Project Title: **Making peanut significantly more drought tolerant**

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Goal of the Project:

Drought stress is a major cause responsible for reduced quality and yield in peanut production in West Texas. In order to develop peanut with significantly improved resistance to drought stress and to maintain the viability of peanut production in West Texas, we proposed to introduce an Arabidopsis gene, *AtKelch*, into peanut to reduce water loss under drought conditions and to make peanut profitable with reduced water input (our early work with *AtKelch* in Arabidopsis indicated that overexpression of *AtKelch* would increase ABA sensitivity and therefore reduce water loss under drought conditions in Arabidopsis, which formed the basis of this project).

Summary of Project:

We first fused water-deficit inducible promoters, the *RD29A* promoter or the pSARK promoter to *AtKelch* cDNA to form the RD29A-AtKelch and the pSARK-AtKelch constructs, respectively, and then we used these constructs to transform wild-type peanut plants. We have obtained 52 putative transgenic peanut lines: with the RD29-AtKelch construct, we obtained 26 independent lines, and among these plants, 18 are in Runner genotype background, and 8 are in Valencia genotype background; with the pSARK-AtKelch construct, we also obtained 26 independent putative transgenic lines and among these plants, 16 are in Runner genotype background, and 10 are in Valencia genotype background. We have achieved our goal in obtaining at least 50 independent putative transgenic peanut plants. We are isolating genomic DNAs and RNAs from these lines. Next we conduct PCR experiments to prove if these plants are transgenic plants and Northern blot analysis to identify high-expression lines. We also plan to generate more seeds from selected high-expression lines for 2011 field-trial experiments to test if these *AtKelch*-expressing peanut plants can tolerate dry conditions in the field.

265
767
2009

Project Title: **Making peanut significantly more drought- and salt-tolerant**

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Final Report:

The goal of this research was to create peanut that can use water more efficiently and increase peanut production in the arid and semi-arid land of West Texas. With the support from TPPB, we generated about 30 transgenic peanut plants. We then isolated DNAs from these putative transgenic peanut plants and conducted PCR experiments to screen for the *AVP1* transgene. Our results indicated that more than half of our putative transgenic peanut plants contain the *AVP1* transgene (Fig. 1A). Subsequently, RNA blot analysis was used to confirm transcription of the *AVP1* transgene and estimate the level of transcript in *AVP1*-expressing plants (Fig. 1C). Transgenic lines showing high levels of *AVP1* expression were selected for further physiological characterization.

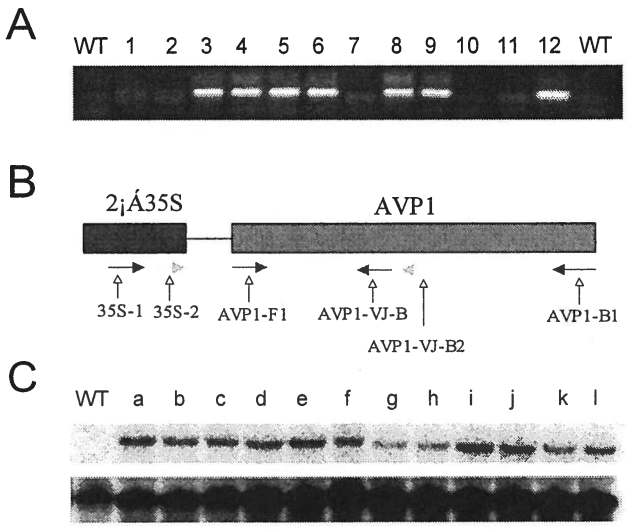

Fig. 1. Molecular analysis of *AVP1*-expressing peanut. **A**. PCR analysis of *AVP1*-expressing peanut. WT, wild-type; lines 1 to 12, 12 independent transgenic peanut plants. **B**. The *AVP1* construct used to transform peanut and the positions of oligonucleotides in *AVP1* that were used for PCR analysis. The oligonucleotides AVP1-F1 and AVP1-B1 were used in the PCR experiment shown in A. **C**. RNA blot analysis of *AVP1*-expressing peanut. Lines a to l, 12 independent *AVP1*-expressing peanut plants that were positive by PCR analysis.

We used several lines that exhibited high levels of *AVP1* transcript for salt tolerance studies, and our data indicated that *AVP1*-expressing peanut plants are indeed more salt-tolerant than wild-type peanut plants (Fig. 2). For example, transgenic peanut plants continued to grow during the salt treatment, whereas the wild-type control plants stopped growing after salt treatment. At the end of the 200 mM NaCl treatment, transgenic plants were visually larger and greener than wild-type plants (Fig. 2). This is an on-going project, and more experiments are planned. We will conduct drought tolerance test in the summer of 2010.

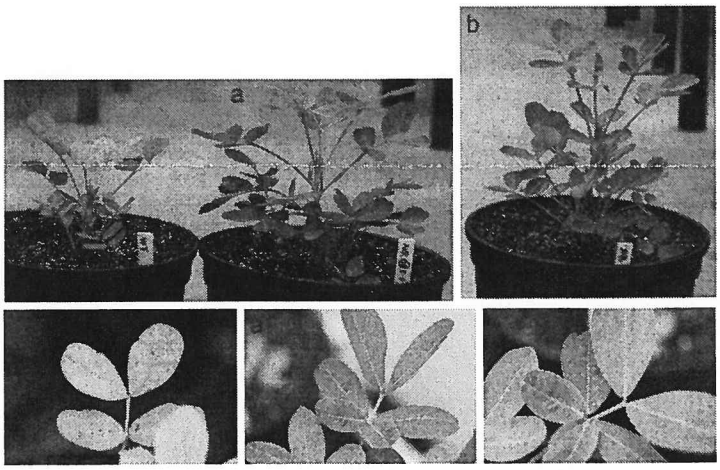

Fig. 2. Phenotype of wild-type and *AVP1*-expressing peanut plants after 200 mM NaCl treatment. WT, wild-type; **a** and **b**, two independent *AVP1*-expressing peanut plants.