



Evaluation of the genetic variation in captive meagre broodstocks

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(Led by FCPCT_ ULPGC)

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Universidad de Las Palmas de Gran Canaria



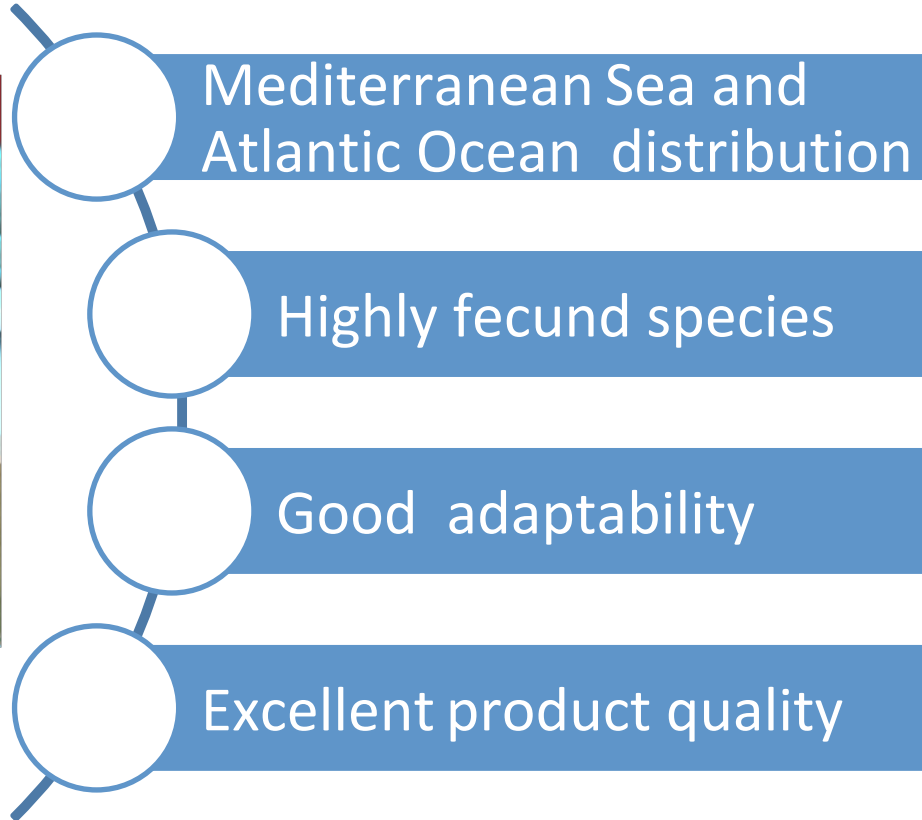
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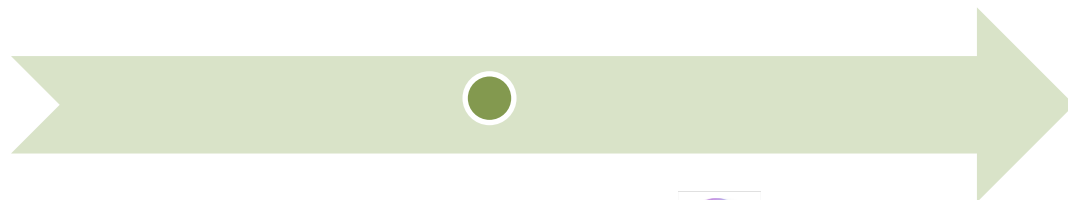
INTRODUCTION



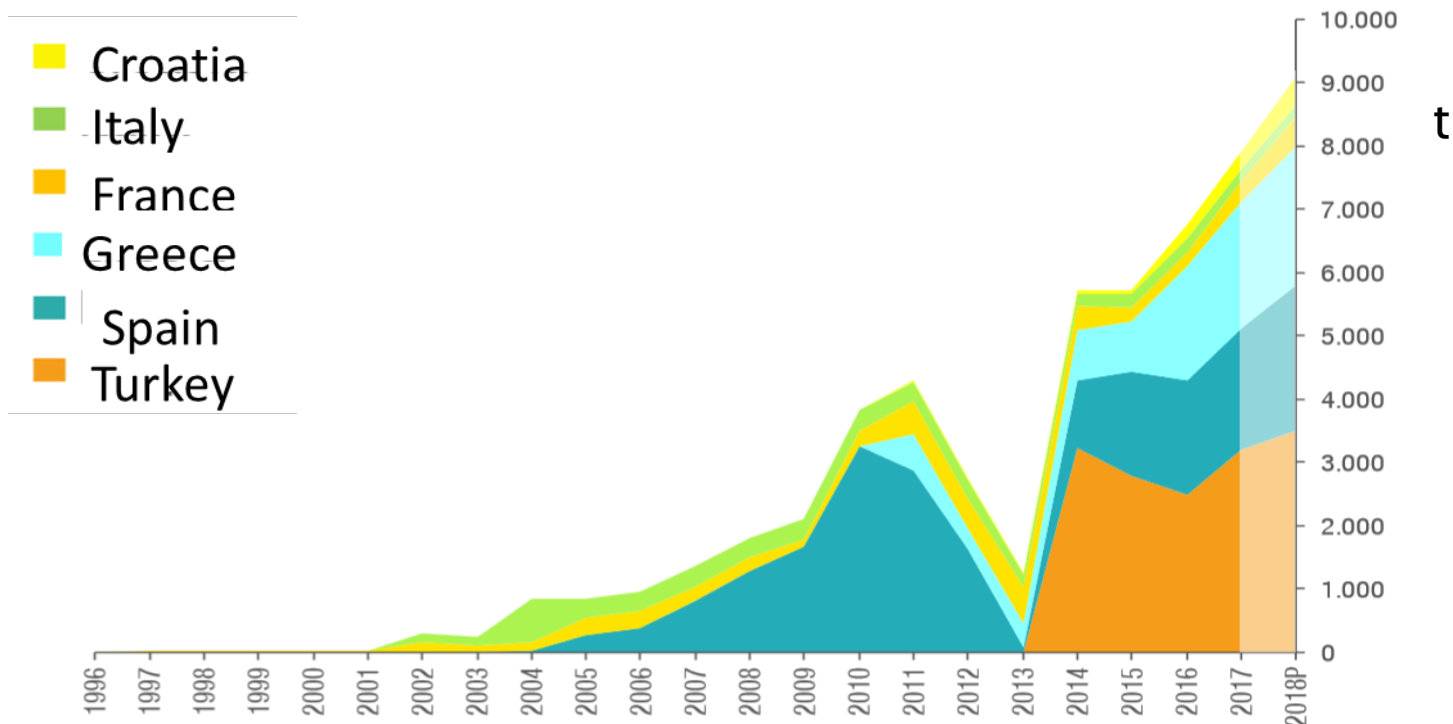
Argyrosomus regius
Scianidae Family



Diversification



PRODUCTION



Evolution of World Meagre Aquaculture Production (without Egypt) from 1996 to 2018. Data from FAO, FEAP, APROMAR. (APROMAR, 2018)

INTRODUCTION

Studies on all aspects of the life-cycle in captivity



INTRODUCTION

GENETIC TOOLS

in order to add value to
the meagre products

GENETIC
IMPROVEMENT



INTRODUCTION

GENETIC IMPROVEMENT

* Base population variability

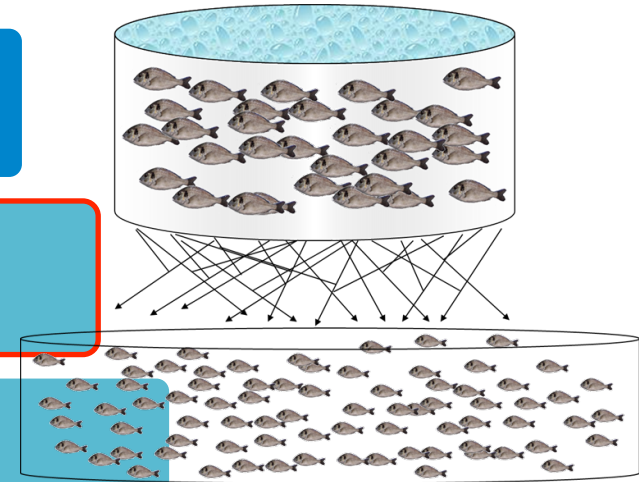
* Genealogy

* Selected traits

* Genetic parameters

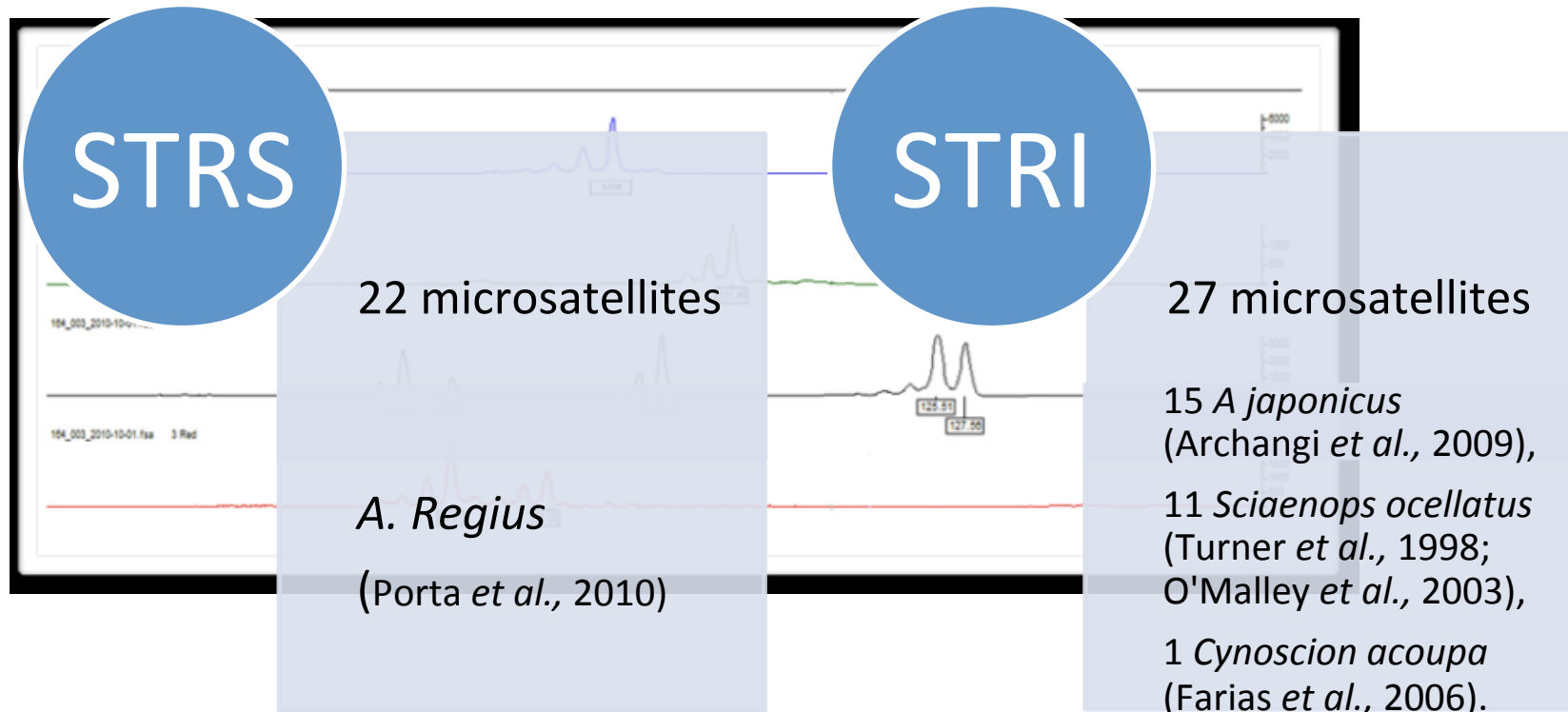
* Genotype-environment interactions

* Breeding value



INTRODUCTION

Two new multiplex of microsatellites Soula et al. (2011)



First estimates of genetic parameters for growth traits reporting a low additive genetic variation, due to the high relationship between animals within populations.



Objective

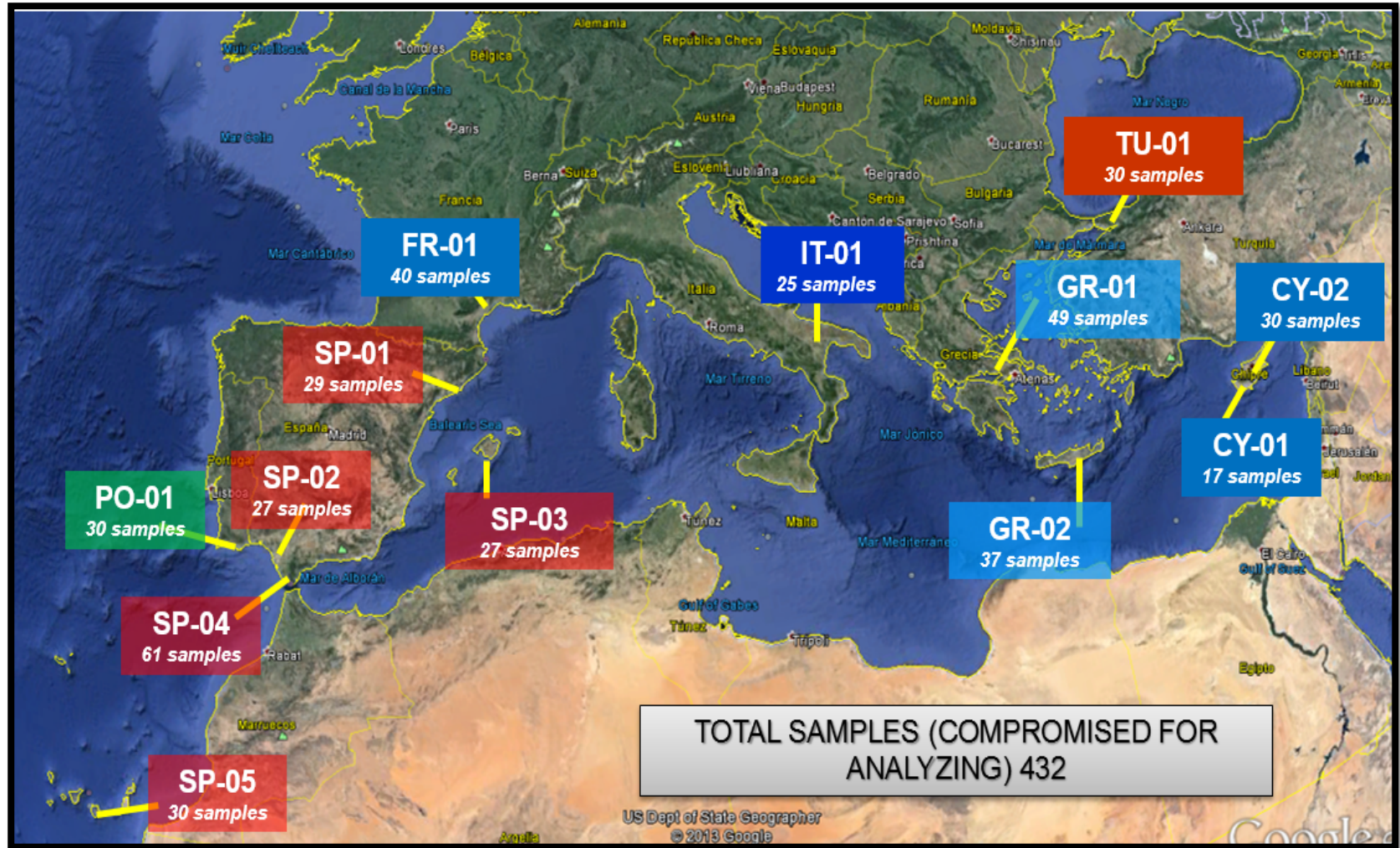
Evaluate the genetic variation in the available captive broodstocks of meagre

It includes breeders of private companies and research centres

Description of work

Describe the status of the identified bottlenecks through the genetic characterization of captive broodstocks

MATERIALS AND METHODS



Geographic distribution of meagre studied samples in Europe, from Canary Islands to Cyprus



MATERIALS AND METHODS

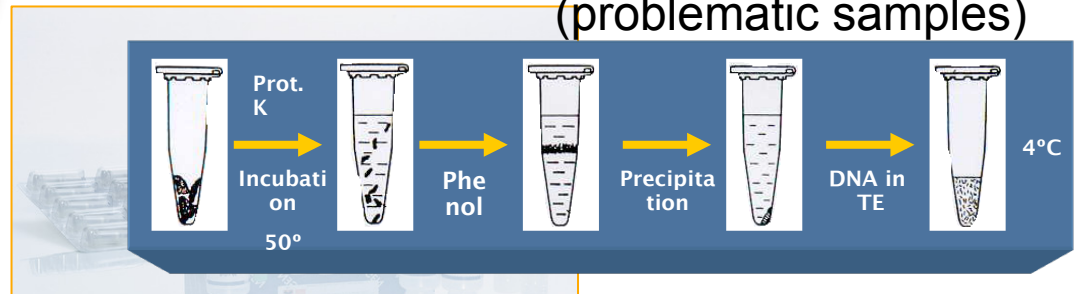
Caudal fin samples



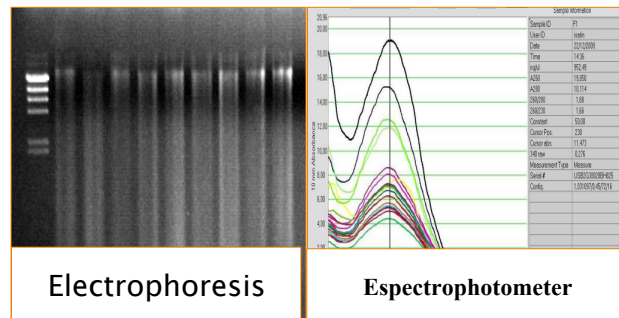
DNA extraction

DNeasy kit (Qiagen)

Phenol-chloroform method
(problematic samples)



DNA quality and quantity



Electrophoresis

Espectrophotometer

✓ 1% Agarose gel GelRed[™] staining

✓ Nanodrop 1000



MATERIALS AND METHODS

Microsatellites

18 microsatellites

Multiplex A Soula et al., 2011						
Multiplex	Marker	Allelic range	5' end Modif	Target species	Acc. No	Motif
STRI	Cac mic14	74-78	5' 6-FAM	<i>Argyrosomus</i>		
STRI	UBA54	127-153	5' 6-FAM	<i>japonicus</i>	EF462926	CA
STRI	UBA50	86-120	5' 6-FAM	<i>japonicus</i>	EF462924	CA
STRI	UBA53		5' VIC	<i>japonicus</i>	EF462925	CA
STRI	Soc431	123-155	5' VIC	<i>Sciaenops ocellatus</i>	AY161032	CA-GT
STRI	UBA042	70-76	5' NED	<i>Argyrosomus</i>	EF462920	GTC
STRI	UBA853	92-112	5' NED	<i>japonicus</i>	EF462929	CT
STRI	UBA5	120-124	5' NED	<i>Argyrosomus</i>	EF462917	CT
STRI	Soc405	113-125	5' PET	<i>Sciaenops ocellatus</i>	AY161014	CA
STRI	UBA6	132-154	5' PET	<i>Argyrosomus</i>	EF462918	CA
STRI	UBA6		5' PET	<i>japonicus</i>	EF462918	CA

Interspecific microsatellites

STRS	GCT15	72-78	5' 6-FAM	<i>Argyrosomus regius</i>	GU724793	GCT
STRS	GA16	82-92	5' 6-FAM	<i>Argyrosomus regius</i>	GU724798	GA
STRS	GA17	106-126	5' 6-FAM	<i>Argyrosomus regius</i>	GU724797	GA
STRS	CA13	94-116	5' VIC	<i>Argyrosomus regius</i>	GU724791	CA
STRS	GA6	98-123	5' NED	<i>Argyrosomus regius</i>	GU724792	CA
STRS	CA3	86-106	5' NED	<i>Argyrosomus regius</i>	GU724783	CA
STRS	CA14	121-133	5' NED	<i>Argyrosomus regius</i>	GU724796	GA
STRS	GA2B	77-111	5' PET	<i>Argyrosomus regius</i>	GU724794	GA

Specific microsatellites



MATERIALS AND METHODS

Multiplex PCRs

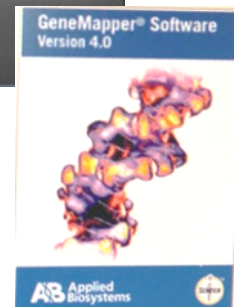
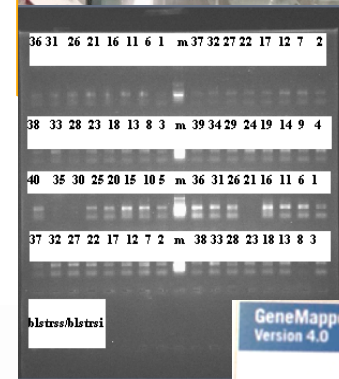
❖ Soula et al., 2011

Amplification

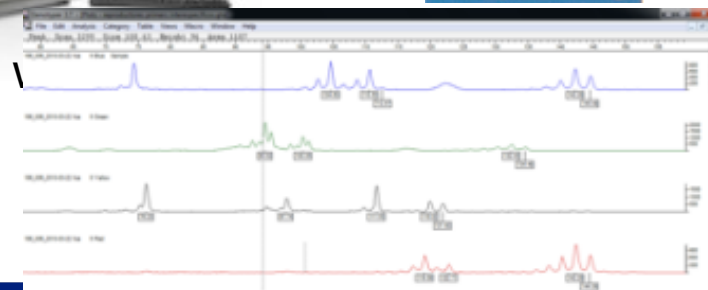
Check 2% agarose gel

Automatic capillar electrophoresis

ABI Prism-3730-XL Genetic Analyzer (*Applied Biosystem*[®])



Electropherograms and genotypes were evaluated with GeneMapper (v4.0) (*Applied Biosystems, Inc.*) software.



MATERIALS AND METHODS

Data analysis

- ✓ Genetic variability parameters

allele number, genotypes, heterozygosities

GENEPOP 4.2

- ✓ To verify geographical subdivision of studied populations



a molecular variance **AMOVA** analysis was conducted

ARLEQUIN 2.0

- ✓ Structuration of populations

GENETIX 4.05



RESULTS AND DISCUSSION

Genetic parameters

Total number of gene copies, alleles and observed and expected heterozygosities

	AVERAGES			
POPULATION	Nº GENE COPIES	Nº ALLELES	HET.OBS.	HET.EXP.
PO-01	27.8	1.8	0.261	0.259
SP-01	54.4	5.2	0.585	0.598
SP-02	45.8	6.3	0.548	0.625
SP-03	51.6	4	0.561	0.505
SP-04	101.5	7.1	0.532	0.595
SP-05	56.3	2.8	0.519	0.446
FR-01	59.5	5.3	0.433	0.507
IT-01	46.3	3.4	0.547	0.502
GR-01-F1	26.88	2.5	0.453	0.449
GR-01-F2	54.7	3	0.388	0.402
GR-02-F1	34.5	3.4	0.473	0.506
GR-02-F2	28.1	2.4	0.493	0.44
TU-01	57.8	2.5	0.375	0.375
CY-01	24	3	0.495	0.509
CY-02	35.3	2.7	0.469	0.455
Arithmetic mean		3.69	0.48	0.48
Weighted mean		4.13	0.48	0.49



RESULTS AND DISCUSSION

- ✓ **The variation of the base population** is essential because the genetic variability influences to the response of selection in short and long term (Falconer and Mackay, 2001).



In gilthead seabream, studies on genetic characterization of populations under commercial exploitation and in research centres, using microsatellites, is very high.

	Ho	Alleles per locus
Gilthead seabream Batargias et al. (1997, 1999)	0.875 one of the highest in teleosts	16,5 (6 loci)
Gilthead seabream Alarcon et al. (2004)	0.864 natural 0.845 culture	15,8 (3 loci)
Gilthead seabream De innocentis et al. (2005)	0.74 0.79 in two companies	17 (4 loci)

RESULTS AND DISCUSSION



- ✓ In meagre only two studies have been conducted to study its genetic structure on wild populations in Atlantic and Mediterranean Sea and commercial exploitation.

	Ho	Alleles per locus
Meagre THIS STUDY	0.48 18% lower	4 (18 loci)
	Ho	Alleles per locus
Meagre Haffray et al. (2012)	0.57 wild	13,2 (11 loci)
		around 3 times lower THIS STUDY
Meagre Haffray et al. (2014)	0.52 Captive French industry	8,5(11 loci)
		around 2 times lower

RESULTS AND DISCUSSION

Genetic parameters

Exclusive allele

7 populations

- ✓ SP-04 and SP-02 the populations with more exclusive alleles, 6 and 4 respectively.

Effective population size

86.79c

3.1% CV

- ✓ under an ideal population, 43.4 males // 43.4 females.
- ✓ It was higher to the minimum recommended to minimize inbreeding depression (50), but lower than the minimum suggested for maintaining enough evolution capacity (500).

RESULTS AND DISCUSSION

Molecular analysis of Variance (AMOVA)

The AMOVA was run for *loci* 1–8

AMOVA for European populations of meagre

Source of variation	Sum of squares	Variance components	Percentage variation
Among populations	358.311	0.47282	18.19220
Within populations	1567.290	2.12622	81.80780
Total	1925.601	2.59905	

Average F-Statistics over all loci
Fixation Indices
FST : 0.18192

High differentiation among populations studied and low gene flow

Other authors suggested that this high genetic structuration reveals a different genetic origin of each population and they need to consider their management regionally

RESULTS AND DISCUSSION

Percentage Variation for locus

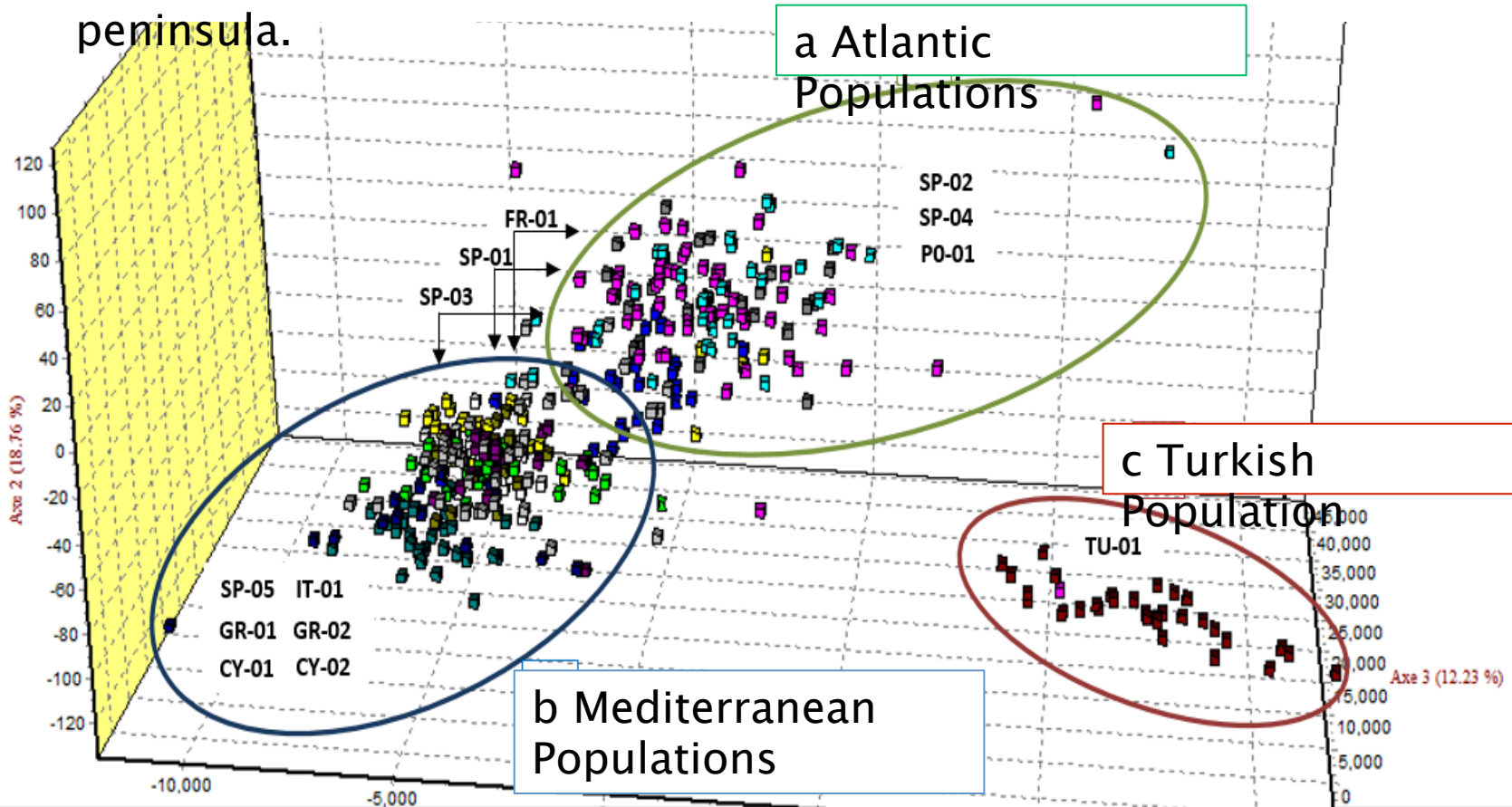
Locus	Among Populations:			Within Populations:				Fixation indices:		
	SSD	d.f.	Va	% variation	SSD	d.f.	Vb	% variation	FST	P-value
1	59.64035	14	0.07853	19.53170	241.68904	747	0.32355	80.46830	0.19532	0.00000
2	72.44379	14	0.09767	27.13830	195.88823	747	0.26223	72.86170	0.27138	0.00000
3	35.87438	14	0.04407	11.59604	251.61908	749	0.33594	88.40396	0.11596	0.00000
4	63.25199	14	0.08802	30.69286	144.88645	729	0.19875	69.30714	0.30693	0.00000
5	31.02726	14	0.04086	14.85683	169.30878	723	0.23418	85.14317	0.14857	0.00000
6	37.82815	14	0.04842	13.55425	226.34431	733	0.30879	86.44575	0.13554	0.00000
7	21.30366	14	0.02571	9.44332	181.19101	735	0.24652	90.55668	0.09443	0.00000
8	36.94136	14	0.04955	18.64083	156.36352	723	0.21627	81.35917	0.18641	0.00000

This fixation index was independent of *loci*

RESULTS AND DISCUSSION

Geographic structuration

✓ The FCA grouping was in concordance with geographic distribution of the European meagre populations



CONCLUSION

- ✓ The significant F_{st} reported, shows a low genetic flow among captive populations studied, producing fragmentation of populations and increasing the effect of the genetics stochastic processes.
- ✓ The main consequence of reduced N_e in meagre populations is related directly with the inbreeding depression, reducing of evolution powerful and enabling the extinction risk.
- ✓ A biological explanation of this genetic structuration of meagre populations could be related with its high fecundity and variance in reproductive success which can decrease N_e , where only small number of families survive to produce descendants.

CONCLUSION

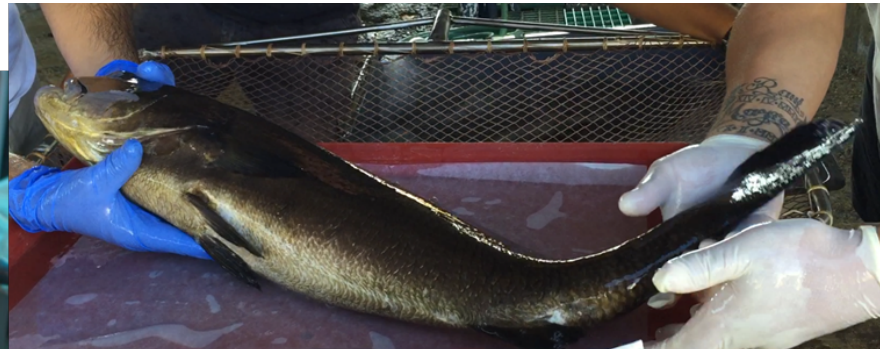
- ✓ One of the most important questions is if this genetic structuration reported on captive meagre populations has any direct consequences or effects on fish performances...
- * is well known that the loss of genetic diversity (low number of alleles, heterozygosity, etc..) due to effective size reduction is affecting to the heritability of quantitative traits Introduction (Franklin 1980; Frankham et al. 2003), and in consequence to change the capacity of populations, wilds or captives.
- * In this way, would be very interesting to constitute new experiments to measure, under the same environmental conditions, the performances of all lines or populations studied.

CONCLUSION

- * This potential genetic differentiation on quantitative genes and traits would be a magnificent tool to constitute the best available base population for a selective breeding program at European scale, especially because meagre is a species in clear disadvantage, in terms of genetic starting point, compared to other important species within the European aquaculture.
- * On the other hand, it is conditioned for the interaction G_E, which has to be studied as well, because it is unknown if populations of meagre under commercial exploiting are adapted to its environmental conditions, which would oblige to established local genetic breeding programs, more than a breeding program to the European scale.



**THANKS FOR YOUR
ATTENTION**



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