ASSOCIATION ANALYSIS USING SSR AND SNP LOCI TO FIND QTL FOR SEED OIL CONTENT IN SOYBEAN

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Figure 1. Different soybean accessions under the same field conditions

Brazil is known by its great variability and seed yield that may be used to extract vegetable oil or biofuel. Nowadays, soybean is the best alternative among all oilseed crops, mainly because of the huge cultivated area throughout the country, the adequate levels of productivity and quality of oil (viscosity and cetane ratio), the short-term cycle (nearly four months from seeding to harvesting) and the network of crushing industries available nationwide.

The objective of this research is to determine the positions of Quantitative Trait Loci (QTL) and the level of polymorphism of candidate genes associated with oil production and fatty acid composition in soybean populations.

The mapping population of 96 accessions, includes Brazilian modern cultivars, Brazilian lines and Plant Introductions from USDA (United States Department of Agriculture) germplasm collection. This population was divided into two subpopulations of 48 accessions each: one with high (more than 22%) and other with low (less than 17%) oil content. Recently, they were planted in the experimental area of ESALQ/USP.

The accessions will be analyzed with 100 SSR (Simple Sequence Repeat) loci that were selected based on their association with QTL for oil content or polymorphism shown in previous studies. Concurrently, we will sequence portions of up to 10 candidate loci involved in oil biosynthesis in a sample of 10 soybean accessions selected to high and low oil. Single Nucleotide Polymorphisms (SNP) identified in the sequences will be used as markers for those candidate genes. SNPs that cause phenotypic variation are priority for genotyping. Collaborators will collect data on the level of fatty acids in each of the accession. Standardized Disequilibrium Coefficients (D') and Squared Allele-frequency Correlations (r2) for pairs of SSR and SNP loci will be used to estimate Linkage Disequilibrium (LD). The seeds' oil content will be tested for significant differences in allele frequencies between the low and high oil groups. Putative QTL may be identified on the basis of highly significant markers, which will certainly become a useful tool in future breeding program to rapidly increase the rates of oil content.

SUMMARY OF RESULTS
TO DATE AND PERSPECTIVES

In a total of 128 SSR loci tested, 100 were polymorphic in a sample of 10 different soybean accessions (*Figure 2*). To each SSR loci the best reaction conditions were optimized in order to reach the better PCR product. Amplicons and marker data of those preliminary tests were detected using a 4300 DNA analyzer and Saga GT software from LI-COR Corporate. The following steps include the determination of allelic frequencies, allele size, and genetic diversity in the mapping population.

In Brazilian field conditions, the 96 soybean accessions have shown high variability in days of flowering, plant maturity, disease resistance, and grain yield.

Preliminary results of the same accessions developed in greenhouse conditions of seed oil content by Ressonance Magnetic Nuclear (RMN) also showed high level of variability (from 11% to up to 25%)(*Figure 3*).

The presence of high genetic variability among soybean cultivars for oil content and seed yield can help to determine regions in LD with SSR and SNP loci. The results suggest that the SSR loci, together with a high standard of variability of the plants will provide a consistent analysis of soybean germplasm.
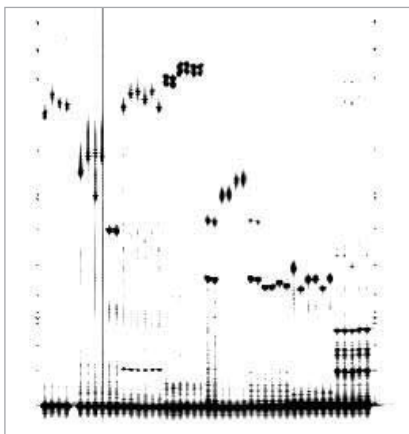

Figure 2. Polymorphism of eight SSR loci showed by electrophoresis in 4300 DNA analyzer
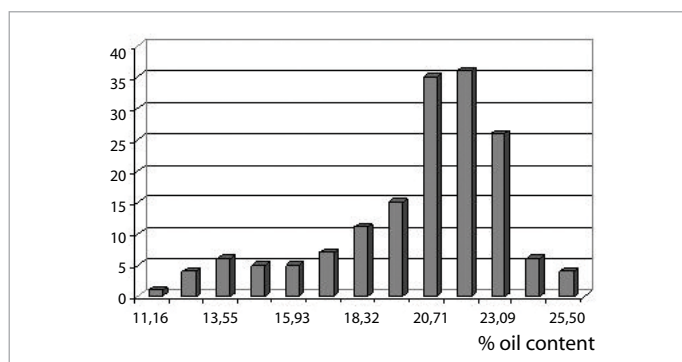

Figure 3. Distribution of oil content (%dry mass) in 96 soybean accession development in Greenhouse conditions

MAIN PUBLICATIONS

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