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Investigation of Entrainment Stresses Using a Power Plant Simulator

Glenn F. Cada
Jean A. Solomon
K. Deva Kumar

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INVESTIGATION OF ENTRAINMENT STRESSES USING A POWER PLANT SIMULATOR

Glenn F. Cada, Jean A. Solomon, and K. Deva Kumar

ENVIRONMENTAL SCIENCES DIVISION

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ABSTRACT

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A Power Plant Simulator (PPS) was constructed at the Oak Ridge National Laboratory to examine the component sources of entrainment mortality. This experimental apparatus circulates temperature-controlled water through a closed loop and has as key components, a pump, a condenser bundle, and vertically adjustable piping. Thus, stresses associated with pump passage, condenser passage, vacuum conditions, and thermal effects can be examined.

Larval bluegill, channel catfish, carp, largemouth bass, and smallmouth bass were exposed to different combinations of pump speed and water temperatures in the PPS. Wide differences among species in their sensitivity to pipe and condenser passage were observed. For most of the species tested, short-term conditional mortalities resulting from the physical stresses of pipe and condenser passage increased with ΔT and/or pumping rate. Pump passage was not a major source of physical damage, and no clear relationship was found between pump efficiency and mortality. Susceptibility to physical stresses associated with entrainment appeared to be inversely related to the size of the entrained organisms.

Delayed mortality frequently occurred among fishes exposed to stresses in the PPS. However, delayed mortality estimates in these experimental groups were significantly greater ($\alpha = 0.05$) than corresponding values in handling control groups in only 15 of 64 comparisons. Like short-term mortalities, relatively higher delayed mortalities were often observed for the smaller species tested.

TABLE OF CONTENTS

	Page
ABSTRACT	v
LIST OF TABLES	ix
LIST OF FIGURES	xiii
1. INTRODUCTION	1
2. DESCRIPTION OF THE POWER PLANT SIMULATOR	4
3. MATERIALS AND METHODS	10
3.1 Experimental Procedure	10
3.2 Data Analysis	16
4. RESULTS	25
4.1 Recovery of Test Fishes	25
4.2 Short-term Mortality	27
4.3 Delayed Mortality	41
5. DISCUSSION	74
6. SUMMARY AND CONCLUSIONS	81
7. LITERATURE CITED	83

LIST OF TABLES

Table		Page
1	Hydraulic parameter comparison between a typical power plant circulating water pump and the Ingersoll-Rand 10 BPM	7
2	Calculated water pressures to which entrained organisms are exposed at various locations in the Power Plant Simulator and at various heights of the top horizontal pipe at a pumping rate of 7.58 m^3/min (91% of maximum), pressures are expressed as kilopascals (kPa)	9
3	Experimental combinations of excess temperature (°C above ambient) and pumping rates used in the 1979 PPS program	13
4	Percent recovery of larval fishes following the 72-h post-test holding period	26
5	Observed short-term (1-h) mortalities and calculated conditional mortalities among carp (*Cyprinus carpio*) larvae exposed to different pumping rates in the Power Plant Simulator	28
6	Observed short-term (1-h) mortalities and calculated conditional mortalities among largemouth bass (*Micropterus salmoides*) larvae exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator	30
7	Observed short-term (1-h) mortalities and calculated conditional mortalities among smallmouth bass (*Micropterus dolomieui*) larvae exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator	32
8	Observed short-term (1-h) mortalities and calculated conditional mortalities among bluegill sunfish (*Lepomis macrochirus*) exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator	33
9	Observed short-term (1-h) mortalities and calculated conditional mortalities among channel catfish (*Ictalurus punctatus*) exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator	35

Table		Page
10	Short-term conditional mortality due to collection stresses ($\hat{m}_3$) for larval fishes exposed to various combinations of T and pumping rate in the Power Plant Simulator	36
11	Short-term conditional mortality due to pipe and condenser passage ($\hat{m}_4$) for larval fishes exposed to various combinations of ΔT and pumping rate in the Power Plant Simulator	37
12	Short-term conditional mortality due to pump passage ($\hat{m}_5$) for larval fishes exposed to various combinations of ΔT and pumping rate in the Power Plant Simulator	38
13	Analysis of variance of observed mortality among fishes exposed to both thermal and physical stresses in the PPS	42
14	Delayed (1-72 h) mortalities among carp (*Cyprinus carpio*) larvae exposed to different pumping rates in the Power Plant Simulator	45
15	Comparisons of delayed mortalities among carp (*Cyprinus carpio*) larvae exposed to stresses in the Power Plant Simulator with delayed mortalities among carp larvae exposed only to handling and natural stresses	47
16	Delayed (1-72 h) mortalities among largemouth bass (*Micropterus salmoides*) larvae exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator	54
17	Comparisons of delayed mortalities among largemouth bass (*Micropterus salmoides*) larvae exposed to stresses in the Power Plant Simulator with delayed mortalities among largemouth bass larvae exposed only to handling and natural stresses	55
18	Delayed (1-72 h) mortalities among smallmouth bass (*Micropterus dolomieui*) larvae exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator	59
19	Comparisons of delayed mortalities among smallmouth bass (*Micropterus dolomieui*) larvae exposed to stresses in the Power Plant Simulator with delayed mortalities among smallmouth bass larvae exposed only to handling and natural stresses	60

Table		Page
20	Delayed (1-72 h) mortality among bluegill sunfish (*Lepomis macrochirus*) exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator	64
21	Comparisons of delayed mortalities among bluegill sunfish (*Lepomis macrochirus*) exposed to stresses in the Power Plant Simulator with delayed mortalities among bluegill sunfish exposed only to handling and natural stresses	65
22	Delayed (1-72 h) mortality among channel catfish (*Ictalurus punctatus*) exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator	71
23	Comparisons of delayed mortalities among channel catfish (*Ictalurus punctatus*) exposed to stresses in the Power Plant Simulator with delayed mortalities among channel catfish exposed only to handling and natural stresses	73

LIST OF FIGURES

Figure		Page
1	Schematic diagram of the ORNL Power Plant Simulator . . .	5
2	Schematic representation of the sources of mortality in the Power Plant Simulator. Refer to text for a description of the experiment types	17
3	Observed cumulative mortalities of carp larvae at various times after exposure to a ΔT of 0°C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types	43
4	Observed cumulative mortalities of carp larvae at various times after exposure to a ΔT of 0°C and a pumping rate of 91% of maximum. Refer to the text for a description of the experiment types	44
5	Observed cumulative mortalities of largemouth bass larvae at various times after exposure to a ΔT of 0°C and a pumping rate of 75% of maximum. Refer to the text for a description of the experiment types	48
6	Observed cumulative mortalities of largemouth bass larvae at various times after exposure to a ΔT of 0°C and a pumping rate of 91% of maximum. Refer to the text for a description of the experiment types	49
7	Observed cumulative mortalities of largemouth bass larvae at various times after exposure to a ΔT of 4°C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types	50
8	Observed cumulative mortalities of largemouth bass larvae at various times after exposure to a ΔT of 10°C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types . .	51
9	Observed cumulative mortalities of largemouth bass larvae at various times after exposure to a ΔT of 10°C and a pumping rate of 91% of maximum. Refer to the text for a description of the experiment types . .	52
10	Observed cumulative mortalities of smallmouth bass larvae at various times after exposure to a ΔT of 4°C and a pumping rate of 91% of maximum. Refer to the text for a description of the experiment types . .	56

Figure		Page
11	Observed cumulative mortalities of smallmouth bass larvae at various times after exposure to a ΔT of 10°C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types	57
12	Observed cumulative mortalities of smallmouth bass larvae at various times after exposure to a ΔT of 10°C and a pumping rate of 91% of maximum. Refer to the text for a description of the experiment types	58
13	Observed cumulative mortalities of bluegill sunfish larvae at various times after exposure to a ΔT of 0°C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types	62
14	Observed cumulative mortalities of bluegill sunfish larvae at various times after exposure to a ΔT of 10°C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types	63
15	Observed cumulative mortalities of channel catfish larvae at various times after exposure to a ΔT of 0°C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types	66
16	Observed cumulative mortalities of channel catfish larvae at various times after exposure to a ΔT of 0°C and a pumping rate of 91% of maximum. Refer to the text for a description of the experiment types	67
17	Observed cumulative mortalities of channel catfish larvae at various times after exposure to a ΔT of 10°C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types	68
18	Observed cumulative mortalities of channel catfish larvae at various times after exposure to a ΔT of 10°C and a pumping rate of 91% of maximum. Refer to the text for a description of the experiment types	69

1. INTRODUCTION

Aquatic organisms that are drawn into the cooling water intake of a power plant and that are small enough to pass through the meshes of its debris screens are subjected to a variety of stresses during passage through the plant. Thermal stresses result from the rapid rise in water temperature (often over 10°C) across the condenser system, while the addition of biocides to control fouling organisms can have toxic effects. Physical stresses to entrained biota arise from a number of sources, including mechanical abrasion in the pump and piping, abrupt pressure changes, and shear forces and buffeting induced by fluid turbulence. A comprehensive review of the biological effects of power plant entrainment is provided in Schubel and Marcy (1978).

The assessment of entrainment mortality resulting from the sum of these stresses at particular power plants can be difficult and equivocal. Differential sampling mortalities at intake and discharge sampling sites will obscure true estimates of entrainment mortality (O'Connor and Schaffer 1977, Cada and Hergenrader 1978, Boreman and Goodyear, in press), and obtaining adequate numbers of a particular species or size class of entrainable organism to ensure statistically valid comparisons can be a problem (Vaughan 1979). Further, the post-entrainment mortalities observed at actual power plants result from the sum of thermal, mechanical, and chemical stresses and their synergisms, and the relative importance of each source is often impossible to determine.

For these reasons, the development of laboratory devices that simulate one or more aspects of power plant cooling water systems has received increasing attention. These research tools are designed to allow quantitative examination of entrainment stresses, either singly or in combination, in a controlled, experimental fashion. Examples of single-factor studies include evaluations of the effects of shear forces (Morgan et al. 1976), hydrostatic pressures (Beck et al. 1975), typical time-excess temperature histories (Schubel 1974, Schubel et al. 1977), and chlorine doses (Latimer et al. 1975). Studies of multiple stresses have involved combinations of excess temperatures and chlorine (Burton et al. 1979), excess temperatures, pressures, and condenser tube passage (Coutant and Kedl 1975, Kedl and Coutant 1976), and excess temperatures, pressures, condenser tube passage, and chlorine doses (Poje et al. 1978, Ginn et al. 1978, O'Connor and Poje 1979).

The ORNL Power Plant Simulator (PPS) was designed to reproduce the internal hydraulics of an open cycle-condenser cooling system (Suffern 1977). In addition to imposing thermal, vacuum, and fluid-induced shear stresses upon entrained organisms, the PPS is unique in that it permits an evaluation of stresses associated with pump passage (cavitation, turbulence, contact with impeller blades). Coutant and Kedl (1975) and Marcy et al. (1978) hypothesized that pumps are the most likely source of physical damage to entrained organisms, and they suggested that future studies pay special attention to pump-induced mortality.

Preliminary biological tests using the PPS were conducted in 1978 (Suffern et al. 1979). The experimental and statistical protocols developed in that document form the basis for subsequent work. This report presents results from the 1979 experimental program, which examined the direct mortality (initial and delayed) of freshwater fish larvae and juveniles passed through the PPS under a variety of pumping rates and excess temperatures.

2. DESCRIPTION OF THE POWER PLANT SIMULATOR

A schematic drawing of the PPS is shown in Fig. 1. This experimental apparatus circulates water through a closed loop and has as key components an 11.3-m^3 reservoir, a pump, a condenser bundle, vertically adjustable piping, and a collection net. Thus, stresses associated with pump passage (cavitation, turbulence, contact with impeller blades), vacuum conditions, thermal effects, and condenser passage can be examined. Experimental organisms can be introduced either ahead of or behind the pump, allowing evaluation of mortality associated with entrainment through the piping and condenser system alone or in combination with pump passage.

The cylindrical reservoir, which contains the collection net as well as the bulk of the water in the PPS, has a diameter of 2.4 m and a height of 2.6 m. It is constructed of stainless steel and is surrounded by thermal control jackets through which either steam or mixtures of hot and cold water can be passed in order to achieve desired experimental temperatures in the circulating water. Thus, the heat load is applied to the PPS reservoir (not the condensers), and water temperatures are the same throughout the closed loop.

The pump is an Ingersoll-Rand Model 10 BPM that was selected for its similarity to circulating water pumps used at actual power plants (Suffern et al. 1979). It has a design-flow capacity of 7.6 m^3/min. Further, it is equipped with a Reeves Vari-Drive, which permits the use of lower pumping rates (lower efficiencies) and the examination of pump-passage stresses associated with the concomitant increases in

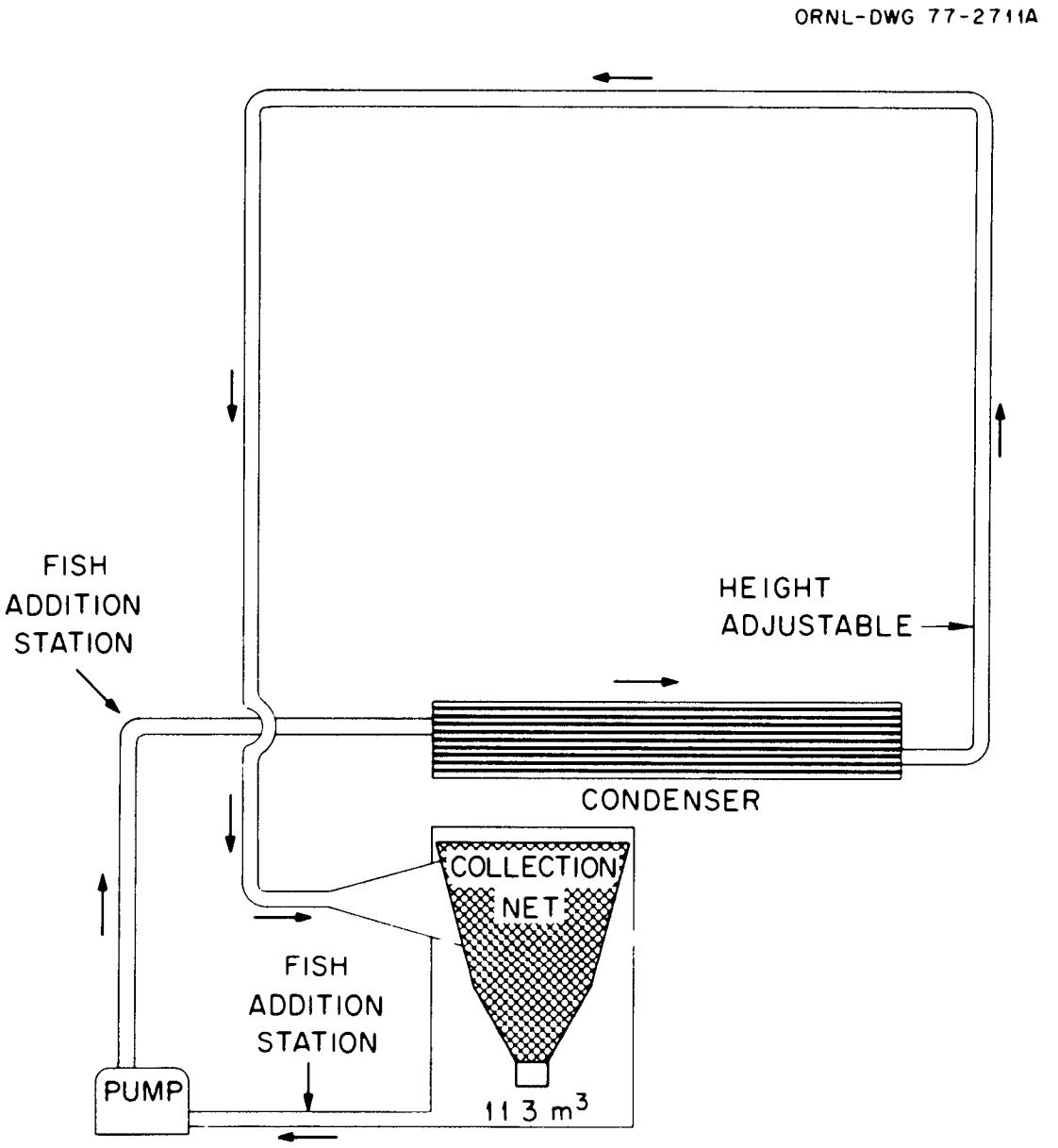

Fig. 1. Schematic diagram of the ORNL Power Plant Simulator.

turbulence. Table 1 is a comparison of important hydraulic parameters of a typical power-plant circulating water pump and the PPS pump. Detailed discussion of these parameters is provided in Suffern et al. (1979).

Aquatic organisms can be introduced into the PPS through addition stations on either side of the pump (Fig. 1). These consist of transparent glass chambers that are connected to the PPS by 6.4-cm diameter valved pipes. Organisms in the pre-pump addition station are introduced into the circulating water flow by simply opening the valves to the chamber and allowing the flow of water from the reservoir to the pump to draw them out of the chamber. These organisms then pass through the pump, condenser system, and eventually into the collection net. Aquatic organisms in the post-pump addition chamber, on the other hand, must be injected into the PPS by means of pressure from an external water line. The water in this chamber is set at the same temperature as the water in the PPS; and by opening appropriate valves, organisms are flushed out of the chamber into the circulating water flow.

The condenser bundle is 9.8 m long and has a diameter of 0.4 m. It contains seventy-three 3.2-cm OD stainless steel tubes with water boxes at each end. At design flow rates of 7.6 m^3/min, estimated water velocities within the condenser tubes average about 2.4 m/s, a value typical of power plant cooling systems (Schubel 1975, Kedl and Coutant 1976).

After passing through the condenser tubes, the circulating flow carries experimental organisms upward and along the top horizontal run of pipe (Fig. 1). The piping in this segment of the PPS is composed of

Table 1. Hydraulic parameter comparison between a typical power-plant circulating water pump and the Ingersoll-Rand 10 BPM, modified from Suffern et al. (1979)

Parameter	Typical circulating water pump (Bull Run Steam Plant)[a]	Ingersoll-Rand 10-BPM
Capacity (m^3/min)	526.1	7.6
Head (m)	6.1	6.7
Tip speed (m/s)	29.0	18.6
NPSH - Required (m)[b]	4.0	4.0
Maximum sphere size (cm)	Large	5.1
Specific speed	9460	5150
Impeller	Mixed-axial	Mixed
Suction	Wet Pit	Flanged Pipe
RPM[c]	235	1170
Diffuser type	Axial diffuser vanes	Volute
Efficiency (%)	90.5	72

[a]The values listed as typical for power plant pumps are those for the pumps at TVA's Bull Run steam plant, and they are comparable to those for eight other TVA plants examined as a basis for this comparison.

[b]Net positive suction head.

[c]Revolutions per minute.

polyvinyl chloride (PVC) and has an outside diameter of 27.4 cm. The height of the horizontal run can be varied to create different pressure conditions by adding spool pieces on the vertical runs. Horizontal-run heights of from 0.0 to 11.3 above the water level of the reservoir can be achieved, with increments as small as 0.9 m. A configuration in which the centerline of the top horizontal run was 5.8 m above the water level of the reservoir was used for all experiments in 1979.

Circulating water was filtered through a 2.0-m diameter, 1.0-m long, conical plankton net composed of 0.5-mm mesh Nitex. The collection net was suspended in the reservoir during operation (Fig. 1), and experimental organisms were retrieved by raising the net out of the water and washing them into a screened collection bucket at the cod end. The PVC collection bucket is 0.31 m long and 0.15 m in diameter.

Water temperatures and pressures at various points in the PPS were monitored during the experiments by means of remote sensors wired to a control panel. Water temperature sensors are located in the top horizontal run, in the descending vertical run, and within the reservoir. Pressure indicators are positioned immediately upstream of the pump (PI-3), upstream of the condenser bundle (PI-9), between the condenser bundle and the ascending vertical run of piping (PI-11), and in the top horizontal run of piping (PI-12). Temperature readings varied less than 0.2 °C among the three sensors, while readings from the four pressure indicators reflected the changing pressure regime at various points in the PPS. Table 2 provides calculated water pressures at these points for various heights of the top horizontal run of piping.

Filtered well water was used in the PPS, pretest stock tanks, and post-test holding aquaria.

Table 2. Calculated water pressures to which entrained organisms are exposed at various locations in the Power Plant Simulator and at various heights of the top horizontal pipe at a pumping rate of 7.58 m^3/min (91% of maximum), pressures expressed as kilopascals (kPa)[a]

Height of top horizontal pipe (m)[c]	Location of pressure gauge[b]			
	PI-3	PI-9	PI-11	PI-12
2.1	12	45	27	- 9
3.3	12	45	27	-21
4.9	12	45	27	-36
5.8	12	45	27	-45
7.3	12	45	27	-60
8.5	12	45	27	-72
10.1	12	45	27	-84

[a]1 atm = 101.3 kPa = 33.9 ft of water = 14.7 psi. Values provided in this table are gauge pressures; i.e., they represent pressures in excess of atmospheric pressure. In order to obtain absolute pressure experienced by entrained organisms at various points in the Power Plant Simulator, add the value for atmospheric pressure (e.g., 101.3 kPa at sea level).

[b]Refer to text for location of pressure gauges.

[c]Height of the centerline of the top horizontal run of pipe (Fig. 1) above the water level in the PPS reservoir. This height was 5.8 m in all 1978 and 1979 experiments.

3. MATERIALS AND METHODS

3.1 Experimental Procedure

The majority of the larval and juvenile fishes examined in the 1979 program were obtained from the Cohutta National Fish Hatchery, Cohutta, Georgia. Species utilized from this source (and their approximate mean total length at the time of testing) were largemouth bass, *Micropterus salmoides* (12 mm), smallmouth bass, *Micropterus dolomieui* (14 mm), bluegill sunfish, *Lepomis macrochirus* (18 mm), and channel catfish, *Ictalurus punctatus* (16-26 mm). Larvae were transferred from the hatchery to the testing facilities at ORNL in large plastic bags, half-filled with hatchery water of the appropriate temperature. After adding the fish, the bags were inflated with oxygen and placed in styrofoam coolers to prevent water temperature fluctuations during the two-hour trip to ORNL.

In addition, carp (*Cyprinus carpio*) larvae were obtained by artificial spawning of adults in the laboratory. Eggs were stripped into saline solution to prevent clumping and were swirled with a few drops of milt. The suspension was immediately poured into a fiberglass holding tank to distribute the eggs uniformly over the bottom. Water temperature was maintained at 24°C and hatching occurred in 3 to 4 days. Carp larvae used in the PPS tests had a mean total length of approximately 7 mm.

All larvae were held in 525-liter rectangular fiberglass tanks prior to testing. The stock tanks were maintained at computer-controlled, constant temperatures by the addition of filtered well

water at a rate of 4 to 5 liters per minute. Holding temperatures varied among species, ranging from 20-22°C for largemouth and smallmouth bass to 24°C for carp, bluegill, and catfish. Illumination was provided by cool white florescent lights controlled by an outside photoswitch to reproduce the seasonal photoperiod. Both pre- and post-test fishes were fed twice a day. Small larvae were fed natural zooplankton collected from an oxidation pond, while larger larvae were fed a mixture of zooplankton and ground trout chow.

Post-test holding facilities consisted of 7-liter, transparent plastic rectangular aquaria fitted onto temperature-controlled water supply manifolds (Suffern et al. 1979). Each aquarium had an independently controlled flow-through rate, and all were connected to a common drain system. Water was maintained in these holding aquaria at the same constant temperatures as the stock tanks. Initially, each group of test fish was placed directly into an individual aquarium. It was found that subsequent post-test observation and enumeration of the unrestricted larvae was difficult and time-consuming. In later tests, fish were confined in 1-liter, cylindrical, brown plastic bottles within the holding aquaria. Large screen panels, composed of 0.5-mm mesh Nitex, allowed water to flow through the bottles. This modification facilitated post-test observations and minimized the handling time required for the removal of dead fish.

The 1979 experimental program consisted of examining the effects of two variables, water temperature and pumping rate, on the post-test survival of each of the five fish species. Water temperatures ranging from ambient holding temperatures to 15°C above ambient were used to

assess the relative effects of thermal stresses on entrained organisms. The relationship of pumping efficiency to entrainment mortality, on the other hand, was examined by varying the pumping rate. The pump used in the PPS operates most efficiently at a rate of 7.58 m^3/min, or 91% of maximum. Lower pumping rates, which result in inefficient pump operation and increased turbulence, were expected to cause greater mortalities among pumped larvae due to increased physical stresses. Accordingly, the effects of pumping rates ranging from 4.16 m^3/min (50% of maximum) to 7.58 m^3/min (91% of maximum) were studied during 1979. The particular combinations of water temperature and pumping rate that each species was exposed to in the PPS are given in Table 3.

In order to separate the effects of condenser tube and pump passage in the PPS from those of handling and natural mortality, three control groups and two treatment groups were examined for each of the combinations listed in Table 3. These five experimental groups, in order of increasing stress, were:

(1) Handling Control (Experiment Type 1): Fish were dipnetted out of the stock tank into a transfer aquarium. The contents of the transfer aquarium were then poured into the appropriate post-test holding aquarium. This control group was used to determine the mortality due to handling effects and natural causes.

(2) Thermal Control (Experiment Type 2): Fish were dipnetted out of the stock tank into a transfer aquarium. Larvae in the transfer aquarium were poured into a screened bottle and exposed to the water in the PPS reservoir for a period of time equivalent to the time necessary to pass an organism through the PPS and retrieve it from the collection net (350-400 seconds). The test fish were

Table 3. Experimental combinations of excess temperature (°C above ambient) and pumping rates used in the 1979 PPS Program

Species	ΔT (°C)	Pumping rate		Number of replicates
		Percent of maximum	m^3/min	
Largemouth bass	0	91	7.58	2
	0	75	6.25	2
	4	50	4.16	1
	10	50	4.16	1
	10	91	7.58	1
Smallmouth bass	4	91	7.58	1
	10	91	7.58	2
	10	50	4.16	1
Carp	0	50	4.16	2
	0	91	7.58	1
Bluegill sunfish	0	50	4.16	2
	10	50	4.16	3
	15	50	4.16	1
Channel catfish	0	50	4.16	6
	0	91	7.58	2
	10	50	4.16	6
	10	91	7.58	2
	15	50	4.16	1

then placed in a post-test holding aquarium. This control group was used to assess additional mortality due to thermal shock (if any) and exposure to the water circulating within the closed system of the PPS.

(3) Collection Control (Experiment Type 3): Fish were dipnetted out of the stock tank into a transfer aquarium. The PPS pump was shut off, and fish were poured into the collection net directly in front of the diffuser. After 15 seconds, the pump was turned on for 60 seconds. After the pump was turned off again, the net was slowly raised out of the water and the contents were washed off the sides by means of a gentle spray of PPS water applied to the outside of the net with a submersible pump and hose. Larvae were retrieved from the collection bucket at the bottom of the net and transferred to a holding aquarium. This control group was used to estimate mortality due to the collection process.

(4) Condenser Passage (Experiment Type 4): Larvae were dipnetted out of the stock tank into a transfer aquarium. Depending on their size, fish were either poured directly or transferred by means of a large, glass syringe into the post-pump addition station. The larvae were then injected into the circulating water flow downstream of the pump, such that they passed through the condenser bundles and elevated return piping, but not the pump. After 90 seconds, the pump was shut off and larvae were recovered in the same manner as collection controls. This treatment group, when corrected for control mortalities, was designed to estimate mortality due to passage through the PPS piping and condenser tube system.

(5) Pump Passage (Experiment Type 5): In this test group, fish larvae were treated in the same manner as the condenser passage group, with the exception that they were introduced into the circulating water flow through an addition station located upstream of the pump. This treatment group, when corrected for control mortalities, permits estimates of mortality due to passage through the entire PPS, as well as evaluation of the effects of pump passage alone when corrected for condenser passage mortality.

Approximately 50 fish larvae were utilized per replicate of each experimental type. An entire series of tests, i.e., three control groups and two treatment groups, could be completed in one hour, and in the majority of cases experimental combinations of temperature and pump speed were replicated at least once. Both the sequence of tests within a given series and the post-test holding aquaria were randomly assigned. Based on an analysis of variance, differences in mortality among test fishes attributable to the effects of different holding aquaria were not significant ($P = 0.05$ level).

All fish were examined at intervals of 1, 24, 48, and 72 hours after testing. At each observation period, fish were classified as either alive (swimming normally), stunned (abnormal behavior, swimming erratically), or dead (no response to probing). Dead fish were removed at each observation period and preserved in 10 percent formalin solution. Larvae that were alive at the end of the 72-hour, post-test holding period were also preserved.

Measurements of total body length were made on fish in each experimental group by means of a grid attached to the base of a

binocular stereoscopic microscope. All preserved fish from a given observation period were recounted, and if the number was less than 25, all were measured. If the number of larvae was greater than 25, the sample was halved with a Folsom plankton splitter (McEwen et al. 1954) and all of the larvae in one of the subsamples was measured.

3.2. Data Analysis

Mortality has been partitioned into five components, corresponding to the five types of experiments (labeled Experiment Types 1 through 5 on pp. 11-14 and in Fig. 2) performed in the 1979 experimental program. The effects of each of these components or sources of mortality (i.e., handling and natural causes, thermal stresses, collection, condenser passage, and pump passage) may be further partitioned into short-term and delayed mortality. Mortality observed during the first hour after exposure i.e., during time interval (0-1 h) is termed short-term mortality, while mortality observed during the time interval (1-72 h) is termed delayed mortality. A description of methods for determining conditional short-term and delayed mortalities and their variances, for each experiment type, is provided in the following paragraphs. A conditional mortality rate (m) (or just conditional mortality) due to a given source of mortality is defined as the fraction of fish dying due to that source of mortality in the absence of any other sources of mortality, and it is equal to 1.0 minus the probability of surviving (S) that source of mortality (Ricker 1975), i.e., $m = 1 - S$.

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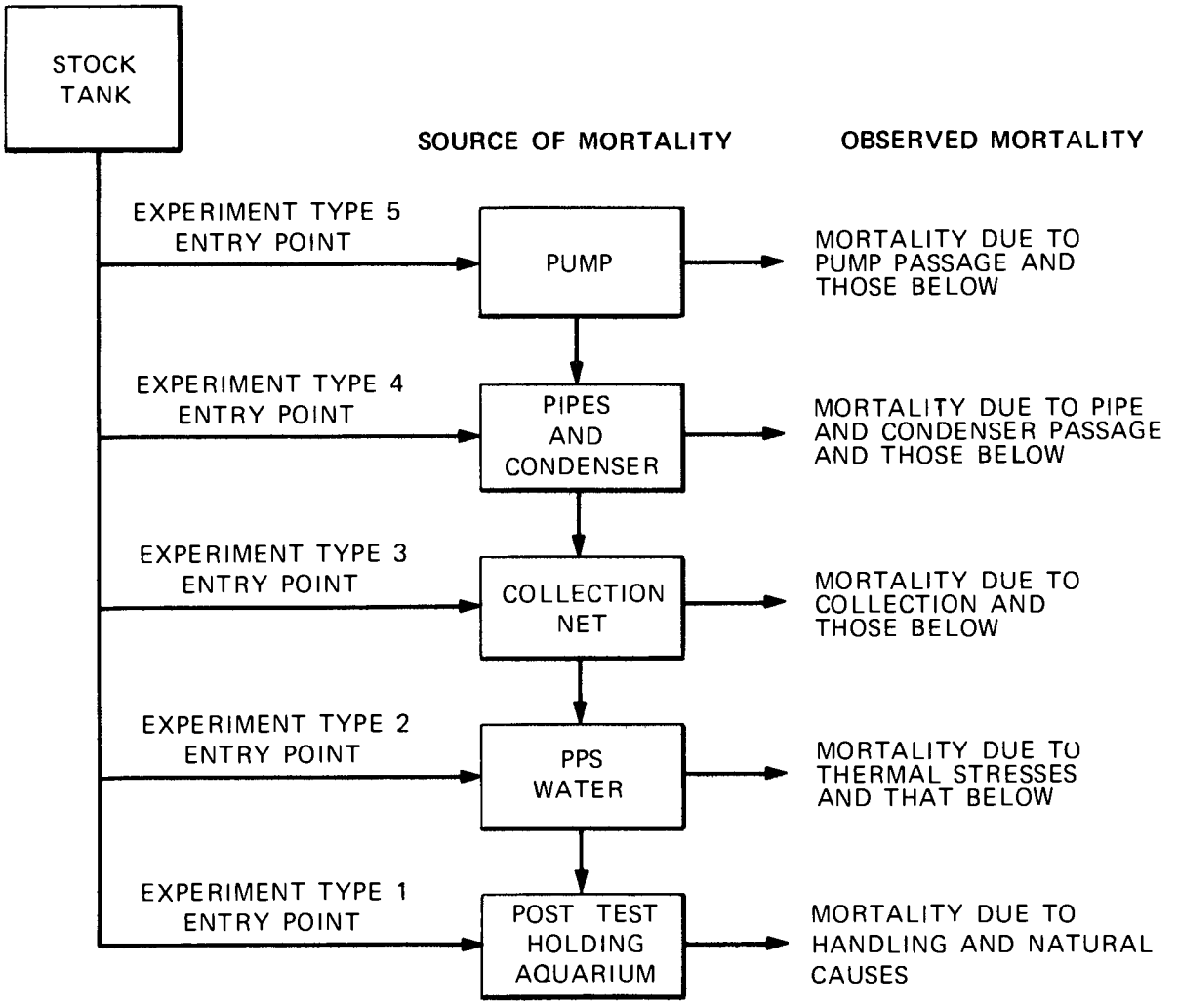

Fig. 2. Schematic representation of the sources of mortality in the Power Plant Simulator. Refer to text for a description of the experiment types.

Short-Term Mortality

Experiment Type 1 (Handling Control)

The estimate of conditional mortality due to natural causes and handling stresses (m_1) is given by

$$\hat{m}_1 = \frac{n_1}{N_1} = P_1 \quad , \tag{1}$$

where P_1 = observed proportion of fish that died due to natural causes and handling,

n_1 = number of fish in Experiment Type 1 dead at 1 hour, and

N_1 = total number of fish in Experiment Type 1.

Experiment Type 2 (Thermal Control)

If n_2 fish were observed dead at 1 hour, then the total short-term mortality due to natural causes, handling stresses, andthermal shock combined (P_2) is given by

$$P_2 = \frac{n_2}{N_2} \quad , \tag{2}$$

where n_2 = number of fish in Experiment Type 2 dead at 1 hour.

N_2 = total number of fish in Experiment Type 2.

Assuming that the probability of surviving natural causes of mortality and of handling stresses combined (S_1) is independent of the probability of surviving thermal shock (S_2), then the probability of surviving both sources of mortality, whether they occur sequentially or simultaneously, is simply

$$S_1 S_2 = 1 - P_2 \quad ,$$

from which

$$S_2 = \frac{1 - P_2}{S_1} \quad .$$

Since by definition, $S_1 = 1 - m_1$ and $S_2 = 1 - m_2$,

$$m_2 = \frac{P_2 - m_1}{1 - m_1} \quad . \tag{3}$$

We obtain an estimate of m_1 ($\hat{m}_1$) from Experiment Type 1 and P_2 is obtained from Experiment Type 2. Equation (3) can then be used to obtain an estimate of m_2, i.e.,

$$\hat{m}_2 = \frac{P_2 - \hat{m}_1}{1 - \hat{m}_1} = \frac{P_2 - P_1}{1 - P_1} \quad . \tag{4}$$

Experiment Type 3 (Collection Control)

Using the same arguments previously advanced, we estimate m_3, the conditional mortality due to the collection process, as

$$\hat{m}_3 = \frac{P_3 - P_2}{1 - P_2} \quad , \tag{5}$$

where $P_2 = \frac{n_2}{N_2}$ = observed proportion of fish that died due to natural causes, handling, and thermal stress,

and $P_3 = \frac{n_3}{N_3}$ = observed proportion of fish that died due to natural causes, handling, thermal stress, and the collection process.

P_2 is observed from Experiment Type 2 and P_3 from Experiment Type 3.

Experiment Type 4 (Condenser Passage)

The estimate of conditional mortality due to pipe and condenser passage (m_4) is given by

$$\hat{m}_4 = \frac{P_4 - P_3}{1 - P_3} \quad , \tag{6}$$

where $P_3 = \frac{n_3}{N_3}$ = observed proportion of fish that died in Experiment Type 3,

and $P_4 = \frac{n_4}{N_4}$ = observed proportion of fish that died in Experiment Type 4.

Experiment Type 5 (Pump Passage)

The estimate of conditional mortality due to pump passage (m_5) is given by

$$\hat{m}_5 = \frac{P_5 - P_4}{1 - P_4} \quad , \tag{7}$$

where $P_4 = \frac{n_4}{N_4}$ = observed proportion of fish that died in Experiment Type 4,

and $P_5 = \frac{n_5}{N_5}$ = observed proportion of fish that died in Experiment Type 5.

Estimates of Variances

The variance of $\hat{m}_1$ is the usual variance of a binomial variable, namely,

$$V(\hat{m}_1) = \frac{\hat{m}_1 \ (1-\hat{m}_1)}{N_1} \quad . \tag{8a}$$

The variances of $\hat{m}_2$, $\hat{m}_3$, $\hat{m}_4$, and $\hat{m}_5$ all have a similar form (Fleiss 1973)

$$V(\hat{m}_2) = \left(\frac{1}{1-\hat{m}_1}\right)^2 \left(\frac{P_2(1-P_2)}{N_2} + (1-\hat{m}_2)^2 \frac{\hat{m}_1(1-\hat{m}_1)}{N_1}\right) \tag{8b}$$

$$V(\hat{m}_3) = \left(\frac{1}{1-P_2}\right)^2 \left(\frac{P_3(1-P_3)}{N_3} + (1-\hat{m}_3)^2 \frac{P_2(1-P_2)}{N_2}\right) \tag{8c}$$

$$V(\hat{m}_4) = \left(\frac{1}{1-P_3}\right)^2 \left(\frac{P_4(1-P_4)}{N_4} + (1-\hat{m}_4)^2 \frac{P_3(1-P_3)}{N_3}\right) \tag{8d}$$

$$V(\hat{m}_5) = \left(\frac{1}{1-P_4}\right)^2 \left(\frac{P_5(1-P_5)}{N_5} + (1-\hat{m}_5)^2 \frac{P_4(1-P_4)}{N_4}\right) \tag{8e}$$

these estimates, $V(\hat{m}_1)$, were used in a normal approximation to the binomial distribution, in calculating the probability that the observed $\hat{m}_1$ is, in fact, 0.

Delayed Mortality

As stated before, delayed mortality is defined as the mortality that occurs during the time interval (1-72 h). If d_1 is the delayed

mortality for Experiment Type i (when i equals Experiment Type 1, 2, 3, 4, or 5), then the estimate of d_1 is given by

$$\hat{d}_1 = \frac{\text{number of fish that died in Experiment Type i in the interval (1-72 h)}}{\text{number of fish alive after 1 hour for Experiment Type i}}$$

$$= \frac{a}{b} \qquad (9)$$

It is assumed that d_1 is not a function of the short-term mortality. $\hat{d}_1$ is an unbiased estimator of the d_1, the "time" delayed mortality.

The variance of d_1 is

$$V(\hat{d}_1) = E[V(\hat{d}_1|b)] + V\left[E[\hat{d}_1|b]\right] \quad , \qquad (10)$$

where $V(\hat{d}_1\ b)$ = conditional variance of $\hat{d}_1$, given b,

$E[\cdot]$ = expectation symbol,

$E[d_1|b]$ = conditional expectation of $\hat{d}_1$, given b, and

b = number of fish alive after 1 hour for Experiment Type i.

The second term on the right hand side of (10) is zero, since

$$E[\hat{d}_1|b] = \frac{b\ d_1}{b} = d_1 .$$

Since d_1 is a constant, $V(d_1) = 0$. Therefore,

$$V(\hat{d}_1) = E[V(\hat{d}_1|b)$$

$$= E\left[\frac{\hat{d}_1(1-\hat{d}_1)}{b}\right]$$

$$= \hat{d}_1(1-\hat{d}_1)\ E[\tfrac{1}{b}] \quad .$$

From Johnson and Kotz (1969), we obtain

$$E\left[\frac{1}{b}\right] \doteq \frac{\left(1-\frac{2}{N_1}\right)}{\left[N_1(1-P_1)-P_1\right]} = \frac{1}{n_1} \quad ,$$

where N_1 = total number of fish in Experiment Type 1 at time 0, and P_1 = proportion of the fish that died in the interval (0-1 h) in Experiment Type 1.

Therefore,

$$V(\hat{d}_1) \doteq \frac{d_1(1-d_1)}{n_1} \quad .$$

The estimate of $V(\hat{d}_1)$ is

$$V(\hat{d}_1) \doteq \frac{\hat{d}_1(1-\hat{d}_1)}{n_1} \quad .$$

Note that as $N1_1$ becomes large, $(1 - \frac{2}{N_1})$ approaches 1, and $[N_1(1-P_1)-P_1]$ approaches $N_1(1-P_1)$. Then n_1 becomes the expected number of fish that would survive the short-term mortality. If there are no residual effects due to exposure to experimental conditions in the PPS, the observed delayed mortality should be equal to natural long-term mortality.

The significance of differences between the delayed mortalities among larvae in test groups exposed to stresses in the PPS and delayed mortalities among larvae in the handling control groups, i.e.,

$\hat{d}_1 - \hat{d}_1$ where $1 = 2, 3, 4, 5$, was tested by means of a two-sided t test. The variance of $\hat{d}_1 - \hat{d}_1$ was estimated as the sum of the variances of the separate delayed mortalities:

$$V(\hat{d}_1 - \hat{d}_1) = V(\hat{d}_1) + V(\hat{d}_1) \quad . \tag{11}$$

4. RESULTS

4.1. Recovery of Test Fishes from the PPS System

A basic consideration when evaluating the PPS and associated post-test holding facilities is the recovery of experimental organisms from the system, i.e., the percent of the total number of fish used in a given experiment that is recovered at the end of the 72-h holding period. Recovery may not be 100 percent for a number of reasons. Counting errors could occur when obtaining fish from the stock tank prior to testing; these errors would result in recoveries either less than or greater than 100 percent. Losses of larval fish within the PPS or from the post-test holding aquaria would result in recoveries of less than 100 percent. Finally, rapid decomposition of small larvae (carp and largemouth bass) that die during the 24-h interval between observation periods may render the larvae unrecognizable, and thus they would not be counted and removed during subsequent observations.

Table 4 shows the percent recovery of fish larvae from the PPS system. These values were calculated by dividing the total number of fish recovered at the end of the experiment (sum of the number dead at each observation period and the number alive 72 h after testing) by the number counted at the beginning of the experiment, generally 50. In 15 of the 25 combinations of species and experimental type, recovery was over 95 percent, and in only one case was it less than 80 percent (Table 4). The low value for condenser-passed carp was due to a single replicate in which only 14 of 50 fish were recovered. The reason for this low recovery is unknown; recovery of other replicates of this

Table 4. Percent recovery of larval fishes following the 72-h, post-test holding period for the five types of experiments

Species	Experiment Type					Weighted mean
	Handling control	Thermal control	Collection control	Condenser passage	Pump passage	
Carp	97.3	102.0	89.3	64.7	88.7	88.4
Channel catfish	99.8	100.0	99.3	100.7	97.3	99.4
Bluegill sunfish	101.0	99.7	99.3	98.7	101.7	100.1
Smallmouth bass	96.5	82.9	96.0	98.0	93.5	93.4
Largemouth bass	85.6	82.9	81.9	85.7	86.8	84.6
Weighted mean	96.8	95.0	94.9	94.3	94.9	

experiment averaged 85 percent. The largest species tested (bluegill sunfish, channel catfish) had the highest recovery percentages, while the smallest larvae (carp, largemouth bass) had the lowest recovery percentages.

A two-way analysis of variance was used to detect significant differences among recovery percentages. It was determined that differences in recovery between experiment types were not significant, while differences between species were significant (α = 0.05). A Duncan's multiple range test indicated that recovery percentages were significantly greater (α = 0.05) for bluegill sunfish and channel catfish than for the other three species.

The calculations of short-term and delayed mortalities in the following sections are based on ratios of the number dead at a particular observation time (hour 1 or 72) to the total number starting the experiment (at hour 0 or 1). Since recoveries of test fishes were often not 100 percent, it was assumed that any fish unaccounted for at the end of the experiment were subject to the same mortality rates as those that were recovered.

4.2. Short-Term Mortality

The observed short-term (1-h) total mortalities (P_1) and estimated conditional mortalities ($\hat{m}_1$) among carp larvae tested in the PPS are given in Table 5. Collection stress was found to be an important factor for this species, as evidenced by estimated conditional mortalities for the collection controls ($\hat{m}_3$) of 0.12 and 0.73. Estimated conditional mortalities due to pipe and condenser passage

Table 5 Observed short-term (1-h) mortalities and calculated conditional mortalities among carp (*Cyprinus carpio*) larvae exposed to different pumping rates in the Power Plant Simulator

Experiment type (or source of stress)	ΔT (°C)	Pumping rate (% of maximum)	Number recovered	Observed mortality[a] P_1	Conditional mortality[b] $\hat{m}_1$	Standard[c] deviation	Critical[d] probability
Handling control	0	-	100	0 00	0 00	0 00	-
Thermal control	0	50	102	0 00	0 00	0.00	-
Collection control	0	50	85	0 12	0 12	0.03	< 0.01
Condenser passage	0	50	51	0.25	0 16	0.08	0.02
Pump passage	0	50	85	0 13	e	e	e
Handling control	0	-	46	0.00	0 00	0.00	-
Thermal control	0	91	51	0.00	0 00	0.00	-
Collection control	0	91	48	0.73	0.73	0.06	< 0 01
Condenser passage	0	91	46	0 96	0.84	0 12	< 0.01
Pump passage	0	91	48	0 73	e	e	e

[a] P_1 = Total short-term mortality observed among fish in Experiment Type 1 (see Materials and Methods)

[b] $\hat{m}_1$ = Conditional mortality due to the ith source of stress (see Materials and Methods)

[c] Standard deviation of $\hat{m}_1$

[d] The probability of observing the given value of $\hat{m}_1$ or greater if, in fact, the true m_1 is equal to zero

[e] The estimated conditional mortality is negative, indicating that a feasible estimate is not obtainable due to high background mortality.

($\hat{m}_4$) were also high (0.16 and 0.84 at pumping rates of 50% and 91% of maximum, respectively). All $\hat{m}_i$'s for the collection control and condenser-passage experimental groups were significantly greater than zero at the $\alpha = 0.05$ level (Table 5).

The significance of conditional mortalities due to pump passage could not be determined from our experimental data at the pumping rates examined (Table 5). Calculated $\hat{m}_5$ values for carp larvae in the pump-passage experimental group were negative because the observed short-term mortalities (P_5) for these fish were lower than the respective values of the condenser-passage group (P_4). When the background mortality (P_i) is greater than the combined mortality due to background and the stress under consideration (P_{i+1}), the estimated conditional mortality ($\hat{m}_{i+1}$) will be negative; see equations (4), (5), (6), and (7). We are unable to estimate the conditional mortality when this occurs.

Table 6 provides short-term, conditional mortality estimates for largemouth bass larvae exposed to various combinations of temperatures and pumping rates. Short-term mortalities resulting from thermal stress ($\hat{m}_2$) ranged from 0.00 at a ΔT of 4°C to a maximum of 0.04 at a ΔT of 10°C, and were not significantly different from zero at the $\alpha = 0.05$ level. Like carp, conditional mortalities among largemouth bass larvae due to collection stresses ($\hat{m}_3$) were variable. Values of $\hat{m}_3$ ranged from 0.00 to 0.31 and were significantly greater than zero in three of the five experimental regimes examined (Table 6). There was no consistent relationship between $\hat{m}_3$ and pumping rate evident from these data, but at a given pumping rate the conditional mortality

Table 6 Observed short-term (1-h) mortalities and calculated conditional mortalities among largemouth bass (*Micropterus salmoides*) larvae exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator

Experiment type (or source of stress)	ΔT (°C)	Pumping rate (% of maximum)	Number recovered	Observed mortality[a] P_1	Conditional mortality[b] $\hat{m}_1$	Standard[c] deviation	Critical[d] probability
Handling control	0	-	99	0.00	0 00	0.00	-
Thermal control	0	75	96	0 00	0.00	0 00	-
Collection control	0	75	95	0 00	0 00	0 00	-
Condenser passage	0	75	102	< 0.01	< 0.01	0.01	0.16
Pump passage	0	75	91	0 11	0 10	0 03	< 0.01
Handling control	0	-	92	0 01	0 01	0 01	0 16
Thermal control	0	91	93	0.02	0 01	0 02	0.28
Collection control	0	91	94	0.21	0.20	0.05	< 0.01
Condenser passage	0	91	85	0 22	0.01	0.08	0 43
Pump passage	0	91	94	0.26	0.04	0 08	0 31
Handling control	0	-	47	0.00	0.00	0 00	-
Thermal control	4	50	42	0.00	0 00	0.00	-
Collection control	4	50	40	0.00	0.00	0 00	-
Condenser passage	4	50	45	0.00	0.00	0.00	-
Pump passage	4	50	49	0.02	0 02	0 02	0.16
Handling control	0	-	23	0 00	0 00	0.00	-
Thermal control	10	50	26	0.04	0 04	0 04	0.15
Collection control	10	50	24	0.33	0.31	0.10	< 0.01
Condenser passage	10	50	30	0.77	0.65	0.13	< 0 01
Pump passage	10	50	33	0.48	e	e	e
Handling control	0	-	37	0.03	0.03	0.03	0.16
Thermal control	10	91	33	0.00	e	e	e
Collection control	10	91	33	0.24	0.24	0 08	< 0.01
Condenser passage	10	91	38	1.00	1.00	0.00	< 0 01
Pump passage	10	91	35	0.94	-	-	-

[a] P_1 = Total short-term mortality observed among fish in Experiment Type 1 (see Materials and Methods)

[b] $\hat{m}_1$ = Conditional mortality due to the 1th source of stress (see Materials and Methods).

[c] Standard deviation of $\hat{m}_1$

[d] The probability of observing the given value of $\hat{m}_1$ or greater if, in fact, the true m_1 is equal to zero.

[e] The estimated conditional mortality is negative, indicating that a feasible estimate is not obtainable due to high background mortality

due to collection stresses appeared to be directly related to temperature.

Estimated short-term conditional mortalities of largemouth bass due to pipe and condenser passage ($\hat{m}_4$) ranged from 0.00 to 1.00, and were statistically significant (α = 0.05) only at a ΔT of 10°C (Table 6). Conditional mortalities due to pump passage ($\hat{m}_5$) were significant only under conditions of 0° ΔT and 75% of maximum pumping rate. Observed high mortalities in the pump-passage experimental group at a ΔT of 10°C were not translated into significant estimated conditional mortalities because of high mortalities in the condenser-passage group under these conditions.

Smallmouth bass larvae were tested at ΔT's of 4° and 10°C and pumping rates of 50 and 91% of maximum. Observed short-term mortalities (P_1) did not exceed 0.06 (Table 7), and estimated short-term conditional mortalities were significantly greater than zero (α = 0.05) only once. Unlike carp and largemouth bass, short-term conditional mortalities due to collection stresses ($\hat{m}_3$) and pipe and condenser passage ($\hat{m}_4$) were low under all conditions examined.

Bluegill sunfish larvae were exposed to ΔT's of 0, 10, and 15°C in the PPS at a single pumping rate, 50% of maximum (Table 8). The estimate of conditional mortality due to thermal stresses ($\hat{m}_2$) was statistically significant (α = 0.05) at a ΔT of 10°C, and complete mortality resulted from exposure of the thermal control group to water temperatures in the PPS of 15°C above ambient. Estimated conditional mortalities due to collection stresses, pipe and condenser passage, and pump passage increased with an increase in ΔT from 0° to 10°C, and all were significantly greater than zero (α = 0.05) at a ΔT of 10°C.

Table 7 Observed short-term (1-h) mortalities and calculated conditional mortalities among smallmouth bass (*Micropterus dolomieui*) larvae exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator

Experiment type (or source of stress)	ΔT (°C)	Pumping rate (% of maximum)	Number recovered	Observed mortality[a] P_1	Conditional mortality[b] $\hat{m}_1$	Standard[c] deviation	Critical[d] probability
Handling control	0	-	48	0 02	0 02	0 02	0 16
Thermal control	4	91	50	0.00	e	e	e
Collection control	4	91	50	0 00	0.00	0 00	-
Condenser passage	4	91	49	0 02	0 02	0 02	0.16
Pump passage	4	91	53	0.06	0 04	0.04	0 17
Handling control	0	-	49	0 00	0 00	0 00	0 50
Thermal control	10	50	47	0 00	0 00	0 00	0 50
Collection control	10	50	49	0 00	0 00	0 00	0.50
Condenser passage	10	50	49	0 00	0 00	0 00	0.50
Pump passage	10	50	48	0 00	0 00	0.00	0 50
Handling control	0	-	96	0.00	0 00	0 00	0 50
Thermal control	10	91	43	0.02	0 02	0 01	< 0 01
Collection control	10	91	93	0 01	e	e	e
Condenser passage	10	91	96	0 01	e	e	e
Pump passage	10	91	48	0.02	0.01	0 02	0 33

[a] P_1 = Total short-term mortality observed among fish in Experiment Type 1 (see Materials and Methods).

[b] $\hat{m}_1$ = Conditional mortality due to the 1th source of stress (see Materials and Methods).

[c] Standard deviation of $\hat{m}_1$.

[d] The probability of observing the given value of $\hat{m}_1$ or greater if, in fact, the true m_1 is equal to zero.

[e] The estimated conditional mortality is negative, indicating that a feasible estimate is not obtainable due to high background mortality

Table 8 Observed short-term (1-h) mortalities and calculated conditional mortalities among bluegill sunfish (Lepomis macrochirus) exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator

Experiment type (or source of stress)	ΔT (°C)	Pumping rate (% of maximum)	Number recovered	Observed mortality[a] P_1	Conditional mortality[b] $\hat{m}_1$	Standard[c] deviation	Critical[d] probability
Handling control	0	-	103	< 0.01	< 0 01	0.01	0.15
Thermal control	0	50	98	0.01	< 0.01	0.01	0.49
Collection control	0	50	101	0.02	< 0 01	0.02	0 29
Condenser passage	0	50	95	0.04	0 02	0.02	0 18
Pump passage	0	50	101	0.07	0 03	0 03	0.20
Handling control	0	-	150	0.00	0 00	0 00	-
Thermal control	10	50	150	0.03	0 03	0.01	0 01
Collection control	10	50	149	0 13	0.10	0.03	< 0.01
Condenser passage	10	50	151	0 20	0 08	0.05	0.04
Pump passage	10	50	152	0.38	0 22	0.06	< 0 01
Handling control	0	-	50	0 00	0.00	0.00	-
Thermal control	15	50	50	1 00	1.00	0.00	0 01
Collection control	15	50	46	1 00	-	-	-
Condenser passage	15	50	49	1 00	-	-	-
Pump passage	15	50	51	1 00	-	-	-

[a] P_1 = Total short-term mortality observed among fish in Experiment Type 1 (see Materials and Methods)

[b] $\hat{m}_1$ = Conditional mortality due to the ith source of stress (see Materials and Methods)

[c] Standard deviation of $\hat{m}_1$

[d] The probability of observing the given value of $\hat{m}_1$ or greater if, in fact, the true m_1 is equal to zero

Channel catfish larvae exhibited relatively low short-term mortalities under most conditions tested (Table 9). Estimates of conditional mortalities due to thermal stresses ($\hat{m}_2$) were 0.00 at ΔT's of 0° and 10°C, but, like bluegill, complete mortality resulted from exposure to 15°C ΔT water. Short-term conditional mortality due to collection stresses was not an important factor; the highest estimated $\hat{m}_3$ in these tests was < 0.01.

At ΔT's of 0° and 10°C, estimated conditional mortalities resulting from pipe and condenser passage ($\hat{m}_4$) ranged from 0.00 to 0.02, while $\hat{m}_5$'s ranged from 0.00 to 0.09 (Table 9). At a given temperature, estimated pump passage conditional mortalities were directly related to pumping rate in these tests. On the other hand, no consistent relationships were apparent between pumping rate and the short-term mortalities of channel catfish larvae arising from either collection stresses or pipe and condenser passage.

Table 10 through 12 compare estimated short-term conditional mortalities among different fish species tested under the same conditions of ΔT and pumping rate in the PPS. Mortalities due to collection stresses (Table 10) were relatively high for carp and largemouth bass and uniformly low (< 0.01) for small mouth bass and channel catfish. These data showed no indication of a consistent relationship between pumping rate and $\hat{m}_3$ at a given temperature, but the magnitude of the estimated short-term conditional mortality due to collection stresses appeared to be directly related to ΔT for bluegill sunfish at 50% and largemouth bass at 91%.

Table 9 Observed short-term (1-h) mortalities and calculated conditional mortalities among channel catfish (*Ictalurus punctatus*) exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator

Experiment type (or source of stress)	ΔT (°C)	Pumping rate (% of maximum)	Number recovered	Observed mortality[a] P_1	Conditional mortality[b] $\hat{m}_1$	Standard[c] deviation	Critical[d] probability
Handling control	0	-	249	0.00	0.00	0.00	-
Thermal control	0	50	300	0 00	0.00	0.00	-
Collection control	0	50	301	< 0.01	< 0 01	< 0.01	0.04
Condenser passage	0	50	245	0 01	< 0.01	< 0.01	0 40
Pump passage	0	50	291	0 04	0 03	0.01	0.03
Handling control	0	-	100	0 00	0 00	0 00	-
Thermal control	0	91	100	0 00	0.00	0.00	-
Collection control	0	91	99	0 00	0 00	0 00	-
Condenser passage	0	91	100	0 00	0.00	0 00	-
Pump passage	0	91	99	0 05	0 05	0 02	0.01
Handling control	0	-	300	0 00	0 00	0.00	-
Thermal control	10	50	300	0 00	0 00	0.00	-
Collection control	10	50	295	0 00	0 00	0.00	-
Condenser passage	10	50	305	0.02	0.02	< 0.01	< 0.01
Pump passage	10	50	292	0 03	0.01	0 01	0 20
Handling control	0	-	99	0.00	0.00	0 00	-
Thermal control	10	91	100	0.00	0 00	0 00	-
Collection control	10	91	98	0 00	0 00	0.00	-
Condenser passage	10	91	99	0 02	0.02	0.01	0.08
Pump passage	10	91	101	0.11	0.09	0.03	< 0 01
Handling control	0	-	50	0 00	0 00	0 00	-
Thermal control	15	50	50	1 00	1 00	0.00	<0.01
Collection control	15	50	50	1 00	-	-	-
Condenser passage	15	50	56	1.00	-	-	-
Pump passage	15	50	44	1.00	-	-	-

[a] P_1 = Total short-term mortality observed among fish in Experiment Type 1 (see Materials and Methods).

[b] $\hat{m}_1$ = Conditional mortality due to the ith source of stress (see Materials and Methods).

[c] Standard deviation of $\hat{m}_1$

[d] The probability of observing the given value of $\hat{m}_1$ or greater if, in fact, the true m_1 is equal to zero.

Table 10. Short-term conditional mortality due to collection stresses ($\hat{m}_3$) for larval fishes exposed to various combinations of ΔT and pumping rate in the Power Plant Simulator

ΔT (°C)	Pumping rate (% of maximum)	Species	Conditional mortality ($\hat{m}_3$)	Critical[a] probability
0	50	carp	0.12	< 0.01
		bluegill sunfish	< 0.01	0.29
		channel catfish	< 0.01	0.04
0	91	carp	0.73	< 0.01
		channel catfish	0.00	-
		largemouth bass	0.20	< 0.01
10	50	bluegill sunfish	0.10	< 0.01
		channel catfish	0.00	-
		smallmouth bass	0.00	-
		largemouth bass	0.31	< 0.01
10	91	channel catfish	0.00	-
		smallmouth bass	b	b
		largemouth bass	0.24	< 0.01

[a]The probability of observing the given value of $\hat{m}_3$ or greater if, in fact, the true m_3 is equal to 0.

[b]The estimated conditional mortality is negative, indicating that a feasible estimate is not obtainable due to high background mortality.

Table 11. Short-term conditional mortality due to pipe and condenser passage ($\hat{m}_4$) for larval fishes exposed to various combinations of ΔT and pumping rate in the Power Plant Simulator

ΔT (°C)	Pumping rate (% of maximum)	Species	Conditional mortality ($\hat{m}_4$)	Critical[a] probability
0	50	carp	0.16	0.02
		bluegill sunfish	0.02	0.18
		channel catfish	< 0.01	0.40
0	91	carp	0.84	< 0.01
		channel catfish	0.00	-
		largemouth bass	0.01	0.43
10	50	bluegill sunfish	0.08	0.04
		channel catfish	0.02	< 0.01
		smallmouth bass	0.00	-
		largemouth bass	0.65	< 0.01
10	91	channel catfish	0.02	0.08
		smallmouth bass	b	b
		largemouth bass	1.00	< 0.01

[a]The probability of observing the given value of $\hat{m}_4$ or greater if, in fact, the true m_4 is equal to 0.

[b]The estimated conditional mortality is negative, indicating that a feasible estimate is not obtainable due to high background mortality.

Table 12. Short-term conditional mortality due to pump passage ($\hat{m}_5$) for larval fishes exposed to various combinations of ΔT and pumping rate in the Power Plant Simulator

ΔT (°C)	Pumping rate (% of maximum)	Species	Conditional mortality ($\hat{m}_5$)	Critical[a] probability
0	50	carp	(2)	(2)
		bluegill sunfish	0.03	0.20
		channel catfish	0.03	0.03
0	91	carp	(2)	(2)
		channel catfish	0.05	< 0.01
		largemouth bass	0.04	0.31
10	50	bluegill sunfish	0.22	< 0.01
		channel catfish	0.01	0.20
		smallmouth bass	0.00	-
		largemouth bass	b	b
10	91	channel catfish	0.09	< 0.01
		smallmouth bass	0.01	0.33
		largemouth bass	-	-

[a]The probability of observing the given value of $\hat{m}_5$ or greater if, in fact, the true m_5 is equal to 0.

[b]The estimated conditional mortality is negative, indicating that a feasible estimate is not obtainable due to high background mortality.

Table 11 contains the estimated short-term conditional mortality rates resulting from the stresses of pipe and condenser passage ($\hat{m}_4$). As with the collection control experimental group, $\hat{m}_4$'s tended to be high for carp and largemouth bass and were uniformly low among smallmouth bass and channel catfish under the experimental conditions tested. At a given temperature, the magnitude of $\hat{m}_4$ was directly related to the pumping rate for carp and largemouth bass larvae but not for channel catfish or smallmouth bass. Estimated conditional mortalities due to pipe and condenser passage were directly related to ΔT at a given pumping rate for bluegill, channel catfish, and largemouth bass.

Estimates of short-term conditional mortalities due to pump passage are given in Table 12. The mortalities resulting from this source of entrainment stress were relatively low in most cases, and among carp, smallmouth bass, and largemouth bass larvae no significant additional mortality due to pump passage was detected in this study. Unlike the pattern exhibited by $\hat{m}_4$'s (Table 11), estimated conditional mortalities due to pump passage were directly related to pumping rate for channel catfish larvae. At a given pumping rate, only bluegill sunfish showed a consistent direct relationship between $\hat{m}_5$ and ΔT.

It is also of interest to compare the short-term mortality responses of fishes exposed to thermal and physical stresses acting in concert, e.g. ($\hat{m}_5$ at a ΔT of 10 °C) with mortalities resulting from the separate stresses (e.g., $\hat{m}_5$ at ambient temperatures (only physical stress) and m_2 at a ΔT of 10 °C (only thermal stress)). If

conditional mortalities resulting from simultaneous thermal and physical stresses are greater than would be expected based on combining the effects of separate stresses, it would be indicative of synergistic effects between elevated temperatures and physical stresses.

Statistically, synergism can be expressed as an interaction between the effects of temperature and mechanical stress in a two-way factorial analysis of variance. The two entrainment stress factors are each represented by two levels, absence and presence:

Physical stress	Temperature stress	
	Absent	Present
Absent	P_3 at $\Delta T = 0°C$	P_3 at $\Delta T = 10°C$
Present	P_5 at $\Delta T = 0°C$	P_5 at $\Delta T = 10°C$

Three sets of data were available to test the null hypothesis of no interaction: bluegill sunfish at 50% of maximum pumping rate (Table 8) and channel catfish at 50% and 91% of maximum pumping rate (Table 9).

Prior to the analysis of variance, an angular transformation (p. 386, Sokal and Rohlf 1969) was applied to the observed mortality data (P_1's). Residuals were examined for any marked deviation from normality and none was detected. All analyses were done using the Statistical Analysis System (SAS 1979).

Results of the analyses of variance are summarized in Table 13. While the main effects (thermal stress and physical stress) were frequently significant sources of mortality, the thermal stress X physical stress interaction was not significant at the $\alpha = 0.05$ level for any of the three data sets. Therefore, synergistic effects between these two entrainment stresses could not be demonstrated by the 1979 PPS data. Although there was an indication of synergistic effects for bluegill sunfish (interaction term was significant at the $\alpha = 0.10$ level), the number of experimental replicates was small. Potential synergistic interactions between entrainment stresses will be examined further in 1981.

4.3. Delayed Mortality

The cumulative delayed mortalities observed among carp larvae at various times following exposure to different pumping rates in the PPS are depicted in Figs. 3 and 4. In both graphs, observed cumulative mortalities in the condenser-passage group (Experiment Type 4) were greater than those in the other test groups throughout the entire post-test holding period. Delayed mortalities among the handling and thermal control groups were low (≤ 0.10), but cumulative delayed mortalities in the collection control groups were only slightly below the respective values for the pump passage groups.

Estimates of delayed mortality in carp larvae were significantly greater than zero ($\alpha = 0.05$) for all experimental types except the thermal control at a pumping rate of 50% (Table 14). On the other hand, only fish in Experiment Type 5 showed significant delayed

Table 13. Analysis of variance of observed mortality among fishes exposed to both thermal and physical stresses in the PPS

Source of variation	df	SS	MS	F
Bluegill sunfish, 50% of maximum pumping rate				
Thermal stress (ΔT)	1	0.241	0.241	44.12[a]
Physical stress	1	0.095	0.095	17.36[a]
Thermal x physical	1	0.021	0.021	3.91
Error	6	0.033	0.005	
Total (corrected)	9	0.413		
Channel catfish, 50% of maximum pumping rate				
Thermal stress (ΔT)	1	0.003	0.003	0.33
Physical stress	1	0.104	0.104	10.16[b]
Thermal x physical	1	0.205	0.010	0.20
Error	20	0.205	0.010	
Total (corrected)	23	0.314		
Channel catfish, 91% of maximum pumping rate				
Thermal stress (ΔT)	1	0.006	0.006	1.04
Physical stress	1	0.148	0.148	24.81[a]
Thermal x physical	1	0.006	0.006	1.04
Error	4	0.024	0.006	
Total (corrected)	7	0.184		

[a]Source of mortality is significant at the $\alpha = 0.01$ level.

[b]Source of mortality is significant at the $\alpha = 0.05$ level.

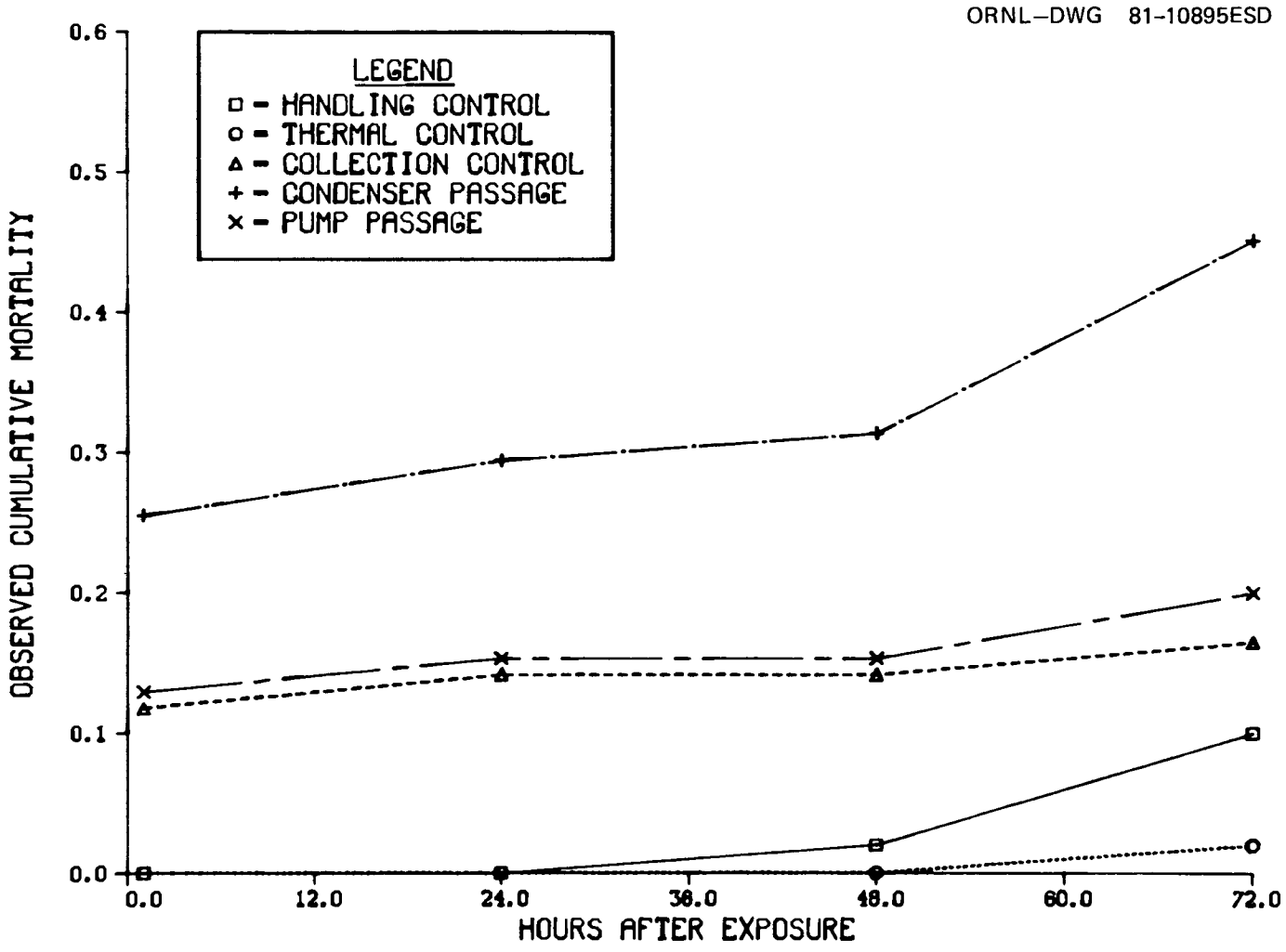

Fig. 3. Observed cumulative mortalities of carp larvae at various times after exposure to a ΔT of $0^{\circ}C$ and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types.

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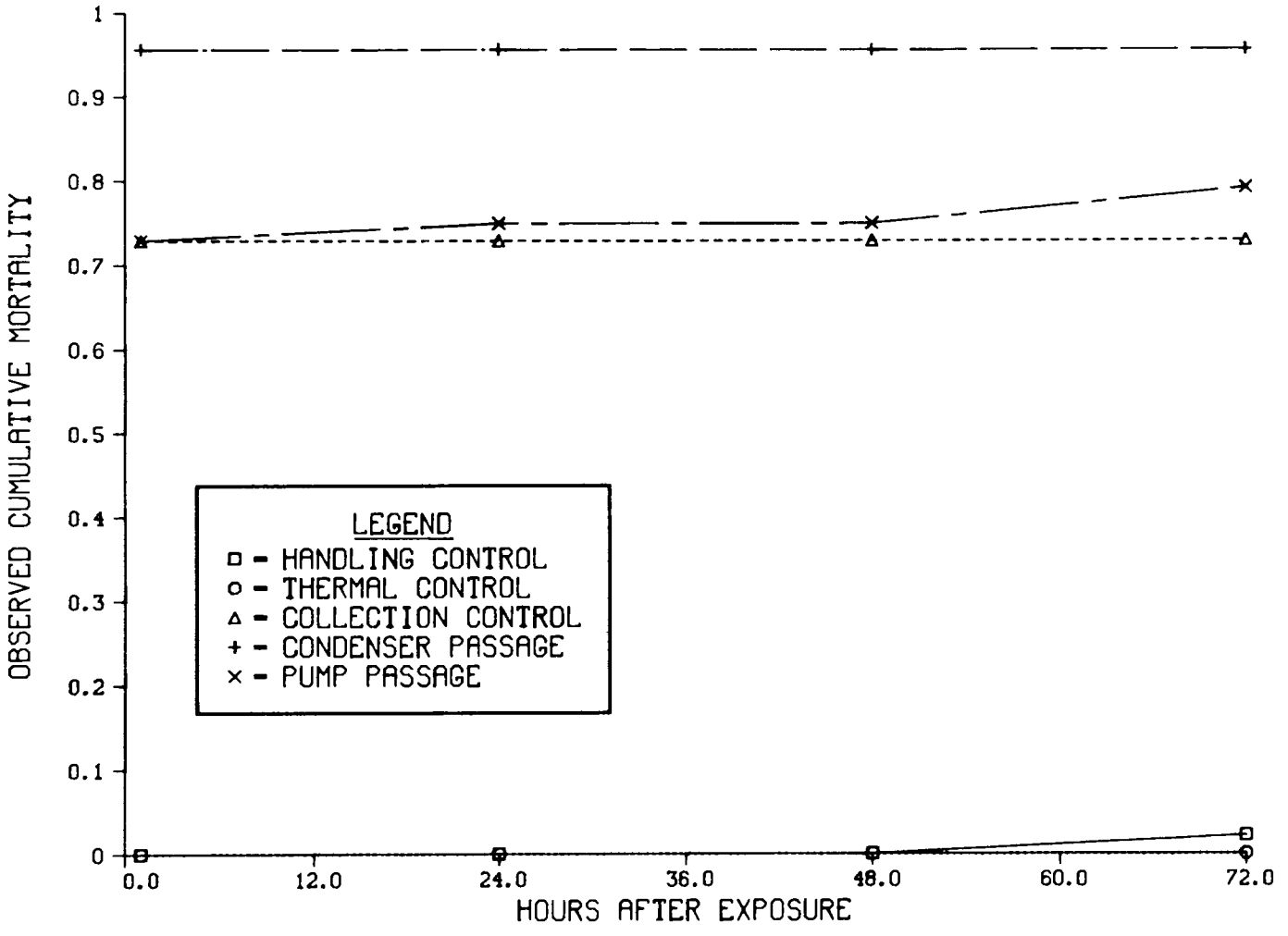

Fig. 4. Observed cumulative mortalities of carp larvae at various times after exposure to a ΔT of 0°C and a pumping rate of 91% of maximum. Refer to the text for a description of the experiment types.

Table 14. Delayed (1-72 h) mortalities among carp (*Cyprinus carpio*) larvae exposed to different pumping rates in the Power Plant Simulator

Experiment type	$\Delta T(^{\circ}C)$	Pumping rate (% of maximum)	Number of fish[a] $(N_1)(1-P_1)$	Delayed mortality[b] $(\hat{d}_1)$	Standard[c] deviation	Critical[d] probability
Handling control	0	-	100	0.10	0.03	< 0.01
Thermal control	0	50	102	0.02	0.01	0.07
Collection control	0	50	75	0.05	0.03	0.02
Condenser passage	0	50	38	0.26	0.07	< 0.01
Pump passage	0	50	74	0.08	0.03	< 0.01
Handling control	0	-	46	0.02	0.02	0.15
Thermal control	0	91	51	0.00	0.00	-
Collection control	0	91	13	0.00	0.00	-
Condenser passage	0	91	2	0.00	0.00	-
Pump passage	0	91	13	0.23	0.12	0.03

[a]Number of fish used in the estimation of delayed mortality, where N_1 = number of fish recovered in Experiment Type 1, and P_1 = proportion dead in Experiment Type 1 in the time interval (0-1 h).

[b]$\hat{d}_1$ = estimated delayed mortality for Experiment Type 1 (see Materials and Methods).

[c]Standard deviation of $\hat{d}_1$ (see Materials and Methods).

[d]Probability of observing the given value of $\hat{d}_1$ or greater if, in fact, d_1 is equal to zero.

mortalities at a pumping rate of 91% of maximum. The results under the latter test conditions must be interpreted with caution, however, in view of the high short-term mortalities (Table 5) and resultant low numbers of fish available to estimate delayed mortalities in Table 14. Table 15 compares delayed mortalities for carp larvae in Experiment Types 2 through 5 to those in Experiment Type 1 (handling and natural mortality). Delayed mortalities of test groups were significantly greater (α = 0.05) than delayed mortalities of handling control groups in two cases (condenser-passage group at 50%); pump-passage group at 91%); in one instance (thermal control at 50%) the delayed mortality of the test group was lower than the handling control. In all other cases, differences in delayed mortalities between fish exposed to stresses in the PPS and those exposed only to handling and natural stresses were not significant at the α = 0.05 level.

Figures 5 through 9 are plots of the cumulative delayed mortalities of largemouth bass larvae following exposure to different combinations of excess temperature and pumping rates in the PPS. Largemouth bass larvae in Experiment Type 2 exhibited cumulative 72-h mortalities of 0.02 to 0.22 following exposure to ΔT's of 0° and 4°C and mortalities of 0.12 and 1.00 following exposure to a ΔT of 10°C. At pumping rates of 91%, cumulative delayed mortalities in the collection control group were generally similar to, or even exceeded, mortalities in Experiment Types 4 and 5, which had received the additional stresses of condenser and pump passage, respectively. In general, the magnitudes of the 72-h cumulative mortalities of largemouth bass larvae in Experiment Types 3 through 5 were directly related to both ΔT and pumping rate (Figs. 5 through 9).

Table 15 Comparisons of delayed mortalities among carp (*Cyprinus carpio*) larvae exposed to stresses in the Power Plant Simulator with delayed mortalities among carp larvae exposed only to handling and natural stresses

Experiment type	$\Delta T(^{\circ}C)$	Pumping rate (% of maximum)	Delayed mortality[a] ($\hat{d}_i$)	Difference from handling control ($\hat{d}_i - \hat{d}_1$)	Standard[b] deviation	Critical[c] probability
Handling control	0	-	0.10	0 00	0 00	-
Thermal control	0	50	0.02	-0.08	0.03	< 0.01
Collection control	0	50	0 05	-0.05	0 04	0 12
Condenser passage	0	50	0 26	0.16	0.08	0 02
Pump passage	0	50	0.08	-0.02	0.04	0.33
Handling control	0	-	0 02	0 00	0 00	-
Thermal control	0	91	0.00	-0 02	0 02	0 15
Collection control	0	91	0 00	-0 02	0.02	0.15
Condenser passage	0	91	0.00	-0 02	0 02	0 15
Pump passage	0	91	0.23	0 21	0.12	0 04

[a] $\hat{d}_i$ = estimated delayed morality for Experiment Type i, where i = 1,2,3,4,5 Experiment Type 1 = Handling Control, Experiment Type 2 = Thermal Control, Experiment Type 3 = Collection Control, Experiment Type 4 = Condenser Passage, Experiment Type 5 = Pump Passage.

[b] Standard deviation of $\hat{d}_i - \hat{d}_1 = V(\hat{d}_i) + V(\hat{d}_1)^{1/2}$ See Materials and Methods.

[c] Probability of observing the absolute value of $\hat{d}_i - \hat{d}_1$ or greater if, in fact, $d_i - d_1$ is equal to zero

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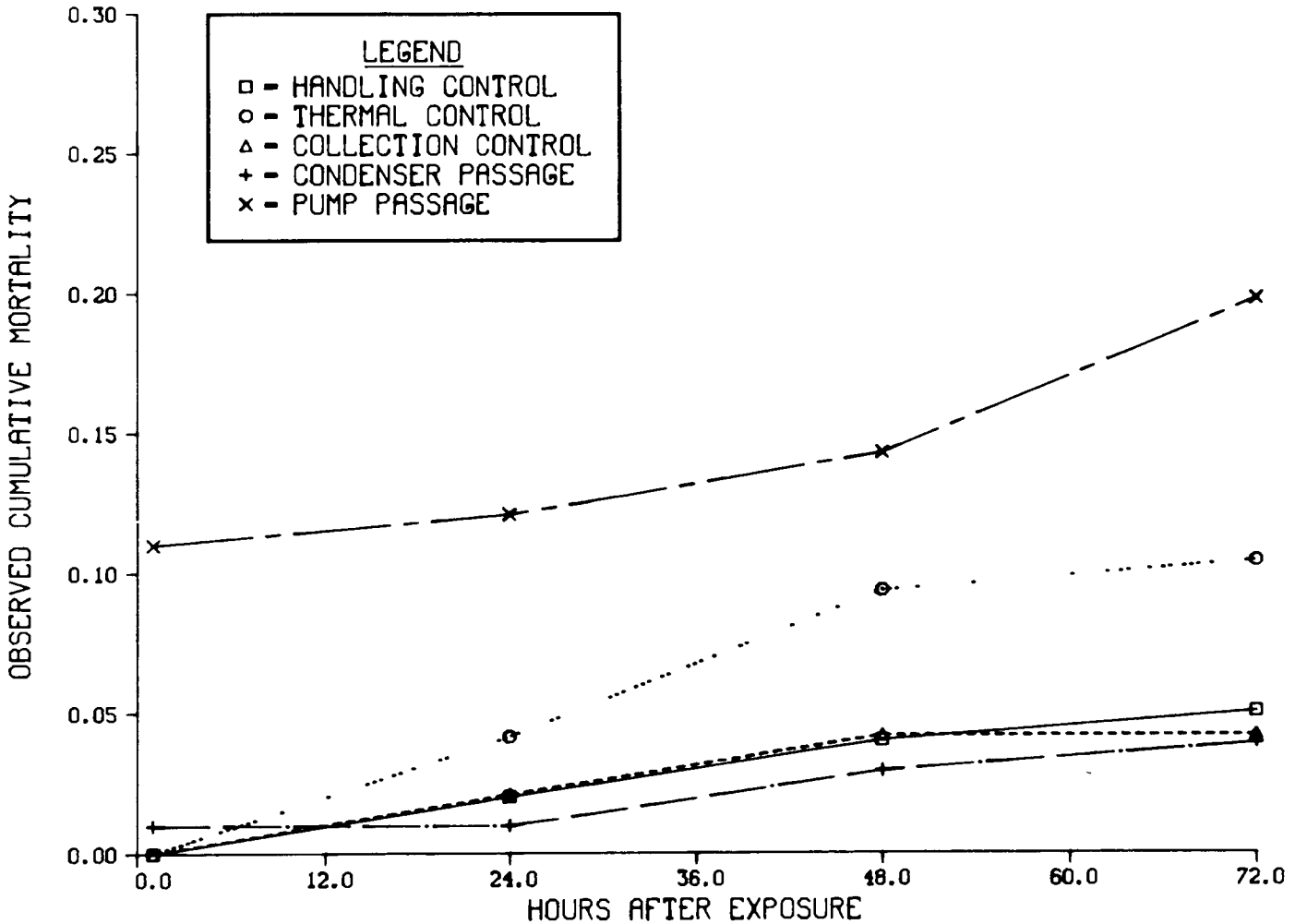

Fig. 5. Observed cumulative mortalities of largemouth bass larvae at various times after exposure to a ΔT of 0°C and a pumping rate of 75% of maximum. Refer to the text for a description of the experiment types.

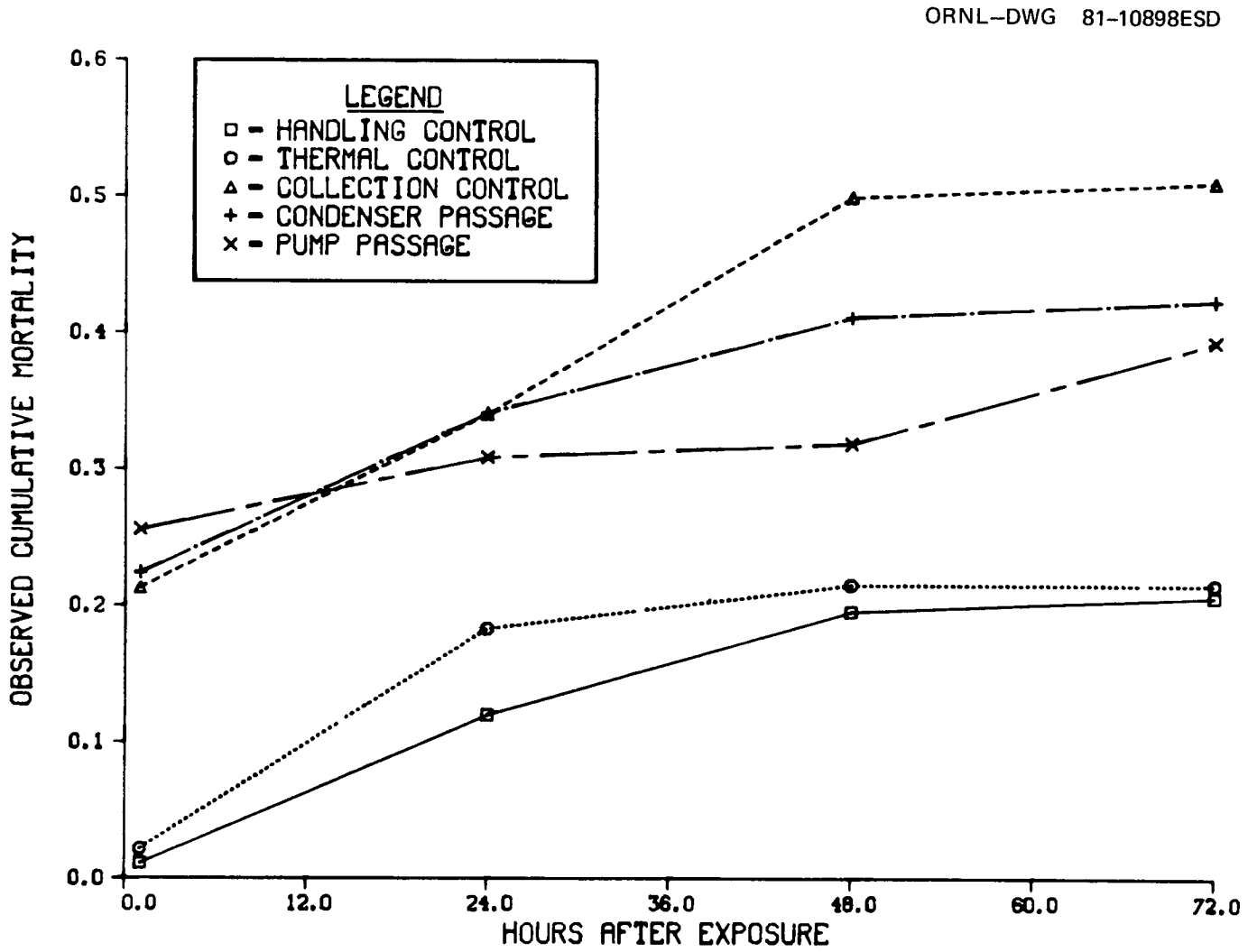

Fig. 6. Observed cumulative mortalities of largemouth bass larvae at various times after exposure to a ΔT of $0\,^{\circ}C$ and a pumping rate of 91% of maximum. Refer to the text for a description of the experiment types.

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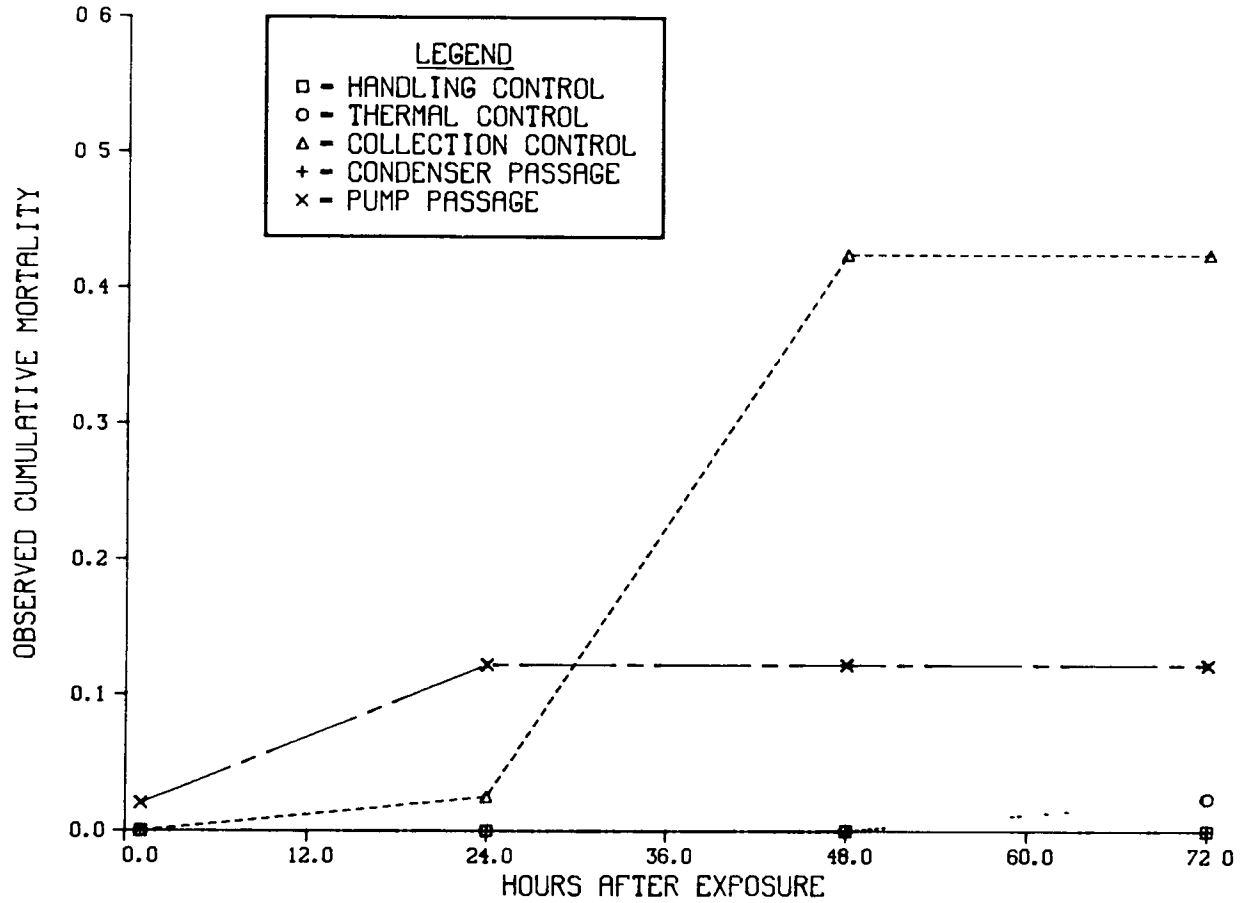

Fig. 7. Observed cumulative mortalities of largemouth bass larvae at various times after exposure to a ΔT of 4 °C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types.

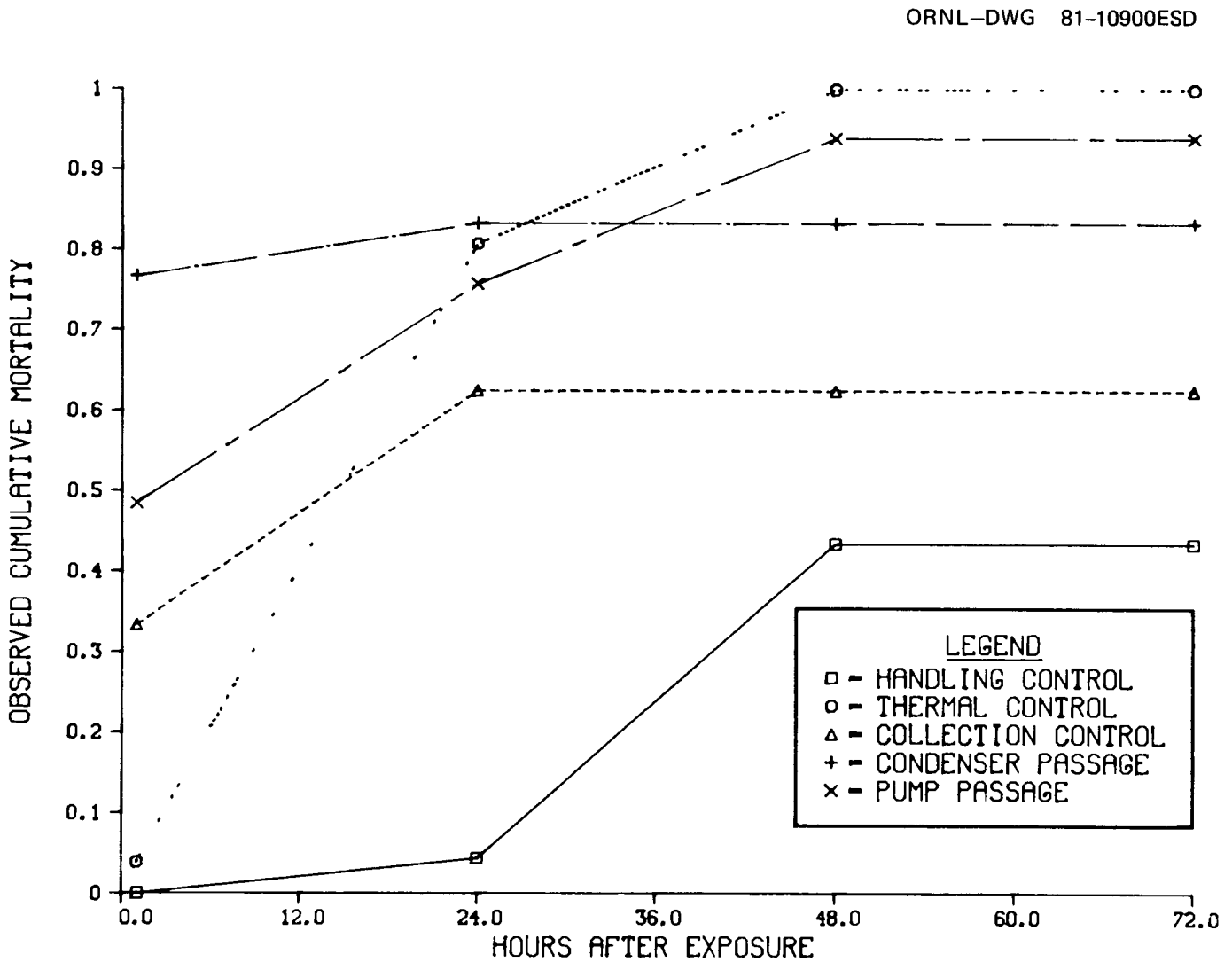

Fig. 8. Observed cumulative mortalities of largemouth bass larvae at various times after exposure to a ΔT of 10°C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types.

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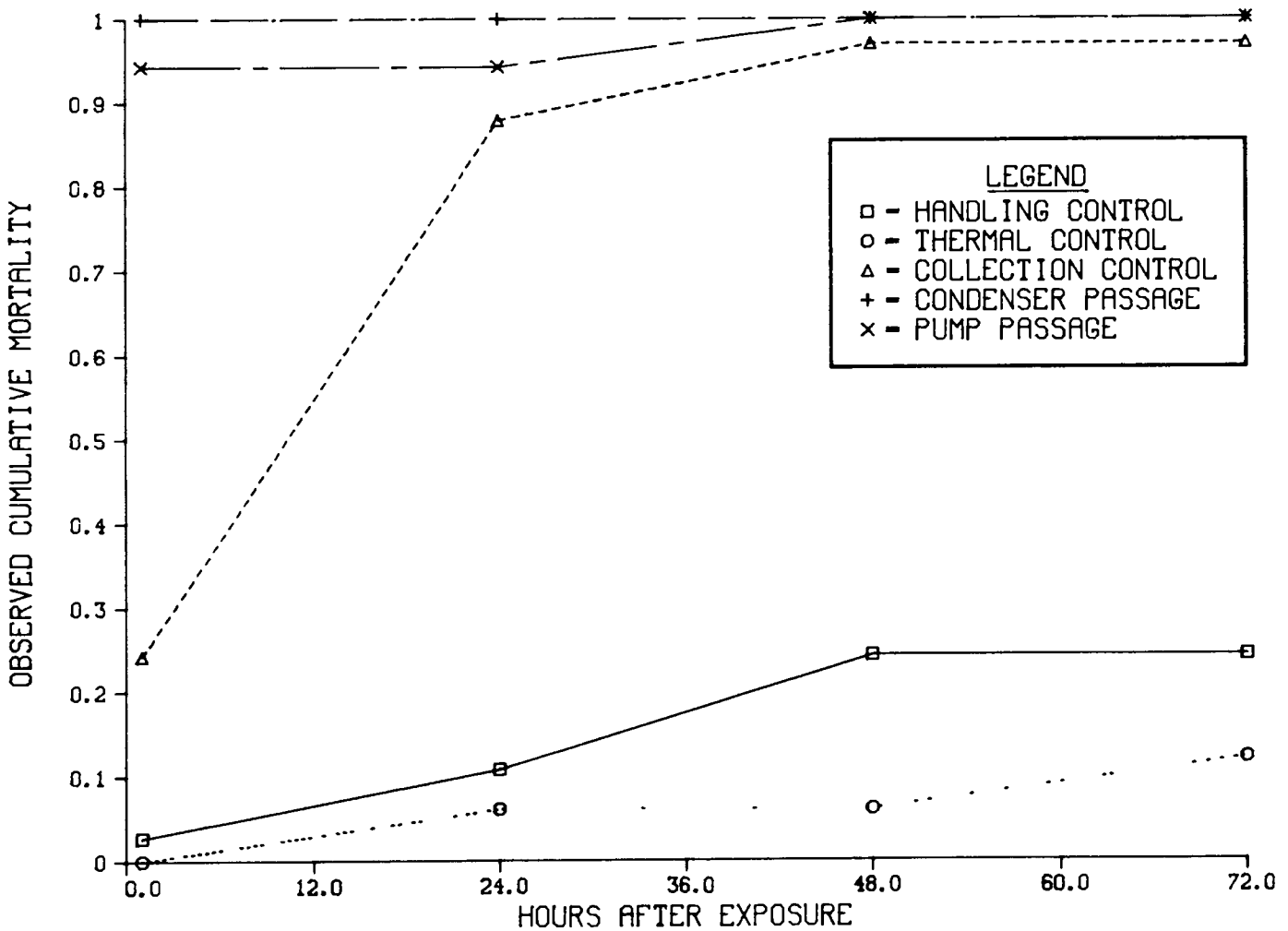

Fig. 9. Observed cumulative mortalities of largemouth bass larvae at various times after exposure to a ΔT of 10°C and a pumping rate of 91% of maximum. Refer to the text for a description of the experiment types.

Delayed mortalities of largemouth bass larvae were significantly greater than zero (α = 0.05) for nearly all comparisons (Table 16). Delayed mortalities in test groups exposed to stresses in the PPS (Experiment Types 2 through 5) were significantly greater (α = 0.05) than mortalities in handling control groups (Experiment Type 1) in 7 of 20 possible comparisons (Table 17). For largemouth bass larvae, the $\hat{d}_1$'s among groups exposed only to handling and natural stresses were never significantly greater (α = 0.05) than the $\hat{d}_1$'s among the other experimental groups in these tests.

Few consistent trends were evident from the cumulative delayed mortality data for smallmouth bass larvae plotted in Figures 10 through 12. Under some experimental conditions (Figs. 10 and 11), delayed mortalities of handling and thermal control groups were similar to or exceeded those of condenser and pump passage groups; under different conditions (Fig. 12), the opposite tendency was observed. Except for condenser and pump passage groups exposed to a ΔT of 10°C and pumping rates of 91%, 72-h post-test mortalities among smallmouth bass larvae were relatively low ($\leq$ 0.15). Delayed mortalities in the collection control group did not exceed 0.04 under any of the conditions tested.

In many cases, delayed mortalities ($\hat{d}_1$) of smallmouth bass were not significantly greater than zero at the α = 0.05 level (Table 18). However, at a 10°C ΔT and 91% pumping rate condenser-passage and pump-passage groups exhibited delayed mortalities that were not only significantly greater than zero (Table 18) but also greater than d_1, i.e., the delayed mortality in the handling control (Table 19). Table 19 also indicates that a ΔT of 4°C and a pumping rate of 91%,

Table 16 Delayed (1-72 h) mortalities among largemouth bass (*Micropterus salmoides*) larvae exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator

Experiment type	ΔT(°C)	Pumping rate (% of maximum)	Number of fish[a] $(N_1)(1-P_1)$	Delayed mortality[b] $(\hat{d}_1)$	Standard[c] deviation	Critical[d] probability
Handling control	0	-	99	0 05	0.02	0.01
Thermal control	0	75	96	0 10	0.03	< 0.01
Collection control	0	75	95	0.04	0.02	0.02
Condenser passage	0	75	101	0.03	0.02	0.04
Pump passage	0	75	81	0 10	0.03	< 0.01
Handling control	0	-	91	0 20	0.04	< 0 01
Thermal control	0	91	91	0.20	0.04	< 0.01
Collection control	0	91	74	0.38	0.06	< 0.01
Condenser passage	0	91	66	0.26	0.05	< 0.01
Pump passage	0	91	70	0.19	0.05	< 0.01
Handling control	0	-	47	0.00	0.00	-
Thermal control	4	50	42	0 02	0.02	0.15
Collection control	4	50	40	0.43	0.08	< 0 01
Condenser passage	4	50	45	0.00	0.00	-
Pump passage	4	50	48	0 10	0.04	< 0 01
Handling control	0	-	23	0.43	0.10	< 0.01
Thermal control	10	50	25	1 00	0.00	< 0.01
Collection control	10	50	16	0 44	0.12	< 0.01
Condenser passage	10	50	7	0 29	0.18	0.05
Pump passage	10	50	17	0 88	0.08	< 0.01
Handling control	0	-	37	0 22	0.07	< 0.01
Thermal control	10	91	33	0.12	0.06	< 0.01
Collection control	10	91	25	0 96	0.04	< 0 01
Condenser passage	10	91	0	-	-	-
Pump passage	10	91	2	1.00	0.00	< 0 01

[a]Number of fish used in the estimation of delayed mortality, where N_1 = number of fish recovered in Experiment Type 1, and P_1 = proportion dead in Experiment Type 1 in the time interval (0-1 h).

[b]$\hat{d}_1$ = estimated delayed mortality for Experiment Type 1 (see Materials and Methods).

[c]Standard deviation of $\hat{d}_1$ (see Materials and Methods).

[d]Probability of observing the given value of $\hat{d}_1$ or greater if, in fact, d_1 is equal to zero

Table 17 Comparisons of delayed mortalities among largemouth bass (*Micropterus salmoides*) larvae exposed to stresses in the Power Plant Simulator with delayed mortalities among largemouth bass larvae exposed only to handling and natural stresses

Experiment type	ΔT(°C)	Pumping rate (% of maximum)	Delayed mortality[a] ($\hat{d}_i$)	Difference from handling control ($\hat{d}_i$-$\hat{d}_1$)	Standard[b] deviation	Critical[c] probability
Handling control	0	-	0 05	0 00	0 00	-
Thermal control	0	75	0 10	0 05	0.04	0 08
Collection control	0	75	0 04	-0 01	0 03	0 39
Condenser passage	0	75	0 03	-0.02	0 03	0 22
Pump passage	0	75	0 10	0 05	0 04	0 11
Handling control	0	-	0 20	0 00	0 00	-
Thermal control	0	91	0 20	0 00	0 06	0 50
Collection control	0	91	0 38	0 18	0 07	0 01
Condenser passage	0	91	0 26	0 06	0 07	0 19
Pump passage	0	91	0 19	-0 01	0.06	0 42
Handling control	0	-	0 00	0 00	0 00	-
Thermal control	4	50	0 02	0 02	0 02	0 15
Collection control	4	50	0 43	0.43	0 08	0 01
Condenser passage	4	50	0 00	0.00	0 00	-
Pump passage	4	50	0 10	0 10	0 04	0 01
Handling control	0	-	0 43	0 00	0 00	-
Thermal control	10	50	1 00	0 57	0 10	0 01
Collection control	10	50	0 44	0 01	0 16	0 49
Condenser passage	10	50	0 29	-0 15	0 20	0 23
Pump passage	10	50	0 88	0 45	0 13	0 01
Handling control	0	-	0 22	0 00	0 00	-
Thermal control	10	91	0 12	-0 10	0 09	0.12
Collection control	10	91	0 96	0 74	0 08	0.01
Condenser passage	10	91	-	-	-	-
Pump passage	10	91	1 00	0 78	0 07	0 01

[a] $\hat{d}_i$ = estimated delayed mortality for Experiment Type i, where i = 1,2,3,4,5 Experiment Type 1 = Handling Control, Experiment Type 2 = Thermal Control, Experiment Type 3 = Collection Control, Experiment Type 4 = Condenser Passage, Experiment Type 5 = Pump Passage

[b] Standard deviation of $(\hat{d}_i-\hat{d}_1) = (V(\hat{d}_i) + V(\hat{d}_1))^{1/2}$ See Materials and Methods

[c] Probability of observing the absolute value of $(\hat{d}_i-\hat{d}_1)$ or greater if, in fact, (d_i-d_1) is equal to zero

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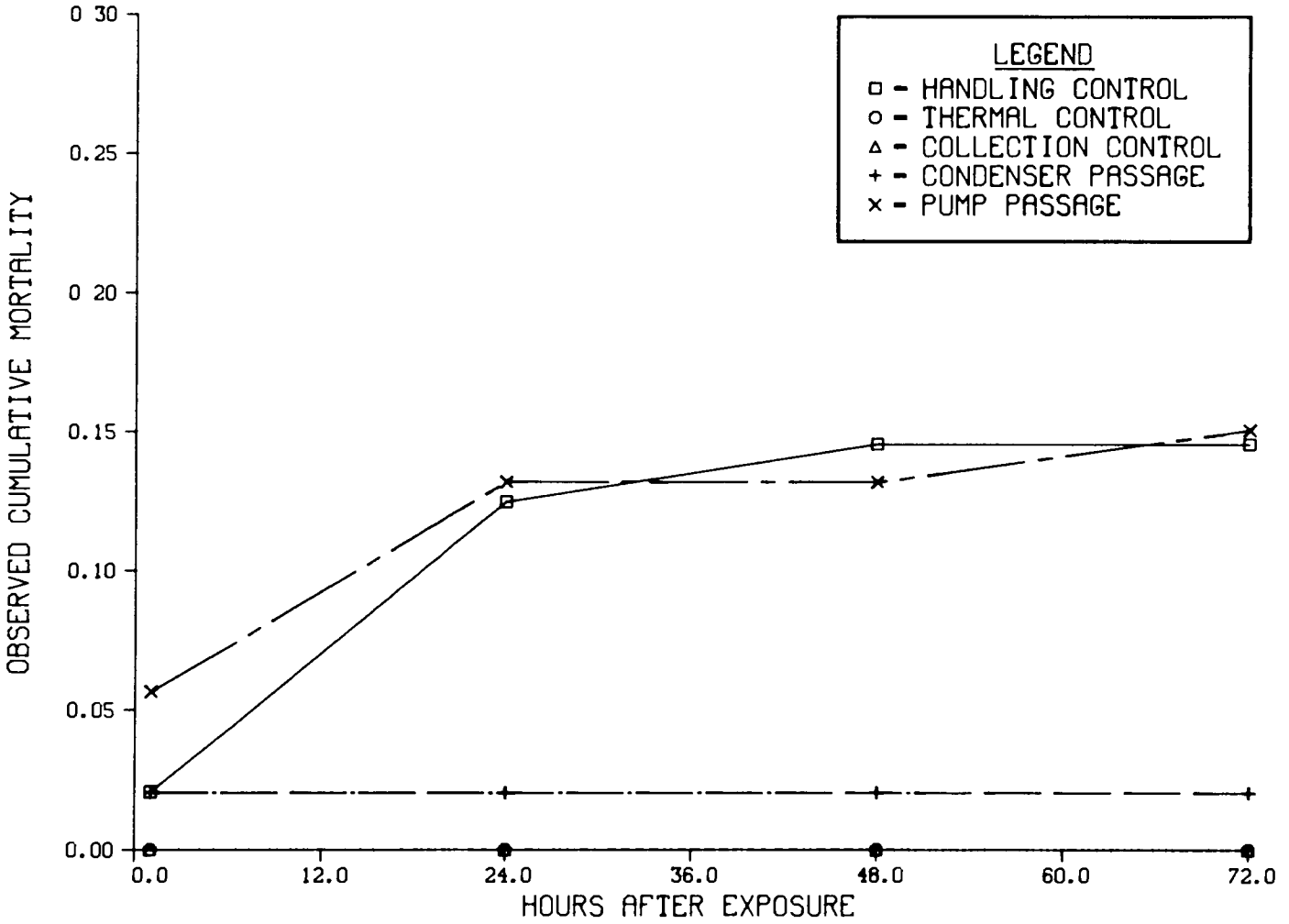

Fig. 10. Observed cumulative mortalities of smallmouth bass larvae at various times after exposure to a ΔT of 4°C and a pumping rate of 91% of maximum. Refer to the text for a description of the experiment types.

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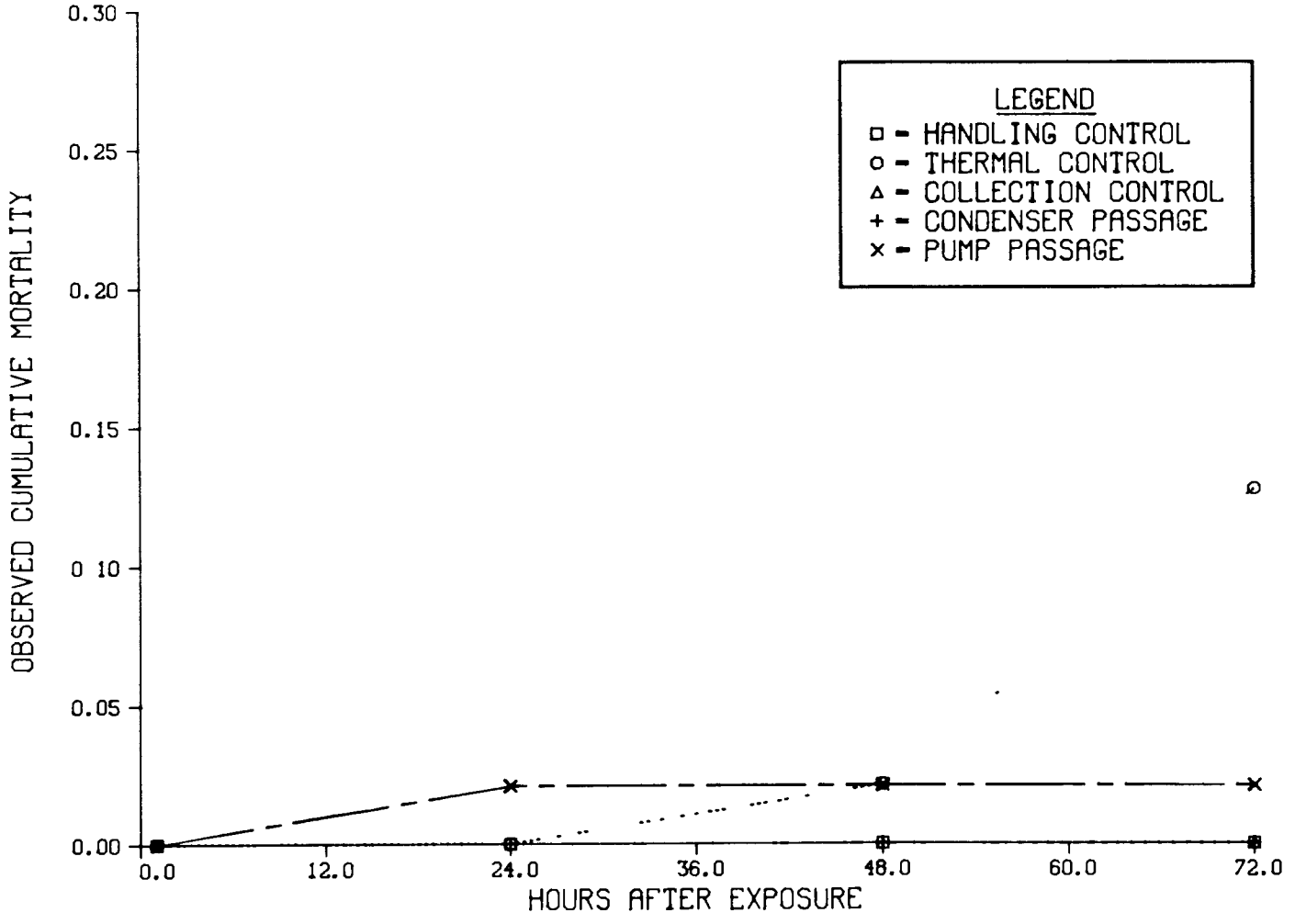

Fig. 11. Observed cumulative mortalities of smallmouth bass larvae at various times after exposure to a ΔT of 10°C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types.

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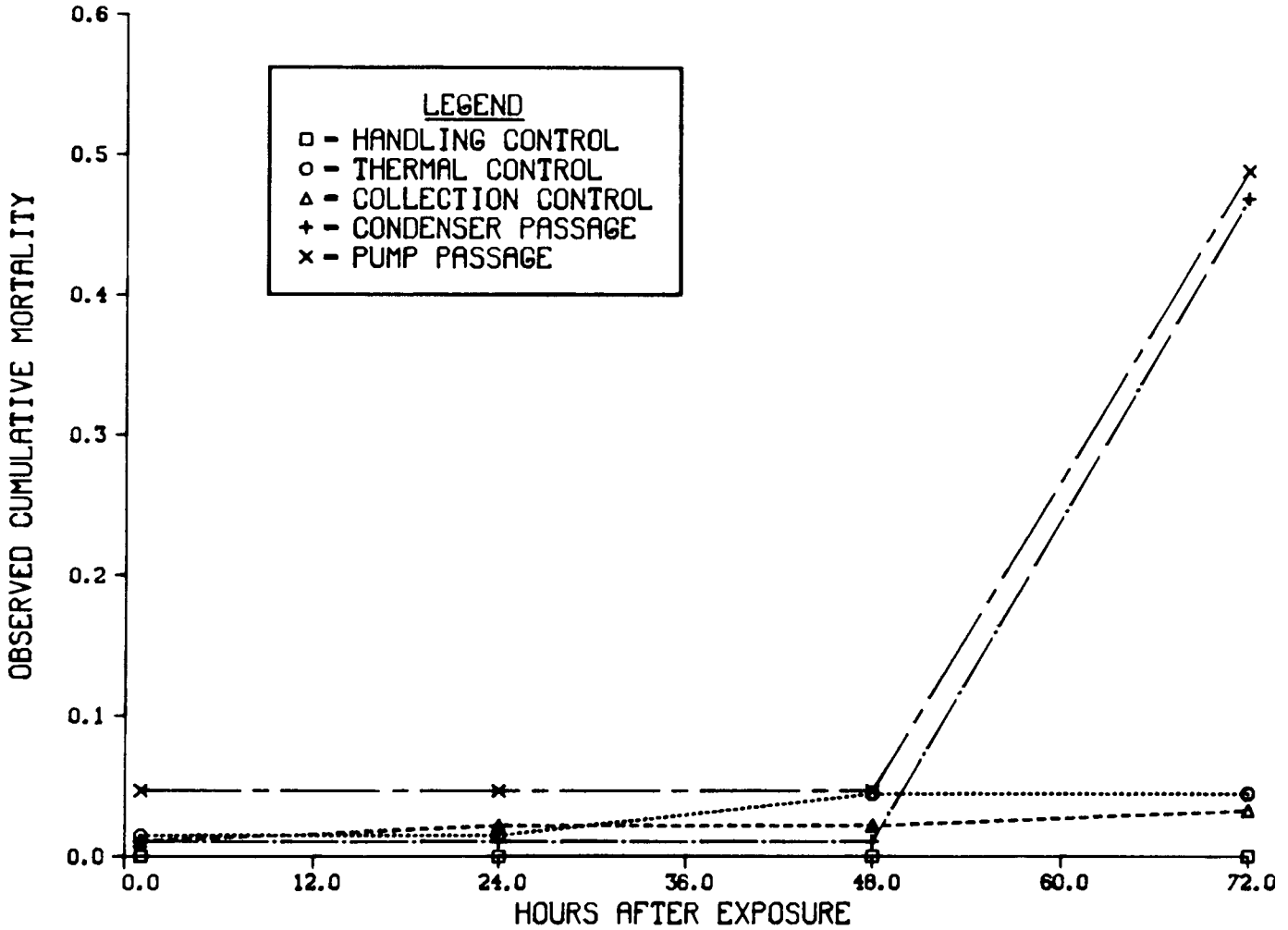

Fig. 12. Observed cumulative mortalities of smallmouth bass larvae at various times after exposure to a ΔT of 10°C and a pumping rate of 91% of maximum. Refer to the text for a description of the experiment types.

Table 18. Delayed (1-72 h) mortalities among smallmouth bass (*Micropterus dolomieui*) larvae exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator

Experiment type	ΔT(°C)	Pumping rate (% of maximum)	Number of fish[a] $(N_1)(1-P_1)$	Delayed mortality[b] $(\hat{d}_1)$	Standard[c] deviation	Critical[d] probability
Handling control	0	-	47	0.13	0.05	< 0.01
Thermal control	4	91	50	0.00	0.00	-
Collection control	4	91	50	0.00	0.00	-
Condenser passage	4	91	48	0.00	0.00	-
Pump passage	4	91	50	0.10	0.04	< 0.01
Handling control	0	-	49	0.00	0.00	-
Thermal control	10	50	47	0.13	0.05	< 0.01
Collection control	10	50	49	0.00	0.00	-
Condenser passage	10	50	49	0.00	0.00	-
Pump passage	10	50	48	0.02	0.02	0.15
Handling control	0	-	96	0.00	0.00	-
Thermal control	10	91	67	0.03	0.02	0.07
Collection control	10	91	92	0.02	0.02	0.07
Condenser passage	10	91	95	0.46	0.05	< 0.01
Pump passage	10	91	82	0.46	0.05	< 0.01

[a]Number of fish used in the estimation of delayed mortality, where N_1 = number of fish recovered in Experiment Type 1, and P_1 = proportion dead in Experiment Type 1 in the time interval (0-1 h).

[b]$\hat{d}_1$ = estimated delayed mortality for Experiment Type 1 (see Materials and Methods).

[c]Standard deviation of $\hat{d}_1$ (see Materials and Methods).

[d]Probability of observing the given value of $\hat{d}_1$ or greater if, in fact, d_1 is equal to zero.

Table 19 Comparisons of delayed mortalities among smallmouth bass (*Micropterus dolomieui*) larvae exposed to stresses in the Power Plant Simulator with delayed mortalities among smallmouth bass larvae exposed only to handling and natural stresses

Experiment type	ΔT(°C)	Pumping rate (% of maximum)	Delayed mortality[a] ($\hat{d}_i$)	Difference from handling control ($\hat{d}_i - \hat{d}_1$)	Standard[b] deviation	Critical[c] probability
Handling control	0	-	0 13	0 00	0 00	-
Thermal control	4	91	0.00	-0.13	0.05	< 0.01
Collection control	4	91	0 00	-0.13	0.05	< 0 01
Condenser passage	4	91	0 00	-0 13	0 05	< 0.01
Pump passage	4	91	0 10	-0 03	0.06	0.33
Handling control	0	-	0 00	0 00	0 00	-
Thermal control	10	50	0.13	0 13	0 05	< 0.01
Collection control	10	50	0 00	0.00	0.00	-
Condenser passage	10	50	0 00	0.00	0 00	-
Pump passage	10	50	0.02	0 02	0.02	0.15
Handling control	0	-	0.00	0 00	0.00	-
Thermal control	10	91	0.03	0.03	0.02	0.07
Collection control	10	91	0 02	0 02	0.02	0.07
Condenser passage	10	91	0.46	0.46	0 05	< 0.01
Pump passage	10	91	0 46	0.46	0 05	< 0.01

[a] $\hat{d}_i$ = estimated delayed mortality for Experiment Type i, where i = 1,2,3,4,5 Experiment Type 1 = Handling Control, Experiment Type 2 = Thermal Control, Experiment Type 3 = Collection Control, Experiment Type 4 = Condenser Passage, Experiment Type 5 = Pump Passage

[b] Standard deviation of $(\hat{d}_i - \hat{d}_1) = (V(\hat{d}_i) + V(\hat{d}_1))^{1/2}$. See Materials and Methods

[c] Probability of observing the absolute value of $(\hat{d}_i - \hat{d}_1)$ or greater if, in fact, $(d_i - d_1)$ is equal to zero

delayed mortalities among smallmouth bass larvae in Experiment Types 2, 3, and 4 were significantly smaller than the delayed mortality in Experiment Type 1. The unexpected negative differences were due to high delayed mortality in the handling control group.

Figures 13 and 14 show the cumulative delayed mortalities of bluegill sunfish larvae tested at different water temperatures in the PPS. Mortality at the end of the 72-h post-test holding period was greater for those experimental groups given a temperature shock of 10°C (Fig. 14) than for corresponding groups exposed to physical entrainment stresses at ambient temperatures (Fig. 13). During most of the post-test holding period, cumulative mortalities of fish in the condenser passage and pump passage groups exceeded those of the controls.

At ΔT's of 0° and 10°C, $\hat{d}_1$'s for all experimental groups were significantly greater than zero (Table 20). However, delayed mortalities of fish exposed to PPS stresses were significantly different ($\alpha = 0.05$) from the relatively high delayed mortalities of fish given only handling and natural stresses in only 1 of 8 comparisons (Table 21).

Among the five fish species examined in the 1979 experimental program, channel catfish were the most resistant to stresses in the PPS from the standpoint of both short-term and delayed mortality. The cumulative mortalities of catfish larvae at various times during the 72-h post-test holding period are shown in Fig. 15 through 18. With the exception of tests conducted at a ΔT of 10°C and a pumping rate of 91% of maximum (Fig. 18), cumulative moralities of all experimental groups remained below 0.10 throughout the holding period. Catfish in

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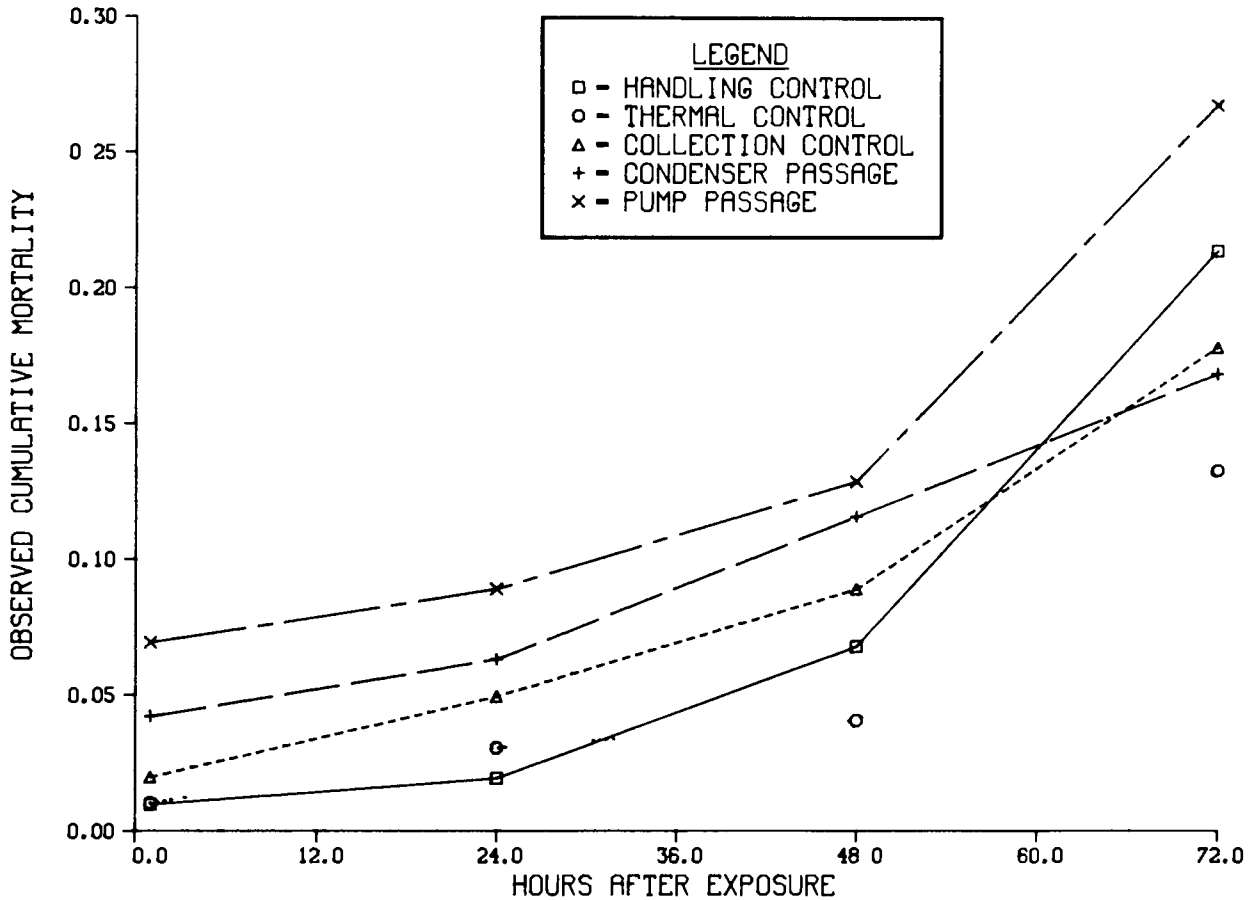

Fig. 13. Observed cumulative mortalities of bluegill sunfish larvae at various times after exposure to a ΔT of 0°C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types.

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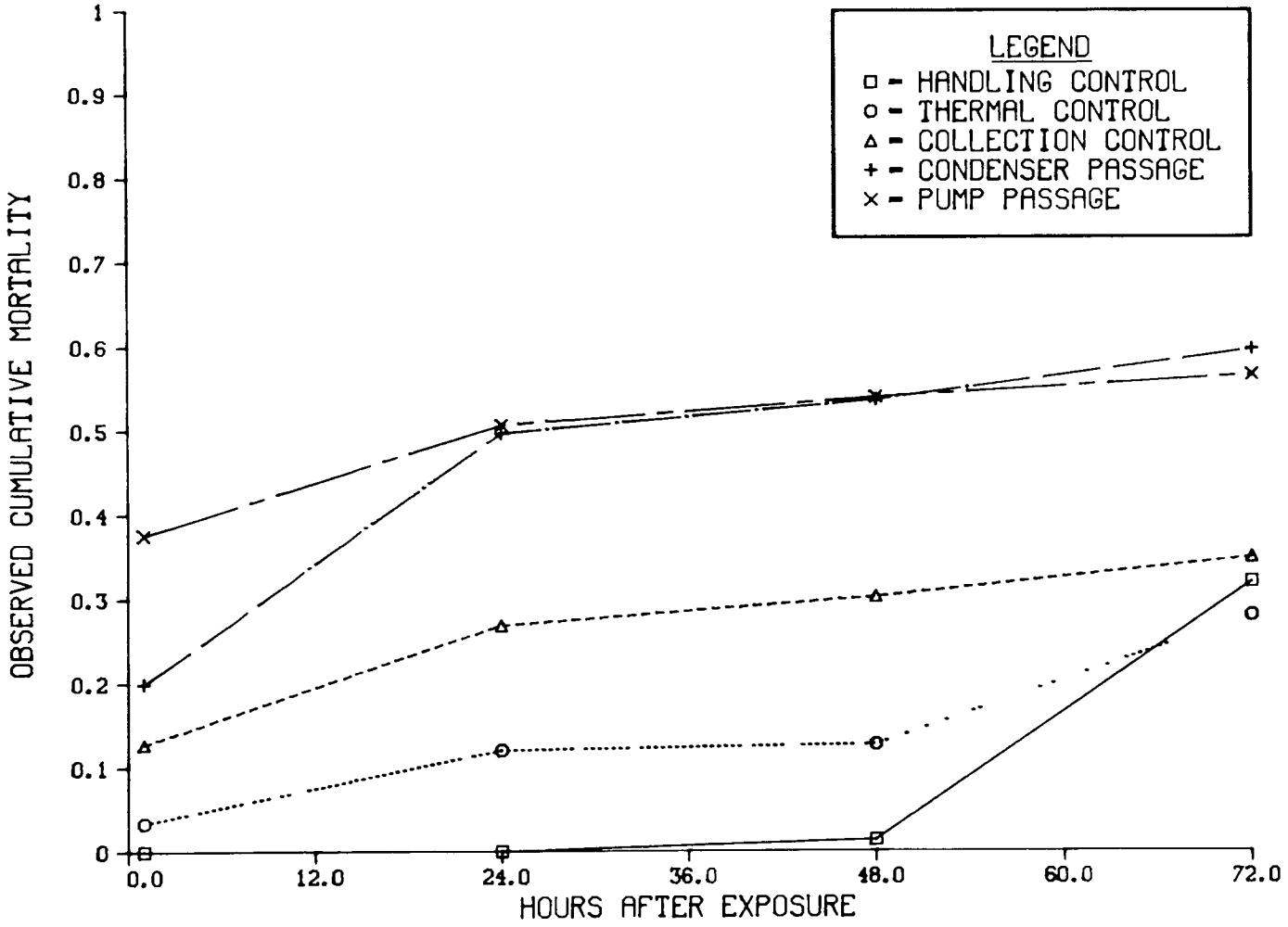

Fig. 14. Observed cumulative mortalities of bluegill sunfish larvae at various times after exposure to a ΔT of 10°C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types.

Table 20. Delayed (1-72 h) mortalities among bluegill sunfish (*Lepomis macrochirus*) exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator

Experiment type	ΔT(°C)	Pumping rate (% of maximum)	Number of fish[a] $(N_1)(1-P_1)$	Delayed mortality[b] $(\hat{d}_1)$	Standard[c] deviation	Critical[d] probability
Handling control	0	-	102	0.21	0.04	< 0.01
Thermal control	0	50	97	0.12	0.04	< 0.01
Collection control	0	50	99	0.16	0.04	< 0.01
Condenser passage	0	50	91	0.13	0.04	< 0.01
Pump passage	0	50	94	0.21	0.04	< 0.01
Handling control	0	-	150	0.32	0.04	< 0.01
Thermal control	10	50	145	0.26	0.04	< 0.01
Collection control	10	50	130	0.25	0.04	< 0.01
Condenser passage	10	50	121	0.50	0.05	< 0.01
Pump passage	10	50	95	0.31	0.05	< 0.01
Handling control	0	-	0	-	-	-
Thermal control	15	50	0	-	-	-
Collection control	15	50	0	-	-	-
Condenser passage	15	50	0	-	-	-
Pump passage	15	50	0	-	-	-

[a]Number of fish used in the estimation of delayed mortality, where N_1 = number of fish recovered in Experiment Type 1, and P_1 = proportion dead in Experiment Type 1 in the time interval (0-1 h).

[b]$\hat{d}_1$ = estimated delayed mortality for Experiment Type 1 (see Materials and Methods).

[c]Standard deviation of $\hat{d}_1$ (see Materials and Methods).

[d]Probability of observing the given value of $\hat{d}_1$ or greater if, in fact, d_1 is equal to zero.

Table 21 Comparisons of delayed mortalities among bluegill sunfish (*Leopmis macrochirus*) exposed to stresses in the Power Plant Simulator with delayed mortalities among bluegill sunfish exposed only to handling and natural stresses

Experiment type	$\Delta T(^{\circ}C)$	Pumping rate (% of maximum)	Delayed mortality[a] ($\hat{d}_i$)	Difference from handling control ($\hat{d}_i - \hat{d}_1$)	Standard[b] deviation	Critical[c] probability
Handling control	0	-	0 21	0 00	0.00	-
Thermal control	0	50	0.12	-0 08	0.05	0 06
Collection control	0	50	0 16	-0 04	0 05	0 21
Condenser passage	0	50	0.13	-0 07	0.05	0.08
Pump passage	0	50	0 21	< 0.01	0 06	0.45
Handling control	0	-	0.32	0 00	0.00	-
Thermal control	10	50	0 26	-0 06	0 05	0 11
Collection control	10	50	0 25	-0.07	0 05	0 11
Condenser passage	10	50	0 50	0 18	0 06	< 0 01
Pump passage	10	50	0 31	-0 01	0.06	0.40
Handling control	0	-	0 28	0 00	0.00	-
Thermal control	15	50	-	-	-	-
Collection control	15	50	-	-	-	-
Condenser passage	15	50	-	-	-	-
Pump passage	15	50	-	-	-	-

[a] $\hat{d}_i$ = estimated delayed mortality for Experiment Type i, where i = 1,2,3,4,5 Experiment Type 1 = Handling Control, Experiment Type 2 = Thermal Control, Experiment Type 3 = Collection Control, Experiment Type 4 = Condenser Passage, Experiment Type 5 = Pump Passage

[b] Standard deviation of $(\hat{d}_i - \hat{d}_1) = (V(\hat{d}_i) + V(\hat{d}_1))^{1/2}$ See Materials and Methods

[c] Probability of observing the absolute value of $(\hat{d}_i - \hat{d}_1)$ or greater if, in fact, $(d_i - d_1)$ is equal to zero.

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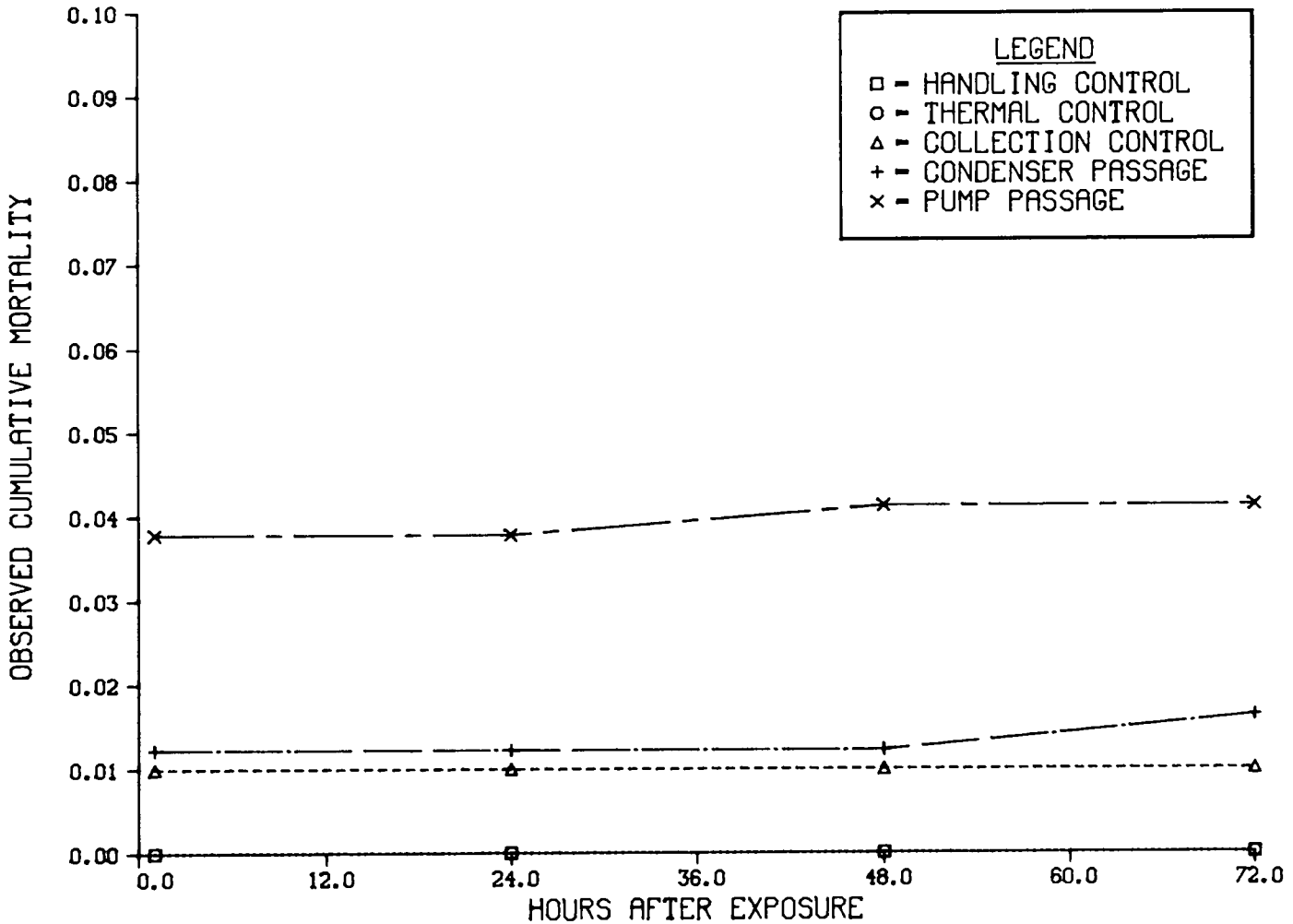

Fig. 15. Observed cumulative mortalities of channel catfish larvae at various times after exposure to a ΔT of 0°C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types.

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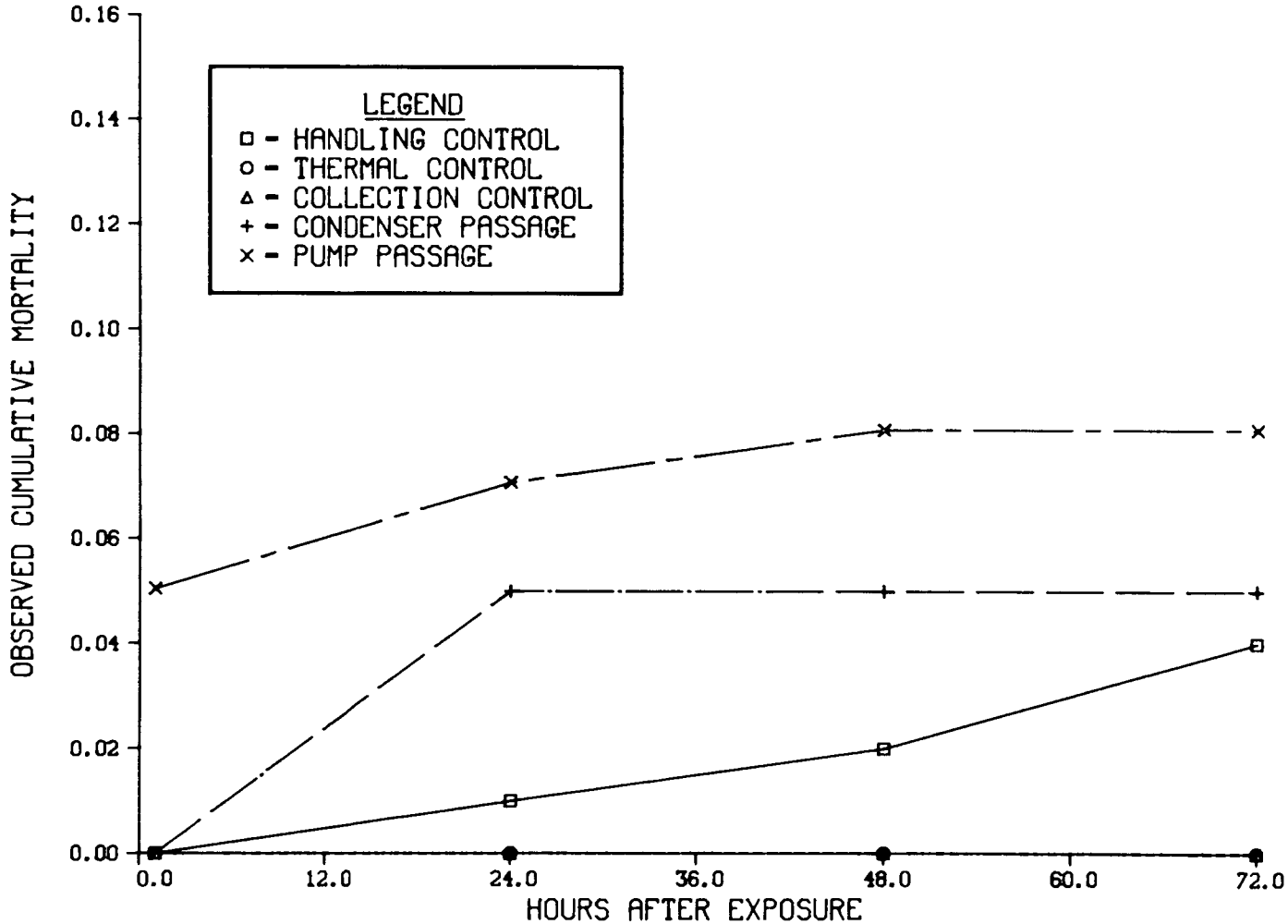

Fig. 16. Observed cumulative mortalities of channel catfish larvae at various times after exposure to a ΔT of 0 °C and a pumping rate of 91% of maximum. Refer to the text for a description of the experiment types.

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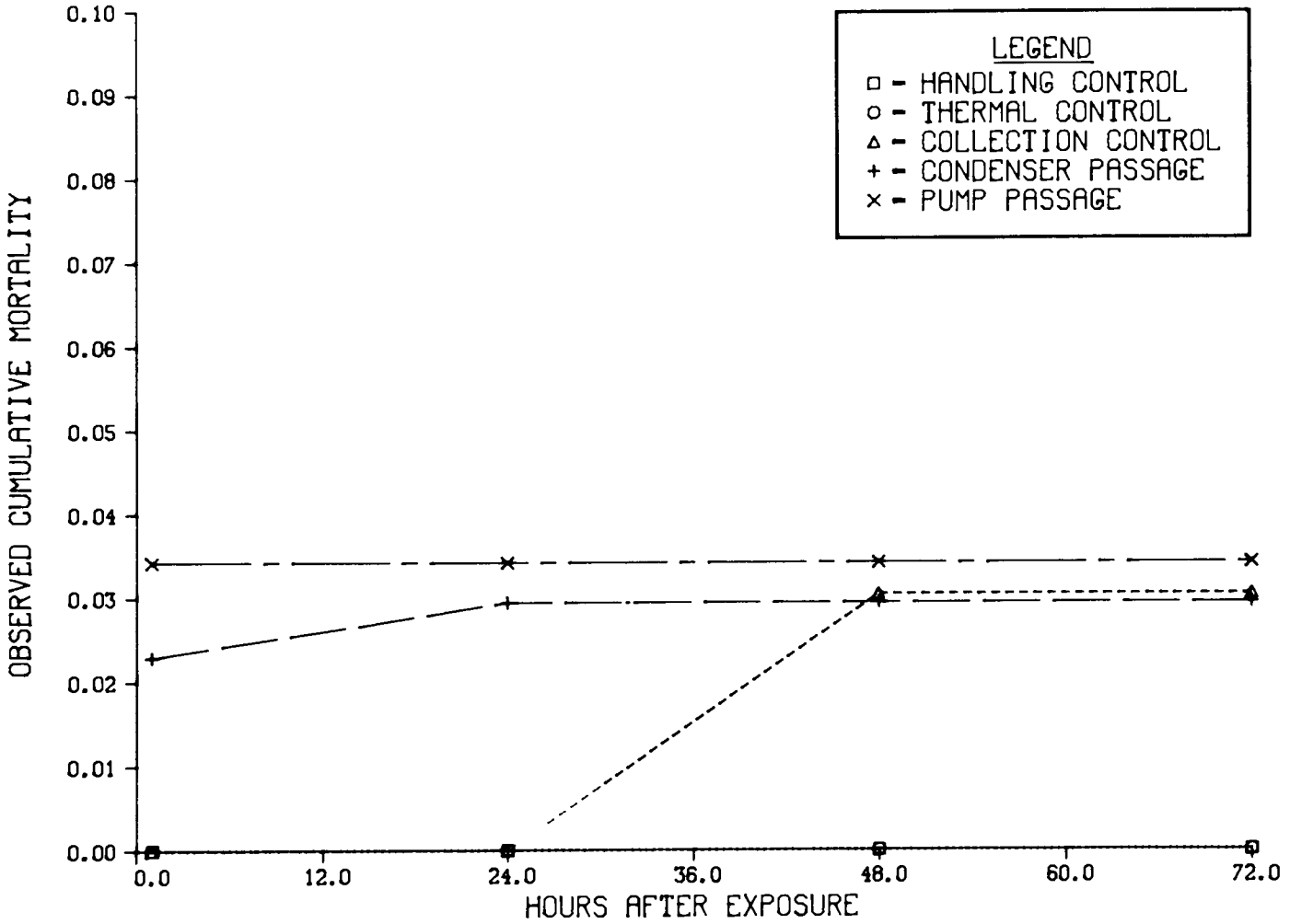

Fig. 17. Observed cumulative mortalities of channel catfish larvae at various times after exposure to a ΔT of 10°C and a pumping rate of 50% of maximum. Refer to the text for a description of the experiment types.

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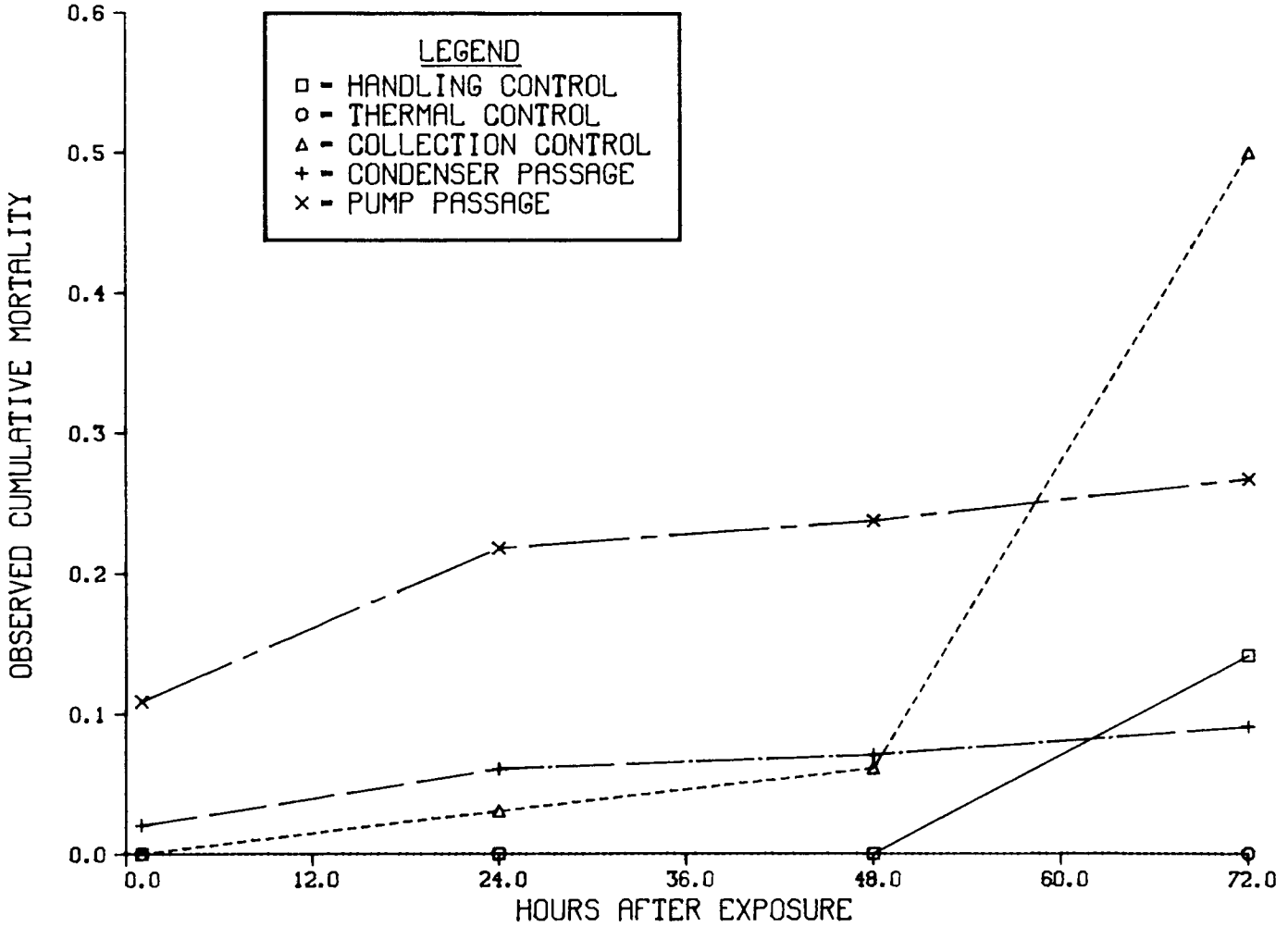

Fig. 18. Observed cumulative mortalities of channel catfish larvae at various times after exposure to a ΔT of 10°C and a pumping rate of 91% of maximum. Refer to the text for a description of the experiment types.

the pump-passage group exhibited the highest cumulative mortalities of all experiment types under the conditions depicted in Fig. 15 through 17, and at many of the post-test observation periods the condenser-passage group also exhibited greater cumulative mortalities than the controls.

The relatively high cumulative delayed mortality of the collection control group in Figure 18, which surpassed that of the pump passage group at the final observation period, may not be representative of the true delayed collection mortality under these conditions. Values plotted for collection control mortality are the combination of two replicates (Table 3), one of which had a total mortality of only 0.04 at the end of the holding period. For unknown reasons, however, 42 of the 49 fish used in the other replicate of this experimental group (which was run on the same date) died between the 48-h and 72-h observation periods. It is likely that the catfish in this replicate died for reasons other than the delayed effects of collection stresses, in view of the relatively low mortalities suffered by catfish in the other replicate of these test conditions and under all other test conditions (Figs. 15 through 17).

Estimates of delayed mortality (d_1) of channel catfish are given in Table 22. At a given temperature, the magnitudes of both $\hat{d}_4$ and $\hat{d}_5$ (delayed mortalities in the condenser- and pump-passage groups, respectively) were directly related to the pumping rate. On the other hand, $\hat{d}_3$, $\hat{d}_4$, and $\hat{d}_5$ all increased with increased temperature at a pumping rate of 91%. Delayed mortality in the collection control group ($\hat{d}_3$) also increased with increased temperature at a pumping

Table 22 Delayed (1-72 h) mortalities among channel catfish (Ictalurus punctatus) exposed to combinations of excess temperature and pumping rates in the Power Plant Simulator

Experiment type	ΔT(°C)	Pumping rate (% of maximum)	Number of fish[a] $(N_1)(1-P_1)$	Delayed mortality[b] $(\hat{d}_1)$	Standard[c] deviation	Critical[d] probability
Handling control	0	-	249	0 00	0 00	-
Thermal control	0	50	300	0 00	0 00	-
Collection control	0	50	298	0.00	0 00	-
Condenser passage	0	50	242	< 0.01	< 0 01	0 16
Pump passage	0	50	280	< 0 01	< 0 01	0 16
Handling control	0	-	100	0.04	0.02	0 02
Thermal control	0	91	100	0.00	0 00	-
Collection control	0	91	99	0.00	0.00	-
Condenser passage	0	91	100	0 05	0.02	< 0 01
Pump passage	0	91	94	0 03	0.02	0 04
Handling control	0	-	300	0 00	0 00	-
Thermal control	10	50	300	0 00	0.00	-
Collection control	10	50	295	0.03	0 01	< 0 01
Condenser passage	10	50	298	< 0 01	< 0 01	0 08
Pump passage	10	50	282	0.00	0 00	-
Handling control	0	-	99	0.14	0.03	< 0 01
Thermal control	10	91	100	0.00	0.00	-
Collection control	10	91	98	0.50	0.05	< 0.01
Condenser passage	10	91	97	0.07	0 03	< 0 01
Pump passage	10	91	90	0 18	0 04	< 0 01
Handling control	0	-	50	0.00	0 00	-
Thermal control	15	50	0	-	-	-
Collection control	15	50	0	-	-	-
Condenser passage	15	50	0	-	-	-
Pump passage	15	50	0	-	-	-

[a]Number of fish used in the estimation of delayed mortality, where N_1 = number of fish recovered in Experiment Type 1, and P_1 = proportion dead in Experiment Type 1 in the time interval (0-1 h)

[b]$\hat{d}_1$ = estimated delayed mortality for Experiment Type 1 (see Materials and Methods)

[c]Standard deviation of $\hat{d}_1$ (see Materials and Methods)

[d]Probability of observing the given value of $\hat{d}_1$ or greater if, in fact, d_1 is equal to zero

rate of 50%, but $\hat{d}_4$ and $\hat{d}_5$ were not greatly affected (Table 22). $\hat{d}_2$, the delayed mortality in the thermal control group, was zero under all conditions tested.

Table 23 presents the results of comparisons between the delayed mortalities of channel catfish exposed to stresses in the PPS ($\hat{d}_2$, $\hat{d}_3$, $\hat{d}_4$, and $\hat{d}_5$) and those exposed only to handling and natural stresses ($\hat{d}_1$). At the α = 0.05 level, d_2 was significantly less than $\hat{d}_1$ in two comparisons. $\hat{d}_3$ was significantly less than $\hat{d}_1$ at a ΔT of 0°C and pumping rate of 91%, but was significantly greater than d_1 in the tests conducted at a ΔT of 10°C. Delayed mortalities in the condenser-passage groups ($\hat{d}_4$) and the pump-passage groups ($\hat{d}_5$) were never significantly greater than delayed mortalities in the handling control at the α = 0.05 level.

In summary, continued mortality was observed among all species following the 1-h observation period. In 22 of the 31 comparisons involving fishes in the condenser-passage and pump-passage experimental groups, these delayed mortalities were significantly greater than zero. Further, delayed mortalities among these 22 were significantly greater than handling control delayed mortalities in 7 comparisons. For most condenser-passage and pump-passage experimental groups, delayed mortalities ($\hat{d}_1$) followed the same patterns as observed for short-term mortalities (P_1), that is, increased experimental stresses tended to result in increases in both P_1 and $\hat{d}_1$.

Table 23. Comparisons of delayed mortalities among channel catfish (Ictalurus punctatus) exposed to stresses in the Power Plant Simulator with delayed mortalities among channel catfish exposed only to handling and natural stresses

Experiment type	$\Delta T(^{\circ}C)$	Pumping rate (% of maximum)	Delayed mortality[a] ($\hat{d}_i$)	Difference from handling control ($\hat{d}_i - \hat{d}_1$)	Standard[b] deviation	Critical[c] probability
Handling control	0	-	0 00	0 00	0.00	-
Thermal control	0	50	0.00	0.00	0 00	-
Collection control	0	50	0 00	0 00	0.00	-
Condenser passage	0	50	< 0 01	< 0.01	< 0 01	0 16
Pump passage	0	50	< 0.01	< 0 01	< 0 01	0 16
Handling control	0	-	0 04	0.00	0.00	-
Thermal control	0	91	0.00	-0 04	0.02	0 02
Collection control	0	91	0 00	-0 04	0.02	0 02
Condenser passage	0	91	0 05	0.01	0.03	0 37
Pump passage	0	91	0.03	-0 01	0.02	0 38
Handling control	0	-	0 00	0.00	0 00	-
Thermal control	10	50	0 00	0.00	0.00	-
Collection control	10	50	0 03	0.03	< 0.01	< 0 01
Condenser passage	10	50	< 0 01	< 0 01	< 0.01	0.08
Pump passage	10	50	0.00	0 00	0.00	-
Handling control	0	-	0.14	0 00	0.00	-
Thermal control	10	91	0 00	-0 14	0.03	< 0 01
Collection control	10	91	0.50	0.36	0.06	< 0 01
Condenser passage	10	91	0 07	-0.07	0.04	0 06
Pump passage	10	91	0.18	0.04	0.05	0 25
Handling control	0	-	0 00	0 00	0.00	-
Thermal control	15	50	-	-	-	-
Collection control	15	50	-	-	-	-
Condenser passage	15	50	-	-	-	-
Pump passage	15	50	-	-	-	-

[a] $\hat{d}_i$ = estimated delayed mortality for Experiment Type i, where i = 1,2,3,4,5 Experiment Type 1 = Handling Control, Experiment Type 2 = Thermal Control, Experiment Type 3 = Collection Control, Experiment Type 4 = Condenser Passage, Experiment Type 5 = Pump Passage

[b] Standard deviation of $(\hat{d}_i - \hat{d}_1) = (V(\hat{d}_i) + V(\hat{d}_1))^{1/2}$ See Materials and Methods

[c] Probability of observing the absolute value of $(\hat{d}_i - \hat{d}_1)$ or greater if, in fact, $(d_i - d_1)$ is equal to zero

5. DISCUSSION

The results of the 1979 experimental program demonstrated wide differences among larval fish species in their sensitivity to stresses of condenser and pump passage. Small, fragile fish, such as the carp and largemouth bass larvae used in these studies, suffered high short-term mortalities as a result of the stresses of passage through the pipes and condenser tubes. Other species (smallmouth bass and channel catfish) exhibited relatively low mortalities following condenser passage in the PPS, even when accompanied by thermal shocks of 10°C above ambient. For most species, short-term conditional mortalities resulting from the physical stresses of pipe and condenser passage increased with increasing ΔT and/or pumping rate.

The short-term mortality responses to the effects of pump passage were relatively low in most of these tests. With a single exception, short-term conditional mortalities were less than 0.10 (Table 12). In the case of smallmouth bass and channel catfish larvae, overall short-term mortalities (P_5) in the pump passage groups were low, and as a result the conditional mortality estimates due to the effects of pump passage ($\hat{m}_5$) were also relatively low. Carp and largemouth bass larvae, on the other hand, exhibited relatively high short-term mortalities in the pump-passage group. However, because of high background mortalities, i. e., mortalities due to the effects of collection and/or pipe and condenser passage, additional short-term conditional mortalities due to pump passage could not be distinguished for these two species in most cases.

Studies of fish mortality resulting from passage through hydroelectric turbines indicated the mortality was directly related to runner speed and inversely related to turbine efficiency (Turbak et al. 1980). Although other sources of turbine mortality may be important at specific installations, runner speed and turbine efficiency were primary factors for both Francis and Kaplan turbines. If these same factors apply to power plant pumps, then the conditional mortalities observed from PPS pump passage would largely depend on high blade speeds (pumping rate of 91% of maximum) or low pump efficiencies (pumping rate of 50% of maximum). Although pump passage mortality in general was relatively low (Table 12), there was some indication from the channel catfish tests that mortality was higher at the higher pump speeds. More information is needed to clarify the relative importance of blade speeds and pump efficiencies to mortality of entrainable fishes.

Delayed mortalities among fishes passed through the pipes and condenser ($\hat{d}_4$) generally followed the same size-dependent pattern as short-term mortalities, i.e., relatively higher delayed mortalities were often observed for the smaller species tested. The instance in which $\hat{d}_4$ for carp was zero (Table 14) was probably not indicative of an absence of delayed mortality, but rather the result of a very high short-term mortality (P_4 = 0.96) which left only two individuals for subsequent observations. Carp larvae in these tests displayed a sensitivity to the mechanical stresses of pipe and condenser passage which, if not exhibited as a short-term mortality response, was eventually manifested as relatively high delayed mortality. In contrast,

channel catfish were shown to be less sensitive to pipe and condenser passage stresses since both short-term and delayed mortalities in the condenser passage groups were low. Delayed mortalities among largemouth bass, smallmouth bass, and bluegill sunfish larvae in the condenser passage groups were generally relatively high.

Comparisons of the proportion of condenser-passed fish that died 1 h after the experiment (P_4) to the proportion of initial survivors that died between 1 and 72 h after the experiment ($\hat{d}_4$) reveal different trends for different species. For bluegill sunfish, smallmouth bass, and largemouth bass, the higher the short-term mortality (P_4), the higher was the delayed mortality ($\hat{d}_4$). The opposite was observed for carp, although as noted earlier the high short-term mortality precluded a reliable assessment of delayed mortality under certain test conditions. Both P_4 and $\hat{d}_4$ were relatively low for channel catfish, and no consistent relationship between these two mortality estimates was evident.

Delayed mortalities among larval fishes in the pump-passage experimental groups followed much the same pattern as those in the condenser-passage groups. Values of $\hat{d}_5$ were relatively high in most cases for carp, bluegill sunfish, largemouth bass, and smallmouth bass. On the other hand, delayed mortalities of channel catfish exceeded 0.03 only at a ΔT of 10°C and a pumping rate of 91%. A direct relationship was observed between the magnitudes of short-term mortalities (P_5) and delayed mortalities ($\hat{d}_5$) for each of the five species. That is, increased experimental stresses (elevated temperature and/or pumping rate) led not only to increased short-term

mortalities but also to increased delayed mortalities among fishes passed through the entire PPS system.

The relative effects on mortality of the various entrainment stresses simulated in the PPS can be assessed by examining the conditional mortality rates due to these stresses. For example, the short-term mortality response to an excess temperature shock of 10°C is estimated by $\hat{m}_2$, calculated from data for larvae exposed to water in the PPS reservoir set at a ΔT of 10°C. The physical effects of pipe and condenser passage, on the other hand, can be estimated by $\hat{m}_4$ calculated from data for larvae passed through the PPS at ambient water temperatures. The data given in Tables 6 through 9 show that for these species the short-term mortalities due to thermal stress ($\hat{m}_2$ at a ΔT of 10°C) and physical stress ($\hat{m}_4$ and $\hat{m}_5$ at a ΔT of 0°C), acting separately, were relatively low and roughly equivalent. Carp larvae (Table 5) were the only test organisms which exhibited relatively high short-term mortalities due to physical stresses at ambient temperatures. Smallmouth bass larvae were not tested at ambient water temperatures (Table 7), but values of m_4 and m_5 were low for this species even at elevated water temperatures.

Various investigators (Coutant and Kedl 1975, Marcy et al. 1978, O'Connor and Poje 1979) have speculated that circulating water pumps are major sources of physical damage to organisms entrained at steam electric power plants. These hypotheses have not been borne out, however, by the results of the 1979 PPS experiments. Short-term conditional mortalities due to pump passage (Table 12) were generally relatively low and in many cases not significantly greater than zero at

the α = 0.05 level. Although obvious physical damage (decapitation, mutilation) was occasionally noted, in most cases the conditional mortalities due to pump passage were even less than would be predicted (Ebey and Beauchamp 1977, Suffern 1977) based on probability of contact with the pump impeller blade.

The results of tests designed to ascertain the effects of different pumping efficiencies on the survival of pump-passed larvae were contrary to expectations. It was hypothesized that the lowest pump-induced mortalities would occur at the highest pump efficiency (pumping rate of 91% of maximum), and that effects of increased turbulence resulting from lower pump efficiencies (= lower pumping rate) would be manifested as increased $\hat{m}_5$'s. In fact, it was found that higher pumping rates resulted in higher short-term conditional mortalities for all species (Table 12). These results must be interpreted with caution, however, in light of the high collection mortalities that were noted among some species (Table 10). It was observed that much of the water flowing into the conical collection net was filtered through a restricted region of the net opposite the point where the descending run of piping entered the reservoir (Fig. 1). The nonuniformity of flow through the filtering surface caused through-mesh velocities in this region to be higher than anticipated and probably resulted in the significant collection mortalities observed among smaller, more fragile larvae (carp, largemouth carp, bluegill sunfish).

Future studies with the Power Plant Simulator will examine both direct and indirect mortality of entrainable fishes. Direct mortality experiments will be a continuation of the 1979 work and will concentrate on refining the estimates of conditional mortalities resulting

from the different entrainment stresses. Potential synergistic interactions between thermal and physical stresses were suggested by the 1979 data for some species, but could not be adequately tested statistically due to the small number of replicates. However, we believe that synergistic effects may play a major role in overall entrainment mortality, and this factor will be studied in greater depth in 1981.

Van Winkle et al. (1979) have suggested that indirect mortality may be an important, although often overlooked, component of entrainment mortality. They defined indirect mortality as the mortality of organisms that survive the entrainment experience itself but that die prematurely due to sublethal effects of entrainment (e.g., increased vulnerability to predation, disease, parasitism, or the effects of toxic materials, and a reduction in competitive ability or propensity to feed). In addition, Van Winkle et al. (1979) provided the results of preliminary experiments that suggested that striped bass larvae may be less likely to feed following sublethal heat shocks.

Since indirect entrainment mortality is virtually impossible to study in the field, laboratory experiments provide the only means of quantifying and assessing the significance of this potential problem. The PPS can be of exceptional value in these efforts by allowing us to deliver thermal and/or physical stresses to organisms and subsequently to examine the sources of indirect mortality in a controlled laboratory setting. Effects of sublethal entrainment stresses on the vulnerability of fish larvae to predation will be the focus of future indirect mortality studies. In these experiments, fish which survive physical and/or thermal stresses in the PPS will be mixed with unstressed,

control fish and offered to predators in laboratory tanks (Coutant 1973, Kania and O'Hara 1974, Sullivan et al. 1978). The relative degrees of predation upon the stressed and unstressed groups will then be examined for statistically significant differences. Other sources of indirect mortality, such as reduced feeding efficiency (Van Winkle et al. 1979), could be studied as time allows.

In summary, conditional mortalities of larval fishes resulting from the various thermal and physical stresses of power plant entrainment revealed a wide range of tolerance among different species. In addition, susceptibility to these stresses appeared to be inversely related to size, although the analysis of size-mortality relationships in these tests is complicated by species-specific differences in sensitivity to entrainment. Unlike the domination of physical components of entrainment stress suggested by field studies (Beck et al. 1978), short-term conditional mortalities calculated from PPS data indicated that, within the range of conditions tested, the relative effects of individual sources of stress (thermal stress, physical stress from pump, pipe, and condenser passage, and collection techniques) were roughly equivalent for a given species. In addition, the results of the 1979 experimental program suggest that although a single factor, such as thermal shock or shear forces accompanying condenser tube passage, may not have a major, direct effect, its influence may still be exerted in some species through interaction with other stress factors that are a part of the entrainment experience.

6. SUMMARY AND CONCLUSIONS

1. The ORNL Power Plant Simulator was used to assess larval fish entrainment mortality resulting from the stresses of excess temperatures, pipe and condenser passage, and pump passage. The 1979 experimental program consisted of exposing the larvae of carp, largemouth bass, smallmouth bass, bluegill sunfish, and channel catfish to different combinations of pumping rates and excess water temperatures in the PPS.
2. Recoveries of test fishes from the PPS and post-test holding facilities were greater than 95 percent in the majority of cases. Differences in recoveries between different experimental types were not statistically significant, but recovery percentages were significantly greater for the largest species tested (bluegill sunfish and channel catfish) than for the other three species.
3. Short-term (0-1 h) conditional mortalities due to the stresses of pipe and condenser passage were relatively high for carp and largemouth bass and consistently low for smallmouth bass and channel catfish. Conditional mortalities resulting from pump passage were low in most cases in which additional mortalities could be detected.
4. Delayed (1-72 h) mortalities following exposure to mechanical stresses in the PPS were relatively high for carp, largemouth bass, smallmouth bass, and bluegill sunfish. Channel catfish, on the other hand, exhibited low delayed mortalities under most experimental conditions.

5. The data from the 1979 experimental program showed a wide range of sensitivity to entrainment stresses among different fish species. Further, short-term conditional mortalities calculated from these experiments suggest that, within the range of conditions tested, the relative effects of individual sources of thermal, physical, and collection stresses were roughly equivalent for a given species.

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