GENOMIC SOLUTIONS FOR SHELLFISH SELECTIVE BREEDING

Workshop Vivaldi

Lucie Genestout & Romain Morvezen

AQUA 2018 Montpellier – 29/08/2018

Overview

- 300 000 analyzes / year
    - 40 000 for aquaculture
- 1 200 m^2 of facilities
- 20 km from Paris
- Since 1956
- 2 million samples in stock
- Subsidiary of the cooperative EVOLUTION Les éleveurs de vie

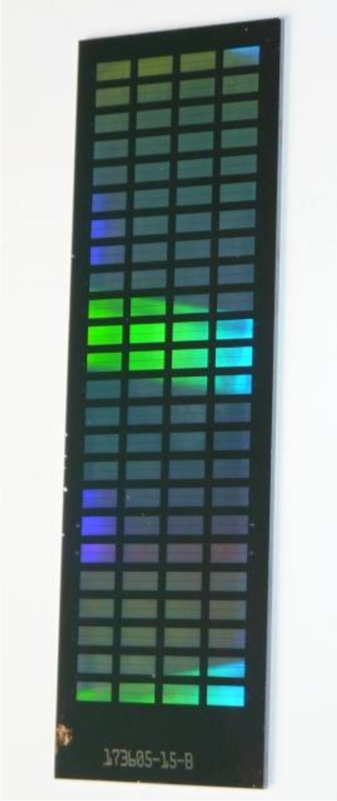
An industrial genotyping platform

LABOGENA
DNA Experts

- High throughput genotyping on DNA chips
  - 8 liquid handling robots (Tecan)
  - 5 extraction robots Qiasymphony (Qiagen)
  - Infinium XT chemistry (Illumina): 96 samples HD Chips
- Management system LIMS
  - Barcode traceability from the sampling to the result
- Standards
  - ISO17025
  - ISAG interlaboratory tests rank 1

An efficient SNP panel for Pacific Oyster

- 384 markers on new high throughput Illumina XT chemistry
- Including OsHV-1 resistance markers
- Assignments made with AccurAssign Labogena software
  - Taking into account mating plans
  - Using both likelihood and exclusion
  - Ranking of parents



96samples DNA chip Illumina

An efficient SNP panel for Pacific Oyster

	marinove	Novostrea Bretagne	Satmar Naturellement…	France Naissain
Assigned to 1 couple	**466**	**514**	**577**	**408**
Assigned to many couples	12	0	0	0
Not assigned	12	47	23	6
Inexploitable	31	35	12	152
Useful assignment rate	89%	86%	94%	72%

Results from GenOyster Project, obtained with GigADN Project markers

SYSAAF Ifremer FranceAgriMer

An efficient SNP panel for Pacific Oyster

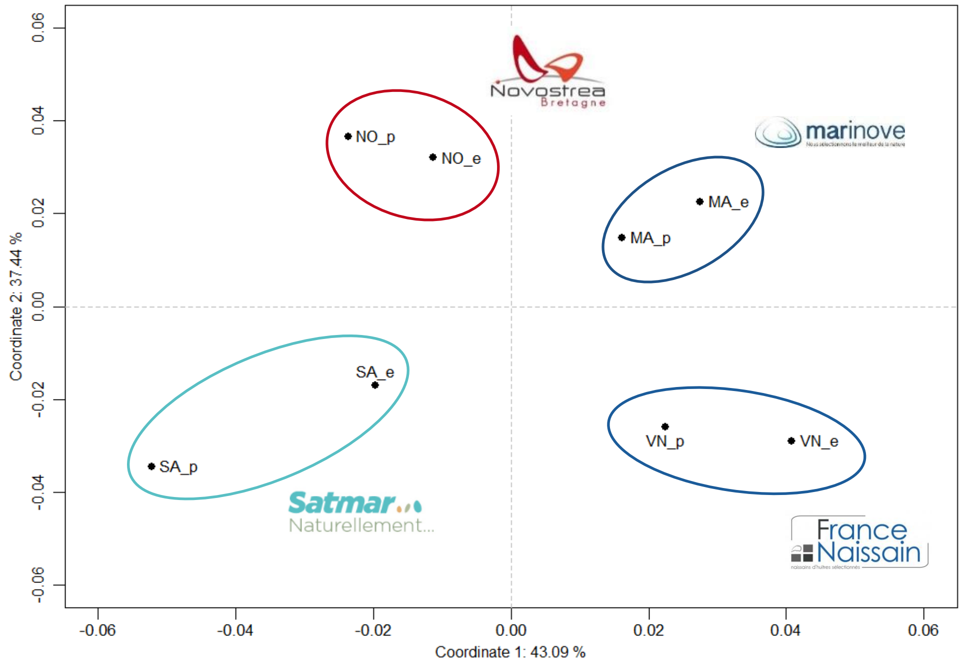
Assigned to 1 couple	**522**	**1098**
Assigned to many couples	2	3
Not assigned	49	55
Inexploitable	225	53
Useful assignment rate	65%	91%

Results from Vivaldi Project, obtained with GigADN Project markers

An efficient SNP panel for Pacific Oyster

- Good tool to manage genetic diversity by identifying over representation of parents
- Helps to keep reproductive success
- Need to improve DNA quality
- More than 13 000 Oysters already analyzed
- Could be used to identify the origin-hatchery

Next SNP panel in progress for Manila Clams !



Results from GenOyster Project

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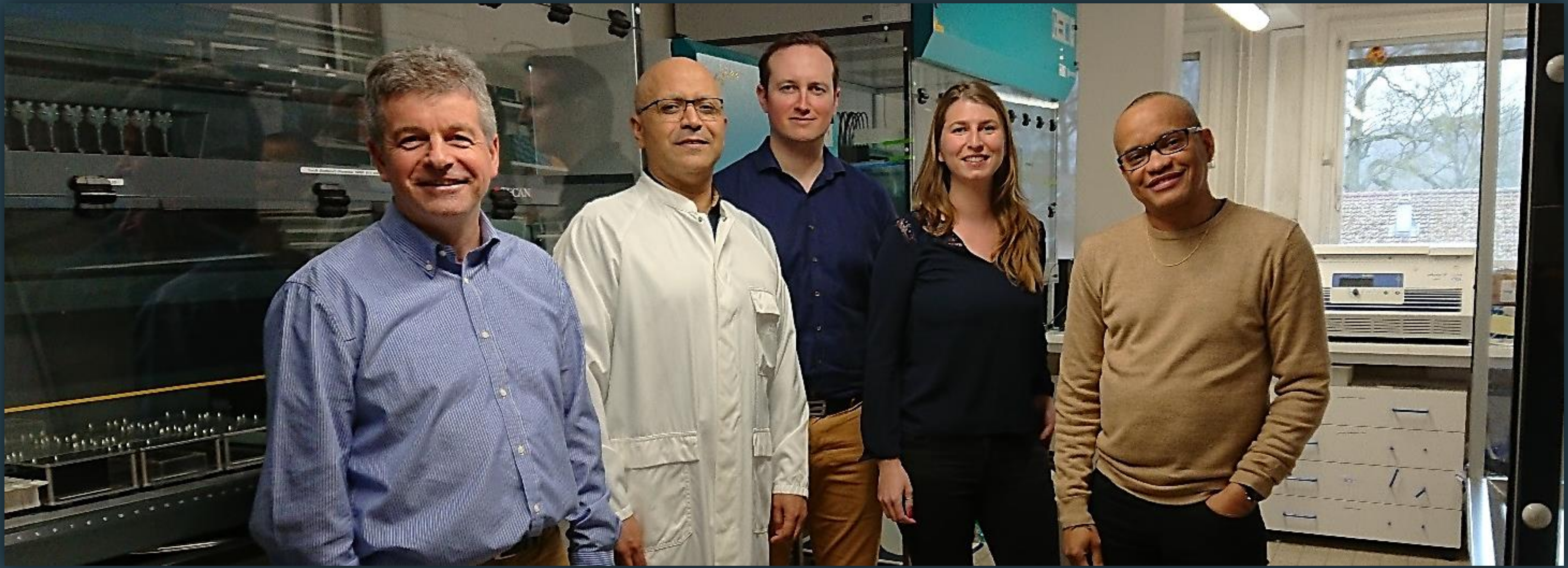
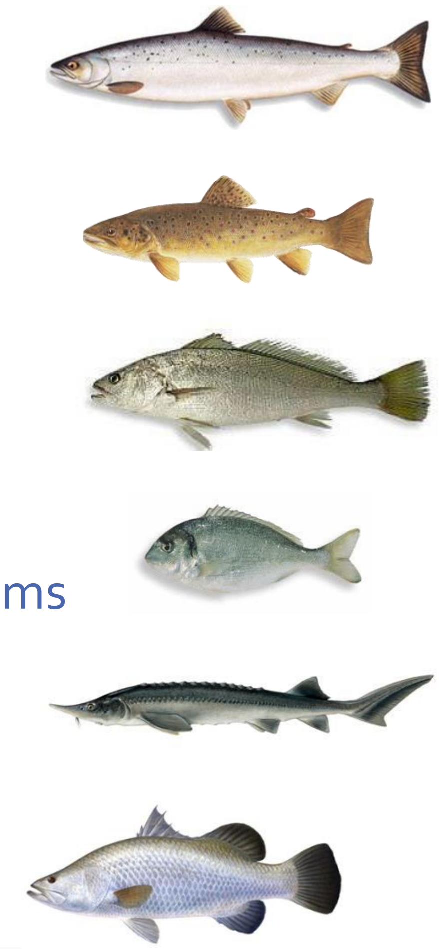
DNA Experts



Visit us at booth #102 !

What’s SYSAAF ?

SYSAAF : **French Poultry and Aquaculture Breeders Association**
18 breeders in aquaculture
10 species of fishes, 4 shellfishes and 3 shrimps
Aquaculture workforce : 7 full-time employees and 3 PhD candidates

Our role :

- Counseling in genetic selection and management of breeding programs
- Genetic and genomic evaluations and inbreeding management
- Research & development
- Ploidy control


Shellfish selection in SYSAAF

5 French shellfish hatcheries members of the SYSAAF – for their selective breeding

Shellfish selection in SYSAAF

Satmar Naturellement...
France Naissain
DRMM
marinove
FRANCE HALIOTIS

5 French shellfish hatcheries members of the SYSAAF – for their selective breeding

Active in research programs :

- GenOyster (2015-2018)

  Validation of DNA-parentage assignment panel for Pacific oyster *C. gigas*

- VIVALDI (2016-2020)

  Development of DNA-parentage assignment panel for Manila clam *R. philippinarum*

  Estimation of genetic parameters for growth and quality traits in *C. gigas*

  Estimation of genetic parameters for resistance to *Perkinsus olseni* and *V. tapetis*, and for growth and quality traits in *R. philippinarum*

  Improvement of breeding strategies by simulations

- RésiPal (2018-2019)

  Estimation of genetic parameters for resistance to *V. tapetis* by controlled challenge

- GenOrmeau (2016-2019)

  Estimation of genetic parameters for growth, quality, color in *Haliotis Turbeculata*

  Development of DNA-parentage assignment panel

Shellfish selection in SYSAAF

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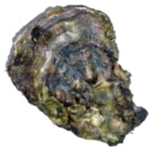

- VIVALDI (2016-2020)

  Development of **DNA-parentage assignment panel** for Manila clam *R. philippinarum*

  Estimation of genetic parameters for growth and quality traits in *C. gigas*

  Estimation of genetic parameters for resistance to *Perkinsus olseni* and *V. tapetis*, and for growth and quality traits in *R. philippinarum*

  Improvement of breeding strategies by simulations

- RésiPal (2018-2019)

  Estimation of genetic parameters for resistance to *V. tapetis* by controlled challenge, with **parentage reconstruction**

- GenOrmeau (2016-2019)

  Estimation of genetic parameters for growth, quality, color in *Haliotis Turbeculata*

  Development **of DNA-parentage assignment panel**

DNA-parentage assignment in selection

Why ?

- Mass selection assisted by marker
- Inbreeding management in hatchery-based selective breeding program (pedigree information for broodstock)
- Pedigree allows for selection on estimated breeding values (EBV)
- Accuracy increased in mixed family design with EBV (Phenotype = Genetic + Environnement)
- Genealogical selection on collateral (on letal traits) for mixed family design

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Example : Rainbow trout in France

Growth possibly measured on candidates

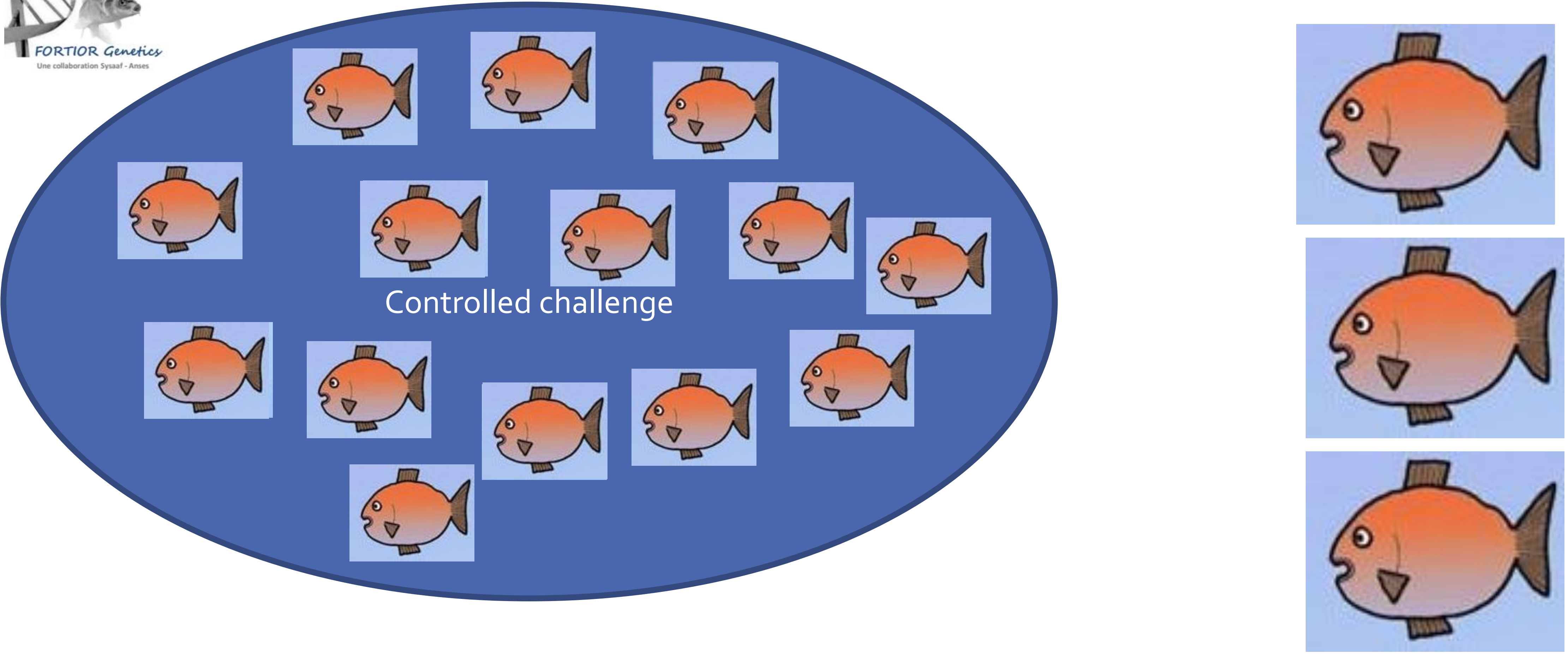
Yield traits measured on sibs

Eggs production measured on sibs

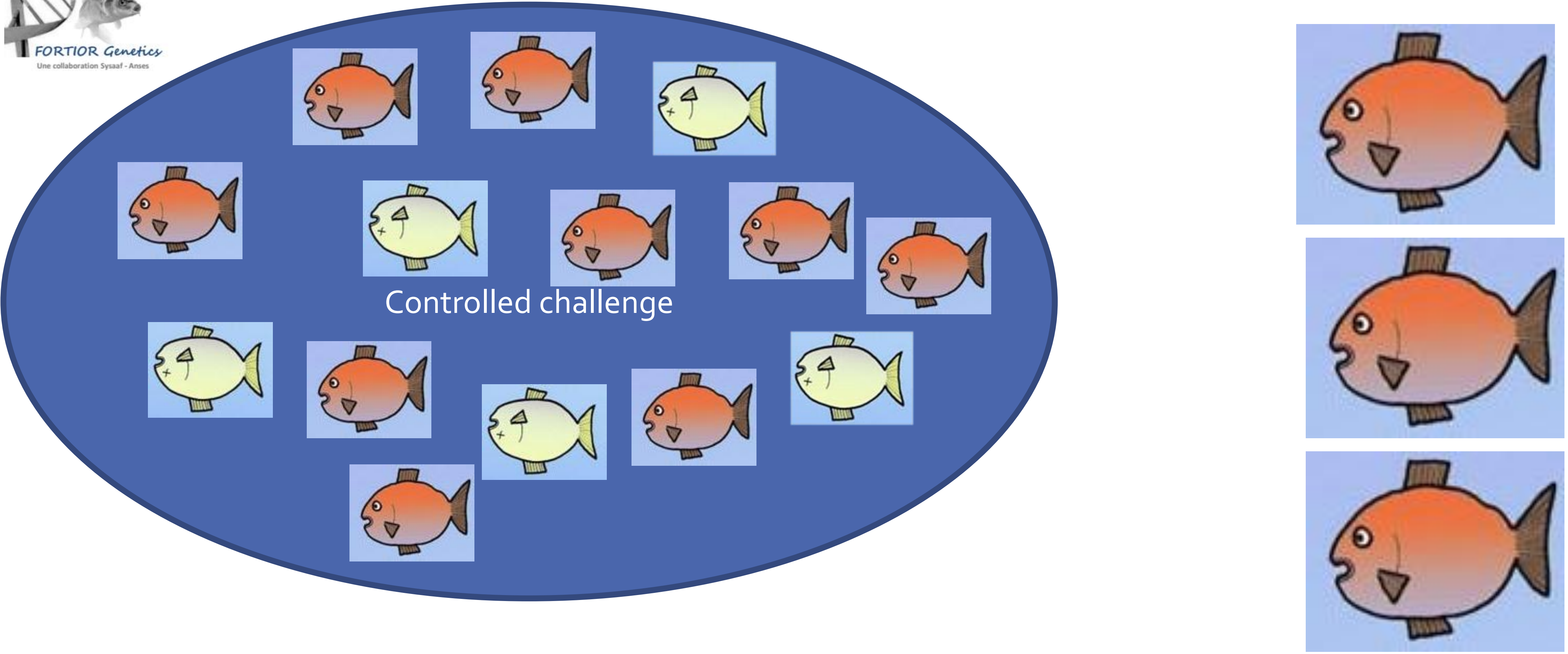
Resistance to many pathogens by controlled or field challenges measured on sibs

Resistance to pathogens by controlled challenges on sibs

FORTIOR Genetics
Une collaboration Sysaaf - Anses



Resistance to pathogens by controlled challenges on sibs



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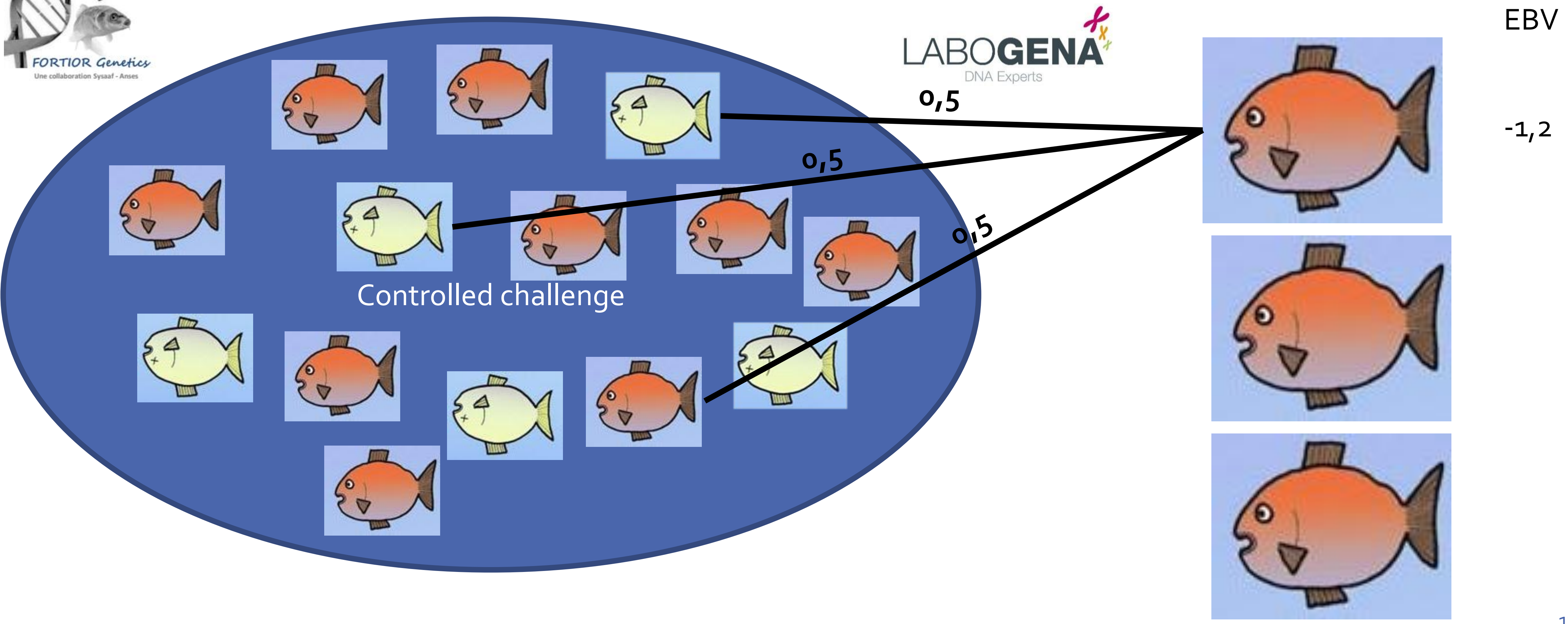

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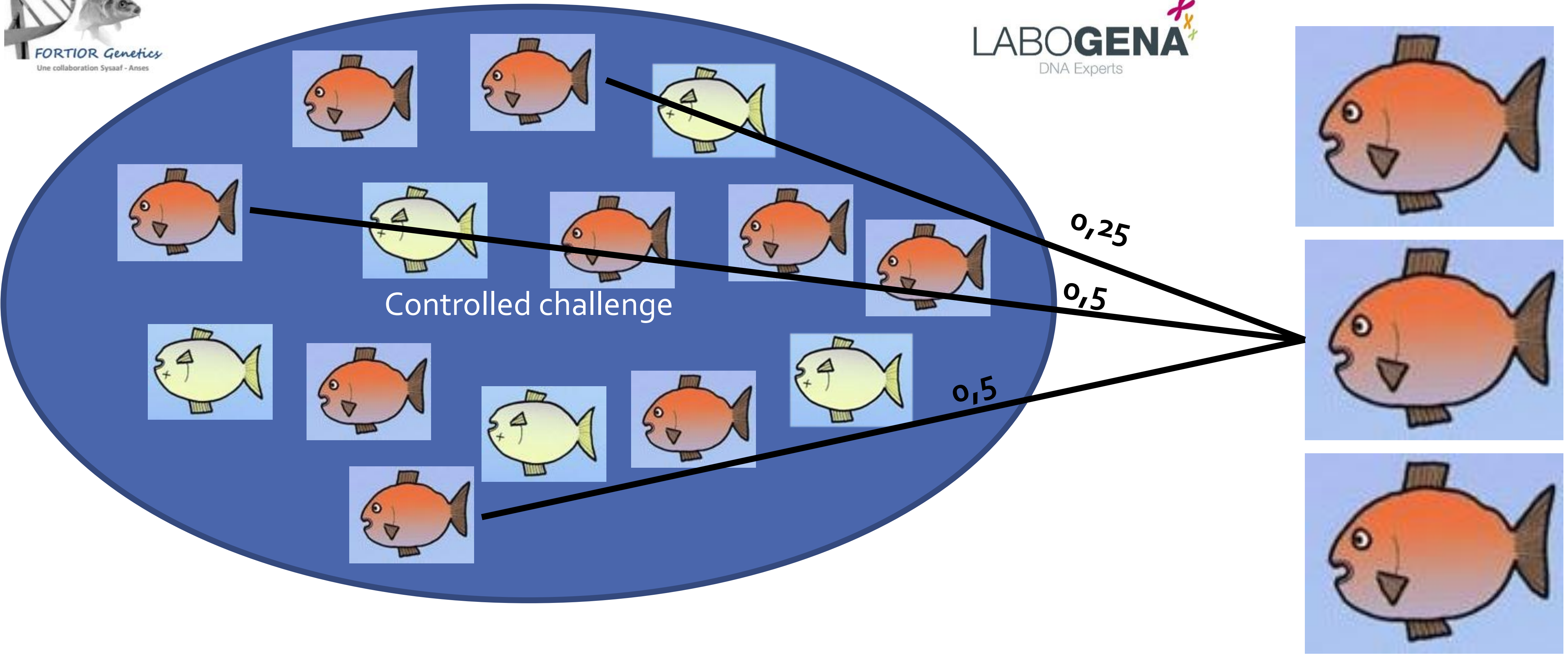

EBV

-1,2

1,6

Resistance to pathogens by controlled challenges on sibs

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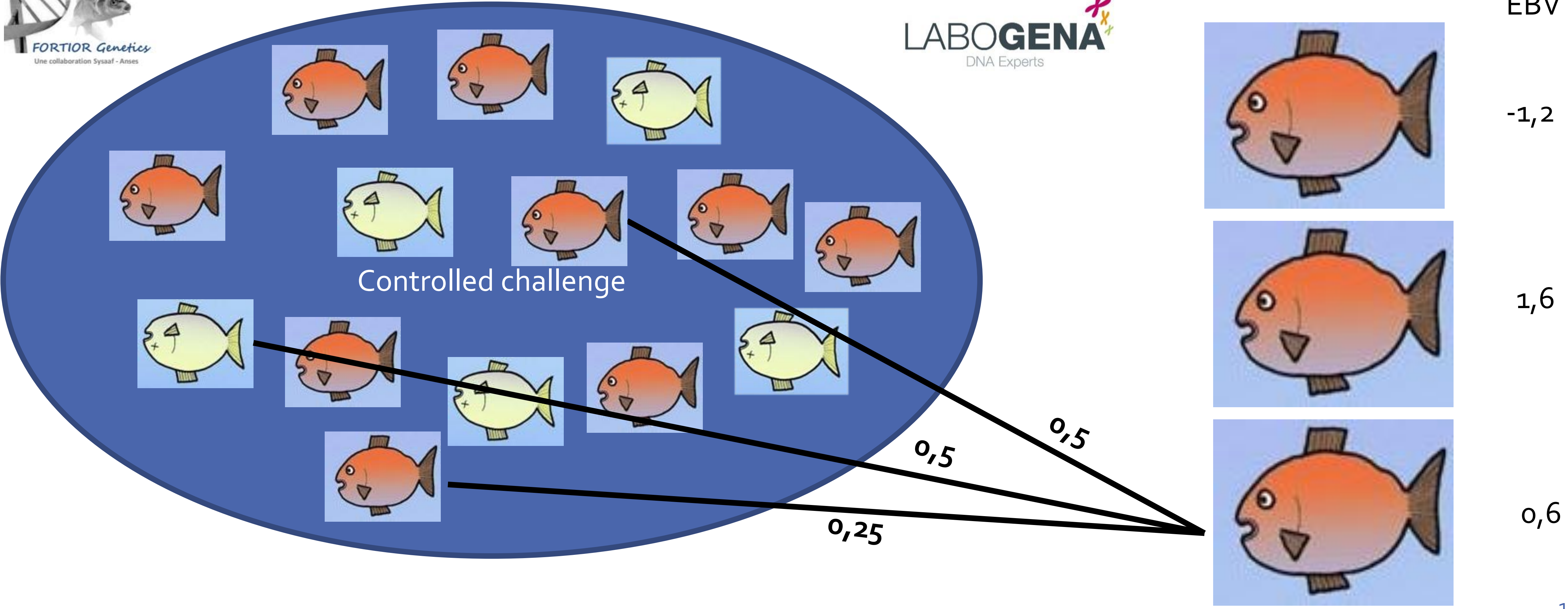
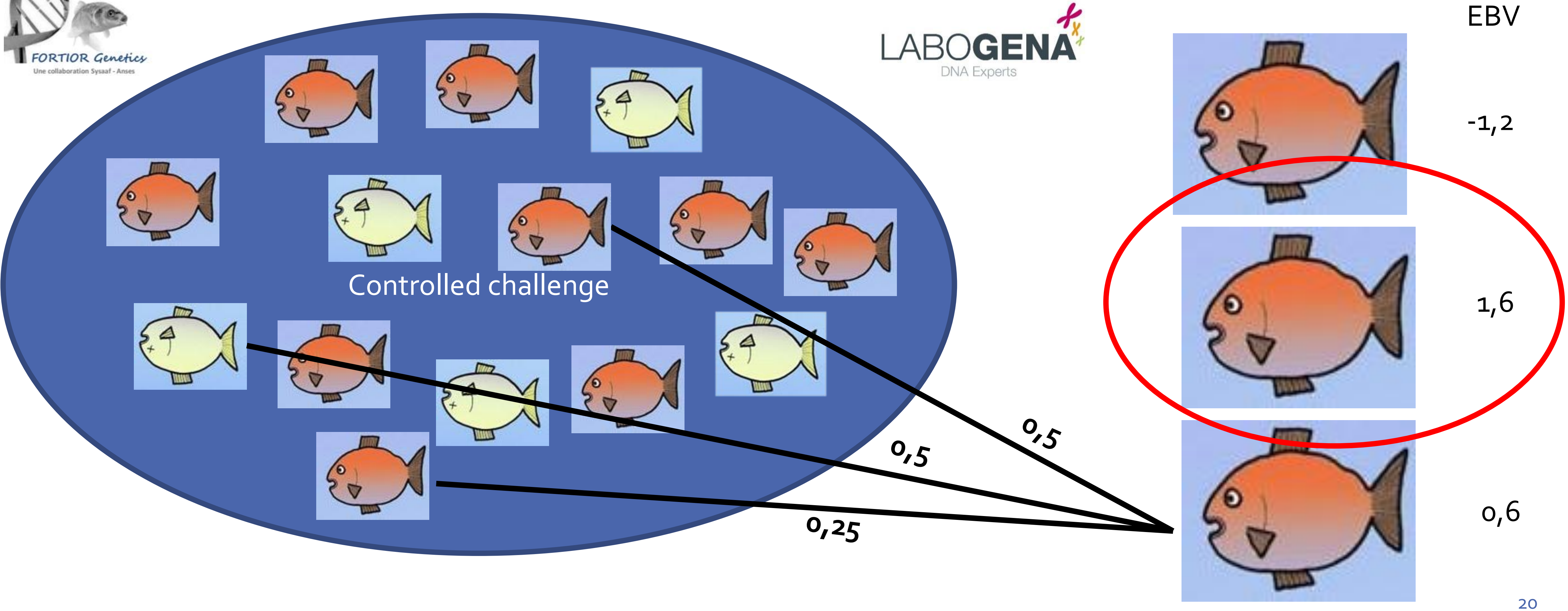


Resistance to pathogens by controlled challenges on sibs



DNA-parentage assignment in selection

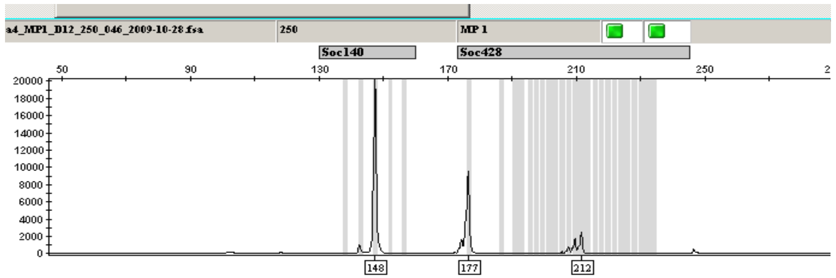
How ?

Microsatellite

Since 1990s,
8 to 15 microsat loci provide > 99% assignment power for finfish
High prevalence of null allele in molluscs multiplex sets of microsatellite (12 to 20 microsat in three multiplex)

Chromosome maternal : GGGCACACACAGCAGC (CA)3

Chromosome paternal : GGGCACACACACACAGCAGC (CA)5

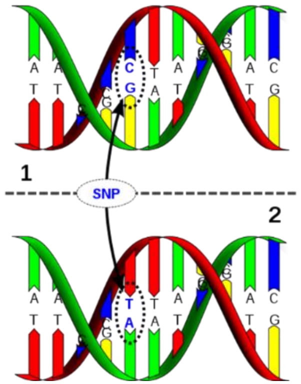


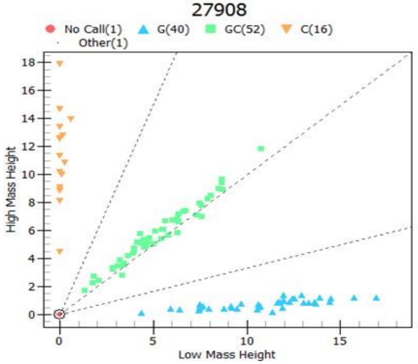
SNP (Single Nucleotid Polymorphism)

Since NGS method SNPs supplanting microsatellites
100-400 SNPs
High degree of polymorphism in mollusc genome
=> massive number of SNPs for marker design

Chromosome maternal : AGGGCAGGCAAGCAT

Chromosome paternal : AGGGCACGCAAGCAT





Numerous panels for parentage assignment

Panel for parentage assignment available in many breeding aquaculture species…

Pacific white shrimp (*Litopenaeus vannamei*)	**Rainbow trout (*Oncorhynchus mykiss*)**	Common carp (*Cyprinus carpio*)
Chinook salmon (*Oncorhynchus tshawytscha*)	Atlantic salmon (*Salmo salar*)	Yellowtail (*Seriola quinqueradiata)*
Kuruma prawn (*Marsupenaeus japonicas*)	Tiger shrimp (*Penaeus monodon*)	**Sea bream (*Sparus auratus*)**
Bighead carp (*Hypophthalmichthys nobilis*)	Coho salmon (*Oncorhynchus kisutch*)	**Turbot (*Scophthalmus maximus*)**
Siberian sturgeon (*Acipenser baerii*)	Sea cucumber (*Apostichopus japonicus*)	Japanese eel (*Anguilla japonica*)
European sea bass (*Dicentrarchus labrax*)	***Blue shrimp (Litopenaeus stylirostris)***	…

… and shellfish species

Grooved carpet shell (*Ruditapes decussatus*)	Triangle sail mussel (*Hyriopsis cumingii*)	Pearl oyster (*Pinctada fucata*)
Silver-lipped pearl oyster (*Pinctada maxima*)	**Green abalone (*Haliotis tuberculata*)**	Great scallop (*Pecten maximus*)
European flat oyster (Ostrea edulis)	Eastern oyster (*Crassostrea virginica*)	**Pacific oyster (*Crassostrea gigas*)**
South-African abalone (*Haliotis midae*)	Blue mussel (Mytilus galloprovincialis)	…

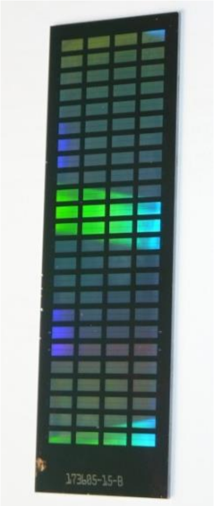
What’s next : Genomic selection

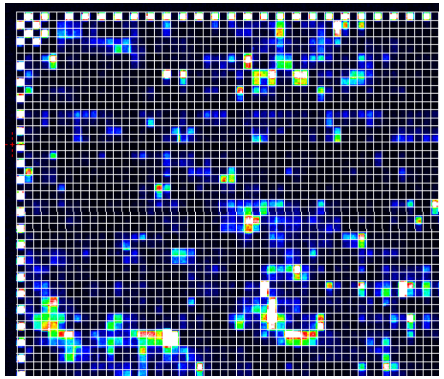
Why ?

- Improvement of accuracy of genetic indexes and decrease generation intervals
- Favorable effect on selection intensities
- New traits as selection criteria : hard to measure, low heritable, sex-limited, **lethal traits** (Hayes et al., 2009)
- Individuals rankings and not family rankings

How ?

High density chip, from 4K to 600K SNPs





Resistance to pathogens by controlled challenges on sibs – WITH GENOMICS !

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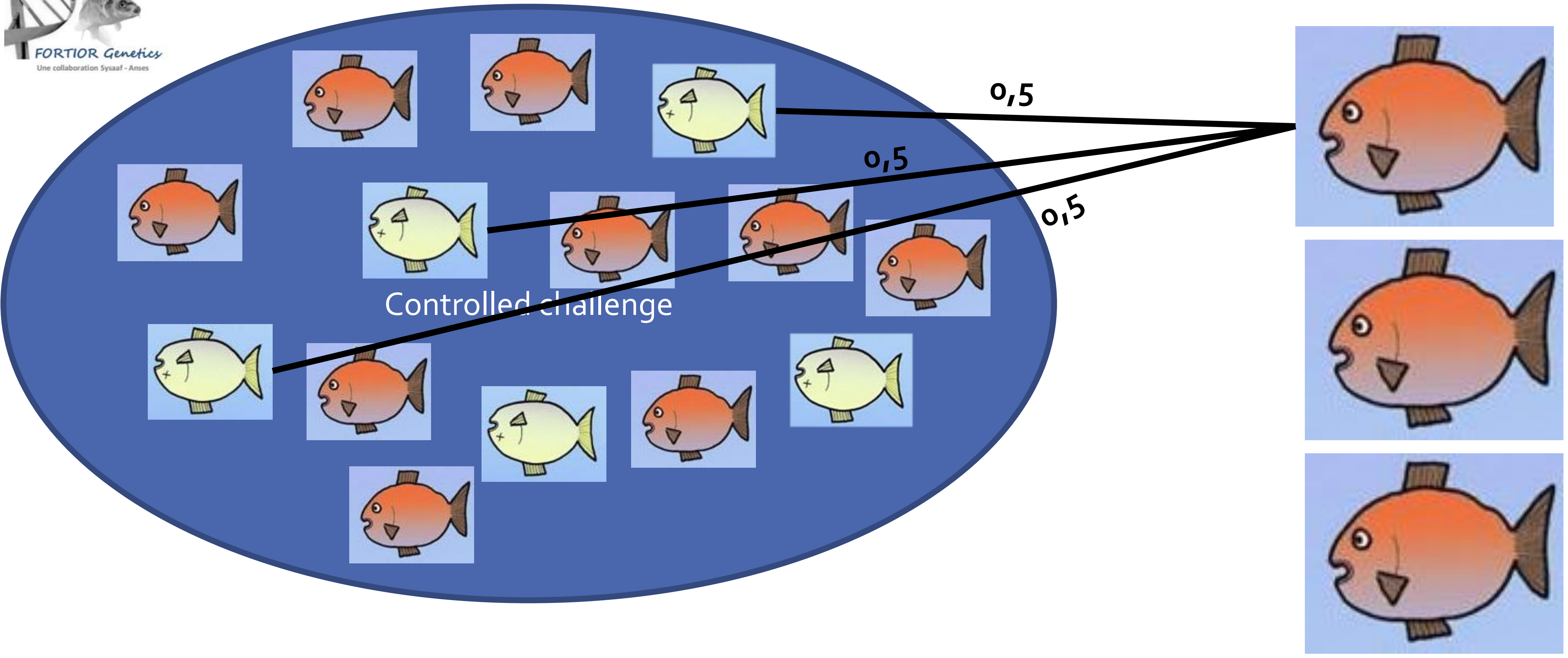

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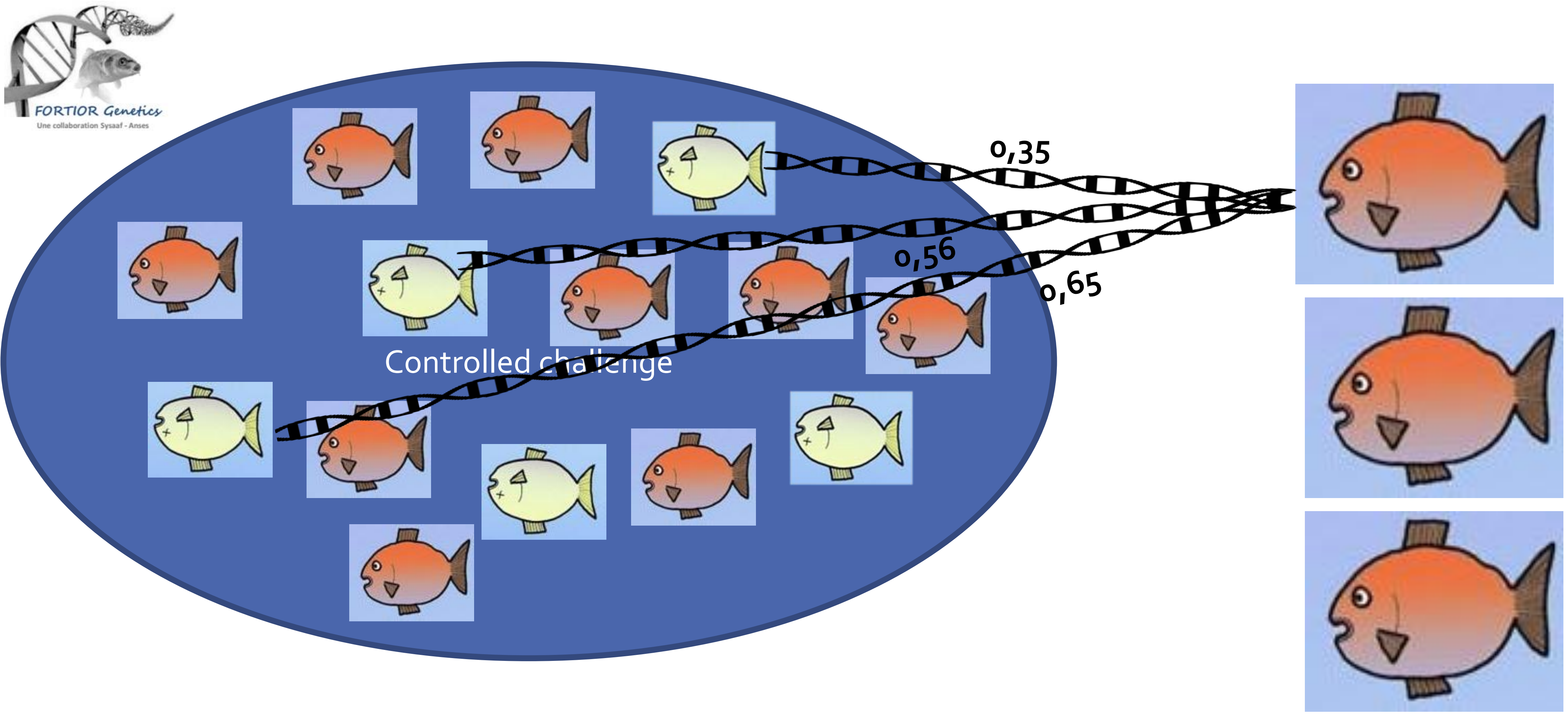


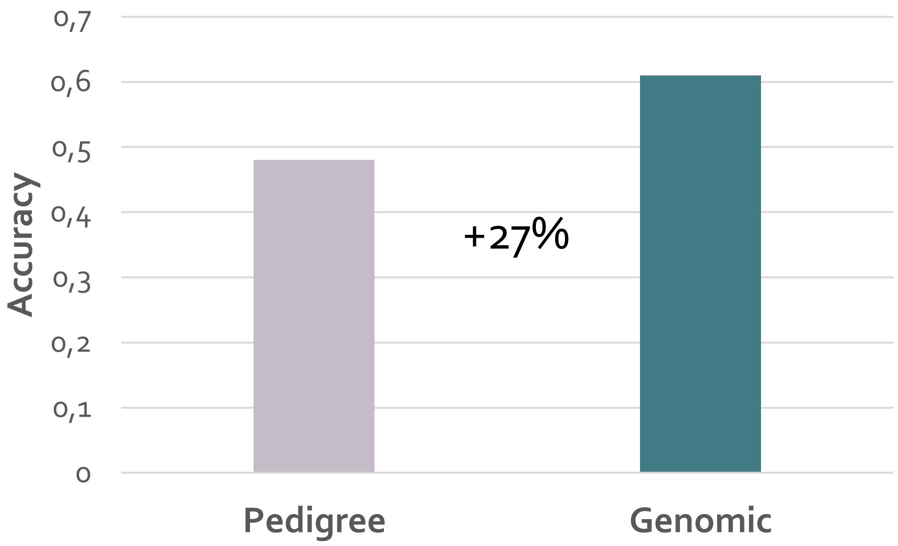
GEBV

-1,5

Genomic selection

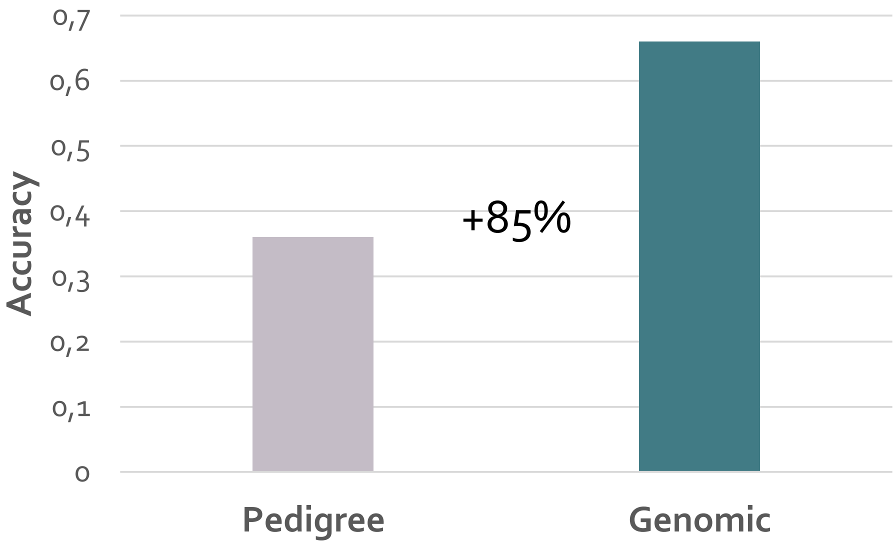
Examples in aquaculture :

Farmed Atlantic salmon resistance to sea lice
(Tsai et al., 2016)



h² = 0,22-0,33
Array: 35K (subset of the 132K array)
1119 individuals with phenotype and genotype

BCWD resistance in rainbow trout
(Vallejo et al. 2017)



h² = 0,33-0,37
Array: 35K
1473 individuals with phenotype and genotype
+ 6420 individuals with phenotype

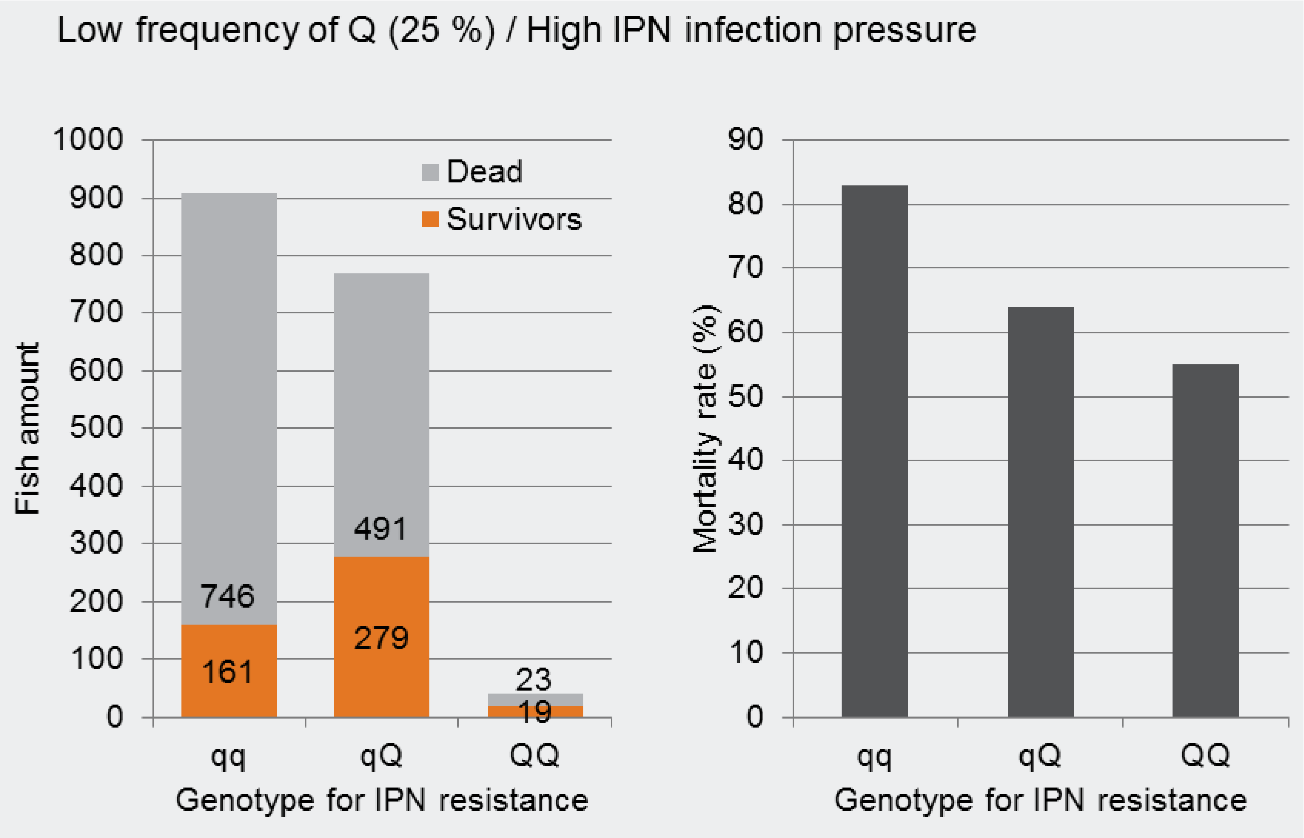
Available tools for genomic selection

High density public SNP arrays available in aquaculture species

Species	Number SNPs	Assay Type	Reference
Atlantic salmon (*Salmo salar*)	132K	Affymetrix Axiom	Houston et al., 2014
Nile tilapia (*Oreochromis niloticus*)	50K	Illumina	Yanez, ISGA 2018
Channel catfish (*Ictalurus punctatus*) & blue catfish (*Ictalurus furcatus*)	250K	Affymetrix Axiom	Liu et al., 2014
Channel catfish (*Ictalurus punctatus*) & blue catfish (*Ictalurus furcatus*)	690K	Affymetrix Axiom	Zeng et al., 2017
Rainbow trout (*Oncorhynchus mykiss*)	57K	Affymetrix Axiom	Palti et al., 2015
European sea bass (*Dicentrarchus labrax*)	57k	Affymetrix Axiom	Allal et al. ISGA 2018
Pacific oyster (*Crassostrea gigas*)	190K	Affymetrix Axiom	Qi et al., 2017
Pacific oyster (*Crassostrea gigas*) & European flat oyster (*Ostrea edulis*)	41K / 15K	Affymetrix Axiom	Gutierrez et al., 2017

Next challenges for shellfish breeding

- Estimation of heritabilities and identification of QTLs for interest traits : production traits, disease resistance…



From documentation of the product :
« AquaGen® Rainbow QTL-innOva® »

Next challenges for shellfish breeding

- Estimation of heritabilities and identification of QTLs for interest traits : production traits, disease resistance…
- Development of methods to phenotype shellfish (**controlled challenges…**)
- Adaptation of breeding programs to new tools – implementing genomic selection
- Genomic ressources for other commercially important shellfish species

The Sysaaf (dream) team

Thank you for your attention

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