University of Alberta

Blasting Effects on Incubating Salmonid Eggs

by

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Abstract

Blasting in or near water has the potential to affect fish negatively. The shaking of the substrate measured as peak particle velocity (PPV) has the potential to cause mortality of incubating eggs. Although a maximum allowable limit (13 mm/s) exists to protect eggs, no studies have related PPVs from blasting to egg mortality. To address this information gap, a field study and laboratory blast simulations were developed to measure egg mortality at different PPV exposures. Despite exposures over double the current guideline, no mortality was observed in lake trout (*Salvelinus namaycush*) eggs in the field. In the laboratory, increased mortality of rainbow trout (*Oncorhynchus mykiss*) eggs was found in exposures greater than 132 mm/s, during a very short period of egg development. Results indicate that the current PPV guideline provides ample protection for spawning beds and could be increased substantially, at least for *Oncorhynchus* species.

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Chapter 2. Effects of Explosives on Incubating Lake Trout Eggs in the Canadian Arctic

Chapter 3. Effects of Simulated Blasting on Mortality of Rainbow Trout Eggs

Chapter 1. General Introduction

Disturbances to fish and their habitat come from many sources, including both natural and anthropogenic. Anthropogenic disturbances to fish and fish habitat can be caused by any activity that leads to the harmful alteration, disruption, or destruction of fish habitat (DF0 1986). These activities include any activity that leads to fish mortality, or pollution, or physical alteration of fish habitat. Some activities have the potential to cause degradation in multiple ways. For example, explosives can cause chemical pollution from the blasting residues, as well as destruction, sedimentation, and shaking of nearby areas. Even the noise generated from continued blasting has the potential to deter fish from the area.

Blasting in or near fish-bearing waters originates from many sources, including geophysical exploration, open-pit mining, construction, and other industrial development. If this blasting is of great enough magnitude, the pressure and mechanical shock waves produced have the potential to negatively impact fish and their incubating eggs in the surrounding area. Past research has shown that the sudden change in pressure that results from an explosion, measured indirectly as overpressure, is responsible for most impacts on juvenile and adult fish, by rupturing the swimbladder or other soft organs (e.g. Wright 1982; Keevin et al. 1999). Conversely, the mechanical shaking of the substrate that follows an explosion, measured as peak particle velocity (PPV), is believed to be most damaging to incubating eggs (Wright 1982). Appropriately, guidelines exist in Canada for blasting in or near fish habitat, which contain a maximum allowable limit for both overpressure (100 kPa) and PPV (13 mm/s) to protect fish and their incubating eggs, respectively (Wright and Hopky 1998).

Many studies have focused on effects of submarine blasting on fish (summaries by Wright 1982; Keevin et al. 1999). This type of blasting has the potential to kill large amounts of fish; however, various methods can be used to minimize the impact, including limiting blast sizes, using bubble curtains to dissipate energy, using noise to scare fish out of the affected area, and removing fish during explosions (Keevin 1998; Wright and Hopky 1998). Although this type of blasting has been shown to be detrimental to juvenile and adult fish, only Kostyuchenko (1973) has investigated effects of submarine blasting on pelagic fish eggs. He found 50 g charges of TNT were capable of reducing survival up to 20 m away.

During out-of-water blasting, energy can travel long distances through the hard substrate and cause shaking of aquatic substrate great distances from the blast source (Figure 1-1). On the other hand, little overpressure is transferred through the substrate and into the lake water and is usually of minimal concern where setback distances are large. For example, if a confined 100 kg charge were detonated out-of-water in hard rock substrate, a setback distance of 50.3 m would be needed to remain below the overpressure limit; however, a distance of 150.9 m would be required to keep below the PPV limit (Wright and Hopky 1998). It is believed that this shaking of the lake substrate may negatively affect fish eggs and larvae that incubate in the substrate (Wright 1982).

The sensitivity of fish eggs to physical disturbance has been well documented, however, this work has primarily focused on determining sensitive stages to disturbances, instead of determining critical exposure levels. For most species, the highest sensitivity is during epiboly, until the mesoderm completely covers the yolk sac (e.g. Jensen and Alderdice 1983 and 1989; Fitzsimons 1994). Because of their focus on sensitive developmental stages, many of these studies use only qualitative means of generating PPVs, and thus provide limited quantification of exposure, e.g., swishing a bucket back and forth (Fitzsimons 1994), inverting a beaker (Johnson et al. 1983 and 1989), pouring eggs in water from different heights into a bowl (Jensen and Collins 2003) or vibrating an incubation tray (Hata 1927; Smirnov 1954 and 1955). Quantification of mechanical disturbance exposures have included measuring the height from which eggs are dropped into water (Battle 1944; Smirnov 1954), dropped onto mesh (Holmefjord and Bolla 1988; Gwo et al. 1995), dropped onto a soft plastic bumper (Dwyer et al. 1993), or are dropped while inside an enclosure (Jensen and Alderdice 1983, and 1989; Crisp 1990; Hilomen-Garcia 1998; Krise 2001).

It has been suggested that the dropping eggs inside an enclosure allows conversion of drop height to multiple units of exposure, such as energy (ergs) (Jensen and Alderdice 1989) or PPV (Jensen 2003), through the use of simple physical principles. Using this latter conversion, it has been estimated that an exposure of 140 mm/s would cause 10 % egg mortality of the most sensitive Pacific salmon. These conversions, however, may not be accurate in estimating PPVs produced by blasting, due to differences between exposure sources. Blasting events may last several seconds whereas the drop-height exposure is produced as a single large pulse upon impact. When dropped, the mass of the egg is also related to the energy imparted on that egg at the end of the fall, whereas the effect of PPVs from blasting, as measured in the spawning substrate, is not dependent on the egg size. Another shortfall of the drop-height approach is that eggs were exposed free of surrounding fluid, and because water is believed to provide protection to the eggs (Jensen and Alderdice 1983), previous estimates may be conservative in estimating PPV effects on eggs in a natural, aquatic environment.

Although the maximum allowable PPV limit for protecting spawning beds is precise, the only known previous attempts to relate mechanical effects of blasting to mortality of incubating eggs have failed to show any increases in mortality. In one study, Dolly Varden (*Salvelinus malma*) eggs, in a sensitive developmental stage, were placed 1,200 m from the source of the Project Long Shot 80-kiloton (72.6×10^6 kg) nuclear detonation on Amchitka, Alaska; although the eggs received a peak vertical acceleration of 30.4 m/s^2, there was no increase in mortality (Seymour and Nakatani 1967; cited in Post et al. 1974). In a second study, associated with the proposed Project Wagon Wheel natural gas stimulation authorization, effects of peak vertical accelerations on rainbow trout eggs were investigated in the laboratory, because a highly valued trout stream was located near the proposed site (Post et al. 1974). For this study, a recording taken during the similar 40-kiloton Rulison gas stimulation nuclear detonation was used to simulate exposures on rainbow trout (*O. mykiss*) eggs. Exposures were carried out at four stages of egg development, including a sensitive stage. Eggs were subjected to five repeated exposures of approximately 10, 20, 49 and 98 m/s^2 by repeatedly dropping artificial redds containing the eggs. Despite the large vertical accelerations, no increase in mortality was detected (Post et al. 1974). Although vertical acceleration is related to PPV, PPV was not reported.

If the equations contained in the Canadian blasting guidelines (Wright and Hopky 1998) can be used to estimate PPVs from these large explosions, Project Long Shot could have produced PPVs of more than 23,000 mm/s at 1,200 m from the source, and exceeded the 13 mm/s guideline for 130 km from the source. The 40-kiloton Rulison gas stimulation detonation produced an acceleration of 9.8 m/s^2 at 6.25 km from the source. This event could have exceeded guidelines up to 90.9 km from the source; at 6.25 km, it could have produced a PPV of 943 mm/s.

The PPV guideline for protecting spawning beds from blasting was thus developed using very limited information. Even though recent authors have suggested that the current guideline provides ample protection to salmonid species (Jensen 2003), the actual PPV levels from blasting that will increase mortality of fish eggs remains unknown. To address this information gap, I undertook both field and laboratory experiments. A study was conducted *in situ* at Diavik Diamond Mine Inc.'s open-pit mine at Lac de Gras, where lake trout eggs were placed in the lake substrate and both mortality and PPV exposure were recorded. To control for natural variations in survival among field sites, effects of simulated blasting events were also examined in the laboratory.

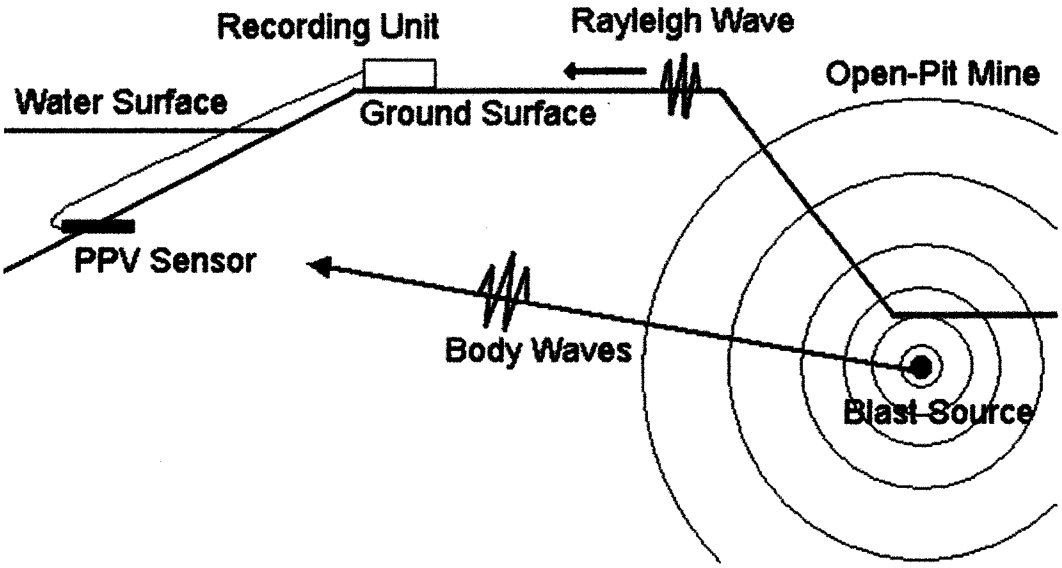

Figure 1-1. Illustration of an out-of-water blasting event in an open-pit mining operation near water. Seismic waves (Rayleigh and body waves) emanate outward from the blasting source, but attenuate in the substrate. Transfer of overpressure into the lake is minimal if setback distances are considerable. The current maximum allowable limit for shaking, measured in the aquatic substrate, is 13 mm/s.

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Chapter 2. Effects of Explosives on Incubating Lake Trout Eggs in the Canadian Arctic

Introduction

Use of explosives in or near fish-bearing waters occurs with many activities, including geophysical exploration, construction, industrial development, and mining. Explosions produce seismic waves, which are manifest as small motions of the substrate, or as pressure within the water column, that radiate outward from the blast location. In engineering practice, the complex waveforms are usually quantified by characteristic measures such as peak particle velocities (PPV) and overpressures. If the blasts are strong enough, the resulting pressure change and mechanical shock waves can negatively impact fish in the surrounding area. Past research has shown that the sudden pressure deficit, measured indirectly as overpressure, is responsible for most impacts on juvenile and adult fish, by rupturing the swimbladder and other soft organs (Wright 1982). In contrast, the mechanical shaking of the substrate, measured as PPV, is believed to be most damaging to incubating eggs (Wright 1982). Appropriately, Canadian Department of Fisheries and Oceans (DFO) developed guidelines for blasting that contain maximum allowable limits for both overpressure (100 kPa) and PPV (13 mm/s) in or near fish habitat (Wright and Hopky 1998).

If explosions generate PPVs that damage or kill incubating eggs, the habitat quality (sensu Marsden et al. 1995a) and productive capacity (sensu DFO 1986) of spawning reefs exposed to explosions may be compromised for species, such as lake trout (*Salvelinus namaycush*), that broadcast their eggs onto those reefs. This, in turn, would require habitat compensation activities, to offset reductions in productive capacity (DFO 1986). Physical shock, of which PPV is one type, has been demonstrated to increase mortality of lake trout eggs in laboratory experiments and was highest when the shock occurred during the final stages of epiboly (Fitzsimons 1994). This is comparable to what has been observed for other salmonid species (Smirnov 1954, 1955; Jensen and Alderdice 1983, 1989). Similarly, Eshenroder et al. (1995) suggested that physical shock during a period that roughly corresponded with this development stage was largely responsible for differences in survival of eggs in buried versus exposed incubators on a Lake Superior spawning reef.

Although these largely qualitative studies of physical shock raise concerns about comparable effects of PPVs on incubating eggs, no research has examined the relationship between PPV and egg mortality and there is, in fact, uncertainty with respect to the 13 mm/s PPV guideline (e.g., Jensen 2003). There is also considerable uncertainty regarding the cumulative effects of repeated blasts on over-wintering, developing eggs of fall-spawning fishes. Because use of explosives in or near waters will likely increase in the future, research is needed to assess the relationship between blast intensity (PPV) and effects on developing fish embryos, to assess the appropriateness of current blasting guidelines.

The objectives of my study were to (1) determine if mortality of lake trout eggs increased where PPVs exceeded the maximum allowable limit, relative to a reference site, and (2) assess if increased egg mortality is primarily due to limited exposure during early development or to repeated exposures throughout the incubation period.

Study Site

Lac de Gras is a large (surface area = 572 km^2), ultra-oligotrophic (total phosphorus < 3 µg/L; DDMI 1998a), low-arctic lake located on the Barrenlands of the Northwest Territories (Canada), approximately 300 km northeast of Yellowknife. My study area extended outwards from the east side of East Island, approximately 64°31' N, 110°20' W (Figure 2-1), where Diavik Diamond Mines Inc. (DDMI) is conducting open-pit mining of kimberlite pipes in their A154 pit. Explosives are used as the primary method of excavating rock. While overpressure was not expected to cause concern (DDMI 1998a), it was predicted that a blast zone, where PPVs exceeded DFO's maximum allowable limit, could extend up to 717 m beyond the pit's dike into Lac de Gras (DDMI 1998a).

Lac de Gras supports a stable, slow-growing community of at least eight coldwater fishes (DDMI 1997a), several of which are fall spawners, e.g., round whitefish, cisco, and lake trout. Lake trout are dominant in Lac de Gras, both numerically and in biomass (DDMI 1997a). Due to their position in the food chain, importance to traditional and sports fishing, and sensitivity to environmental change or disturbance, lake trout was identified as a "sentinel species" for the lake (DDMI 1998b) and was the focus of my research. Eight potential lake trout spawning shoals were identified within the predicted blast zone of the A154 pit (DDMI 1997b). Eggs deposited on these shoals (late August – late September) incubate over winter and hatch approximately 8-10 months later (May to July). Over this long incubation period, they are exposed to both ambient environmental conditions and PPVs from blasts in the A154 pit.

Four sites with suitable spawning habitat were chosen for this study (Figure 2-1). Spawning habitat suitability was based on water depth (2.5 - 7 m, but next to deeper water), substrate size (4 - 30 cm, but up to 1 m) and shape (angular), interstitial space (0.2 - 1 m deep), and a location away from depositional effects (Scott and Crossman 1973; Gunn 1995; Marsden et al. 1995b; DDMI 1997b). Of the four sites, three were located within the predicted blast zone. Two of the three, East Dike and South Dike, were located close to the dike wall; the South Dike site was, in fact, composed entirely of crushed rock from the outer dike wall. In contrast, East Dike, although directly connected to the dike wall, was composed entirely of natural spawning substrate. The third blast-zone site (Tern Island) was approximately 220 m east of the dike wall. A reference site (Reference Island) was located approximately 2,200 m east of the dike wall, well outside of the predicted blast zone (Figure 2-1). Both Tern Island and Reference Island were also composed of natural spawning substrate. All sites were at depths of 3-5 m and were close to land to enable deployment of geophysical blast monitoring equipment.

Methods

Experimental Procedure

Lake trout were obtained using small mesh gill-nets (19-38 mm bar mesh) between 2300 and 0130 hours on September 16 - 17, 2003. Eggs and seminal fluid were removed from three females and four males, respectively, with the eggs from individual females mixed with the sperm from at least two males. Fertilized eggs were allowed to water-harden in lake water over night (8 h), disinfected with a weak iodine solution (1:600 by volume), and rinsed in fresh lake water, before being mixed into a bulk sample.

Lake trout eggs from this bulk sample were then loaded into Plexiglas "sandwich" incubators. These incubators were developed for investigations of survival and development of lake trout eggs (Kennedy 1980; Gunn and Keller 1984; Manny et al.

1989). Each incubator consisted of a 12.5 cm x 25 cm x 1 cm Plexiglas piece containing 50 separate cells, each holding a single trout egg. These cells were enclosed on both sides with 2-mm Nitex mesh, which was held in place and protected by thinner pieces of Plexiglas (see Figure 1 in Manny et al. 1989). The incubators allow eggs and fry to be exposed to ambient conditions (including PPVs) in the shallow interstitial waters, but protect them from other sources of mortality, such as predation, thereby allowing differences in survival rates among sites to be related to differences in local ambient conditions. Incubators were wedged into substrate interstices and held in place by side chains, spaced approximately 1 m apart, that were attached to a main chain anchored to the bottom (see Figure 3 in Manny et al. 1989).

After the incubators were loaded with lake trout eggs, 20 incubators were randomly assigned to each of the four sites. Incubators were inserted by divers into the substrate at the appropriate sites, following the procedure of Manny et al. (1989). Deployment at all sites was completed on the same day, starting with Reference Island, followed by Tern Island, South Dike, and finally East Dike. Each site received two chains of 10 incubators each, located in close proximity and in similar suitable habitat conditions (described above).

Given that the period in which lake trout embryos are most sensitive to physical shock is early in development (Fitzsimons 1994), half of the incubators on each chain (n = 5) were retrieved after 20 days of exposure (September 17 - October 7) to assess mortality during this early period (early retrieval). The remaining incubators were left in the lake until ice-off the following summer (July 16-18; final retrieval). During retrievals, sedimentation at the sites was noted qualitatively, as was whether each incubator had remained in the substrate or been dislodged. When incubators were brought to the surface, they were immediately placed in containers of lake water.

Survival Data Analysis

During early retrieval, the contents of two of the five incubators per chain were preserved in Davidson's solution for histological analysis at DFO's Freshwater Institute in Winnipeg. Preserved eggs were examined at 40× magnification and the presence and stage of development of the preserved embryos (Balon 1980) were determined for each egg. Any deformities of the embryos were also noted. Survival was calculated per incubator as the number of eggs alive when preserved divided by the initial number of eggs (50). The other early retrieval incubators (n = 3 per chain) were transported by air to the University of Alberta's BioSciences Aquatics Facility. There, eggs were kept in the incubators and held in a recirculation chiller unit at 4°C until after hatch. To assess survival of early retrieval lake trout eggs incubated at the university, the incubators were monitored over the remainder of the incubation period (October 8 until the following spring) at which time the fates of all eggs were classified as dead, deformed or hatched. Hatched fry were monitored in the incubator cells for an additional 45 days after hatch, for better comparison to incubators remaining in the lake until final retrieval was possible. Survival of lab-incubated eggs was calculated per incubator as the number of hatched but not deformed embryos divided by the initial number of eggs (50).

At final retrieval, lake trout embryos still in the lake had already hatched, so contents of incubator cells were classified as dead, alive or empty. Empty cells were subsequently classified as either dead (eaten or decomposed) or alive (escaped) to allow comparison with early retrieval results. Two approaches were used for this classification. (a) 100% escape correction. With my incubators maintained in the laboratory, fry were frequently observed escaping from the incubator cells as the contents of yolk sacs were used up. Because I know fry were capable of escaping, I assumed in this approach that all empty cells at final retrieval resulted from escapement of successfully hatched embryos; therefore, all empty cells were classified as alive. (b) 50% escape correction. Casselman (unpublished data cited in Casselman 1995) noted that half (50.3%) of the empty cells in his study had contained embryos that had either hatched prematurely, died and had decomposed or had been consumed by invertebrate scavengers; in both cases, only mucoid remains of highly decomposed fry remained in some of the empty cells. There was no explanation for the other half (49.7%) of the empty cells. Therefore, I also used a correction factor assuming that only half of the empty cells were the result of escapees. Survival at final retrieval was then calculated for each incubator as the number of live embryos assuming either that 50% or 100% of the empty cells had contained a live embryo that escaped, all divided by the initial number of eggs (50).

Survival data was arcsine square root transformed prior to analysis, to meet test assumptions (Zar 1999). This transformation is used on percentage or proportion data to meet the assumption of normality (Zar 1999). Survival between chains within sites and among sites was analyzed using a nested analysis of variance (ANOVA) design. ANOVA is an analysis for making simultaneous comparisons between two or more means to determine if they are from different populations. When a significant difference in survival among sites was found, least-significant-difference (LSD) multiple comparison was used to identify sites with survival different from the reference site. Results from the two retrieval periods were analyzed separately. Differences in survival between the early and final retrieval groups at each site were evaluated by a Student's *t*-test (Bonferroni corrected $\alpha = 0.0125$). The Student's *t*-test is a method used to assess differences between two means to determine if they are statistically different. All statistical tests were performed using SYSTAT version 10 (SPSS 2000). For all statistical tests, a significance level α of 0.05 was used unless otherwise noted.

Blast Data

Blasts were monitored and recorded at all four sites using an *OYO Sea Array 4*™ 4-component underwater geophone sensor attached to an Instantel Mini Blast Mate™ monitor. The sensor, attached to one of the chains at each site, was wedged between large boulders, in a manner similar to the incubators, to ensure coupling to the lake substrate. The sensor was then attached to a monitor placed on shore. Due to short battery life and long time between blasts, monitors needed to be retrieved between blasting events so that data could be downloaded and batteries could be recharged. As many blasts as possible were measured over the incubation period; however, due to poor weather conditions and equipment malfunction, some blasts were missed. These missed events were greatest at sites located away from the dike wall (Tern Island and Reference Island) due to difficulty in travel under arctic field conditions, especially at times of extreme cold and at lake freeze-up and thaw. To estimate the exposures of blast events not recorded, a PPV verses scaled distance plot was created for each site using blasting records over the entire incubation period, using the methods of Dowding (1985).

Scaled distance plots are commonly used for prediction of blast strength. They are based on an empirical relationship between blast size and location and measured PPV at a site. The measured PPV is plotted against the scaled distance, which is a normalized value that takes into account both the blast weight per delay and the distance from the blast. With the scaled distance relation in place, a PPV prediction can be made based on blast weight and distance from the blast. Upper and lower limits that contain 95% of the data were also calculated according to Dowding (1985). The upper limit is generally used to predict safe distances from a blast.

Additional details of how the PPV and overpressures are determined will be provided in a later contribution. Briefly, however, the geophone sensor includes three orthogonal geophones and one hydrophone. Different seismic detectors are sensitive to different aspects of the ground motion, such as particle displacement, particle velocity, or particle acceleration. The geophones are sensitive to the particle velocity component, which is the actual speed at which the ground is moving in response to the passing of a seismic wave. The particle velocity must not be confused with the speed at which the seismic wave itself propagates. The PPV is derived from the appropriately corrected motions of the three geophones and is the largest peak to peak value for a given event. Similarly, the overpressure is the positive or negative pressure change relative to ambient caused by motion of the seismic wave in the water. Neither of these values gives any indication as to the length and magnitude of the total exposure, but are frequently used (in this case, for blasting guidelines) for logistical reasons.

From the measured and estimated PPV exposures, I calculated two different measures of blast exposure. *Mean PPV* for each site was the average of all measured and estimated blast events over the appropriate exposure period (early and final retrieval exposures). *Max PPV* was the highest measured or estimated exposure at a site during an exposure period. The number of exposures measured or estimated (within the 95 % confidence interval) to exceed the 13 mm/s guideline was also reported for each site. Overpressure data were not recorded for the early exposure period, but because overpressure values were so low, only maximum measured values are presented for the final retrieval exposure period.

Results

General Observations

Lake trout appeared to show no avoidance of the blasting area. Both juvenile and adult lake trout were observed within the blast zone, even shortly after blasting events. Lake trout in spawning condition were, in fact, caught within 100 m of the dike, suggesting that they spawned within the blast zone. During both retrievals, no significant sedimentation was observed at any site, and no incubators became dislodged.

Early Retrieval

All developing lake trout embryos preserved at early retrieval had completed epiboly and were into Balon's (1980) $E^2 5$ developmental stage, which corresponded to the beginning of organogenesis, where the tail bud is free from the yolk. Survival was similar between incubators preserved immediately at early retrieval and the incubators subsequently held at the University of Alberta until after hatch for all chains within sites (t-tests, $P > 0.05$; Table 2-1). After combining both sets of incubators, survival did not differ between chains within sites ($F = 0.055$; df = 4,32; $P = 0.994$) but did differ among sites ($F = 38.715$; df = 3,4; $P = 0.002$). Survival at Tern Island was slightly, but not significantly, lower than at the reference site (LSD; $P = 0.156$). In contrast, survival at South Dike and East Dike was higher than at the reference site (LSD, $P < 0.02$; Figure 2-2).

Final Retrieval

The percentage of cells classified as dead was very similar within and among sites (50.0 - 55.6%) except for South Dike, which had a higher amount of dead eggs (60.0 – 69.2%; Table 2-2). In contrast, variation was large among cells classified as alive (0.8 - 33.3%) and empty (15.6 - 47.2%; Table 2-2) and the two categories were highly negatively correlated ($r = -0.75$, $n = 40$, $P < 0.001$), whereas the values of "alive" and "dead" cells ($r = -0.36$, $n = 40$, $P = 0.074$), and of "empty" and "dead" cells ($r = -0.35$, $n = 40$, $P = 0.077$) were each only weakly correlated. Furthermore, a sample of empty cells ($n = 100$) examined at 40× magnification showed no remnants of embryos.

50% Escape Correction. – Using the 50% escape correction at final retrieval, survival did not differ between chains within sites (nested ANOVA; $F = 1.75$; df = 4,32; $P = 0.164$) or among sites ($F = 4.57$; df = 3,4; $P = 0.088$; Figure 2-3a). Survival was lower at final verses early retrieval for all sites. Decreases in survival relative to early retrieval ranged from 13% at East Dike ($t = 3.60$, df = 18, $P = 0.002$) to 30% at South Dike ($t = 7.41$, df = 18, $P < 0.001$; Figure 2-4a).

100% Escape Correction. – With the 100% escape correction for empty cells at final retrieval, survival between chains within sites was similar ($F = 0.59$; df = 4,32; $P = 0.672$), but survival differed among sites ($F = 7.12$; df = 3,4; $P = 0.044$; Figure 2-3b). Survival was lower (by 10%) than the reference site only at South Dike (LSD; $P = 0.042$). Likewise, survival decreased between early and final retrieval only at South Dike (18%; $t = 5.413$, df = 18, $P < 0.001$; Figure 2-4b).

Blast Exposures

During the early exposure period, six blasting events occurred. No measured or estimated blasts exceeded the Canadian guideline of 13 mm/s, although only one or two blasts per site were measured directly. The 95% CIs of the estimated *Max PPVs*, however, were large and marginally included 13 mm/s at all blast-zone sites (Table 2-3). Estimated *Mean PPV* for the three blast-zone sites did not differ ($F = 0.031$; df = 2, 15; $P = 0.970$). The highest recorded PPV at the reference site was only 0.58 mm/s.

During the entire incubation period there were 96 blasting events; measured PPVs exceeded guidelines at all three exposed sites. *Max PPV* was highest at East Dike, more than double the maximum allowable PPV limit (Table 2-4). Consistent with the early retrieval period, however, the three exposed sites did not differ in *Mean PPV* over the entire exposure period ($F = 0.785$; df = 2, 270; $P = 0.457$; Table 2-4). Exposures at the reference site were similar to those during the early retrieval period, with a maximum of only 0.7 mm/s. In total, blasts may have exceeded guidelines more than 20 times at exposed sites (Table 2-4). The maximum observed overpressure (0.011 kPA), which was measured at South Dike, was well below the Canadian guidelines of 100 kPA.

Discussion

During the early retrieval exposure period, recorded and estimated PPVs at all three blast-zone sites were very similar, although measurements were obtained for only 25% of the blasts. The highest recorded PPV, 4.4 mm/s at Tern Island was still well below the guidelines. All lake trout eggs preserved at early retrieval either showed no signs of development, indicating they had not been viable, or contained embryos at Balon's (1980) $E^2 5$ development stage, corresponding to early organogenesis. This suggests that there was no additional mortality after incubators were placed into the lake and that the embryos were past the developmental stage most sensitive to physical shock (Fitzsimons 1994) at the time of retrieval. Early retrieval survival at South and East dikes was actually higher than at the reference site, and survival at Tern Island was similar to the reference site, indicating clearly that there were no negative effects of this low-level blast exposure during the early stages of egg development. I suggest that factors other than blasting (e.g., natural variation among sites or handling effects) caused the observed differences in survival. Indeed, during incubator installation, one cooler held incubators destined for South and East dikes (having higher early survival) and another cooler was used for Tern Island and Reference Island (with the lower survival). Even though attempts were made to ensure that each cooler and all incubators were treated equally, I cannot rule out this as a possible source of the small variation. Regardless, there were no measurable negative effects of blasting during the early exposure period, when the eggs were most sensitive to physical shock.

Exposures at all three blast-zone sites over the entire incubation period were similar. All had exposures greater than the maximum allowable limit, with the highest recorded PPV, 28.5 mm/s, at East Dike. Final retrieval results were not as straightforward, however, due to the presence of empty cells. Previous studies using similar incubators with the same mesh size have also noted empty cells, and they were explained, albeit only in part, by lake trout eggs or embryos that had been eaten or had disintegrated (Casselman 1995; Eshenroder et al. 1995; Manny et al. 1995). In my laboratory-reared eggs, however, I observed that the screen mesh used to cover the incubator cells was large enough to allow young fish to escape when their yolk sacs were almost completely absorbed. Because final retrieval was not possible under my arctic field conditions until mid July (verses March-June in other studies), when yolk sac absorption was nearly complete, I suggest that for my study, empty cells were largely or entirely the result of escapees, since (a) the incubation period was sufficiently long to allow the lake trout larvae to absorb their yolk sacs and increase their chance of escape, (b) I observed post-absorption escapes in the lab, (c) there was no physical evidence, in the form of residual organic matter, that would be expected from predation or disintegration (Casselman 1995), (d) the number of live fish and empty cells at final retrieval were strongly negatively correlated, and (e) survival between the early and final retrievals did not generally differ, assuming the 100% escape correction. Eshenroder et al. (1995) also reported that the number of empty cells (up to 88.8%) increased with duration of incubation, suggesting that the presence of empty cells may increase with reduction in the size of the yolk sac. As well, Claramunt et al. (2005) showed that there was minimal loss of eggs after the initial two weeks of exposure in a natural setting, even while the eggs are exposed to outside predation. Nevertheless, because I could not rule out the possibility that some embryos were eaten or had completely disintegrated, I analyzed the final retrieval data using both 50% escape and 100% escape possibilities.

The 50% escape correction suggested that mortality increased significantly between early and final retrieval at all sites, even though the most sensitive stage of development had passed. On the other hand, this correction provided no evidence of blast effects on over-winter survival of lake trout eggs, as survival did not differ among all four sites at final retrieval. Thus, the apparent increase in mortality between early and final retrievals would have only resulted from natural mortality, something that was not observed in the early retrieval incubators that were subsequently held in the laboratory. The lack of organic residue or of predators in the cells also does not support this possibility.

As discussed above, I believe the 100% escape correction provides the best estimate of survival at final retrieval. Under this scenario, South Dike was the only site where survival was lower (by 10%) than at the reference site and where survival decreased between early and final retrievals (by 18%). Because neither mean nor maximum exposure to PPVs were higher at South Dike than the other blast-zone sites, I suggest that survival during this period was related more to the difference in spawning substrate characteristics than to blasting. Indeed, only at South Dike was the substrate not composed of natural spawning substrate and was composed of large jagged pieces of rock that had recently been crushed. Furthermore, the largest PPV was recorded at East Dike, which also had the highest survival.

The highest PPV exposure recorded at the reference site over the entire incubation period was 0.70 mm/s, which is only 0.19 mm/s above the detection limit and far below the Canadian guideline limit of 13 mm/s. An exposure of this level should not have any negative effects on incubation, as it is likely comparable to background vibrations, e.g., from wave action. The trigger levels of the Blast Mate recorders at the exposed sites were, in fact, set at more than double the 0.51 mm/s detection limit to reduce such false triggers. As such, the reference site provided a good estimate of lake trout egg survival at a site free from blasting effects. Survival at the reference site (early retrieval 48.3%, final retrieval 50% escape and 100% escape 25.8% and 45.4% respectively) was also comparable to studies done using similar incubators (1-49%; Casselman 1995, Eshenroder et al. 1995 and Manny et al. 1995).

Difference in mean PPV exposure among all exposed sites was small (0.6 mm/s) and not significant over the entire incubation period. For this estimate of exposure, I used every blasting event regardless of PPV size, as all sites were exposed to all blasting events. There are, however, numerous alternative ways in which an index of overall PPV exposure could be calculated, but without understanding how, or at what level, a single blast exposure causes mortality in fish eggs, it is difficult to develop a meaningful measurement of cumulative exposure.

The largest blast exposure, 28.5 mm/s at East Dike, was more than twice the maximum allowable level in the Canadian blasting guidelines (Wright and Hopky 1998). However, this blast occurred when the lake trout eggs were past the developmental stage most sensitive to mechanical disturbance (Fitzsimons 1994). Because East Dike also showed the highest survival over the incubation period, it appears that an exposure of 28.5 mm/s does not increase mortality of eggs at that stage of development. I cannot rule out that this exposure can increase mortality when eggs are at an earlier, more sensitive stage.

Exposure levels at each site depend not only on the amount of explosives used in any given blast, but also on the location of the blast within the pit, the size of each blast, and possibly, on environmental conditions (e.g., presence of ice), all of which vary among blasting events. Because of these differences, there can be no simple relation between exposure and distance from the pit. Together with changes in sensitivity of the lake trout eggs over time, and natural variation in survival of eggs among sites, this variation in exposure means that determining a specific distance or blast level that increases mortality is very difficult under operational field conditions, even if some exposures are large enough to cause significant mortality, in some locations at some times.

Although the level of blasting conducted at DDMI's A154 pit appeared not to result in reduced survival of lake trout eggs during my study, the level at which PPVs from blasting will increase mortality of eggs remains unknown. A recent study suggested that the maximum allowable PPV level contained in the Canadian blasting guidelines may be an order of magnitude lower than that required to cause 10% mortality in Pacific salmon (*Oncorhynchus spp.*) eggs (Jensen 2003). Additionally, Post et al. (1974) found that peak vertical accelerations up to ten times the force of gravity (g), repeated five times, did not increase mortality of rainbow trout (*O.* mykiss) eggs regardless of developmental stage tested (37.5, 75, 125, and 250 degree-days post fertilization). Given that the largest peak vertical acceleration recorded at DDMI was equivalent to a gravitational force of only 0.212 g, this suggests that Post et al.'s PPVs were much greater than 13 mm/s. Unknown, however, is how the sensitivity to PPV varies among different species (but see Jensen 2003).

A laboratory study that could control the size, number, and timing of exposure relative to fish egg development is needed to more accurately determine the PPV levels that increase mortality of incubating eggs. Such information may help predict the effects of blasts on fish egg mortality in the field, within the constraints described above. Development of a reliable and standardized blast-simulation procedure in the laboratory would also facilitate the testing of additional species of interest that should result in improved guidelines for protecting developing fish embryos from blasting.

Table 2-1. Percentage survival (mean +/- SE) of lake trout eggs per incubator (out of 50) for the laboratory raised ($n = 3$ per chain) and preserved incubators ($n = 2$ per chain) after 20 days of exposure in the lake substrate (early retrieval). Survival did not differ between preserved and lab raised eggs for any chain (t-tests; $P > 0.05$).

Site	Chain	Survival (lab raised)	Survival (preserved)
South Dike	1	55.3 +/- 2.7	53.0 +/- 5.0
	2	56.0 +/- 5.3	48.8 +/- 8.0
East Dike	1	52.7 +/- 6.7	52.0 +/- 2.0
	2	54.7 +/- 7.4	50.0 +/- 2.0
Tern Island	1	43.3 +/- 4.7	46.0 +/- 6.0
	2	46.0 +/- 4.2	47.0 +/- 3.0
Reference Island	1	45.3 +/- 2.7	50.0 +/- 2.0
	2	42.0 +/- 10.3	56.0 +/- 2.0

Table 2-2. Percentage of incubator cells classified as containing a dead embryo, being empty or containing a live fry (mean +/- SE), for incubators retrieved from each chain (*n* = 5) after ice out (final retrieval). Each incubator was originally seeded with 50 lake trout eggs.

Site	Chain	Dead	Empty	Alive
South Dike	1	60.0 +/- 3.8	22.0 +/- 4.6	18.0 +/- 7.0
	2	69.2 +/- 1.0	24.0 +/- 5.6	6.8 +/- 5.8
East Dike	1	50.0 +/- 2.7	22.4 +/-3.6	27.6 +/- 5.0
	2	51.2 +/- 4.1	15.6 +/- 5.2	33.3 +/- 3.8
Tern Island	1	53.6 +/- 7.0	30.0 +/- 3.8	16.4 +/- 3.8
	2	52.0 +/- 4.1	47.2 +/- 3.4	0.8 +/- 0.8
Reference Island	1	53.6 +/- 3.2	36.0 +/- 7.6	10.4 +/- 5.4
	2	55.6 +/- 4.5	42.4 +/- 4.4	2.0 +/- 1.6

Table 2-3. Measured and estimated peak particle velocities (PPVs; in mm/s) at the reference and three blast-zone sites in Lac de Gras during the early retrieval exposure period. *N* is the number of blasting events during this period, and *n* is the number of blasting events recorded. Below detection indicates the number of blasting events that were below the trigger levels of the recorders at each site (1.5 mm/s at the exposed sites, and 0.51 mm/s at Reference Island). The remaining events were missed due to poor weather or equipment malfunction. Exposures during these missed events were estimated according to Dowding (1985). Blasts > 13 mm/s refers to all blasts measured or estimated within 95 % CI to exceed the 13 mm/s guideline.

Site	*N*	*n*	Below detection	PPV Mean (SE)	Measured max. PPV	Estimated max. PPV (95% CI)	Blasts > 13 mm/s
South Dike	6	2	1	3.8 (0.5)	3.7	5.7 (2.3-14.3)	1
East Dike	6	1	2	4.0 (0.5)	3.1	5.6 (2.2-16.2)	3
Tern Island	6	2	1	4.0 (0.7)	4.4	6.1 (1.5-13.5)	1
Reference Island	6	1	2	0.57 (0.0)	0.58	0.6 (0.08-4.59)	0

Table 2-4. Measured and estimated peak particle velocities (PPVs; in mm/s) for the entire incubation period at the reference site and three blast-zone sites in Lac de Gras. *N* is the number of blasting events during this period, and *n* is the number of blasting events recorded. Below detection indicates the number of blasting events that were below the trigger levels of the recorders at each site (1.5 mm/s at the exposed sites, and 0.51 mm/s at Reference Island). The remaining events were missed due to poor weather or equipment malfunction. Exposures during these missed events were estimated according to Dowding (1985). Estimated Max. PPVs (and 95% CI) only reported for sites where the values estimated for missed events and their 95% CI exceeded the largest measured event. Blasts > 13 mm/s refers to all blasts measured or estimated within 95 % CI to exceed the 13 mm/s guideline.

Site	*N*	*n*	Below detection	PPV Mean (SE)	Measured max. PPV	Estimated max. PPV (95% CI)	Blasts > 13 mm/s
South Dike	91	64	4	6.4 (0.4)	24.9	≤ 24.9	24
East Dike	91	61	8	5.9 (0.4)	28.5	≤ 28.5	23
Tern Island	91	25	3	5.8 (0.3)	17.4	9.9 (2.4-21.8)	33
Reference Island	91	5	23	0.59 (0.0)	0.70	0.66 (0.09-5.00)	0

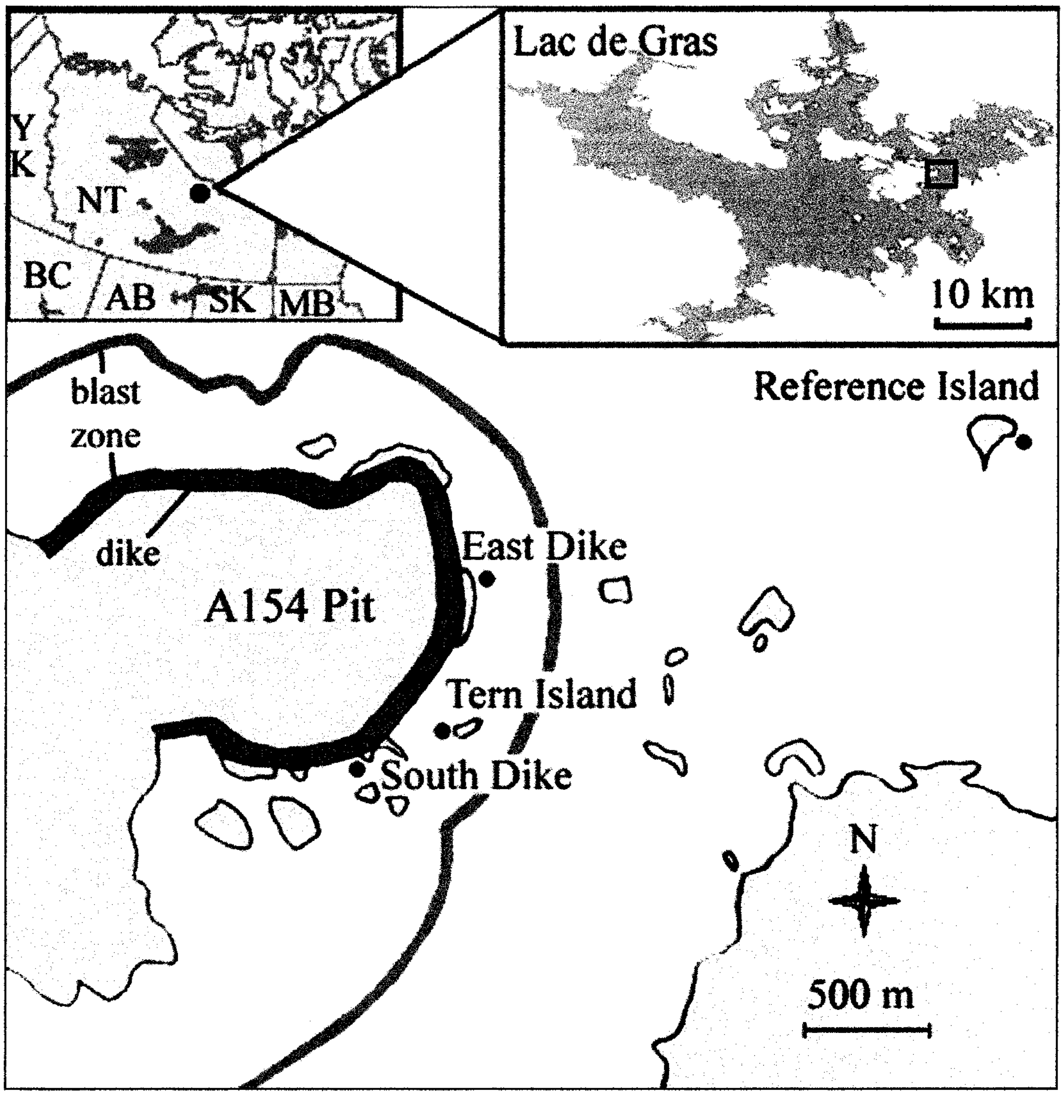

Figure 2-1. Map of Las de Gras, Northwest Territories (NT), and of the four sites chosen for deployment of incubators and blast monitoring equipment. The light grey line around the dike indicates the predicted blast zone, where maximum Peak Particle Velocity (PPV) was expected to exceed 13mm/s for the A154 pit and a second, proposed pit, to be located to the south of A154.

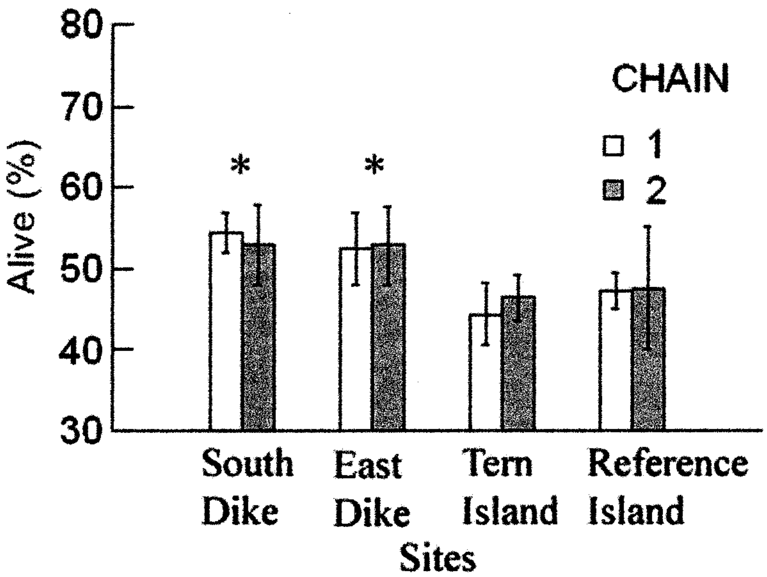

Figure 2-2. Percentage of live lake trout eggs (mean +/- SE) recovered from five incubators per chain at early retrieval (laboratory raised and preserved combined). Sites where survival differed from Reference Island (control site; LSD, $P < 0.05$) are marked with asterisks.

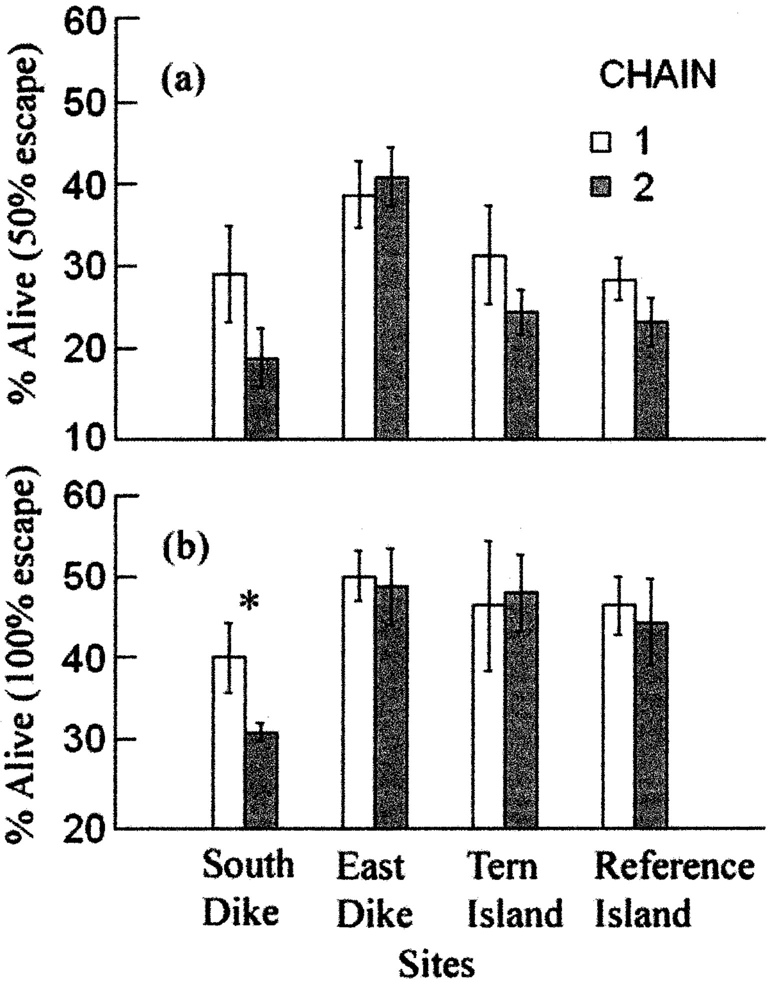

Figure 2-3. Percentage of live lake trout embryos (mean +/- SE) at final retrieval estimated using the (a) 50% escape correction; and (b) 100% escape correction, from 5 incubators per chain. Sites where survival differed from Reference Island (control site; LSD, $P < 0.05$) are marked with asterisks. Survival between chains within sites did not differ.

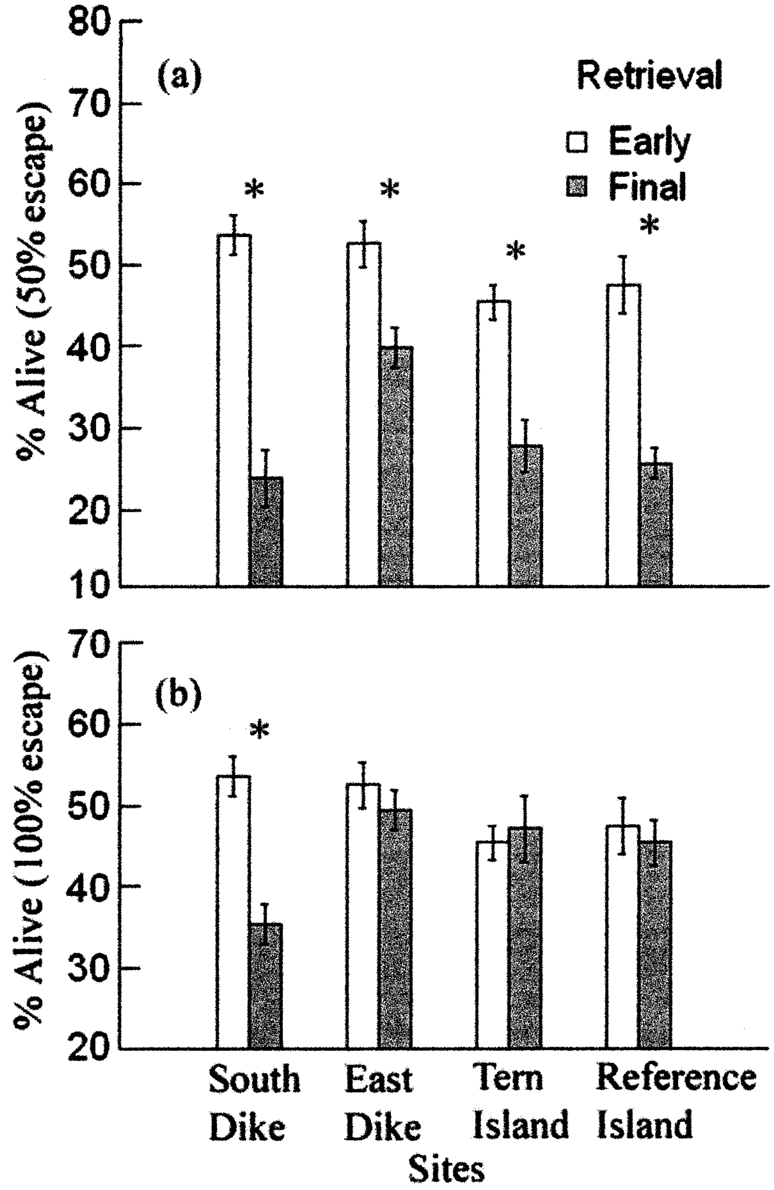

Figure 2-4. Survival of lake trout embryos (mean +/- SE) at early (n = 10 per site) and final (n = 10 per site) retrieval in incubators at each site using the (a) 50% escape correction and (b) 100% escape correction on the final retrieval. Sites with significant differences in survival between retrievals (t-tests, $P < 0.0125$) are marked with asterisks.

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Chapter 3. Simulated Blasting Effects on Mortality of Rainbow Trout Eggs

Introduction

Explosives are used in or near fish-bearing waters for many activities, including geophysical exploration, open-pit mining, construction, and other industrial development. If the resulting blasts are great enough, the pressure and shock waves produced can negatively affect fish and their incubating eggs in the vicinity. The sudden pressure deficit (measured indirectly as overpressure) that results from an explosion can, in particular, rupture the swimbladder and other soft organs of juvenile and adult fish (Wright 1982; Keevin et al. 1999). In contrast, developing eggs may be damaged more by the shaking of the substrate, measured as peak particle velocity (PPV; Wright 1982). Appropriately, guidelines in Canada provide maximum allowable limits for both overpressure (100 kPa) and PPV (13 mm/s) to protect fish and their incubating eggs, respectively (Wright and Hopky 1998).

When blasting occurs, energy emanates outward from the source. The intensity (i.e., energy per unit area per unit time) of the released energy decays due to geometrical spreading and is inversely proportional to the square of the distance from the source (Newman 1973). When blasting occurs in water, both pressure and energy waves are transmitted through the water and can be detrimental to fish and fish eggs (e.g., Kostyuchenko 1973, Wright 1982). Energy is also lost at geologic boundaries due to reflection, as only some is transmitted through the interface. For example, at a hard rock/water boundary, approximately 34 % of the energy is reflected back into the substrate (Telford et al. 1990). As such, when blasting occurs out of water, there is loss of energy (overpressure) as it is transferred into the water. On the other hand, energy waves (PPVs) measured at the interface do not have this loss of energy and are capable of exceeding the maximum allowable limit (13 mm/s) at a much greater distance from the source compared to overpressure. This shaking of the aquatic substrate may have negative effects on the fish eggs and the larvae that incubate in that substrate, due to direct physical shock, large distances from the source.

Sensitivity of fish eggs to physical shock has been documented for many species of cultured fish. This work has primarily focused on identifying developmental stages most sensitive to disturbances in order to minimize mortality during handling. As such, this work has not directly examined critical exposure levels. The majority of this work has focused on Pacific salmon (*Oncorhynchus* spp.; Smirnov 1954, 1955; Jensen and Alderdice 1983, 1989; Johnson et al. 1983, 1989; Dwyer 1993; Jensen and Collins 2003), although other cultured species have also been investigated (Holmefjord and Bolla 1988; Crisp 1990; Fitzsimons 1994; Hilomen-Garcia 1998; Gwo et al. 1995; Krise 2001). A study on non-cultured species was conducted by Battle (1944) who related sensitivity to differences in chorion thickness and the spawning life history characteristics. These studies indicate that egg sensitivity starts soon after fertilization and ends when the eggs have reached the "eyed" stage, characterized by the eyes becoming pigmented. For most species, the period of peak sensitivity occurs during epiboly, until the mesodermal sheath has completely replaced the vitelline membrane around the yolk (Velsen 1987). The duration of this sensitive stage, however, varies among species.

Because of their focus on sensitive developmental stages, many of these studies provide only qualitative assessments of the physical shock exposures. "Quantification" of mechanical disturbance exposures have included measuring the height that eggs are dropped into water (Battle 1944; Smirnov 1954), dropped onto mesh (Gwo et al. 1995), or are dropped inside an enclosure (Jensen and Alderdice 1983, 1989; Crisp 1990; Krise 2001). It has been suggested that this last technique allows conversion from drop height to more explicit units of exposure, such as energy (ergs; Jensen and Alderdice 1989) or PPV (Jensen 2003), via simple physical principles.

No actual blasting studies have demonstrated that PPVs above the Canadian guideline increase mortality of incubating eggs. In the only study to quantify PPV exposure due to blasting, Faulkner et al. (in press) found no elevated mortality of lake trout eggs, despite exposures (28.5 mm/s) more than double the PPV limit. Seymour and Nakatani (1967; cited in Post et al. 1974) and Post et al. (1974) recorded peak vertical accelerations experienced by eggs at a sensitive developmental stage when exposed to an actual or simulated nuclear detonation. Although the eggs received up to five repeated exposures of up to 98 m/s^2, no increase in mortality was detected. Acceleration is related to PPV via straightforward integration, but as knowledge of the frequency and material properties is required, it is difficult to convert their value to PPV. For comparison, however, the PPV of 28.5 mm/s measured by Faulkner at al. (in press) had a peak vertical acceleration of only 2.1 m/s^2, suggesting that the PPVs in the nuclear tests were well above the Canadian guideline.

Using a non-blasting, physical shock exposure, Jensen (2003) suggested that the lowest PPV to cause 10% mortality (LD10) of the most sensitive Pacific salmon (chinook; *O. tshawytscha*) eggs is 140 mm/s, more than an order of magnitude greater than the PPV guideline of 13 mm/s. This estimate, however, was based on studies (Jensen and Alderdice 1983, 1989) in which eggs held in an enclosure, free of surrounding water, were dropped and simple physical principles were used to estimate the velocity of the eggs at the end of the fall. This is similar to, but not exactly the same as, the particle velocities associated with a propagating wave. How the findings from this technique compare to blasting exposure in an aquatic environment is unknown. For example, Jensen and Alderdice (1983) provided evidence of reduced mortality for eggs held in water suggesting that the LD10 values reported by Jensen (2003) may, in fact, be conservative for naturally incubating eggs. Also unclear, however, is whether this drop-height technique, with or without water, is a realistic simulation of PPVs from blasting.

Although it has recently been suggested that the PPV guideline provides ample protection to incubating eggs (Jensen 2003, Faulkner et al. in press), its accuracy (i.e., its relation to actual LD10 levels) remains unknown. To address this concern, I developed a blast simulation technique to examine effects of PPV on egg survival. My technique aimed to produce PPVs representative of well-described out-of-water blasting events (Welz et al. in press), as well as those with much higher PPVs both in terms of amplitudes, frequencies, and total exposure levels. Both single and repeated exposures were examined, with eggs (in water) held loose in containers. A comparison was also done with eggs loose in containers and placed in spawning gravel. I also explored the accuracy of the LD10s of Jensen (2003) by exposing eggs both in and out-of-water to the same drop-height technique.

Methods

I focused my research on rainbow trout eggs due to their availability and relevance to previous studies (Post et al. 1974; Jensen and Alderdice 1989). Eggs from an anadromous form of rainbow trout were found to be intermediate in sensitivity to mechanical disturbance (drop-height; Jensen and Alderdice 1989).

I obtained fertilized and water-hardened rainbow trout eggs over the fall of 2004 and 2005 from the Raven Brood Trout Station, Caroline, Alberta. These eggs were placed in a cooler of oxygenated water and transported by vehicle (3 hrs) to the Biosciences Aquatics Facility at the University of Alberta. Upon arrival, eggs were disinfected using a weak iodine solution (1:600 by volume). Eggs were then apportioned randomly to the different exposure treatments. Eggs for the drop-height exposures were loaded loosely into a tray of a vertical stack incubator (Heath trays), whereas eggs for the blast simulation studies were loaded into containers that had previously been loaded into a separate vertical stack incubator. These plastic containers are open at the top and covered on the bottom with screening. Both vertical stack incubators were hooked up to the same recirculation tank kept at 8 °C (+/- 0.5 °C) in 2004 and 10 °C (+/- 0.5 °C) in 2005. The water was changed frequently to prevent high ammonia and low oxygen concentrations. Egg samples were preserved daily to monitor development.

Blast Simulation (Weight-Drop Apparatus)

I designed a weight-drop apparatus to conduct the simulated blasting events. The base of the weight-drop apparatus was constructed of a 1 m x 2 m x 0.64 cm steel plate that was insulated from the floor by three evenly spaced pieces of 5-cm foam padding (Figure 3-1). A 1.8 m long, 1.9 cm diameter pole was mounted in the middle of the plate 25 cm from one end (175 cm from the other end), and was secured at the top by three straps that were attached to nearby walls. The pole had a series of holes spaced 15 cm apart to allow for drop heights ranging from 2 cm to 137 cm. To ensure a smooth drop, an 18.1 kg weight was attached to 30-cm of oversized aluminium pipe, which slid freely over the pole. The weight was held at a desired height by fitting a pin into an L-shaped slot cut into the top of the aluminium sleeve. To release the weight, the pin was quickly pulled out allowing the weight to fall. A 0.64-cm rubber mat was placed at the base of the pole to cushion the shock of the weight and lower the frequencies transmitted into the steel plate. At the other end of the steel base, a 60 cm x 40 cm x 35 cm fibreglass tank was placed on top of two 0.64-cm rubber mats. In 2004, the back of the tank was 2.5 cm from the end of the plate and the front was 112.5 cm from the base of the pole. In 2005, the tank was rotated 90 degrees and the front was 112.5 cm from the base of the pole, and 22.5 cm from the end of the plate.

Exposures were recorded with an *OYO Sea Array 4*™ 4-component underwater geophone sensor that was coupled to the bottom of the tank. This sensor contains three self-orienting *30CT 10 Hz OYO* geophones to monitor movement in the longitudinal, vertical, and transverse directions. The sensor was attached to an *Instantel Mini Blast Mate*™ monitor, which was used to record exposures. The maximum PPV that can be recorded in any channel is 256 mm/s. In 2004, the sensor was kept in the tank during exposure whereas in 2005, the desired exposures were measured repeatedly before eggs were placed into the tank and the sensor was removed prior to egg exposure.

The weight-drop apparatus aimed not only to produce PPVs, but also to emulate the duration, frequency, acceleration, and energy content of actual blasting events recorded during the Faulkner et al. (in press) field study (Welz et al. in prep.). The PPV levels were reported as the peak vector sum of the movement in the three geophone channels. Duration was defined as the period (in seconds) of significant shaking. Frequency (Hz) was reported as the zero crossing frequency for each of the three geophone channels, which is the frequency of the largest wave peak. Peak acceleration (m/s^2) was reported for all three channels. To compare the energy content of the blasting events, energy was estimated using the formula for kinetic energy (KE):

$$KE = \left(\frac{1}{2}\right) m \sum_{t0}^{tn} v^2$$

where m = mass, and v = particle velocity (not wave propagation velocity) and t_0 and t_n are time zero and time final. Because in my application mass is constant, energy is directly proportional to v^2. To calculate the relative power of the event, the squared particle velocities of the three geophone channels were summed and plotted over time, and the area under the curve was calculated (Welz et al. in prep.).

Although more than one delay is commonly used during blasting events (i.e., more than one detonation is set off with a short interval between detonations), the current Canadian guideline does not address this. Therefore, in 2004, the simulation focused on single blasts with no delays. In 2005, however, repeated exposures were used to simulate multiple-delay blasting events that produce continued vibrations with durations in excess of 3 seconds.

2004: single exposure trials. – In 2004, exposures were carried out three times during egg development, at 51, 59, and 67 degree-days, all of which correspond to early epiboly stages (Vernier's stages 9, 9, and 10, respectively; Velson 1987). At each developmental stage, there were six PPV treatments (exposures; 12.4 – 219.3 mm/s) and a control (Table 3-1). Other parameters of the blast simulations are also given in Table 3-1. Replicates for each exposure consisted of four 6 x 6 x 5 cm containers, each holding an average of 132 (SD 21, N = 84) eggs. No dead eggs, characterized by a white and opaque appearance, were loaded into the containers. There were also six non-handled control containers, which were loaded with eggs, but received no further manipulation. During the exposures, 65 litres of water was transferred from the recirculation tank into the fibreglass tank. Water was changed during exposures to ensure temperature did not change by more than 2 °C. At each developmental stage and PPV level, four containers were placed in the tank, two on each side of the sensor, parallel to the exposure source. These containers were held tightly on both sides with Plexiglas spacers and all containers were coupled to the bottom of the tank. After exposure, the containers were loaded back into incubation trays to allow for further development.

2005: repeat exposure trials. – In 2005, exposures were carried out at 75 and 95 degree-days, which corresponds to one half epiboly and early organogenesis (Vernier's stages 11 and 14, respectively; Velsen 1987). For both stages, there were four PPV treatments (36.3 – 245.5 mm/s; Table 3-1). Examples of each level of exposure are shown in Figure 3-2.There were also six levels of repeat exposures for each developmental stage and PPV, i.e., eggs in each stage-PPV combination experienced 0 (handled control), 1, 2, 3, 5, or 7 "blasts" at 30-second intervals. Replicates for each exposure consisted of three 6 x 6 x 5 cm containers each containing an average of 99 (SD 27, N = 144) live eggs. There were also six non-handled controls. Water was transferred to the exposure tank as described above. During exposure trials, containers were placed in three rows of six, parallel to the exposure source, and were coupled to the bottom. An exposure of appropriate magnitude was then generated. At the end of the repetitions, the containers were returned to the vertical stack incubator to allow for further development.

Spawning-gravel effects. – In 2005, a separate set of trials examined effects of spawning gravel on survival of eggs exposed to simulated blasting. Spawning gravel was smooth river rock, ca. 3 cm in the longest axis, similar to that used by Post et al. (1974). For the with-gravel treatment, 8 x 6 x 5 cm containers were used, whereas 6 x 6 x 5 cm containers were used for the without-gravel treatment. The 8 x 6 x 5 cm containers were first half-filled with gravel, newly fertilized eggs were then loaded into the container, falling into the interstitial spaces between the gravel. Additional gravel was carefully added to fill the container and ensure the eggs were properly covered. The with-gravel containers had an average of 112 (SD 13) eggs, the without-gravel containers had an average of 123 (SD 28) eggs. Exposures were conducted at 92 degree-days (3/4 epiboly; Vernier's stage 12, Velson 1987). During exposures, four containers of each treatment were placed into the exposure tank in two alternating rows, parallel to the exposure source, and were coupled to the bottom of the tank. Five repeat exposures were carried out at the same PPV levels in other 2005 experiments (Table 3-1). After exposures, all containers were returned to the incubators for continued incubation. There were also eight non-handled controls for both the with and without-gravel treatments.

Mortality. – After all exposures, eggs were allowed to incubate until at least the eyed stage. It is believed that the majority of mortality due to physical shock will have occurred before this stage (Smirnov 1954, 1955, Johnson et al. 1989, Jensen and Collins 2003). Eggs were classified as dead (white, partially white in appearance, or without noticeable development) or alive. Eggs were subsequently preserved in Davidson's solution for later verification of classification. In some cases, unfertilized eggs were difficult to distinguish from eggs that died during exposures; therefore, they were included as dead eggs. Mortality was calculated on a per-container basis as the number of eggs that were classified as dead, when examined at the eyed stage, divided by the total number of eggs in the container.

Data analyses. – All mortality rates were arcsine square root transformed before statistical analysis to meet test assumptions (Zar 1999). This transformation is used on percentage or proportion data to meet the assumption of normality (Zar 1999). Control mortality was analysed both within and among treatments to assess mortality due to handling. Experiments in 2004, 2005, and the spawning-gravel study were analysed separately using analysis of variance (ANOVA). ANOVA is an analysis for making simultaneous comparisons between two or more means to determine if they are from different populations (Zar 1999). In 2004, differences in mortality were assessed among exposure levels, and development stages. For 2005, differences in mortality among repetitions of exposures were also assessed and, for the spawning-gravel study, differences in mortality were also assessed between treatments (with and without-gravel). For all treatments, Fisher's least-significance-difference (LSD) multiple comparison was used to identify exposure levels that differed from the handled control (Zar 1999).

For all statistical tests, a significance level α of 0.05 was used. Statistical analyses were performed using SYSTAT version 10 (SPSS 2000), unless otherwise noted. If the exposure treatments generated sufficient mortality compared to the handled controls, the exposures levels that cause 5 % and 10 % mortality (LD5, and LD10; respectively) were calculated using Probit analysis (PriProbit version 1.63 © 1998-2000 Masayuki Sakuma, Kyoto University, Kyoto Japan; Sakuma 1998), correcting for mortality due to natural response and handling (Finney 1971).

Drop-Height Apparatus

A drop-height apparatus was built according to specifications provided by Jensen and Alderdice (1989). The apparatus (see Figure 1 of Jensen and Alderdice 1989) has an aluminium carrier with a compartment to carry eggs in a small petri dish (60-mm diameter x 15 mm). This carrier is attached to a release platform with a quick release mechanism, which can be moved to any height (0 – 1000 mm). Upon release, two wires guide the carrier to ensure a smooth, straight drop. Friction was minimized by using oversized sleeves in the carrier guides and using lubricant on the wires. The carrier was also designed to come to an abrupt stop upon landing by weighting the carrier with lead shot.

Exposures were conducted at 51, 59, 67, 75, and 95 degree-days, which corresponds to different early epiboly to early organogenesis stages (Vernier's development stages 9, 9, 10, 11, and 14, respectively; Velson 1987). The first three exposure stages were conducted in 2004, while the last two exposures were in 2005. Exposures were conducted by dropping eggs 0 (control), 10, 20, 50, 100, 400, and 800 mm at each developmental stage. Assuming no effect from friction, these drop heights were translated into final velocities using the following equation:

$$V_t = \sqrt{V_0^2 + 2gh}$$

where V_t is final velocity (mm/s), V_0 is initial velocity (0 mm/s), g is acceleration due to gravity (9806 mm/sec^2), and h is drop height (mm). Using this equation, the final velocities (PPV) of the exposures were 0, 443, 626, 990, 1,400, 2,801 and 3,961 mm/s. Limitation of the apparatus made drop heights less than 10 mm (440 mm/s) unreliable.

The average weight of exposed eggs was calculated in 2005 from six samples of 30 eggs. This was used to estimate energy imparted on the eggs, as energy applied to an object at the end of a fall may be estimated by the following equation:

$$E = Mgh$$

where E = energy (ergs), M = mass of egg (grams), g = acceleration due to gravity 980.6 cm/s^2, h = drop height (cm) (after Jensen and Alderdice 1989).

Exposures were conducted in a manner similar to Jensen and Alderdice (1983, 1989). For each exposure, an average of 39 eggs (SD 11, N = 72) that were healthy in appearance (non-white) were placed in a petri dish free of surrounding fluid, which was then fitted into the slot at the top of the carrier. The carrier was released from the appropriate height to achieve the desired exposure (out-of-water treatment). Exposures were also performed with the eggs submersed in water (in-water treatment). All exposures were done in triplicate. After exposure, the egg samples were loaded into separate incubation compartments and allowed to incubate until at least the eyed stage was reached. Mortality was calculated as described above.

Mortality rates were arcsine square root transformed before statistical analysis to meet test assumptions (Zar 1999). Due to differences in stock source, each year of exposure was analysed separately. ANOVA was used to assess differences in mortality among the exposure levels, treatments (out-of-water and in-water), and developmental stages. The LD5 and LD10 PPV values were calculated as above for each year and treatment. These LD5 and LD10 PPV values were also transformed into energy (ergs) for comparison with previous studies. All other statistical tests were performed as described above.

Results

Blast Simulation (Weight-Drop Apparatus)

Overall, the blast simulations were adequate in reproducing real blasting events. The simulated blasts did, however, differ in some parameters from actual blasts at an open-pit mine (Table 3-1). At similar PPVs, frequencies and accelerations were higher, but durations lower in the simulated blasts, relative to actual blasts (Figure 3-3). The latter was addressed in 2005 using up to seven repeated exposures. Simulated blasts had lower relative power compared to actual blasting events at similar PPVs; however, this was more than compensated for by the repeated exposures in 2005. The highest simulated exposures had a relative power of 1846, compared to 73 at DDMI (Table 3-1).

Control Mortality. – Mortality of handled controls was similar among the three stages of development in 2004 ($F = 1.575$; df = 2, 9; $P = 0.259$) and two stages in 2005 ($t = 0.178$, df = 22, $P = 0.860$). There were, however, small increases in mortality between the non-handled and handled controls of 6.58 % in 2004 ($t = 1.947$, df = 16, $P = 0.035$) and 5.41 % in 2005 ($t = 1.430$, df = 28, $P = 0.084$; Figure 3-4); however, the latter was not statistically significant. During the comparison study there was no increase in mortality between the handled and non-handled controls for either the with or without-gravel treatments ($t = 1.769$, df = 12, $P = 0.051$ and $t = 0.498$, df = 23, $P = 0.312$; respectively; Figure 3-5).

2004: single exposure trials. – In 2004, mortality did not differ among the three stages of egg development ($F = 1.835$; df = 2, 75; $P = 0.167$), or among all seven PPV exposures ($F = 1.168$; df = 6, 75; $P = 0.332$). With the stages combined, mortality ranged from 21.1 % (SE 2.3 %) in the handled control to 27.5 % (SE 2.2 %) in the 219.3 mm/s exposure (Figure 3-4).

2005: repeat exposure trials. – In 2005, mortality differed between developmental stages ($F = 15.925$; df = 1, 134; $P < 0.001$), with 95 degree-days having higher mortality; therefore, each stage was analyzed separately. At 75 and 95 degree-day stages, mortality was similar among the different repeat exposures ($F = 2.447$; df = 4, 52; $P = 0.058$ and $F = 1.725$; df = 4, 52; $P = 0.159$; respectively). At 75 degree-days, mortality differed among exposure levels ($F = 5.331$; df = 4, 67; $P = 0.001$), although no exposures had mortality higher than the handled control (Figure 3-4). At 95 degree-days, mortality did not differ among exposure sizes ($F = 2.259$; df = 4, 67; $P = 0.072$) and was actually slightly less than the handled control in all but the 199.1 mm/s exposures (Figure 3-4). Overall, there was no increase in mortality due to exposure size or repetition of exposures.

Spawning-gravel effects. – Mortality was higher in the with-gravel than the without-gravel treatment ($F = 9.495$; df = 1, 39; $P = 0.004$). Unlike the other five developmental stages tested (all tested in the absence of gravel), mortality differed among exposure levels for the eggs in the without-gravel treatment ($F = 2.932$; df = 4, 18; $P = 0.050$), being greater than the handled control at both 199.1 and 245.4 mm/s (LSD $P < 0.05$; Figure 3-5). Mortality also differed among exposure levels in the with-gravel treatment ($F = 4.949$; df = 4, 17; $P = 0.008$); however, mortality was only greater than the handled control at 245.4 mm/s (LSD $P < 0.50$).

An LD5 of 145 mm/s (106 – 167; 95 % CI) and an LD10 of 168 mm/s (134 – 185; 95 % CI) was determined for the with-gravel treatment (Probit Analysis). Mortality was not sufficiently high in the without-gravel exposure (or any other exposure) to generate accurate LD values.

Drop-Height Apparatus

Mortality of handled controls did not differ among stages of development within years or exposure types (*t*-tests, ANOVA, $P > 0.05$). Mortality of the handled controls did differ, however, between years ($t = 3.336$, df = 28, $P = 0.002$), most likely due to a difference between egg batches. Consequently, each year was analysed separately.

In 2004 (51, 59 and 67 degree-days), egg mortality differed among exposure treatments (in and out-of-water; $F = 47.297$; df = 1, 115; $P < 0.001$), and exposure sizes ($F = 31.286$; df = 6, 115; $P < 0.001$), but not developmental stages ($F = 0.859$; df = 2, 115; $P = 0.426$). The out-of-water and in-water treatments were therefore analyzed separately with stages combined. Out-of-water mortality differed among exposure sizes ($F = 75.246$; df = 6, 55; $P < 0.001$), and all exposures ≥ 1,400 mm/s had higher mortality than the handled control (LSD, $P < 0.05$; Figure 3-6a). For the in-water treatment, mortality also differed among exposure sizes ($F = 10.331$; df = 6, 56; $P < 0.001$), with only the 2,801, and 3,961 mm/s exposures having higher mortality than the handled control (LSD; $P < 0.05$; Figure 3-6b).

In 2005 (75 and 95 degree-days), as in 2004, egg mortality also differed among treatments ($F = 28.053$; df = 1, 75; $P < 0.001$), exposure size ($F = 112.012$; df = 6, 75; $P < 0.001$), but not developmental stage of exposure ($F = 2.258$; df = 1, 75; $P = 0.137$). Analysed separately, mortality in the out-of-water treatment differed among exposure sizes ($F = 296.106$; df = 6, 35; $P < 0.001$), with mortality higher than the handled control for exposures ≥ 990 mm/s (LSD, $P < 0.05$; Figure 3-6a). For the in-water treatment, mortality also differed among exposure sizes ($F = 60.731$; df = 6, 35; $P < 0.001$); exposures ≥ 2,801 had higher mortality than the handled control (LSD; $P < 0.05$; Figure 3-6b).

The out-of-water treatment had lower LD exposure levels than the in-water treatment for both 2004 and 2005 (Table 3-2). Estimated LD5s for the out-of-water treatment (794 – 821 mm/s) were 66 – 77 % of the levels for the in-water treatment. Estimated LD10 exposures for the out-of-water treatment (925 – 970 mm/s) were 55 – 74 % of the LD10s for eggs in water (Table 3-2). Although my LD10 PPV values in the current study are greater than those of Jensen and Alderdice (1989), the LD10 for energy (ergs) was within the range they reported for Pacific salmon (Table 3-3).

Discussion

Blast Simulation (Weight-Drop Apparatus)

The weight-drop blast simulation attempted to replicate the effects of an out-of-water blast on spawning substrate, based on the field measurements in Lac de Gras, NWT

(Welz et al. in prep.). The most important component is believed to be PPV, and is used as the standard for protecting fish eggs around blasting operations (Wright and Hopky 1998). Although PPV is used as the guideline, other factors of blasting may also be important, such as durational exposure, the peak acceleration of the substrate, and energy content of the seismic waves.

The weight-drop procedure produced higher peak accelerations than those measured in Lac de Gras. In response, I added padding and made other improvements to the apparatus; however, frequencies remained higher, in large part due to the small scale of the apparatus. Higher frequencies likely result in greater exposure at a given PPV, due to the increased acceleration and the small mass of each egg.

The relative power of single simulated blasting events was lower than real blasting events, due to shorter durations and higher frequencies of events. By using the repeated exposures in 2005, the lab simulations achieved similar durations and energy as the actual blasts. Because mortality of eggs did not increase from these repeated exposures, it is likely that the peak energy content of the wave, estimated by PPV, is most important in predicting egg mortality due to blasting.

Because mortality was calculated at the eyed stage there is a possibility of increased mortality prior to hatching. Increases in mortality over this time were found to be relatively small for both the weight-drop and drop-height treatments, with a maximum increase of only 3.6 % and most importantly, the increased mortality was similar among all exposure sizes within all treatments (unpublished data). Therefore, using mortality calculated at the eyed stage should not bias results.

It is also important to note that calculations of mortality at the eyed stage included damaged eggs, characterized as being partially white, marbled in appearance, or containing a small white streak. Previous studies indicate that most mortality occurs within two days of exposure (e.g. Smirnov 1954, 1955, Johnson et al. 1989), although data on delayed mortality are not reported. Others have suggested that mortality is notable, with eggs turning white, within minutes after exposures (Jensen and Collins 2003); however, time of egg death has not been studied explicitly. In the current study, many damaged eggs developed embryos with pigmented eyes, indicating that development had continued for many days after exposure. These eggs continued to develop further to the hatching stage; however, all eventually died at or just before hatching. Thus, including damaged eggs as dead eggs is imperative to accurately assess mortality in early developmental stage exposures.

All six stages of development exposed to my weight-drop simulations are within the sensitive period (Jensen 2003; Jensen and Collins 2003; Johnson et al. 1983, and 1989), and corresponded to all stages of epiboly. Of course, care must be taken when handling eggs at this time, due to the potential to cause mortality (e.g. Johnson et al. 1983 and 1989). Comparison of handled and non-handled controls indicated that handling did cause mortality (up to 6.6 %), however, using exposure- and stage-specific handled controls, my experiments minimized this potential bias.

When eggs were free in the test containers (i.e., without gravel), neither single exposures up to 219.3 mm/s (during early epiboly), nor repeated exposures up to 245.4 mm/s (mid and late epiboly) increased mortality relative to handled controls. Although the increased mortality (4.9 %) at exposures ≥ 199.1 mm/s when eggs were at three-quarters epiboly was statistically significant, this outcome was because exceptionally low variability in the handled controls at this stage resulted in an exceptionally sensitive analysis. However, when comparing actual increases in mortality (effect size) above handled controls among exposure stages, no exposures lower than 199.1 mm/s showed an increase in mortality above 4.9 %.

Spawning gravel increased egg mortality, but also variability in mortality. As a result, only at 245.4 mm/s was mortality significantly above the handled control, although mean mortality was also above the handled control (by 10.3%) at 199.1 mm/s. Therefore, with-gravel exposures at least as high as 132.3 mm/s showed no increased mortality. The reason for the increased mortality with gravel is not known, but could have been due to movement of the gravel substrate in some of the containers.

Our spawning gravel results were generally consistent with those of Post et al. (1974), who used similarly sized gravel in their blast simulation. Those authors found no increase in mortality despite five repeated vertical acceleration exposures up to approximately 98.0 m/s^2, which is comparable to my findings that vertical accelerations > 51.3 m/s^2 (and probably closer to 184 m/s^2) were required to increase mortality at the most sensitive developmental period.

Drop-Height Apparatus

In the out-of-water treatment, the LD10 PPV for early-epiboly stages (2004 exposures) was higher than the average of three similar stages reported by Jensen (2003). For the mid- to late-epiboly stages (2005 exposures) the LD10 was again higher than the average for similar stages reported by Jensen (2003). It's not clear what was behind these differences; one possibility is differences in sensitivity between the source populations. Jensen's (2003) eggs were from a wild anadromous source, whereas my source was a domestic strain, although Krise (2001) found no difference in egg sensitivity between wild and domestic stocks. Although all attempts were made to replicate Jensen and Alderdice's (1989) methodology, I can also not rule out subtle differences between the drop-height apparatuses or the exposure procedures.

The increased LD10 PPV for the in-water treatment, relative to the out-of-water exposures, also seen for chinook salmon (*O. tshawytscha*; Jensen and Alderdice 1983), was considerably larger for the early-epiboly trials than for the trials with later-stage eggs. This suggests that although water consistently provides protection, the amount of this protection is reduced when the eggs are at a more sensitive stage. The protection provided by water is most likely the result of the eggs becoming less deformed on impact, since the water's density and viscosity is much greater than that of air. This, in turn, suggests that Jensen's (2003) LD10 estimates are conservative for eggs in an aquatic environment, assuming the estimated PPV accurately simulates blasting produced PPVs.

Conclusion

Lacking overlap in either PPV exposure or mortality for similar stages tested by the weight-drop and drop-height procedures, I cannot assess the latter's ability to realistically simulate a blasting event. The lowest exposure to cause significant mortality in the weight-drop simulations was 199.1 mm/s, corresponding to a drop-height of only 2 mm. Because the drop-height apparatus cannot reliably produce PPV exposures at such low heights, direct comparisons between methods cannot be made.

Although the accuracy of the drop-height method remains unproven in an absolute sense, it does provide a relatively simple method for comparing sensitivity of eggs among species and stages. It has already been used to describe sensitivity of Atlantic salmon (Krise 2001), milkfish (Hilomen-Garcia 1998) and chinook, chum, coho, pink, sockeye, and rainbow trout (anadromous) (Jensen and Alderdice 1983, 1989). However, egg mass must be taken into account when comparing sensitivities because the energy imparted on an egg in the drop-height procedure is directly related to its mass.

When drop-height exposures are converted to energy, rather than PPV, the most sensitive *Oncorhynchus* species tested during its most sensitive stages had an LD10 of 140 ergs (Jensen and Alderdice 1989). Using the same energy-based calculations, my rainbow trout eggs were intermediate in sensitivity, with an LD10 of 264 ergs. If the relative sensitivities are similar for blasting induced PPVs, then this relationship could be used with my weight-drop results to identify a data-based PPV guideline. The highest exposure that caused no increase in mortality in my weight-drop trials was 132.3 mm/s, which would then translate to an exposure of 70 mm/s as a conservative level below which no mortality is expected for *Oncorhynchus*. Thus, the current Canadian guideline of 13 mm/s should provide ample protection for *Oncorhynchus* spp. even during the most sensitive period of egg development. Although this limit may provide protection to incubating eggs from shaking of the substrate, further research may also be needed to ensure structural integrity of spawning habitats. Future work on blasting effects should focus on determining the egg sensitivity of other species of interest using both the weight-drop and simpler drop-height method.

Table 3-1. Characterization of weight-drop simulations during 2004 and 2005 and actual blasting events recorded at Diavik Diamond Mine Inc.'s (DDMI) A154 pit on Lac de Gras, NWT (see Faulkner et al. In Press). Parameters include peak particle velocity (PPV), reported as the peak vector sum of movement in the transverse (Tran), vertical (Vert), and longitudinal (Long) directions, duration of the shaking event, peak acceleration measured in each direction, zero-crossing frequency for each direction, and a relative power estimate of the waveforms. All values are reported as mean (SE). See text for additional details.

Year	N	PPV (mm/s)	Duration (s)	Acceleration (m/s^2)			Zero-Crossing Frequency (Hz)			Relative
				Tran	Vert	Long	Tran	Vert	Long	Power
2004	3	12.4 (1.0)	1.2 (0.4)	5.9 (0.6)	2.9 (0.6)	5.4 (0.2)	67.7 (7.8)	63 (20)	107 (61)	4.6 (0.3)
2004	3	31.0 (2.1)	0.84 (0.03)	12.3 (0.5)	20.3 (3.4)	22.2 (2.0)	41.5 (2.5)	59 (11)	133 (38)	46.3 (1.2)
2004	3	107.3 (7.4)	0.68 (0.04)	22.7 (2.0)	52.9 (5.6)	32.9 (2.8)	10.6 (1.0)	39.5 (7.0)	70.0 (3.1)	319 (27)
2004	3	117.7 (3.3)	0.71 (0.06)	25.1 (3.8)	62.1 (2.2)	39.4 (4.1)	9.8 (0.6)	38.5 (6.5)	100 (14)	435 (21)
2004	3	141 (12)	0.80 (0.03)	25.7 (2.6)	66.2 (6.0)	40.6 (5.0)	21.3 (6.7)	35.9 (5.4)	78 (32)	618 (97)
2004	3	219.3 (1.2)	0.89 (0.05)	27.7 (1.8)	137 (12)	39.7 (3.2)	18.4 (1.9)	41.4 (7.1)	70 (24)	1313 (56)
2005	15	36.3 (0.4)	0.53 (0.01)	3.3 (0.1)	12.4 (0.1)	10.5 (0.3)	44.2 (3.0)	48.0 (0.5)	91.4 (2.6)	43.7 (0.5)
2005	15	132.3 (2.4)	0.90 (0.34)	11.6 (0.5)	51.3 (1.8)	34.9 (0.7)	19.0 (1.2)	35.3 (0.6)	71.8 (0.3)	385.1 (7.0)
2005	15	199.1 (2.6)	0.82 (0.01)	27.3 (3.8)	184.5 (4.5)	41.7 (0.7)	11.9 (1.7)	38.0 (1.5)	77.1 (6.3)	1466 (21)
2005	15	245.4 (3.0)	0.88 (0.02)	25.9 (2.0)	243.5 (3.1)	48.0 (0.5)	12.9 (2.3)	26.1 (2.6)	109.8 (1.3)	1846 (16)
DDMI	10	15.9 (1.9)	3.4 (0.22)	1.2 (0.1)	1.2 (0.1)	1.4 (0.1)	9.2 (0.8)	12.7 (1.9)	9.2 (0.4)	73 (14)

Table 3-2. The drop-height exposures (PPV, mm/s; 95 % CI) estimated by Probit analysis to cause 5 % and 10 % mortality (LD5 and LD10, respectively) for the out-of-water and in-water treatments at 51, 58, and 67 degree-days and 75 and 95 degree-days of development.

Treatment	Stage	LD5	LD10
Out-of-water	51-67	794 (464 – 1,059)	970 (620 – 1,238)
In-water	51-67	1,195 (459 – 1,663)	1,756 (983 – 2,182)
Out-of-water	75-95	821 (747 – 884)	925 (856 – 985)
In-water	75-95	1,060 (902 – 1,200)	1,254 (1,095 – 1,393)

Table 3-3. Comparison of drop-height peak particle velocity (PPV), and energy estimated by Probit analysis to cause 10 % mortality (LD10) in eggs of *Oncorhynchus* spp., including anadromous (a) and domesticated (d) forms of rainbow trout (*O. mykiss*). Data are from (1) the current study, and (2) Jensen and Alderdice (1989). Most exposures were done with eggs free of surrounding water (no-water). The in-water treatment is also included for comparison.

Species	Source Study	Exposure	Height (cm)	PPV (mm/s)	Energy (ergs)
O. mykiss (a)	2	No-water	0.94	429	140
O. tshawytscha	2	No-water	0.39	276	140
O. kisutch	2	No-water	0.65	357	180
O. mykiss (d)	1	No-water	4.37	925	264
O. keta	2	No-water	1.8	594	440
O. gorbuscha	2	No-water	2.4	686	480
O. nerka	2	No-water	3.8	863	500
O. mykiss (d)	1	In-water	8.02	1,254	484

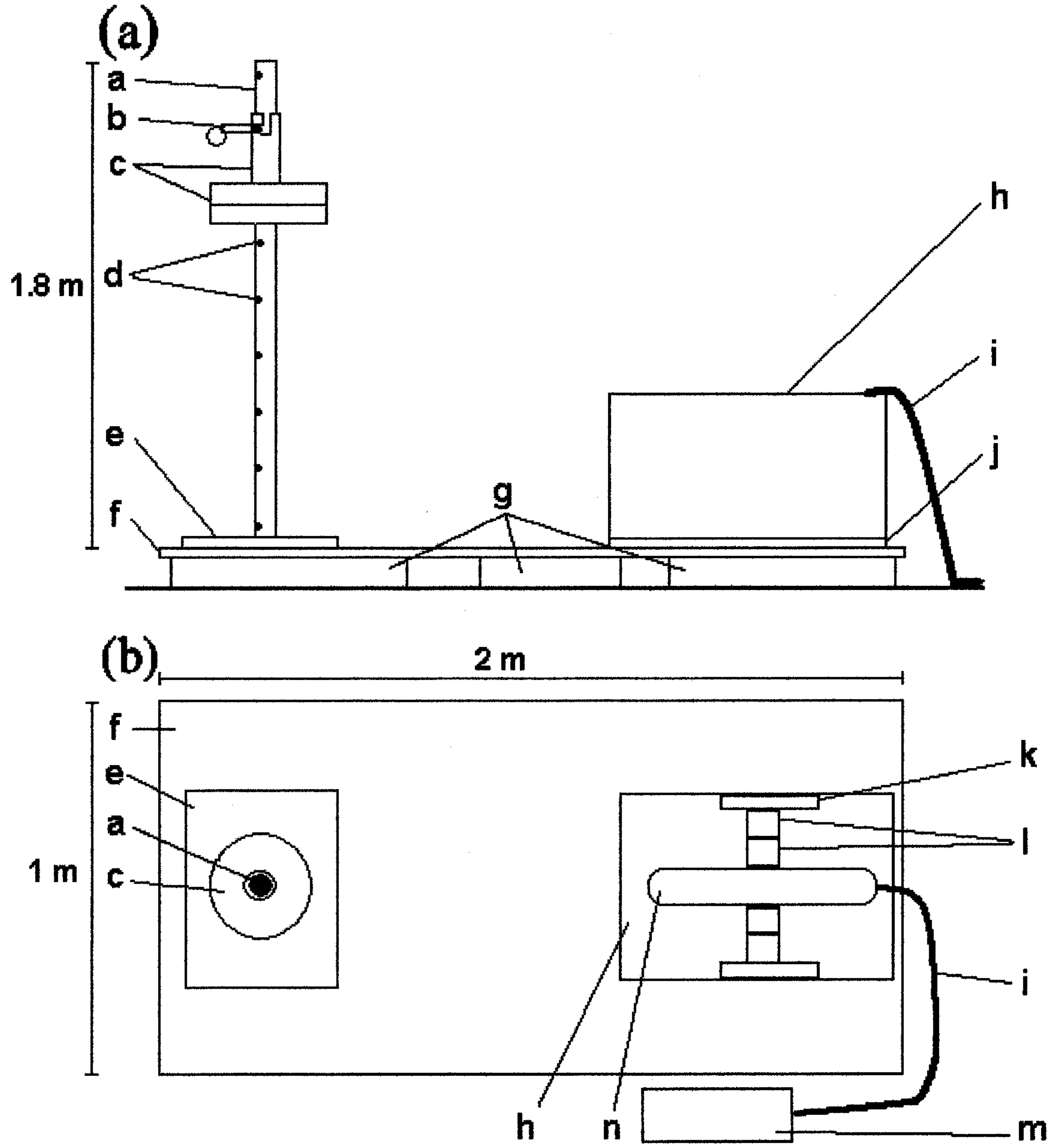

Figure 3-1. The weight-drop apparatus (a) side-view, and (b) top-view showing the 2004 exposure set-up. a: steel pole, secured at the top with straps (not shown); b: pin to release the weight apparatus; c: the weight set-up, including an aluminium sleeve and cast iron weights; d: holes drilled in the pole spaced 15 cm apart; e: 1 cm rubber mat; f: 0.64 cm thick steel base plate; g: 5 cm foam padding; h: fibreglass tank; i: sensor cable; j: 2.5 cm rubber mat, k: Plexiglas spacer; l: 6 x 6 x 5 cm containers; m: Instantel Mini Blast Mate™ monitor; n: *OYO Sea Array 4*™ 4-component underwater geophone sensor. For the 2005 exposure set-up, the exposure tank was rotated 90 degrees. See text for additional details.

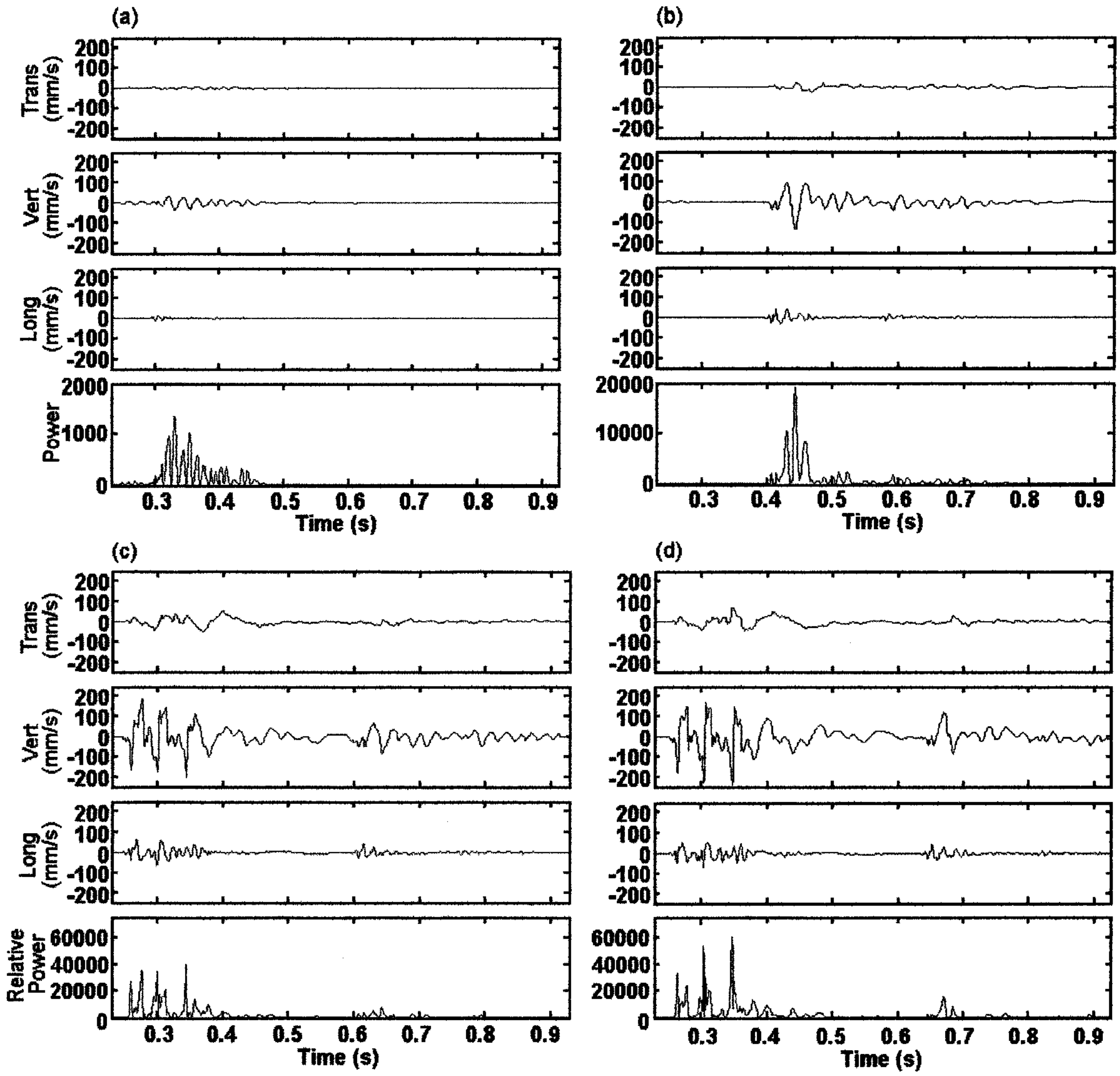

Figure 3-2. Examples of the four exposure levels (in PPV, mm/s): (a) 36.3 (SE 0.4), (b) 132.3 (SE 2.4), (c) 199.1 (SE 2.6), and (d) 245.4 (SE 3.0), used in 2005 (75, 92, and 95 degree-days). Measurements of peak particle velocity are given for the transverse (Trans), vertical (Vert), and longitudinal (Long) directions. Relative Power is an estimate of the magnitude of the combined movement (see text for additional information). Note the differences in the Relative Power axis scale. Actual peak particle velocities of these events were (a) 36.7 mm/s, (b) 138 mm/s, (c) 200 mm/s, and (d) 245 mm/s. To calculate error each exposure level was repeated 15 times prior to actual exposures.

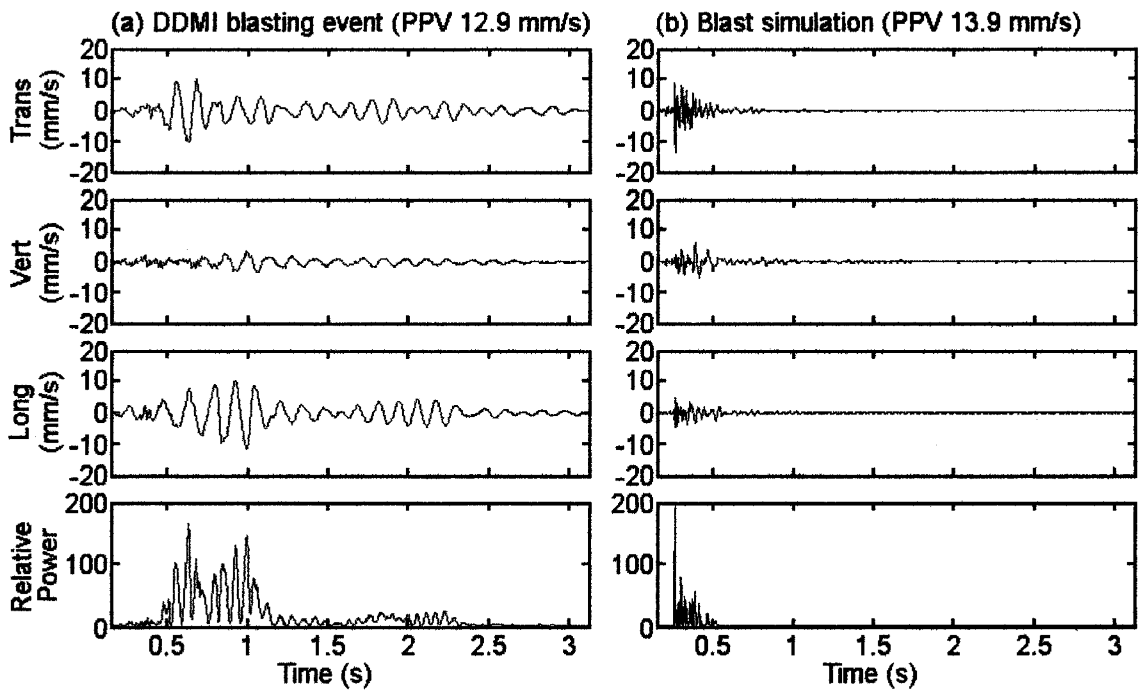

Figure 3-3. Comparison between (a) a blasting event measured in the lake substrate at Diavik Diamond Mine Inc.'s open-pit mining operation on Lac de Gras and (b) a laboratory blast simulation with a similar peak particle velocity. The transverse (Trans), vertical (Vert) and longitudinal (Long) components of peak particle velocity are shown, along with an estimate of Relative Power (see text for additional information). In 2005, multiple exposures were used to simulate longer events.

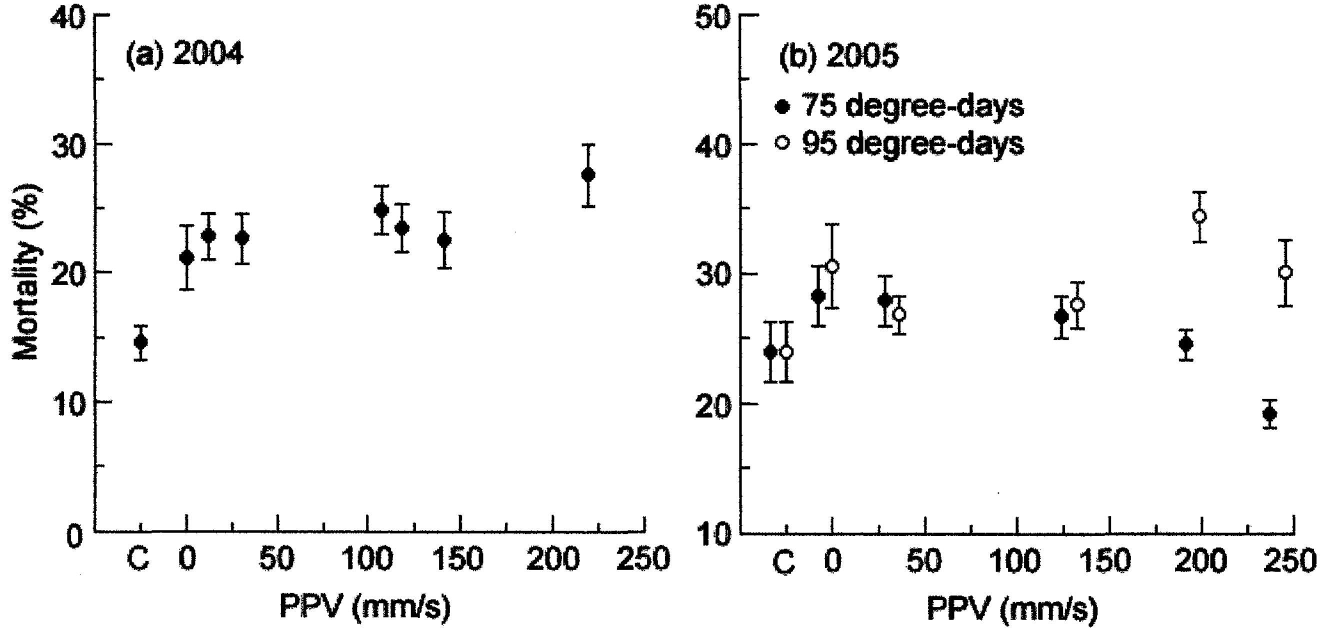

Figure 3-4. Mortality (mean +/- SE) of rainbow trout embryos in weight-drop exposures for (a) 2004 exposure trials (51, 59 and 67 degree-days, developmental stages combined), and (b) 2005 exposure trials (75 and 95 degree-days; offset to facilitate comparison). Handled control mortality is plotted at 0 mm/s; non-handled control mortality is at C. No exposures had mortality greater than the handled control (ANOVA, $P > 0.05$).

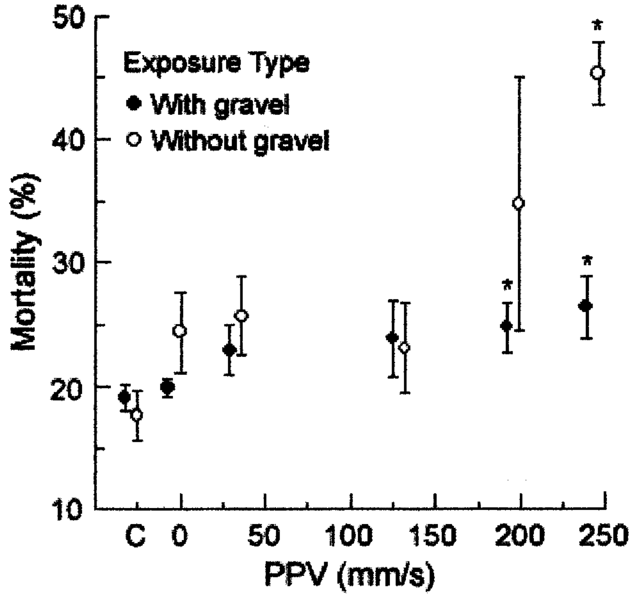

Figure 3-5. Mortality (mean +/- SE) of rainbow trout embryos (at 92 degree-days) in weight-drop exposures with and without spawning gravel. Handled control mortality is plotted at 0 mm/s; non-handled control mortality is at C. To facilitate comparison, the treatments are offset. *: exposures with mortality higher than the handled control (LSD $P<0.05$).

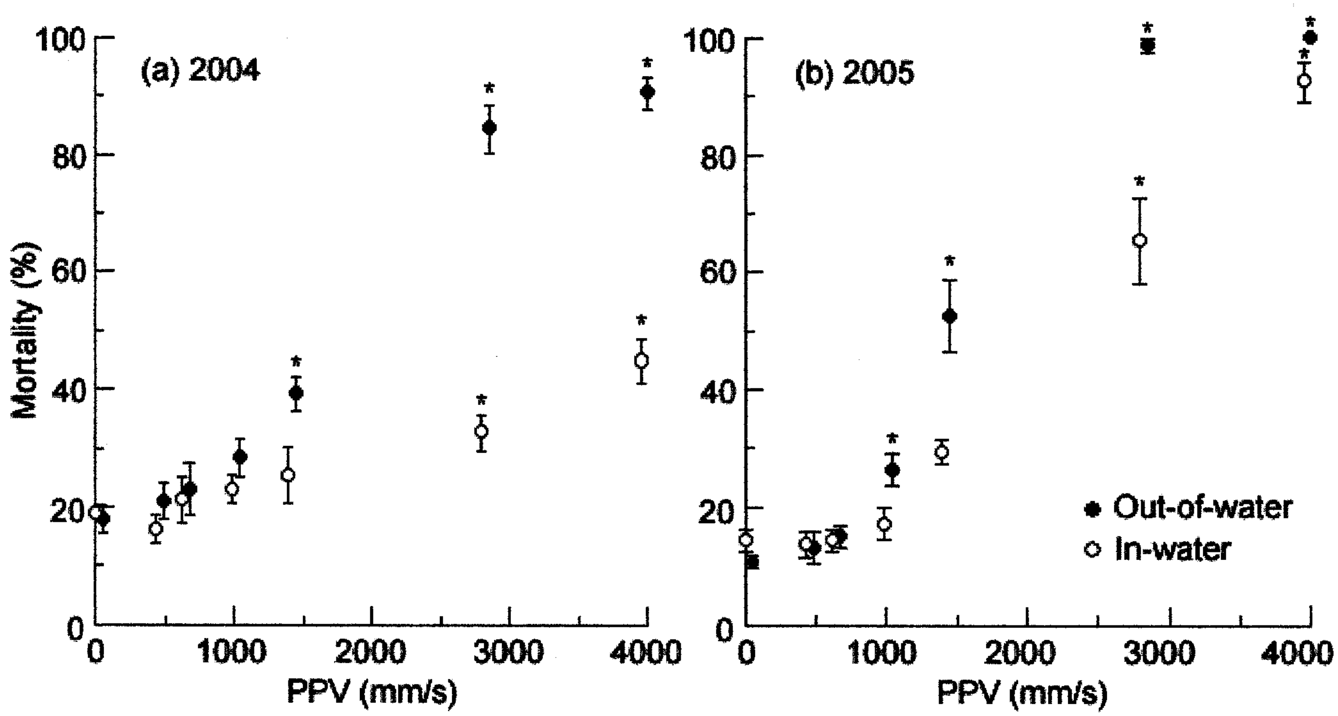

Figure 3-6. Mortality (mean +/- SE) of rainbow trout embryos in the out-of-water and in-water drop-height exposures in (a) 2004 (51, 59 and 67 degree-days, results combined) and (b) 2005 (75 and 95 degree-days, results combined). To facilitate comparison, the treatments are offset. Handled control is shown at 0 mm/s. *: exposures with mortality higher than the handled control (LSD P<0.05).

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Chapter 4. General Discussion

Blasting occurs in and near fisheries habitat from many sources. If blasting is of great enough magnitude, the corresponding shaking of the substrate has the potential to cause mortality of fish eggs that incubate in or on that substrate (Wright 1982). The Department of Fisheries and Oceans (DFO) have a maximum allowable limit for this shaking to ensure protection of fish habitat (Wright and Hopky 1998). This peak particle velocity (PPV) guideline of 13 mm/s, however, was based on minimal scientific information. Although the sensitivity of fish eggs to physical shock has been well documented (Battle 1944; Smirnov 1954, 1955; Johnson et al. 1983, 1989; Jensen and Alderdice 1983, 1989; Holmefjord and Bolla 1988; Crisp 1990; Dwyer 1993; Fitzsimons 1994; Gwo et al. 1995; Hilomen-Garcia 1995; Krise 2001; Jensen and Collins 2003), the level of PPV that will induce mortality of fish eggs was unknown, since no previous exposure studies have related mortality of fish eggs to blasting induced PPV. To address this information gap I conducted both a field experiment at Diavik Diamond Mine Inc. (DDMI) and blast-simulation experiments in the laboratory to determine the relationship between PPV and mortality of fish eggs.

At DDMI's mining operation at Lac de Gras, lake trout (*Salvelinus namaycush*) eggs in Plexiglas incubators (Kennedy 1980; Gunn and Keller 1984; Manny et al. 1989) were held in the lake substrate at four different sites. Three sites were in the predicted blast zone, where they experienced shaking from blasting in the open-pit mine. Under blasting conditions at DDMI's A154 open-pit, the maximum recorded blast had a PPV of 28.5 mm/s. Although this exposure is more than double the current PPV guideline, it did not cause an increase in mortality of lake trout eggs. This blast, however, occurred after the most sensitive stage of development (Fitzsimons 1994). During the most sensitive period of lake trout egg development (Fitzsimons 1994) the highest recorded exposure was only 4.4 mm/s. Not surprisingly, there was no increase in mortality.

Over the entire 10-month, over-winter incubation period of lake trout in Lac de Gras, survival was only lower at one of the three sites within the blast zone, relative to the reference site. This exposed site, however, did not have a larger exposure than any other blast-zone site but was, in fact, the only site composed of non-natural substrate used in construction of the dike. Natural variability among the sites likely contributed to differences in survival. Survival of lake trout eggs within all sites (35.4-49.4 %), including the one with reduced survival, was consistent with survival found in other studies on lake trout egg incubation in the Laurentian Great Lakes (1-49 %; Casselman 1995; Eshenroder et al. 1995; Manny et al. 1995). Natural variability in survival of lake trout eggs among sites, and the lack of control on the timing and sizes of the blasts, made it very difficult to demonstrate increases in mortality due to blasting. Therefore, I shifted effort to the laboratory, where the afore mentioned factors could be controlled.

Because of egg availability, I used rainbow trout (*Oncorhynchus mykiss*) eggs in the laboratory blast-simulations to define a PPV mortality threshold. Previous physical shock exposures had also focused on rainbow trout, which facilitated comparisons (Post et al. 1974; Jensen 2003). How sensitivities compare between lake and rainbow trout is unknown. Future work should focus on comparing egg sensitivity of these species.

In the laboratory, a weight-drop technique was developed to simulate blasts. Both single and repeated exposures were tested at several points during the sensitive developmental period, and an exposure was also conducted on eggs with and without spawning gravel. Repeated exposures did not increase mortality. The smallest exposure to increase mortality of rainbow trout eggs was 199.1 mm/s; however, mortality increased at only one stage of development (92 degree-days). The window of sensitivity thus appears to be very short. At this same developmental stage, mortality increased at the 199.1 mm/s treatment for both the with-gravel and without-gravel treatments, however, the increase was higher with eggs in spawning gravel.

The window of sensitivity that I measured at 92 degree-days (three quarters epiboly) appears to extend, at most from 76 to 94 degree-days. This range is consistent with Johnson et al. (1989), who found increased mortality during simulated handling exposures at only one day of developmental (98 – 99 degree-days) for rainbow trout eggs incubated at two different temperatures. Those authors, however, did not describe the corresponding development stage, so differences in developmental rates among stock source could have led to the small differences in degree-days of the most sensitive stages with my results. With much larger exposures and a different methodology (drop-height; see below), Jensen (2003) estimated the most sensitive period to occur earlier (78 degree-days). This may simply be due to the effects of the much larger exposures and the fact that Jensen (2003) corrected all egg mortality for mortality in the handled controls before any analysis (discussed by Johnson et al. 1989).

Jensen and Alderdice's (1983,1989) drop-height apparatus cannot reliably produce PPVs below 440 mm/s; thus, determining an exposure – mortality relation at these low exposure levels is not reliable. With no overlap in mortality or exposures between the weight-drop simulation and drop-height simulation, the appropriateness of the latter method for simulating blasts in an aquatic environment cannot be determined. Although the drop-height apparatus may not accurately predict mortality from blasting-induced PPVs, it does provide a relatively simple method of comparing sensitivity of eggs from different species, but only relative sensitivity, until the assumption that the relationship between "true" sensitivity and "drop-height" sensitivity is comparable among species is tested.

Jensen and Alderdice (1989) discussed parameters that may relate to egg sensitivity, such as egg mass, egg volume, egg density, internal hydrostatic pressure, and tension in zona radiata (a function of pressure and egg size). These parameters are often related to spawning life history characteristics. Species with pelagic eggs have been found to be extremely sensitive to physical shock compared to species with demersal eggs (Battle 1944; Hilomen-Garcia 1998). For the pelagic spawning milkfish, an LD10 (exposure level that causes 10 % mortality) was estimated to be only 0.8 ergs (Hilomen-Garcia 1998), compared to a minimum of 140 ergs in Pacific salmon (Jensen and Alderdice 1989). Battle (1944) also demonstrated that species with parental care of eggs (e.g. three-spine stickleback, *Gasterosteus aculeatus*) may also be more sensitive to physical shock than species without (killifish, *Fundulus heteroclitus*), and suggested that chorion thickness may be the factor that explains differences in sensitivity. Although these factors may influence egg sensitivity and be used to identify species of concern, the only parameter for which a correction is needed when comparing species with the drop-height treatment is egg mass, since the energy imparted on the egg at the end of the fall is related to the mass of the egg (Jensen and Alderdice 1989).

Using the drop-height energy exposures, the most sensitive *Oncorhynchus* species (*O. tshawytscha*, and *O. mykiss*) during the sensitive period had an LD10 of 140 ergs (Jensen and Alderdice 1989). The rainbow trout eggs in the current study had an LD10 of

263 ergs using a similar out-of-water exposure. In the drop-height simulations, exposing eggs in-water provided additional protection to eggs compared to exposures out-of-water. Because an in-water treatment may provide a better comparison, future work should focus on defining these in-water LD10s for other species. The difference in LD10 values mentioned above suggests that the most sensitive *Oncorhynchus* species is approximately twice (1.89 times) as sensitive as the rainbow trout in the current study assuming a comparable relationship.

The smallest exposure that increased mortality in the weight-drop simulations was 199.1 mm/s; exposures up to 132.3 mm/s did not increase mortality. This latter exposure could provide a conservative maximum allowable limit for protecting incubating eggs of rainbow trout. To ensure protection of other *Oncorhynchus* species, I will tentatively suggest that a PPV of 70 mm/s (132.3/1.89) should protect all Pacific salmon that have been tested. Future work should focus on determining the egg sensitivity of other species of interest using the simple drop-height method and weight-drop method and testing this "1.89" factor.

Increasing the PPV guidelines would greatly reduce setback distances for confined explosives. Using the equations provided in the guideline for use of explosions in Canadian waters (Wright and Hopky 1998), an increase in the PPV guideline from 13 mm/s to 70 mm/s would reduce the setback distance requirements for a 100 kg explosion in hard rock substrate from 150.9 to 52.7 m. While this setback distance is greatly reduced, it is still greater than what is needed to stay below the overpressure guideline (100 kPA) of 50.3 m for the same out-of-water explosion in hard rock substrate. While this guideline should protect incubating eggs of *Oncorhynchus* species, other species of interest need to be tested.

Although a 70 mm/s guideline may provide ample protection to developing eggs, this level might physically damage the habitat. This, in turn, might reduce the quality of the habitat. This effect would likely be more important for limnetic spawning sites, given their relatively static structure, than in the dynamic spawning substrate of most lotic systems. To ensure that productive capacity of the habitat is not lowered as a result of a higher PPV guideline, a structural guideline would also have to be considered for vulnerable habitat.

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