

# Investigating the Genetics of Sex Determination in European Sea Bass (*Dicentrarchus labrax*) using RADseq

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# Outline

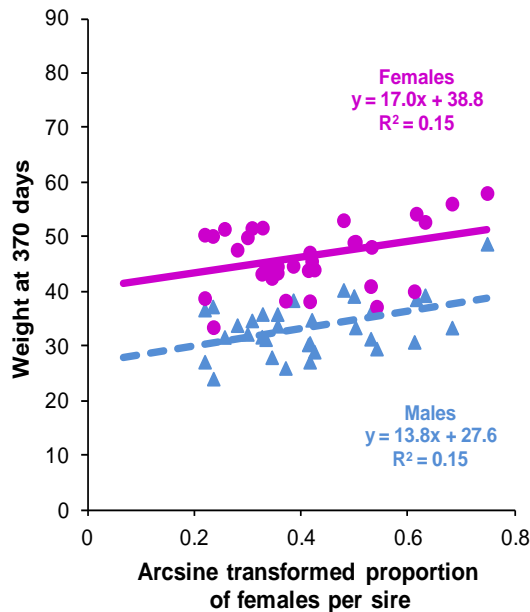
- \* Brief intro to sea bass aquaculture industry
- \* Introducing RAD-seq and some of its key points
- \* Application of RAD-seq in studying sex determination

# Sexual dimorphism

- \* Females grow faster (~40% higher body weight) and mature later
- \* Under culture conditions a high percentage of males (70-90%) is commonly observed
- \* Temperature influences sex ratio (temperatures  $>17^{\circ}\text{C}$  early in development favouring males)



# European seabass: polygenic + temperature effects



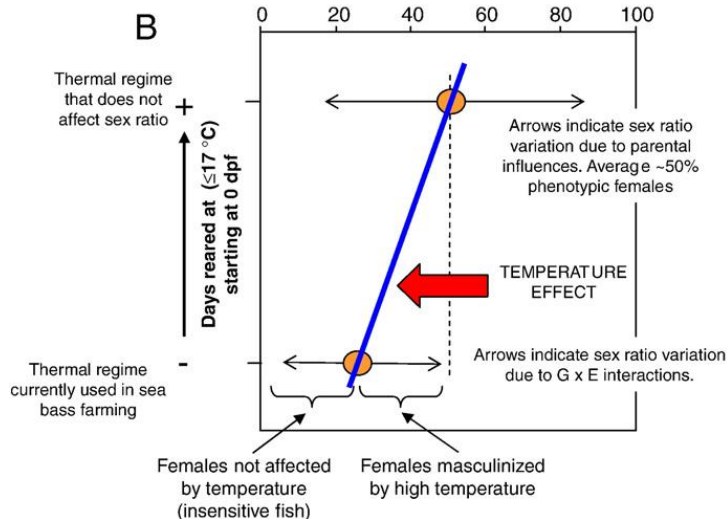
$h^2$  of sex tendency =  $0.62 \pm 0.12$   
 Gen corr with BW:  $r_A = 0.50 \pm 0.09$

Select for faster growth



Increased proportion of females

Females grow faster and mature later



Change early thermal regime

Vandeputte et al. (2007)  
 Navarro-Martin et al. (2009)

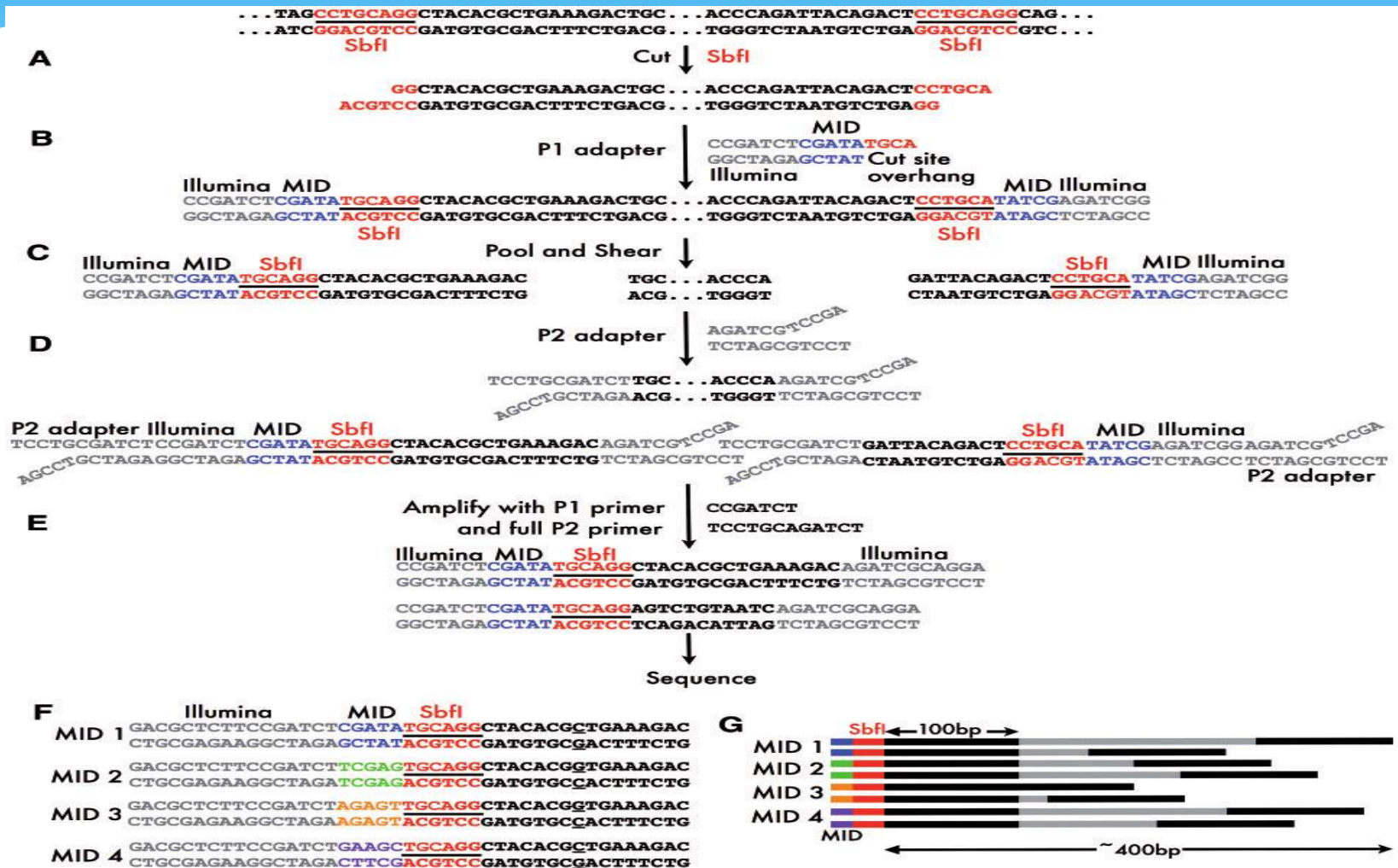
# Genomic resources

- \* Reference genome available since 2014
- \* Linkage map based on microsatellites
- \* No SNP chip
- \* No attempt of medium-high density genotyping at the moment





# RAD Sequencing

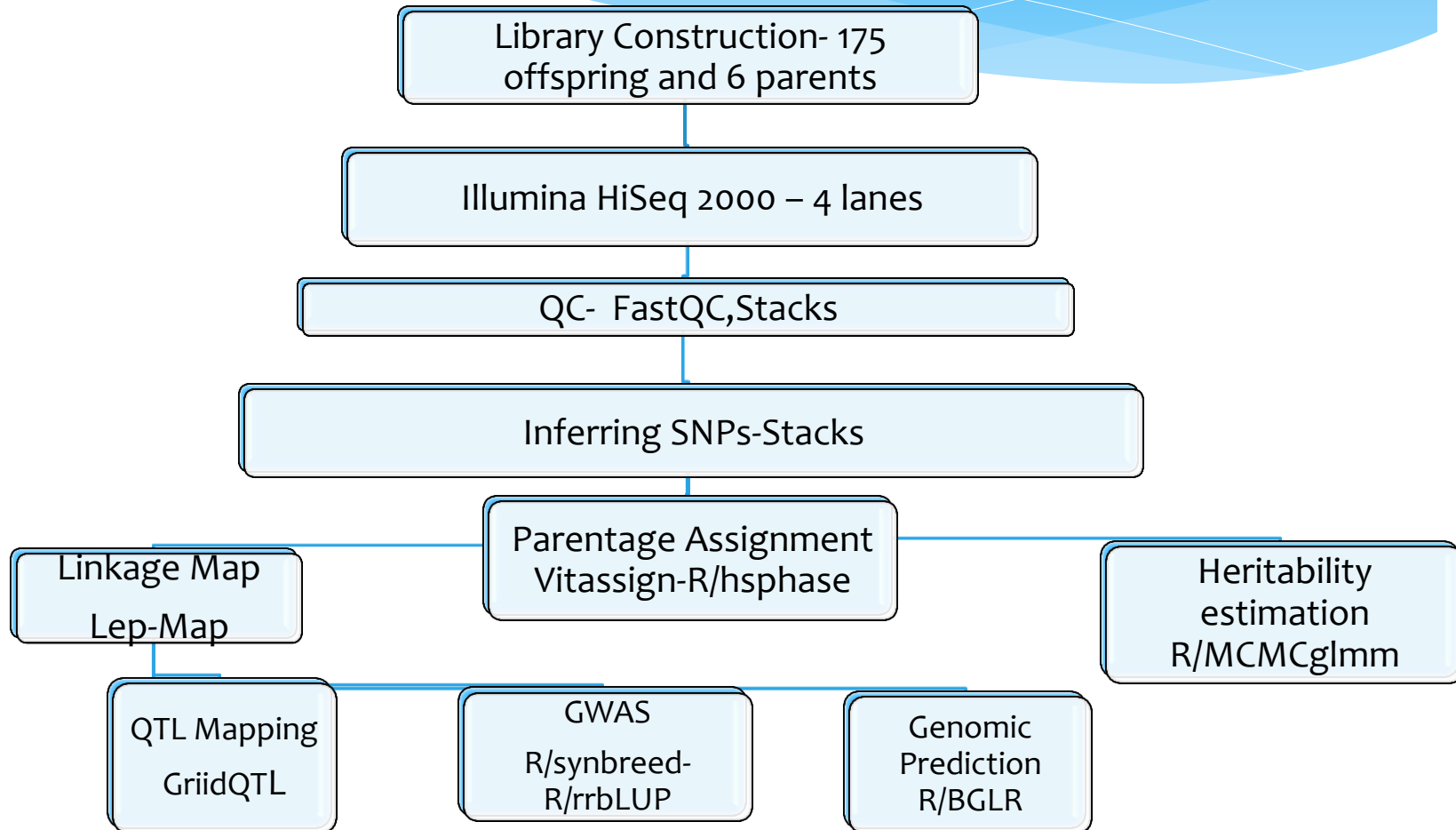


# General stats (175 F<sub>2</sub> progeny)

Dam	Sire	Family	Contribution %	Female Percentage%	Average Weight (g)±se	Average Length (mm)	P-value
Dam 1	Sire 1	1	7.4%	69	194.2 (58.7)	256.5 (20.9)	0.27
Dam 1	Sire 2	2	10.8%	26	179.3 (34.1)	249.5 (15.3)	0.066
Dam 1	Sire 3	3	9.7%	71	181.9 (65.2)	248.6 (31.4)	0.15
Dam 1	Sire 4	4	17.7%	45	178.2 (48.2)	248.7 (19.9)	0.719
Dam 2	Sire 1	5	9.1%	69	148.1 (37)	231.5 (16.1)	0.21
Dam 2	Sire 2	6	12.6%	36	150.2 (54.9)	233.0 (26.8)	0.28
Dam 2	Sire 3	7	10.9%	84	170.0 (67.5)	240.2 (28.3)	0.0059**
Dam 2	Sire 4	8	21.7%	34	160.5 (52.8)	239.6 (24.7)	0.074

\*  $\alpha=0.05$  \*\*  $\alpha=0.01$  \*\*\*  $\alpha=0.001$

# Workflow





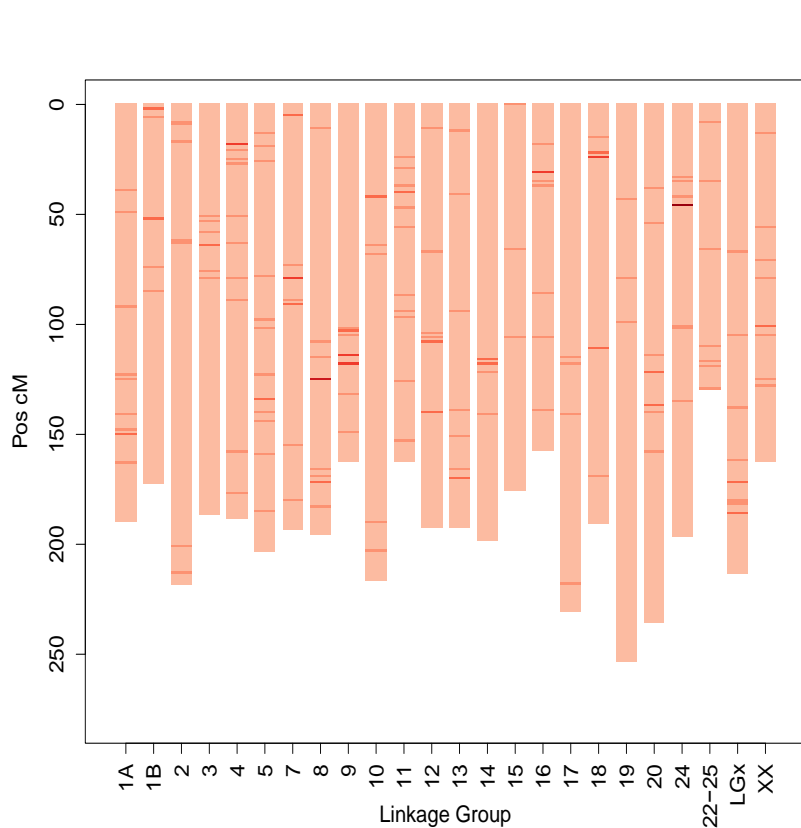
# Sequenced RAD-tags

- \* 1,156,659,542 raw reads
- \* QC
  - \* quality score under 30
  - \* ambiguous barcodes
  - \* orphaned paired-end reads
- \* 76.7% retained
  - \* 86,927,866
- \* 56,696 unique RAD-tags
- \* RAD-tags retrieved in at least 75% and carrying just one or two SNPs

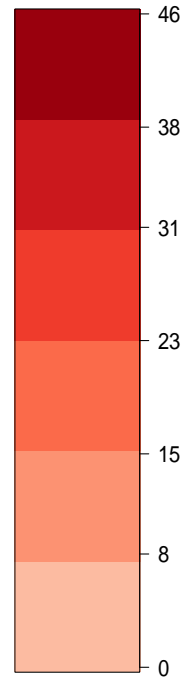
# Parentage Assignment-Heritability estimation

- \* All 175 offspring were uniquely assigned to parental pairs
- \* Heritability estimated on the liability scale using R/MCMCglmm
- \* Vitassign 200 SNPs
- \*  $h^2 = 0.47$
- \* R/hsphase allowed for 3% genotypic error

# Sea bass SNP linkage map



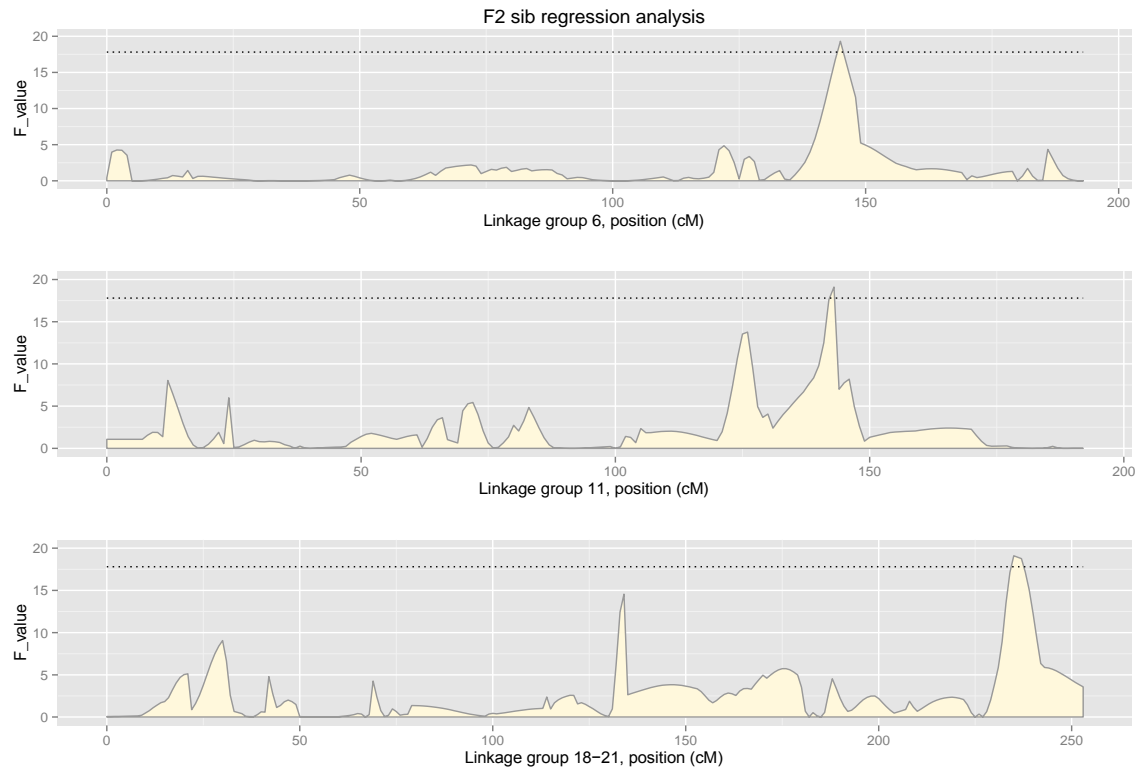
No. of SNPs  
per cM



- \* Lep-Map software
- \* 6,706 SNPs
- \* 24 LGs
- \* Correspondence with reference genome
- \* 856 SNPs (>10%) located on unanchored contigs

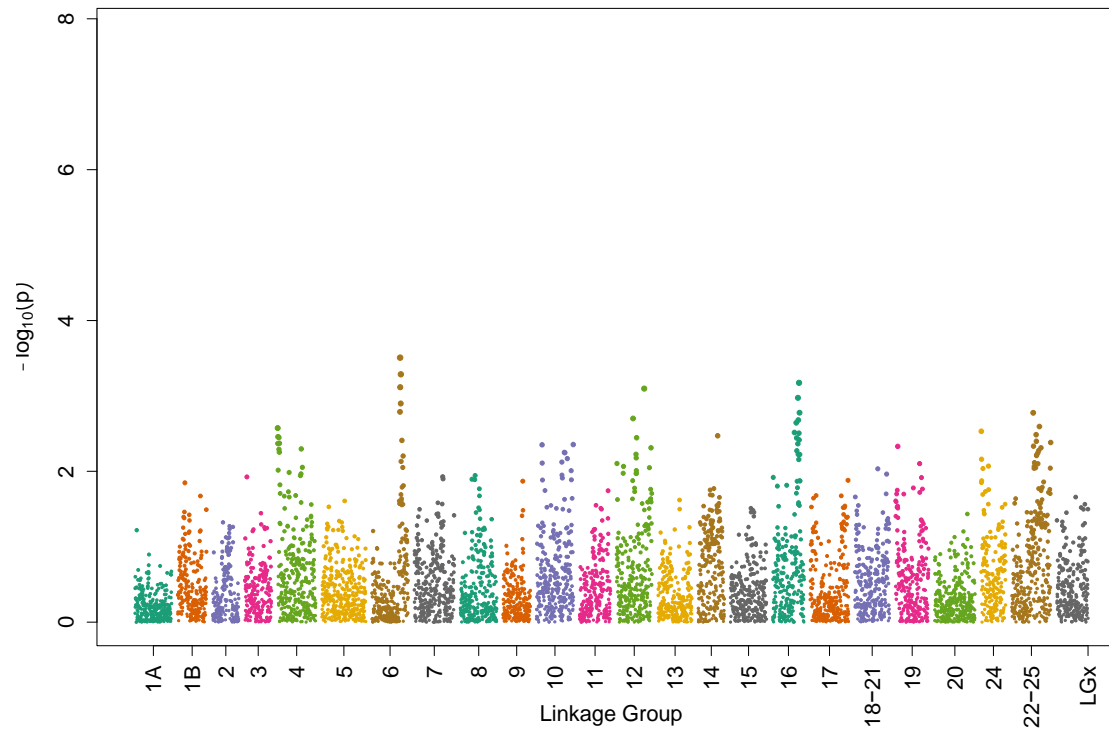
Chromosome (seabass_v1.0)	No. markers	Unique Positions	Length (cM)
LG1A	302	125	191.61
LG1B	238	84	163.33
LG2	216	96	204.47
LG3	219	89	174.05
LG4	315	119	191.19
LG5	363	147	206.11
LG6	299	133	243.5
LG7	330	126	245.49
LG8	301	90	156.75
LG9	226	108	259.7
LG10	305	108	218.23
LG11	255	98	192.49
LG12	292	105	181.85
LG13	280	140	188.76
LG14	217	102	216.2
LG15	298	110	173.17
LG16	259	137	227.55
LG17	318	104	199.54
LG18-21	286	147	210.52
LG19	272	111	202.01
LG20	333	113	229.37
LG24	203	76	164.14
LG22-25	321	117	194.33
LGx	258	91	182.57
	6,706	2,676	4,816.93

# Putative sex-determining QTL



# SNPs associated with phenotypic sex

GWAS European sea bass sex determination





# Genomic prediction – phenotypic sex

Proportions of offspring with correctly assigned sex in the validation sets (25 animals; 100 replicates) and testing of prediction deviations from expected by chance using t-tests.

Model	Assigned Correctly (%)	P-value
pBLUP	64	$2.2 \times 10^{-16}$
GBLUP	58	$2.913 \times 10^{-15}$
BayesA	59	$3.364 \times 10^{-15}$
BayesB	58	$5.394 \times 10^{-14}$
BayesC	59	$2.16 \times 10^{-16}$
BayesLasso	58	$8.056 \times 10^{-16}$

# Genomic prediction - length

Mean accuracies of predicted length for animals in the validation sets (25 animals; 100 replicates).

Model	Mean accuracy (s.e.)
pBLUP	0.32 (0.018)
GBLUP	0.37 (0.017)
BayesA	0.37 (0.02)
BayesB	0.38 (0.016)
BayesC	0.38 (0.015)
BayesLasso	0.38 (0.018)

# Conclusions

- \* High density SNP linkage map developed – 6700 markers
- \* Three QTL associated with sex, but barely significant
- \* No individual SNPs showed significant association with sex
- \* Supports polygenic hypothesis for sea bass
- \* No advantage of SNP-based models of prediction of sex compared to pedigree-based models
- \* Advantage of SNP-based models of prediction of length compared to pedigree-based models