BIOTECH TRAITS IN ALFALFA – WHAT’S NEXT?

Mark McCaslin, Stephen Temple and Peter Reisen

In June 2005, USDA/FDA/EPA deregulated the Roundup Ready® trait in alfalfa. In August 2005, seed of the first Roundup Ready alfalfa varieties was offered for sale. This was the first biotech trait to be commercialized in alfalfa, and paved the way for new research supporting a second wave of biotech traits enhancing value in alfalfa. These new biotech traits can be divided into two categories: output traits that enhance forage quality and input traits that improve efficiency of crop production.

Output Traits
The Consortium for Alfalfa Improvement1 (CAI) was formed in 2004 to explore the use of biotechnology for improvement of alfalfa as a dairy feed. The CAI has focused on alfalfa improvements in two areas: improved fiber digestibility and increased efficiency of protein utilization.

Alfalfa is an important source of fiber in most dairy rations. Lignin is an anti-quality compound in alfalfa cell walls that increases with advanced plant maturity, and reduces the digestibility of alfalfa fiber. There are various biotech tools that can be used to turn off or “silence” native genes. Scientists at the Noble Foundation have now systematically silenced virtually each of the genes that code enzymes required for lignin synthesis in alfalfa. CAI scientists are in the process of evaluating transgenic plants with reduced expression of one or more lignin biosynthetic genes. Although virtually all of the transgenic plants contain reduced lignin content, they vary widely in lignin composition. Increased fiber digestibility is also a common feature of the transgenic plants, but agronomic performance varies significantly by transgene. Based on multiple lab and field studies initiated since 2000, we have learned that transgenic plants with reduced expression of two key lignin enzymes, COMT and CCOMT have decreased lignin, increased fiber digestibility and acceptable agronomic performance. Elite alfalfa populations containing the COMT- or CCOMT- transgene have been developed. In 2007 hay will be produced to enable CAI sheep and dairy feeding studies to confirm improved animal performance of these reduced lignin alfalfa plants. Positive results from these feeding trials would move the project into an accelerated development mode. Reduced lignin alfalfa may provide an important new genetic tool for hay producers, providing more flexibility in harvest management and increasing forage quality and/or forage yield.

1 The Consortium for Alfalfa Improvement partners are the U.S. Dairy Forage Research Center, the Samuel Roberts Noble Foundation and Forage Genetics International.
Alfalfa produces more protein per acre than any other crop grown in the U.S. However, alfalfa protein is often inefficiently utilized by high producing dairy cows. This inefficient utilization leads to increased nitrogen losses to the environment and a requirement for feeding supplemental protein. The problem with alfalfa protein can be traced to two factors: post-harvest proteolysis (haylage) and the rapid rate of protein degradation in the rumen (hay and haylage). The CAI is exploring two strategies for increasing protein stability in haylage and in the rumen.

**PPO Alfalfa**
In red clover, polyphenol oxidase (PPO) combines with a specific phenolic substrate to protect protein from post harvest proteolysis. The red clover PPO gene was cloned and has now been expressed in alfalfa. Various biotech approaches for endogenous expression of a suitable PPO substrate are being explored.

**Tannin Alfalfa**
Condensed tannins are a class of phenolic compounds found in many plants. Tannins bind with proteins and slow the rate of protein degradation in the rumen. Tannin containing forages (e.g. birdsfoot trefoil and sanfoin) have more bypass protein and are non-bloating when grazed by ruminants. Alfalfa produces condensed tannins, but only in the seedcoat. Various biotech strategies are being explored for production of condensed tannins in leaves and stems of alfalfa. The U.S. Dairy Forage Research Center estimates that tannin alfalfa could decrease protein feed supplement costs for dairy by 60% and significantly decrease N losses to the environment. In addition, UC Davis scientists are exploring strategies for production of hydrolysable tannins in alfalfa, unrelated compounds with positive attributes similar to condensed tannins.

**Input traits**
Industry, non-profit and public research institutions are investing several hundred million dollars per year in gene discovery programs aimed at improving crop performance. These genomics-based gene discovery programs are turning up hundreds of gene candidates for numerous value-added traits.

More than half of the alfalfa grown in the U.S. is produced under irrigation or under dryland conditions where moisture commonly limits productivity. Several biotech companies are currently exploring and testing transgenes that increase drought tolerance and water use efficiency when expressed in crop plants. Several of these gene candidates for drought tolerance are now being expressed in alfalfa. In collaboration with Monsanto, Forage Genetics will begin testing several new potential drought resistant transgenic alfalfa plants in 2007.

Although genes for increased biomass or delayed flowering are of little interest for grain crops, they offer exciting potential for alfalfa. Several such genes have been identified in general phenotypic assays of new gene candidates and are now being inserted into alfalfa. These new transgenes may offer our best opportunity to significantly increase forage yield in alfalfa.
SUMMARY

It’s an exciting time for those involved in alfalfa improvement. Using conventional breeding techniques, alfalfa breeders continue to make incremental progress in improving yield, persistence and forage quality. Biotech traits offer a new and exciting tool for break-through improvements in crop performance and crop value. Several new traits are currently in testing and development in alfalfa. We see significant potential for these to increase forage yield, improve forage quality and/or increase the role of alfalfa in animal diets.