to refine and evaluate fathead minnow and *Ceriodaphnia* test techniques at field research sites (Mount et al. 1984, Mount and Norberg 1985). We frequently communicate with these and other researchers around the United States regarding ambient toxicity tests. However, to our knowledge no one has yet used these tests to detect and evaluate the effects of contaminated ground water entering streams and rivers.

METHODS

Site Descriptions

Two surface waters in southeastern Wyoming were chosen for this study. Both the Laramie River and Crow Creek flow past industrial sites where groundwater flow patterns indicate discharge from the industrial property to the surface water. Because both sites have significant subsurface contamination, ground water is a potential pollution source for the adjacent stream or river.

Laramie River. The Union Pacific Tie Treatment Plant (UPTTP) is a U.S. EPA Superfund site located 1 km southwest of Laramie, Wyoming, adjacent to the Laramie River (Fig. 1). From the UPTTP site, the river flows northward through Laramie. Approximately 32 ha (80 acres) of the UPTTP property are heavily contaminated with creosote wastes emanating from a series of unlined waste ponds (CH2M/Hill 1985). The site is bordered on the north by Interstate 80 and on the west by the Laramie River and is underlain by contaminated alluvial sediments that extend from ground surface to bedrock, 5 to 10 m below ground surface. Ground water within the alluvial aquifer travels northwesterly across the site and discharges to the river along the west and northwest borders of the site. This alluvial aquifer is a major source of contaminants to the Laramie River (see CH2M/Hill 1984, 1985 for a complete geologic description).

In October 1983 an oily seep was discovered in the Laramie River adjacent to the UPTTP site, and free oil could be detected in the river up to 3 km downstream (Crossey and Bergman 1985). Subsequently, mini-piezometers were used to monitor and define the extent of the seep. In 1984, an oil body was

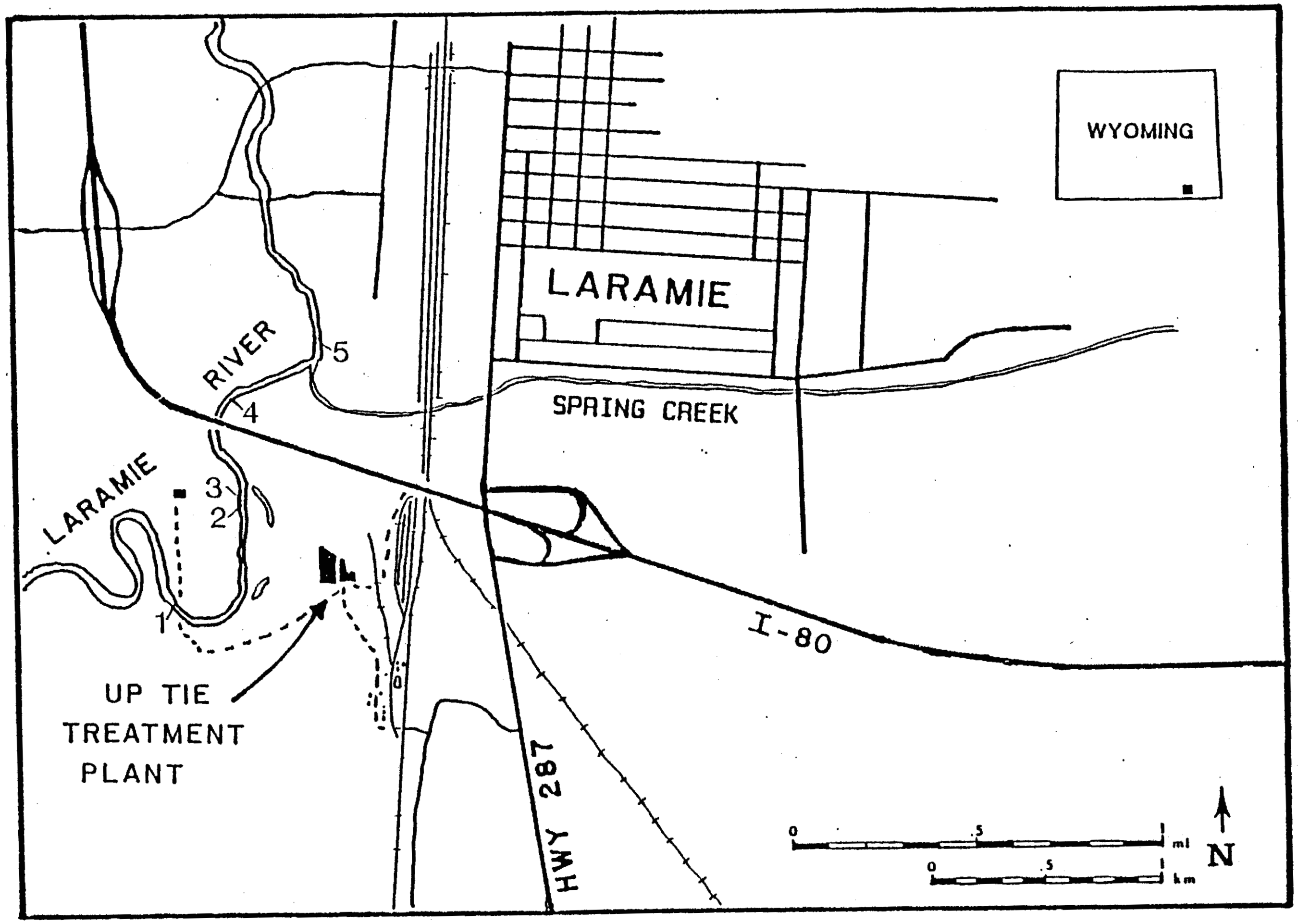


Figure 1. Location of the Laramie River study site, Laramie, Wyoming. Numbers denote sampling locations: 1 = Upstream Control and Upstream piezometer (October sampling only), 2 = Above seep, 3 = Site piezometer, 4 = I-80, 5 = Spring Creek.

located 1 m below the surface of the river sediments at the same location (Crossey and Bergman 1985). It extended approximately 30 m along the river bank and halfway across the river channel and contained an estimated 4000 to 6000 L of creosote oil. As a result, the Laramie River was relocated approximately 50 m west of its former channel in September 1985, in order to control further contamination entering the river (Fig. 2). The former river channel was covered by several meters of fill and is no longer accessible for sampling. Additional remedial cleanup activities have been initiated at the UPTTP site by the Union Pacific Railroad.

Crow Creek. Frontier Oil Refinery is adjacent to Crow Creek, a small third-order stream that flows through Cheyenne, Wyoming (Fig. 3). Effluent from the refinery is regulated under an NPDES (National Pollutant Discharge Elimination System) discharge permit and enters Crow Creek on the downstream (eastern) border of the refinery property. Although the effluent does not exceed those discharge permit limitations, Steadman (1986) reported adverse effects on rainbow trout placed in Crow Creek.

Since 1930, daily refinery operation, spills and leaking waste ponds have contaminated ground water beneath the property considerably. Abandoned waste ponds from the original refinery facility are buried at the southwest corner of the site. This complex of old ponds is designated the Southwest Surface Impoundment (SWSI Site, Fig. 4) and is being decontaminated by the current operators of the refinery. Ground water flows northwest to southeast, traversing the site and discharging to Crow Creek (Bill Payne, Frontier Oil Refinery, personal communication). Hence, in addition to the regulated effluent, Crow Creek may also be contaminated by ground water upstream from the NPDES discharge.

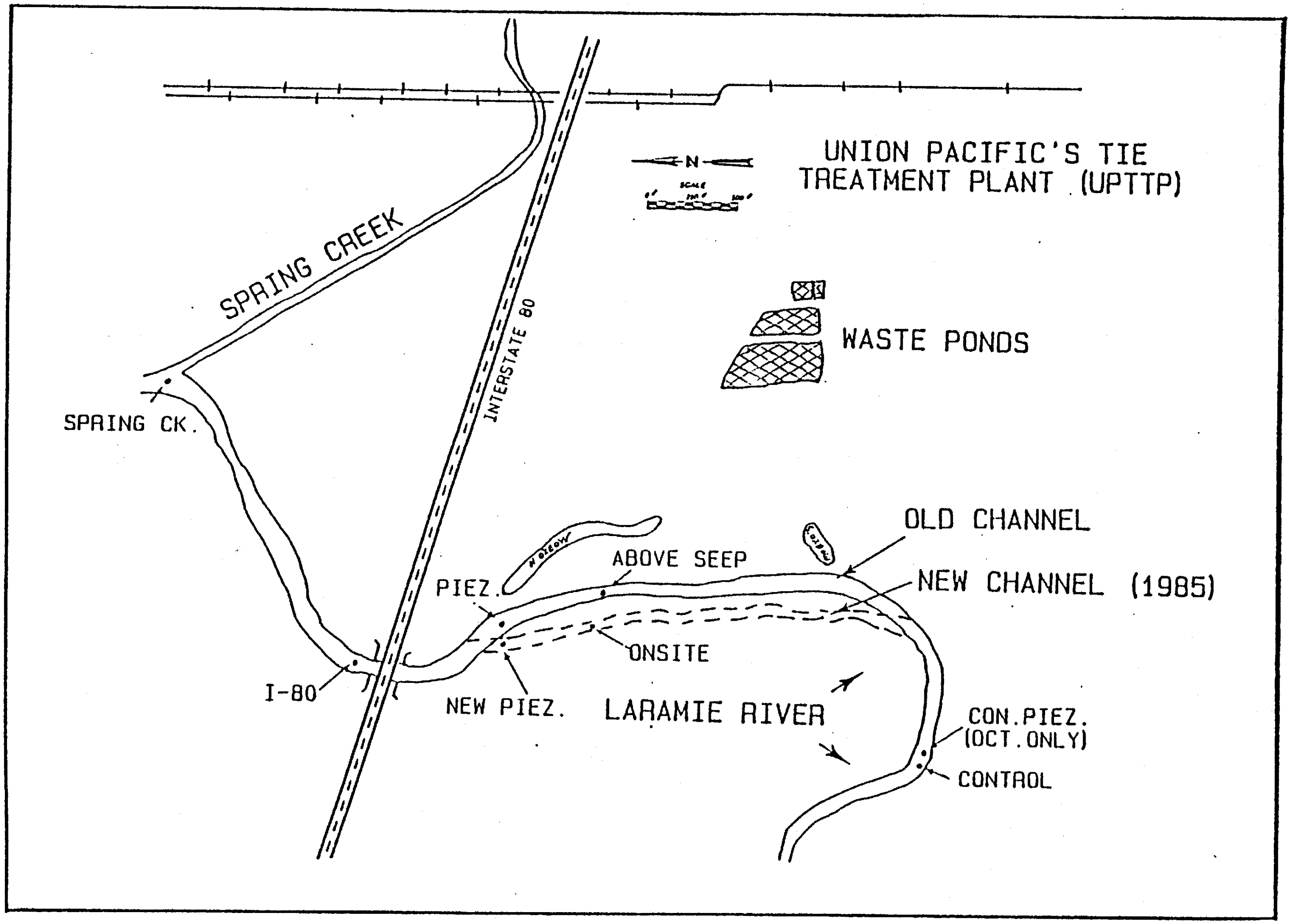


Figure 2. Laramie River sampling locations adjacent to the UPTTP. October samples were taken from the new channel; PIEZ. = interstitial water samples collected using mini-piezometer.

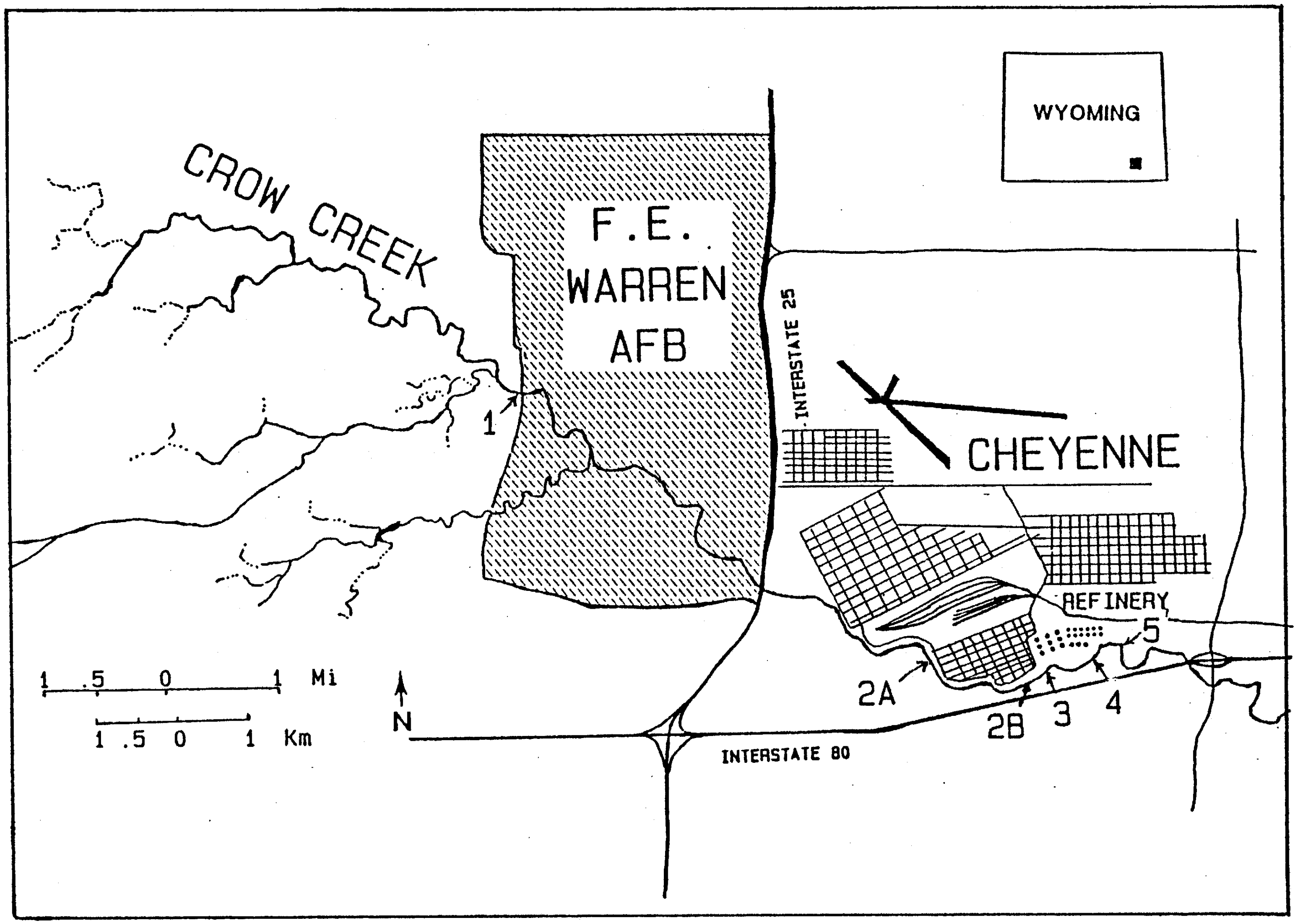


Figure 3. Location of the Crow Creek study site, Cheyenne, Wyoming. Numbers denote sampling locations: 1 = Upstream control, 2A = Optimist Park, 2B = Upstream Morrie Avenue, 3 = Morrie Avenue, 4 = Refinery, and 5 = NPDES.

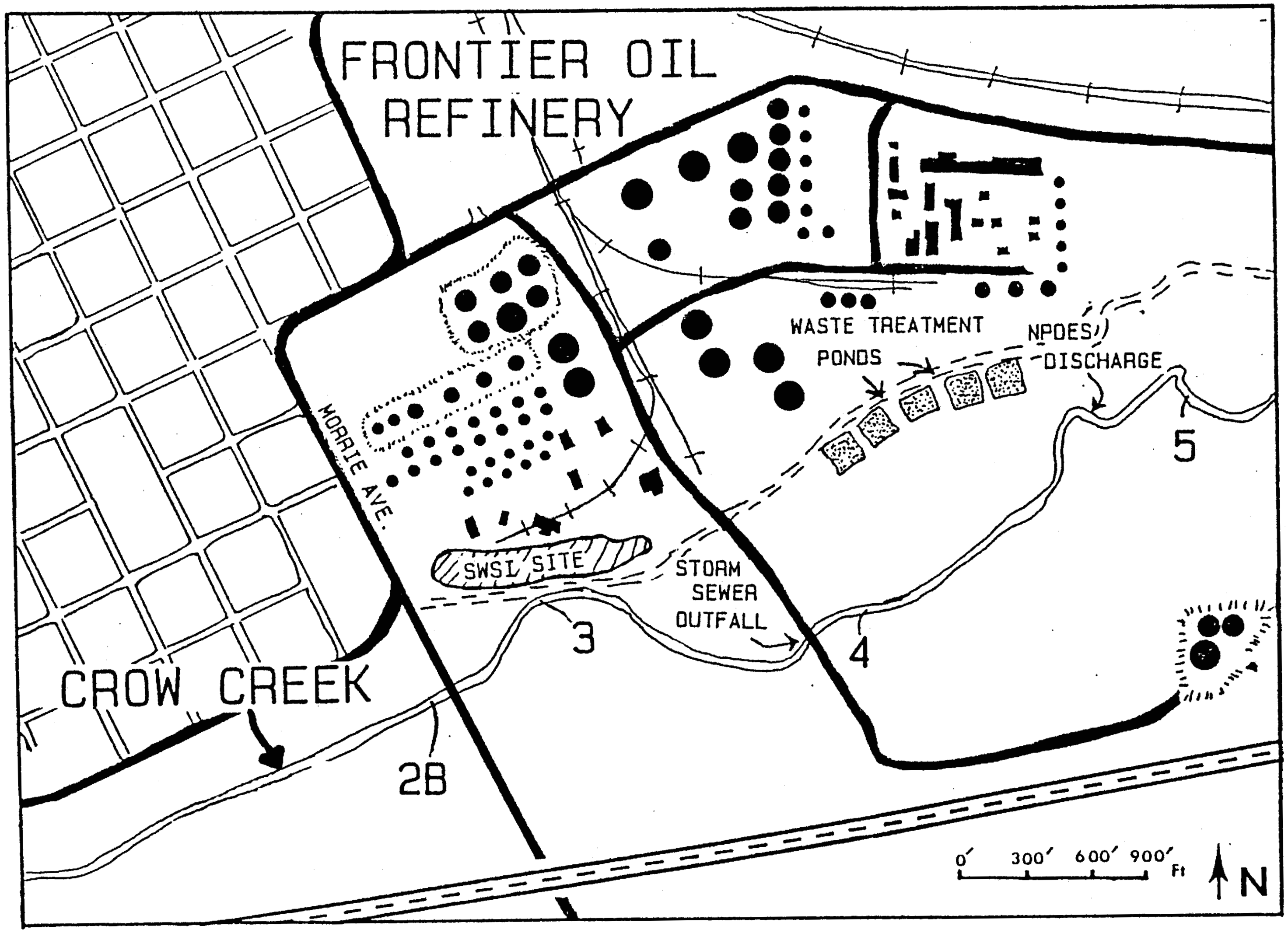


Figure 4. Crow Creek sampling locations adjacent to Frontier Oil Refinery. Numbers denote sampling locations: 2B = Upstream Morrie Avenue, 3 = Morrie Avenue, 4 = Refinery, 5 = NPDES. The Morrie Avenue site lies immediately adjacent to the buried waste ponds (SWSI Site).

On August 1, 1985, torrential hail and rain storms centered over downtown Cheyenne caused a major flood in Crow Creek. Peak stream flows during the flood were 65 m^3/s (2300 cfs) at the Interstate 25 bridge, approximately 5.8 km upstream from the oil refinery's NPDES discharge; 234 m^3/s (8260 cfs) at the Morrie Avenue bridge, approximately 0.9 km upstream from the NPDES discharge; and 211 m^3/s (7470 cfs) at the Interstate 80 bridge, approximately 1.2 km downstream from the NPDES discharge (Druse et al. 1986). For comparison, the flow rate of Crow Creek at Optimist Park on July 23, one week before the flood, was 0.10 m^3/s (3.6 cfs) and on August 7, one week after the flood, was 0.17 m^3/s (5.9 cfs; U.S. Geological Survey, Cheyenne, Wyoming, unpublished file data). Flood waters covered low-lying ground on the refinery property and abutted waste treatment ponds and the SWSI Site. Hence, groundwater flow probably increased considerably through the areas of heaviest surface and subsurface contamination at the refinery during and immediately after the flood.

Sample Collection, Preservation and Storage

Water samples from the Laramie River site were collected on June 14, July 18, August 9, and October 3, 1985. June, July and August samples were taken from the original river channel, whereas October samples adjacent to the UPTTP site were taken from corresponding locations in the new river channel (Figs. 1 and 2). On each sampling date, four river water samples and either one (in June, July and August) or two (in October) interstitial water samples were collected. The river water samples were (1) an upstream control approximately 0.5 km above the UPTTP site; (2) river water flowing directly over the location of the oil body that lay beneath the river sediments (or at a corresponding location in the new river channel in October); (3) approximately 0.5 km downstream from the oil body, at the Interstate 80 bridge; and (4) approximately 1.3 km downstream from the oil body, below the confluence of the Laramie River and Spring Creek, a small stream that flows through Laramie. Interstitial water was withdrawn from sediments using Teflon mini-piezometers (Lee and Cherry 1978) inserted 1 m below the riverbed, from which water was siphoned by a hand-operated vacuum pump. In June, July and August, the piezometer was placed approximately 30 m downstream from the river-water sampling location that was directly over the oil body (sample 2 described above), in order to avoid sampling creosote oil in the sediments. The two interstitial waters sampled in October were withdrawn from river sediments at (1) the upstream control location, and (2) in the new channel at the same location as the river water sample.

During Year 1 at Crow Creek, water samples were collected on June 13, July 17, August 20, October 24, and December 12, 1985 and February 24 and April 29, 1986. On each sampling date, five surface-water samples were collected at the following locations: 1) an upstream control above the Round Top Road bridge west of F.E. Warren Air Force Base; 2) at Optimist Park, approximately 8.1 km downstream from the control and immediately downstream from the Union Pacific Railroad yards; 3) 50 m below the Morrie Avenue bridge and adjacent to the upstream end of the SWSI site on the refinery property, approximately 10.1 km downstream from the control; 4) below the county road bridge midway along the refinery property, approximately 10.6 km downstream from the control; and 5) 50 m below the refinery's NPDES discharge pipe at the downstream end of the refinery property, approximately 11.0 km downstream from the control (Figs. 3 and 4).

During Year 2 at Crow Creek, water samples were collected on June 24, July 5, July 21, August 4, August 18, September 3, and September 18, 1986. On June

24 and July 5, surface-water samples were collected at the (1) upstream control, (2) Morrie Avenue bridge, (3) county road bridge midway along the refinery, and (4) NPDES discharge sampling locations described in the previous paragraph. Additionally, we withdrew interstitial water from sediments using mini-piezometers inserted 1 m below the creek bed at the Morrie Avenue, Refinery, and NPDES sampling locations. From July 21 to September 18 we included an extra sampling location approximately 50 m upstream from the Morrie Avenue bridge, where surface and ground water were collected in the same manner as at the other downstream sampling locations. That sampling location was added for comparison with the refinery sampling locations, because we anticipated that groundwater upstream from the refinery property would not be affected by surface and subsurface wastes at the refinery.

At each surface-water and groundwater sampling location, a 19-L grab sample was collected in a polyethylene jug, stored in a cooler, and returned to the University of Wyoming Red Buttes Environmental Biology Lab south of Laramie in < 2 h. Samples were refrigerated at $4^{o}C$ for subsequent toxicity tests. A 1-L aliquot was drawn from each sample for chemical analyses. Subsamples for cation and trace element analyses were filtered (0.45 µm cellulose acetate filter) and acidified with redistilled HNO_3 (1 ml/L). Subsamples for anion analyses, dissolved organic carbon, and high performance liquid chromatography were filtered (0.45 µm cellulose acetate filter) and refrigerated at $4^{o}C$.

Toxicity Tests

Test Organisms. Stock cultures of fathead minnows (*Pimephales promelas*) and *Ceriodaphnia dubia* (an aquatic invertebrate) were originally obtained from the U.S. EPA Environmental Research Lab in Duluth, Minnesota, and have been cultured at the Red Buttes Environmental Biology Lab for several years.

Neonates from those stock cultures were used for all toxicity tests conducted in this study. Fathead minnow adults are maintained in our laboratory at 25°C in brood tanks that contain plastic spawning tiles. Brood tanks are checked daily for newly fertilized eggs, which adhere to the undersides of tiles. One day prior to beginning a test, tiles to which unhatched eggs are attached are placed in a basin containing clean culture water. The following day, fathead minnow fry that have hatched (and thus are < 24 h old) are removed from the basin and used to begin a toxicity test.

Ceriodaphnia are cultured at 25°C in our laboratory in glass dishes. These brood cultures are transferred to fresh water three times per week. Four h before a test is started, adult *Ceriodaphnia* in a brood culture are transferred to clean water and the remaining young are discarded. Offspring born to the transferred adults within the next 4 h are removed from the culture dish and used to begin the toxicity test.

Ambient Toxicity Tests. To test instream toxicity, we adopted two short-term, sublethal tests that were recently developed by the U.S. EPA for assessing effects of industrial and municipal effluents. The 7-d survival and growth test using fathead minnows and the 7-d survival and reproduction test using *Ceriodaphnia dubia* were first described by Mount and Norberg (1984) and Norberg and Mount (1985). Standardized protocols for these tests have been published more recently by Horning and Weber (1985).

In ambient toxicity tests, water samples are not serially diluted as is done in traditional toxicity tests. Instead, the water sample is tested as collected, and upstream water serves as an "ambient control" for the sample of interest. Additionally, laboratory water is used as a "laboratory control" in case no test organisms survive in any instream sample. However, statistical comparisons of downstream water samples are made only to the upstream control, since the objective of the ambient test in a regulatory framework often is to determine if the influx of a pollutant source changes the response of the test organisms relative to the upstream control. Pairwise comparisons between all possible combinations of sampling locations are sometimes also made, in order to test whether water at a given sampling location causes adverse or beneficial effects relative to the sampling location immediately upstream.

Temperature for all 7-d fathead minnow and *Ceriodaphnia* tests was maintained at 25°C using recirculating water baths, and tests were run under a 16-h light/8-h darkness photoperiod.

We conducted fathead minnow tests using 1-L glass beakers that contained 500 ml of test water. Either two or four replicate beakers were tested for each water sample and the laboratory control. We began the study using two replicates per sample, as was the practice at several other laboratories at that time (Horning and Weber 1985 recommend a minimum of two replicates per sample). However, we expanded to four replicates per sample after August 1985 when we discovered that our ability to resolve significant differences among treatments was low and that variances often were not homogeneous using only two replicates.

To begin a test, ten neonate fathead minnow larvae were placed in each beaker. Beakers were covered with watch glasses to decrease sample evaporation, and the fish were fed 0.1 ml (approximately 700-1000 shrimp) of a concentrated suspension of newly hatched brine shrimp (*Artemia salina*) three times per day. The number of live larvae in each beaker was recorded every 24 h, and dead larvae were removed at that time. Additionally, all but approximately 75 ml of the exposure solution was siphoned daily out of each beaker to remove feces and uneaten brine shrimp. Then, 500 ml of fresh exposure water warmed to 25°C was gently poured into the beakers. At the end of Day 7, all surviving fathead minnows in each beaker were placed as a group into a small aluminum weighing boat and oven dried at 100°C. Dry weight of each group of fish (biomass in each replicate beaker) was determined on a Sauter electronic micro-balance. Finally, endpoints of survival and growth (average dry weight per fish) in downstream waters were compared to the upstream control as described below in the Statistical Analyses section.

We conducted Ceriodaphnia tests using 30-ml plastic cups that contained 15 ml of test water. Ten replicate beakers were tested for each water sample and the laboratory control. To begin a test, one neonate Ceriodaphnia was placed in each beaker. Beakers were covered with watch glasses to decrease sample evaporation, and a yeast/algae suspension (50 µl, containing approximately 10^6 algal cells and 200 µg of yeast) was added to each beaker once a day as food for the Ceriodaphnia. Survival and reproduction were monitored every 24 h; and on Days 3 and 5, live adults were transferred to beakers containing fresh exposure water. Ceriodaphnia usually begin reproducing by Day 4 of a test and have at least three broods by the end of Day 7. Tests were terminated on Day 7, and endpoints of survival and reproduction (average number of offspring/female) in downstream waters were compared to the upstream control as described below in the Statistical Analyses section.

The reproduction endpoint in Ceriodaphnia tests can be computed two ways (Hamilton 1986). MOA (mean overall) reproduction is the traditional measure of average number of offspring produced per female, and is calculated by dividing the total number of offspring produced in a test by the number of females that started the test. If a female dies before reproducing, her offspring total is recorded as zero. Standard errors of the estimate of total reproduction are computed by the usual method for estimating a mean value (Hamilton 1986). MIM (mean ignoring mortality) reproduction calculations isolate reproductive effects of a toxicant from survival effects. To estimate the MIM value, the mean number of offspring produced per live adult is computed for each day of the test. Then those means are summed over all seven days of the test to compute average total reproduction. An adult death is treated mathematically as though it occurred halfway through that 24-h period. Standard errors of the estimate of total reproduction are computed using a Bootstrap technique (Hamilton 1986), which requires high-speed computer calculations.

The MOA statistic integrates both survival and reproduction into an estimate that is more interpretable as a population-level response to a toxicant, whereas the MIM statistic subtracts out the effects of differential survival and is more interpretable as an organism-level, physiological/reproductive response to a toxicant. Both measures were computed in this study and the results are compared in this report. To compute these statistics, we used BSVAR, a computer program available from Dr. John Rodgers at the U.S. EPA Environmental Research Lab in Duluth, Minnesota.

Acute Toxicity Tests. Acute toxicity tests using fathead minnows and *Ceriodaphnia dubia* were performed for Laramie River interstitial water collected in June 1985 and for Crow Creek water collected below the oil refinery NPDES discharge in June and July 1985. All tests followed ASTM (1980) standard practices and were conducted at 25°C under a 16-h light/8-h darkness photoperiod. Fathead minnows were tested for 96 h in 80 ml of test solution contained in a 150-ml glass beaker, whereas *Ceriodaphnia* were tested for 48 h in 15 ml of test solution contained in a 30-ml plastic cup. Exposure concentrations were 10, 18, 32, 56 and 100% of the full-strength water sample; in addition, a laboratory control was tested. All exposure concentrations <

100% were diluted with laboratory control water (Table 1).

Acute toxicity test procedures for both species were similar. To begin a test, 10 neonate fathead minnows or Ceriodaphnia were placed in each of three replicate beakers for each exposure level and the control. Beakers were covered with watch glasses to decrease sample evaporation, and no food was added during the test. Every 24 h, survival of test animals was observed, dead animals were removed from the beakers, and live animals were transferred to fresh exposure solutions. At the end of each test, percentage survival at each exposure level was computed. LC50 (median lethal concentration) values were then calculated using the trimmed Spearman-Karber method (Hamilton et al. 1977) and expressed as percent of full-strength ground water or surface water. [Note that as LC50 values decrease, toxicity of the water increases.]

Chemical Analyses

Routine chemical parameters, including temperature, pH, conductivity, alkalinity, hardness and total ammonia, were analyzed at the Red Buttes Environmental Biology Lab by standard methods (APHA 1980). Methods for these measurements were (1) pH using a Corning Model 10 pH meter, (2) conductivity using a Extech Model 440 conductivity meter, (3) alkalinity and hardness by titration, and (4) total ammonia using an Orion Ionalyzer Model 407A equipped with a selective-ion ammonia probe. Free ammonia (NH_3) was computed from temperature, pH and total ammonia values using equilibrium calculations described by Emerson et al. (1975).

Major inorganic cations (Na^+, K^+, Ca^{2+}, Mg^{2+} and Sr^{2+}) were analyzed by the University of Wyoming Plant Sciences Department, using a Perkin-Elmer Model 5500 inductively coupled argon plasma emission spectrophotometer (ICP). Major inorganic anions (Cl^-, NO_3^-, F^- and SO_4^{2-}) were analyzed at the Red Buttes

Table 1. Quality of dilution water used for fathead minnow (*Pimephales promelas*) and *Ceriodaphnia dubia* acute toxicity tests in June and July 1985.

Parameter	Filtered fathead minnow tank water[a]	50% raceway/ 50% deionized water
pH (units)	8.3 (7.8 - 8.5)	8.4
Conductivity (µS/cm at 25°C)	498 (487 - 514)	255
Alkalinity (mg/L as $CaCO_3$)	197 (178 - 208)	164
Hardness (mg/L as $CaCO_3$)	254 (238 - 265)	230

[a]Values expressed as means for five acute toxicity tests; ranges of values presented in parentheses.

Environmental Biology Lab using a Dionex Model 2110i ion chromatograph equipped with an electrical conductivity detector. The carrier eluant was a 0.0025 N Na_2CO_3/0.0027 N $NaHCO_3$ buffer. We computed NH_4^+ concentrations by subtracting free ammonia from total ammonia, and we computed HCO_3^- and CO_3^{2-} concentrations from temperature, pH and alkalinity values using equilibrium calculations described by Drever (1982).

Dissolved concentrations of eleven inorganic trace elements (Al, As, Cd, Cr, Cu, Fe, Hg, Ni, Pb, Se and Zn) were also analyzed by the UW Plant Sciences Department using ICP. Because ICP methods have relatively high detection limits (10-100 μg/L), we additionally analyzed Cd, Cr, Cu and Zn using a Perkin-Elmer Model 2380 atomic absorption spectrophotometer (AA) at the Red Buttes Environmental Biology Lab. AA analyses are considerably more expensive and time-consuming than ICP analyses, but they allowed us to obtain detection limits of 0.1 μg/L for Cd and Zn and 1.0 μg/L for Cr and Cu. We expected that concentrations of these four elements might be high in some samples and that they might contribute to instream toxicity.

Organic analyses were performed at three levels of resolution. Dissolved organic carbon (DOC) concentrations were determined using an Oceanography International organic carbon analyzer equipped with an infrared CO_2 detector, located in the UW Geology Department. Reverse-phase high performance liquid chromatography (HPLC) gradients of water samples were run on a Waters Model 402 HPLC equipped with ultraviolet and fluorescence detectors, located at the Red Buttes Environmental Biology Lab. HPLC samples (50 μl) were injected onto a C_{18} Radial Pak column and eluted over a 30-min run, using a linear gradient from 100% H_2O to 100% CH_3CN at 2.0 ml/min. Blank gradients (no injection) and a standard mixture containing several aromatic hydrocarbons were also run for comparison with the test samples. Finally, the Laramie River interstitial water collected in June 1985 was analyzed using gas chromatography/mass spectrometry (GC/MS) by Rocky Mountain Analytical Lab in Denver, Colorado. Because of the high cost of GC/MS scans compared to DOC and HPLC analyses, only the Laramie River interstitial water collected in June 1985 was analyzed by this method.

Statistical Analyses

Horning and Weber (1985) recommend that results of *Ceriodaphnia* survival tests be analyzed by Fisher's Exact Test and that results of fathead minnow survival and growth tests and *Ceriodaphnia* reproduction tests be analyzed by Analysis of Variance (ANOVA). Fisher's Exact Test is based on the assumption of a multinomial distribution of mortalities (Sokal and Rohlf 1981). Since it provides a conservative estimate of the probability associated with a difference in survival between two treatments (Horning and Weber 1985) we adopted the assumption of a multinomial distribution for these *Ceriodaphnia* survival data without testing. However, ANOVA is based on two more sensitive assumptions -- normality of data and homogeneity of variances. We tested both of these assumptions, as described below, before proceeding with ANOVA or a more appropriate statistical method.

Normality. Data from fathead minnow survival and growth tests and *Ceriodaphnia* MOA reproduction tests were tested for normality using a Chi-Square Goodness of Fit Test at $\alpha = 0.01$ (Horning and Weber 1985). The assumption of normally distributed data was rejected in only two data sets (August 1985 Laramie River and July 21, 1986 Crow Creek *Ceriodaphnia* MOA reproduction). For those data, we performed the following nonparametric statistical tests: 1) Steel's Many-One Rank Test at $\alpha = 0.05$ (Horning and Weber 1985) to test for decreased reproduction at downstream locations relative to the upstream control, and 2) the Kruskal-Wallis Test followed by nonparametric multiple comparisons of treatment pairs at $\alpha = 0.05$ (Sokal and Rohlf 1981) to test for significant differences in reproduction between any two sampling locations. Steel's Test and the Kruskal-Wallis Test are analogous, respectively, to the parametric one-tailed and two-tailed tests for differences among means that are described below.

Homogeneity of Variances. Homogeneity of variances was tested using Bartlett's Test at $\alpha = 0.01$ (Horning and Weber 1985) for all fathead minnow survival and growth and *Ceriodaphnia* MOA reproduction data sets in which the assumption of normally distributed data could not be rejected. Although ANOVA is relatively robust to non-homogeneity of variances, Milliken and Johnson (1984) suggest using paired *t* tests instead of ANOVA when the null hypothesis of homogeneous variances can be rejected at $\alpha = 0.01$. Therefore, we did not use ANOVA to analyze results when (1) variances were significantly non-homogeneous (October 1985 and February, April, June, July 21, August and September 1986 Crow Creek fathead minnow survival tests; June, July and October 1985 Laramie River and June, July, October and December 1985 and February, June, July 21, August and September 1986 Crow Creek *Ceriodaphnia* MOA reproduction tests); or (2) when $\max_i\{var_i\}/\min_i\{var_i\} > 100$ in fathead minnow tests using only 2 replicate beakers per sample (June, July and August 1985 Laramie River samples and July 1985 Crow Creek samples), because of the large differences in variances that could obscure otherwise significant differences between two treatments with low variances. In those cases, we used paired *t* tests based on the following statistic to compare treatment means (Milliken and Johnson 1984):

$$t^* = \frac{\overline{X}_1 - \overline{X}_2}{\sqrt{[s.e.(\overline{X}_1)]^2 + [s.e.(\overline{X}_2)]^2}}$$

where $\overline{X}_1$ = mean value for sampling location 1, $\overline{X}_2$ = mean value for sampling location 2, s.e.($\overline{X}_1$) = standard error of the estimate of $\overline{X}_1$, and s.e.($\overline{X}_2$) = standard error of the estimate of $\overline{X}_2$.

For sampling dates on which variances were non-homogeneous among the sampling locations, survival or reproduction at a downstream location was judged to be significantly less than survival or reproduction in the upstream control when $\underline{t}^*$ was greater than Dunnett's one-tailed critical value at α = 0.05 (Dunnett 1964). Dunnett's Method is a post hoc multiple comparison procedure designed only for comparisons of a control with several treatments, in which a specified overall confidence level (1 - α) is to be maintained for a family of non-independent comparisons (Dunnett 1955).

Additionally, we wanted to compare any given sampling location to any other sampling location on the same sampling date. For that analysis, survival or reproduction was judged to be significantly different between any two sampling locations when the absolute value of t^* was greater than the following two-tailed critical value for Tukey's HSD Method at α = 0.05 (Neter et al. 1985):

$$T = \frac{q_{[0.05;r,N-r]}}{\sqrt{2}}$$

where q = tabulated value of the studentized range, r = total number of treatments plus control, and N = total number of replicates tested. Tukey's HSD Method is a post-hoc multiple comparison procedure designed for all possible pairwise comparisons among treatments, in which a specified overall confidence level (1 - α) is to be maintained for a family of non-independent comparisons.

Dunnett's Method and Tukey's Method were originally designed for use with ANOVA, but their critical values can also be used as approximate critical values for the paired $\underline{t}$ tests described above. ANOVA methods used for tests in which the homogeneity assumption could not be rejected are described below.

Survival. Because only 10 *Ceriodaphnia* (10 replicate beakers x 1 animal/beaker) were tested per sample, their 7-d survival in Laramie River and Crow Creek waters was analyzed using Fisher's Exact Test (Horning and Weber 1985). We also used Fisher's Exact Test to analyze 7-d survival in fathead minnow tests in which only 20 animals (2 replicate beakers x 10 animals/beaker) were tested (all Laramie River samples and June, July and August 1985 Crow Creek samples). Survival at a downstream location was judged to be significantly less than survival in the upstream control when the one-tailed probability associated with that comparison was < 0.05. Likewise, for all possible pairwise comparisons among sampling locations, survival at a given sampling location was judged to be significantly different than survival at another sampling location when the two-tailed probability associated with that pairwise comparison was < 0.05 (Sokal and Rohlf 1981).

For fathead minnow tests in which 40 animals (4 replicate beakers x 10 animals/beaker) were tested and the assumptions of normality and homogeneity could not be rejected (December 1985 and July 5, 1986 Crow Creek samples), we compared 7-d survival using MINNOW, a statistical package for analyzing fathead minnow survival and growth tests that was programmed for IBM personal computers by Dr. Jeffrey Giddings of Oak Ridge National Laboratory (ORNL). Briefly, that program computes an ANOVA on arcsine-square-root-transformed percent survival data and then tests for decreased survival in downstream waters relative to the upstream control, using Dunnett's one-tailed critical values at $\alpha = 0.05$. We also tested all possible pairwise comparisons of survival among sampling locations, using the same arcsine-square-root-transformed data and two-tailed critical values for Tukey's HSD Method at $\alpha = 0.05$.

Reproduction. MOA total numbers of offspring produced by *Ceriodaphnia* were analyzed by ANOVA computed on untransformed data when the assumptions of normality and homogeneity could not be rejected (August 1985 and April and July 5, 1986 Crow Creek samples). We then tested for (1) decreased MOA reproduction in downstream waters relative to the upstream control using Dunnett's one-tailed critical values at $\alpha = 0.05$, and (2) differences in MOA reproduction among all possible pairs of treatments using Tukey's HSD two-tailed critical values at $\alpha = 0.05$. Since means and standard errors of estimates of MIM total numbers of offspring were estimated using a Bootstrap procedure, ANOVA comparisons were not possible for MIM total reproduction on any sampling date. Therefore, we performed paired *t* tests between all possible pairs of sampling locations and compared those t^* values to Dunnett's one-tailed critical values at $\alpha = 0.05$ and Tukey's HSD two-tailed critical values at $\alpha = 0.05$. If all *Ceriodaphnia* females died in water from a given sampling location, that location was not included in the post hoc comparison of MIM total reproduction for that sampling date.

Growth. Seven-day fathead minnow weights were analyzed for all sampling dates using the ORNL computer program, MINNOW. It computes an ANOVA on untransformed weights and then tests for decreased growth in downstream waters relative to the upstream control, using Dunnett's one-tailed critical values at $\alpha = 0.05$. Two-tailed comparisons of growth between all possible pairs of treatments were made using Tukey's HSD Method at $\alpha = 0.05$. If all fathead minnow larvae died in water from a given sampling location, that location was not included in the post hoc comparison of growth.

Ammonia-Toxicity Correlations. Because low survival, growth and reproduction usually occurred in waters with high concentrations of unionized ammonia (NH_3), we correlated NH_3 concentrations versus fathead minnow survival and growth and *Ceriodaphnia* survival and MOA reproduction, in order to test associations between those variables. Non-parametric Spearman rank correlations (Sokal and Rohlf 1981) were performed for those four associations using the NONPAR CORR routine in the Statistical Package for the Social Sciences (Nie et al. 1975), since the relationships of interest appeared not to be linear.

RESULTS

Laramie River Study Site

Toxicity Tests. All fathead minnow larvae and *Ceriodaphnia* tested in the interstitial water on June 14, 1985 died in < 24 h (Figs. 5 and 7 and Appendix Table A-1). That interstitial water was collected from a mini-piezometer inserted 1 m deep in river sediments adjacent to the UPTTP, 30 m downstream from the creosote oil body. Fathead minnow survival and growth and *Ceriodaphnia* survival and reproduction at all other sampling locations on June 14, 1985 were not significantly lower than in the upstream control (Figs. 5, 6, 7 and 8 and Appendix Table A-1), although there was a trend toward decreased fathead minnow survival in river water collected directly over the oil body and downstream at the I-80 bridge. Additionally, survival, growth and reproduction at all of those locations was significantly greater than in the interstitial water (Appendix Table A-2). Acute lethality tests conducted on the interstitial water on June 14, 1985 showed the 96-h LC50 (median lethal

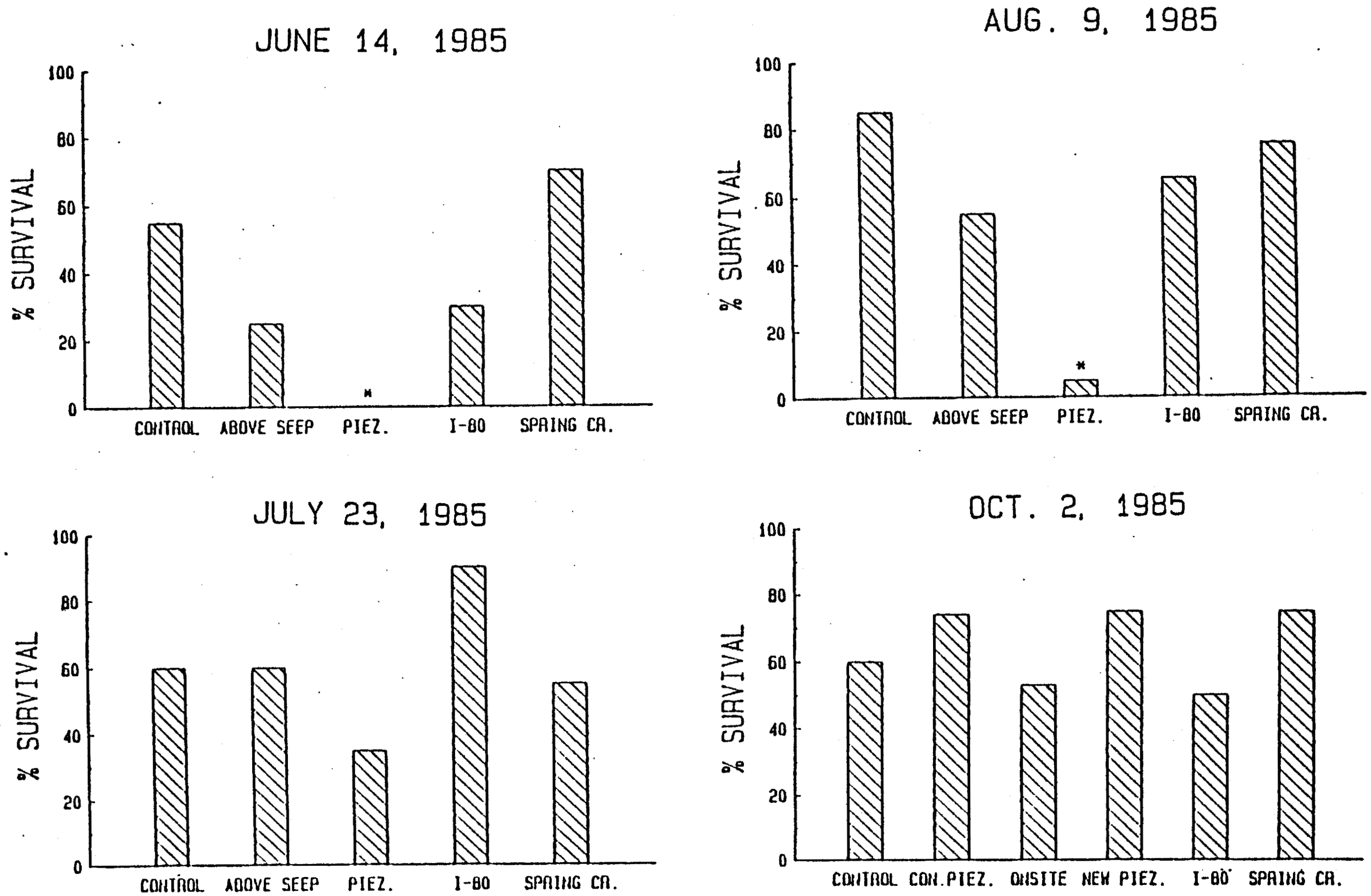


Figure 5. Fathead minnow (Pimephales promelas) survival in Laramie River water and interstitial water (PIEZ.) from June to October 1985. * = significantly lower survival than upstream control ($P < 0.05$).

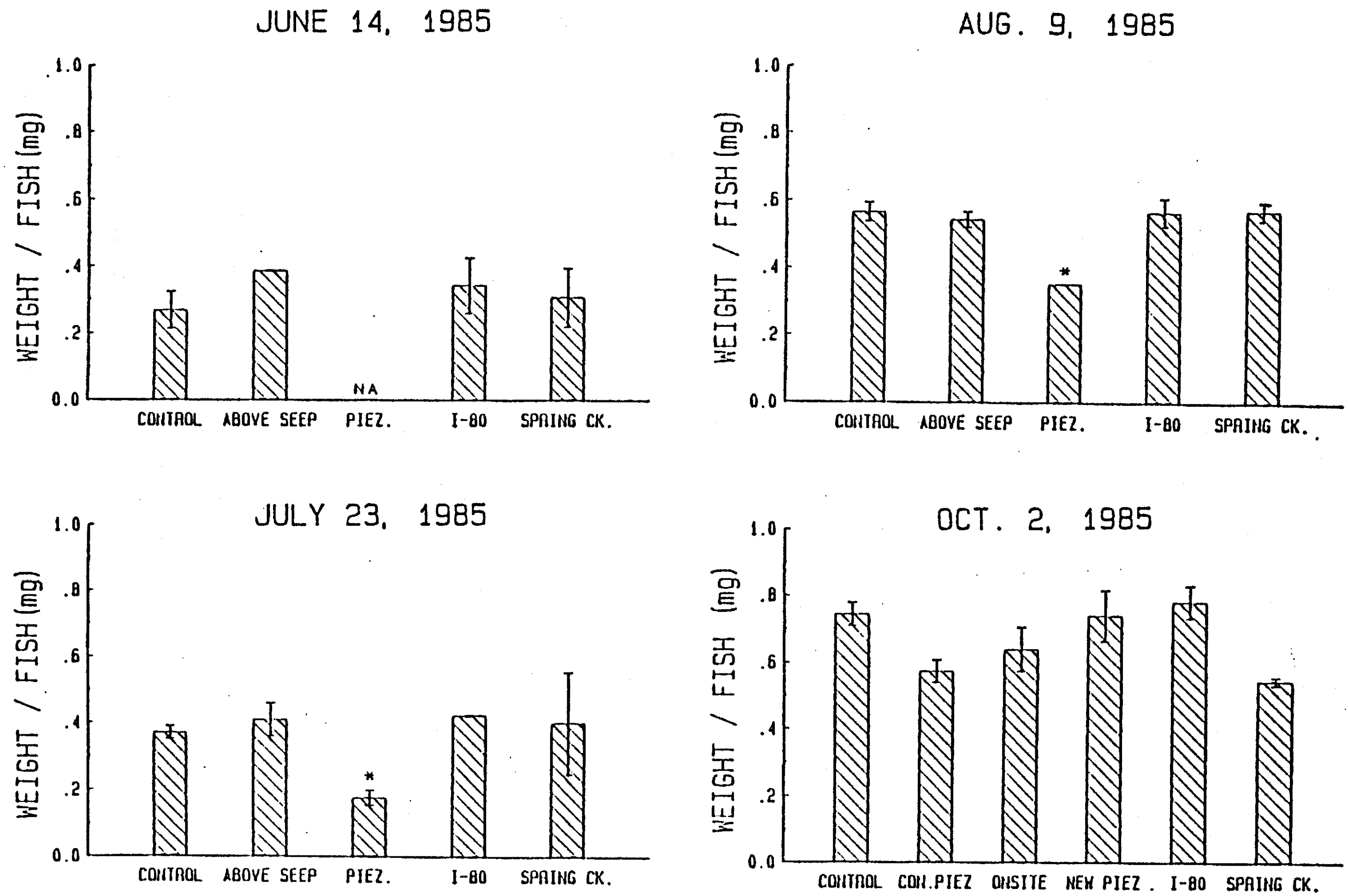


Figure 6. Fathead minnow (Pimephales promelas) growth (mean ± one standard error of the mean) in Laramie River water and interstitial water (PIEZ.) from June to October 1985. NA = value could not be calculated because all larvae died; * = significantly lower weight/fish than upstream control ($P < 0.05$).

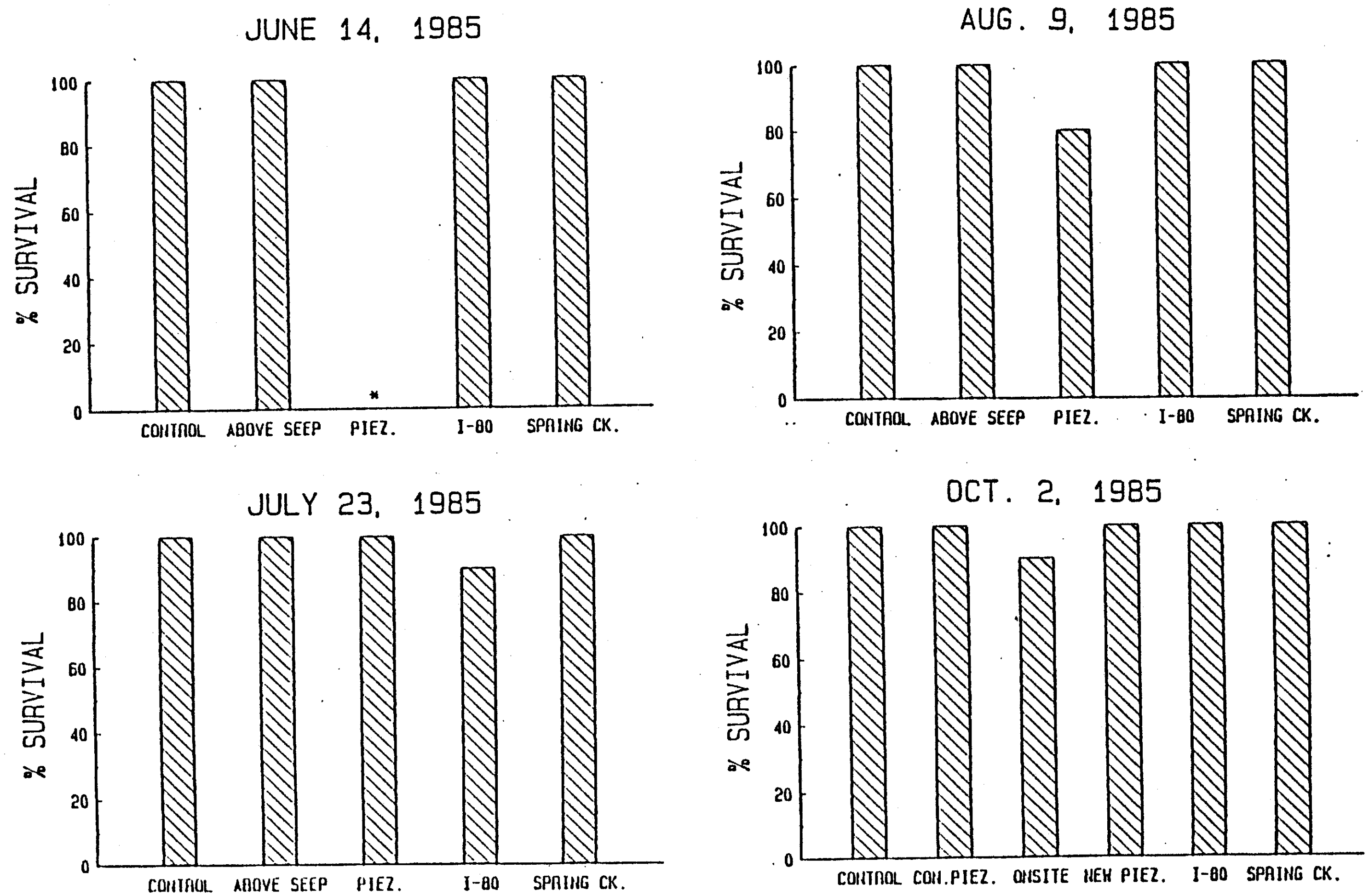


Figure 7. Ceriodaphnia dubia survival in Laramie River water and interstitial water (PIEZ.) from June to October 1985. * = significantly lower survival than upstream control ($P < 0.05$).

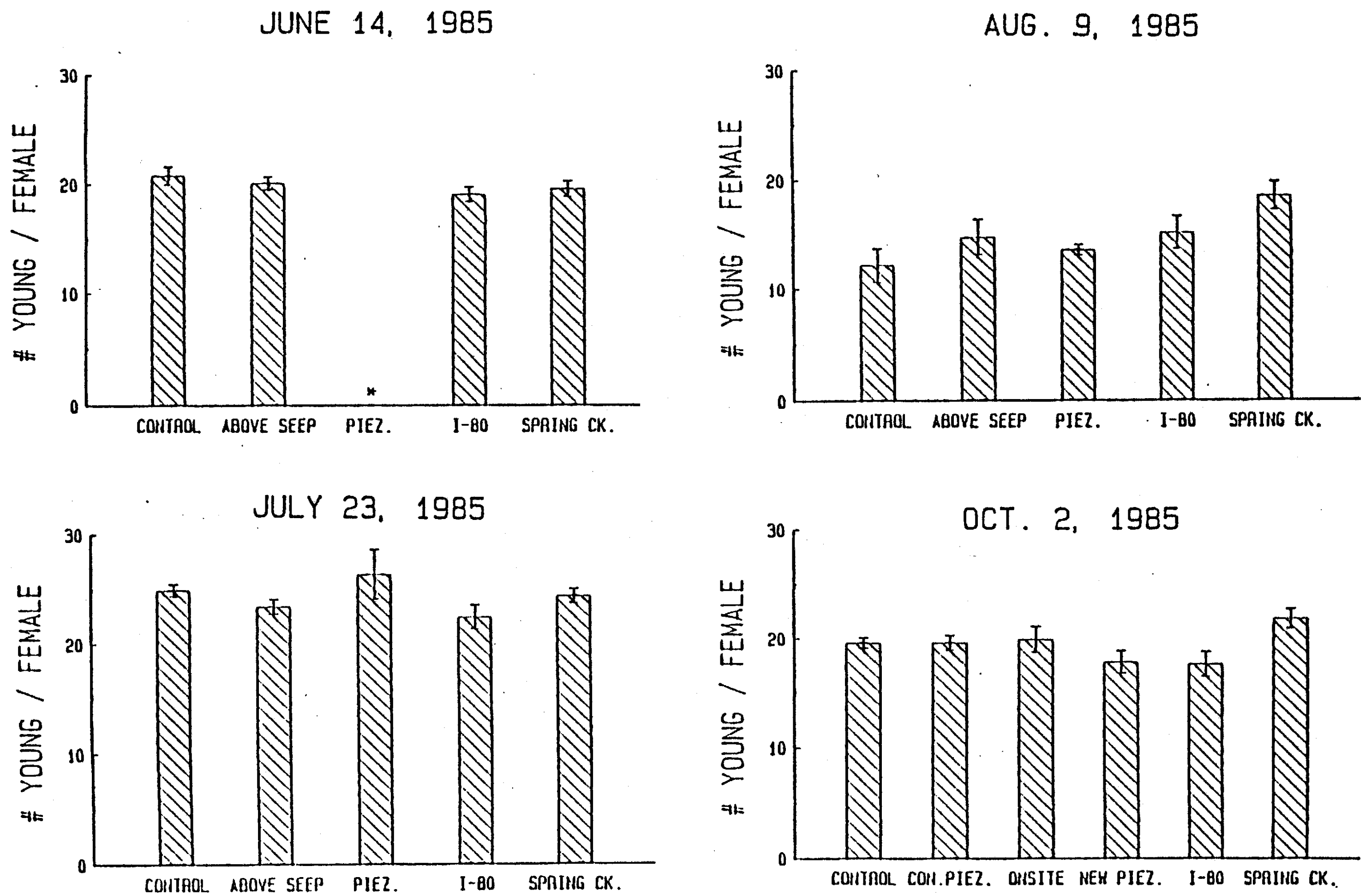


Figure 8. Ceriodaphnia dubia MOA reproduction (mean ± one standard error of the mean) in Laramie River water and interstitial water (PIEZ.) from June to October 1985. * = significantly lower # young/female than upstream control ($P < 0.05$).

concentration) for fathead minnows was 20% of full-strength interstitial water and the 48-h LC50 for Ceriodaphnia was 18% of full-strength interstitial water.

On July 18, 1985, fathead minnow growth in interstitial water was significantly lower than in the upstream control and all other downstream sampling locations except below the Spring Creek confluence; and on August 9, 1985, fathead minnow survival and growth in interstitial water were also significantly lower than in the upstream control and all other downstream sampling locations (Figs. 5 and 6 and Appendix Tables A-1 and A-2). Relative to the upstream control, Ceriodaphnia survival and reproduction were not adversely affected at any sampling location on July 18 and August 9, 1985 (Figs. 7 and 8 and Appendix Table A-1). However, Ceriodaphnia MOA reproduction below the Spring Creek confluence was significantly greater than in the interstitial water on August 9, 1985, and MIM reproduction below the Spring Creek confluence was significantly greater than in the upstream control and interstitial water on August 9, 1985 (Appendix Table A-2). After the Laramie River was rechanneled in September 1985, fathead minnow survival and growth and Ceriodaphnia survival and reproduction were not adversely affected in interstitial and river waters collected at corresponding locations in the new river channel adjacent to the tie treating plant (Figs. 5, 6, 7 and 8 and Appendix Tables A-1 and A-2).

Chemical Analyses. Routine water chemistry parameters and major inorganic ions at all Laramie River sampling locations were within normal ranges and were similar to the upstream control (Table 2 and Appendix Tables B-1 and B-2). Of the 11 trace elements analyzed, concentrations of Al, As, Cd, Fe, Hg, Ni, Pb and Se remained relatively low and did not vary considerably (Table 2 and Appendix Table B-3). Most concentrations of chromium and copper also were low;

Table 2. Ranges of values for chemical parameters measured in Laramie River water and interstitial water from June 1985 to October 1985 and in Crow Creek water and interstitial water from June 1985 to September 1986.[a]

Parameter[b]	Range of values	
	Laramie River	Crow Creek
Routine chemical parameters		
pH (units)	7.6 - 8.4	7.4 - 8.6
Conductivity (μS/cm at 25°C)	607 - 1220	347 - 1604
Alkalinity (as $CaCO_3$)	103 - 172	134 - 438
Hardness (as $CaCO_3$)	230 - 499	95 - 654
Ammonia, total (as N)	< 0.10	< 0.10 - 12.0
Ammonia, unionized (as NH_3)	< 0.01	< 0.01 - 0.90
DOC	2.1 - 32.9	2.0 - 24.9
Major inorganic ions		
Na^+	38 - 86	12 - 122
Ca^{2+}	67 - 137	52 - 265
Mg^{2+}	10 - 38	4 - 32
K^+	2.6 - 5.8	4 - 26
Sr^{2+}	0.5 - 1.2	0.2 - 1.2
NH_4^+	< 0.13	< 0.13 - 14.8
Cl^-	7 - 48	7 - 168
SO_4^{2-}	171 - 514	14 - 308
NO_3^-	< 1.0	< 0.1 - 18.5
F^-	0.4 - 0.8	0.4 - 10.9
HCO_3^-	125 - 206	161 - 532
Trace elements		
Al	< 0.1	< 0.1 - 0.2
As	< 0.1	< 0.1
Cd	0.0007 - 0.0035	0.0001 - 0.0021
Cr	< 0.001 - 0.0139	< 0.001 - 0.0399
Cu	< 0.001 - 0.0261	< 0.001 - 0.0417
Fe	0.01 - 0.15	< 0.01 - 0.22
Hg	< 0.1	< 0.1
Ni	< 0.01	< 0.01 - 0.04
Pb	< 0.1	< 0.1
Se	< 0.1	< 0.1
Zn	0.0003 - 0.0187	< 0.0001 - 0.44

[a]See Appendix Tables B-1 to B-6 for detailed listings of values.

[b]Values expressed as mg/L, unless otherwise noted.

however, Cr concentration was 0.0139 mg/L in the Laramie River below its confluence with Spring Creek on October 3, 1985, and Cu concentration was 0.0261 mg/L in the Laramie River at the I-80 bridge on June 14, 1985. Zn concentrations varied considerably (Table 2) and were > 0.005 mg/L at least once at each sampling location.

Low concentrations of anthracene, phenanthrene and chrysene were detected in reverse-phase HPLC gradients of the toxic interstitial water in June 1985 (Fig. 9(a)). These three organic compounds were also the three highest peaks detected in reverse-phase HPLC gradients of the creosote oil that underlaid the river sediments (Fig. 9(b); see also Crossey and Bergman 1985). However, organics were not detected in interstitial water using HPLC on the other sampling dates. And no priority pollutants were detected in GC-MS analyses of the base-neutral fraction of interstitial water collected in June 1985.

Crow Creek Study Site

Toxicity Tests. All fathead minnow larvae and Ceriodaphnia tested in Crow Creek water below the oil refinery's NPDES discharge on June 13, July 17, and October 24, 1985 and February 24, July 21, August 4, August 18, September 3, and September 18, 1986 died by Day 7 of the tests (Figs. 10 and 12 and Appendix Table A-3). On June 13, 1985 the 96-h LC50 of Crow Creek water below the NPDES discharge was 53% for fathead minnows, and the 48-h LC50 for Ceriodaphnia was between 56% and 100%. Corresponding July 17, 1985 LC50 values were 26% for fathead minnows and 73% for Ceriodaphnia. Additionally, all Ceriodaphnia died in Crow Creek water below the NPDES discharge on December 12, 1985, and fathead minnow survival in Crow Creek water below the NPDES discharge was significantly less than in the upstream control on December 12, 1985 and June 24, 1986 (Figs. 10 and 12 and Appendix Table A-3). On August 20, 1985 and April 29 and July 5,

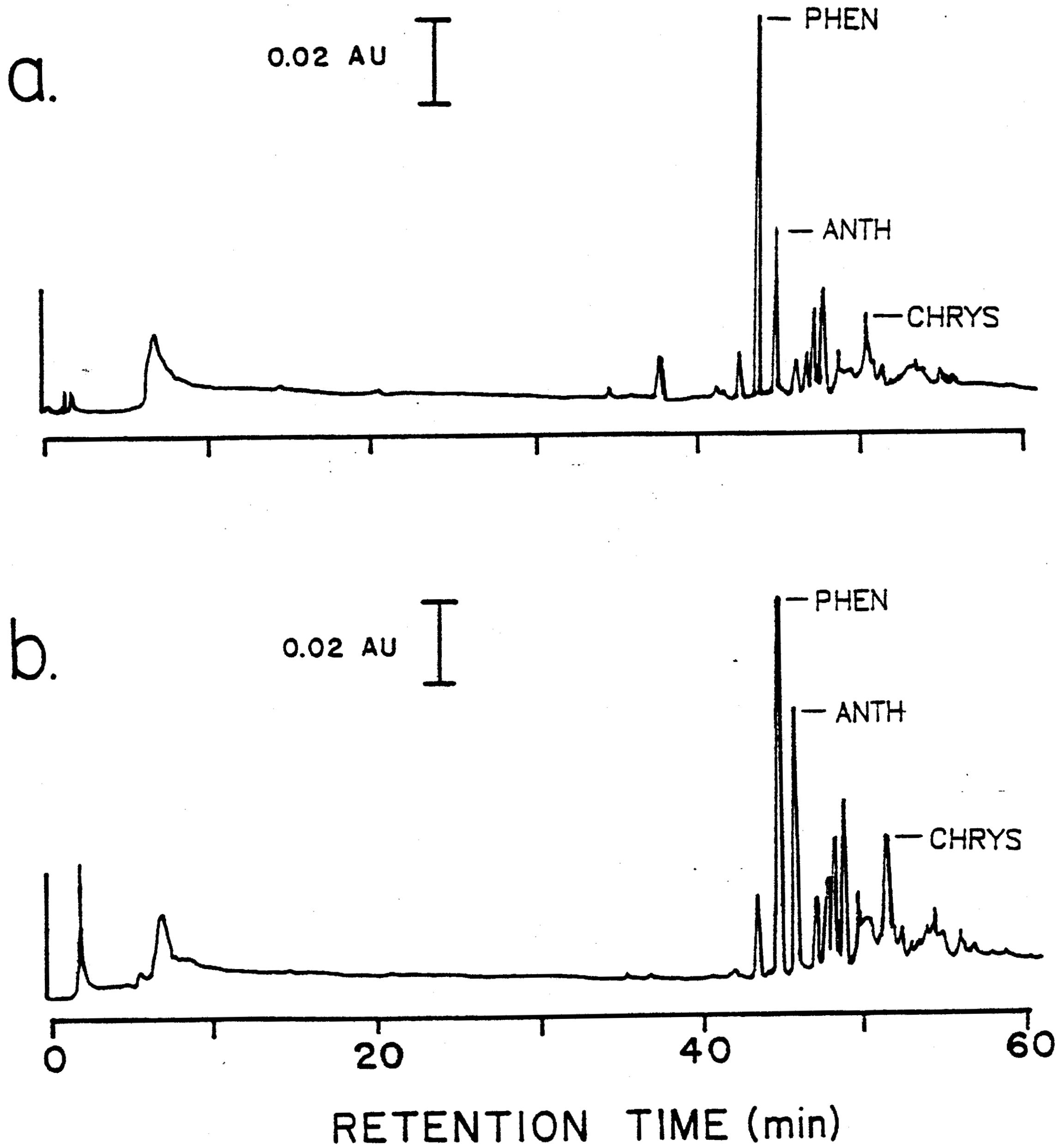


Figure 9. Reverse-phase HPLC chromatograms of (a) June 1985 interstitial water collected from a mini-piezometer inserted 1 m deep in Laramie River sediments, 30 m downstream of the oil body (see Fig. 2 for sampling location); and (b) creosote oil. Phen = phenanthrene, Anth = anthracene, Chrys = chrysene.

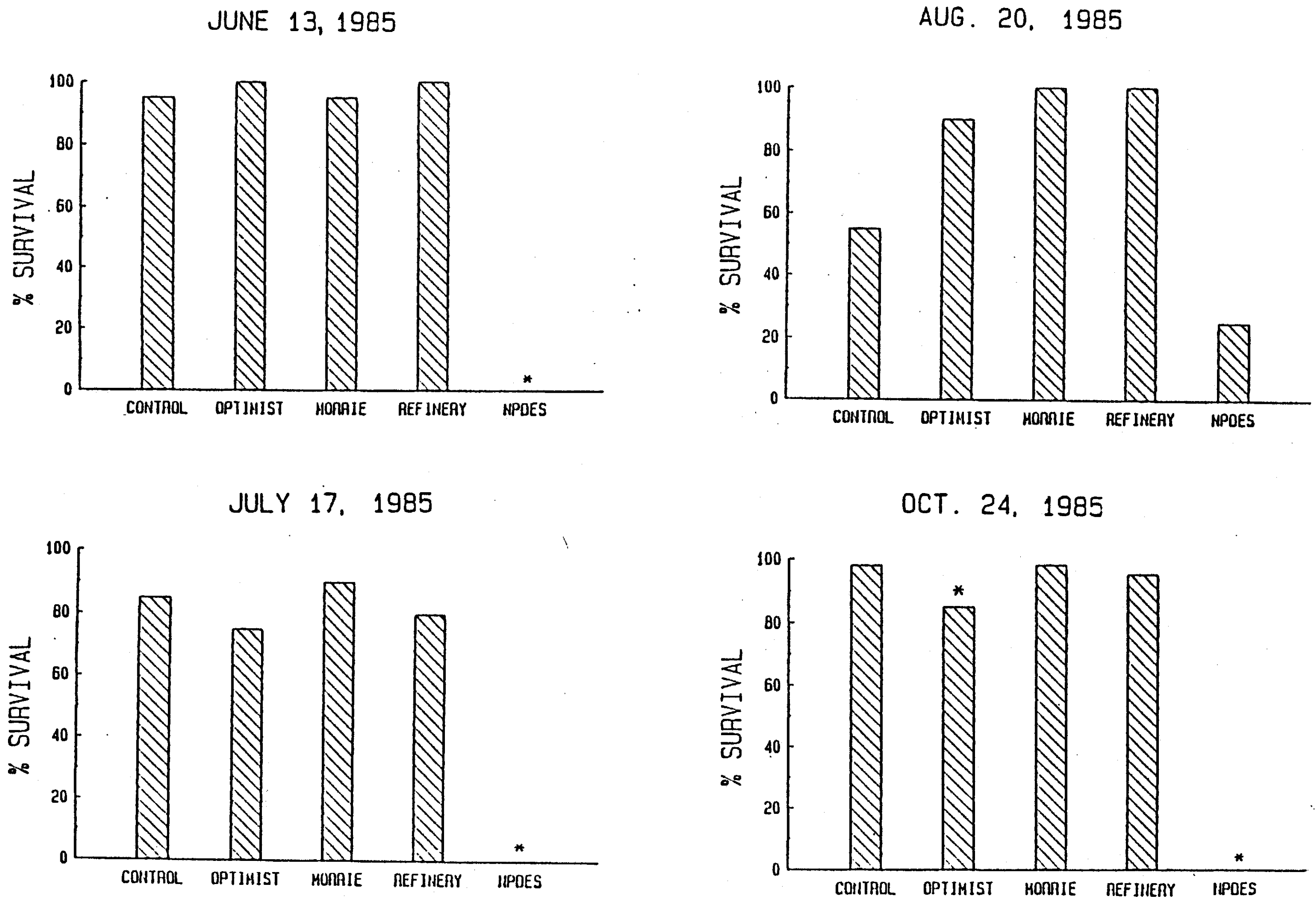
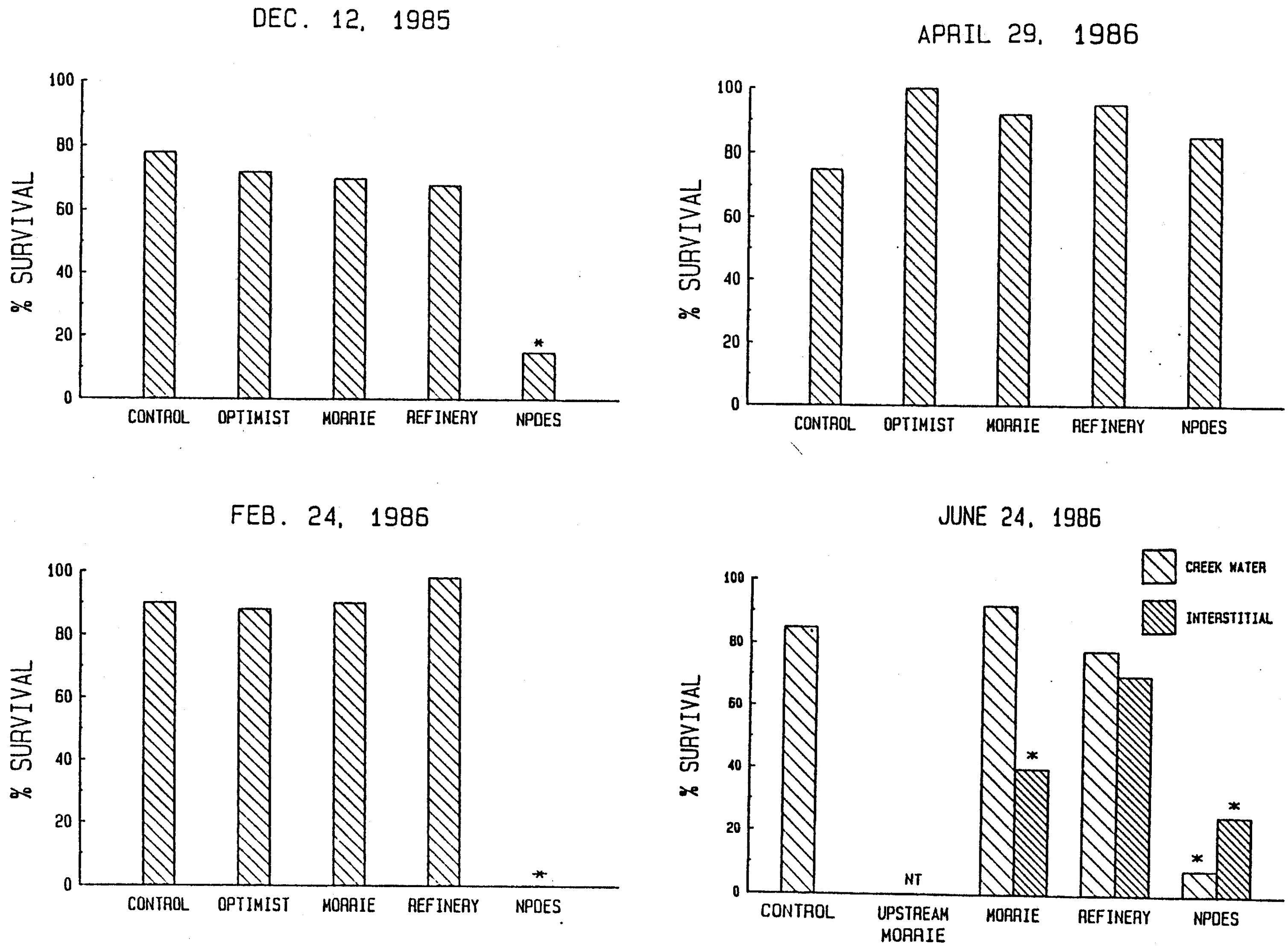


Figure 10. Fathead minnow (Pimephales promelas) survival in Crow Creek water and interstitial water from June 1985 to September 1986. NT = not tested; * = significantly lower survival than upstream control (P<0.05).



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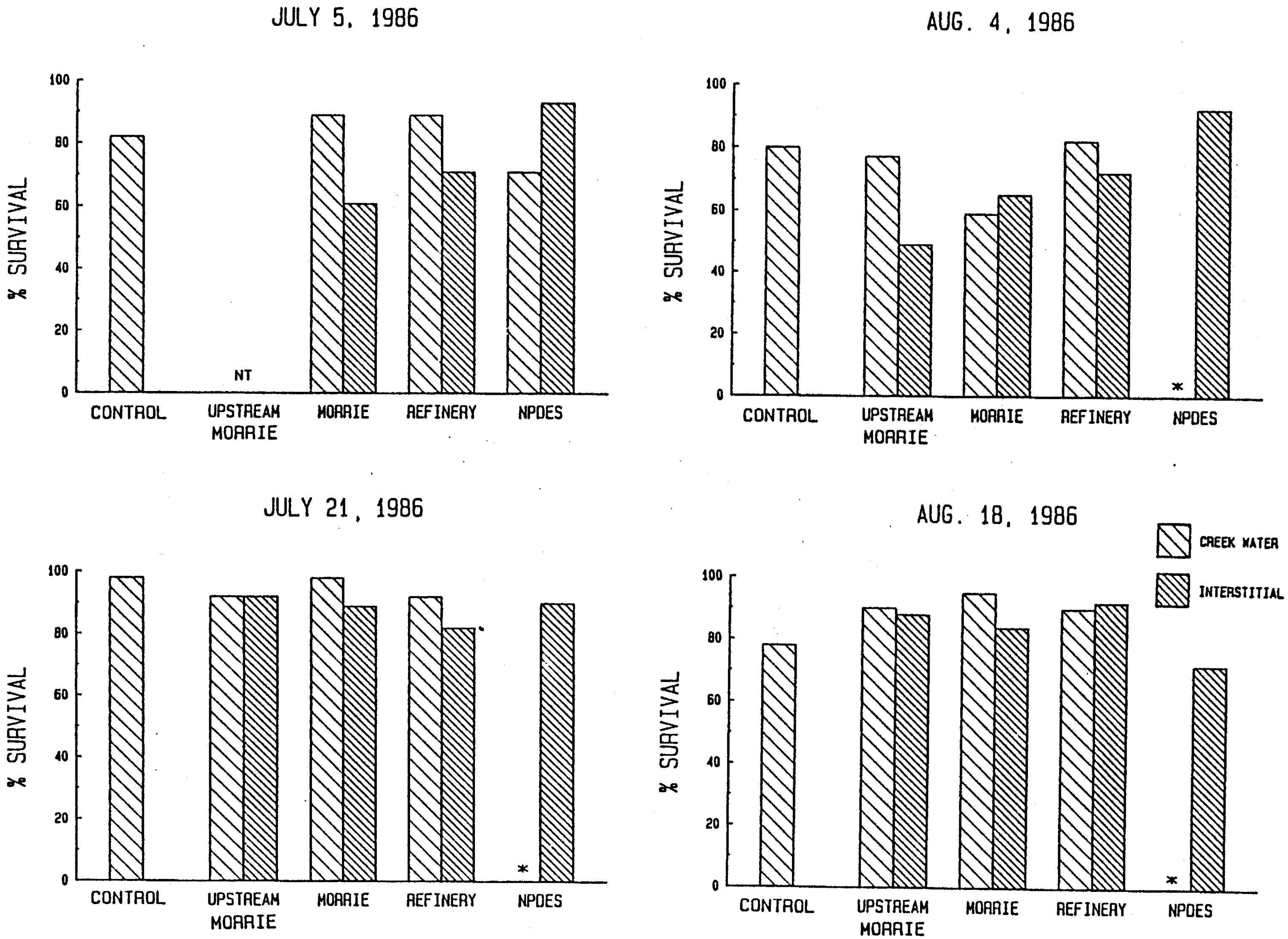

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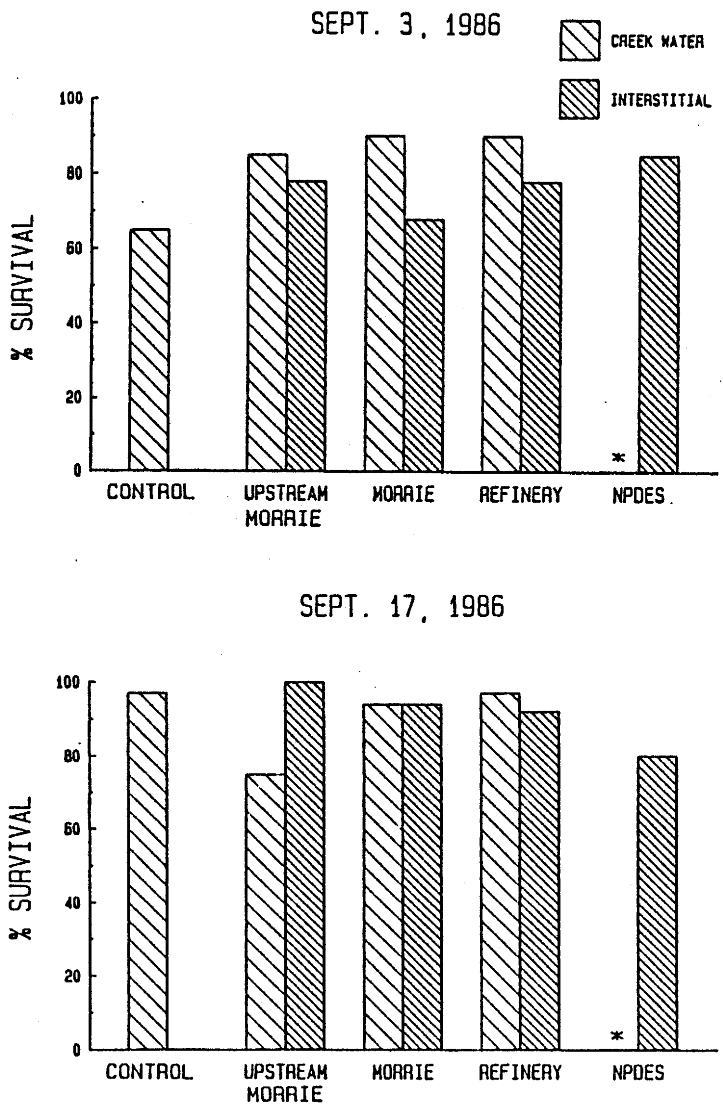

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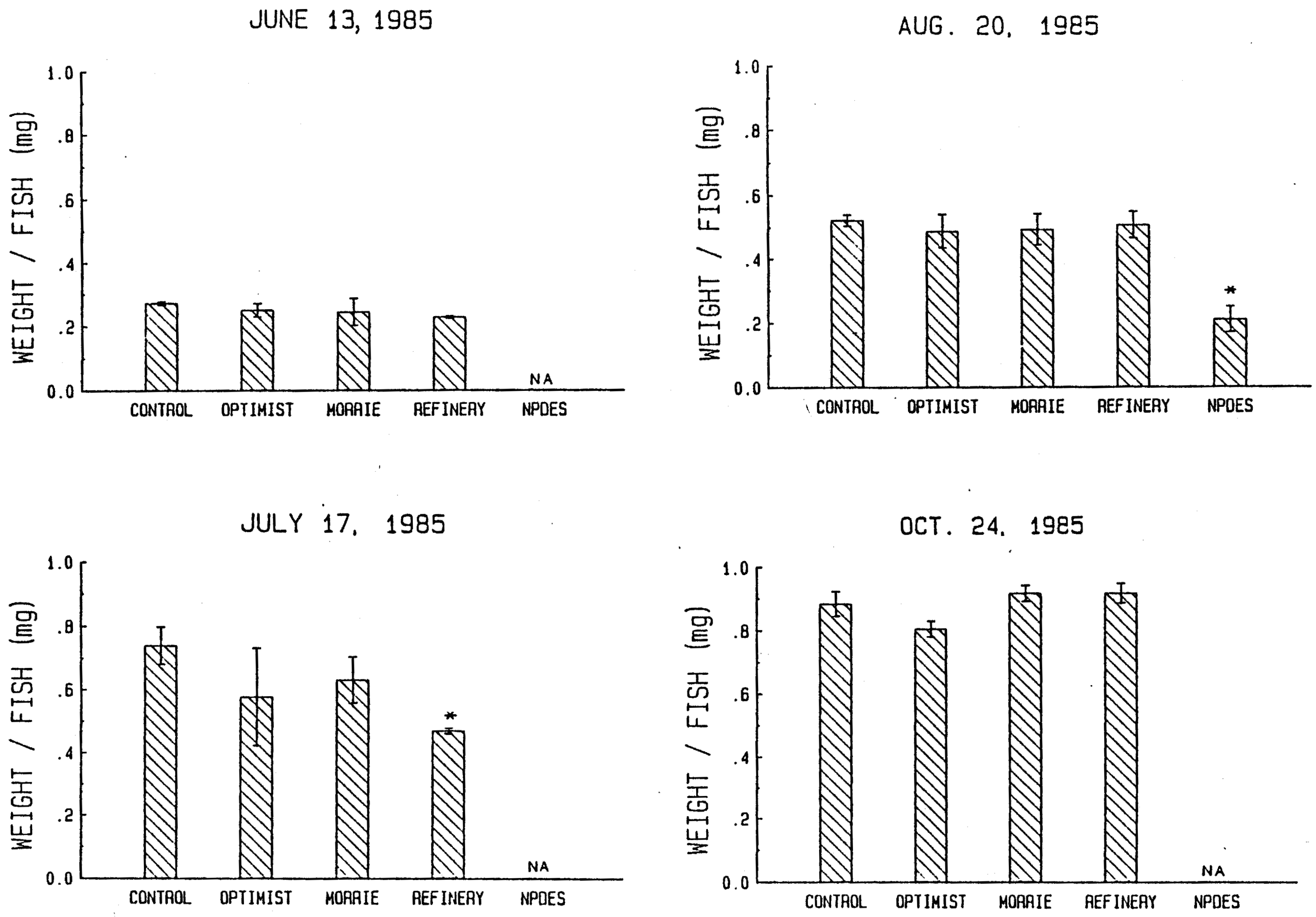
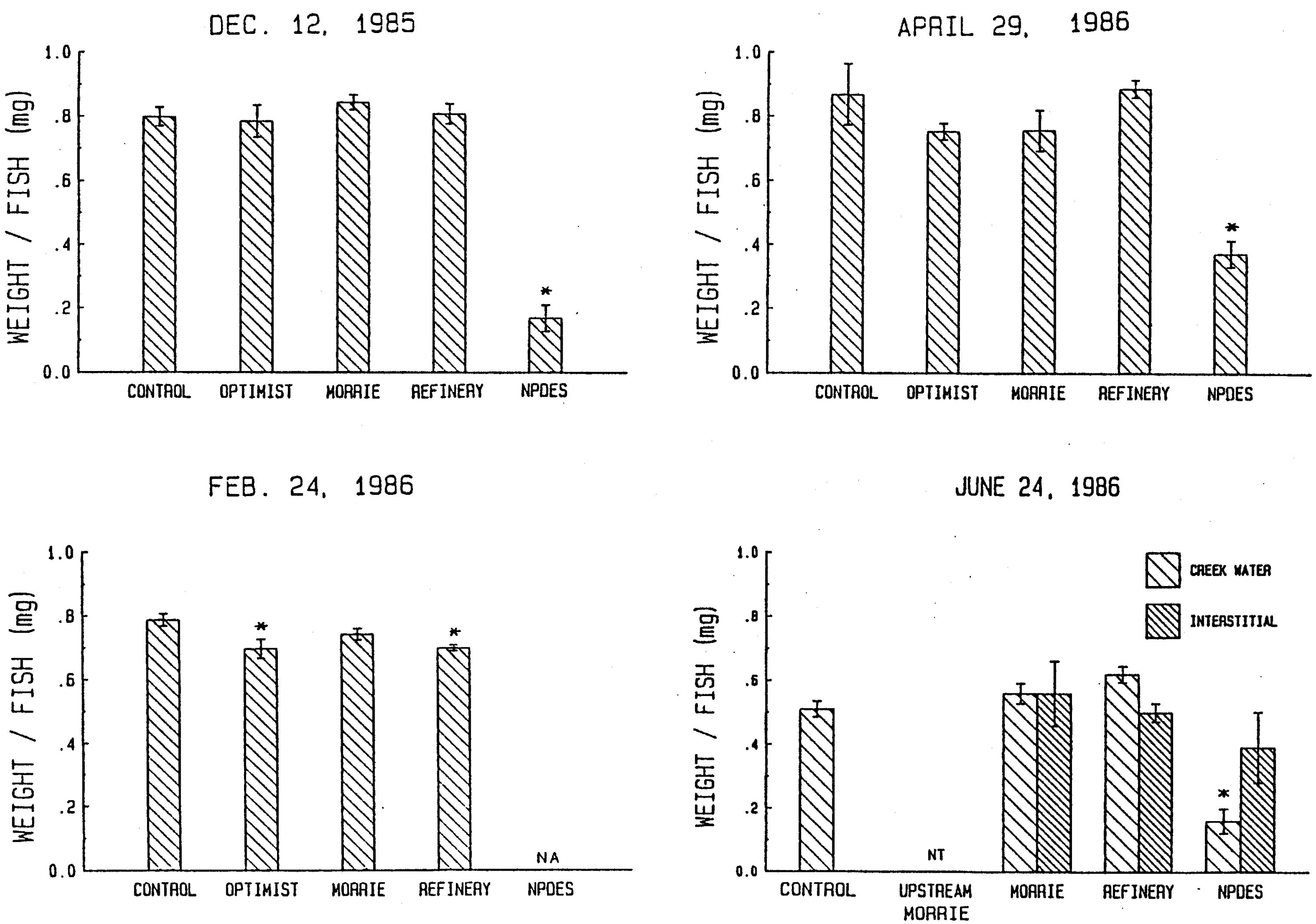


Figure 11. Fathead minnow (Pimephales promelas) growth (mean ± one standard error of the mean) in Crow Creek water and interstitial water from June 1985 to September 1986. NT = not tested; NA = value could not be calculated because all larvae died; * = significantly lower weight/fish than upstream control ($P < 0.05$).



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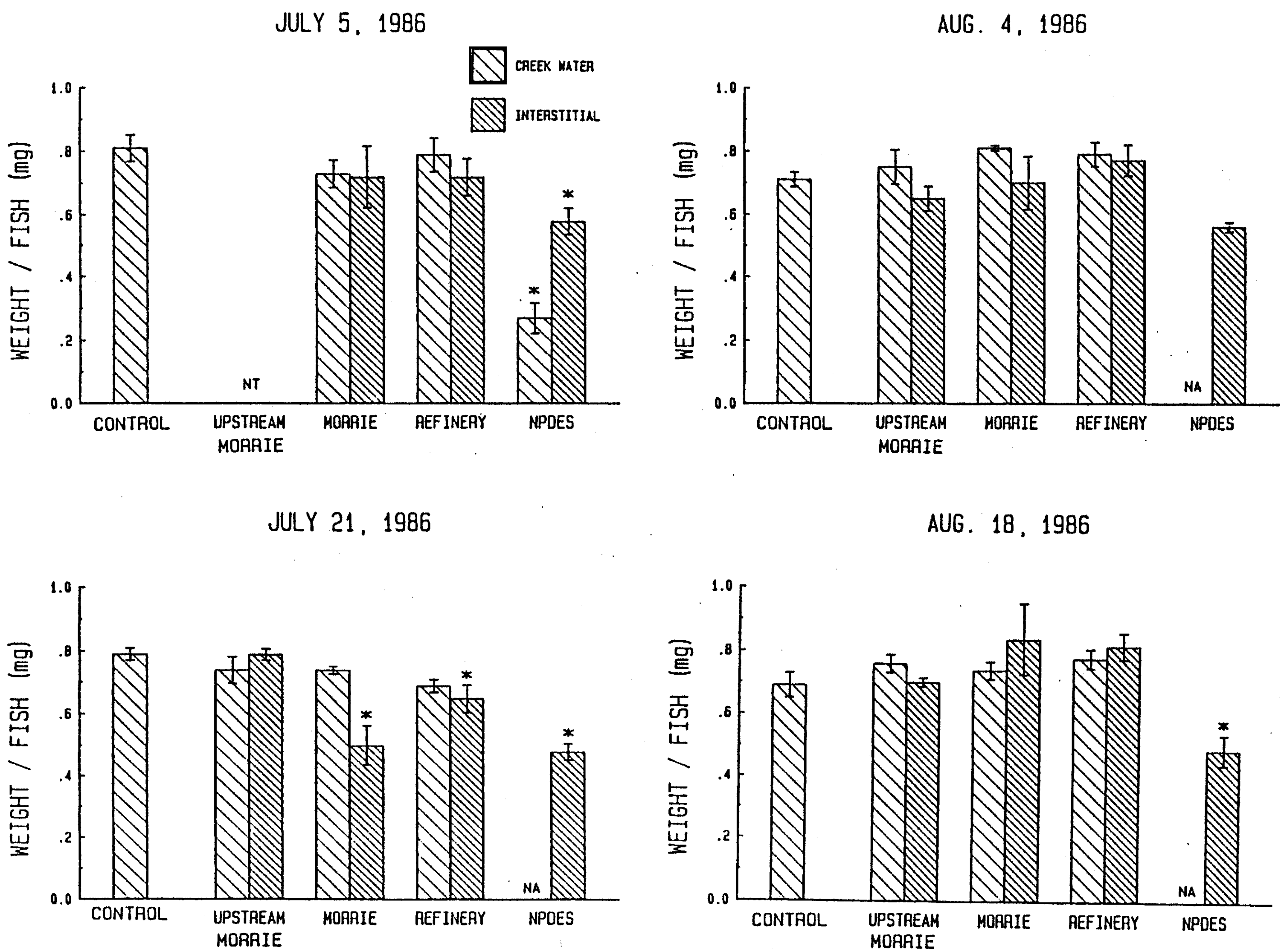

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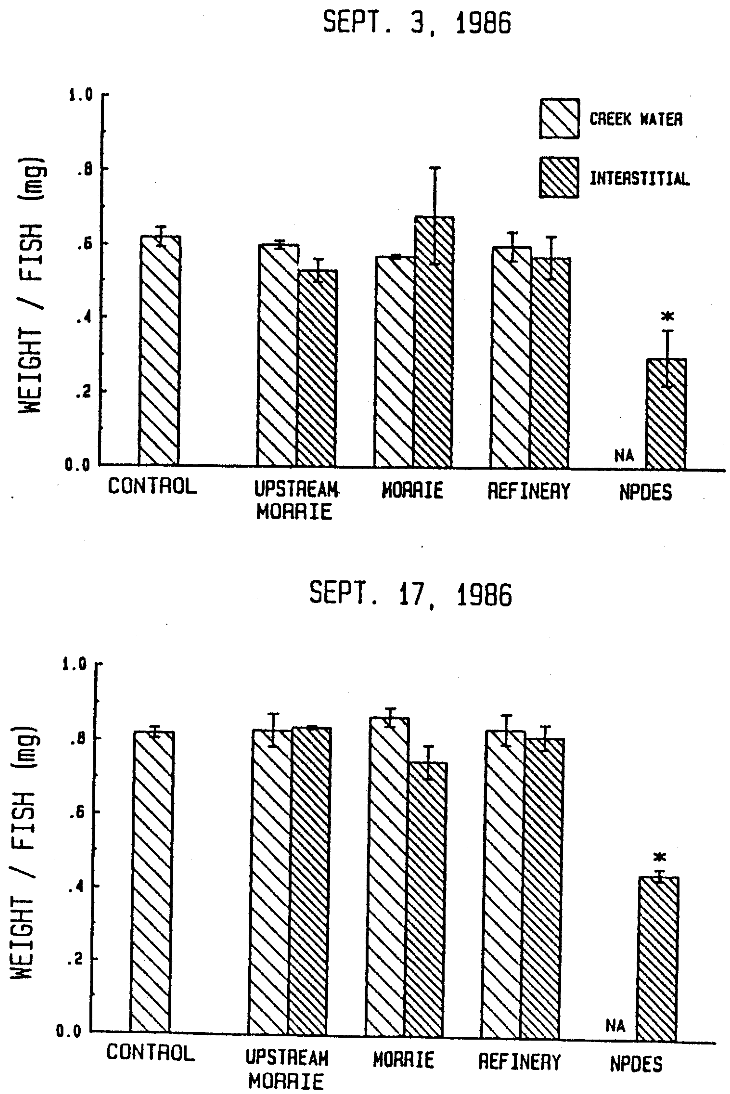

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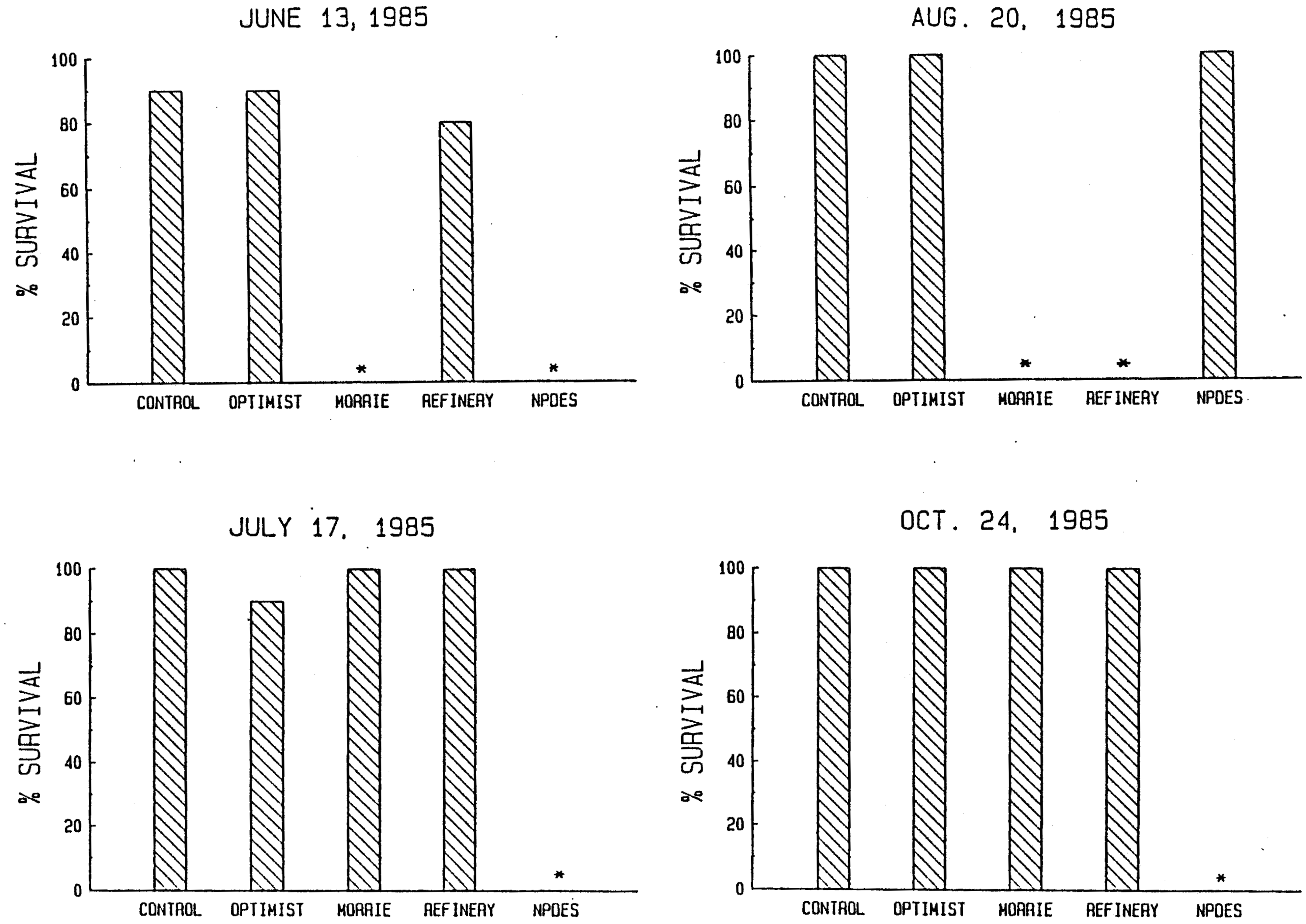
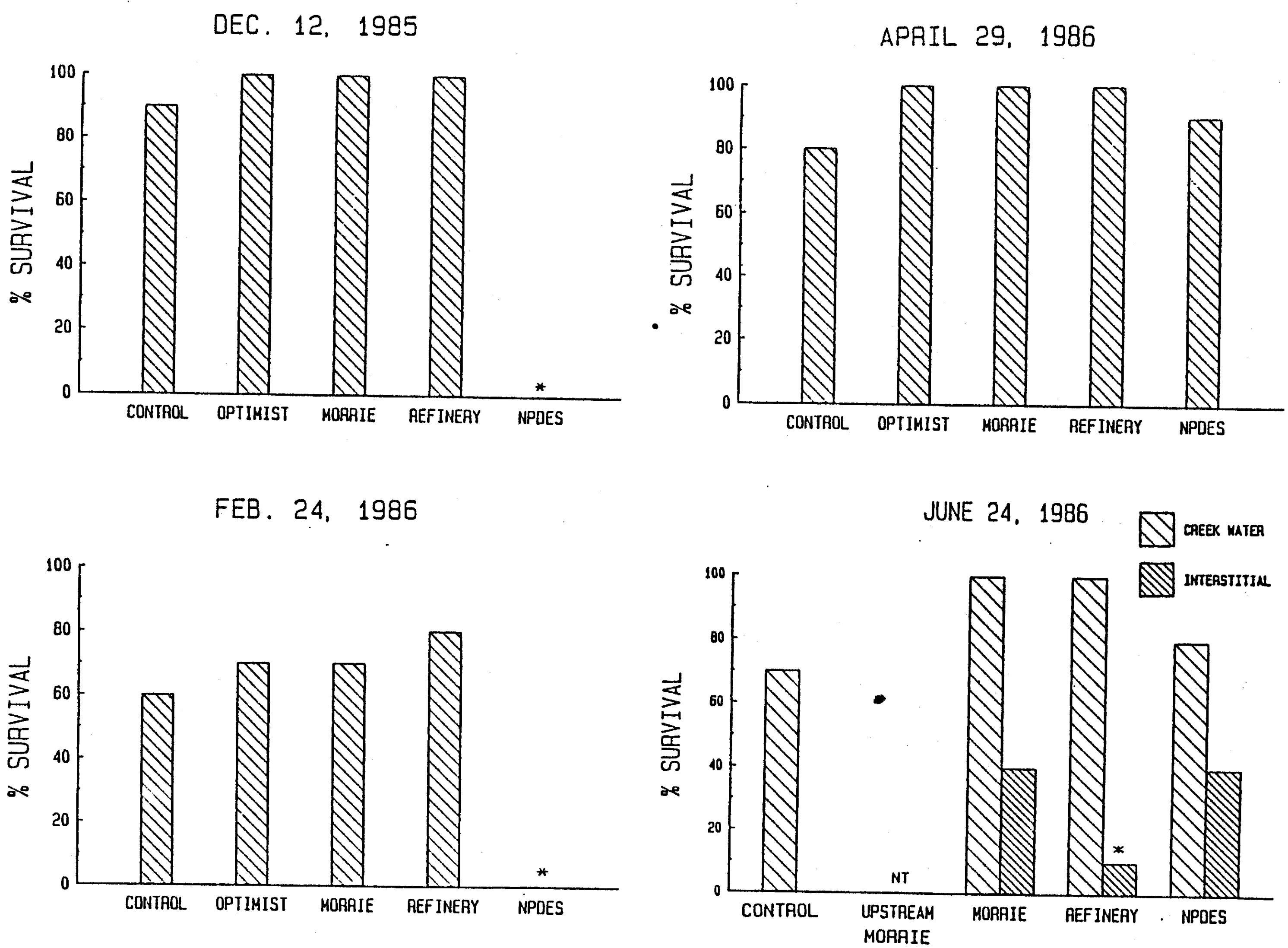


Figure 12. Ceriodaphnia dubia survival in Crow Creek water and interstitial water from June 1985 to September 1986. NT = not tested; * = significantly lower survival than in upstream control (P <0.05).



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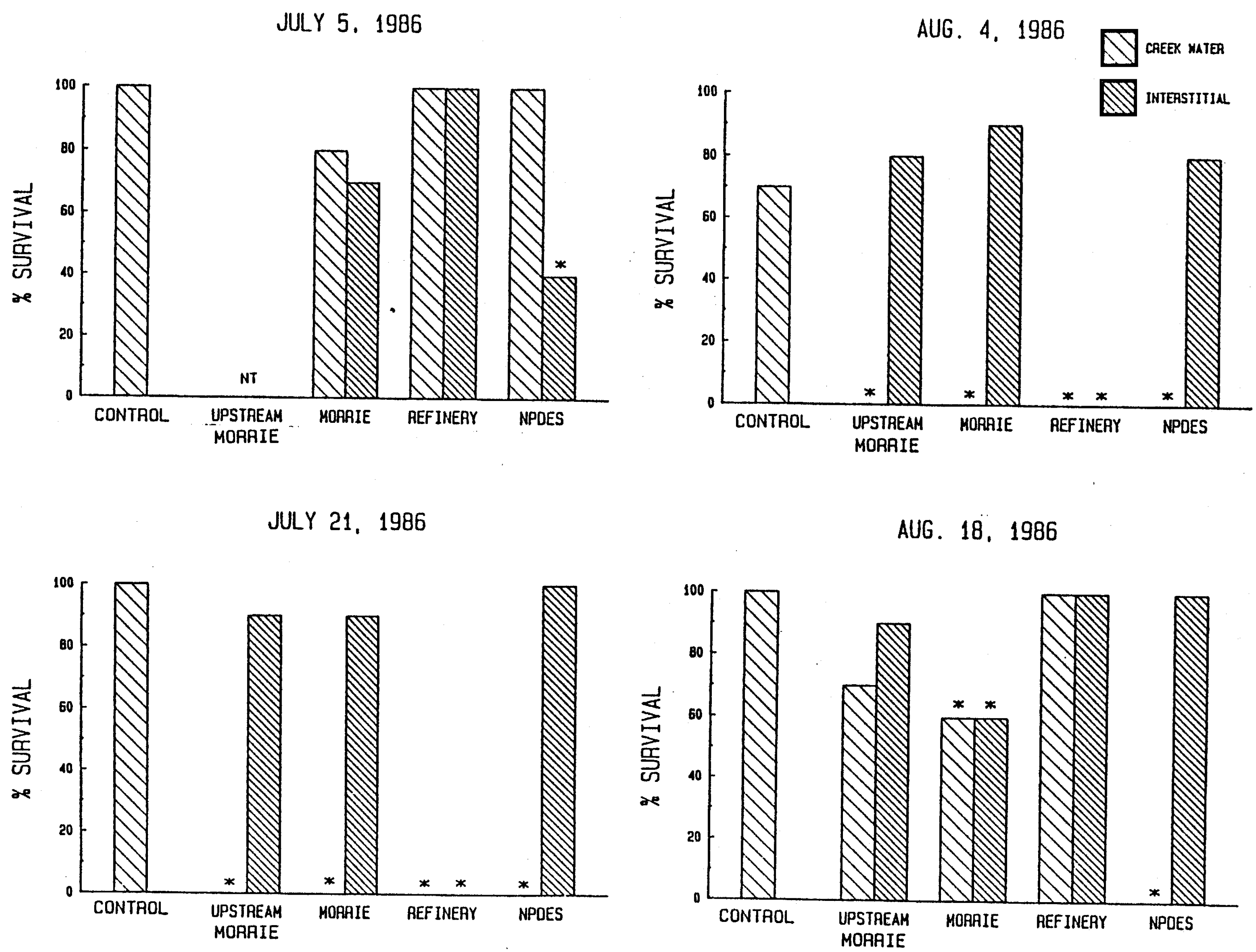

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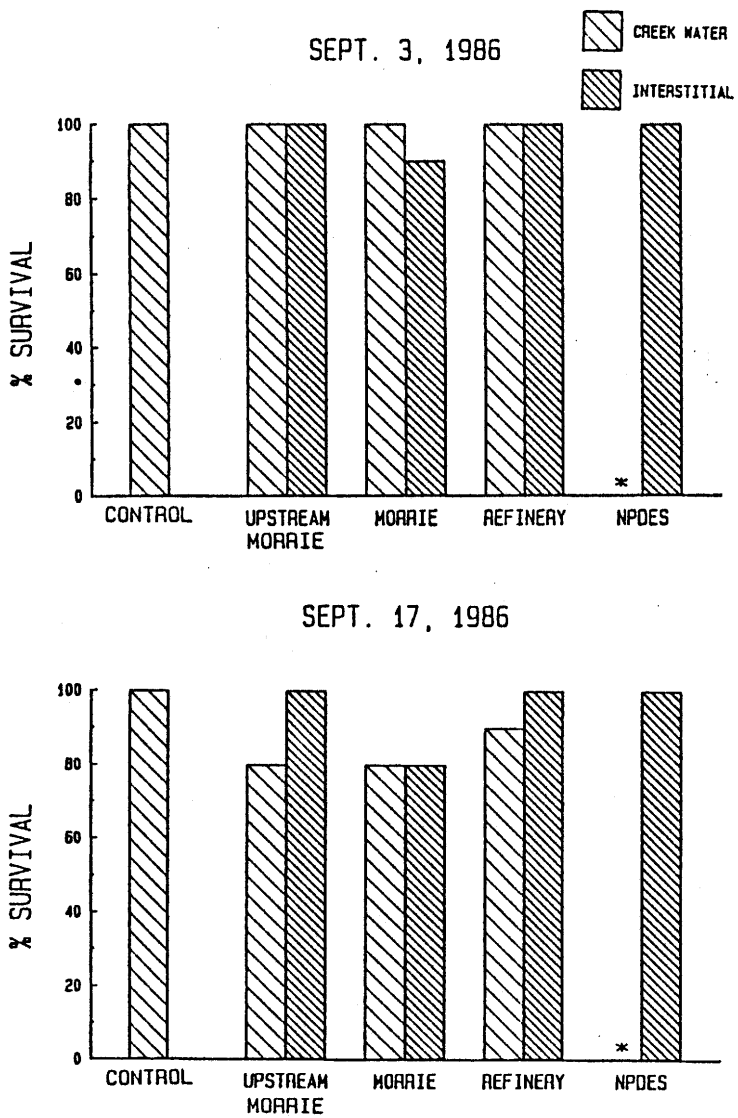

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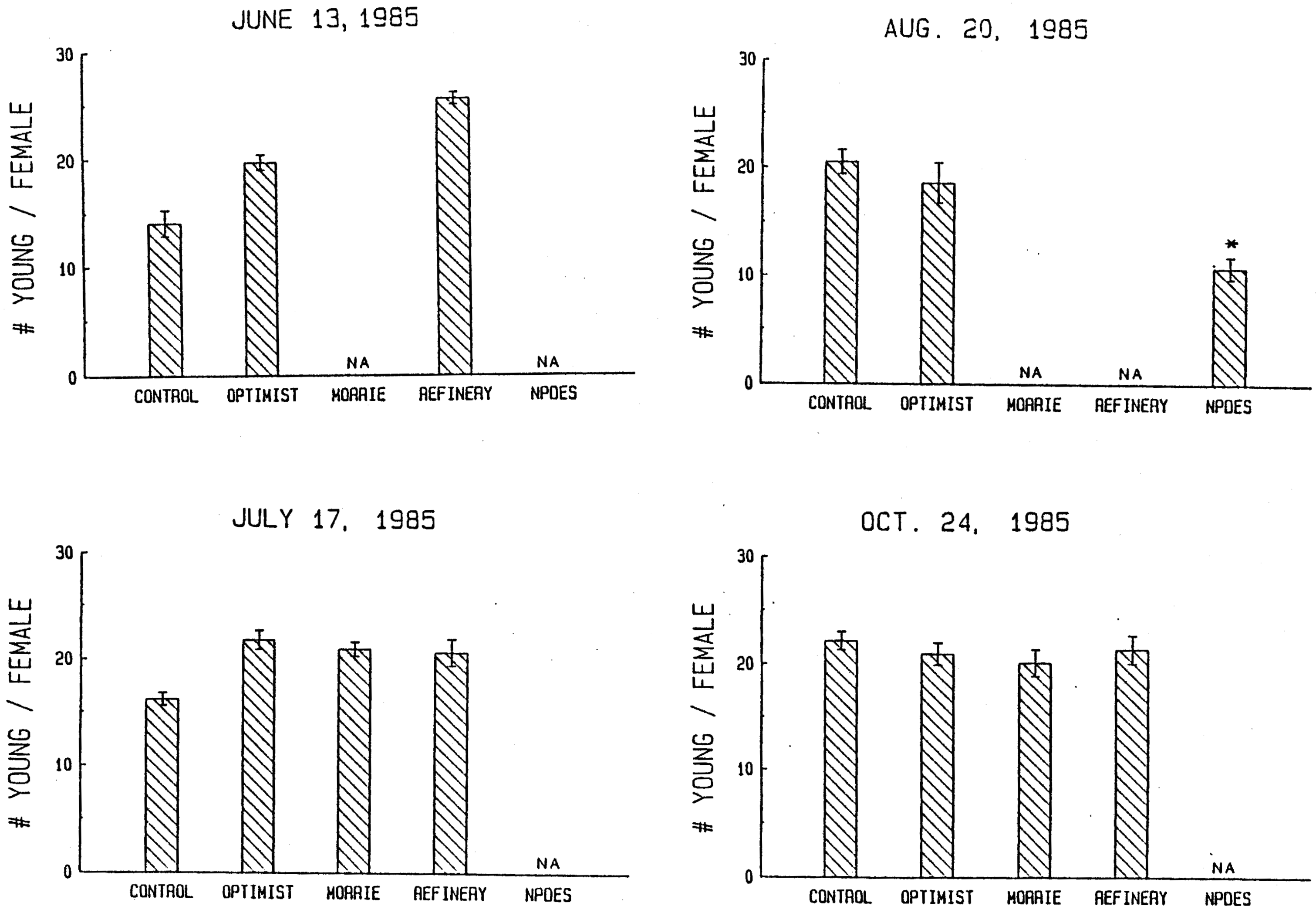
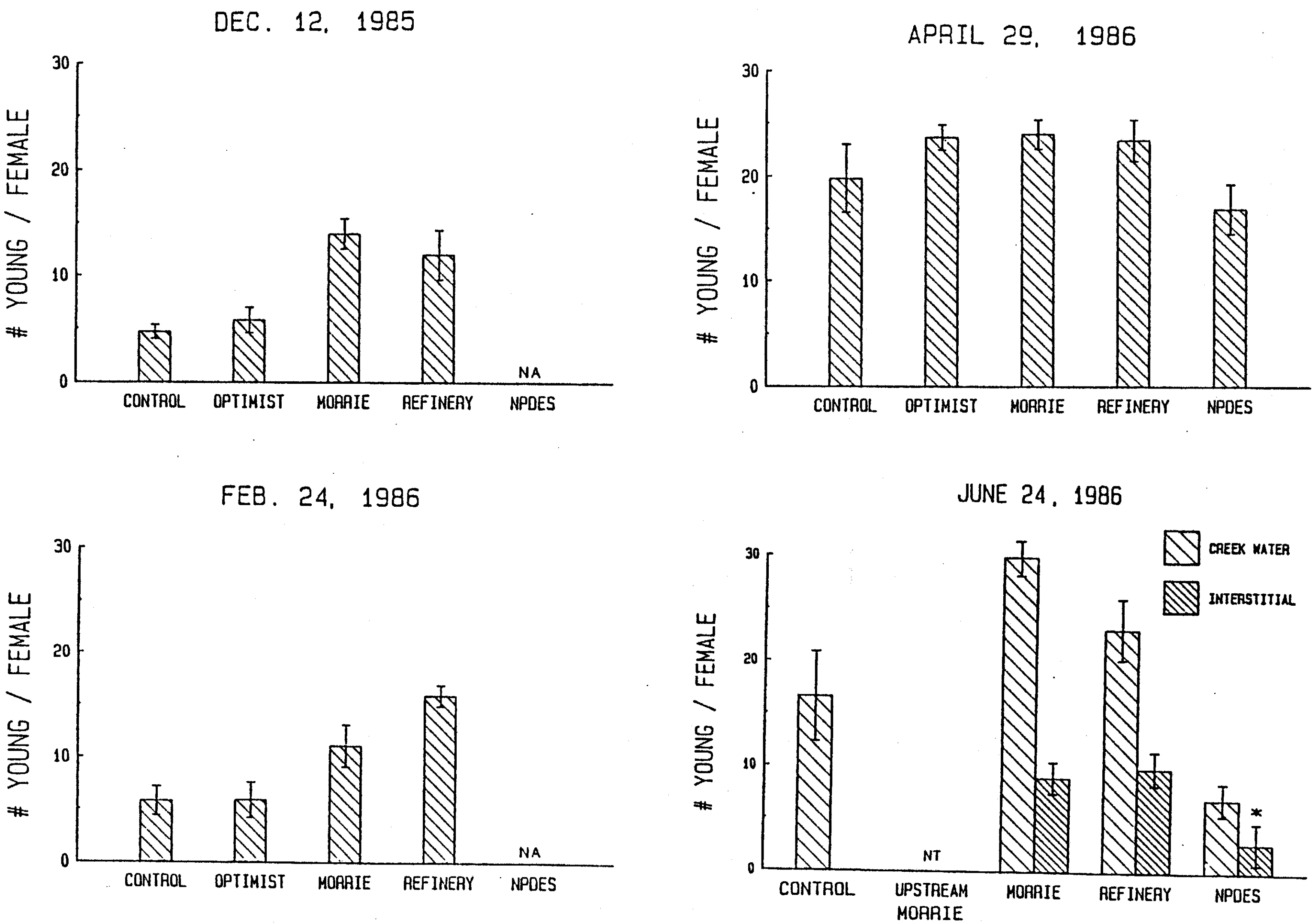


Figure 13. Ceriodaphnia dubia MOA reproduction (mean ± one standard error of the mean) in Crow Creek water and interstitial water from June 1985 to September 1986. NT = not tested; NA = value could not be calculated because survival was significantly lower than control; * = significantly lower reproduction than in upstream control ($P < 0.05$).



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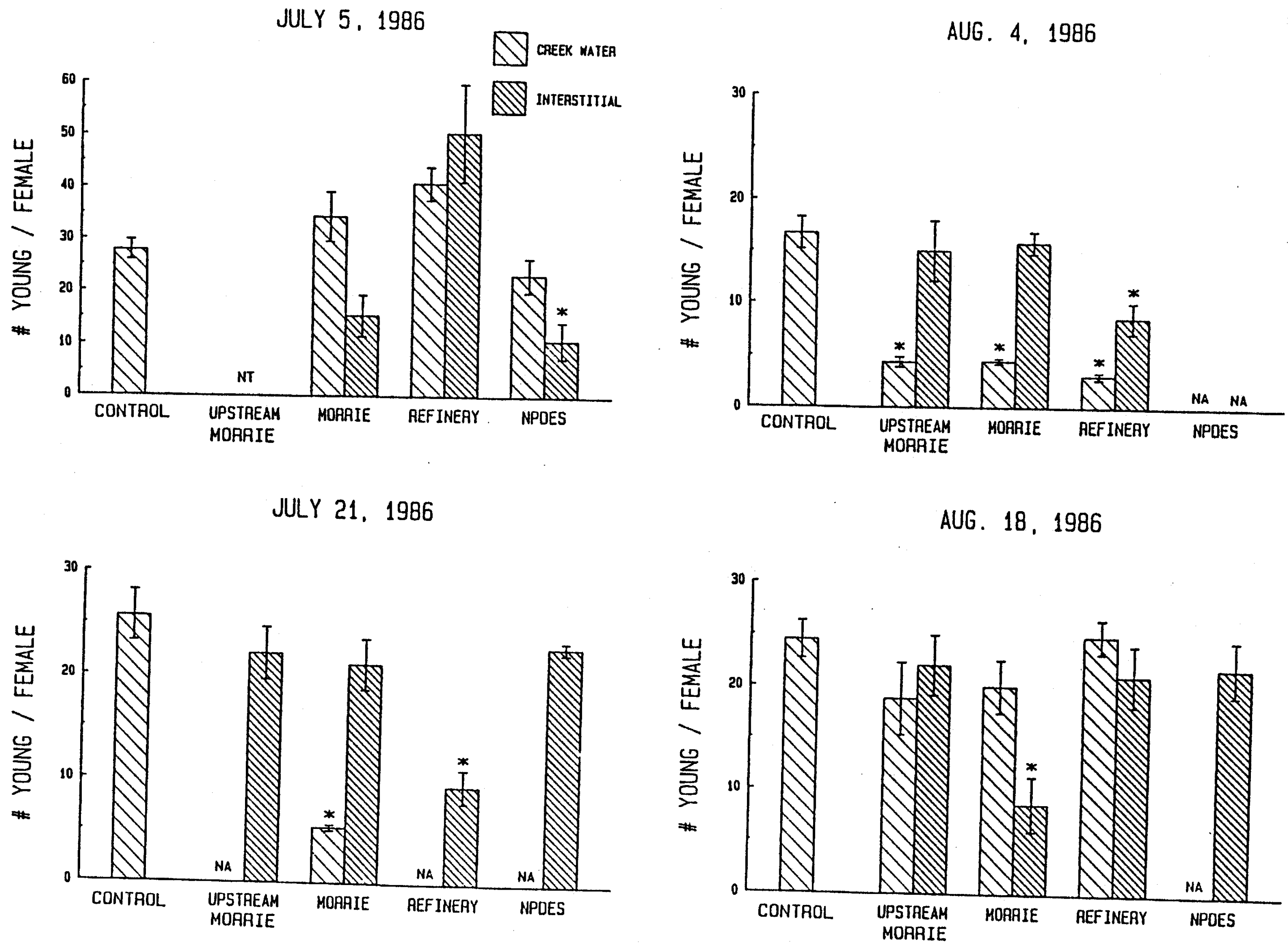

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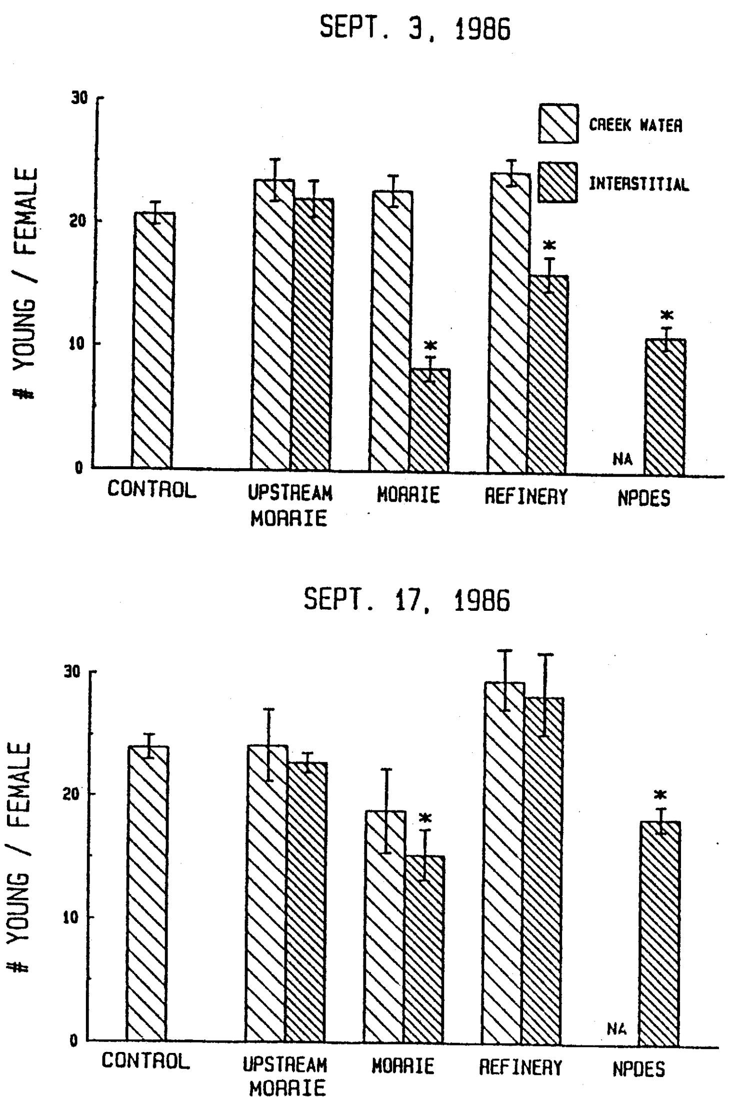


Figure 13 (continued).

1986, either fathead minnow growth or Ceriodaphnia MIM reproduction below the NPDES discharge was significantly lower than in the upstream control, even though survival was not significantly decreased (Figs. 10, 11, 12 and 13 and Appendix Table A-3). Thus, in one-tailed comparisons with the upstream control, Crow Creek water downstream from the NPDES discharge adversely affected fathead minnows and Ceriodaphnia on every sampling date.

Crow Creek water collected at the refinery sampling location (adjacent to the refinery but upstream from the NPDES discharge) significantly reduced fathead minnow growth relative to the upstream control on July 17, 1985 and February 24, 1986, whereas it killed all Ceriodaphnia by Day 7 of the test and significantly reduced their reproduction relative to the upstream control only on August 20, 1985 and July 21 and August 4, 1986 (Figs. 11, 12 and 13 and Appendix Table A-3).

Water collected at the Morrie Avenue bridge (the upstream boundary of the oil refinery property, adjacent to the SWSI Site) on June 13, 1985 killed all Ceriodaphnia between Days 6 and 7 of the test (Fig. 12 and Appendix Table A-3). And water collected at the Morrie Avenue bridge on August 20, 1985 and July 21 and August 4, 1986 also killed all Ceriodaphnia by Day 7 of the test and significantly reduced their MOA reproduction relative to the upstream control (Figs. 12 and 13 and Appendix Table A-3). However, samples from Morrie Avenue bridge never adversely affected fathead minnows (Figs. 10 and 11 and Appendix Table A-3).

Crow Creek water samples were collected just upstream from the Morrie Avenue bridge only during Year 2 (June to September 1986). At that sampling location, all Ceriodaphnia died by Day 7 and their MOA reproduction was significantly less than in the upstream control on July 21 and August 4, 1986 (Figs. 12 and 13 and Appendix Table A-3). Fathead minnow survival and growth were never decreased relative to the upstream control (Figs. 10 and 11 and Appendix Table A-3).

Crow Creek water samples were collected adjacent to Optimist Park only during Year 1 (June 1985 to April 1986). At that sampling location, fathead minnow growth was significantly lower than in the upstream control on February 24, 1986 (Fig. 11 and Appendix Table A-3). However, that decrease in growth in water collected at Optimist Park and along the refinery was not large (0.70 mg/fish at Optimist Park and the refinery vs. 0.79 mg/fish in the upstream control). Therefore, the February 24, 1986 significant decreases in fathead minnow growth at Optimist Park and the refinery sampling locations were probably artifacts of unusually small variances in weights among replicate beakers in the ANOVA computations (compare the low standard errors of estimates of mean weights on February 24, 1986 versus all other sampling dates in Appendix Table A-3). Optimist Park samples never adversely affected fathead minnow survival or <u>Ceriodaphnia</u> survival or reproduction relative to the upstream control (Figs. 10, 12 and 13 and Appendix Table A-3).

Similar to Crow Creek water collected downstream from the refinery's NPDES discharge pipe, interstitial water withdrawn 1 m deep in the creek sediments at this same location reduced fathead minnow survival or growth or <u>Ceriodaphnia</u> survival or reproduction relative to the upstream control on every sampling date (June to September 1986; Figs. 10, 11, 12 and 13 and Appendix Table A-3).

Interstitial waters collected at the refinery sampling location and downstream from the Morrie Avenue bridge also caused adverse effects, but not as predictably as at the NPDES sampling location. Refinery interstitial water reduced fathead minnow growth or <u>Ceriodaphnia</u> survival or growth relative to the upstream control on June 24, July 21, August 4, and September 3, 1986, whereas interstitial water collected downstream from the Morrie Avenue bridge reduced fathead minnow survival or growth or *Ceriodaphnia* survival or reproduction relative to the upstream control on June 24, July 21, August 18, and September 3, 1986 (Figs. 10, 11, 12 and 13 and Appendix Table A-3).

Interstitial water collected just upstream from the Morrie Avenue bridge never adversely affected fathead minnow survival and growth or *Ceriodaphnia* survival and reproduction relative to the upstream control (Figs. 10, 11, 12 and 13 and Appendix Table A-3).

Two-tailed comparisons of all possible pairwise combinations of sampling locations within a test showed additional differences and similarities not indicated by one-tailed comparisons of downstream sampling locations only with the upstream control (Appendix Table A-4). For example, fathead minnow weights were sometimes ambiguously similar. On July 17, 1985, fathead minnow weight in Crow Creek water collected adjacent to the refinery was significantly less than in the upstream control but not significantly less than at Optimist Park and Morrie Avenue, the two sampling locations immediately upstream from the refinery; yet fathead minnow weights for those two locations were not significantly less than the upstream control (Appendix Table A-4). Similarly on September 3, 1986, fathead minnow weight in interstitial water collected downstream from the NPDES discharge was significantly less than in the upstream control but not significantly less than in interstitial waters collected adjacent to the refinery and upstream from the Morrie Avenue bridge, in which fathead minnow weights were not significantly less than in the upstream control (Appendix Table A-4). And finally, on August 18, 1986, fathead minnow weight in interstitial water collected downstream from the NPDES discharge was not significantly less than in the upstream control, based on the two-tailed comparisons (Appendix Table A-4); yet the same weight was significantly less than the weight in the upstream control, based on one-tailed comparisons (Appendix Table A-3).

Two-tailed comparisons of Ceriodaphnia reproduction tended to show additional significant differences not indicated in one-tailed comparisons of downstream sampling locations only with respect to the upstream control. For example, on June 13, July 17 and December 12, 1985 and February 24 and July 5, 1986, MOA total offspring per female increased significantly in waters collected from at least one downstream sampling location. Increases in reproduction at downstream locations were largest on December 12, 1985 (4.2 offspring/female in the upstream control vs. 12.0 offspring/female in Crow Creek water adjacent to the refinery) and February 24, 1986 (3.6 offspring/female in the upstream control vs. 13.0 offspring/female in Crow Creek water adjacent to the refinery).

Chemical Analyses. Except for unionized ammonia (NH_3) and ammonium ion (NH_4^+), routine water chemistry parameters and major inorganic ions at all Crow Creek sampling locations were generally within normal ranges for alkaline surface waters in the western United States (Table 2 and Appendix Tables B-4 and B-5). NH_3, the species of ammonia most toxic to aquatic animals, was calculated to be > 0.07 mg/L and as high as 0.90 mg/L in Crow Creek water downstream from the refinery's NPDES discharge in every month total ammonia was measured (Appendix Table B-4). Additionally, NH_3 concentrations in interstitial water collected 1 m deep in Crow Creek sediments downstream from the NPDES discharge ranged from 0.07 to 0.29 mg/L (June to September 1986). Total ammonia was not measured on July 17 and August 20, 1985 due to instrument malfunction.

Concentrations of Na^+, Ca^{2+}, K^+, NH_4^+, Cl^-, SO_4^{2-}, NO_3^- and F^- in Crow Creek water tended to increase downstream from the control, especially Na^+, K^+,

NH_4^+, Cl^-, SO_4^{2-} and F^- downstream from the refinery's NPDES discharge (Appendix Table B-5). However, the NO_3^- concentration in Crow Creek water downstream from the NPDES discharge often was lower than in Crow Creek water collected adjacent to the refinery, only 0.4 km upstream. Concentrations of major inorganic ions in interstitial waters did not tend to increase from upstream to downstream sampling locations. Instead, interstitial water collected downstream from the Morrie Avenue bridge (adjacent to the SWSI Site) always had the highest concentrations of Na^+, Ca^{2+}, Mg^{2+}, Sr^{2+}, Cl^- and SO_4^{2-} and almost always had the highest concentrations of K^+ and NO_3^-, compared to the other three interstitial-water sampling locations. Interstitial water collected downstream from the NPDES discharge always had the highest concentrations of NH_4^+ and F^-. Additionally, HCO_3^- concentrations were always higher in interstitial water collected downstream from the Morrie Avenue bridge and the NPDES discharge than in all other surface waters and interstitial waters on dates when interstitial-water samples were collected. However, those HCO_3^- concentrations may not be reliable, since they were calculated from total alkalinity values that may reflect the presence of weak organic acids as well as carbonic acid.

Of the 11 trace elements analyzed, only chromium, copper and zinc were unusually high in some samples (Appendix Table B-6). Total dissolved Cr concentration was 0.0399 mg/L below the refinery's NPDES discharge on June 13, 1985, and between 0.0126 and 0.0392 mg/L from June 24 to September 17, 1986; however, Cr^{6+} (the most toxic chromium species) was not analyzed in any samples. All other total dissolved Cr values below the NPDES discharge were $\leq$ 0.0056 mg/L. Concentrations of Cu were 0.0417 mg/L on July 17, 1985 and 0.0296 mg/L on July 5, 1986 in the upstream control; and 0.0165 mg/L along the oil refinery on August 20, 1985. Since their respective downstream samples had much lower Cu concentrations, those two high values may have been caused by sample contamination. The zinc concentration was 0.44 mg/L in the interstitial water collected adjacent to the oil refinery on July 21, 1986; all other values were $\leq$ 0.09 mg/L.

No organic compounds were detected in any Crow Creek waters or interstitial waters using reverse-phase HPLC in our laboratory. Similarly, none of the 13 phenolic priority pollutants monitored by GC-MS analyses in Crow Creek waters collected by Frontier Refinery personnel in October and December 1985 and July 1986 was above detection limits (Rocky Mountain Analytical Laboratory 1986a, 1986b, 1986c). Dissolved organic carbon concentrations ranged between 2.0 and 24.9 mg/L, and DOC concentrations usually were higher downstream from the upstream control. However, the range of DOC values in Crow Creek from June 1985 to September 1986 was narrower than the range of DOC values recorded for the Laramie River from June 1985 to October 1985 (Table 2).

Ammonia-Toxicity Correlations. Because high concentrations of unionized ammonia (NH_3) were present in Crow Creek water and interstitial water downstream from the refinery's NPDES discharge, we tested whether NH_3 concentrations correlated significantly with fathead minnow and *Ceriodaphnia* toxicity in those waters. Figures 14 and 15 show trends of decreasing survival, growth and reproduction as NH_3 concentrations increased. But generally, those responses were less adversely affected (i.e., survival, growth and reproduction were higher) in interstitial waters than in Crow Creek waters at similar NH_3 concentrations.

When data for Crow Creek waters and interstitial waters were combined, Spearman coefficients of rank correlation were negative for NH_3 concentrations versus each of the four response variables and were highly significant

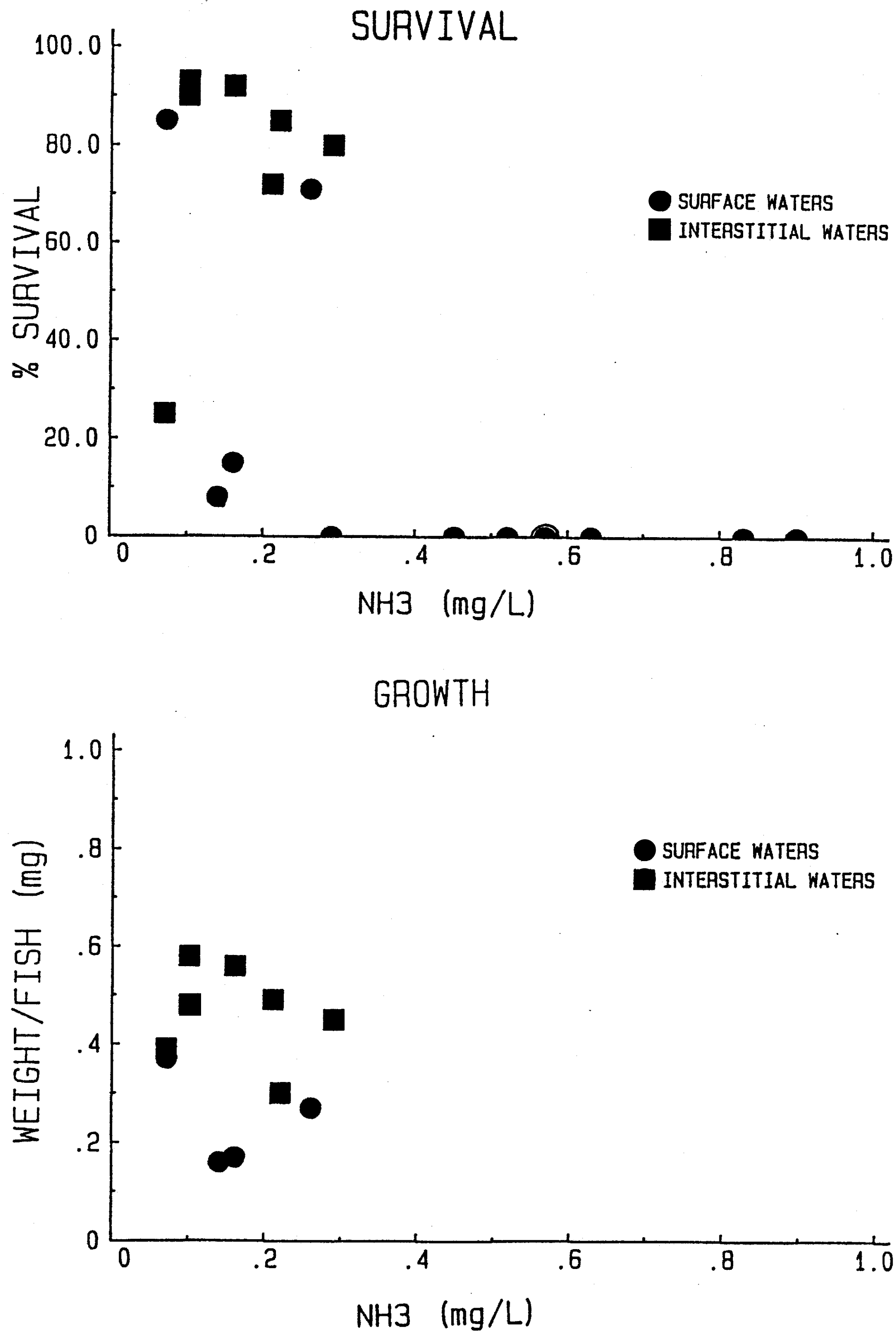
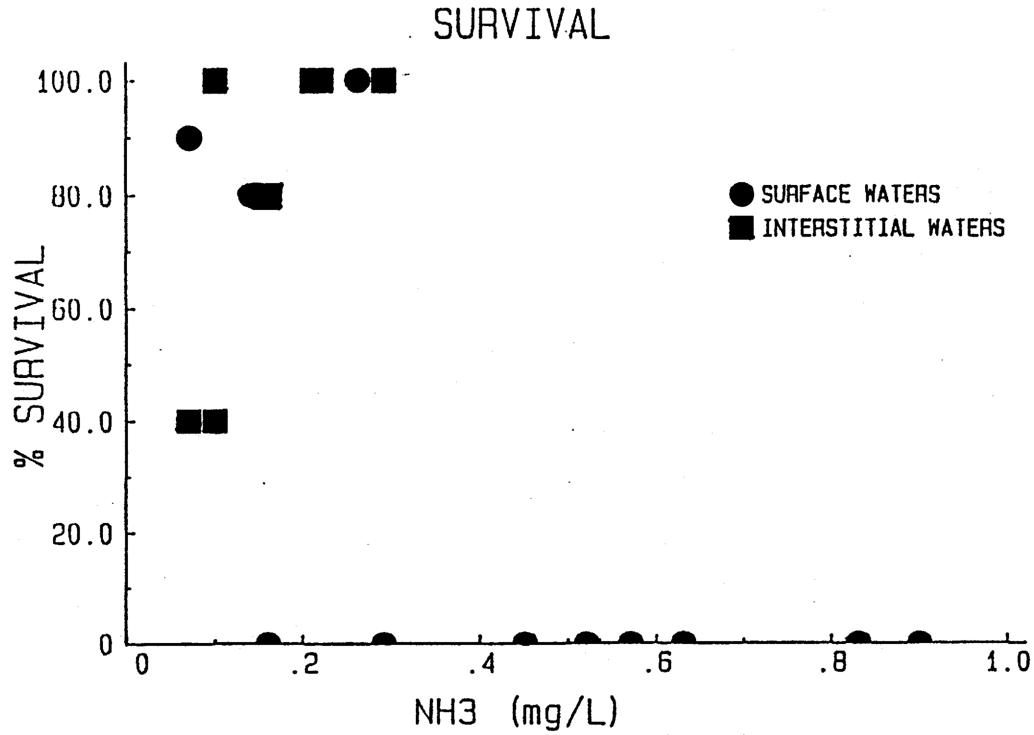


Figure 14. Fathead minnow (Pimephales promelas) survival and growth versus unionized ammonia (NH_3) concentrations in Crow Creek water and interstitial water collected downstream from the Frontier Oil Refinery NPDES discharge. Values taken from Appendix Tables A-3 and B-4.



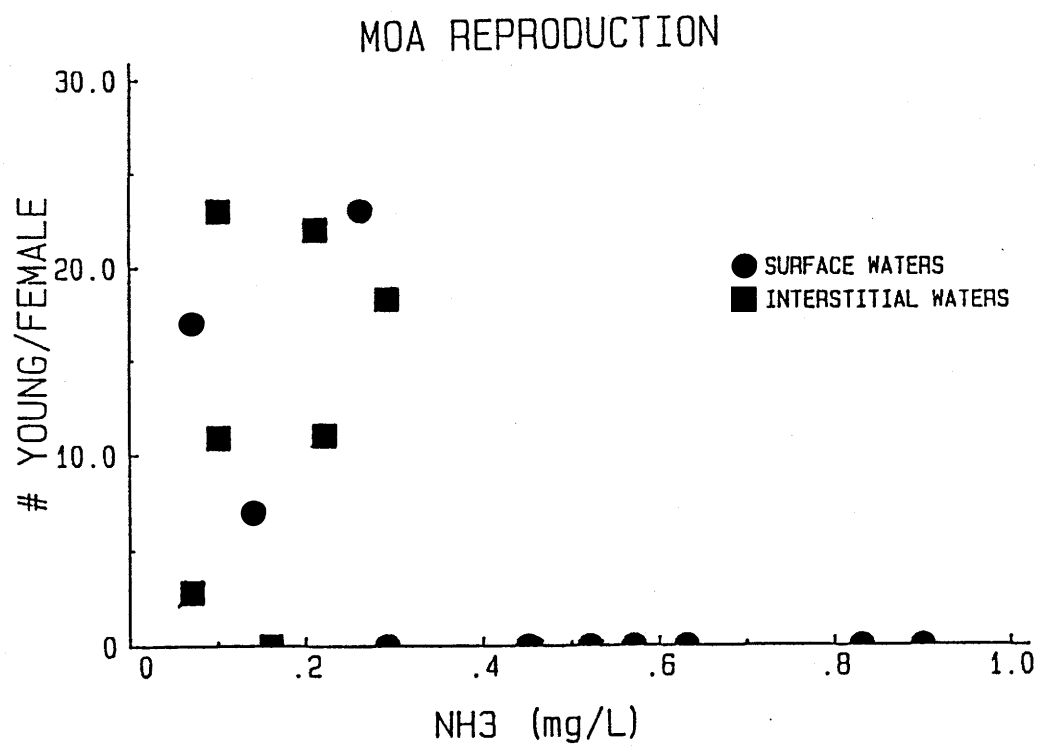


Figure 15. Ceriodaphnia dubia survival and reproduction versus unionized ammonia (NH_3) concentrations in Crow Creek water and interstitial water collected downstream from the Frontier Oil Refinery NPDES discharge. Values taken from Appendix Tables A-3 and B-4.

($P < 0.01$) for the following relationships: [NH_3] vs. fathead minnow survival, [NH_3] vs. Ceriodaphnia survival, and [NH_3] vs. Ceriodaphnia MOA reproduction (Table 3). The significance levels associated with those coefficients were strongly influenced by responses in waters with NH_3 concentrations > 0.4 mg/L, where survival was always 0% and reproduction was always 0 offspring/female. Spearman coefficients of rank correlation were much less negative and were not significant ($P > 0.05$) when data for waters only containing < 0.4 mg NH_3/L were included in the correlations. Hence, the lack of a significant coefficient of rank correlation for [NH_3] vs. fathead minnow growth in Table 3 may be partly because no waters with NH_3 concentration > 0.4 mg/L could be included in the correlation, since no fathead minnow larvae survived through Day 7 in those tests (Figure 14).

Similar results occurred when the data were analyzed separately as Crow Creek waters and interstitial waters. None of the Spearman coefficients of rank correlation was significant ($P > 0.05$) in interstitial waters (Table 3), in which NH_3 concentrations were always < 0.4 mg/L. But correlations of [NH_3] versus fathead minnow survival and Ceriodaphnia survival and reproduction in Crow Creek waters were significant ($P < 0.05$), mainly because survival and reproduction were zero in those waters when NH_3 concentration was > 0.4 mg/L.

DISCUSSION

Laramie River Study Site

The Union Pacific Tie Treatment Plant illustrates several common problems encountered at hazardous waste sites. This industrial site was operated adjacent to a major river in southeast Wyoming for nearly 100 years. Waste

Table 3. Spearman coefficients of rank correlation for unionized ammonia (NH_3) concentrations versus fathead minnow (Pimephales promelas) survival and growth and Ceriodaphnia dubia survival and reproduction in Crow Creek water and interstitial water collected downstream from the Frontier Oil Refinery NPDES discharge.

Comparison[b]	Spearman coefficient of rank correlation[a] Crow Creek waters	Interstitial waters	Combined data
$[NH_3]$ vs. fathead minnow survival	-0.81 ** (12)	-0.07 (7)	-0.78 *** (19)
$[NH_3]$ vs. fathead minnow growth	-0.20 (4)	-0.23 (7)	-0.14 (11)
$[NH_3]$ vs. Ceriodaphnia survival	-0.67 * (12)	0.72 (7)	-0.60 ** (19)
$[NH_3]$ vs. Ceriodaphnia MOA reproduction	-0.67 * (12)	0.29 (7)	-0.58 ** (19)

[a]Sample sizes are shown in parentheses below the coefficients of rank correlation. Significance levels are indicated as follows: * = P < 0.05, ** = P < 0.01, P < 0.001.

[b]For these correlations, concentrations of unionized ammonia (NH_3) were expressed as mg/L; fathead minnow and Ceriodaphnia survival values were expressed as percent survival; fathead minnow growth values were expressed as mg/fish; and Ceriodaphnia MOA reproduction values were expressed as total offspring/female.

management practices for the various liquids and solids used in several wood preservation processes did not exist or were haphazard. Accumulation of this material on the surface and below ground led to severe groundwater/riverine pollution. Because the contaminants are a complex mixture of organic and inorganic chemicals, potential environmental fates and effects are difficult to predict at this site.

In this study, we used aquatic organisms to integrate the effects of the numerous biologically available pollutants in river water and interstitial sediment water. Fathead minnow 96-h acute toxicity and 7-d survival and growth tests demonstrated that toxic ground waters underlaid the Laramie River adjacent to the UPTTP. During June, July and August 1985, interstitial water withdrawn from river sediments decreased either survival or growth of the fish. Relocating the Laramie River in September 1985 to a new channel west of its previous location appeared to remove the immediate sediment pollution problem, as evidenced by no adverse effect on survival or growth of fathead minnows in the October groundwater sample. However, further studies would be required to confirm that groundwater contaminants do not migrate to the new river channel in the future. Although the Ceriodaphnia 48-h LC50 for interstitial water in June 1985 was approximately equal to the fathead minnow 96-h LC50, Ceriodaphnia 7-d survival and reproduction appeared to be less sensitive than fathead minnow 7-d survival and growth at the Laramie River site. Ceriodaphnia survival and reproduction were significantly decreased only in the June interstitial water. Because the waters used for the Ceriodaphnia tests were the same as those used for the fathead minnow tests, it appears that Ceriodaphnia were simply more tolerant of the pollutants at the UPTTP. Therefore, based on Ceriodaphnia tests only, it was not possible to determine whether rechannelization changed the quality of ground water underlying the two river channels in August and

October 1985.

Chemistry data suggest that trace organic compounds, as indicated by higher dissolved organic carbon concentrations (Appendix Table B-1) and the polynuclear aromatics identified in the HPLC scan of the June 1985 interstitial water (Fig. 9), may have been a major cause of toxicity in the interstitial waters. No routine water chemistry parameters or trace elements in the interstitial water and river water differed considerably from their concentrations in corresponding upstream control waters.

Survival and reproduction of control animals in the *Ceriodaphnia* 7-d tests were less variable than survival and growth of control animals in the fathead minnow 7-d tests at the Laramie River study site. Control *Ceriodaphnia* survival was always 100%, and average reproduction ranged from 25 offspring/female in July to 12 offspring/female in August. Control fathead minnow survival ranged from 55% in June to 85% in August, whereas average weights ranged from 0.27 mg/fish in June to 0.74 mg/fish in August.

Monthly sampling during the summer of 1985 demonstrated two important aspects of groundwater/riverine systems, such as the Laramie River. First, ground water entering the river adjacent to the UPTTP was consistently more toxic than river water, indicating considerable dilution by the river water. Second, toxicity at all sampling locations, including the interstitial-water sample, varied temporally. This is not surprising, because the flow rate of the Laramie River (Fig. 16) follows a hydrograph pattern typical of surface waters in the western United States, wherein flow rates are much higher during late spring and early summer than during other seasons. Thus, stream discharge and hydrologic flow of ground water into the adjacent river vary considerably. There are periods during high flow when the UPTTP site gains water from the river (CH2M/Hill 1985), temporarily reversing the normal flow of ground water.

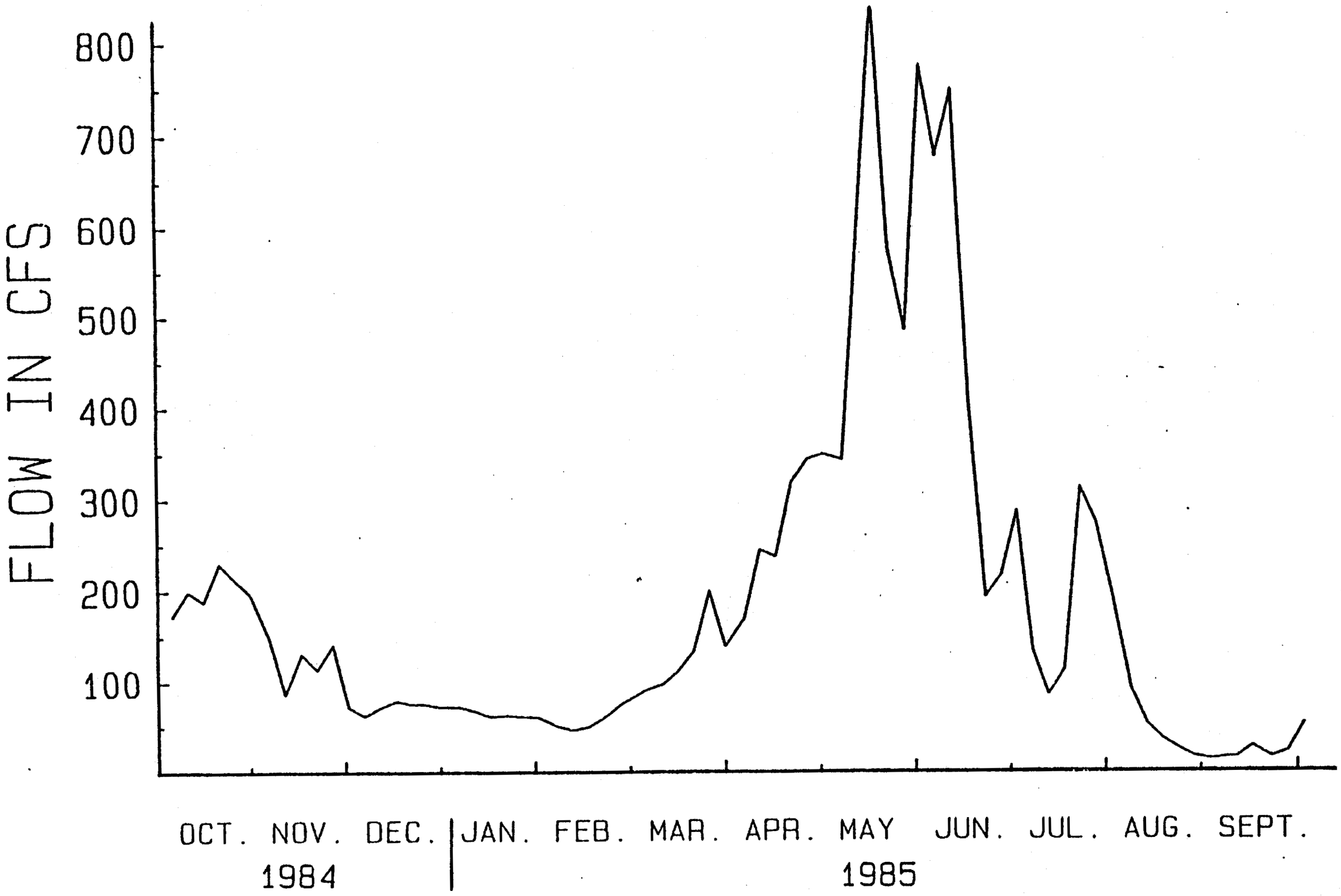


Figure 16. Laramie River hydrograph from October 1984 to September 1985. Data were compiled from unpublished files at the USGS office in Cheyenne, Wyoming.

Hence, toxicity of ground water and river water can be expected to vary temporally.

Crow Creek Study Site

In Crow Creek adjacent to Frontier Oil Refinery, surface water may be contaminated due to groundwater pollutants from the old refinery facilities and due to current surface-water releases at a permitted NPDES discharge. Crow Creek has been judged by the Wyoming Game and Fish Department to be without sufficient hydrologic qualities to support fish life (John Wagner, Wyoming Department of Environmental Quality, personal communication). Because of that assessment, Crow Creek is classified as a Class IV (lowest ranking) stream by the Wyoming Department of Environmental Quality and is not protected for aquatic life. Instead, NPDES discharge limitations for the refinery are computed according to U.S EPA Best Available Technology (BAT) treatment guidelines for a refinery of that design and size. BAT guidelines are based on concentrations of individual chemicals in effluents and not on biological effects in receiving waters. For the refinery effluent, limitations are placed on average and maximum discharges for several parameters including pH, biological oxygen demand, chemical oxygen demand, total phenols, ammonia, sulfide, oil and grease, total suspended solids, total chromium, and hexavalent chromium.

Although monthly NPDES reports filed by Frontier Oil Refinery (available for inspection at the Wyoming Department of Environmental Quality, Cheyenne, Wyoming) indicate that the NPDES discharge was always in compliance with limitations imposed by the Wyoming Department of Environmental Quality during this two-year study, acute toxicity tests and 7-d ambient toxicity tests indicated consistent adverse effects on survival, growth or reproduction of fathead minnows and *Ceriodaphnia* in Crow Creek water and interstitial water collected downstream from the discharge.

The magnitude of the biological response variables in waters collected downstream from the NPDES discharge appeared to be negatively related to the concentration of unionized ammonia (Figs. 14 and 15). For example, Spearman coefficients of rank correlation were negative and highly significant ($P < 0.01$) for the following relationships: [NH_3] vs. fathead minnow survival, [NH_3] vs. *Ceriodaphnia* survival, and [NH_3] vs. *Ceriodaphnia* MOA reproduction (Table 3). Furthermore, NH_3 concentrations > 0.4 mg/L always caused 100% mortality in fathead minnow and *Ceriodaphnia* 7-d tests. But at NH_3 concentrations < 0.4 mg/L, survival, growth and reproduction varied widely (Figs. 14 and 15).

Thurston et al. (1983) reported 96-h LC50 values between 1.8 and 3.4 mg/L of unionized ammonia at 22°C for fathead minnow fry and adults, while John W. Arthur and coworkers (U.S. EPA, Monticello, Minnesota, unpublished data) determined a 96-h LC50 of 2.6 mg/L of unionized ammonia at 26°C for fathead minnow fry. No toxicity data are available for fathead minnow larvae exposed to ammonia in 7-d survival and growth tests. Thus, there appeared to be sufficient unionized ammonia (0.07-0.90 mg/L) present in Crow Creek water and interstitial water below the NPDES discharge to account for some sublethal effects on the fish. However, the percent contributions of NH_3 to observed toxicity cannot be computed based on the limited data available in the literature.

We are aware of no similar toxicity test data for *Ceriodaphnia*. But Arthur and coworkers determined a 96-h LC50 of 1.3 mg/L of unionized ammonia at 20°C for adults of another cladoceran, *Simocephalus vetulus*, and DeGraeve et al. (1980) reported a 48-h LC50 of 1.16 mg/L for unionized ammonia at 14°C for *Daphnia pulicaria*. Although it is difficult to infer toxic effects concentrations across species, those data suggest that there may also have been sufficient unionized ammonia present to account for some sublethal effects on the Ceriodaphnia in our tests.

This apparent dominance of ammonia toxicity, especially at NH_3 concentrations > 0.4 mg/L, should be interpreted cautiously because other inorganic or organic contaminants could also have contributed to the adverse effects on survival, growth, and reproduction. The high variability in biological responses observed at < 0.4 mg NH_3/L could be explained in several ways, including: 1) NH_3 concentrations were too low to cause the observed biological responses, which were instead caused by other toxicants whose concentrations were not highly correlated with NH_3 concentrations; 2) bioavailability of NH_3 differed among the samples due to their complex and variable chemical composition, thus causing high variability in the intensity of biological responses at similar ammonia concentrations; or 3) synergistic and/or antagonistic interactions between NH_3 and other chemical species present in those waters caused high variability in the observed biological responses. Given the chemical complexity of the water samples, we cannot currently reject any of those explanations.

For example, Cr concentrations were > 10 ug/L in Crow Creek water below the NPDES discharge from June 24 to September 17, 1986, providing another possible chemical explanation for the observed toxicity. However, only one sample at this same location contained a Cr concentration > 10 ug/L from June 13, 1985 to April 29, 1986. Furthermore, Cr concentrations in the corresponding groundwater samples collected from June 24 to September 17, 1986 were $\leq$ 4 ug/L. Zinc concentrations were > 10 ug/L but $\leq$ 60 ug/L in Crow Creek water below the NPDES discharge on June 24 and July 21, 1986.

The consistent adverse biological effects observed in Crow Creek water below the NPDES discharge demonstrate that the fathead minnow and Ceriodaphnia 7-d ambient toxicity tests were capable of detecting the presence of instream contaminants. However, in this study the migration of contaminated ground water is difficult to infer from surface-water toxicity tests alone. Temporal variability in biological response at a given sampling location precluded assigning a biological response "fingerprint" to a given pollution source, and the chemical complexity of these waters made it difficult to assign responsibility for observed biological responses to individual toxicants.

Several times, Crow Creek water upstream from the NPDES discharge, but still adjacent to the refinery, caused adverse biological effects. For example, fathead minnow growth was significantly lower at the refinery sampling location compared to the upstream control on July 17, 1985 and February 24, 1986, whereas Ceriodaphnia survival and growth were lower at the refinery and at Morrie Avenue bridge (adjacent to the SWSI Site) on August 20, 1985 and July 21 and August 4, 1986. Additionally, Ceriodaphnia survival at Morrie Avenue bridge was significantly less than at the upstream control on June 13, 1985 and August 18, 1986.

On each of those dates, we found no obvious chemical constituent in Crow Creek water that would have been responsible for the observed biological effects. Although Zn concentrations in Crow Creek water ranged up to 90 ug/L, the EPA Criteria Document for Zn (USEPA 1980) lists 570 ug/L as the value for protecting aquatic life in waters with a hardness of 200 mg/L as $CaCO_3$ (near the lower end of the range of hardness values for Crow Creek water during this study). Additionally, chemical analyses by other investigators support this contention. High concentrations of several U.S. EPA organic priority pollutants occur in ground water < 0.5 km from Crow Creek, yet none have been found in groundwater wells immediately next to Crow Creek. For example, benzene, ethylbenzene, toluene, xylene and 2,4-dichlorophenol were detected as high as 12,000, 9900, 8500, 7600 and 1500 μg/L, respectively, in ground water on the refinery property; but they were reported below detection limits at the creek in a recent summary of chemical analyses of monitoring wells drilled at the refinery (Robert Elbert and Associates 1986). In the same report, total organic halogen (TOH) concentrations were always < 100 μg/L and total organic carbon (TOC) concentrations were always < 100 mg/L adjacent to Crow Creek.

The lack of identifiable toxicants upstream from the NPDES discharge does not invalidate the toxicity test results. Storm sewer runoff or unknown spills and discharges upstream may account for some of the toxicity. However, the August 20, 1985 samples were collected following the massive flood in Crow Creek. The SWSI Site, where wastes from previous refinery operations were buried or stored at the surface, was most likely saturated during the torrential rain and hail storms. Hence, polluted ground water from that site may have contributed at least some of the contamination that was detected biologically in Crow Creek in August 1985. And although Crow Creek water killed all Ceriodaphnia at Morrie Avenue bridge and the refinery sampling locations, its toxic and reproductive effects were lessened below the NPDES discharge, less than 1 km downstream from the Morrie Avenue bridge (Figs. 3 and 4). Either physical/chemical processes (e.g., sediment adsorption, photolysis, volatilization, complexation with chemical constituents in the NPDES discharge) or biological processes (e.g., microbial degradation) are probably responsible for the decreased downstream effects.

Fathead minnows and Ceriodaphnia varied in their relative sensitivity to toxicants in Crow Creek water, depending on sampling location and sampling date. A potential reason for this variability in sensitivity would be the presence of different contaminants at different sites and at different times of the year. But results from Crow Creek illustrate that it is useful to have data from more than one species when evaluating toxic effects in receiving waters. As a minimum, we believe that the fathead minnow and an invertebrate should be tested.

Temporal and spatial patterns for fathead minnow growth at the control and the three refinery sampling locations in Crow Creek were relatively simple (Fig. 17). Mean weight per fish in surface waters collected at the upstream control, downstream from the Morrie Avenue bridge, and adjacent to the refinery did not vary considerably between October 1985 (when we switched to four replicate beakers per sampling location) and September 1986, and all three sampling locations tracked each other well. However, significant decreases in growth occurred in surface water collected downstream from the NPDES discharge, and the temporal variations at that sampling location did not track the temporal variations at the other locations.

With the exception of the July 21, 1986 sampling location downstream from the Morrie Avenue bridge (adjacent to the SWSI Site), fathead minnow growth in all of the interstitial waters (including the sampling location downstream from the NPDES discharge) tracked the growth in Crow Creek waters upstream from the NPDES discharge (Figs. 17 and 18). This could indicate migration of ground water into surface water, or vice versa. However, it appears that surface-water toxicity downstream from the NPDES discharge is dominated by the effluent and not by underlying ground water.

Contrary to fathead minnow growth, Ceriodaphnia reproduction varied considerably in surface waters (Fig. 19). Reproduction in Crow Creek water collected downstream from the Morrie Avenue bridge and adjacent to the refinery track each other well. However, reproduction at those sampling locations did not track reproduction in Crow Creek water collected at the upstream control

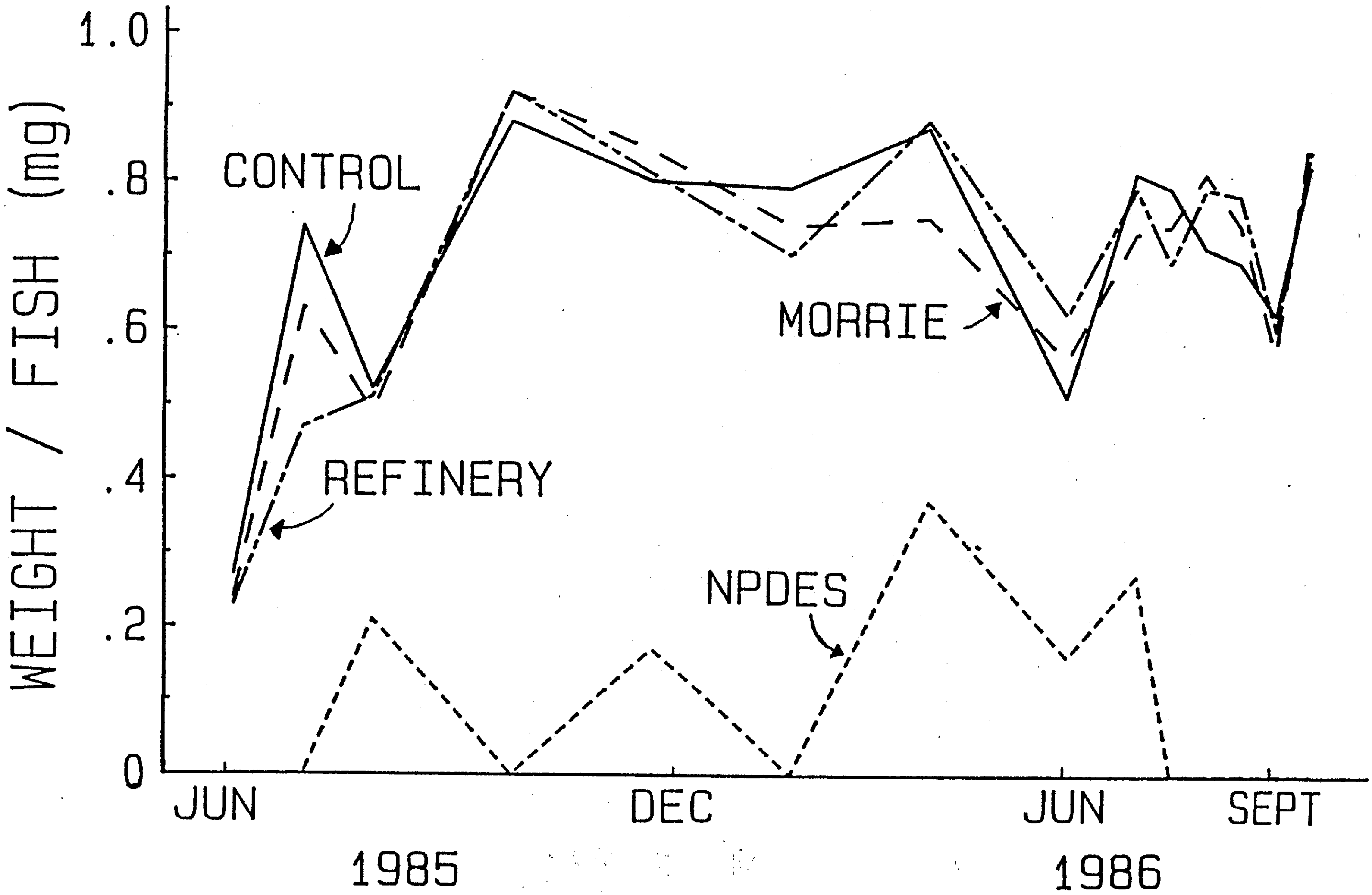


Figure 17. Temporal and spatial trends in fathead minnow (Pimephales promelas) growth in Crow Creek water from June 1985 to September 1986.

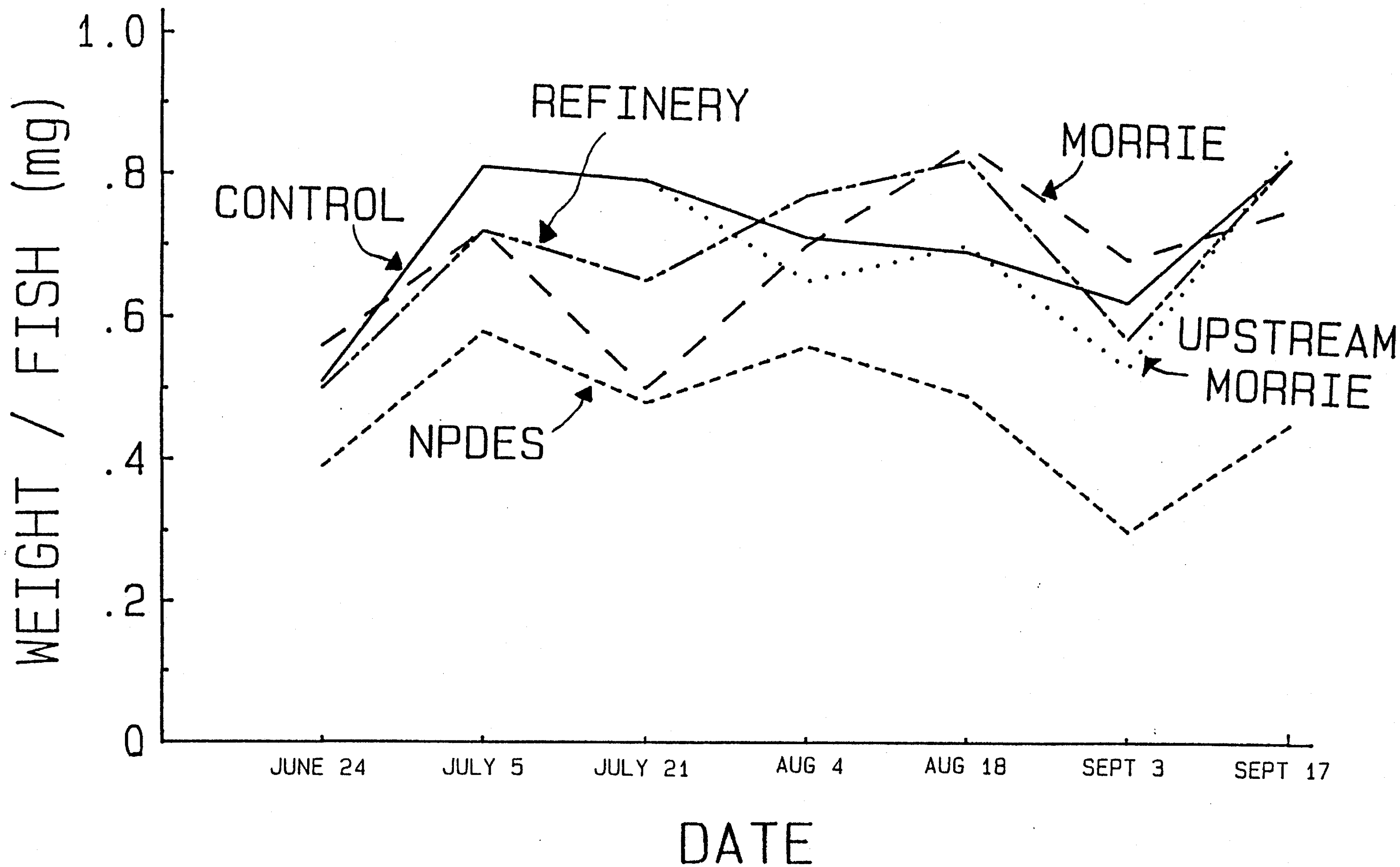


Figure 18. Temporal and spatial trends in fathead minnow (Pimephales promelas) growth in Crow Creek upstream control and interstitial water from June to September 1986.

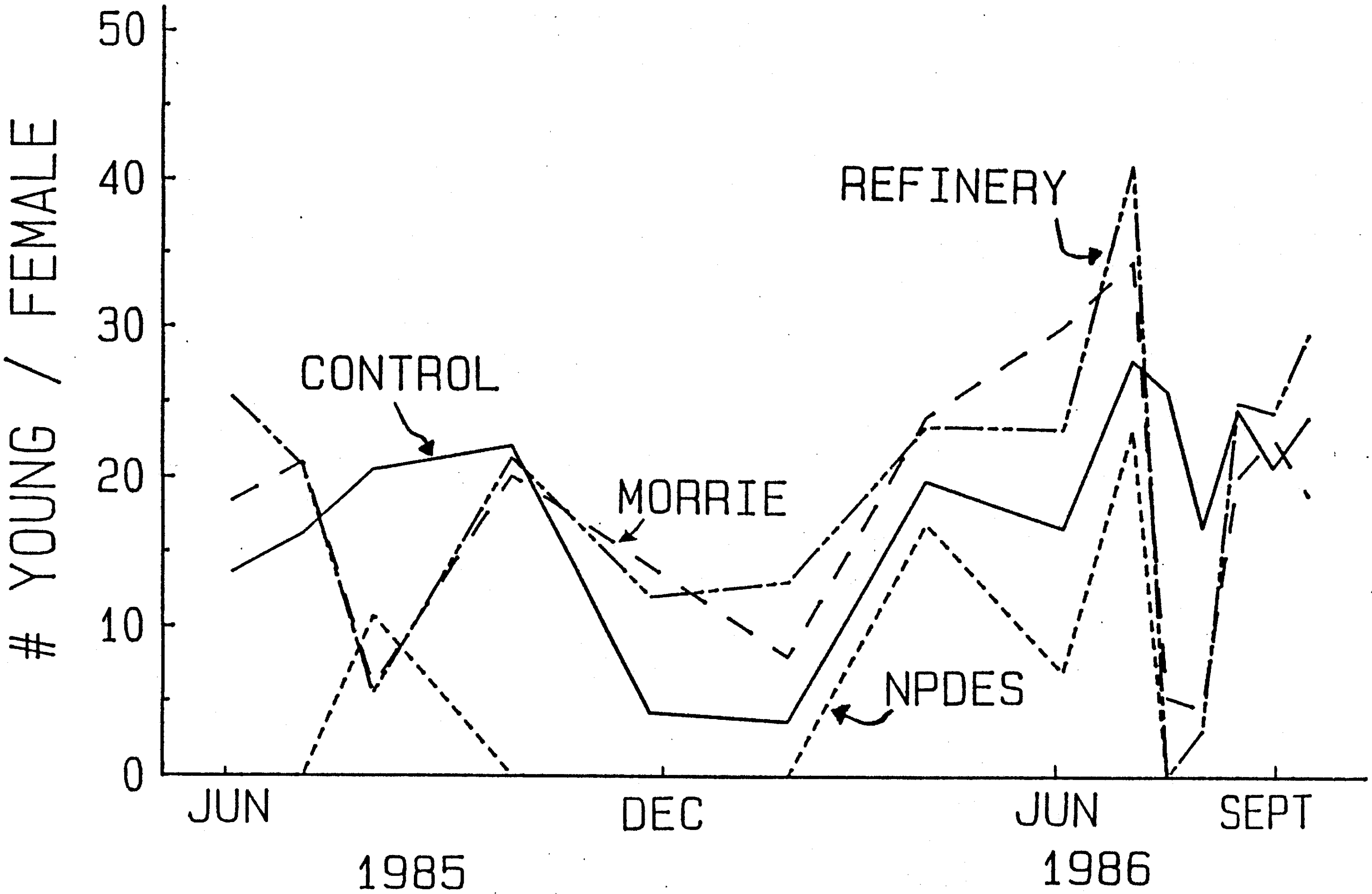


Figure 19. Temporal and spatial trends in Ceriodaphnia dubia MOA reproduction in Crow Creek water from June 1985 to September 1986.

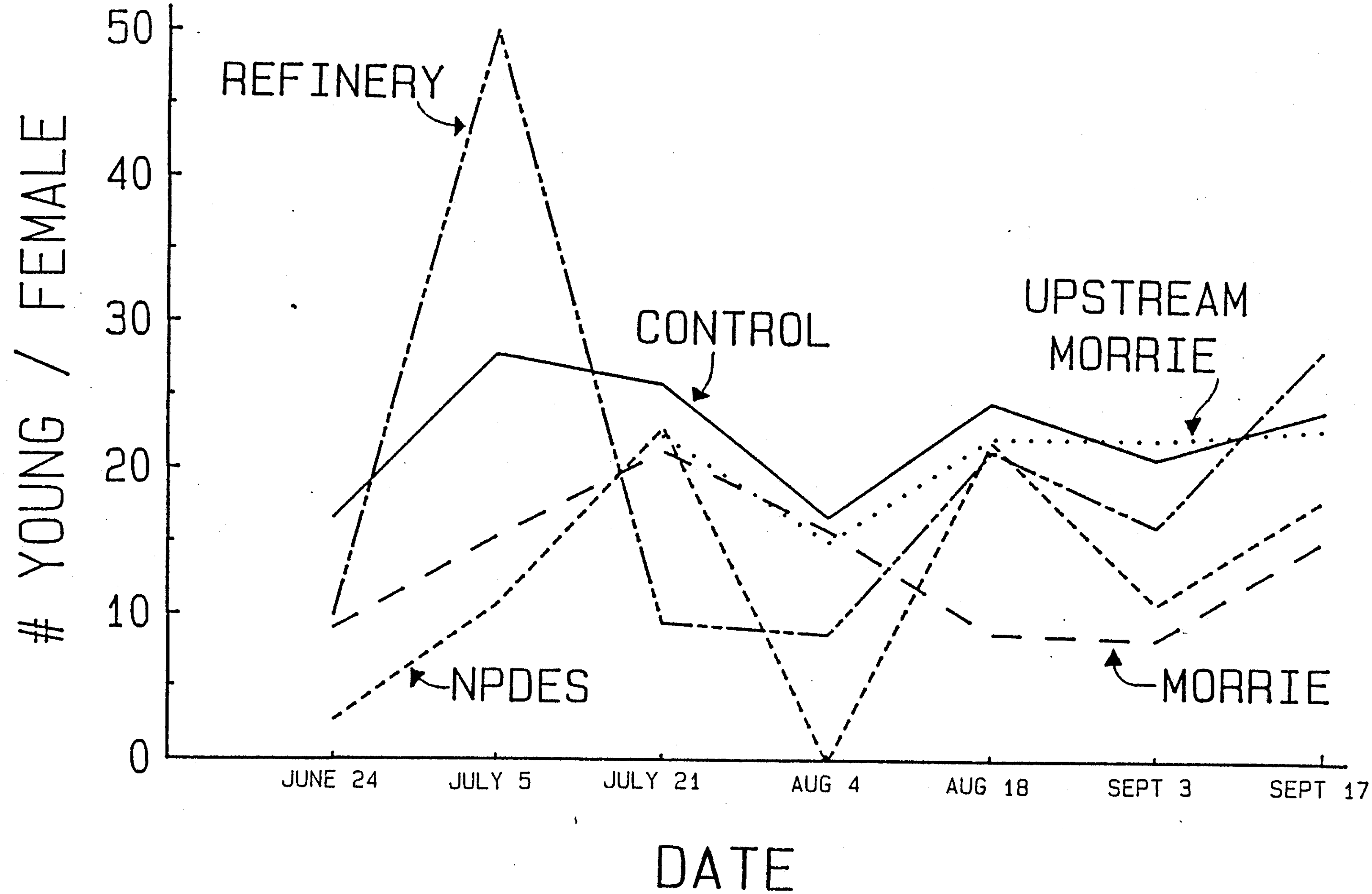


Figure 20. Temporal and spatial trends in <u>Ceriodaphnia dubia</u> MOA reproduction in Crow Creek upstream control and interstitial water from June to September 1986.

and downstream from the NPDES discharge; those latter two locations usually were significantly different from each other and did not track each other well. Reproduction in interstitial waters also varied and did not track each other or upstream surface water very well (Figs. 19 and 20). Based on Ceriodaphnia results alone, it does not appear that surface water and ground water communicated with each other at any sampling location. Figures 19 and 20 shows considerable variability in Ceriodaphnia reproduction between samples collected approximately every two weeks from June to September 1986. But if the July 21 and August 4, 1986 samples had not been collected, the amount of temporal variability would have appeared to be much less. Similarly, removing the July 5, 1986 sample would have decreased the apparent variability. We might also have documented wide excursions in biological effects during Summer 1985 if we had sampled more frequently than once every one or two months.

As discussed for the Laramie River study site, temporal variability in toxicity at Crow Creek sites would be expected to depend on stream flow rates and amount of groundwater infiltration. The variability in storm sewer and surface runoff from a municipal setting such as Cheyenne increases the difficulty of ascribing adverse instream biological effects to permitted discharges or specific groundwater pollution sources. Without detectable chemical "fingerprints" of contaminants to complement the toxicity test results, statements can only be made concerning the quality of the receiving water for supporting aquatic life. Such high-sensitivity chemical analyses are not routinely performed in the initial phases of a pollution investigation and thus may limit the extent of responsibility for observed toxicity that can be atributed to potential pollution sources.

Moreover, the temporal and spatial variability we observed during this two-year study demonstrates that instream toxicity at an apparently simple industrial site can vary widely. Monthly sampling at only one upstream location could easily have missed important pollution events or confounded interpretations of downstream pollution. These results present a challenge to regulators and industry representatives to select cost-effective monitoring plans for suspected pollution sources.

We encountered one major problem with *Ceriodaphnia* tests at Crow Creek. Although survival was always > 60% at the upstream control, MOA reproduction varied from approximately 28 offspring/female on July 5, 1986 to 4 offspring/female on December 12, 1985 and February 24, 1986. Because of low upstream control reproduction during the winter months, it was difficult to demonstrate adverse reproductive effects at downstream sampling locations. This may have been due to a chemical constituent that was present in the water at the upstream control, but was degraded or not bioavailable by the time the stream water reached the downstream sampling locations. Or it may also have been due to fewer bacteria in upstream control water during winter, thus providing less food for *Ceriodaphnia* adults to eat. We did not notice a similar problem of low reproduction in upstream control water at the Laramie River study site, perhaps because we did not test that water during winter months.

Evaluation of Toxicity Tests

In addition to establishing the potential for migration of contaminated ground water into surface waters at the Laramie River and Crow Creek study sites, an important objective of this study was to evaluate the utility of fathead minnow and *Ceriodaphnia* ambient toxicity tests as routine biological monitoring techniques. In the following sections, we address difficulties with (1) current procedures for culturing and testing these animals and (2) evaluation of toxicity test results.

Culturing and Testing Procedures. A large amount of fathead minnow acute and chronic toxicity test data for single compounds and effluents is available in the published literature (e.g., Brooke et al. 1984). Fathead minnows are easily maintained in a laboratory, and an abundant supply of newly hatched larvae can be produced routinely for starting ambient toxicity tests. Larvae are easy to count with an unaided eye as long as the test water is not turbid. Siphoning the exposure chambers without removing and/or injuring the fish is tedious, but with practice the daily time requirement for handling a test comprising four replicate beakers for each of six test waters is approximately three hours. Necessary equipment to conduct the fathead minnow test is available or can be easily fabricated in most aquatic biology laboratories.

An important aspect of the fathead minnow test that we changed during Year 1 of this study was the number of replicates per exposure water. For all of the Laramie River sampling dates and for June 13, July 17, and August 20, 1985 at Crow Creek, we tested only two replicate chambers containing ten fish for each test water. However, variability of average fish weights between replicates was sometimes high. Furthermore, variances among exposure waters often were not homogeneous. Therefore, we changed to four replicates per exposure water in the Crow Creek study after August 1985 and obtained much more consistent results. This allowed us to simplify the statistical analyses of fish weights and increased our theoretical statistical power by a factor of approximately 1.7. Based on results of a recent round-robin interlaboratory study of the fathead minnow 7-d growth and reproduction test, DeGraeve et al. (1987) recommended three replicates per exposure water as the most cost-effective test design.

Not as much acute and chronic toxicity data are available in the published literature for Ceriodaphnia dubia as there are for fathead minnows. But due to the increasing popularity of the Ceriodaphnia 7-d test, this will not be a major limitation in the near future.

A major drawback to the Ceriodaphnia test is that nutritional requirements for Ceriodaphnia are not well known. Occasionally, cultures in our laboratory and laboratories of other investigators around the country "crash" (die off) or produce males and diapause eggs, both of which indicate stress. New cultures must be started when that occurs. But with proper rotation of cultures, there are usually enough neonates available to begin a test at any time. Current research at several laboratories across the country should help to resolve problems related to Ceriodaphnia nutrition and culturing (see RELATED RESEARCH).

Related to the culturing problem, we also had difficulty maintaining adequate laboratory-water controls during the Ceriodaphnia 7-d toxicity tests. Survival and reproduction were erratic and often much less than in the upstream control waters. When upstream controls perform well, there is no need for laboratory controls. However, good survival and reproduction in laboratory water are needed to demonstrate that experimental techniques are not to blame when survival or reproduction in upstream controls is low.

Daily observation of Ceriodaphnia adult survival and numbers of offspring is not difficult, but it requires a low-power dissecting microscope to view the animals. Adults can be transferred to fresh test water every other day fairly quickly, so the daily requirement for a test with ten replicates of six or seven test waters is approximately two hours. As with the fathead minnow test, equipment needed for the Ceriodaphnia 7-d test is available in most aquatic biology laboratories.

Interpretation of Results. Ambient toxicity testing poses several problems related to experimental design and statistical analysis that are not encountered in traditional serial-dilution toxicity tests. These problems should be resolved before data are analyzed, and ideally even before sampling and toxicity testing are begun.

In traditional serial-dilution toxicity testing, survival, growth or reprduction in several dilutions of a given toxicant are compared to a laboratory control water containing no toxicant. One-way Analysis of Variance (ANOVA) is usually used to test for significant differences, and a one-tailed post hoc comparison method (e.g., Dunnett's) is used to test for significant decreases in each toxicant dilution relative to the laboratory control. Usually, there is no conflict regarding appropriate controls and statistical analysis techniques.

As we mentioned in the Statistical Methods section of this report, though, there can be several choices for a control water in ambient toxicity tests, depending on the purpose of the investigation. Upstream surface water is an obvious choice for comparisons with downstream surface waters near potential pollution sites. But in our study, *Ceriodaphnia* in upstream controls produced few offspring on December 12, 1985 and February 24, 1986. This made it difficult to detect significant decreases in reproduction in downstream waters, and some downstream waters significantly increased reproduction relative to the upstream control. Furthermore, the comparison of interest may be water collected immediately downstream from a potential pollution source versus water collected immediately upstream. Both of those waters could adversely affect survival, growth or reproduction relative to the upstream control; yet if they are not different from each other, the potential pollution source may not be causing additional adverse biological effects in the stream or river. And finally, if ground water is tested in conjunction with surface waters, a control for comparison with the ground water may be difficult to select. Therefore, we suggest that all comparisons of interest be identified and an appropriate control be chosen before the sampling locations are selected. In some cases, more than one control may need to be identified (e.g., a different control for ground waters and surface waters). Most important, it should be decided in advance under what conditions a given water will be classified as causing significant adverse biological effects.

Once the comparisons of interest are identified, the choice of a statistical method will become easier. Dunnett's Method (Dunnett 1955, 1964) is designed to compare several treatments to one control (e.g., several downstream sampling locations to one upstream sampling location). It is slightly more conservative than a traditional $\underline{t}$ test, but is more liberal (i.e., it is more likely to indicate that a given difference between treatment means is significant) than are Tukey's HSD Method or Scheffe's Method (Neter et al. 1985). A one-tailed Dunnett's test is appropriate when testing only for significant decreases relative to the control; a two-tailed Dunnett's test is appropriate when testing for significant differences (decreases or increases) relative to the control.

Tukey's HSD Method is more appropriate when adjacent sampling locations are to be compared, which will often occur when there are several potential pollution sources along the sampling transect or when surface waters and ground waters are analyzed concurrently. But because it is a more conservative test, Tukey's HSD Method is less likely than Dunnett's Method to indicate significant decreases relative to the upstream control. Additionally, only two-tailed comparisons are possible with Tukey's HSD Method.

Finally, it should be remembered that as more sampling locations are added to an analysis, it usually becomes more difficult to identify significant differences between any two sampling locations in the post hoc ANOVA comparisons. This occurs because the critical distance between treatment means must be increased to compensate for the increased number of non-independent comparisons that the additional sampling locations will entail.

Therefore, inferences about potential pollution sources can easily be confounded by (1) inappropriate selection of the type and location of samples, (2) the total number of samples tested, and (3) the post hoc ANOVA method used to identify significant differences. Given the variety of situations that might be encountered at different study sites where ambient toxicity tests are conducted, inflexible guidelines for study designs and statistical analyses would be counter-productive. However, we recommend that these decisions be made as early as possible in the design of a study.

Toxicity Testing vs. Chemical Analyses

One of the goals of the National Pollutant Discharge Elimination System (NPDES) is to control the discharge of "toxics in toxic amounts" into the nation's waterways. In the past, it has been based on water quality criteria for 129 pollutants. Many contract analytical laboratories are now capable of routinely analyzing soil and water samples for these compounds. In addition, regulatory agencies and environmental consultants have used the 129 priority pollutants as a focus for assessing a wide range of environmental contamination problems.

Yet cumulative experience with this system has indicated that many pollution problems are not addressed adequately. Although 129 priority pollutants are identified, thousands of other chemicals are discharged to aquatic systems and can cause adverse biological effects. And because the

NPDES system is currently based on laboratory toxicity data for single compounds, discharge limitations are not always environmentally realistic. Contaminant exposures in aquatic systems often occur as complex mixtures of compounds, containing perhaps some priority pollutants and many compounds not on the priority list. Therefore, previous methods for determining discharge limitations based on Best Available Technology and concentrations of individual chemicals may have underprotected or overprotected aquatic life in receiving waters.

Interest has now turned toward addressing the biological impacts of receiving-water contamination. For example, assessing the hazard of effluent mixtures was the topic of a recent workshop attended by many prominent aquatic toxicologists (Bergman et al. 1986). And results of field research and test standardization related to effluent testing have been published by the U.S. EPA (e.g., Mount et al. 1984, Horning and Weber 1985). Results of those meetings and toxicity studies indicate that biological tests contribute considerable information cost-effectively for evaluating the potential or realized effects of surface-water contamination. And our studies on the Laramie River and Crow Creek demonstrate that ambient toxicity tests can be used to detect contaminated ground water and surface water. We do not believe that toxicity tests will supplant chemical analyses in pollutant studies. Instead, toxicity tests complement chemical analyses. Without chemical analyses, it is difficult to identify sources of toxicity; without toxicity tests, it is difficult to interpret the biological significance of the presence of pollutants in an aquatic system.

Cost Comparisons

Current costs of fathead minnow and *Ceriodaphnia* 7-d ambient toxicity tests, fathead minnow and invertebrate acute toxicity tests, and various chemical analyses are listed in Table 4. Ranges of prices for toxicity tests were obtained from price lists and a telephone survey of five university and private toxicity testing laboratories. Higher prices were quoted by laboratories that anticipated having to run an ambient toxicity test more than once to satisfy some clients. Prices in Table 4 indicate that the 7-d ambient tests cost approximately two to three times as much as corresponding acute toxicity tests. Based on results of this study, we believe that 7-d ambient tests are cost-effective compared to shorter-duration acute tests. For example, fathead minnow and *Ceriodaphnia* 7-d tests showed an adverse effect of Crow Creek water collected below the refinery discharge on all fourteen sampling dates during the two-year study. However, 96-h fathead minnow and 48-h *Ceriodaphnia* acute tests would have identified an LC50 < 100% of full-strength stream water in only ten of those fourteen tests. And in upstream waters where only fathead minnow growth or *Ceriodaphnia* reproduction was adversely affected, acute toxicity tests would not have indicated the presence of toxicants.

A suite of routine chemistry parameters (pH, conductivity, alkalinity, hardness, etc.) and major inorganic ions would cost approximately $1000 to $1800 for six water samples, the same number analyzed in an acute or ambient toxicity test. The cost of one acute test is therefore approximately one-half the cost of this suite of routine chemical analyses, and one ambient toxicity test costs approximately the same to twice as much as the routine chemical analyses. Since total ammonia below the NPDES discharge on Crow Creek was the only major inorganic toxicant that we could identify in this study, much of the observed toxicity would not have been predicted from routine chemical analyses. Trace elements did not account for much of the toxicity either, yet a suite of

Table 4. Comparison of costs for toxicity tests and chemical analyses.

Test	Range of costs[a]
Toxicity tests	
Fathead minnow 7-d survival and growth (4 replicates x 6 exposure waters)	\$1100 - \$1800
Ceriodaphnia 7-d survival and reproduction (10 replicates x 6 exposure waters)	\$800 - \$1800
Fathead minnow 96-h LC50 (3 replicates x 6 exposure waters)	\$550 - \$825
Invertebrate 48-h LC50 -- *Ceriodaphnia* or *Daphnia* (3 replicates x 6 exposure waters)	\$400 - \$825
Chemistry analyses	
Routine chemistry parameters (pH, conductivity, alkalinity, hardness, ammonia, total phenols, oil and grease, solids; 6 samples)	\$630 - \$1200
Major inorganic ions (K^+, Na^+, Ca^{2+}, Mg^{2+} by ICP or AA, and Cl^-, NO_3^-, SO_4^{2-} by ion chromatography; 6 samples)	\$390 - \$578
Trace elements (20 elements by AA; 6 samples)	\$1200 - \$3000
Dissolved organic carbon (6 samples)	\$130 - \$210
Reverse-phase HPLC gradient fingerprints (6 samples)	\$510
GC-MS scan of major organics (6 samples)	\$300 - \$600
GC-MS priority pollutant organics (6 samples)	\$3750 - \$7650

[a]Toxicity test costs were compiled from price lists and a telephone survey of six university and private toxicity testing laboratories. Chemical analysis costs were compiled from price lists of five private analytical chemistry laboratories; not all laboratories reported prices for all chemical analyses listed.

20 trace elements would have cost from the same to twice as much as one ambient toxicity test. Organic chemicals explained much of the toxicity of ground water at the Laramie River site. Relatively simple dissolved organic carbon analyses and reverse-phase HPLC fingerprints would have cost about $500 total for 6 samples and would have indicated the presence of potentially toxic organics, although their identities would still not have been known. A GC-MS scan of only the ten major organics in all 6 water samples would have cost $300 to $600, approximately equal to the cost of an acute toxicity test. And finally, a complete priority pollutant scan of six water samples, which would probably still not have identified all potential inorganic and organic toxicants in Laramie River or Crow Creek waters, would have cost approximately $3750 to $7650.

Therefore, we believe that ambient toxicity tests are cost-competitive with chemical analyses and provide additional information concerning potential biological effects of toxicants that cannot be predicted reliably from a list of all chemical constituents in a water sample.

SUMMARY AND CONCLUSIONS

In this two-year study, we (1) evaluated the utility of U.S. EPA fathead minnow and *Ceriodaphnia dubia* ambient toxicity tests as monitors of the effects of groundwater pollution that enters surface waters, (2) compared the sensitivity of those biological tests to the sensitivity of chemical analyses for detecting the presence of groundwater contaminants, and (3) assessed temporal variability of groundwater and surface-water contamination in the Laramie River and Crow Creek in southeast Wyoming. Major results are as follows:

- Toxic ground water underlaid Laramie River sediments adjacent to the former Union Pacific Tie Treatment Plant. This ground water was heavily contaminated with water soluble organic compounds typical of creosote oil that permeated the soils adjacent to the river and also underlaid Laramie River sediments approximately 30 m upstream from the groundwater sampling location.
- Laramie River water flowing directly over the sediments in which the oil body lay and at two downstream locations did not adversely affect survival, growth or reproduction of fathead minnows and Ceriodaphnia, compared to the upstream control during June, July and August 1985.
- The Laramie River was rechanneled in September 1985 to avoid oil seeps and contaminated ground water. Ground water and river water collected at corresponding locations in the new river channel in October 1985 did not adversely affect survival, growth, and reproduction.
- Low concentrations of anthracene, phenanthrene and chrysene were detected in the toxic interstitial water withdrawn from Laramie River sediments in June 1985, using reverse-phase HPLC gradients. However, organics were not detected using HPLC on other sampling dates. And no trace elements, major inorganic ions or routine water chemistry parameters differed considerably from the upstream control on any sampling date.
- Crow Creek water collected 50 m downstream from Frontier Oil Refinery's NPDES discharge adversely affected fathead minnow survival or growth or Ceriodaphnia survival or reproduction relative to the upstream control on every sampling date. Often, this toxicity appeared to be

caused by high unionized ammonia (NH_3) concentrations allowed in the NPDES permit for that discharge.

- Interstitial water collected 1 m deep in Crow Creek sediments downstream from Frontier Oil Refinery's NPDES discharge adversely affected fathead minnow survival or growth or Ceriodaphnia survival or reproduction on every sampling date during Year 2, the only times interstitial waters were collected. Unionized ammonia (NH_3) may also have contributed to the observed biological effects in these waters, indicating a general area of ammonia contamination in surface-water and groundwater downstream from the NPDES discharge. However, survival, growth and reproduction were usually higher in interstitial waters than in Crow Creek waters at similar NH_3 concentrations suggesting that chromium concentrations > 10 ug/L may also have contributed to the observed toxicity in Crow Creek waters during Year 2.
- Crow Creek waters collected on August 20, 1985 upstream from the NPDES discharge, but still adjacent to the oil refinery property, were toxic to Ceriodaphnia. Those samples were collected after Crow Creek flooded because of an intense hail and rain storm on August 1, 1985; hence, the toxicity may have been caused by contaminated ground water emanating from buried wastes at the old refinery facility or by storm-sewer runoff entering Crow Creek upstream from the refinery site. No inorganic or organic contaminants could be identified as possible toxicants in those samples.
- During June, July and August 1986, Crow Creek waters and interstitial sediment waters collected adjacent to the oil refinery property adversely affected fathead minnow survival or growth or Ceriodaphnia survival or reproduction on at least one sampling date. Adverse

effects in those surface waters often occurred when the corresponding interstitial water also caused adverse effects. Yet on two consecutive sampling dates in July and August, Crow Creek water collected immediately upstream from the refinery sampling locations (above Morrie Avenue bridge) also was toxic. Therefore, it was not always possible to conclude that contaminated ground water migrated into surface waters.

These results indicate that (1) ambient toxicity tests can be used in alkaline surface waters of the western U.S.; (2) they are sensitive enough to detect contaminated ground water and surface water; (3) they may be more sensitive in some cases than routine, inexpensive chemical analyses for detecting the presence of contaminants; and (4) toxicity of contaminated ground water and an industrial discharge varied considerably during the study.

REFERENCES

American Public Health Association (APHA). 1980. Standard Methods for the Examination of Water and Wastewater. 15th Edition. American Public Health Association, Washington, DC.

American Society for Testing and Materials (ASTM). 1980. Standard practice for conducting acute toxicity tests with fishes, macroinvertebrates, and amphibians. Protocol E729-80, Annual Book of ASTM Standards. ASTM, Philadelphia, PA.

Bergman, H.L., R.A. Kimerle and A.W. Maki. 1986. Environmental Hazard Assessment of Effluents. Pergamon Press, Elmsford, NY.

Brooke, L.T., D.J. Call, D.L. Geiger and C.E. Northcott (eds.). 1984. Acute toxicities of organic chemicals to fathead minnows (Pimephales promelas). Center for Lake Superior Environmental Studies, University of Wisconsin-Superior, Superior, WI.

CH2M/Hill. 1984. Phase III report. Remedial action feasibility study: Laramie Railroad Tie Treating Plant. Draft report. Submitted to Union Pacific Railroad, Omaha, NE.

CH2M/Hill. 1985. Phase II report. Investigative research and remedial action program: Laramie Railroad Tie Treating Plant. Revised draft report. Submitted to Union Pacific Railroad, Omaha, NE.

Crossey, M.J. and H.L. Bergman. 1985. Organic contaminant transport in groundwater, surface water and surface water sediments: a case study of a wood-treating plant. Wyoming Water Research Center, University of Wyoming, Laramie, WY.

DeGraeve, G.M., R.L. Overcast and H.L. Bergman. 1980. Toxicity of underground coal gasification condenser water and selected constituents to aquatic biota. Arch. Environ. Contam. Toxicol. 9:543-555.

DeGraeve, G.M., J.D. Cooney, T.L. Pollock, N.G. Reichenbach and J.H. Dean. 1987. Round-robin study to determine the reproductibility of the 7-day fathead minnow larval survival and growth test. Draft report - January 7, 1987. Battelle Columbus Division, Columbus, OH.

Drever, J.I. 1982. The Geochemistry of Natural Waters. Prentice-Hall, Inc., Englewood Cliffs, NJ.

Druse, S.A., M.E. Cooley, S.L. Green and H.W. Lowham. 1986. Flood of August 1, 1985, in Cheyenne, Wyoming. Hydrologic Investigations Atlas HA-699, U.S. Geological Survey, Reston, VA.

Dunnett, C.W. 1955. A multiple comparison procedure for comparing several treatments with a control. J. Amer. Statist. Assoc. 50:1096-1121.

Dunnett, C.W. 1964. New tables for multiple comparisons with a control. Biometrics 20:482-491.

Emerson, K., R.C. Russo, R.E. Lund and R.V. Thurston. 1975. Aqueous ammonia equipibrium calculations: Effect of pH and temperature. J. Fish. Res. Board Can. 32:2379-2383.

Hamilton, M.A. 1986. Statistical analysis of the cladoceran reproductivity test. Environ. Toxicol. Chem. 5:205-212.

Hamilton, M.A., R.C. Russo and R.V. Thurston. 1977. Trimmed Spearman-Karber method for estimating median lethal concentrations in toxicity bioassays. Environ. Sci. Technol. 11:714-719. [See also correction in Environ. Sci. Technol. 12:417 (1978).]

Horning, W.B., III and C.I. Weber (eds.). 1985. Short-term methods for estimating the chronic toxicity of effluents and receiving waters to freshwater organisms. U.S. Environmental Protection Agency Publication No. EPA/600/4-85/014, Environmental Monitoring and Support Laboratory, Cincinnati, OH.

Lee, D.R. and J.A. Cherry. 1978. A field exercise on groundwater flow using seepage meters and mini-piezometers. J. Ceol. Ed. 27:6-10.

Milliken, G.A. and D.E. Johnson. 1984. Analysis of Messy Data. Volume I: Designed Experiments. Lifetime Learining Publications, Belmont, CA.

Mount, D.I. and T.J. Norberg. 1984. A seven-day life-cycle cladoceran toxicity test. Environ. Toxicol. Chem. 3:425-434.

Mount, D.I., N.A. Thomas, T.J. Norberg, M.T. Barbour, T.H. Roush and W.F. Brandes. 1984. Effluent and ambient toxicity testing and instream community response on the Ottawa River, Lima, Ohio. U.S. Environmental Protection Agency Publication No. EPA-600/3-84-080, Environmental Research Laboratory, Duluth, MN.

Mount, D.I. and T.J. Norberg (eds.). 1985. Validity of effluent and ambiant toxicity tests for predicting biological impact, Scippo Creek, Circleville, Ohio. U.S. Environmental Protection Agency Publication No. EPA/600/3-85/044, Environmental Research Laboratory, Duluth, MN.

Neter, J., W. Wasserman and M.H. Kutner. 1985. Applied Linear Statistical Models. Second Edition. Richard D. Irwin, Inc., Homewood, IL.

Nie, N.H., J.G. Jenkins, K. Steinbrenner and D.H. Bent (eds.). 1975. Statistical Package for the Social Sciences. Second Edition. McGraw-Hill Book Company, New York.

Norberg, T.J. and D.I. Mount. 1985. A new fathead minnow (Pimephales promelas) subchronic toxicity test. Environ. Toxicol. Chem. 4:711-718.

Robert Elbert and Associates. 1986. Assessment of groundwater conditions: Frontier Oil and Refining Company Cheyenne Refinery, Cheyenne, Wyoming. Report filed with Wyoming Department of Environmental Quality, Water Quality Division, Cheyenne, WY.

Rocky Mountain Analytical Laboratory. 1986a. Analytical results for Husky/RMT Properties, Inc., January 31, 1986. Report filed with Wyoming Department of Environmental Quality, Water Quality Division, Cheyenne, WY.

Rocky Mountain Analytical Laboratory. 1986b. Analytical results for Husky/RMT Properties, Inc., March 25, 1986. Report filed with Wyoming Department of Environmental Quality, Water Quality Division, Cheyenne, WY.

Rocky Mountain Analytical Laboratory. 1986c. Analytical results for Frontier Oil and Refining, September 19, 1986. Report filed with Wyoming Department of Environmental Quality, Water Quality Division, Cheyenne, WY.

Steadman, B.L. 1986. Xenobiotic metabolism in rainbow trout: toxicology, biochemistry and biomonitoring. Ph.D. Dissertation, University of Wyoming, Laramie, WY.

Thurston, R.V., R.C. Russo and G.E. Phillips. 1983. Acute toxicity of ammonia to fathead minnows. Trans. Amer. Fish. Soc. 112:705-711.

U.S. Environmental Protection Agency (USEPA). 1980. Ambient Water Quality Criteria for Zinc. Office of Water Regulations and Standards, Criteria and Standards Division, Washington, D.C.

APPENDIX A

Chronic Toxicity Test Results

- Laramie River: June 1985 - October 1985
- Crow Creek: June 1985 - September 1986

Table A-1. Seven-day survival and growth of fathead minnows (Pimephales promelas) and seven-day survival and reproduction of Ceriodaphnia dubia in Laramie River water and interstitial (piezometer) water from June 1985 to October 1985. Statistical comparisons were made using one-tailed tests for significant decreases in survival, growth or reproduction relative to the upstream control.[a]

		Fathead minnows		Ceriodaphnia		
Date	Site	Percent survival	Weight (mg)[b]	Percent survival	MOA total offspring/ female[b,c]	MIM total offspring/ female[b,c]
June 14, 1985	Upstream control	55	0.27 ± 0.055	100	20.8 ± 0.84	20.8 ± 0.84
	Above seep	25	0.39 ± 0.003	100	20.1 ± 0.60	20.1 ± 0.60
	Piezometer onsite	0 *	---[d] ---	0 *	0 ± 0 *	--- ---
	I-80	30	0.34 ± 0.084	100	19.1 ± 0.69	19.1 ± 0.69
	Spring Creek	70	0.31 ± 0.088	100	19.6 ± 0.73	19.6 ± 0.73
July 18, 1985	Upstream control	60	0.37 ± 0.020	100	24.9 ± 0.57	24.9 ± 0.57
	Above seep	60	0.41 ± 0.050	100	23.4 ± 0.69	23.4 ± 0.69
	Piezometer onsite	35	0.18 ± 0.023 *	100	26.3 ± 2.27	26.3 ± 2.27
	I-80	90	0.42 ± 0.004	90	20.5 ± 2.18	22.4 ± 1.11
	Spring Creek	55	0.40 ± 0.154	100	24.3 ± 0.67	24.3 ± 0.67
Aug. 9, 1985	Upstream control	85	0.56 ± 0.029	100	12.2 ± 1.60	12.2 ± 1.60
	Above seep	55	0.54 ± 0.024	100	14.8 ± 1.64	14.8 ± 1.64
	Piezometer onsite	5 *	0.35 ± 0 *	80	11.8 ± 1.29	13.6 ± 0.53
	I-80	65	0.56 ± 0.042	100	15.2 ± 1.52	15.2 ± 1.52
	Spring Creek	75	0.57 ± 0.028	100	18.5 ± 1.28	18.5 ± 1.28
Oct. 3, 1985[e]	Upstream control	60	0.74 ± 0.035	100	19.6 ± 0.50	19.6 ± 0.50
	Upstream piezometer	70	0.57 ± 0.035	100	19.6 ± 0.69	19.6 ± 0.69
	New channel onsite	53	0.64 ± 0.066	90	18.4 ± 1.83	19.9 ± 1.20
	New piezometer onsite	75	0.74 ± 0.076	100	17.8 ± 1.06	17.8 ± 1.06
	I-80	50	0.78 ± 0.049	100	17.6 ± 1.17	17.6 ± 1.17
	Spring Creek	75	0.54 ± 0.013	100	21.8 ± 0.92	21.8 ± 0.92

[a]* = significantly less than upstream control, using Fisher's Exact Test (survival tests) and Dunnett's Method (growth and reproduction tests) for comparisons of all treatments with the control at $\alpha = 0.05$.

[b]Values expressed as mean ± one standard error of the mean.

[c]MOA = mean reproduction of all females that started the test; MIM = mean reproduction of surviving females (see Toxicity Tests in METHODS for calculation procedures).

[d]--- = value could not be calculated because all of the test animals died.

[e]Laramie River was rechanneled in September 1985 (see Site Descriptions in METHODS); October 1985 stream samples were collected at corresponding locations along the new river channel.

Table A-2. Seven-day survival and growth of fathead minnows (<u>Pimephales promelas</u>) and seven-day survival and reproduction of <u>Ceriodaphnia dubia</u> in Laramie River water and interstitial (piezometer) water from June 1985 to October 1985. Statistical comparisons made using two-tailed tests for significant differences in survival, growth or reproduction among all treatments.[a]

		Fathead minnows		Ceriodaphnia		
Date	Site	Percent survival	Weight (mg)[b]	Percent survival	MOA total offspring/ female[b,c]	MIM total offspring/ female[b,c]
June 14, 1985	Upstream control	55 r	0.27 ± 0.055 r	100 r	20.8 ± 0.84 r	20.8 ± 0.84 r
	Above seep	25 r	0.39 ± 0.003 r	100 r	20.1 ± 0.60 r	20.1 ± 0.60 r
	Piezometer onsite	0 s	---[d] ---	0 s	0 ± 0 s	--- ---
	I-80	30 r	0.34 ± 0.084 r	100 r	19.1 ± 0.69 r	19.1 ± 0.69 r
	Spring Creek	70 t	0.31 ± 0.088 r	100 r	19.6 ± 0.73 r	19.6 ± 0.73 r
July 18, 1985	Upstream control	60 r,s	0.37 ± 0.020 r	100 r	24.9 ± 0.57 r	24.9 ± 0.57 r
	Above seep	60 r,s	0.41 ± 0.050 r,s	100 r	23.4 ± 0.69 r	23.4 ± 0.69 r
	Piezometer onsite	35 r	0.18 ± 0.023 t	100 r	26.3 ± 2.27 r	26.3 ± 2.27 r
	I-80	90 s	0.42 ± 0.004 s	90 r	20.5 ± 2.18 r	22.4 ± 1.11 r
	Spring Creek	55 r	0.40 ± 0.154 r,s,t	100 r	24.3 ± 0.67 r	24.3 ± 0.67 r
Aug. 9, 1985	Upstream control	85 r	0.56 ± 0.029 r	100 r	12.2 ± 1.60 r,s	12.2 ± 1.60 r
	Above seep	55 r	0.54 ± 0.024 r	100 r	14.8 ± 1.64 r,s	14.8 ± 1.64 r,s
	Piezometer onsite	5 s	0.35 ± 0 s	80 r	11.8 ± 1.29 r	13.6 ± 0.53 r
	I-80	65 r	0.56 ± 0.042 r	100 r	15.2 ± 1.52 r,s	15.2 ± 1.52 r,s
	Spring Creek	75 r	0.57 ± 0.028 r	100 r	18.5 ± 1.28 s	18.5 ± 1.28 s
Oct. 3, 1985[e]	Upstream control	60 r	0.74 ± 0.035 r	100 r	19.6 ± 0.50 r,s	19.6 ± 0.50 r
	Upstream piezometer	70 r	0.57 ± 0.035 r	100 r	19.6 ± 0.69 r,s	19.6 ± 0.69 r
	New channel onsite	53 r	0.64 ± 0.066 r	90 r	18.4 ± 1.83 r,s	19.9 ± 1.20 r
	New piezometer onsite	75 r	0.74 ± 0.076 r	100 r	17.8 ± 1.06 r	17.8 ± 1.06 r
	I-80	50 r	0.78 ± 0.049 r	100 r	17.6 ± 1.17 r,s	17.6 ± 1.17 r
	Spring Creek	75 r	0.54 ± 0.013 r	100 r	21.8 ± 0.92 s	21.8 ± 0.92 r

[a]The letters r, s and t denote statistical inferences among treatments; treatments within the same test on the same date that share a common letter are not significantly different from each other, using Fisher's Exact Test (survival tests) and Tukey's HSD Method (growth and reproduction tests) for all possible pairwise comparisons among treatments at $\alpha = 0.05$. For some tests, downstream sampling locations shown as significantly lower than the upstream control in Appendix Table A-1 are not shown as significantly different from the upstream control in this table because two-tailed comparisons of all possible combinations of treatment pairs (using either Fisher's Exact Test or Tukey's HSD Method) is more conservative than one-tailed comparisons of all non-control treatments only with the upstream control (using either Fisher's Exact Test or Dunnett's Method).

[b]Values expressed as mean ± one standard error of the mean.

[c]MOA = mean reproduction of all females that started the test; MIM = mean reproduction of surviving females (see Toxicity Tests in METHODS for calculation procedures).

[d]--- = value could not be calculated because all of the test animals died.

[e]Laramie River was rechanneled in September 1985 (see Site Descriptions in METHODS); October 1985 stream samples were collected at corresponding locations along the new river channel.

Table A-3. Seven-day survival and growth of fathead minnows (<u>Pimephales promelas</u>) and seven-day survival and reproduction of <u>Ceriodaphnia dubia</u> in Crow Creek water and interstitial water from June 1985 to September 1986. Statistical comparisons were made using one-tailed tests for significant decreases in survival, growth or reproduction relative to the upstream control.[a]

		Fathead minnows		Ceriodaphnia		
Date	Site[b]	Percent survival	Weight (mg)[c]	Percent survival	MOA total offspring/ female[c,d]	MIM total offspring/ female[c,d]
June 13, 1985	Upstream control: S	95	0.27 ± 0.006	90	13.6 ± 1.20	14.1 ± 1.22
	Optimist Park: S	100	0.25 ± 0.021	90	18.7 ± 1.16	19.7 ± 0.71
	Morrie Avenue: S	95	0.24 ± 0.043	0 *	18.4 ± 1.91	--- ---
	Refinery: S	100	0.23 ± 0.005	80	25.3 ± 0.68	25.6 ± 0.60
	Below NPDES: S	0 *	---[e] ---	0 *	0 ± 0 *	--- ---
July 17, 1985	Upstream control: S	85	0.74 ± 0.059	100	16.2 ± 0.61	16.2 ± 0.61
	Optimist Park: S	75	0.58 ± 0.156	90	20.8 ± 1.26	21.8 ± 0.89
	Morrie Avenue: S	90	0.63 ± 0.074	100	21.0 ± 0.67	21.0 ± 0.67
	Refinery: S	80	0.47 ± 0.009 *	100	20.7 ± 1.24	20.7 ± 1.24
	Below NPDES: S	0 *	--- ---	0 *	0 ± 0 *	--- ---
Aug. 20, 1985	Upstream control: S	55	0.52 ± 0.018	100	20.5 ± 1.10	20.5 ± 1.10
	Optimist Park: S	90	0.49 ± 0.053	100	18.6 ± 1.84	18.6 ± 1.84
	Morrie Avenue: S	100	0.49 ± 0.049	0 *	6.0 ± 0.89 *	--- ---
	Refinery: S	100	0.51 ± 0.042	0 *	5.5 ± 0.78 *	--- ---
	Below NPDES: S	25	0.21 ± 0.040 *	100	10.7 ± 1.05 *	10.7 ± 1.05 *
Oct. 24, 1985	Upstream control: S	98	0.88 ± 0.039	100	22.1 ± 0.86	22.1 ± 0.86
	Optimist Park: S	85	0.81 ± 0.025	100	20.9 ± 1.04	20.9 ± 1.04
	Morrie Avenue: S	98	0.92 ± 0.026	100	20.1 ± 1.26	20.1 ± 1.26
	Refinery: S	95	0.92 ± 0.032	100	21.3 ± 1.32	21.3 ± 1.32
	Below NPDES: S	0 *	--- ---	0 *	0 ± 0 *	--- ---
Dec. 12, 1985	Upstream control: S	78	0.80 ± 0.030	90	4.2 ± 0.76	4.7 ± 0.67
	Optimist Park: S	72	0.78 ± 0.051	100	5.8 ± 1.21	5.8 ± 1.21
	Morrie Avenue: S	70	0.84 ± 0.024	100	14.0 ± 1.46	14.0 ± 1.46
	Refinery: S	68	0.81 ± 0.032	100	12.0 ± 2.37	12.0 ± 2.37
	Below NPDES: S	15 *	0.17 ± 0.041 *	0 *	0 ± 0 *	--- ---
Feb. 24, 1986	Upstream control: S	90	0.79 ± 0.020	60	3.6 ± 1.22	5.8 ± 1.40
	Optimist Park: S	88	0.70 ± 0.030 *	70	4.1 ± 1.47	5.9 ± 1.71
	Morrie Avenue: S	90	0.74 ± 0.018	70	8.0 ± 2.11	11.1 ± 2.00
	Refinery: S	100	0.70 ± 0.010 *	80	13.0 ± 2.08	15.9 ± 1.01
	Below NPDES: S	0 *	--- ---	0 *	0 ± 0 *	--- ---
April 29, 1986	Upstream control: S	75	0.87 ± 0.089	80	19.8 ± 3.22	24.4 ± 1.28
	Optimist Park: S	100	0.75 ± 0.026	100	23.7 ± 1.20	23.7 ± 1.20
	Morrie Avenue: S	92	0.75 ± 0.064	100	24.0 ± 1.39	24.0 ± 1.39
	Refinery: S	95	0.88 ± 0.028	100	23.4 ± 1.96	23.4 ± 1.96
	Below NPDES: S	85	0.37 ± 0.042 *	90	16.9 ± 2.37	18.7 ± 1.62 *
June 24, 1986	Upstream control: S	85	0.51 ± 0.025	70	16.6 ± 4.30	22.8 ± 3.26
	Morrie Avenue: S	92	0.56 ± 0.032	100	30.0 ± 1.75	30.0 ± 1.75
	P	40 *	0.56 ± 0.102	40	9.0 ± 1.50	12.9 ± 1.02 *
	Refinery: S	78	0.62 ± 0.026	100	23.2 ± 2.94	23.2 ± 2.94
	P	70	0.50 ± 0.029	10 *	9.9 ± 1.61	14.3 ± 3.09
	Below NPDES: S	8 *	0.16 ± 0.039 *	80	7.0 ± 1.54	8.8 ± 1.26 *
	P	25 *	0.39 ± 0.112	40	2.8 ± 1.98 *	5.8 ± 3.96 *
July 5, 1986	Upstream control: S	82	0.81 ± 0.042	100	27.8 ± 1.95	27.8 ± 1.95
	Morrie Avenue: S	89	0.73 ± 0.043	80	34.4 ± 4.78	35.9 ± 4.21
	P	61	0.72 ± 0.097	70	15.4 ± 3.91	19.2 ± 3.80
	Refinery: S	89	0.79 ± 0.053	100	40.9 ± 3.19	40.9 ± 3.19
	P	71	0.72 ± 0.058	100	50.6 ± 9.33	50.6 ± 9.33
	Below NPDES: S	71	0.27 ± 0.048 *	100	23.2 ± 3.29	23.2 ± 3.29
	P	93	0.58 ± 0.043 *	40 *	10.9 ± 3.43 *	16.0 ± 4.79

Table A-3 (continued).

Date	Site[b]	Fathead minnows: Percent survival	Fathead minnows: Weight (mg)[c]	Ceriodaphnia: Percent survival	Ceriodaphnia: MOA total offspring/female[c,d]	Ceriodaphnia: MIM total offspring/female[c,d]
July 21, 1986	Upstream control: S	98	0.79 ± 0.020	100	25.7 ± 2.43	25.7 ± 2.43
	Upstream Morrie: S	92	0.74 ± 0.043	0 *	0 ± 0 *	--- ---
	P	92	0.79 ± 0.019	90	22.2 ± 2.56	24.1 ± 0.95
	Morrie Avenue: S	98	0.74 ± 0.013	0 *	5.3 ± 0.30 *	--- ---
	P	89	0.50 ± 0.064 *	90	21.2 ± 2.50	23.6 ± 0.95
	Refinery: S	92	0.69 ± 0.021	0 *	0 ± 0 *	--- ---
	P	82	0.65 ± 0.044 *	0 *	9.4 ± 1.63 *	--- ---
	Below NPDES: S	0 *	--- ---	0 *	0 ± 0 *	--- ---
	P	90	0.48 ± 0.028 *	100	23.0 ± 0.59	23.0 ± 0.59
Aug. 4, 1986	Upstream control: S	80	0.71 ± 0.023	70	16.7 ± 1.56	17.4 ± 1.29
	Upstream Morrie: S	77	0.75 ± 0.055	0 *	4.4 ± 0.50 *	--- ---
	P	49	0.65 ± 0.039	80	15.0 ± 2.96	17.2 ± 2.98
	Morrie Avenue: S	59	0.81 ± 0.009	0 *	4.5 ± 0.27 *	--- ---
	P	65	0.70 ± 0.084	90	15.8 ± 1.09	15.8 ± 1.09
	Refinery: S	82	0.79 ± 0.039	0 *	3.1 ± 0.35 *	--- ---
	P	72	0.77 ± 0.050	0 *	8.6 ± 1.45 *	--- ---
	Below NPDES: S	0 *	--- ---	0 *	0 ± 0 *	--- ---
	P	92	0.56 ± 0.016	80	0 ± 0 *	0 ± 0 *
Aug. 18, 1986	Upstream control: S	78	0.69 ± 0.040	100	24.5 ± 1.81	24.5 ± 1.81
	Upstream Morrie: S	90	0.76 ± 0.029	70	18.8 ± 3.53	23.6 ± 3.52
	P	88	0.70 ± 0.015	90	22.1 ± 2.93	24.5 ± 1.83
	Morrie Avenue: S	95	0.74 ± 0.029	60 *	20.1 ± 2.60	23.6 ± 1.84
	P	84	0.84 ± 0.113	60 *	8.7 ± 2.69 *	11.9 ± 2.96 *
	Refinery: S	90	0.78 ± 0.031	100	25.0 ± 1.62	25.0 ± 1.62
	P	92	0.82 ± 0.043	100	21.2 ± 2.96	23.6 ± 1.99
	Below NPDES: S	0 *	--- ---	0 *	0 ± 0 *	--- ---
	P	72	0.49 ± 0.049 *	100	22.0 ± 2.67	22.0 ± 2.67
Sept. 3, 1986	Upstream control: S	65	0.62 ± 0.027	100	20.7 ± 0.89	20.7 ± 0.89
	Upstream Morrie: S	85	0.60 ± 0.012	100	23.5 ± 1.71	23.5 ± 1.71
	P	78	0.53 ± 0.032	100	22.0 ± 1.48	22.0 ± 1.48
	Morrie Avenue: S	90	0.57 ± 0.007	100	22.7 ± 1.27	22.7 ± 1.27
	P	68	0.68 ± 0.131	90	8.3 ± 0.99 *	8.9 ± 0.80 *
	Refinery: S	90	0.60 ± 0.040	100	24.3 ± 1.05	24.3 ± 1.05
	P	78	0.57 ± 0.059	100	16.1 ± 1.40 *	16.1 ± 1.40 *
	Below NPDES: S	0 *	--- ---	0 *	0 ± 0 *	--- ---
	P	85	0.30 ± 0.076 *	100	11.0 ± 0.94 *	11.0 ± 0.94 *
Sept. 17, 1986	Upstream control: S	97	0.82 ± 0.015	100	24.0 ± 1.01	24.0 ± 1.01
	Upstream Morrie: S	75	0.83 ± 0.045	80	24.2 ± 2.94	27.1 ± 1.15
	P	100	0.84 ± 0.006	100	22.8 ± 0.81	22.8 ± 0.81
	Morrie Avenue: S	94	0.87 ± 0.025	80	18.9 ± 3.44	23.1 ± 1.98
	P	94	0.75 ± 0.045	80	15.3 ± 2.09 *	17.1 ± 1.38 *
	Refinery: S	97	0.84 ± 0.042	90	29.5 ± 2.29	30.6 ± 1.88
	P	92	0.82 ± 0.034	100	28.3 ± 3.11	28.3 ± 3.11
	Below NPDES: S	0 *	--- ---	0 *	0 ± 0 *	--- ---
	P	80	0.45 ± 0.016 *	100	18.3 ± 1.04 *	18.3 ± 1.04 *

[a]* = significantly less than upstream control, using Fisher's Exact Test (June, July and August 1985 FHM and all Ceriodaphnia survival tests) and Dunnett's Method (all other FHM survival tests and all growth and reproduction tests) for comparisons of all treatments with the control at $\alpha = 0.05$.

[b]S = surface water; P = interstitial water collected from mini-piezometer inserted 1 m below creek bed.

[c]Values expressed as mean ± one standard error of the mean.

[d]MOA = mean reproduction of all females that started the test; MIM = mean reproduction of surviving females (see Toxicity Tests in METHODS for calculation procedures).

[e]--- = value could not be calculated because all of the test animals died.

Table A-4. Seven-day survival and growth of fathead minnows (Pimephales promelas) and seven-day survival and reproduction of Ceriodaphnia dubia in Crow Creek water and interstitial water from June 1985 to September 1986. Statistical comparisons were made using two-tailed tests for significant differences in survival, growth or reproduction among all treatments.[a]

		Fathead minnows		Ceriodaphnia		
Date	Site[b]	Percent survival	Weight (mg)[c]	Percent survival	MOA total offspring/ female[c,d]	MIM total offspring/ female[c,d]
June 13, 1985	Upstream control: S	95 r	0.27 ± 0.006 r	90 r	13.6 ± 1.20 r	14.1 ± 1.22 r
	Optimist Park: S	100 r	0.25 ± 0.021 r	90 r	18.7 ± 1.16 s	19.7 ± 0.71 s
	Morrie Avenue: S	95 r	0.24 ± 0.043 r	0 s	18.4 ± 1.91 r,s	--- ---
	Refinery: S	100 r	0.23 ± 0.005 r	80 r	25.3 ± 0.68 t	25.6 ± 0.60 t
	Below NPDES: S	0 s	---[e] ---	0 s	0 ± 0 u	--- ---
July 17, 1985	Upstream control: S	85 r	0.74 ± 0.059 r	100 r	16.2 ± 0.61 r	16.2 ± 0.61 r
	Optimist Park: S	75 r	0.58 ± 0.156 r,s	90 r	20.8 ± 1.26 s	21.8 ± 0.89 r
	Morrie Avenue: S	90 r	0.63 ± 0.074 r,s	100 r	21.0 ± 0.67 s	21.0 ± 0.67 r
	Refinery: S	80 r	0.47 ± 0.009 s	100 r	20.7 ± 1.24 s	20.7 ± 1.24 r
	Below NPDES: S	0 s	--- ---	0 s	0 ± 0 t	--- ---
Aug. 20, 1985	Upstream control: S	55 r	0.52 ± 0.018 r	100 r	20.5 ± 1.10 r	20.5 ± 1.10 r
	Optimist Park: S	90 s	0.49 ± 0.053 r	100 r	18.6 ± 1.84 r	18.6 ± 1.84 r
	Morrie Avenue: S	100 s	0.49 ± 0.049 r	0 s	6.0 ± 0.89 s,t	--- ---
	Refinery: S	100 s	0.51 ± 0.042 r	0 s	5.5 ± 0.78 s	--- ---
	Below NPDES: S	25 r	0.21 ± 0.040 s	100 r	10.7 ± 1.05 t	10.7 ± 1.05 s
Oct. 24, 1985	Upstream control: S	98 r	0.88 ± 0.039 r	100 r	22.1 ± 0.86 r	22.1 ± 0.86 r
	Optimist Park: S	85 r	0.81 ± 0.025 r	100 r	20.9 ± 1.04 r	20.9 ± 1.04 r
	Morrie Avenue: S	98 r	0.92 ± 0.026 r	100 r	20.1 ± 1.26 r	20.1 ± 1.26 r
	Refinery: S	95 r	0.92 ± 0.032 r	100 r	21.3 ± 1.32 r	21.3 ± 1.32 r
	Below NPDES: S	0 s	--- ---	0 s	0 ± 0 s	--- ---
Dec. 12, 1985	Upstream control: S	78 r	0.80 ± 0.030 r	90 r	4.2 ± 0.76 r	4.7 ± 0.67 r
	Optimist Park: S	72 r	0.78 ± 0.051 r	100 r	5.8 ± 1.21 r,s	5.8 ± 1.21 r,s
	Morrie Avenue: S	70 r	0.84 ± 0.024 r	100 r	14.0 ± 1.46 t	14.0 ± 1.46 t
	Refinery: S	68 r	0.81 ± 0.032 r	100 r	12.0 ± 2.37 s,t	12.0 ± 2.37 s,t
	Below NPDES: S	15 s	0.17 ± 0.041 s	0 s	0 ± 0 u	--- ---
Feb. 24, 1986	Upstream control: S	90 r	0.79 ± 0.020 r	60 r	3.6 ± 1.22 r	5.8 ± 1.40 r
	Optimist Park: S	88 r	0.70 ± 0.030 s	70 r	4.1 ± 1.47 r	5.9 ± 1.71 r
	Morrie Avenue: S	90 r	0.74 ± 0.018 r,s	70 r	8.0 ± 2.11 r,s	11.1 ± 2.00 r,s
	Refinery: S	100 r	0.70 ± 0.010 s	80 r	13.0 ± 2.08 s	15.9 ± 1.01 s
	Below NPDES: S	0 s	--- ---	0 s	0 ± 0 t	--- ---
April 29, 1986	Upstream control: S	75 r	0.87 ± 0.089 r	80 r	19.8 ± 3.22 r	24.4 ± 1.28 r
	Optimist Park: S	100 r	0.75 ± 0.026 r	100 r	23.7 ± 1.20 r	23.7 ± 1.20 r
	Morrie Avenue: S	92 r	0.75 ± 0.064 r	100 r	24.0 ± 1.39 r	24.0 ± 1.39 r
	Refinery: S	95 r	0.88 ± 0.028 r	100 r	23.4 ± 1.96 r	23.4 ± 1.96 r
	Below NPDES: S	85 r	0.37 ± 0.042 s	90 r	16.9 ± 2.37 r	18.7 ± 1.62 r
June 24, 1986	Upstream control: S	85 r	0.51 ± 0.025 r,s	70 r,s	16.6 ± 4.30 r,s	22.8 ± 3.26 r,s,t,u
	Morrie Avenue: S	92 r	0.56 ± 0.032 r	100 r	30.0 ± 1.75 r	30.0 ± 1.75 r
	P	40 s	0.56 ± 0.102 r	40 s,t	9.0 ± 1.50 s	12.9 ± 1.02 s,t,v
	Refinery: S	78 r	0.62 ± 0.026 r	100 r	23.2 ± 2.94 r	23.2 ± 2.94 r,u
	P	70 r	0.50 ± 0.029 r,s	10 t	9.9 ± 1.61 s	14.3 ± 3.09 t,u,v
	Below NPDES: S	8 t	0.16 ± 0.039 s	80 r,s	7.0 ± 1.54 s	8.8 ± 1.26 v
	P	25 t	0.39 ± 0.112 r,s	40 s,t	2.8 ± 1.98 s	5.8 ± 3.96 v
July 5, 1986	Upstream control: S	82 r	0.81 ± 0.042 r	100 r	27.8 ± 1.95 r,s,t	27.8 ± 1.95 r,s,t,u,v,w
	Morrie Avenue: S	89 r	0.73 ± 0.043 r	80 r,s	34.4 ± 4.78 r,s,u	35.9 ± 4.21 s,x,u,y
	P	61 r	0.72 ± 0.097 r	70 r,s	15.4 ± 3.91 s,t	19.2 ± 3.80 t,v,w
	Refinery: S	89 r	0.79 ± 0.053 r	100 r	40.9 ± 3.19 r,u	40.9 ± 3.19 x,z
	P	71 r	0.72 ± 0.058 r	100 r	50.6 ± 9.33 u	50.6 ± 9.33 u,y,z
	Below NPDES: S	71 r	0.27 ± 0.048 s	100 r	23.2 ± 3.29 r,s,t	23.2 ± 3.29 v,w,y
	P	93 r	0.58 ± 0.043 r	40 s	10.9 ± 3.43 t	16.0 ± 4.79 w

Table A-4 (continued).

		Fathead minnows		Ceriodaphnia		
Date	Site[b]	Percent survival	Weight (mg)[c]	Percent survival	MOA total offspring/ female[c,d]	MIM total offspring/ female[c,d]
July 21, 1986	Upstream control: S	98 r	0.79 ± 0.020 r	100 r	25.7 ± 2.43 r,s	25.7 ± 2.43 r
	Upstream Morrie: S	92 r	0.74 ± 0.043 r	0 s	0 ± 0 u	--- ---
	P	92 r	0.79 ± 0.019 r	90 r	22.2 ± 2.56 s,t	24.1 ± 0.95 r
	Morrie Avenue: S	98 r	0.74 ± 0.013 r	0 s	5.3 ± 0.30 t	--- ---
	P	89 r	0.50 ± 0.064 s,t	90 r	21.2 ± 2.50 s,t	23.6 ± 0.95 r
	Refinery: S	92 r	0.69 ± 0.021 r	0 s	0 ± 0 u	--- ---
	P	82 r	0.65 ± 0.044 r,s	0 s	9.4 ± 1.63 t	--- ---
	Below NPDES: S	0 s	--- ---	0 s	0 ± 0 u	--- ---
	P	90 r	0.48 ± 0.028 t	100 r	23.0 ± 0.59 r,s	23.0 ± 0.59 r
Aug. 4, 1986	Upstream control: S	80 r,s	0.71 ± 0.023 r,s	70 r	16.7 ± 1.56 r	17.4 ± 1.29 r
	Upstream Morrie: S	77 r,s	0.75 ± 0.055 r,s	0 s	4.4 ± 0.50 s,t	--- ---
	P	49 r	0.65 ± 0.039 r,s	80 r	15.0 ± 2.96 r,u	17.2 ± 2.98 r
	Morrie Avenue: S	59 r,s	0.81 ± 0.009 r	0 s	4.5 ± 0.27 s,t	--- ---
	P	65 r,s	0.70 ± 0.084 r,s	90 r	15.8 ± 1.09 r	15.8 ± 1.09 r
	Refinery: S	82 r,s	0.79 ± 0.039 r	0 s	3.1 ± 0.35 s	--- ---
	P	72 r,s	0.77 ± 0.050 r,s	0 s	8.6 ± 1.45 t,u	--- ---
	Below NPDES: S	0 t	--- ---	0 s	0 ± 0 v	--- ---
	P	92 s	0.56 ± 0.016 s	80 r	0 ± 0 v	0 ± 0 s
Aug. 18, 1986	Upstream control: S	78 r	0.69 ± 0.040 r,s	100 r	24.5 ± 1.81 r	24.5 ± 1.81 r
	Upstream Morrie: S	90 r	0.76 ± 0.029 r	70 r	18.8 ± 3.53 r,s	23.6 ± 3.52 r,s
	P	88 r	0.70 ± 0.015 r,s	90 r	22.1 ± 2.93 r	24.5 ± 1.83 r
	Morrie Avenue: S	95 r	0.74 ± 0.029 r	60 r	20.1 ± 2.60 r,s	23.6 ± 1.84 r
	P	84 r	0.84 ± 0.113 r	60 r	8.7 ± 2.69 s	11.9 ± 2.96 s
	Refinery: S	90 r	0.78 ± 0.031 r	100 r	25.0 ± 1.62 r	25.0 ± 1.62 r
	P	92 r	0.82 ± 0.043 r	100 r	21.2 ± 2.96 r,s	23.6 ± 1.99 r
	Below NPDES: S	0 s	--- ---	0 s	0 ± 0 t	--- ---
	P	72 r	0.49 ± 0.049 s	100 r	22.0 ± 2.67 r	22.0 ± 2.67 r,s
Sept. 3, 1986	Upstream control: S	65 r	0.62 ± 0.027 r	100 r	20.7 ± 0.89 r,s	20.7 ± 0.89 r,s
	Upstream Morrie: S	85 r	0.60 ± 0.012 r	100 r	23.5 ± 1.71 r	23.5 ± 1.71 r
	P	78 r	0.53 ± 0.032 r,s	100 r	22.0 ± 1.48 r,s	22.0 ± 1.48 r,s
	Morrie Avenue: S	90 r	0.57 ± 0.007 r,s	100 r	22.7 ± 1.27 r	22.7 ± 1.27 r
	P	68 r	0.68 ± 0.131 r	90 r	8.3 ± 0.99 t,u	8.9 ± 0.80 t
	Refinery: S	90 r	0.60 ± 0.040 r	100 r	24.3 ± 1.05 r	24.3 ± 1.05 r
	P	78 r	0.57 ± 0.059 r,s	100 r	16.1 ± 1.40 s,v	16.1 ± 1.40 s,u
	Below NPDES: S	0 s	--- ---	0 s	0 ± 0 w	--- ---
	P	85 r	0.30 ± 0.076 s	100 r	11.0 ± 0.94 u,v	11.0 ± 0.94 t,u
Sept. 17, 1986	Upstream control: S	97	0.82 ± 0.015 r	100 r	24.0 ± 1.01 r	24.0 ± 1.01 r,s
	Upstream Morrie: S	75	0.83 ± 0.045 r	80 r	24.2 ± 2.94 r,s,t	27.1 ± 1.15 r,s
	P	100	0.84 ± 0.006 r	100 r	22.8 ± 0.81 r	22.8 ± 0.81 r
	Morrie Avenue: S	94	0.87 ± 0.025 r	80 r	18.9 ± 3.44 r,s,t	23.1 ± 1.98 r,s,t,u
	P	94	0.75 ± 0.045 r	80 r	15.3 ± 2.09 s,t	17.1 ± 1.38 t,u
	Refinery: S	97	0.84 ± 0.042 r	90 r	29.5 ± 2.29 r	30.6 ± 1.88 s
	P	92	0.82 ± 0.034 r	100 r	28.3 ± 3.11 r,t	28.3 ± 3.11 r,s,v
	Below NPDES: S	0 *	--- ---	0 s	0 ± 0 u	--- ---
	P	80	0.45 ± 0.016 s	100 r	18.3 ± 1.04 t	18.3 ± 1.04 u,v

[a]The letters r, s, t, u, v, w, x, y and z denote statistical inferences among treatments; treatments within the same test on the same date that share a common letter are not significantly different from each other, using Fisher's Exact Test (June, July and August 1985 FHM and all Ceriodaphnia survival tests) and Tukey's HSD Method (all other FHM survival tests and all growth and reproduction tests) for all possible pairwise comparisons among treatments at $\alpha = 0.05$. For some tests, downstream sampling locations shown as significantly lower than the upstream control in Appendix Table A-3 are not shown as significantly different from the upstream control in this table because two-tailed comparisons of all possible combinations of treatment pairs (using either Fisher's Exact Test or Tukey's HSD Method) is more conservative than one-tailed comparisons of all non-control treatments only with the upstream control (using either Fisher's Exact Test or Dunnett's Method).

[b]S = surface water; P = interstitial water collected from mini-piezometer inserted 1 m below creek bed.

[c]Values expressed as mean ± one standard error of the mean.

[d]MOA = mean reproduction of all females that started the test; MIM = mean reproduction of surviving females (see Toxicity Tests in METHODS for calculation procedures).

[e]--- = value could not be calculated because all of the test animals died.

APPENDIX B

Water Chemistry

- Laramie River: June 1985 - October 1985
- Crow Creek: June 1985 - September 1986

Table B-1. Routine water chemistry parameters in Laramie River water and interstitial (piezometer) water from June 1985 to October 1985.

Date	Site	pH	Conductivity ($\mu S/cm$)	Alkalinity (mg/L as $CaCO_3$)	Hardness (mg/L as $CaCO_3$)	Total ammonia (mg N/L)	Union-ized ammonia (mg NH_3/L)	Dissolved organic carbon (mg/L)
June 1985	Upstream control	7.8	612	106	235	< 0.10	< 0.01	11.7
	Above seep	7.7	622	104	240	< 0.10	< 0.01	10.5
	Piezometer onsite	7.8	1158	158	461	< 0.10	< 0.01	23.0
	I-80	7.6	607	103	230	< 0.10	< 0.01	10.2
	Spring Creek	7.8	647	108	250	< 0.10	< 0.01	10.0
July 1985	Upstream control	8.4	1150	156	490	---[a]	---	6.6
	Above seep	8.3	1160	156	499	---	---	6.9
	Piezometer onsite	8.1	1220	172	482	---	---	9.9
	I-80	8.3	1160	162	459	---	---	9.8
	Spring Creek	8.4	1150	157	455	---	---	32.9
August 1985	Upstream control	8.2	1036	134	396	---	---	9.5
	Above seep	8.1	1038	134	402	---	---	7.5
	Piezometer onsite	7.8	1033	139	402	---	---	9.4
	I-80	8.2	1032	134	392	---	---	7.9
	Spring Creek	8.1	1026	138	412	---	---	8.4
October 1985[b]	Upstream control	8.1	763	114	313	< 0.10	< 0.01	6.5
	Upstream piezometer	8.0	741	110	286	< 0.10	< 0.01	8.0
	New channel onsite	8.1	788	113	317	< 0.10	< 0.01	7.3
	New piezometer onsite	8.3	762	112	298	< 0.10	< 0.01	8.0
	I-80	8.0	797	110	309	< 0.10	< 0.01	8.4
	Spring Creek	8.0	798	119	305	< 0.10	< 0.01	2.1

[a]--- = value not determined.

[b]Laramie River was rechanneled in September 1985 (see Site Descriptions in METHODS); October 1985 stream samples were collected at corresponding locations along the new river channel.

Table B-2. Concentrations of major inorganic ions in Laramie River water and interstitial (piezometer) water from June 1985 to October 1985.[a]

		Cations						Anions					
Date	Site	Na^+	Ca^{2+}	Mg^{2+}	K^+	Sr^{2+}	NH_4^+	Cl^-	SO_4^{2-}	NO_3^-	F^-	HCO_3^-	CO_3^{2-}
June 1985	Upstream control	42	77	10	3.7	0.6	< 0.13	7	176	< 1	0.5	128	0.5
	Above seep	43	68	10	2.8	0.6	< 0.13	7	176	< 1	0.5	126	0.4
	Piezometer onsite	73	90	34	5.8	1.2	< 0.13	48	299	< 1	0.8	191	0.8
	I-80	38	67	12	2.8	0.5	< 0.13	7	171	< 1	0.5	125	0.3
	Spring Creek	38	74	12	2.9	0.6	< 0.13	8	185	< 1	0.5	131	0.5
July 1985	Upstream control	86	137	22	4.6	1.0	---[b]	20	508	< 1	0.8	184	3.2
	Above seep	83	134	22	4.4	1.1	---	21	503	< 1	0.8	185	2.6
	Piezometer onsite	81	136	23	5.0	1.2	---	25	510	< 1	0.8	206	1.8
	I-80	82	136	22	4.5	1.0	---	21	503	< 1	0.8	192	2.7
	Spring Creek	76	126	21	4.4	0.9	---	21	514	< 1	0.7	185	3.2
August 1985	Upstream control	73	128	20	3.9	0.9	---	19	314	< 1	0.7	160	1.7
	Above seep	67	128	20	3.9	0.8	---	19	312	< 1	0.6	161	1.4
	Piezometer onsite	75	113	22	5.0	1.0	---	21	306	< 1	0.4	168	0.7
	I-80	78	124	19	3.9	0.8	---	19	309	< 1	0.4	160	1.7
	Spring Creek	69	128	20	4.1	0.9	---	20	306	< 1	0.4	165	1.4
October 1985[c]	Upstream control	48	66	32	2.9	0.5	< 0.13	10	228	< 1	0.6	137	1.1
	Upstream piezometer	45	65	31	2.6	0.5	< 0.13	10	218	< 1	0.5	132	0.9
	New channel onsite	49	69	34	2.8	0.5	< 0.13	12	237	< 1	0.5	136	1.1
	New piezometer onsite	48	69	31	2.7	0.5	< 0.13	11	225	< 1	0.5	133	1.7
	I-80	51	67	33	2.7	0.5	< 0.13	12	237	< 1	0.5	132	0.9
	Spring Creek	47	70	38	2.8	0.6	< 0.13	11	235	< 1	0.5	143	0.9

[a]Values expressed as mg/L.

[b]--- = value not determined.

[c]Laramie River was rechanneled in September 1985 (see Site Descriptions in METHODS); October 1985 stream samples were collected at corresponding locations along the new river channel.

Table B-3. Concentrations of trace elements in Laramie River water and interstitial (piezometer) water from June 1985 to October 1985.[a]

Date	Site	Element										
		Al	As	Cd	Cr	Cu	Fe	Hg	Ni	Pb	Se	Zn
June 1985	Upstream control	< 0.1	< 0.1	0.0015	0.0041	< 0.0010	0.12	< 0.1	< 0.01	< 0.1	< 0.1	0.0075
	Above seep	< 0.1	< 0.1	0.0030	0.0030	< 0.0010	0.14	< 0.1	0.02	< 0.1	< 0.1	0.0036
	Piezometer onsite	< 0.1	< 0.1	0.0023	0.0017	< 0.0010	0.13	< 0.1	< 0.01	< 0.1	0.1	0.0124
	I-80	0.1	< 0.1	0.0019	0.0078	0.0261	0.15	< 0.1	< 0.01	< 0.1	< 0.1	0.0018
	Spring Creek	< 0.1	< 0.1	0.0014	0.0036	< 0.0010	0.12	< 0.1	< 0.01	< 0.1	< 0.1	0.0025
July 1985	Upstream control	< 0.1	< 0.1	0.0023	0.0022	< 0.0010	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.0045
	Above seep	0.1	< 0.1	0.0022	0.0028	< 0.0010	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.0035
	Piezometer onsite	0.1	< 0.1	0.0029	< 0.0010	< 0.0010	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.0132
	I-80	< 0.1	< 0.1	0.0026	0.0080	0.0023	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.0112
	Spring Creek	< 0.1	< 0.1	0.0035	0.0043	0.0016	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.0064
August 1985	Upstream control	0.1	< 0.1	0.0022	0.0028	< 0.0010	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.0139
	Above seep	< 0.1	< 0.1	0.0029	< 0.0010	< 0.0010	0.03	< 0.1	< 0.01	< 0.1	< 0.1	0.0187
	Piezometer onsite	0.1	< 0.1	0.0024	< 0.0010	< 0.0010	0.07	< 0.1	< 0.01	< 0.1	< 0.1	0.0096
	I-80	0.1	< 0.1	0.0021	0.0053	< 0.0010	0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0003
	Spring Creek	0.1	< 0.1	0.0020	< 0.0010	< 0.0010	0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0018
October 1985[b]	Upstream control	< 0.1	< 0.1	0.0007	< 0.0010	< 0.0010	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.0008
	Upstream piezometer	< 0.1	< 0.1	0.0010	< 0.0010	< 0.0010	0.02	< 0.1	0.01	< 0.1	< 0.1	0.0075
	New channel onsite	< 0.1	< 0.1	0.0008	< 0.0010	< 0.0010	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.0052
	New piezometer onsite	< 0.1	< 0.1	0.0008	0.0055	< 0.0010	0.02	< 0.1	0.01	< 0.1	< 0.1	0.0062
	I-80	< 0.1	< 0.1	0.0008	< 0.0010	< 0.0010	0.02	< 0.1	0.01	< 0.1	< 0.1	0.0130
	Spring Creek	< 0.1	< 0.1	0.0007	0.0139	< 0.0010	0.05	< 0.1	< 0.01	< 0.1	< 0.1	0.0033

[a]Values expressed as mg/L. Al, As, Fe, Hg, Ni, Pb and Se were analyzed by inductively coupled plasma emission spectroscopy (ICP); Cd, Cr, Cu and Zn were analyzed by atomic absorption spectroscopy (detection limits using ICP for these four elements were only 0.01 mg/L).

[b]Laramie River was rechanneled in September 1985 (see Site Descriptions in METHODS); October 1985 stream samples were collected at corresponding locations along the new river channel.

Table B-4. Routine water chemistry parameters in Crow Creek water and interstitial water from June 1985 to September 1986.

Date	Site[a]	pH	Conductivity (μS/cm)	Alkalinity (mg/L as $CaCO_3$)	Hardness (mg/L as $CaCO_3$)	Total ammonia (mg N/L)	Union-ized ammonia (mg NH_3/L)	Dissolved organic carbon (mg/L)
June 13, 1985	Upstream control: S	8.0	403	176	182	< 0.10	< 0.01	5.8
	Optimist Park: S	8.4	543	188	221	0.14	0.02	12.3
	Morrie Avenue: S	8.2	560	182	230	< 0.10	< 0.01	8.5
	Refinery: S	8.1	626	188	336	< 0.10	< 0.01	8.8
	Below NPDES: S	7.9	802	163	288	12.00	0.63	22.4
July 17, 1985	Upstream control: S	8.2	347	134	141	---[b]	---	22.4
	Optimist Park: S	8.4	640	180	240	---	---	6.5
	Morrie Avenue: S	8.3	671	179	249	---	---	8.3
	Refinery: S	7.9	696	182	259	---	---	8.9
	Below NPDES: S	7.7	934	158	232	---	---	15.1
Aug. 20, 1985	Upstream control: S	8.1	492	197	218	---	---	4.4
	Optimist Park: S	8.2	796	230	305	---	---	17.8
	Morrie Avenue: S	8.1	797	224	306	---	---	18.5
	Refinery: S	8.0	802	223	309	---	---	21.6
	Below NPDES: S	7.9	895	208	290	---	---	24.9
Oct. 24, 1985	Upstream control: S	8.1	539	224	258	< 0.10	< 0.01	11.2
	Optimist Park: S	8.2	746	243	329	< 0.10	< 0.01	9.4
	Morrie Avenue: S	8.1	770	226	227	< 0.10	< 0.01	11.1
	Refinery: S	7.9	807	247	188	< 0.10	< 0.01	6.9
	Below NPDES: S	8.0	906	220	243	6.90	0.45	2.4
Dec. 12, 1985	Upstream control: S	8.0	532	230	200	< 0.10	< 0.01	2.2
	Optimist Park: S	8.0	778	266	180	0.17	0.01	2.6
	Morrie Avenue: S	8.0	818	275	205	0.21	0.01	3.2
	Refinery: S	8.0	865	280	182	0.28	0.02	3.4
	Below NPDES: S	8.0	920	262	228	2.50	0.16	4.6
Feb. 24, 1986	Upstream control: S	8.1	453	197	194	< 0.10	< 0.01	2.0
	Optimist Park: S	8.2	607	224	220	< 0.10	< 0.01	2.7
	Morrie Avenue: S	8.2	643	234	194	< 0.10	< 0.01	2.9
	Refinery: S	8.2	662	228	197	< 0.10	< 0.01	3.1
	Below NPDES: S	8.6	710	224	273	4.00	0.90	4.6
April 29, 1986	Upstream control: S	8.1	497	209	95	< 0.10	< 0.01	2.6
	Optimist Park: S	8.2	618	235	103	< 0.10	< 0.01	4.2
	Morrie Avenue: S	8.1	601	224	106	< 0.10	< 0.01	4.0
	Refinery: S	8.0	675	232	110	< 0.10	< 0.01	4.5
	Below NPDES: S	8.0	790	217	118	1.10	0.07	6.6

Table B-4 (continued).

Date	Site[a]	pH	Conductivity ($\mu S/cm$)	Alkalinity (mg/L as $CaCO_3$)	Hardness (mg/L as $CaCO_3$)	Total ammonia (mg N/L)	Unionized ammonia (mg NH_3/L)	Dissolved organic carbon (mg/L)
June 24, 1986	Upstream control: S	8.1	441	202	177	< 0.10	< 0.01	5.7
	Morrie Avenue: S	8.4	627	192	220	< 0.10	< 0.01	7.2
	P	7.3	1596	380	654	< 0.10	< 0.01	7.4
	Refinery: S	7.8	732	212	262	0.28	0.01	6.8
	P	7.5	732	210	277	0.98	0.02	14.8
	Below NPDES: S	7.5	956	182	247	6.40	0.14	23.7
	P	7.3	1070	424	433	5.00	0.07	13.5
July 5, 1986	Upstream control: S	8.0	408	194	190	< 0.10	< 0.01	4.0
	Morrie Avenue: S	8.4	684	232	262	0.30	0.05	8.4
	P	7.6	1572	404	646	< 0.10	< 0.01	8.5
	Refinery: S	7.8	766	244	296	0.41	0.02	8.6
	P	7.7	742	238	277	0.48	0.02	9.0
	Below NPDES: S	7.8	964	210	266	6.20	0.26	15.1
	P	7.5	1090	438	441	4.60	0.10	15.6
July 21, 1986	Upstream control: S	8.1	413	196	182	< 0.10	< 0.01	4.6
	Upstream Morrie: S	8.2	541	184	209	0.10	0.01	7.9
	P	7.7	659	228	228	0.42	0.01	6.8
	Morrie Avenue: S	8.5	553	192	209	< 0.10	< 0.01	7.7
	P	7.4	1571	398	604	< 0.10	< 0.01	8.7
	Refinery: S	8.0	597	184	205	0.14	0.01	19.2
	P	8.0	481	168	205	0.20	0.01	8.4
	Below NPDES: S	7.8	836	174	201	6.80	0.29	9.7
	P	7.6	929	304	342	3.70	0.10	12.7
Aug. 4, 1986	Upstream control: S	7.9	423	182	179	< 0.10	< 0.01	4.6
	Upstream Morrie: S	7.8	498	146	156	0.28	0.01	10.8
	P	7.6	658	230	240	0.78	0.02	6.7
	Morrie Avenue: S	7.6	505	144	175	0.38	0.01	10.6
	P	7.5	1602	402	619	< 0.10	< 0.01	9.2
	Refinery: S	7.9	525	150	179	0.35	0.02	10.5
	P	7.9	492	148	167	0.34	0.02	7.9
	Below NPDES: S	8.0	767	162	168	8.70	0.57	10.7
	P	7.7	886	262	277	4.70	0.16	11.1
Aug. 18, 1986	Upstream control: S	8.0	372	160	167	< 0.10	< 0.01	5.2
	Upstream Morrie: S	8.1	682	214	236	< 0.10	< 0.01	9.7
	P	7.7	651	218	224	0.80	0.03	8.5
	Morrie Avenue: S	8.3	690	218	239	< 0.10	< 0.01	5.0
	P	7.5	1580	412	577	< 0.10	< 0.01	9.5
	Refinery: S	8.2	709	232	255	< 0.10	< 0.01	9.7
	P	8.0	700	218	247	0.22	0.01	8.2
	Below NPDES: S	8.2	963	200	236	8.30	0.83	16.8
	P	7.8	914	288	300	5.00	0.21	12.5

Table B-4 (continued).

Date	Site[a]	pH	Conductivity ($\mu S/cm$)	Alkalinity (mg/L as $CaCO_3$)	Hardness (mg/L as $CaCO_3$)	Total ammonia (mg N/L)	Union-ized ammonia (mg NH_3/L)	Dissolved organic carbon (mg/L)
Sept. 3, 1986	Upstream control: S	8.3	496	204	240	< 0.10	< 0.01	3.8
	Upstream Morrie: S	8.3	586	188	171	0.26	0.03	11.8
	P	8.3	620	166	232	< 0.10	< 0.01	8.2
	Morrie Avenue: S	8.2	592	172	186	0.25	0.03	12.1
	P	7.7	1604	396	357	< 0.10	< 0.01	8.5
	Refinery: S	8.1	602	170	403	0.26	0.02	12.2
	P	8.2	477	158	163	0.15	0.01	10.4
	Below NPDES: S	8.1	843	172	198	6.45	0.52	14.5
	P	7.8	960	322	304	5.40	0.22	13.5
Sept. 17, 1986	Upstream control: S	8.1	421	178	167	< 0.10	< 0.01	5.9
	Upstream Morrie: S	8.2	693	204	234	< 0.10	< 0.01	6.9
	P	8.0	708	214	228	0.59	0.04	6.4
	Morrie Avenue: S	8.4	687	204	228	< 0.10	< 0.01	6.4
	P	7.6	1602	388	581	< 0.10	< 0.01	8.5
	Refinery: S	8.3	725	208	258	< 0.10	< 0.01	7.6
	P	8.2	750	214	255	0.10	0.01	6.7
	Below NPDES: S	8.3	936	200	229	4.60	0.57	13.3
	P	7.9	1014	330	319	5.60	0.29	13.6

[a]S = surface water; P = interstitial water collected from mini-piezometer inserted 1 m below creek bed.

[b]--- = value not determined.

Table B-5. Concentrations of major inorganic ions in Crow Creek water and interstitial water from June 1985 to September 1986.[a]

		Cations						Anions					
Date	Site[b]	Na^+	Ca^{2+}	Mg^{2+}	K^+	Sr^{2+}	NH_4^+	Cl^-	SO_4^{2-}	NO_3^-	F^-	HCO_3^-	CO_3^{2-}
June 13, 1985	Upstream control	18	67	4	6	0.3	< 0.13	7	27	5.2	0.4	212	1.3
	Optimist Park	37	81	6	8	0.5	0.16	30	46	4.7	0.9	223	3.1
	Morrie Avenue	40	80	6	8	0.5	< 0.13	34	51	4.7	0.9	218	1.9
	Refinery	42	85	6	9	0.5	< 0.13	39	57	8.7	0.9	226	1.6
	Below NPDES	68	82	5	16	0.5	14.79	97	108	5.2	5.9	197	1.0
July 17, 1985	Upstream control	16	52	4	6	0.3	---[c]	9	17	1.7	1.0	161	1.3
	Optimist Park	36	81	7	10	0.5	---	37	60	3.7	0.9	213	3.3
	Morrie Avenue	40	91	8	9	0.4	---	43	64	4.2	0.9	213	2.4
	Refinery	40	115	8	9	0.5	---	45	66	7.2	0.9	220	1.1
	Below NPDES	62	107	7	19	0.4	---	94	130	5.7	7.4	191	0.6
Aug. 20, 1985	Upstream control	19	103	5	7	0.4	---	10	66	2.2	0.9	236	1.8
	Optimist Park	55	145	8	12	0.5	---	58	93	9.2	1.0	275	2.9
	Morrie Avenue	50	145	8	12	0.5	---	51	97	9.7	0.9	269	2.2
	Refinery	48	146	8	12	0.5	---	46	96	9.7	0.9	268	1.8
	Below NPDES	58	137	8	16	0.5	---	66	109	8.2	2.9	251	1.3
Oct. 24, 1985	Upstream control	16	86	14	6	0.3	< 0.13	12	38	1.0	0.7	269	2.1
	Optimist Park	36	106	20	8	0.5	< 0.13	39	88	8.5	0.9	290	3.0
	Morrie Avenue	38	98	21	9	0.5	< 0.13	43	93	12.5	0.9	271	2.2
	Refinery	40	103	22	9	0.5	< 0.13	47	95	14.0	0.9	298	1.5
	Below NPDES	54	95	20	16	0.5	8.41	48	117	10.5	4.3	265	1.7
Dec. 12, 1985	Upstream control	13	84	11	5	0.3	< 0.13	11	35	3.9	0.9	277	1.7
	Optimist Park	37	101	19	9	0.5	0.21	40	93	11.9	0.9	320	2.1
	Morrie Avenue	40	112	19	8	0.6	0.26	49	71	16.1	0.9	331	2.1
	Refinery	44	117	20	9	0.6	0.34	56	109	18.5	0.9	337	2.2
	Below NPDES	56	114	19	12	0.6	3.05	61	123	15.7	1.8	315	2.1
Feb. 24, 1985	Upstream control	12	72	10	4	0.2	< 0.13	9	34	1.1	1.0	237	1.8
	Optimist Park	26	84	14	6	0.4	< 0.13	28	67	4.9	0.9	268	2.7
	Morrie Avenue	29	88	15	7	0.4	< 0.13	34	75	7.2	0.9	280	2.5
	Refinery	30	89	15	7	0.4	< 0.13	36	79	8.2	0.9	273	2.4
	Below NPDES	36	88	15	8	0.4	4.20	48	93	7.7	1.4	260	6.6
April 29, 1986	Upstream control	15	78	11	4	0.2	< 0.13	10	29	5.0	1.0	251	1.7
	Optimist Park	38	87	17	6	0.4	< 0.13	34	50	3.2	0.9	281	2.5
	Morrie Avenue	39	86	18	6	0.4	< 0.13	38	57	4.6	0.9	269	2.1
	Refinery	41	88	18	6	0.4	< 0.13	42	61	5.1	1.0	279	1.8
	Below NPDES	63	83	17	15	0.4	1.34	97	86	4.2	3.1	261	1.7

Table B-5 (continued).

Date	Site[b]	Cations						Anions					
		Na^+	Ca^{2+}	Mg^{2+}	K^+	Sr^{2+}	NH_4^+	Cl^-	SO_4^{2-}	NO_3^-	F^-	HCO_3^-	CO_3^{2-}
June 24, 1986	Upstream control: S	17	66	10	7	0.4	< 0.13	15	25	1.4	1.2	243	1.6
	Morrie Avenue: S	31	53	10	8	0.4	< 0.13	41	80	4.0	1.1	227	3.5
	P	110	238	27	14	1.2	< 0.13	168	300	13.2	1.6	462	0.7
	Refinery: S	51	88	16	11	0.6	0.35	47	92	9.2	1.1	256	1.0
	P	46	90	15	11	0.6	1.24	47	90	0.7	1.0	255	0.5
	Below NPDES: S	82	85	15	25	0.7	8.10	100	141	5.0	10.8	221	0.5
	P	62	159	23	13	0.9	6.37	77	46	0.1	2.1	516	0.7
July 5, 1986	Upstream control: S	21	66	10	10	0.5	< 0.13	10	16	1.2	0.9	234	1.4
	Morrie Avenue: S	53	86	14	8	0.5	0.34	39	64	3.4	1.1	274	4.4
	P	122	265	32	15	1.2	< 0.13	100	282	0.6	1.5	490	1.5
	Refinery: S	60	97	17	12	0.7	0.51	41	72	6.7	1.0	295	1.2
	P	58	85	17	12	0.6	0.60	39	71	< 0.1	1.0	288	0.9
	Below NPDES: S	87	99	15	26	0.7	7.71	103	93	2.2	10.9	254	1.1
	P	67	163	24	13	0.9	5.82	54	30	< 0.1	1.6	532	1.1
July 21, 1986	Upstream control: S	19	71	10	9	0.4	< 0.13	10	16	6.0	0.9	236	1.6
	Upstream Morrie: S	33	75	12	10	0.5	0.12	26	52	4.1	0.7	220	2.1
	P	50	89	13	10	0.5	0.53	41	52	< 0.1	1.0	276	0.9
	Morrie Avenue: S	35	76	12	10	0.5	< 0.13	27	56	4.3	0.7	225	4.4
	P	117	246	30	15	1.1	< 0.13	98	279	< 0.1	1.5	483	0.9
	Refinery: S	33	69	10	10	0.5	0.17	28	55	5.8	0.7	222	1.4
	P	25	99	9	8	0.4	0.24	23	43	< 0.1	0.8	202	1.3
	Below NPDES: S	80	73	10	22	0.5	8.45	64	80	2.4	8.3	210	0.8
	P	72	120	17	13	0.7	4.66	77	51	< 0.1	3.8	369	1.0
Aug. 4, 1986	Upstream control: S	20	65	10	4	0.2	< 0.13	11	14	13.1	1.1	220	1.0
	Upstream Morrie: S	42	62	9	7	0.2	0.35	31	56	5.2	1.7	177	0.7
	P	58	87	12	6	0.2	0.98	39	46	< 0.1	1.0	279	0.7
	Morrie Avenue: S	44	64	9	7	0.2	0.48	32	57	4.4	0.7	175	0.4
	P	121	243	31	12	0.8	< 0.13	101	308	< 0.1	1.5	488	1.2
	Refinery: S	45	70	10	7	0.2	0.43	32	59	6.2	0.6	181	0.9
	P	38	64	12	5	0.2	0.42	32	114	< 0.1	0.6	179	0.9
	Below NPDES: S	71	70	10	20	0.2	10.60	57	81	4.7	5.2	195	1.2
	P	70	112	16	10	0.5	5.89	56	53	< 0.1	3.8	317	1.0
Aug. 18, 1986	Upstream control: S	21	55	10	4	0.2	< 0.13	10	18	0.7	0.9	193	1.1
	Upstream Morrie: S	49	89	14	7	0.4	< 0.13	33	63	3.0	1.0	257	2.0
	P	48	87	12	6	0.2	1.00	43	59	< 0.1	1.0	264	0.8
	Morrie Avenue: S	56	89	15	7	0.4	< 0.13	29	63	2.6	1.0	259	3.3
	P	118	245	31	11	0.8	< 0.13	99	285	< 0.1	1.5	500	1.2
	Refinery: S	53	95	15	7	0.4	< 0.13	62	91	6.0	1.0	277	2.8
	P	52	94	15	8	0.3	0.27	31	70	< 0.1	1.0	262	1.7
	Below NPDES: S	94	88	14	26	0.4	9.81	74	85	12.2	10.6	239	2.5
	P	79	111	16	11	0.4	6.22	49	48	< 0.1	5.0	348	1.4

Table B-5 (continued).

Date	Site[b]		Cations						Anions				
		Na^+	Ca^{2+}	Mg^{2+}	K^+	Sr^{2+}	NH_4^+	Cl^-	SO_4^{2-}	NO_3^-	F^-	HCO_3^-	CO_3^{2-}
Sept. 3, 1986	Upstream control: S	21	70	10	4	0.2	< 0.13	9	15	1.7	0.8	243	2.9
	Upstream Morrie: S	48	73	11	8	0.3	0.30	36	58	5.3	0.7	224	2.8
	P	47	83	12	6	0.2	< 0.13	34	43	0.2	1.0	197	2.4
	Morrie Avenue: S	45	74	11	7	0.3	0.29	37	60	5.1	0.7	205	2.1
	P	112	244	29	11	0.8	< 0.13	118	274	0.5	1.3	479	1.8
	Refinery: S	44	75	11	7	0.2	0.31	37	42	5.5	0.7	204	1.7
	P	38	58	9	5	0.3	0.18	25	37	< 0.1	0.9	189	1.8
	Below NPDES: S	83	74	10	21	0.3	7.75	86	77	0.3	6.7	206	1.7
	P	72	122	18	11	0.4	6.72	77	42	0.6	4.3	389	1.6
Sept. 17, 1986	Upstream control: S	18	66	10	4	0.2	< 0.13	10	18	2.0	0.9	214	1.6
	Upstream Morrie: S	46	90	16	7	0.4	< 0.13	37	75	4.5	0.8	244	2.2
	P	52	92	13	7	0.3	0.72	40	70	0.4	0.9	258	1.6
	Morrie Avenue: S	49	88	15	7	0.4	< 0.13	38	76	4.3	0.8	241	3.8
	P	121	229	29	10	0.9	< 0.13	120	273	0.5	1.2	470	1.4
	Refinery: S	54	95	16	7	0.4	< 0.13	41	88	5.6	0.9	247	3.2
	P	54	97	15	8	0.5	0.12	42	88	0.2	0.8	256	2.3
	Below NPDES: S	88	87	14	20	0.4	5.32	90	94	4.8	5.6	238	3.1
	P	78	120	17	12	0.5	6.90	83	48	< 0.1	4.4	398	2.1

[a]Values expressed as mg/L.

[b]S = surface water; P = interstitial water collected from mini-piezometer inserted 1 m below creek bed.

[c]--- = value not determined.

Table B-6. Concentrations of trace elements in Crow Creek water and interstitial water from June 1985 to September 1986.[a]

Date	Site[b]	Element										
		Al	As	Cd	Cr	Cu	Fe	Hg	Ni	Pb	Se	Zn
June 13, 1985	Upstream control	< 0.1	< 0.1	0.0015	0.0027	< 0.0010	0.07	< 0.1	< 0.01	< 0.1	< 0.1	0.0017
	Optimist Park	< 0.1	< 0.1	0.0015	0.0054	0.0032	0.05	< 0.1	0.02	< 0.1	< 0.1	0.0013
	Morrie Avenue	< 0.1	< 0.1	0.0013	0.0024	< 0.0010	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.0095
	Refinery	< 0.1	< 0.1	0.0016	0.0032	< 0.0010	0.05	< 0.1	< 0.01	< 0.1	< 0.1	0.0072
	Below NPDES	< 0.1	< 0.1	0.0018	0.0399	0.0011	0.10	< 0.1	< 0.01	< 0.1	< 0.1	0.0085
July 17, 1985	Upstream control	< 0.1	< 0.1	0.0010	< 0.0010	0.0417	0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0005
	Optimist Park	0.1	< 0.1	0.0013	0.0019	< 0.0010	0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0062
	Morrie Avenue	< 0.1	< 0.1	0.0015	0.0030	< 0.0010	0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0078
	Refinery	0.1	< 0.1	0.0020	0.0022	< 0.0010	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.0060
	Below NPDES	< 0.1	< 0.1	0.0019	< 0.0010	< 0.0010	0.03	< 0.1	< 0.01	< 0.1	0.1	0.0074
Aug. 20, 1985	Upstream control	0.1	< 0.1	0.0009	< 0.0010	< 0.0010	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0046
	Optimist Park	< 0.1	< 0.1	0.0021	0.0032	0.0025	0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0059
	Morrie Avenue	< 0.1	< 0.1	0.0020	0.0022	0.0016	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.0050
	Refinery	< 0.1	< 0.1	0.0014	0.0063	0.0165	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0099
	Below NPDES	< 0.1	< 0.1	0.0011	< 0.0010	0.0013	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.0051
Oct. 24, 1985	Upstream control	< 0.1	< 0.1	0.0003	0.0068	0.0025	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0008
	Optimist Park	< 0.1	< 0.1	0.0007	0.0075	0.0042	0.03	< 0.1	< 0.01	< 0.1	< 0.1	0.0034
	Morrie Avenue	< 0.1	< 0.1	0.0008	< 0.0010	0.0068	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0088
	Refinery	< 0.1	< 0.1	0.0008	< 0.0010	0.0016	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.0014
	Below NPDES	< 0.1	< 0.1	0.0009	0.0056	0.0056	0.03	< 0.1	< 0.01	< 0.1	< 0.1	0.0058
Dec. 12, 1985	Upstream control	< 0.1	< 0.1	0.0005	< 0.0010	0.0016	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0028
	Optimist Park	< 0.1	< 0.1	0.0006	< 0.0010	< 0.0010	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0005
	Morrie Avenue	< 0.1	< 0.1	0.0006	< 0.0010	0.0014	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0006
	Refinery	< 0.1	< 0.1	0.0009	< 0.0010	0.0012	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.0019
	Below NPDES	< 0.1	< 0.1	0.0010	0.0014	< 0.0010	< 0.01	< 0.1	< 0.01	0.1	< 0.1	< 0.0001
Feb. 24, 1986	Upstream control	< 0.1	< 0.1	0.0002	< 0.0010	0.0019	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	< 0.0001
	Optimist Park	< 0.1	< 0.1	0.0005	< 0.0010	0.0053	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0005
	Morrie Avenue	< 0.1	< 0.1	0.0005	< 0.0010	0.0025	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0021
	Refinery	< 0.1	< 0.1	0.0007	< 0.0010	0.0019	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.0049
	Below NPDES	< 0.1	< 0.1	0.0009	0.0031	0.0020	0.03	< 0.1	< 0.01	< 0.1	< 0.1	0.0049
April 29, 1986	Upstream control	< 0.1	< 0.1	0.0004	< 0.0010	< 0.0010	0.05	< 0.1	0.02	< 0.1	< 0.1	< 0.0001
	Optimist Park	< 0.1	< 0.1	0.0007	< 0.0010	< 0.0010	0.05	< 0.1	0.02	< 0.1	< 0.1	0.0007
	Morrie Avenue	< 0.1	< 0.1	0.0006	< 0.0010	< 0.0010	0.07	< 0.1	0.03	< 0.1	< 0.1	0.0020
	Refinery	0.1	< 0.1	0.0008	< 0.0010	< 0.0010	0.07	< 0.1	0.03	< 0.1	< 0.1	0.0067
	Below NPDES	< 0.1	< 0.1	0.0008	< 0.0010	< 0.0010	0.09	< 0.1	0.04	< 0.1	< 0.1	0.0044

Table B-6 (continued).

Date	Site[b]	Element										
		Al	As	Cd	Cr	Cu	Fe	Hg	Ni	Pb	Se	Zn
June 24, 1986	Upstream control: S	< 0.1	< 0.1	0.0001	0.0013	0.0012	0.03	< 0.1	< 0.01	< 0.1	< 0.1	< 0.01
	Morrie Avenue: S	< 0.1	< 0.1	0.0001	0.0012	0.0017	0.03	< 0.1	< 0.01	< 0.1	< 0.1	< 0.01
	P	< 0.1	< 0.1	0.0001	0.0030	0.0007	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	Refinery: S	< 0.1	< 0.1	0.0002	0.0024	0.0012	0.04	< 0.1	0.01	< 0.1	< 0.1	0.01
	P	< 0.1	< 0.1	0.0002	0.0022	0.0024	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	Below NPDES: S	0.1	< 0.1	0.0005	0.0392	0.0029	0.10	< 0.1	0.02	< 0.1	< 0.1	0.03
	P	< 0.1	0.1	0.0001	0.0029	0.0011	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.09
July 5, 1986	Upstream control: S	< 0.1	< 0.1	0.001	0.0039	0.0296	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.10
	Morrie Avenue: S	0.2	< 0.1	0.0005	0.0028	0.0081	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	< 0.01
	P	< 0.1	0.1	0.0001	0.0042	0.0013	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.09
	Refinery: S	< 0.1	< 0.1	0.0001	0.0030	0.0019	0.05	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	P	< 0.1	< 0.1	0.000	0.0026	0.0014	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	Below NPDES: S	< 0.1	< 0.1	0.000	0.0296	0.0033	0.10	< 0.1	0.03	< 0.1	< 0.1	0.01
	P	< 0.1	< 0.1	0.0001	0.0022	0.0006	0.01	0.1	0.03	< 0.1	0.1	0.01
July 21, 1986	Upstream control: S	< 0.1	< 0.1	0.000	0.0011	0.0031	0.05	< 0.1	0.03	< 0.1	< 0.1	0.01
	Upstream Morrie: S	< 0.1	< 0.1	0.000	0.0013	0.0031	0.05	< 0.1	0.03	< 0.1	< 0.1	0.01
	P	< 0.1	< 0.1	0.000	0.0019	0.0013	0.05	< 0.1	0.04	< 0.1	< 0.1	0.02
	Morrie Avenue: S	< 0.1	< 0.1	0.000	0.0019	0.0074	0.07	< 0.1	< 0.01	< 0.1	< 0.1	< 0.01
	P	0.1	0.1	0.000	0.0026	0.0013	0.05	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	Refinery: S	< 0.1	0.1	0.0001	0.0037	0.0048	0.01	0.1	< 0.01	< 0.1	0.1	< 0.01
	P	< 0.1	0.1	0.0004	0.0035	0.0052	0.10	0.1	< 0.01	< 0.1	0.1	0.44
	Below NPDES: S	< 0.1	0.1	0.000	0.0163	0.0070	0.08	0.1	< 0.01	< 0.1	0.1	0.06
	P	< 0.1	0.1	0.0002	0.0042	0.0027	0.03	0.1	< 0.01	< 0.1	0.1	0.06
Aug. 4, 1986	Upstream control: S	< 0.1	< 0.1	0.000	0.0012	0.0017	0.02	< 0.1	< 0.01	< 0.1	< 0.1	< 0.01
	Upstream Morrie: S	< 0.1	< 0.1	0.0002	0.0019	0.0024	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.03
	P	< 0.1	< 0.1	0.0001	0.0016	0.0001	0.02	< 0.1	< 0.01	< 0.1	< 0.1	< 0.01
	Morrie Avenue: S	< 0.1	< 0.1	0.0004	0.0020	0.0040	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.05
	P	< 0.1	< 0.1	0.0002	0.0026	0.0003	0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.02
	Refinery: S	< 0.1	< 0.1	0.0003	0.0020	0.0022	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.06
	P	< 0.1	< 0.1	0.0001	0.0014	0.0028	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.04
	Below NPDES: S	< 0.1	< 0.1	0.0003	0.0141	0.0017	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	P	< 0.1	< 0.1	0.0002	0.0030	0.0038	0.05	< 0.1	0.01	< 0.1	< 0.1	0.01
Aug. 18, 1986	Upstream control: S	< 0.1	< 0.1	0.000	0.0008	0.0009	0.05	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	Upstream Morrie: S	< 0.1	< 0.1	0.000	0.0020	0.0015	0.05	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	P	< 0.1	< 0.1	0.000	0.0015	0.0000	0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	Morrie Avenue: S	< 0.1	< 0.1	0.000	0.0042	0.0016	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	P	0.1	< 0.1	0.000	0.0028	0.0007	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.05
	Refinery: S	< 0.1	< 0.1	0.0001	0.0022	0.0011	0.03	< 0.1	< 0.01	< 0.1	< 0.1	0.07
	P	< 0.1	< 0.1	0.000	0.0020	0.0003	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.10
	Below NPDES: S	0.1	< 0.1	0.0004	0.0301	0.0024	0.13	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	P	< 0.1	< 0.1	0.0001	0.0037	0.0002	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.09

Table B-6 (continued).

Date	Site[b]	Element										
		Al	As	Cd	Cr	Cu	Fe	Hg	Ni	Pb	Se	Zn
Sept. 3, 1986	Upstream control: S	< 0.1	< 0.1	0.0001	0.0010	0.0029	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	Upstream Morrie: S	< 0.1	< 0.1	0.000	0.0022	0.0015	< 0.01	< 0.1	< 0.01	< 0.1	< 0.1	0.02
	P	< 0.1	< 0.1	0.000	0.0015	0.0007	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.02
	Morrie Avenue: S	< 0.1	< 0.1	0.000	0.0026	0.0011	0.22	< 0.1	< 0.01	< 0.1	< 0.1	0.08
	P	< 0.1	< 0.1	0.000	0.0037	0.0007	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.02
	Refinery: S	< 0.1	< 0.1	0.000	0.0027	0.0046	0.04	< 0.1	< 0.01	< 0.1	< 0.1	0.02
	P	< 0.1	< 0.1	0.000	0.0015	0.0008	0.02	< 0.1	< 0.01	< 0.1	< 0.1	< 0.01
	Below NPDES: S	< 0.1	< 0.1	0.000	0.0142	0.0020	0.11	< 0.1	< 0.01	< 0.1	< 0.1	< 0.01
	P	< 0.1	< 0.1	0.000	0.0036	0.0005	0.09	< 0.1	< 0.01	< 0.1	< 0.1	< 0.01
Sept. 17, 1986	Upstream control: S	< 0.1	< 0.1	0.0003	0.0012	0.0014	0.01	< 0.1	< 0.01	< 0.1	< 0.1	< 0.01
	Upstream Morrie: S	< 0.1	< 0.1	0.000	0.0009	0.0018	0.02	< 0.1	< 0.01	< 0.1	< 0.1	< 0.01
	P	< 0.1	< 0.1	0.000	0.0008	0.0018	0.02	< 0.1	< 0.01	< 0.1	< 0.1	< 0.01
	Morrie Avenue: S	< 0.1	< 0.1	0.000	0.0010	0.0021	0.05	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	P	< 0.1	< 0.1	0.0001	0.0009	0.0015	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	Refinery: S	< 0.1	< 0.1	0.0001	0.0007	0.0021	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	P	< 0.1	< 0.1	0.000	0.0003	0.0003	0.14	< 0.1	0.01	< 0.1	< 0.1	0.04
	Below NPDES: S	0.1	< 0.1	0.0006	0.0126	0.0013	0.08	< 0.1	< 0.01	< 0.1	< 0.1	0.01
	P	0.1	< 0.1	0.0001	0.0024	0.0002	0.02	< 0.1	< 0.01	< 0.1	< 0.1	0.01

[a]Values expressed as mg/L. Al, As, Fe, Hg, Ni, Pb and Se were analyzed by inductively coupled plasma emission spectroscopy (ICP); Cd, Cr and Cu were analyzed by atomic absorption spectroscopy (detection limits using ICP for these three elements were only 0.01 mg/L; Zn was analyzed by atomic absorption spectroscopy from June 1985 to April 1986 and by ICP from June to September 1986 (due to unreliable atomic absorption spectroscopy analyses during Year 2 of the study).

[b]S = surface water; P = interstitial water collected from mini-piezometer inserted 1 m below creek bed.