

and together lead to a better insight into the hybrid seed purity.

**F202** Molecular cloning and characterization of disease-resistant genes from apple

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Environmental stress is the major limiting factor in plant productivity. Naturally, plants have evolved a wide range of mechanisms to cope with such stresses, e.g. disease resistant gene (R-gene). Unfortunately, the crop and fruit plants developed by traditional breeding tend to have good food qualities but poor disease resistance, especially compared to wild type plants. Poor disease resistance can be partly originated by loss of certain R-genes. Once certain R-genes or other relevant genes are available, the use of gene transfer technology can provide the direct solution for this problem. As the approaches to provide the solution, R-genes from a wild type plant were isolated and characterized. To obtain more R-genes, a cDNA library of Jung-Sun- Mae-Ju was constructed and utilized for the isolation of the R-genes. For the cloning of NBS domain DNA fragment, PCR clones from the genomic DNA of a wild type apple (root stock) were selected. 5 clones in the 3 group out of 4 groups. From the screening of the cDNA library with probes of NBS DNA fragments, six new R-genes were isolated. The genes are supposed to generate 3~4.9 kb mRNAs and were expressed in the leaves of Yesan-Samyup, Hoengsung-Hwanyup and certain apple cultivars. These genes encode functional motifs NBS-LRR family, TIR-NBS-LRR or truncated form of TIR-NBS that lacks the LRRs. The similarity of the nucleotide sequences with tobacco N gene and potato NL25 is extremely high. Next step can be the application of the isolated genes to improve the apple cultivars against biotic stress like fungal or viral pathogens by the gene manipulation of the isolated R-genes.

**F203** Chromosomal assignment of

the garlic BAC clones using FISH technique

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Two BAC libraries of Danyang garlic cultivar were constructed. First one has been constructed using the pIndigoBAC536 vector and HMW garlic DNA extracted from leaf protoplasts. Second BAC library was constructed using the pBAC1/SACB1 vector, which is designed to give zero background, and HMW garlic DNA extracted from leaf nuclei. Forty seven clones from first BAC library were characterized by Southern hybridization with garlic genomic DNA or HRY4, a repetitive sequence of garlic, as a probe. Several BAC inserts, contained low copy of repetitive sequence, were then labeled as probes for FISH analysis. GBC5e (100 kb), one of the BAC clones which does not interrupted by repetitive sequence, was detected on the chromosome 7 in garlic. And, GBC4d (110 kb) gave rise to two different hybridization signals on the prometaphase chromosome. However, the location of hybridization can not be assigned due to the difficulty of karyotyping in the FISH condition used.

**F204** Genotyping of Field-Collected Soybean Mosaic Virus by RT-PCR and Restriction Fragment Length Polymorphism Analysis of the P1 Gene

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The genomic variability of soybean mosaic

virus (SMV) was examined by reverse transcription-polymerase chain reaction (RT-PCR) and restriction fragment length polymorphism (RFLP) analysis. The P1 protein is the least conserved region of the entire polyprotein of SMV, particularly in the N-terminal half having hypervariation in length and in sequence, which is the best target for strain discrimination. Among 14 symptomatic collected soybean plants, 12 yielded an array of amplification products of expected size 924 bp fragments following RT-PCR with specific primers. Successful amplification produced the entire P1 gene except cleavage site between P1 and HC-Pro. The amplified products were digested with *AfIII*, *BclI*, *HhaI*, *MboI*, and *XhoI*, the restriction enzyme chosen to discriminate between the collected isolates and known strains. The distinctive restriction patterns generated by the listed enzymes classified in certain types and subtypes and showed as a marker to discriminate between PCR products generated from field isolates including 4 reference strains.

#### **F205** Characterization and cloning of SUF1 (suppressor of FRI 1) that regulates flowering time in *Arabidopsis*

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Flowering of *Arabidopsis* is promoted by several interacting genetic pathways: photoperiod, vernalization and autonomous pathways. In the autonomous pathway, *FLOWERING LOCUS C (FLC)* is known as a central molecule, which acts as a floral repressor. *FLC* is positively regulated by *FRI*, while it is negatively regulated by *LD*, *FVE*, *FCA* and vernalization treatment. By fast neutron mutagenesis of *FRI-Col*, a very late flowering line, we isolated mutants that flower as early as Columbia ecotype. By genetic complementation of these mutants, five mutants (FN6, 24, 108,

202 and 225) were shown to be allelic and the mutants were named as *suf1-1* to *suf1-5* (suppressor of *FRI*). Our physiological data showed that the *suf1-1* allele is sensitive to photoperiods (short day vs long day) and vernalization (4?, 8 weeks). The RT-PCR analysis showed that the expression of *FLC* in *suf1-1* was not change in comparison to *FRI-Col*, but *ALG20* and *FT* were expressed as *Col*. Thus *SUF1* acts located between *FLC* and *AGL20* on the autonomous pathway. The mapping of *suf1* showed that *suf1* is tightly linked to a molecular marker *AtGABab* on chromosome III. Currently, the cloning of *SUF1* gene is in progress.

#### **F206** AGAMOUS-LIKE 20 gene integrates gibberellin signals for flowering in *Arabidopsis thaliana*

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Flowering in *Arabidopsis* is triggered by endogenous and environmental signals. Genes that regulate this transition have been assigned to separate genetic pathways that control the response to stimuli. A MADS-box gene, *AGAMOUS-LIKE 20 (AGL20)* promotes flowering by responding to the signals from autonomous, vernalization and photoperiod pathways. In this study, we show that the *AGL20* gene expression also responds to gibberellins which are absolutely required for flowering in short day conditions. Grown under non-inductive conditions, *gal* mutants never produce flowers and also showed low levels of *AGL20*. When the mutants were treated with gibberellin, they produced flowers and this phenotype was also accompanied with an increase in *AGL20* expression. Transgenic plants constitutively overexpressing *AGL20* gene (*35S::AGL20*) were insensitive to gibberellins and paclobutrazol, an inhibitor of gibberellin biosynthesis. Our results indicate that