14th International Symposium on the Biology and Management of Coregonid Fishes

WebCoregonid2020

June 22–26, 2020

Oral presentation

Session: Theme: 2. Genetics & evolutionary ecology

Organised by University of Jyväskylä, Finland

Sponsored by: Luke NATURAL RESOURCES INSTITUTE FINLAND JYVÄSKYLÄN YLIOPISTO UNIVERSITY OF JYVÄSKYLÄ Federation of Finnish Learned Societies

ddRADseq for European whitefish (*Coregonus lavaretus*): Genome wide markers for genomic selection and studies of genome and population evolution

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Natural Resources Institute Finland (Luke)

Production systems

31600 Jokioinen, Finland

This project has received funding under Kolarctic CBC programme 2014-2020 with a contract number 4/2018/095/KO4058 'ArctAqua - Cross-border innovations in Arctic aquaculture'

15.6.2020

Objective: A method to generate genomic data for whitefish genomic selection

Motivation:

- The 2nd most valuable farmed fish species in Finland.

Objective:

- Genotyping by Sequencing method

1. Laboratory protocol
2. Bioinformatic protocol

Impact:

- Enable genomic selection.

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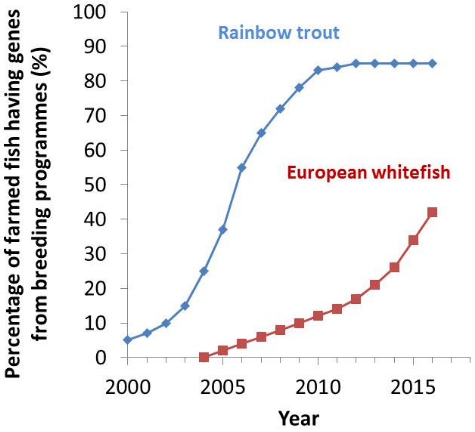
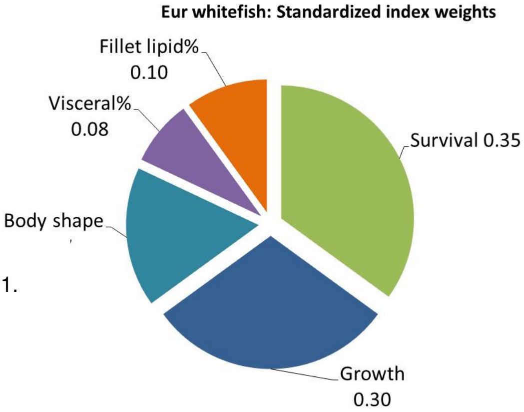
European whitefish *Coregonus lavaretus* breeding programme

Luke's breeding programme has major impact on industry



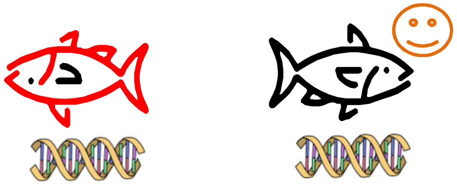
Started in 2002
4th generation now
Around 70 families every year

Janhunen et al. (2017). *Aquaculture* 481: 229-238.
Kause et al. (2011). *Journal of Animal Science* 89: 959-971.
Antti.Kause@luke.fi



Genomic selection: DNA variation of individuals is used to predict the genetic superiority of parents for improved product quality, disease resistance and production efficiency

- More accurate selection, new traits



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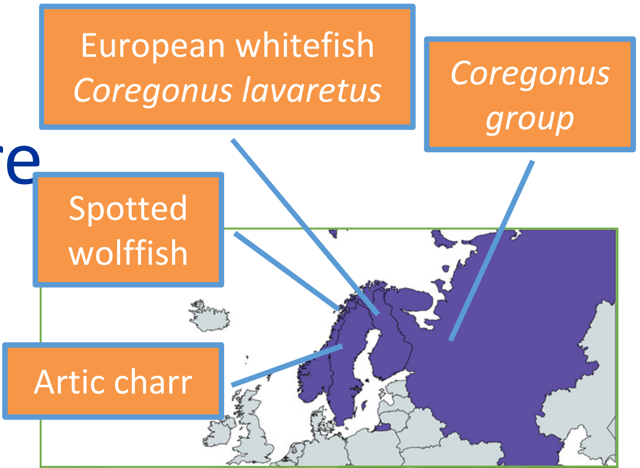
ArctAqua – Cross-border innovations in Arctic aquaculture

- KOLARTIC EU CBC Cross Border Cooperation project
- November 2019 – November 2022
- Coordinator: Jose Beirão (Nord Uni, Norway)
- 8 industry and research partners
- To intensify SMART SPECIALISATION of aquaculture in artic areas
- Species of national interest

Luke's main research task: Develop reproduction technologies and genomic tools for genomic selection in European whitefish (siika) national breeding programme



European whitefish *Coregonus lavaretus*

Coregonus group

Spotted wolffish

Artic charr

Luke team: Tapio, Kause, Tapio, Iso-Touru, Fischer, Bitz, Koskinen, Nousiainen.

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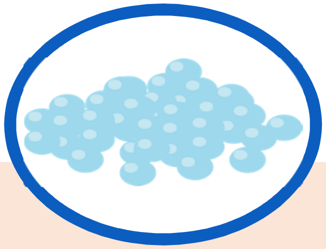
Blue bioeconomy

Genomics and breeding

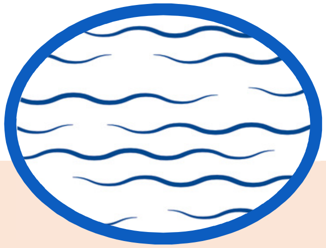


Breeding and genetics of aquaculture species

Broodstock management concepts

Genome based tools for breeding

Sustainable use of aquatic ecosystems



Sustainable and profitable fisheries

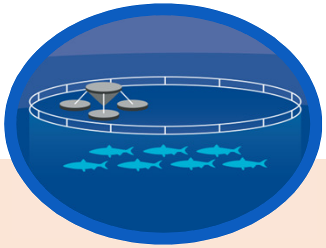
Rebuilding migratory fish populations

Viable fish for releases

Conservation and management of threatened species

Spatial planning in waters

Blue Production



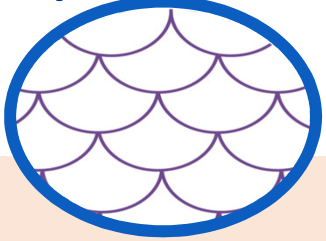
Sustainable production of aquatic organisms

Recirculating and offshore aquaculture production, methods and technologies

Aquatic feed solutions

Symbiosis concepts

Value-added food and aquabiomass products



Fractionation and valorisation of aquatic biomass to added value products

Aquatic value chains

Valorisation of algae production

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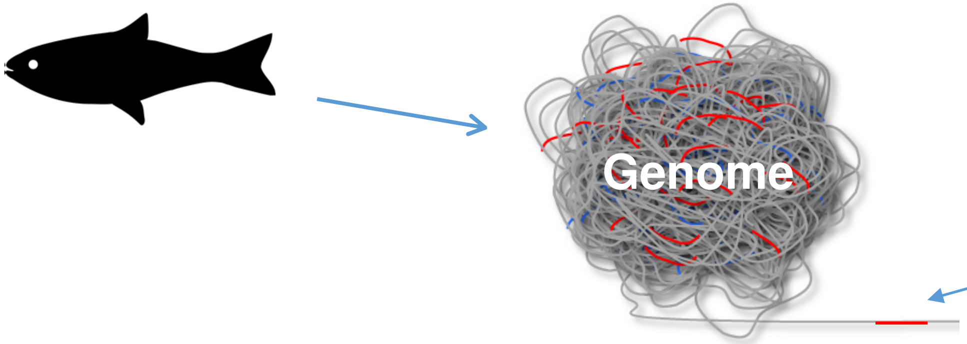
Genotyping By Sequencing (GBS)



5%

Variable site

```
TACGAGGGCACTGGTACGCATTA
TACGAGGGCACTGGTACGCATTA
TACGAGGGCACTGGTACGCATTA
TACGAGGGCACTGGTACGCATTA
TACGAGGGCACAGGTACGCATTA
TACGAGGGCACAGGTACGCATTA
```

15000 – 70000 genome segments

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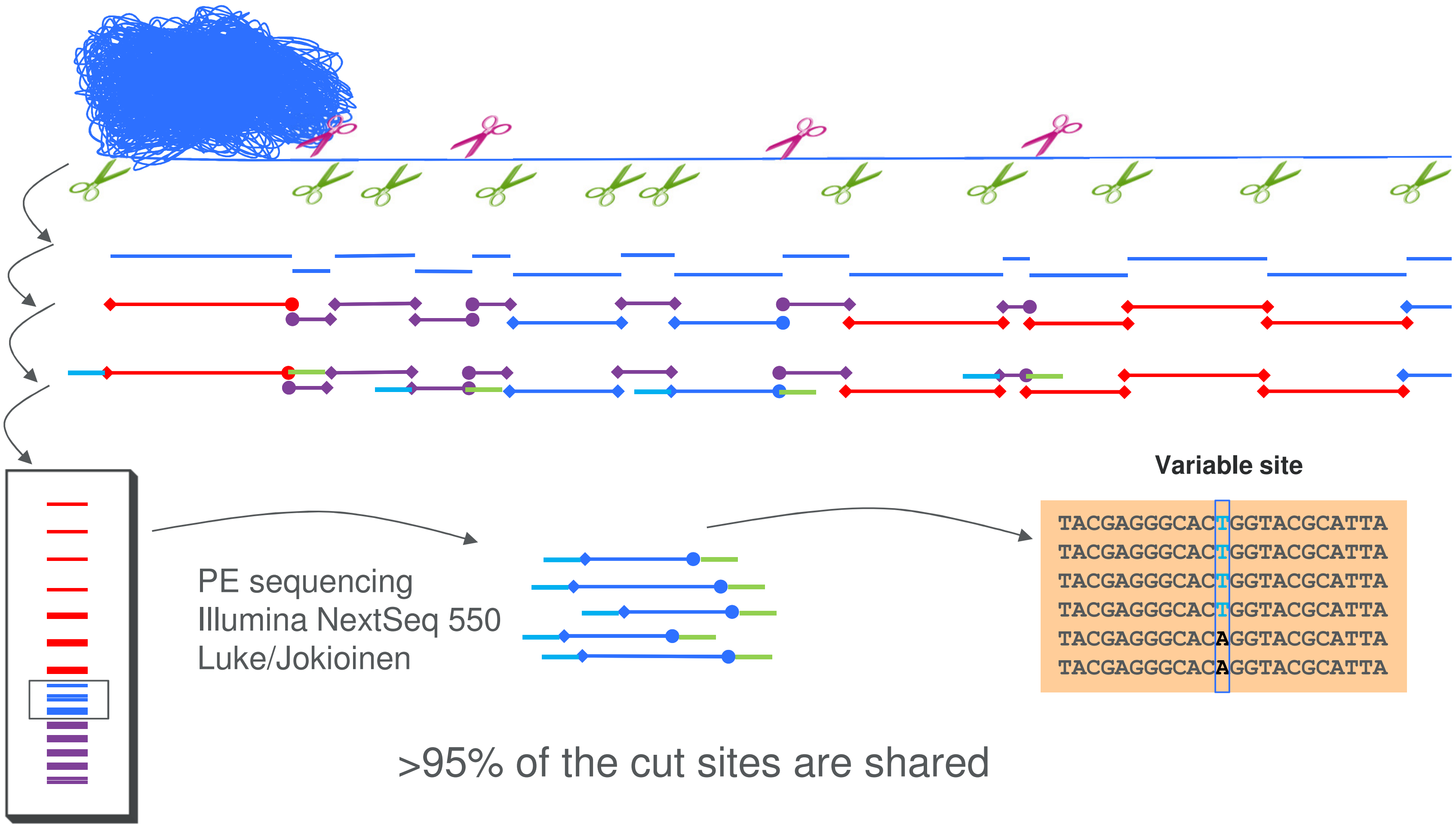

RE digestion

Adaptor
ligation

Index
PCR

Size
selection

PE sequencing
Illumina NextSeq 550
Luke/Jokioinen

Variable site

TACGAGGGCACTGGTACGCATTA
TACGAGGGCACTGGTACGCATTA
TACGAGGGCACTGGTACGCATTA
TACGAGGGCACTGGTACGCATTA
TACGAGGGCACAGGTACGCATTA
TACGAGGGCACAGGTACGCATTA

>95% of the cut sites are shared

Kolarctic

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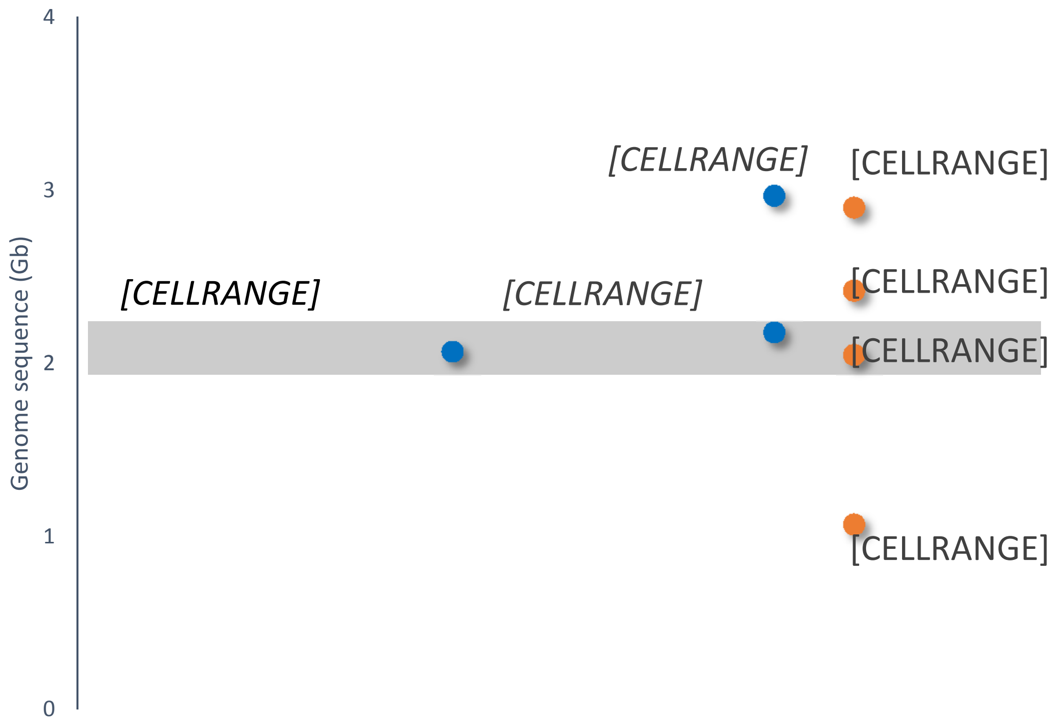
Whitefish genome



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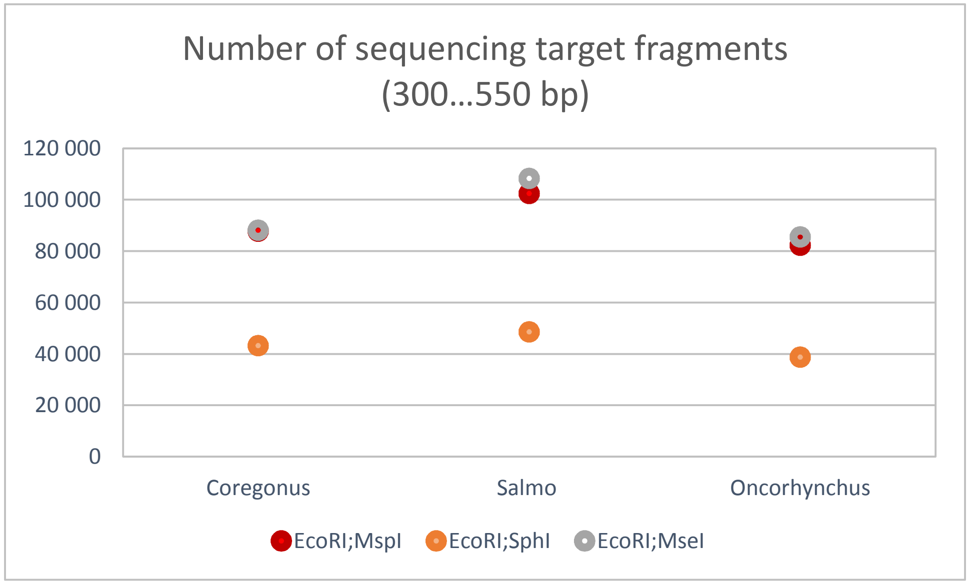
Enzyme choice

- Optimal target length and number
    - Length 2xRL… 500 bp
    - 10…100x sequencing coverage
- Combining iteratively:
    1. Enzymes
        - Short enough (300…550bp)
        - Optimal number (50…100k)
    2. Included size range of fragments
    3. Sequencing protocol
        - Single end / paired end
        - Read length (RL)



SimRAD (Lepais & Weir (2014))

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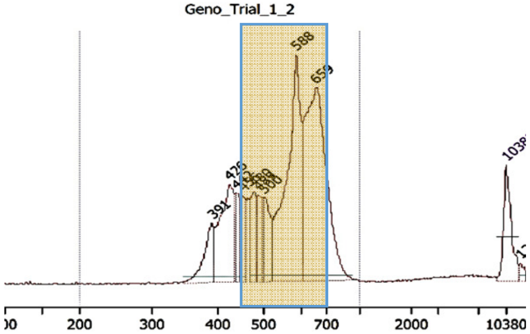
Data generation protocol

- Double Digestion with EcoRI and SphI
- Ligation with common adapters
- Library preparation using Phusion-polymerase and Nextera XT indices
- Size selection on a gel: 450–700 bp fragment length
- Sequencing on NextSeq550: Paired-end (2x150)



Salas-Lizana, R., & Oono, R. (2018).

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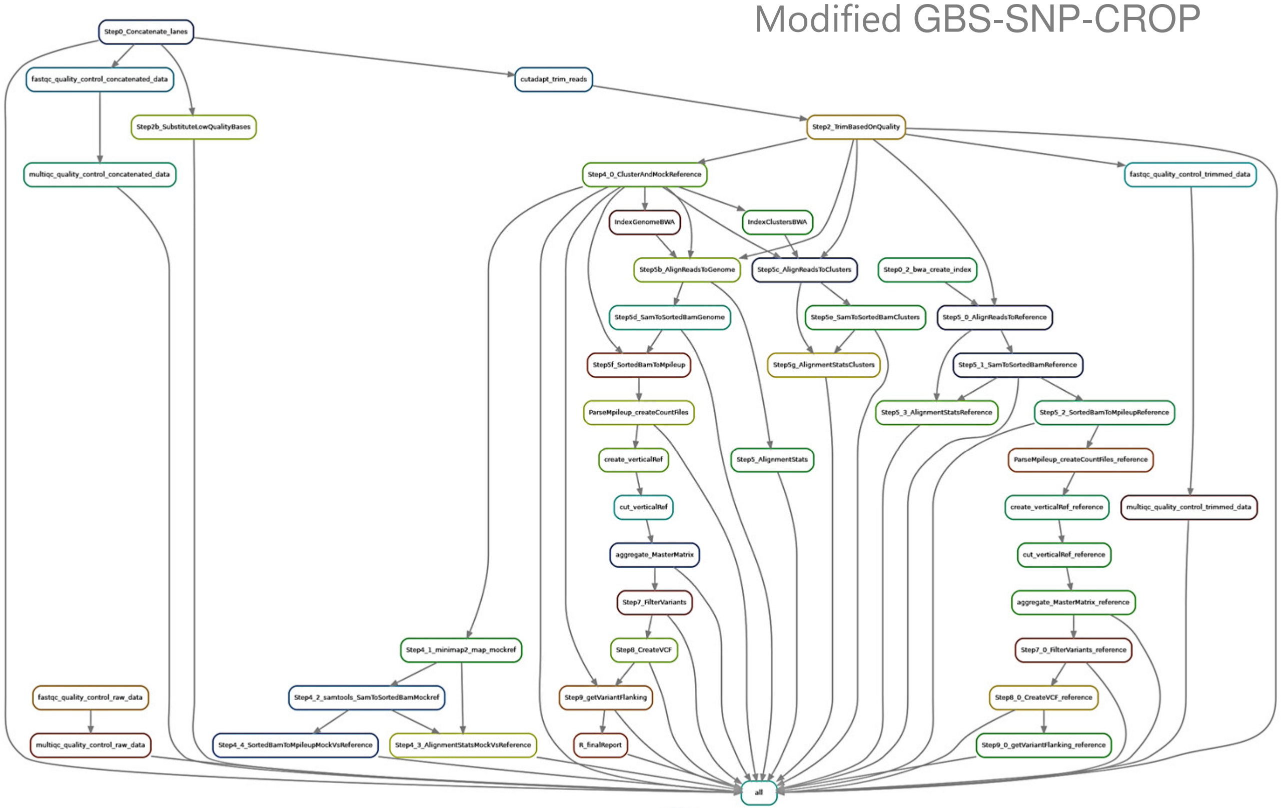
Bioinformatic pipe

Modified GBS-SNP-CROP



Tools in each stage

1. FastQC, MultiQC
2. PEAR, vsearch
3. bwa
4. samtools
5. R

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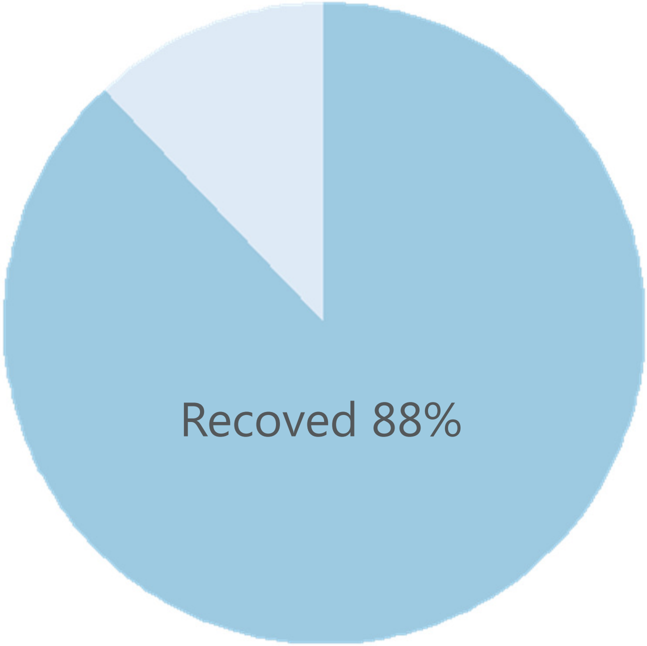
Experimental fragment recovery

Expectation:
43,241 fragments



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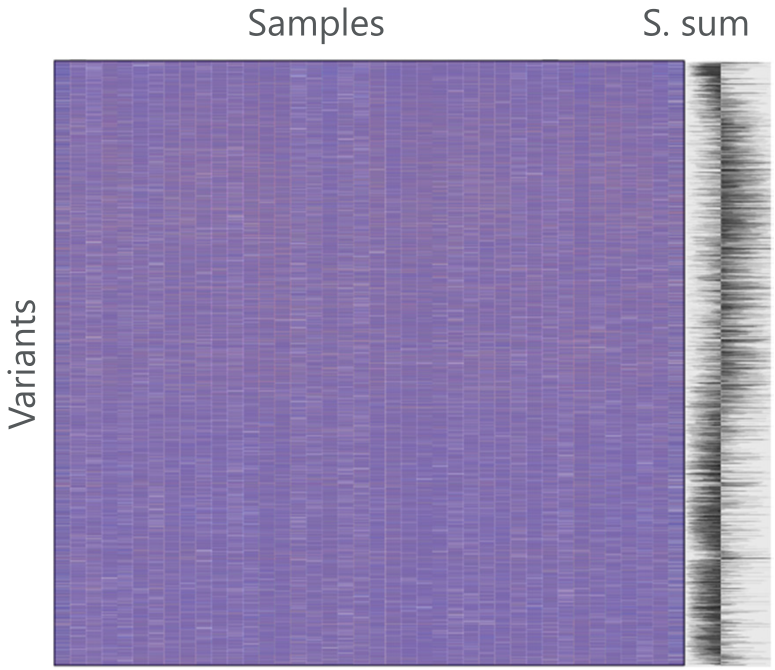
Data set pruning

- Excluding samples with >10% missing
- Removing exceptionally low/high read count loci

-> 14 485 variable markers



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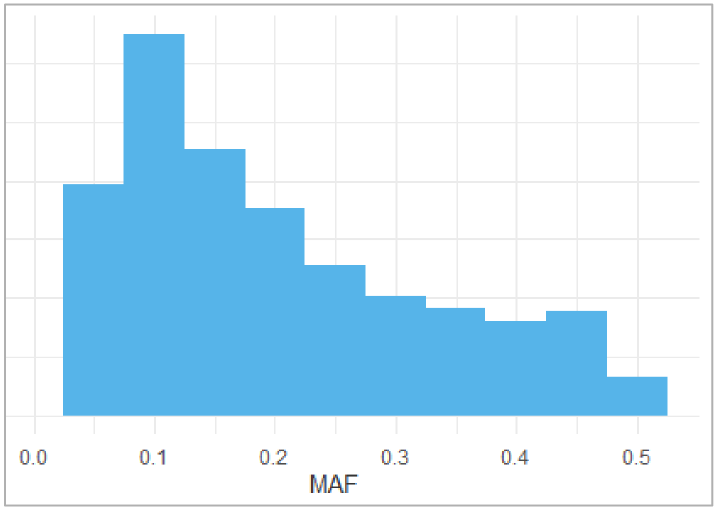
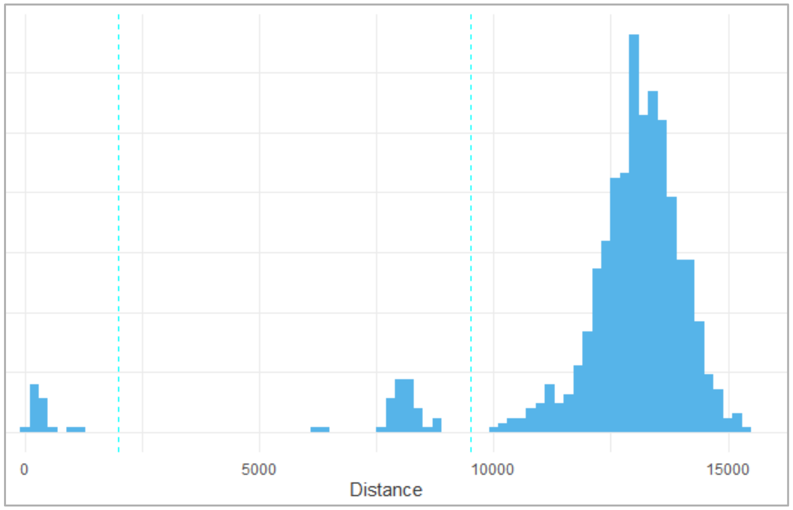
Assessing marker patterns

Distribution of Minor Allele Frequencies



Distribution of Manhattan distances



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Summary

- Established 15 000 marker candidates
- Developed a method to score.
- These can be used as markers for breeding, identification and management purposes.

-> Facilitates genomic breeding and research immediately.

-> Enables generating other genotyping methods.

- Next steps
    - Further optimization to improve consistency and reduce the price.
    - Assigning to genome.
    - Generating genomic data for Finnish whitefish breeding program for genomic selection.

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Thank you!

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