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ENVIRONMENTAL SCIENCES DIVISION

EVALUATION OF FISH KILLS DURING NOVEMBER 1986 AND
JULY 1987 IN UPPER EAST FORK POPLAR CREEK
NEAR THE Y-12 PLANT

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ABSTRACT

RYON, M. G., J. M. LOAR, G. R. SOUTHWORTH, A. J. STEWART, S. M. ADAMS, and L. A. KSZOS. 1990. Evaluation of fish kills during November 1986 and July 1987 in upper East Fork Poplar Creek near the Y-12 Plant. ORNL/TM-11578. Oak Ridge National Laboratory, Oak Ridge, Tennessee. 128 pp.

The Environmental Sciences Division (ESD) investigated two fish kills that occurred on November 21, 1986, and July 9, 1987, in upper East Fork Poplar Creek at the outfall of New Hope Pond (NHP) below the Oak Ridge Y-12 Plant. A 5.5-km area downstream of NHP was initially surveyed following each kill, and a 1.6-km reach was routinely surveyed for an additional 23 to 25 d. The fish kills were limited primarily to the stoneroller (*Camptostoma anomalum*); 1148 fish died in the first kill, and 747 fish, in the second. Affected fish showed extensive scale loss, fin rot, and hemorrhaging from the gills, fins, and anus. Numerous unaffected fish were observed in the survey areas during and following both kills.

Investigative procedures included sampling of water at the inlet and outfall of NHP for water quality, examination of operating procedures at the Y-12 Plant and in the biomonitoring program that may have adversely affected the fish populations, review of results of concurrent ambient toxicity tests of the inlet and outfall water of NHP, autopsy investigations of the cause of death of the stonerollers, and laboratory experimentation to evaluate potential causes. This investigation involved ESD staff, Y-12 Plant staff, and a disease specialist with the U.S. Fish and Wildlife Service and continued for several months following the kills.

The investigations revealed that the cause of death was bacterial hemorrhagic septicemia caused by *Aeromonas hydrophila*, which is a stress-mediated disease. The specific stressor responsible for the outbreak of the disease was not identified. Several possible stresses were indicated, including elevated concentrations of mercury and

chlorine, excessive electroshocking activity, and elevated levels of the pathogen. Cumulative stress due to the combination of several factors was also suggested. Elevated temperatures and overcrowding may have enhanced the spread of the epizootic but were not the primary causes. The impact on the stoneroller population below NHP was not ecologically significant.

1. BACKGROUND INFORMATION

On Friday, November 21, 1986, at 9:20 a.m., J. M. Loar of the Environmental Sciences Division (ESD) at Oak Ridge National Laboratory, received a call from J. D. Gass of the Health, Safety, Environment, and Accountability Division at the Y-12 Plant concerning dead fish in East Fork Poplar Creek (EFPC) below the outfall of New Hope Pond (NHP). An investigation of the kill was initiated immediately and continued for approximately 3 weeks. On Thursday, July 9, 1987, at approximately 12:00 p.m., M. G. Ryon observed moribund fish below NHP during routine sampling. The sampling was promptly halted, and a survey of the fish kill was initiated. A summary of the findings of these investigations is presented herein.

1.1 NOVEMBER 1986 FISH KILL

Following notification of the kill on November 21, ESD staff examined the fish below NHP outfall and conducted a reconnaissance of the upper reaches of EFPC to determine the extent of the kill. The reconnaissance involved crews walking upstream through each site collecting and, later, counting all dead and dying fish and aquatic vertebrates. A 5.5-km reach of EFPC below NHP was surveyed that morning. Observations were made at the five bridges between East Fork Poplar Creek kilometer (EFK) 18.2 (at the intersection of Jefferson Avenue and Oak Ridge Turnpike) and EFK 22.3 (behind Dean Stallings Ford), where the first dead and dying fish were observed (Fig. 1). No dead fish were found in approximately 100-m reaches of stream near the bridges downstream of EFK 22.3. Areas near these bridges were also checked on five other days (November 22, 24-26, and December 1), and only one dead fish was found (on December 1 at the upper Tulsa Avenue bridge at EFK 21.4, 0.9 km below Dean Stallings Ford).

Systematic surveys of a 1.4-km reach of EFPC between EFK 22.3 and NHP were conducted between November 21 and December 15 (Table 1). All dead or dying fish were collected by a one- or two-person crew from ESD. On November 21-23, Wayne Schacher, Anderson County Wildlife Officer of

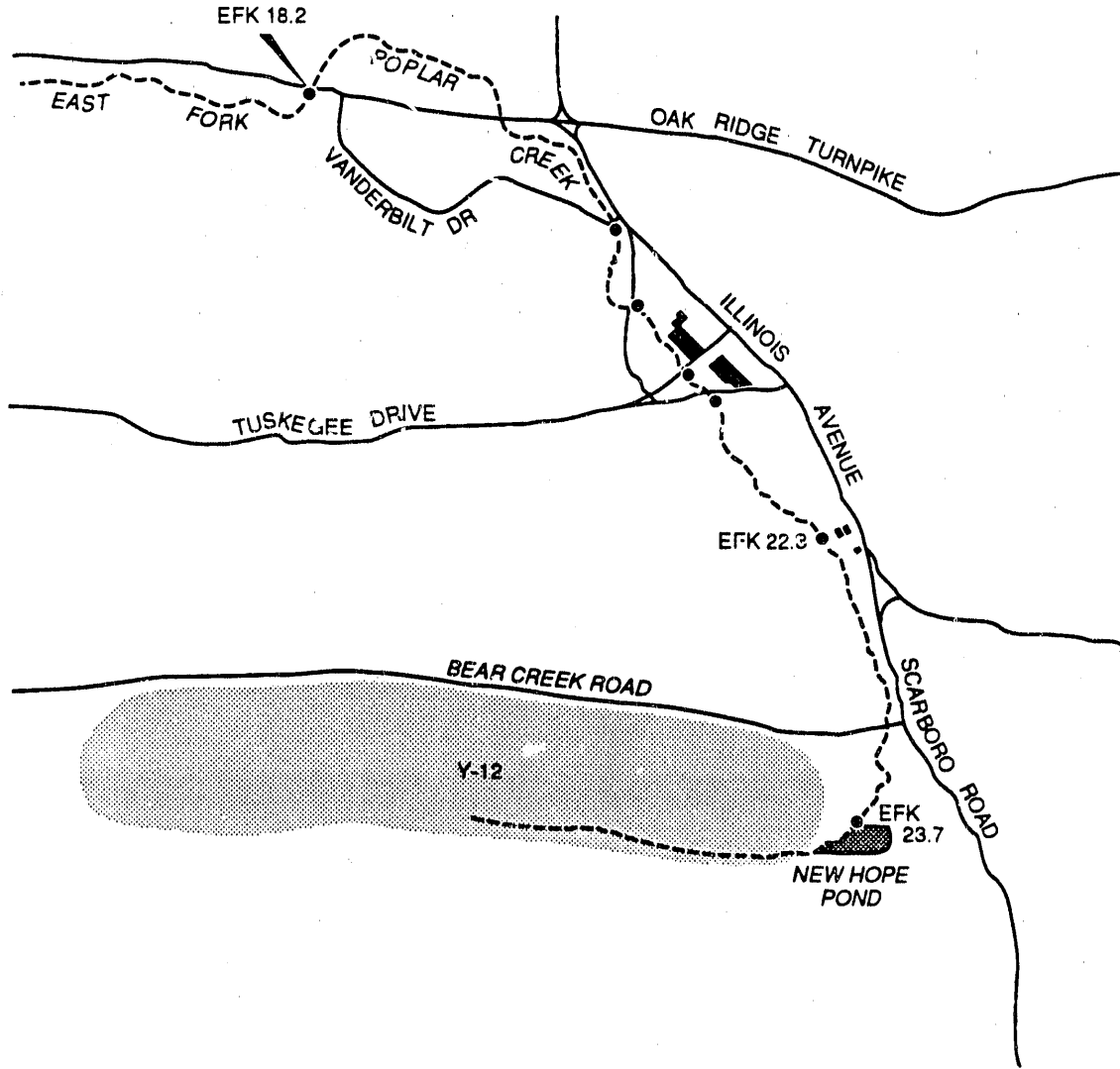


Fig. 1. Map of upper East Fork Poplar Creek indicating area of fish kills and sites surveyed. EFK = East Fork kilometer.

Table 1. Number of dead and dying fish collected over a 1.4-km reach of East Fork Poplar Creek downstream of New Hope Pond during November/December 1986

Date	No. of fish collected		Cumulative No. of Fish
	Stonerollers	Others	
11/21/86	673	1 ^a	674
11/22/86	355		1029
11/23/86	77	1 ^b	1107
11/24/86	13 ^c		1120
11/25/86	17		1137
11/26/86	0 ^c		1137
11/27/86	NS ^d		
11/28/86	NS		
11/29/86	NS		
11/30/86	NS		
12/1/86	4		1141
12/2/86	0 ^c		1141
12/3/86	2		1143
12/4/86	1		1144
12/5/86	4		1148
12/6/86	NS		
12/7/86	NS		
12/8/86	NS ^e		
12/9/86	NS ^e		
12/10/86	NS ^e		
12/11/86	NS ^e		
12/12/86	0		1148
12/15/86	0		1148

^aBlacknose dace (*Rhinichthys atratulus*).

^bStriped shiner (*Notropis chrysocephalus*).

^cActual number of dead fish was probably underestimated because of turbid water.

^dNS = no survey was conducted.

^eSurvey was not conducted because of high flows and turbidity.

the Tennessee Wildlife Resources Agency (TWRA) assisted the ESD crew in surveying. To meet the reporting requirements of the TWRA, fish were identified by species and categorized by 2-in. size classes. Except for the first day (November 21), all fish were also categorized by post-mortem changes: (1) no loss of color, indicating recent death (less than a few hours); (2) pale color, rigid body without signs of decomposition (died within past 24 h); and (3) decomposed, with only fragments of the skeleton present in worse cases (died prior to past 24 h). Fish in the latter category were added to the counts of fish in the first two categories for the preceding day to arrive at the total number killed per day as shown in Table 1.

Based on these surveys, the November 1986 fish kill can be described as follows.

1. A total of 1148 dead or dying fish were collected from EFPC, and all (99.8%), but two individuals were stonerollers, *Campostoma anomalum*,
2. Most of the fish collected were hemorrhaging (bleeding) from the gills, pectoral fins, pelvic fins, and/or anus,
3. The kill began early in the morning of November 21 and continued over a 2-week period through at least December 5; no evidence, such as decomposed remains, was found to indicate that fish died before November 21,
4. Ninety percent of the fish died on the first 2 d,
5. Numerous live fish, including stonerollers, were observed behaving normally throughout the 1.4-km reach of EFPC below NHP during the November/December survey period,
6. Dead crayfish were also found in relatively low numbers; a total of 26 was found for the 24-d period, a maximum of 13 on November 23.

1.2 JULY 1987 FISH KILL

Immediately following discovery of the July 9 kill, ESD staff conducted a survey for dead or dying fish in EFPC between NHP and Bear Creek Road and then alerted the appropriate Y-12 Plant staff and the ESD Aquatic Toxicology group. Later that day, checks at the five downstream bridges and a survey of the upper 1.4 km of EFPC were done in

the same manner as in November 1986. Systematic surveys of this upper section of EFPC were continued over the period of July 9 to July 31 (Table 2). Checks of the downstream bridges were made on 6 d (July 10-12, 16-17, and 21), and only one dead fish, a striped shiner, was found (on July 16 at the upper Tulsa Avenue bridge). Because of rainfall with associated turbidity, mortality was probably underestimated on July 12. Also, a blocknet was used at the lower end of the survey reach from July 15 through 17, which increased the number of dead fish recovered on those days.

Based on these surveys, the July 1987 fish kill can be described as follows.

1. A total of 747 dead or dying fish were collected from EFPC, and all but 18 were stonerollers (97.6%),
2. Most of the fish were hemorrhaging, showed extensive scale loss or fin rot, or were swimming erratically in a spiral motion; fewer fish showed hemorrhaging and more showed fin rot than during the November 1986 kill,
3. The kill began before its discovery on July 9, as indicated by decomposed fish, and the mortality pattern was extended over a 3-week period,
4. Forty-eight percent of the fish died during the first week; 96% died within the first 10 d,
5. Numerous live fish, including stonerollers, were observed behaving normally throughout the 1.4-km reach,
6. Low numbers of dead crayfish were also found; a total of 40 were found for the 23-d period, a maximum of 9 on July 15,
7. Predation on dead or dying fish by kingfishers (*Megaceryle alcyon*), great white herons (*Ardea herodias*), and green-back herons (*Butorides striatus*) was observed during the entire July 1987 fish kill (in contrast to the November 1986 fish kill).

1.3 SITE DESCRIPTION

The 1.4-km area in which the fish kills occurred is part of upper EFPC below NHP and east of the Y-12 Plant (Fig. 1). The headwaters of EFPC consist of springs that originate on the

Table 2. Number of dead and dying fish collected over a 1.4-km reach of East Fork Poplar Creek downstream of New Hope Pond during July 1987

Date	No. of fish collected		Cumulative No. of Fish
	Stonerollers	Others	
7/8/87 (and earlier) ^a	24		24
7/9/87	55	1 ^b	80
7/10/87	41		121
7/11/87	94		215
7/12/87	14 ^c	1 ^d	230
7/13/87	32		262
7/14/87	93	2 ^e	357
7/15/87	67	1 ^f	425
7/16/87	216	8 ^g	649
7/17/87	63	3 ^h	715
7/18/87	5		720
7/19/87	2		722
7/20/87	4		726
7/21/87	4		730
7/22/87	4	1 ⁱ	735
7/23/87	4	1 ^d	740
7/24/87	6		746
7/25/87	NS ^j		
7/26/87	NS		
7/27/87	1		747
7/28/87	NS		
7/29/87	0		747
7/30/87	NS		
7/31/87	0		747

^aFish found on July 9 that were decomposed and died prior to past 24 h.

^bRedbreast sunfish (*Lepomis auritus*).

^cWater was high and turbid after heavy rainfall.

^dBluegill sunfish (*Lepomis macrochirus*).

^eOne white sucker (*Catostomus commersoni*) and one mosquito fish (*Gambusia affinis*).

^fCreek chub (*Semotilus atromaculatus*).

^gOne creek chub, four mosquitofish, two blacknose dace (*Rhinichthys atratulus*), and one striped shiner (*Notropis chrysocephalus*) (at upper Tulsa Avenue bridge).

^hOne redbreast sunfish, one mosquitofish, and one white sucker.

ⁱWhite sucker.

^jNS = no survey made.

northwest slope of Chestnut Ridge. The stream is contained in culverts through much of the west end of the Y-12 Plant before entering a rip-rap channel that is approximately 2.4 m wide and 2.6 m high (Kasten 1986). EFPC receives effluents from numerous outlets in this 1.5-km section within the boundaries of the Y-12 Plant. Included in the discharges are cooling water and cooling-tower blowdown (64% of discharge volume), process wastewater (32%), coal-yard runoff, and storm drainage. The process wastewaters include effluents from waste-treatment facilities, photographic laboratories, fire-fighter training zones, plating operations, miscellaneous laboratories, and chemical preparation and make-up areas (Loar 1988).

At the time of the fish kills, EFPC entered NHP, a 2.2-ha retention basin designed to equalize pH of the plant effluents (Pritz and Sanders 1982). Although the pond has now been drained, filled, and capped, it was used for neutralization, sediment retention, and spill control prior to November 1988. After passing over a concrete weir at the outfall of the pond, EFPC flowed 23.7 km to its confluence with Poplar Creek.

The fish kills occurred in the 1.4 km reach of EFPC from the NHP outfall to below Bear Creek Road near Dean Stallings Ford, about 300 m downstream of the DOE Oak Ridge Reservation (ORR) boundary. The average width of the stream in this reach is 4.8 m, and the average depth is 27 cm (Loar 1988). Substrate is typically gravel and cobble interspersed with rough bedrock outcrops and occasional patches of algae or *Potamogeton* sp. The section immediately below the NHP outfall and on either side of Bear Creek Road is lined with rip-rap banks, but the remaining sections have natural clay banks, often with undercut areas. Vegetated cover above Bear Creek Road is limited to meadow grasses and large deciduous trees (canopy cover of 0 to 90%). Below the road, a more consistently enclosed canopy exists with heavily wooded banks. No obvious ecological impacts on EFPC from commercial development are evident in the reach below the ORR boundary.

2. INVESTIGATIVE STUDIES

2.1 NOVEMBER 1986 FISH KILL

Intensive sampling of water quality at the outfall of NHP and downstream was initiated by Y-12 Plant staff immediately after notification of the November fish kill (Table 3). The preliminary data were reviewed by representatives of the U.S. Department of Energy (DOE), ESD, and the Y-12 Plant on November 24. Elevated levels of total mercury (Hg) of 30 to 40 ppb were recorded on November 19, 20, and 21 by the on-line mercury analyzer at the outfall of NHP; normal concentrations range from 1 to 3 ppb. Levels of Hg in the November 21 grab samples had declined to a more typical range (2 to 9 ppb, Table 1). Some of the peaks coincided with the removal of a downstream plug in the storm-sewer line immediately south of Building 9201-4. This long, mercury-contaminated line was being cleaned as part of the remedial action program to reduce mercury in plant effluents. Removal of the plug caused the release of approximately 100 to 200 L of water. Cleaning operations were terminated on November 21 but resumed again on December 1, following a second meeting held that morning between staff from DOE, ESD, and the Y-12 Plant. Criteria that were considered in the decision to resume the cleaning operation included (1) knowledge of the pathogenic cause of fish death, (2) return of water quality at the outfall of NHP to prekill conditions, (3) information indicating that either the kill was over or near termination, and (4) low turbidity in EFPC to permit monitoring of the fish populations in the stream after cleaning operations resumed.

To investigate the hypothesis that mercury releases at the Y-12 Plant were associated with the fish kill, laboratory toxicity tests were conducted on November 25 on stonerollers collected from an uncontaminated region of White Oak Creek above ORNL (Appendix A). Sludge removed from the contaminated sewer line and deposited in the northwest sludge basin was added to several aerated aquaria in the Aquatic Ecology Laboratory of ESD at approximate concentrations of 30, 15, and 0.5 g of sludge per liter. A control aquarium (without any type of sediment added) had a turbidity equivalent to that in the

Table 3. Concentrations of ICAP^a metals in one grab sample per site collected the morning of November 21, 1986, at the outfall of New Hope Pond (NHP) and four sites downstream in the City of Oak Ridge

Parameter (mg/L)	Sampling site ^b			
	EFK ^c 23.7 ^d	EFK 22.6	EFK 21.3	EFK 16.5
Ag	0.02	0.02	0.02	0.02
Al	0.22	0.20	0.17	0.23
As	0.2	0.2	0.2	0.2
B	0.04	0.04	0.04	0.04
Ba	0.053	0.055	0.054	0.055
Be	0.0005	0.0005	0.0005	0.0005
Ca	53.6	54.9	55.8	62.8
Cd	0.02	0.02	0.02	0.02
Ce	0.08	0.08	0.08	0.08
Co	0.01	0.01	0.01	0.01
Cr	0.03	0.03	0.03	0.03
Cu	0.01	0.01	0.01	0.01
Fe	0.6	0.5	0.3	0.2
Ga	0.05	0.05	0.05	0.05
Hg	0.0090	0.0065	0.0032	0.0016
K	4.0	4.0	4.0	4.0
La	0.01	0.01	0.01	0.01
Li	0.022	0.022	0.023	0.024
Mg	13.5	13.7	13.6	13.6
Mn	0.095	0.077	0.056	0.048
Mo	0.12	0.12	0.11	0.06
Na	35.6	34.7	31.8	28.2
Nb	0.05	0.05	0.05	0.05
Ni	0.04	0.04	0.04	0.04

Table 3. (continued)

Parameter (mg/L)	Sampling site ^b			
	EFK ^c 23.7 ^d	EFK 22.6	EFK 21.3	EFK 16.5
P	0.4	0.4	0.3	0.3
Pb	0.1	0.1	0.1	0.1
Se	0.022	0.0002	0.002	0.002
Sr	0.157	0.159	0.163	0.165
Th	0.05	0.05	0.05	0.05
Ti	0.01	0.01	0.01	0.01
U	0.028	0.023	0.015	0.085
V	0.02	0.02	0.02	0.02
Zn	0.092	0.071	0.046	0.022
Zr	0.01	0.01	0.01	0.01
CN ^e	0.003	0.006	0.003	0.002
TOC ^f	5.0	5.2	4.8	5.2
			EFK 18.0	EFK 16.5

^aICAP - inductively-coupled plasma spectrophotometry.

^bEFK 23.7 is NHP outfall; EFK 22.6 is behind dentist office; EFK 21.3 is behind K-Mart;

EFK 18.2 is across from Weigel's; and EFK 16.5 is behind Fire Hall No. 1.

^cEFK - East Fork kilometer, with EFK 0.0 located at the confluence of East Fork Poplar Creek and Poplar Creek.

^dSample collected at 0955 h.

^eCN - cyanide radical.

^fTOC - total organic carbon.

low-concentration aquarium (23 NTU). No measurements were made of actual Hg concentrations in the aquaria. All 10 fish at each of the two higher levels died within 67 h; 3 of 10 died at the lowest level after 111 h. Although mortality was related to concentration, the fish probably died from the high turbidity (approximately 200 NTU) at the two higher concentrations. Fish dying in this test did not exhibit the same symptoms (e.g., hemorrhaging) observed in fish dying in EFPC, and observations of the gills of the fish that died in the high turbidity aquaria showed higher than normal secretions of mucus, a general indicator of gill irritation. Although the test results did not support the hypothesis that mercury-containing sludge discharged during the storm-sewer cleaning operations caused the fish kill, the results must be regarded as inconclusive because of the mortality observed in the test.

As part of the Y-12 Plant Biological Monitoring and Abatement Program (BMAP) for EFPC, as specified in Part III(C): Special Condition No. 7 of the National Pollutant Discharge Elimination System (NPDES) permit for the Y-12 Plant, ambient toxicity testing is routinely conducted at the outfall of NHP. Water from the outfall of NHP is tested for toxicity with a microcrustacean (*Ceriodaphnia dubia/affinis*) and fathead minnow larvae (*Pimephales promelas*). Each test lasts for 7 d and uses water samples composited daily over the test period. Portions of each daily sample of water are also analyzed for pH, alkalinity, hardness, conductivity, and chlorine.

Water from the outfall of NHP was evaluated on a routine basis for toxicity once in October, twice in November, and once in December of 1986. The second test in November (November 18-24), spanned the period of the fish kill; the first (November 4-11) preceded the kill by 10 d. The December 3-10 test followed the first reported kill by 12 d. Water quality data acquired in conjunction with the toxicity tests and results of toxicity tests based on survival and growth of fathead minnow larvae are shown in Table 4. In the two November tests and one December test, survival of fathead minnow larvae reared in water from NHP was significantly ($p < 0.05$) reduced to 56.0%, 78.0%, and 72.0%,

Table 4. Water quality parameters and toxicity of water from the outfall of New Hope Pond for various 7-d consecutive periods in October, November, and December 1986. (Survival of minnow larvae tested over each 7-d period is given as a daily mean from five replicates; mean values for two summer 7-d periods are given for comparison)

Date	pH	Water quality parameter ^a					Mean survival (%)	Growth (mg/larvae) ^c
		Conductivity (μ mho/cm)	Alkalinity (mg as CaCO ₃)	Hardness (mg as CaCO ₃)	Total (mg/L)	Flow ^d (L/s)		
10/07/86	8.29	501	123.0	164.0	0.00	98.0		
10/08/86	8.66	519	119.0	166.0	0.00	96.0		
10/09/86	8.25	389	124.5	178.0	0.00	94.0	0.505 ± 0.053	
10/10/86	8.03	395	115.0	163.0	0.00	94.0	[97.8%]	
10/11/86	8.21	316	113.5	170.0	0.00	92.0	(0.516 ± 0.044)	
10/12/86	8.17	569	111.0	176.0	0.00	86.0		
10/13/86	8.26	368	95.0	156.0	0.00	82.0		
11/04/86	8.06	459	118.0	156.0	0.00	98.0		
11/05/86	7.95	498	110.0	174.0	0.00	98.0		
11/06/86	8.13	607	118.0	190.0	0.00	94.0	0.324 ± 0.078 ^d	
11/07/86	8.30	485	120.0	176.0	0.00	94.0	[68.9%]	
11/08/86	8.15	398	105.0	152.0	0.00	84.0	(0.477 ± 0.028)	
11/09/86	8.21	543	116.0	178.0	0.00	76.0		
11/10/86	8.16	413	98.0	154.0	0.00	56.0 ^d		
11/18/86	8.13	560	109.0	160.0	0.00	98.0	0.287 ± 0.072 ^d	
11/19/86	8.13	547	110.0	148.0	0.00	98.0	[70.5%]	
11/20/86	8.15	451	114.0	164.0	0.00	98.0	(0.407 ± 0.021)	
11/21/86	7.99	468	102.0	172.0	0.00	88.0		
11/22/86	8.19	439	111.0	186.0	0.00	86.0		
11/23/86	8.21	419	111.0	170.0	0.00	78.0		
11/24/86	8.04	300	84.0	143.0	0.00	78.0 ^d		
12/03/86	7.98	355	108.0	158.0	0.00	100.0		
12/04/86	8.05	312	106.0	170.0	0.04	100.0		
12/05/86	8.10	371	104.0	170.0	0.04	90.0	0.397 ± 0.100 ^d	
12/06/86	8.19	413	106.0	180.0	0.02	86.0	[64.1%]	
12/07/86	8.24	320	106.0	162.0	0.02	80.0	(0.619 ± 0.156)	
12/08/86	8.08	408	101.0	170.0	0.04	76.0		
12/09/86	7.88	276	70.0	108.0	0.00	1168		
12/10/86	7.99	382	105.0	164.0	0.00	262		
08/7-14/86 ^e	8.43	336	110.5	166.3	0.00	98.0	[94.7%]	
06/26-7/4/86 ^e	8.23	371	115.0	174.0	0.00	98.0	[82.2%]	

^aAll analyses were for samples composited over 24-h periods except on 12/10/86, when a grab sample was used.

^bIn excess of the base flow of 431 L/s to indicate possible high-flow conditions.

^cMean growth over the 7-d test, ± 1 SD. Values in parentheses are controls; growth in NHP water is given as percentage of controls

(in brackets).

^dEnd of test values significantly lower ($p < 0.05$) than controls.

^eValues are means for the indicated 7-d periods.

respectively; survival of the larvae in control water did not fall below 96.0% in any of these tests. In the same three tests, growth of the larvae in water from NHP was also significantly reduced to 68.9%, 70.5%, and 64.1%, respectively, of growth of larvae in controls. In the test conducted in October and in tests during the summer of 1986 (Table 4), water from the pond did not show evidence of toxicity to fathead minnow larvae. In the same test periods, *Ceriodaphnia* reared in water from the outfall of NHP typically did about as well as, or in some cases somewhat better than, *Ceriodaphnia* reared in control (reconstituted hard) water (Table 5). The larval fish toxicity tests indicated the presence of an unknown toxic agent(s) in EFPC water during the time of the November fish kill.

Mercuric salts exhibit acute toxicity to freshwater fish in the 15- to 500-ppb (as Hg) range and to *Daphnia magna* at 5 to 13 ppb (Cushman et al. 1977). The lower limits of these ranges approximate levels of total Hg present in the NHP discharge during the week of November 17. Toxicity tests are performed using dissolved mercuric salts; however, Hg measurements made on NHP discharge water represent the sum of dissolved and particle-associated Hg. The latter form is relatively inert with respect to acute toxicity. Thus, assessing the toxicological significance of Hg in these waters requires reasonable estimates of the distribution of Hg between dissolved and particulate phases.

Studies of the phase distribution of Hg indicate that 10 to 25% of the total Hg in NHP discharge was in the dissolved form under summer temperature and flow conditions and at normal (1 to 3 ppb) Hg concentrations (Elwood et al. 1987). Abnormally high Hg levels at the outfall of NHP could result from (1) disturbance of mercury-contaminated sediments, which would increase the fraction of Hg that is particle-associated or (2) mobilization of dissolved Hg by oxidants, etc., which would result in a higher-than-normal fraction of the Hg being in the dissolved state. A conservative assumption is that 50% of the Hg is in the dissolved form. Using this assumption, total Hg levels in excess of 25 ppb in the NHP discharge would be at levels shown to be

Table 5. Reproduction of *Ceriodaphnia* in water from the outfall of New Hope Pond and in control (reconstituted hard) water in 7-d static-renewal tests conducted in June-July, August, October, and November 1986 (Values are the mean number of offspring (± 1 SD) per female surviving the full 7-d test period)

Test period	Offspring per female	Survival (%)	No. of males excluded ^a
06/27-07/04/86			
Control ^b	15.8 \pm 3.6	90	0
NHP water ^b	31.3 \pm 10.4	90	1
08/07-14/86			
Control ^b	13.2 \pm 3.3	100	0
NHP water ^b	14.8 \pm 2.6	100	0
10/07-13/86			
Control ^c	24.7 \pm 3.6	90	0
Control ^b	22.2 \pm 9.3	100	1
NHP water ^c	22.6 \pm 4.1	100	1
NHP water ^b	27.2 \pm 7.8	100	1
NHP water ^{b,d}	28.4 \pm 10.1	100	1
11/04-11/86			
Control ^c	16.0 \pm 1.8	100	1
NHP water ^c	16.2 \pm 5.1	100	1
11/18-24/86			
Control ^c	6.0 \pm 6.1	90	0
Control ^b	19.6 \pm 6.0	90	0
NHP water ^c	12.0 \pm 5.6	80	0
12/03-10/86	No test done; the cultures did not produce enough neonates to use as test animals.		

^aMale animals were included in computations of survival but were excluded from computations of fecundity.

^bAnimals fed digested trout-chow.

^cAnimals fed digested, flake-based food.

^dWater filtered through 0.5 μ m pore-size glass-fiber filters before being tested.

acutely toxic to channel catfish (*Ictalurus punctatus*), a micro-crustacean (*Daphnia magna*), and the European dace (*Phoxinus phoxinus*) (Cushman et al. 1977). However, this level would be more than a factor of 10 below Hg concentrations acutely toxic to other species, such as rainbow trout (*Oncorhynchus mykiss*).

In addition to monitoring water quality, conducting toxicity tests, and evaluating the toxicological significance of elevated mercury levels in the outfall of NHP, an investigation of other possible causes of death was initiated on November 24. Charles Carlson, a fish disease specialist with the U.S. Fish and Wildlife Service (USFWS), was contacted by S. M. Adams for assistance with the investigation. Mr. Carlson visited ORNL on November 26. He examined dead fish collected and frozen on November 21 and 25 and live fish collected on November 26. After the fish were examined for internal and external parasites, bacterial samples from the kidneys were obtained for culture and returned to the USFWS laboratory near Asheville, North Carolina. After the cultures had incubated for several days, Mr. Carlson called S. M. Adams with the results on Monday, December 1, prior to the meeting held that day; written confirmation of these results was obtained several days later (Appendix B). From these studies and other investigations (e.g., Pippy and Hare 1969; Wedemeyer et al. 1976), as well as individual conversations with Mr. Carlson by S. M. Adams and by J. M. Loar on December 1, the cause of death can be summarized as follows.

1. The direct cause of death was a bacterial hemorrhagic septicemia (BHS), also called motile aeromonad septicemia, an infectious bacterial disease often encountered in intensive fish culture systems,
2. The disease-causing organism (or pathogen) was *Aeromonas hydrophila*, which is normally present in most waters,
3. BHS is a stress-mediated disease [i.e., the host (fish) must interact with the pathogen in a stressful environment, which lowers the resistance of the fish to disease],
4. Environmental factors that promote epizootics of aeromonad/pseudomonad hemorrhagic septicemia include

(a) pre-existing protozoan infections, (b) inadequate pond cleaning leading to increased bacterial loads in water, (c) increased particulate matter in water, (d) handling, (e) crowding, (f) low oxygen, and (g) chronic sublethal exposure to a variety of contaminants, including heavy metals, pesticides, and PCB (Wedemeyer and McLeay 1981),

5. No evidence of an association between the dead crayfish and BHS was collected.

2.2 JULY 1987 FISH KILL

Following discovery of the July 1987 fish kill, ESD and Y-12 staff members were consulted concerning additional water quality sampling. Because water quality at the NHP inlet and outlet was already being monitored by both groups and the additional sampling of lower EFPC in November 1986 failed to yield significant new information (Table 3), additional water quality sampling was not conducted. However, as in November 1986, releases of elevated levels of mercury were monitored and were traced to cleaning of mercury-contaminated sewer lines. Because of these releases, mercury levels were also monitored more extensively in fish below NHP.

A review of mercury toxicity information published after the November 1986 fish kill (Etnier et al. 1987) confirmed the acute toxicity ranges of mercuric salts given in Sect. 2.1. No information was found on mercury toxicity to stonerollers or other fish found in EFPC.

Fortunately, ambient toxicity testing using the microcrustacean *Ceriodaphnia dubia/affinis* and the fathead minnow *Pimephales promelas* was conducted on NHP outfall water at the time of the fish kill. Using the methods described in Sect. 2.1, water from the inlet and outfall of NHP was tested at least once a month from January through July 1987 (Table 6). The results indicated that *Ceriodaphnia* and/or fathead minnow larvae occasionally showed effects of toxicity but only during the week of July 9-16 did both demonstrate effects from the inlet and/or outfall samples. In fact, more-severe effects were noted at the outfall, suggesting additional toxic inputs from NHP. These effects

Table 6. Survival (%) and reproduction (offspring per surviving female) of *Ceriodaphnia* and survival and growth (mg dry weight per fish) of fathead minnow larvae reared for 7-d periods in water from the inlet and outfall of New Hope Pond (NHP)

Sample	Test date	<i>Ceriodaphnia</i>		Fathead minnow		
		Concentration (%)	Survival (%)	Reproduction (mean \pm s.d.)	Survival (%)	Growth (mean \pm SD)
Inlet ^a	1/9-16/87	100	0		5.0	0.085 \pm 0.170
		90	0		22.5	0.082 \pm 0.163
		100	80	20.00 \pm 7.21	90.0	0.339 \pm 0.056
Outfall ^b	1/9-16/87	80	100	24.50 \pm 3.34	97.5	0.340 \pm 0.098
		100	100	26.80 \pm 2.82	98.0	0.422 \pm 0.059
Control ^c	1/9-16/87					
Outfall	2/5-12/87	100	0		90.0	0.360 \pm 0.050
		80	60	19.50 \pm 5.21	97.5	0.400 \pm 0.020
Control	2/5-12/87		90	19.00 \pm 5.81	92.5	0.480 \pm 0.030
Inlet	3/5-12/87	100	0		92.5	0.334 \pm 0.033
		80	0		90.0	0.330 \pm 0.062
Outfall	3/5-12/87	100	20	2.00 \pm 2.83	97.5	0.302 \pm 0.035
		80	100	19.70 \pm 3.06	87.5	0.299 \pm 0.039
Control	3/5-12/87		100	4.70 \pm 4.27	100.0	0.277 \pm 0.019
Outfall	4/9-16/87	100	60	18.83 \pm 4.07	82.5	0.399 \pm 0.042
		80	80	19.13 \pm 4.61	95.0	0.502 \pm 0.026
Control	4/9-16/87		70	17.71 \pm 2.56	95.0	0.551 \pm 0.035
Inlet	5/7-14/87	100	100	18.80 \pm 6.29	82.5	0.308 \pm 0.068
		80	100	21.20 \pm 3.05	97.5	0.387 \pm 0.078
Outfall	5/7-14/87	100	100	24.30 \pm 5.44	90.0	0.391 \pm 0.064
		80	100	24.20 \pm 7.87	90.0	0.396 \pm 0.025
Control	5/7-14/87		90	22.44 \pm 2.79	97.5	0.327 \pm 0.069

Table 6. (continued)

Sample	Test date	Concentration (%)	Ceriodaphnia		Fathead minnow	
			Survival (%)	Reproduction (mean \pm s.d.)	Survival (%)	Growth (mean \pm SD)
Outfall	5/21-8/87	100	100	24.20 \pm 4.94	97.5	0.247 \pm 0.082
Control	5/21-8/87	80	90	24.00 \pm 7.31	82.5	0.387 \pm 0.095
			100	22.70 \pm 4.47	90.0	0.329 \pm 0.040
Outfall	6/11-8/87	100	90	24.33 \pm 3.12	77.5	0.254 \pm 0.027
Control	6/11-8/87	80	90	21.78 \pm 6.72	77.5	0.258 \pm 0.029
			90	22.56 \pm 5.94	87.5	0.212 \pm 0.026
Inlet	7/9-16/87	100	0		65.0	0.226 \pm 0.021
		80	50	5.25 \pm 2.87	75.0	0.262 \pm 0.036
Outfall	7/9-16/87	100	0		30.0	0.194 \pm 0.032
Control	7/9-16/87	80	0		72.5	0.276 \pm 0.054
			60	11.50 \pm 9.99	97.5	0.566 \pm 0.051

^aInlet - East Fork Poplar Creek just upstream from NHP.

^bOutfall - outfall of NHP.

^cControl - hard reconstituted water.

included decreased survival of *Ceriodaphnia* and fathead minnow larvae, decreased reproduction of *Ceriodaphnia*, and reduced growth of fathead minnow larvae. All significant effects were noted at 100% and 80% concentrations, with a corresponding decrease in the magnitude of the effect with decreasing concentration.

A review of water quality data collected concurrently with the July 9-16 toxicity test indicated that concentrations of residual chlorine were slightly elevated but decreased after passage through NHP (Table 7). Water quality data collected during the toxicity tests in 1987 are given in Appendix C.

In addition to these investigations, kidney smears were taken from dead or dying fish to confirm the presence of the bacteria *Aeromonas hydrophila*. These smears were sent to Charles Carlson of the USFWS, who confirmed that the bacteria were indeed present. Thus, the cause of death in the July 1987 kill. The same as that in the November 1986 fish kill. The summary given in Sect. 2.1 accurately describes both incidents.

Table 7. Comparison of 24-hr composite vs grab samples taken from the inlet and outfall of New Hope Pond (NHP) during July 9-15, 1987

Day	Sample type	pH	Conductivity (μ S/cm)	Alkalinity (mg/L as CaCO ₃)	Hardness (mg/L as CaCO ₃)	Total residual Cl (mg/L)
			Inlet ^a : composite samples ^b			
09	Composite	8.04	381	116.0	180	0.12
10	Composite	8.01	319	115.0	190	0.07
11	Composite	8.30	345	105.0	176	0.02
12	Grab	8.06	371	110.0	168	0.18
13	Grab	8.05	319	113.0	176	0.19
14	Grab	8.14	356	117.0	184	0.12
15	Composite	8.14	307	110.0	162	0.13
	Mean ^d	8.12	338	111.5	177.0	0.09
	SD	0.13	33	5.1	11.6	0.05
			Inlet ^a : grab samples			
09	Grab	8.03	373	119.0	176	0.51
10	Grab	8.03	355	115.0	168	0.18
11	Grab	8.03	371	119.0	172	0.03
12	Grab	8.06	371	110.0	168	0.18
13	Grab	8.02	375	113.0	170	0.13
14	Grab	8.13	354	115.0	200	0.12
15	Grab	8.19	340	121.0	170	0.13
	Mean	8.06	363	116.0	174.8	0.19
	SD	0.07	13	3.9	11.4	0.15

Table 7. (continued)

Day	Sample type	pH	Conductivity ($\mu\text{S}/\text{cm}$)	Alkalinity (mg/L as CaCO_3)	Hardness (mg/L as CaCO_3)	Total residual Cl (mg/L)
			Outfall ^b : composite samples			
09	Composite	8.15	393	118.0	178	0.00
10	Composite	8.13	370	122.0	172	0.00
11	Composite	8.09	377	115.0	180	0.08
12	Composite	8.05	338	98.0	167	0.00
13	Composite	8.18	394	115.0	174	0.00
14	Composite	8.23	375	116.0	196	0.01
15	Composite	8.30	332	115.0	166	0.00
	Mean	8.16	368	114.1	176.1	0.01
	SD	0.08	25	7.6	10.2	0.03
			Outfall ^b : grab samples			
09	Grab	7.96	375	120.0	174	0.00
10	Grab	7.93	368	120.0	172	0.00
11	Grab	8.09	361	116.0	168	0.20
12	Grab	7.99	383	111.0	172	0.02
13	Grab	8.04	384	117.0	174	0.03
14	Grab	8.02	361	116.0	184	0.02
15	Grab	8.06	384	114.0	178	0.04
	Mean	8.01	374	116.3	174.6	0.04
	SD	0.06	11	3.2	5.1	0.07

^aInlet = East Fork Poplar Creek just upstream from NHP.

^bBecause of a compositor malfunction at the inlet site, composite samples collected at this site on July 12-14 were actually grab samples.

^cOutfall = the outfall of NHP.

3. EVALUATION OF POTENTIAL CAUSES OF THE FISH KILL (EPIZOOTIC)

In developing hypotheses about the cause of the epizootic, several potential sources of stress were identified and evaluated, including (1) overcrowding, (2) high temperatures and rapid changes in temperature, (3) electroshocking, (4) exposure to elevated levels of pollutants, especially Hg, and (5) a combination of stresses. The role of the pathogen and the environmental conditions existing in NHP in initiating the epizootic was also considered. Each of these hypotheses is discussed in the following.

3.1 OVERCROWDING

The high population density required for intensive fish culture is a primary factor responsible for disease outbreaks, or epizootics (Wedemeyer et al. 1976). Stoneroller densities below NHP increased by a factor of 10 between March 1986 and March 1987 (Table 8); observable increases in the population were first noted in this region of EFPC in July 1986 (Ryon 1986). The small population size may account, in part, for the absence of BHS in 1985. Although the densities of most species also increased over this same period in this region of EFPC, only the striped shiner (*Notropis chrysocephalus*) had a density similar to that of the stoneroller, and it was essentially unaffected by BHS. Such an occurrence may not be unusual. The common shiner (*Notropis cornutus*), a close relative of the striped shiner, was apparently unaffected by an epizootic of *Aeromonas liquefaciens* (taxonomically the same as *A. hydrophila*, Wolke 1975) in the Miramichi River in New Brunswick (Pippy and Hare 1969).

The effect of density on mortality rate of stonerollers was tested experimentally in the laboratory. The experimental design consisted of six 20-L aquaria; four tanks received no bacteria and two tanks received a known concentration of viable *Aeromonas hydrophilia*. Within each of these two groups, stonerollers from an uncontaminated reference stream were stocked at densities of 10 and 20 fish per tank. Thus, the four treatment groups were: (1) no bacteria, low-fish density; (2) no bacteria, high-fish density; (3) bacteria and low-fish density; and (4) bacteria and high-fish density.

Table 8. Estimated size of the stoneroller population (N_S) and the total fish population (N_T) in a 116-m reach of East Fork Poplar Creek. The study site is located approximately 200 m below the outfall of New Hope Pond. Population estimates are based on the three-pass removal method (Carle and Strub 1978)

Sampling date	Stoneroller population (N_S)	Fish population (N_T)	Percentage stonerollers (N_S/N_T)
May 14, 1985	0	51	0
October 22, 1985	6	341	2
January 29, 1986	0	104	0
March 11, 1986	11	278	4
May 15, 1986	0	81	0
November 10, 1986	1047	2450	43
March 3, 1987	827	1718	48
October 26, 1987 ^a	531	1754	30
March 13, 1988 ^a	766	1949	38

^aData collected for a 90-m reach but adjusted to represent 116-m reach.

Cultures of pure *A. hydrophilia* were obtained from the USFWS National Fish Health Research Laboratory located in Kearneysville, West Virginia. Bacterial suspensions of known density and purity were prepared by streaking a small amount of stock bacteria onto Bacto-agar plates. After sufficient growth of colonies, the bacteria were transferred to trypticase soy broth in 2000 mL Erlenmeyer flasks and allowed to grow. When an optimum density was reached, the culture fluid was centrifuged to recapture the bacteria. These bacteria were resuspended in a phosphate buffer solution at pH 7.2 to 7.3 before they were added to two of the experimental tanks. For each of the four treatment groups, the time to mortality was recorded for all fish in each tank. The mean time to death was calculated for each group as the mean number of days required for mortality of all fish in each tank.

The experimental combination of high fish density and bacteria had the greatest effect on mortality rate (Fig. 2). Without bacteria, mean time to death was 12.8 d at the low fish density and 10.1 d at the higher density, indicating a slight effect of density alone on mortality. At the low fish density, the mean time to death in the tanks having bacteria was about the same as that in the tanks without bacteria. However, at the high densities, mean time to death decreased substantially (Fig. 2). These results suggest that fish density could have been one of the major factors influencing both fish kills.

High densities of fish in the presence of the ubiquitous *Aeromonas* pathogen may not, however, initiate an epizootic. Disease outbreaks usually occur when fish are stressed by additional environmental factors such as increases of a contaminant or temperature (Meyer 1970; Wedemeyer 1970). Some evidence suggests that at high fish densities specific biochemical inhibitory factors produced by the fish themselves can cause stress or mortality. Arzapalo et al. (1980) working with *Tilapia* spp. at high densities, showed that these fish displayed an autoimmune response to hemagglutinins in the mucus. According to Smith (1977), hemagglutinins in fish mucus can induce anaphylactic (secondary) responses of varying degrees of severity in fish. In some instances, these responses predispose fish to disease from secondary etiologic agents and in other cases they cause death directly. Thus,

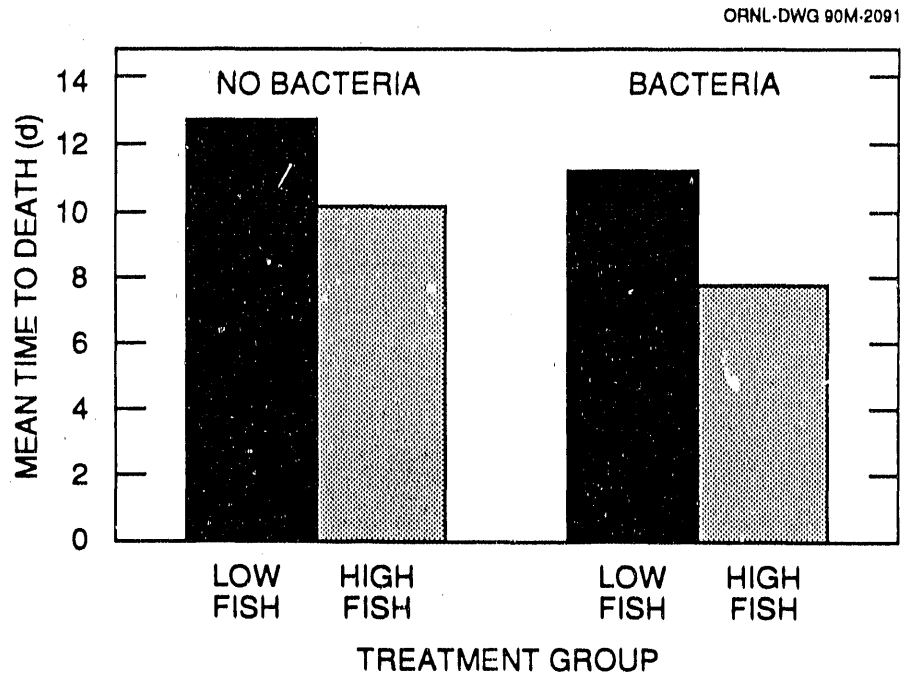


Fig. 2. Effect of bacteria and fish density on the mortality rate (mean number of days required to kill the population in each tank) of stonerollers obtained from an unpolluted stream.

overcrowding is not only a stressor but also an agent that enhances the spread of infectious diseases, such as BHS.

The significance of high densities or overcrowding as a source of stress to the stoneroller population below NHP, however, is unclear because the species normally forms large schools while foraging. Moreover, the densities of 1.93 and 1.42 individuals per square meter observed on November 10, 1986, and March 3, 1987, respectively, in EFPC approximately 200 m below NHP was similar to that found in May 1986 at other sites located 5 to 15 km downstream and was more than a factor of 2 lower than the density observed in White Oak Creek east of the ORNL site boundary (Ryon 1987). No evidence of BHS in the White Oak Creek population has been reported.

In addition to their recent high densities below NHP, the stoneroller has a unique feeding habit that may explain why this species was the only one affected by the epizootic. It is an herbivore and grazes on periphyton (attached algae) that is abundant on rock surfaces below NHP. While feeding, stonerollers ingest particulate matter, including fish feces, that settles on the stream bottom. Ingestion is a major pathway in the transmission of BHS between individuals in a population (Wolke 1975). Crowding of a bottom-dwelling species may also explain why the disease is commonly observed in pond culture of catfishes. Although other "bottom-dwelling" fishes inhabit upper EFPC, including the white sucker (*Catostomus commersoni*), a species that is susceptible to bacterial disease caused by *Aeromonas hydrophila* (Pippy and Hare 1969), their densities are at least one or two orders of magnitude below the density of stonerollers.

3.2 TEMPERATURE

Like overcrowding, temperature can be a source of environmental stress and can enhance the transmission of BHS. For example, salmonid epizootics caused by BHS can increase dramatically at temperatures above 7 to 10°C (Wedemeyer et al. 1976). Also, salmonid mortality rates from *Aeromonas* infections increase as temperatures increase, with high rates at 17.8 to 20.5°C (Fryer et al. 1976). Available data suggest that the thermal environment below NHP could have enhanced the spread of the

pathogen. For example, average daily water temperatures for a 2-week period prior to November 21 were generally above 15°C at a site approximately 200 m below NHP, and maximum temperatures often exceeded 20°C (Fig. 3). Mean and maximum daily temperatures were generally 6 to 7°C lower in Brushy Fork, a reference stream located just north of Oak Ridge. A similar trend occurred in July 1987; mean daily temperatures were 4 to 5°C higher and daily maximums were 8°C higher in EFPC than in Brushy Fork (Fig. 3).

The thermograph record was also reviewed to determine if a significant drop in air temperatures on November 13-14 (as a result of passage through the region of the first major cold front of the fall) could have caused a correspondingly abrupt change in water temperature and triggered the outbreak of BHS. The maximum water temperature below NHP just prior to the cold front was 20.8°C at 1600 h on November 12. Except for a slight rise in temperature in late afternoon of the following day, the temperature declined steadily over the next 36 h (from 20.8° to 13.1°C at 0600 h on November 14). The maximum rate of change (0.5°C/h) occurred between 1800 h and midnight on November 13, when the water temperature fell from 17.0° to 14.0°C. The maximum rate of change in Brushy Fork was 0.3°C/h over approximately the same time period. The abrupt decline in water temperatures in EFPC, although greater than that observed in Brushy Fork, was probably not sufficient to have stressed the stoneroller populations and initiated the epizootic (Elliott 1981). These conclusions regarding the November 1986 temperatures were further supported by the lack of any major temperature shifts associated with the fish kill in July 1987.

3.3 ELECTROSHOCKING

Another potential source of stress on the fish populations below NHP was electroshocking, a technique routinely used to estimate fish population size. Although Mr. Carlson suggested the electroshocking conducted on October 24, 1986, may have caused the outbreak of BHS approximately 4 weeks later, the evidence is not convincing.

A review of the electroshocking pattern in upper EFPC for the period spanning the fish kills (Table 9) indicates a continual source of

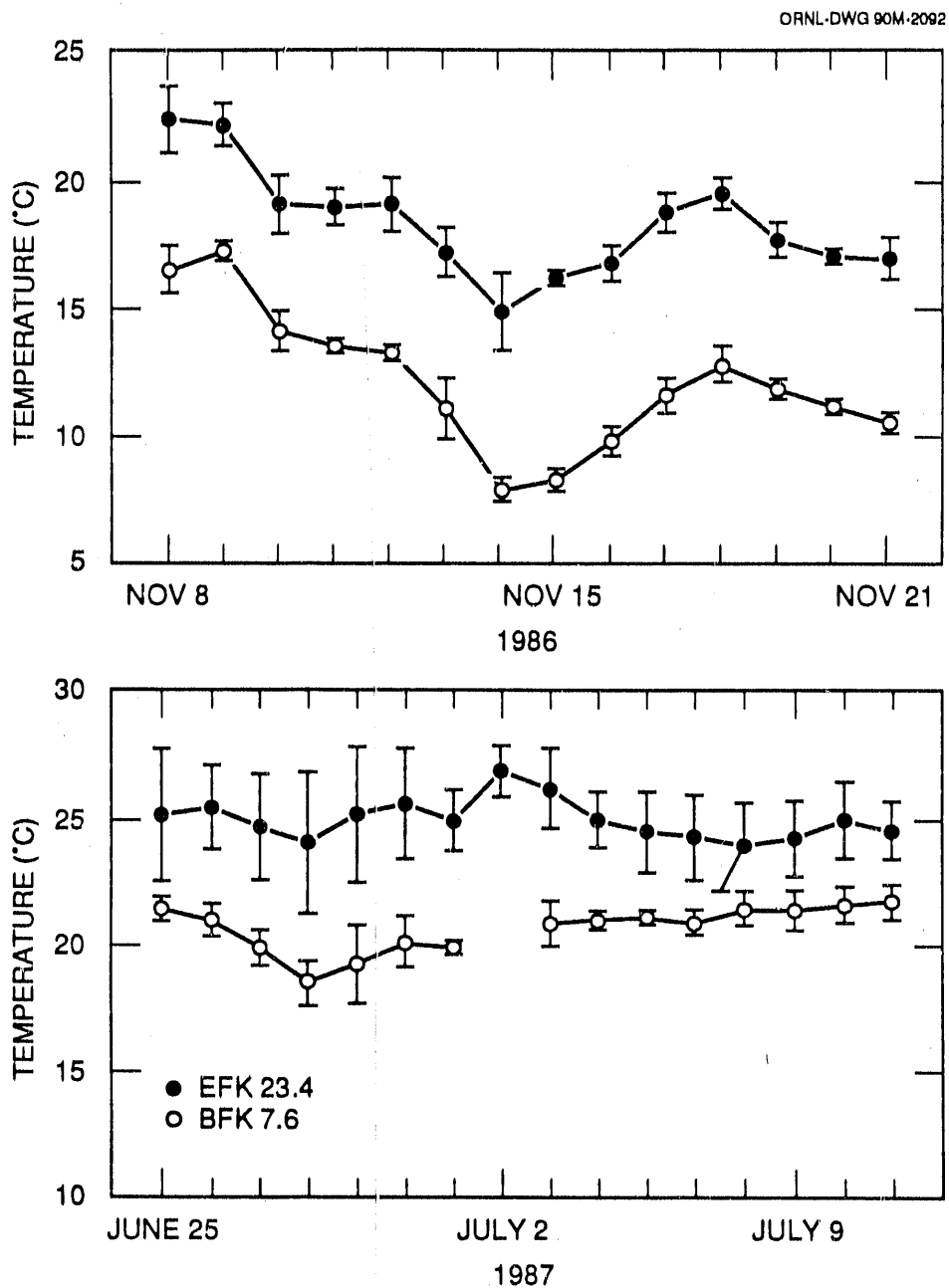


Fig. 3. Daily mean temperatures in East Fork Poplar Creek (EFK) and Brushy Fork (BFK) prior to fish kills on November 21, 1986, and July 9, 1987. Temperature data were recorded prior to the fish kill at 2-h intervals using a Ryan-Peabody thermograph (Model J 90). The temperature data for the second fish kill were recorded at 1-h intervals using a Ryan TempMentor digital thermograph.

Table 9. Electros shocking activities in upper East Fork Poplar Creek below New Hope Pond during the period associated with the fish kills

Date	Activity	Sampling reach (EFK ^a)	Number of passes through reach	Duration of electroshock activity (h)
10/09/86	Tagging ^b	22.0-23.6	1	2
10/24/86	Fish collection for blood/enzyme analyses ^b	23.0-23.6	1	1
11/10/86	Population estimation ^c	23.3-23.4	3	3
11/21/86	Fish kill			
03/03/87	Population estimation ^c	23.3-23.4	3	2.5
03/20/87	Fish collection for blood/enzyme analyses ^b	23.0-23.6	1	1
03/31/87	Tagging ^b	22.0-23.6	1	2
05/22/87	Fish collection for contaminant analyses ^b	22.0-23.6	1	1
06/18/87	Fish collection for blood/enzyme analyses ^b	23.0-23.6	1	0.5
06/25/87	Fish collection for parasite studies ^b	23.1-23.5	1	0.5
07/07/87	Tagging ^b	22.0-23.6	1	2
07/09/87	Fisk kill			

^aEFK - East Fork Poplar Creek kilometer.

^bSunfish only.

^cAll species.

stress. Electroshocking is used below NHP for various collecting purposes, and all incidents could adversely affect the stoneroller population. Before both fish kills, electroshocking occurred on several dates. However, similar periods of electroshocking occurred in March 1987 (some at temperatures similar to those of the November 1986 kill) and had no adverse effect on the stoneroller population.

An assumption associated with the electroshocking hypothesis is that stonerollers received more stress or were more susceptible to electroshocking at one period than at another. Otherwise, BHS would occur following every electroshocking event. Because the mortality of stonerollers was high during the October 24, 1986, sampling trip, it is possible that the population in only this area was under stress from some unknown source (e.g. extreme overcrowding). The additional stress of the electroshocking could have had both immediate and latent effects (i.e., initiation of the epizootic 4 weeks later, as suggested by Mr. Carlson). However, more extensive electroshocking than that used on October 24 was employed on November 10, 1986, during quantitative sampling of the fish populations in a 116-m reach of EFPC approximately 200 m below NHP. At this time, three consecutive passes with two electroshocking units (compared with one pass with a single unit in October) were made through the study reach. Fish were anesthetized with tricaine methanesulfonate, weighed, measured, and then returned to the stream. Even with these additional sources of stress, stoneroller mortality was less than 5%, whereas that of the striped shiner, a species unaffected by the epizootic, was substantially higher (43%). Sampling mortality of the other ten species combined was 4%.

Another problem with designating the October 24 and June 18 electroshocking periods as the primary stress, is related to the length of the incubation period for BHS. At the temperatures that existed in late October 1986 and mid-June 1987, the incubation period between the first stress and the *Aeromonas* infection would have been a week or less. This would place the fish kill substantially before November 21 or July 9. Although the electroshocking hypothesis cannot be rejected, additional evidence would be required for electroshocking to be accepted as the stress that caused the outbreaks of BHS.

3.4 POLLUTANTS

3.4.1 Mercury

Some data indicate that the epizootic may have been caused by elevated levels of mercury (Hg) associated with a storm-sewer cleaning and relining project at the Y-12 Plant. Total Hg concentrations in EFPC at the inlet to NHP exceeded 1000 ppb and approached 100 ppb just prior to the November 1986 and July 1987 fish kills, respectively (Appendix D). On both occasions, Hg concentrations during the preceding 3 weeks ranged from 2 to 4 ppb, which is typical for this site. Although there is always some Hg in the water and sediments below NHP, the elevated levels in November 1986 and July 1987 may have provided enough additional stress to have caused the fish kills. No fish kills have been observed from the time the sewer relining project was completed in late 1987 until the present (approximately 18 months).

Bioaccumulation of Hg was measured in November 1986 and July 1987. The Hg concentration in the alimentary tract of a dead stoneroller collected on November 22 from below NHP was only 8 $\mu\text{g/g}$, wet weight (Appendix E), whereas Hg concentrations in the surface sediments of the stream in this area range from 20 to 150 $\mu\text{g/g}$. Analysis of more extensive samples taken from stonerollers and a white sucker collected in July 1987 also indicated low Hg concentrations in intestine, muscle, and whole-body samples (Table 10). Also, the mercury content of periphyton, the food source for stonerollers, was determined to be $159 \pm 44.0 \mu\text{g/g}$ (Appendix F). Consequently, stress caused by the uptake of Hg adsorbed to suspended particulates seems unlikely. However, like many other possible explanations for the kill, elevated Hg levels in water cannot be refuted as the cause of the fish kill.

Table 10. Total mercury levels ($\mu\text{g/g}$) in intestine, muscle, and whole-body samples of fish taken from upper East Fork Poplar Creek during July 1987

Sample date	Sample type	Total Hg
Stoneroller (<i>Campostoma anomalum</i>)		
July 16	Whole body ^a	3.39
July 16	Whole body	2.04
July 23	Whole body	1.77
July 11	Intestine	3.4
July 11	Intestine	80.0
July 16	Intestine	6.0
July 22	Intestine	12.0
July 22	Intestine	8.9
July 11	Muscle ^b	1.2
July 11	Muscle	1.7
July 16	Muscle	1.5
July 22	Muscle	1.2
July 22	Muscle	1.2
White sucker (<i>Catostomus commersoni</i>)		
July 22	Intestine	7.3
July 22	Muscle	0.97

^aWhole body minus intestine and alimentary tract contents.

^bMuscle tissue represented in filet.

3.4.2 Other Inorganics

Most other inorganics varied within normal (for NHP outfall) ranges during October–December 1986 (Appendix G and H). Higher-than-normal levels of lithium (approximately 0.2 vs 0.02 ppm) were noted on several occasions in October and November, and uranium was about ten times higher (0.13 to 0.28 vs 0.03 ppm) in NHP outfall during October 7–13 than that observed in November and December. Prior to and during the fish kill, abnormal levels of nitrate (53 and 25 ppm on November 18–19, 1986, vs 2 to 3 ppm on other dates) and residual chlorine (0.45 ppm on November 21, 1986, vs <0.1 ppm on other dates) were observed at NHP outfall. The high level of chlorine coincides with the start of the fish kill; however, similar levels were not observed in composited water samples taken for toxicity testing on that date (Table 4).

Concentrations of inorganics in the NHP outfall in June and July 1987 differed from those found in October and December 1986 (Appendix I). Lithium and nitrates had not increased noticeably. However, aluminum levels were increased from normal levels of 0.1 to 0.3 ppm to 1.09, 3.24, and 1.43 ppm on June 19, July 7, and July 8, respectively. Ammonia levels were also elevated to 2.2, 1.4, and 0.9 mg/L on July 8–10, respectively. Total residual chlorine concentrations (mg/L) measured in the outfall of NHP during the ambient toxicity tests were not substantially higher during July 9–15 test (0.0 to 0.20) in comparison to other test periods (0.0 to 0.14) (Table 7 and Appendix C).

3.4.3 Organics

Numerous organic compounds were detected in NHP inlet and outlet samples between October and December 1986, most at very low levels (<0.05 ppm). (Compounds included in this analysis are listed in Appendices J and K.) The only organic found in excess of 1 ppm was acetone, which exhibited a seemingly random pattern of occasional spikes to levels as high as 47 ppm (October 7, 1986) amidst a normal day-to-day concentration below 0.01 ppm. The very high level of acetone observed on October 7 was not corroborated by the total organic carbon (TOC)

analyses of that sample (6 ppm), suggesting an artifact or sample contamination. There did not appear to be any increase in either TOC or biological oxygen demand (BOD) associated with acetone levels in excess of 1 ppm, also suggesting an artificial source.

The other organics most common in NHP inlet and outlet samples were butylcarbitol and another (unidentified) polymeric ether. When summed, levels of these two organics approached 0.5 ppm on some dates. During the week of the first fish kill (November 18-24), butylcarbitol ranged from 0.006 to 0.41 ppm, averaging approximately 0.2 ppm, while levels of the other polymeric ether averaged 0.1 ppm. It is unlikely that such levels of these compounds were acutely toxic to fish, but data are not available. Levels approaching 0.5 ppm could act as a source of nutrition for microorganisms and promote their growth in NHP (Sect. 3.6).

Other organics found at lower concentrations and less frequently in these samples included tributyl phosphate, 4-tert-butylphenol, diacetone alcohol, prometone (an herbicide used at the Y-12 Plant for weed control), a substituted diphosphoric acid ester, substituted octadecanoic and hexadecanoic esters, tetrahydrofuran, methylene chloride, methyl chloride, bromoform, phthalates, polyaromatic hydrocarbons, polychlorinated biphenyls, and alkylhydrocarbons.

High levels of TOC (86 to 380 ppm) were observed in NHP outfall samples on November 28, 29, and December 4, 1986. These values were corroborated by high values of chemical oxygen demand but not high BOD. Analysis by gas chromatography/mass spectroscopy did not detect any organics at levels even approximating this high TOC. The nature and source of the high TOC in these samples remains unknown.

Analyses of NHP inlet and outfall samples for organics was also conducted in June and July 1987 as part of a toxicity assessment. No significant concentrations of any organic compounds were detected.

Fish in the reach of EFPC below NHP received intermittent exposures to low levels of a number of inorganic and organic chemicals in the 8 weeks prior to the November fish kill and the month prior to the July fish kill. The most consistent pattern of exposure occurred in the case of mercury, and large peaks in concentration coincided with both

fish kills. The most toxicologically significant exposure (in terms of known toxic concentrations) was the 0.45 ppm chlorine noted on November 21, the day of the first fish kill. This exposure may have played a role in exacerbating the effects of the bacterial infection already present in the fish population.

3.5 CUMULATIVE STRESS

The epizootic might have been caused by cumulative stress. According to this hypothesis, none of the previously mentioned factors (overcrowding, temperature, electroshocking, and elevated Hg levels) was sufficient to have initiated the outbreak of BHS. However, if it is assumed that they occurred within the same general time frame (approximately a 4-week period prior to the kills), then the fish experienced a cumulative or synergistic stress, eventually resulting in a weakened condition characterized by reduced resistance to disease. Under stress, the immune system becomes dysfunctional, rendering fish more susceptible to mortality from numerous causes (Shul'man 1974; Wedemeyer and McLeay 1981; Anderson, in press).

3.6 ROLE OF THE PATHOGEN AND NEW HOPE POND

The last hypothesis regarding the cause of the epizootic below NHP emphasizes the role of the pathogen *Aeromonas hydrophila* and the environmental conditions in NHP. Hazen (1979) found that the occurrence of infected largemouth bass (*Micropterus salmoides*) was significantly correlated with the density of *A. hydrophila* in a South Carolina reservoir. The comparison was even more significant when a time-lag of a month occurred between high pathogen densities and largemouth bass infections. Peak densities of *A. hydrophila* occurred in June and October.

The environment in EFPC below NHP is assumed to have been stressful for some time prior to the kills as a result of overcrowding and/or chronic exposure to pollutants. Although fish may have been stressed prior to outbreaks of BHS on November 21 and July 9, as indicated by the high mortality that occurred during an electroshocking episode on October 24, no epizootic was observed. This may have been because a

presumably low pathogen population existed at that time. A later increase in pathogen levels in NHP may have triggered the epizootic that resulted in the November 1986 fish kill.

The BMAP documented significant changes (i.e., senescence) in the aquatic macrophytes in NHP between September and December 1986. Such natural seasonal decomposition of the pond vegetation can, in turn, affect the redox potential of the sediments and the types and concentrations of dissolved and particulate organic matter present in and exported from the pond. In a South Carolina reservoir, high densities of *A. hydrophila* were found in decaying mats of aquatic vegetation (Hazen 1979). If a similar situation existed in NHP, an increase in pathogen densities could have occurred in EFPC during the time of the fish kill(s). Drying commercial fish ponds at least once a year is recommended as one method of reducing the buildup of organic matter and fish pathogens in pond bottoms (Wedemeyer et al. 1976). By comparison, NHP was last cleaned (i.e., dredged) almost 15 years ago. Thus, it is conceivable that pathogen populations in the pond increased significantly, ultimately triggering the outbreak of BHS and resulting in the fish kills. However, this hypothesis lacks direct verification. Virtually nothing is known of microbial population dynamics in NHP.

4. ENVIRONMENTAL CONSEQUENCES OF THE EPIZOOTIC

The impact of the epizootic on the stoneroller population in EFPC can be estimated from BMAP census data. A population size of 1047 individuals was calculated for a 116-m reach of EFPC downstream of NHP in November 1986 prior to the fish kill (Table 8). Assuming that the stoneroller population density in this study section is representative of that over a much larger reach of upper EFPC, the estimated stoneroller population in the 0.5-km reach of EFPC between Bear Creek Road (EFK 23.15) and NHP (EFK 23.65) was 4513 individuals for November 1986. Although dead fish were actually collected over a much larger reach (1.4 km), it was assumed that many had drifted downstream and that the actual area of the kill was probably more limited. Therefore, a 0.5-km rather than 1.6-km reach was used in the calculations.

The loss of 1146 individuals as a result of the epizootic (Table 1) represented a 25% reduction in the stoneroller population of upper EFPC in November 1986. Similar calculations of the fish kill in July 1987 showed that 729 stonerollers died out of an estimated population of 3560, representing 20% mortality. Recognizing that (1) not all of the fish that died from the epizootic were enumerated [ESD staff and W. Schacher of TWRA agree, however, that the collection efficiency was relatively high (Schacher 1986)] and (2) actual densities just below NHP may have been much higher than 7 to 9 fish per meter of stream, the estimated mortality probably ranged from 20 to 30% of the population. This level of impact on the stoneroller population could be significant.

Although the proportion of the total fish population represented by stonerollers increased from 43 to 48% after the November fish kill, the proportion of stonerollers decreased to only 30% after the July fish kill. Overall, the stoneroller population in the 116-m study section of EFPC below NHP declined by 50% between samples taken before and after the two fish kills (Table 8). Recovery of the stoneroller population from the fish kills was indicated by sampling on March 13, 1988, in which both proportion and total stoneroller population increased (Table 8).

5. INTERAGENCY INTERACTIONS

Successful investigation of these fish kills required extensive interaction and cooperation between several parties, including various organizations within the investigation team (ORNL and Y-12 Plant staffs) and the appropriate regulatory agencies (TWRA and Tennessee Department of Health and Environment). These interactions are summarized in Tables 11 and 12.

Table 11. Chronological sequence of interactions between various parties responsible for the investigation of the fish epizootic that occurred in November 1986 in East Fork Poplar Creek below New Hope Pond. Interactions between staff members of the Environmental Sciences Division (ESD) at Oak Ridge National Laboratory are not included

Date	Type of interaction		Principal parties ^a	Subject
	Phone Call	Meeting		
11/21/86	X		J. D. Gass (Y-12) J. M. Loar (ESD)	Notification of fish kill
11/21/86		X	J. M. Loar (ESD) G. R. Southworth (ESD) C. Kimbrough (Y-12)	Analysis of water quality data
11/21/86	X		D. Melgaard (TDHE) J. M. Loar (ESD)	Call received but no interaction
11/21/86		X	W. Schacher (TWRA) J. Evans (TWRA) J. M. Loar et al. (ESD)	Assist with fish survey
11/22/86		X	W. Schacher (TWRA) J. M. Loar (ESD) G. R. Southworth (ESD)	Assist with fish survey
11/22/86	X		J. M. Loar (ESD) T. R. Butz (Y-12)	Update on fish kill
11/23/86		X	W. Schacher (TWRA) J. M. Loar (ESD)	Assist with fish survey
11/23/86	X		J. M. Loar (ESD) T. R. Butz (Y-12)	Update on fish kill
11/24/86	X		J. M. Loar (ESD) B. Clark (TDHE)	Update on fish kill
11/24/86		X	ESD/Y-12/DOE	Exchange of information on fish kill

Table 11. (continued)

Date	Type of interaction		Principal parties ^a	Subject
	Phone Call	Meeting		
11/24/86	X		C. L. Stair (ESA) J. M. Loar (ESD)	Update on fish kill
11/25/86	X		J. M. Loar (ESD) C. C. Hill (Y-12)	Update on fish kill
11/26/86	X		J. M. Loar (ESD) C. C. Hill (Y-12)	Update on fish kill
12/01/86		X	ESD/Y-12/DOE	Exchange of information on fish kill
12/01/86	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Content of press release
12/08/86	X		J. M. Loar (ESD) W. Schacher (TWRA)	Update on dead fish count
12/16/86		X	ESD/Y-12/DOE/TDHE/EPA	Summary of fish kill

^aDOE - Department of Energy; ESA - Environmental and Safety Activities, Martin Marietta Energy Systems, Inc.; TWRA - Tennessee Wildlife Resources Agency; TDHE - Tennessee Department of Health and Environment; Y-12 - Health, Safety, Environment and Accountability Division, Y-12 Plant.

Table 12. Chronological sequence of interactions between various parties responsible for the investigation of the fish epizootic that occurred in July 1987 in East Fork Poplar Creek below New Hope Pond. Interactions between staff members of the Environmental Sciences Division (ESD) at Oak Ridge National Laboratory are not included

Date	Type of interaction		Principal parties ^a	Subject
	Phone Call	Meeting		
7/9/87	X		C. W. Gehrs (ESD) J. D. Gass (Y-12)	Notification of fish kill
7/9/87	X		J. D. Gass (Y-12) C. W. Gehrs (ESD)	Update on fish kill
7/10/87	X		J. D. Gass (Y-12) C. W. Gehrs (ESD)	Update on fish kill
7/10/87	X		S. M. Adams (ESD) C. Carlson (USFWS)	Discussion of pathogen smears
7/11/87	X		J. D. Gass (Y-12) J. M. Loar (ESD)	Discussion of water quality data
7/11/87	X		J. M. Loar (ESD) C. C. Hill (Y-12)	Update on fish kill
7/12/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Update on fish kill
7/13/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Update on fish kill
7/13/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Discussion of water quality data
7/14/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Update on fish kill
7/14/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Discussion of press release
7/14/87	X		J. M. Loar (ESD) B. Clark (TDHE)	Update on fish kill

Table 12. (continued)

Date	Type of interaction		Principal parties ^a	Subject
	Phone Call	Meeting		
7/15/87	X		B. Clark (TDHE) J. M. Loar (ESD)	Update on fish kill
7/15/87	X		J. M. Loar (ESD) C. C. Hill (Y-12)	Update on fish kill
7/16/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Update on fish kill
7/17/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Update on fish kill
7/18/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Update on fish kill
7/19/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Update on fish kill
7/20/87		X	ESD Staff	Discussion of fish kill
7/20/87	X		J. M. Loar (ESD) C. C. Hill (Y-12)	Update on fish kill
7/21/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Update on fish kill
7/21/87	X		J. M. Loar (ESD) B. Clark (TDHE)	Update on fish kill
7/22/87	X		J. M. Loar (ESD) C. C. Hill (Y-12)	Update on fish kill
7/23/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Update on fish kill
7/27/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Update on fish kill

Table 12. (continued)

Date	Type of interaction		Principal parties ^a	Subject
	Phone Call	Meeting		
7/29/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Update on fish kill
7/29/87	X		C. Carlson (USFWS) J. M. Loar (ESD)	Discussion of bacterial smears
8/4/87	X		J. M. Loar (ESD) C. C. Hill (Y-12)	Update on fish kill
8/5/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Update on fish kill
8/6/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Update on fish kill
8/7/87	X		C. C. Hill (Y-12) J. M. Loar (ESD)	Update on fish kill

^aTDHE - Tennessee Department of Health and Environment;
Y-12 - Health, Safety, Environment and Accountability Division,
Y-12 Plant; USFWS - U.S. Fish and Wildlife Science.

6. CONCLUSIONS/RECOMMENDATIONS

The direct cause of the fish kills that were first observed on November 21, 1986, and July 9, 1987, just below the outfall of NHP is attributed to an outbreak of BHS, a stress-mediated disease common in commercial fish culture operations. The pathogen was identified as *Aeromonas hydrophila* by Charles Carlson, a fish disease specialist with the USFWS, who analyzed kidney samples taken from stonerollers collected below NHP. The disease resulted in a 20 to 30% reduction in the stoneroller population within a 0.5-km reach of EFPC below NHP. Their high population densities, strong schooling behavior, and unique benthic feeding habit, in combination with elevated stream temperatures, probably explain why only stonerollers were affected and how the disease was so rapidly transmitted among individuals in the population.

The cause of the epizootic (i.e., the events or stresses that resulted in an outbreak of BHS) cannot be identified. Overcrowding and temperature were probably more important in promoting the rapid spread of the disease than in initiating the epizootic. The rapid decrease in water temperature that occurred approximately 1 week prior to the outbreak of the disease in November 1986 was probably not a significant factor. Instead, the outbreak may have been caused by

(1) electroshocking, (2) elevated levels of mercury associated with the cleaning and relining of highly contaminated storm-sewer lines, (3) cumulative stress from these and other unidentified physical or chemical stressors, or (4) changes in environmental conditions within NHP that enhanced the growth of the pathogen population.

For an epizootic of BHS to occur, the host (fish) must interact with sufficient densities of a pathogen in a stressful environment. Consequently, to control future outbreaks of the disease, information is needed on both the population dynamics of the pathogen and the source(s) of stress in the environment. For example, additional studies would be required to identify environmental conditions that promote the growth of *Aeromonas hydrophila* populations in NHP. However, such studies are not recommended now because the source of stress that triggered the outbreak could have been elevated Hg levels. These levels were associated with a

remedial action (storm sewer cleaning and relining project) that was completed more than 18 months ago. To reduce stress on stoneroller populations, the collection of redbreast sunfish (*Lepomis auritus*) by electroshocking was combined for the bioaccumulation and biological indicator studies of the BMAP. Techniques were also modified to minimize the exposure of dense schools of stonerollers to electric current. Finally, if total Hg levels in the discharge from NHP exceed 25 ppb, it is recommended that Y-12 Plant staff (1) notify appropriate personnel responsible for biological monitoring (currently J. M. Loar of the Environmental Sciences Division at ORNL) and (2) make frequent observations near the discharge for evidence of an incipient fish kill. Because the cause of the epizootics was not conclusively identified, future outbreaks may be difficult to eliminate. However, the completion of the storm-sewer cleaning and relining project, the closing of NHP, and the restriction of electroshocking activities probably reduced the likelihood of future outbreaks of BHS in upper EFPC.

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APPENDIX A

MEMO FROM L. M. ADAMS (KSZOS), ENVIRONMENTAL SCIENCES DIVISION (ORNL),
TO J. M. LOAR, ENVIRONMENTAL SCIENCES DIVISION (ORNL),
DESCRIBING RESULTS OF TOXICITY TESTS ON SEWER LINE
SLUDGE, USING STONEROLLERS (*Campostoma Anomalum*)

APPENDIX A

MARTIN MARIETTA

Internal Correspondence

MARTIN MARIETTA ENERGY SYSTEMS, INC.

To: Jim Loar

From: Lynn M. Adams *LM Adams*

Subject: Toxicity tests with sludge from Y-12 Northwest sludge basin.

*Rec'd 12-2-86
XO GWS
GRS
AJS*

I. Five aquaria were set up on November 25 containing 10 L of dechlorinated tap water and ten Campostoma. Sludge from the Y-12 northwest sludge basin was added to four of the aquaria in approximate concentrations of 30 g/L, 15 g/L, and 0.5 g/L. One aquaria with dechlorinated tap water served as a control. After 6 hours of the test, aeration was added to maintain sufficient dissolved oxygen. The two aquaria containing 30 g/L were terminated after 24 hours due to high mortality. I believe the mortality in these aquaria may have been due to high turbidity (>200 NTU).

Conc. (g/L)	No. Fish Beginning	Number Surviving (hours)					Turbidity (NTU)
		17	24	67	87	111	
30	10	5	1	—	—	—	> 200
30	10	4	2	—	—	—	> 200
15	10	8	5	0	0	0	> 200
0.5	10	10	9	8	8	7	23
Control	10	10	10	10	10	10	23

II. One additional aquaria was set up on November 26 containing 10 L of dechlorinated tap water, six Campostoma, and approximately 1 g/L of the sludge.

Conc. (g/L)	No Fish Beginning	Number Surviving (hours)		Turbidity (NTU)
		43	75	
1.0	6	4	4	35
Control	10	10	10	23

APPENDIX B

LETTER FROM C. P. CARLSON, U.S. FISH AND WILDLIFE SERVICE, TO
S. M. ADAMS, ENVIRONMENTAL SCIENCES DIVISION (ORNL),
DESCRIBING RESULTS OF THE LABORATORY TESTS ON
STONEROLLERS (*Campostoma Anomalum*)



APPENDIX B

United States Department of the Interior

FISH AND WILDLIFE SERVICE

Hatchery Biologist-Area

P. O. Box 158

Pisgah Forest, North Carolina 28768

December 2, 1986

Rec'd 12-5-86 JMU

Marshall Adams
Building 1505
Mail Stop 36
Oak Ridge National Laboratory
P. O. Box X
Oak Ridge, TN 37831

Dear Marshall,

I have completed the laboratory tests of kidney material taken from the stonerollers (Campostoma anomalum) at your laboratory on November 26, 1986.

As I reported to you yesterday during our telephone conversation the stonerollers were infected with the bacterium Aeromonas hydrophilia which causes the fish disease bacterial hemorrhagic septicemia (also known as abdominal dropsy, motil aeromonad septicemia, red pest, etc.). These bacteria were isolated on tryptic soy agar (TSA) slants inoculated with kidney material from the stonerollers. 4/4 cultures were positive for Aeromonas hydrophilia from frozen stonerollers collected on November 25, 1986, 2/3 positive from frozen stonerollers collected on November 21, 1986, and 1/2 positive from fresh stonerollers collected on November 26, 1986.

This group of bacteria are found in all natural water and also compose part of the normal intestinal microflora of healthy fish. They can cause the death of fish when they invade internal organs and begin to produce toxins. Stress can initiate an active epizootic of bacterial hemorrhagic septicemia in feral and cultured fish populations. Low oxygen, high water temperatures, handling, crowding, are some of the stress factors which can weaken the fish. The two pools in which the stonerollers died were electrofished last month. This may have produced enough stress on the fish to provoke the epizootic.

Sincerely

Charles P. Carlson
Charles P. Carlson

cc: James M. Loar ✓

APPENDIX C

**WATER QUALITY PARAMETERS MEASURED DURING THE AMBIENT TOXICITY
TESTS ON WATER FROM EAST FORK POPLAR CREEK ABOVE AND BELOW
NEW HOPE POND, JANUARY-JULY 1987**

Appendix C. pH, conductivity, alkalinity and hardness, and total residual chlorine in water from East Fork Poplar Creek just upstream from New Hope Pond and immediately below New Hope Pond (inlet and outfall, respectively) for various 7-d periods in 1987

Day	Sample type	pH	Conductivity ($\mu\text{mho/cm}$)	Alkalinity (mg/L as CaCO_3)	Hardness (mg/L as CaCO_3)	TRC (mg/L)
<u>Inlet</u>						
January						
09	Composite	8.08	364	103.0	162	0.37
10	Grab	8.23	344	109.0	168	0.34
11	Composite	8.16	428	104.0	164	0.17
12	Composite	8.10	432	107.0	154	0.26
13	Composite	8.05	329	104.0	144	0.64
14	Composite	8.08	398	100.0	170	0.27
15	Composite	8.16	312	100.0	130	0.28
<u>Outfall</u>						
January						
09	Grab	8.06	372	102.0	164	0.14
10	Grab	8.20	365	112.0	164	0.04
11	Composite	8.12	367	100.0	173	0.02
12	Composite	8.11	410	98.0	146	0.10
13	Composite	8.10	357	102.0	160	0.06
14	Composite	8.12	427	97.0	170	0.13
15	Composite	8.11	341	95.0	136	0.12
<u>Outfall</u>						
February						
05	Composite	8.01	438	96.0	154	0.04
06	Composite	8.08	395	108.0	178	0.04
07	Composite	8.24	416	118.0	176	0.03
08	Composite	8.23	331	105.0	157	0.03
09	Composite	8.18	418	114.0	162	0.02
10	Grab	8.07	388	105.0	152	0.11
11	Grab	8.02	357	106.0	160	0.11
<u>Inlet</u>						
March						
05	Composite	8.07	374	103.0	168	0.15
06	Grab	8.00	369	112.0	182	0.28
07	Grab	7.83	604	92.0	242	0.25
08	Grab	8.12	381	111.0	157	0.27
09	Grab	7.83	530	98.0	196	0.22
10	Grab	8.03	322	110.0	174	0.29
11	Grab	8.03	349	107.0	147	0.34

Appendix C. (continued)

Day	Sample type	pH	Conductivity ($\mu\text{mho/cm}$)	Alkalinity (mg/L as CaCO_3)	Hardness (mg/L as CaCO_3)	TRC (mg/L)
<u>Outfall</u>						
March						
05	Composite	8.10	395	82.0	164	0.03
06	Composite	8.01	368	112.0	166	0.04
07	Composite	8.15	344	112.0	170	0.02
08	Composite	8.23	358	108.0	174	0.00
09	Composite	8.03	376	112.0	158	0.00
10	Composite	7.98	360	103.0	166	0.00
11	Composite	8.02	412	109.0	172	0.05
<u>Outfall</u>						
April						
09	Composite	8.24	440	116.5	186	0.00
10	Composite	8.16	390	110.0	142	0.00
11	Composite	8.47	307	115.0	170	0.00
12	Composite	8.35	337	109.0	160	0.00
13	Composite	8.14	331	109.0	162	0.00
14	Composite	8.34	341	107.0	154	0.00
15	Composite	7.99	330	128.0	122	0.00
<u>Inlet</u>						
May						
07	Composite	8.21	471	119.0	178	0.06
08	Composite	8.04	477	114.0	178	0.04
09	Composite	8.05	461	112.0	186	0.03
10	Composite	8.11	457	112.0	160	0.00
11	Composite	8.22	491	107.0	198	0.00
12	Composite	8.13	461	109.0	178	0.02
13	Composite	8.18	461	112.0	186	0.01
<u>Outfall</u>						
May						
07	Composite	8.70	480	118.0	184	0.00
08	Composite	8.70	426	116.0	204	0.00
09	Composite	8.76	455	116.0	170	0.00
10	Composite	8.52	480	112.0	168	0.02
11	Composite	8.70	472	111.0	188	0.00
12	Composite	8.59	429	110.0	164	0.00
13	Composite	8.34	447	111.0	190	0.00

Appendix C. (continued)

Day	Sample type	pH	Conductivity ($\mu\text{mho/cm}$)	Alkalinity (mg/L as CaCO_3)	Hardness (mg/L as CaCO_3)	TRC (mg/L)
<u>Outfall</u>						
May						
21	Composite	8.03	470	115.0	182	0.00
22	Composite	8.22	554	104.0	178	0.00
23	Composite	8.26	498	108.0	184	0.00
24	Composite	8.16	427	105.0	174	0.00
25	Composite	8.32	407	107.0	174	0.00
26	Composite	8.23	472	102.0	184	0.00
27	Composite	8.35	493	106.0	106	0.00
<u>Outfall</u>						
June						
11	Composite	8.11	353	104.0	158	0.00
12	Composite	8.24	307	109.0	170	0.00
13	Grab	8.01	349	105.0	158	0.00
14	Grab	7.94	403	100.0	176	0.00
15	Composite	8.16	322	105.0	170	0.00
16	Composite	8.16	323	105.0	164	0.00
17	Composite	8.20	332	105.0	162	0.00

APPENDIX D

**CONCENTRATION OF TOTAL MERCURY (mg/L) IN GRAB SAMPLES COLLECTED
AT NEW HOPE POND FROM OCTOBER 27 THROUGH DECEMBER 15, 1986,
AND FROM JUNE 1 THROUGH JULY 20, 1987**

Appendix D

Date	Time	Inlet	Outlet
10/27/86	0930	0.0024	0.0000
	0935	0.0000	0.0011
	1500	0.0026	0.0010
10/28/86	1050	0.0024	0.0013
	1530	0.0042	0.0019
10/29/86	0915	0.0026	0.0013
	1435	0.0027	0.0014
10/30/86	0950	0.0028	0.0012
	1530	0.0021	0.0022
10/31/86	1035	0.0024	0.0013
	1430	0.0022	0.0019
11/03/86	1000	0.0022	0.0014
	1445	0.0035	0.0019
11/04/86	0845	0.0021	0.0013
	1500	0.0026	0.0020
11/05/86	0915	0.0020	0.0013
	1425	0.0019	0.0015
11/06/86	0915	0.0018	0.0013
	1440	0.0018	0.0018
11/07/86	0945	0.0013	0.0014
	1430	0.0015	0.0011
11/10/86	0925	0.0020	0.0018
	1445	0.0017	0.0013
11/11/86	0920	0.0023	0.0014
	1450	0.0019	0.0016
11/12/86	1020	0.0016	0.0010
	1505	0.0022	0.0014
11/13/86	0945	0.0021	0.0014
	1540	0.0018	0.0015
11/14/86	1001	0.0036	0.0043
	1500	0.0022	0.0022
11/17/86	0900	0.0015	0.0012
11/18/86	0845	0.0032	0.0022
	1415	0.0045	0.0020
11/19/86	1000	0.0033	0.0019
	1500	1.400	0.0022
11/20/86	0900	0.0110	0.0100
	1510	0.0420	0.0640
11/21/86	0915	0.0200	0.0120
11/24/86	0915	0.0140	0.0076
	1550	0.0070	0.0040
11/25/86	1000	0.0060	0.0032
	1415	0.0082	0.0037
11/26/88	0900	0.0044	0.0027
	1400	0.0036	0.0025

Appendix D. (continued)

Date	Time	Inlet	Outlet
12/01/86	0950	0.0044	0.0043
	1435	0.0042	0.0029
12/02/86	0845	0.0030	0.0021
	1320	0.0030	0.0020
12/03/86	0855	0.0040	0.0024
	1445	0.0220	0.0027
12/04/86	0905	0.0038	0.0056
	1430	0.0190	0.0039
12/05/86	0920	0.0700	0.0380
12/06/86	0920	0.0170	0.0110
	2015	0.0170	0.0110
12/07/86	1450	0.0180	0.0078
	1930	0.0660	0.0092
	2240	0.0200	0.0088
12/08/86	0915	0.0110	0.0100
	1445	0.0110	0.0060
	1450	0.0710	0.0330
12/09/86	0910	0.0070	0.0050
	1415	0.0090	0.0080
12/10/86	0855	0.0046	0.0026
	1500	0.0044	0.0044
12/11/86	0915	0.0036	0.0020
	1430	0.0030	0.0019
12/12/86	0850	0.0036	0.0027
	1430	0.0078	0.0031
12/15/86	0955	0.0046	0.0024
	1430	0.0400	0.0031
Min		0.0013	0.0000
Max		1.400	0.0640
Mean		0.0288	0.0051
6/01/87	0910	0.0032	0.0019
	1530	0.0050	0.0021
6/02/87	0820	0.0028	0.0120
	1400	0.0054	0.0019
6/03/87	1015	0.0030	0.0014
	1400	0.0022	0.0018
6/04/87	0910	0.0026	0.0012
	1510	0.0020	0.0012
6/05/87	0930	0.0032	0.0026
	1330	0.0120	0.0030
6/08/87	0945	0.0036	0.0013
	1600	0.0030	0.0016
6/09/87	1100	0.0030	0.0012
	1500	0.0026	0.0014

Appendix D. (continued)

Date	Time	Inlet	Outlet
6/10/87	0950	0.0032	0.0011
	1445	0.0028	0.0019
6/11/87	0915	0.0030	0.0012
	1530	0.0046	0.0022
6/12/87	0850	0.0036	0.0014
	1300	0.0032	0.0016
6/15/87	0850	0.0026	0.0013
	1410	0.0034	0.0018
6/16/87	0955	0.0038	0.0011
	1420	0.0064	0.0018
6/17/87	0830	0.0030	0.0015
	1230	0.0024	0.0020
6/18/87	0830	0.0024	0.0010
	1315	0.0026	0.0017
6/19/87	0955	0.0022	0.0013
	1500	0.0058	0.0015
6/22/87	0955	0.0020	0.0011
	1300	0.0024	0.0013
6/23/87	1120	0.0150	0.0014
	1300	0.0130	0.0020
6/24/87	1105	0.0110	0.0024
	1430	0.0052	0.0025
6/25/87	0915	0.0042	0.0016
	1430	0.0084	0.0024
6/26/87	1115	0.0120	0.0021
	1535	0.0060	0.0038
6/29/87	0850	0.0054	0.0020
	1410	0.0080	0.0032
6/30/87	0755	0.0054	0.0019
	1430	0.0066	0.0023
7/01/87	0750	0.0060	0.0018
	1410	0.0120	0.0025
7/02/87	0820	0.0044	0.0020
	1410	0.0098	0.0024
7/07/87	0820	0.0036	0.0062
	1415	0.0230	0.0079
7/08/87	0900	0.0200	0.0090
	1330	0.0900	0.0100
7/09/87	0815	0.0250	0.0120
	1325	0.0220	0.0088
7/10/87	0820	0.0210	0.0086
	1335	0.0240	0.0110
7/13/87	0910	0.0088	0.0072
	1400	0.0000	0.0045
	1420	0.0090	0.0054
	1500	0.0110	0.0000

Appendix D. (continued)

Date	Time	Inlet	Outlet
7/14/87	0830	0.0240	0.0120
	1415	0.0170	0.0140
7/15/87	0830	0.0140	0.0065
	1315	0.1400	0.0840
7/16/87	0900	0.0520	0.0270
	1420	0.0310	0.0240
7/17/87	1050	0.0290	0.0140
	1430	0.0180	0.0130
7/18/87	0830	0.0200	0.0100
	0945	0.0180	0.0085
	1115	0.0270	0.0095
7/19/87	0815	0.0140	0.0070
	1100	0.0170	0.0088
7/20/87	0850	0.0230	0.0170
	1450	0.0170	0.0050
Min		0.0000	0.0000
Max		0.1400	0.0840
Mean		0.0129	0.0060

APPENDIX E

CONCENTRATIONS OF INORGANIC CHEMICALS ($\mu\text{g/g}$) IN THE GUT CONTENTS OF
STONEROLLERS (*Campostoma Anomalum*) COLLECTED ON NOVEMBER 22, 1986

Appendix E

REQ: A02194-
 HNO3 DISSOLUTION AND FILTERED
 UNITS: mg/kg (=μg/g)
 DF = 11.7683
 PREP BY: MJF
 DATE: 11/22/86
 ID: ENV ICP

BASIS: AS RECEIVED
 PROC \$: 556
 ANAL BY: MJF
 FROM: DLI: FISH3.BRNINSTR

Concentrations (μg/g)		Concentrations (μg/g)	
Ag	2.2	Mg	440
Al	615	Mn	177
As	<3	Mo	0.7
B	<0.5	Na	1390
Ba	16.3	Nb	<0.6
Be	0.072	Ni	6.4
Ca	1770	P	1900
Cd	1.4	Pb	3
Ce	1.4	Sc	0.13
Co	2.4	Sr	3.64
Cr	3.1	Th	<0.6
Cu	30.2	Ti	4.4
Fe	1390	V	0.7
Ga	<0.6	Zn	109
K	1570	Zr	0.4
La	0.4	Hg	8
Li	1.22		

APPENDIX F

MERCURY CONTENT OF PERIPHYTON, FEBRUARY 10, 1987

MERCURY CONTENT OF PERIPHYTON

MARCH 7, 1987

Rocks containing periphyton were collected from shallow riffle areas in East Fork Poplar Creek and several streams in White Oak Creek watershed. These sites are included in the Biological Monitoring and Abatement Programs for the Y-12 Plant and ORNL, respectively. The rocks were collected by H. L. Boston and R. D. Bailey on February 10, 1987, and were taken to the laboratory in plastic containers of stream water from the collection sites. In the laboratory, the periphyton was scraped from the rock surfaces using a plastic bristled toothbrush. Triplicate samples were scraped into three aluminum pans and oven-dried for 42 h at 65°C. A 30- to 50-mg subsample of each dried sample was scraped into a clean scintillation vial and submitted for Hg analysis.

The Hg content was determined by cold vapor extraction and atomic absorption spectroscopy. The results are presented in the following table as ppm Hg on a dry weight basis. The data shown are the mean concentration (± 1 SD) for three replicates.

Site	Hg	Site	Hg
BF ^a	0.29 \pm 0.06	WCK 6.8 ^a	3.81 ^b
EFK 24.4	607 \pm 98.7	WCK 3.9	5.00 \pm 1.74
EFK 22.9	159 \pm 44.0	WCK 3.4	7.21 \pm 0.70
EFK 17.0	56.0 \pm 3.5	WCK 2.9	5.87 \pm 0.80
EFK 13.0	23.5 \pm 13.0	WCK 2.3	3.72 \pm 0.32
EFK 10.9	13.9 \pm 3.2	MEK 0.6	0.60 \pm 0.04
EFK 7.7	16.2 \pm 6.2	NTK 1.0 ^a	0.09 \pm 0.03 ^c

^aDenotes reference sites.

^bOnly one sample analyzed.

^cValues near detection limit of analysis and may be an overestimate.

- Notes:
1. Sediment in New Hope Pond contains about 150 ppm Hg.
 2. Sediment at EFK 22.9 contains approximately 60 ppm Hg.
 3. Hg values may be correlated with the organic content and the age (turnover time) of the periphyton (e.g., greater concentrations at EFK 7.7 may reflect greater organic content or age of periphyton).

APPENDIX G

CONCENTRATIONS OF INORGANIC COMPOUNDS (mg/L) IN COMPOSITE SAMPLES
OF WATER COLLECTED AT THE OUTFALL OF NEW HOPE POND DURING 7-d
TOXICITY TESTS AT NEW HOPE POND ON OCTOBER 7-14,
NOVEMBER 4-11, AND NOVEMBER 18-25, 1986

Table G-1. New Hope Pond Toxicity Test of 10/7 - 10/14/86

Dates:	10/07	10/08	10/09	10/10	10/11	10/12	10/13	Min	Max	Avg	Std Dev
Composites:											
Flow (mgd)	8.48	8.48	8.82	9.17	10.60	12.28	16.10	8.48	16.10	10.56	2.60
Hg	0.0019	0.0024	0.0011	0.001	0.0012	0.0014	0.0012	0.001	0.0024	0.0015	0.0005
Se	1.3	1.2	1.1	1.2	1.1	1.1	0.9	0.9	1.3	1.13	0.12
F	2.1	2.2	2	2	2.5	2.7	2.6	2.0	2.7	2.3	0.27
MNO3	62	80	110	58	66	85	68	58.0	110.0	75.6	16.6
SD4	14	10	7	8	14	6	13	6.0	14.0	10.3	3.1
TSS											
U	0.258	0.246	0.276	0.133	0.167	0.142	0.142	0.133	0.276	0.195	0.058
TOC	6	8	4	4	7.1	7.4	3.3	3.3	8.0	5.7	1.8
CM	0.005	0.003	0.007	0.007	0.004	<0.002	<0.002	0.002	0.007	0.004	0.002
pH (std units)	7.1	6.9	7.8	7.8	8.1	6.6	7.3	6.6	8.1	7.4	0.5
Al	0.39	0.3	0.11	0.21	0.28	0.25	1.03	0.11	1.03	0.37	0.28
As	<0.04	<0.04	<0.04	<0.04	<0.04	<0.04	<0.04	0.04	0.04	0.04	0.02
Ba	0.0609	0.0524	0.0488	0.051	0.0478	0.0521	0.0435	0.0435	0.0609	0.0509	0.0050
Be	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.0001	0.0001	0.0001	0.0000
B	0.032	0.033	0.036	0.03	0.034	0.036	0.034	0.030	0.036	0.034	0.002
Cd	<0.003	<0.003	<0.003	<0.003	<0.003	<0.003	<0.003	0.003	0.003	0.003	0.001
Ca	45.4	51.1	51.8	47.6	46.7	48.4	37.1	37.1	51.8	46.9	4.5
Ce	<0.02	<0.02	<0.02	<0.02	<0.02	<0.02	<0.02	0.02	0.02	0.02	0.01
Cr	<0.006	<0.006	<0.006	<0.006	<0.006	<0.006	<0.006	0.006	0.006	0.006	0.003
Co	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	0.002	0.002	0.002	0.001
Cu	0.014	0.01	0.006	0.007	0.007	0.008	0.008	0.006	0.014	0.009	0.002
Ga	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	0.01	0.00
Fe	0.56	0.45	0.16	0.31	0.41	0.35	1.01	0.16	1.01	0.46	0.25
Ia	<0.003	<0.003	<0.003	<0.003	<0.003	<0.003	<0.003	0.003	0.003	0.003	0.001
Pb	<0.02	<0.02	<0.02	<0.02	<0.02	<0.02	<0.02	0.02	0.02	0.02	0.01
Li	0.022	0.195	0.039	0.021	0.157	0.032	0.023	0.021	0.195	0.070	0.068
Mg	11.4	12.3	12.8	11.4	9.83	11.2	8.56	8.56	12.80	11.07	1.34
Mn	0.161	0.058	0.028	0.042	0.038	0.045	0.052	0.028	0.161	0.061	0.042
Mo	0.126	0.143	0.127	0.175	0.139	0.136	0.096	0.096	0.175	0.135	0.022
Ni	<0.007	<0.007	<0.007	<0.007	<0.007	<0.007	<0.007	0.007	0.007	0.007	0.003
Nb	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	0.01	0.00
P	0.5	0.54	0.51	0.43	0.33	0.40	0.36	0.33	0.54	0.44	0.07
K	4.2	4.7	3.9	2.5	2.9	2.8	2.5	2.5	4.7	3.4	0.8
Sc	<0.0004	<0.0004	<0.0004	<0.0004	<0.0004	<0.0004	<0.0004	0.0004	0.0004	0.0004	0.0002
Ag	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	0.004	0.004	0.004	0.002
Na	30	35.2	38.3	21.1	22.5	26.8	18.7	18.70	38.30	27.53	6.82
Sr	0.131	0.143	0.149	0.143	0.132	0.149	0.120	0.120	0.149	0.138	0.010
Th	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	0.01	0.00
Tl	<0.002	<0.002	<0.002	0.004	<0.002	<0.002	0.002	0.002	0.004	0.002	0.001
V	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	0.004	0.004	0.004	0.002
Y	0.089	0.057	0.040	0.048	0.039	0.047	0.051	0.039	0.089	0.054	0.019
Zn	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	0.002	0.002	0.002	0.001
Zr											
Grabs:											
CM	0.007	0.005	0.005	0.015	0.008	0.006	<0.002	0.002	0.015	0.007	0.004
Oil	3	<2	2	2	<2	<2	<2	2.0	3.0	2.1	1.1
CL2 (residual)	<0.1	<0.1	<0.1	<0.1	<2	*	<0.1	<0.1	0.1	0.1	0.0

Table G-2. New Hope Pond Toxicity Test of 11/4 - 11/11/86

Dates:	11/04	11/05	11/06	11/07	11/08	11/09	11/10	11/10	Min	Max	Avg	Std Dev
Composites:												
Flow (mgd)	8.48	9.34	8.65	11.33	9.34	11.15	8.82	8.82	8.48	11.33	9.59	1.09
Hg	0.0015	0.0014	0.0009	0.0011	0.0013	0.0000	0.0018	0.0018	0.0009	0.0018	0.0013	0.0003
Se	1.0	1.0	0.85	0.85	0.80	<0.07	0.002	0.002	0.002	1.0	1.0	0.001
F	2.2	2.4	2.6	3.0	3.0	3.2	3.4	3.4	2.2	3.4	2.8	0.4
NN03	95	80	94	90	75	82	80	80	75	95	85	7
S04	<5.0	<5.0	6.0	6.0	<5.0	5.0	6.0	6.0	5.0	6.0	5.4	2.3
ISS	0.017	0.018	0.016	0.015	0.026	0.046	0.028	0.028	0.015	0.046	0.024	0.010
U	5.0	4.8	5.0	5.0	7.0	5.0	4.0	4.0	4.0	7.0	5.1	0.8
TOC	0.002	0.01	0.95	0.006	0.006	0.011	0.007	0.007	0.002	0.950	0.142	0.330
CN	6.9	7.3	8.0	8.1	7.4	8.8	6.5	6.5	6.5	8.8	7.6	0.7
pH (std units)												
Al	0.33	0.16	0.15	0.37	0.44	0.16	0.49	0.49	0.15	0.49	0.30	0.13
As	<0.04	<0.04	<0.04	<0.04	<0.04	<0.04	<0.04	<0.04	0.04	0.04	0.04	0.02
Ba	0.0500	0.0489	0.0342	0.0534	0.0472	0.0547	0.0534	0.0534	0.0472	0.0534	0.0520	0.0030
Be	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.0001	0.0001	0.0001	0.0000
B	0.034	<0.003	0.045	0.038	0.039	0.046	0.046	0.046	0.00	0.05	0.04	0.02
Cd	<0.003	<0.003	<0.003	<0.003	<0.003	<0.003	<0.003	<0.003	0.003	0.003	0.003	0.001
Ce	44.7	44.8	54.7	57.0	47.3	55.4	51.0	51.0	45	57	51	5
Co	<0.006	<0.006	<0.006	<0.006	<0.006	<0.006	<0.006	<0.006	0.006	0.006	0.006	0.001
Cr	<0.002	<0.002	<0.002	<0.002	0.002	<0.002	<0.002	<0.002	0.002	0.002	0.002	0.001
Co	0.007	0.006	0.006	0.010	0.008	0.006	0.008	0.008	0.006	0.010	0.007	0.001
Cu	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	0.01	0.00
Ga	0.78	0.55	0.56	0.92	0.89	0.34	0.83	0.83	0.34	0.93	0.71	0.22
Fe	<0.003	<0.003	<0.003	<0.003	<0.003	<0.003	<0.003	<0.003	0.003	0.003	0.003	0.001
La	<0.02	<0.02	<0.02	<0.02	<0.02	<0.02	<0.02	<0.02	0.02	0.02	0.02	0.01
Pb	0.018	0.017	0.117	0.028	0.081	0.039	0.036	0.036	0.02	0.12	0.05	0.03
Li	11.60	11.20	12.50	14.10	10.80	13.60	11.90	11.90	10.60	14.10	12.24	1.14
Mg	0.068	0.058	0.076	0.086	0.086	0.075	0.105	0.105	0.058	0.105	0.079	0.014
Mn	0.168	0.161	0.147	0.139	0.108	0.113	0.083	0.083	0.08	0.17	0.13	0.03
Mo	0.008	0.009	0.012	0.012	0.011	0.009	0.009	0.009	0.009	0.012	0.010	0.001
Ni	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	0.01	0.00
Nb	0.32	0.36	0.30	0.43	0.33	0.30	0.24	0.24	0.2	0.4	0.3	0.1
P	2.6	2.4	2.8	3.2	2.9	3.2	3.0	3.0	2.4	3.2	2.9	0.3
K	<0.0004	<0.0004	<0.0004	<0.0004	<0.0004	<0.0004	<0.0004	<0.0004	0.0004	0.0004	0.0004	0.0002
Sc	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	0.004	0.004	0.004	0.002
As	25.5	25.1	28.4	32.2	22.3	30.9	23.9	23.9	22	32	27	3
Na	0.140	0.145	0.166	0.166	0.139	0.176	0.158	0.158	0.139	0.176	0.156	0.013
Sr	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	0.01	0.00
Th	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	0.002	0.002	0.002	0.003
Ti	<0.004	0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	0.004	0.004	0.004	0.002
V	0.067	0.042	0.053	0.081	0.066	0.046	0.052	0.052	0.042	0.081	0.058	0.013
Zn	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	0.002	0.002	0.002	0.001
Zr	0.006	0.007	0.007	0.005	0.008	0.008	0.006	0.006	0.005	0.008	0.007	0.001
Grabs:												
CW	<2	<2	<2	<2	4	<2	<2	<2	2	4	2	1
Oil	<0.1	<0.1	<0.1	<0.1	*	*	<0.1	<0.1	0.1	0.1	0.0	0.0
CL2 (residual)	<0.1	<0.1	<0.1	<0.1	*	*	<0.1	<0.1	0.1	0.1	0.0	0.0

Table G-3. Few Hope Pond Toxicity Test of 11/18 - 11/25/86

Composites: Flow (mgd)	Dates:							Avg	Std Dev		
	11/18	11/19	11/20	11/21	11/22	11/23	11/24				
U	8.89	8.82	11.33	9.34	8.65	10.60	13.25	8.65	13.25	10.14	1.57
TOC	0.0029	0.0016	0.013	0.0400	0.0096	0.0043	0.0072	0.0016	0.0400	0.0112	0.0012
CN	0.007	0.006	0.007	0.005	0.010	0.006	0.012	0.005	0.012	0.008	0.002
pH (std units)	7.5	6.8	7.0	7.0	6.7	7.2	7.8	6.7	7.8	7.1	0.4
Al	0.19	0.12	0.33	0.52	0.24	0.25	1.40	0.12	1.40	0.44	0.41
As	<0.04	<0.04	<0.04	<0.04	<0.04	<0.04	<0.04	0.04	0.04	0.04	0.02
Ba	0.0500	0.0462	0.0454	0.0537	0.0454	0.5170	0.0442	0.0442	0.5170	0.1146	0.1843
Be	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.0001	0.0001	0.0001	0.0001	0.0000
B	0.041	0.043	0.038	0.042	0.042	0.045	0.039	0.038	0.045	0.041	0.002
Cd	<0.003	<0.003	<0.003	<0.003	<0.003	<0.003	<0.003	0.003	0.003	0.003	0.001
Ca	46.1	45.7	41.8	46.8	48.8	48.8	33.0	33.0	48.9	44.4	5.2
Co	<0.02	<0.02	<0.02	<0.02	<0.02	<0.02	<0.02	0.02	0.02	0.02	0.01
Cr	<0.006	<0.006	<0.006	<0.006	<0.006	<0.006	<0.006	0.006	0.006	0.006	0.003
Cu	0.005	0.004	0.010	0.013	0.010	0.011	0.012	0.004	0.013	0.008	0.003
Ga	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	0.01	0.00
Fe	0.54	0.44	0.66	1.11	0.69	0.65	1.78	0.44	1.78	0.84	0.43
La	<0.003	<0.003	<0.003	<0.003	<0.003	<0.003	<0.003	0.003	0.003	0.003	0.001
Pb	<0.02	<0.02	<0.02	<0.02	<0.02	<0.02	<0.02	0.02	0.02	0.02	0.01
Li	0.019	0.020	0.018	0.023	0.049	0.023	0.027	0.018	0.049	0.026	0.010
Mg	11.8	11.9	9.9	11.5	11.5	12.8	7.4	7.4	12.9	11.1	1.6
Nb	0.081	0.078	0.368	0.098	0.080	0.075	0.086	0.068	0.099	0.081	0.009
Po	0.122	0.133	0.109	0.090	0.107	0.127	0.047	0.047	0.133	0.105	0.027
Ni	0.011	0.012	0.009	0.010	0.010	0.008	0.012	0.008	0.012	0.010	0.001
Nb	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	0.01	0.00
P	0.37	0.35	0.35	0.48	0.53	0.32	0.33	0.32	0.53	0.39	0.08
K	2.8	3.3	3.0	2.6	3.2	3.3	2.7	2.6	3.3	3.0	6.3
Sc	<0.0004	<0.0004	<0.0004	<0.0004	<0.0004	<0.0004	<0.0004	0.0004	0.0004	0.0004	0.0002
Ag	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	0.004	0.004	0.004	0.002
Na	31.8	34.9	30.6	25.7	29.6	34.1	15.0	15.0	34.9	28.8	6.3
Sr	0.149	0.143	0.132	0.149	0.140	0.156	0.108	0.108	0.156	0.140	0.015
Th	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	0.01	0.00
Ti	0.003	0.002	0.005	0.006	0.003	0.003	0.012	0.002	0.012	0.005	0.003
V	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	0.004	0.004	0.004	0.002
Zn	0.068	0.058	0.062	0.093	0.080	0.087	0.087	0.058	0.093	0.074	0.012
Zr	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	0.002	0.002	0.002	0.001
Grabs:											
CN	0.006	0.005	0.010	0.004	0.009	0.007	0.009	0.004	0.010	0.007	0.002
Oil	<2	<2	<2	<2	<2	<2	<2	2	2	2	1
CL2 (residual)	<0.1	<0.1	<0.1	0.45	*	*	<0.1	0.1	0.5	0.2	0.1

APPENDIX H

LIST OF INORGANIC CONSTITUENTS MEASURED DAILY IN COMPOSITE SAMPLES
COLLECTED FROM THE INLET AND OUTLET OF NEW HOPE POND,
OCTOBER 1 THROUGH DECEMBER 15, 1986

Appendix H

Determination	Determination
<u>Composites</u>	
Flow	pH
Mercury	Selenium
Uranium, total	Ammonia
Biochemical oxygen demand	Chemical oxygen demand
Chloride	Cyanide
Fluoride	Total organic carbon
Filterable residue	Sulfate
Butyl carbitol	Total dissolved solids
Nitrates	Total nitrogen
Kjeldahl nitrogen	
<u>ICP Sweep</u>	
Aluminum	Arsenic
Barium	Beryllium
Boron	Cadmium
Calcium	Cerium
Chromium	Cobalt
Copper	Gallium
Iron	Lanthium
Lead	Lithium
Magnesium	Manganese
Molybdenum	Nickel
Niobium	Phosphorus
Potassium	Scandium
Silver	Sodium
Strontium	Titanium
Vanadium	Zinc
Zirconium	
<u>Grabs</u>	
Cyanide	Oil
Residual chlorine	Mercury
Perchloroethylene	Settleable solids
<u>Field Readings</u>	
Dissolved oxygen	Specific conductivity
Temperature	

APPENDIX I

CONCENTRATIONS OF INORGANIC COMPOUNDS (mg/L) IN COMPOSITE WATER COLLECTED
AT THE OUTFALL OF NEW HOPE POND DURING JUNE 1 TO JULY 2
AND JULY 7 TO JULY 20, 1987

Table I-6

Parameter	Date sample type duration						
	July 6, 1987 composite	July 7, 1987 composite 120 hr	July 8, 1987 composite 24 hr	July 9, 1987 composite 24 hr	July 10, 1987 composite 24 hr	July 11, 1987 composite	July 12, 1987 composite
Mercury		0.0088	0.006	0.0072	0.0069		
Selenium		<0.002	<0.002	<0.002	<0.002		
Uranium-235(x)		0.66	0.52	0.6	0.5		
Uranium, Total		0.02	0.042	0.024	0.019		
Ammonia			2.2	1.4	0.9		
BOD			<5	<5	<5		
COD			8	8.6	8		
Chloride			17	18	18		
Cyanide			0.004	0.003	0.005		
Fluoride			0.7	0.9	1		
Nitrate			4	4.1	3.6		
Nitrogen, Kjeldahl				2			
Nitrogen, Total				6.1			
TOC			3.2	3	4.2		
TDS			270	260	230		
ISS			12	<5	<5		
Sulfate			72	73	46		
Surfactants				<0.05			
Aluminum		3.24	1.43	0.39	0.25		
Arsenic		<0.04	<0.04	<0.04	<0.04		
Barium		0.0563	0.0535	0.0463	0.0438		
Beryllium		<0.0001	<0.0001	<0.0001	<0.0001		
Boron		0.025	0.042	0.028	0.026		
Bromine		<0.003	<0.003	<0.003	<0.003		
Calcium		41.7	52.6	52.3	48.5		
Calcium		<0.02	<0.02	<0.02	<0.02		
Cerium		<0.006	<0.006	<0.006	<0.006		
Chromium		<0.002	<0.002	<0.002	<0.002		
Cobalt		0.008	0.007	0.004	0.006		
Copper		<0.01	<0.01	<0.01	<0.01		
Gallium		2.42	1.31	0.42	0.33		
Iron		<0.003	<0.003	<0.003	<0.003		
Lanthanum		<0.02	<0.02	<0.02	<0.02		
Lead		0.065	0.022	0.018	0.015		
Lithium		8.61	10.6	12.1	11.6		
Magnesium		0.071	0.138	0.119	0.108		
Manganese		0.123	0.086	0.136	0.17		
Molybdenum		<0.007	<0.007	<0.007	<0.007		
Nickel		<0.01	<0.01	<0.01	<0.01		
Niobium		0.3	0.24	0.22	0.17		
Phosphorus		2.7	2.7	2	2		
Potassium		0.0005	<0.0004	<0.0004	<0.0004		
Scandium		<0.004	<0.004	<0.004	<0.004		
Silver		9.86	11	13.5	10.5		
Sodium		0.117	0.15	0.146	0.137		
Strontium		<0.01	<0.01	<0.01	<0.01		
Thorium		0.012	0.009	0.002	0.003		
Titanium		0.006	0.004	<0.004	0.006		
Vanadium		0.042	0.042	0.029	0.024		
Zinc		<0.002	<0.002	<0.002	<0.002		
Zirconium							

APPENDIX J

LIST OF INORGANIC AND ORGANIC CHEMICALS MEASURED DAILY IN COMPOSITE
WATER SAMPLES COLLECTED AT THE OUTFALL OF NEW HOPE POND
DURING 7-d TOXICITY TESTS ON DECEMBER 2-8
AND DECEMBER 9-15, 1986

Appendix J

Determination	Determination
Mercury	Selenium
Uranium, total	Ammonia
Biochemical oxygen demand	Chemical oxygen demand
Chloride	Cyanide
Fluoride	Total organic carbon
Filterable residue	Sulfate
Butyl carbitol	
	<u>ICP Sweep</u>
Aluminum	Arsenic
Barium	Beryllium
Boron	Cadmium
Calcium	Cerium
Chromium	Cobalt
Copper	Gallium
Iron	Lanthium
Lead	Lithium
Magnesium	Manganese
Molybdenum	Nickel
Niobium	Phosphorus
Potassium	Scandium
Silver	Sodium
Strontium	Titanium
Vanadium	Zinc
Zirconium	
	<u>PCBs</u>
PCB (Aroclor-1016)	PCB (Aroclor-1221)
PCB (Aroclor-1232)	PCB (Aroclor-1242)
PCB (Aroclor-1248)	PCB (Aroclor-1254)
PCB (Aroclor-1260)	

Priority Chemicals - Volatile Organics

Benzene	Bromoform
Carbontetrachloride	Chlorodibromomethane
Chloroethane	Chloroform
cis-1,3-Dichloropropylene	Dichlorobromomethane
Ethyl benzene	Methyl bromide
Methyl chloride	Methylene chloride
1,1-Dichloroethane	1,1-Dichloroethylene
1,1,1-Trichloroethane	1,1,2-Trichloroethane

Appendix J. (continued)

Determination	Determination
1,1,2,2-Tetrachloroethane	1,2-Dichlorobenzene
1,2-Dichloroethane	1,2-Dichloropropane
1,3-Dichlorobenzene	1,4-Dichlorobenzene
Tetrachloroethylene (Perk)	Toluene
trans-1,2-Dichloroethylene	trans-1,3-Dichloropropylene
Trichloroethylene	Trichlorofluoromethane
2-Chloroethyl vinyl ether	Vinyl chloride
<u>Priority Chemicals - Base Neutrals</u>	
bis-2-Ethylhexylphthalate	Benzidine
Di-n-butylphthalate	Diethylphthalate
Fluoranthene	Phenanthrene
<u>Priority Chemicals - Acid Extraction</u>	
Phenol	4-Nitrophenol
4,6-Dinitro-o-cresol	p-Chloro-m-cresol
Pentachlorophenol	2-Chlorophenol
2-Nitrophenol	2,4-Dichlorophenol
2,4-Dimethylphenol	2,4-Dinitrophenol
2,4,6-Trichlorophenol	

APPENDIX K

LIST OF ORGANIC CHEMICALS MEASURED IN DAILY COMPOSITE WATER
SAMPLES COLLECTED AT THE OUTFALL OF NEW HOPE POND DURING 7-d
TOXICITY TESTS ON OCTOBER 7-14, NOVEMBER 3-11,
AND NOVEMBER 18-25, 1986

Appendix K

Compounds	Compounds
Dibromomethane	Freon 113
Butyl carbitol	Diacetone alcohol
Chloromethane	Bromomethane
Vinyl chloride	Chloroethane
Methylene chloride	Acetone
Carbon disulfide	1,1-Dichloromethane
1,1-Dichloroethane	trans-1,2-Dichloroethane
Chloroform	1,2-Dichloroethane
2-Butanone	1,1,1-Trichloroethane
Carbon tetrachloride	Vinyl acetate
Bromodichloromethane	1,2-Dichloropropanone
cis-1,3-Dichloropropene	Trichloroethene
Dibromochloromethane	1,1,2-Trichloroethane
Benzene	trans-1,3-Dichloropropene
2-Chloroethylvinylether	Bromoform
4-Methyl-2-pentanone	2-Hexanone
Tetrachloroethene	1,1,2,2-Tetrachloroethane
Toluene	Chlorobenzene
Ethylbenzene	Styrene
Total xylenes	Phenol
bis(2-Chloroethyl)ether	2-Chlorophenol
1,3-Dichlorobenzene	1,4-Dichlorobenzene
Benzyl alcohol	1,2-Dichlorobenzene
2-Methylphenol	bis(2-Chloroisopropyl)ether
4-Methylphenol	n-Nitroso-di-n-propylamine
Hexachloroethane	Nitrobenzene
Isophorone	2-Nitrophenol
2,4-Dimethylphenol	Benzoic acid
bis(2-Chloroethoxy)methane	2,4-Dichlorophenol
1,2,4-Trichlorobenzene	Naphthalene
4-Nitroaniline	Hexachlorobutadiene
4-Chloro-3-methylphenol	2-Methylnaphthalene
Hexachlorocyclopentadiene	2,4,6-Trichlorophenol
2,4,5-Trichlorophenol	2-Chloronaphthalene
2-Nitroaniline	Dimethylphthalate
Acenaphthylene	3-Nitroaniline
Acenaphthene	2,4-Dinitrophenol
4-Nitrophenol	Dibenzofuran
2,4-Dinitrotoluene	2,6-Dinitrotoluene
Diethylphthalate	4-Chlorophenyl-phenylether

Appendix K. (continued)

Compounds	Compounds
Fluorene	4-Nitroaniline
4,6-Dinitro-2-methylphenol	n-Nitrosodiphenylamine
4-Bromophenyl-phenylether	Hexachlorobenzene
Pentachlorophenol	Phenanthrene
Anthracene	Di-n-butylphthalate
Fluoranthrene	Pyrene
Butylbenzylphthalate	3,3'-Dichlorobenzidine
Benzo(a)anthracene	bis(2-Ethylhexyl)phthalate
Chrysene	Di-n-octylphthalate
Benzo(b)fluoranthene	Benzo(k)fluoranthene
Benzo(a)pyrene	Indeno(1,2,3-cd)pyrene
Dibenzo(a,h)anthracene	Benzo(g,h,i)perylene
PCB (Aroclor-1016)	PCB (Aroclor-1221)
PCB (Aroclor-1232)	PCB (Aroclor-1242)
PCB (Aroclor-1248)	PCB (Aroclor-1254)
PCB (Aroclor-1260)	

Appendix L

Compounds	Compounds
Butyl carbitol	PCB, total
<u>Priority Chemicals - Volatile Organics</u>	
Benzene	Bromoform
Carbontetrachloride	Chlorodibromomethane
Chloroethane	Chloroform
cis-1,3-Dichloropropylene	Dichlorobromomethane
Ethyl benzene	Methyl bromide
Methyl chloride	Methylene chloride
1,1-Dichloroethane	1,1-Dichloroethylene
1,1,1-Trichloroethane	1,1,2-Trichloroethane
1,1,2,2-Tetrachloroethane	1,2-Dichlorobenzene
1,2-Dichloroethane	1,2-Dichloropropane
1,3-Dichlorobenzene	1,4-Dichlorobenzene
Tetrachloroethylene (Perk)	Toluene
trans-1,2-Dichloroethylene	trans-1,3-Dichloropropylene
Trichloroethylene	Trichlorofluoromethane
2-Chloroethyl vinyl ether	Vinyl chloride
<u>Priority Chemicals - Base Neutrals</u>	
Acenaphthene	Acenaphthylene
Anthracene	Benzidine
Benzo(a)anthracene	Benzo(a)pyrene
Benzo(ghi)perylene	Benzo(k)fluoranthene
bis(2-Chloroethoxy)methane	
bis(2-Chloroethyl)ether	
bis(2-Chloroisopropyl)ether	
bis-2-Ethylhexylphthalate	
Butyl benzylphthalate	Chrysene
Di-n-butylphthalate	Di-n-octylphthalate
Dibenzo(a,h)anthracene	Diethylphthalate
Dimethylphthalate	Fluoranthene
4-Bromophenyl phenyl ether	Fluorene
4-Chloro-phenyl phenyl ether	Hexachlorobenzene
Hexachlorobutadiene	Hexachlorocyclopentadiene
Hexachloroethane	Indeno(1,2,3-cd)pyrene
Isophorone	
N-Nitrosodi-n-propylamine	N-Nitrosodiphenylamine
N-Nitrosodimethylamine	1,2-Dichlorobenzene
Napthalene	1,3-Dichlorobenzene
1,2,4-Trichlorobenzene	Phenanthrene
1,4-Dichlorobenzene	3,3'-Dichlorobenzidine
Pyrene	

Appendix L. (continued)

Compounds	Compounds
3,4-Benzofluoranthene	2-Chloronaphthalene
2,4-Dinitrotoluene	2,6-Dinitrotoluene
<u>Priority Chemicals - Acid Extraction</u>	
Phenol	4-Nitrophenol
4,6-Dinitro-o-cresol	p-Chloro-m-cresol
Pentachlorophenol	2-Chlorophenol
2-Nitrophenol	2,4-Dichlorophenol
2,4-Dimethylphenol	2,4-Dinitrophenol
2,4,6-Trichlorophenol	

END

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