

# Genomics Based Approaches to Genetic Improvement in Sugarcane

Robert Henry



# Centre for Plant Conservation Genetics



*Life Enriched by Plant Biodiversity*

# Plant Diversity and Evolution

GENOTYPIC AND PHENOTYPIC  
VARIATION IN HIGHER PLANTS

Edited by R.J. Henry,  
Centre for Plant Conservation Genetics,  
Southern Cross University, Australia

An understanding of plant diversity at both the genome and phenome levels is important for biodiversity conservation and plant breeding. Recent advances in genomics have also resulted in a growth of the subject of plant functional genomics. This book brings these areas together, by reviewing aspects of plant evolution as it relates to variation in plant genomes and associated variations in plant phenomes. Topics covered include chloroplast and mitochondrial genomes, reticulate evolution, polyploidy, population genetics within a species, the evolution of the flower, diversity in plant cell walls and in secondary metabolism, and the importance of plant diversity in ecology and agriculture. Contributors include leading authorities from Europe, the USA, Australia and New Zealand. It will be of significant interest to those working within the areas of plant genetics, evolution, biodiversity, botany and agriculture.

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# Plant Resources FOR FOOD, FUEL OR CONSERVATION



ROBERT HENRY

# Food



# Biomass options

- Sorghum
- Sugarcane
- Grasses
- Shrubs
- Trees

**biobank**

# Special Journal Issues- Biofuels

Plant  
Biotechnology  
Journal

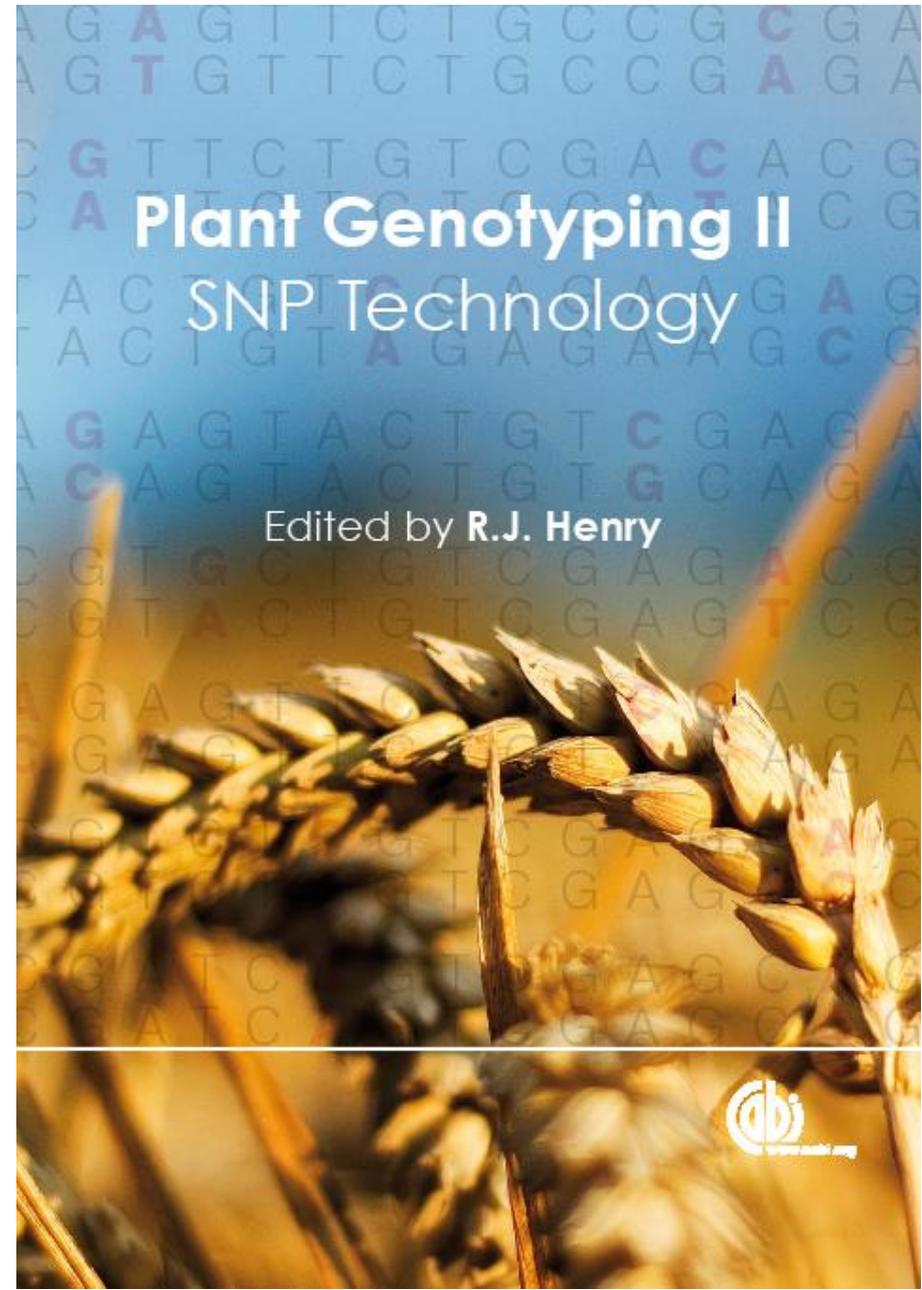
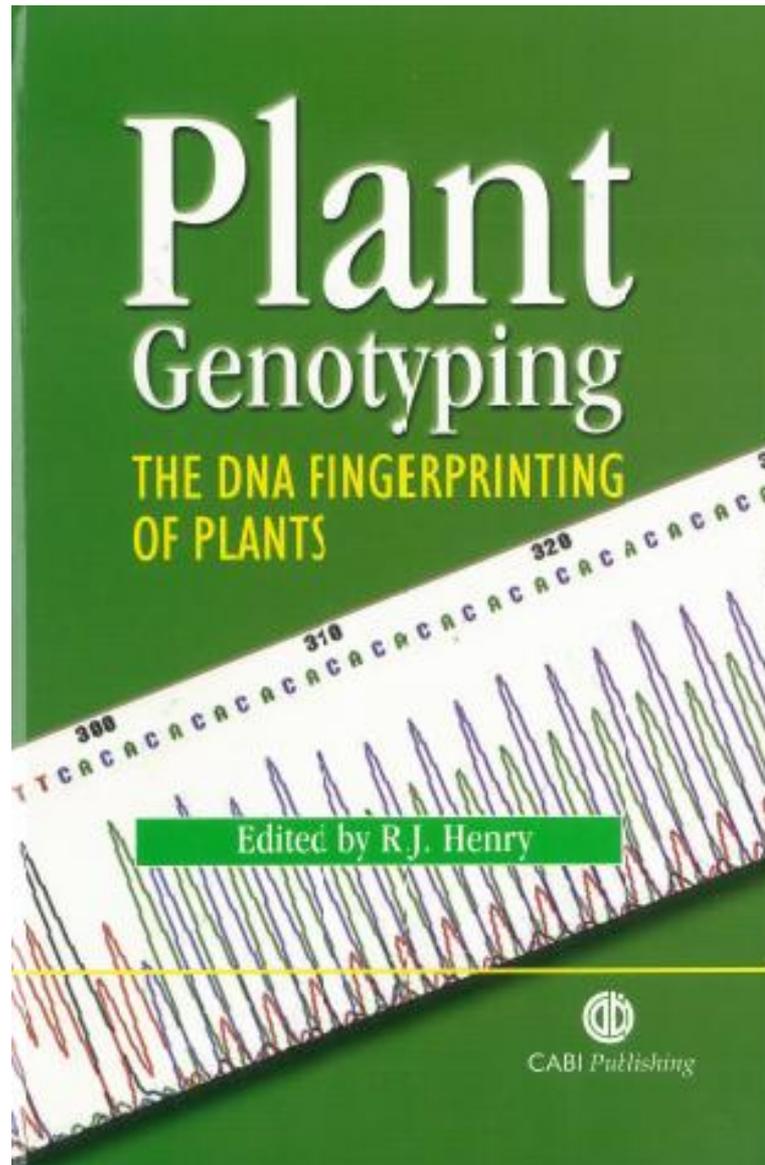
Volume 3 Issue 3  
May 2005

The diagram consists of a vertical bar on the left and a horizontal bar on the right. The vertical bar has a scale from 0.0 to 1.0. The horizontal bar has a scale from 0 to 140. The diagram is divided into several colored sections (orange, blue, yellow, pink) and contains various labels and numbers.

Labels on the vertical bar (left): 0.0, 0.2, 0.4, 0.6, 0.8, 1.0. Labels on the horizontal bar (right): 0, 20, 40, 60, 80, 100, 120, 140. Labels in the middle: 0.0, 0.2, 0.4, 0.6, 0.8, 1.0. Labels on the right side of the horizontal bar: 0.0, 0.2, 0.4, 0.6, 0.8, 1.0. Labels on the far right: 0.0, 0.2, 0.4, 0.6, 0.8, 1.0.

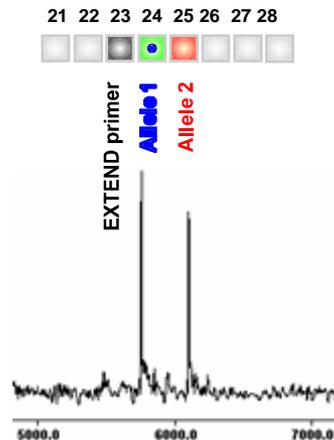
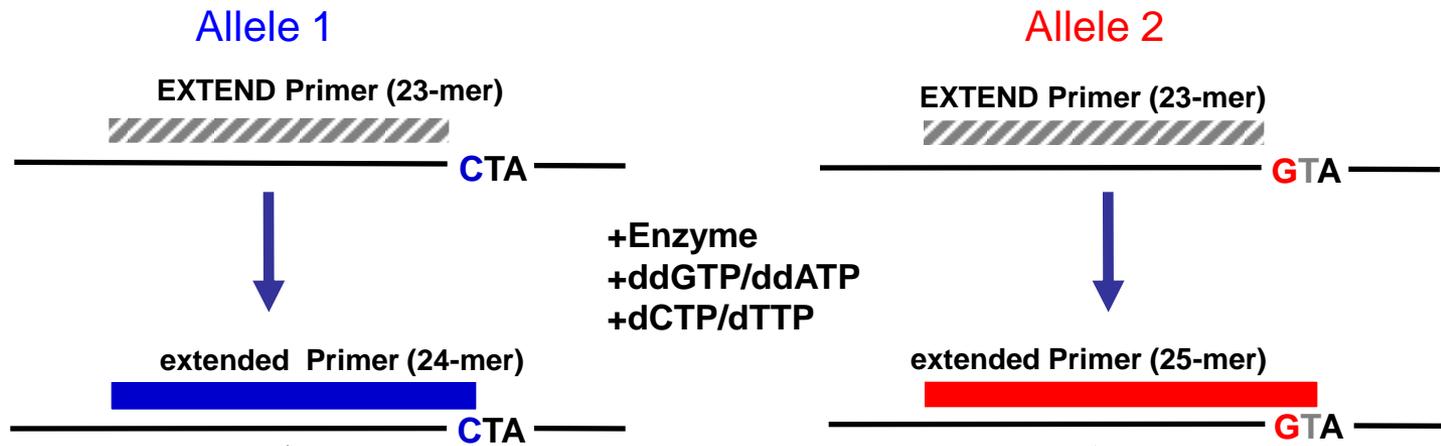
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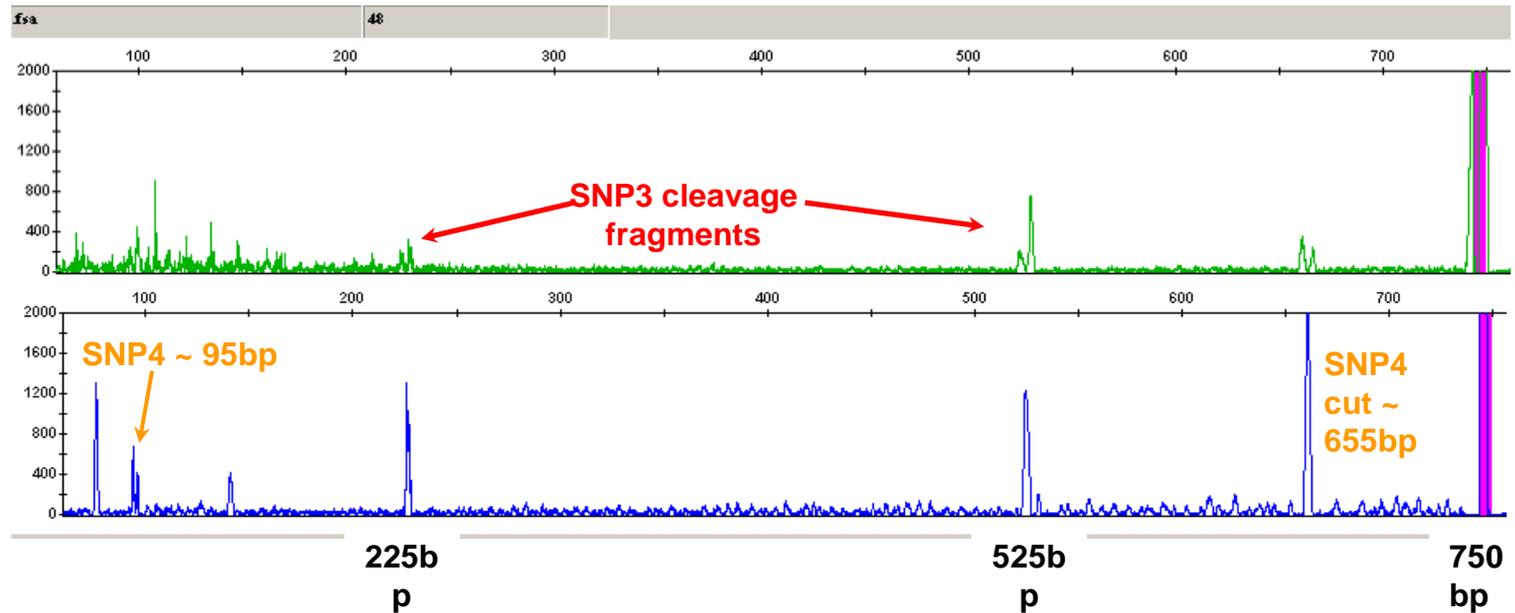
# Genotyping by mass analysis





# Advanced molecular screening

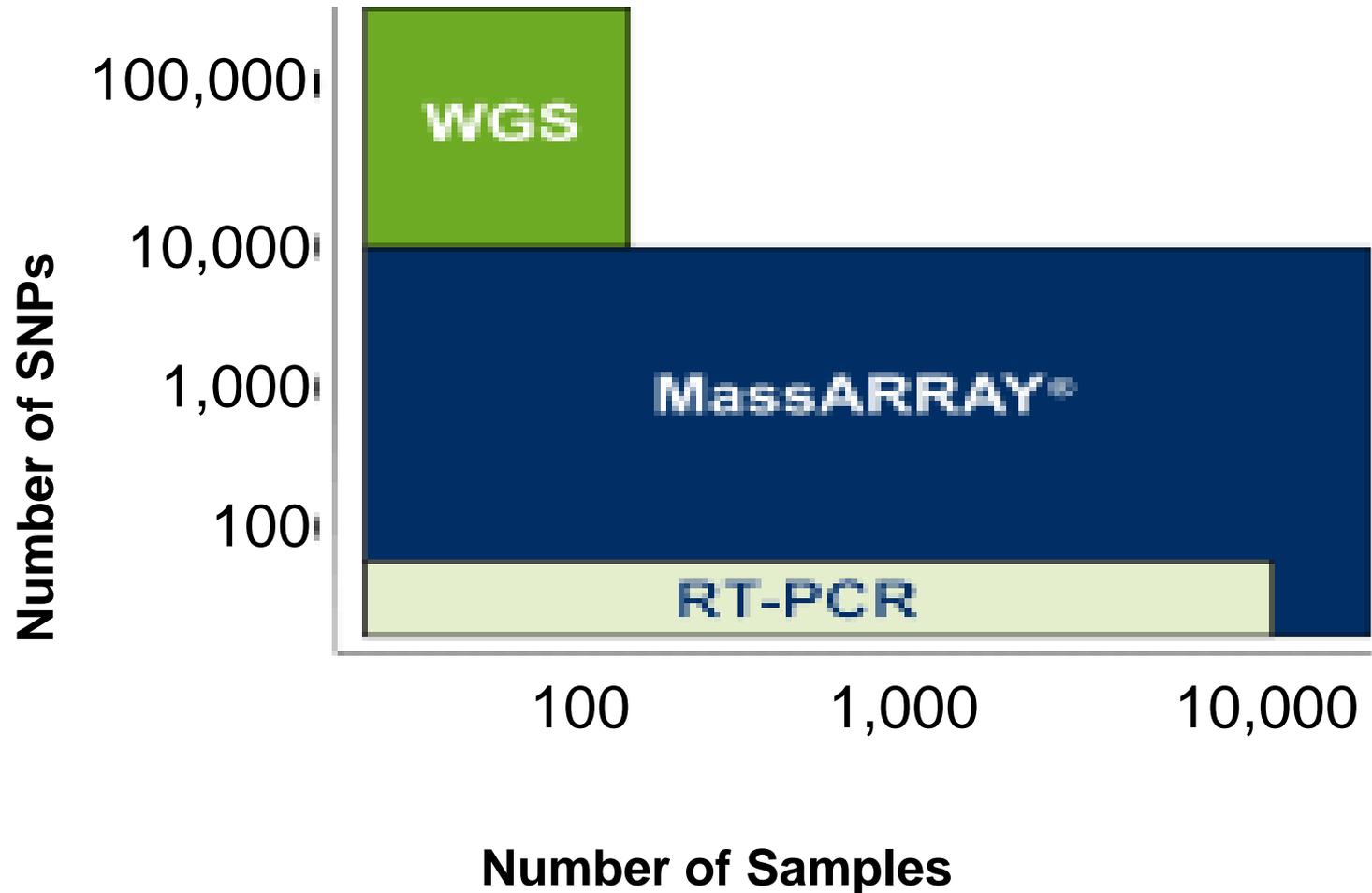
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3' C T A G G U G C G U G A G G C U G A A G G C G U G A G C U G G T A G G C U G A G C U G G T A G G T 5'



## Endonucleolytic mutation analysis by internal labelling (EMAIL)\*

\*Michael J. Cross, Daniel L. E. Waters, L. Slade Lee, and Robert J. Henry.  
Endonucleolytic mutation analysis by internal labelling (EMAIL).  
Electrophoresis 2008, 29, 1291–1301

# Technologies for different applications

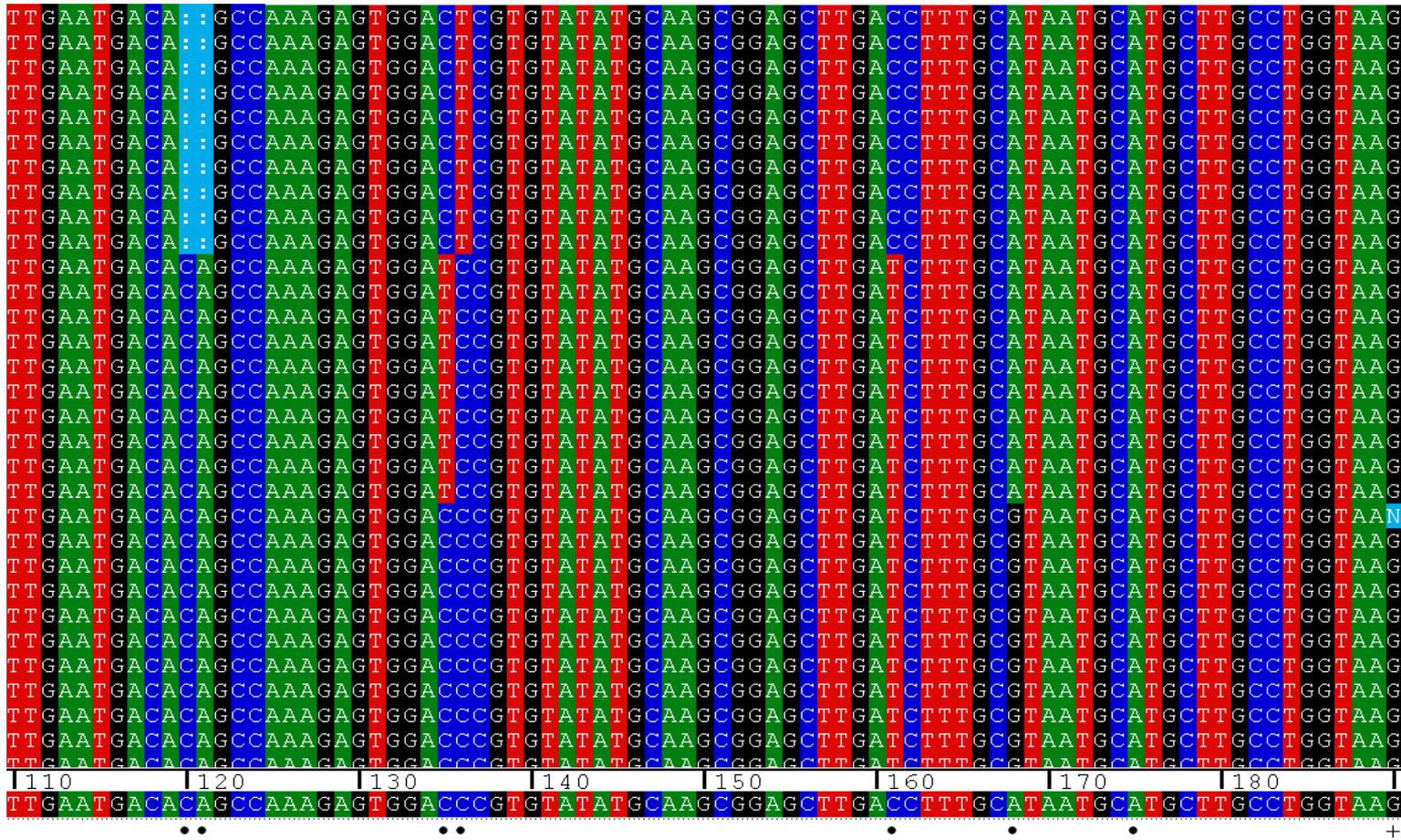




# NGS applications for SNP discovery

- Amplicon sequencing
- cDNA sequencing
- Sequencing of gene rich regions
- Whole genome sequencing

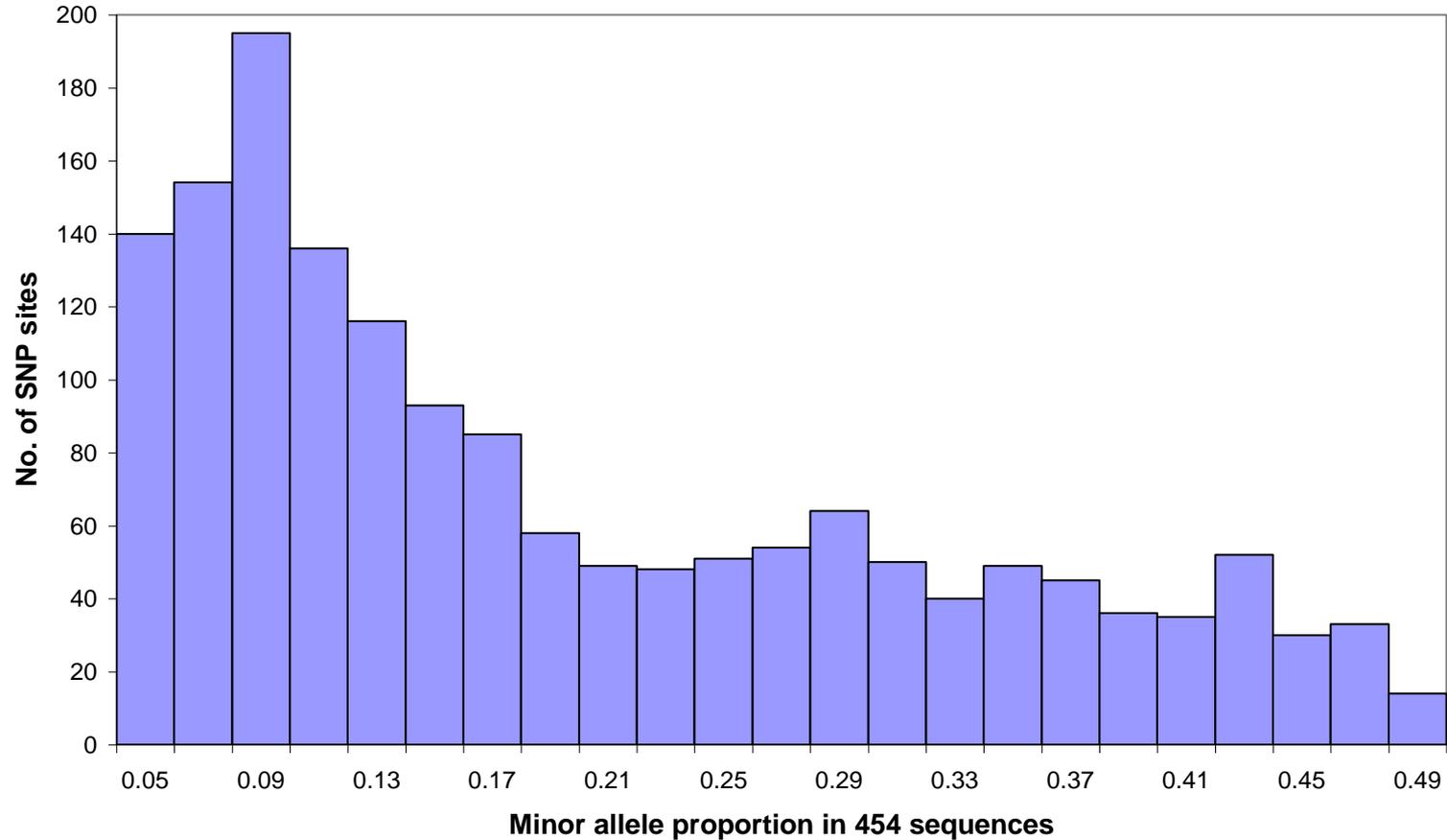
# SNP discovery in sugarcane



# Summary of SNP detection in 454 sequences of 300 amplicons

	<b>Female (<i>S. officinarum</i>) parent</b>	<b>Male (offic. X spont.) Parent</b>
<b>Products with one consensus seq.</b>	<b>247</b>	<b>242</b>
<b>Total length of amplicons</b>	<b>58 kb</b>	<b>57 kb</b>
<b>Products with one or more SNPs</b>	<b>213</b>	<b>227</b>
<b>Products SNPs absent</b>	<b>34</b>	<b>15</b>
<b>SNPs with rare allele frequency <math>\geq 4\%</math></b>	<b>1,013</b>	<b>1,632</b>
<b>Mean SNPs per amplicon</b>	<b>4.76</b>	<b>7.19</b>
<b>Average sequence depth at SNP sites</b>	<b>279</b>	<b>257</b>
<b>Candidate SD SNPs (4% to 15% freq.)</b>	<b>216</b>	<b>788</b>

# Frequency of rare SNP allele



# 454 SNP discovery

- One SNP every: 35/58 bases
- Developed SNP identification macros that also produce a consensus sequence with SNP sites coded ready for MassARRAY Assay Design software
- ~ 90% of assays validated a SNP
- Published in Plant Biotechnology Journal

# Pipeline for mapping genes of interest using SNPs

Discover & evaluate SNPs

454 re-sequencing



Develop/test SNP marker assays

Sequenom

MassARRAY platform



Genotype mapping population

# Impact of NGS

- Gene discovery
- Promoter analysis
- Expression profiling
- Conventional breeding
- GM sugarcane

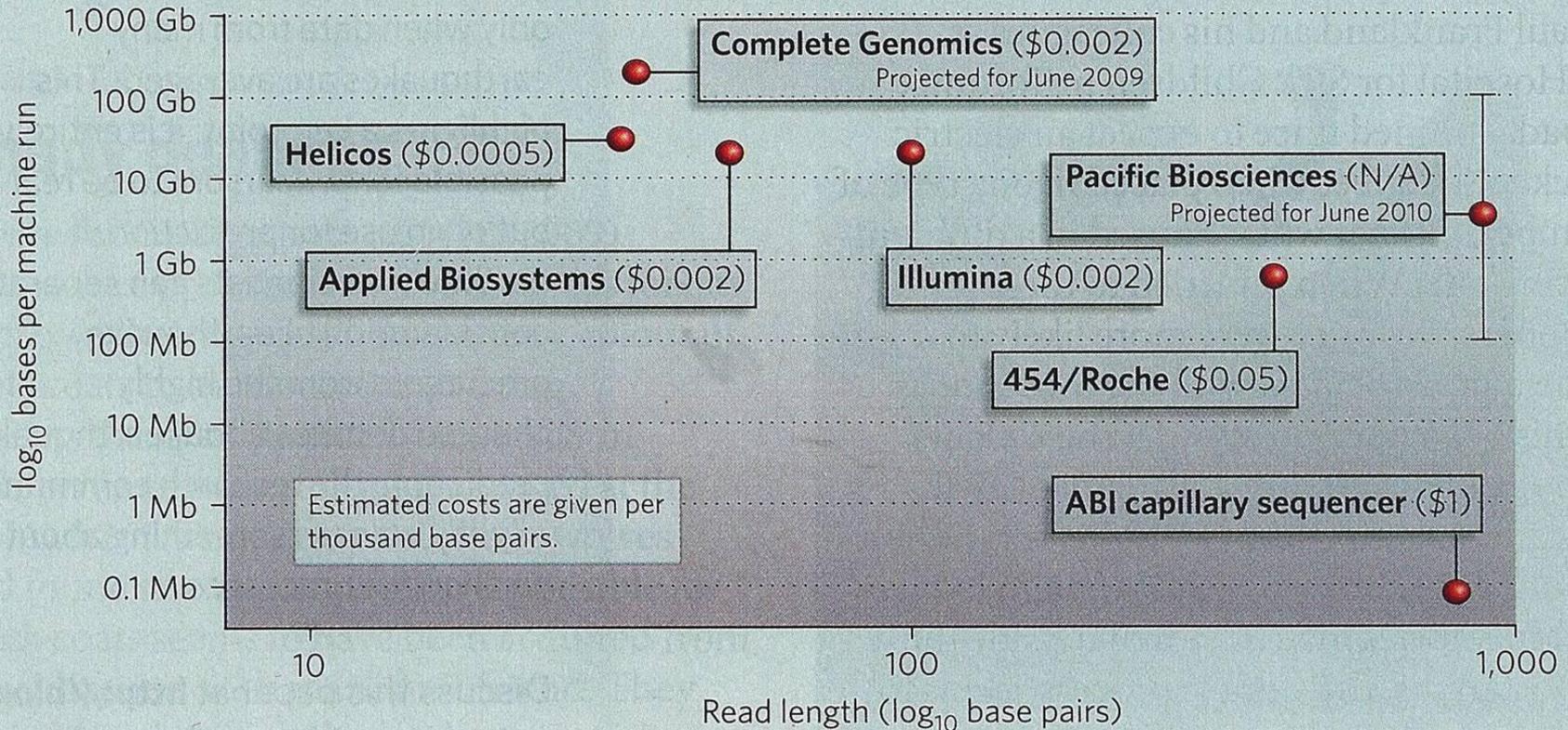


# Southern Cross Plant Genomics

- NGS sequencing facility (ARC)
- Bioplatforms Australia (NCRIS)
- Intersect (NSW)

# THE SEQUENCING RACE

The increasingly crowded market for genome-sequencing machines includes new entrants looking to push the boundaries in both speed and accuracy.



E. C. Hayden (2009) Genome sequencing: the third generation in Nature 12 February 2009

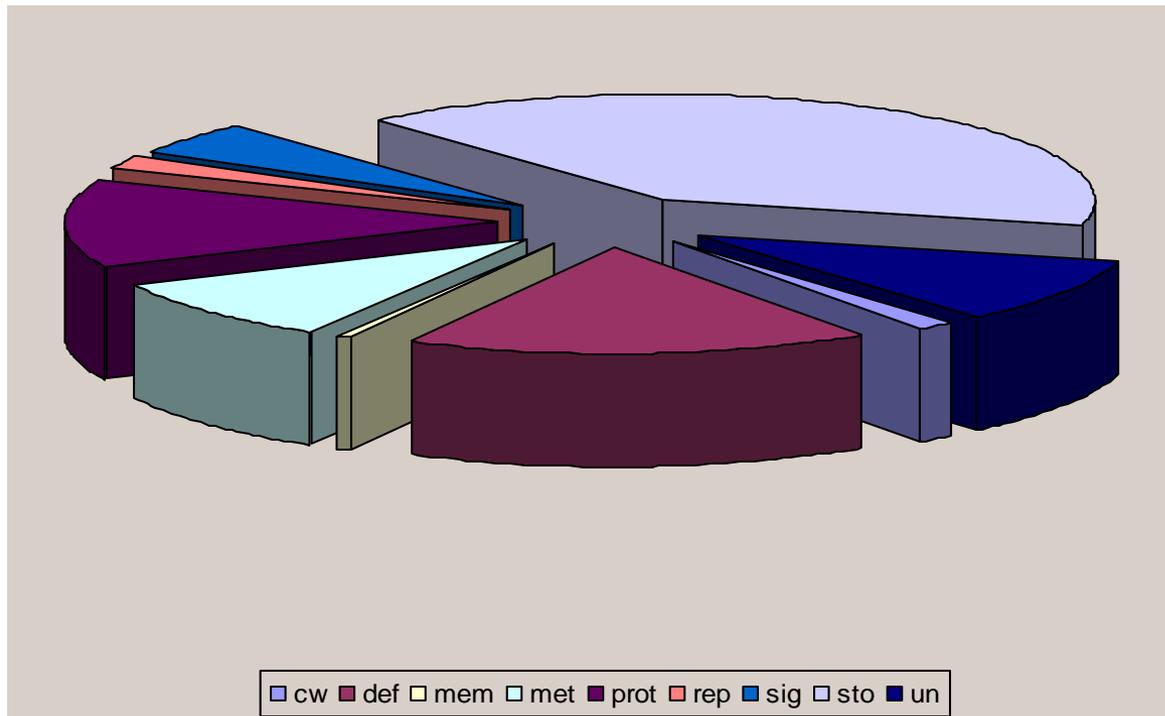
# SOLiD 3 R&D, now and future direction

- Currently exceeding 40Gb
- Sequencing to 100 bp
- 1B tags through greater bead density
- 100Gb by end of year
- Road map for 250Gb
- \$10K genome at high coverage by end of year

# NGS experiments

- Wheat genome sequencing
- *Oryza* reference genome sequencing
- Sugarcane gene sequencing
- Wheat transcriptome analysis
- Rainforest biodiversity
- Eucalypt genome re-sequencing

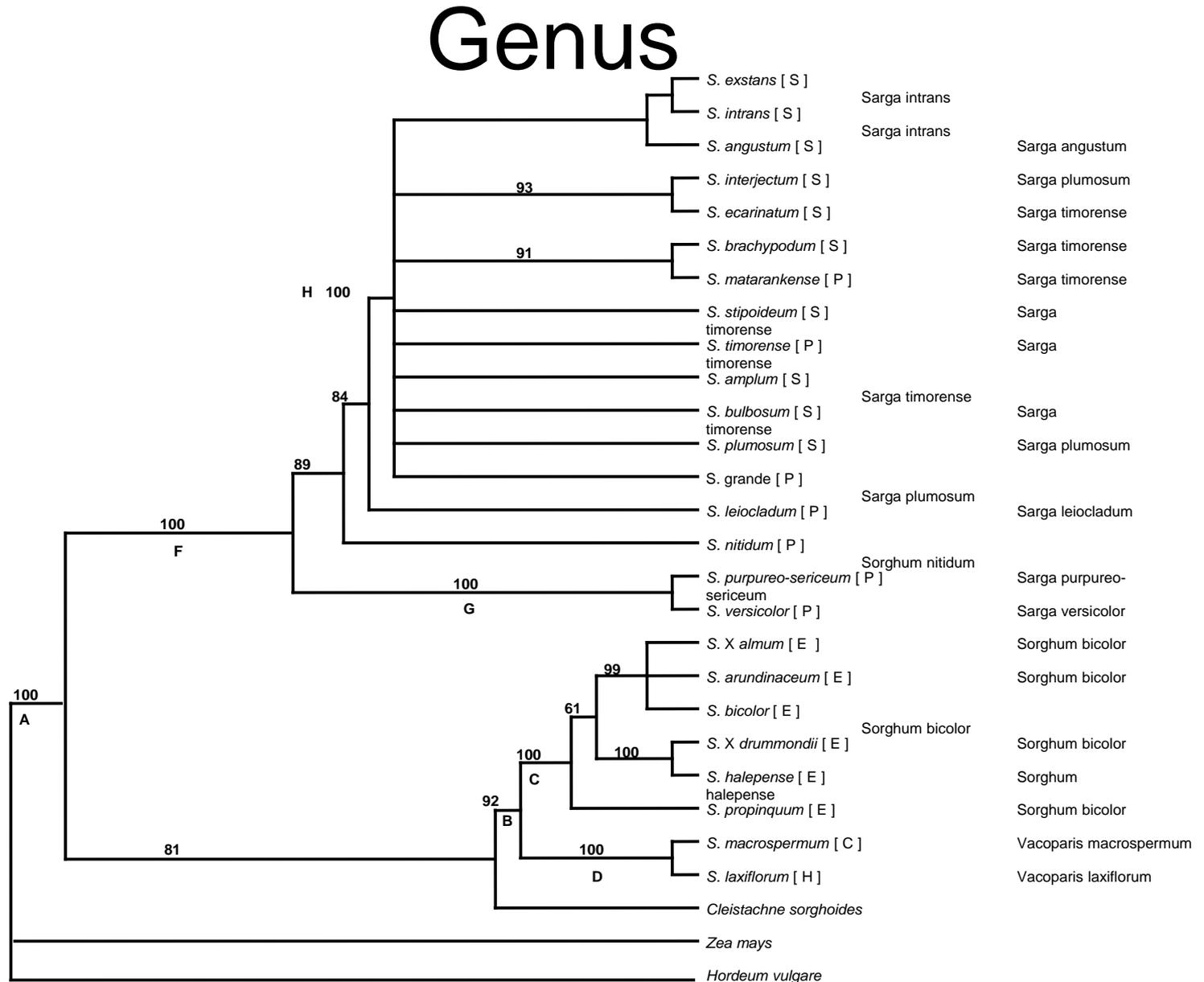
# Next Generation DNA sequencing accelerated discovery of genes for food and energy traits



# Sugarcane genome sequencing



# Relationships within the Sorghum Genus



# Sugarcane Genome Sequencing- CRC Project

- SCU/CSIRO
- Focus on gene rich parts of genome
- Establishes allelic diversity within and between genotypes
- Assignment of alleles to genomes
- Target SNP mining
- Facilitates gene mapping
- International collaboration

# Sugarcane volume

- **GENETICS, GENOMICS AND BREEDING IN CROP PLANTS**

**Publishers: Science Publishers, Inc., New Hampshire, Jersey, Plymouth**

- **Volume 10**

**Sugarcane Robert Henry Editor**

# Biofuels CRC

## Mission:

Transport energy self  
sufficiency and reduced  
carbon emissions from  
Australian transport

# Acknowledgements



Australian Government  
Rural Industries Research and  
Development Corporation



Australian Government  
Australian Research Council



Grains  
Research &  
Development  
Corporation



# Acknowledgements

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