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**Pacific Northwest  
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**Simulated Passage Through a  
Modified Kaplan Turbine Pressure  
Regime: A Supplement to “Laboratory  
Studies of the Effects of Pressure and  
Dissolved Gas Supersaturation on  
Turbine-Passed Fish”**

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Prepared for the:  
Hydropower Program  
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Idaho Falls, Idaho



Pacific Northwest National Laboratory  
Richland, Washington 99352  
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**Simulated Passage Through A Modified Kaplan Turbine  
Pressure Regime: A Supplement to “Laboratory Studies  
of the Effects of Pressure and Dissolved Gas  
Supersaturation on Turbine-Passed Fish”**

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## Executive Summary

Migratory and resident fish in the Columbia River basin are exposed to stresses associated with hydroelectric power production, including pressure changes during turbine passage and dissolved gas supersaturation (resulting from the release of water from the spillway). The responses of fall Chinook salmon and bluegill sunfish to these two stresses, both singly and in combination, were investigated in the laboratory. A previous test series (Abernethy et al. 2001) evaluated the effects of passage through a Kaplan turbine under the “worst case” pressure conditions. For this series of tests, pressure changes were modified to simulate passage through a Kaplan turbine under a more “fish-friendly” mode of operation. The results were compared to results from Abernethy et al. (2001).

Fish were exposed to total dissolved gas (TDG) levels of 100%, 120%, or 135% of saturation for 16-22 hours at either surface (101 kPa) or 30 ft (191 kPa) of pressure, then held at surface pressure at 100% saturation for a 48-hour observation period. Sensitivity of fall Chinook salmon to gas supersaturation was slightly higher than in the previous test series, with 15% mortality for surface-acclimated fish at 120% TDG, compared to 0% in the previous tests. All fish died in the first five hours in both test series at 135% TDG at surface pressures. Bluegill sunfish were again less sensitive to TDG than fall Chinook salmon. Only 10% of surface-acclimated bluegills died at a 135% TDG level, slightly more than died in the previous test series under the same conditions. No bluegills died from GBT if exposed at a water pressure equivalent to 30 feet of depth.

The two species were exposed to a modified pressure regime simulating passage through a Kaplan turbine on the Columbia River, i.e., gradual pressure increase to 4 atmospheres pressure, followed by a rapid (0.1 second) decrease to 0.5 atmosphere, followed by gradual return to 1 atmosphere (surface water pressure). No fall Chinook salmon died during or after exposure to the modified turbine-passage pressures, and no significant injuries were observed. As with the previous test series, it cannot be determined whether fall Chinook salmon were able to acclimate to the greater water pressure during the pre-test holding period. Bluegill sunfish exposed to a modified turbine-passage pressures had lower injury and mortality rates than bluegills subjected to the “worst case” pressure conditions in previous Kaplan turbine pressure tests. Injury and mortality rates for bluegills were again higher than for fall Chinook salmon, especially if they had first been acclimated to water pressures equivalent to 30 feet of depth.

These data indicate that altered operating conditions that raise the nadir (low point) of the turbine-passage pressure regime could reduce the injury and mortality rates of fish during turbine passage. Fall Chinook salmon were not injured or killed when subjected to the modified pressure scenario. Bluegills were more sensitive to pressure effects than fall Chinook salmon, but injury and mortality rates were lower under the modified Kaplan pressure regime. This improvement was particularly significant among fish that were acclimated to greater water pressures (traveling at greater depth).



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# Glossary

<b>AHTS</b>	Advanced Hydropower Turbine System Program of the U.S. Department of Energy
<b>GBT</b>	Gas Bubble Trauma
<b>gpm</b>	gallons per minute
<b>Kaplan turbine</b>	An axial-flow (propeller-type) turbine with adjustable runner blades and adjustable guide vanes
<b>kPa</b>	kilopascals; a measure of pressure. 101 kPa = 1 atmosphere = 14.73 psi
<b>nadir</b>	the lowest pressure in the time vs. pressure regime experienced by fish in these experiments.
<b>physoclistous</b>	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.
<b>physostomous fish</b>	Fish that have a duct (pneumatic duct) which 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.
<b>pneumatic duct</b>	The duct that connects the swim (gas) bladder and the gut in physostomous fish
<b>pressure spike</b>	In these experiments, the rapid water pressure decrease from several times atmospheric pressure to a low point (nadir) of less than one atmosphere
<b>psi</b>	pounds per square inch, a measure of pressure
<b>spill</b>	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 supersaturated with air.
<b>swim bladder</b>	An internal gas bladder that has a weight-regulating (hydrostatic) function in higher fishes
<b>TDG</b>	Total Dissolved Gas
<b>TGP</b>	Total Gas Pressure
<b>vapor pressure</b>	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

Migratory and resident fish in the Columbia River basin are exposed to a variety of stresses associated with hydroelectric power production, including pressure changes during turbine passage and dissolved gas supersaturation (resulting from the release of water from the spillway). The responses of rainbow trout, Chinook salmon, and bluegill sunfish to these two stresses, both singly and in combination, were investigated in the laboratory and reported in Abernethy et al. (2001).

The three species were exposed to a pressure regime simulating passage through a Kaplan turbine on the Columbia River, i.e., pressure was gradually increased to 4 atmospheres (atm), or 3 atm of hydrostatic pressure, followed by a rapid (0.1 second) decrease to sub-atmospheric pressure (less than 0.1 atm), followed by gradual return to surface pressure (1 atm). When acclimated to surface water pressures, neither rainbow trout nor Chinook salmon died after exposure to turbine-passage pressures, although some injuries were observed. More pressure-related injuries were noted among rainbow trout that had been held in greater pre-test water pressures. Fall Chinook salmon and rainbow trout did not appear to have a substantially greater “turbine-passage” mortality rate if they were held at greater water pressures before exposure. However, we are not certain whether these salmonids were able to acclimate to the greater water pressure during the pre-test holding period; if not, their experience would have been similar to that of surface-pressure-acclimated fish. Bluegill exposed to turbine-passage pressures had higher injury and mortality rates than salmonids, especially if they had first been acclimated to water pressures equivalent to 30 feet of depth. For all species, the combination of gas supersaturation and turbine-passage pressure regime was more damaging than either stress separately. The highest injuries and mortalities were experienced by bluegills acclimated to a combination of water pressures characteristic of 30-foot depths, dissolved gas supersaturation, and turbine-passage pressures.

These data indicate that fish in the Columbia River basin may be killed by pressure changes associated with turbine passage, especially if they are entrained from greater depths. Bluegills were more sensitive to pressure effects than the two salmonid species, but less sensitive to gas supersaturation. Chinook salmon traveling at the surface experienced low injury and mortality rates from turbine-passage pressure changes. Advanced turbine designs, or altered operating conditions, that raise the nadir (low point) of the turbine-passage pressure regime could reduce the injury and mortality of fish. This improvement would be particularly significant among fish that are acclimated to greater water pressures (traveling at greater depth) because of dissolved gas supersaturation.

Based on the outcome of tests documented in Abernethy et al. (2001), we conducted another test series using a modified Kaplan turbine pressure regime with a nadir of 0.5 atm (50 kPa), representing the most “fish-friendly” operating conditions for Kaplan turbines without major impacts on power generation. Our objective was to compare mortality and injury rates for fall Chinook salmon and bluegills under more “fish-friendly” Kaplan turbine operating conditions to results of previous tests using typical Kaplan pressure regimes occurring at most hydroelectric dams on the Columbia River.

The modified pressure regime is similar to pressures expected for the new advanced turbine runner design developed by Alden Research Laboratory, Inc. and Northern Research and Engineering

Corporation (ARL/NREC team). Based on a consideration of salmonid data in USACE (1991), the ARL/NREC turbine is also designed to produce a slow rate of pressure change and a nadir that is at least 30% of the acclimation pressure (Odeh, 1999). For ARL/NREC's turbine, the nadir is expected to be approximately 10 psi (70 kPa) (Cook, et al. 1997), in contrast to the 7-10 kPa for typical Kaplan turbine operation and 50 kPa in the modified Kaplan pressure sequence. The effects of reducing the rate of pressure change were not evaluated in this test series.

Time/pressure sequences for all phases of the turbine passage scenarios (acclimation, entry into the intake, rate of pressure change, exit in the draft tube and tailrace) were identical to previous Kaplan pressure simulations. Modifications in pressure regimes, test fish, and methods as compared to the Kaplan test series reported in Abernethy et al. (2001) are described below. Results were evaluated independently and also compared to test results for the same species under the previous Kaplan turbine pressure regimes.

## 2.0 Methods

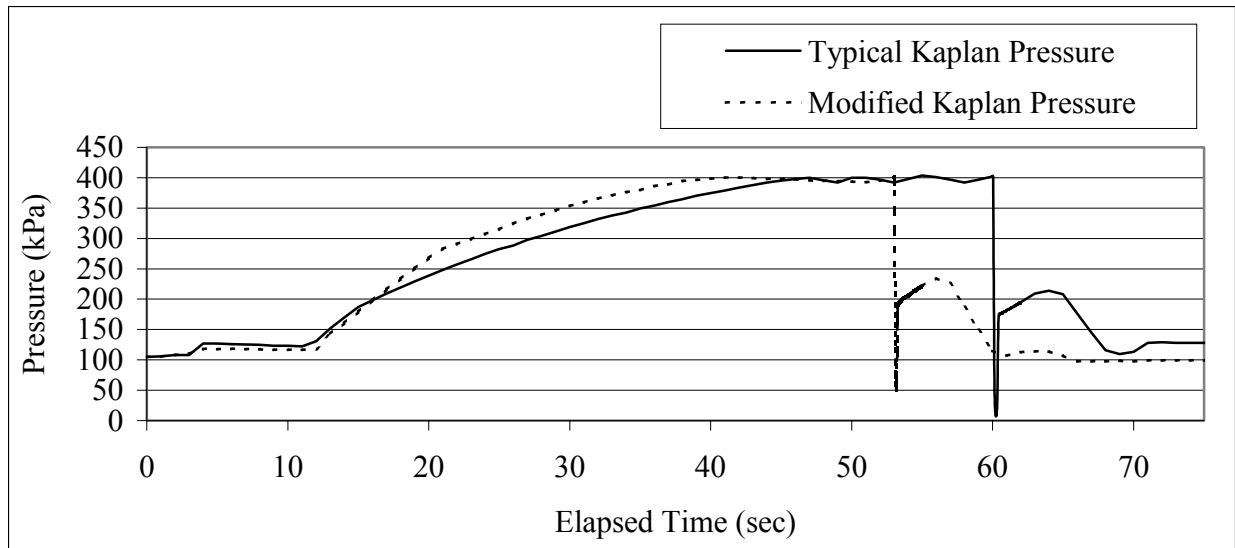
### 2.1 Gas Supersaturation and Turbine Passage Systems

A pressurized packed cell column was used to generate gas-supersaturated water. The system was designed by Point Four Systems of Port Moody, British Columbia for use in previous studies at PNNL and is described in Aspen Applied Sciences (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.

The Turbine Passage System 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 hydroelectric turbine. The exposure chambers for the turbine passage system consisted of two 11-inch (27.5cm) diameter acrylic tubes, 22-inches (55 cm) long. The volume of each cylinder was about 34 L.

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 feet of head (3 atm, or ~400 kPa). The system can drop the pressure from 100 feet (~400 kPa) of head to close to the vapor pressure of water (~1 psi or 2-10 kPa) in 0.1 second.

The Labtech software sub-program controlling the sequence simulating the turbine spike was altered to produce a nadir of 50 kPa (26% of acclimation of depth-acclimated group). Due to the hyperbaric chamber's design, achieving an exact 50 kPa nadir was difficult; the sensitivity and response time of pressure sensors controlling piston movement in the hyperbaric chambers affected the precision. In addition, small bubbles suspended in the water due to elevated TDG levels further affected the reproducibility and precision of the nadir during tests. Several tests were aborted and repeated due to mechanical malfunctions resulting in poor pressure spike results. A sequence with a nadir of 41-55 kPa (0.6 to 0.8 atmospheres) was considered to be a "good" test if no other pressure anomalies occurred during the sequence. The typical and modified Kaplan turbine passage pressure scenarios are compared in Figure 2.1. All pre-test holding and acclimation conditions, and the rest of the turbine passage regime and the depth acclimation (101 and 191 kPa) conditions remained the same as in previous tests reported in Abernethy et al. (2001).



**Figure 2.1.** Comparison of the expected pressure regimes during typical Kaplan turbine operations and operations modified to reduce the nadir.

## 2.2 Fish Stocks

Juvenile fall Chinook salmon were from the same stock (Priest Rapids hatchery stock) as were used in previous tests, but were larger (14 cm FL versus 10 cm) and older (12 months instead of 8 months). Bluegills were acquired from the same vendor (Osage Catfisheries) but were younger (exact age unknown) and smaller (4-7 cm FL versus 7-10 cm FL).

## 2.3 Injury Assessment

The condition of fish (alive, dead, other external symptoms) was checked immediately before and after the pressure sequence. Fish were also checked 1, 24, and 48 h after the pressure sequence was completed. Observations included the numbers dead, experiencing loss of equilibrium, abnormal swimming behavior or buoyancy, and external signs of trauma. Dead fish were examined at the first opportunity (usually at 1 h) to determine cause of death. Fish surviving 48 h were euthanized in MS-222 and examined under a dissecting scope (5-50 power) with the aid of optical fiber lighting and a base light. Examples of various injuries were captured as digital images. Examinations included both external and internal examination. External examination consisted of looking for signs of gas bubble trauma in the fins, skin, eye, and gill filaments. Swim bladder rupture, internal bleeding, and gas bubbles in the heart were determined by internal examination. For bluegills, swim bladder rupture and internal bleeding could be determined without incising the fish by using backlighting.



## 3.0 Results

### 3.1 Fall Chinook Salmon

A series of 17 tests simulating passage through the modified Kaplan turbine pressure sequence was completed during mid-January to mid-March, 2001. 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. Comparison of these results to previous results with Kaplan turbine pressure spike simulations are made in the Discussion section.

#### 3.1.1 Gas Bubble Trauma-Related Death and Injuries

Fall Chinook salmon exposed to elevated total dissolved gas (TDG) levels and held at pressures simulating 30 feet of depth showed no signs of gas bubble trauma (GBT) during acclimation, turbine passage simulation, or the 48-hr post-test observation period (Table 3.1). However, all fall Chinook salmon (80 fish) exposed to 135% TDG levels at surface acclimation died during the acclimation period. At 120% TDG, a small number of fish died (18 of 120 fish) during acclimation. In addition, 3 of 120 fish had signs of GBT (bubbles in the eye) at the end of the 48-hr post-exposure observation period. None of the fish held at 100% TDG at surface acclimation showed signs of GBT.

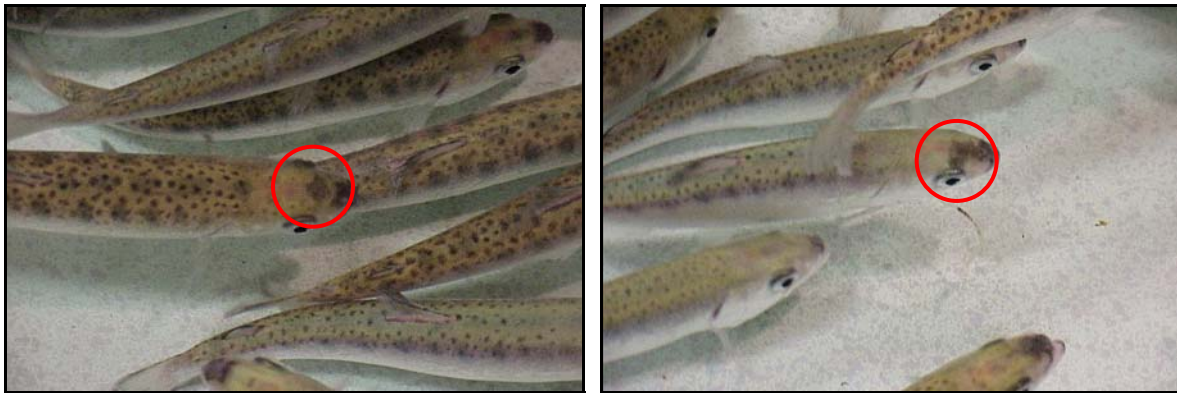
#### 3.1.2 Pressure-Related Death and Injuries

No mortalities occurred for fall Chinook salmon exposed to a simulated turbine passage with a 50 kPa nadir, regardless of acclimation depth or TDG level (TDG levels of 135% were not tested; Table 3.1). In addition, there were no signs of serious injuries (i.e., swim bladder rupture) attributable to the pressure spike associated with turbine passage.

**Table 3.1.** Mortality and injury rates for fall Chinook salmon based on TDG (% saturation), 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	4	5	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
Injured	0	0	0	0	1	1	-	-	0	0	0	0	0	0	0	0	0
OK - no injuries	20	20	20	20	15	14	0	0	20	20	20	20	20	20	20	20	20
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	6	3	20	20	0	0	0	0	0	0	0	0	0
Injured	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0
OK - no injuries	20	20	20	20	14	16	0	0	20	20	20	20	20	20	20	20	20

The only other observed “injury” was the occurrence of a faint cranial spot on the top of the head in some fish ( Figure 3.1). As was reported in Abernethy et al. (2001), the spot occurred in both spiked and non-spiked groups, at all TDG levels, and in both surface and depth-acclimated groups. Fish acclimated to depth had more head spots than fish acclimated to surface pressure, and fish acclimated at elevated TDG levels had more head spots than fish acclimated at 100% TDG. However, the only significant result ( $p$ -value = 0.0015 using AOV) for the frequency of occurrence of the cranial spot was for spiked fish (10.6%) compared to control (non-spiked) fish (1.6%).



**Figure 3.1.** Cranial spot observed on fall Chinook salmon following exposure to simulated turbine spike scenarios.

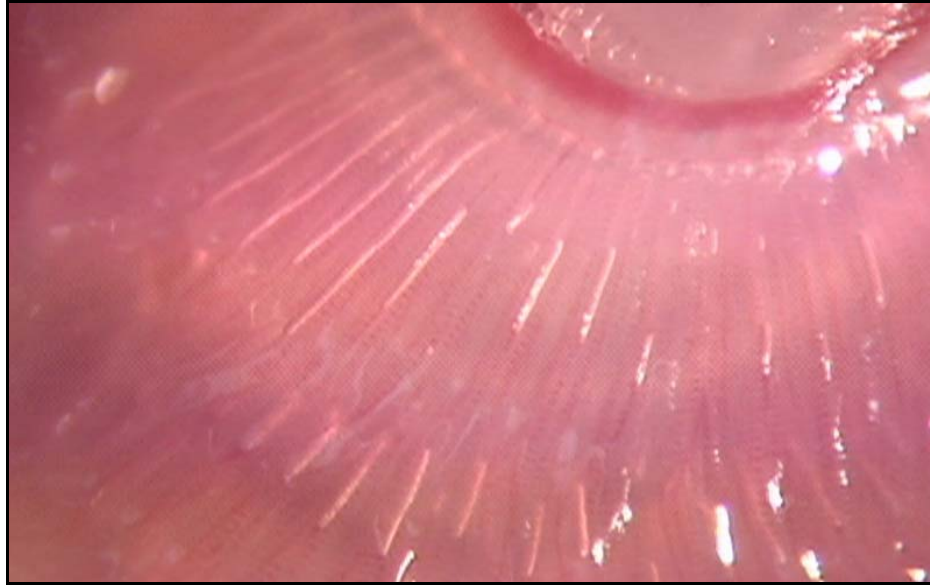
## 3.2 Bluegills

A series of 18 tests was completed during mid-March to mid-April, 2001. 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. Comparison of these results to previous results with Kaplan turbine pressure spike simulations are made in the Discussion section.

### 3.2.1 Gas Bubble Trauma-Related Death and Injuries

Bluegills exposed to elevated TDG levels and held at pressures simulating 30 feet of depth showed no signs of GBT during acclimation. However, two (2.2%) control (non-spiked) fish died. One control fish acclimated at 30 ft depth and 135% TDG developed gas bubbles in the gills (Figure 3.2) and died within one hour after removal from the hyperbaric chamber. Another control fish acclimated at 30 ft depth and 100% TDG that died within the first hour had a ruptured swim bladder and hemorrhaging along the vertebral column. The exact cause of death could not be determined. However, there was no evidence of gas bubbles in the heart or gill filaments.

For surface-acclimated bluegills, 39 of 120 fish exposed to 135% TDG died during the acclimation period (Table 3.2). Others showed external signs of GBT (bubbles in the eyes and fins) but no counts



**Figure 3.2.** Elongated bubbles observed in the gill filaments of a control (non-spiked) bluegill held at surface pressure following acclimation to 30 ft depth and 135% TDG.

**Table 3.2.** Mortality and injury rates for bluegills based on TDG (% saturation), 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 <sup>(a)</sup>	2	3	1 <sup>(a)</sup>	2	3	1	2	3	1	2	3	1 <sup>(a)</sup>	2	3	1	2	3
Dead From GBT	0	0	0	0	0	0	7	5	8	0	0	0	0	0	0	0	0	0
Dead from Spike	0	2	1	0	0	0	0	0	0	2	0	6	0	0	1	8	2	1
Injured	3 <sup>(a)</sup>	17	19	1 <sup>(a)</sup>	20	20	7 <sup>(a)</sup>	6	11	8 <sup>(a)</sup>	13	12	5 <sup>(a)</sup>	15	10	8 <sup>(a)</sup>	12	8
OK – no injuries	17 <sup>(a)</sup>	1	0	19 <sup>(a)</sup>	0	0	6 <sup>(a)</sup>	9	1	10 <sup>(a)</sup>	7	1	15 <sup>(a)</sup>	5	9	4 <sup>(a)</sup>	6	11
Control Group	Surface Acclimation (101 kPa)									30 ft Acclimation (191 kPa)								
	100% TDG			120% TDG			135% TDG			100% TDG			120% TDG			135% TDG		
Dead From GBT	0	0	0	0	0	0	5	6	8	0	0	1	0	0	0	0	0	0
Died During 48-hr period	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	1
Injured	0 <sup>(a)</sup>	13	11	1 <sup>(a)</sup>	12	10	8 <sup>(a)</sup>	3	8	4 <sup>(a)</sup>	8	8	3 <sup>(a)</sup>	10	4	11 <sup>(a)</sup>	14	14
OK – no injuries	20 <sup>(a)</sup>	7	9	19 <sup>(a)</sup>	8	10	7 <sup>(a)</sup>	11	4	16 <sup>(a)</sup>	12	10	17 <sup>(a)</sup>	10	16	9 <sup>(a)</sup>	6	5

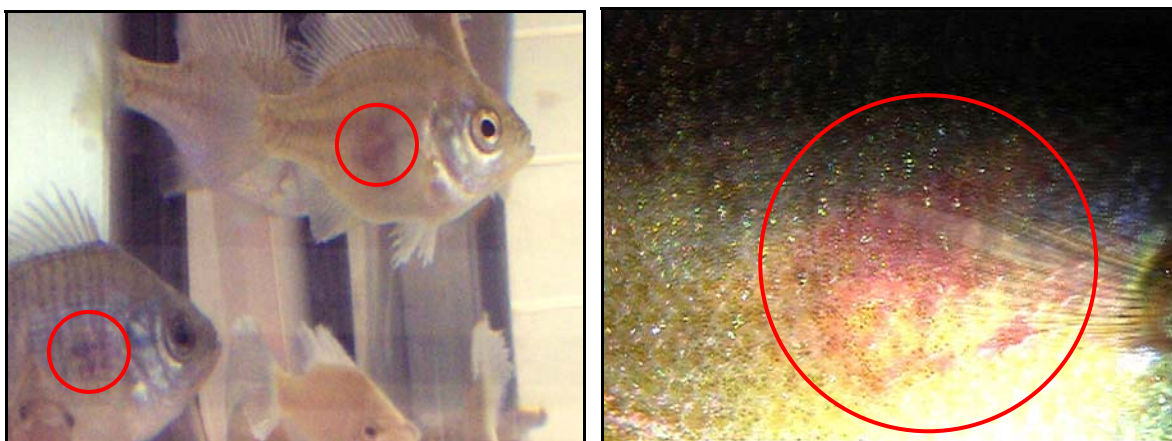
(a) These tests were evaluated for injury using less stringent methods than the subsequent tests. Injured and OK results were not used in the final summary.

were made until after the 48-hour post-exposure observation. By then, most GBT symptoms had disappeared. No surface-acclimated fish exposed to 100% or 120% TDG died or showed external signs of GBT.

### 3.2.2 Pressure-Related Death and Injuries

Effects from exposure to the modified Kaplan turbine pressure sequence were variable (Table 3.2). Mortalities directly attributable to the pressure spike ranged from 0 to 40%. Few surface-acclimated bluegills died as a result of the pressure spike. Only 3 of 160 spiked fish (1.9%, and all were from 100% TDG groups) and none of 161 surface-acclimated control fish died during the post-exposure holding period. Bluegills acclimated to 30 feet of depth had significantly higher ( $p$  value = 0.0289) but more variable mortality rates. For 100% TDG depth-acclimated fish, 8 of 60 fish (13.3%) died, with 0 to 6 fish dying in each 20-fish test group. For 120% TDG depth-acclimated fish, only 1 of 60 fish (1.7%) died. For depth-acclimated fish held at 135% TDG, 11 of 60 fish (18.3%) had spike-related mortality, with a range of 1 to 8 fish dying in each 20-fish test group. Most mortalities had ruptured swim bladders and massive internal hemorrhaging. In addition, mortalities of spiked, depth-acclimated fish exposed to elevated TDG levels gas sometimes had gas bubbles in their hearts.

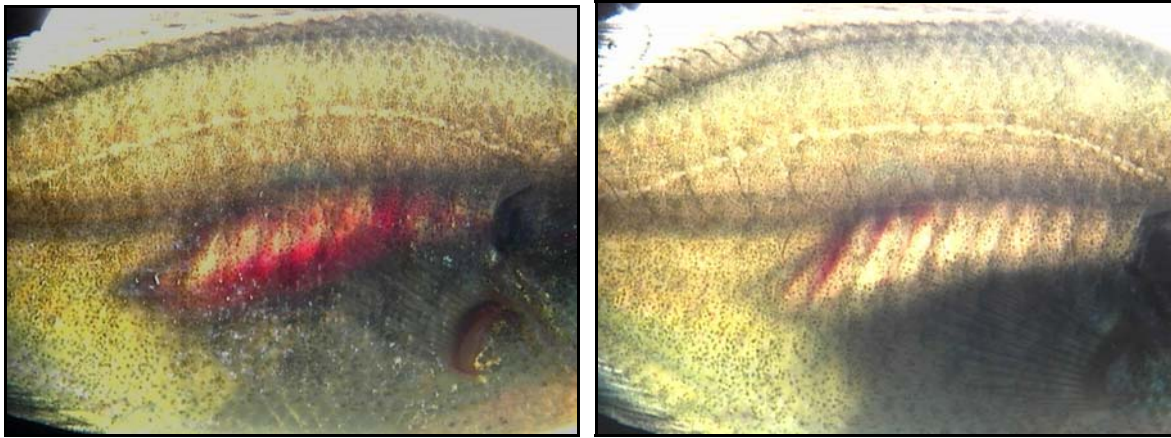
The only observed external signs of injury during initial examination of surviving fish were occasional minor hemorrhaging in the pericardial cavity (visible through the transparent membrane behind the gills) and a “rosy” discoloration (Figure 3.3) on the side of the fish (just posterior to the pectoral fin), which was believed to be evidence of internal hemorrhaging. Midway through the first set of replicates, it was discovered that small bluegills were semi-transparent, and by using backlighting (strong light from under the fish) during macroscopic examination, internal organs were sufficiently illuminated to “see” internal structures without making an incision. Using the backlighting technique, it was confirmed that the “rosy” coloration was due to internal hemorrhaging near the swim bladder, but the



**Figure 3.3.** External view of hemorrhaging observed in bluegills immediately after pressure spike (left photo) and after 48 hours (right photo).

swim bladder itself was not ruptured (Figure 3.4). Because the first set of replicates was not evaluated using the backlighting technique, only the last two replicates were used to evaluate and compare injury rates.

The frequency of “injuries” was high and variable for both spiked and non-spiked fish. Most injuries were limited to light internal hemorrhaging and there was no significant correlation between injury rate and either depth acclimation or TDG level. However, when the number of fish that died was added to the number injured, a significantly higher number of bluegills acclimated at 30 ft were not dead or injured (i.e., OK) compared to surface-acclimated fish ( $p$  value = 0.0289). Control (non-spiked) fish also had internal hemorrhaging, but the number and severity were usually lower than for the corresponding test (spiked) group (Table 3.2).



**Figure 3.4.** External view of internal hemorrhages in juvenile bluegill as viewed with backlighting.

## 4.0 Discussion

The time/pressure sequences used in this test series approximated the pressure regime experienced by fish passing through the modified Kaplan turbine pressure sequence with an estimated nadir of 50 kPa (0.5 atm) compared to  $\leq 10$  kPa (0.1 atm) estimated for peak operation of a Kaplan turbine. The hyperbaric chamber system was originally designed and built to generate pressure spikes approaching vapor pressure. Under ideal conditions, pressures as low as 3 kPa (0.03 atm) were possible. However, achieving an exact 50 kPa nadir was difficult because precision was affected by the sensitivity and response time of pressure sensors controlling piston movement in the hyperbaric chambers. Several tests were aborted and repeated due to mechanical malfunctions resulting in poor pressure spike results. In addition, small bubbles suspended in the water due to elevated TDG levels further affected the reproducibility and precision of the nadir during tests.

The TDG level causing death or injury for surface-acclimated fish varied between species. TDG levels used in the modified turbine passage simulations were the same as were used in previous tests. Although some surface-acclimated fall Chinook salmon died at 120% TDG in this test series (whereas none died in the prior Kaplan turbine series), the difference in overall response of fish to elevated TDG levels was not statistically significant.

After removing fish mortalities attributable to acute GBT, the mortality for fall Chinook salmon and bluegills subjected to either the typical or modified Kaplan turbine pressure scenarios was compared (Table 4.1). Mortality levels were very low for fall Chinook salmon in both scenarios. For bluegills, the percent mortality was consistently lower for depth-acclimated fish exposed to the modified pressure regime, although mortalities for surface-acclimated bluegills were variable.

**Table 4.1.** Percent mortality for fall Chinook salmon and bluegills subjected to typical or modified pressures during passage through a Kaplan turbine.

	TDG Level	Fall Chinook Salmon		Bluegills	
		Typical	Modified	Typical	Modified
		% dead/trial	% dead/trial	% dead/trial	% dead/trial
Surface	100	0, 0, 0	0, 0, 0	0, 0, 5	0, 10, 5
	120	0, 0, 0	0, 0, 0	0, 15, 5	0, 0, 0
	135	(all GBT dead)	(all GBT dead)	0, 0, 20	0, 0, 0
Depth	100	0, 0, 0	0, 0, 0	30, 40, 35	10, 0, 30
	120	10, 0, 5	0, 0, 0	30, 35, 45	0, 0, 5
	135	0, 0, 15	0, 0, 0	35, 30, 65	40, 10, 5



## 4.1 Fall Chinook Salmon

Death and injury rates attributable to acute GBT were similar to previous tests with juvenile fall Chinook salmon (Abernethy et al. 2001). At 135%, all fish died. At 120% TDG, 15% of fall Chinook salmon died during acclimation whereas none died in the previous Kaplan test series. Overall, the difference in susceptibility to GBT was not statistically significant.

No fall Chinook salmon died or suffered serious injuries when subjected to the modified Kaplan turbine pressure sequence, regardless of acclimation depth or TDG acclimation conditions. However, some salmon developed black head spots identical to those observed in rainbow trout in earlier tests simulating Kaplan turbine time/pressure scenarios. The incidence of head spots was significantly higher for spiked fish (10.6%) than for control fish (1.6%). Head spots observed on fall Chinook salmon were not as pronounced or persistent as they were on rainbow trout in earlier tests (Abernethy et al. 2001). Although head spots were visible when the fish were alive and submerged in water, the spots quickly faded when the fish were euthanized and examined under a dissecting scope. Efforts to photograph head spots on euthanized fish were generally unsuccessful.

## 4.2 Bluegills

Although the bluegills used in the modified Kaplan turbine pressure sequence were smaller than those used in previous Kaplan turbine passage simulations, death and injury rates attributable to acute GBT were similar in both test series (Abernethy et al. 2001). When acclimated at surface pressures at 135% TDG, 65% of the fish died, whereas 42% died in the previous Kaplan test series. No fish in either test series showed external signs of GBT when acclimated to surface pressures at 120% TDG. Overall, the difference in susceptibility to GBT was statistically insignificant.

The mortality rate for turbine-spiked bluegills was significantly lower ( $p$  value = 0.0328) for fish subjected to the modified Kaplan turbine pressure sequence (5.5%) than to the typical Kaplan turbine spike (21.7%). When broken down by acclimation depth, the mortality rate for depth-acclimated bluegills was significantly higher than for surface-acclimated bluegills for both turbine types. With the Kaplan turbine scenario, 5.0% of surface-acclimated bluegills died, compared to 38.3% of depth-acclimated bluegills, for a 1:7.7 ratio. For the modified Kaplan turbine pressure sequence, the mortality rates were reduced to 1.7% and 11.1%, respectively, for a 1:6.5 ratio.

Although the mortality rate was lower for the modified Kaplan turbine pressure sequence than for the typical Kaplan pressure sequence, the injury rate was significantly higher ( $p$  value = 0.0025). However, different criteria were used to evaluate injuries in the two series. In the original Kaplan turbine series, the most prevalent injury was a ruptured swim bladder, identified by viewing bubbles and hemorrhages in the pericardial cavity through a membrane behind the last gill arch. In the modified Kaplan turbine pressure series, fewer fish had ruptured swim bladders or major internal hemorrhaging. However, with the smaller, almost transparent bluegills used, it was possible to detect minor internal hemorrhaging, which was classified as an “injury”. The internal hemorrhage was sometimes visible externally as a “rosy” appearance on the side of the fish just posterior to the pectoral fin. Such minor internal hemorrhaging would not have been detected when examining larger fish used in the original Kaplan test series.

Internal hemorrhaging was frequently seen in “control” fish, including bluegills held at surface pressures and 100% TDG where pressure changes and effects of elevated gas were not factors. After completing such a test and finding 13 of 17 control fish with minor internal hemorrhaging, bluegills were gently netted directly from the main stock tank, anesthetized, and immediately examined under the dissecting scope in an attempt to determine the cause of the symptom. None of the 12 bluegills examined had visible internal hemorrhaging. Based on the examination, we concluded that handling (netting, loading, or unloading) may have affected both the frequency and severity of internal injuries in both control and test fish groups. The most probable source of injury was during the removal of fish from the hyperbaric chamber, when fish were flushed from the chamber into a plastic garbage can. Efforts to reduce the frequency of injuries associated with removal by partially filling the garbage can prior to flushing fish out of the chambers were unsuccessful.

The source of the hemorrhage appeared to be the gas gland on the posterior side of the swim bladder and/or “*rete mirabile*”, the network of blood vessels supplying the gas gland. The extent of hemorrhaging ranged from severe, where the entire peritoneal cavity was discolored, to very minor, where only a few small vessels were affected, as depicted in Figure 5.



## 5.0 Conclusions

Observations during the acclimation of fish to depth were the same as in prior tests. Over a 24-hr acclimation period, fall Chinook salmon were unable to reach equilibrium and attain neutral buoyancy to pressures simulating 30 ft of depth (salmon had no access to the surface while in the sealed test chambers). In contrast, bluegills became neutrally buoyant in about 2 hr through gas exchange from the blood.

The frequency, type, and severity of injuries varied between species. In addition, the number of fish injured or killed was lower for fish subjected to the modified Kaplan turbine pressure sequence as compared to the typical Kaplan turbine scenario. The differences are summarized below:

- There was no evidence of swim bladder rupture in fall Chinook salmon when subjected to the modified Kaplan turbine pressure sequence. A small number of salmon had ruptured swim bladders in the typical Kaplan series.
- No fall Chinook salmon developed instantaneous gas bubble formation, a cause of death or injury in a small number of fish in the typical Kaplan test series.
- Some fall Chinook salmon developed mild cranial lesions. The discolorations were not nearly as distinct as those observed in rainbow trout in the previous Kaplan series. Although cranial lesions were sometimes observed on fall Chinook salmon in the previous Kaplan series, they were very light, short-lasting, and dismissed as being insignificant.
- The effect of TDG on bluegills was nearly identical in both the modified Kaplan turbine pressure sequence and typical Kaplan test series, except that 2 depth-acclimated bluegills (both controls for the pressure spike) developed instantaneous bubble formation when returned to surface acclimation pressure, resulting in immediate death.
- Swim bladder rupture still occurred in depth-acclimated bluegills in the modified Kaplan turbine pressure sequence; however, the magnitude and frequency of occurrence were much lower than for bluegills subjected to the typical Kaplan pressure spike simulation. Observations with high-speed video showed that bluegills acclimated to 30 ft of depth and subjected to the modified Kaplan turbine pressure sequence did not experience a catastrophic rupture or “blow-out” as was documented with the typical Kaplan pressure spike.
- The use of smaller bluegills in the modified Kaplan test series made it easier to detect and document swim bladder rupture and internal hemorrhaging injuries.
- Fewer bluegills died as a result of the modified Kaplan pressure spike, although a high percentage of spiked fish still had injuries in the form of internal hemorrhaging. The occurrence of internal

hemorrhaging in surface-acclimated control fish suggests that handling contributed to the reported injury rate, and actual injuries attributable to the pressure spike may be much lower than reported.

The effects of dissolved gas supersaturation did not appear to have a significant synergistic effect with pressure under the modified Kaplan pressure simulation. The most severe injuries in bluegills (swim bladder rupture and internal hemorrhaging) were affected more by pressure change. Both rapid pressure change in test fish (turbine passage pressure spike) and gradual change in control fish (depth-acclimated fish returned to surface pressures) resulted in significant injury rates for bluegills. “Stretching” of the swim bladder beyond its “normal” volume or other trauma (handling) can easily rupture capillaries surrounding the swim bladder, resulting in internal hemorrhaging.

The results suggest that death/injury rates were slightly lower for salmonids and presumably other physostomous fish if the turbine pressure spike was reduced to 50 kPa (0.5 atm) instead of  $\leq 10$  kPa (0.1 atm), although statistically, the mortality/injury rates were virtually the same due to the small proportion of trout and salmon injured during the Kaplan turbine simulations. For bluegills and presumably other physoclistous fish, death and serious injury rates associated with the 50 kPa modified Kaplan turbine scenario were substantially lower than for the typical Kaplan pressure scenario; however swim bladder rupture still occurred under the modified pressure regime for some depth-acclimated fish. Swim bladder ruptures were much more frequent and violent with the typical Kaplan pressure simulation, resulting in “blow-outs” under some conditions.

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