

was PCR amplified and sequenced using the 454-FLX platform. The sequence reads were aligned to the reference genome for assembly and SNP detection. Cleaved amplified polymorphic sequence (CAPS) markers are being designed and tested from SNPs that are located within restriction enzyme cut sites. This method and the resulting molecular markers will be particularly valuable for breeding and genetic research that involves closely related and genetically similar tomato cultivars.

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### (366) Screening of *Arabidopsis* Ecotypes for Glucosinolates Accumulated in Shoots and Seeds

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Interest in glucosinolates (GS), sulfur-containing secondary metabolites in plants, has increased due to reported anti-carcinogenic properties. We investigated the amount of variation among types of GS along with total GS content in *Arabidopsis* ecotypes. We acquired 58 *Arabidopsis* ecotypes collected from Japan, China, southern Asia, eastern Europe, western Europe, Africa, South America, and North America. All ecotypes were grown in a walk-in growth chamber with a 14/10 photoperiod at 20 °C to produce shoots and seeds. Shoots were harvested just before anthesis and stored until further analysis. Freeze-dried seeds and shoots were ground and GS were extracted using methanol, benzyl glucosinolate, and barium-lead acetate. Desulfoglucosinolates were separated with a Hewlett-Packard high performance liquid chromatograph. Across all ecotypes the average total GS content was 518.60 mole per gram of dry weight. However, GS content ranged from 0.25 to 5691.83 mole per gram of dry weight among the individual ecotypes. Average total GS in seeds and shoots was 1456 and 164 mole per gram of dry weight, respectively. The range of GS concentrations in shoots and seeds was 0.25 to 2438.28 and 2.57 to 5691.83 mole per gram of dry weight, respectively. There was also a significant difference among ecotypes for GS content. Extremely high standard deviations for GS in seeds (1854) and shoots (482) further indicated the large amount of variation in *Arabidopsis* for Glu concentration. The high level of variation observed in this study might be because of the very wide area of collection of ecotypes. There was a strong positive correlation between total GS and aliphatic ( $r=0.89^{***}$ ) and indole-3-methyl GS ( $r=0.85^{***}$ ) whereas total GS was only moderately correlated with aromatic GS ( $r=0.54$ ). Lack of correlations between some of the GS (such as nasturtiin and napole) and total GS indicates that their levels may be manipulated without affecting the total Glu levels in *Arabidopsis*. High and low GS concentration ecotypes of *Arabidopsis* identified in this study may provide a basis for further genetic analysis at the molecular level in *Arabidopsis* and eventually to manipulation of GS levels.

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### (367) Cloning and Characterizing Full Length cDNA Encoding S-RNase (S39) in Chinese White Pear

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Recently, much progress in cultivars S-genotyping and S-alleles identification of chinese white pear (*Pyrus bretschneideri* Rehd.) was achieved. But gene expression patterns of majority alleles were not confirmed yet and their sequences obtained were non-functional. To clone and characterize cDNAs of chinese white pear S-RNases, an old cultivar, 'Xuehuali' (S4S16), was used. Consequently, a new S39-RNase was identified and two functional S-RNase cDNAs (S4 and S39) were cloned from styles of 'Xuehuali' by RT-PCR and RACE technology. The new S39-RNase cDNA contained 928 nucleotides including a complete open reading frame (ORF) predicted to encode a protein of 228 amino acids. The S39-RNase displayed the basic structural features of pear S-RNases, i.e., five conserved regions and one hypervariable (HV) region. At the deduced amino acid level, S39-RNase showed 67% to 79% similarity with other S-RNases of Maloideae. Phylogenetic analysis revealed that rosaceous S-RNases formed a subfamily-specific S-RNase group but not a species-specific group, suggesting that S-RNases occurs after divergence of subfamily but before the divergence of species in Rosaceae. We also found that 'Xuehuali' did not represent one cultivar but one cultivar group of different genotypes. This finding indicated that much more potential compatible combinations in chinese white pear should exist, which would be usefully for breeding of new cultivars.

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### (368) Asymmetric Hybridization: A Tool to Be Used in *Citrus* Radiation Map Creation

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*Citrus* small genome size ( $n\sim 385$  Mb) makes it highly amenable to genomics. However, no public physical maps of the *Citrus* genome are currently available. Asymmetric somatic hybridization (gamma radiation) could be used to build a radiation hybrid map of the *Citrus* genome. Via protoplast fusion, alien chromosomes fragments (broken by gamma radiation) may be inserted into receptor genomes. If insertions become permanent such alien chromosome introgression makes the produced somatic hybrid cells a most efficient route to the production of ordered maps with non- or minimally polymorphic markers. The idea of combining gamma irradiation with the microprotoplast mediated chromosome transfer (MMCT) technique is interesting since gene introgression could be more efficiently achieved by using smaller parts of the donor genome instead of whole genome. Microprotoplasts not only present much less chromosomes, significantly reducing destabilization