Translational research in sugarcane

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Sugarcane in Brazil

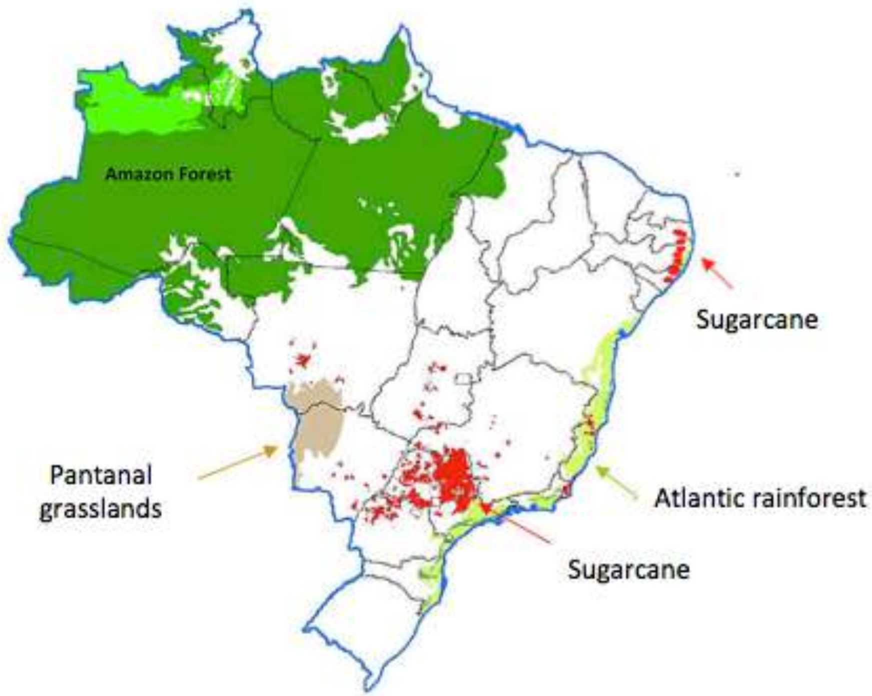
Brazil is the biggest grower among 80 countries that cultivate sugarcane

São Paulo state: produces 60% of sugarcane cultivated in Brazil

Revenues: U$ 43.8 billions (2012)

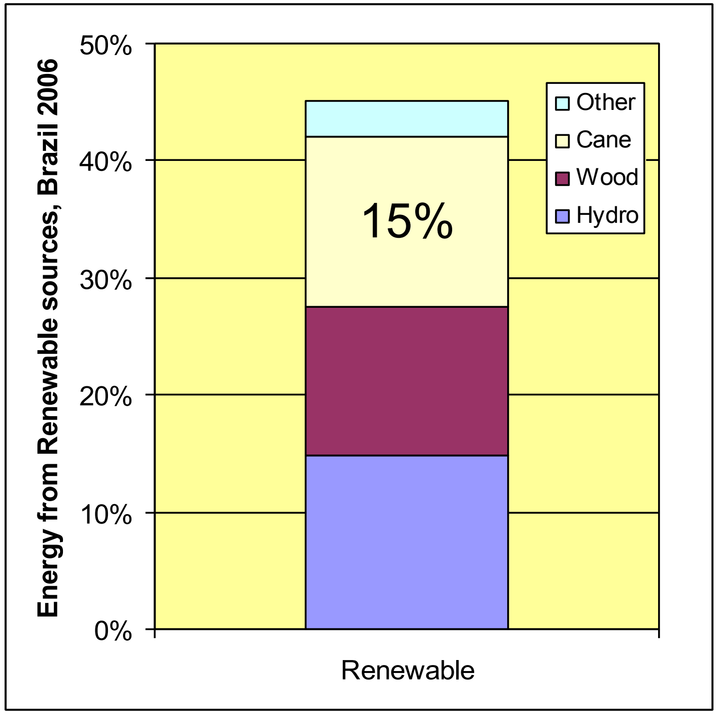
Jobs: 1.09 million workers

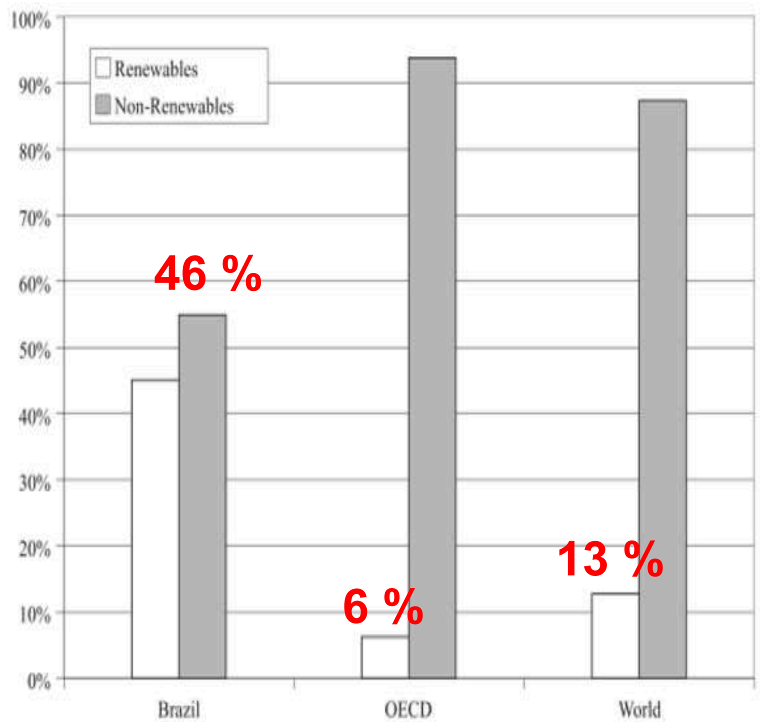
Sugarcane is 3-6 m high and accumulates sugar in the stalks (up to 42% dry weight after 12 months)



Source: Goldemberg, 2008

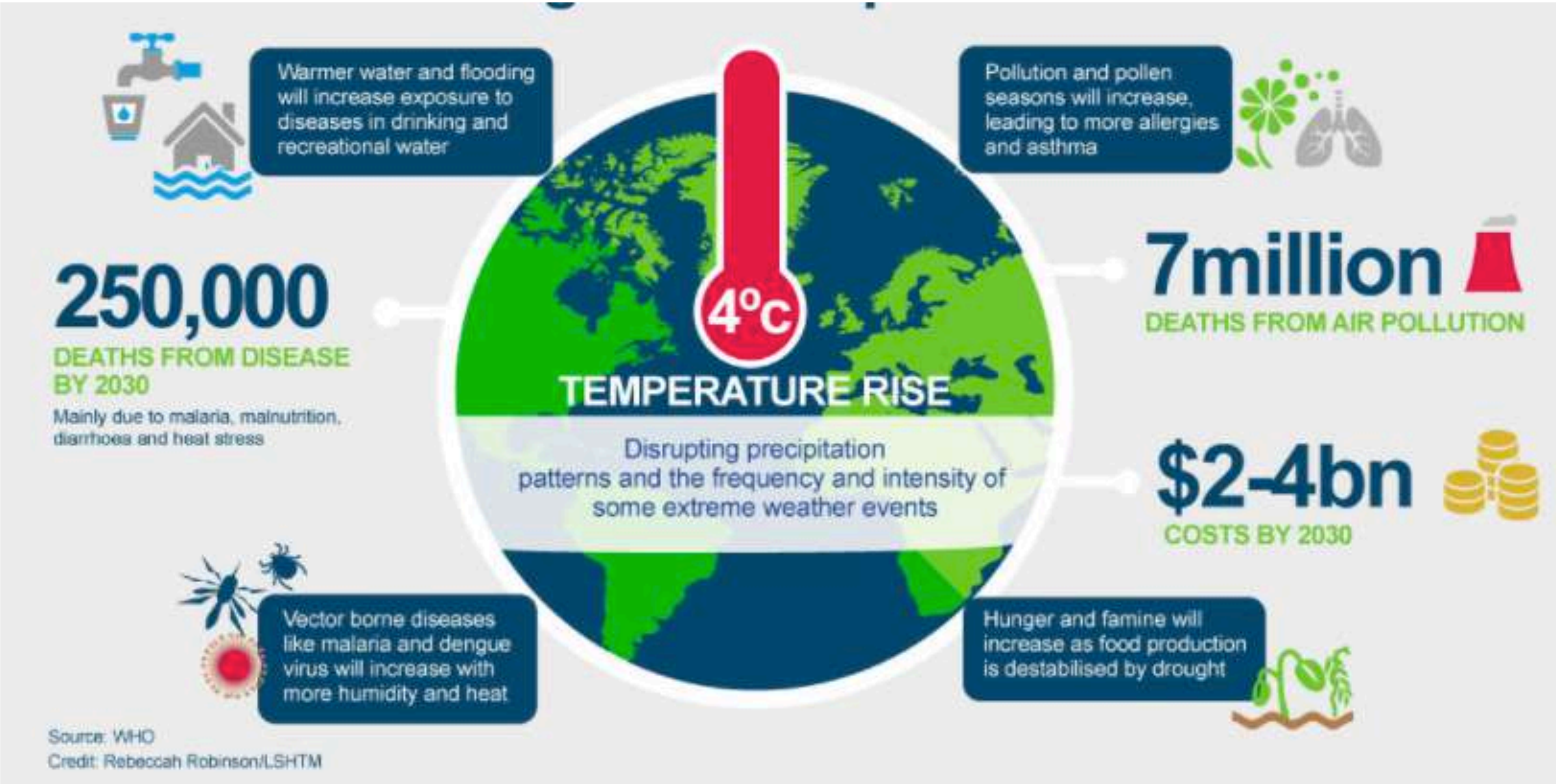
Energy from renewable sources in Brazil





Source: Leite (2009). Energy in Brazil: Towards a Renewable Energy Dominated System.

How climate change could impact the world



Drought effects on sugarcane production

From time to time, water deficits develop in several regions, reducing productivity

Table 1 | Estimated losses in sugarcane fields due to drought stress.

Year	Region	Losses	Rain (mm H_20)/ percentage of the expected rain
2008	São Paulo State	6.3% (Castro, 2008)	419.5/49%
2010	Zona da Mata (Pernambuco State)	40% (Cavalcanti, 2010)	300/50%
2012	Alagoas State	20% (Agência Globo, 2012; Sindaçucar, 2012)	774/48.6%
2012	Pernambuco State	35% (Associação dos fornecedores de cana de pernambuco, 2012; Camarotto, 2012)	629.4/50.7%
2013	Paraiba State	30% (G1 Agency, 2012; Silva, 2013)	n.a./up to 58.7%
2013	Zona da Mata (Pernambuco State)	25% (Brasilagro, 2013)	821/48.7%
2014	Ribeirão Preto (São Paulo State)	15% (Palhares, 2014)	480/51.6%

Source: Gentile et al., 2015

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Main questions

- What is the genetic and physiological basis underlying the different levels of biomass accumulation and of drought tolerance observed in sugarcane cultivars?

- What are the genes/proteins that will help us to enhance biomass accumulation and drought tolerance in sugarcane?




- Are these genes useful to enhance drought tolerance in *transgenic* sugarcane ?

- How can we transfer this knowledge to sugarcane growers?

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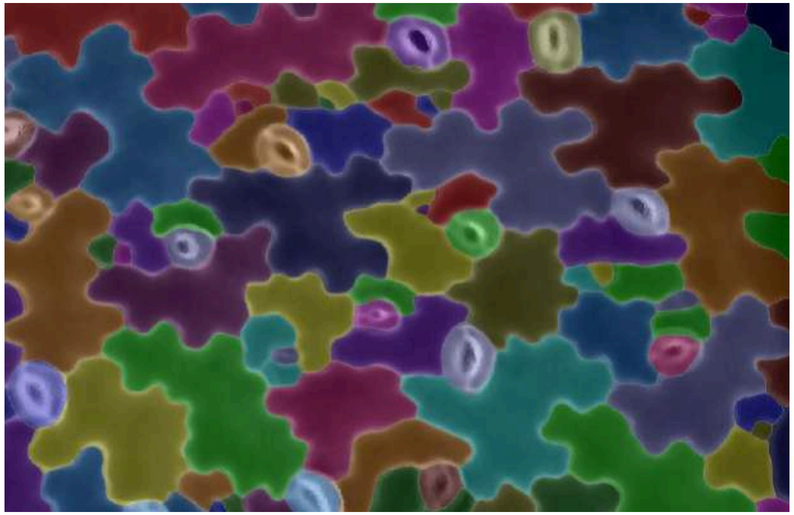
Translational approach for sugarcane research



MPI f. Plant Breeding Research/ Smith

Biotechnology

Proteomics

Transcriptomics &
Microtranscriptomics

Physiology &
Breeding

Metabolomics

Genomics

www.prospects.co.uk

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DELLA, the protein of the green revolution, controls internode development in sugarcane

Rafael Tavares
UNICAMP (Brazil)/SRA (Australia)

Prakash Lakshmanan
SRA (Australia)

Edgar Peiter
(Martin Luther University, Germany)

Antony O´Connell
SRA (Australia)

Camila
Caldana
CTBE (Brazil)

Renato Vicentini
UNICAMP (Brazil)

José Sérgio Soares
UNICAMP (Brazil)

FAPESP

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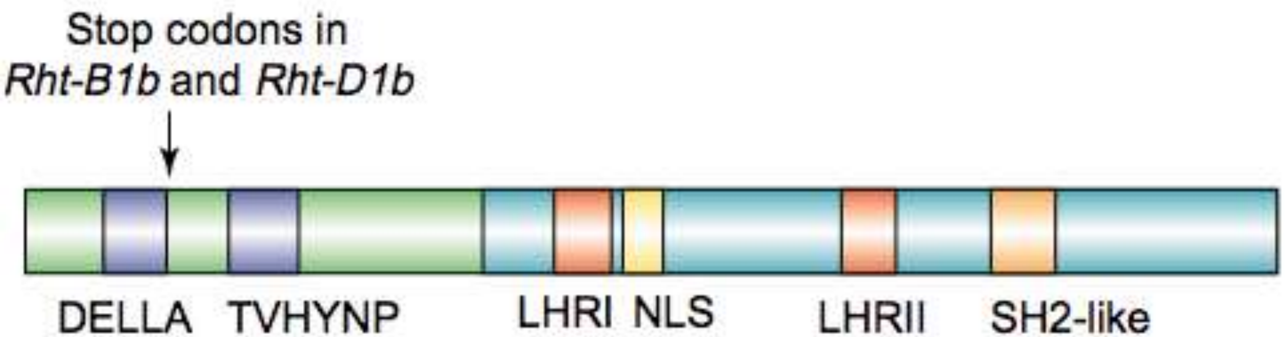
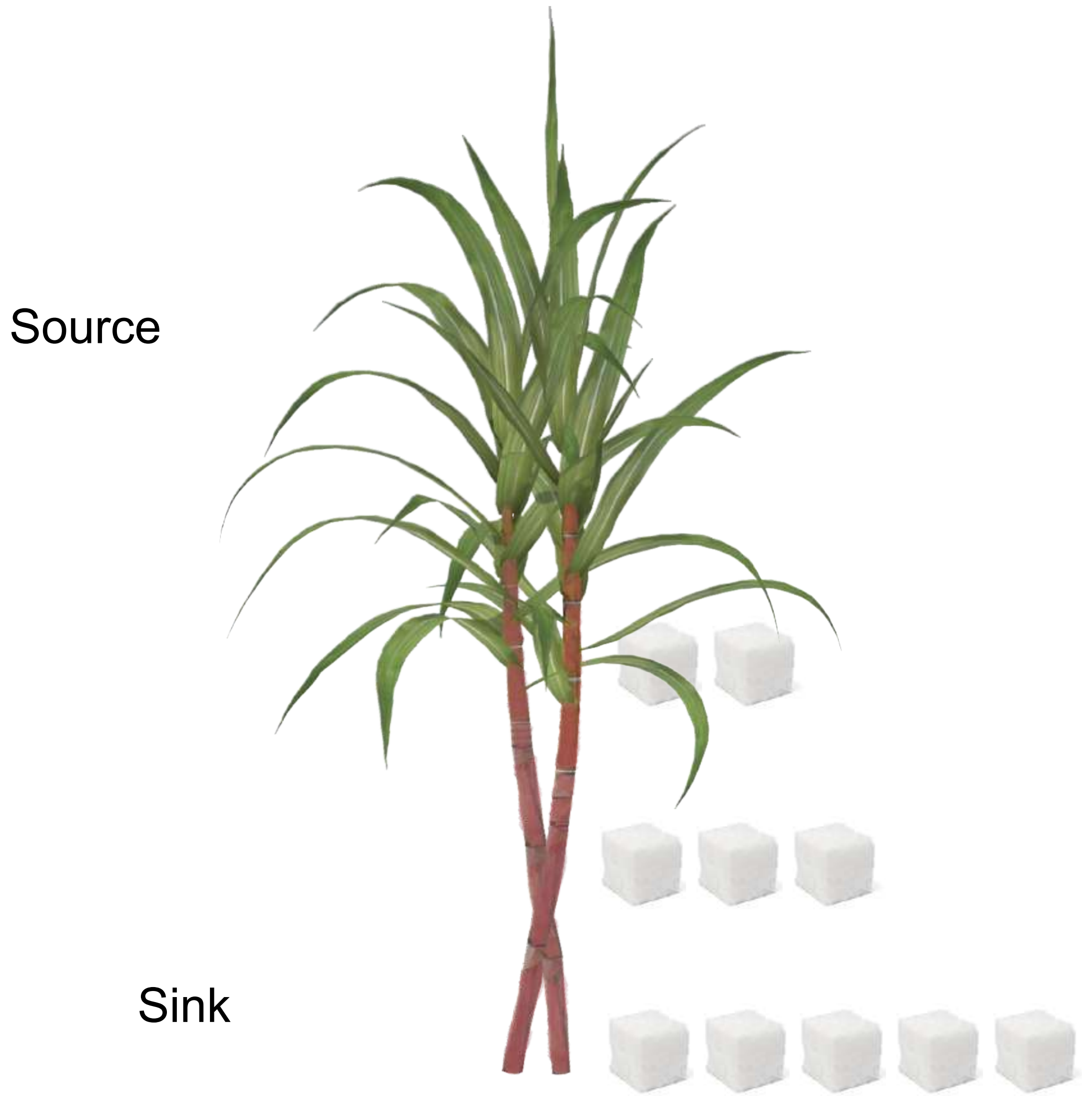
DELLA - the protein of the green revolution

Deegeo-woo-gen | Woo-gen | Calrose 76 | Calrose

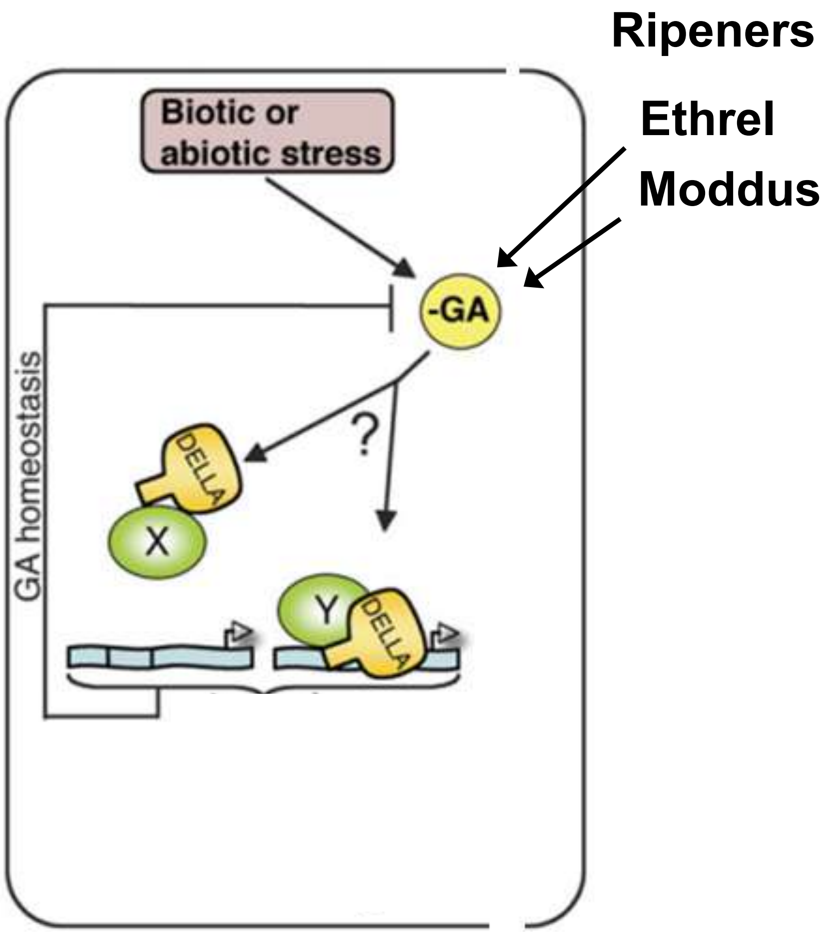


Hedden (2002). Trends Genet 19:5-9

Sucrose accumulation in sugarcane

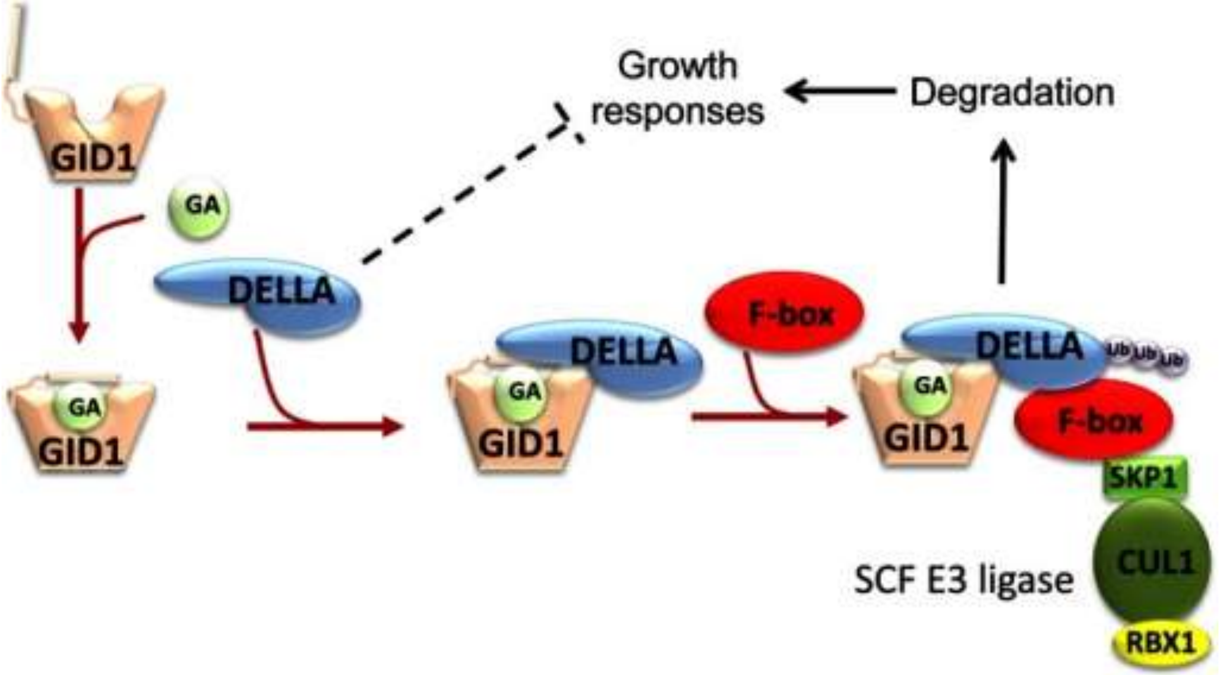


Abiotic stress and ripeners restrain growth in sugarcane



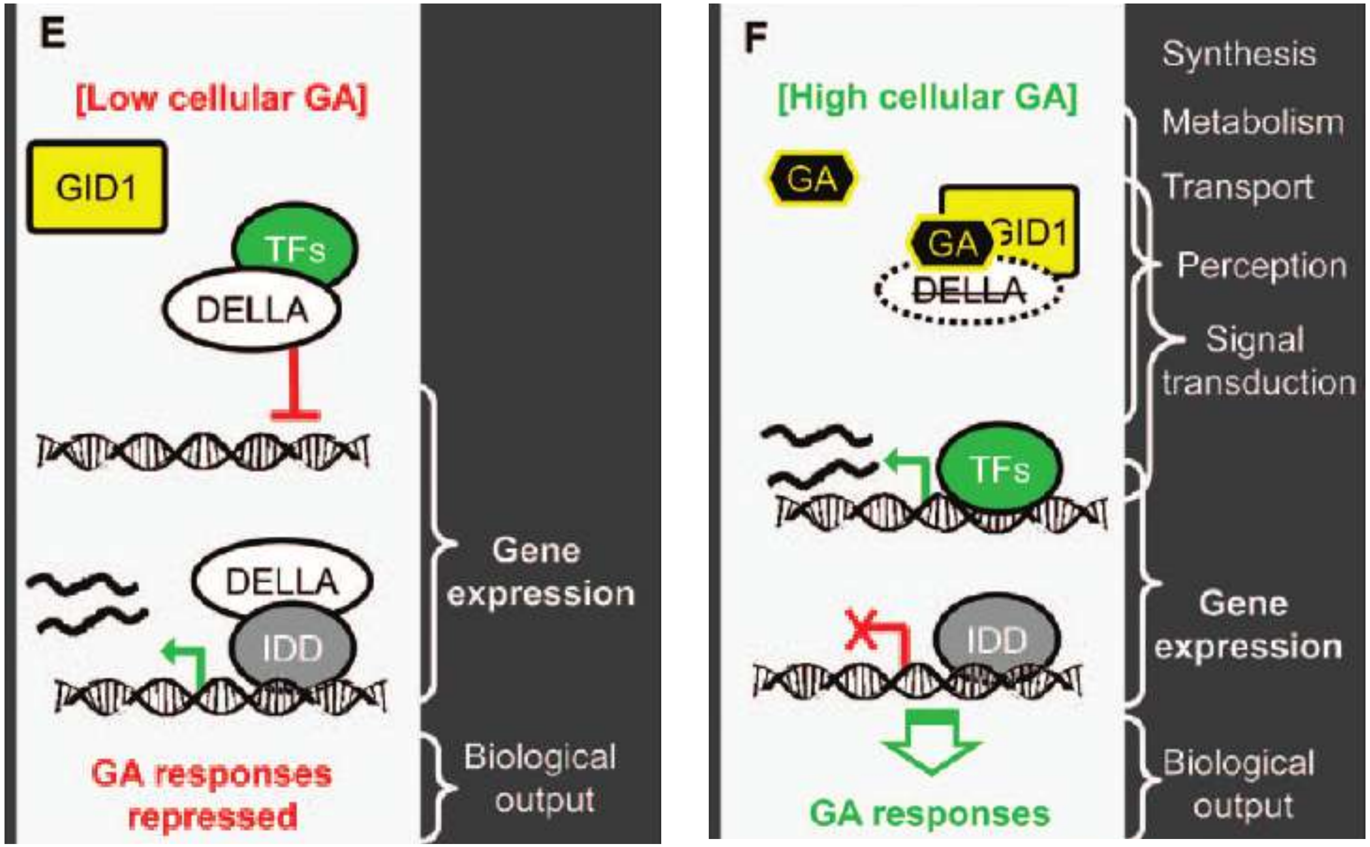
Journal of Experimental Botany, Vol. 60, No. 4, pp. 1085–1092, 2009
doi:10.1093/jxb/ern301 Advance Access publication 28 November, 2009
Journal of Experimental Botany
REVIEW PAPER
Releasing the brakes of plant growth: how GAs shutdown DELLA proteins

GA perception and signal transduction



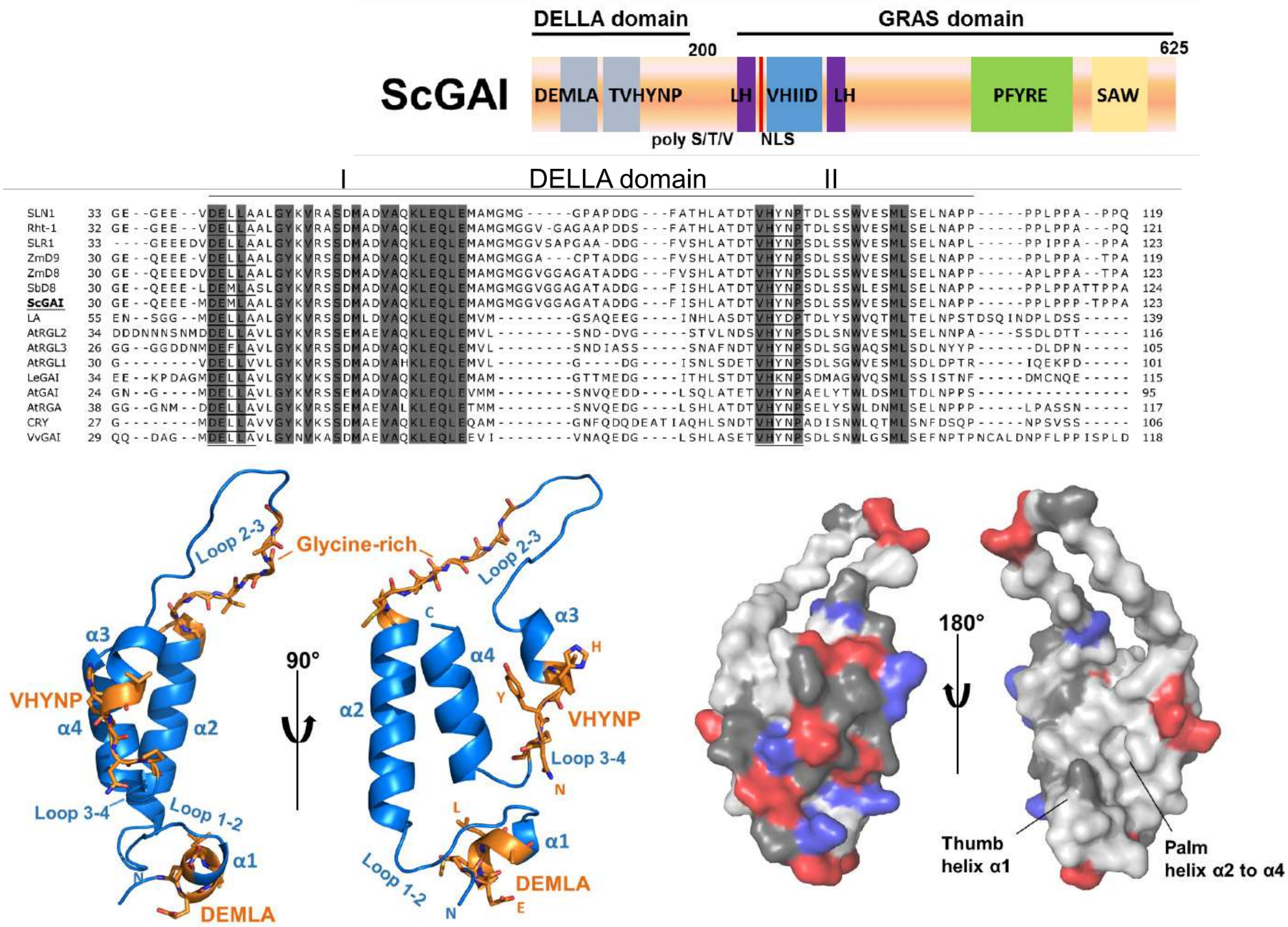
J Plant Growth Regul. 2015; 34: 740–760.

DELLAs interact with key regulatory proteins to modulate plant development

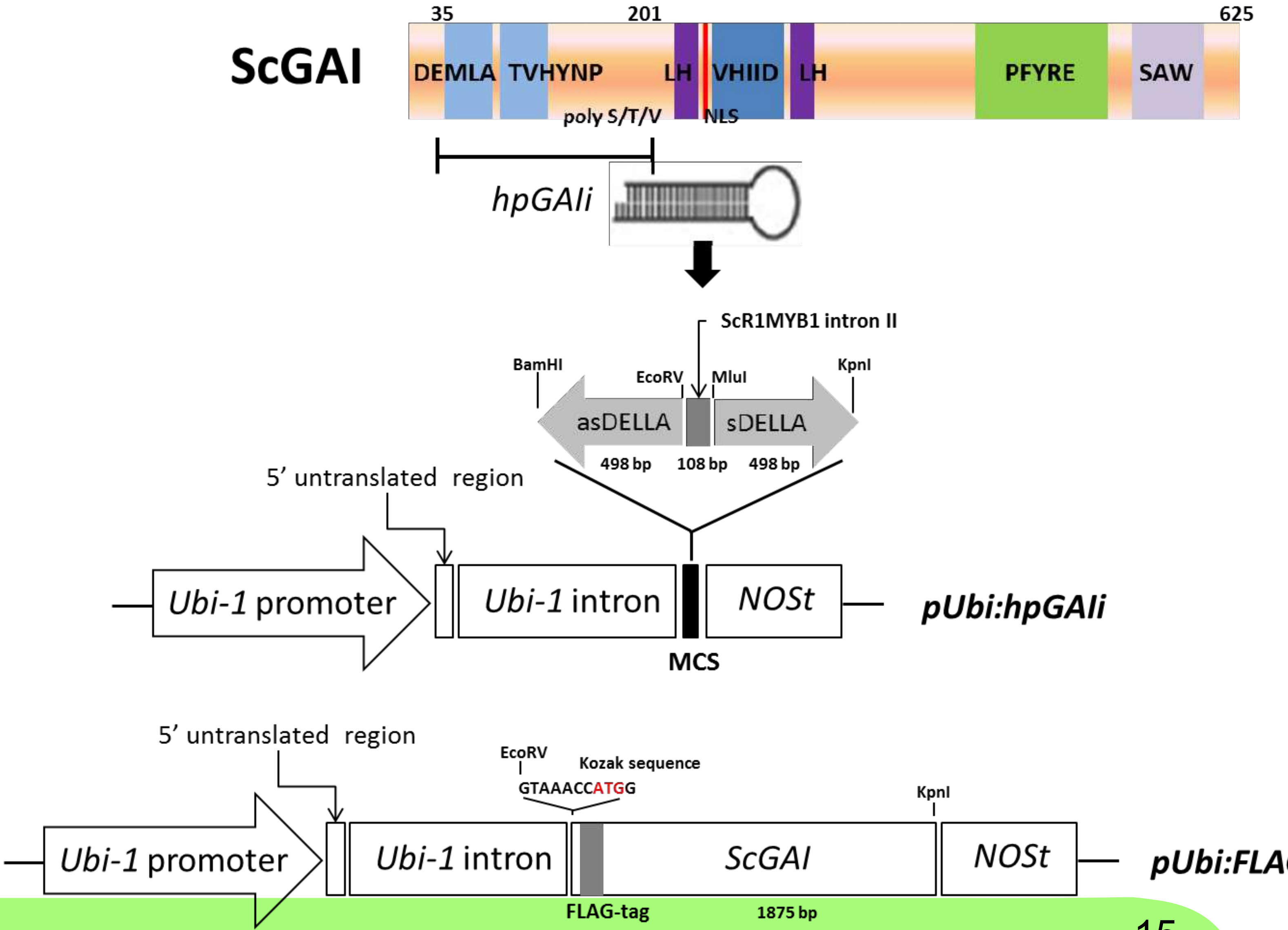


Stability and structure are essential for DELLA plays its role

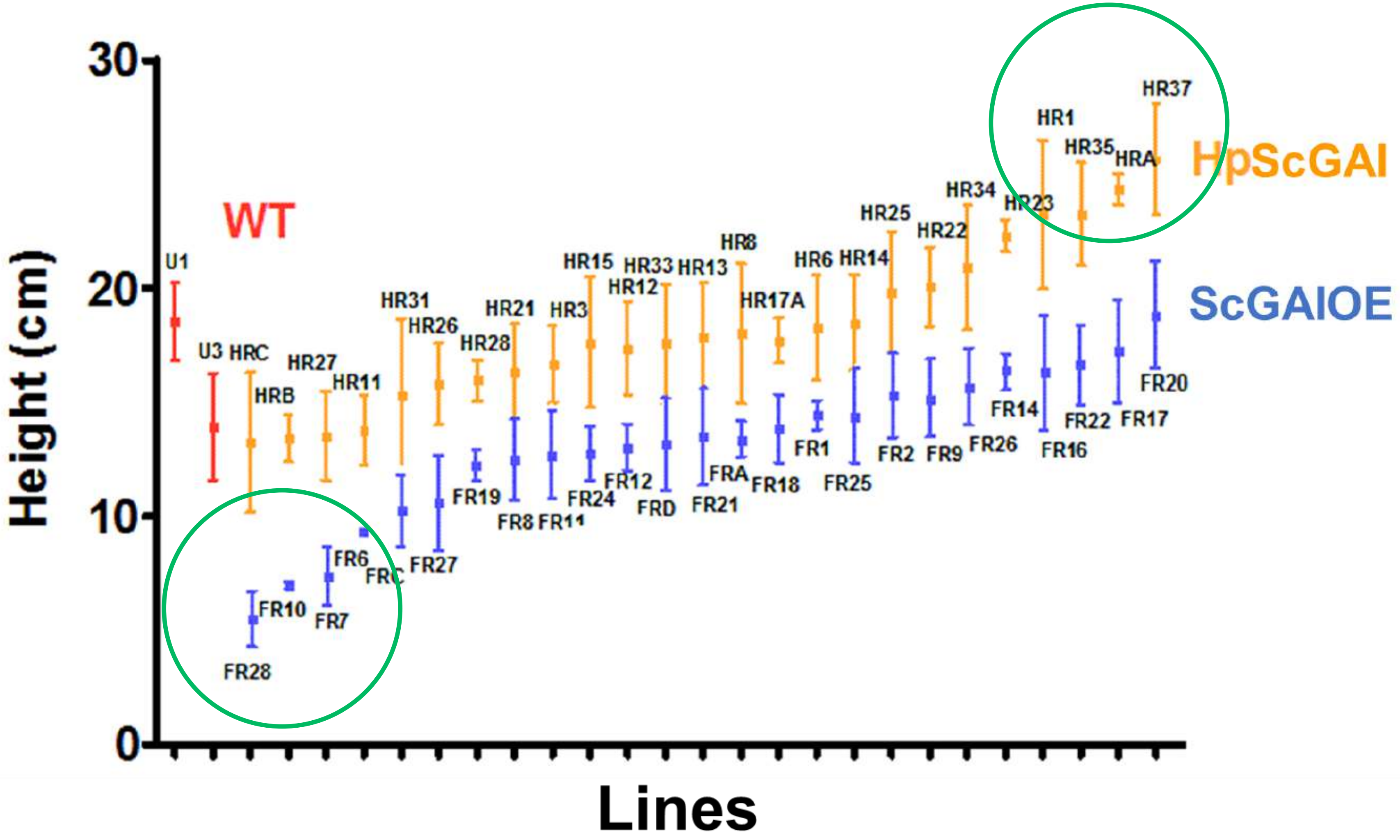
ScGAI encodes the nuclear DELLA protein in sugarcane



Overexpression and hairpin-mediated silencing of *ScGAI* gene in transgenic sugarcane plants



Phenotype screening of 3-months-old transgenic sugarcane plants



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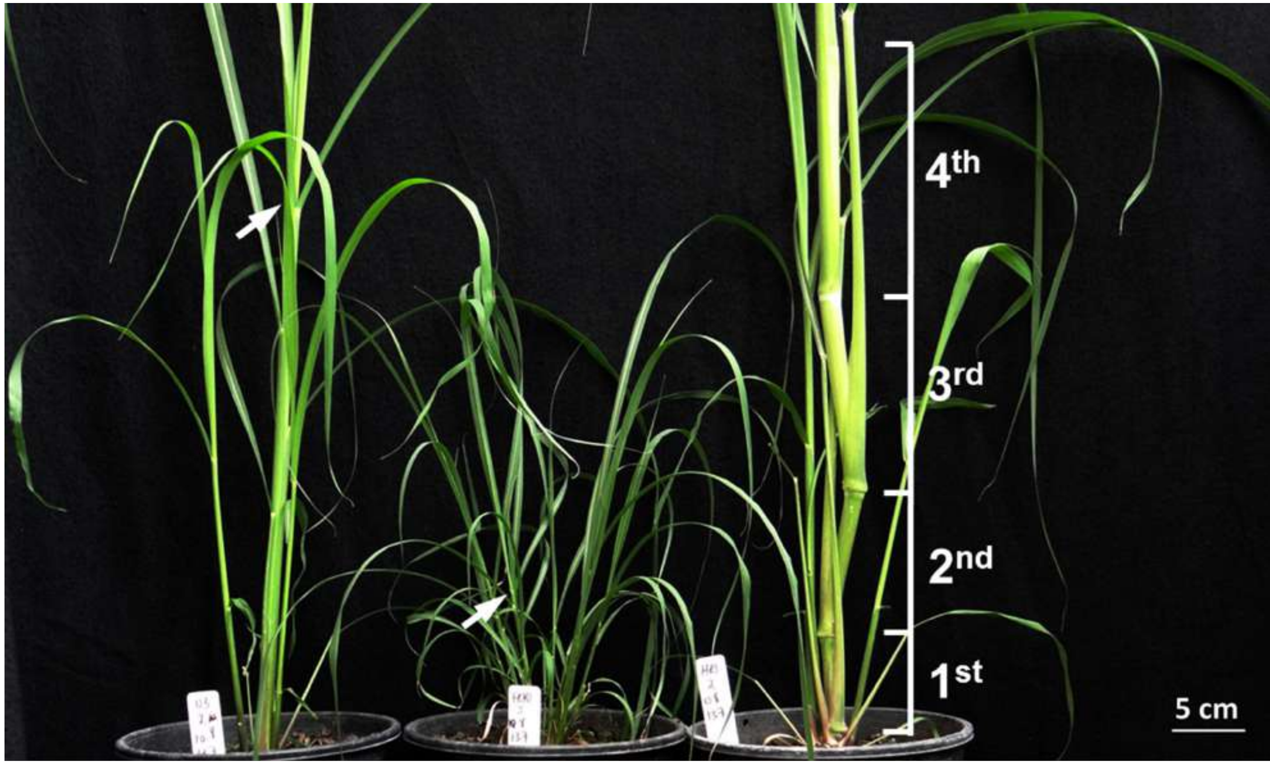
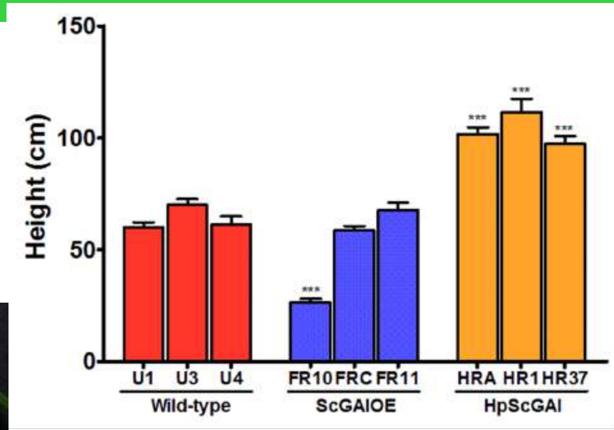
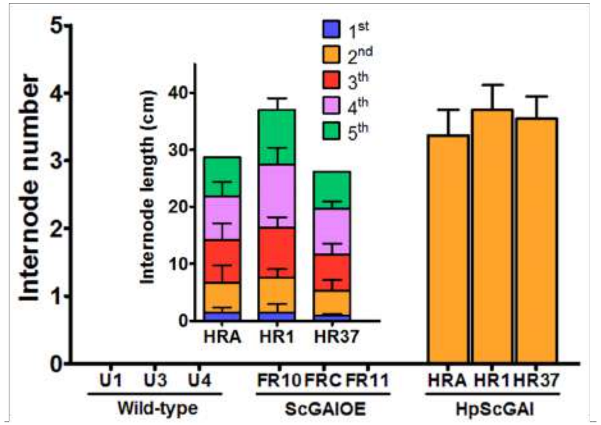
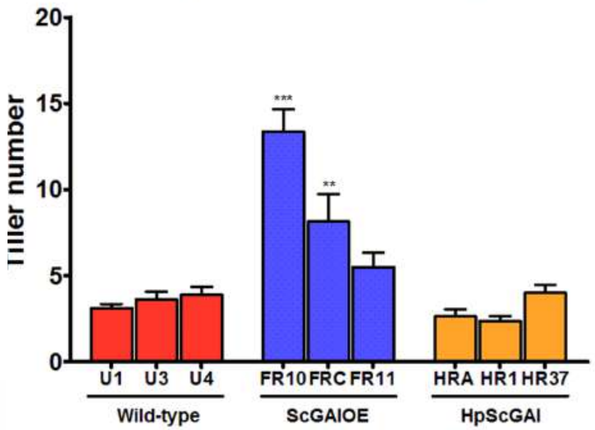
ScGAI regulates growth, tillering and internode elongation in 3-months-old sugarcane



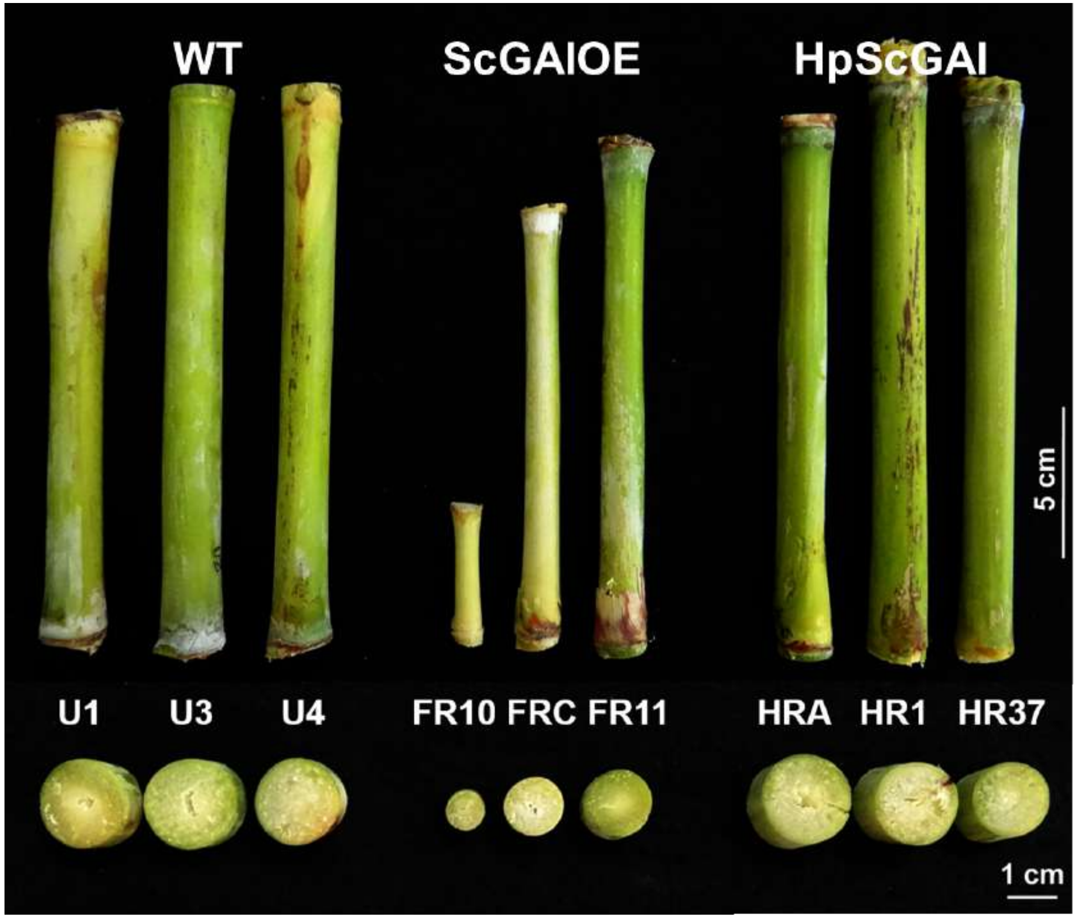
WT ScGAIOE HpScGAI

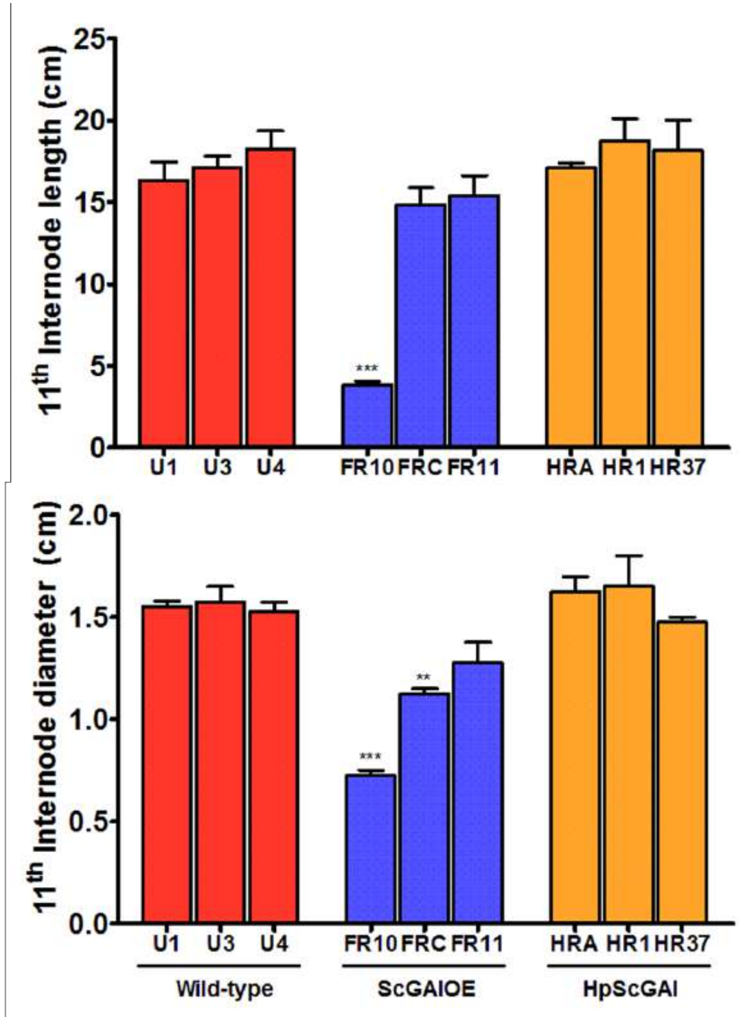




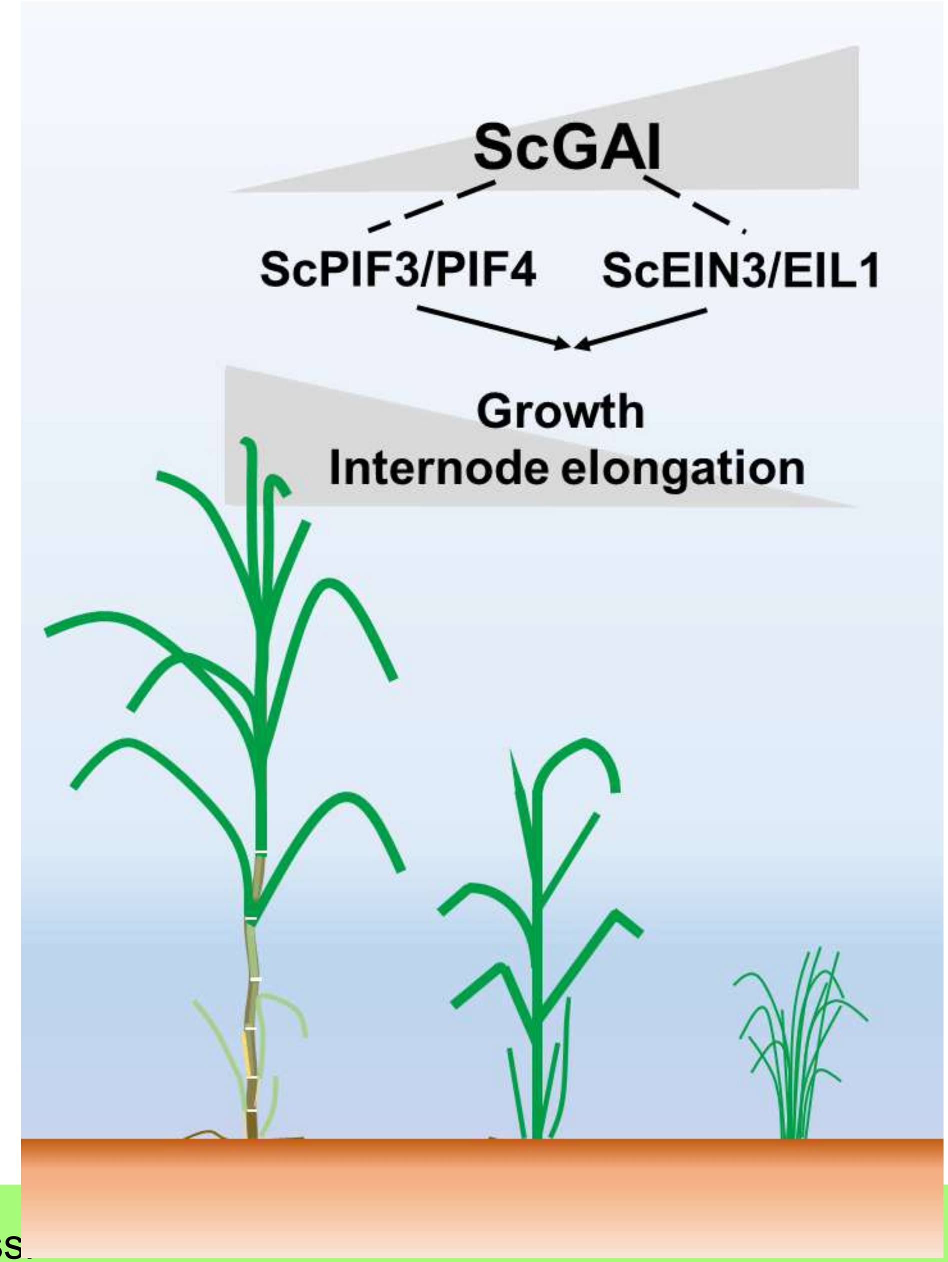


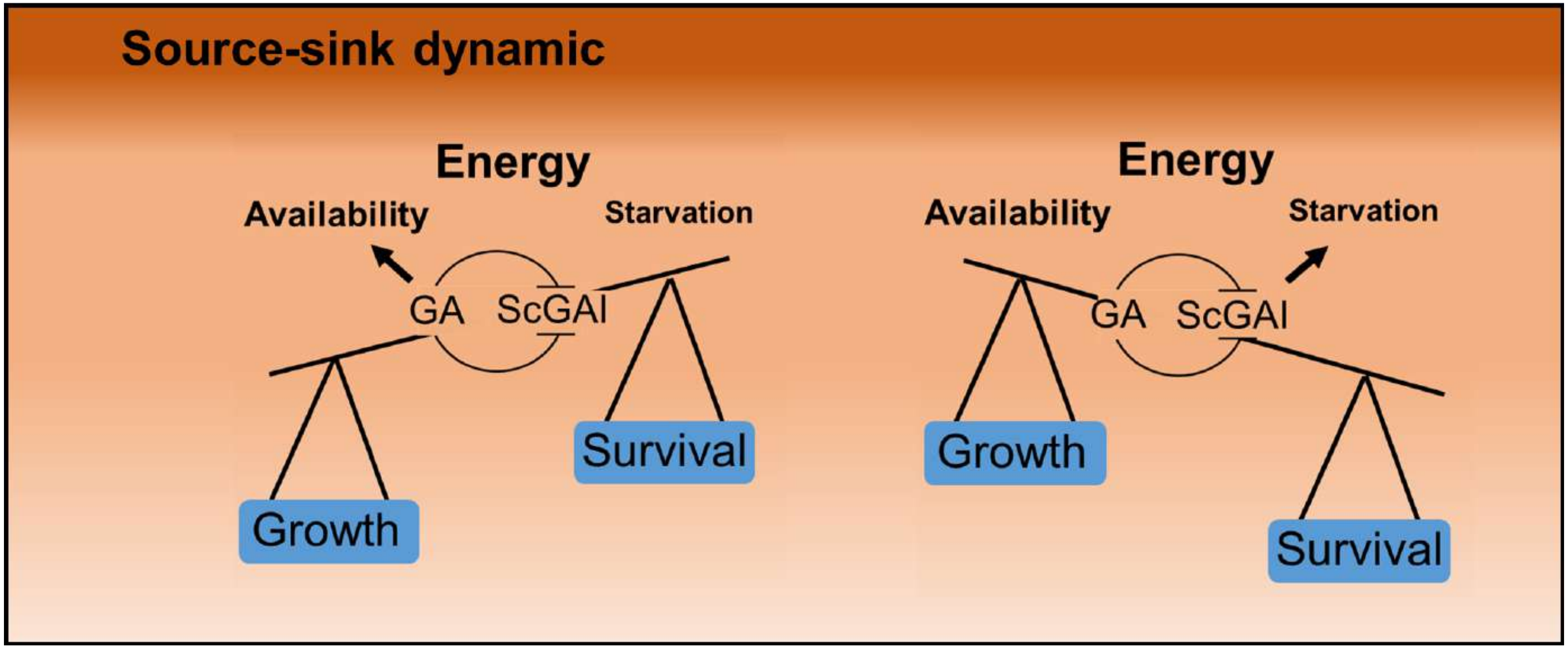
11th Internode from 6-months-old HpScGAI plants presented an inaltered diameter





ScGAI orchestrates growth and internode elongation




Source-sink dynamic
Energy
Availability
Starvation
GA
ScGAI
Survival
Growth
Energy
Availability
Starvation
GA
ScGAI
Growth
Survival

2016 dry at Pernambuco, Brazil
Reduction of 25% (tons of harvested cane)

Consortium for physiology, genomics, proteomics and biotechnology associated to drought stress in sugarcane

Laurício Endres (UFAL)

Monalisa S. Carneiro (UFSCAR)

Rejane J.M.C. Mansur (UFRPE)

Tercílio Calsa Jr. (UFPE)

Helaine Carrer (ESALQ/USP)

Glaucia M. Souza (IQ-USP)

Marcelo Menossi (Coordinator, IB-UNICAMP)

UFAL ufscar UFRPE UFPE USP UNICAMP

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Field experiment

Lower tolerance: RB72454, RB855536 e RB855113
Higher tolerance: RB92579, RB867515 e SP79-1011

Plants cultivated in the field in AL, PE and SP,
Irrigated or rainfed (sequeiro)

Leaves and internodes collected after 3, 7 and 11 months;

Several parameters were evaluated: gas exchange, proline content, leaf area, internode diameter, productivity, etc)

Drought (sequeiro)

Irrigated

RB867515, 7 months after planting

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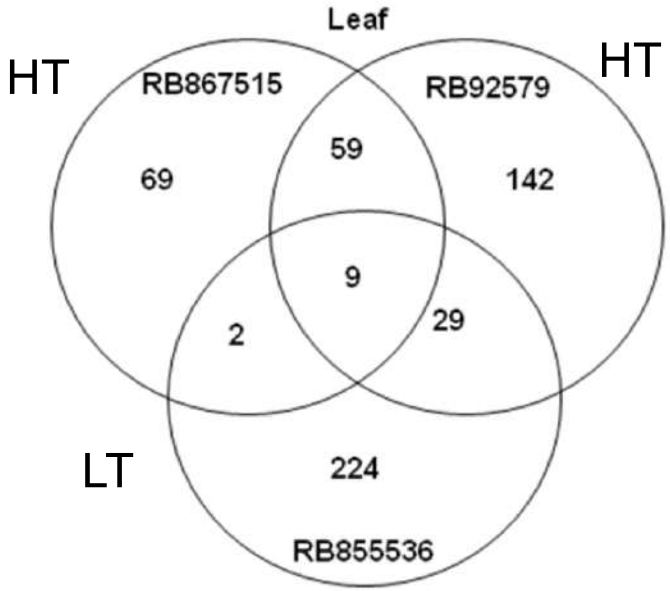
Sugarcane transcriptome under drought stress (7 MAP)

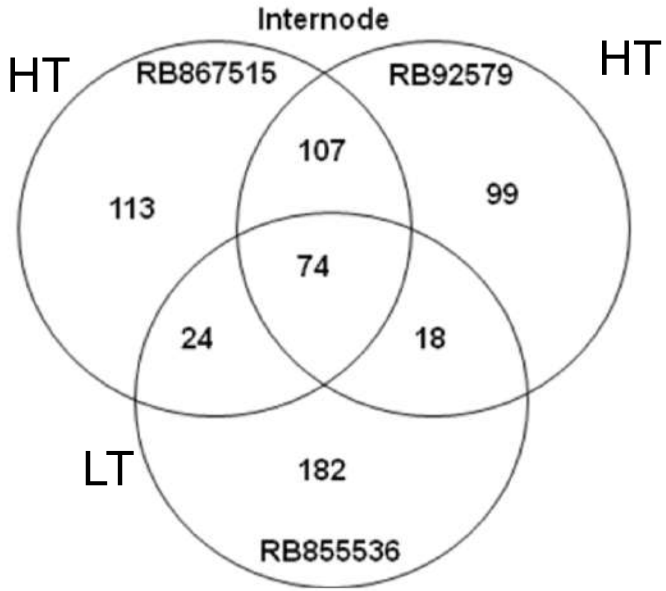
Agilent chips with 14,522 sugarcane genes
Leaves and first internode


Glaucia Souza


Maximiller
Dal-Bianco



534 genes



787 genes

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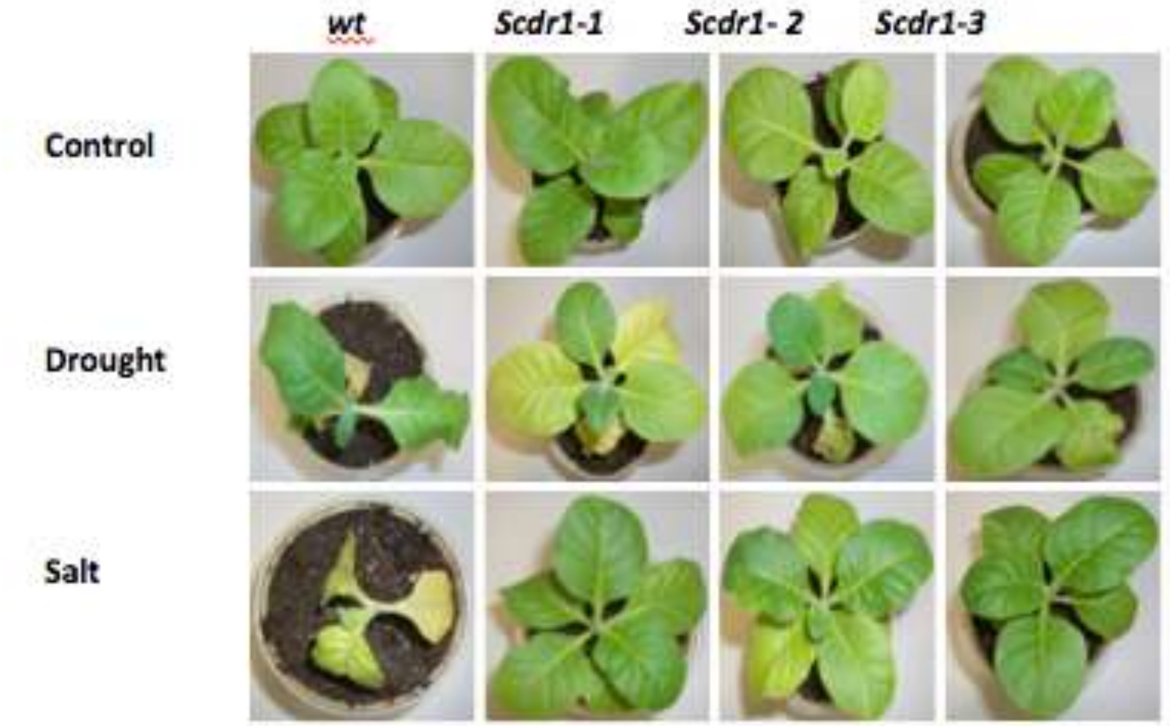
Gene *Scdr1* – tolerância à seca

Kevin Begcy

Planta de tabaco selvagem

Plantas transgênicas de tabaco contendo o gene *Scdr1* de cana



Plantas de tabaco foram cultivadas por 13 semanas sob condição controle, com irrigação.

Em seguida, as plantas foram divididas em três grupos:

Control: plantas mantidas sob irrigação;

Drought (seca): regadas com solução de polietileno glicol para criar déficit hídrico, e

Salt: irrigadas com NaCl para criar estresse salino.

FOX hunting system: large scale functional genomics


the plant journal

The Plant Journal (2009) 57, 883–894 doi: 10.1111/j.1365-313X.2008.03733.x

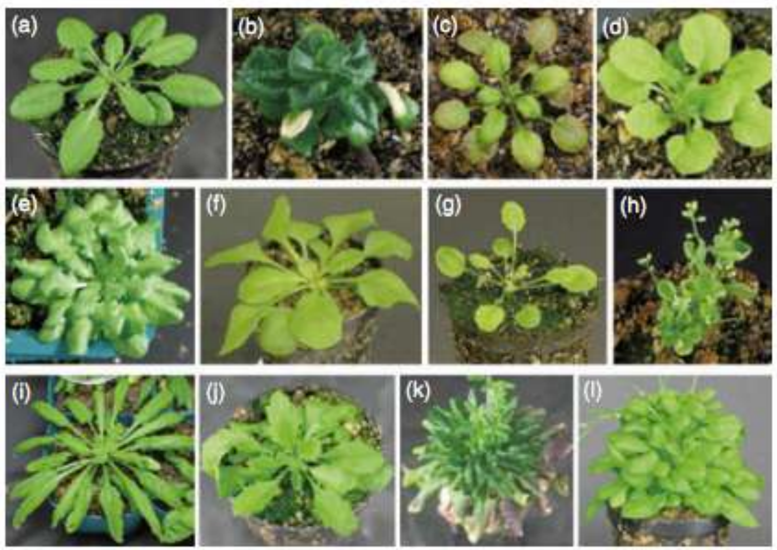
Systematic approaches to using the FOX hunting system to identify useful rice genes

Youichi Kondou[1,†], Mieko Higuchi[1,†], Shinya Takahashi[1,‡], Tetsuya Sakurai[1], Takanari Ichikawa[1], Hirofumi Kuroda[1], Takeshi Yoshizumi[1], Yuko Tsumoto[1], Yoko Horii[1], Mika Kawashima[1], Yukako Hasegawa[1], Tomoko Kuriyama[1], Keiko Matsui[1], Miyako Kusano[1], Doris Albinsky[1], Hideki Takahashi[1], Yukiko Nakamura[1], Makoto Suzuki[1], Hitoshi Sakakibara[1], Mikiko Kojima[1], Kenji Akiyama[1], Atsushi Kurotani[1], Motoaki Seki[1], Miki Fujita[1], Akiko Enju[1], Naoki Yokotani[2], Tsutomu Saitou[2], Kozue Ashidate[2], Naka Fujimoto[2], Yasuo Ishikawa[2], Yayoi Mori[2], Rie Nanba[2], Kazumasa Takata[2], Kuniko Uno[2], Shoji Sugano[3], Jun Natsuki[3], Joseph Gogo Dubouzet[3], Satoru Maeda[3], Miki Ohtake[3], Masaki Mori[3], Kenji Oda[2], Hiroshi Takatsuji[3], Hirohiko Hirochika[3] and Minami Matsui[1,*]

[1]*RIKEN Plant Science Center, 1-7-22 Suehiro-cho, Tsurumi-ku, Yokohama, Kanagawa 230-0045 Japan,*
[2]*Research Institute for Biological Sciences Okayama, 7549-1 Yoshikawa, Kibi-chuo, Okayama 716-1241 Japan, and*
[3]*Disease Resistance Research Unit, National Institute of Agrobiological Sciences, 2-1-2 Kan-nondai, Tsukuba, Ibaraki 305-8602, Japan*

- 11.000 rice genes
- 23.000 transgenic Arabidopsis plants

WT →



Vanessa Gonçalves

Dirk Inzé
VIB

Nathalie Gonzalez
VIB

Giovanna Guidelli

Goal: To overexpress 200 sugarcane genes in Arabidopsis

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Moderate water deficit using a robotic platform

WIWAM platform

http://bit.ly/WIWAMSC

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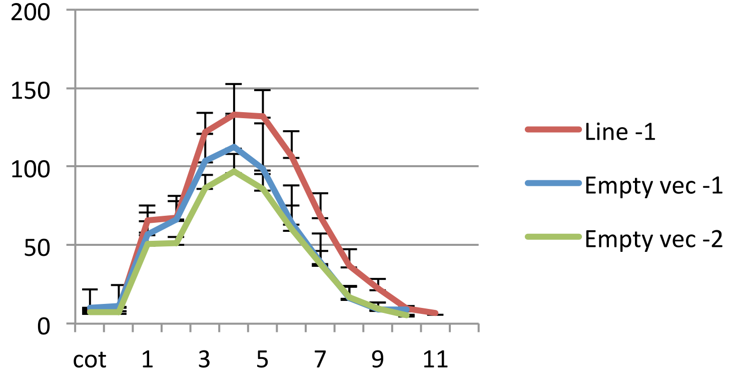
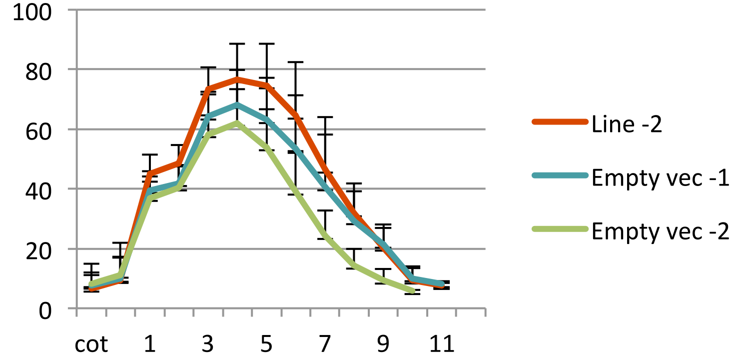
Leaf area under moderated water deficit

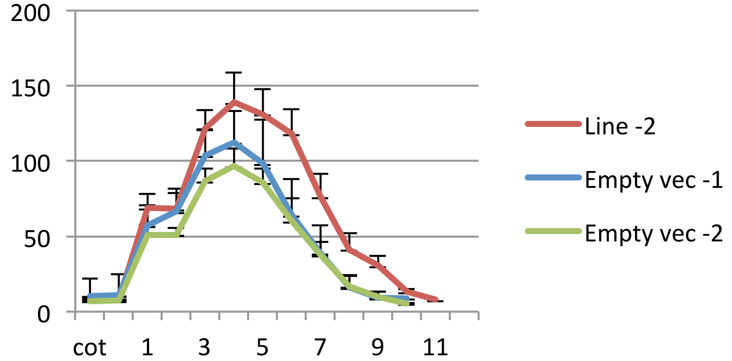
Control | Drought

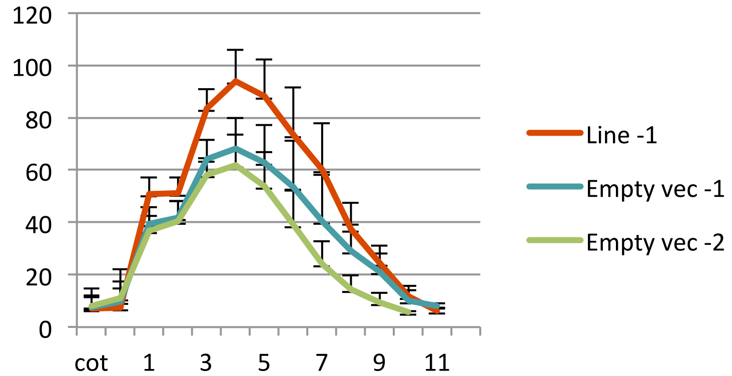
Pathogen response



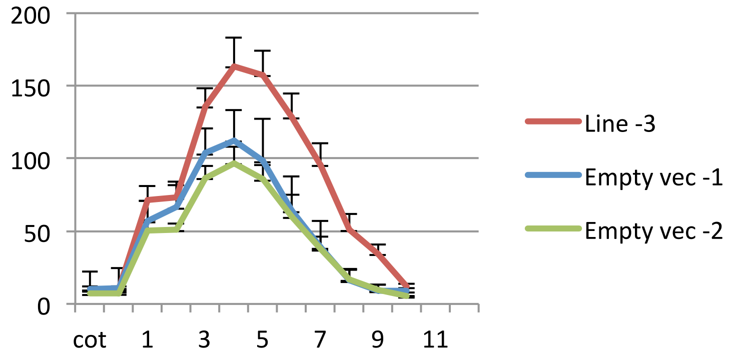


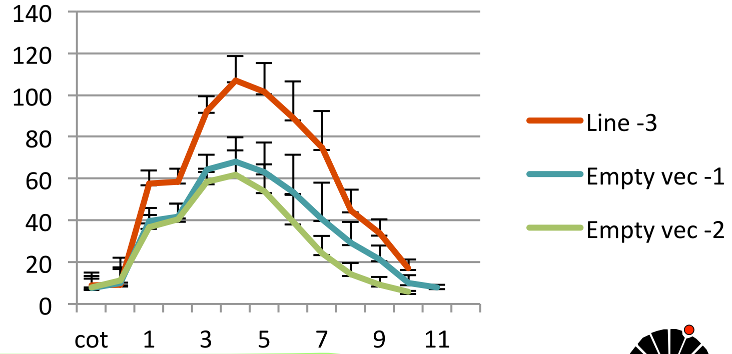
Transcription factor





Transcription factor





Plant survival under severe stress

Protein involved in ion transport

14 days without watering

One day after rewatering

52.3% (11 plants) of the transgenic lines survived
7.1% (1 plant) of control lines (empty vector) survived.

15 distinct genes evaluated so far, and 4 conferred drought stress tolerance in transgenic Arabidopsis

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Technology transfer of sugarcane genes

Genes with proof of concept in model plants do not meet the expectations of private companies.

Proof of concept in the real crop is a must

Transformation and field assays in cooperation with companies to solve this bottleneck

PangeiaBiotech
Plant Transformation Facility

Embrapa

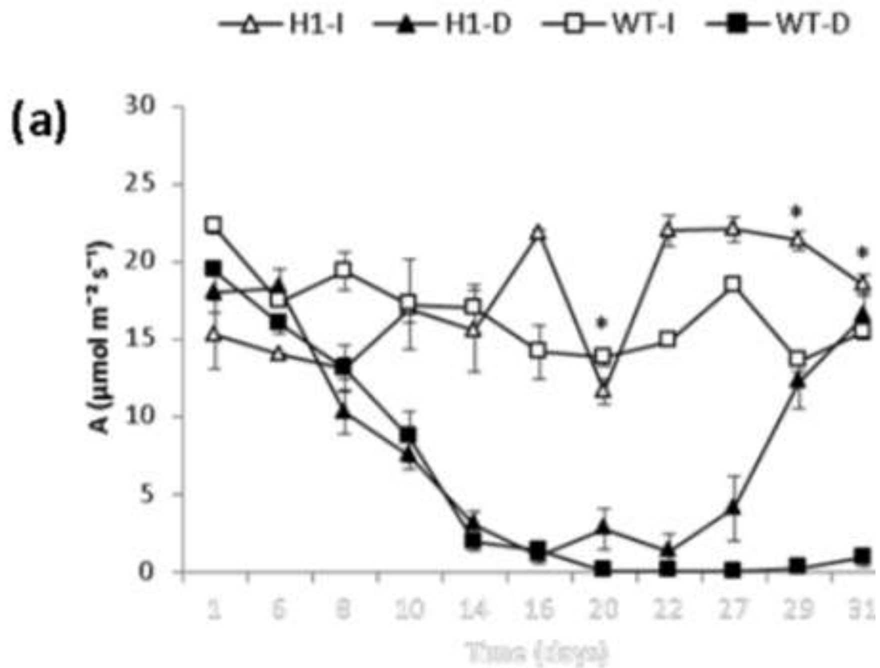
Brazilian Agricultural Research Corporation

Superexpressão de genes da própria cana em cana GM

Plantas transgênicas e do tipo selvagem (WT) de cana foram submetidas a suspensão de rega por 21 dias e então irrigadas por 5 dias.

As plantas transgênicas superexpressando um gene da própria cana que codifica uma proteína associada com tolerância a metais pesados apresentaram menos dano que as plantas WT

Fotossíntese



Transgênicas WT

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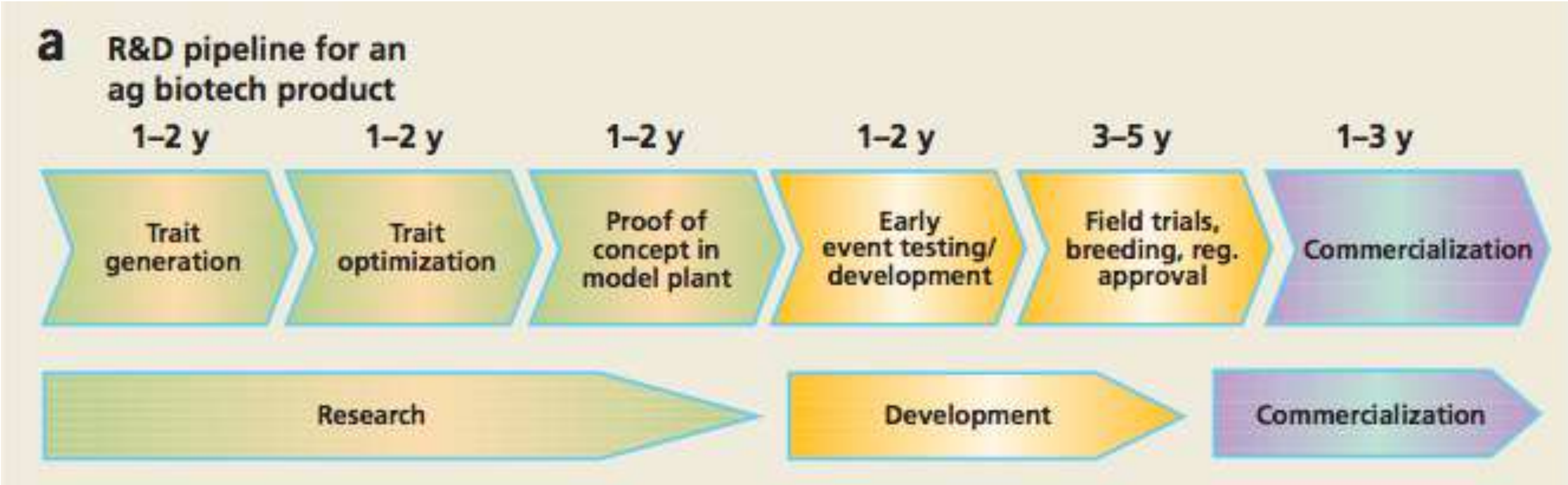
Field trials in the near future

Hugo Molinari
EMBRAPA

Pipeline for GM sugarcane



McElroy (2004) Nature Biotechnology 22:817- 822.

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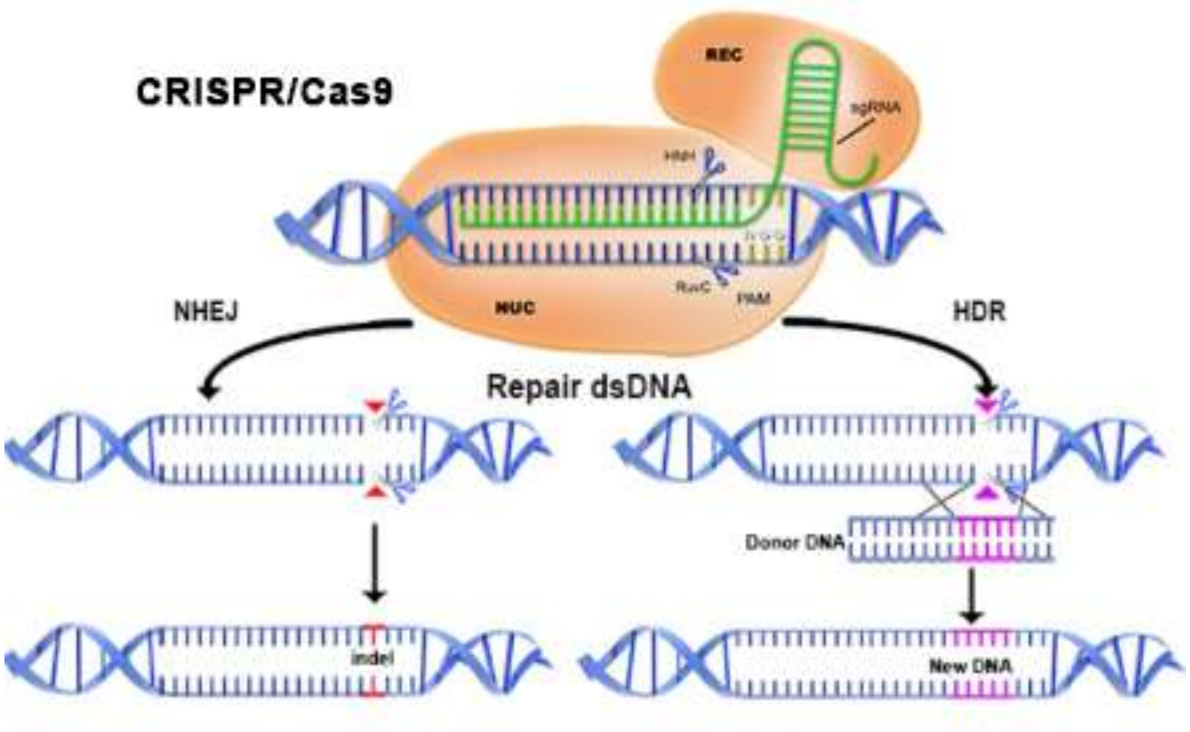
CRISPR will change everything



From Advanced Analytical.

Conclusions

• The gene encoding the DELLA protein controls internode development in sugarcane

• Sugarcane cultivars have different responses to drought at the molecular level

• Gene expression profiling have allowed the discovery of proteins that can enhance drought tolerance in transgenic plants

Thank you!