

Next Generation Bioenergy

*Technologies for Feedstocks for
Cellulosic Ethanol Production from Non-Food Sources*



German Spangenberg



World Energy Consumption – Status and Forecasts

- Fossil fuels account for 80% of the world energy consumption and their use is the main cause of global warming
- World primary energy consumption grew on average 2.1% per year over the last 30 years
- Over 10 billion tons oil equivalents of primary energy are consumed per year
- 1.6 billion people, a quarter of the world population, has no access to electricity
- Conventional oil reserves will inexorably concentrate in the Middle East, which holds 65% of the world proven reserves
- Coal is the most abundant fossil energy for the next century



Rationale for Bioenergy on Global Basis - 1

Bioenergy is being targeted as key contributor to global fuel supplies, displacing fossil fuels, in the decades ahead - since

- Population is increasing
- Per capita GDP is increasing (standard of living)
- World energy consumption is rising
- Burning of fossil fuels is a major source of greenhouse gases
- Fossil energy resources are finite



3

Rationale for Bioenergy on Global Basis - 2

- Petroleum resources are most limiting
- Petroleum is the major source of transportation fuel
- Modern economies cannot function without abundant and inexpensive transportation fuel
- The majority of proven petroleum reserves are in the politically unstable Middle East

Alternative, renewable and sustainable bioenergy must be developed if we intend to maintain our high standard of living in the longer term.



4

Energy in Victoria - Challenges

Victoria is facing significant environmental challenges to the economic advantages it derives from utilisation of its very low-cost brown coal resources - since

- Victoria accounts for 22% of Australia's greenhouse gas emissions
- Approximately 52% of these arise from the use of brown coal for electricity generation in the State
- Need to contain greenhouse gas emissions from supply and use of energy to develop over time a sustainable energy sector



5

Feeding the World

- Human population doubled since 1960 to 6 billion
- World population predicted to increase to 8 billion by 2030 and to 9 billion by 2050
- In the next 2 generations the world will consume twice as much food as consumed in the entire history of humankind
- Multiple new technologies of food production created and adopted to keep up with population growth



6

Sustainable Bioenergy Development

- **Need to follow long term strategy of decoupling the biofuel and food industries**
- **Next generation of biofuels will need to come from non-food sources**
- **Produced from dedicated bioenergy crops**
(e.g. low input, water and nutrient use efficient perennial grasses – not competing for arable land for cropping)
- **Produced from by-products of other human activities**
- **Use of enzyme fermentation of crop residues such as straw and grass biomass as feedstocks for cellulosic ethanol**



7

Sustainable Bioenergy Development - Cellulosic Ethanol

- **About half of the above ground biomass of grain crops is 'wasted' as straw**
- **Globally, ca. 2 billion tons of wheat, rice and maize straw are produced annually**
- **Australia could generate c. 40 M tons cereal residue per year**
(as feedstock potential >15 BL bioethanol)
- **Biodegradation of cellulose and hemicellulose in straw is heavily prevented by lignin**



- **Solution: Recycling of agricultural waste through cellulosic ethanol**

Cellulosic Ethanol – Feedstock Issues

- **Solar collection efficiency**
- **Biomass conversion**



- **Quantity**
- **Quality**
- **Sustainability**



Solar Collection Efficiency – Requirements and Issues

- **High energy content plants** (cellulose)
- **High 'cellulose-intensity' crops** (available cellulose/hectare yield)
- **Minimisation of agricultural input requirements** (water, fertilizer)
- **Tolerance to abiotic stress** (low/irregular rainfall, salinity)
- **Seasonal variation in biomass production**
- **Deliberate development of perennials**
- **Feedstock diversity to minimise supply disruption**
- **Suitability of the local climate/environment**

Biomass Conversion – Requirements and Issues

- Convert biomass into 'free sugar' in time, energy and resource efficient manner
- Achieved by accessing cellulose and hemicellulose and exposing them to fermentation using enzymes
- Problems:
 - a) lignin interferes physically & chemically with enzyme activity
 - b) cellulose hydrolysis is slow and costly
- Potential Solutions:
 - a) reduce lignin content and modify its composition
 - b) enable plants to produce their own hydrolysis enzymes

Technologies for Feedstocks for Cellulosic Ethanol Production from Non-Food Sources

- **Cell wall modification to assist in deconstruction**
 - altering lignin content, composition & cleavability
 - reducing lignin-polysaccharide cross-links
 - replacing lignin with polysaccharides
- **Improvement of agronomic traits for sustainable bioenergy plant production systems, optimising**
 - biomass yield
 - vegetative growth patterns
 - biotic and abiotic stress tolerances
 - nutrient use efficiency
 - others
- **Production of cell wall polysaccharides degrading enzymes in plants**
 - enhanced cellobiohydrolases
 - enhanced endoglucanases
- **Generation of value added products in plant biomass**

Cell Wall Modification to Assist in Deconstruction - 1

Developmental Stages in Perennial Grasses



V1 V2 V3
Vegetative

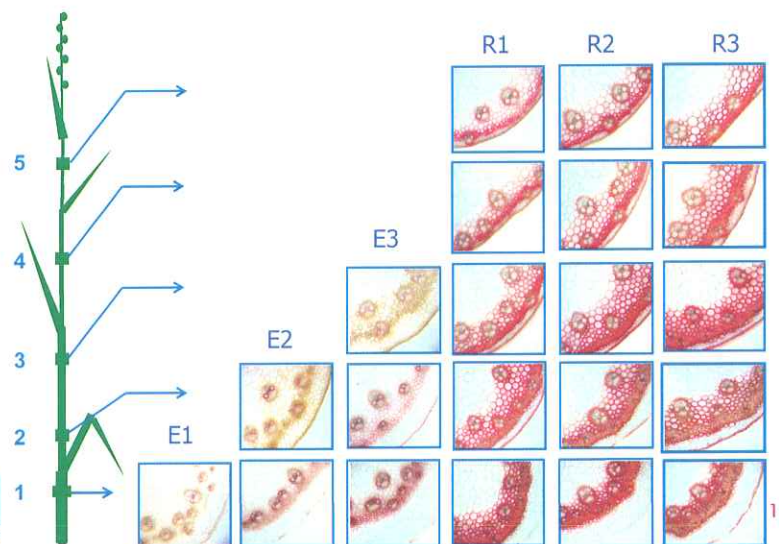
E1 E2 E3
Elongation

R1 R2 R3
Reproductive

13

Cell Wall Modification to Assist in Deconstruction - 2

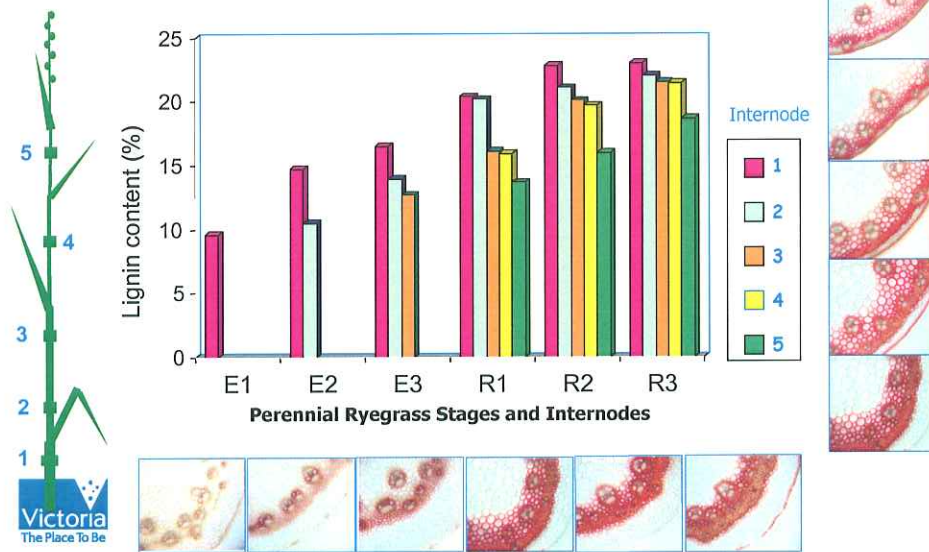
Lignin Deposition in Perennial Grasses



14

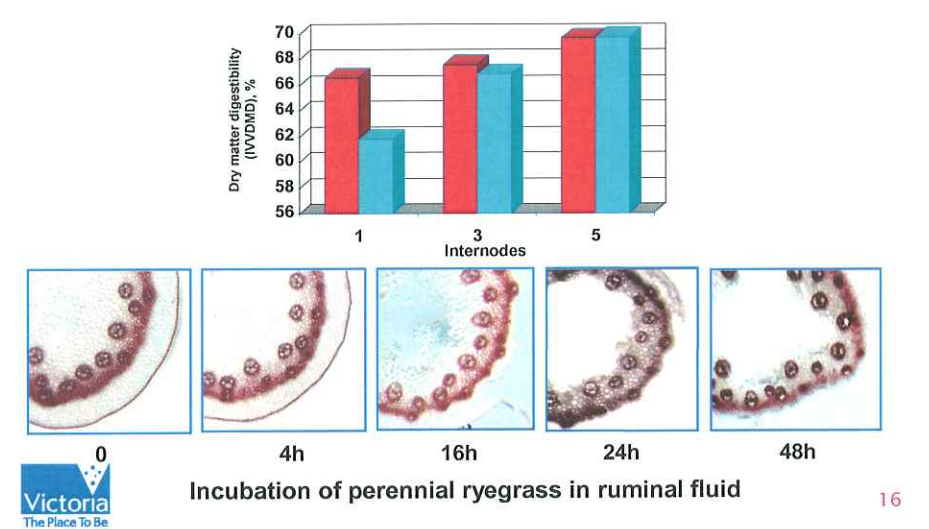
Cell Wall Modification to Assist in Deconstruction - 3

Lignin Content in Perennial Grasses



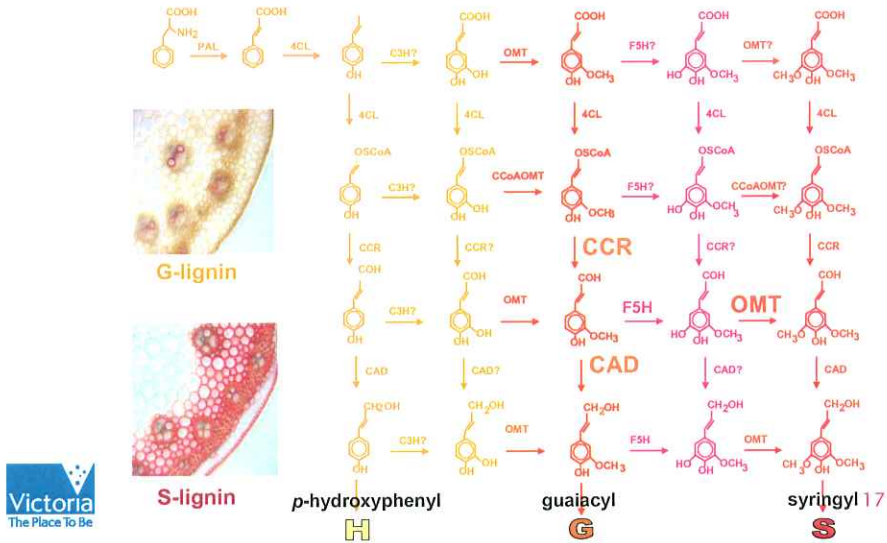
Cell Wall Modification to Assist in Deconstruction - 4

Lignin Interferes with Cell Wall Fermentation



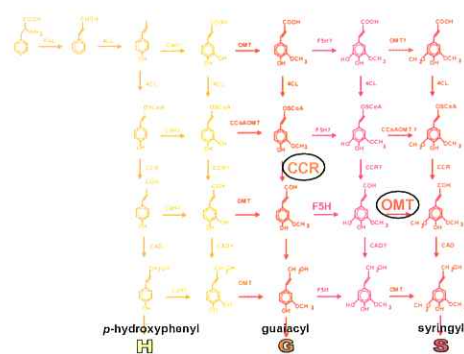
Cell Wall Modification to Assist in Deconstruction - 5

Lignin Biosynthesis in Grasses

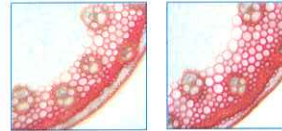


Cell Wall Modification to Assist in Deconstruction - 6

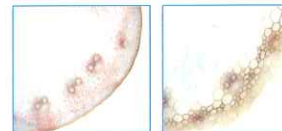
Technology 1: Genetic Modification Technology for Cell Wall Modification in Wheat Straw and Perennial Grasses



Control Grass



GM Grass with Reduced OMT



GM Grass with Reduced CCR

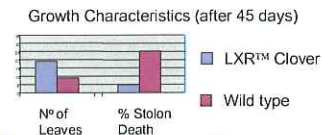
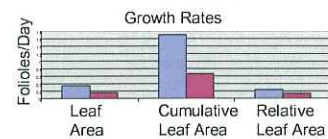
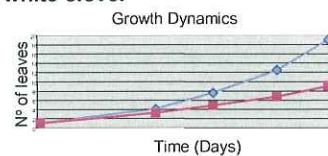


- **Cell wall modification to assist in deconstruction**
 - altering lignin content, composition & cleavability
 - reducing lignin-polysaccharide cross-links
 - replacing lignin with polysaccharides
- **Improvement of agronomic traits for sustainable bioenergy plant production systems, optimising**
 - biomass yield
 - vegetative growth patterns
 - biotic and abiotic stress tolerances
 - nutrient use efficiency
 - others
- **Production of cell wall polysaccharides degrading enzymes in plants**
 - enhanced cellobiohydrolases
 - enhanced endoglucanases
- **Generation of value added products in plant biomass**

Optimised Biomass Yield and Vegetative Growth Patterns



LXR white clover



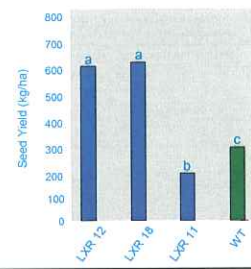
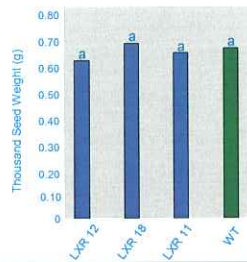
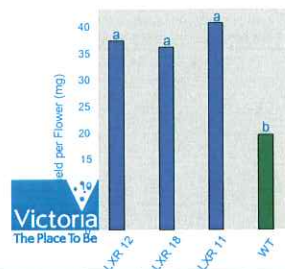
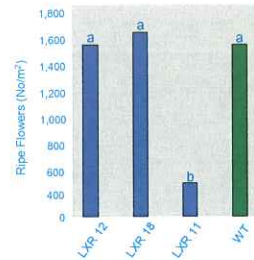
Cytokinins → Senescence



Technology 2: Genetic Modification Technology for Yield Enhancement in Wheat and Perennial Grasses



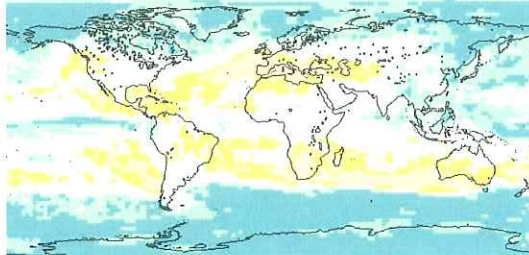
Field
evaluation
of LXR
white clover



- **Cell wall modification to assist in deconstruction**
 - altering lignin content, composition & cleavability
 - reducing lignin-polysaccharide cross-links
 - replacing lignin with polysaccharides
- **Improvement of agronomic traits for sustainable bioenergy plant production systems, optimising**
 - biomass yield
 - vegetative growth patterns
 - biotic and abiotic stress tolerances
 - nutrient use efficiency
 - others
- **Production of cell wall polysaccharides degrading enzymes in plants**
 - enhanced cellobiohydrolases
 - enhanced endoglucanases
- **Generation of value added products in plant biomass**

Climate Change and Variability

- Projected climate change implications for Australia suggest



Report to the Australian Greenhouse Office, DEH, March 2005

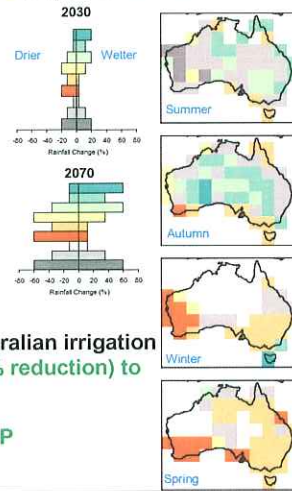
- A likelihood of decreasing rainfall over most of Australia

- GDP scenario losses due to reduction in Australian irrigation allocations estimated between **\$136 million (5% reduction)** to **\$751 million (20% reduction)**

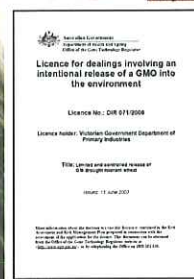
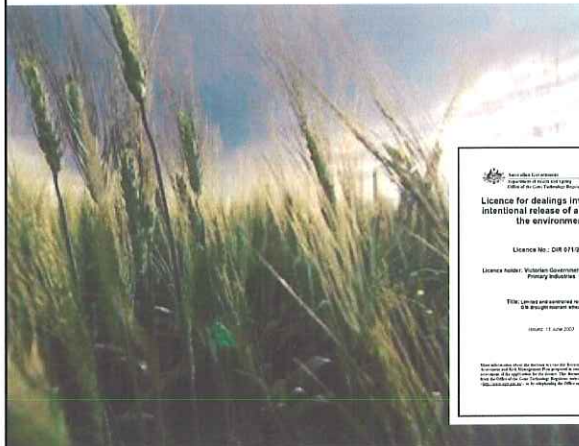


- Drought subtracted c. **1% from Australia's GDP** in 2002/03; equal to ca. **\$6.6 billion**

Precipitation increase in ≥90% of simulations
Precipitation increase in ≥5% of simulations
Precipitation decrease in ≥5% of simulations
Precipitation decrease in ≥90% of simulations

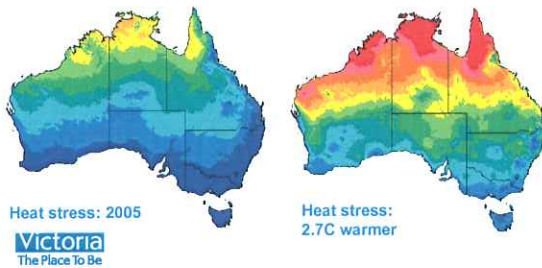


Technology 3: Genetic Modification Technology for Drought Tolerance in Wheat and Perennial Grasses

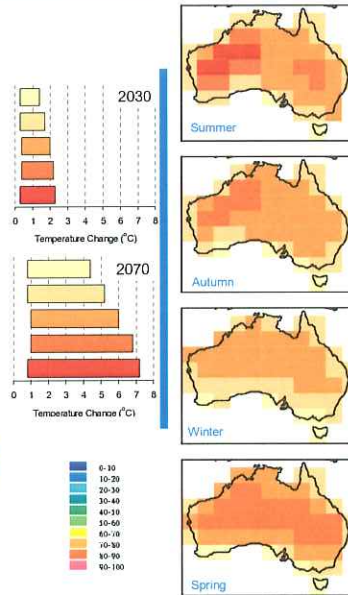


Climate Change and Variability

- Projected climate change implications for Australia suggest
- Annual average warming over much of inland Australia of **1-6°C by 2070**
- Increased heat stress frequency

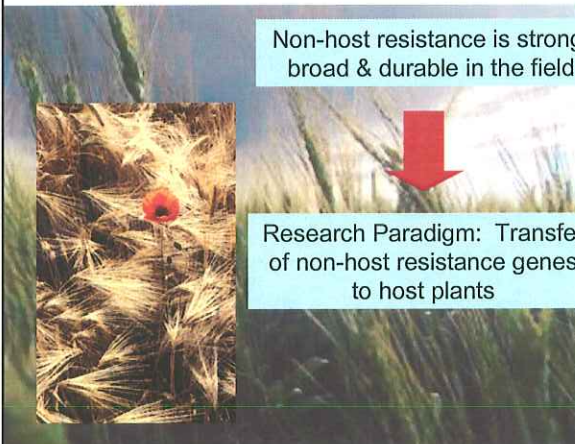


Report to the Australian Greenhouse Office, DEH, March 2005



Non-Host Resistance

Plants Are Surprisingly Healthy!



Estimated national trait values of diseases in wheat

- Stripe rust **\$102 million**
- Crown rot **\$90 million**
- *Septoria tritici* **\$86 million**
- *Septoria nodorum* **\$83 million**
- Stem rust **\$57 million**
- Leaf rust **\$56 million**

Technology 4: Genetic Modification Technology for Biotic Stress Tolerance in Wheat and Perennial Grasses



Yellow leaf spot
(*Pyrenophora tritici-repentis*)

Powdery mildew
(*Blumeria graminis*)

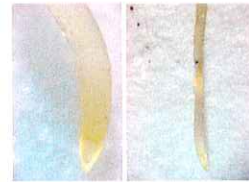
27

- **Cell wall modification to assist in deconstruction**
 - altering lignin content, composition & cleavability
 - reducing lignin-polysaccharide cross-links
 - replacing lignin with polysaccharides
- **Improvement of agronomic traits for sustainable bioenergy plant production systems, optimising**
 - biomass yield
 - vegetative growth patterns
 - biotic and abiotic stress tolerances
 - nutrient use efficiency
 - others
- **Production of cell wall polysaccharides degrading enzymes in plants**
 - enhanced cellobiohydrolases
 - enhanced endoglucanases
- **Generation of value added products in plant biomass**

28

Nutrient Use Efficiency

- P forms insoluble unavailable compounds
- 30 million tons of P fertiliser applied yearly worldwide
- Australian farmers spend \$600 million in P fertiliser per year
- Up to 80% of applied P fertiliser lost
- \$10 billion of P in Australian soils



OA-Transgenic



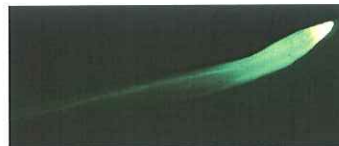
Control

29

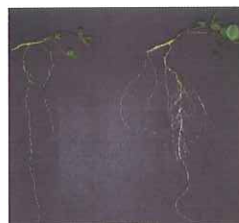


**Technology 5:
Genetic Modification Technology for Nutrient Use Efficiency in Wheat and Perennial Grasses**

Root tip-prevalent promoter from white clover (*TrPT1::gfp*)

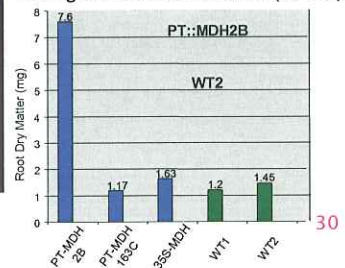


Isogenic Control vs Transgenic 500 uM Al



Isogenic Control vs Transgenic 500 uM Al

Root growth under Al-stress (10 uM)



30

- **Cell wall modification to assist in deconstruction**
 - altering lignin content, composition & cleavability
 - reducing lignin-polysaccharide cross-links
 - replacing lignin with polysaccharides
- **Improvement of agronomic traits for sustainable bioenergy plant production systems, optimising**
 - biomass yield
 - vegetative growth patterns
 - biotic and abiotic stress tolerances
 - nutrient use efficiency
 - others
- **Production of cell wall polysaccharides degrading enzymes in plants**
 - enhanced cellobiohydrolases
 - enhanced endoglucanases
- **Generation of value added products in plant biomass**