

P2.24 - Targeted association analysis for tolerance to salinity in rice using SSR markers

Negrão S¹, Courtois B², Ahmadi N², Babo P¹, Frouin J², Greco R³, Bruschi G^{3,4}, Vancoppenolle S², Katsantonis D⁵, Lupotto E⁴, Oliveira MM¹, Piffanelli P³

¹ ITQB-UNL, Quinta do Marquês, 2784-505, Oeiras, Portugal

² CIRAD, Avenue Agropolis, 34398 Montpellier Cedex 5, France

³ PTP- Rice Genomics Unit, Via Einstein, Località Cascina Codazza, 26900 Lodi, Italy

⁴ CRA- Rice Research Unit, s.s. 11 per Torino, km 2.5, 13100 Vercelli, Italy

⁵ NAGREF, Thermi-Thessaloniki, P.O. Box 60411, Greece

Rice is considered as a crop sensitive to salinity; however, it is grown mainly in deltaic areas with salt problems all over Europe. European rice breeding programs (France, Greece, Italy, Portugal and Spain) have established an European Rice Genetic Resources Collection (ERGRC) of some 450 accessions, mainly temperate *japonica*, extensively characterised for agronomic traits and maintained by CIRAD. The main goal of the present study is to identify within this collection, a set of best performing genes and alleles for salinity tolerance, as well as the associated donors and molecular markers for use in breeding programs. In order to obtain the general organisation structure of the ERGRC we assessed its genetic diversity through a Bayesian analysis of genotypic data over 26 SSR loci. A sub-sample of 200 accessions maximizing simultaneously allele number and allelic associations was then extracted for association analysis. The sub-sample was phenotyped for salinity tolerance at an early vegetative stage under controlled conditions, being leaf Na⁺/K⁺ ratio the most discriminating trait. Based on literature review, we assembled a list with more than 100 rice candidate genes for salt tolerance, which are involved in signaling, ion homeostasis, stress tolerance, transcription regulation, general metabolism and unknown functions. With this information, we developed a database of rice QTLs and candidate genes for salinity tolerance (http://tropgenedb.cirad.fr/html/rice_QTL.html). We selected 16 of these candidate genes for association analysis. SSR markers were used for the association analysis. A first set of 58 common SSR (www.gramene.org) covering these particular genes was used. In addition, we designed 320 SSR markers covering 100 kb up and downstream each candidate gene. In the end, we selected four of the designed SSR markers, the two closest to the gene and one in each end of the linkage disequilibrium region (100 kb each gene side). Results of association analysis between 16 target areas (using 60 SSR) and a dozen salinity tolerance traits are presented. Methodological constraints stem from the use of multi-allelic markers are discussed.

P2.25 - Involvement of alternative oxidase (AOX) in adventitious rooting of *Olea europaea* L.

Santos Macedo E¹, Cardoso HG¹, Peixe A², Arnholdt-Schmitt Birgit¹

¹ EU Marie Curie Chair - Laboratory of Molecular Biology, ICAAM, University of Évora, Apartado 94, 7002-554 Évora, Portugal

² Laboratory of Biotechnology and Plant Breeding, ICAAM, University of Évora, Apartado 94, 7002-554 Évora, Portugal

AOX was proposed as a functional marker candidate for efficient adventitious rooting of olive (*Olea europaea* L.) shoot cuttings (Arnholdt-Schmitt et al., 2006, Proceedings of the 2nd International Seminar Olivebioteq, Marsala, Mazara del Vallo, Italy, Vol I, pp 249–254; Arnholdt-Schmitt et al., 2006, Trends Plant Sci., 11 (6):281-287). Recently, this hypothesis was strengthened by results showing the involvement of AOX activity in adventitious rooting in semi-hardwood olive cuttings of the easy-to-root cultivar 'Cobrançosa'. Additionally, a high degree of sequence polymorphisms and 3'-UTR length variability had been identified for *OeAOX2* as potential sources for differential gene regulation (Santos Macedo et al. 2009. Physiologia Plantarum 137:532-552). The cultivar 'Galega vulgar' is a bad rooting olive genotype presenting average rooting rates in shoot cuttings of only 5-20%. However, under optimized *in vitro* culture conditions this cultivar demonstrates the high rate of 60–75 % adventitious rooting. After 5-7 days in culture, some cells from the cortex and also from the sub-epidermal tissue reveal a dense cytoplasm and present high mitosis rates and the first morphogenetic root fields are observed after 12-16 days in culture. Root primordial, become visible after 20 days. (Peixe et al. in preparation). To study the general relationship of AOX transcript accumulation to rooting, RT-PCR analyses of *OeAOX2* have been performed with cv. *Galega vulgar* in both optimized systems for olive rooting, in shoot cuttings and micro shoots. The results will be discussed in view of the potential of *OeAOX2* as functional marker candidate for efficient root induction.