GRAASP:  a genetic tool for advancing understanding of the marine ecology of Atlantic salmon *Salmo salar* in the Eastern Atlantic Ocean

Eric Verspoor   Phil McGinnity
Genetically-based
Regional Assignment of
Atlantic Salmon Protocol

IASRB sponsored
EU funded

Contributing Consortium Partners

- Marine Scotland, Freshwater Laboratory, Pitlochry, Scotland
- Department of Zoology, University College, Cork, Ireland,
- Norwegian Institute for Nature Research Norway
- GENINDEXE, La Rochelle, France,
- Agri-Food and Biosciences Institute of Northern Ireland,
  Fisheries and Aquatic Ecosystems Branch, Belfast, UK
- School of Biosciences, University of Exeter, UK
- Departament of Functional Biology and Genetics, University of
  Oviedo, Spain
- National Institute of Aquatic Resources, Technical University of
  Denmark
- Icelandic Fisheries Laboratories, Reykjavik, Iceland
- Department of Biology, University of Turku, Finland
- School of Biological Sciences, Queens University, Belfast, UK,
- Institute of Marine Research, Bergen, Norway,
Genetically-based Regional Assignment of Atlantic Salmon Protocol

GRAASP Talk Outline

- Background
- Development
- Application
- Enhancement

Genetically-based Regional Assignment of Atlantic Salmon Protocol
Background

Marine Mortality

Recent Declines in Adult Returns

Friedland et al 2010
North Atlantic
(~50,000,000 km²)

~0.1 to 1 post-smolt for every 1 km sq

~2300 rivers
~5-10 million smolts

Background

Stock specific Migratory Routes and feeding areas
GRAASP Background

**Population Differentiation**

- 2300 rivers, \(? \times 2300\) populations
- Extensive, fragmented spawning distribution
- Homing
- 1000s of years of reproductive isolation
- To varying degrees for some but not all genome characteristics and locations

**Genome**

- 7-8 billion base pairs distributed across 56-58 chromosomes plus mitochondrial DNA

**Challenge:** to find the genome locations with the greatest population differentiation that can be exploited as markers of natal origin
For brown trout *Salmo trutta* it has been estimated that, on average, one variable microsatellite locus provides the same discriminatory power as eight allozyme loci (Estoup et al. 1998).
Microsatellite-based Assignment

**Background**

Assignment of Atlantic salmon from 2002 West Greenland fishery to continent of origin

<table>
<thead>
<tr>
<th>Molecular Marker</th>
<th>Resolving Power</th>
<th>Relative Problems</th>
</tr>
</thead>
<tbody>
<tr>
<td>Proteins</td>
<td>99.9%</td>
<td>Need for fresh or fresh frozen tissue and to kill fish; need 4-6 loci to be screened</td>
</tr>
<tr>
<td>mtDNA</td>
<td>~95%</td>
<td>Relatively high error rate</td>
</tr>
<tr>
<td>Satellite DNA</td>
<td>99.999%</td>
<td>None</td>
</tr>
</tbody>
</table>

West Greenland: to North American and European origin

1 microsatellite+ 1 mtDNA RFLP 99+% assignment (Gilbey et al. 2005)

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</thead>
<tbody>
<tr>
<td>Proteins</td>
<td>~50% in most cases</td>
<td>Need for fresh or fresh frozen tissue and to kill fish; need 4-6 loci to be screened; poor resolution</td>
</tr>
<tr>
<td>mtDNA</td>
<td>~50% in most cases</td>
<td>Relatively high error rate</td>
</tr>
<tr>
<td>Satellite DNA</td>
<td>&gt;80% where studied</td>
<td>Need to identify best loci and to use large numbers of loci</td>
</tr>
</tbody>
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North American to region or river of origin

European to region or river of origin


Proportions of Atlantic salmon stock groups in Finnish catches


Baltic Sea

Southern Europe

Ireland

National GSI (unpublished)
Selection of Microsatellite Loci

SALMAN Meeting Virginia

Evaluation of Irish and Scottish Data Set

Virginia Panel

SALSEA-Merge 14
GRAAASP Development

Development of Genetic Baseline

12 laboratories
12 political regions
~2300 salmon rivers

Design optimal sampling plan and extend existing data with respect to loci and rivers covered

Link completed data sets by inter-laboratory calibration

Develop computer data base, standardize data sets, enter data

Check and validate quality of data, sample sizes, eliminate poor quality samples

Integrating GRAAASP Baseline

26,813 fish from 466 locations, 284 rivers encompassing ~85% of wild salmon production

Table:

<table>
<thead>
<tr>
<th>Country</th>
<th>Rivers</th>
<th>Sample sites</th>
<th>Fish</th>
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</thead>
<tbody>
<tr>
<td>Denmark</td>
<td>2</td>
<td>3</td>
<td>180</td>
</tr>
<tr>
<td>England</td>
<td>22</td>
<td>33</td>
<td>1409</td>
</tr>
<tr>
<td>France</td>
<td>9</td>
<td>9</td>
<td>450</td>
</tr>
<tr>
<td>Iceland</td>
<td>16</td>
<td>22</td>
<td>1986</td>
</tr>
<tr>
<td>Ireland</td>
<td>29</td>
<td>40</td>
<td>2053</td>
</tr>
<tr>
<td>Northern Ireland</td>
<td>7</td>
<td>18</td>
<td>1302</td>
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<tr>
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<td>84</td>
<td>104</td>
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<td>Russia</td>
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<td>33</td>
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<td>71</td>
<td>187</td>
<td>8973</td>
</tr>
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<td>Spain</td>
<td>4</td>
<td>4</td>
<td>190</td>
</tr>
<tr>
<td>Sweden</td>
<td>4</td>
<td>4</td>
<td>172</td>
</tr>
<tr>
<td>Wales</td>
<td>6</td>
<td>9</td>
<td>347</td>
</tr>
<tr>
<td>Total</td>
<td>284</td>
<td>466</td>
<td>26813</td>
</tr>
</tbody>
</table>
GRAASP Development

Identification of assignment regions

- hierarchical assessment of relatedness of individuals within and among rivers using STRUCTURE

POSTER
J. Gibey et al. Geographical differentiation and structuring of European Atlantic salmon stocks at microsatellite DNA loci in relation to the regional assignment of marine fish.

3 highly divergent regional groups: North, South, Iceland

17 divergent regional groups: North (7), South (8), Iceland (2)
Selection of 1000 blind samples not in baseline from baseline rivers and from non-baseline rivers

Initial studies of marine samples

Application to the Study of Spatial and Temporal Distributional Heterogeneity of Regional Stocks

- detailed in the talk that follows by

J. Gilbey, J. Coughlan Marine distribution of regional Atlantic salmon post-smolt stocks in the NE Atlantic ascertained by microsatellite DNA based assignment.
GRAASP Enhancement

Resolving smaller regions, rivers and populations

- increased number of loci needed with decreased differentiation
- SNPs offer potentially cost-effective way of identifying larger numbers of high resolution markers

Allozymes: lack of inter-laboratory calibration – excludes some regionally restricted and potentially informative variation and too few loci to give power to assign individuals.

microsatellites

River specific signals
Identifying further markers for resolving smaller regions, rivers and populations

1. different or larger set of microsatellites

Väha, Primmer unpublished

2. Identification mtDNA SNP markers

43% mtDNA genome, 137 variants in study of 330 individuals from 29 rivers across Europe

E. Verspoor et al. Regional mtDNA SNP differentiation in European Atlantic salmon (Salmo salar): an assessment of potential utility for determination of natal-origin

3. Identification of nuclear SNP markers

CIGENE

…ACTTGGCAGTC...

…ACTTGGCAGTC...

POSTERS

J. Coughlan et al. Geographical structuring of European Atlantic salmon as revealed by nuclear SNPs: potential for application in the assignment of origin of marine fish.

J. Coughlan et al. A preliminary evaluation of use of nuclear SNPs for the assignment to origin of marine post-smolt Atlantic salmon captures in the NE Atlantic
GRAASP

SUMMARY

PROGRESS

- Genetic assignment tool for European salmon now exists
- Comprehensive capacity to assign a high proportion of fish to regional stock groups
- Ready to be applied to new and archival marine samples e.g. WG
- Provides basis for understanding stock specific movements, marine utilisation, behaviour, ecology, performance.
- Stock heterogeneity may mean response heterogeneity to climate change
- Potential for refinement to achieve higher resolution river stock or population assignment by extending baseline and refining marker set
- Increased understanding of diversity within European stock complex
  Baseline can be used to understand the evolution of different migratory behaviours, of post-glacial colonization, glacial refugia, etc
Contributing Individuals

John Gilbey, Jamie Coughlan, Paulo Prodohl, Vidar Wennevik