

EFFECTS OF HATCHERY STRESS
ON HEAT SHOCK PROTEIN INDUCTION
IN JUVENILE ATLANTIC SALMON, *SALMO SALAR*

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EXTENDED ABSTRACT ONLY – DO NOT CITE

The development of sensitive and rapid methods to identify stressful conditions in animal husbandry has been the goal of numerous studies. The expression of heat shock proteins (hsps) has been considered as a possible candidate as a stress indicator. To test this hypothesis, juvenile Atlantic salmon (*Salmo salar*) were exposed to common forms of hatchery stress and the response of hsps 90, 70 and 30 determined. Treatments included exposure to:

- 1) two types of anesthesia (tricaine methanesulfonate, 75 ppm and 2-phenoxyethanol 250 ppm; fish fully anesthetized for 10 minutes followed by a one hour recovery period)
- 2) formalin (250 ppm for two hours followed by a one hour recovery period)

- 3) hypoxia (fish maintained in a static tank without aeration until gasping at surface, followed by a one-hour recovery period)
- 4) hyperoxia (99% O₂ bubbled into static tank for 30 min (approximately 22 ppm dissolved oxygen) followed by a one hour recovery period)
- 5) capture stress (fish netted and allowed to struggle in net out of water for 20 seconds, returned to the water for 20 seconds, repeated three times followed by a one-hour recovery period)
- 6) crowding, (15 fish (mean weight 43 g) in 20 L of water for 3 hours followed by a one hour recovery period)
- 7) starvation (fish fasted for one week)
- 8) cold shock (two hours at 4°C, followed by a one hour recovery period at ambient temperature 19°C)
- 9) Positive control - 15-minute heat shock at 26°C ($\Delta T=10^{\circ}\text{C}$), followed by a one hour recovery period
- 10) Negative control – unhandled fish.

Total RNA was isolated from gill tissue following treatment, and subjected to Northern analysis with cDNA probes generated to hsp90, 70 and 30 by polymerase chain reaction (PCR) or cloning. Assay of mRNA for actin, a prominent constitutive protein, was used to normalize hsp mRNA. Hsp mRNA was not upregulated in response to the different hatchery stresses, whereas heat shock resulted in upregulation of hsp90 (Fig. 1) and 70. Although hsp90 mRNA levels were unaffected by the heat shock treatment used in this experiment, a subsequent investigation revealed that a 30 minute exposure to 26°C water stimulated upregulation. These data suggest that hsp90, 70 and 30 are not sensitive indicators of hatchery stress in Atlantic salmon.

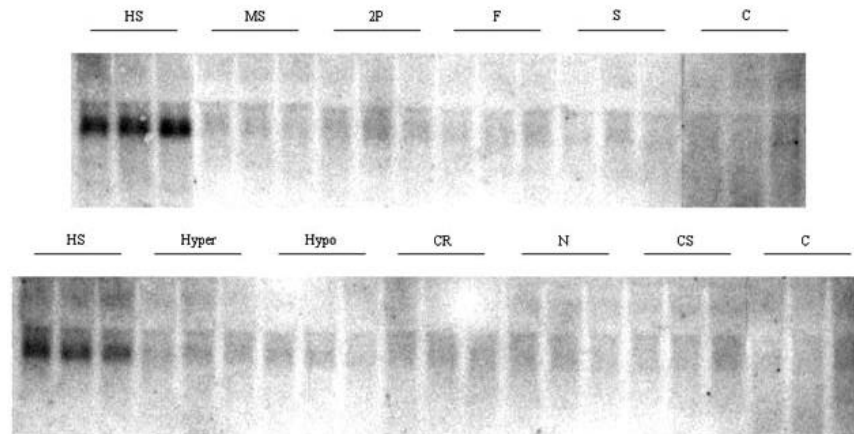


Figure 1. Northern blot of gill tissues from juvenile Atlantic salmon exposed to control and stress conditions as described in text and probed with α - ^{32}P dATP labeled hsp30 cDNA. HS = heat shock, MS = tricaine methanesulfonate, 2P = 2 -phenoxyethanol, F = formalin, S = starvation, C = negative control, Hyper = hyperoxia, Hypo = hypoxia, CR = crowding, N = capture stress, CS = cold shock.

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