Mapping QTL for Resistance to Summer Mortality in the Pacific Oyster, *Crassostrea gigas*

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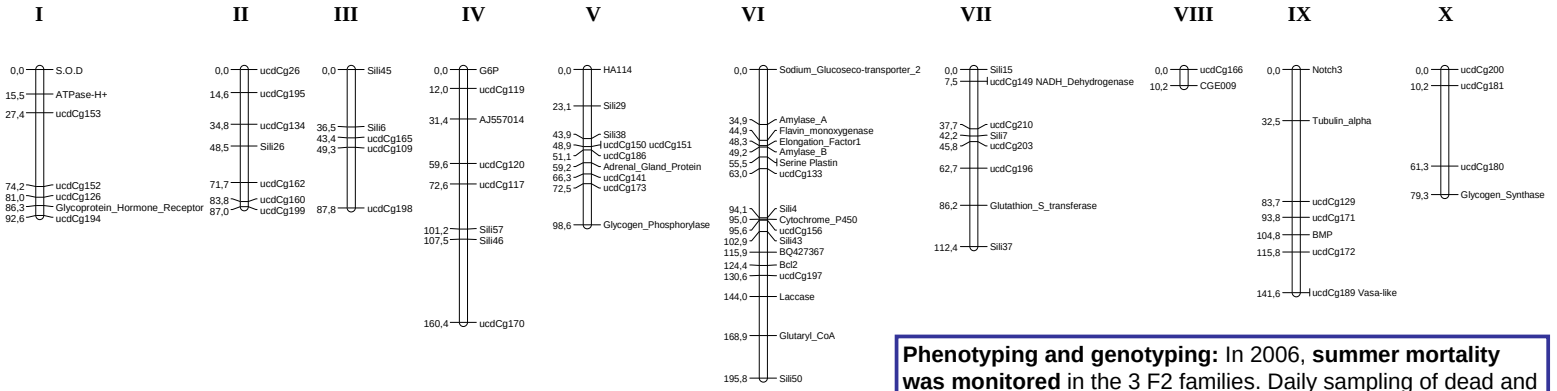
Introduction: Current knowledge about the genetic basis underlying traits of interest is limited in the Pacific oyster. Thanks to the French research project "MOREST", **lines resistant and sensitive to summer mortality** were obtained by divergent selection [1]. In 2006, **F2 segregating families** were produced to be monitored during a summer mortality event.

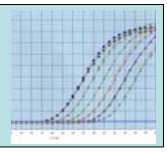


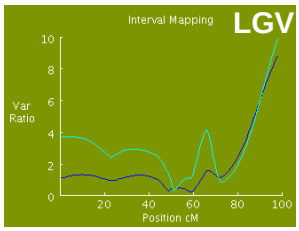
Linkage Mapping: Crimap [2] was used to build a consensus linkage map based on the analysis of three F2 families of 300 individuals each. The map is composed of 82 type I markers forming **10 linkage groups**, which matches the haploid number of chromosomes and. Sex specific maps show a higher recombination rate in females (1250cM) than in males (1050cM) as previously described [3].

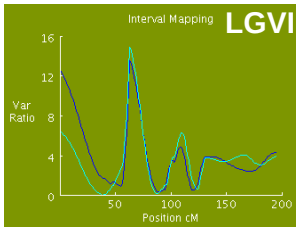
Phenotyping and genotyping: In 2006, **summer mortality was monitored** in the 3 F2 families. Daily sampling of dead and live oysters allowed the **individual quantification of** OsHV-1, a virus known to be associated to mass mortalities and has important economical loss for oysters farmers [4], by a newly developed Q-PCR assay [5]. Mortality was correlated with the presence of OsHV1. A set of 114 markers composed of 47 newly developed **SNP markers** [6]; 18 ***In silico* developed SSR** markers and 50 previously **published SSR** [3,7] were selectively genotyped for a total of 900 individuals.

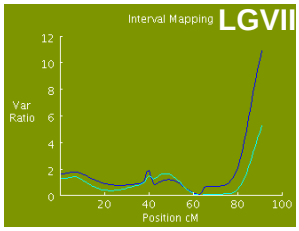
Individual Mortality Recording

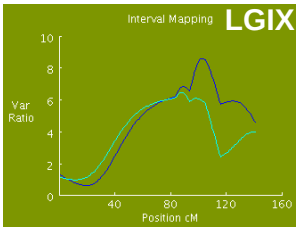
Individual Virus Load











LGV Position (cM)	LGV Lod score	LGV Percent. variance	LGVI Position (cM)	LGVI Lod score	LGVI Percent. variance		LGVII Position (cM)	LGVII Lod score	LGVII Percent. variance	LGIX Position (cM)	LGIX Lod score	LGIX Percent. variance
98	3.779	7.7	63/112	12.061	24	Mortality	91	4.691	9.7	103	3.7	7.6
98	4.248	8.6	63/112	12.198	24.3	Viral Load	91	NS	NS	88	NS	NS

QTL identification: QTL detection was successful for the two studied traits. Over the three F2s, **five regions were identified** in Linkage Groups V, VI, VII, IX. These explained **49% and 32.9% of mortality and viral load variances** respectively. Most of the QTL co-localized for the two traits. When analyzing each QTL individually and setting the viral load as a covariate, only part of the variation in mortality was explained by the 'viral load' QTL. This suggests that the viral infection is not the only cause of mortality in our experiment (non significant viral load QTL in LGVII and IX). These results **supports the high heritability of resistance to summer mortality** [8] and opens new perspectives in terms of MAS and localization of genes differentially expressed in resistant and sensitive lines [9].

REFERENCES:
[1] Samain, J.F. and McCombie, H. (2007): Mortalités estivales de l'huître creuse *Crassostrea gigas*. Défi Morest. Ifremer/Quae.
[2] Lander, E.S. and Green, P., 1987. Construction of Multilocus Genetic Linkage Maps in Humans. PNAS. 84, 2363-2367.
[3] Hubert, S. and Hedgecock, D., 2004. Linkage maps of microsatellite DNA markers for the Pacific oyster *Crassostrea gigas*. Genetics. 168, 351-62.
[4] Renault T, Le Deuff RM, Cochennec N, Chollet B, Maffart P. (1995) Herpes-like viruses associated with high mortality levels in larvae and spat of Pacific oysters, Crassostrea gigas: a comparative study, the thermal effects on virus detection in hatchery-reared larvae, reproduction of the disease in axenic larvae. Veterinary research 26(5-6):539-43.
[5] Pepin, J.-F., Riou, A. and Renault, T., Submitted. Rapid and sensitive detection of ostreid herpesvirus 1 in oyster samples by realtime PCR. Journal of Virological Methods.
[6] Sauvage, C., Bierne, N., Lapegue, S. and Boudry, P., 2007. Single Nucleotide polymorphisms and their relationship to codon usage bias in the Pacific oyster *Crassostrea gigas*. Gene. 406, 13-22.
[7] Yu, H. and Li, Q.I., 2007. EST-SSR markers from the Pacific oyster, *Crassostrea gigas*. Molecular Ecology Notes. 7, 860-862.
[8] Degremont, L., Ernande, B., Bedier, E. and Boudry, P., 2007. Summer mortality of hatchery-produced Pacific oyster spat (*Crassostrea gigas*). I. Estimation of genetic parameters for survival and growth. Aquaculture. 262, 41-53.
[9] Fleury, E., Fabioux, C., Lelong, C., Favrel, P. and Huvet ,A. (2008) Gene. In press. Characterization of a gonad-specific transforming growth factor-β superfamily member differentially expressed during the reproductive cycle of the oyster *Crassostrea gigas*.

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