Expression marker development based on SuperSAGE transcription profiling

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SuperSAGE is an universal functional genomics tool for any Eukaryotes. This powerful, open-architecture transcription profiling platform enables in-depth analysis of changes in abundance of virtually all poly-(A)-containing mRNAs expressed from eukaryotic genomes (1) (see the presentation of KAHL et al., these proceedings).

We applied SuperSAGE for transcription profiling of drought and salt-stressed roots and nodules from chickpea and cold-stressed stems and leaves from chickpea and lentil.

Our approach yielded more than 360.000 cDNA signatures representing more than 24.000 single genes from stressed material and non-stressed controls including more than 2900 significantly stress-regulated protein-coding genes. SuperSAGE tags (SuperTags) of genes significant for drought-, salt- and cold-stress are currently applied in 3'-rapid amplification of cDNA ends (3'RACE) and 5'RACE for Expression Marker design.

We aim at SuperTag-Arrays for legumes. The direct use of significant 26bp SuperTags for transcription profiling in populations of interest was recently demonstrated (MATSUMURA et al. 2006). Customized assays or SuperTag-Arrays can be designed by exploiting SSR and SNPs in the 5' and 3' UTR of differentially expressed isoforms up- or downregulated under stress.

The first Expression Markers for profiling drought, salt- and cold-stress in legumes will be based on SuperTags found in chickpea and lentil with 100% match in *Medicago truncatula* (*Table 1*).

Table 1: **Annotation of Chickpea & Lentil SuperTags in Legumes via Basic Local Alignment Search Tool (BLAST) in NCBI & TIGR. Total tags (salt stressed roots): 24,456; total tags annotated 5,332. Total tags (cold stressed leaves: 10,151; total tags annotated 4,013.**

Legume Species	Source: Coldstressed Leaves BLAST hits	Source: Salt stressed Roots BLAST hits
Medicago truncatula	907	2464
Medicago sativa	254	656
Vicia spp.	34	102
Lotus corniculatus	310	965
Lotus japonicus	309	934
Glycine max	459	1250
Glycine soja	180	748
Phaseolus vulgaris	179	508
Phaseolus coccineus	101	442
Trifolium pratense	515	1367
Lupinus albus	35	199
Pisum sativum	148	396
Arachis hypogaea	49	204

Besides the discovery of new genes interesting for traits of agronomic interest, SuperSAGE also allows the simultaneous analysis of expressed genes of two organisms during interaction e.g. of Parasite/Host (MATSUMURA et al. 2003).

Tolerance or resistance to various parasites (e.g. broomrape, *Orobanche crenata*) and pathogens (e.g. *Ascochyta*, *Fusarium*) are of complex nature. SuperSAGE analyses can decipher these complex interactions by profiling all genes during interaction. These data can be exploited for Expression Marker design.

SuperTag based Expression Markers as well as SSR and SNP markers suitable for screening susceptibility or resistance/tolerance to pathogens are new tools in studies on genetic diversity, for Marker assisted breeding, and selection of germplasm.

References

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