

Functional Genomics of Rice by T-DNA Tagging

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During the past four years we have produced over 100,000 transgenic rice lines that carry T-DNA insertion. T-DNA was inserted into rice chromosome at an average of 1.4 genetic loci per plant. It can be estimated that the probability of finding a T-DNA insertion within a given gene from our insertional lines is approximately 60%. The binary T-DNA vector used in the insertion contained the promoterless β -glucuronidase (*gus*) reporter gene. This gene trap vector is designed to detect a gene fusion between *gus* and the endogenous gene that is tagged by the T-DNA. It was shown that 2–5% transgenic plants were GUS positive in the tested organs. Analysis of the T-DNA flanking sequences resulted in identification of knockout genes, which are disrupted by T-DNA insertion. Some of these lines showed co-segregation of T-DNA and a mutant phenotype. For reverse genetics approach, DNA pools have been prepared from the T-DNA tagged lines. For more efficient use of the lines, we are systemically determining the border sequences of each T-DNA insertion. The data will be valuable in identifying insertional mutant lines and understanding functional roles of rice genes.

Present and Future of Hot Pepper Genomics

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We have been conducting a genome research of hot pepper since 1996 as a comprehensive group effort. The objectives of our research is to construct a molecular linkage map based on RFLP, AFLP and SSR. Up to now, we have constructed an interspecific molecular linkage map of pepper using an F₂ population from a cross between *Capsicum annuum* cv.TF68 and *C. chinense* cv.Habanero. For physical analysis, we also constructed a 15 × genome size BAC library from *C. annuum* cv.CM334, which is known for strong resistance against *Phytophthora capsici*. Based on this molecular linkage map, the mature fruit color determined by carotenoid pigments was investigated genetically and shown to be cosegregated with *psy* encoding phytoene synthase of carotenoid biosynthesis pathway. Understanding pungency at molecular level, placenta-specific cDNA clones were isolated from a highly pungent cultivar cv.Habanero using the suppression subtractive hybridization (SSH) method and are now being further investigated genetically and biochemically. To study quantitative resistance of *Phytophthora* and virus, we have analysed 100 F₂ plants as mapping population derived from a cross between cultivar CM334 and Chilsuncho.