Selecting sugarcane with higher transpiration efficiency

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Prologue – ways to use physiological understanding in breeding*

3 pathways:

1. Identifying traits for indirect selection
   - Repeatable, cheap to measure, high genetic correlation with breeding objective

2. Identifying trait targets for introgression breeding

3. Identifying environments for selection
   - Eg. conditions that maximise expression of desired genetic variation

Jackson, Cooper, Robertson, Hammer. 1996. *Field Crops Research*

Other concepts, definitions:

“PREDICTION” of yield or sugar content

- for a breeder, this usually refers to predicting the *relative* performance or *ranking* of economic value of a set of genotypes across the targeted environments, not absolute levels of performance of genotypes.

Selection index theory

A selection index is a single number used to rank a set of candidate clones being selected using several measurements at the same time:

$$SI_i = b_1*X_{1i} + b_2*X_{2i} + ... + b_n*X_{ni}$$

where SI_i = the selection index of genotype i ; b_1, b_2, …, b_n are the index coefficients to be estimated (below) for trait 1, trait 2, …, trait n; and X_{1i}, $X_{2i,, …,}$ X_{2n} are the measurements trait 1, trait 2, …, trait n

For example, in early stage selection, may measure yield, sugar content, canopy temperature via UAV at different times…

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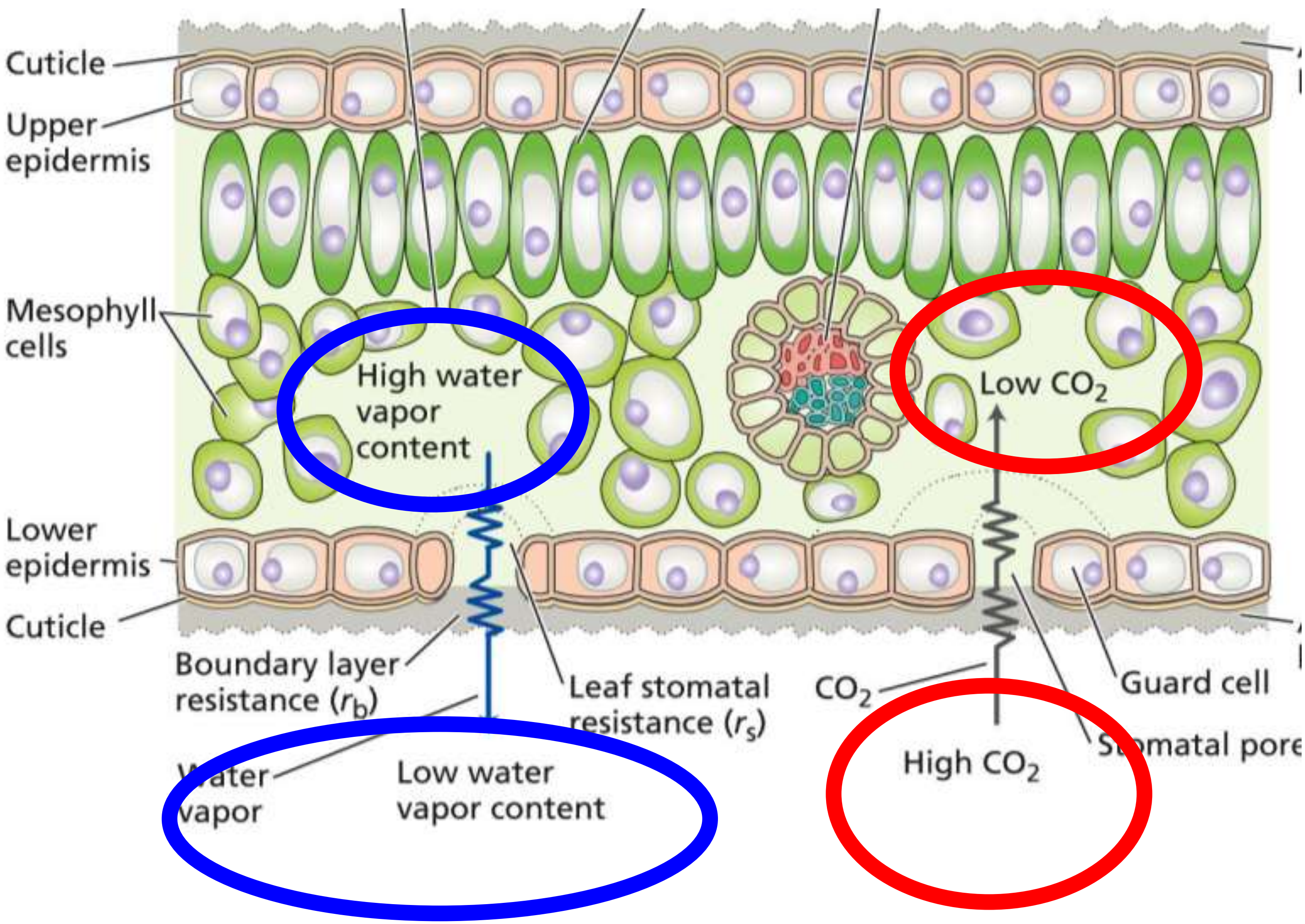
Transpiration efficiency (TE)

TE = biomass growth/water lost through stomata


Cuticle
Upper epidermis
Mesophyll cells
High water vapor content
Low CO_2
Lower epidermis
Cuticle
Boundary layer resistance (r_b)
Leaf stomatal resistance (r_s)
CO_2
Guard cell
Stomatal pore
High CO_2
Water vapor
Low water vapor content

Transpiration efficiency (TE)

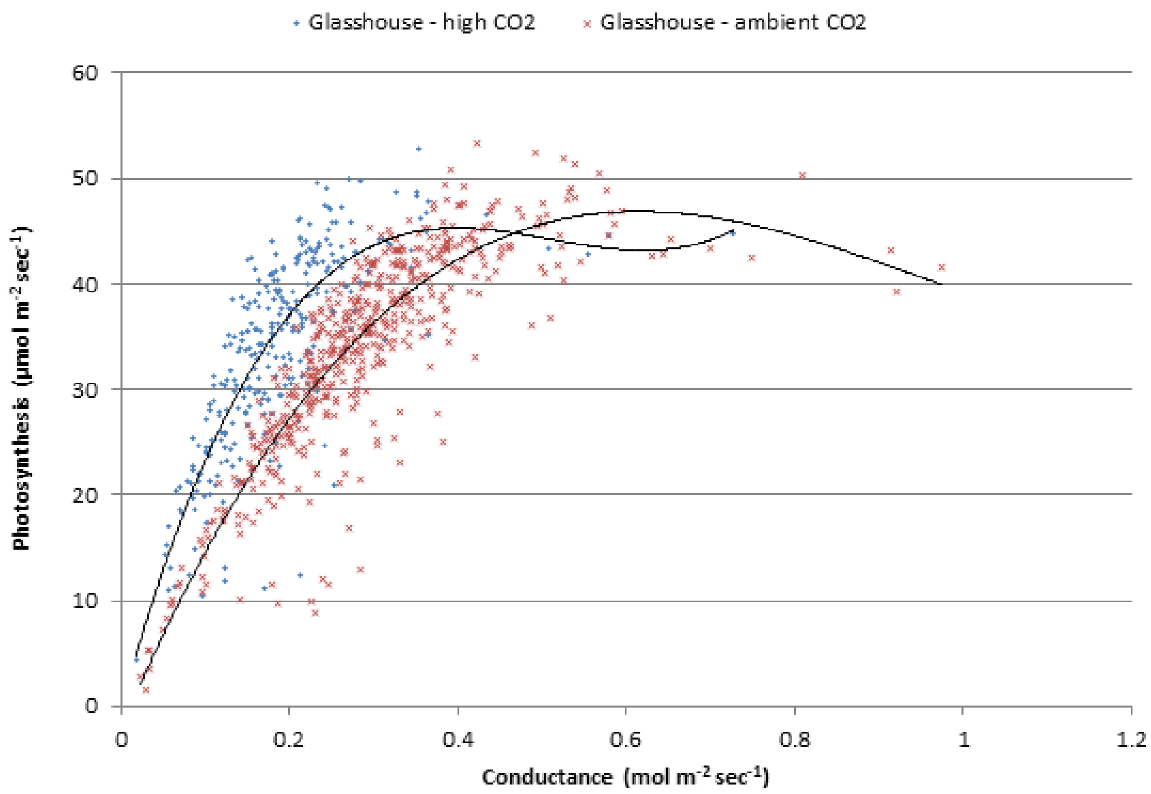
TE = biomass growth/water lost through stomata

Biomass yield = Transpiration x TE

Importance of TE – a range of industry issues

- Water availability is biggest limitation in rain-fed areas in sugarcane
- Costs of water (electricity) increasing for irrigated farms
- Expansion of industry limited by amount of cane per water
  - Expansion on fringes of existing rainfed regions
  - Water use efficiency is a major driver on return on investment for new major industry areas
- What are the implications of rising CO2? (currently ~400ppm, increasing at ~2ppm per year and accelerating)

High CO2 levels decrease conductance, have little impact on photosynthesis, and therefore increase TE.

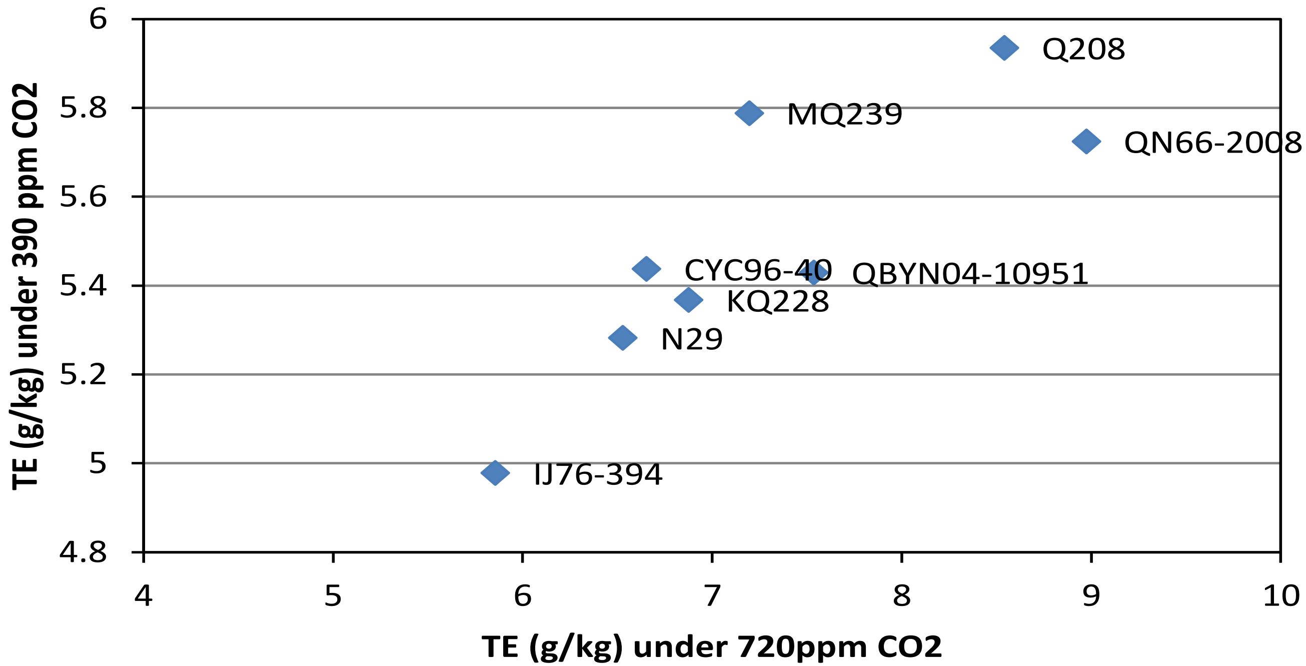


720ppm versus 390ppm shown

Location	Photosynthesis ($\mu mol\ m^{-2}\ sec^{-1}$)	Conductance ($mol\ m^{-2}\ sec^{-1}$)	Ci (ppm)
390 ppm CO_2	33.8	0.29	151
720 ppm CO_2	34.1	0.20	318

Loc x Water	Water use (l/pot)	Growth (g/pot)	TE (total) (g/l)
720 ppm			
Dry	34.7	273	8.1
Wet	47.6	310	6.5
390 ppm			
Dry	53.3	309	5.9
Wet	63.2	325	5.1

Effects of increasing CO2 level on genotype ranking



Learning from past work on genetic improvement in transpiration efficiency…

- Lots of research in other crops
- Experience from other crops – not yet major impact on cultivar development
- Why? Largely because of negative genetic correlation between TE and transpiration
    - Is this the case for sugarcane?
    - If yes, can we/how to/ address this?

Examples of genetic variation in TE

Clone	Type	TE (g/L)
IJ76-394	*E. arundinaceus*	8.63
QN66-2008	Commercial parent	8.28
Q253	Commercial cultivar	7.34
Q208	Commercial cultivar	7.47
KQ228	Commercial cultivar	5.95
QS04-772	Commercial parent	5.76
	Mean	6.86
	LSD (P<0.05)	1.46

Around ±20% of the mean found

Jackson *et al* (2015) J.Exp.Bot;
Stokes *et al* (2016) ASSCT

Change in biomass (yield) by changing TE by 20%

Location	Irrigation	% change	
		- 20%	+ 20%
Bambaroo	None	-14.9	13.1
Bundaberg	None	-17.8	16.5
	Irrigation	-14.1	10.7
Kuttabul	None	-15.2	13.3
	Irrigation	-11.5	8.2
Mackay	None	-14.9	13.5
	Irrigation	-10.9	7.3
Macknade	None	-14.2	11.8
Meringa	None	-18.1	17.0
Mirani	None	-16.2	14.6
	Irrigation	-13.2	10.2
Plane Creek	None	-14.6	12.9
	Irrigation	-12.1	8.5
Tully	None	-8.2	6.2

Details in:
Stokes *et al* (2016) ASSCT.

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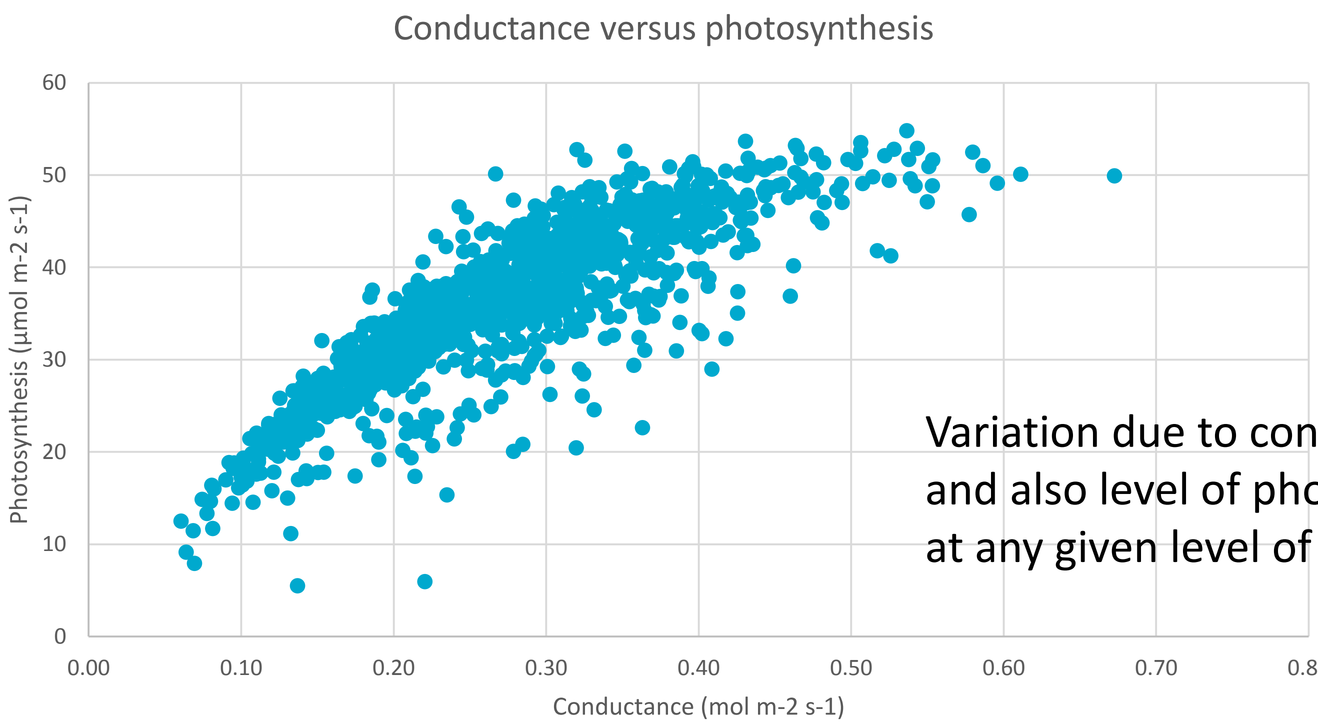
BUT…high TE tends to come with reduced transpiration and less biomass…

	Leaf area	Shoot DW	Root DW	R/S ratio	Total DW	Water use
a) Experiment 1a (49 genotypes)						
Leaf area	1					
Shoot DW	0.79	1				
Root DW	0.62	0.88	1			
R/S ratio	0.03	0.23	0.64	1		
Total DW	0.74	0.98	0.96	0.41	1	
Water use	0.75	0.96	0.94	0.39	0.98	1
TE	-0.41	-0.36	-0.3	-0.02	-0.35	-0.50

Details in: Jackson *et al* (2015) *J.Exp.Bot*

Reason for negative genetic correlation – curvilinear relation between conductance and photosynthesis



Variation due to conductance and also level of photosynthesis at any given level of conductance

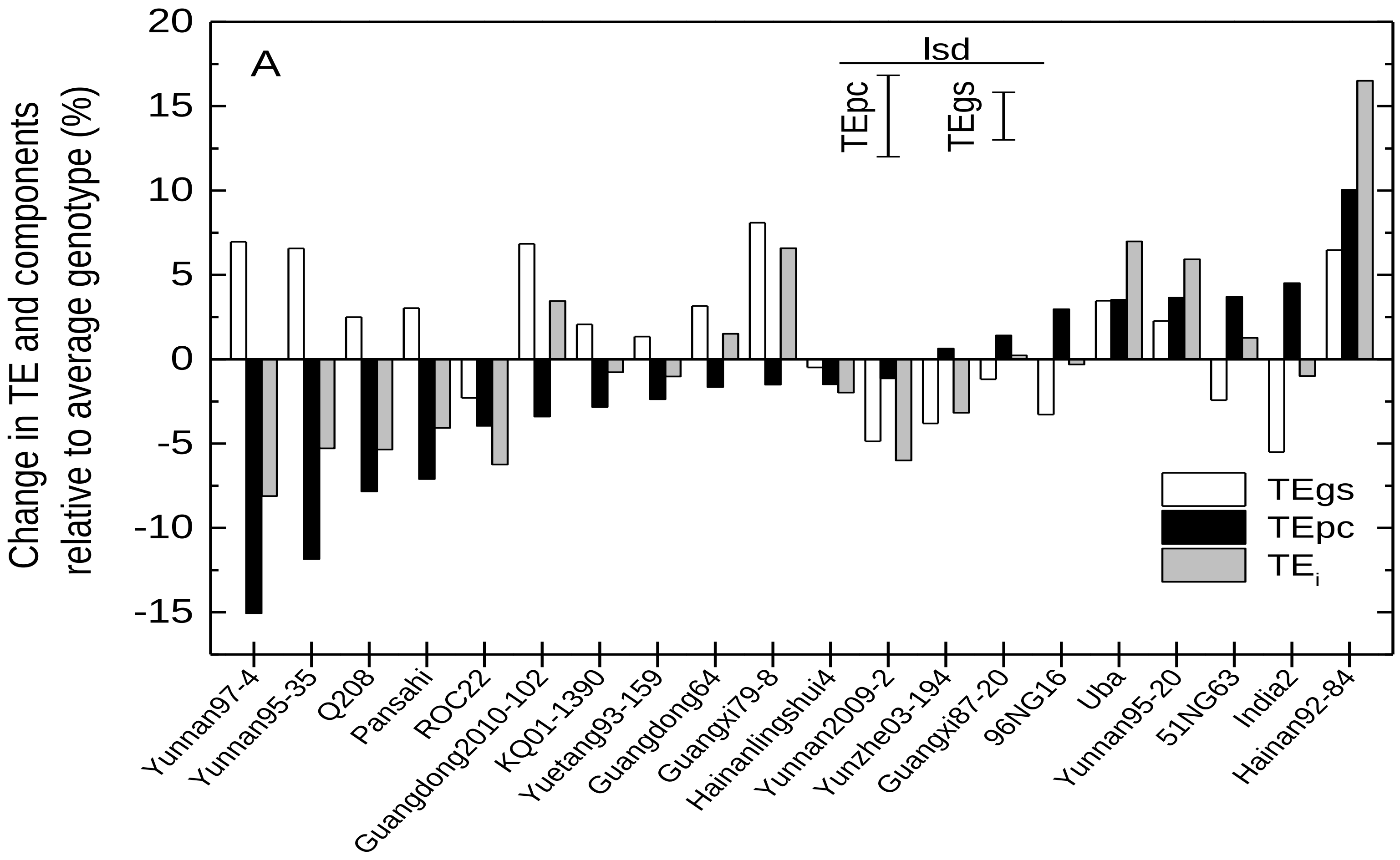
Variation in TE can be partitioned into that due to conductance variation and that due to photosynthesis capacity

Both components are important, conductance causes most variation, but highly significant variation due to photosynthesis capacity does exist:

Statistic	A (µmol m^{-2} s^{-1})	g_s (mol m^{-2} s^{-1})	C_i (µL L^{-1})	TE_i (A/g_s) (µmol mol^{-1})	TEgs (µmol mol^{-1})	TEpc (µmol mol^{-1})
GCV (%)	25.3	27.4	8.6	5.3	4.0	2.9
σ_{clones}^2	19.1***	0.00122***	132***	55.8***	32.1***	16.9***
$\sigma_{clone\ x\ dates}^2$	4.81**	0.00029**	5.1 ns	7.9 ns	4.92**	0.52 ns
σ_{error}^2	36.3	0.00278	1198	470	63.0	183.1
H_b (all data basis)	0.96	0.96	0.87	0.88	0.94	0.91
H_b (single measure basis)	0.32	0.29	0.10	0.10	0.21	0.14

Li *et al*, 2017 (*J. Exp. Bot.*)

Is possible to look at TE components (conductance vs photosynthesis capacity) for individual clones



Li *et al*, 2017 (*J. Exp. Bot*. in press)

Some key points from physiological research:

- “Naturally occurring” genetic variation in breeding populations exist at approx. ± 20%
  - Likely larger variation with targeted crossing and selection

- A 1% increase in TE (assuming no negative impact on transpiration rate) changes cane yield overall by 0.5-0.9% in rainfed and supplementary irrigation environments

- Genetic variation in TE due to conductance changes and photosynthesis changes.   Variation in both.   Need to separate the two in selection.

Application in breeding programs?

- Why is selecting for TE better than just selecting for yield directly?
- Selecting for TE alone will reduce yield?
- If the trait is useful (ie. promotes high yield) it should automatically be selected for indirectly anyway…
- The measurements are labour intensive…

Competition in early stages – probably selects for vigour (high conductance) and against transpiration efficiency

Early stage of selection

- Only one selection environment, small plots
- Wish to use data to predict performance across a range of environments
- Currently use yield + sugar content
- Use low stress environments normally (need to grow well for planting material, reduce error variation…)

Current line of thinking for application:

- Early phase selection (stages 1,2) – using single low stress environment only

- Hypothesis:
  - for selecting for water limited environments, want both high yield + high TE (combined)
  - Eg. two clones with similar growth rates and high yield – if one has a high TE it will run into water stress later
  - An index combining (yield + TE) will be predictive of yield under water stress

Evidence supporting this hypothesis

- Field experiment:
  - 22 clones (later stage selections)
  - Planted at two sites
  - Each site has irrigated and rainfed treatments
  - 3 reps, 10m x 4 row plots
  - Plant + 2 ratoon crops

- Measured leaf temperature several times
- Cane yield + sugar content at harvest

Comment on leaf temperature

- Shown to strongly relate to relative rate of transpiration (evaporation cools down the leaf: low temp = high transp.)
- Could be used as a measure of relative rate of water use in plots and therefore an index for relative TE (TE ~ yield/rel water use)
- Can be estimated using UAV imaging, therefore amenable to practical application in large scale selection trials

Correlation between measurements of clones made in irrigated treatment and cane yield in dry treatment:

Measurements in irrigated treatment	Correlation with cane yield in dry treatment
Yield alone	0.52**
Leaf temperature alone	-0.10(ns)
Yield + leaf temperature	0.64**

Both yield and leaf temp. have positive coefficients – ie. clones with high yield and high temp (low rate of water loss) perform best in dry treatment

Correlation between measurements of clones made in irrigated treatment and cane yield in dry treatment:

Measurements in irrigated treatment	Correlation with cane yield in dry treatment
Yield alone	0.39 (ns)
S. Conductance alone	-0.25 (ns)
Yield + conductance	0.57**

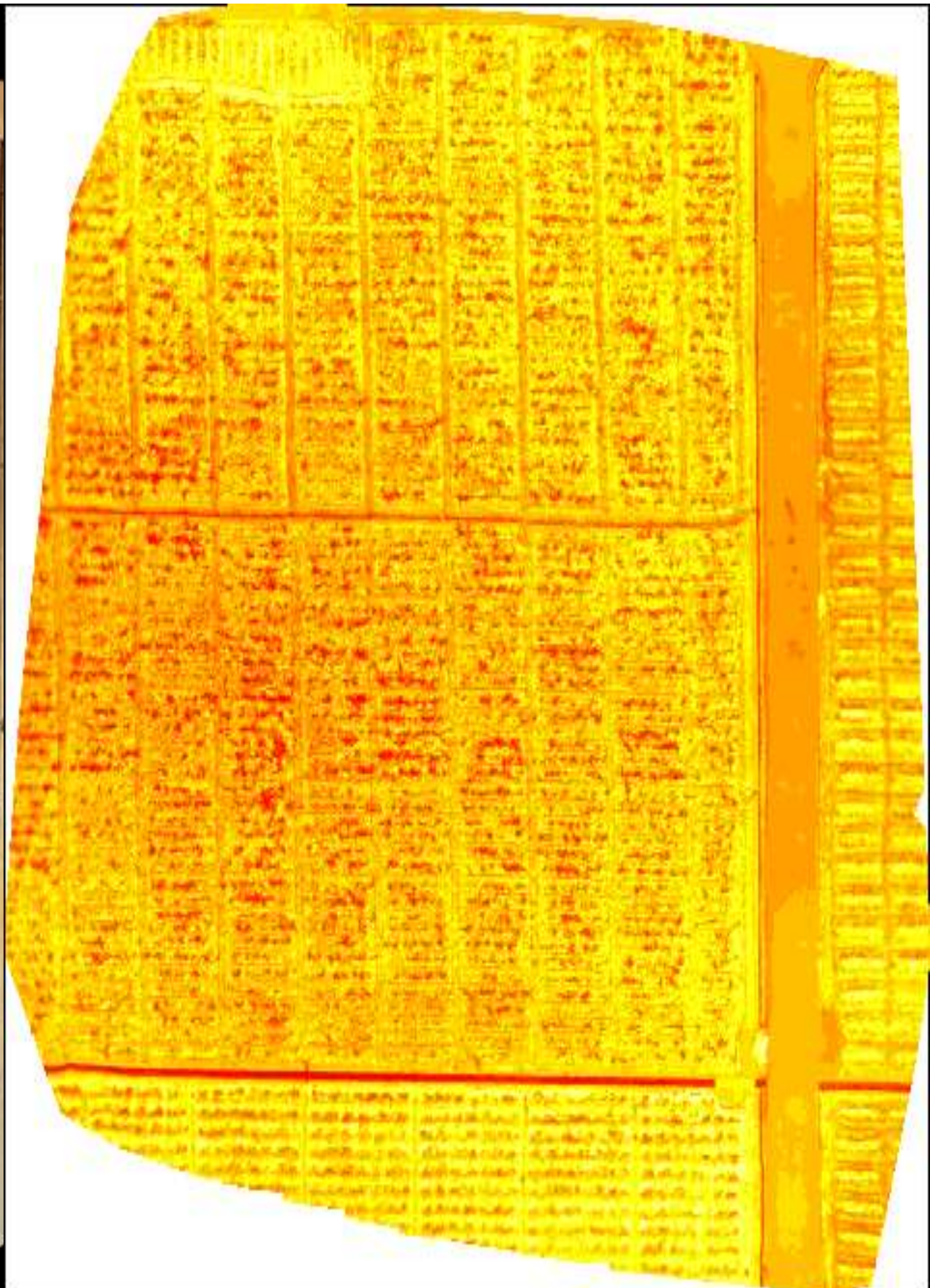
Yield and conductance have positive and negative coefficients respectively – ie. clones with high yield and low conductance (low rate of water loss) perform best in dry treatment

Key points:

- Theoretical and preliminary empirical results support hypothesis that yield + water use rate could provide a useful selection index for environments with range of water limitation (despite data limitations)

- Needs lots of testing & lots of optimising:
  - Low cost measurement technology (UAV based estimates of leaf temp and canopy cover)
  - Sampling issues (Right conditions for measuring canopy temp: time of day, weather; how many times of measurement for accuracy, etc…)

- But, if it works, this approach could provide a valuable and practical index in early stages of selection in breeding programs

Next steps

- Australian project currently underway (SRA/CSIRO) looking at using UAV imaging for estimating relative conductance
- Workshop in Yunnan/China (July) to review current available image technology options and directions of this area of work
- The focus of next steps should be on technology and concept testing within practical selection systems (sugarcane, possibly other crops)

Potential area of collaboration:

- This is a possible area for more mutually beneficial international collaboration

- For others – utilize expertise being developed in CSIRO and SRA
- For CSIRO/SRA – acquisition of more data and environments for developing and testing the technology and selection indices

Thank you

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