

# Development of SNPs and their use for the identification of summer mortality resistance QTLs in the Pacific oyster (*Crassostrea gigas*).

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## 1. AQUACULTURAL IMPORTANCE

Due to its aquacultural potential, the Pacific oyster, originating from Japan, has been introduced in many countries. Its annual production represents the highest of all aquatic organism: 4.4 millions metric tons for a value of \$3.7 billions in 2003 (FAO, 2004). (1)



## 3. DEVELOPMENT OF SNPs

- Genes of interest are selected within the 2800 ESTs available in Genbank
- Primers are designed using PRIMER 3 on cDNA available sequences (3)
- PCR products are directly sequenced using Big Dye v3 Technology on an ABI 3100

## 4. RESULTS

DNA from 24 F0 mapping parents is sequenced to detect true SNP Polymorphism and to select the most informative loci. Our first results show a HIGH LEVEL OF POLYMORPHISM (Fig 1 & 2), compared to other species like human with 1SNP every 1kb (4) or in maize with 1 SNP every 70 bp (5). In *Crassostrea gigas*, one SNP is detected every 61 bp in coding regions whereas, in non coding regions, we have detected one SNP every 41 bp.

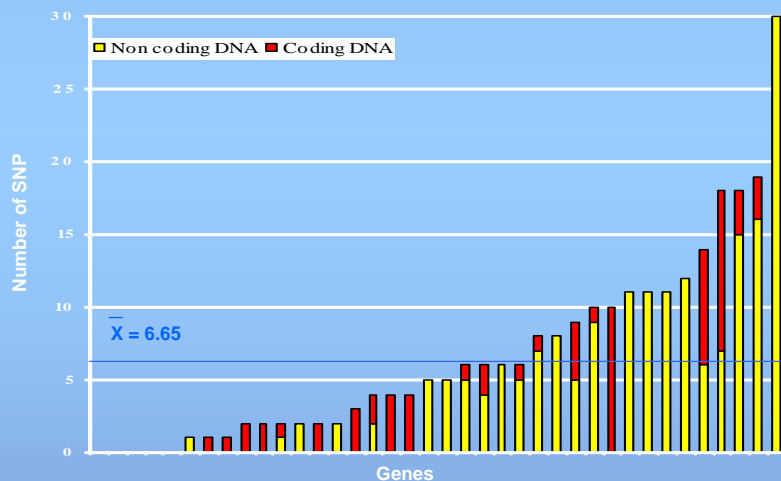


Fig 1: Level of polymorphism in Intron and Exon for each selected EST

To go further, other parameters were analysed such as the non synonymous diversity or the rate of G/C at third position in all ESTs. Negative correlation is observed between these two parameters (Spearman's Rho = -0.47, p=0.006). These results can be related to the hypothesis that selective constraints for optimal codon usage are likely to be correlated with selective constraints on the protein both, between codons in *C. gigas* (6).

## 5. PERSPECTIVES

On one side, further analysis of molecular evolution will be carried out. On the other side, each putative SNP will be scored by Maldi-ToF mass spectrometry to validate each marker and to detect heterozygote loci in F1 progenies. The same method will be used to genotype the F2 mapping families. This will allow the establishment of a HIGHER DENSITY LINKAGE MAP based on developed SNPs and available microsatellites (7). The final goal is to identify Quantitative Trait Loci (QTL) for summer mortality and other traits of aquacultural interest in *C. gigas*.

## 2. SUMMER MORTALITY

Summer mortality of the Pacific oyster has been described in the literature for decades. This is an important problem for oyster production in several countries. Studies showed that it results from the complex interaction of the environment, the oyster and pathogens.

As this trait was shown to be highly heritable ( $h^2 \approx 0.6$  (2)), one possible solution to overcome this problem would be to select animals tolerant to summer mortality. The identification of markers closely linked to the resistance would be an important step towards the establishment of a marker-assisted selection program.

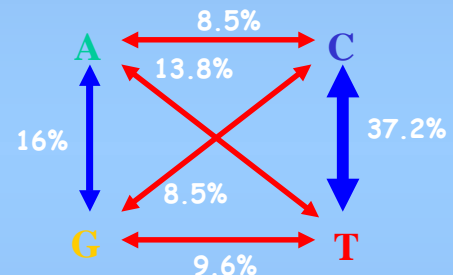


Fig 2: Percentage and Nature of SNPs found in selected ESTs

Among detected SNP, 73% are in intron and 27% in exons. Surprisingly, in coding regions 41% of markers are synonymous changes whereas, 59% are non synonymous.

Moreover, some biological functions are more polymorphic than other ones (Fig 3).

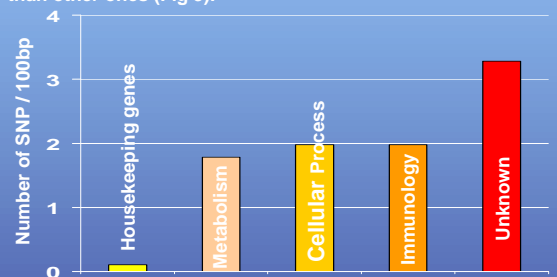


Fig 3: Average Number of SNPs by Biological Function

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