

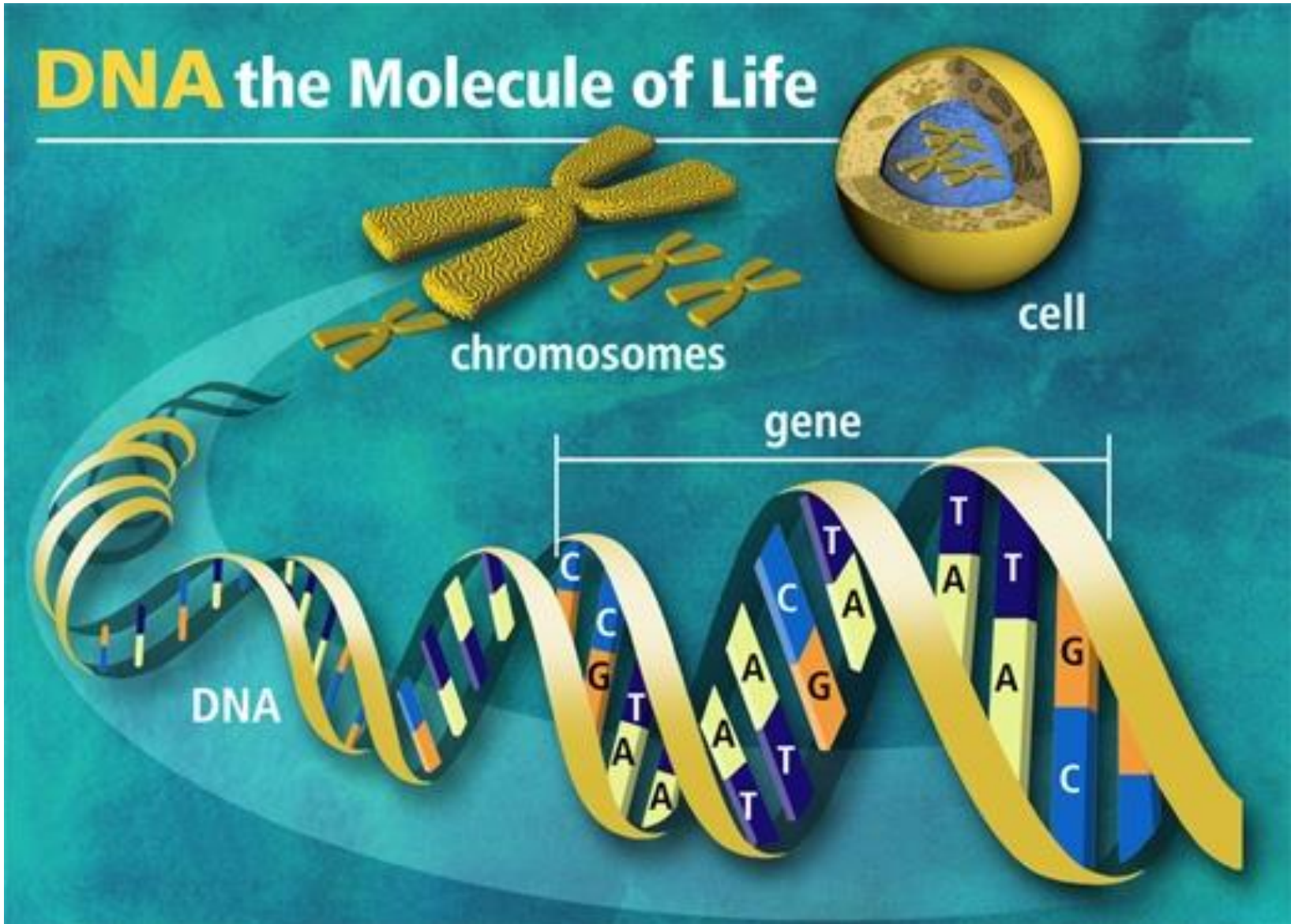


Genome sequencing and its practical application for fisheries management

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DTU Aqua
National Institute of Aquatic Resources





'Genetics' to 'Genomics'



10-100 DNA
markers

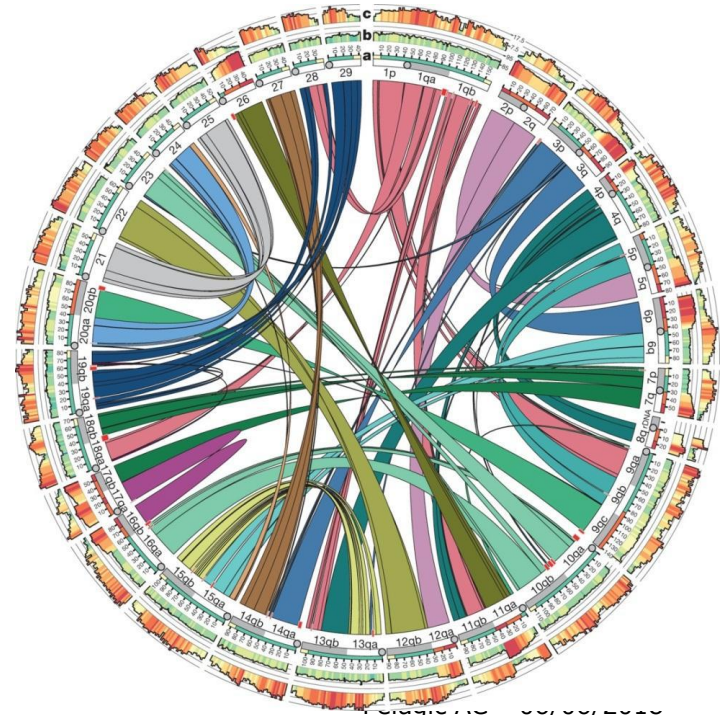
>>1000 DNA
markers

Application for fisheries management

- Define management units
- Quantify the extent of divergence and connectivity between stock units
- Allow mixed-stock analysis with substantially increased resolution relative to morphology based data
- Species identification in time and space (including prey species)

Genomic analysis example: Atlantic salmon

- Genome: 2.97 billion bases
 - ACTGCTGAAGGT.....
- 46.598 genes (37.206 coding for a specific protein)
- Links between genes and life histories



Example from Atlantic salmon

Size matters - sometimes

LETTER

doi:10.1038/nature16062

Sex-dependent dominance at a single locus maintains variation in age at maturity in salmon

Nicola J. Barson^{1*}, Tutku Aykanat^{2*}, Kjetil Hindar³, Matthew Baranski⁴, Geir H. Bolstad³, Peder Fiske³, Céleste Jacq⁴, Arne J. Jensen³, Susan E. Johnston⁵, Sten Karlsson³, Matthew Kent¹, Thomas Moen⁶, Eero Niemelä⁷, Torfinn Nome¹, Tor F. Næsje³, Panu Orell⁷, Atso Romakkaniemi⁷, Harald Sægrov⁸, Kurt Urdal⁸, Jaakko Erkinaro⁷, Sigbjørn Lien¹ & Craig R. Primmer²

Males and females share many traits that have a common genetic basis; however, selection on these traits often differs between the sexes, leading to sexual conflict^{1,2}. Under such sexual antagonism, theory predicts the evolution of genetic architectures that resolve this sexual conflict²⁻⁵. Yet, despite intense theoretical and empirical interest, the specific loci underlying sexually

known genetic architecture means that the evolutionary consequences of sexual conflict, particularly its importance in maintaining adaptive variation^{3,6,16}, remains largely unknown^{14,16}.

The age at which an individual reproduces is a critical point in its life history. Age at maturity affects fitness traits including survival, size at maturity and lifetime reproductive success¹⁷. Age at maturity

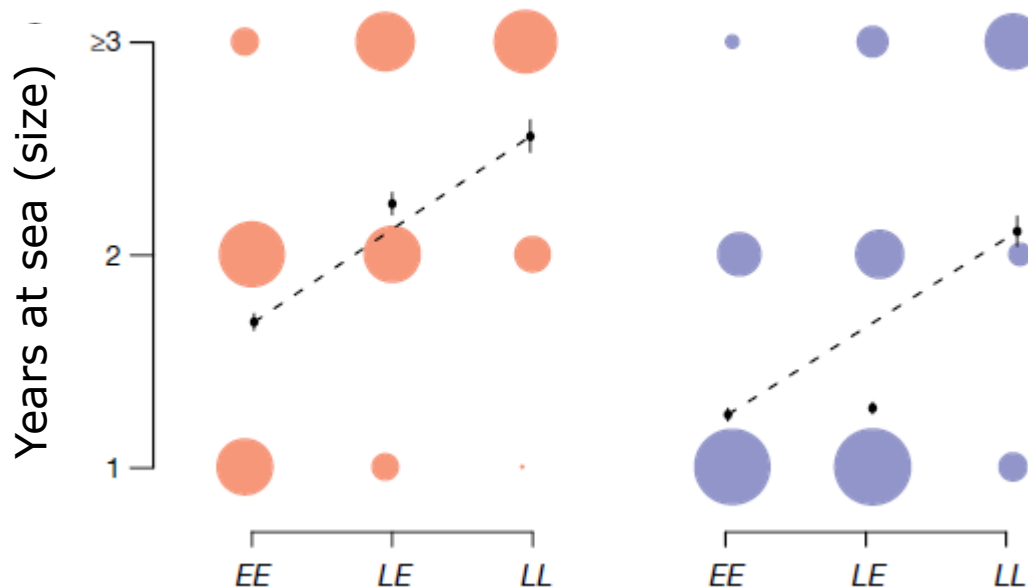
Genetics and size in Atlantic salmon

- A single gene (*VGLL3*) explains 39% of the variation in age at maturation (= size at river return)

- Gen
- Gen

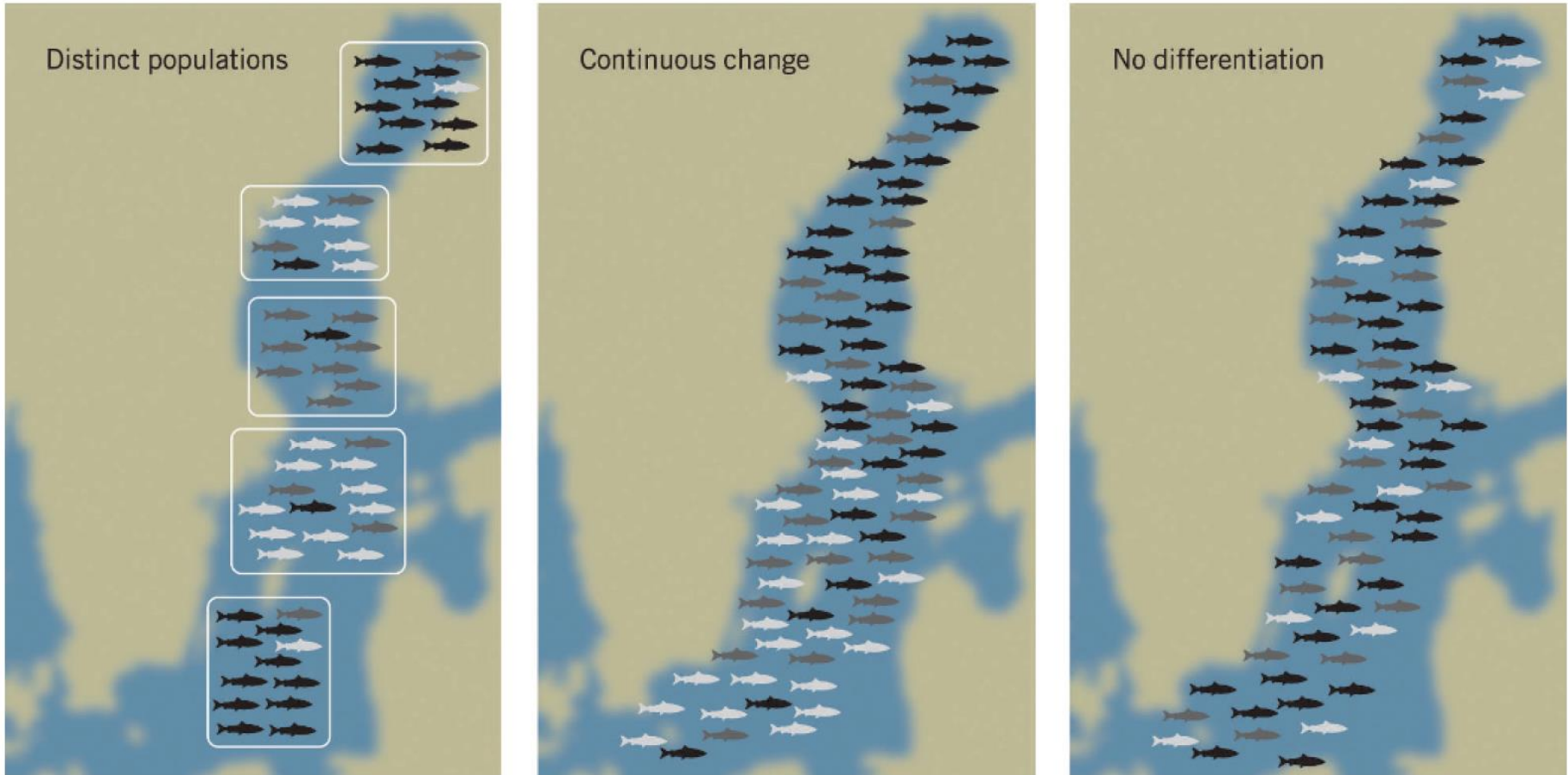
Implications for management and stocking programmes

Barson et al.
2015



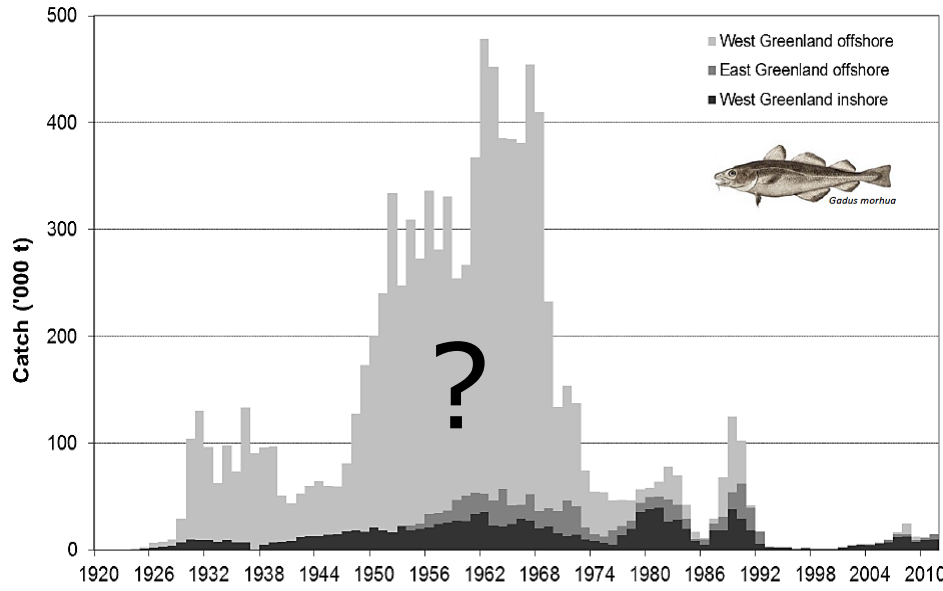
Types of population/stock structure – a theoretical example

Laikre et al. 2005

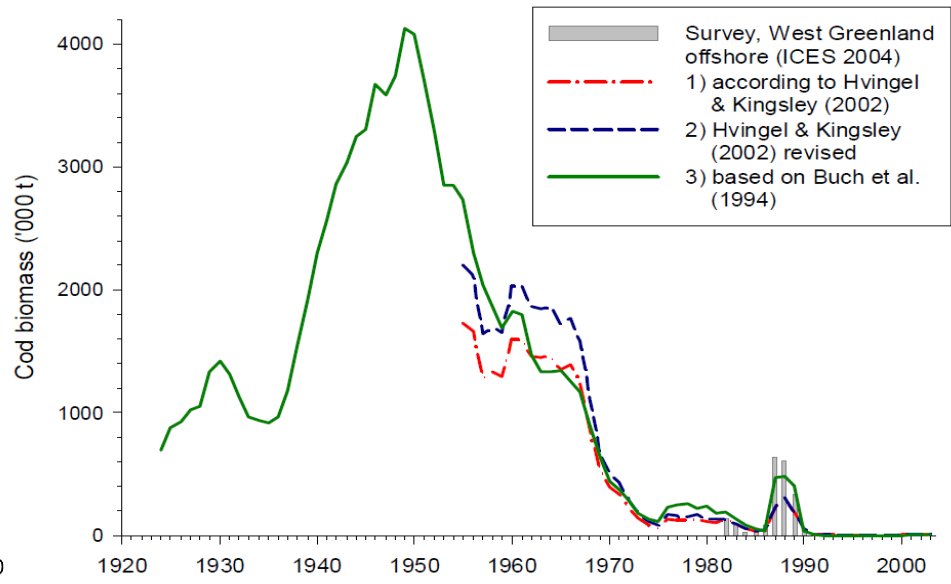


Genetic differences arise due to random and evolutionary processes

Greenland cod fishery



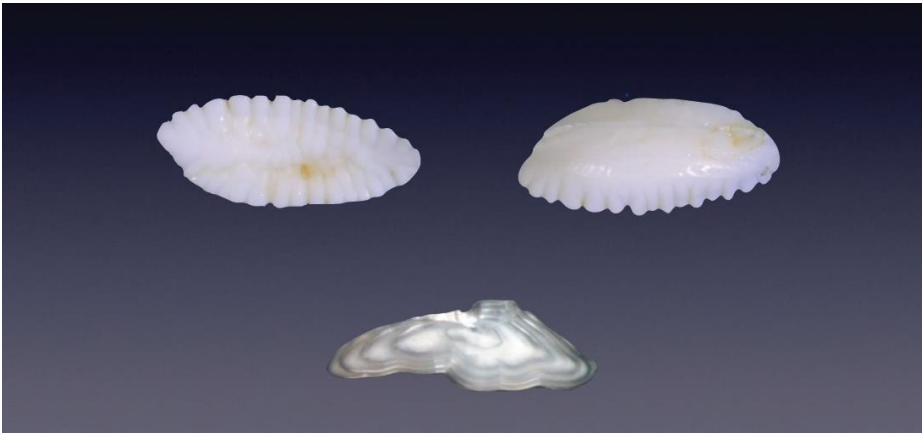
ICES Advice 2011



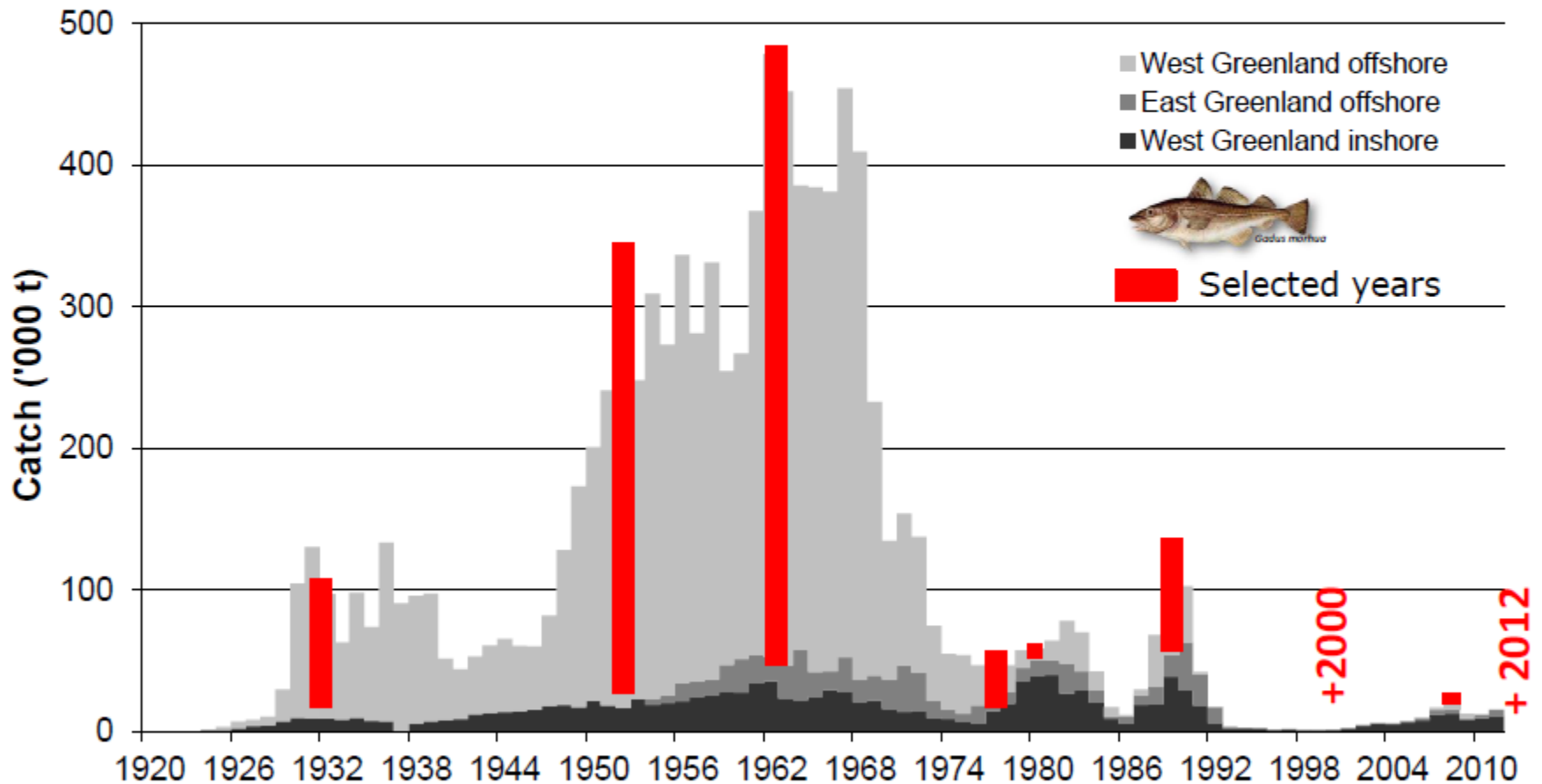
Wieland & Paulsen 2004



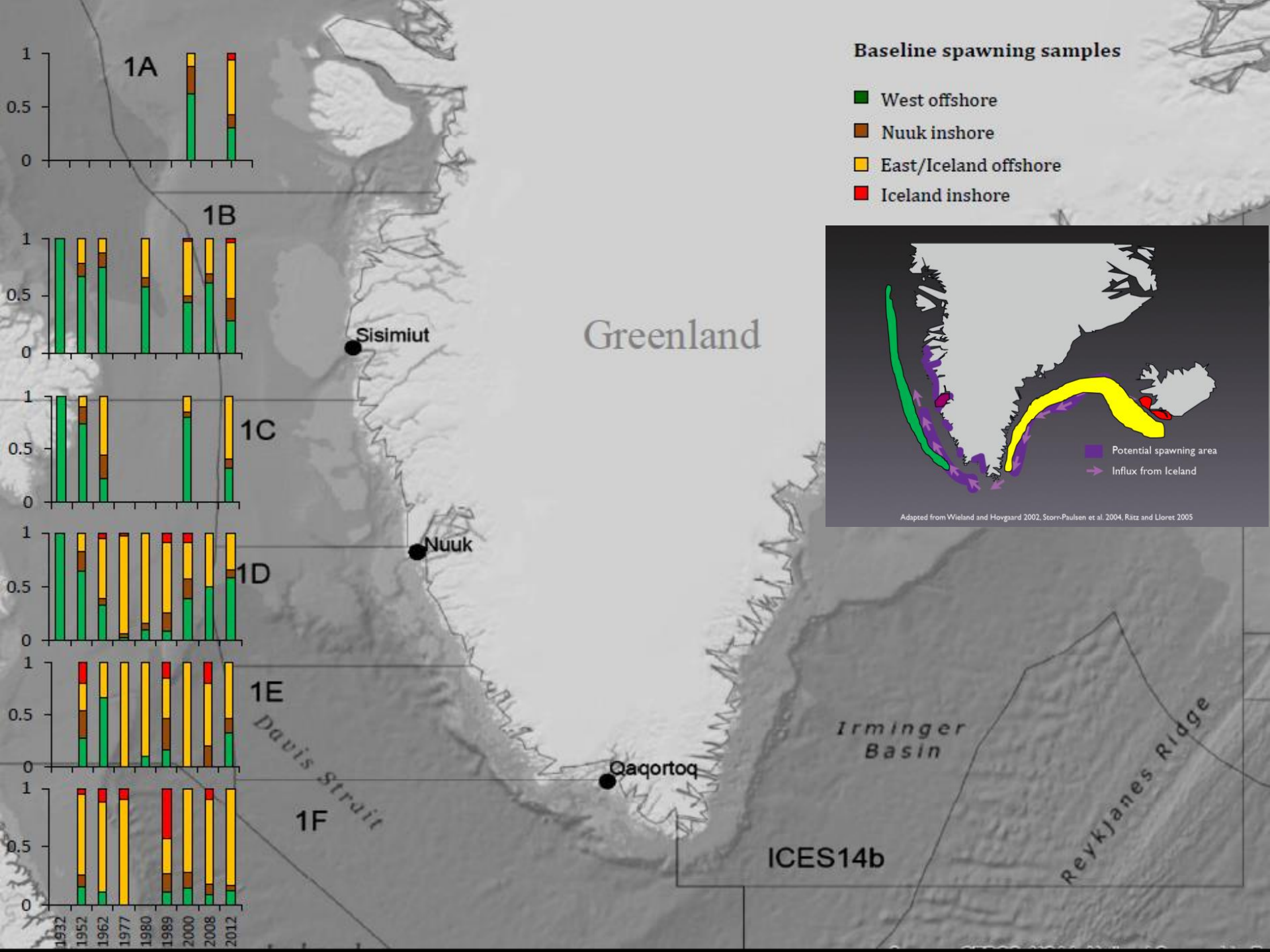
Sara Bonanomi



Historical commercial catches of Atlantic cod (*Gadus morhua*) in Greenlandic waters



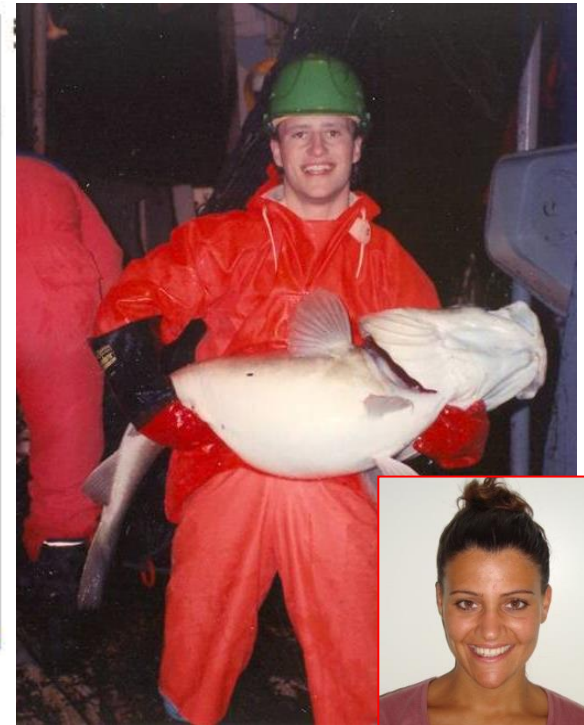
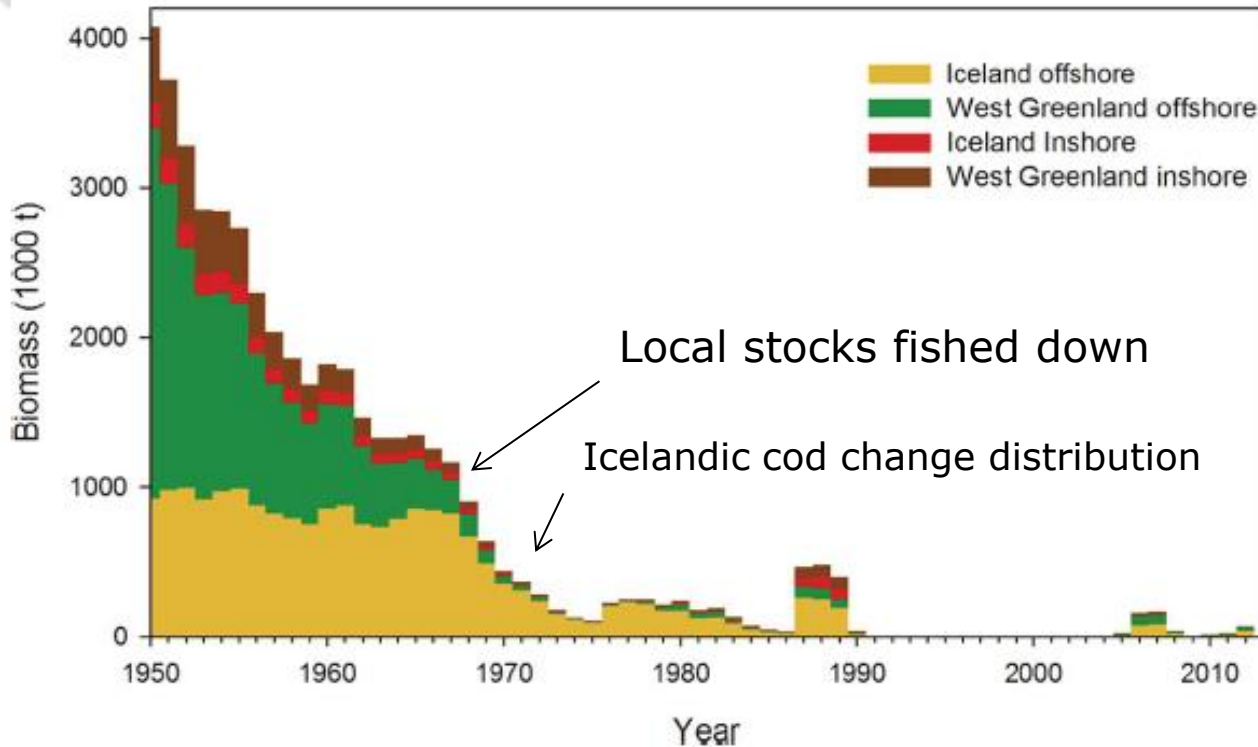
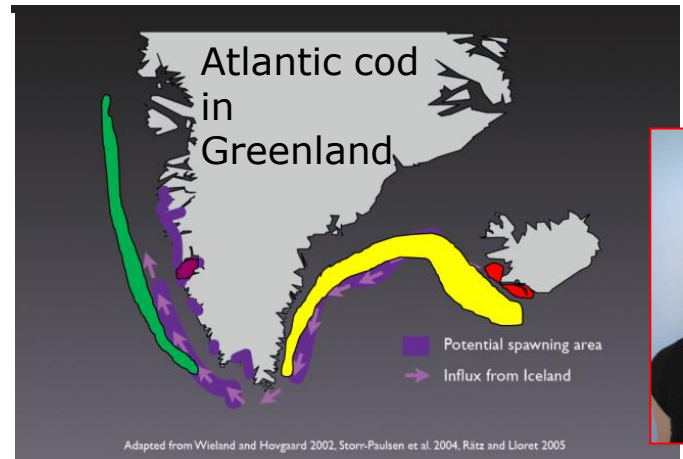
DNA from 100 archived otoliths for each year



OPEN Archived DNA reveals fisheries and climate induced collapse of a major fishery

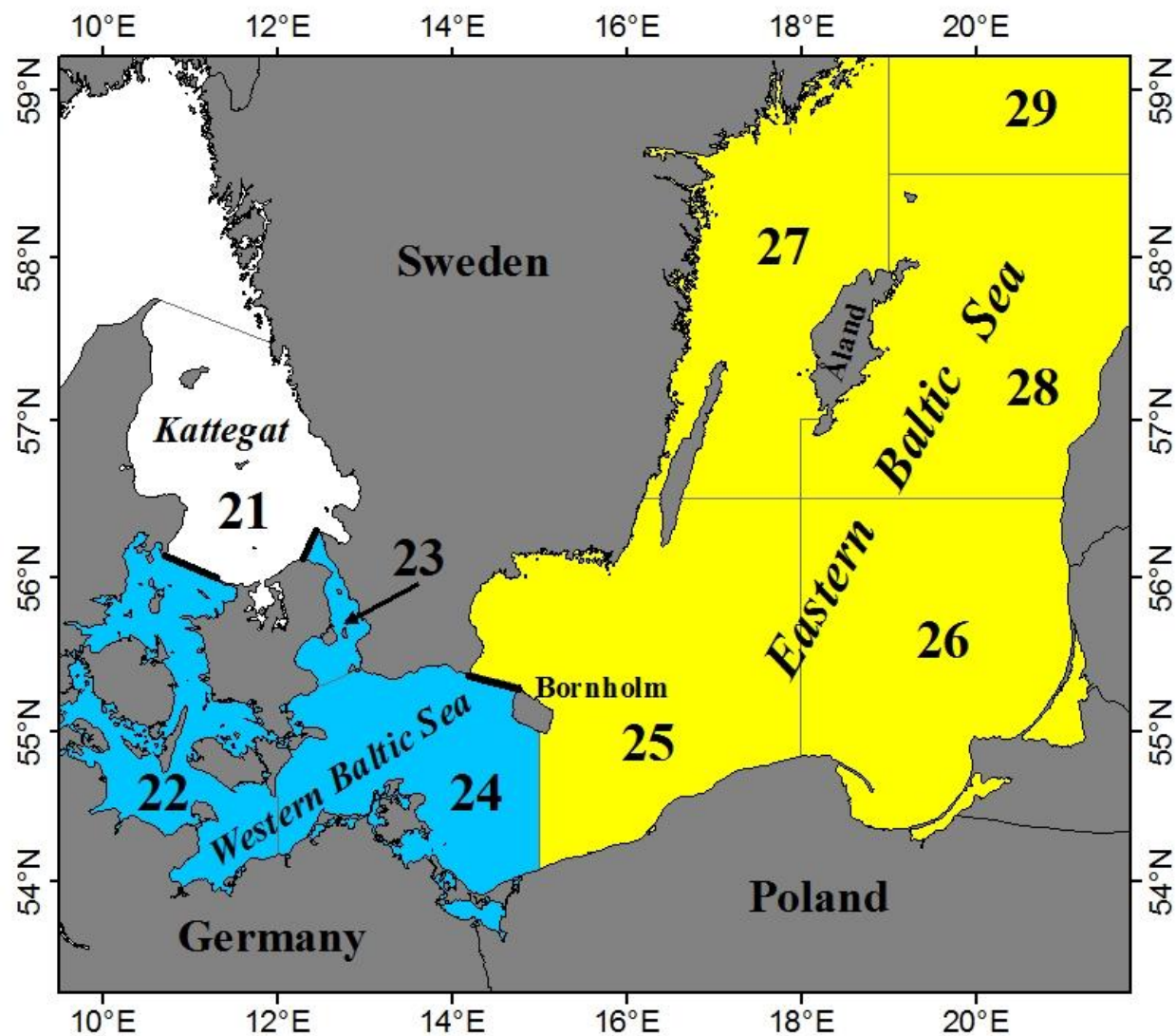
Sara Bonanomi^{1,2,†}, Loïc Pellissier^{3,†}, Nina Overgaard Therkildsen^{2,4}, Rasmus Berg Hedeholm^{2,5}, Anja Retzel^{2,5}, Dorte Meldrup², Steffen Malskær Olsen⁶, Anders Nielsen⁷, Christophe Pampoulie⁸, Jakob Hemmer-Hansen², Mary Susanne Wisz^{9,†}, Peter Grønkjær^{2,10,†}

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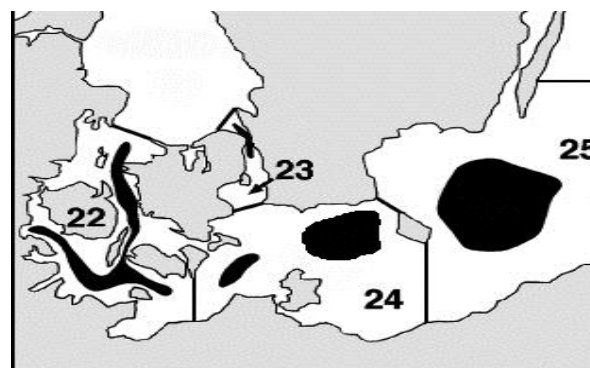


Cod example 2

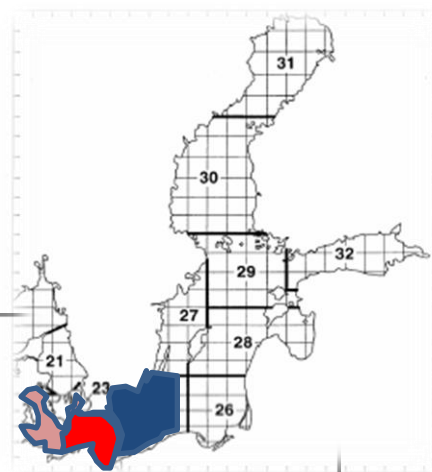
Atlantic cod in the Baltic Sea



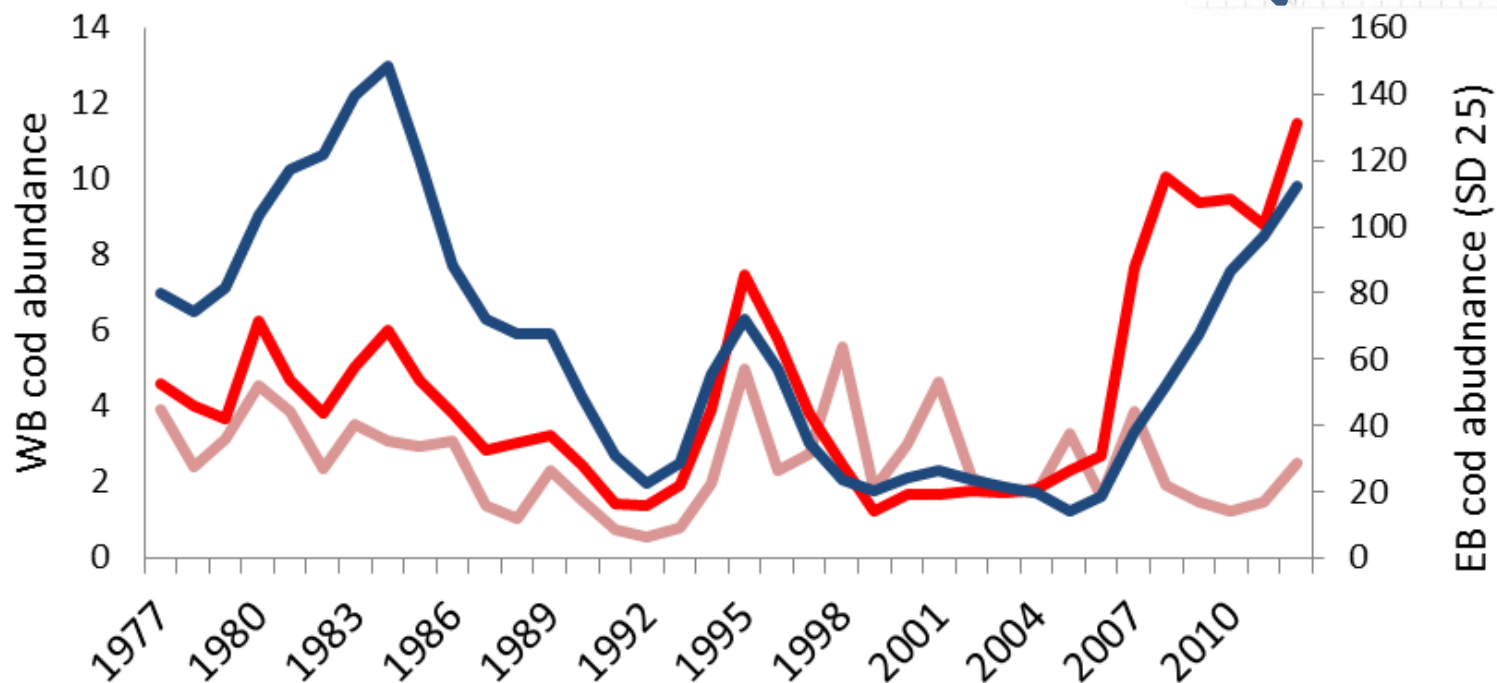
Stock assessment until 2015



Atlantic cod in the Baltic Sea



Trends in WB compared to EB

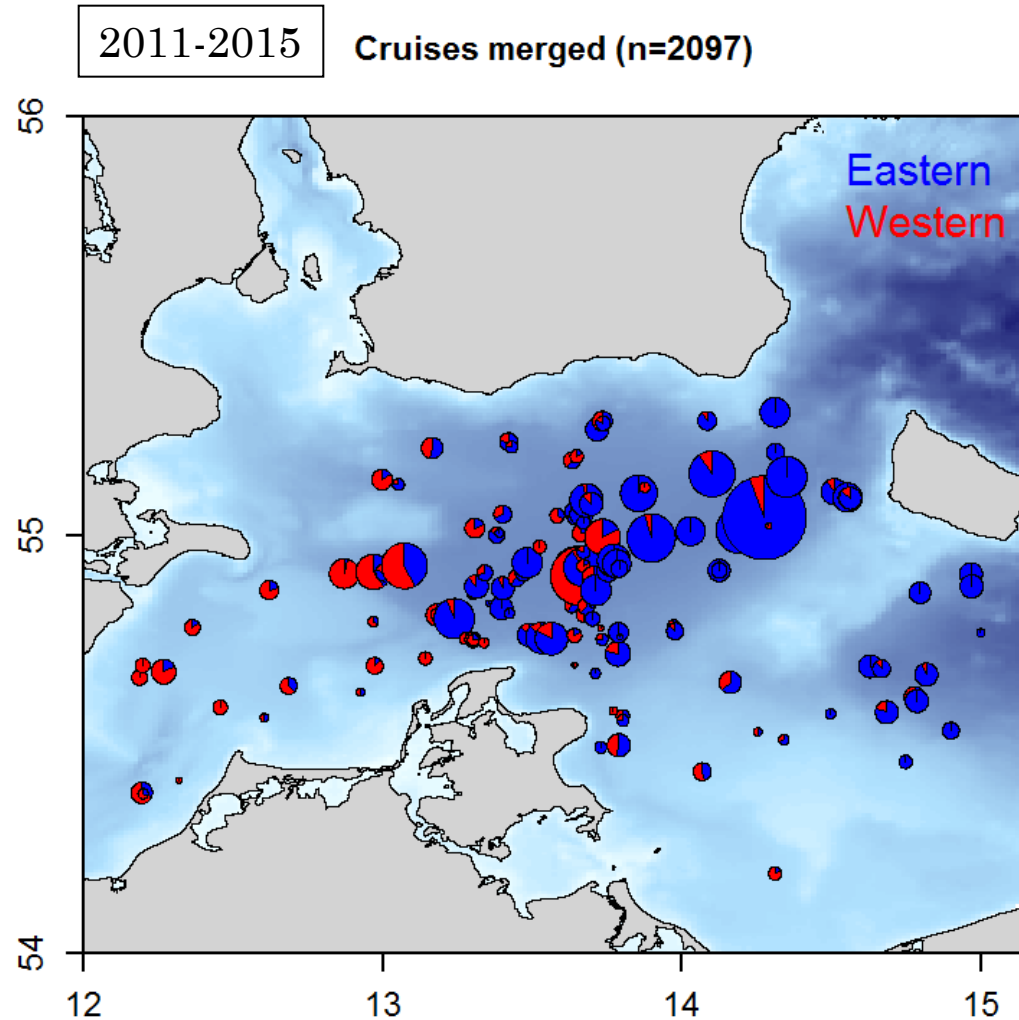


Atlantic cod in the Baltic Sea

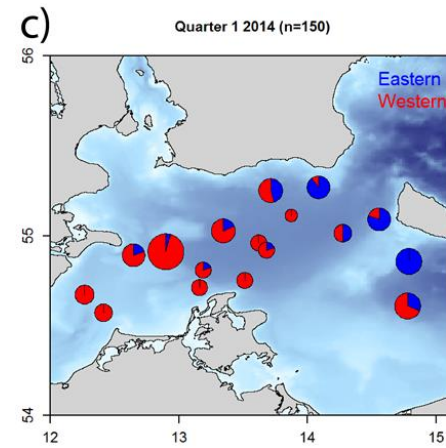
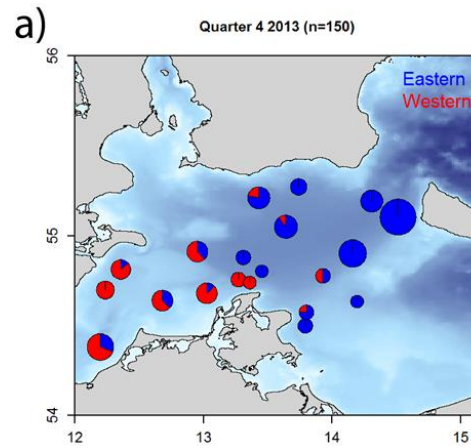
Scientific cruises sample mixed stocks



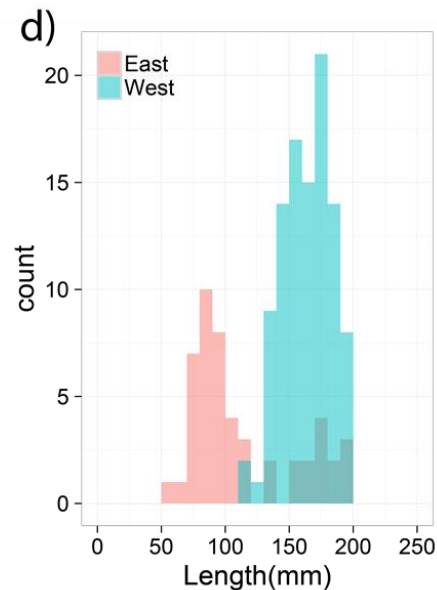
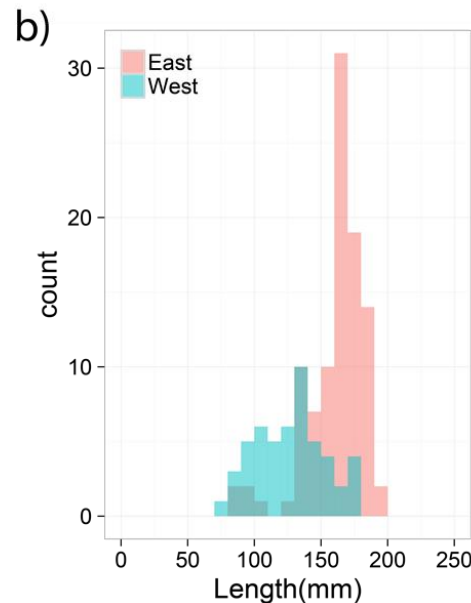
Genetically determined stock affiliation as either 'Eastern' (blue) or 'Western' (red)
Baltic cod



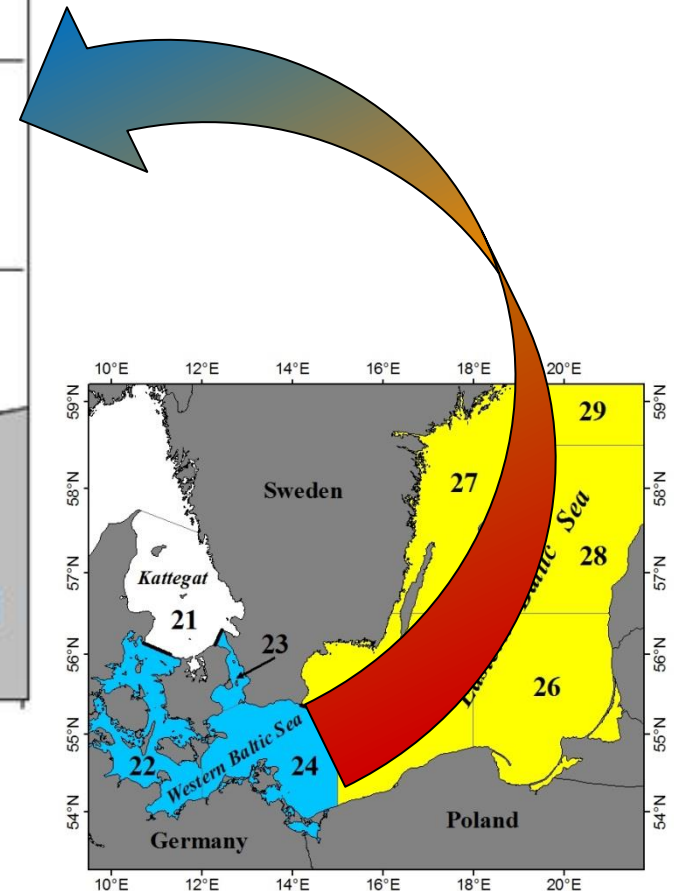
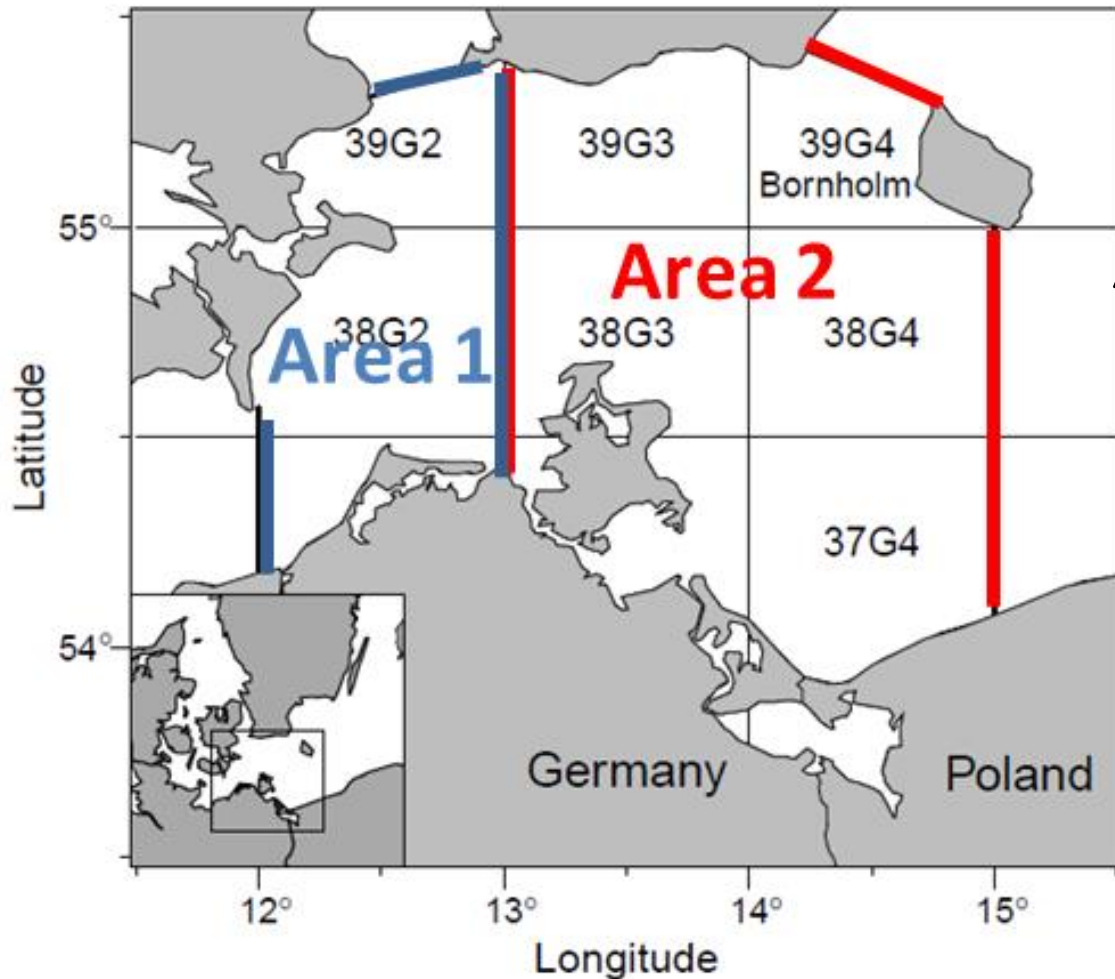
Samples of juveniles (<20cm) reveal recruitment dynamics



Highly dynamic distribution in time and space, likely also in response to climatic and biological drivers

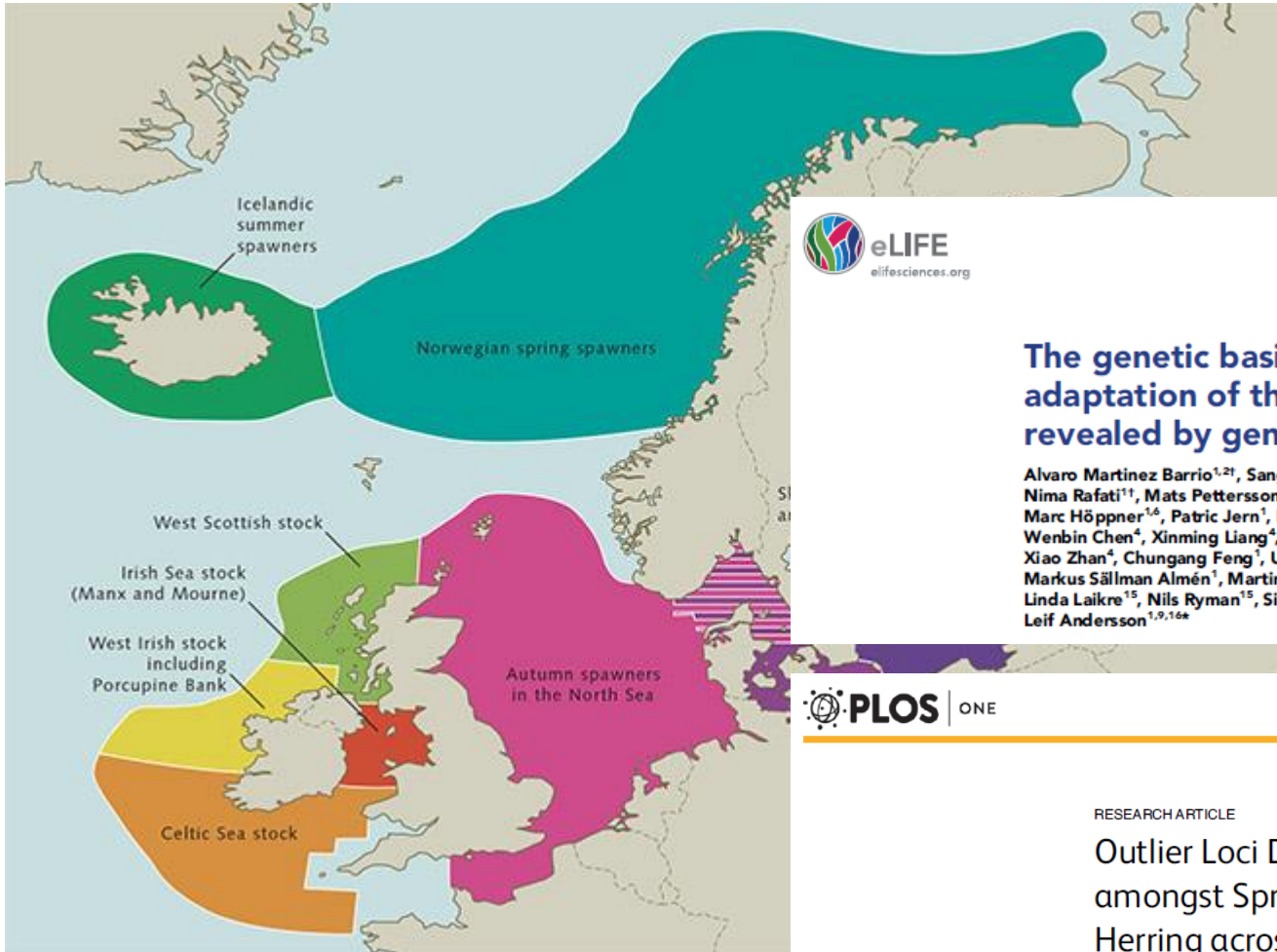


Atlantic cod in the W Baltic Sea – genetic marker based split stock assessment



The next 'perfect' example

Atlantic herring stocks and their mixing is now assessable with genetic markers throughout species' distribution



RESEARCH ARTICLE



The genetic basis for ecological adaptation of the Atlantic herring revealed by genome sequencing

Alvaro Martinez Barrio^{1,2†}, Sangeet Lamichhane^{1†}, Guangyi Fan^{3,4†}, Nima Rafati^{1†}, Mats Petterson¹, He Zhang^{4,5}, Jacques Dainat^{1,6}, Diana Ekman⁷, Marc Höppner^{1,6}, Patric Jern¹, Marcel Martin⁷, Björn Nystedt², Xin Liu⁴, Wenbin Chen⁴, Xinming Liang⁴, Chengcheng Shi⁴, Yuanyuan Fu^{4,8}, Kailong Ma⁴, Xiao Zhan⁴, Chungang Feng¹, Ulla Gustafson⁹, Carl-Johan Rubin¹, Markus Sällman Almén¹, Martina Blass¹⁰, Michele Casini¹¹, Arild Folkvord^{12,13,14}, Linda Laikre¹⁵, Nils Ryman¹⁵, Simon Ming-Yuen Lee³, Xun Xu⁴, Leif Andersson^{1,9,16*}



RESEARCH ARTICLE

Outlier Loci Detect Intraspecific Biodiversity amongst Spring and Autumn Spawning Herring across Local Scales

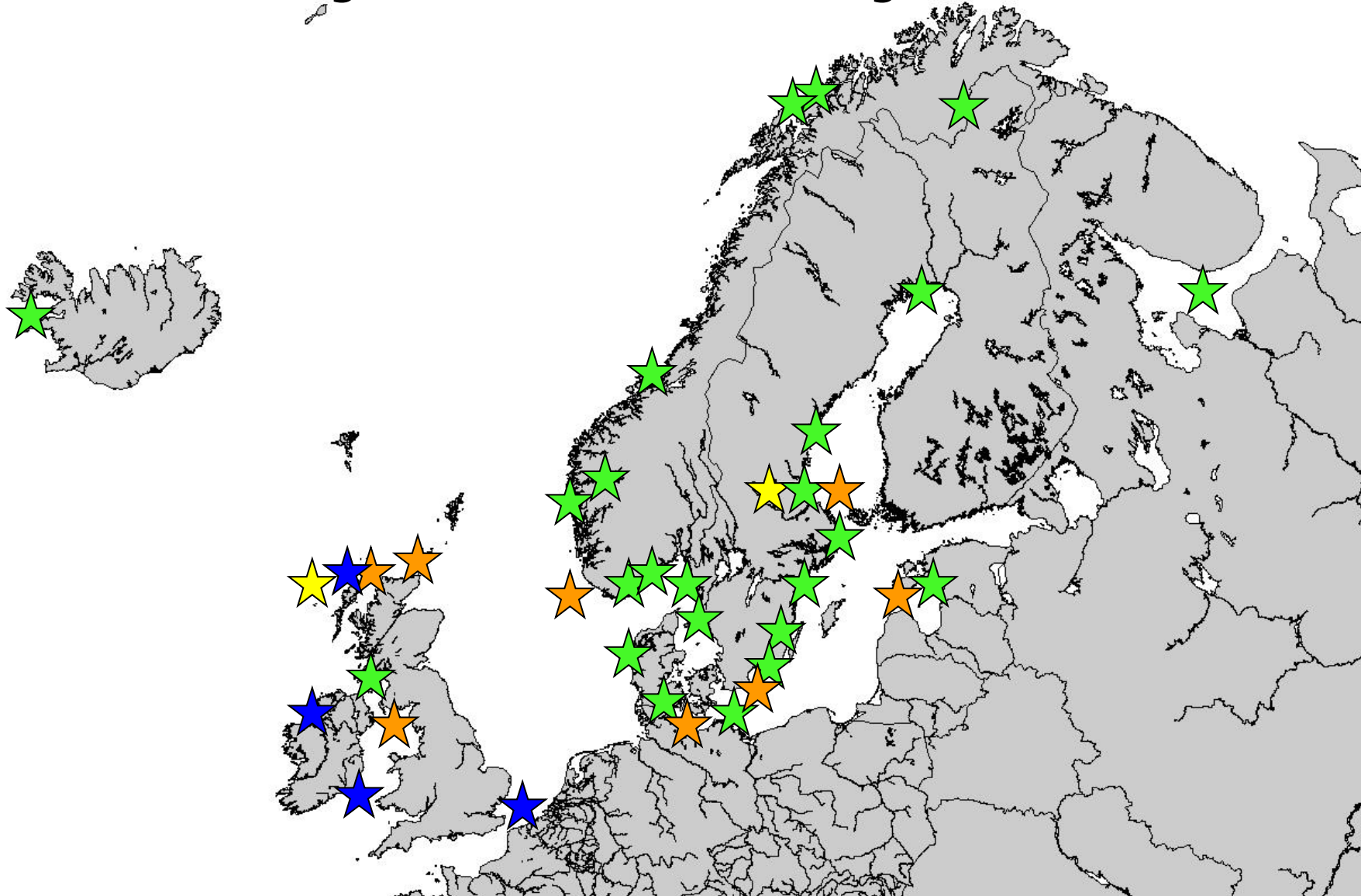
Dorte Bekkevold^{1*}, Riho Gross², Timo Arula³, Sarah J. Helyar⁴, Henn Ojaveer³

¹ Technical University of Denmark, National Institute of Aquatic Resources, Charlottenlund, Denmark, ² Estonian University of Life Sciences, Institute of Veterinary Medicine and Animal Sciences, Department of Aquaculture, Tartu, Estonia, ³ University of Tartu, Estonian Marine Institute, Pärnu, Estonia, ⁴ Institute for Global Food Security, Queen's University Belfast, Belfast, United Kingdom



★ Greenland

Samples of herring in spawning condition, representative of the biological units contributing to fisheries



State of the art: Taking advantage of genomics to do genetics

```

ATGCCCTAAAGGACTACTGA
CCCTGCTTTTAGAAATGCTC
TCGATTGCATATGCCCTAAA
GGGTACTGACCCTGCTTGT
AGAAATGCTCTCGATGCATA
TGCCCTAAAGGGTACTGAC
CCTGCTATTAGAAATGCTCT
CGATGCATATGCCCTAAATG
GGTACTGACCCTGCTAGTA
GAAATGCTCTCGATGCATAT
GCCCFAAAGGGTACTGACC
CTGCTCTTAGAAATGCTCTC
GATGCATATC
  
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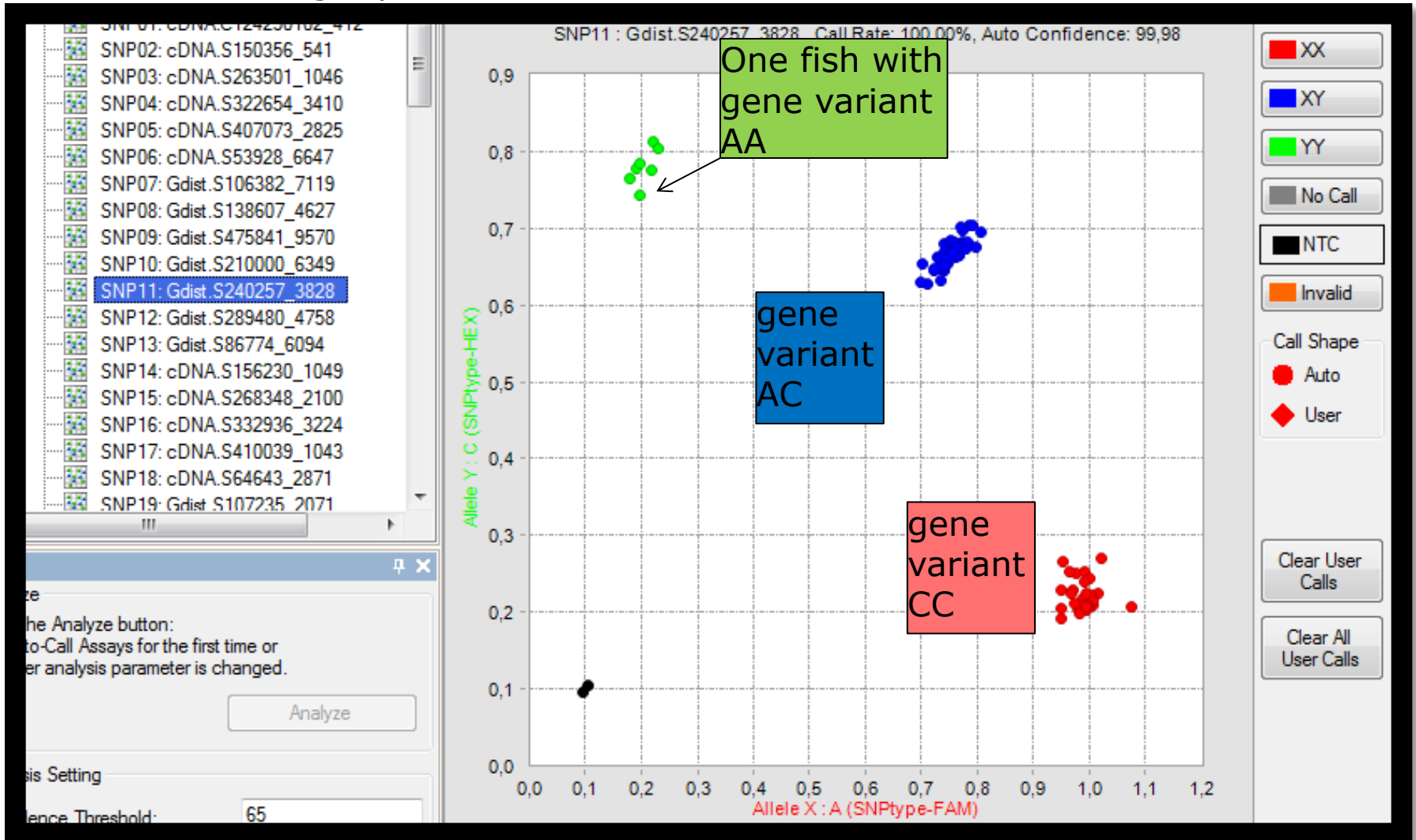
Sequencing genomes (~800*10⁶ bases) to obtain information about stock specific differences

From 9 mio. DNA sites to 48



Example of genetic data – one herring SNP marker out of 96

BioMark Fluidigm platform



Gene frequency differences and how they are used to determine the origin of a fish – an example

North Sea autumn spawning herring

Gene 1
AA = 25% of fish
AC = 22%
CC = 53%

Gene 2
TT = 99% of fish
TG = 1%
GG = 0%

Norwegian spring spawning herring

AA = 81% of fish
AC = 18%
CC = 1%

TT = 1% of fish
TG = 1%
GG = 98%



Probability of genotype
(Gene 1 & Gene 2)

in:

North Sea = 0

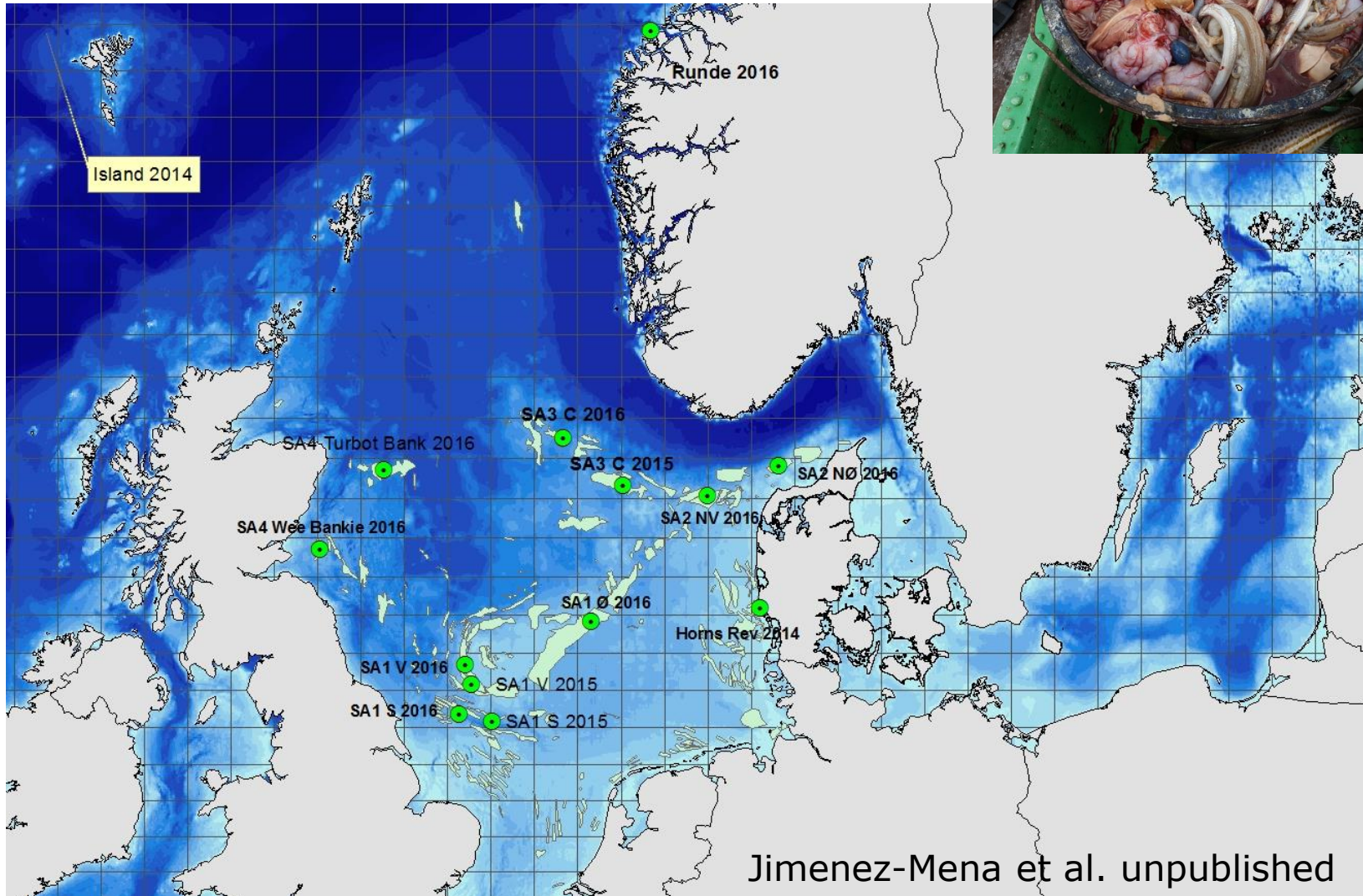
Norwegian = 0.8

Population splits addressable with new markers



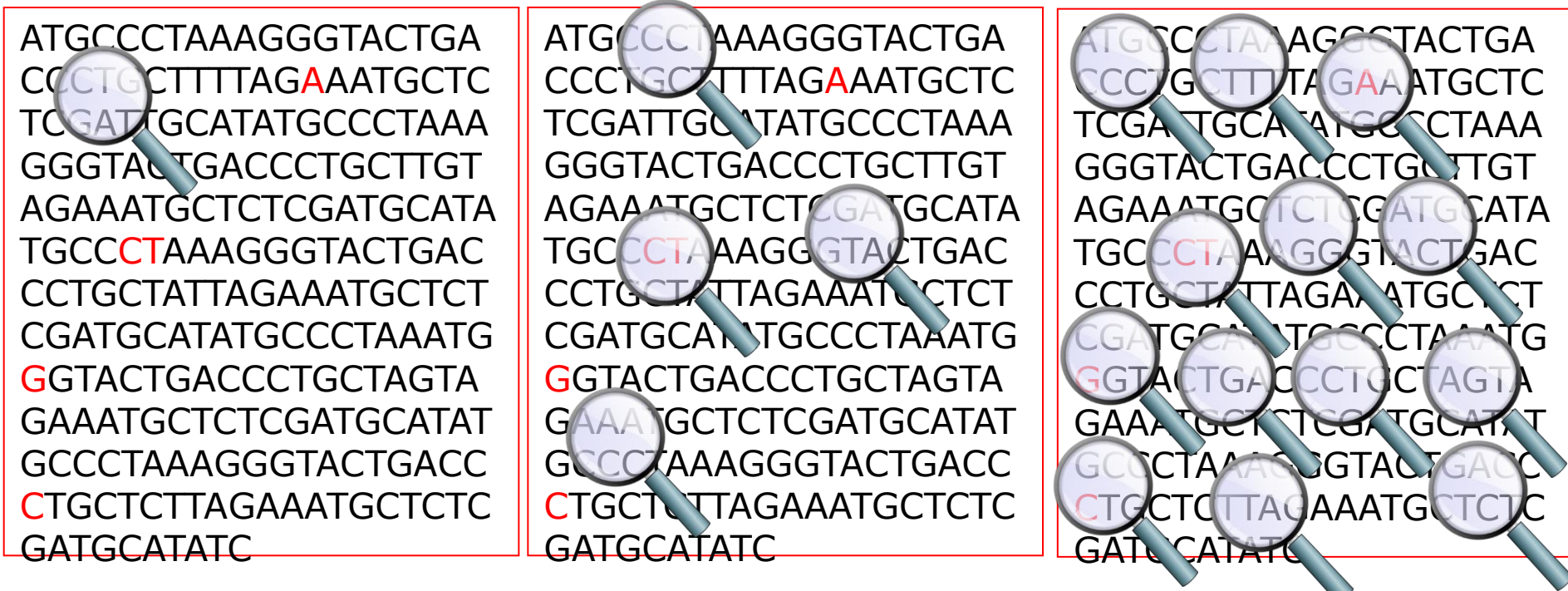
Area	ICES management area/sub area	Stocks/populations assumed to contribute to mixed aggregation fisheries	
Norwegian Sea	Div 2.a	ISSH, NSSH, NASH, NSAS, and potentially WBIH	
North Sea and English Channel	Sub Area 4 + Div 7.d	WBSS, NSAS, NSSH, and potentially ISSH, NASH, WBIH, Downs	In testing
Skagerrak	Div 3.a Skagerrak	WBSS, NSAS, Downs, NSSH, CBH	In testing
Western Baltic Sea	Div 3.a Kattegat and SD 22, 23, 24	WBSS, NSAS, NSSH, CBH	In testing
Ireland and West of the British Isles	Div 6.a, Div 7.b-c	WBIH, and potentially NSSH and NSAS	In testing

Lesser sandeel genomic study



When can we trust genetic data on stock units?

Robustness of result depends on the depth of the analysis



The image displays three panels of DNA sequence alignment, each enclosed in a red border. The sequences are as follows:

Panel 1 (Left):
ATGCCCTAAAGGGTACTGA
CCCTGCTTTTAGAAATGCTC
TCGATTGCATATGCCCTAAA
GGGTACTGACCCTGCTTGT
AGAAATGCTCTCGATGCATA
TGCCCTAAAGGGTACTGAC
CCTGCTATTAGAAATGCTCT
CGATGCATATGCCCTAAATG
GGTACTGACCCTGCTAGTA
GAAATGCTCTCGATGCATAT
GCCCTAAAGGGTACTGACC
CTGCTCTTAGAAATGCTCTC
GATGCATATC

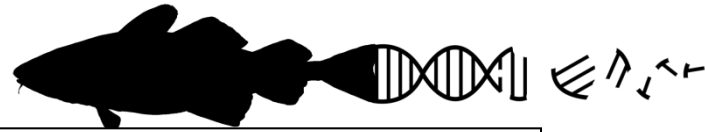
Panel 2 (Middle):
ATGCCCTAAAGGGTACTGA
CCCTGCTTTTAGAAATGCTC
TCGATTGCATATGCCCTAAA
GGGTACTGACCCTGCTTGT
AGAAATGCTCTCGATGCATA
TGCCCTAAAGGGTACTGAC
CCTGCTATTAGAAATGCTCT
CGATGCATATGCCCTAAATG
GGTACTGACCCTGCTAGTA
GAAATGCTCTCGATGCATAT
GCCCTAAAGGGTACTGACC
CTGCTCTTAGAAATGCTCTC
GATGCATATC

Panel 3 (Right):
ATGCCCTAAAGGGTACTGA
CCCTGCTTTTAGAAATGCTC
TCGATTGCATATGCCCTAAA
GGGTACTGACCCTGCTTGT
AGAAATGCTCTCGATGCATA
TGCCCTAAAGGGTACTGAC
CCTGCTATTAGAAATGCTCT
CGATGCATATGCCCTAAATG
GGTACTGACCCTGCTAGTA
GAAATGCTCTCGATGCATAT
GCCCTAAAGGGTACTGACC
CTGCTCTTAGAAATGCTCTC
GATGCATATC

Magnifying glasses are placed over the sequences to highlight differences. In the first panel, a magnifying glass is over the 'A' in the second position of the second line. In the second panel, magnifying glasses are over the 'A' in the second position of the second line, the 'G' in the second position of the third line, and the 'A' in the second position of the fourth line. In the third panel, magnifying glasses are over the 'A' in the second position of the second line, the 'G' in the second position of the third line, and the 'A' in the second position of the fourth line.

And the list of species and stocks with genetic data keeps getting longer: Atlantic mackerel, sprat, mesopelagics,

Novel approaches to the application of molecular methods in ecosystem and fisheries assessment



Environmental DNA



om water

ORIGINAL ARTICLE

WILEY FISH and FISHERIES

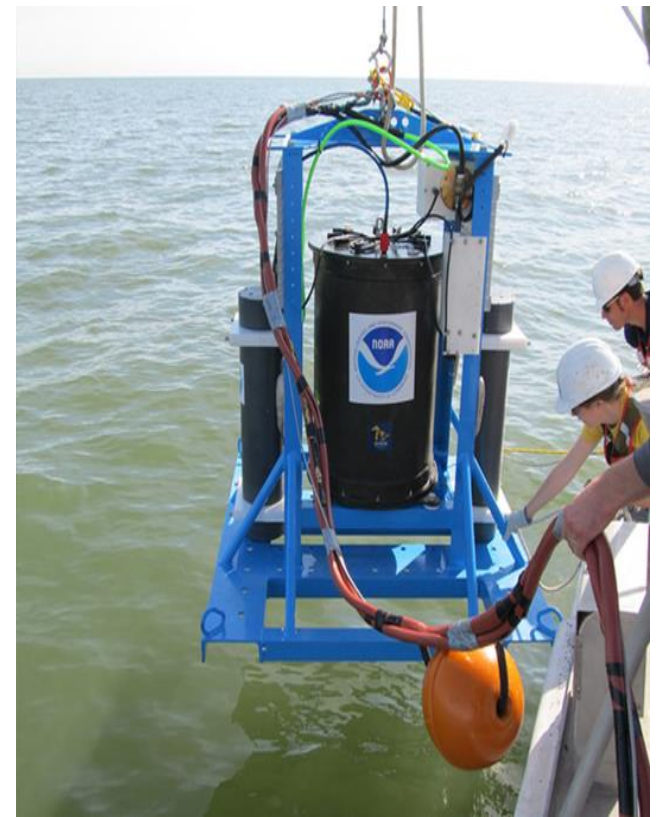
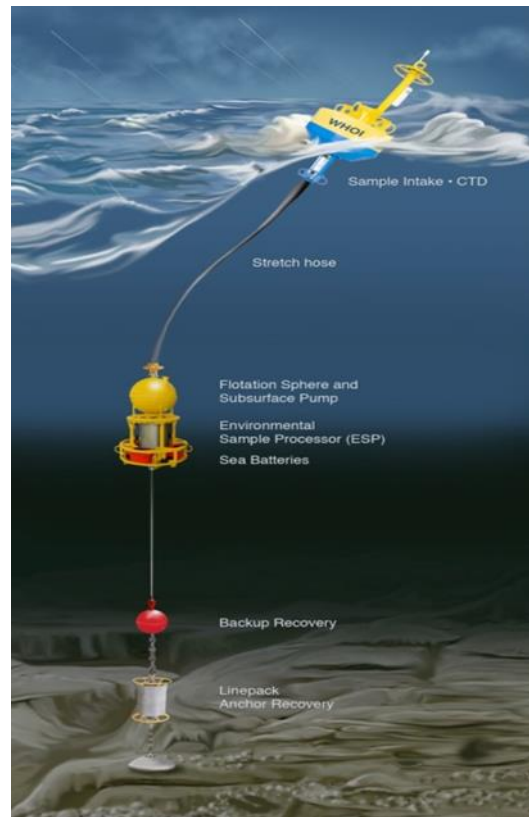
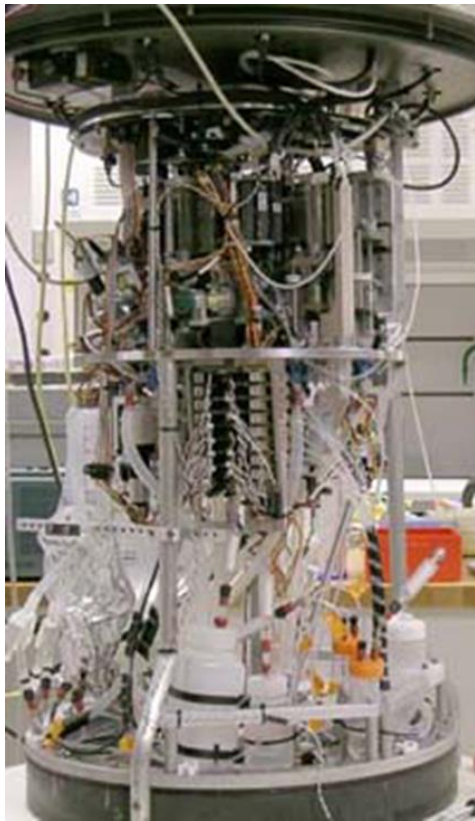
The sceptical optimist: challenges and perspectives for the application of environmental DNA in marine fisheries

Brian Klitgaard Hansen¹ | Dorte Bekkevold¹ | Lotte Worsøe Clausen^{1,2} | Einar Eg Nielsen¹



ments





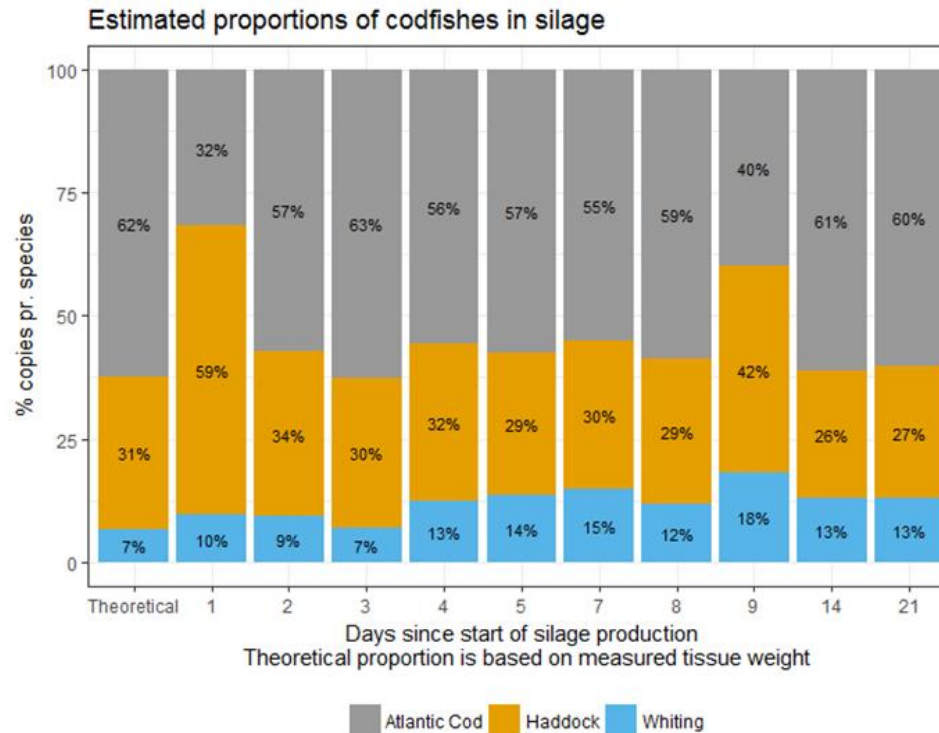
AAGE V. JENSENS FOND

DTU Aqua
National Institute of Aquatic Resources



Silage

- Fish was dissolved in $\sim 10\text{L}$ of formic acid
- 250 μl samples taken out and analyzed on qPCR.



Hansen et al. (unpublished data)

Costings of SNP analyses for input to assessment

Platform	Cost per individual (@ 2000 fish)			Bench & bio-informatics time 2000 fish
# markers typed	50 SNP	100 SNP	200 SNP	100 SNPs
SNP array based (Fluidigm)	14 €	20 €	34 €	31 days
GT sequencing based*	4 €	5 €	6 €	~17 days

(* Estimate from Aykanat *et al.*, J Fish Biol., 2016)

- Genetic studies are expensive – Not really and prices are constantly decreasing
- Genetic results are often 'oversold' – We have several cases proving the opposite. Genetic analyses cannot answer all management relevant questions, but lack of basic biological information leads to risk of less efficient management strategies
- Inconsistent interpretation of genetic results - Structural, historical sampling issues
- Importance of genetic information is far outweighed by other inputs to management decisions – Substantially better resolution with genetic markers. Time series are emerging. Potential for front user-based analyses on-site.
- Obstacle is implementation into assessment and management

