Re-Designing Alfalfa for Increased Yield and Quality
Trait Targets

• Agronomic Traits (input traits)
  – Herbicide tolerance
  – Pest resistance
  – Abiotic stress tolerance
  – Increased yield per se

• Quality Traits (output traits)
  – Fiber digestibility
  – Efficiency of protein utilization
Quality Traits

Plant Science ↔ Animal Science

Dairy Producers
Consortium for Alfalfa Improvement

USDFRC

Noble Foundation

Pioneer

Forage Genetics International
Re-Designing Alfalfa for Improved Forage Quality

• The CAI concept:
  – Interdisciplinary, inter-institutional collaboration
  – Ruminant nutrition
  – Biochemistry
  – Plant molecular biology
  – Plant breeding/agronomy

• CAI target traits – “re-designing alfalfa”
  – Increased cell wall digestibility
  – Improved efficiency of protein utilization
Reduced Lignin Alfalfa

- Lignin increases with advanced maturity in alfalfa.
- Lignin is indigestible, and binds with cellulose/hemicellulose – reducing fiber digestibility.

**Consortium for Alfalfa Improvement**
Gene knockout to modify lignin pathway

Use genetic engineering to knockout/silence “lignin gene(s)” in alfalfa

Compound A

\[ \text{Enzyme 1} \]

\[ \text{Gene 1} \]

Compound B

\[ \text{Enzyme 2} \]

\[ \text{Gene 2} \]

Lignin

Reduced lignin alfalfa
Lignin Biosynthetic Pathway

phenylalanine → cinnamate → 4-coumarate → 4-coumaroyl CoA → 4-coumaraldehyde → 4-coumaryl alcohol → H lignin

4-coumaroyl shikimate → caffeoyl shikimate → caffeoyl CoA → feruloyl CoA → coniferaldehyde → coniferyl alcohol → G lignin

5-hydroxyconiferaldehyde → 5-hydroxyconiferyl alcohol → S lignin

Measure affect on lignin content and composition

Noble Foundation gene knockouts
2008 Summary – RL1 Alfalfa

24% increase in fiber digestibility

28% lignin reduction

Note: lower stem samples, CAI equation, average of 25 elite events
Nampa, ID May 30, 2007

3.63 T/A

3.58 T/A

3.29 T/A

3.23 T/A

RL1

RL1 NULL

RL2

RL2 NULL
USDFRC Sheep Feeding Studies
RL1 Alfalfa

Changes in NDF Digestibility over Time

- RL Alfalfa = Increased flexibility in harvest timing

- Reduced lignin

- Null control

- Early bud
- Late bud
- 10% bloom

NDFD%
Cutting Management Trial at Arlington, WI
2008 Cutting Management Trials (WI, MN)

Forage Quality (NDFD%)

Forage Yield (lbs/plot)

3 location mean

Vegetative     Early bud      Late Bud    10% bloom    Full bloom

10-14 days

57%

RL1
null
RL Construct Comparison

2008 RL Event Sorting Nursery – West Salem, WI

% Null Control

- RL1
- RL2
- RL3
- Commercial

Yield
NDFD

Holy Cow!
Reduced Lignin Summary

- Gene knockouts can be used to significantly change lignin content and composition
- RL alfalfa identified with good agronomic performance
- RL alfalfa has increased NDFD and slower change in quality with advancing maturity
  - More flexibility in harvest timing
  - Fewer harvests per season = $$$
Tannin Alfalfa

- FGI/Noble/USDFRC collaboration to improve efficiency of alfalfa protein utilization for dairy.
- Tannins bind with protein, slowing rate of rumen degradation and increasing RUP (bypass protein).
  - Increasing RUP 20% → 30% would eliminate need for protein supplements
- Condensed tannins can be found in alfalfa seed coats, but not leaves or stems.
  - Genetic engineering can be used to modify gene expression, producing Tannin Alfalfa.
Transgenic Tannin Alfalfa – Research Approach

MtLAP in Medicago

Lc MtLAP

Increase metabolic feed for pathway

anthocyanins

BAN

catechin epicatechin

Production of tannin monomers

Condensed tannin

Tannin polymerization

Tannin Alfalfa
Tannin Alfalfa

- USDFRC MW dairy model for tannin alfalfa:
  - 60% reduction in protein feed supplementation
  - Up to 12% increase in net return for dairy
  - 25% reduction in N losses

- Tannin containing forages are non-bloating
  - Worldwide alfalfa related bloat losses > $200M
Agronomic Traits

• Roundup Ready Alfalfa
• Abiotic Stress Tolerance
  – Drought tolerance/WUE
• Delayed flowering
Drought Tolerance

- Drought tolerance and water use efficiency will become critical with increasing cost and decreasing availability of irrigation water in the West.
- Drought tolerance genes have been identified and will be commercialized in several crops, including alfalfa.
Drought Tolerance Field Trials

Drought tolerant corn trials: Grain yield on 80% moisture

Drought tolerance in soy

Monsanto website
Drought tolerance trial
Nampa, ID April 8, 2009
• Genetic basis of flowering control is well understood.
• Transgenic approaches to modify flowering time are being evaluated.
• Important tool for increasing forage yield in alfalfa.
Delayed flowering in soybeans