

# Retrospective Assessment of the Direct Genetic Impacts of Escaped Farm Salmon on a Wild Scottish Stock

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## Background

The potential for direct or indirect impacts on wild Atlantic salmon populations from conspecific farm escapes is well known<sup>1</sup>

The Loch na Thull (LnT) catchment is a small river system in northwest Scotland with an anecdotal historical record of a small stock of wild Atlantic salmon, believed to spawn predominantly in the outflow burn.

Cages for rearing farmed salmon to the smolt stage, prior to their transfer to marine cages, were located in the Loch from 1997 to 2004.

The analysis of scales from salmon smolts caught in the Loch in 1998, 1999, 2000 and 2004 showed farm escapes to be present

Local fisheries managers wanted to know if -

- the Loch na Thull system contains wild native salmon populations?
- if the Loch na Thull stock is a physical or reproductive mixture of wild native and farmed fish, or strays from neighbouring rivers?

## Methodology

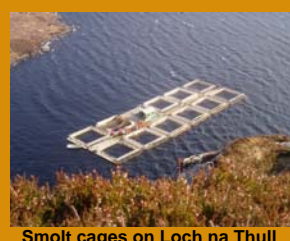
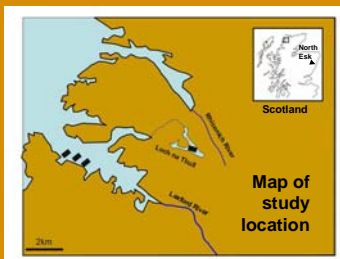
A retrospective analysis of direct genetic impact was carried out using a limited historical collection of scales along with some contemporary collections of fin tissue as well as relevant existing molecular genetic data on regional farm and wild salmon stocks

The samples were screened for genetic variation for a standard random set of molecular markers to assess the extent of genetic relatedness of the samples and draw inferences about the farm or wild ancestry of individual fish. The primary analysis encompassed 291 fish:

- adult salmon caught in the system in 2004 and 2005,
- fry electro-fished in the Loch na Thull outflow burn in 2006,
- parr collected in 2006 in Loch na Thull
- escaped farm smolts from Loch.
- fry and parr from the neighbouring Rhiconich and Laxford rivers

Existing data from other studies came from samples of

- farm salmon reared in cages in Loch na Thull in 1997 and 1998
- Norwegian Mowi salmon from 1997, 1998 and 2006
- Juvenile salmon from the main Rhiconich river collected in 2005
- juvenile salmon from the North Esk river system



Smolt cages on Loch na Thull

DNA was extracted using standard methods<sup>2</sup> and, where required, further purified using Illustra DNA purification kits (GE Healthcare). Genetic screening for microsatellite and mitochondrial DNA variation was carried out using polymerase chain reaction (PCR) methods<sup>3</sup>.

Microsatellite variation was typed for 14 microsatellite loci - *SSsp2210*, *SSspG7*, *SSsp2201*, *SSsp1605*, *SSsp2216*, *SSsp3016*, *SsaD144*, *SsaD486*, *Ssa202*, *Ssa171*, *Ssa289*, *Ssa14*, *Ssa197*, and *SsaF43*. Existing data for four - *SSsp1605*, *SSsp2201*, *SSsp2210* and *SSsp2216* - were available for the additional samples included in the analysis.

Mitochondrial variation was characterised in the ND1 gene region using the restriction enzymes *DraI*, *HaeIII*, *HinfI*, *RsaI* by standard methods<sup>4</sup> or methods for degraded DNA<sup>2</sup>.

## ABSTRACT

The potential exists for negative impacts on wild Atlantic salmon populations (*Salmo salar*) from interbreeding with escaped farm fish but its actual extent remains uncertain and controversial. Information on numbers of escapes and gene introgression is generally lacking, in part due to a lack of historical baseline information. A molecular genetic study was carried out of impacts in the Loch na Thull catchment, a small river system in northwest Scotland containing a small wild stock and used for freshwater cage rearing of farm smolts from 1997-2004 with a documented presence of escapes. The study was retrospective and based on an eclectic set of DNA sources and genetic data that precluded a simple before and after comparison. The study found significant genetic differences between wild and farm salmon and that farm salmon appeared to contribute little to the genetic character of the current wild stock through interbreeding and introgression. The study does not rule out that ecological interactions with farm escapes such as competition for food and space or increased pathogen transfers, may have led to indirect genetic impacts (e.g. loss of genetic variability), due to increased mortality and smaller numbers of returning spawners.

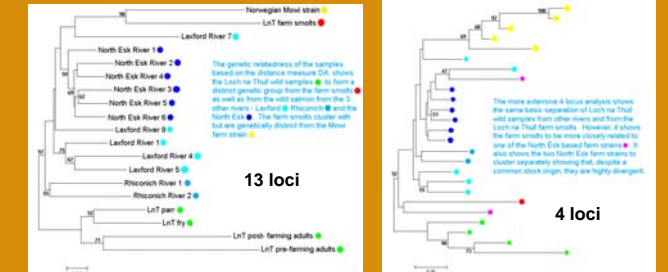
## Results

### Microsatellite Analysis

- no significant deviation from Hardy-Weinberg within samples, or linkage disequilibrium except within wild LnT parr sample
- heterozygosity and allelic diversity lower in LnT samples with latter proportionately lower suggesting population bottlenecked
- general genetic heterogeneity except among wild samples within the LnT and Laxford catchments
- based on Nei's genetic distance, separate clustering with strong bootstrap support of wild samples from each catchment distinct from farm samples, for both 14 locus and 4 locus analyses
- higher level of genetic divergence among LnT samples than among samples in other catchments
- self-assignment showed >95% of salmon correctly identified to sample of origin with a  $p > 0.5$ ;
- in LnT samples, one fish was indicated to be of farm origin and one from the River Laxford; no mixed origin fish were identified.
- LnT samples show lowest estimated effective breeding sizes

### Mitochondrial Analysis

- overall heterogeneity of haplotype frequencies among samples
- significant differences among catchments
- significant differences between LnT wild and farm smolt samples
- clustering of samples into catchment and farm groups is less marked and robust than in microsatellite analysis
- higher divergence among LnT samples compared to other rivers



## Conclusions

Despite the absence of good pre-farming samples of wild fish, the results obtained are informative. The results make it possible to conclude that:

- the Loch na Thull catchment retains a distinct wild salmon population
- offspring of farm stock, or wild spawners straying from the neighbouring Laxford or Rhiconich systems, represent only a small proportion of the contemporary stock.
- no substantive genetic impact from farm escapes was detected
- genetic impacts from ecological interactions (e.g. competition for food and space or increased pathogen transfers) with farm escapes in Loch na Thull causing an overall population decline can not be ruled out.

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### References

1. Ferguson A., Fleming I. A., Hindar K., Skaala Ø., McGinnity P., Cross T. and Prodahl, P. (2007). Farm escapes, Chapter 7, in: "The Atlantic salmon: genetics, conservation and management". Verspoor E., Stradmeyer L. and Nielsen J.L. (eds). Blackwell Publishing, Oxford, pp. 357-398.
2. Knox, D.K. Lehmann, D.G. Reddin & E. Verspoor. (2002). Genotyping of archival Atlantic salmon scales from northern Quebec & West Greenland using novel PCR primers for degraded DNA. Journal of Fish Biology 60: 266-270.
3. Hansen, M.M., Villanueva, Nielsen, E.E. and Bekkevold, D. (2007). Farm escapes, Chapter 4, in: "The Atlantic salmon: genetics, conservation and management". Verspoor E., Stradmeyer L. and Nielsen J.L. (eds). Blackwell Publishing, Oxford, pp. 86-114.
4. Verspoor, E., McCarthy, E., Knox, D., Bourke, E. & Cross, T.F. (1999). Phylogeography of European Atlantic salmon (*Salmo salar* L.) based on variation in the ND1/16S rRNA region of the mtDNA. Biological Journal of the Linnean Society 68: 129-146.