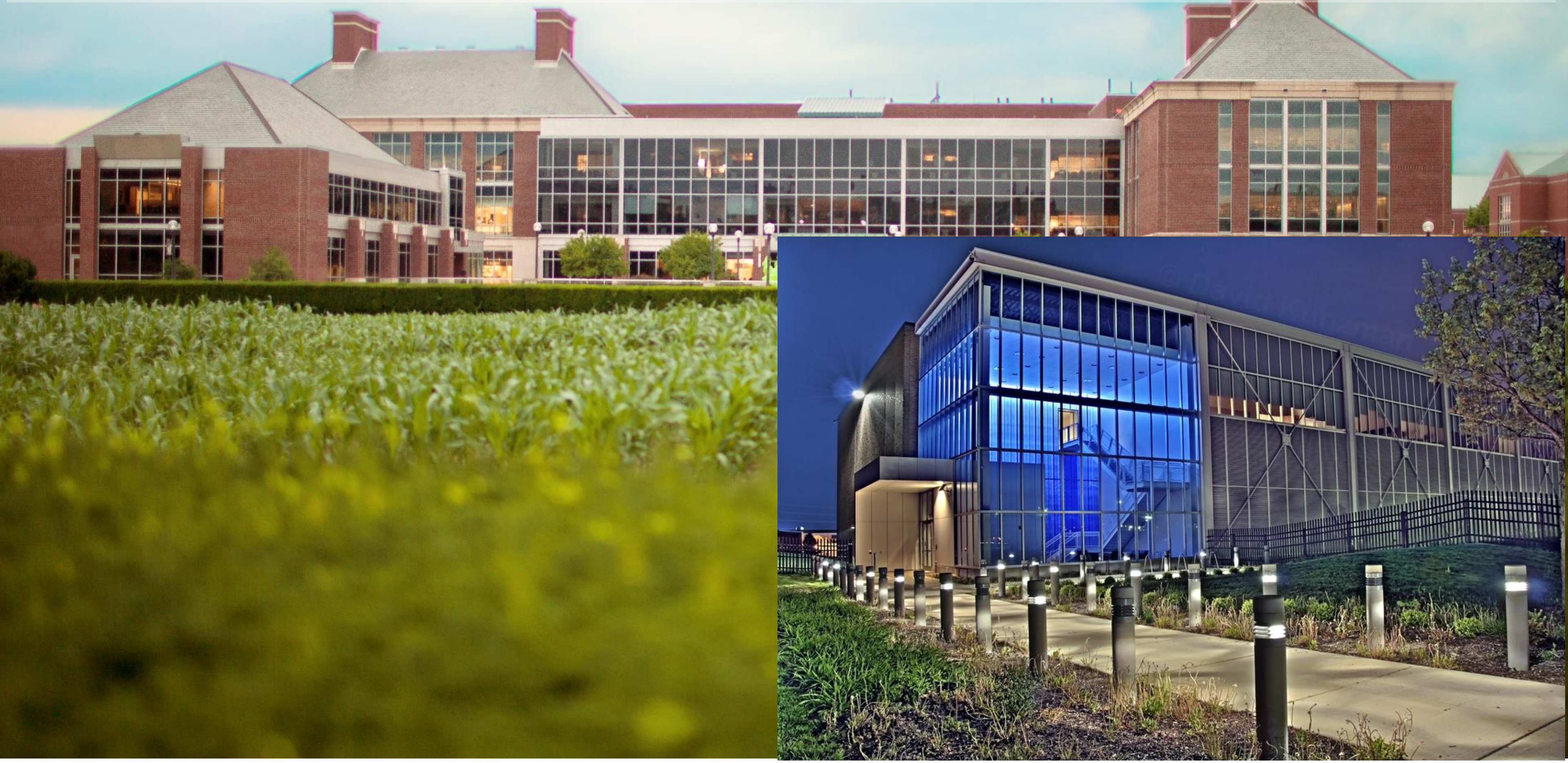
# Progress in Increasing Carbon Uptake into Managed Systems via Engineering of Photosynthesis.

Steve Long, Carl R. Woese Institute of Genomic Biology, University of Illinois, USA and Lancaster Environment Centre, Lancaster University UK.



INSTITUTE FOR GENOMIC BIOLOGY
WHERE SCIENCE MEETS SOCIETY

BILL & MELINDA GATES foundation

1867 ILLINOIS
UNIVERSITY OF ILLINOIS AT URBANA-CHAMPAIGN

## ROADMAP

- Why photosynthesis?
- The Gates Foundation RIPE Project.
- Examples of reduction to practice.
- The plant as the factory.

# ROADMAP

- Why photosynthesis?
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# WHY PHOTOSYNTHESIS – M X GIGANTEUS ILLINOIS.

Harvested yield t $ha^{-1}$

PAR GJ $ha^{-1}$

Interception efficiency

Conversion efficiency

Partitioning efficiency

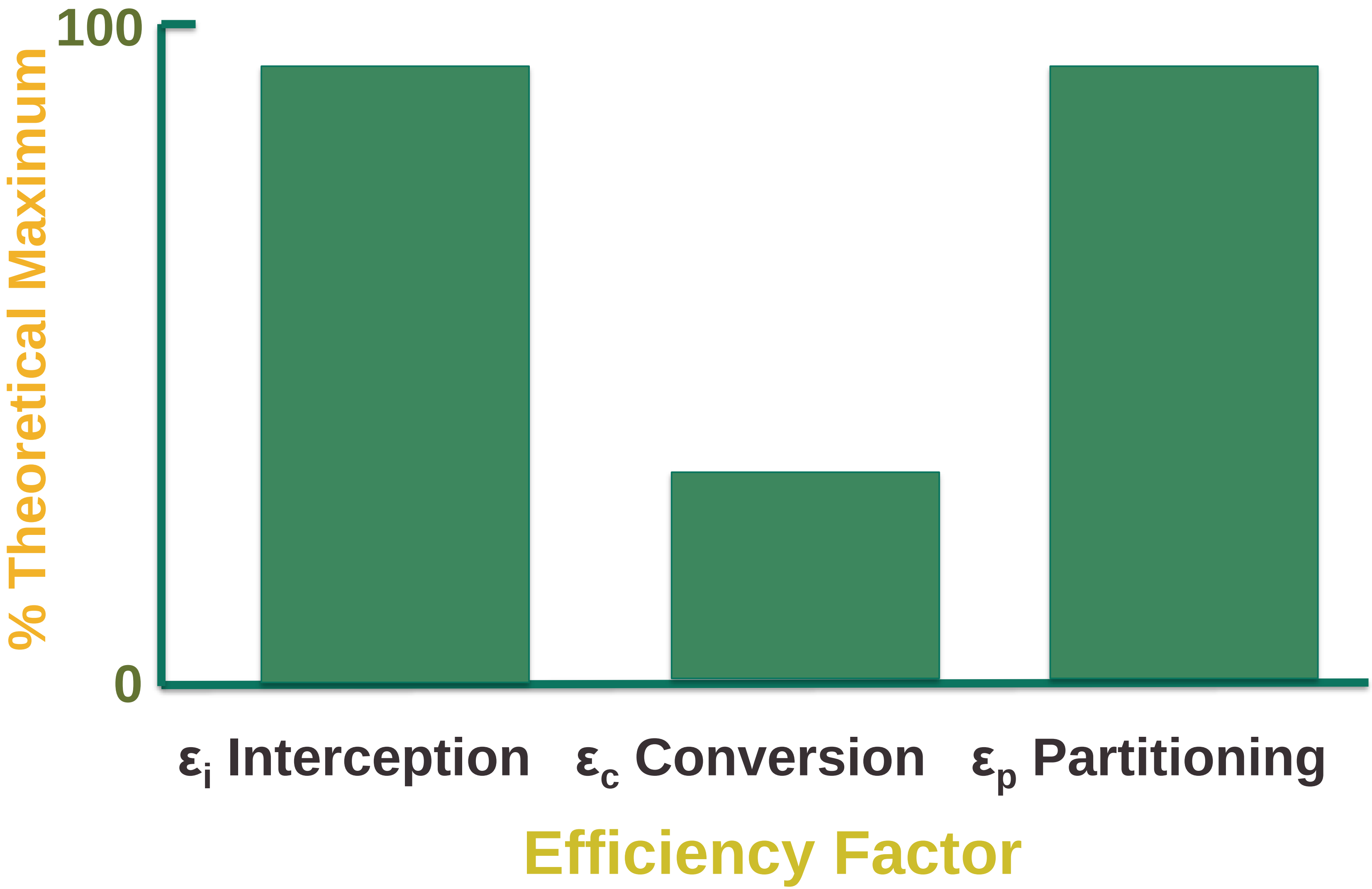
Energy content of biomass GJ $t^{-1}$

$$W_h = S \times \varepsilon_i \times \varepsilon_c \times \varepsilon_p / c$$

$$30.3 = 19530 \times 0.81 \times 0.042 \times 0.75/16.5$$

- Dohleman & Long (2009) Plant Physiology **150**, 2104–2115
- Dohleman, Heaton, Arundale & Long (2012) GCB Bioenergy **4**, 534–544

# WHY PHOTOSYNTHESIS?




Zhu, Long & Ort (2010) Ann. Rev. Pl. **Biology** 2010, 61: 235-261

# OPEN-AIR ENRICHMENT OF $CO_2$ – FACE

Leaf photosynthesis increased 25% across growing season.


- Rogers, Allen, Bernacchi, Ainsworth & Long (2004) Plant, Cell and Environment 27, 449–458
- Long, Ainsworth, Leakey, Noesberger & Ort (2006) Science 312 : 1918-1921

# INCREASE IN YIELD WITH ELEVATED $CO_2$ IN FIELD

Long et al. (2006) Science 312, 1918-1921; (2007) Science 315, 460.

Soybeans

16%

Rice

12%

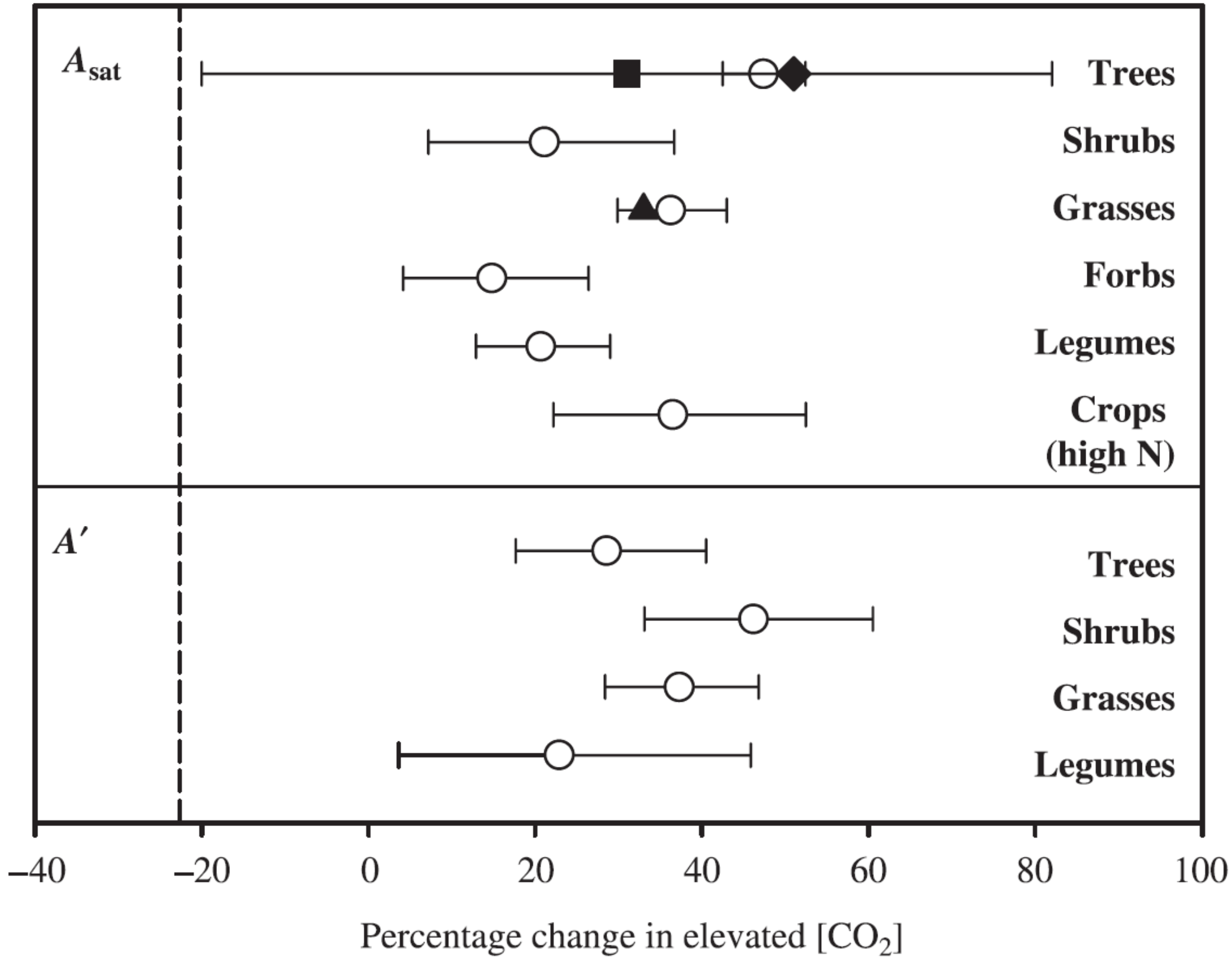
Wheat

15%

Munching_bugs_2.jpg

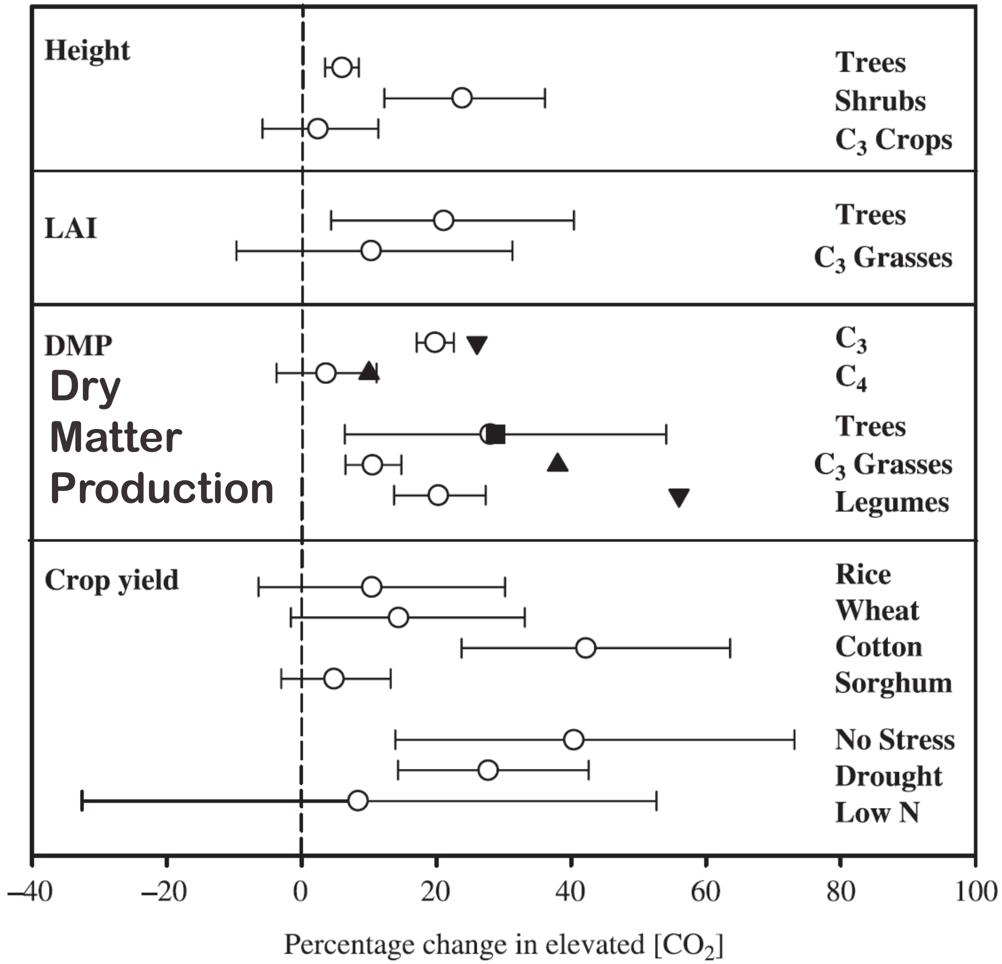
# WHY PHOTOSYNTHESIS?

# WHY PHOTOSYNTHESIS?



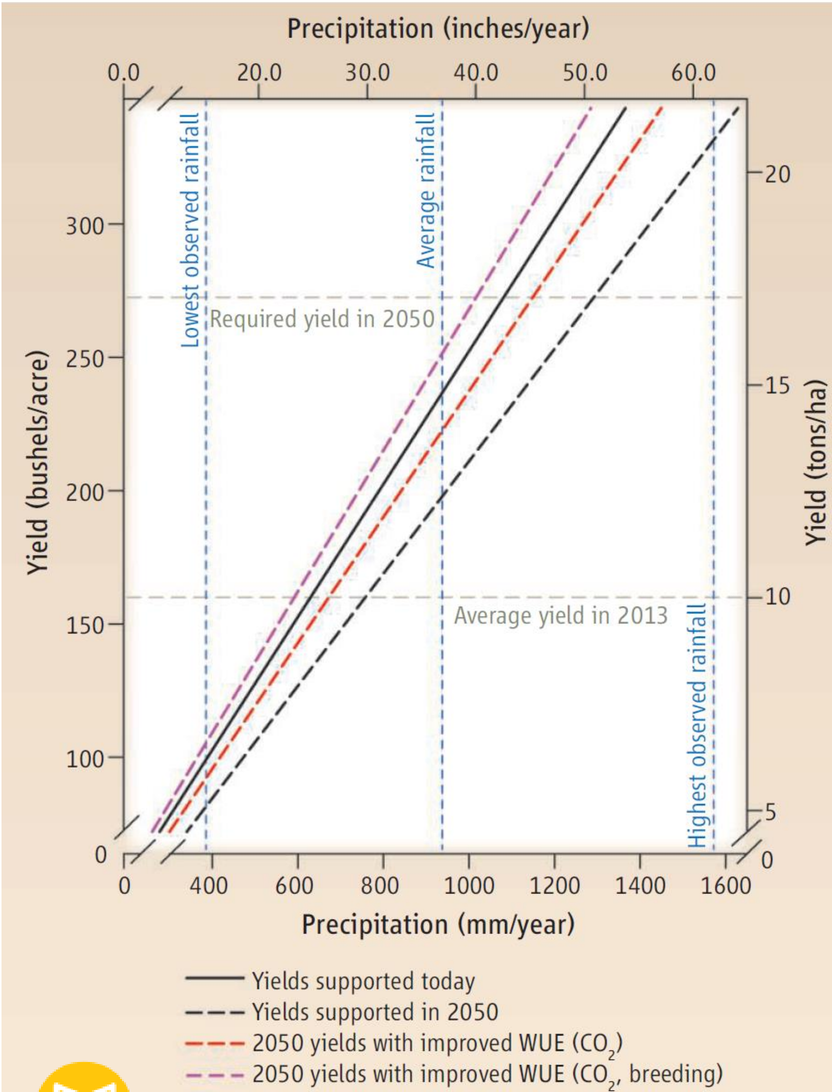

Ainsworth & Long (2005) New Phytologist 165: 351–372

# WHY PHOTOSYNTHESIS?




Ainsworth & Long (2005) New Phytologist 165: 351–372

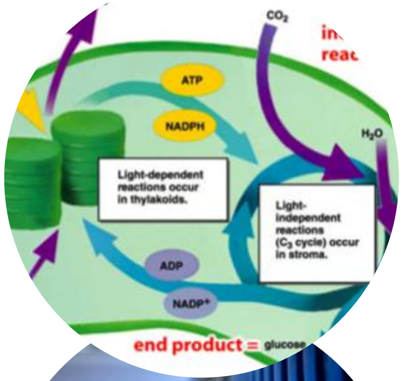
# RAIN-FED MIDWEST MAIZE AND CLIMATE CHANGE





Ort & Long (2016) **Science 344,** 484-5.

# WHY IS ENGINEERING OF CROP PHOTOSYNTHESIS SCIENTIFICALLY AND TECHNOLOGICALLY TIMELY?




Photosynthesis known in more detail than any other plant process, and is highly conserved.

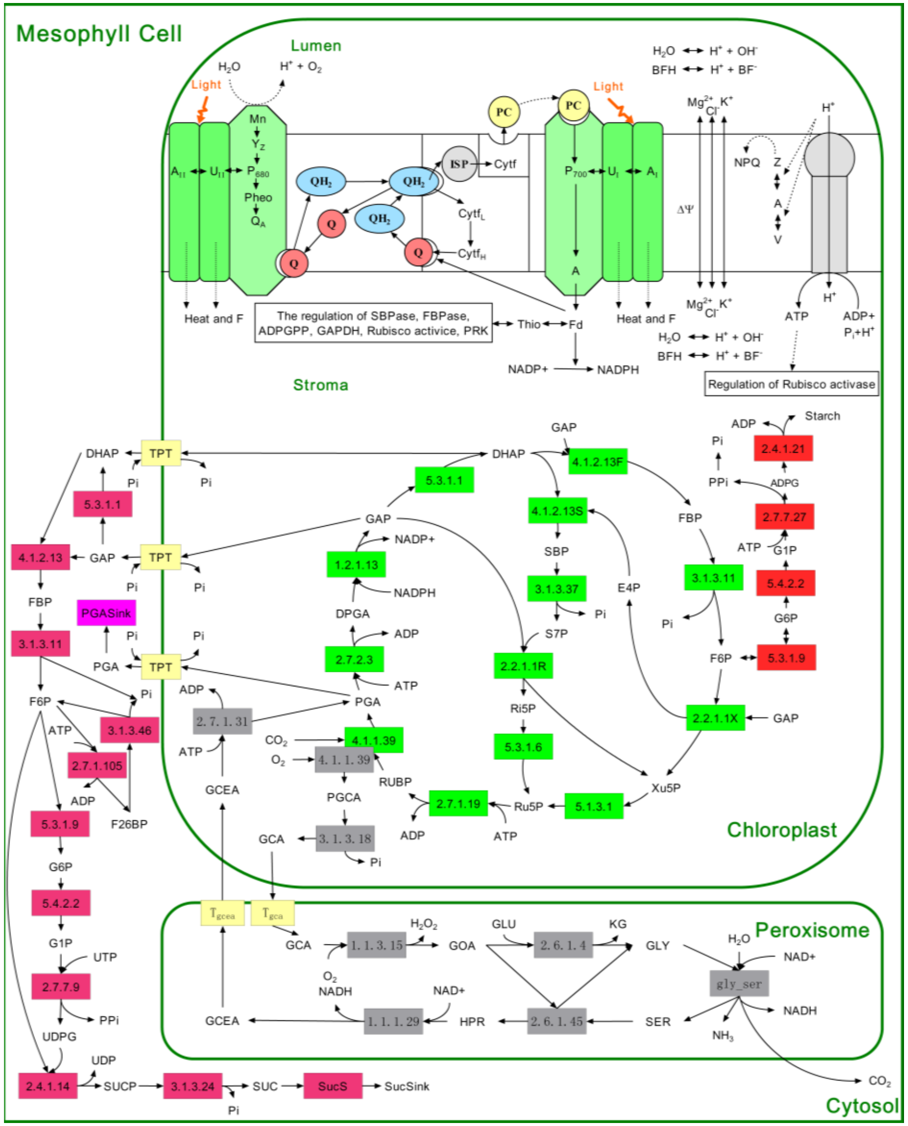
High performance computing

Crop transformation becoming increasingly routine.

# 160 QUANTITATIVELY VARIABLE STEPS – A FEW TRILLION PERMUTATIONS – WHERE DO WE START!?!

# DYNAMIC MODEL OF CELLULAR PHOTOSYNTHESIS

Represented as a system of differential equations to guide improvement of existing system




Zhu, DeSturler & Long (2007) **Plant Physiology** 145: 513-526
Zhu, Ort & Long (2013) **Plant, Cell & Env.** 36: 1711-1727

Cell

Long S.P., Marshall-Colon A. & Zhu X.G. (2015) Meeting the Global Food Demand of the Future by Engineering Crop Photosynthesis for Yield Potential. *Cell*, 161, 56-66.

**Table 1. This Table Lists the Manipulations That Could Be Undertaken to Improve Photosynthetic Efficiency in C3 Crops, the Type of Manipulation, and the Model Estimated Improvement in Efficiency of Conversion of Received Light Energy into Crop Biomass Relative to Today's Best Cultivars**

| | Manipulation | Type | Efficiency Gain | Timescale | Additional Benefits |
|---|---|---|---|---|---|
| 1 | extend usable spectrum of crop photosynthesis into NIR | CSyn | 10%–30%[a] | L | could be used to power 3 or improve value of 10. C4 |
| 2 | more rapid relaxation of heat dissipation at PSII | Syn | 30%[i] | S | synergistic with all other changes. C4 |
| 3 | convert C3 crops to C4 | Syn | 30%[c,e] | L | improved WUE and NUE |
| 4 | add cyanobacterial or microalgal CO2/HCO3 pumps | Syn | 5%–10%[d] | M | improved WUE and NUE |
| 5 | add cyanobacterial carboxysome system | CSyn | 60%[d] | L | improved WUE and NUE |
| 6 | add algal pyrenoid CO2 concentrating system | CSyn | 60%[d] | L | improved WUE and NUE |
| 7 | substitute forms of Rubisco better adapted to today's $CO_2$ | CSyn, B | 15%–30%[h,j] | L | improved WUE and NUE |
| 8 | synthetic photorespiratory bypasses | Syn | 15%[c,f] | S | improved WUE and NUE |
| 9 | optimize regeneration of RubP | Sys, B | 60%[g] | S | synergistic with all; improved NUE. C4 |
| 10 | transmit more light to lower canopy leaves | B, Syn | 15%–60%[b,c] | S | synergistic with 1, and 3 thru 9. improved WUE and albedo. C4 |

CSyn indicates synthetic addition of foreign genes to the chloroplast or plastid genome; Syn indicates synthetic addition to the nuclear genome; Sys indicated up- or down-regulation of existing genes; and B indicates that the improvement may be tractable by breeding given adequate molecular markers for the specific genes. The efficiency gains are from modeled estimates and are largely untested; these vary greatly depending on different assumptions and can vary with environmental conditions. For example, the benefits of items three through eight will increase with temperature and so

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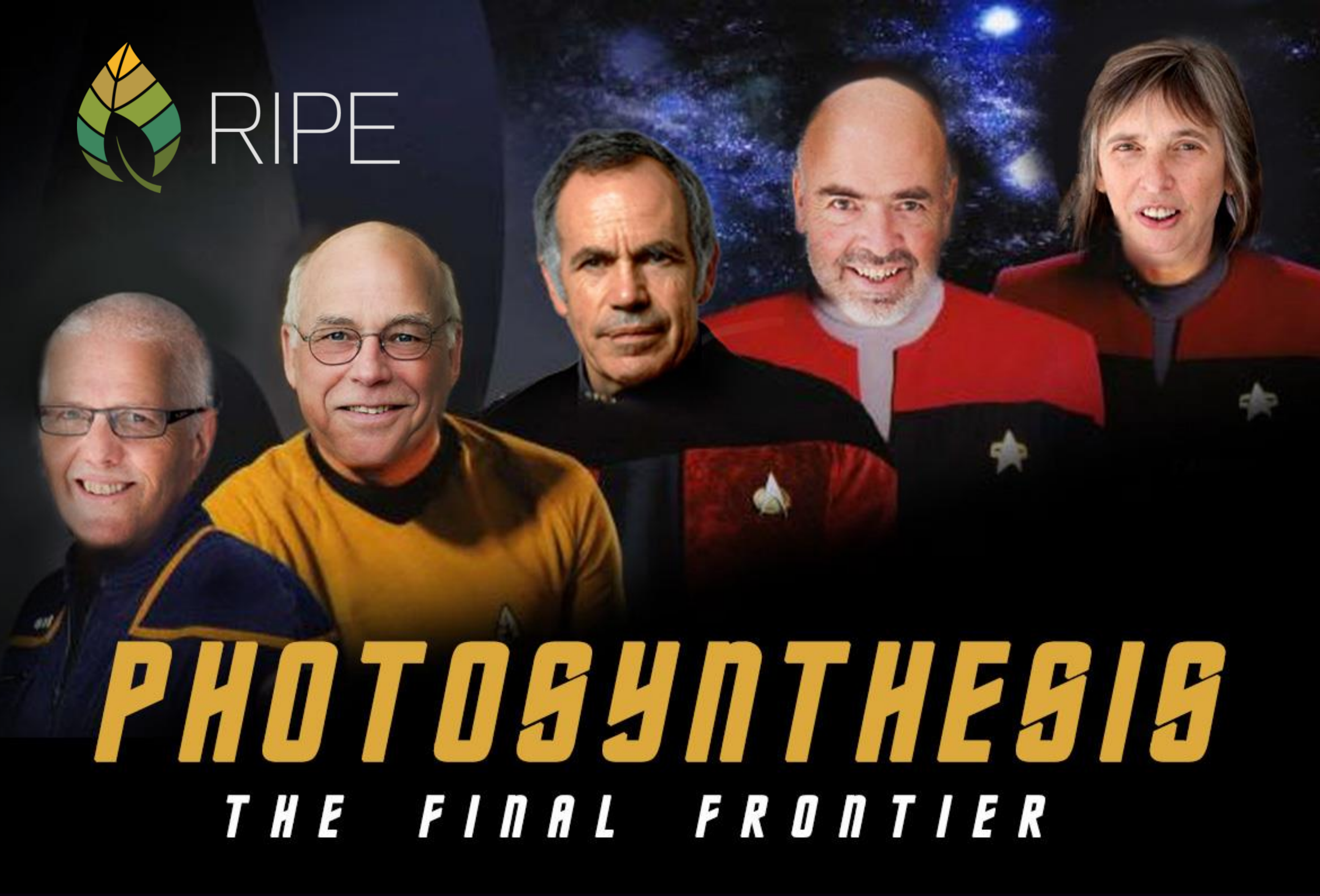
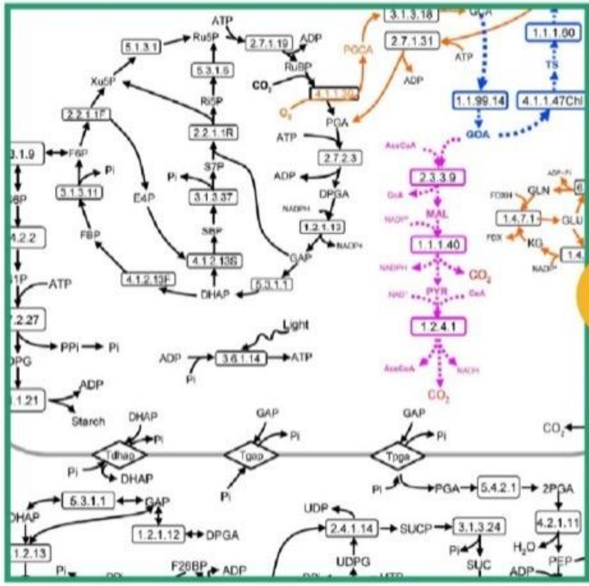
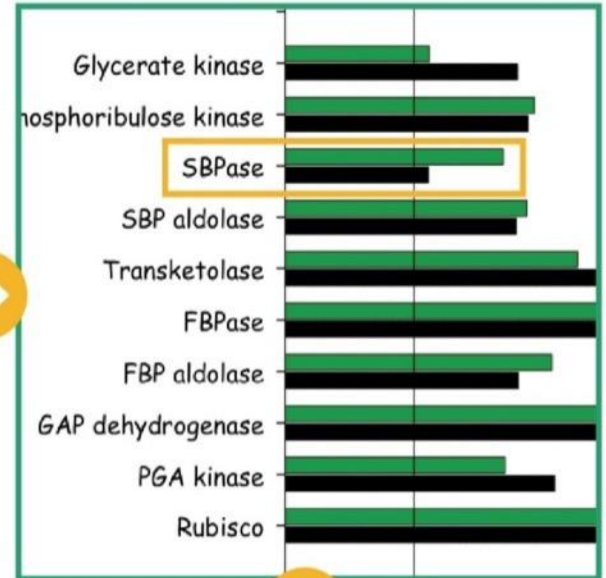
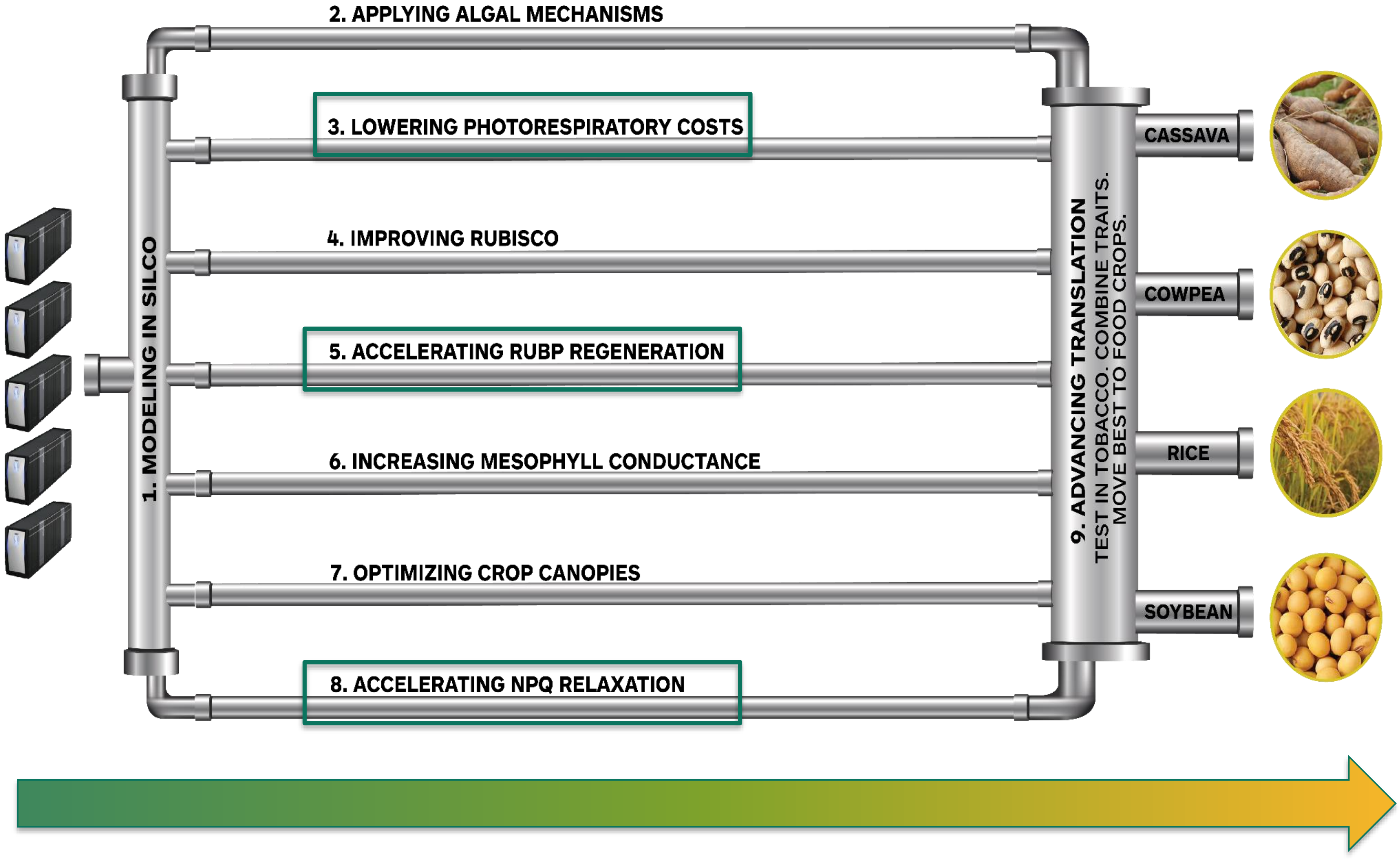

RIPE
PHOTOSYNTHESIS
THE FINAL FRONTIER






THE AUSTRALIAN NATIONAL UNIVERSITY
LSU
Lancaster University
Berkeley UNIVERSITY OF CALIFORNIA
University of Essex
USDA ARS Agricultural Research Service
ILLINOIS UNIVERSITY OF ILLINOIS AT URBANA-CHAMPAIGN



2. APPLYING ALGAL MECHANISMS
3. LOWERING PHOTORESPIRATORY COSTS
4. IMPROVING RUBISCO
5. ACCELERATING RUBP REGENERATION
6. INCREASING MESOPHYLL CONDUCTANCE
7. OPTIMIZING CROP CANOPIES
8. ACCELERATING NPQ RELAXATION
1. MODELING IN SILCO
9. ADVANCING TRANSLATION
TEST IN TOBACCO. COMBINE TRAITS.
MOVE BEST TO FOOD CROPS.
CASSAVA
COWPEA
RICE
SOYBEAN

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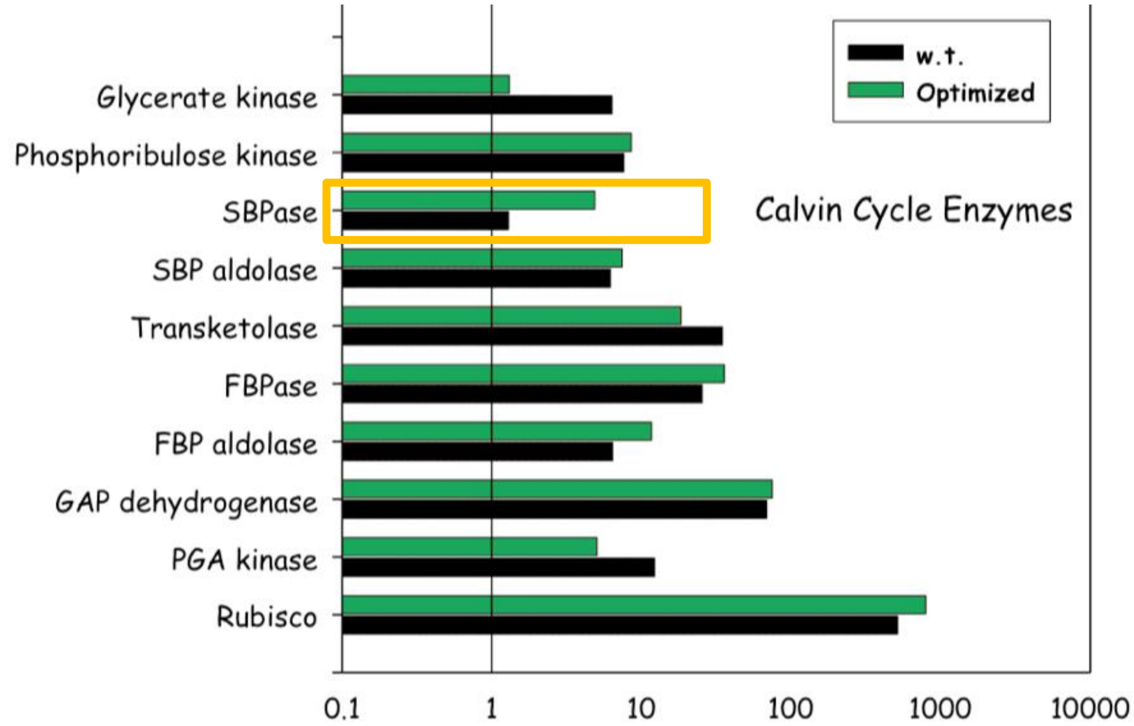
# EVOLUTIONARY ALGORITHM TO PREDICT OPTIMUM INVESTMENT

Increased carbohydrate production




Zhu, DeSturler & Long (2007) **Plant Physiology 145,** 513–526

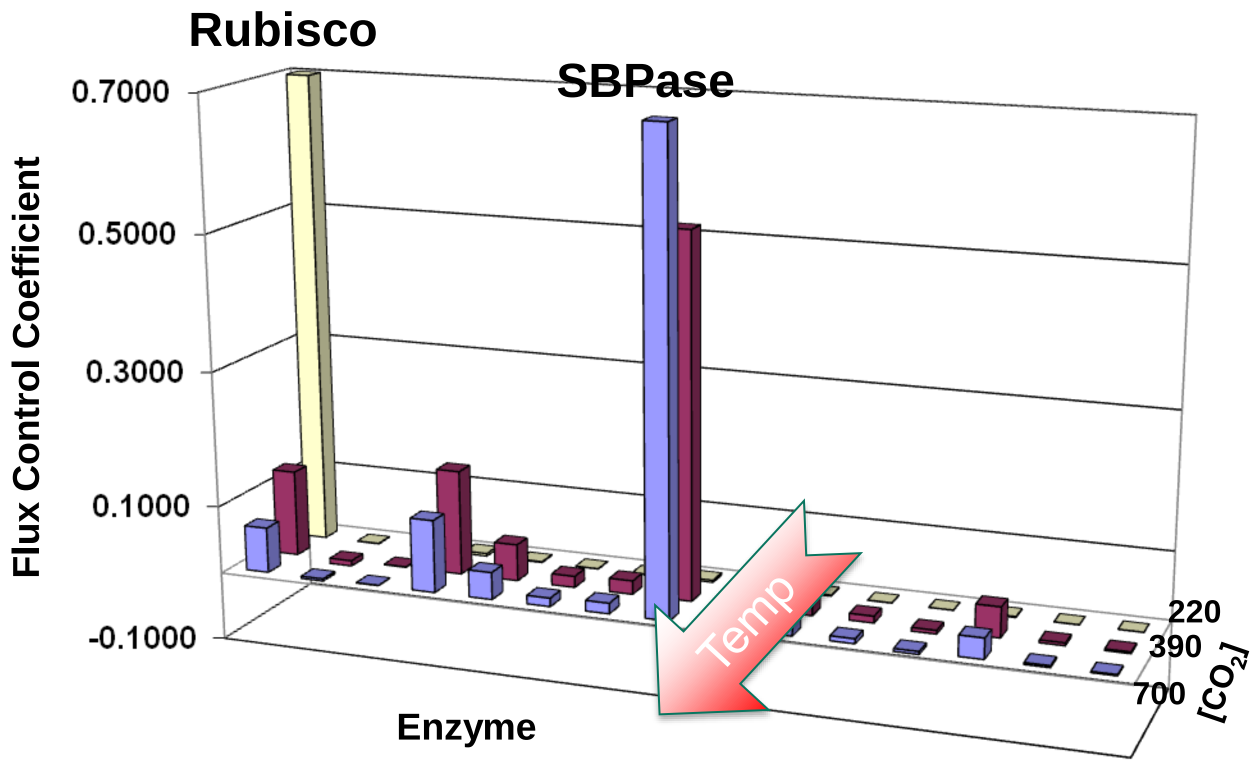
# SBPASE TESTING THE REALITY


WT4 WT12 WT14 11.5 60.12 30.15

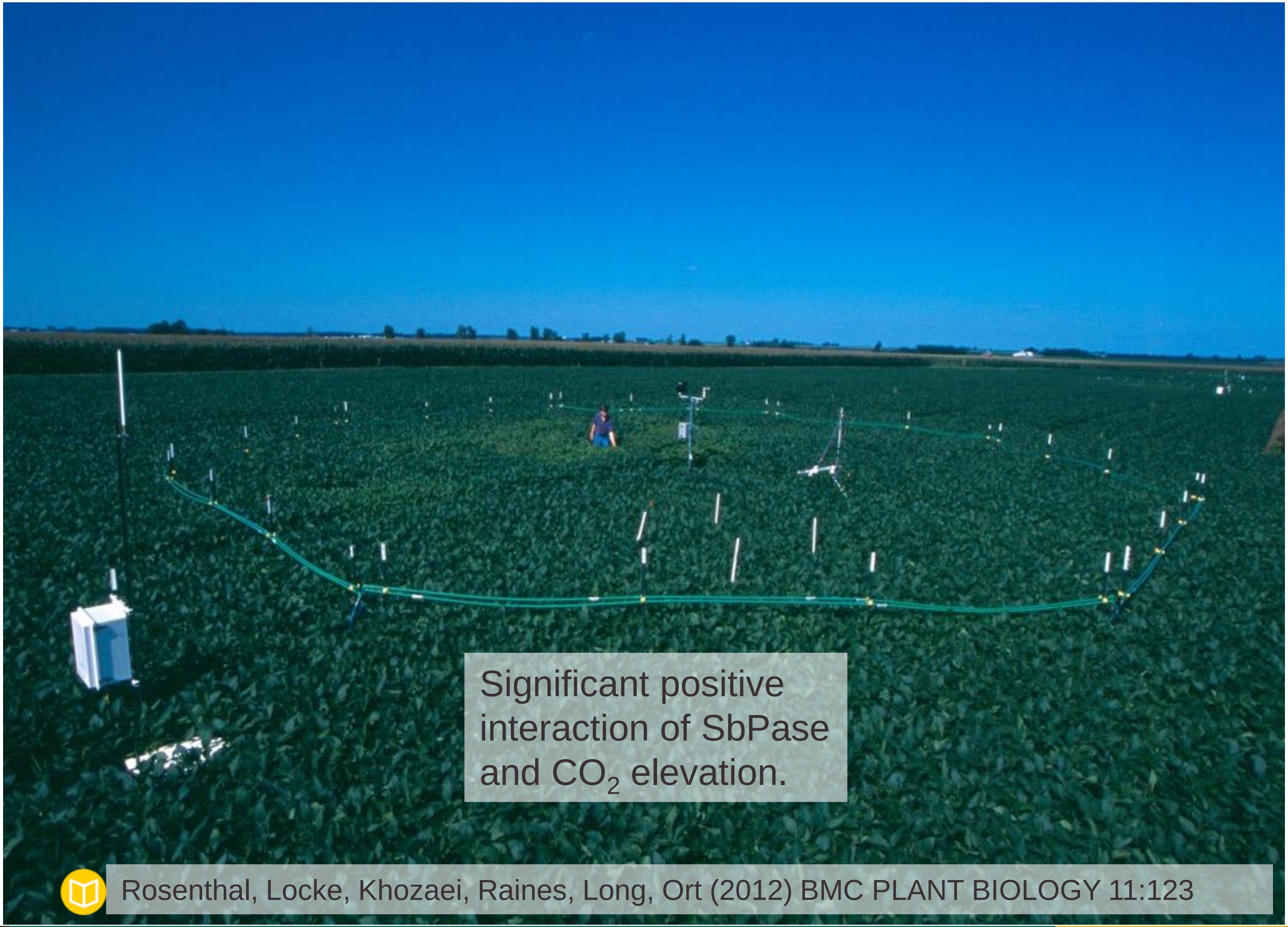
# Why is SbPase limiting the system?

From: Zhu, DeSturler & Long (2007) *Plant Physiology* **145,** 513

$$\varepsilon_{Ei}^{A} \approx \frac{\frac{\delta A}{A}}{\frac{\delta E_i}{E_i}} = \frac{\partial \ln A}{\partial \ln E_i}$$



# SBPASE: TESTING THE HYPOTHESIS



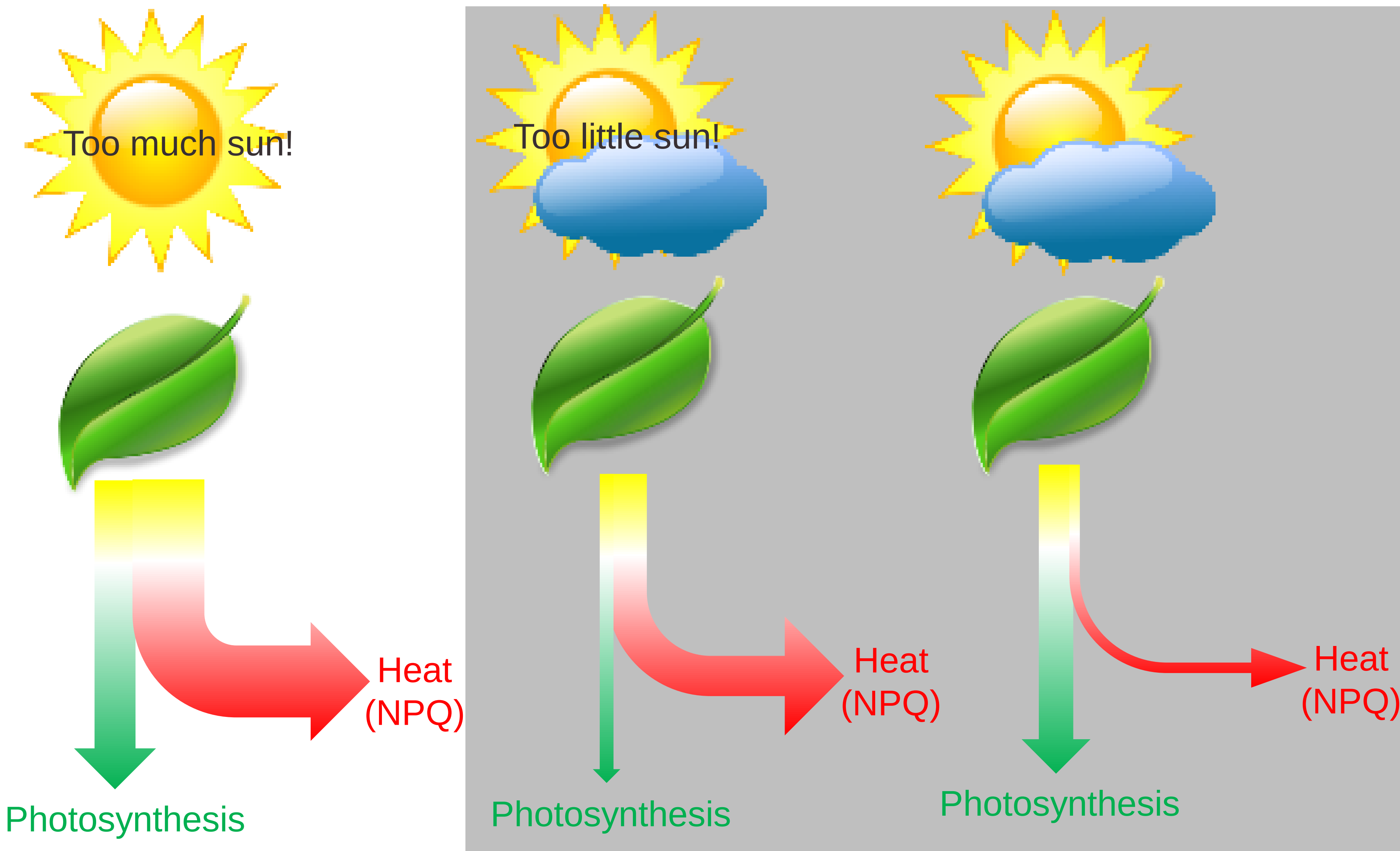
Significant positive interaction of SbPase and $CO_2$ elevation.

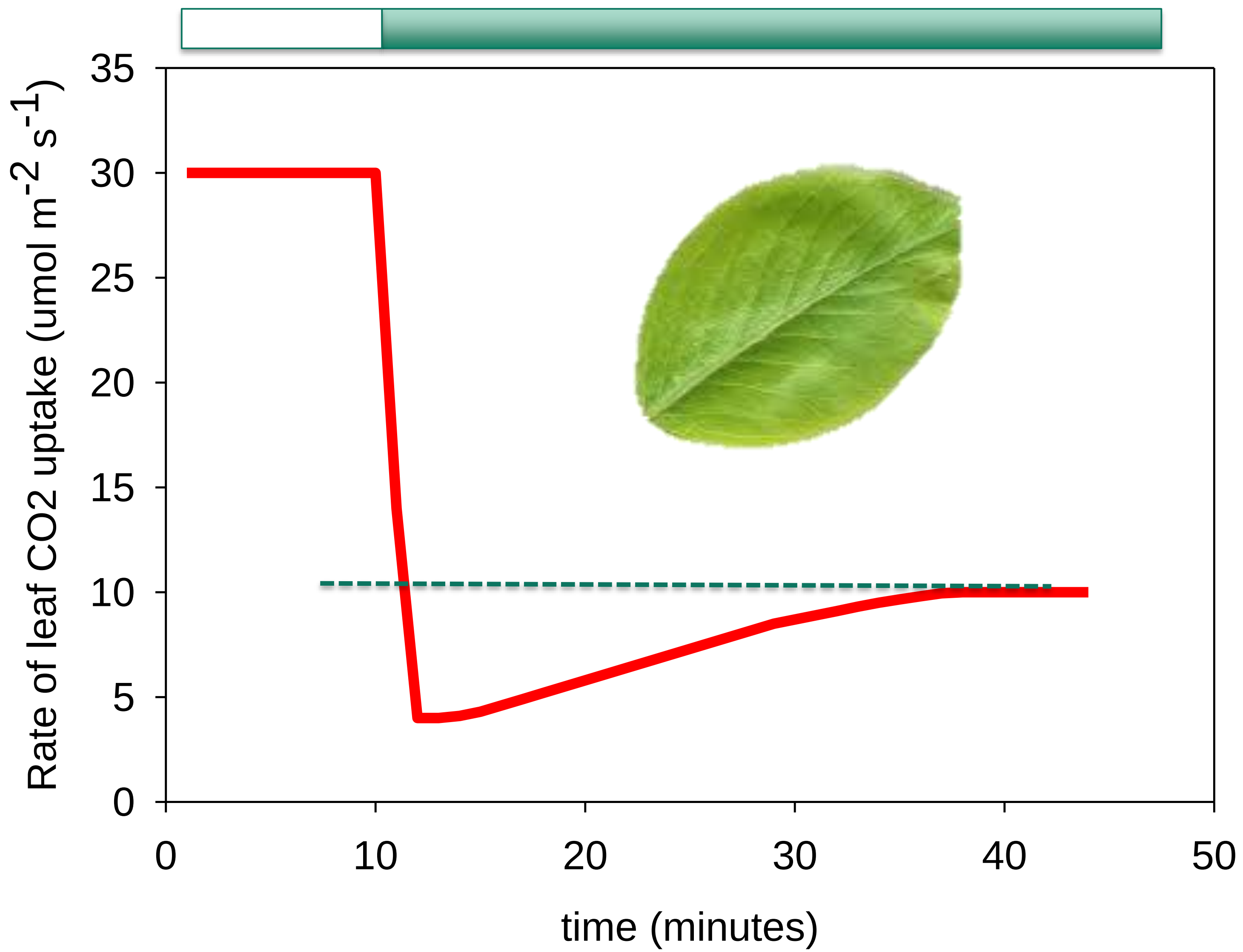
Rosenthal, Locke, Khozaei, Raines, Long, Ort (2012) BMC PLANT BIOLOGY 11:123

# SBPASE: TESTING THE HYPOTHESIS

Significant positive interaction of elevated SbPase with increase in $CO_2$ and in temperature.

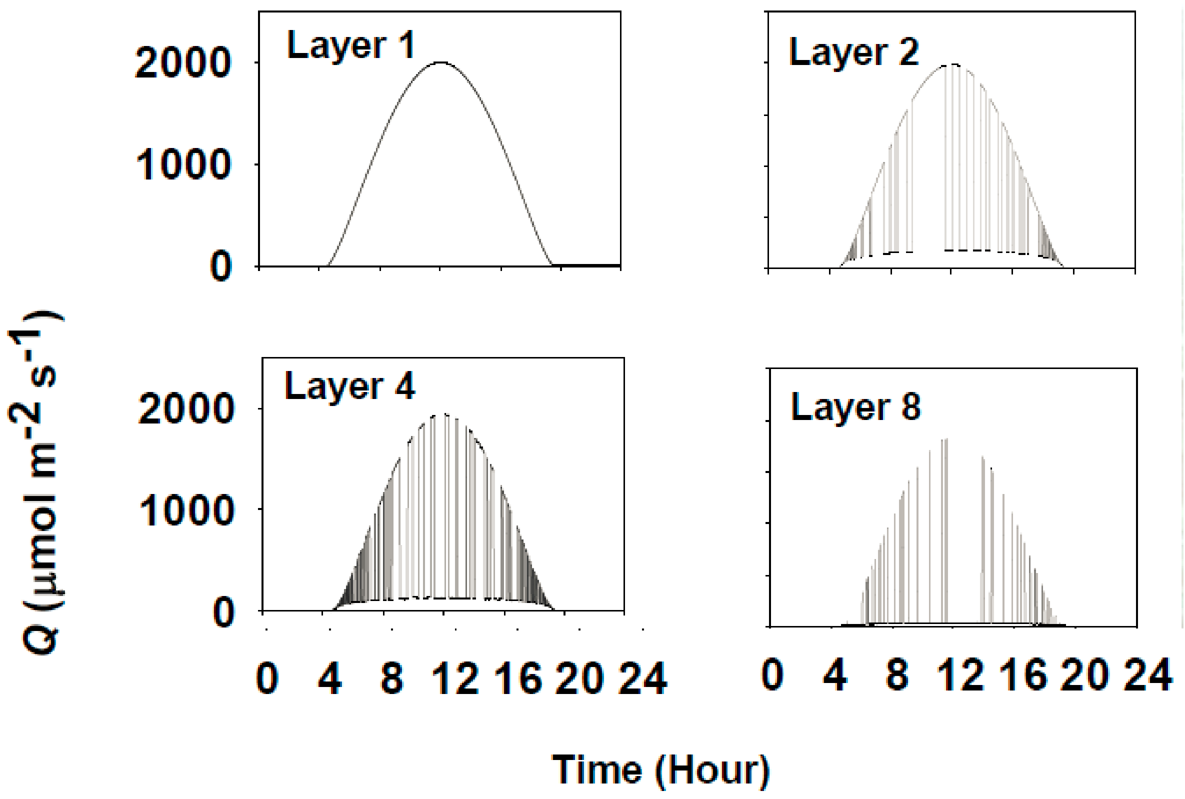
Kohler et al. (2017) Journal of Experimental Botany 68: 715.


Too much sun!
Heat
(NPQ)
Photosynthesis
Too little sun!
Heat
(NPQ)
Photosynthesis
Heat
(NPQ)
Photosynthesis



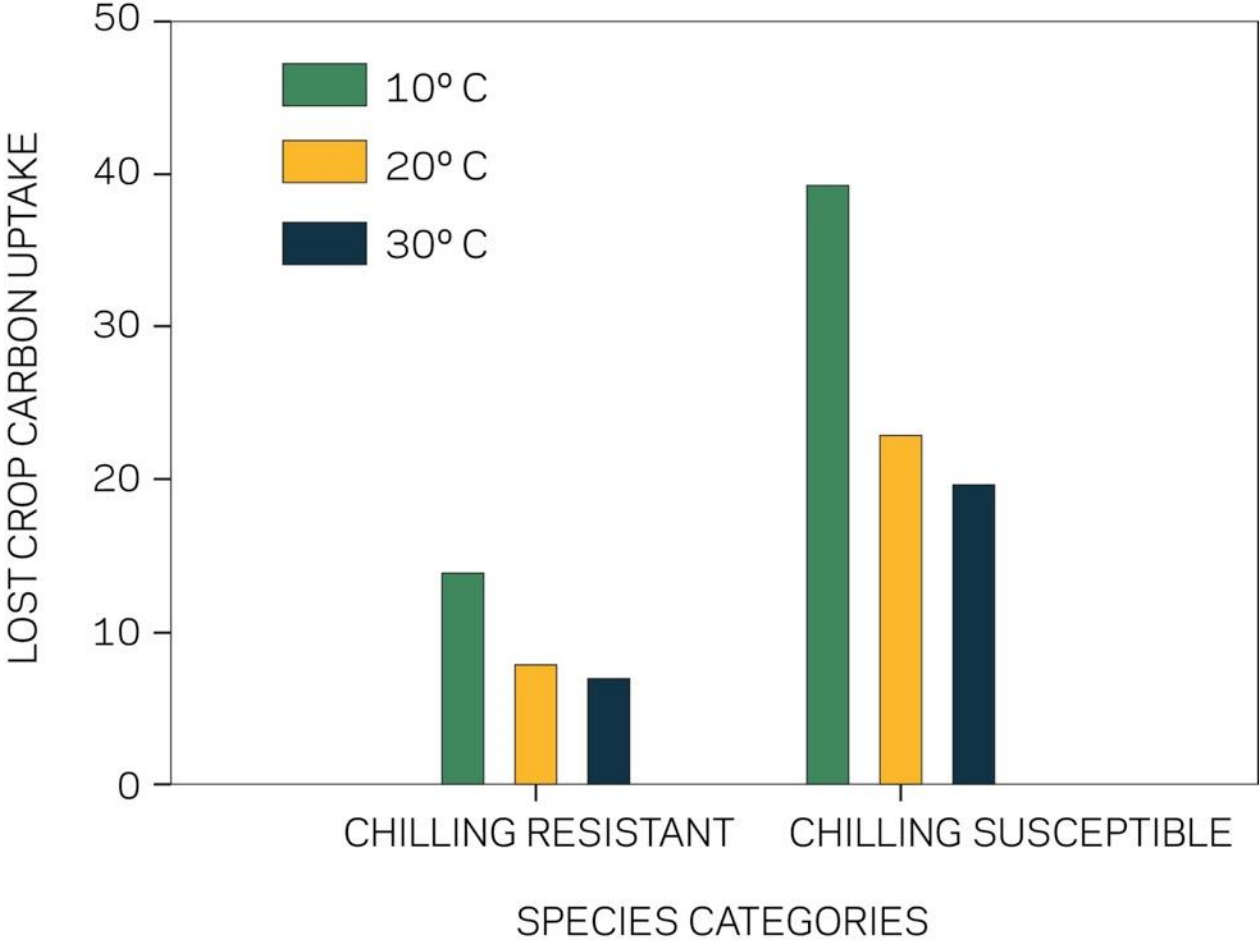
Rate of leaf CO2 uptake (umol m-2 s-1)
35
30
25
20
15
10
5
0
0
10
20
30
40
50
time (minutes)

# SLOW RECOVERY OF EFFICIENCY

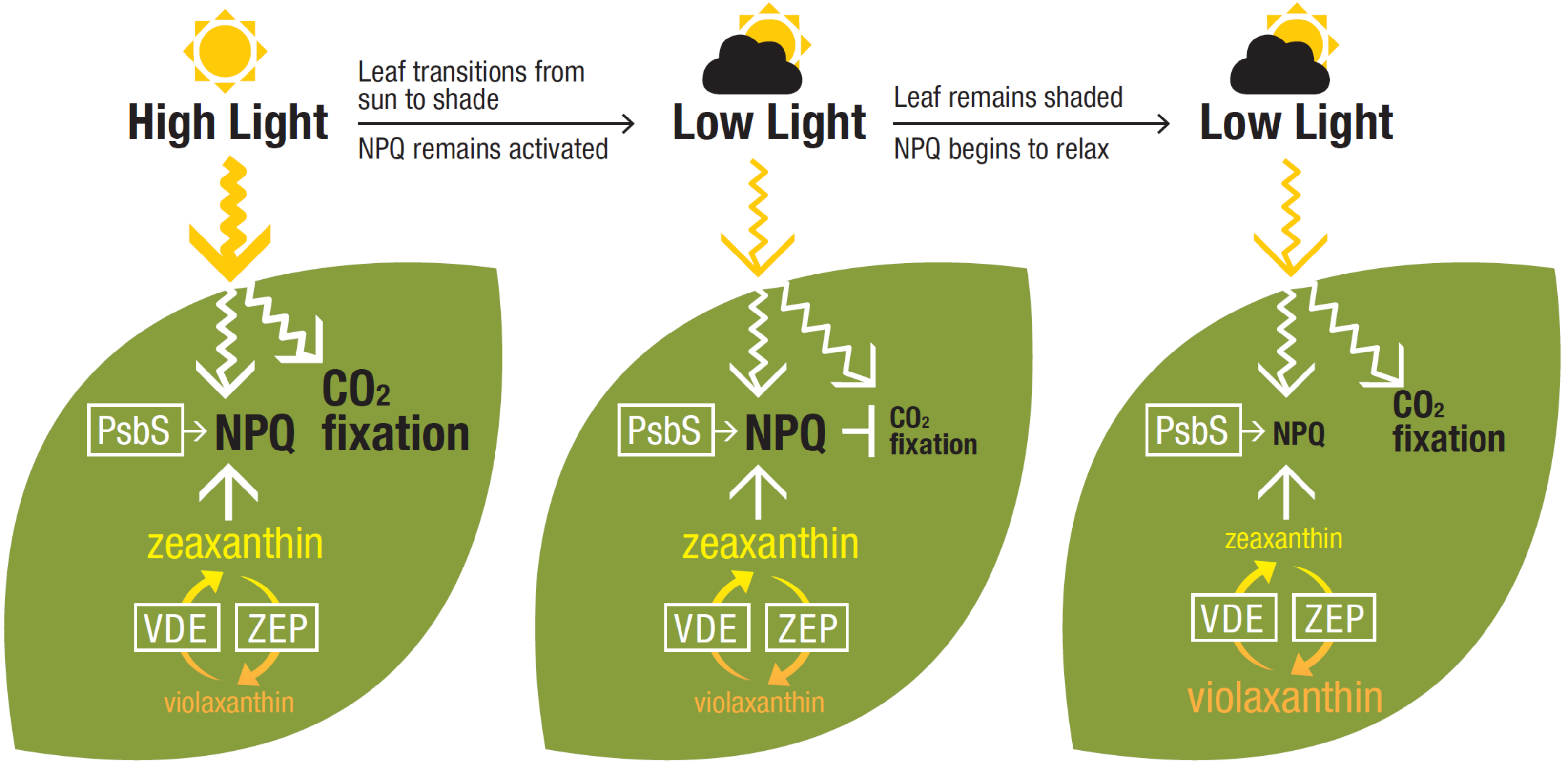



Zhu & Long (2004) J. Exp. Bot. 55, 1167-1175.

# COST OF SLOW RELAXATION IN SHADE




Zhu & Long (2004) J. Exp. Bot. 55, 1167-1175.

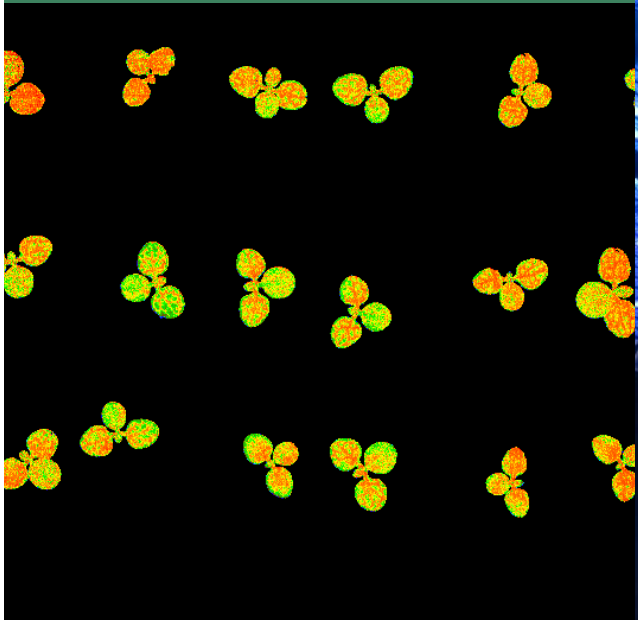


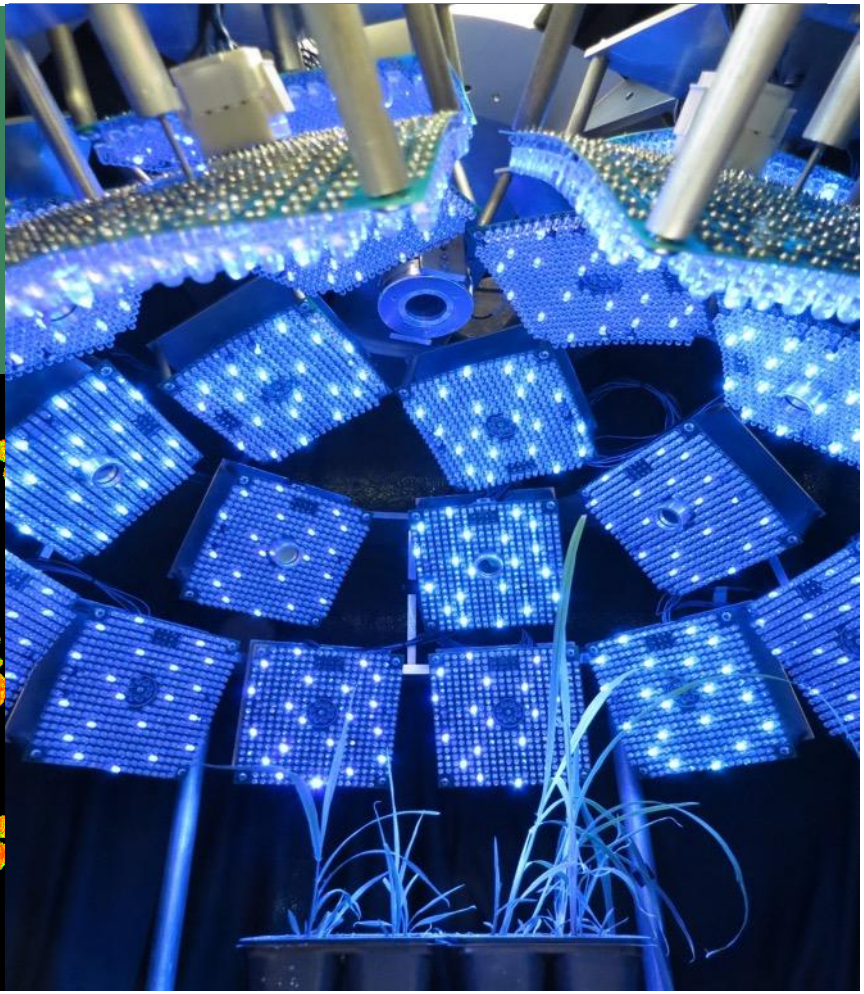

ZEP speeds up NPQ relaxation
VDE balances ZEP activity during NPQ induction
PsbS adjusts NPQ level to maintain WT amplitude

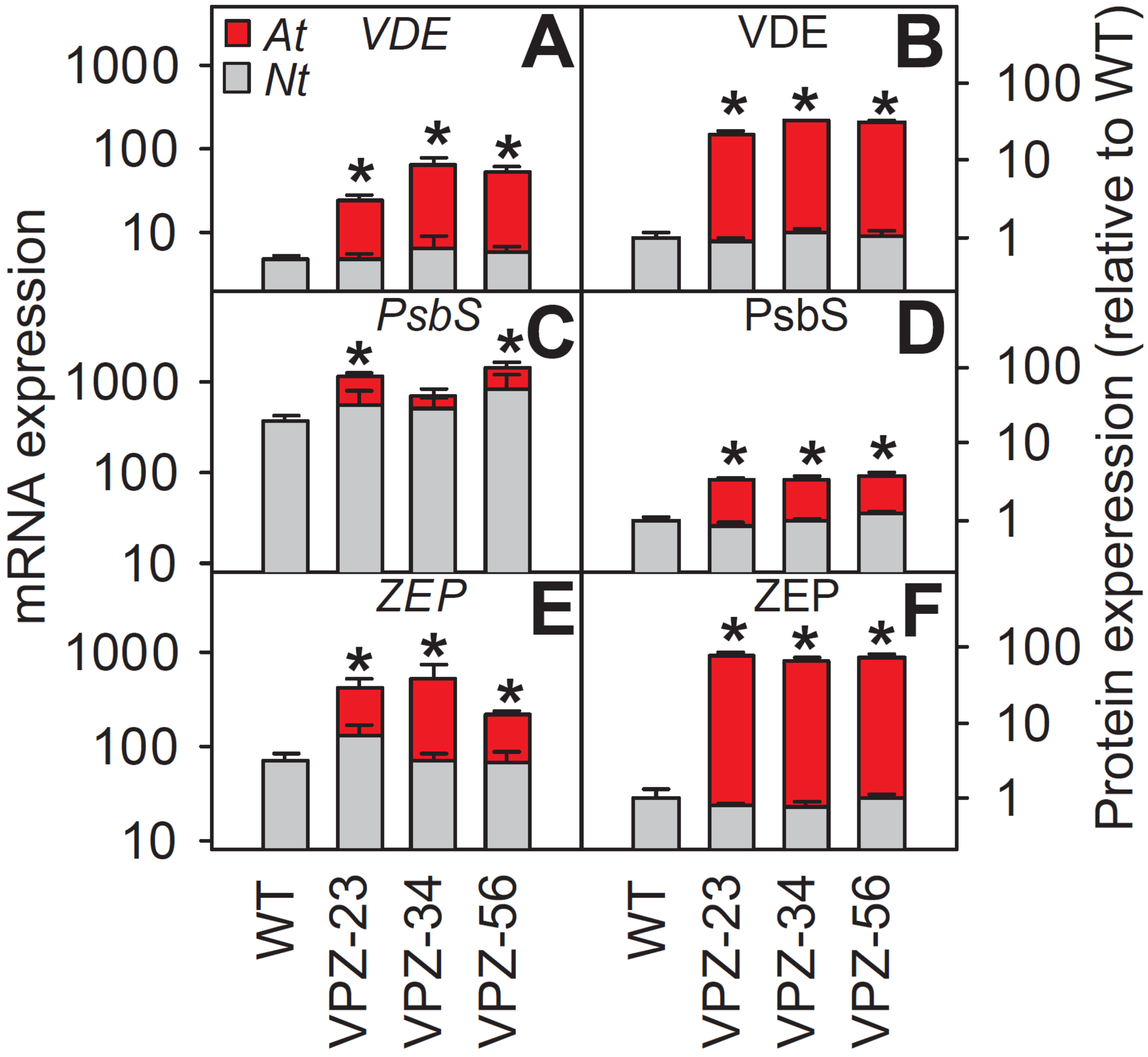
Kromdijk, Głowacka, Leonelli, Iwai, Niyogi & Long (2016) **Science 354,** 857-861

# SELECTING FASTER RELAXATION OF NPQ IN ENGINEERED PLANTS

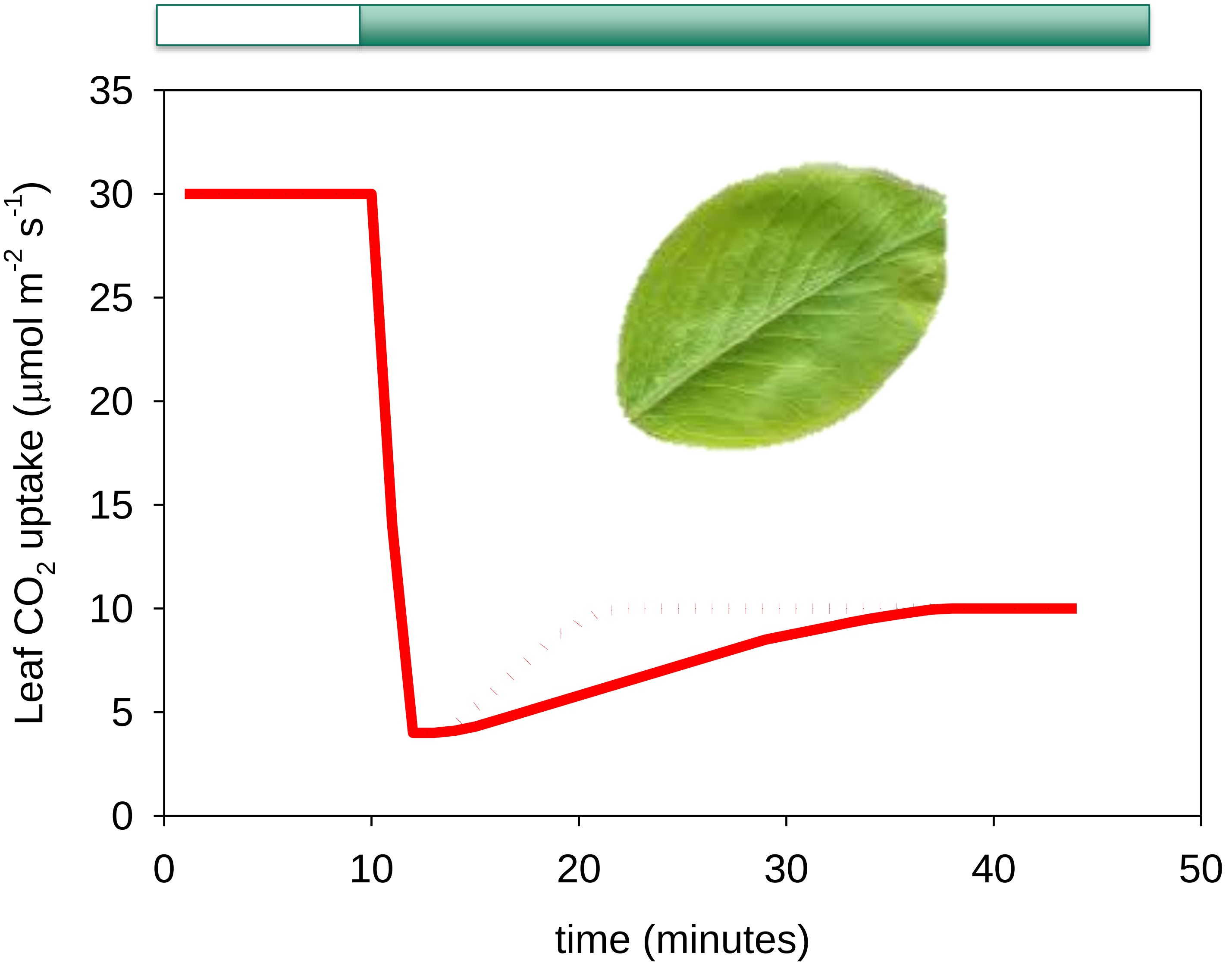
Chlorophyll fluorescence of young seedlings





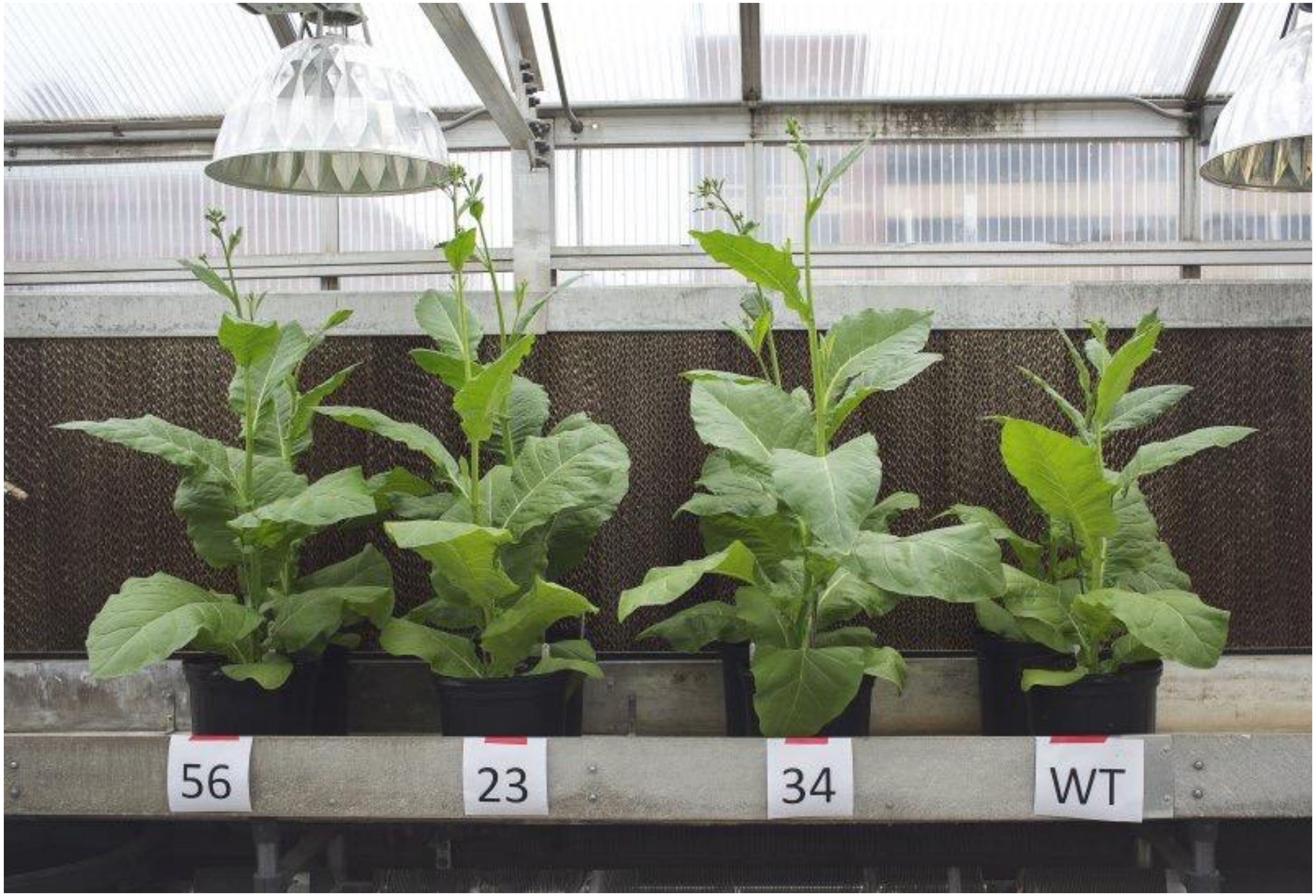


At
Nt
VDE
A
B
PsbS
C
D
ZEP
E
F
mRNA expression
Protein expereession (relative to WT)
1000
100
10
1
WT
VPZ-23
VPZ-34
VPZ-56

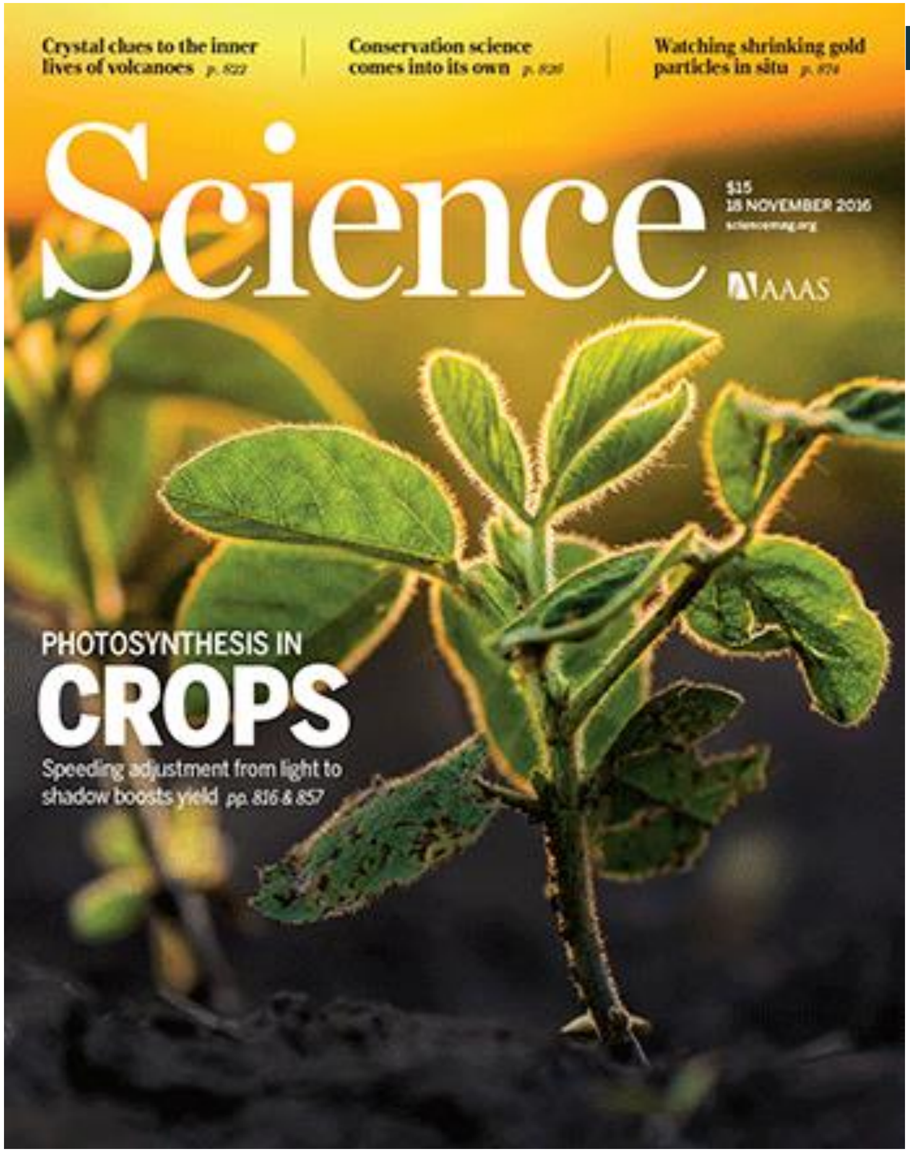


35
30
25
20
15
10
5
0
Leaf CO2 uptake (μmol m-2 s-1)
0
10
20
30
40
50
time (minutes)

# RELAXING PHOTOPROTECTION



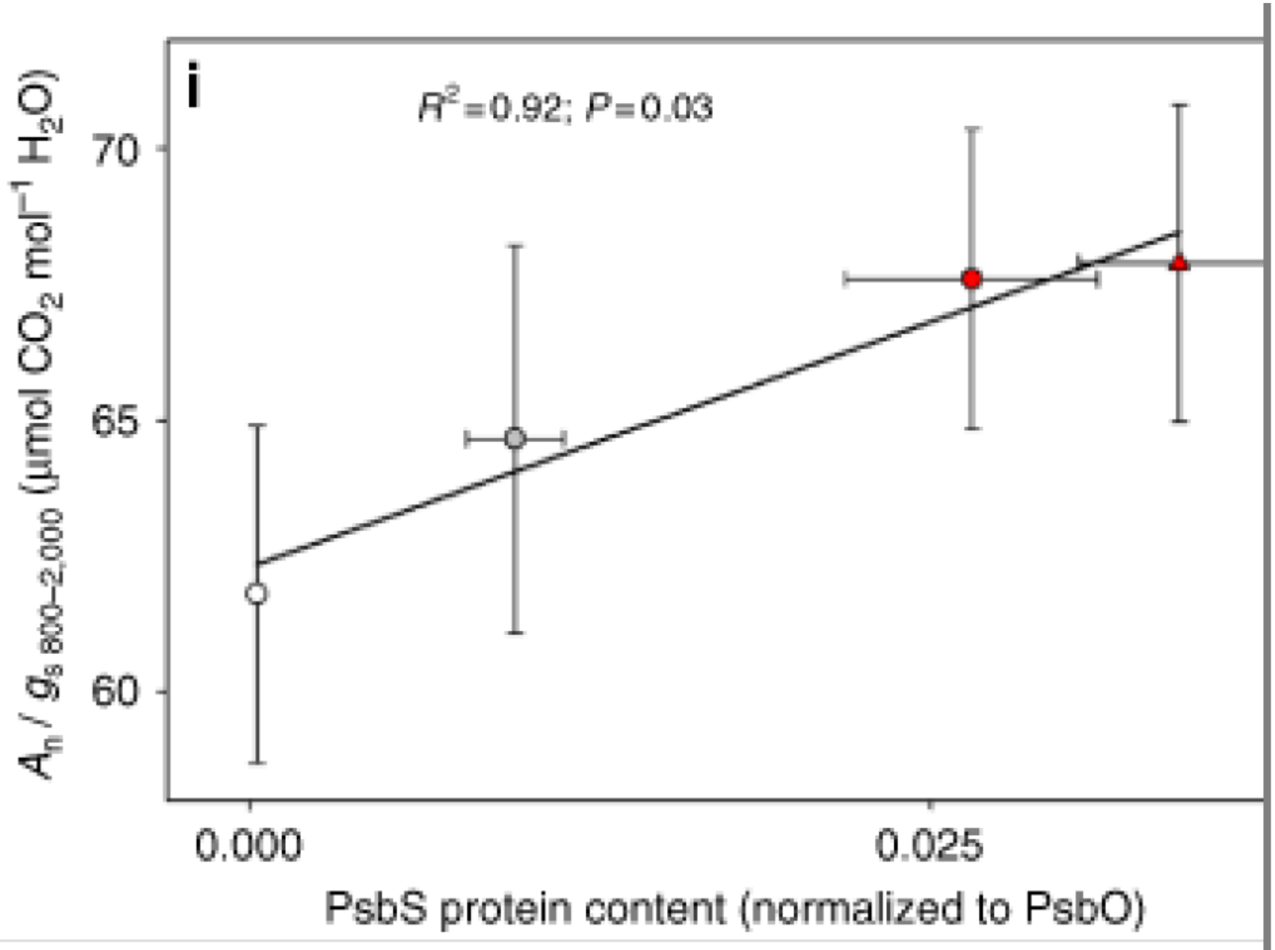
Niyogi_Science_cover-111716.png




# D TRIALS

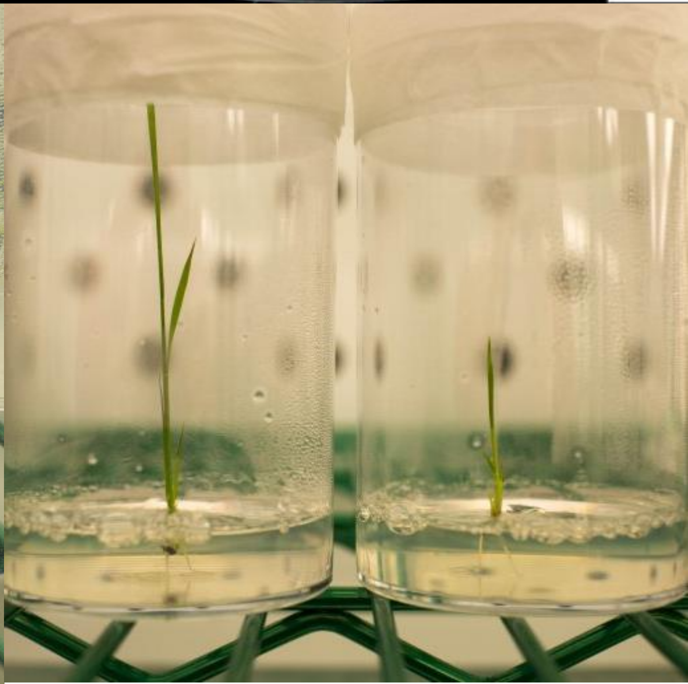

Kromdijk, Głowacka, Leonelli, Iwai, Niyogi & Long (2016) **Science 354,** 857-861.

Glowacka, Kromdijk, Niyogi & Long (2017) Nature Comms. **9**, 868, 1-11

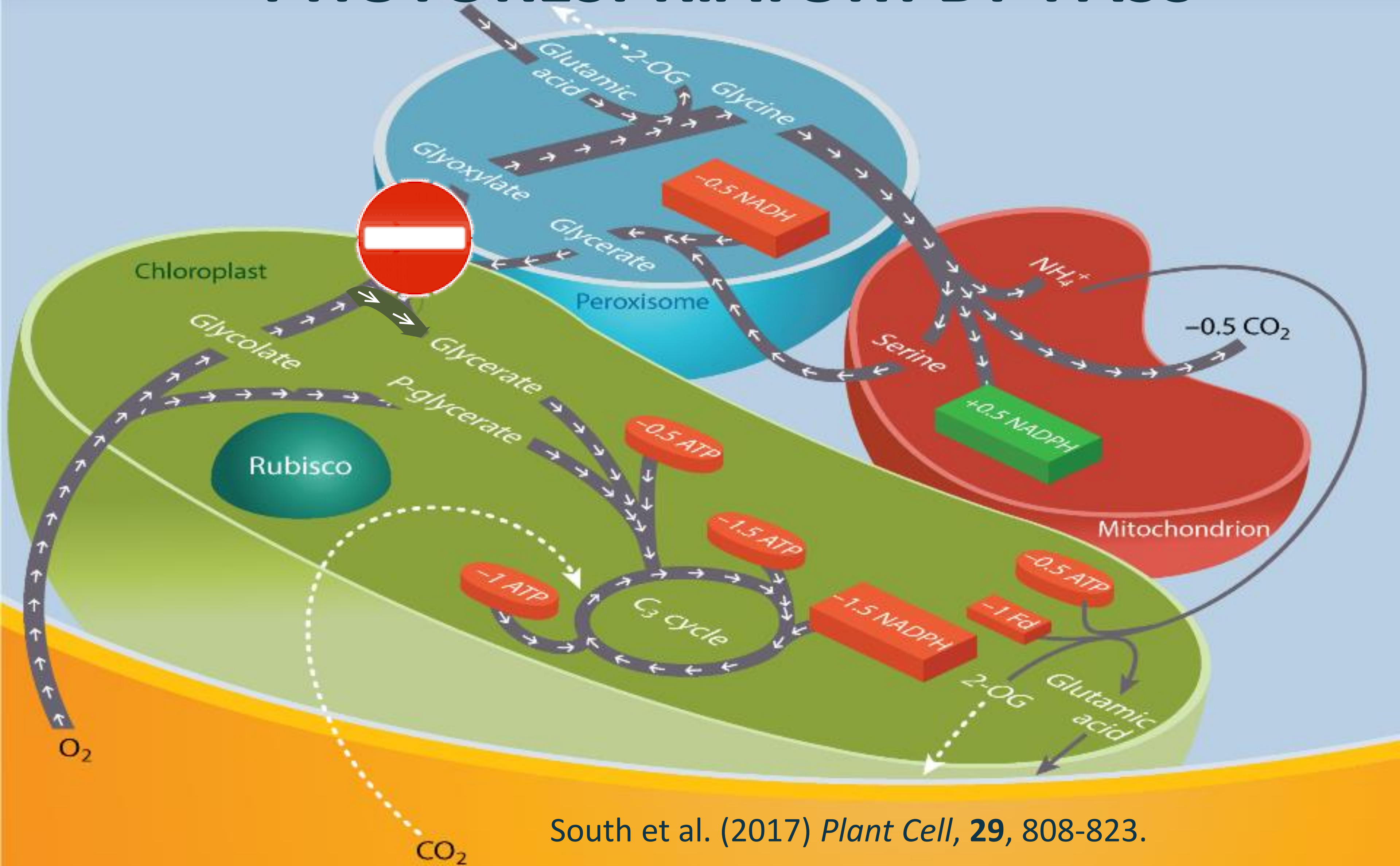




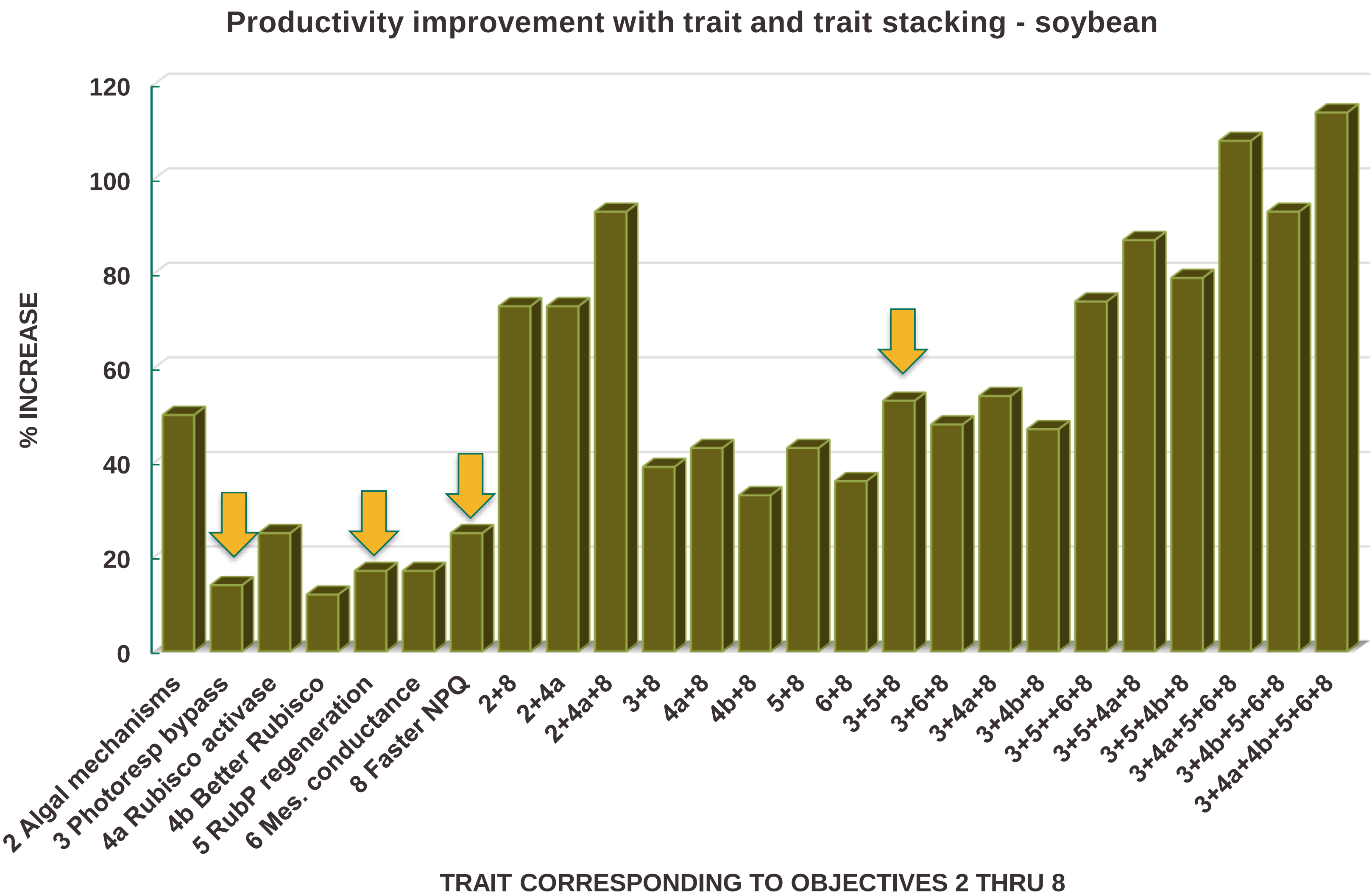
g



# PHOTORESPRIATORY BY-PASS




South et al. (2017) *Plant Cell*, **29**, 808-823.


Productivity improvement with trait and trait stacking - soybean
% INCREASE
0
20
40
60
80
100
120
2 Algal mechanisms
3 Photoresp bypass
4a Rubisco activase
4b Better Rubisco
5 RubP regeneration
6 Mes. conductance
8 Faster NPQ
2+8
2+4a
2+4a+8
3+8
4a+8
4b+8
5+8
6+8
3+5+8
3+6+8
3+4a+8
3+4b+8
3+5++6+8
3+5+4a+8
3+5+4b+8
3+4a+5+6+8
3+4b+5+6+8
3+4a+4b+5+6+8
TRAIT CORRESPONDING TO OBJECTIVES 2 THRU 8

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- Why photosynthesis?
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easy
hard

# BUILD ON THE GREATEST SUCCESS TO DATE IN DECARBONIZING ENERGY SUPPLY, BY USING AN ESTABLISHED DEPLOYABLE TECHNOLOGY.

LETTERS
PUBLISHED ONLINE: 23 OCTOBER 2017 | DOI: 10.1038/NCLIMATE3410

nature climate change

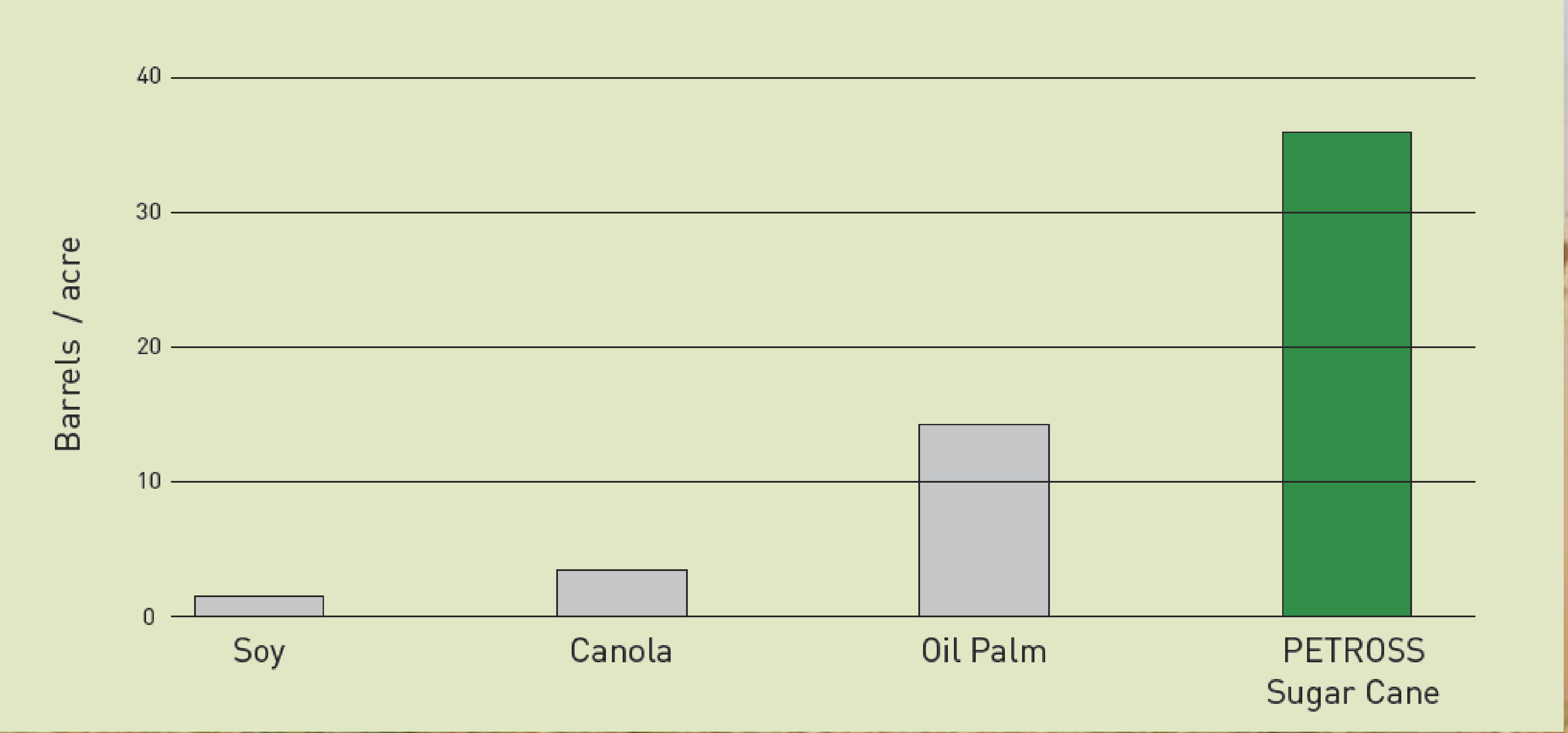
## Brazilian sugarcane ethanol as an expandable green alternative to crude oil use

Deepak Jaiswal[1†], Amanda P. De Souza[1,2], Søren Larsen[3,4,5], David S. LeBauer[1,6], Fernando E. Miguez[7], Gerd Sparovek[3], Germán Bollero[8], Marcos S. Buckeridge[2] and Stephen P. Long[1,8,9,10★]

**Reduction of $CO_2$ emissions will require a transition from fossil fuels to alternative energy sources. Expansion of Brazilian sugarcane ethanol[1,2] provides one near-term scalable solution to reduce $CO_2$ emissions from the global transport sector. In contrast to corn ethanol, the Brazilian sugarcane ethanol system may offset 86% of $CO_2$ emissions compared to oil use, and emissions resulting from land-use change to sugarcane are paid back in just 2–8 years[3,4]. But, it has been uncertain how much further expansion is possible given increasing demand for food and animal feed, climate change impacts and protection of natural ecosystems. We show that Brazilian sugarcane ethanol can provide the equivalent of 3.63–12.77 Mb $d^{-1}$ of crude oil by 2045 under projected climate change while protecting forests under conservation[5] and accounting for future land demand for food and animal feed production. The corresponding range**

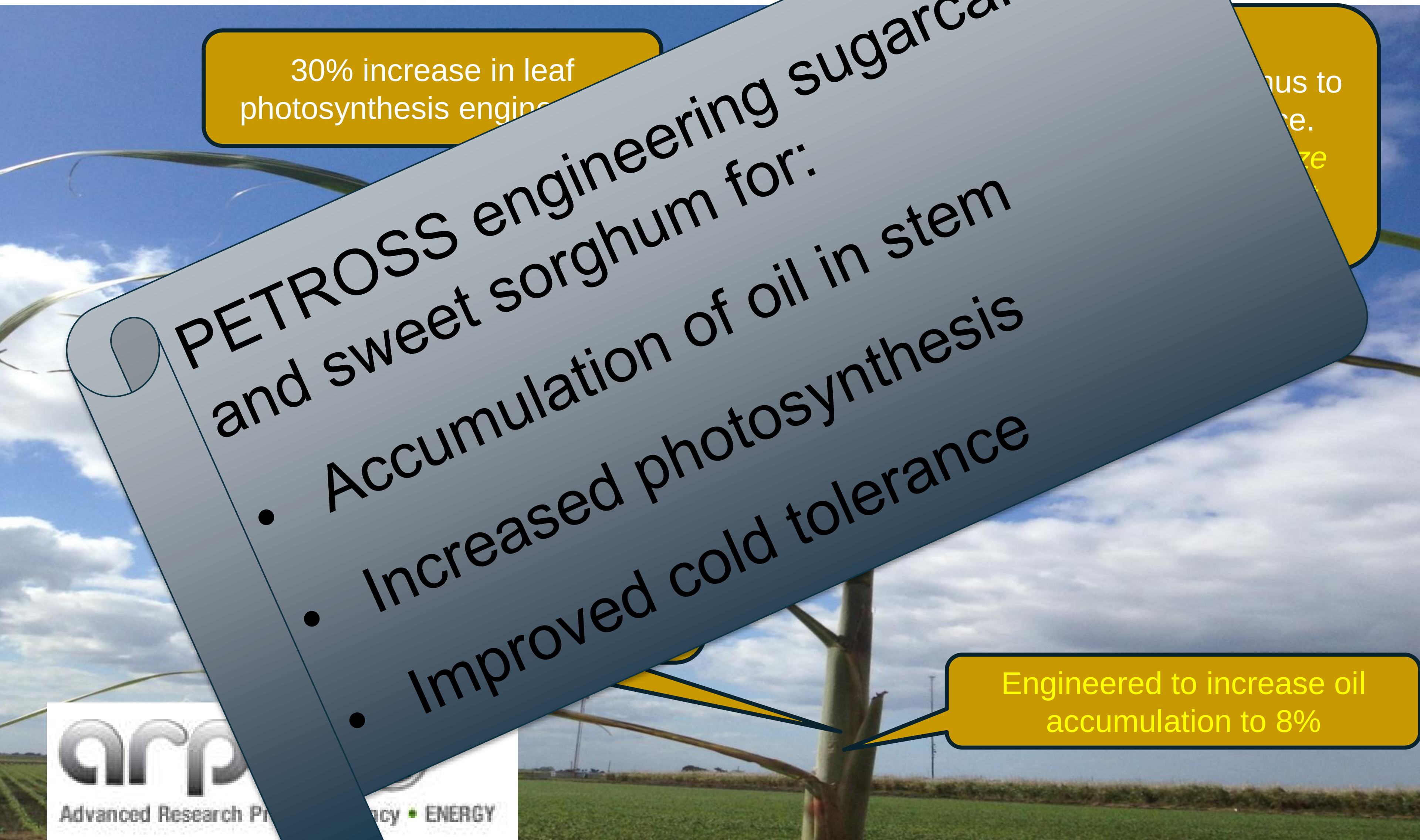
BioCro model (Supplementary Fig. 2 and Supplementary Table 1). Rather than project yield from empirical relationships, BioCro simulates plant growth hour by hour on the basis of underlying biophysical and biochemical mechanisms, using site-specific soil properties and hourly weather records[13]. Importantly, it includes the processes that respond interactively to increasing $CO_2$, temperature and drought incidence. The model was parameterized and calibrated at leaf, canopy, and field scales for gas exchange rates and biomass partitioning (Supplementary Figs 3–5 and Supplementary Table 1). BioCro performance was evaluated against independently measured yield data (Supplementary Fig. 6; r.m.s. error = 29 tons $ha^{-1}$; concordance correlation coefficient = 0.90) from multiple research sites in Brazil (Supplementary Table 2). Section 1 in the Methods details the parameterization and validation of BioCro

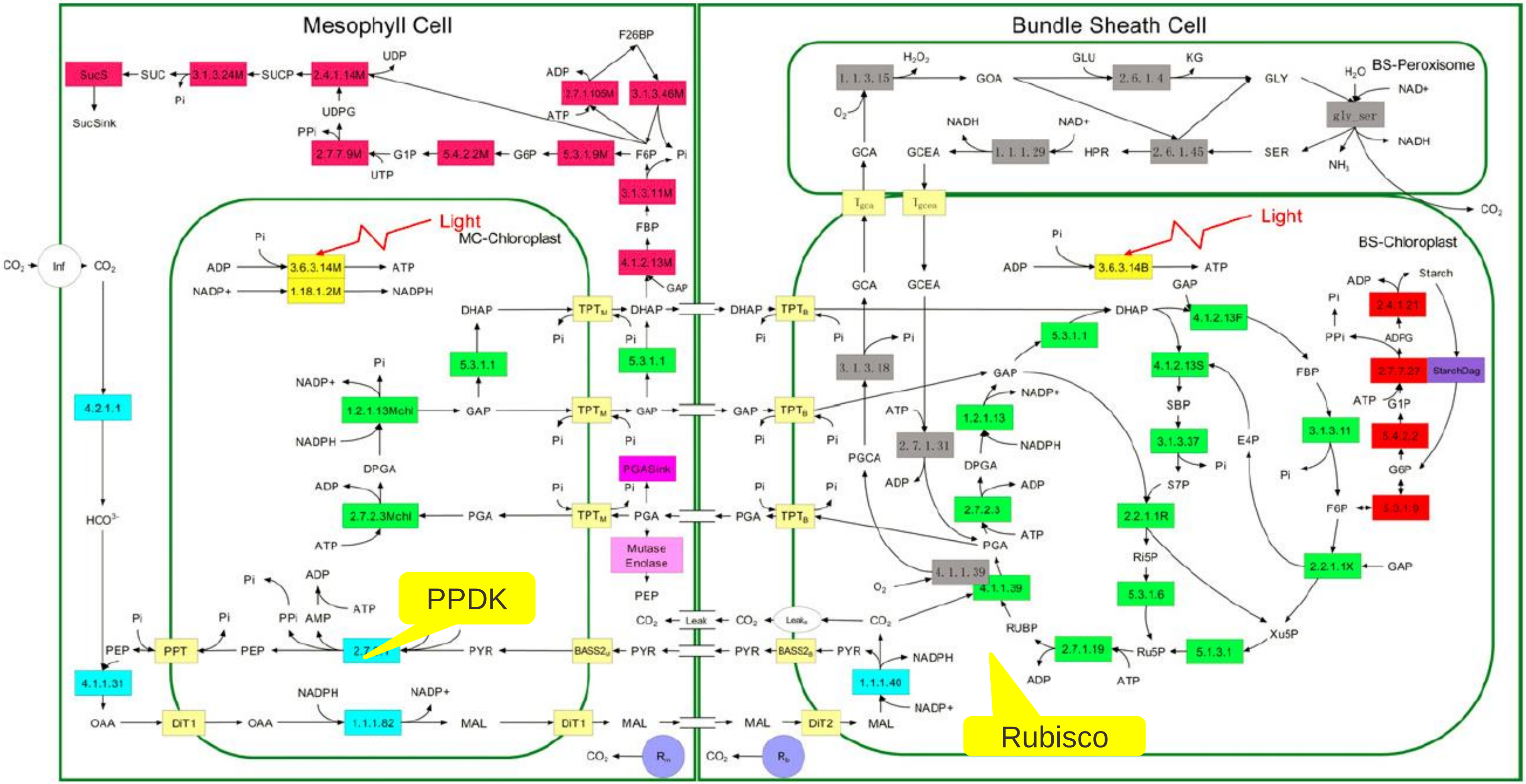
# PETROSS – U.Illinois-BNL-U.Florida-U.Nebraska
# Advantaged Oil-Producing Sugarcane – Our goal:
## MAKING BIODIESEL/BIOJET PRODUCTION VIABLE FOR LAND-USE




Advanced Research Projects Agency • ENERGY

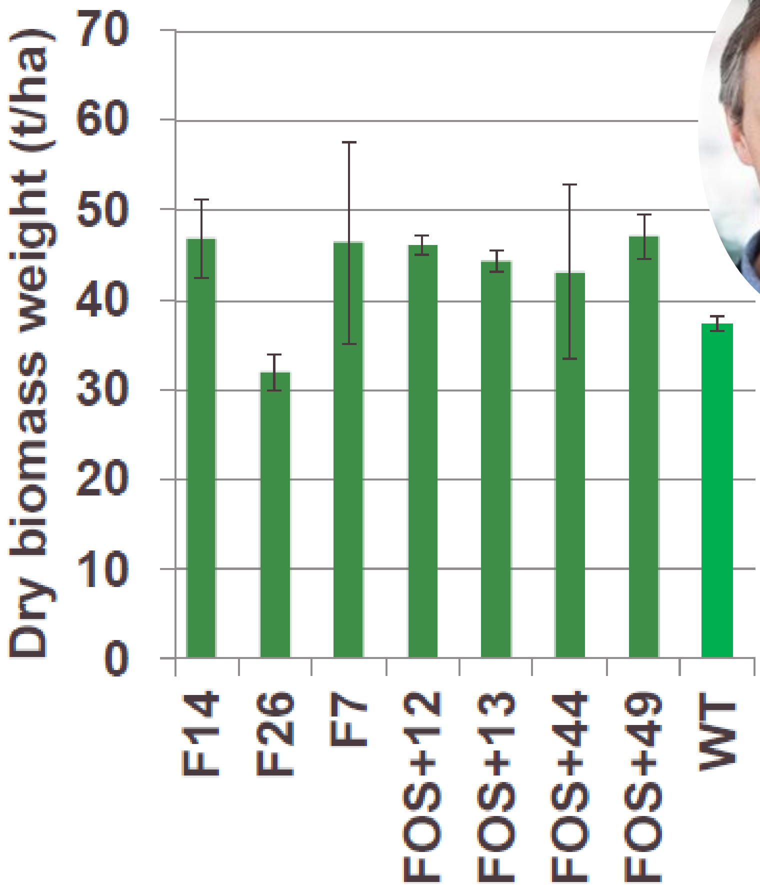
# PETROSS AT 4 YEARS





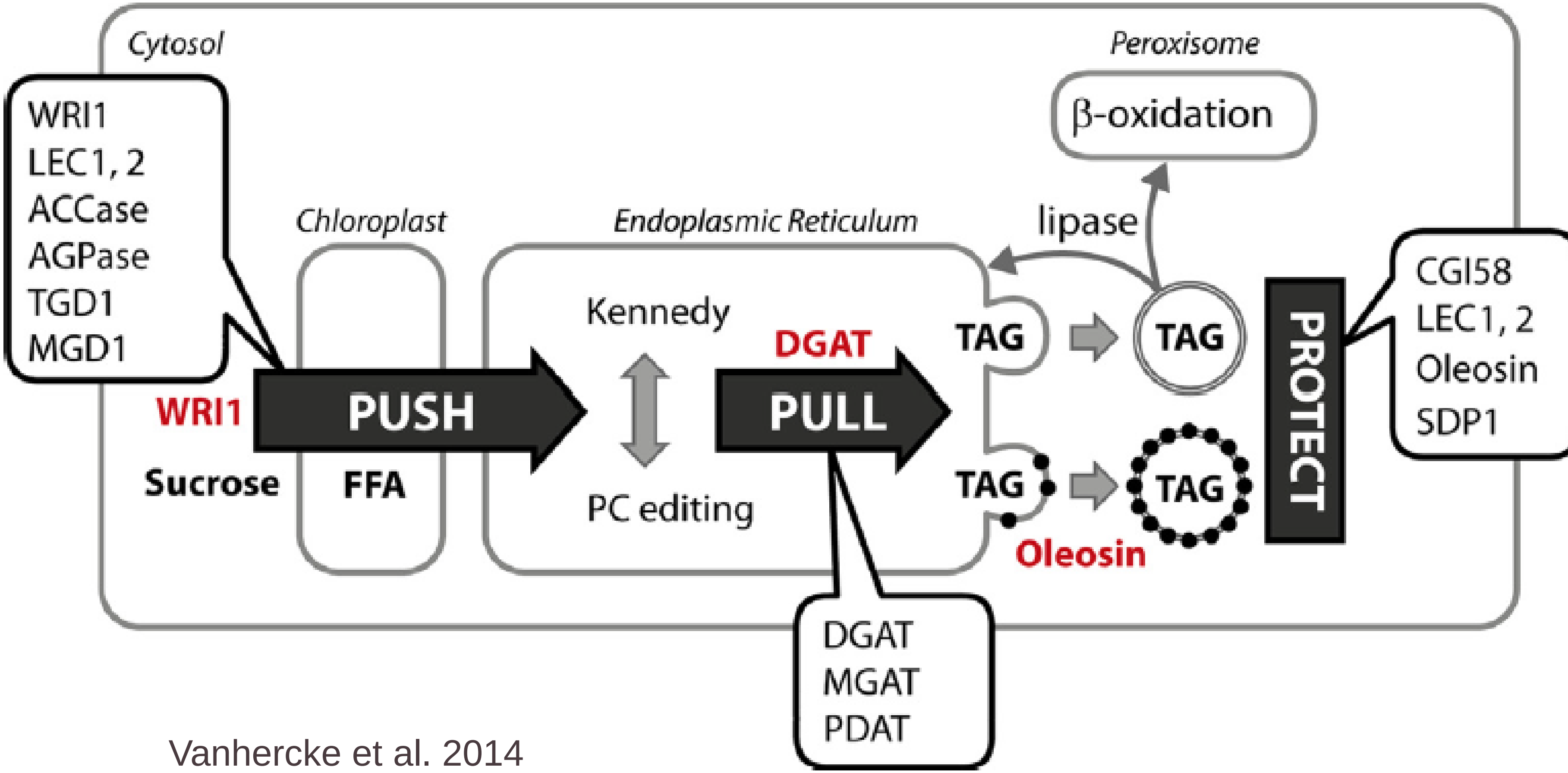

Wang, Long & Zhu (2014**) Plant Physiology 164**, 2231–2246

6 of 7 events; > +6 t/ha




# STRATEGY TO INCREASE OIL

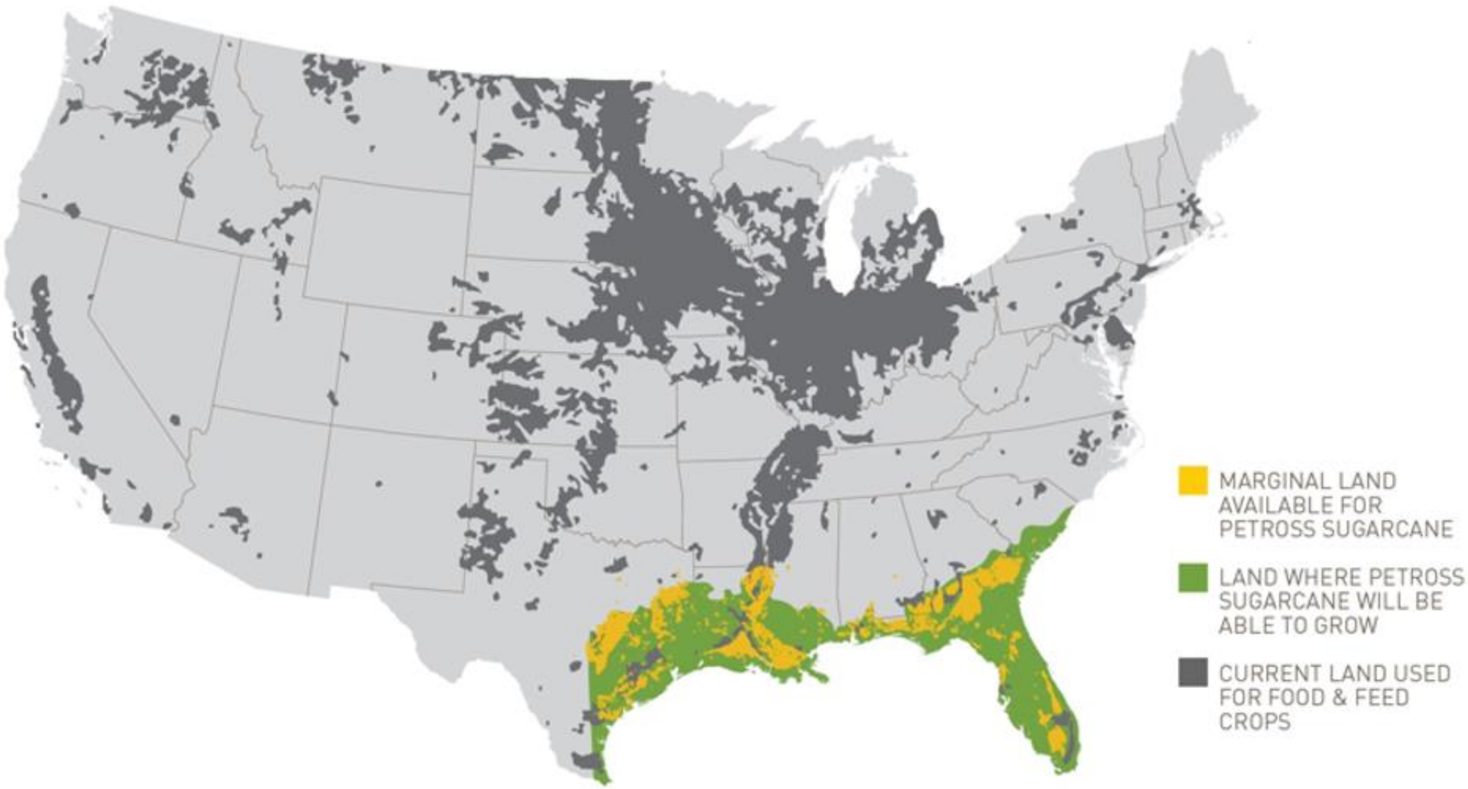
Metabolic engineering of sugarcane to increase TAG biosynthesis




Vanhercke et al. 2014

Genes described to influence accumulation of TAG in vegetative tissues

# WHERE COULD IT BE GROWN AND HOW MUCH?

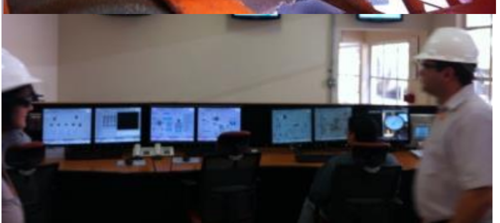



Using only the 23 million acres of marginal land in the area on the map, we estimate the PETROSS crops will be able to produce 24.67 billion gallons of biodiesel/biojet – easily meeting 2/3 of the needs set forth by the Renewable Fuel Standard mandate of 36 billion gallons of biofuel by 2022.  Excludes Hawaii and Puerto Rico!

# TECHNOECONOMIC ANALYSIS



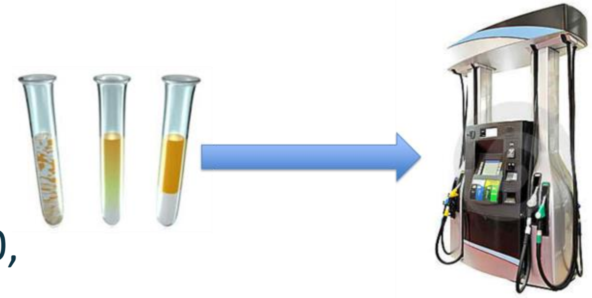


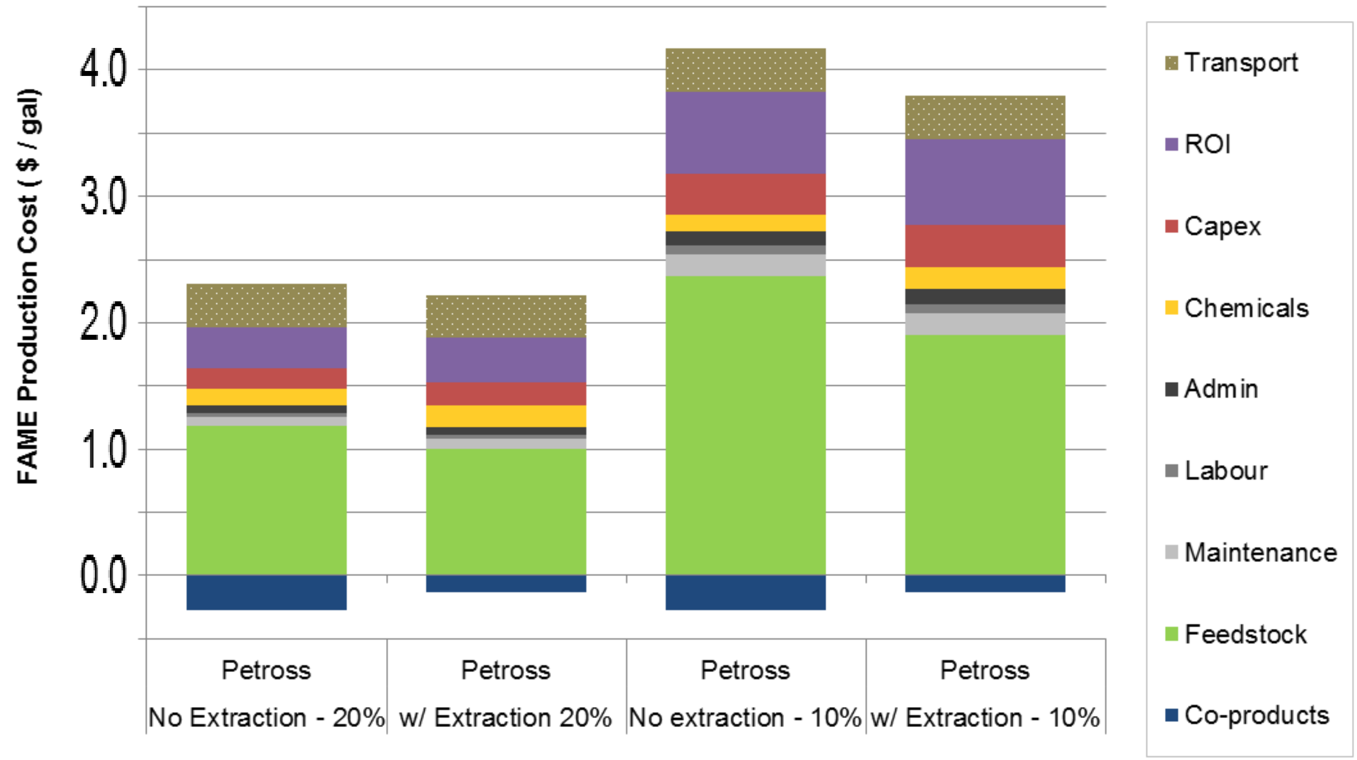


PETROSS Sugarcane 33 barrels/acre
Soybean 0.8 barrels/acre

Kumar D, Long SP, Singh V (2018) Global Change Biology Bioenergy, 10, 92-107

Long SP, Huang H, Singh V (2017) Patent US2015105546-A1 US9394503-B2





SoyFACE
Free-air Concentration Enrichment facility

soyface.illinois.edu

# MORE INFORMATION:

LONG LAB

lab.igb.illinois.edu/long

RIPE
Realizing Increased Photosynthetic Efficiency

ripe.illinois.edu

PETROSS
GENETICALLY ADVANTAGED OIL-PRODUCING SUGARCANE & SWEET SORGHUM

petross.illinois.edu

**An Incomplete list of credits**

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David LeBauer
Deepak Jaiswal
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Kasia Glowacka
Liana AcevedoSiaca
Lisa Emerson
Lindsay Clark
Lynn Massenburg
Mitch Altschuler
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