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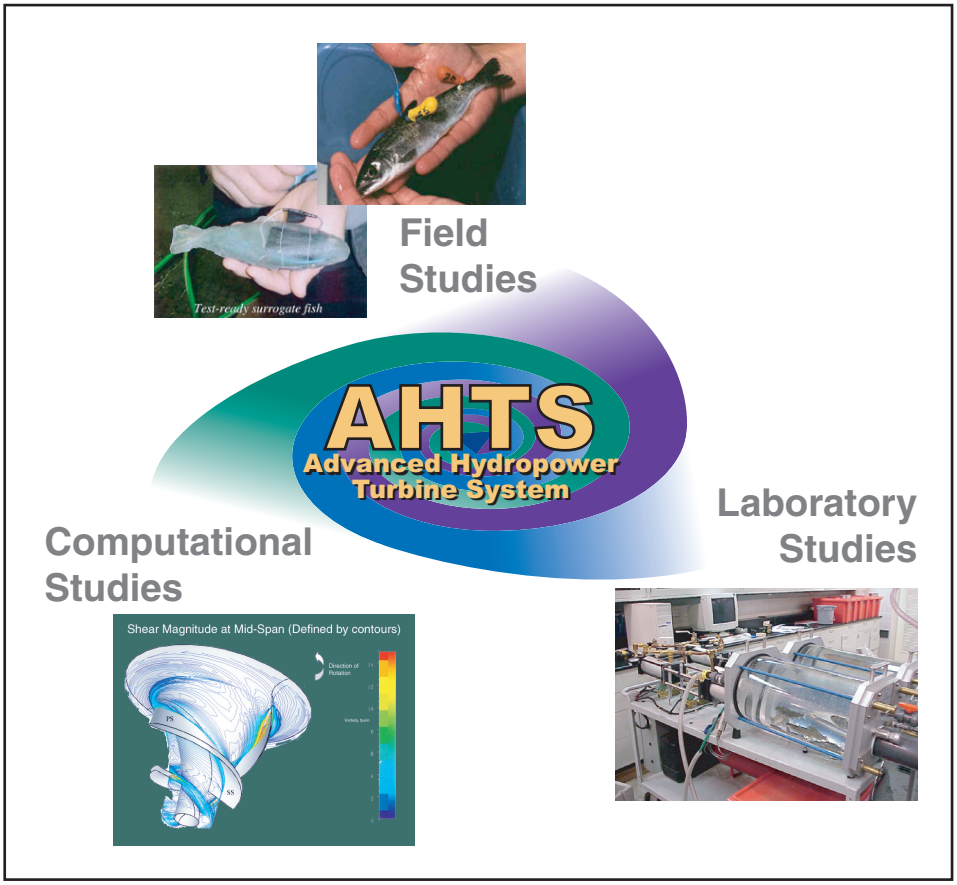
Laboratory Studies of the Effects of Pressure and Dissolved Gas Supersaturation on Turbine-Passed Fish



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Laboratory Studies of the Effects of Pressure and Dissolved Gas Supersaturation on Turbine-Passed Fish

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Executive Summary

The U.S. Department of Energy's Advanced Hydropower Turbine Systems (AHTS) Program supports development of environmentally friendly turbines, i.e., turbine systems in which attributes such as fish passage survival are emphasized. It is expected that these advanced turbines could permit the efficient generation of electricity while minimizing the damage to fish and their habitats.

Designing advanced turbine systems requires knowledge of environmental conditions that injure or kill fish such as the stresses associated with hydroelectric power production, including pressure changes fish experience during turbine passage and dissolved gas supersaturation (resulting from the release of water from the spillway).

The objective of this study was to examine the relative importance of pressure changes as a source of turbine-passage injury and mortality. Specific tests were designed to quantify the response of fish to rapid pressure changes typical of turbine passage, with and without the complication of the fish being acclimated to gas supersaturated water.

We investigated the responses of rainbow trout (*Oncorhynchus mykiss*), chinook salmon (*O. tshawytscha*), and bluegill sunfish (*Lepomis macrochirus*) to these two stresses, both singly and in combination.

Based on results of our laboratory studies, we reached the following conclusions:

- The gas supersaturation level that causes acute gas bubble trauma (GBT) varies among species. Resistance to acute GBT, from greatest to least, is bluegill >fall chinook salmon > rainbow trout. Bluegills also had a lower incidence of chronic GBT symptoms than did fall chinook salmon and rainbow trout.

- The frequency, type, and severity of injuries related to pressure changes during turbine passage vary among species.

  - Bluegills, and presumably most physoclistous fish, are extremely susceptible to swim bladder rupture when exposed to the sudden pressure change during turbine passage. The total dissolved gas level had only a small additive effect on the injury/death rate due to the pressure spike.

  - Fall chinook salmon suffered ruptured swim bladders, but at a much lower rate than bluegills. When acclimated to elevated gas levels at 191 kPa, the turbine passage sequence also caused instantaneous bubble formation in a small number of fish, resulting in immediate death.

  - Swim bladder rupture was not observed in rainbow trout, regardless of total dissolved gas (TDG) level or acclimation pressure.

- If dissolved gas supersaturation is not a problem, our experiments suggest that the brief low pressure spike to about 0.1 atmosphere downstream from the turbine runner will cause little direct mortality among surface-acclimated salmonids. If fish are entrained from greater depths, such that their swim bladders contain more gas and will expand more during the low pressure spike, the injury and mortality rates will be higher.

- Injury/mortality rates would likely be reduced or eliminated if the nadir of the turbine pressure spike was higher, as is expected to be the case with new fish-friendly turbine designs. A follow-up series of tests is needed under a modified pressure regime that more closely reflects conditions expected in new turbine designs, or with a nadir of ~50 kPa.

- The low pressure spike is especially a problem if the water is highly supersaturated with gases (well beyond water quality standards), and the fish respond to the supersaturation by depth compensation.

Acknowledgments

Many people contributed to this project. Peggy Brookshier, Chair of the U.S. Department of Energy Advanced Hydropower Turbine System (AHTS) Program, and John Flynn, Office of Biopower and Hydropower Technologies, U.S. Department of Energy, provided guidance and support. Mike Sale of the Oak Ridge National Laboratory helped develop the project and acted as a reviewer. Dr. Ralph Elston, Aquatechnics, Inc., provided valuable technical assistance and training in monitoring and describing gas bubble disease in fish. Susan Blanton and Joanne Duncan of Pacific Northwest National Laboratory (PNNL) provided technical assistance, and Dennis Dauble and Duane Neitzel, PNNL, provided comments and advice during the development of the methods and protocols used to accomplish this work and reviewed draft reports. We appreciate the valuable guidance and reviews of the AHTS Technical Committee, an advisory group of biologists and engineers who represent federal agencies, Tribes, and the hydropower industry.

The work was conducted at the Pacific Northwest National Laboratory (PNNL) in Richland, Washington. PNNL is managed by Battelle Memorial Institute for the U.S. Department of Energy. The work was conducted under a Related Services Agreement with the U.S. Department of Energy, Contract DE-AC06-76RL01830.

Glossary

AHTS	Advanced Hydropower Turbine System Program of the U.S. Department of Energy
bulb turbine	An axial flow turbine often used in low-head applications. In large units the generator is housed within the submerged bulb and is driven by a variable pitch propeller at the trailing end of the bulb.
Francis turbine	A radial-inflow reaction turbine, where the flow through the runner is radial to the shaft
GBT	Gas bubble trauma
gpm	Gallons per minute
Kaplan turbine	An axial-flow (propeller-type) turbine with adjustable runner blades and adjustable guide vanes
kPa	Kilopascals; a measure of pressure. 101 kPa = 1 atmosphere = 14.73 psi
nadir	The lowest pressure in the time versus pressure regime experienced by fish in these experiments
physoclistous	Fish that lack a direct connection (pneumatic duct) between the swim bladder and the esophagus. These species must adjust the pressures within the swim bladder by the relatively slow process of diffusion of gases from the blood.
physostomous fish	Fish that have a duct (pneumatic duct) that connects the swim bladder with the esophagus. In these species, gas can be quickly taken into or vented from the swim bladder through the duct, so that adjustment to changing water pressures can take place rapidly.
pneumatic duct	The duct that connects the swim (gas) bladder and the gut in physostomous fish
pressure spike	In these experiments, the rapid water pressure decrease from several times atmospheric pressure to a low point (nadir) of less than one atmosphere
psi	Pounds per square inch, a measure of pressure

spill	Passing water over the spillway of a hydroelectric dam, rather than through the turbine. Spill allows surface-oriented fish to move downstream without passing through the turbines, but it may also cause the river water to become super-saturated with air.
swim bladder	An internal gas bladder that has a weight-regulating (hydrostatic) function in higher fishes
TDG	Total dissolved gas
TGP	Total gas pressure
vapor pressure	Pressure exerted by a vapor when the vapor is in equilibrium with the liquid form of the same substance—i.e., when conditions are such that the substance can exist in both phases.

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1.0 Introduction

The U.S. Department of Energy's Advanced Hydropower Turbine System (AHTS) Program supports the development of "environmentally friendly" turbines, i.e., turbine systems in which environmental attributes such as fish passage survival are emphasized. Advanced turbines would be suitable for installation at new hydropower facilities and potentially suitable for replacing aging turbines at existing plants. It is expected that these turbines could permit the efficient generation of electricity while minimizing the damage to fish and their habitats.

A successful design program for the AHTS requires up-front, pre-design specifications for the environmental conditions that occur within the turbine system. Precise knowledge of conditions that kill fish will assist engineers in the design of an advanced turbine system. Two recent reviews of biological information provide the starting point for determining the environmental conditions that must be studied (Wittinger et al. 1995; Čada et al. 1997). Both reviews concluded that additional biological studies are needed to answer questions related to the potential direct and indirect effects of pressure stresses on fish during turbine passage under varying dissolved gas concentrations. To help answer these questions, the AHTS Program asked Pacific Northwest National Laboratory (PNNL) to study the effects of pressure and dissolved gas supersaturation on turbine-passed fish. Information from these studies will provide the basis for communicating biological specifications to turbine designers.

PNNL tested the responses of rainbow trout (*Oncorhynchus mykiss*), Chinook salmon (*O. tshawytscha*), and bluegill sunfish (*Leponis macrochirus*) to these stresses, singly and in combination, in the laboratory. Our overall objective was to examine the relative importance of pressure changes as a source of turbine-passage injury and mortality. Specific tests were designed to quantify the response of fish to rapid pressure changes typical of turbine passage, with and without the complication of the fish being acclimated to gas supersaturated water.

This report contains seven sections. In Section 2.0 we present background information on water pressure, fish responses to pressure changes, and complications of gas supersaturation. Methods and materials are described in Section 3.0. Results are presented in Section 4.0, discussion in 5.0, and conclusions in 6.0. References are provided in Section 7.0. Appendix A includes supplementary information.

2.0 Background

Changes in water pressure and dissolved gas concentrations can affect the survival of fish passing through turbines or inhabiting tailwaters of hydroelectric dams. Čada (1999) presented a summary of information on the pressure regimes and dissolved gas concentrations that might be expected at hydroelectric power plants, summarized past and ongoing studies, and suggested an experimental protocol for resolving the issue.

In natural, unimpounded rivers, fish experience water pressures ranging from near atmospheric (at the surface) to about 2 atmospheres (water pressure increases by 1 atmosphere with every 34-ft increase in depth). Dissolved gases are normally at or below 100% saturation; water is rarely supersaturated (>100% saturation) in natural rivers.

The construction of hydroelectric dams on river systems alters the range of both water pressures and dissolved gas concentrations to which riverine fish may be exposed (Čada 1999). Within the depths of the hydroelectric reservoir, fish may become acclimated to water pressures that are greater than any that occurred in the natural river. Fish entrained in the turbine intake flow experience rapid pressure increases and decreases of a greater magnitude than exists in nature. Dissolved gas concentrations may be lowered or raised by the presence of hydroelectric dams. For example, oxidation of organic matter in deep waters of a reservoir may lead to low oxygen concentrations in the hypolimnion and discharged waters. On the other hand, spilling water from dams often increases the concentration of dissolved gases. Although this may have an important benefit by increasing the amount of dissolved oxygen, water spilled from high dams into deep plunge pools can become supersaturated with dissolved air and cause gas bubble trauma (gas bubble disease) in fish. Passage of water through a conventional hydroelectric turbine does not increase the amount of dissolved gas (because it is a closed system), but the percent saturation changes as water pressures change in different portions of the turbine system. That is, even though the concentration of dissolved gas does not change in the closed system, surface water entering the gatewell that is equilibrium with the atmosphere will become undersaturated as water pressures increase approaching the runner, and then become briefly supersaturated in the subatmospheric pressures downstream from the runner. The turbine discharge returns to equilibrium with the atmosphere in the tailrace.

Water pressure is expressed in kilopascals (kPa), where 101.3 kPa = 1 atmosphere = 760 mm Hg = 14.73 psi. Water pressure increases with depth at a rate of 9.799 kPa/m (= 0.0294 atmosphere/ft = 0.434 psi/ft = 73.49 mm Hg/m). Thus, a fish residing at the water surface experiences about 101 kPa, whereas a fish residing at 10 m depth experiences a water pressure of about 200 kPa. Because fish can control their depth, they can ensure that the rate of pressure change they experience in natural rivers is small and not damaging.

Surface-acclimated fish that are entrained in the turbine flow experience increasing water pressures as they descend to the upstream side of the runner. This period of pressure increase may occur in a time frame measured in seconds to minutes, depending on whether the fish resists entrainment by swimming against the flow in the forebay and intake. As the fish passes through the runner, pressure drops very rapidly on the downstream side of the runner and into the draft tube, often to less than atmospheric

pressure. Occasionally, pressures downstream from the runner may momentarily drop below the vapor pressure of water, resulting in cavitation. Passage through this low pressure zone occurs in no more than a few seconds. After leaving the draft tube, the fish is again exposed to near atmospheric pressures at the surface of the tailrace or greater pressures if the fish swims to deeper water.

Čada (1990) reported that water pressures in one bulb turbine (horizontal propeller-type turbine) varied from a high of 210 kPa to a low of about 80 kPa. In such a turbine, a surface-acclimated fish (101 kPa) would experience a doubling of pressure upstream from the runner, followed by a momentary pressure decrease to about 80% of the acclimation pressure, all within as little as 15 seconds.

Bell (1991) calculated pressures near the Francis runners at the Lequille, Cushman No. 2, and Shasta hydroelectric plants. From the turbine entrance (i.e., leading edge of the runner blades, at mid-height of the wicket gates) to the bottom of the turbine (entrance to the draft tube), calculated pressures dropped from 582 to 56 kPa at Lequille, from 830 to 68 kPa at Cushman No. 2, and from 736 to 95 kPa at Shasta hydroelectric plant.

Calculated pressures experienced by fish passing through a Kaplan turbine at the McNary Dam on the Columbia River would be as high as 460 kPa (Montgomery Watson 1995). A fish passing along the upper surface of the turbine blade near the hub would be exposed to pressures that are estimated to be no lower than 115 kPa. The most damaging pressure changes would be experienced by fish entering at the ceiling depth and passing along the bottom side of the blade near its tip; in this case, pressures would drop from 340 kPa to 2 kPa in less than 1 sec. A value of 2 kPa is approximately the vapor pressure of water, i.e., the pressure at which cavitation occurs. Exposure to the lowest pressures, on the downstream side of the runner, were estimated to last no more than 0.25 sec before pressure rapidly returned to near atmospheric pressures in the draft tube and tailwaters.

2.1 Fish Responses to Pressure Changes

Among fish with swim bladders, the response to rapid pressure changes encountered within a turbine is affected by whether the fish is *physostomous* or *physoclistous*. Physostomous fish have a duct, the pneumatic duct, which connects the swim bladder with the esophagus. Gas can be quickly taken into or vented from the swim bladder through the mouth and pneumatic duct, so that adjustment to changing water pressures can take place rapidly, often on the order of seconds. As a general rule, physostomes include the soft-rayed fishes like salmon, trout, catfish, minnows, shad, and gar. On the other hand, physoclists lack a direct connection between the swim bladder and the esophagus. In these fish, the contents and pressures within the swim bladder must be adjusted by diffusion into the blood, a process measured on the order of hours. Physoclistous fish include many of the spiny-rayed fishes such as perch, bass, and bluegill sunfish.

Once inside a turbine, surface-acclimated physoclistous fish cannot adjust the volume of their swim bladder rapidly enough to compensate for changing water pressures; the swim bladder will be compressed and the fish will become more dense under increasing water pressures. Conversely, in a region of low pressure, downstream from the runner, the swim bladder will expand rapidly, potentially to the point of bursting. Physostomes have more control over the volume of gas in the swim bladder than physoclists. If

a deep-water-adapted physostome is drawn toward a surface intake, decreasing water pressure will cause the swim bladder to expand. Excess gas can be vented through the pneumatic duct if the rate of ascent is sufficiently slow. However, even physostomous fish may not be capable of venting excess gas in response to the rapid pressure reductions (often less than 1 sec) that occur within the turbine and draft tube.

Čada et al. (1997) reviewed a large number of experiments that examined the effects of pressure increases and decreases on fish. They concluded that pressure increases of the magnitude found in hydroelectric turbines are unlikely to injure or kill entrained fish. Rapid, brief pressure increases caused little or no direct mortality in a variety of studies using a variety of fish. However, high pressures may alter the behavior of fish such that they may have increased susceptibility to other, non-pressure-related sources of mortality. Some investigators have noted that fish exposed to high pressures were momentarily stunned. Although the test fish fully recovered in the laboratory holding tanks, temporarily stunned fish may be more susceptible to predators in the tailwaters of a hydroelectric dam.

The pressure decreases that fish experience downstream from the runner occur rapidly and may be large. From a direct mortality standpoint, laboratory studies suggest that the brief exposure to subatmospheric pressures within the turbine is more likely to be damaging to fish with swim bladders. Although there is considerable variation in the response of fish to pressure reductions, the highest mortalities occurred when the pressure reduction was greatest, i.e., when the exposure pressure was a relatively small fraction of the acclimation pressure. On the other hand, three tests in which exposure pressure was no less than 60% of the acclimation pressure resulted in little or no mortality. Based on these limited studies of a variety of fish, Čada et al. (1997) suggested that pressures within the turbine should fall to no less than 60% of the value to which entrained fish are acclimated. For surface-oriented fish, a pressure of 60 kPa or greater at all points within the turbine and draft tube would be expected to protect most fish from direct mortality resulting from low pressures.

Based on a consideration of salmonid data in USACE (1991), ARL (1996) suggested that minimum pressures within the turbine be no less than 30% of the fish's initial acclimation pressure. For fish distributed within the top 34 ft of water, this would dictate a minimum pressure of about 10 psi (69 kPa). ARL's suggested minimum pressure criterion (30% of acclimation) is less restrictive than the "60 percent of acclimation" criterion suggested by Čada et al. (1997). Whereas it may protect deep-adapted salmonids (and other physostomes) that are able to vent some of the expanding gases in the swim bladder as they are drawn upwards toward the intake, the 30% criterion may not be sufficient to protect other species of physoclistous fish.

2.2 Complications of Gas Supersaturation

Supersaturation of dissolved gases, leading to gas bubble trauma (gas bubble disease) has been most commonly reported at Columbia River Basin dams. However, it has also been observed at dams in the southeastern U.S. Air is entrained in water spilled from high dams as numerous small bubbles with a great increase in air-water interfacial area. The air in the bubbles dissolves under pressure in deep tailwater pools. When this water subsequently surfaces downstream, the gases are supersaturated relative to local atmospheric pressures. Once the bubbles have left the water, the rate to reach equilibrium is

considerably slower, so supersaturation may persist in flowing waters for days, and excessive dissolved gas levels may persist far from the source of supersaturation (APHA/AWWA/WEF 1995).

Montgomery Watson (1995) summarized the history of gas supersaturation problems in the Columbia River. Briefly, total gas pressures in the range of 115 to 143% have occurred in the Columbia and Snake rivers during periods of high spilling. Gas supersaturation problems were first identified in the 1960s, but declined in the 1970s and 1980s as a result of 1) installation of turbines, thereby reducing the amount of water that was spilled, 2) installation of flip-lip spill deflectors on some spillways, and 3) generally low levels of precipitation and runoff. With the increased use of spill in the 1990s to enhance downstream fish passage in the Columbia River Basin, gas supersaturation has once again become a problem. For example, during the emergency spill program of 1994, maximum total gas pressure ranged from 120 to 133%. Under heavy spill conditions that could occur under possible future reservoir drawdowns, total gas pressure could increase to as much as 140% (Montgomery Watson 1995).

In years of low flow, there may be considerable dilution of spilled (supersaturated) water by turbine flow, such that total gas pressure (TGP) may decrease downstream from the dams. Conversely, AASI (1998) notes that in years of high river flows, supersaturated spill flows will be much greater than turbine flows, so that dilution of supersaturated water in the mixing zone below the dam will be small. Total gas pressure may be elevated throughout the river (due to high spill-to-turbine flow ratios), leading to high TGP in the forebay of the next dam downstream.

To examine the phenomenon of entrainment of fish acclimated to supersaturated water, Montgomery Watson (1995) exposed smolt-size rainbow trout to different levels of water pressure and dissolved gas saturation in laboratory chambers. The pressure exposure system consisted of two acrylic cylinders, each 55 cm long and 27.5 cm in diameter, connected to a system of hydraulic and pneumatic cylinders and their controls and water supply (detailed schematics are provided in the report). The chambers were connected to hydraulic cylinders, which in turn were connected to pneumatic cylinders. A computer-controlled gas pressurization system caused the pneumatic cylinders to change the position of the hydraulic cylinders, thereby pressurizing or depressurizing the test chambers while maintaining control over dissolved gas concentrations. Pressure could be dropped from 300 kPa to the vapor pressure of water in 0.1 sec.

In the Montgomery Watson (1995) experiments, groups of age 0, 9- to 10-cm-long rainbow trout were exposed to the following pressure regime in the test chambers: Initial Pressurization Phase (atmospheric pressure to 300 kPa in 30 to 60 sec); Transient Phase (drop to the vapor pressure of water, 2 kPa, in 0.10 sec); Low Pressure Phase (close to the vapor pressure of water for 0.25 sec); and Recovery Phase (return to 115 to 120 kPa in 30 to 60 sec). This was estimated to be the worst-case pressure condition for a fish passing close to a turbine blade at McNary Dam. Groups of 20 test fish in each chamber were exposed to the pressure transients (and different gas saturations) and held in the chambers for an additional 30 min. After the 30 min were up, treatment and control fish were removed from the chambers, combined, and introduced to a tank containing adult rainbow trout predators. Some fish exposed to pressure changes at 125 and 130% of saturation lost equilibrium and/or died (direct mortality). However, differences between treatment and control groups in the mortality due to predation (indirect mortality) were not statistically significant.

As can be seen from the foregoing discussion, the changes in water pressure and dissolved gas concentrations associated with hydroelectric facilities could affect the survival of fish passing through turbines or inhabiting tailwaters below the dams. The U.S. Department of Energy's Hydropower Program supported experiments that examined the injury and mortality to fish caused by these stresses. This report presents the results of the initial set of test conditions.

3.0 Methods and Materials

Čada (1999) suggested 12 possible test conditions for resolving pressure-related concerns about fish passage at hydropower projects throughout the United States. In six of the conditions, the nadir of the turbine pressure spike was "near zero" (0 kPa) as might occur at dams on the lower Columbia River operating at high efficiency, whereas the other six conditions would simulate a less severe nadir (somewhere between 0 and 101 kPa) representing turbines operating at reduced efficiency. These test conditions are explained below and prioritized in Table 3.1. We conducted the tests in November and December 1999, January and February 2000, and June and July 2000.

3.1 Test Species

Rainbow trout ~13 cm in length were obtained from Troutlodge, Inc. in Soap Lake, Washington. Bluegills ~7 to 10 cm in length were purchased from Osage Catfisheries, Osage Beach, Missouri. Eyed fall chinook salmon eggs were obtained from the Washington Department of Fish and Wildlife's Priest Rapids Hatchery on the Columbia River near Mattawa, Washington. The eggs were hatched and reared at PNNL's aquatic lab until the juvenile fall chinook salmon reached ~10 cm in length.

3.2 Test Conditions

In an early experiment to examine the phenomenon of entrainment of fish acclimated to supersaturated water, Montgomery Watson (1995) exposed smolt-size rainbow trout to different levels of water pressure and dissolved gas saturation in laboratory chambers. Our experiments used the same hyperbaric chambers and experimental procedures, and are an expansion of the experiment in the following areas:

- **Test a greater number of species**. Montgomery Watson (1995) tested only rainbow trout. We conducted tests with rainbow trout, fall chinook salmon, and bluegill, with future plans to test American shad (*Alosa sapidissima*), Pacific lamprey (*Entosphenus tridentatus*), and other game fish species.

- **Expand the range of turbine-passage pressures**. Montgomery Watson (1995) tested a "typical" turbine-passage pressure regime, i.e., what would be expected at McNary Dam turbines (test conditions 2, 6, and 10 in Table 3.1). This range will be modified to include other pressures scenarios to see whether the turbine designers could trade off pressure increases if necessary to reduce other stresses (test conditions 1, 5, and 9 in Table 3.1).

Table 3.1. Twelve Possible Test Conditions for PNNL's Turbine Passage Pressure Studies Based on Different Combinations of Pre-Turbine Acclimation Pressures (101 or 191 kPa), Total Dissolved Gas Concentrations (100, 120, and 135% saturation), and Pressure Changes to Which Fish Would be Exposed in a Turbine. Pressures as high as 460 kPa and as low as 2 kPa can be expected in Kaplan turbines on the Columbia River.

Nadir of Turbine Pressure Spike	Acclimation Pressure 101 kPa			Acclimation Pressure 191 kPa		
	% Saturation			% Saturation		
	100	120	135	100	120	135
Near 0 kPa (Maximum Turbine Efficiency) Expected for Lower Columbia River Dam	Test Priorities					
	2	6	10	4	8	12
~50 kPa (Reduced Turbine Efficiency) Outside the Expected Operations of a Lower Columbia River Dam	1	5	9	3	7	11

- **Test depth-compensated, gas-loaded fish**. Assuming that most downstream migrants travel in the upper 3 m, Montgomery Watson (1995) tested fish that were acclimated to surface water pressures and varying degrees of gas supersaturation (test conditions 2, 6, and 10 in Table 3.1). NMFS (2000; Section E.6.3) pointed out that gas-loaded fish may compensate for the high dissolved gas levels by swimming at greater depths (10 m test conditions 4, 8, and 12 in Table 3.1). The swim bladders of these fish are acclimated to greater pressures, and the fish may suffer greater injury when suddenly exposed to low pressures in a turbine.

Tests were conducted at ambient well water temperature (~17°C). Forty fish (20 in each of two chambers) were acclimated to one of three gas saturation levels (100, 120, and 135% saturation) at either near surface pressure (101 kPa, 14.7 psi) or the equivalent of 30 ft of pressure (191 kPa, 27.7 psi) for a period of 16 to 22 hr. One group was then subjected to a pressure spike simulating passage through a hydroelectric turbine, while the second group acted as a paired control (same gas exposure history, but no pressure spike). During the computer-controlled program sequence, pressure increased to ~58 psi (~400 kPa) over 30 to 60 sec to simulate fish entering the turbine intake and approaching the runner. Fish then were subjected to a sudden decrease in pressure to ~1 psi (2-10 kPa), the pressure scenario expected at lower Columbia River dams as described in scenarios 2, 4, 6, 8, 10, and 12 in Table 1. After the spike, the pressure transition simulated a fish passing out the draft tube and into the tailrace. After completion of the simulated turbine passage, fish were removed from the chambers and placed in a holding trough for a 48-hr post-exposure observation. The second group of 20 fish (the handling control, undergoing identical treatment, except they were not subjected to the turbine spike) was also held for a 48-hr post-exposure observation.

3.3 Gas Supersaturation System

A pressurized packed cell column was used to generate gas-supersaturated water. The system (Figure 3.1) was designed by Point Four Systems of Port Moody, British Columbia, for use in previous studies at PNNL and is described in AASI (1998). Pressurized water and air were added to the column. Total dissolved gas levels were achieved by controlling pressure within the column and by adjusting the position of a proximity switch on a sight glass.

For our tests, 10 gpm of well water was pumped into the top of the column. Backpressure was created inside the column by restricting the discharge line at the bottom of the column. As the water level increased within the column, a proximity switch sensor mounted on a sight glass controlled a valve allowing pressurized air to enter the column to maintain the desired level within the column. The combination

Figure 3.1. Gas Supersaturation System Showing Controls (lower right) and Saturometer (upper right)

of compressed air acting against the pressurized water source and the restricted outlet created gas-supersaturated water within the column. The water level inside the column affected the water/air interface. Therefore, adjusting water level and/or internal column pressure were used to change and control gas supersaturation levels.

As the system was set up for our tests, an internal pressure of ~11 psi at a flow of 10 gpm was needed to produce total dissolved gas (TDG) levels of ~135%. A pressure of ~6 psi resulted in TDG level of ~120%. For "normal" saturated water, the outlet was opened wide so that no backpressure was created. In this condition, the TDG level was <105%. Gas levels were monitored with a Sweeney Saturometer, Model DS-1B (Sweeney Aquamatic, Stony Creek, Connecticut). As described in Aspen Applied Sciences (1998), the manufacturer calibrated our instrument before delivery to PNNL. In the laboratory, the instrument readings were compared to other instruments by the same manufacturer and found to be within tolerances required for the study (±1%).

3.4 Turbine Passage System

The Turbine Passage System (Figure 3.2) was designed and built by Reimers Engineering in 1994 and is described in Montgomery Watson (1995). The system can create a variety of pressure regimes, and for this study, was used to simulate the pressure history that fish would experience in passing through a typical Kaplan turbine on the mid-Columbia River. The exposure chambers for the turbine passage system consisted of two 27.5-cm-diameter acrylic tubes, 55 cm long. The volume of each cylinder was about 34 L.

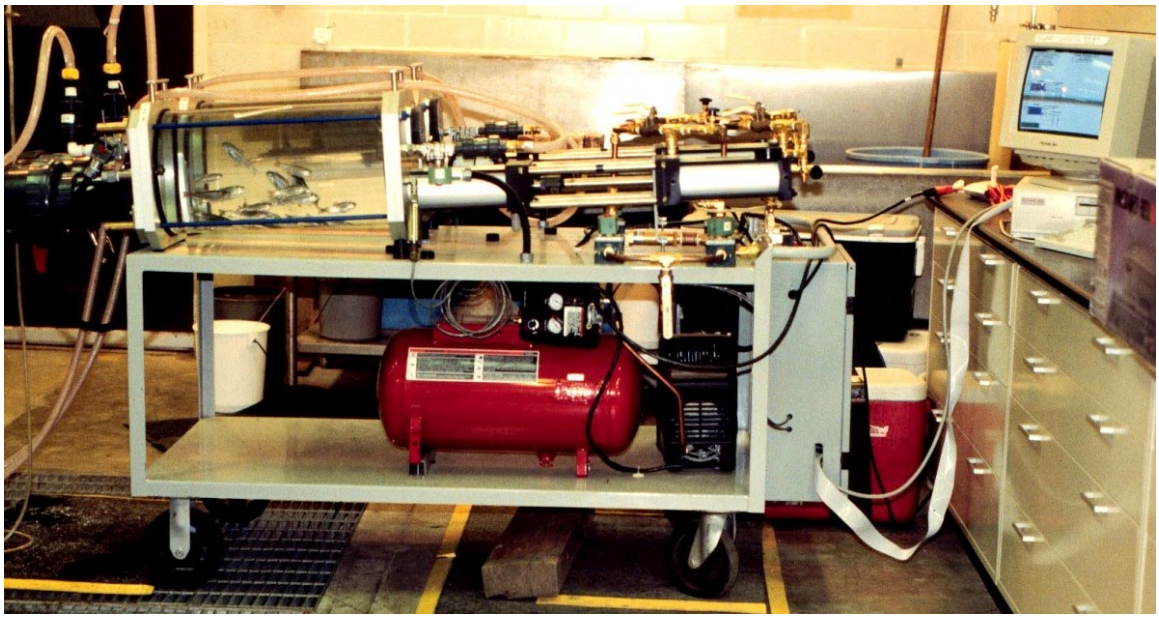


Figure 3.2. Turbine Passage System with Rainbow Trout in Hyperbaric Chambers

The chambers were connected to hydraulic cylinders, which in turn were connected to pneumatic cylinders. Through a computer-controlled gas pressurization system attached to the pneumatic cylinders, the positions of the hydraulic cylinders were moved to either pressurize or depressurize the chambers. The maximum pressure of the chamber was 100 ft of head (3 atm, or ~400 kPa). The system can drop the pressure from 100 ft (~400 kPa) of head to close to the vapor pressure of water (~1 psi or 2-10 kPa) in 0.1 sec.

A computer program, the Labtech Control Program (Labtech Control Version 4.2.0 for Microsoft® Windows™, Laboratory Technologies Corporation), controlled the gas cylinders used in the pressurization/depressurization sequence. Sub-programs within the Labtech Control program were used for various chamber operations. Water was pumped from the gas supersaturation system to the chambers at the desired TDG level (100, 120, or 135%), depending on the test scenario.

3.5 Water Delivery System

Water from the gas supersaturation system flowed into a trough. A second centrifugal pump withdrew water from the trough and pumped it to the hyperbaric chambers. A valve on the outlet side of the pump was used to precisely control the quantity of water delivered. Water from the supply line was split and entered both hyperbaric chambers. Flow was equalized through the two chambers by using a restriction (round orifice) in the end of drain tubes leading from each chamber. During the acclimation period for fish, pressure within the hyperbaric chambers was set by adjusting the quantity of pressurized water delivered to the hyperbaric chambers, with back pressure determined by the size of the outlet orifice. For holding fish at 30 ft of depth (191 kPa), a 3/32-in. orifice was used. For holding fish at the surface (101 kPa) a 3/8-in. orifice was used. Flow through each hyperbaric chamber was ~10 and ~14 L/min, respectively, and turnover rate for each chamber was 3.4 and 2.5 min, respectively.

3.6 Fish Introduction

Once the TDG levels had stabilized for the ensuing test, the hyperbaric chambers were partially filled with water, and the delivery pump was turned off. Twenty fish were netted from the acclimation tank and placed in ~10 L of water in a bucket. The fish were then poured into a chamber through the 4-in. PVC pipe and valve on the end of the hyperbaric chamber. After fish were added to each chamber, the valves were closed, the pump was turned on, and all the air was evacuated from each chamber through a vent tube. During acclimation, both the vent tubes and the outlets remained wide open to prevent buildup of gas bubbles in the hyperbaric chambers. Pressure was adjusted within the hyperbaric chambers by making slight adjustments to the flow control valve at the outlet of the supply pump. When the desired pressure was achieved, a dark cover was placed over the chambers to calm the fish. An “acclimation” file was initiated to record pressure at ~90-sec intervals within a chamber during the acclimation period (16-22 hr).

3.7 Pressure Spike

After the acclimation period, one of the two fish groups was subjected to the simulated turbine passage pressure spike shown in Figure 3.3. To initiate the sequence, the piston was first moved all the way in to purge any air that may have accumulated in the cylinder. Then, the piston was positioned in the middle of the stroke. When the sequence was started, the inlet, outlet, and vent tube valves were quickly

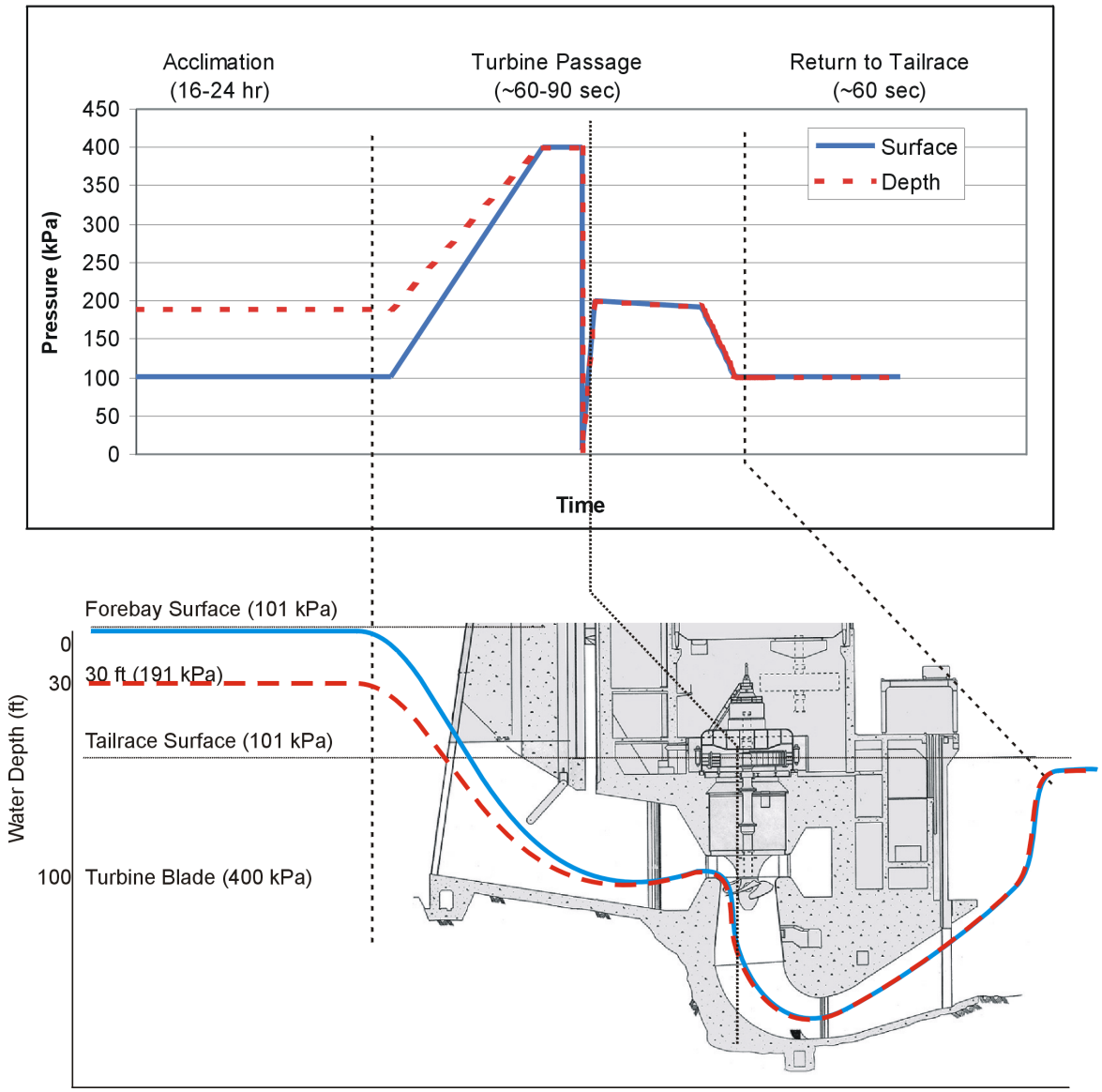


Figure 3.3. Surface (101 kPa) and 30 ft Depth (191 kPa) Acclimation and Hyperbaric Chamber Pressure Exposure Simulation of Turbine Passage. Pressure increases as the fish's depth increases. Pressure spike occurs as fish pass the turbine blades. Pressures then return to surface pressure as fish pass through the draft tube and enter the tailrace.

closed, and the computer controlled the piston to maintain the appropriate pressure (101 or 191 kPa) within the hyperbaric chamber. After a brief period (~15 sec), the "dive" was initiated. The entire pressure sequence lasted about 90 sec. At the end of the sequence, the chambers were taken to "surface" pressure (101 kPa), and water flow was restored to the chambers. Two minutes after completion of the pressure spike, the fish were removed from the chambers and placed in holding troughs.

3.8 Fish Removal

To remove fish from the hyperbaric chambers, a large plastic garbage can was placed under the 4-in. PVC valve outlet. The valve was opened, and the water and fish in the chamber were flushed out. Incoming water helped in flushing the fish. Fish remaining in the chamber after 1 to 2 min were retrieved by removing the PVC valve and "sweeping" the fish from the chamber with a plastic mesh crowding device. Fish were then netted from the garbage can and placed in a partitioned trough for a 48-hr observation. Fish were checked (alive [OK], loss of equilibrium [LE], or dead) at 1, 24, and 48 hrs after the pressure spike.

3.9 Fish Necropsy

Fish that died during acclimation or during/immediately after the pressure spike were examined immediately to determine the cause of death. Fish that died during the 48-hr post-exposure holding period were examined for signs of injuries related to gas bubble trauma or the pressure spike, and survivors were examined at the end of each test. Examinations were performed using a dissecting microscope (up to 40X magnification) with the aid of optical fiber lights. Injuries were documented with digital imagery. Necropsy included examination for bubbles in fins, bubbles or hemorrhaging in the eyes or gill filaments, and internal examination for bubbles and/or hemorrhaging in the heart and major arteries, swim bladder rupture, and hemorrhaging or rupturing of other internal organs.

3.10 Monitoring

The stability of the gas supersaturation and delivery systems was monitored before and during each test. Tests were not initiated until parameters were stable and within ≤5% of desired test conditions. Conditions monitored included those described below.

- Flow through and pressure within the gas supersaturation system was checked at the beginning, 2 to 6 hr later, and at the end of the acclimation period. Flow and pressure readings were for reference only to set the system and maintain proper TDG levels.

- TDG level of supply water (from gas supersaturation system to the hyperbaric chambers) was checked before, during, and at the end of the acclimation period.

- Acclimation pressure was monitored continuously at 90-sec intervals and stored as an "acclimation file" on the computer. Pressure "drift" of <2 psi was accepted, but larger pressure changes voided the test. Pressure gauges built into the hyperbaric chambers were calibrated by the manufacturer prior to the initiation of our tests.

- Flow through each hyperbaric chamber was measured at the beginning, once during, and at the end of the acclimation period by timing the discharge of a known volume from each chamber with a stopwatch.

- Temperature of the gas supersaturation supply water was checked before, during, and at the end of the acclimation period. Temperatures in the holding troughs were monitored daily. Since all systems were using ambient well water at 17°, there was little temperature variation throughout the tests.

- Simulated turbine passage pressure sequence was recorded for each test. A "slow" file measured pressure at 1-sec intervals. A "fast" file measured pressure at 0.1-sec intervals for a 2-sec period that documented the rapid change in pressure from ~400 kPa to <7 kPa. Instrumentation was calibrated by the hyperbaric chamber manufacturer prior to the initiation of our test series.

- Mortalities, injuries, and abnormal swimming behavior were monitored at 1, 24, and 48 hr post-exposure.

- Fish were examined for gas bubble trauma (GBT) under a dissecting microscope, and digital images of injuries were captured to document injuries. Digital "movies" were also recorded with a high-speed camera (250 frames/sec) during the pressure spike to observe fish reaction during rapid decompression.

3.11 Statistical Methods

The purpose of statistical analyses is to determine factors that cause differences in injuries or mortalities between pressure-exposed fish (referred to as "Spike") and control fish ("Non-spike"). In each analysis, four factors were studied. These factors and the levels of each factor are: 1) ***test*** - the test (Spike) and control (Non-spike); 2) ***species*** - bluegill, rainbow, and fall chinook; 3) ***gas*** - 100, 120, and 135% TDG; and 4) ***depth*** - the surface-acclimated (0 ft) and depth-acclimated (30 ft).

Four different response variables were investigated, each individually. These responses included: 1) **GBT** - proportion of fatalities due to GBT; 2) **Spike** - proportion of fatalities due to a turbine-passage pressure regime; 3) **Injured** - proportion of fish that were injured; and 4) **Uninjured** - proportion of fish that were uninjured.

In each analysis, the response variable is a proportion. In many cases, that proportion is close to 0 or 1, because it either happened to a large majority of fish in that test group, or it rarely occurred. When this is the case, the normality assumption when using Analysis of Variance is suspect. For this purpose, the non-parametric Kruskall-Wallis test was used for each analysis. The significance level of $\alpha = 0.05$

was used to determine significance (any p-value below 0.05 is considered significant). Unfortunately, the Kruskall-Wallis test only tests factors individually and does not allow for testing interactions. Therefore, interactions were tested using Analysis of Variance. To attempt to compensate for the lack of normality, only highly significant interactions were reported (p-value <0.01). Only two-way interactions were examined.

When testing each factor individually the null hypothesis (H_o) is: there is no difference in the response variable between the levels of the given factor. The alternative hypothesis (H_a) is: at least one of the levels of the factor is significantly different than the other levels for that specific response variable. The hypotheses for the interactions are: H_o – no significant interaction between the two factors, and H_a – a significant interaction between the two factors.

4.0 Results

4.1 Biological Effects

4.1.1 Rainbow Trout

A series of 17 tests representing test conditions 2, 4, 6, 8, 10, and 12 of Table 3.1 was completed during November and December 1999. The results of these tests are summarized based on injuries/deaths associated with acute and/or chronic GBT, and pressure-spiked versus non-spiked (control) fish.

4.1.1.1 Gas Bubble Trauma

Since all fish were subjected to identical TDG levels, both the test and control groups were used to evaluate GBT. Rainbow trout acclimated at surface pressures (101 kPa) showed signs of acute and chronic GBT at both 120 and 135% TDG levels (Table 4.1, Figure 4.1). At 120% TDG, 17 of 120 fish died from acute GBT. Mortality ranged from 1 to 6 fish per 20-fish group during the acclimation period preceding turbine passage simulation. Other fish had visible gas bubbles in fins (Figure 4.2) and eyes, but after the 48-hr observation period, there were no visible external signs of GBT except that bubbles persisted in the eyes of three fish (Figure 4.3). At 135% TDG, fish went into convulsions and started dying about 2 hr into the acclimation period. Most fish were dead after 5 hr, and all (80 fish total) were dead by the end of the acclimation period. Because of the 100% mortality rate during acclimation, only two test replicates were conducted under 135% TDG surface acclimation conditions.

Table 4.1. Mortality and Injury Rates for Rainbow Trout Based on TDG Level, Acclimation Depth, and Pressure Spike from Turbine Passage

Test Group	Surface Acclimation (101 kPa)								30 ft Acclimation (191 kPa)								
	100% TDG			120% TDG			135% TDG		100% TDG			120% TDG			135% TDG		
Replicate	1	2	3	1	2	3	1	2	1	2	3	1	2	3	1	2	3
Dead From GBT	0	0	0	1	4	6	20	20	0	0	0	0	0	0	0	0	0
Dead from Spike	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Injured	0	1	3	2	0	1	0	0	0	4	5	5	2	3	6	4	5
OK - no injuries	20	19	17	17	16	13	0	0	20	16	15	15	18	17	14	16	15
Control Group	Surface Acclimation (101 kPa)								30 ft Acclimation (191 kPa)								
	100% TDG			120% TDG			135% TDG		100% TDG			120% TDG			135% TDG		
Replicate	1	2	3	1	2	3	1	2	1	2	3	1	2	3	1	2	3
Dead From GBT	0	0	0	1	4	1	20	20	0	0	0	0	0	0	0	0	0
Injured	0	0	0	1	1	0	0	0	0	1	0	0	4	0	0	0	0
OK - no injuries	20	20	20	18	15	19	0	0	20	19	20	20	16	20	20	20	20

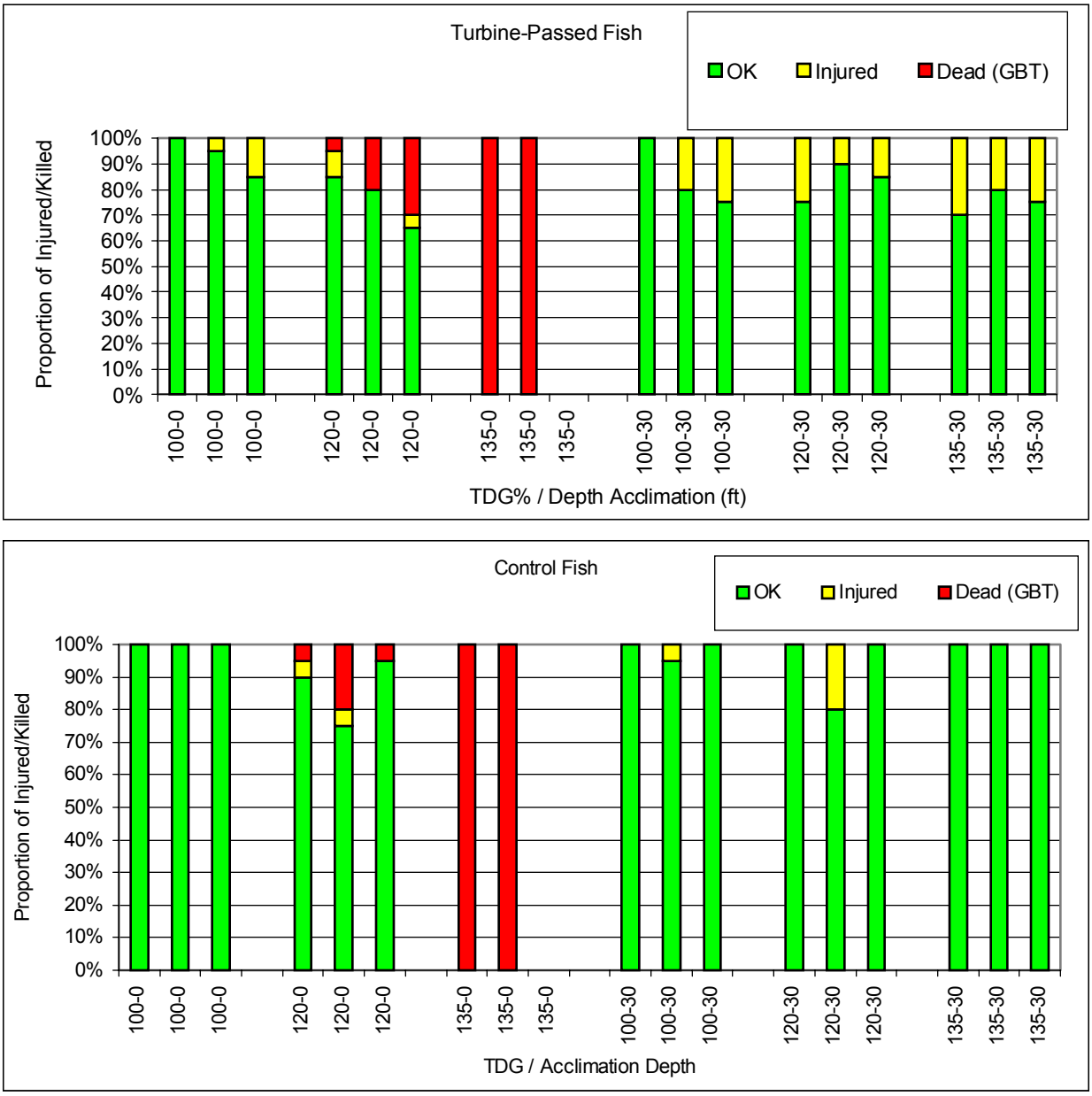


Figure 4.1. A Comparison of the Injury and Mortality Rates for Rainbow Trout Based on TDG Level, Acclimation Depth, and Pressure Spike from Turbine Passage

Rainbow trout exposed to 100, 120, and 135% TDG at acclimation pressures simulating 30 ft of depth (191 kPa) showed no signs of GBT during or after the acclimation period (Table 4.2, Figure 4.1). When the chambers were pressurized at the beginning of the acclimation period, all fish had negative buoyancy (i.e., sank to the bottom of the chamber). All fish were still "negative" at the end of the acclimation period. Swimming behavior (head higher than the tail and constantly "swimming" up) indicated that rainbow trout were unable to achieve neutral buoyancy during the 16- to 22-hr acclimation

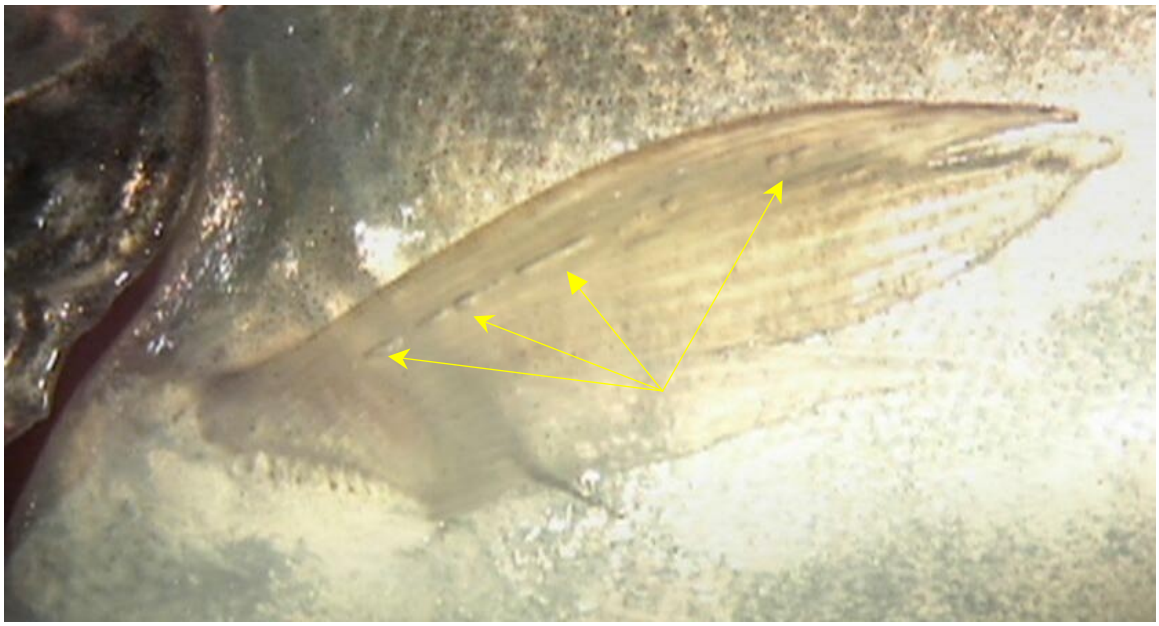


Figure 4.2. Bubbles in the Pectoral Fin of a Rainbow Trout Acclimated to 120% TDG, 101 kPa Pressure

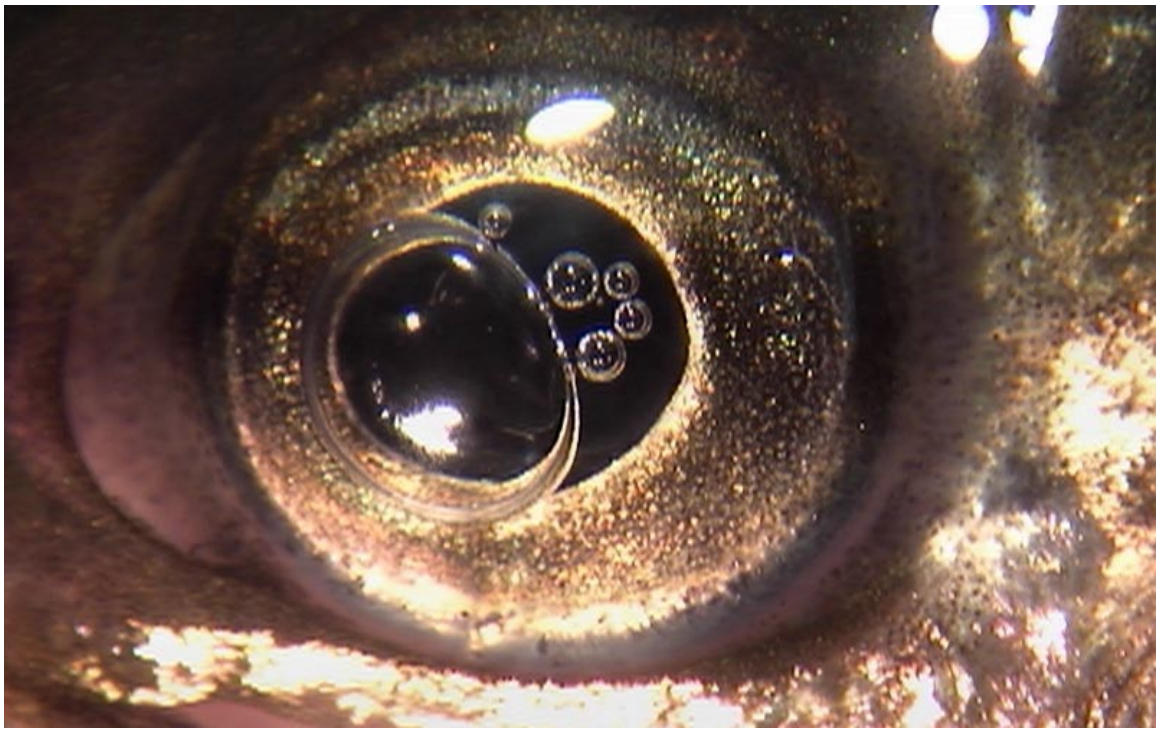


Figure 4.3. Bubbles in the Eye of a Rainbow Trout Acclimated to 120% TDG, 101 kPa Pressure

period by filling the swim bladder with gases dissolved in the blood. This suggests that fish surfacing to feed or as part of normal diurnal behavior will never achieve neutral buoyancy at 30 ft of depth. However, fish held at 191 kPa appeared to be totally protected from the effects of acute and chronic GBT prior to the turbine passage sequence.

4.1.1.2 Effects of Turbine Passage Simulation

Rainbow trout showed little reaction to the pressure spike during the turbine passage simulation. Fish were startled by the sudden decompression but were otherwise unaffected (Table 4.1). No fish lost equilibrium or went into convulsions prior to removal from the chambers, and no fish died (0 of 290 fish that were alive when the pressure spike occurred) in the 48-hr post-exposure holding period. Fish that died from GBT during the acclimation phase (black bars in Figure 4.1) were not used to evaluate effects of the pressure spike.

Fish were startled and darted around when first placed in the holding troughs for 48-hr observation. Initially, most fish, especially turbine-spiked fish, were negatively buoyant. However, within the 1 to 2 hr, most fish were neutrally buoyant after gulping air to inflate their swim bladders, and maintained a normal swimming position (facing upstream) in the trough. A few fish remained negatively buoyant for up to 24 hr.

No external signs of injury or trauma were evident 1 hr after the turbine passage simulation. However, during the first 24 hr in holding troughs, 38 of 543 of necropsied fish (~7%) had developed a black spot on the top of their heads (Figure 4.4). The discoloration appeared to be immediately above the cranial cavity. In some fish, the spot was in the center of the skull between the eye orbits, while in other fish, the spot was off to the side and appeared to reach toward an eye orbit (Figure 4.5). The spot usually persisted throughout the 48-hr holding period.

The black spot was more frequent in depth-acclimated fish than in surface-acclimated fish (33 of 360 fish and 5 of 183 fish, respectively), and more frequent in spiked fish than controls (31 of 269 fish and 7 of 274 fish, respectively) (Table 4.2). Therefore, the highest incidence of the trauma occurred in depth-acclimated, spiked fish. Among the depth-acclimated, spiked fish, higher TDG levels produced a progressively higher occurrence of the injury (5 of 60 fish at 100%, 9 of 60 fish at 120%, and 14 of 60 at 135% TDG).

No ruptured swim bladders were observed in internal necropsies of 520 fish (internal examinations were performed after 13 of 15 tests with surviving fish). Of the 680 fish used in the test series, 9 of 623 fish surviving the acclimation period developed over-inflation of the swim bladder (Figure 9), resulting in fish floating excessively high in the holding trough. All 9 fish were from groups that had undergone the turbine passage simulation. However, 8 of the 9 fish were from 100% TDG acclimated groups (4 from one surface-acclimated group and 4 from one depth-acclimated group). The other fish was from a depth-acclimated 120% TDG group. The over-inflation developed gradually during the 48-hr holding period in the troughs.

Figure 4.4. Discoloration of the Head on Rainbow Trout Observed During Post-Exposure Holding Period

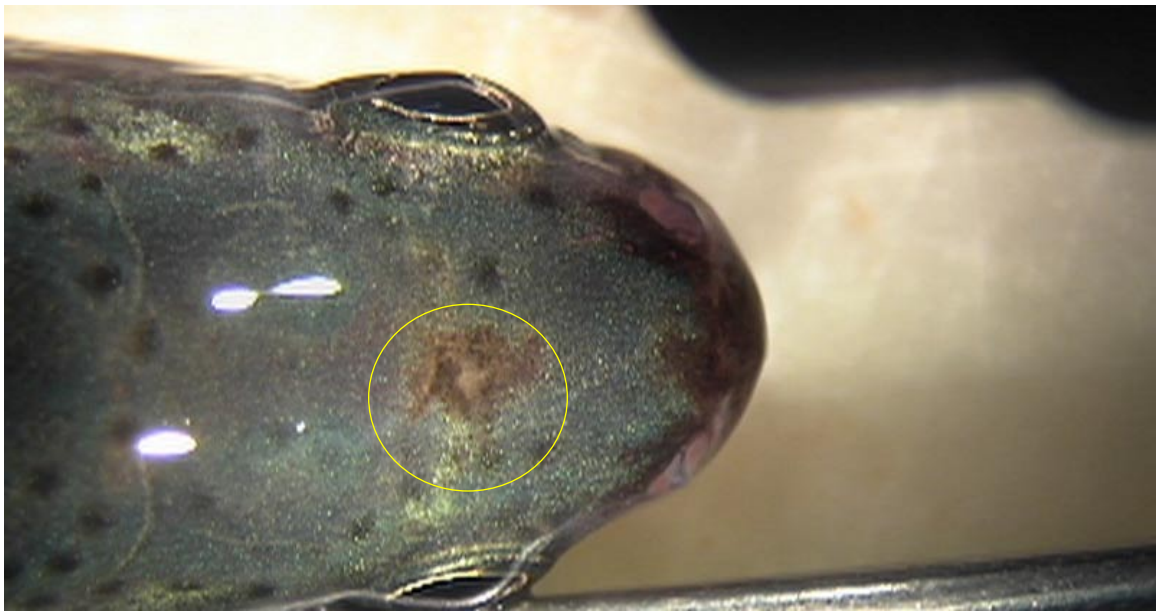


Figure 4.5. Close-Up of Black Spot on Head of Rainbow Trout at Necropsy 48 Hr After Exposure to Turbine Passage Sequence

Table 4.2. Frequency of Black Cranial Spot in Rainbow Trout as Related to TDG Level, Acclimation Depth, and Pressure History

	Pressure-Spiked				Controls			
	# Tested	# Dead (GBT)	# Examined	# with Cranial Spot	# Tested	# Dead (GBT)	# Examined	# with Cranial Spot
100%-0 ft	60	0	40[(a)]	2	60	0	40[(a)]	1
120%-0 ft	60	11	49	1	60	6	54	1
135%-0 ft	40	40	0	-	40	40	0	-
Total	160	51	89	**3**	160	46	94	**2**
100%-30 ft	60	0	60	5	60	0	60	1
120%-30 ft	60	0	60	9	60	0	60	4
135%-30 ft	60	0	60	14	60	0	60	0
Total	180	0	180	**28**	180	0	180	**5**

(a) In one test, a trough divider was dislodged at the 24-hr check, mixing the spiked and control groups. No necropsies were performed on these 40 fish, 20 from each group.

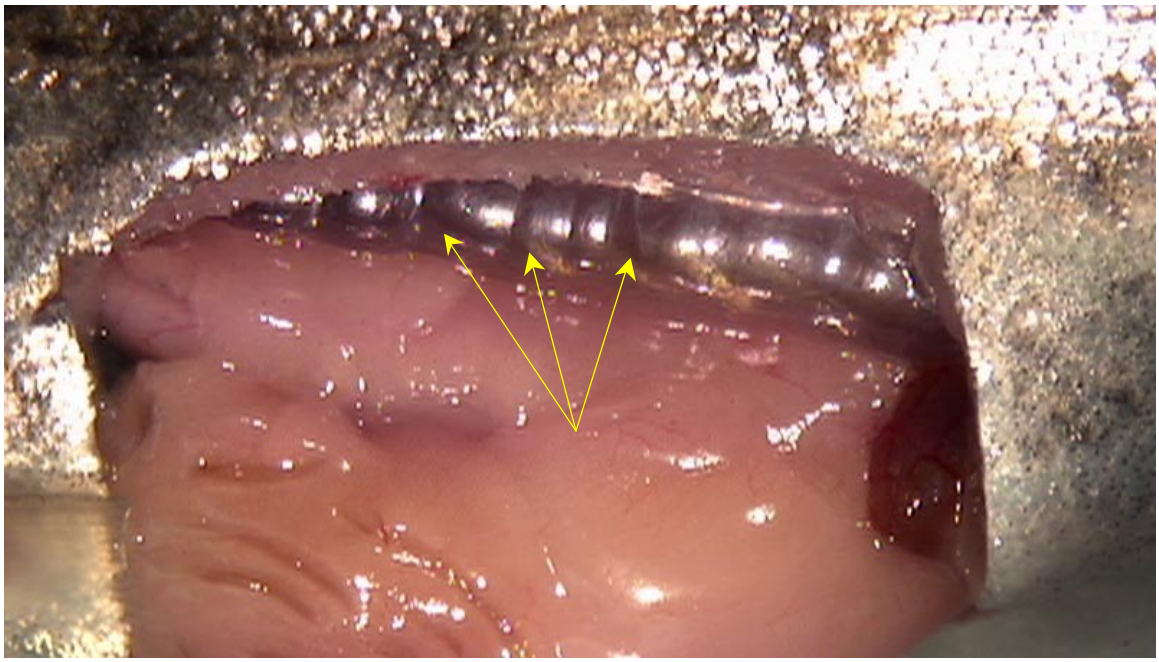


Figure 4.6. Over Inflated Swim Bladder ("ropey" appearance) in Rainbow Trout 48 Hr After Turbine Passage Sequence

4.1.2 Fall Chinook Salmon

A series of 17 tests was completed during June and July, 2000. The results of these tests are summarized based on injuries/deaths associated with acute and/or chronic GBT, and pressure-spiked versus non-spiked (control) fish.

4.1.2.1 Gas Bubble Trauma

Fall chinook salmon acclimated at surface pressures (101 kPa) showed signs of acute GBT at 135% TDG levels (Table 4.3, Figure 4.7). At 120% TDG, no GBT mortalities occurred; however, some fish developed gas bubbles in their fins during the acclimation period. No fish acclimated to 100% TDG showed signs of GBT.

Table 4.3. Mortality and Injury Rates for Fall Chinook Salmon Based on TDG Level, Acclimation Depth, and Pressure Spike from Turbine Passage

Test Group	Surface Acclimation (101 kPa)								30 ft Acclimation (191 kPa)								
	100% TDG			120% TDG			135% TDG		100% TDG			120% TDG			135% TDG		
Replicate	1	2	3	1	2	3	1	2	1	2	3	1	2	3	1	2	3
Dead From GBT	0	0	0	0	0	0	20	20	0	0	0	0	0	0	0	0	0
Dead from Spike	0	0	0	0	0	0	0	0	0	0	0	2	0	1	0	0	3
Injured	0	0	0	1	0	0	0	0	0	0	1	0	0	1	1	4	0
OK - no injuries	20	20	20	17	19	20	0	0	20	20	19	18	20	18	19	16	17
Control Group	Surface Acclimation (101 kPa)								30 ft Acclimation (191 kPa)								
	100% TDG			120% TDG			135% TDG		100% TDG			120% TDG			135% TDG		
Replicate	1	2	3	1	2	3	1	2	1	2	3	1	2	3	1	2	3
Dead From GBT	0	0	0	0	0	0	20	20	0	0	0	0	0	0	0	0	0
Injured	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
OK - no injuries	20	20	20	20	20	20	0	0	20	20	20	20	20	20	20	20	20

At 135% TDG, fish went into convulsions and started dying about 5-6 hr into the acclimation period. All fish were dead by the end of the 16 to 22 hr acclimation period. Because of the 100% mortality rate during acclimation, only two test replicates were completed under 135% surface acclimation conditions.

Fall chinook salmon exposed to 100, 120, and 135% TDG at pressures simulating 30 ft of depth (191 kPa) showed no external signs of GBT during or after the 16- to 22-hr acclimation period (Table 4.3, Figure 4.7). When the chambers were pressurized, all fish had negative buoyancy (sank to the bottom of the chamber). All fish were still "negative" at the end of the acclimation period. Swimming behavior (head higher than the tail and constantly "swimming" up) indicated that fall chinook salmon were unable to achieve neutral buoyancy during the acclimation period by filling the swim bladder with gases

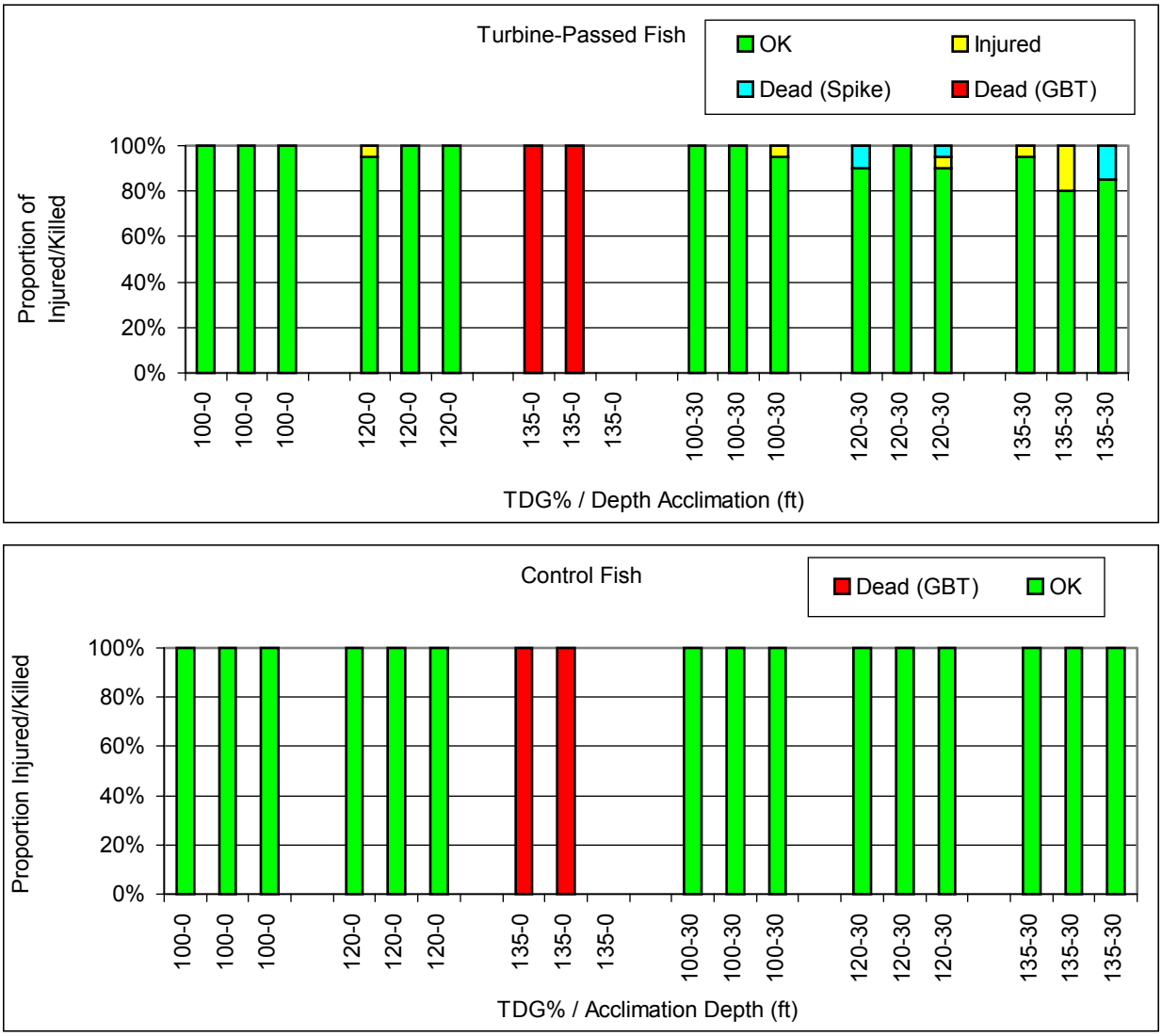


Figure 4.7. A Comparison of the Injury and Mortality Rates for Fall Chinook Salmon Based on TDG Level, Acclimation Depth, and Pressure Spike from Turbine Passage

dissolved in the blood. This suggests that fish surfacing to feed or as part of normal diurnal behavior will never achieve neutral buoyancy at 30 ft of depth. However, depth-acclimated fish appeared to be totally protected from the effects of acute and chronic GBT prior to the turbine passage sequence.

4.1.2.2 Effects of Turbine Passage Simulation

Fall chinook salmon were startled by the sudden decompression. In addition, some fish lost equilibrium and/or went into convulsions immediately following the spike in the turbine passage simulation.

At 135% TDG, 5 of 60 depth-acclimated fish subjected to the turbine passage simulation lost equilibrium, with 2 of the 5 fish dying within 1 hr. Another fish that did not lose equilibrium immediately also died within the first hour. Necropsies of the 3 mortalities revealed massive gas bubbles in the heart (atrium) blocking blood flow to the gills (Figure 4.8). The 3 other fish that lost equilibrium had ruptured swim bladders.

At 120% TDG, 3 of 60 depth-acclimated fish lost equilibrium and all died within 1 hr (Table 4.3). Necropsies again revealed massive gas bubble blockage in afferent lamellar arteries of the gills, blocking blood flow to the gills (Figure 4.9). Another fish had a ruptured swim bladder.

No depth-acclimated fish at 100% TDG died from the turbine passage simulation; however, one fish had a ruptured swim bladder. Necropsies of the non-spiked, depth-acclimated controls showed that none had ruptured swim bladders or any signs of trauma.

Fish were startled and darted around when first placed in the holding troughs for 48-hr observation. Initially, most fish were negatively buoyant, and some fish had problems maintaining an upright position. However, within 1 to 2 hr, most fish were neutrally buoyant after gulping air to inflate their swim bladders, and maintained a normal swimming position (facing upstream) in the trough. A few fish remained negatively buoyant for the entire 48-hr observation period.

Necropsies were performed on a total of 600 fish. The only external sign of injury or trauma during the 48-hr holding period (with the exception of the 6 mortalities) was partial loss of equilibrium in some fish. Necropsies revealed that most of these fish had ruptured swim bladders. Only 1 of 120 of surface-acclimated fish subjected to the turbine passage simulation suffered a ruptured swim bladder, whereas

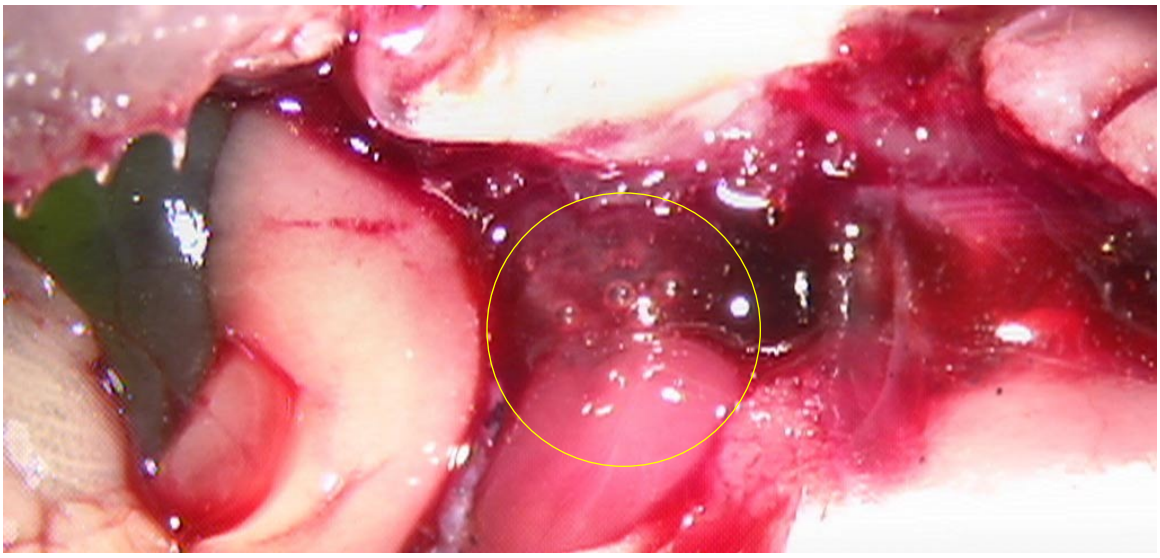


Figure 4.8. Gas Bubble Accumulation in the Atrium of Fall Chinook Salmon Acclimated to 135% TDG at 191 kPa Pressure and Subsequently Subjected to Turbine Passage Pressure Spike

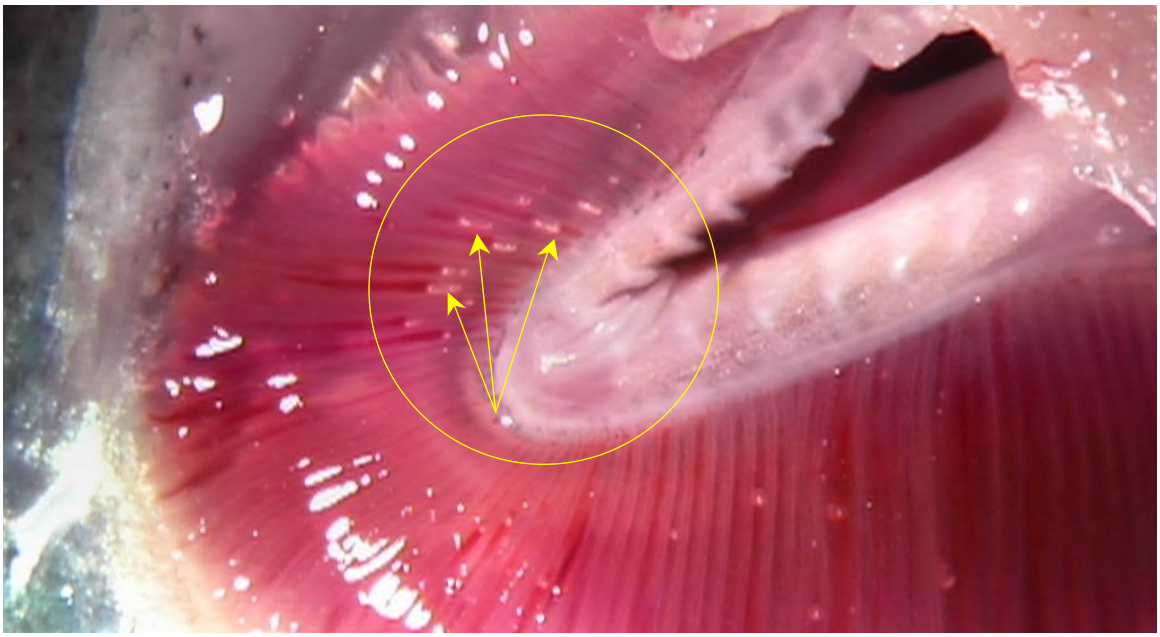


Figure 4.9. Bubble Formation in the Afferent Arteries of the Gills in Fall Chinook Salmon Acclimated at 120% TDG at 191 kPa and Subsequently Subjected to Turbine Passage Sequence

7 of 174 (and all 6 mortalities) of the depth-acclimated fish subjected to the turbine spike had swim bladder ruptures. None of the 120 surface-acclimated, non-spiked fish or 180 depth-acclimated, non-spiked fish showed any signs of GBT or distress after 48 hr.

4.1.3 Bluegill

A series of 18 tests was completed during January and February, 2000. The results of these tests are summarized based on injuries/deaths associated with acute and/or chronic GBT, and pressure-spiked versus non-spiked (control) fish.

4.1.3.1 Gas Bubble Trauma

Bluegills acclimated at surface pressures (101 kPa) showed signs of acute GBT at 135% TDG and chronic GBT at 120% TDG. At 135%, 25 of 120 fish died from acute GBT (Table 4.4, Figure 4.10). Mortality ranged from 1 to 6 fish per 20-fish group during the 16- to 22-hr acclimation period preceding the turbine passage simulation. Other fish had visible gas bubbles in fins and eyes, but after the 48-hr post-exposure observation period, there were no visible external signs of GBT except that some fish had minor hemorrhages in the eyes that may have been the result of GBT (Figure 4.11). At 135% TDG, fish began to die about 10-12 hr into the acclimation period. At the end of the acclimation period, many fish had external GBT signs (bubbles in fins and eyes).

Table 4.4. Mortality and Injury Rates for Bluegills Based on TDG Level, Acclimation Depth, and Pressure Spike from Turbine Passage

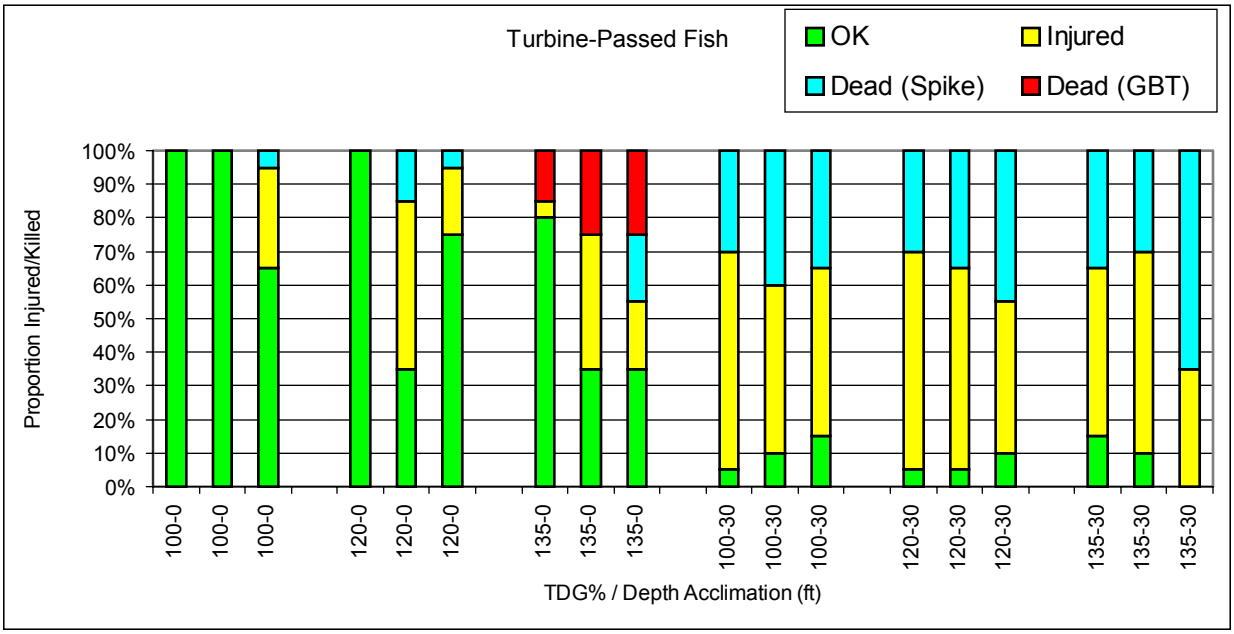
Test Group	Surface Acclimation (101 kPa)									30 ft Acclimation (191 kPa)								
	100% TDG			120% TDG			135% TDG			100% TDG			120% TDG			135% TDG		
Replicate	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3
Dead From GBT	0	0	0	0	0	0	3	5	5	0	0	0	0	0	0	0	0	0
Dead from Spike	0	0	1	0	3	1	0	0	4	6	8	7	6	7	9	7	6	13
Injured	0	0	6	0	10	4	1	8	4	13	10	10	13	12	9	10	12	7
OK – no injuries	20	20	13	20	7	15	16	7	7	1	2	3	1	1	2	3	2	0
	Surface Acclimation (101 kPa)									30 ft Acclimation (191 kPa)								
Test Group	100% TDG			120% TDG			135% TDG			100% TDG			120% TDG			135% TDG		
Replicate	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3
Dead From GBT	0	0	0	0	0	0	6	1	5	1	0	0	0	0	0	1	1	0
Injured	0	0	0	0	0	0	1	0	1	0	0	0	0	0	0	0	0	0
OK – no injuries	20	20	20	20	20	20	13	19	14	19	19	19	20	20	20	19	19	20

Bluegill exposed to 100, 120, and 135% TDG at pressures simulating 30-ft of depth (191 kPa) showed no external signs of GBT during or after the 16- to 22-hr acclimation period. When the chambers were pressurized all fish had negative buoyancy (sank to the bottom of the chamber). However, after about 2 hr, fish were again neutrally buoyant, able to float motionless inside the acclimation chambers. This swimming behavior suggests that bluegills can rapidly inflate their swim bladders from gases dissolved in the blood. Fish appeared to be totally protected from the effects of acute and chronic GBT prior to the turbine passage sequence.

4.1.3.2 Effects of Turbine Passage Simulation

As the turbine passage scenario was initiated, bluegills sank to the bottom of the chamber as pressure within the chamber built and compressed the gas in their swim bladders (Figure 4.12). Bluegills reacted violently to the pressure spike during the turbine passage simulation (Figure 4.13). At the instant of the turbine spike, a large bubble was observed (with the aid of a high-speed video camera) near the heads of most fish (Figure 4.14). Many fish convulsed and settled to the bottom of the chamber. When the fish were removed from the chambers and placed in the holding troughs, most of the spiked fish sank to the bottom of the trough (Figure 4.15). Almost all spiked fish lost equilibrium. Non-spiked, depth-acclimated fish (controls) floated at the surface.

The mortality caused by the pressure spike ranged from 0 to 4 fish for surface-acclimated groups and 6 to 13 for depth-acclimated groups (Table 4.4). Most fish that died as a result of the pressure spike were dead at the 1-hr post-exposure observation check. Most spiked fish (whether dead or alive and experiencing loss of equilibrium) were negatively buoyant, lying on their sides on the bottom of the trough. Depth-acclimated controls (non-spiked fish) were buoyant, often floating on their sides on the surface, struggling to swim upright (Figure 4.16). Surface-acclimated controls exhibited normal buoyancy and behavior when placed in troughs. Over a period of several hours, the depth-acclimated control bluegills were able to maintain a normal, neutral position in the troughs.



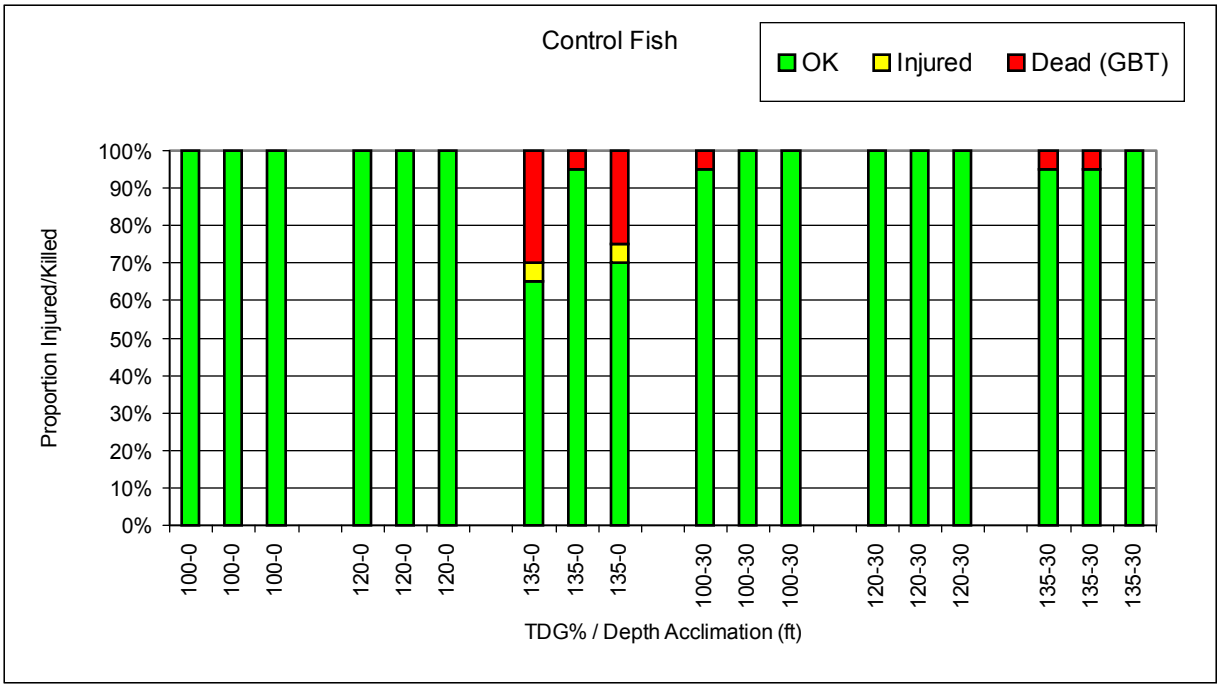


Figure 4.10. Comparison of the Injury and Mortality Rates for Bluegills Based on TDG Level, Acclimation Depth, and Pressure Spike from Turbine Passage

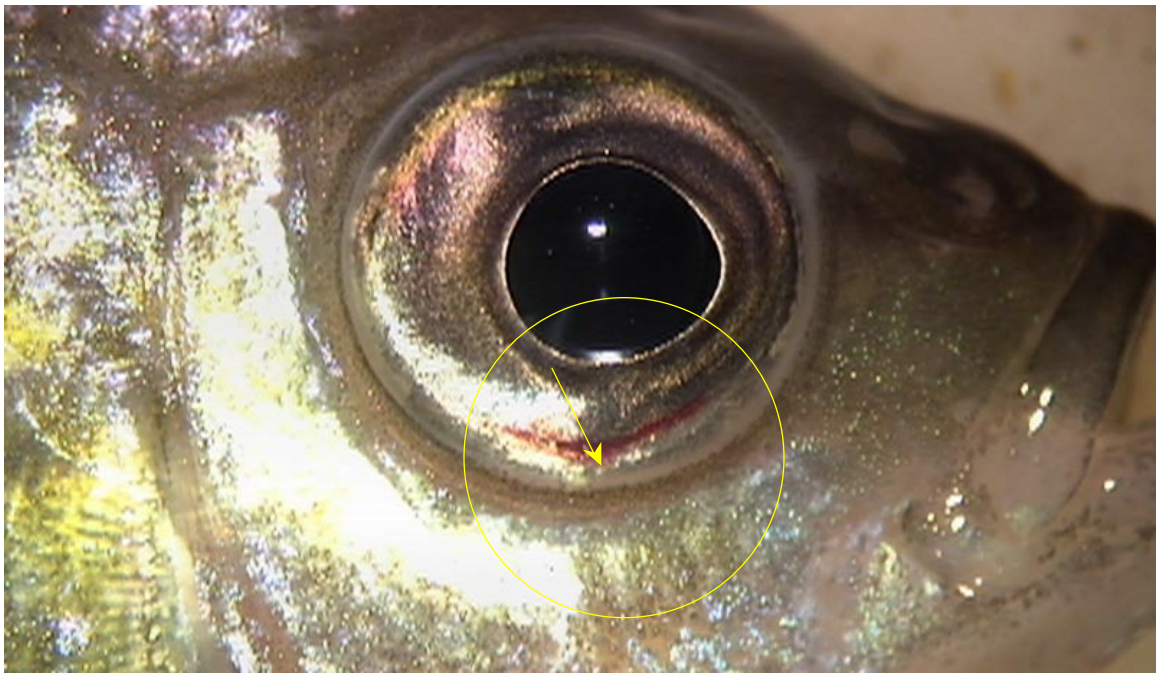


Figure 4.11. Hemorrhage in the Eye of a Bluegill After Acclimation at 135% TDG at 191 kPa and Subsequently Subjected to Turbine Passage Sequence. Fish died from other injuries.

Figure 4.12. Bluegill Sinking to the Bottom as Pressure Increases During the Turbine Passage Sequence

Figure 4.13. Bluegill Reacting to Pressure Spike During Turbine Passage Sequence

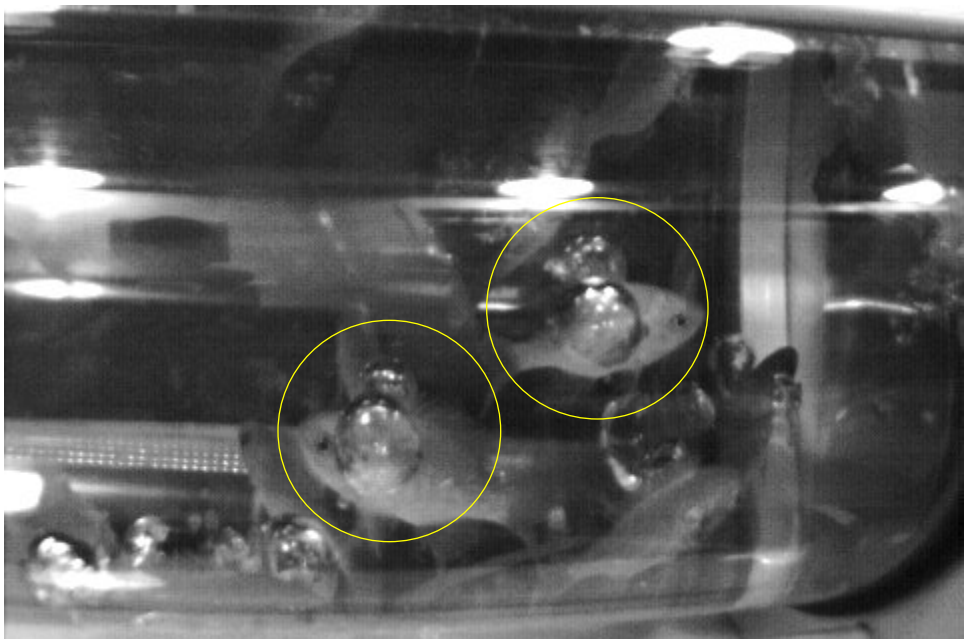


Figure 4.14. Large Bubbles Appearing from Under the Gill Operculum During Turbine Passage Sequence at the Instant When Pressure is Reduced from 400 to 3-7 kPa

Figure 4.15. Spiked Bluegills Sinking to the Bottom of the Holding Trough Immediately Following Turbine Passage Sequence

Figure 4.16. Response of "Control" (non-spiked) Bluegill Acclimating to 191 kPa Immediately After Returning to 101 kPa When Removed from the Hyperbaric Chamber

Over the course of the 48-hr observation period, some fish appeared to recover while others remained on the bottom struggling to maintain an upright position. Necropsies were performed on 680 fish. Depth-acclimated bluegills, regardless of TDG level, suffered high mortality and injury rates. Of the 180 depth-acclimated, "turbine-passed" bluegills (60 each at 100, 120, and 135% TDG), 69 died and 96 were injured. Only 15 fish appeared to be uninjured. A ruptured swim bladder, indicated by the presence of bubbles floating freely in the viscera (Figure 20), was the most common injury, accounting for 95 of the 96 observed injuries. Some of these fish had other injuries as well (i.e., hemorrhages in one or both eyes). Surface-acclimated bluegills exposed to the turbine passage simulation also had swim bladder rupture, but at a lower rate. Of 167 surface-acclimated bluegills alive at the end of the acclimation period that were turbine-spiked, 9 fish that later died and 33 surviving fish (42 total), had ruptured swim bladders, leaving 125 uninjured.

Most of the mortalities or injuries observed in control (non-spiked) bluegills attributable to test parameters was in the 135% surface-acclimated groups. A total of 12 of 60 of these fish died from acute GBT (Table 4.4), and two other fish had eye injuries (exophthalmia and/or hemorrhage). Of the 180 non-spiked acclimated at 30 ft of depth, a total of 3 fish (1 fish acclimated at 100% TDG and 2 fish acclimated at 135% TDG) died of embolism and/or swim bladder rupture during the 48-hr holding period.

A small number (4 of 360 fish, including one surface acclimated and 3 depth-acclimated fish) died during the 48-hr post-test holding period from a bacterial (*Flexibacter columnaris*) infection of the gills.

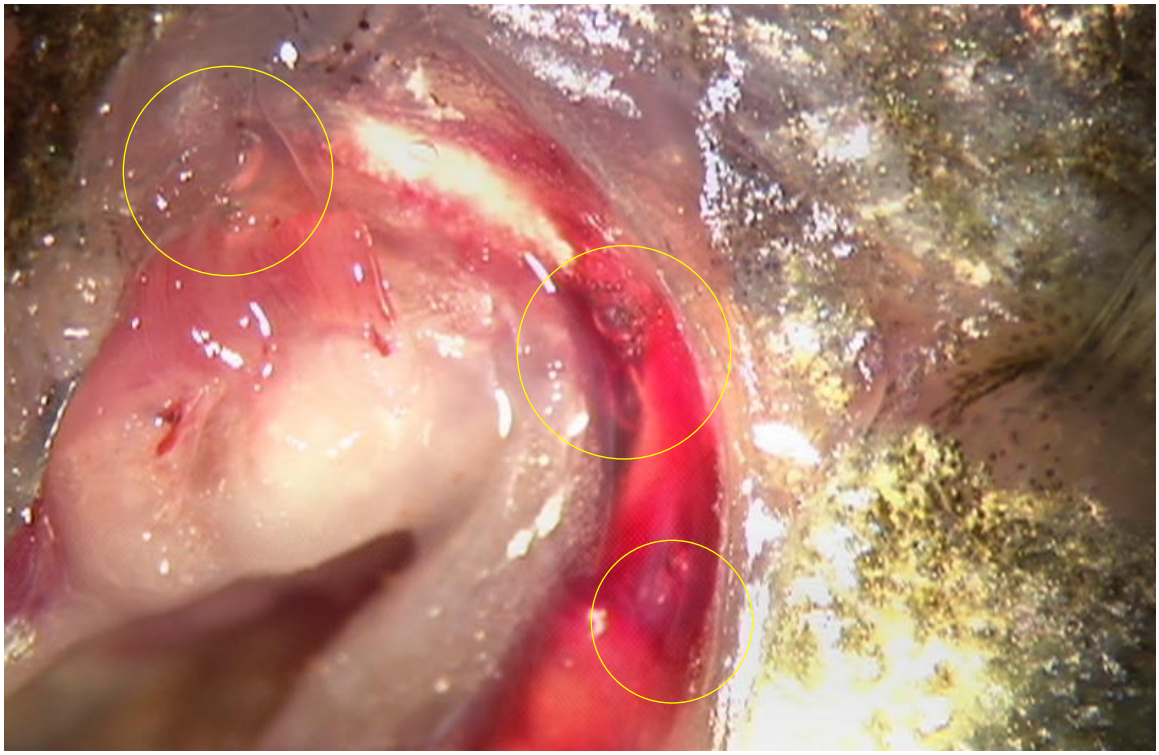


Figure 4.17. Hemorrhaging and Loose Bubbles Visible in the Pericardial Cavity of Bluegills One Hour After Exposure to the Turbine Passage Sequence

4.2 Statistical Analyses

Table 4.5 summarizes the results of the analyses testing an overall difference in each of the four factors for each of the four responses. P-values that have been bolded indicate significance and show there is a significant difference in the levels of that factor for that particular response. Table 4.6 shows the proportions tested in Table 4.5 and can be used to show how the levels differ within a factor.

As expected, the ***test*** factor was significant for Spike Mortality, Injured, and Uninjured. When looking at the GBT mortality, there was no difference between the Spike and Non-Spike runs. The factor ***species*** was also significant for Spike Mortality, Injured, and Uninjured. The Bluegill appears to be having the worst time with higher Spike and injuries. The ***gas*** level of 135 had a significantly higher proportion of GBT, although there were no differences in the ***gas*** levels for Spike fatalities and injuries. For the factor ***depth***, surface level runs had a significantly higher proportion of GBT fatalities, while 30-foot depth runs had a significantly higher proportion of Spike Mortalities.

Table 4.5. p-Values for Each Response and Each Factor (bolding means a significant difference was found)

Factor	GBT Mortality	Spike Mortality	Injured	Uninjured
Test (spike & non-spike)	0.4725	**<0.0001**	**<0.0001**	**<0.0001**
Species	0.2654	**<0.0001**	**0.0013**	**0.0191**
Gas	**<0.0001**	0.6679	0.4784	**0.0001**
Depth	**<0.0001**	**0.0299**	0.0567	0.5961

Table 4.6. Overall Proportions

Factor	GBT Mortality	Spike Mortality	Injured	Uninjured
Test				
Spike	0.100	0.081	0.171	0.648
Non-spike	0.098	0	0.009	0.893
Species				
Bluegill	0.040	0.108	0.182	0.669
Fall Chinook	0.118	0.009	0.012	0.862
Rainbow	0.143	0	0.071	0.787
Gas				
100	0.003	0.031	0.075	0.892
120	0.024	0.040	0.096	0.840
135	0.292	0.052	0.101	0.556
Depth				
0 ft (surface)	0.202	0.009	0.045	0.744
30 ft	0.004	0.069	0.131	0.795

The significant interactions are listed for each response variable in Table 8. Only GBT and Uninjured have significant interactions. In each case, these interactions were with ***species*** and ***gas***, and with ***gas*** and ***depth***. These interactions are visible on Figure 4.18. As the gas increased, the bluegill had significantly fewer GBT mortalities than did the other species. Also, those fish at the surface had significantly more GBT fatalities at the higher gas levels than those fish acclimated at pressures found at 30 ft depth.

Table 4.7. Significant Interactions for Each Response Variable

GBT Mortality	Spike Mortality	Injured	Uninjured
Species – gas *gas - depth*	None	None	*Species – gas* *gas - depth*

Each species was also analyzed individually, without including the control (Non-spike) data. These results are summarized in Table 4.8. Figure 4.18 can be used to better understand significant differences found in Table 4.8.

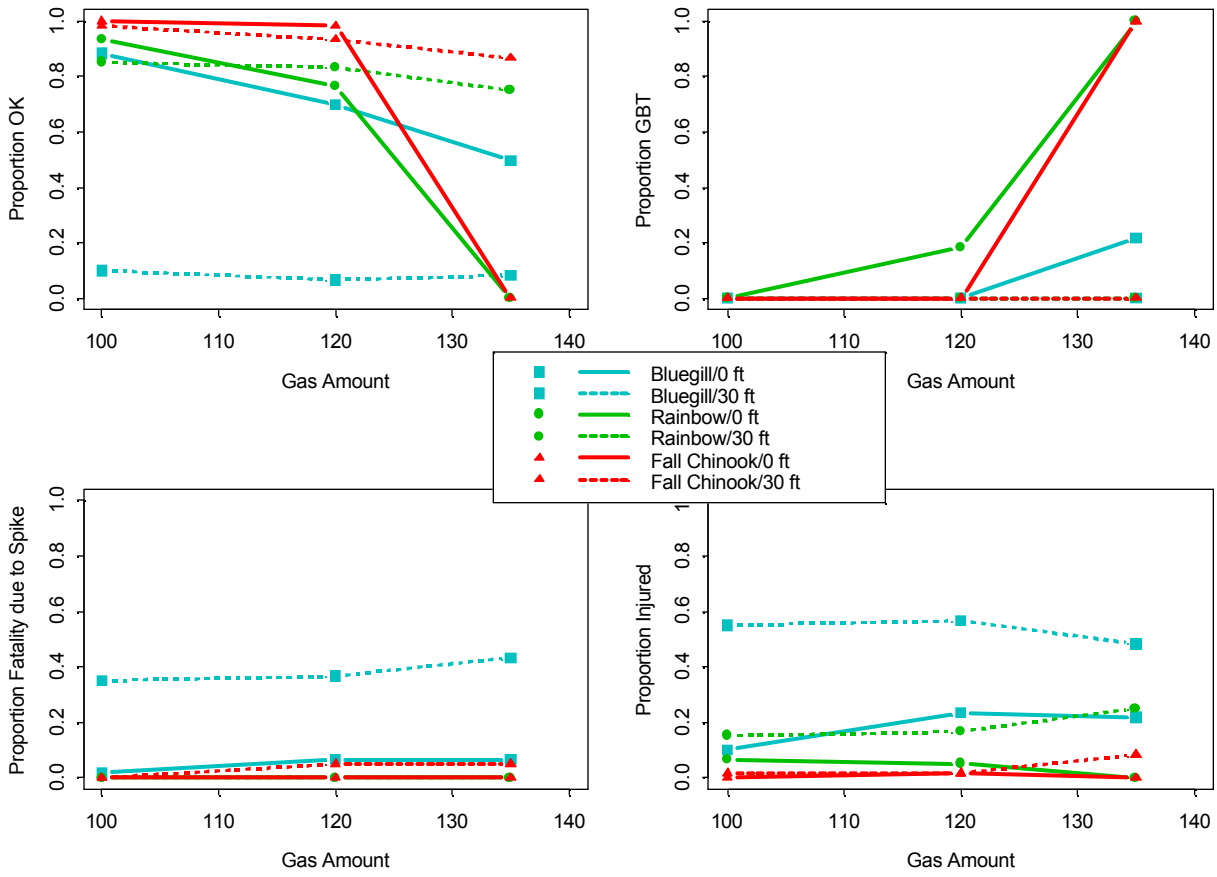


Figure 4.18. Proportion OK (uninjured), Mortality due to GBT and Spike, and Proportion Injured Plotted Against ***Gas***, for Each ***Species/Depth***

Table 4.8. p-Values for Analyses Finding Significant Differences in Gas and Depth for Each Species (no control data included in the analyses)

Factor	GBT Mortality	Spike Mortality	Injured	Uninjured
Bluegill				
Gas	**0.0342**	0.9429	0.8503	0.7753
Depth	0.0662	**0.0003**	**0.0013**	**0.0003**
Fall Chinook				
Gas	0.0773	0.3694	0.6201	**0.0066**
Depth	0.1213	0.0826	0.1480	0.5073
Rainbow				
Gas	0.1724	No Effect	0.8371	**0.0276**
Depth	**0.0072**	No Effect	**0.0061**	0.8842

For the bluegill, there was a significant difference between the ***gas*** levels only for GBT fatalities. GBT fatalities only occurred at ***gas*** level 135. Significant differences between depths were found in proportion of Spike fatalities and injuries. As Figure 4.18 shows, the depth of 30 ft had significantly higher proportions of Spike fatalities and injuries.

For the fall chinook salmon, significant differences at the 0.05 significance level were only found for the factor ***gas*** and the response proportion uninjured. The ***gas*** level of 135 had significantly fewer fish uninjured than the other levels. There were two other factors that were close to being significant. The factor ***gas*** was nearly statistically significant for the GBT mortality proportion for (p-value=0.0773), and the factor ***depth*** was nearly statistically significant for the Spike mortality proportion (p-value=0.0826). Although these were not quite statistically significant, these differences are apparent on Figure 4.18. Figure 4.18 shows an increase in the GBT mortality proportion at the ***gas*** level of 135, especially for those at the surface. Figure 4.18 also shows a slight increase in the Spike mortality proportion at the 3-ft depth.

For the rainbow trout, there was a significant difference between the ***gas*** levels only for proportion uninjured. The ***gas*** level of 135 was significantly lower than the other levels. Significant differences between depths were found in proportion of GBT fatalities and injuries. Surface level runs had significantly higher GBT fatalities, while 30-foot depth runs had significantly higher injuries. There were no Spike fatalities for the rainbow trout.

5.0 Discussion

5.1 Gas Level Effects

The effects of gas supersaturation on fish have been well documented since the 1960s when GBT became a concern due to the construction and operation of mainstem dams on the Columbia and Snake rivers. Weitkamp and Katz (1980) reviewed literature on dissolved gas supersaturation and gas bubble disease and summarized bioassay results for different fish species, gas levels, water temperatures, and water depths. In 1967, juvenile salmonid smolts held at Priest Rapids Dam during the spring outmigration had a high incidence of external gas bubble signs and mortalities (Ebel 1969). However, fish held in submerged pens 2.5 m or deeper did not show GBT symptoms. Beiningen and Ebel (1970) reported that gas supersaturation levels of 123 to 143% downstream of John Day Dam caused mortalities in juvenile and adult salmon and steelhead. Bentley et al. (1976) found that susceptibility of squawfish to elevated gas levels was similar to juvenile salmonids, with 100% mortality occurring in 20 hr at 126% TDG. Montgomery and Becker (1980) observed external gas bubble disease in 72 and 84% of smallmouth bass and northern squawfish, respectively, when TDG levels exceeded 115%.

By 1975, gas supersaturation problems associated with mainstem dams on the Columbia River were greatly reduced (Ebel 1969), and research on the effects of gas supersaturation declined. However, interest in gas supersaturation resurfaced in the 1990s when fisheries managers implemented increased spill at dams to improve passage conditions for salmon smolts during the spring outmigration. Elevated TDG levels came with increased spills. Mesa et al. (2000) assessed GBT in juvenile chinook salmon and steelhead at 110, 120, and 130% TDG. The LT_{20} (time for 20% to die) at 120% TDG was 40 to 120 hr for chinook salmon and 20 to 35 hr for steelhead. At 130%, LT_{20}'s were 3 to 6 hr and 5 to 7 hr, respectively. Counihan et al. (1998) exposed larval white sturgeon to TDG levels of 118 and 131%. Gas bubble signs were observed, but no mortalities occurred in 10 days at 118%, whereas 50% died in 13 days at 131% TDG. Ryan et al. (2000) found that signs of GBT in fish exposed at <120% TDG were rare, and at levels >120%, the researchers were able to develop a model to predict the extent to which fish displayed external GBT signs.

External GBT in fish is reversible. Elston et al. (1997) found that gas bubbles in the fins, gills, and lateral line can be reabsorbed and their presence go undetected due to pressure increases when fish sound in reservoirs or holding ponds or during dam passage. The significance of these findings raised questions about the accuracy of GBT monitoring programs at dams. AASI (1998) conducted extensive research on the physiology of gas bubble formation in fish.

Of the three species tested, bluegills were the most resistant to acute and chronic GBT, and rainbow trout were the least resistant (bluegill > fall chinook salmon > rainbow trout). A TDG level of 135% for up to 22 hr resulted in 100% mortality for rainbow trout and fall chinook salmon and only partial mortality (~21%) for bluegills.

At 120% TDG, some rainbow trout (~14%) died during the acclimation period; however, all fall chinook salmon and bluegills survived, and few showed external signs of GBT. Typical chronic (sub-lethal) GBT signs included gas bubbles in the fins, bubbles in an eye, and occasionally exophthalmia of one or both eyes. All external signs of GBT disappeared during the 48-hr post-exposure holding period except for bubbles inside the eye. Rainbow trout held at 135% TDG at surface pressure (101 kPa) started dying ~2 hr into the acclimation period, whereas fall chinook salmon did not start dying until ~5 hr into the acclimation. Although the onset of deaths with bluegills was not observed directly, acclimation pressure records indicated that mortalities might have started about 9 to 10 hr into the acclimation period.

Gas levels resulting in acute and chronic GBT for fall chinook salmon and rainbow trout were consistent with results from previous GBT studies using our hyperbaric chamber reported in Montgomery Watson (1995). No visible signs of GBT were observed for any species when held at 191 kPa (equivalent of 30 ft of depth). However, when bluegills were returned to and held at surface pressure (101 kPa), most had over-inflated swim bladders and struggled to achieve neutral buoyancy in the holding trough. Rainbow trout and chinook salmon, which remained negatively buoyant when held at 191 kPa, quickly adapted when returned to surface pressure.

In response to the National Marine Fisheries Service's (NMFS) Biological Opinion on the operation of the federal Columbia River Power System (NMFS 1995), the U.S. Army Corps of Engineers (Corps) initiated a Dissolved Gas Abatement Program that is intended to reduce dissolved gas supersaturation associated with spills at federal hydroelectric dams on the Columbia and Snake rivers. A Gas Abatement Workshop was held during October 1996 to review potential structural and operational changes that could be used to improve fish survival during spill events. The Corps is currently pursuing three avenues for gas abatement: 1) reducing the mass of total dissolved gas produced (by the use of flow deflectors and raised stilling basins); 2) minimizing new gas production (by the use of submerged passageways, submerged gates, and turbine flows); and 3) flow degassing in an elevated tailrace (R2 1998). The workshop participants cautioned that some of these gas abatement measures could cause mechanical injury to fish in spillways and stilling basins. A review of available literature (R2 1998) was conducted to assess the negative aspects of spill and gas abatement measures.

NMFS feels that it would be difficult to stay below the U.S. Environmental Protection Agency (EPA) gas saturation criterion of 110% in the Columbia River system without major disruption of the power system, and suspects that values of ≤120% may be sufficiently protective (NMFS 1998, 2000). However, there are many unanswered questions about the effects of gas supersaturation on both juvenile and adult salmonids. Our experiments suggest that fish may be killed by dissolved gas supersaturation in two ways: 1) the direct effect of GBT, and 2) TDG-caused changes in behavior that lead to higher turbine-passage mortality. Gas saturation values of ≤120% are unlikely to cause lethal GBT in the three species we tested. However, our data indicate that if fish change their distribution from the surface to greater depths to compensate for dissolved gas supersaturation (NMFS 2000), the low pressure spike associated with turbine passage may be more lethal. In view of our findings, it would be desirable both to reduce the amount of gas supersaturation (by reducing spill or the amount of gas added during spill) and develop advanced turbines that operate efficiently at a higher pressure downstream from the runner.

5.2 Turbine Passage Effects

Weitkamp and Katz (1980) listed several researchers who confirmed the benefits of hydrostatic compensation for protection from acute and chronic GBT effects as was observed in our tests. However, potential complications for fish equilibrated to elevated TDG levels at depth as it relates to dam passage was not addressed in early studies. Montgomery Watson (1995) evaluated the effects of turbine passage on rainbow trout acclimated to shallow (surface) water conditions and elevated gas levels (115 to 130%). Although swim bladder rupture during turbine passage was a concern, it was not observed. Fish exposed to turbine passage simulations with TDG levels of 125% or higher were more susceptible to predation than controls (turbine passage at 100% TDG). No turbine passage tests were completed with depth-acclimated fish.

Our tests are among the first to combine the effects of elevated TDG, hydrostatic compensation, and turbine passage. Although we expected acute GBT at 135% TDG for surface-acclimated rainbow trout and fall chinook salmon, tests were completed at this level to compare our results to previous studies and observations and as a baseline comparison for depth-acclimated fish. In addition, there is little information on the effects of GBT and turbine passage on bluegills.

In the turbine passage simulation, pressure built from acclimation pressure (101 or 191 kPa) to ~400 kPa over a 30- to 60-sec period. Pressure held at 400 kPa for 15 sec, then suddenly dropped to 4 to 7 kPa in about 0.1 sec. At the instant of the pressure spike, bubbles sometimes briefly appeared within the chamber. Some of these bubbles floated to the surface and accumulated, while other bubbles "flashed" and disappeared. Pressure stayed at this sub-atmospheric level for ~0.2 sec before increasing to ~200 kPa over the next ~5 sec, then gradually returned to 101 kPa over ~30 sec, completing the sequence.

Fish reacted when the "spike" (pressure change from 400 kPa to <7 kPa) occurred, but we don't know if their reaction was due to the sudden pressure change, noise, or mechanical jarring of the hyperbaric chambers. Since both chambers were mounted on the same frame, the noise and jarring was similar in both chambers. Fish in the non-spiked chamber reacted less, indicating that the response was likely due to sudden pressure change.

No loss of equilibrium or other signs of injury were apparent for rainbow trout. All fish appeared normal when removed from the chambers and moved to the holding trough for 48-hr observations. When first placed in the troughs, most of the rainbow trout were negatively buoyant, but they soon filled their swim bladders by gulping air from the surface and swam normally. In three tests (100% TDG surface-acclimated, 100% TDG depth-acclimated, and 120% TDG depth-acclimated), a few rainbow trout became overly buoyant ("floaters") over the 48-hr holding period and struggled to swim normally. The "floaters" occurred only in the "spiked" groups and not in the "control" groups, but were not related to TDG level. Although the condition appears to be directly related to the pressure spike, the exact cause is unknown.

Reaction of fall chinook salmon to the pressure spike was similar to that of rainbow trout; however, in four tests (two tests at 120% TDG depth-acclimated and two tests at 135% TDG depth-acclimated), some fish experienced loss of equilibrium and went into convulsions immediately after the pressure spike. In

the 120% tests, all 3 fish experiencing loss of equilibrium died within 1 hr. In one of the 135% tests, a single fish that lost equilibrium recovered. In the other 135% TDG depth-acclimated test, 2 fish that lost equilibrium and one additional fish died within 1 hr. Macroscopic examination revealed that all mortalities were caused by acute GBT. Bubbles were usually visible in the atrium, and blood flow to the gills was blocked, as indicated by very pale gill filament color. Many depth-acclimated fall chinook salmon also suffered ruptured swim bladders. This condition was determined by necropsy when fish died or after 48 hr. Ruptured swim bladders were observed in only 1 of 60 fish depth-acclimated at 100% TDG, 4 of 60 fish depth-acclimated at 120% TDG, and 8 of 60 fish depth-acclimated at 135% TDG. No depth-acclimated "control" fish had ruptured swim bladders.

The turbine passage simulation was especially harmful to bluegills. High injury and mortality rates were observed in all depth-compensated (191 kPa) groups, regardless of TDG level. The most common injury was swim bladder rupture and hemorrhaging within the body cavity. The source of the hemorrhage was not determined. Surface acclimated bluegill also suffered higher injury and mortality rates than did rainbow trout or chinook salmon, but less than depth-acclimated bluegills. Unlike depth-compensated fish, injury/mortality rates were proportionately higher in groups acclimated at 120 and 135% TDG levels (12, 30, and 36% injury rates for 100, 120, and 135% TDG, respectively). Non-spiked (control) bluegills did not have ruptured swim bladders or hemorrhaging within the body cavity. However, many non-spiked, depth-compensated fish were excessively buoyant when first place in the holding troughs.

It should be noted that the 7- to 10-cm-long bluegills tested in these experiments are larger than those frequently entrained at hydroelectric power plants in the U.S. Juvenile bluegills with an average length of about 5 cm are more commonly entrained. We do not know what effect size has on the response of bluegills to the pressure changes associated with turbine passage. Also, at most hydroelectric power plants that entrain bluegills, the fish are likely to be surface-oriented and the water is not supersaturated with dissolved gases. Our worst case experimental conditions (combinations of gas supersaturation, depth acclimation, and turbine passage) are probably rare outside of the Columbia River Basin.

5.3 Fish Behavior Within the Hyperbaric Chambers

At surface pressure acclimation (101 kPa) all three species were neutrally buoyant during the acclimation period. Fish swam freely within the hyperbaric chambers, although sometimes fish rested on the bottom. At 30 ft depth acclimation (191 kPa), all fish were negatively buoyant when the hyperbaric chambers were first pressurized, sinking to the bottom. Rainbow trout and fall chinook salmon remained negative throughout the acclimation period, swimming "head-up/tail-down" to rise from the bottom of the chamber. However, bluegills were able to inflate their swim bladders to reach neutral buoyancy within 1 to 2 hr and suspend motionless within the water column and off the bottom of the chamber.

When the turbine pressure simulation was initiated and pressures within the hyperbaric chamber increased from either 101 or 191 kPa to ~400 kPa, bluegills were again negative and sank to the bottom of the chamber. Since rainbow trout and fall chinook salmon were already negatively buoyant, swimming behavior did not change significantly as pressure increased.

In response to increasing pressures, fish may actively swim within the turbine to areas that would not be predicted based on modeling of flow fields and neutrally buoyant objects. Harvey (1963) observed an increase in the rate of pectoral fin movements and angle of the body (head upwards) among sockeye salmon in response to pressure increases. Many investigators have observed a tendency for salmonids to swim downwards (sound) in response to increased pressure (Harvey 1963; Muir 1959). This sounding behavior would reinforce the natural tendency of the fish to sink under increased pressures (because the swim bladder becomes compressed). Consequently, actively swimming salmonids may not act like neutrally buoyant objects within the high-pressure region of turbines, but rather, may move to regions of the turbine that pose relatively greater or lesser risk. The effects of the combination of increased body density, sounding behavior, and other directed and random fish movements on turbine-passage mortality are unknown.

6.0 Conclusions

The gas supersaturation level that causes acute GBT varies among species. Resistance to acute GBT, from greatest to least, is bluegill >fall chinook salmon > rainbow trout. Bluegills also had a lower incidence of chronic GBT symptoms than fall chinook salmon and rainbow trout.

Salmonids are not able to quickly fill their swim bladders with gases dissolved in the blood, and therefore, cannot easily inflate their swim bladders without access to air at the surface. However, bluegills have the ability to fill their swim bladders from gases dissolved in the blood and can become neutrally buoyant without surfacing to gulp air.

The frequency, type, and severity of injuries related to pressure changes during turbine passage vary among species, as described below.

- Bluegills, and presumably most physoclistous fish, are extremely susceptible to swim bladder rupture when exposed to the sudden pressure change during turbine passage. The "worst case" turbine passage scenario (pressure dropping to 2-10 kPa) was more harmful to bluegills acclimated at 191 kPa than at 101 kPa. TDG level had only a small additive effect on the injury/death rate due to the pressure spike.

- Fall chinook salmon suffered ruptured swim bladders, but at a much lower rate than bluegills. When acclimated to elevated gas levels at 191 kPa, the turbine passage sequence also caused instantaneous bubble formation in a small number of fish, resulting in immediate death.

- A black discoloration appeared on the top of the head of some rainbow trout, signs of an unknown trauma. The trauma was not observed in chinook salmon. The frequency of this trauma was correlated to acclimation at 191 kPa in fish subjected to the turbine passage sequence. The discoloration rarely appeared in fish acclimated at 101 kPa or in non-spiked fish. Swim bladder rupture was not observed in rainbow trout, regardless of TDG level or acclimation pressure. Although listed as an "injury," the discoloration may not have actually affected the health and/or survivability of rainbow trout.

If dissolved gas supersaturation is not a problem, our experiments suggest that the brief low pressure spike to about 0.1 atmosphere downstream from the turbine runner will cause little direct mortality among surface-acclimated salmonids. If fish are entrained from greater depths, such that their swim bladders contain more gas and will expand more during the low pressure spike, the injury and mortality rates will be higher.

The results suggest that injury/mortality rates would likely be reduced or eliminated if the nadir of the turbine pressure spike was higher, as is expected to be the case with new fish-friendly turbine designs. A follow-up series of tests is needed under a modified pressure regime that more closely reflects conditions expected in new turbine designs, or with a nadir of ~50 kPa.

The low pressure spike is especially a problem if the water is highly supersaturated with gases (well beyond water quality standards), and the fish respond to the supersaturation by depth compensation. While this occurs in the Columbia River Basin when water is being spilled to transport fish downstream (NMFS 2000), dissolved gas supersaturation is probably relatively rare in other river basins.

7.0 References

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Appendix A

Appendix A

The pressure histories for fish acclimated at surface depth (14.7 psi, or 101 kPa) and at a depth of 30 ft (27.7 psi, or 191 kPa) are shown in the following five figures. The first two figures are representative of "acclimation files" for surface and depth-acclimated fish, respectively. The computer software recorded surface acclimation pressure as 14.7 psi instead of 0 psi. During the 22-hr acclimation period, pressure within the hyperbaric chamber was maintained slightly above 14.7 psi to prevent the formation and buildup of bubbles in the chamber. A ventilation tube at the top of the hyperbaric chamber was left open at all times to allow bubbles to flow from the chambers. The vent was closed only to measure flow at the hyperbaric chamber discharge line. Closing the ventilation tubes caused a brief "spike" in the pressure. The spike was more noticeable in the acclimation files for depth-acclimated fish because a high proportion of the total flow exited through the ventilation tubes.

The next two graphs show the pressure history during the time when the computer program had control. When the spike sequence was initiated, inflow, outflow, and ventilation tubes were shut, trapping a volume of water within each chamber. When the baseline pressure stabilized, the "Dive" pressure spike sequence was initiated. Over period of about 30 sec, pressure increases from either 14.7 or 27.7 to ~58 psi. After 15 sec, the spike occurs, where pressures drop to near vapor pressure in ~0.1 sec. After the spike, pressures represent passage through the draft tube. When the pressure sequence approaches surface pressure (14.7 psi) the program is terminated, water supply and drain valves are open, and flow through the hyperbaric chambers is restored.

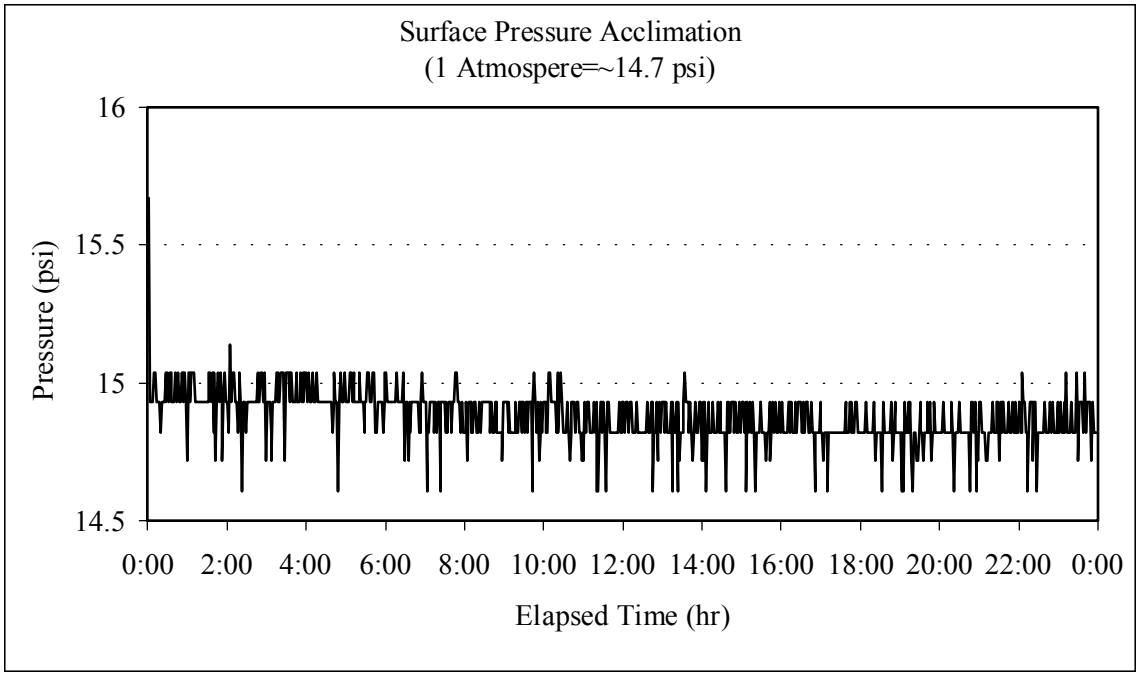

Figure A.1. Example of Surface Acclimation Pressure History

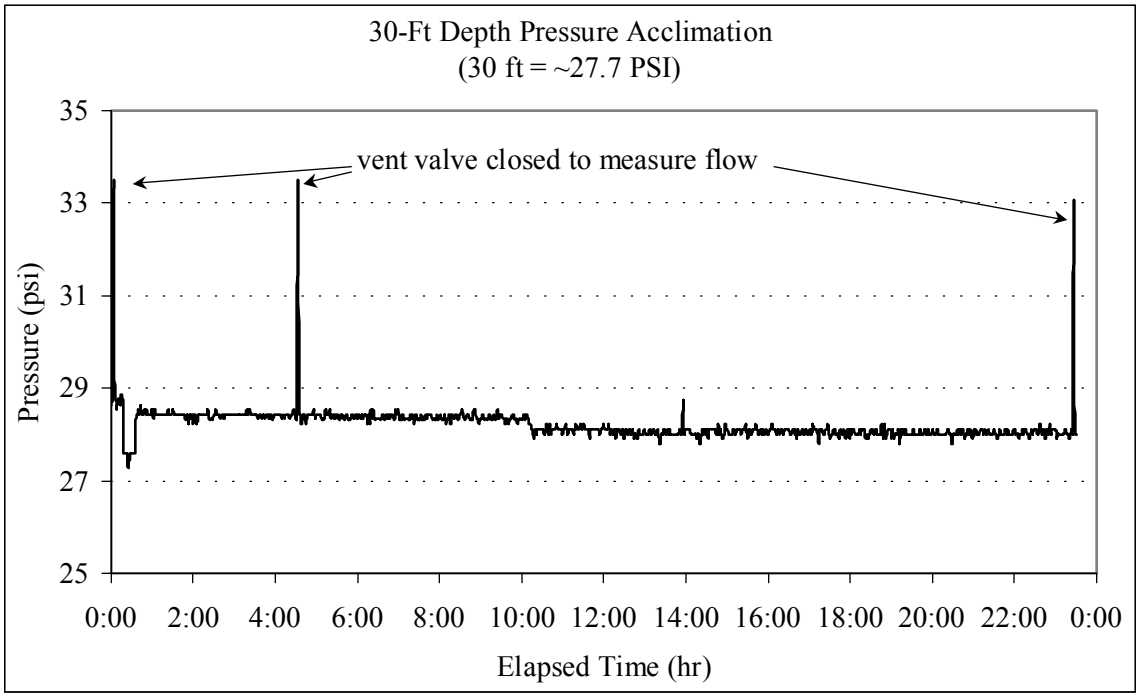

Figure A.2. Example of Depth-Acclimation (30 ft) Pressure History

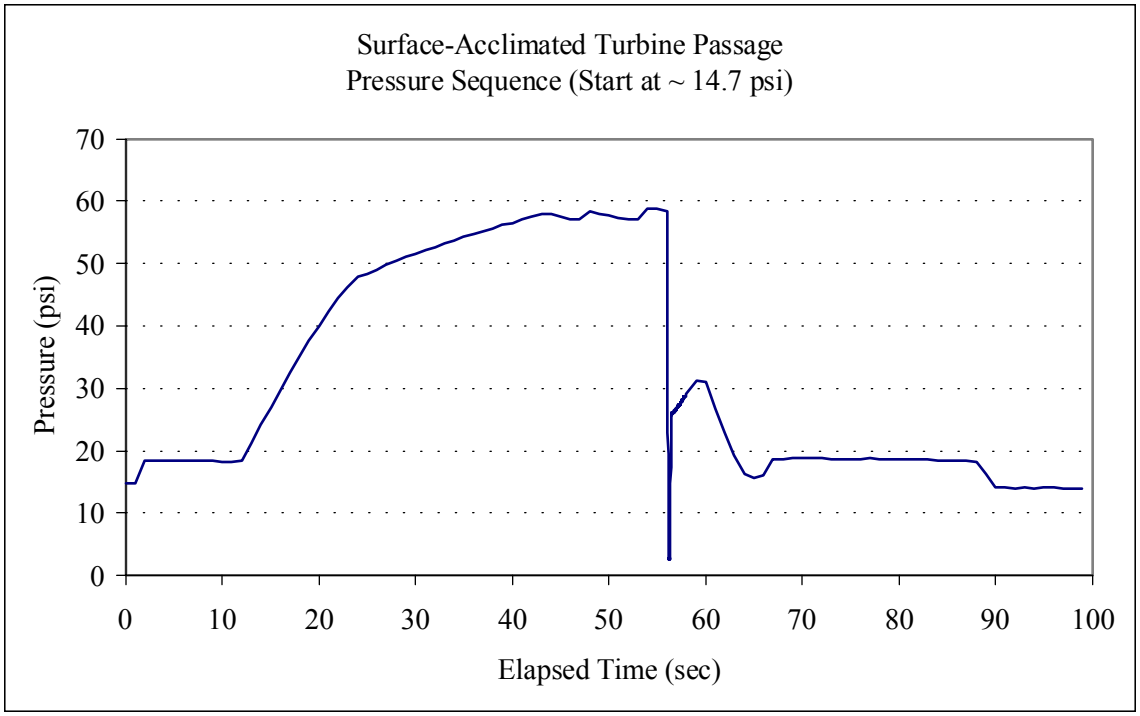

Figure A.3. Example of Computer-Controlled Pressure Sequence for Surface-Acclimated Tests

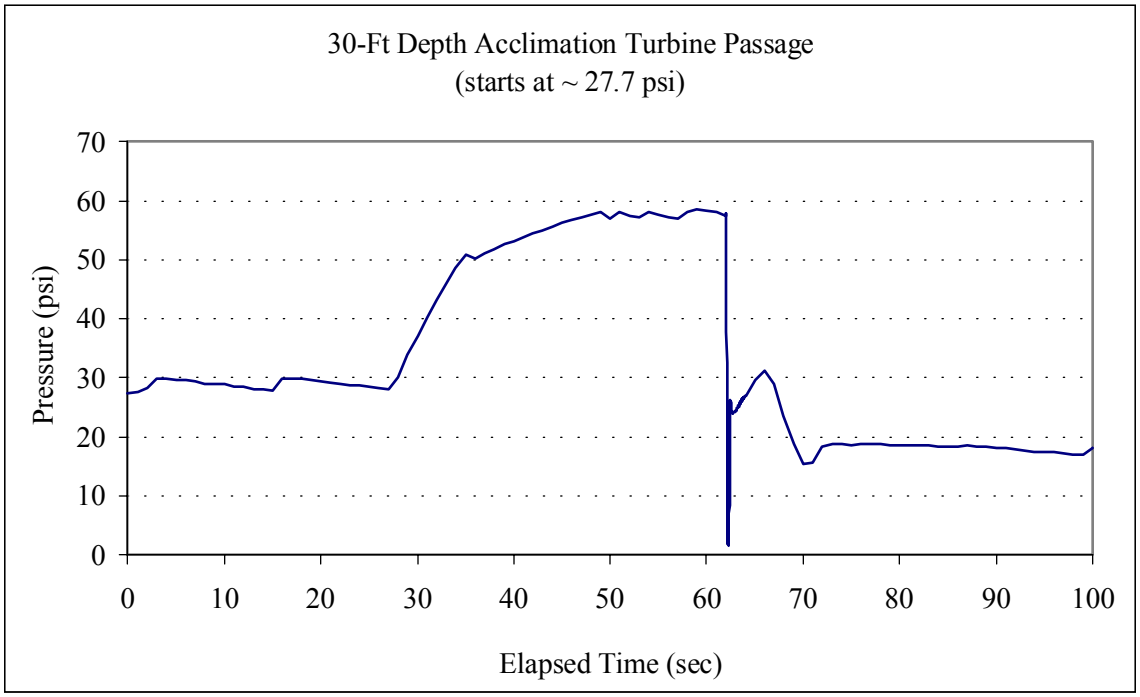

Figure A.4. Example of Computer-Controlled Pressure Sequence for Depth-Acclimated Tests

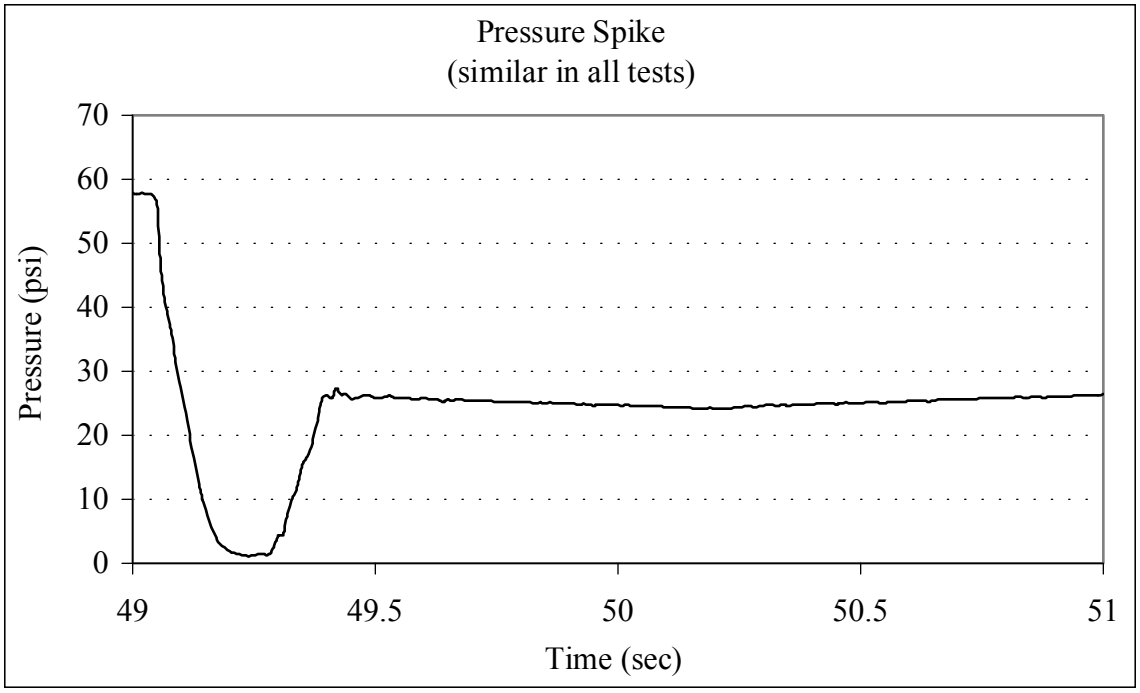

Figure A.5. Example of 2-Second Interval Showing Pressure Sequence When Turbine Passage Spike Occurs