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Peanut Genetics

March 15, 2006
NPB Final Report

Title: **Accelerating Development of Peanut Varieties through Molecular Markers**

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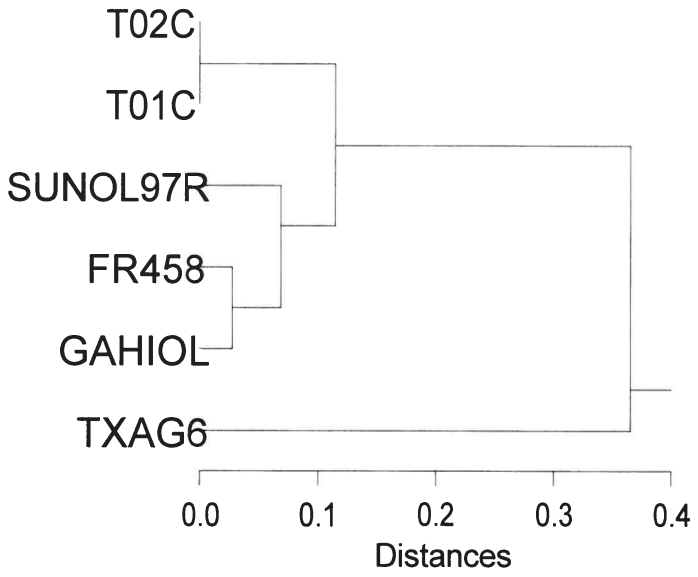
Results to date:

Map published microsatellites (SSRs) on the Florunner x TxAG-6 population. We have used both agarose gels and DNA analyzer-based detection to measure polymorphism. In the case of these parents, many of the polymorphisms are visible by moth detection methods. To data, approximately 40% of the primers used have detected at least one polymorphism between these two parents. Frequently we have observed two bands in each lane, as would be expected for an AABB disomic tetraploid. We have also found these polymorphisms among BC1 progeny of the mapping population. we have seen differences among plants of the mapping population. Based on the 144 primer pairs that we have had synthesized, we would expect insufficient polymorphism to make a map based on these primers alone. However, these can be mapped against the RFLP marker map. In addition, several publications recently have reported additional sets of primers, which when added to the current set should make it possible to construct an independent map of peanut using only SSRs.

Identify microsatellite markers that can be used in our peanut improvement program. The markers are being tested for ability to distinguish between the varieties and populations segregating for early maturity, high O/L, seed size, and plant type. Approximately one quarter of the primers have been able to distinguish differences between this set of cultivated peanut genotypes. Use of agarose gels has not proved satisfactory, because most of the polymorphisms among cultivated accessions are on the order of 2 base pairs (bp) to 4bp difference, too small to resolve. Therefore, we have switched to use of a DNA analyzer for differences. We have been able to distinguish different peanut varieties (see Fig. 1), although plant-to-plant variation of composite lines has been detected, and we are attempting to find a statistical measurement that will allow us to distinguish differences between plant of a variety from differences among varieties. This can be seen especially among Tamrun OL01 and Tamrun OL02, where the two varieties are sister lines, and variation among plants of the same variety so far has appeared to be similar to the variation between the varieties. This means that we need to run additional primers and plants of each variety.

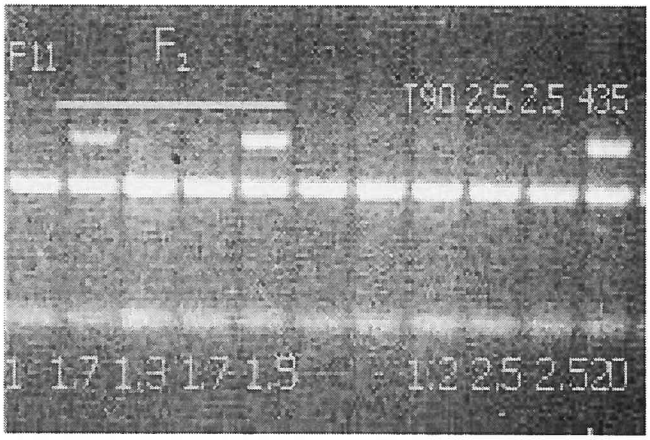
This will be important for using markers to identify hybrids, and also for quality control in seed multiplication.



1Figure 1. Classification of runner varieties using microsatellite markers. TxAG-6 is an interspecific hybrid, and markers show a great difference between this and the cultivated runner varieties.

Markers for the high-O/L trait. We can tell high-O/L from low-O/L parents using markers, but the F2 generation is more difficult. We have identified a pattern that is always present in high-oleic seeds, but this pattern is sometimes also present in low oleic seeds also. Our assumption that we had markers for two independent genes appears to have been incorrect, and so we need to find markers for a second gene for the markers to work. Additional evidence is that two F2 seeds that had O/L ratios of 1.7:1 were planted; the two seeds had different marker patterns. The F3 seeds were harvested and analyzed for their O/L ratios. One plant produced large few high-oleic seeds, and the other produced many such seeds. Therefore these two seeds were genetically different, even though the O/L ratios were the same.

We have used primers to amplify additional desaturase genes, and we have cloned these fragments. These need to be sequenced to confirm that they are indeed desaturase genes, and to look for possible polymorphisms between high-oleic and low-oleic parents that can be used to make markers.



2Figure 2. Markers for the high-oleic trait. The top marker is present in all high-oleic accessions, including UF-435 (rightmost lane), which is the donor parent for the high-oleic trait. Two lanes with a 1.7:1 ratio can be seen (the second and fourth from the left).

Acknowledgments. We express our sincere appreciation to the National Peanut Board, the Texas Peanut Producers Board, and the Texas Peanut Producers for assisting our program again in 2005. A large part of the work reported here would not have been possible without this generous support.

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continue to 2005

Final Report

Peanut Genetics
March 13, 2005

Accelerating Development of Peanut Varieties through Molecular Markers

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Problem and Need

Enhanced resistance to disease, early maturity, water-use efficiency, and oleic:linoleic ratios are important peanut traits requiring improvement in Texas. Breeding for these traits is difficult because of our lack of understanding of the genetics of these traits, and the variability in field conditions which means that the needed weather or pathogens are not present in some years. In all cases, research is needed to identify the genes controlling these traits and to develop molecular markers to accelerate varietal development for Texas producers.

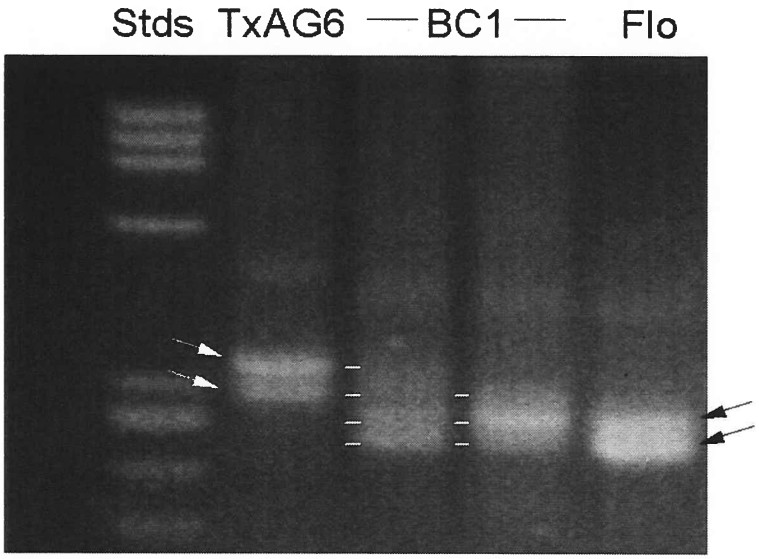
Objectives

The proposed research aims to develop markers for cultivated × cultivated crosses to complement the peanut breeding programs in Texas. The applications expected to benefit the soonest are early maturity, disease resistance, and high O/L content.

Results

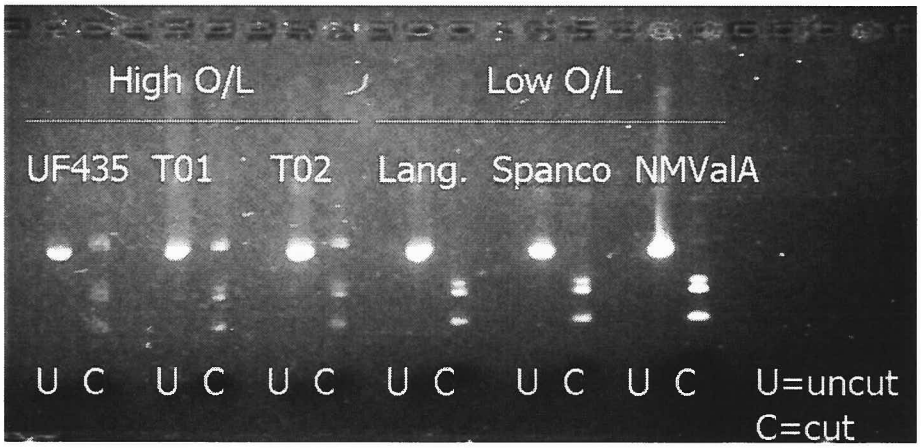
(1) Test microsatellite primers for detection of polymorphism in cultivated peanut and begin mapping of markers on a peanut population. Approximately 110 microsatellites that have been reported to reveal differences among cultivated peanut have been synthesized commercially. We have been able to identify differences among the BC1 progeny of the Florunner x TxAG-6 mapping population. We will use this cross for mapping microsatellite markers on the RFLP map of peanut.

Many of these markers will also be useful for breeding work in cultivated peanut, with the potential to be associated with diverse traits including early maturity, nematode, leafspot, TSWV, and Sclerotinia resistance.

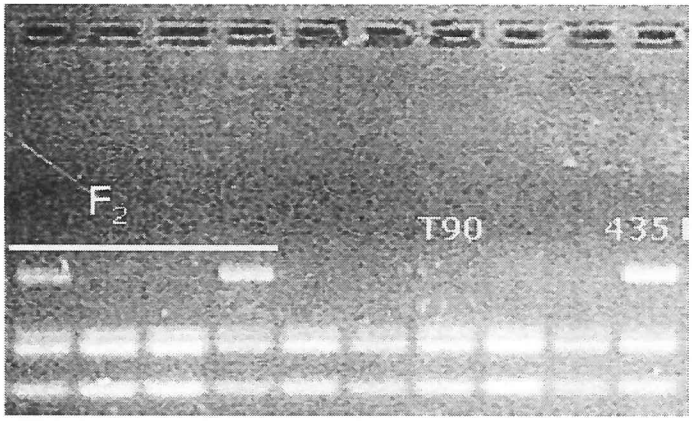


(2) Development of markers for the high-O/L trait. The high-oleic trait has known to be controlled by 2 recessive genes, but there is evidence that more genes are involved. This means additional complexity in breeding for this trait. We are in the process of developing markers that can be used for this trait. Also, success for this trait will signify that this approach is workable in peanut.

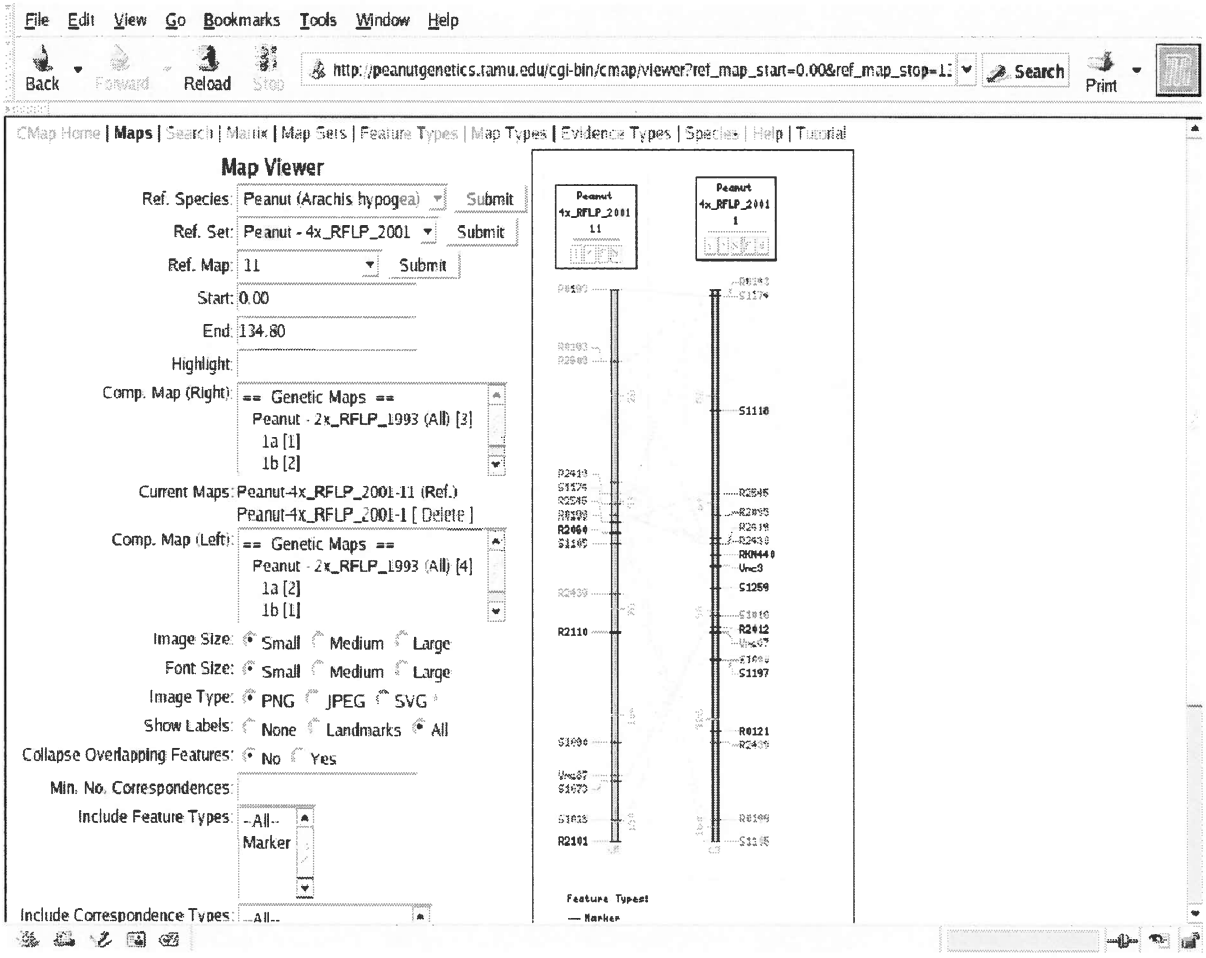
We have used differences in the sequences of the normal (low oleic) and mutant (high oleic) genes to design DNA primers for CAPS (cleaved amplified polymorphic sequence) markers. These markers can distinguish both high-O/L and low-O/L varieties (see figure below).



To test whether these work in a breeding population, we have made F2 crosses between high-O/L and low-O/L varieties, and are now testing the markers on DNA from this population. This can be seen in the following figure:



(3) Development of a marker database and installation of DNA analysis software. We have published an online graphic-oriented database of DNA marker maps using the open source CMAP software. This is the only marker database for peanut, and data will be made available in the Legume Information System (LIS) database for comparison of peanut and other legumes. The benefit of this database is that it will allow easy access to peanut marker data for breeding and genetics work.



We have added both marker maps of peanut (Halward *et al.*, 1993; Burow *et al.*, 2001). The database is publicly available on the internet at http://peanutgenetics.tamu.edu/cmap. The database shows the markers mapped on peanut, and places them on schematic drawings of peanut chromosomes.

ACKNOWLEDGMENTS

We express our sincere appreciation to the National Peanut Board and the Texas Peanut Producers Board for assisting our program in 2004. The work reported here would not have been possible without their generous support.