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Acoustic conditioning for recall/recapture of escaped Atlantic salmon and rainbow trout

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Abstract

Escape of salmon from sea cages is a problem that continues to plague the aquaculture industry. Data collected during the past 15 years from Norway, Scotland, Ireland, Canada and U.S. suggest significant impacts on natural runs of fish and economic losses to producers. The present report investigated the feasibility of using acoustic conditioning as a means of recalling/recapturing escaped fish. Atlantic salmon (*Salmo salar*) and rainbow trout (*Oncorhynchus mykiss*) were found to respond to frequencies in the range of 50 – 400 Hz equally well. Subsequently, both species were conditioned to a 250 Hz acoustic tone during feeding. Juvenile and sub-adult fish readily conditioned to the acoustic signal within four days, with the maximum number of fish responding (85% salmon, 96% trout) by day seven. To assess retention of conditioning, fish were exposed to a single tone without feed reinforcement every one, two or four weeks. Salmon and trout continued to respond for a seven month period with no significant decrease (88% salmon, 97% trout) in response. No significant differences were observed in the response of either species to tones differing in frequency by up to 200 Hz (89% salmon, 96% trout) and intensity by 20 dB (91% salmon, 96% trout). Both species were reproducibly recalled to a cage or feeding ring in a 3.7 m tank, but were reluctant to re-enter the cage. The findings indicate that salmon and trout are readily conditioned to acoustic signals and retain that conditioning for an extended period of time without reinforcement. These characteristics suggest that acoustic conditioning has potential as a means to recall escaped salmon and when coupled with recapture, can reduce interactions with wild stocks and losses to the producer.

Keywords: Atlantic salmon; Rainbow trout; *Salmo salar*; *Oncorhynchus mykiss*; Escape; Acoustics; Conditioning; Recapture

1. Introduction

The marked expansion of aquaculture has increased concern about potentially deleterious effects on the environment including: pollution from waste matter and feed, reduction of fishing and recreational areas and the spread of pathogens to native fish. One of the more biologically significant impacts is the escape of cultured fish and potential interbreeding with or displacement of native stocks (Arthington and Bluehdorn, 1996; Jonsson and Jonsson, 2006). Escapement is particularly troublesome for Atlantic salmon (*Salmo salar*) with some 94% of all adult fish reared in cages in the marine environment (Milewski, 2001). With global production of Atlantic salmon reaching 1.24 million MT in 2004 (FIDI, 2006), even a small percentage of escapement can be significant. Many escape events, particularly those related to catastrophic failures, involve a large number of animals. Norway has recorded an average of 500,000 fish escaping annually since 1992 with upwards of one million fish in 2005 (Hindar, 2001; intrafish.com September 2005) and British Columbia reported 26 escape events in a four year period. In January of 2005 a single storm event resulted in the release of some 685,000 Atlantic salmon off the coast of Scotland (The Scotsman, 23 February 2005). However, repeated small escape events may have greater impacts on wild populations (Skilbrei and Wennevik, 2006). Farmed salmon have been found in 77 rivers in British Columbia (Volpe et al., 2000) and outnumber native populations in several rivers in the Gulf of Maine (Whoriskey and Carr, 2001). Recent

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investigations suggest that escapees are genetically affecting populations in Norway (Skaala et al., 2006), Ireland (Coughlan et al., 2006) and Maine (O'Reilly et al., 2006).

Although development of more secure containment systems would reduce escapement, the current costs of fail-safe retention systems are not economically viable for salmon farming (Volpe, 2001). Furthermore, many escape events are associated with handling mishaps, e.g., transferring fish from net to net, that system design cannot readily prevent. In six escape events in Maine in 2000, three were storm related, one was a boat collision, another was vandalism and the cause of one remains unknown. In Norway, human error (including propeller damage, handling, and installation or technical malfunctions) accounted for 53% of escapes with another 30% related to storms and 12% caused by predators (Holm et al., 2003). In British Columbia, of the 26 escape events reported during 1997-2000, 38% were from net failures (7 related to predator attacks), 42% from handling error, 15% from boat events and 3% from system failure (Ministry of Agriculture and Lands, 2003). Land based facilities are often touted as an alternative to eliminate this problem but the cost of production is excessive and escapes into adjacent rivers and subsequent migration to the ocean have been documented (Stokesbury and Lacroix, 1997; Carr and Whoriskey, 2006).

Means to reduce the incidence of escapes is being given high priority within the aquaculture industry, and there have been improvements in procedures and policies (Porter, 2005). However, given the constraints outlined above it remains likely that Atlantic salmon will continue to escape from aquaculture operations well into the future. As a means to alleviate this problem, the potential of acoustic conditioning for recapture/recall of salmon that have escaped from marine aquaculture facilities was investigated. The approach entails conditioning the fish to associate feeding with a specific acoustic tone and has been found to aggregate several species of farmed fish including Atlantic cod (*Gadus morhua*; Oiestad et al., 1987; Ings and Schneider, 1997), rainbow trout (*Oncorhyncus mykiss*; Abbot, 1972), tilapia (Cichlidae; Levin and Levin, 1994), red sea bream (*Pagrus major*; Tateda et al., 1985), thicklipped mullet (*Creimugil labrosus*; Wright and Eastcott, 1982) and common carp (*Cyprinus carpio*; Wright and Eastcott, 1982). In the present report, development of a conditioned response in rainbow trout and Atlantic salmon was explored. The factors assessed include the duration of the training period, retention of conditioning and the ability to discriminate tones. Conditioning behavior was examined in both fresh and salt water, depending on the size of the fish. Additionally, the ability to recall fish to a pen within a tank also was examined.

2. Materials and methods

2.1. Fish and rearing conditions

Salmon fry (Merrimack River Stock) were provided by the North Attleboro National Fish Hatchery (North Attleboro, MA, USA) and cultured at the University of Rhode Island Aquaculture Center (Kingston, RI, USA). Eyed rainbow trout eggs were obtained from Trout Lodge (Sumner, WA) and hatched and reared at the same facility. Fish were maintained using standard husbandry practices (Piper et al., 1982) under a 12 L:12D photoperiod regime in tanks supplied with single-pass, aerated freshwater ranging seasonally from 8 to 16 °C with supplemental aeration. Animals were fed a standard floating salmonid diet (Nelson Silvercup, Murray, UT) three times daily. Tanks were siphoned as needed and scrubbed weekly.

For experiments in freshwater, three groups each of 40 salmon and trout from 15 to 25 cm in length were selected and transferred to six 3.7 × 1.2 m raceways (1150 l each) with a 25 cm water depth and a flow rate of 20 lpm. For seawater experiments, fish from 20 to 40 cm in length were maintained in 1.5 m diameter tanks in the Blount Aquaculture Research Laboratory (Narragansett Bay Campus, URI, Narragansett, RI) supplied with seawater filtered to 10 µm and heated or chilled to maintain a seasonal temperature range of 10 to 18 °C. Acoustic conditioning and experiments in seawater were conducted on replicate groups of 40 fish in a 3.7 m diameter tank (7200 l) with a flow rate of 50 lpm and water depth of 68 cm. Two 0.9 m high airlift pumps were placed in the tank to circulate water in a clockwise direction. No fish was used in more than one experiment.

2.2. Acoustic equipment

Acoustic signals were generated on a Dell Inspiron 1000 notebook computer (PC) using digital audio editing software (GW; GoldWave Inc., St. John's, Newfoundland, Canada). Pure sine waves were programmed using the formula sin(2*π*f*t) where f = frequency and t = time. Tones were produced with a commercial amplifier (Peavey IPA 1502, Lubell Labs, Columbus, OH) and broadcast from a 20 cm underwater speaker (AQ339 Clark Synthesis, Littleton, CO; frequency range 35 – 17,000 Hz) suspended in the water column at a constant location in the raceway or tank. Frequency, intensity and duration of the tones were controlled using the GW software. To avoid variation in sound pressure thresholds, acoustic signals were generated using a standardized method throughout the investigations.

Acoustic wavelengths at low frequencies are ≥ the dimensions of the tanks used in these investigations, limiting precise calibration of the acoustic source. However, it was possible to use a calibrated hydrophone to characterize the sound pressure level (SPL) broadcast in the tanks. The SPL was received and measured using a hydrophone (Model 6083; International Transducer Corporation, Santa Barbara, CA) with the output filtered and amplified using an Ithaco 4113 filter and 451 amplifier (Ithaca, NY). The signal was subsequently digitized using a NI DAQ 5102, 8 bit digitizer (National Instruments, Austin, TX). The hardware was controlled by Virtual Scope software (National Instrument) which enabled the capture and storage of the received signals. A series of trials was conducted by varying the volume setting on the PC software and the volume on the IPA 1502 amplifier. For each trial a signal was recorded and stored for subsequent SPL evaluation.

For evaluation trials the PC and amplifier volumes were simultaneously adjusted to create the range of sound pressure levels. The received signals were band pass filtered between 1 Hz and 2000 Hz, and digitized at 5 kHz. The receiver amplifier gain, G, was set between 10 and 43 dB. The source sound level (SL) in dB was calculated as:

$$\mathrm{SL} = \text{sound pressure level (SPL)} - \text{transmission loss (TL)}$$

where SPL = | M_x | - G + 20 × $\log_{10}$ (V_{rms}) with | M_x | = hydrophone sensitivity, G = amplifier gain, and (V_{rms}) = the root mean square voltage from the received signal, and

$$\mathrm{TL} = 20 \times \log_{10}(\mathrm{r}),$$

where r = range in meters between the hydrophone and source.

The SPL varied from 110 dB to 160 dB contingent on the volume settings and the frequency. Once calibrated, the system was validated routinely in the experimental tanks with a Dolphin-Ear 100 series hydrophone (www.dolphinear.com). The system allowed for sound intensity to be adjusted from 110 to 150 dB and frequency from 50 to 400 Hz in 50 Hz step increments. The signal strength was maintained at 134 dB in all experiments except as noted.

2.3. Conditioning

Forty fish were transferred to each of three raceways and acclimated to the size, shape and color of the underwater speaker before initiation of conditioning.

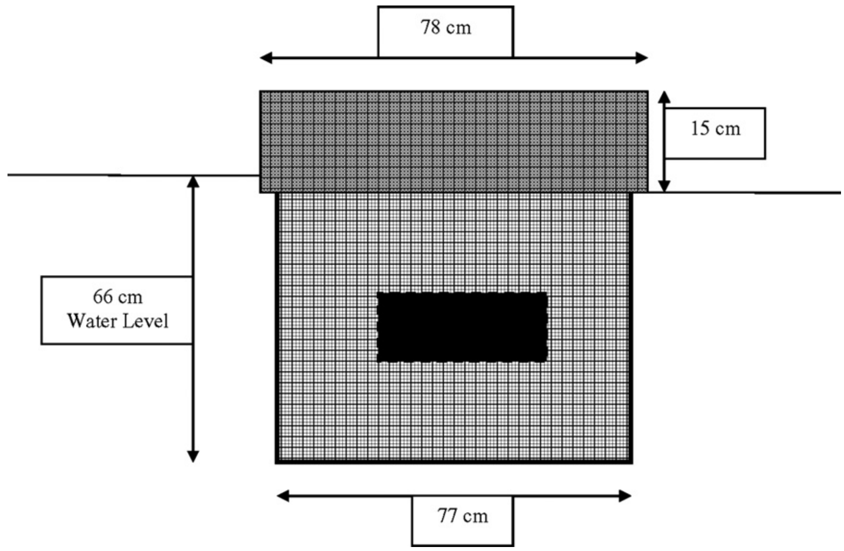

Fig. 1. Line drawing of the cage used for recapture investigations. The body of the cage was constructed of coated wire mesh with rigid polyethylene mesh extending 13 cm above the surface of the water to retain the floating feed. Diagram is not to scale.

Fish were trained to feed at a constant location using floating feeding rings (26 cm diameter) that remained in the raceways at all times. The ring was not an attractant, and the fish would distribute themselves evenly throughout the tank in the absence of food. To condition the fish, an acoustic tone was initiated and the floating feed was placed into the ring by a researcher maintaining a low profile alongside the tank. The tone was repeated three times at 134 dB for a one minute duration with a period of five minutes between each, three times daily during the conditioning period. All feed was consumed during the duration of the tones. Upon demonstration of a positive response, i.e., active swimming into and milling about the feeding ring when the tone was played without a food reward, the fish were considered conditioned. The response was measured as the percentage of the 40 fish that entered the ring to feed during the one minute duration of the tones. In addition to the conditioning trials, the fish were fed to satiation twice daily (broadcast throughout the tank). Both the standard feeding and the acoustic conditioning were conducted at varying times of the day to avoid temporal conditioning. To reduce the potential for fish to habituate to visual cues, movement of personnel in the vicinity of the raceways occurred during non-feeding periods. Conditioning of fish in the 3.7 m tank in seawater was conducted in an identical manner.

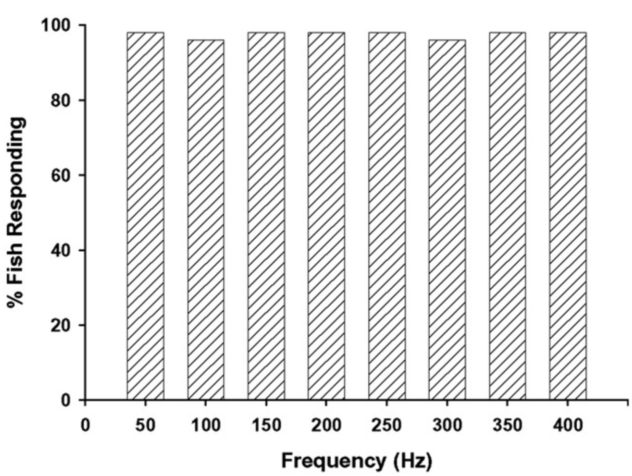

Fig. 2. Initial trials to determine the frequencies Atlantic salmon were capable of sensing. Values are the percentage of fish (n = 40) responding to a novel acoustic signal over the range of 50 – 400 Hz during a single exposure. Response was indicated by a marked increase in activity, cupping of the body and movement away from the sound source.

2.4. Experimental regimens

Retention of the response was assessed in three raceways each containing 40 salmon or trout conditioned to a 250 Hz tone at 134 dB for 14 days and subsequently exposed to the tone at weekly, bi-weekly or monthly intervals for a period of seven months without feed reinforcement. The percentage of individuals returning to the feeding ring in the absence of feed was measured. Only data from the monthly intervals, when all three groups of animals were tested, were used for statistical analyses.

The ability of fish to discriminate between specific frequency tones was assessed in three raceways each containing 40 salmon or trout conditioned to a 250 Hz tone with feed for 14 days as above, then exposed to a 250 or 100 Hz signal (3 × 1 min with 5 min intervals between stimuli) in the absence of feed. A second series of experiments examined the scenario in which fish were exposed to a 100 Hz signal for 14 days without feed reinforcement followed by

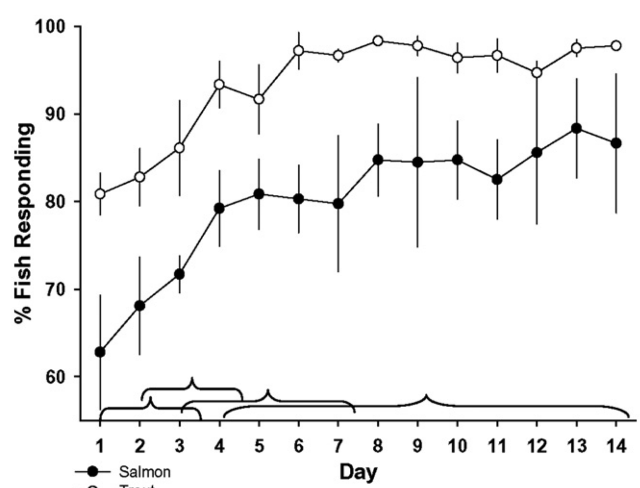

Fig. 3. The response (aggregation at feeding ring) of Atlantic salmon (closed circles) and rainbow trout (open circles) to a 250 Hz tone over a period of 14 days of conditioning. Three replicates of each species were examined daily; values are the mean + SEM. The brackets grouping days indicate statistical similarity (RM ANOVA with a Holm-Sidak method for paired comparisons, $P > 0.05$). Untransformed data are presented as the patterns of statistical significance did not differ between untransformed and ranked data.

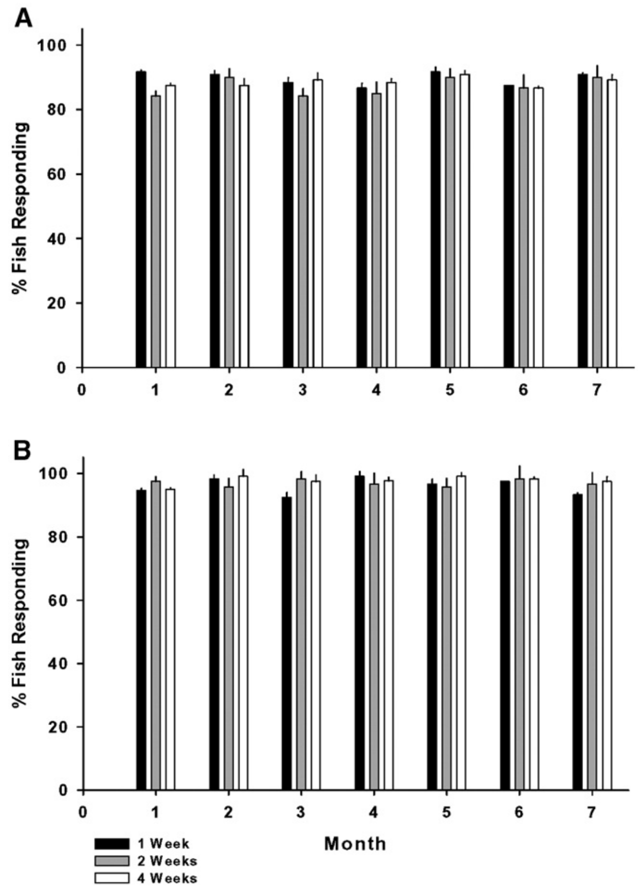

Fig. 4. Retention of conditioning in (A) salmon and (B) trout exposed to a 250 Hz tone without feed reinforcement at intervals of 1 (black), 2 (grey) or 4 (white) weeks monthly. Values (mean + SEM) are expressed as percentage of positive responses for 40 fish per time point with 3 replicates per point. No significant differences were detected.

conditioning for five days to 300 Hz with feeding. Subsequent trials (i.e., replicates) were conducted to determine whether the fish responded to both tones, with the degree of response to the "non-conditioned" 100 Hz tone used as the measure of the ability to discriminate.

The potential to recall salmon to a confined area was examined within the 3.7 m circular tank. Fish were held inside a 77 cm square pen (0.4 m^3) constructed of plastic-coated 5 cm mesh steel wire with four 15 h × 78 w cm sections of rigid, fine mesh plastic wire attached to the perimeter. The top of the pen was open and extended 13 cm above the surface of the water to retain feed (Fig. 1). Groups of 10 trout or salmon were conditioned to a 250 Hz tone for 14 d in the pen as above. The number of fish feeding within the pen during the conditioning regimen was assessed to determine whether the space was adequate to allow for "normal" behavioral responses, i.e., absence of aggressive interactions. Following completion of the conditioning regimen, a side panel was removed to allow movement in and out of the pen. Active entry into the pen during the tone playback was considered a positive response to the specific tone. Fish movement was recorded using an underwater video camera system (Aqua-Vu ZT-50, Nature Vision, Inc., Brainerd, MN). Three separate groups of both salmon and trout were examined sequentially in the single 3.7 m tank.

2.5. Statistical Analyses

Triplicate experimental units were utilized in the initial conditioning experiments (Section 2.3). Data were analyzed as a two-way RM ANOVA with repeated measures on the group of fish within the raceway or tank, and the two factors being species (trout vs. salmon) and time (days). The Holm –Sidak method was used to assess differences between paired comparisons. Retention of the conditioned response (Section 2.4.1) was analyzed as a two-way ANOVA with month of observation (one to seven) and weeks between stimulus (one, two or four) examined separately for salmon and trout. Transformations were not required because the number of individuals per replicate/experiment remained constant and thus certain percentages were not overrepresented. The differences in response to conditioned or novel tones (Section 2.4.2) were assessed as a paired comparison of the average response per day to each tone. Data that failed a test of normality or equal variance were analyzed with Mann-Whitney Rank Tests. Data analyses were conducted using SigmaStat 3.1 software (Richmond, CA). Values reported in the text and figures are expressed as mean percentage ± SEM unless otherwise noted and all differences discussed in the text are significant at a level of $P < 0.05$.

3. Results

3.1. Time required to condition

Fish responded robustly to acoustic tones ranging in frequency from 50 to 400 Hz (Fig. 2). The initial reaction to the acoustic signal was a startle response in which the body curved in a C shape (cupping) and the fish moved away from the speaker. Within 2 - 3 d, acclimation to the signal was apparent with no presence of a startle response. A

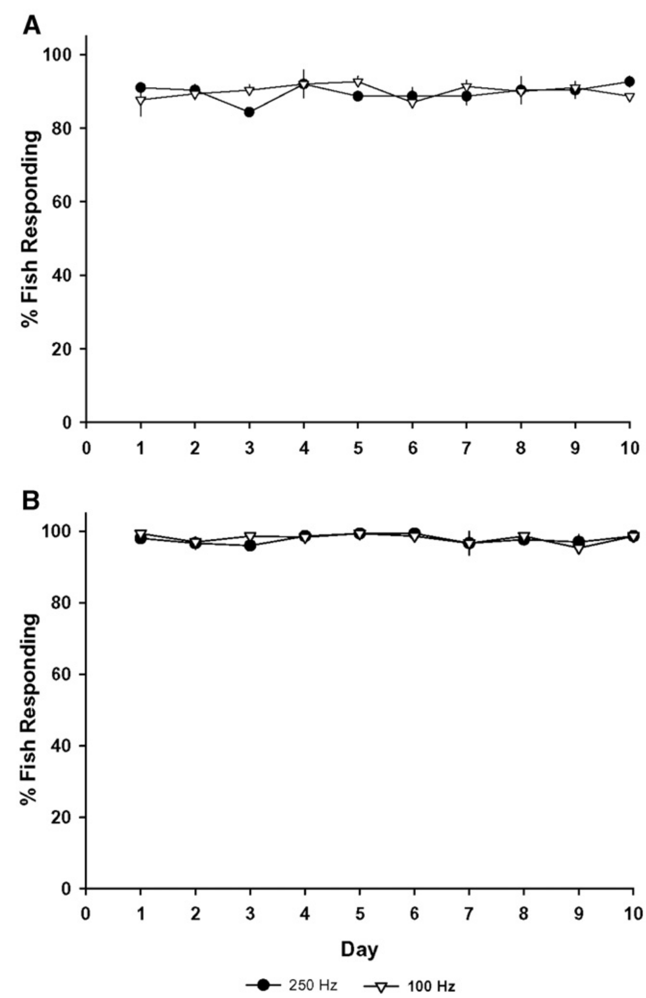

Fig. 5. The response of (A) salmon or (B) trout conditioned to a 250 Hz tone for 14 days prior to being exposed to the 250 Hz tone (solid circle) or a novel 100 Hz (open triangle) in the absence of feed (Day 1). Values are the percentage (mean + SEM) of 40 individuals responding over 3 different trials. No significant differences were observed.

frequency of 250 Hz provided a “clean” signal in the tanks and was used as the standard for the majority of the subsequent studies. Using feed as a positive reinforcement the fish were readily trained to return to a designated area delineated by the feeding ring within four days, and by day seven, the percentage of individuals responding did not increase further (Fig. 3). Upon initiation of the signal fish immediately and vigorously swam to the feeding ring. Following cessation of the signals fish vacated the vicinity of the feeding ring and again dispersed throughout the tank. Following 7 to 14 d conditioning, a higher percentage of trout (96%) responded to the acoustic stimulus than did salmon (85%).

3.2. Retention of conditioning

After an initial two week period of conditioning to a 250 Hz signal, neither salmon nor trout exhibited a decrease in the response to the signal over a period of 7 months (Fig. 4). Both species responded in a similar manner regardless of whether the stimulus was presented at one, two or four week intervals following the initial conditioning period. The overall performance of both species was similar to that observed in the first experiment with 96.9±0.4% and 88.4±0.5% of the trout and salmon responding, respectively.

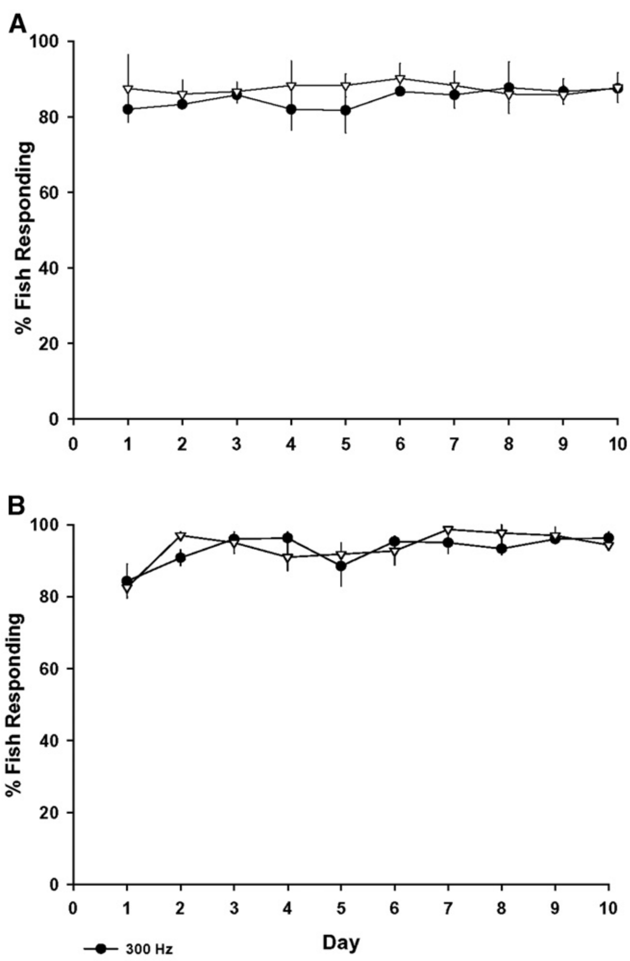

Fig. 6. Percentage of (A) salmon or (B) trout initially exposed to a 100 Hz signal for 14 d, conditioned with feed to a 300 Hz tone for 5 d and subsequently exposed to 300 Hz (solid circle) or 100 Hz (open triangle) in the absence of feed. Values are the percentage (mean+SEM) of 40 individuals responding over three different trials. No significant differences were observed.

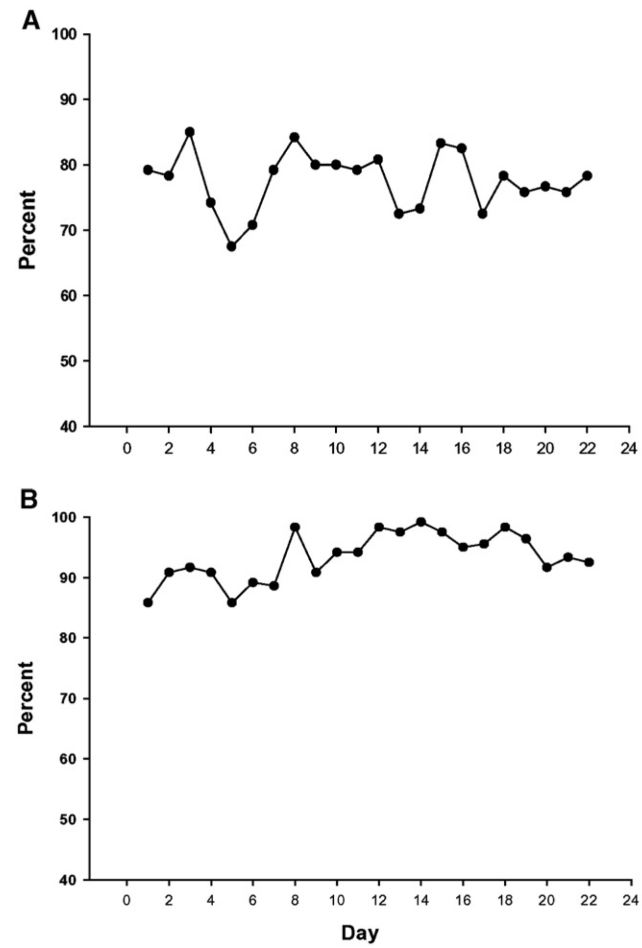

Fig. 7. Percentage of (A) salmon or (B) trout returning to the pen or attached feeding ring in response to an acoustic tone following release. Fish were conditioned to a 250 Hz tone for 14 days in the pen prior to release into the tank. Values are the percentage of 30 individuals returning daily.

3.3. Discrimination of frequency and intensity

Salmon conditioned to 250 Hz (89.8±0.6%) responded equally well to the novel 100 Hz tone (89.3±0.7%; Fig. 5). Similarly, there was no difference between the trout response to the two tones (98.1±0.4% and 97.3±0.6%). There also was no differential response of trout or salmon to 100 and 300 Hz signals with the reverse conditions, i.e., exposure to a 100 Hz signal for 14 d without feed reinforcement followed by conditioning for 5 d to 300 Hz with feeding (Fig. 6). Varying the intensity of the tone by 21 dB did not alter the response of either species with response rates of salmon to decibel levels of 147 and 126 at 90±1 and 92±1, and trout responding at 96±1 and 97±1%.

3.4. Return potential

Salmon maintained within the pen in the 3.7 m tank successfully conditioned to the 250 Hz tone as evidenced by increased activity and aggregation at the top of the pen during the acoustic signal. The density of the fish in the pen was high (3 – 4 Kg for salmon) and when the side panel was removed the majority of individuals migrated out and were reluctant to re-enter the confined space of the pen during the tone stimulus (<50%). To contend with this, a 56 cm diameter feeding ring was attached to the pen as an alternative return site and aggregation at either site was considered a positive response. The average percentage of individuals returning daily to the feeding sites in response to the tone over a period of 14 d was 76.3±1.3% for salmon, and 97.2±0.4% for trout (Fig. 7). While the percentage of fish responding remained the same before and after release, there was a visible decrease in aggressive feeding behavior when the fish were not confined to the pen.

4. Discussion

The ability of fish to detect sound varies markedly from terrestrial animals. The most significant factors impacting sound perception in fish are the large differences in the physical characteristics and sound propagation of air and water. The greater density and viscosity of water require a higher energy to propagate a sound wave. Once initiated the sound wave travels at 4.5 – 5 times the speed in air (1500 m per second versus 330) and persists for longer distances. Of note, sound waves in water generate both particle displacement/acceleration (near field) and pressure (far field), potentially providing fish with greater information than terrestrial organisms where air pressure is the primary component of sound waves (Jobling, 1995). While the nature of the sound waves within the confined space of the tank in the current experiment was likely particle acceleration which decreases with distance from the source, the results indicate that both salmon and trout conditioned to a sound stimulus in a short period of time and retained that conditioning for several months in the absence of the stimulus.

Although salmonids lack accessory auditory structures and have comparatively limited aural acuity (Hawkins and Johnstone, 1978), in the studies presented here both salmon and trout were able to detect acoustic signals ranging from 50 to 400 Hz. Of greater significance, both species were readily conditioned to return to a specific location in response to a 250 Hz tone of 134 dB. Indeed, the response plateaued within four days of initiation of conditioning. These results are in agreement with previous studies in several species of fish, including rainbow trout that were conditioned to respond to a 150 Hz tone by swimming to a specific pond site in anticipation of feed (Abbott, 1972). Thick lipped mullet and common carp were successfully conditioned together to associate an acoustic signal of 150 Hz with food (Wright and Eastcott, 1982).

Escape of sea cage reared salmon is a problem with which the aquaculture industry continues to struggle (Jonsson and Jonsson, 2006). Data from Norway, Scotland, Ireland, Canada and U.S. suggest varying impacts on wild runs of salmon (Naylor et al., 2005). Differences in the life history of native and cultured fish, including age and size at maturation, time of spawning and migratory behavior have the potential to disrupt natural spawning. In Norway, several populations of native Atlantic salmon have been reduced or replaced by cultured fish escaped from net pens (Heggberget et al., 1993; Fiske et al., 2006; Skaala et al., 2006). Similarly, in the British Isles escapees from aquaculture operations may comprise up to 35% of the commercial salmon fisheries in some eastern rivers (Youngson et al., 1997), but more typically make up <5% of the population (Walker et al., 2006). Interbreeding of wild and farmed fish in Ireland has been documented, and found to affect both single-locus and high-heritability quantitative traits, e.g., growth and age at maturity (McGinnity et al., 1997, see review in Jonsson and Jonsson, 2006). Investigations in Canada revealed that for nearly a decade, aquaculture escapees have outnumbered wild fish returning to the Magaguadavic River, (Whoriskey et al., 2006). The escaped salmon entered the river later than wild salmon and may have been responsible for up to 55% of the redds examined (Carr et al., 1997). Escaped fish also have been implicated in spread of pathogens and the decline of more than 30 populations of salmon in Norway by introduction of the monogean parasite, *Gyrodactylus salaries* (Heggberget et al., 1993). Similarly, high mortality of adult salmon has been observed as a result of introduction of furunculosis into wild stocks, potentially through escaped fish (Johnsen and Jensen, 1994). Whether justifiable or not, escapees from the salmon aquaculture industry are seen by many as contributing significantly to the global decline in populations of wild salmon.

The findings of the current investigations suggest the potential for use of acoustic conditioning to recapture/recall escaped salmon. Both trout and salmon were readily trained in a short period of time to return to specific locations in response to an acoustic tone and retained conditioning for the seven month duration of the study with a single monthly reinforcement. The limited training and reinforcement required to maintain conditioning could be integrated readily into standard husbandry or feeding practices. The higher rate at which rainbow trout were conditioned (96% vs 88%) is likely related to the feral/wild nature of the Atlantic salmon employed. The strain of trout used has been domesticated for more than six generations and selected for production traits. Domesticated stocks of Atlantic salmon (Gross, 1998) may condition at the higher rate characteristic of trout.

Although acoustic conditioning was demonstrated to be an effective method of recalling salmon and trout in a laboratory setting, the utility on a commercial scale has yet to be demonstrated. One potential constraint is developing an efficacious means of recapture as fish would not readily enter the small pen in the current study. However, several methods to capture free-swimming salmon have been developed, including weir systems with leaders that should obviate/reduce this concern (Brothers, 1999). Noise pollution in a congested marine environment also might reduce the efficacy of this approach. Fortunately, the isolated location of most fish farms reduces the potential for introduction of confounding signals or dilution of the “homing signal” from boat traffic and other sources. The salmon in the current report were likely cuing on particle displacement, as Hawkins and Johnstone (1978) determined that this was the primary mechanism for sound reception in salmon. If audition in salmon is solely dependent on particle displacement cues, the effective range of this approach for recalling salmon would be limited. However, for trout this might not be as much of a constraint as trout have been observed to remain in close proximity to pens in estuarine fjords following escape (Bridger and Garber, 2002). By way of comparison, Atlantic salmon in the high the energy environment of the Bay of Fundy were found to disperse from the cage site shortly after escape (Whoriskey et al., 2006). These findings suggest that local environmental conditions may influence the efficacy of this approach and will have to be accounted for in the development and implantation of a recall and recapture system.

The trout and salmon in the current investigations did not discriminate among the signal frequencies and intensities examined. Fish conditioned to a 250 Hz signal responded equally to a novel 100 Hz tone and those conditioned to a 300 Hz tone responded to a 100 Hz tone they were previously exposed to without feed reinforcement. Similarly, there was no differential response to tones with signal intensities ranging from 126 – 147 dB. Given that both species were able to detect frequencies from 50 Hz to 400 Hz it remains possible that a frequency differential of greater than 200 HZ is required to discriminate tones. Alternatively, the fish failed to discriminate between the two tones because any pronounced tone may have been associated with food. Longer periods of training might be required to condition the fish to discriminate between these tones. Dijkgraaf (1960) suggests that in some species of fish pitch discrimination can occur at 3/4 of a tone (9% difference). Other hearing generalists, such as the olive flounder (*Paralichthys olivaceus*), also failed to discriminate between tones. Flounder conditioned to a 300 Hz signal responded equally to tones between 200 and 800 Hz (Ankaru et al., 1998). In contrast, some commercially cultured species, such as cod, are hearing specialists and better able to discriminate tones as well as directionality (Buwalda et al., 1983, Schuijf and Hawkins 1983). The functional result of this absence of discrimination by salmon and trout to the signals used in the conditioning regimes employed here is that site specific acoustic conditioning and recall for farms may be limited. Farms located in close proximity may be unable to train fish to a specific tone that would enable recall of only their escapees. While this might reduce recovery by the actual producer, it would not decrease the effectiveness of conditioning for recapture and ultimately could decrease the environmental impact of escapees.

The success with the recall of salmon demonstrated here suggests that acoustic conditioning could be of value for use with other species of finfish cultured in marine net pens such as European sea bass (*Dicentrarchus labrax*) and Atlantic cod. Selective breeding of these species, a critical aspect for successful commercial aquaculture, alters the gene pool of cultured fish. As the number of species cultured in the marine environment continues to rise, the potential for escape and interaction with wild conspecifics or disruption of local ecosystems will increase (Youngson et al., 2001). Concurrent with this expansion, methods to recover fish and reduce impacts on wild stocks and economic losses to producers will become of greater importance.

5. Conclusions

Salmon and trout were readily conditioned to acoustic signals to return to a specific location and retained this conditioning for a period of up to 7 months. The findings suggest that acoustic conditioning could be used as a cost effective method for recapture of salmon that have escaped from marine farms, reducing the impact on wild stocks and economic loss to producers.

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