Problem Formulation for the Environmental Risk Assessment of GE Growth-enhanced Coho Salmon

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Regulation of Genetically Engineered Fish in Canada

Environmental Risk Assessment – Fisheries and Oceans Canada (DFO)

- Risk $\propto$ Exposure x Hazard, including measurement of uncertainty
- Peer reviewed and published

Science Advice to Environment and Climate Change Canada and Health Canada
- Regulatory decision on use of GE fish under CEPA NSNR(O)

Risk Assessment begins with Problem Formulation

- Defines scope of risk assessment and protection goals
- Provides description of procedures for concluding on risk
- Provides a characterization of the organism and comparator species
- Provides a characterization of ecological parameters for the receiving environment

Regulated under Canadian Environmental Protection Act (CEPA)
New Substances Notification Regulations (Organisms) [NSNR(O)]
GE Fish Approved for Commercial Use in Canada

- **AquAdvantage™ salmon**
  - Atlantic salmon containing a growth hormone transgene (GH) to produce faster growth for land-based aquaculture
  - Currently under assessment for expansion

Currently under assessment:

- **GloFish® Electric Green® Tetra**
  - White tetra containing a Cnidarian green fluorescent protein transgene to produce a new colour morph for the ornamental aquarium trade

- **GloFish® Tetra Lines**
  - White tetra containing different coloured Cnidarian fluorescent protein transgenes to produce new colour morphs for the ornamental aquarium trade
TEST CASE: ERA of GH Transgenic Coho Salmon

- Research line containing a sockeye growth hormone (GH) transgene driven by a metallothionin promoter for fast growth
- Developed to examine impacts of accelerated growth in salmonids (not for commercial use)
- Physiology and ecological impacts well studied

Use scenario:
- Theoretical land-based aquaculture of triploid (sterile) GH transgenic coho salmon for domestic food market + diploid (fertile) broodstock
- Located in Burrard inlet, BC, Canada within natural marine and freshwater ranges of wild stocks of coho salmon

- Risk protection goals focus on impacts to wild coho salmon through hybridization
- Also examine: impacts through trophic interactions, environmental toxicity, horizontal gene transfer, as a vector of disease, and to habitat, biogeochemical cycling, and biodiversity
Protection Goal: Maintenance of Wild Coho Salmon Stocks

Coho salmon:
- Anadromous: spawn in rivers, juveniles live in rivers or lakes
- Migrate to Pacific Ocean at 1.5 y
- Return to home river to spawn and die at 3 y of age
- Home to specific rivers to spawn: many genetically and ecologically distinct populations

Ecological Importance:
- Important in marine and freshwater food webs as both predator and prey
- Massing spawn die-off plays a large role in riparian ecosystem nutrient cycling and biodiversity
Protection Goal: Wild Coho Salmon Stocks

Socioeconomic Importance:

• CND$ 2.5-4 million landed in commercial fisheries yearly
• Contribute to CND$ 8 billion recreational fisheries
• Cultural importance to Indigenous communities

Current threats to wild coho salmon populations

• Threatened by overfishing, climate change, habitat disruption, invasive species, competition with hatchery fish (COSEWIC 2016)

Could GH transgenic coho salmon further threaten wild coho populations?
Characterization of GH Transgenic Coho Salmon

- **TAREGETED PHENOTYPE**: Fast growth and improved feed conversion efficiency

- **OFF-TARGET EFFECTS**: results in pleiotropic unintended phenotypes

- Alterations in every major characteristic examined (Devlin et al. 2015), e.g.:
  - life history timing (e.g. mature at 2 versus 3 years of age)
  - gene expression
  - disease resistance
  - behaviour

➢ Potential effects from targeted as well as off-target effects must be considered in ERA
Genotype x Environment Interactions: Uncertainty in Using Laboratory Data to Predict Effects in Nature

- Transgenic and wild-type coho salmon respond to different environments (e.g., hatchery versus semi-natural) very differently.
- Strong genotype x environment interactions (GxE) make predictions of success in nature using laboratory data problematic with high uncertainty.
- Identifies the importance on not relying solely on data from culture environment.

Small-scale culture: 4000L

Mesocosm: 350,000L

Nature: \( \approx 7.14 \times 10^{14} \text{L} \)
Pathway to Harm: Hybridization with Wild Populations

- What steps would need to be completed for GH transgenic salmon to harm wild populations?

- Example Scenario: Pathway to harm through hybridization with wild populations:

  - Release of immature GE coho
  - Survival to maturation
  - Migrate to spawning grounds
  - Successful mating wild coho
  - F1 hybrids
  - Poor survival of hybrids
  - ↓ wild coho productivity
  - Introduce GH transgene to wild pop
  - Altered genetic structure of wild pop

- What scientific research is needed/available to address each step?

Modified from Leggatt et al. 2010
Pathway to Harm: Hybridization with Wild Populations

- Survival in culture is generally much higher than in nature
- Studies in semi-natural conditions indicate:
  - Survival will likely depend on time of release, disease status of released fish, availability of food, presence of predators, etc.

**Survival is possible**, although expected to vary relative to wild populations depending on conditions.
Pathway to Harm: Hybridization with Wild Populations

- Release of immature GE coho → Survival to maturation → Migrate to spawning grounds → Successful mating wild coho → F1 hybrids → Poor survival of hybrids → ↓ wild coho productivity
- Release of mature GE coho

- Has not been examined in GH transgenic coho salmon
- 3N females not expected to migrate
- Domesticated Atlantic salmon as surrogate organism: while many farmed escapes do not return to freshwater, some do (see Glover et al. 2017)
- Aquaculture site is within migration range of many wild coho populations
- Cannot exclude the potential for escaped 2N GH transgenic salmon to migrate to spawning grounds

- Migration is predicted, but not empirically tested
Early studies found laboratory-reared GH transgenic salmon had very poor ability to reproduce with nature-reared wild salmon.

Wild-type laboratory-reared also had poor spawning success: Large culture effect.

Rearing in seawater mesocosms decreased impacts of culture, and under these conditions GH transgenic coho salmon had similar reproductive success as wild type.

 успех скрещивания с дикой кохо предсказывается, уровень может зависеть от времени выпуска, уровня конкуренции при спаривании.

2N broodstock and failed triploid fish only
Pathway to Harm: Hybridization with Wild Populations

- GH transgene may decrease survival in some conditions, increase it in others – depend on availability of prey, presence of predators, level of competition, etc.
- Modeling of transgene introgression found the GH transgene in a wild population could potentially shift genetic backgrounds in transgenic and non-transgenic individuals, which could direct phenotypes away from naturally selected optima (Ahrens and Devlin 2011)
- QTL mapping demonstrated the GH transgene alters genetic basis of growth-related traits in coho salmon: postulated to influence evolutionary changes with coho salmon with ecological consequences (Kodama et al 2018)

- Poor survival of hybrids in some conditions predicted
- Altered genetic structure of wild populations predicted
Pathway to Harm: Hybridization with Wild Populations

- All steps post-release have been corroborated to some extent through laboratory studies or use of surrogate organism

- Potential harm to wild populations through hybridization with GH coho salmon is predicted should GE coho salmon escape

- Predicted level of success/harm of GH transgenic fish differs when examined under laboratory versus semi-natural conditions
  - Importance of simulating relevant natural conditions when examining environmental risk

- Genotype x Environment interactions and limited studies in some steps
  - Moderate Uncertainty in predicting occurrence/level of harm in nature
Other Pathways to Harm to Wild Fish Populations

- Release of GH transgenic (GE) coho salmon
  - Competition with wild fish for food
  - Predation on wild fish
  - Release of infected GH salmon, transfer of disease
  - Hybridization with wild fish
  - Reproductive interference with wild fish

- Altered mass spawning mortality in wild fish
- Change in fish habitat
- Change in food availability
- Change in wild fish populations/communities
- Change in wild fish health

- Hybridization is only one of several interacting pathways to harm to wild fish populations should GE coho salmon escape aquaculture facilities
- Importance of Containment to prevent harm to environmental components from GE GH transgenic coho salmon

Modified from Leggatt et al. 2010
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References


