International Center for Tropical Agriculture
Since 1967 Science to cultivate change

6. June 2023 – NIAB/ABC/GCRF

The climate change-driven challenges and opportunities between physiology and breeding – focus on Phaseolus

Milan Oldrich Urban, PhD

WE means: **Jonatan Soto, Jaumer Ricaurte, Duvan Pineda,** Diego Conejo, Camilo Preciado, Karol Sanchez, Javier Gereda, Ramiro Sabogal, Mauricio Bechara, Ana Salgado, Estephania Ortiz, Jorge Aragon, Seider Culchac, Edilfonso Melo, Esther Torres, Herika Pinta, Yeison Grajales, Jarden Molina, Marta Nupan

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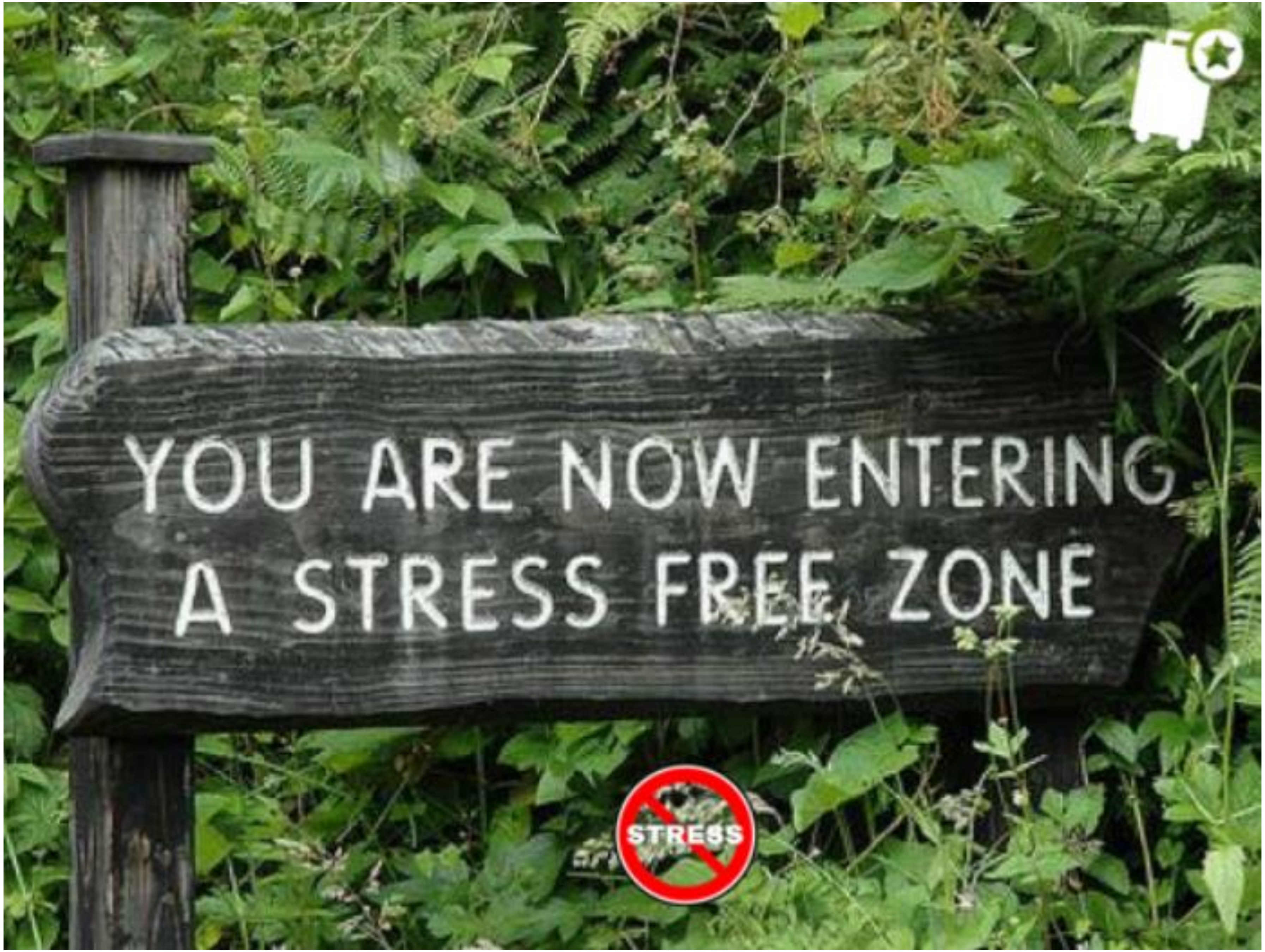
YOU ARE NOW ENTERING
A STRESS FREE ZONE
STRESS

Acknowledgements

- GIZ Germany
- Bean Physiology team
- Climate change modelers – Prakash Jha, Chetan Deva
- University of Amazonia – Juan Salazar
- Michael Selvaraj - Phenomics team
- Dr. STEVE BEEBE + others

- **General introduction**
- **What do we do/use?**
- **Recommendations, Ideotype, Conclusion**

Plant breeding is grounded in prediction

- Plant breeding programs are the operational implementations of coordinated sequences of prediction methods, organized to continuously create, evaluate, and select new genotypes over multiple breeding program cycles (Duvick et al., 2004; Cobb et al. 2019; Technow et al. 2021)

SOCIETY FOR EXPERIMENTAL BIOLOGY

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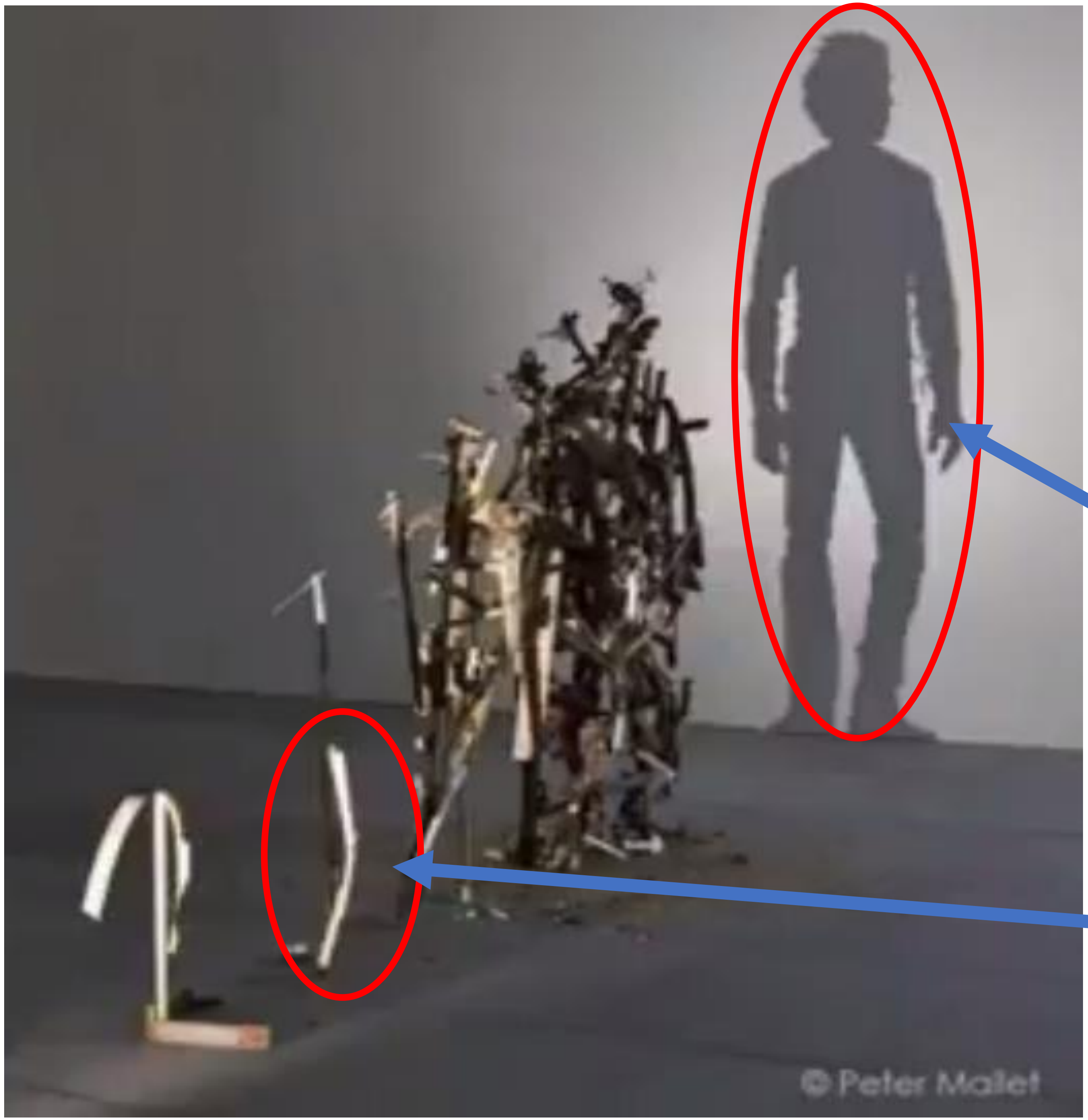
REVIEW PAPER

In pursuit of a better world: crop improvement and the CGIAR

Jana Kholová[1,*], Milan Oldřich Urban[2,*], James Cock[2,*,†], Jairo Arcos[3], Elizabeth Arnaud[4], Destan Aytekin[5], Vania Azevedo[1], Andrew P. Barnes[6], Salvatore Ceccarelli[7], Paul Chavarriaga[2], Joshua N. Cobb[8], David Connor[9], Mark Cooper[10], Peter Craufurd[11], Daniel Debouck[2], Robert Fungo[12,13], Stefania Grando[7], Graeme L. Hammer[10], Carlos E. Jara[14], Charlie Messina[15], Gloria Mosquera[2], Eileen Nchanji[16], Eng Hwa Ng[17], Steven Prager[2], Sindhujan Sankaran[18], Michael Selvaraj[2], François Tardieu[19], Philip Thornton[20], Sandra P. Valdes-Gutierrez[2], Jacob van Etten[4], Peter Wenzl[2] and Yunbi Xu[21,22]



Relevant phenotyping is needed and implies deep understanding of biological mechanisms and their interactions

Lucky breeders ??

Poor physiologists
HOW TO FIND IT?

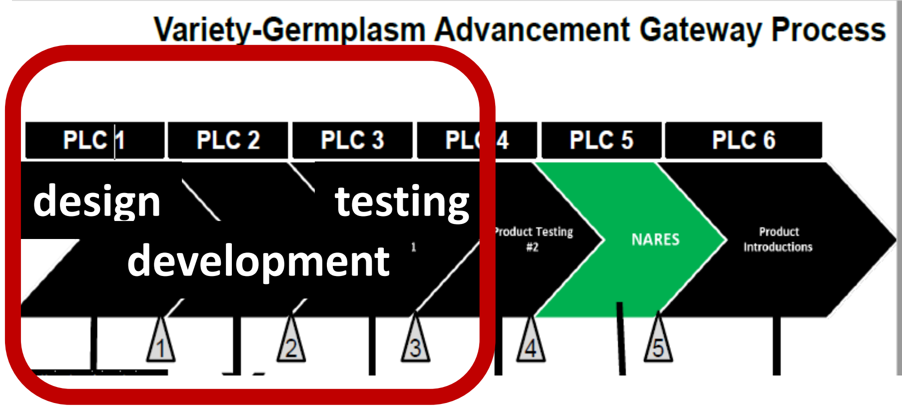
1. Crop improvement strategy - CGIAR

Demand for crop products

example; post-rainy sorghum

Sorghum bread

Livestock feed



genotyping

Accelerated selection

phenotyping

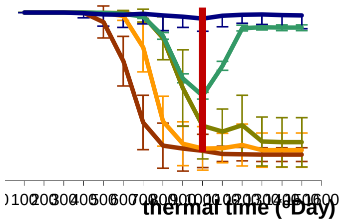
Markets & socioeconomic research

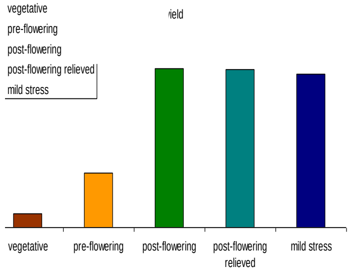
Agro-ecology specific breeding

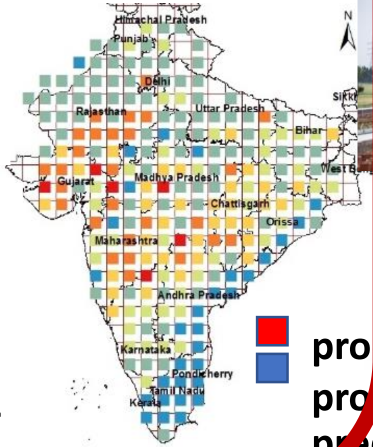
Strategic crosses

TPE

GxExMxS options







production gain

production loss

predictions

targetted phenotyping

Early Populations (1000s)

Advanced populations (100s)

Agronomy package (10s)

Yield quantity & quality

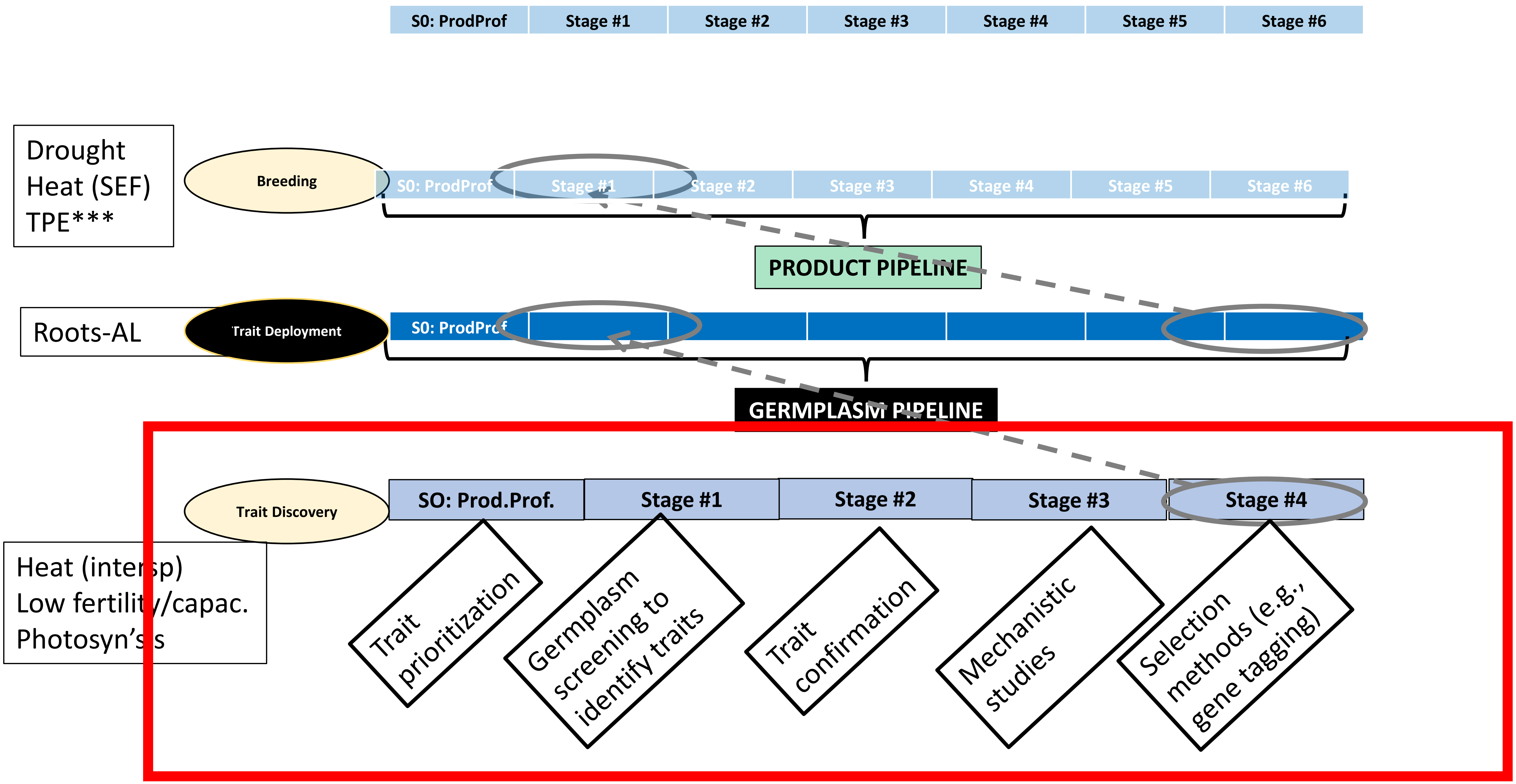
phenotyping pipeline

adaptations –> yield –> quality

product acceptability in TPEs

Objectives of “good” physiology team

- Obtain **proof of concepts** for physiological trait combinations that boost yield – theoretical,
- **Realistic** breeding context (TPE)
- **Broaden the genepool** (wise and selective)
- Deliver novel sources of **traits in acceptable** agronomic background
- Provide **selection protocols** complementing breeding



S0: ProdProf
Stage #1
Stage #2
Stage #3
Stage #4
Stage #5
Stage #6
Drought
Heat (SEF)
TPE***
Breeding
S0: ProdProf
Stage #1
Stage #2
Stage #3
Stage #4
Stage #5
Stage #6
PRODUCT PIPELINE
Roots-AL
Trait Deployment
S0: ProdProf
GERMPLASM PIPELINE
Trait Discovery
SO: Prod.Prof.
Stage #1
Stage #2
Stage #3
Stage #4
Heat (intersp)
Low fertility/capac.
Photosyn's is
Trait prioritization
Germplasm screening to identify traits
Trait confirmation
Mechanistic studies
Selection methods (e.g., gene tagging)

Why physiology is/could be so important?

- Trait is in relationship to stress resistance
- expressed when the crop is exposed to stress

  (constitutive vs induced vs hormesis)
- timing coincides with the plant most sensitive stage of development
- response gradually to stress
- correlated with grain yield with acceptable heritability
- easily measurable or observable
- needs to be verified

- Architecture, seed (size, color, cooking time), yield, seed quality,

Back to the basics: *Phaseolus* spp. originated in contrasting agro-ecologies

coccineus
dumosus
costaricensis

HUMID-SUB HUMID

Secondary Gene Pool

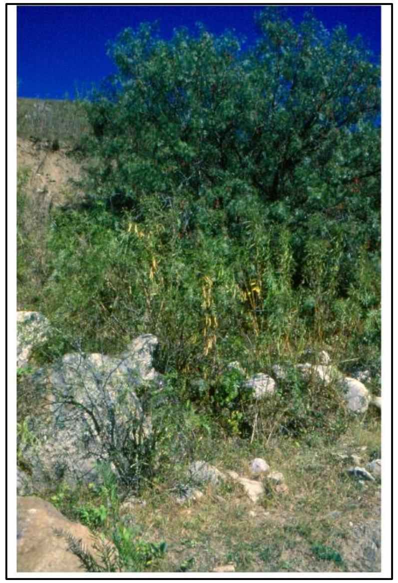
SUB-HUMID to SEMI-ARID

Primary Gene Pool

SEMI-ARID to ARID

Tertiary Gene Pool

acutifolius
parvifolius



Survival is Vegetative vigor;
Disease resistance
This results in poor harvest index

Survival is quick seed production;
Drought, heat resistance
This results in high harvest index

Lessons from Darwinian Agriculture:
the advantage of wilds

- High plant performance is **not essential for plant evolution**
- Protection favoured by evolution have **negative effects on plant performance in most stress scenarios**
- “Risky strategies” genes
  - may have been lost during catastrophes.
- Breeders counteracting the conservative strategies chosen by evolution **(high risk, high productivity).**

Tardieu F. Any trait or trait-related allele can confer drought tolerance: just design the right drought scenario, JXB 63 (1), 2012

ALL Traits have dual effect

1) Does a given trait confer a positive effect on yield in an appreciable proportion of years/scenarios in TPE?

2) In a TPE, what is the trade-off between risk and performance?

3) Will a given trait have positive effect with climate change?

Trait	Abundance of known genes/ alleles affecting the trait	Variable for phenotyping	Positive effect	Drawbacks	Scenario for maximum positive effect	Scenario for maximum negative effect
Short crop cycle	***	Duration (°Cd)	Escape : end of cycle occurs with non-depleted soil water reserve	Lower cumulative photosynthesis during the crop cycle	Very dry year	End of cycle with favourable conditions
Cell protection against stress	***	Aspect, biomass	Controversial[1] probably minor except in very severe stresses	Controversial and variable	Very severe	?[2]
Avoidance via stomatal closure	*	Aspect, biomass gas exchange, thermography	Keep soil water, ↘hydraulic gradients	↘ photosynthesis ↗ leaf temperature (heat stress)	Terminal severe stress	End of cycle with favourable conditions
Avoidance via reduced leaf area	**	Aspect, biomass	Keep soil water, ↘ hydraulic gradients	↘ photosynthesis	Terminal severe stress	End of cycle with favourable conditions
Water use efficiency	**	$\Delta^{13}C$, ratio biomass/ transpiration	↗ Crop for drop ; Avoidance	↘ photosynthesis	Terminal severe stress	End of cycle with favourable conditions
Maintained photosynthesis /stomatal conductance	*	Gas exchange thermography[3]	↗ biomass	↗ risk of stress at end of cycle	Medium/mild stress	Terminal severe stress
Maintained vegetative growth	**	NDVI, proxidetection[4]	↗ biomass	↗ risk of stress at end of cycle	Medium/mild stress	Terminal severe stress
Increased root growth	**	DNA, imaging thermography[5]	↗ water uptake	Competition for C; ↗ risk of stress at end of cycle	Deep water available	Shallow soil
Root architecture : Deeper roots without change in biomass	?[2]	Rhizotrons	↗ water uptake	↘ nutrient uptake	Deep water available	Low nutrient availability in upper layers
Reduced seed abortion	*	Direct observation seed number	↗ yield	↘ quality	Stress during flowering, relieved afterwards	Terminal severe stress

HOW to understand where the effect will be +/- ?

Bean Physiology team – what we are working on

- Only a small part of the methodologies (re)invented by(in) the team.
- Shortened, strongly modified
- All of the connected to (potential) phenotyping of wild acc and other species

frontiers | Frontiers in Plant Science

TYPE Original Research
PUBLISHED 08 December 2022
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SPECIALTY SECTION
This article was submitted to Plant Breeding,
a section of the journal

Using phenomics to identify and integrate traits of interest for better-performing common beans: A validation study on an interspecific hybrid and its Acutifolii parents

Diego Felipe Conejo Rodriguez[1*], Milan Oldřich Urban[2*], Marcela Santaella[1], Javier Mauricio Gereda[1], Aquiles Darghan Contreras[3] and Peter Wenzl[1]

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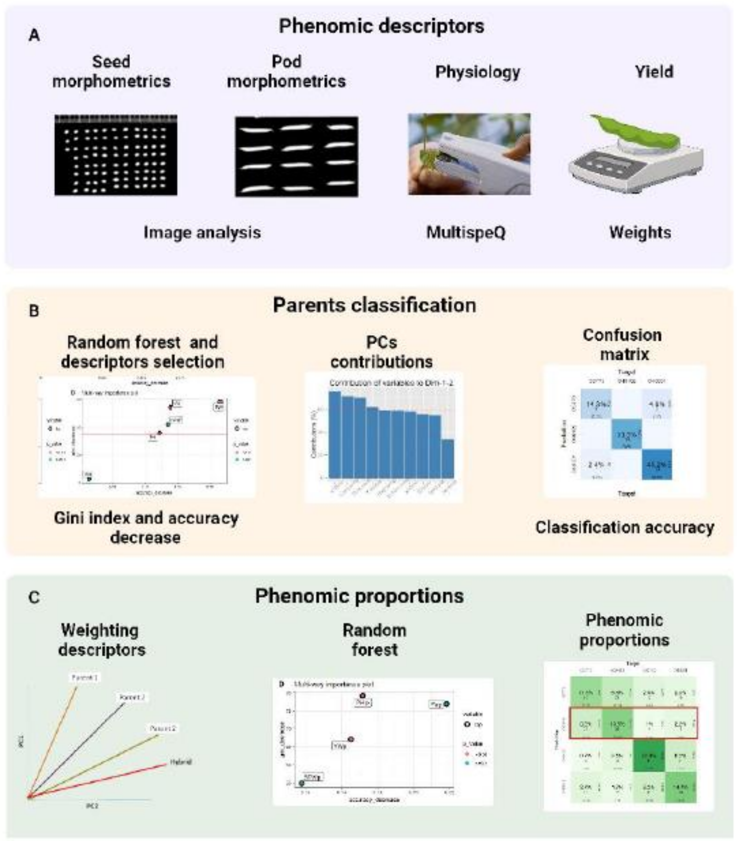
Phenomics descriptors reveal phenotypic ratio between interspecific hybrids with parental lines

Detección de rasgos asociados a tolerancia a altas temperaturas en Phaseolus



Fuente: Conejo-Rodriguez et al (2022)

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CITATION

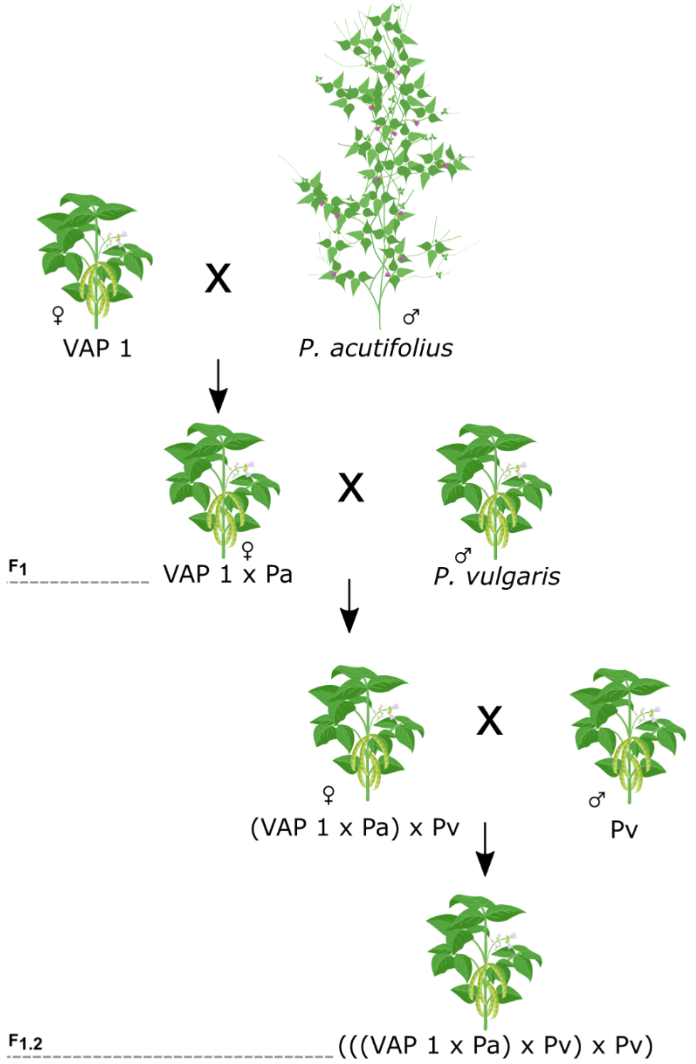
Interspecific common bean population derived from *Phaseolus acutifolius* using a bridging genotype demonstrate useful adaptation to heat tolerance

Cruz Sergio, Lobatón Juan, Urban O. Milan, Daniel Ariza-Suarez, Raatz Bodo, Aparicio Johan, Mosquera Gloria and Beebe Stephen*

Bean Breeding Program, International Center for Tropical Agriculture (CIAT), Palmira, Colombia

Interspecific Mesoamerican Wild Tepary Population (IMAWT)

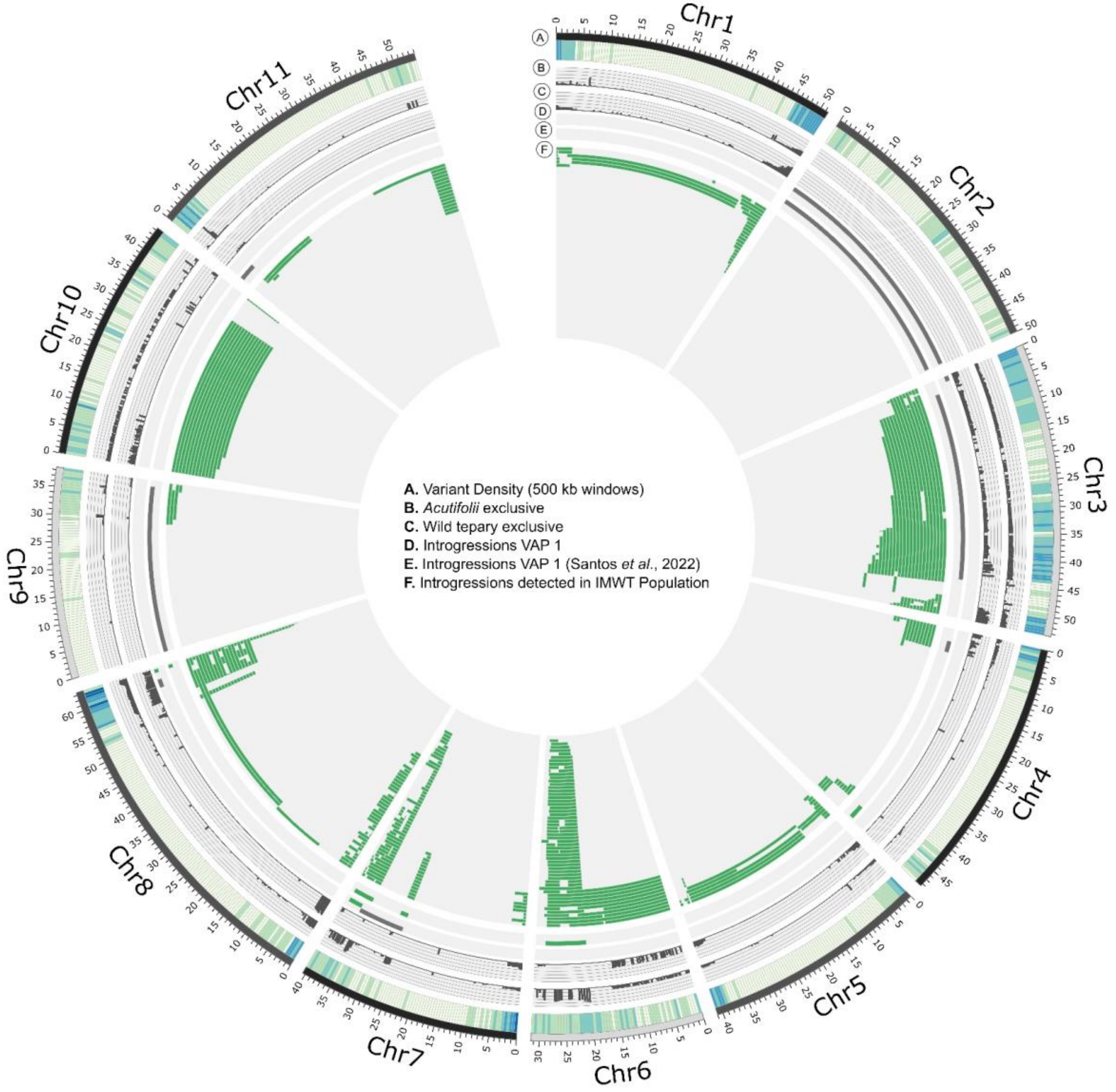
Figure 1: Population Parental lines



((VAP 1x *P. acutifolius*) X *P. vulgaris*) X *P. vulgaris*

12.5% 12.5% 25% 50%

Introgression Analysis



The population in general showed **59.8% of introgressions coming from the** ***Acutifolii parentals***

Phenobox: Standardize images capture

Photographic record

1

Photo box

Cannon SX60

30 seeds/parental
30 seeds/hybrid genotype

Contrasting background

Color Chart

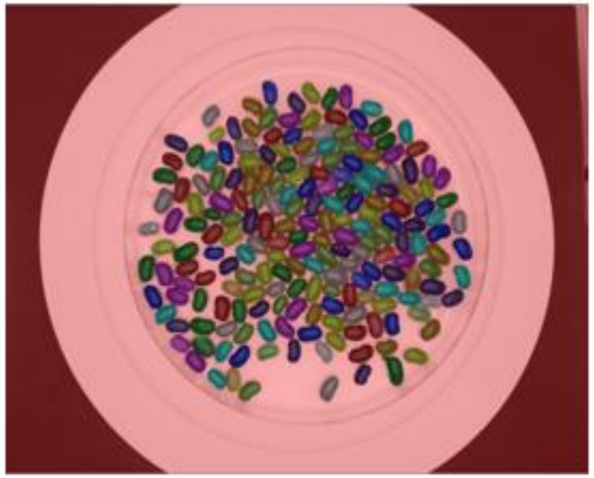
High quality image. (RAW)

Image processing
Pretraining model – Deep learning

1. Image features

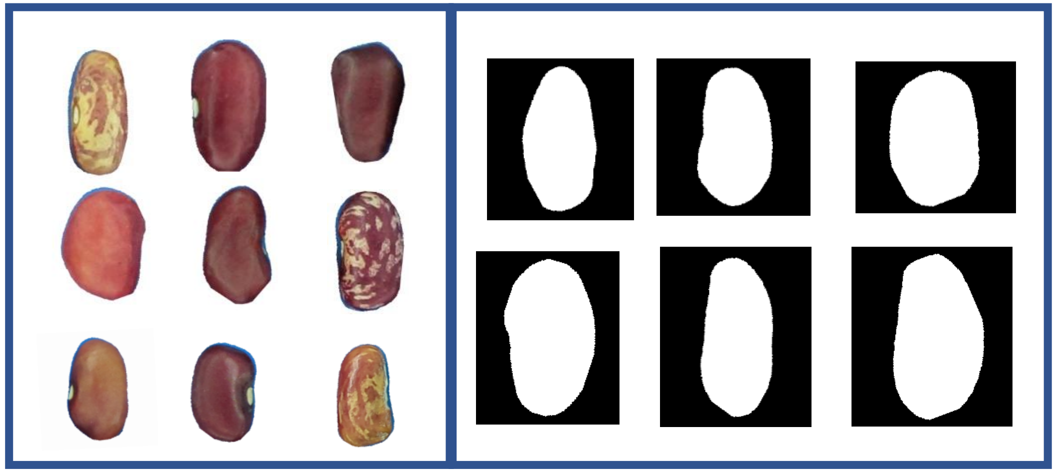
a. QR code
b. jpeg format input
c. Color card
d. > 30 seeds

2. Seed detection



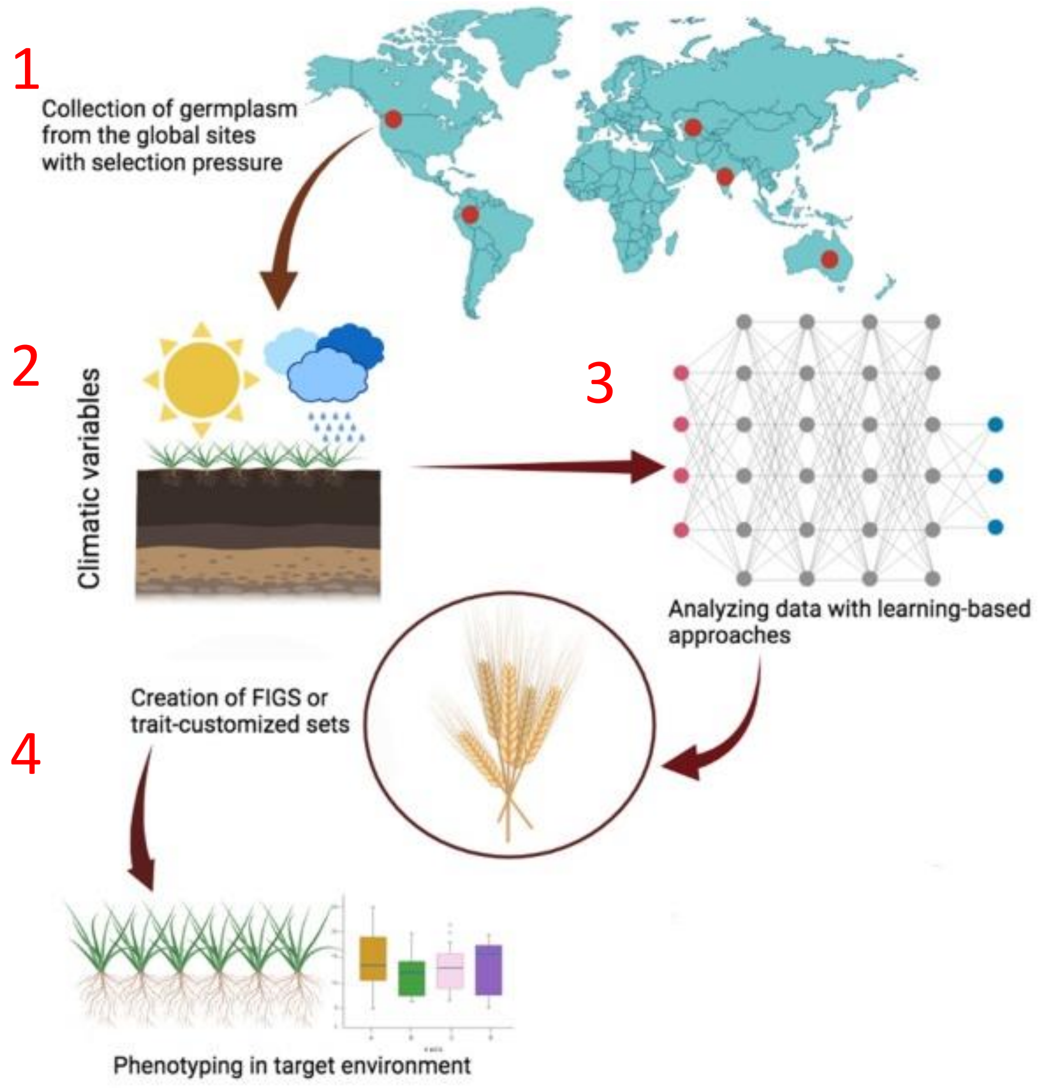
a. **Phyton script**
b. Image segmentation
c. Select seeds
d. Individual seed image

3. Color individual and binarize - contour seed



a. White Background color images
b. Black background binarize images
c. Individual image for seeds
d. jpeg format image output

Focused Identification of Germplasm Strategy (FIGS) contribute traits mining genebanks collections



The absence of phenotypic data on accessions restricts the exploration and use of valuable genetic resources (Furbank & Tester 2011)

Fuente: Stenberg, J. A., & Ortiz, R. (2021); Bohra *et al* (2021)

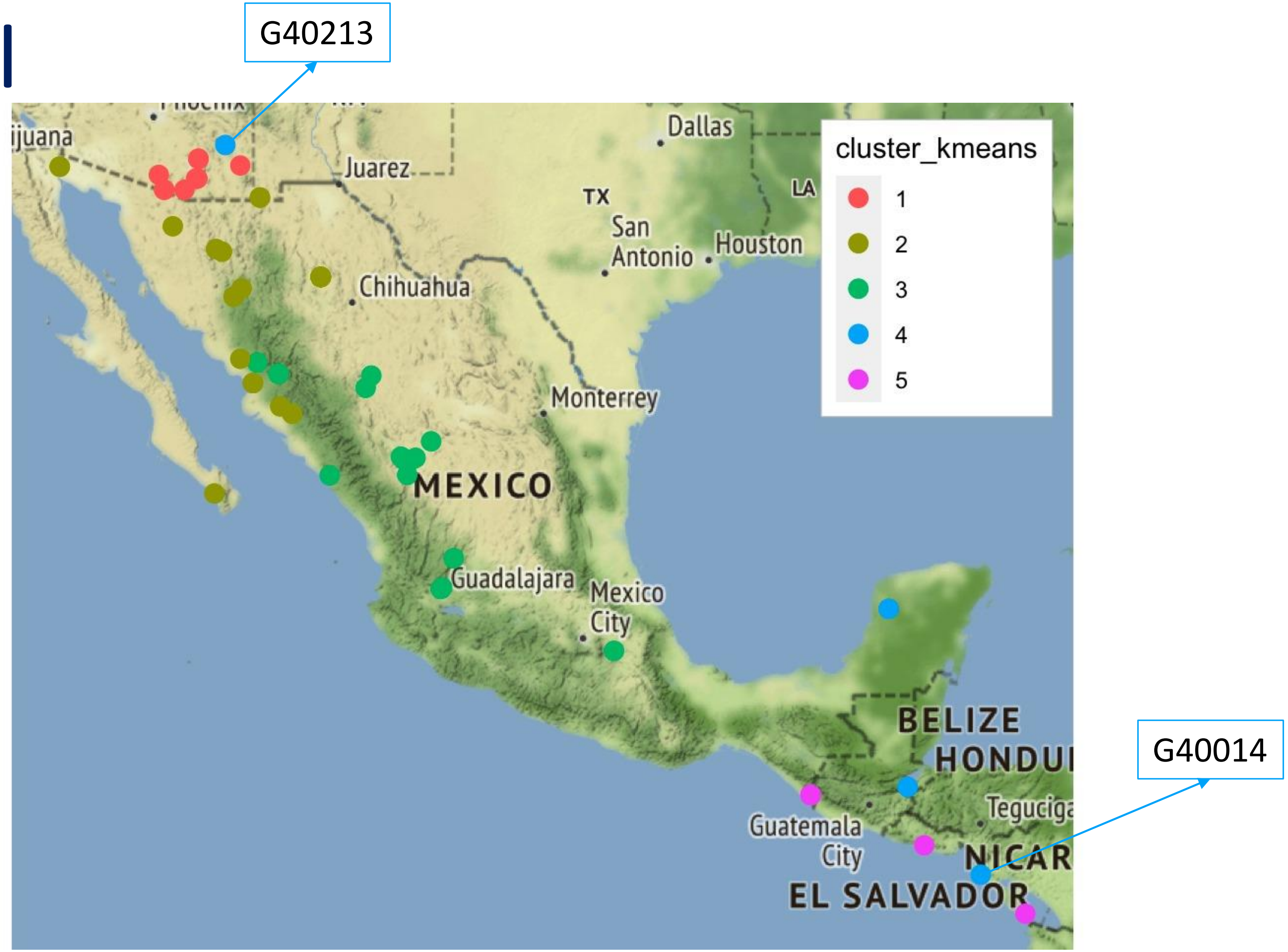
Plant material

- ✓60 Accessions

- ✓23 Cultivated
- ✓3 Weedy
- ✓34 Wild

- ✓43 var. acutifolius
- ✓17 var. tenuifolius

- ✓41 Mexico
- ✓15 United states
- ✓2 Nicaragua
- ✓1 Guatemala
- ✓1 El Salvador



Methodology

53 Classic descriptors: **6 flower, 12 fruit, 14 seed and 20 Vegetative descriptors**

- ✓ Calyx color
- ✓ Corolla color
- ✓ Days to flowering
- ✓ Flowering period

- ✓ Days to first mature pods
- ✓ Mature pod color
- ✓ Ovules per pod
- ✓ Pod curvature
- ✓ Pod dehiscence
- ✓ Pod pubescence
- ✓ Pod width

- ✓ Hilum
- ✓ Number of seeds per pod
- ✓ Seed coat pattern
- ✓ Seed color
- ✓ Seed dimensions
- ✓ Seed shape
- ✓ Seed weight

- ✓ Days to emergence
- ✓ Hypocotyl color
- ✓ Leaflet shape
- ✓ Terminal leaflet length
- ✓ Terminal leaflet width

Morphometric descriptors

MultispeQ: A potential tool for functional trait discovery

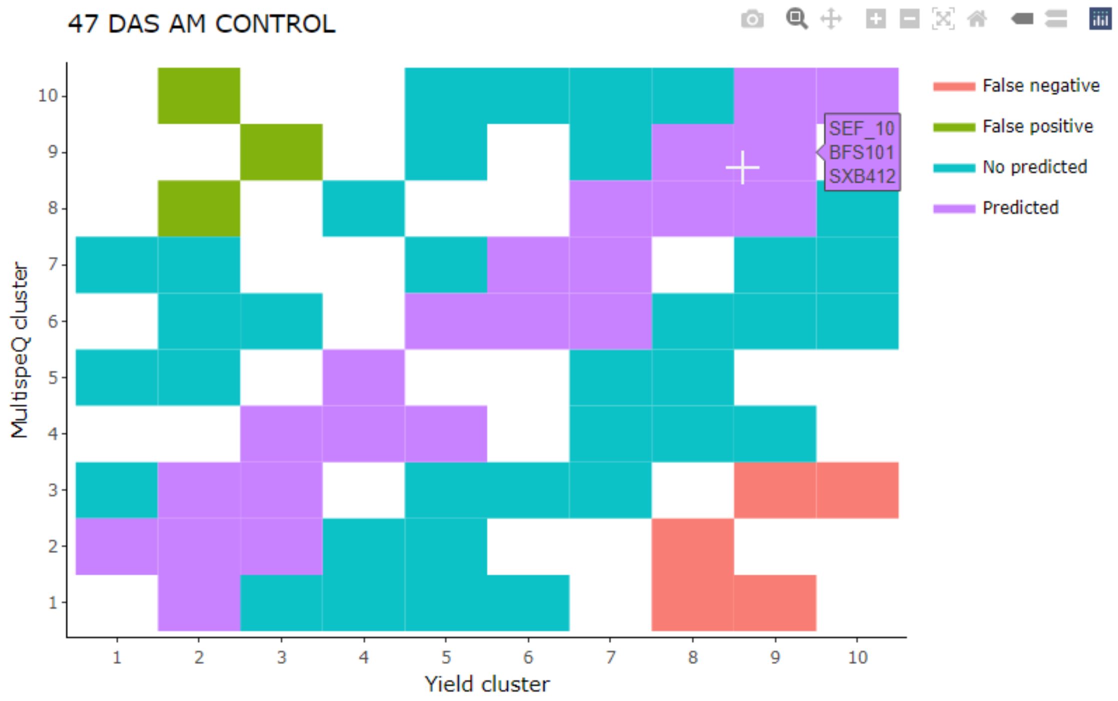
https://www.photosynq.com/multispeq

Trait	PCC
LEF	-0.68
NPQt	-0.43
PhiNPQ	-0.72
PS1.Oxidized.Centers	-0.54
Vh.	-0.46
v_initial_P700	-0.38
P700_DIRK_ampl	-0.39
gH.	-0.39
Phi2	0.64
PhiNO	0.58
FmPrime	0.61
FvP_over_FmP	0.71
phi_index	0.64
PS1.Active.Centers	0.35
PS1.Over.Reduced.Centers	0.27
FoPrime	0.28
Fs	0.39
kP700	0.19
tP700	0.23
Relative.Chlorophyll	0.19
value1	0.33

Handheld device for large scale and non-invasive phenomics evaluations

Leaf-related traits such as photosynthetic efficiency, chlorophyll fluorescence, among others as well as environmental variables.

RankspeQ: Contrast of MSPQ ranks with final yield



Show 10 entries Search:

conf_matrix	Variable	Count
CONTROL 47 DAS AM	Predicted	40
CONTROL 28 DAS AM	Predicted	33
DROUGHT 26 DAS PM	Predicted	33
DROUGHT 48 DAS AM	Predicted	33
DROUGHT 26 DAS AM	Predicted	32
DROUGHT 34 DAS AM	Predicted	30
DROUGHT 48 DAS PM	Predicted	30
DROUGHT 34 DAS PM	Predicted	28
CONTROL 47 DAS PM	Predicted	23
DROUGHT 42 DAS PM	Predicted	23

Showing 1 to 10 of 104 entries Previous 1 2 3 4 5 … 11 Next

4 categories:

- Predicted, the genotypes behaved the same on both MSPQ and yield.
- False negative, the genotypes were the best in yield but low MSPQ score.
- False positive, the genotypes were the best in MSPQ but low yield.
- No predicted. The yield behavior could not be explained by MultispeQ

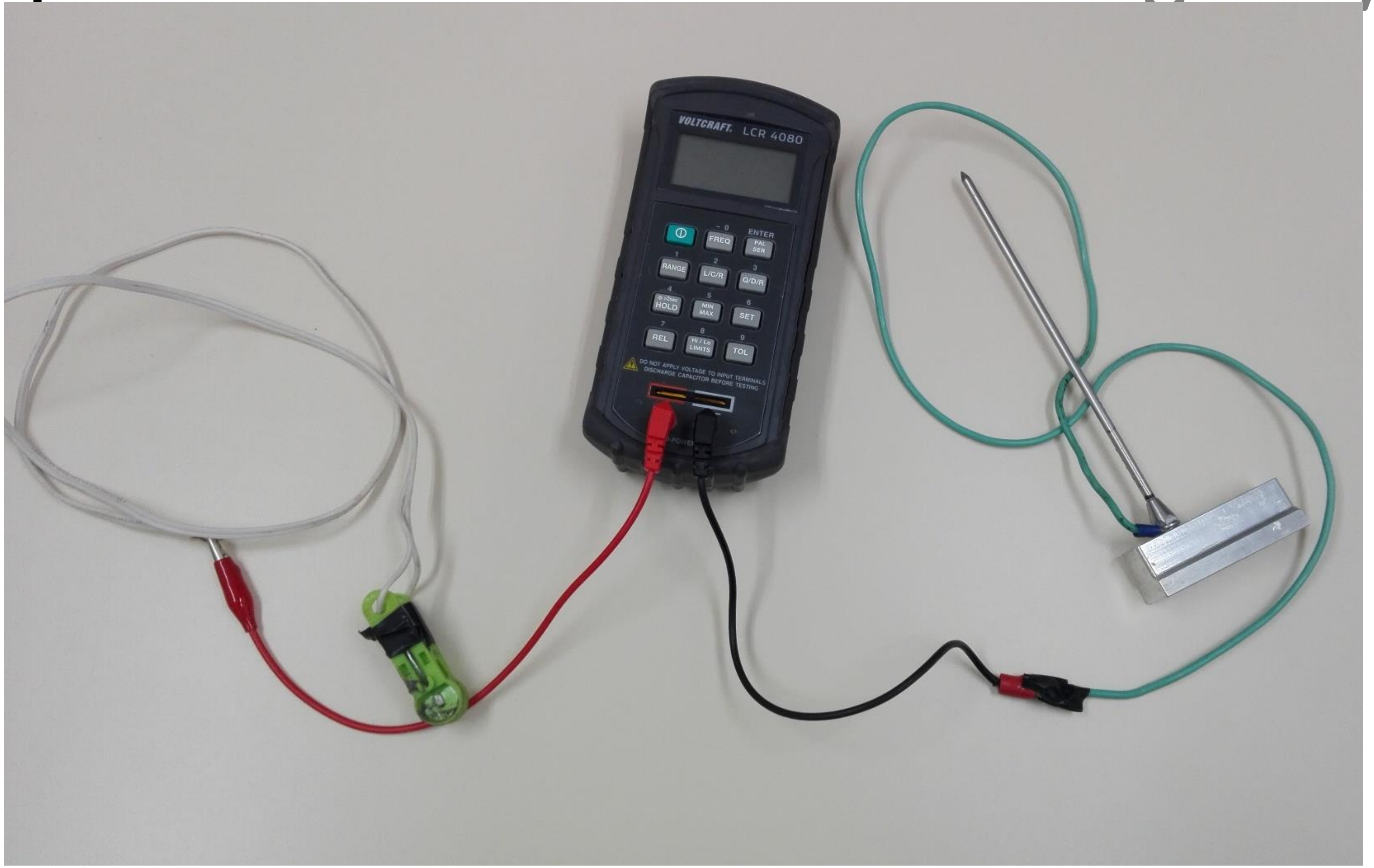
Soto et al. 2023, submitted

How physiology can help to find the best partners in saving precious GB accessions

Figura 3. Injerto de *P. albicarminus* (G40901) sobre *P. dumosus* (G35684).

Figura 4. Injerto de *P. chiapasanus* (G40790) sobre *P. oligospermus* (G40542).

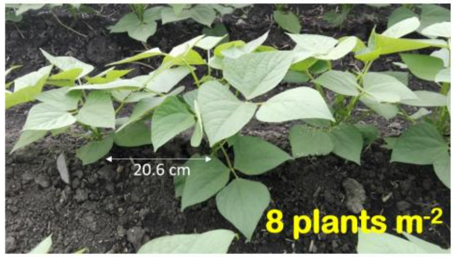
Capacitance relation to root and grain yield



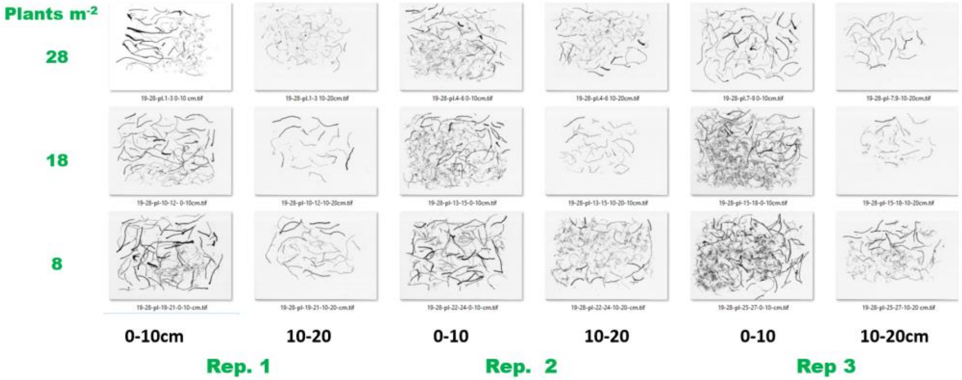
O. CHLOUPEK 1972, Dietrich et al, 2012

Root Capacitance

Field experiment of common bean
(cv. Amadeus)
26 days after planting Jul-24-2019
Lot M2 CIAT Palmira Colombia



Plant density in Amadeus genotype

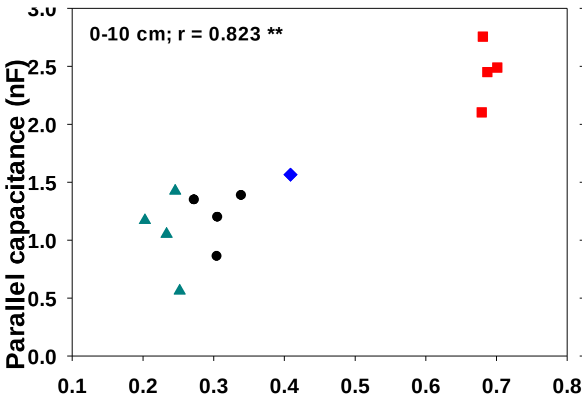


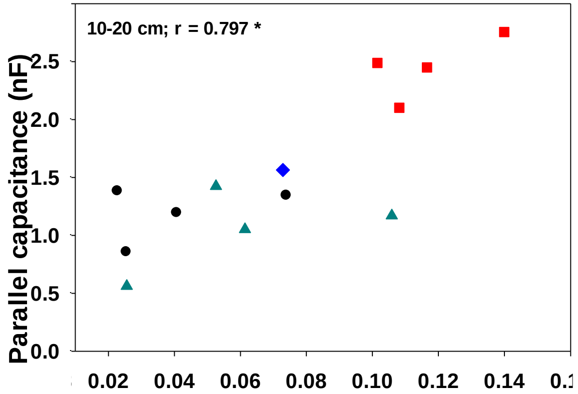
Ricaurte J, Pineda D, Otero M, Urban M , 2019

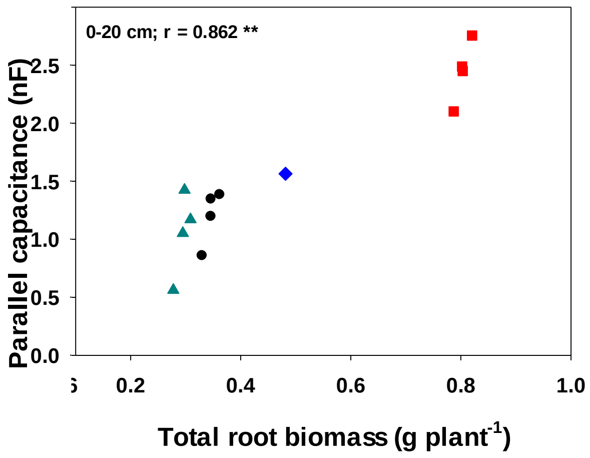
Capacitance correlation to grain yield (g plant^{-1})

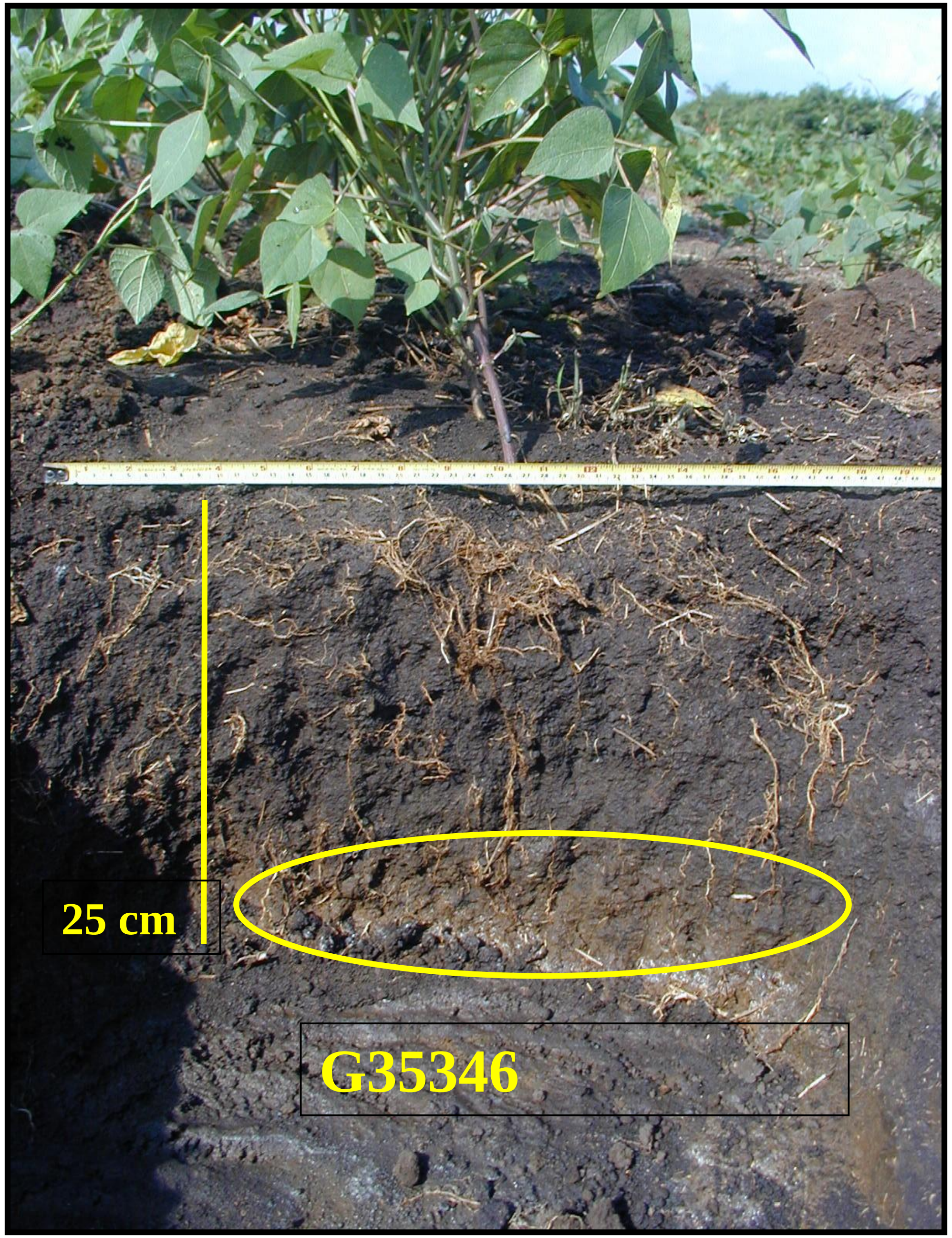
Time after planting (day)	Parallel C (nF)
63	0.460
70	0.255

Red squares, **black circles** and **green triangles** indicate plant densities of **8**, **18** and **29** plants m^{-2}, respectively. Central **blue diamond** the global mean from 9 datapoints.









P. coccineus in an aluminum toxic soil

← 65+ % Al saturation

← 80% Al saturation

ALB 91 x [G35346 x ALB 91]

[SER 16 x (SER 16 x G35346)]

Tolerance to Acid Soil

(pH 4)

Automatic pH correction

Use: Al, macro/micro, root hairs, pH etc.

Qiao, S., Fang, Y., Wu, A. *et al.* Dissecting root trait variability in maize genotypes using the semi-hydroponic phenotyping platform. *Plant Soil* **439,** 75–90 (2019). https://doi.org/10.1007/s11104-018-3803-6

High Al_A774_1,15

High Al_A774_1,3

Low P_A774_1,15

Low P_A774_1,3

Control_A774_1,3

20 Days after planting

High Al_ALB91_1,15

High Al_ALB91_1,3

Low P_ALB91_1,15

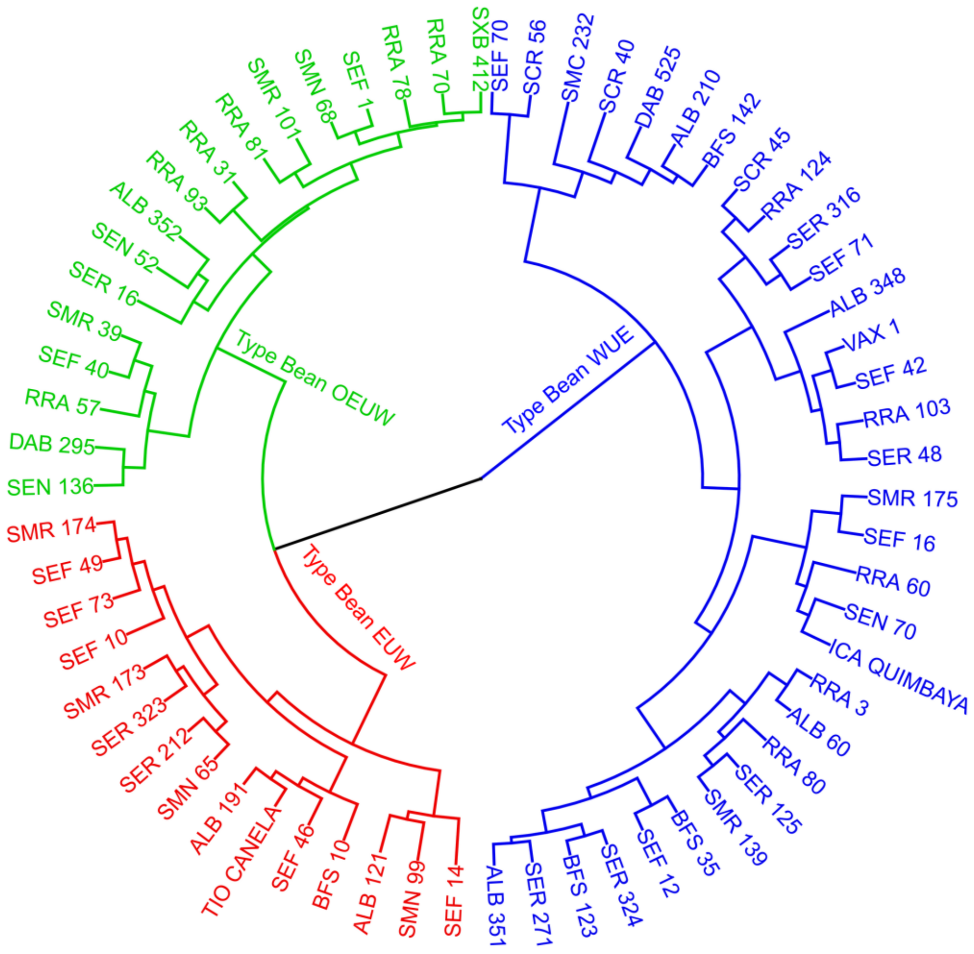
Low P_ALB91_1,3

Control_ALB91_1,3

Water use, leaf cooling and carbon assimilation efficiency of heat resistant common beans evaluated in Western Amazonia

Juan Carlos Suárez[1,2,3*], Milan O. Urban[4], Amara Tatiana Contreras[1,2], Jhon Eduar Noriega [1,2], Chetan Deva[5], Stephen E. Beebe[4], José A. Polanía[6], Fernando Casanoves[7], Idupulapati M. Rao[4]

FPS, 2021, vol 12



A

Measured traits in Western Amazonia

	EUW	WUE	OEUW
A	⇈	⇊	↑
gs	⇈	⇊	↓
E	⇈	↓	=
Ls	⇊	⇈	↑
WUE	⇊	⇈	↓
Ci	↑	↓	↑
LTD	⇊	⇈	=
fPSII	=	=	=
LSP	↑	↓	=
GY	↑	↓	↑
Rd	↑	=	↓
LCP	=	⇊	⇈
Vcmax	↓	=	⇈
Jmax	↓	↑	=

B

Hypothesized bean "heat ideotype" for Western Amazonia

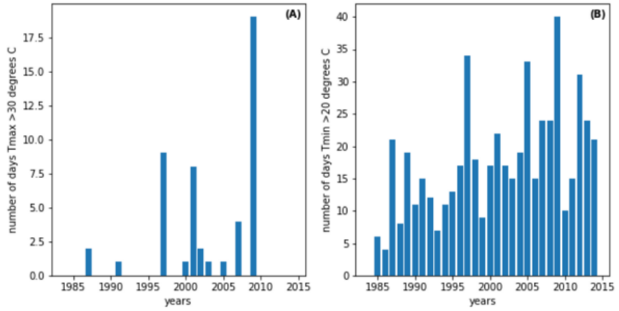
SENSITIVE TO HEAT => CONSERVATIVE

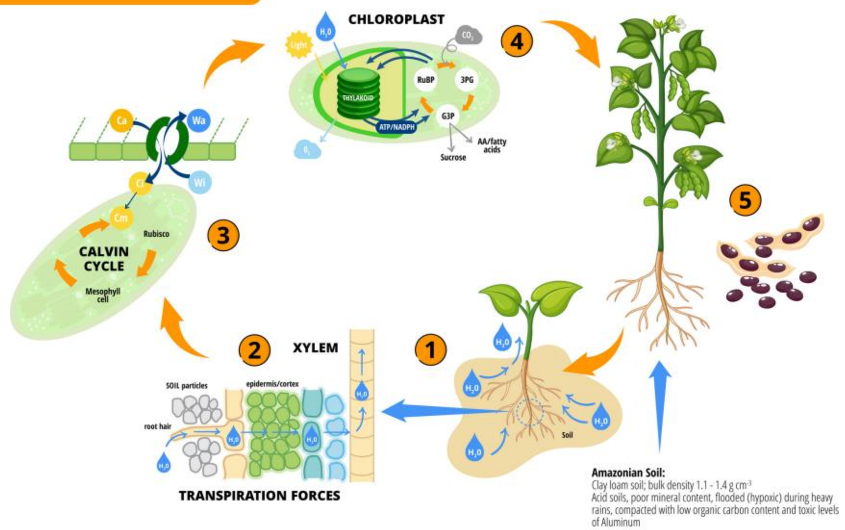
- 1: Roots
  - Decreased root hydraulics
  - Root prolongation rate and root hair density decreased
  - Sensitivity to hypoxia
- 2: Transpiration continuum
  - Low stem biomass
  - "Open" canopy architecture
- 3: Stomatal regulation
  - Stomatal transpiration is highly reduced => WUE increase
  - Stomatal limitation value increased
- 4: Photosynthetic efficiency
  - Ci reduced
  - Enzymatic heat instability => RuBisCO regeneration enhanced
  - RuBisCO carboxylation rate not influenced
- 5: Seed yield
  - Lower grain yield

ADAPTED TO HEAT => OPPORTUNISTIC

- 1: Roots
  - Higher root water uptake
  - Higher resistance to flooding
  - Active role of root hairs (Al^{3+})
- 2: Transpiration continuum
  - Increased root-stem-leaf hydraulic conductivity => cooler leaves
  - Smaller but thicker leaves
  - Anisohydric leaf type
- 3: Stomatal regulation
  - High water loss
  - Enhanced thermal dissipation
- 4: Photosynthetic efficiency
  - High CO_2 assimilation
  - Photosynthetic apparatus fully acclimatized
  - Light compensation and saturation points increased
  - High RuBisCO specifity
- 5: Seed yield
  - Higher grain yield

BEANS IN WESTERN AMAZONIA



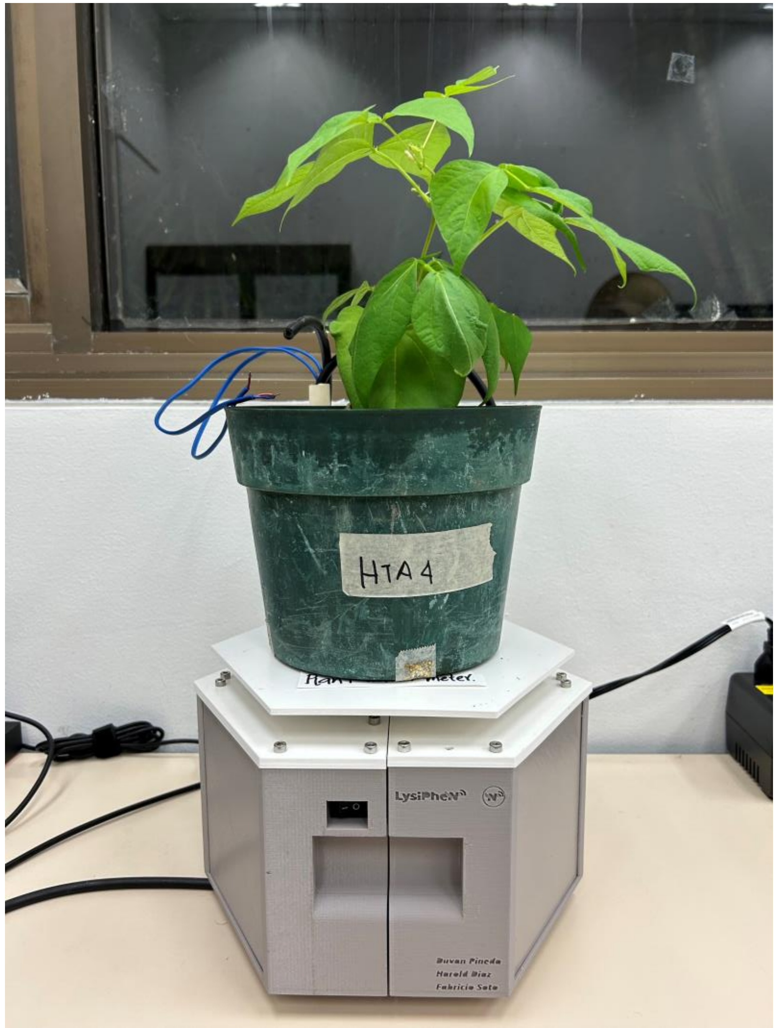


	$+H_2O$	$-H_2O$
High E	X WATER SPENDER - <u>dynamic</u>	X WATER SPENDER, RISK taking
Low E	X TRUE WATER SAVER - <u>conservative</u>	X <u>Typical reaction</u>, ADVANTAGE if…

DIURNAL
Vs
SEASONAL CHANGES

LysiPheN

sophisticated lysimeter at very low cost



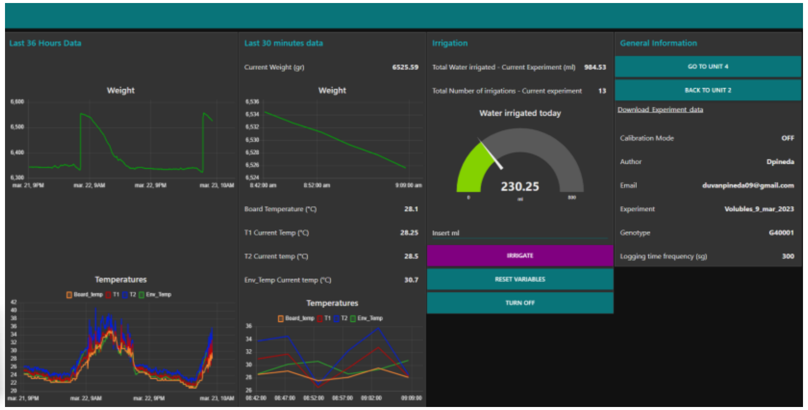
LysipheN is an IoT & automated prototype for **near Real-time High Frequency Crop Phenotyping** available to everyone.

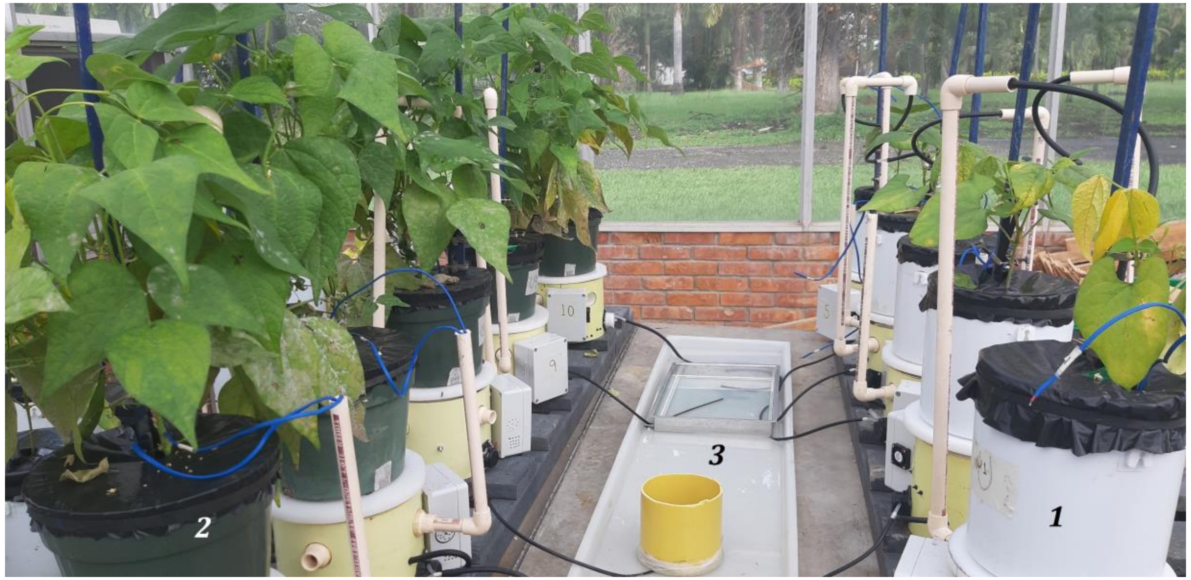
LysiPheN

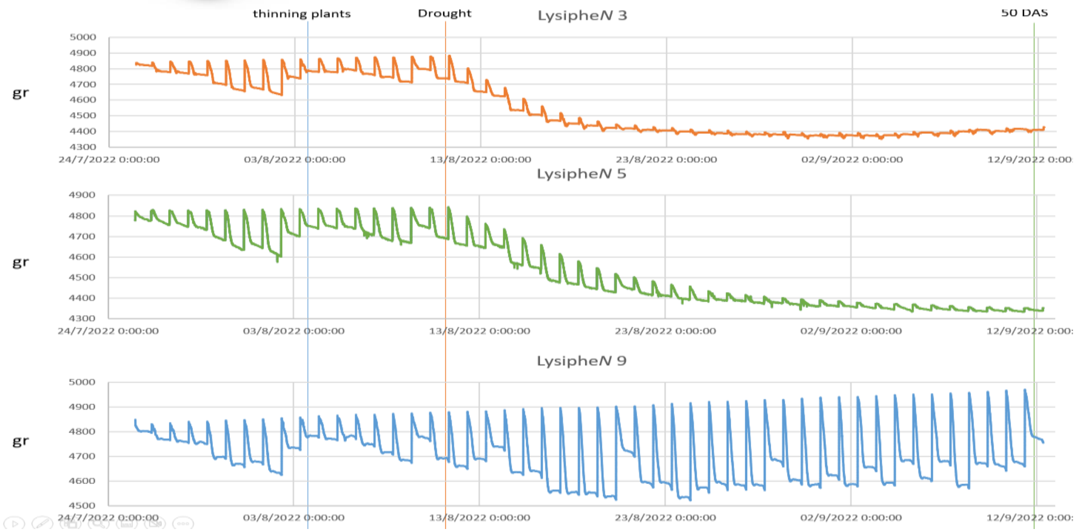
More than 12k data per prototype in a bean experiment season.

Weight system up to 100kg.

Connected with solar panels or conventional electrical sources in any country (Africa, Central America, etc.).







LysipheN 3: irrigation at 50% of transpired water **LysipheN 5:** irrigation at 75% of transpired water **LysipheN 9:** irrigation at 100% of transpired water

Interspecific hybridizations resilient to high temperature increases

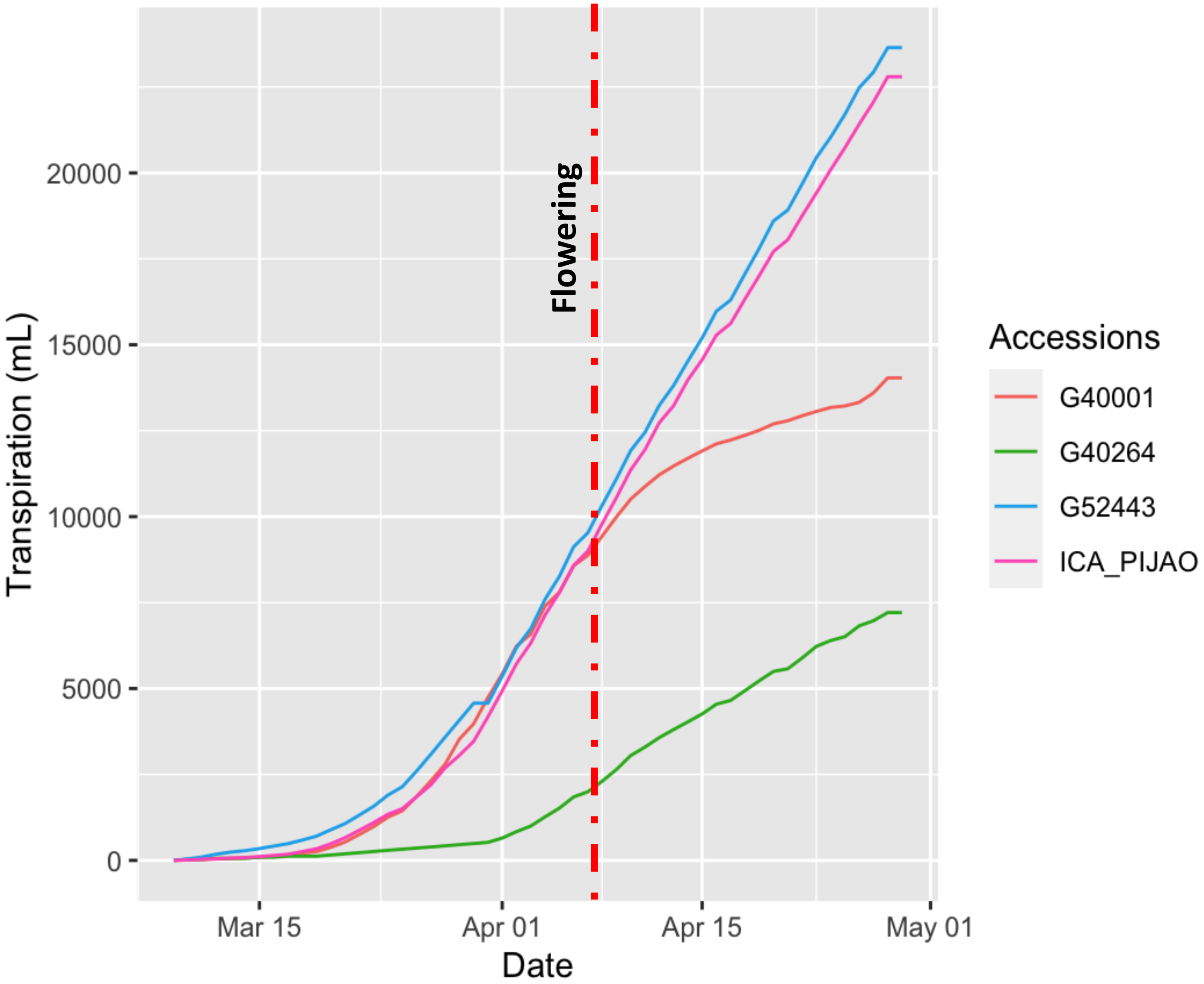
***P. acutifolius* G40001**

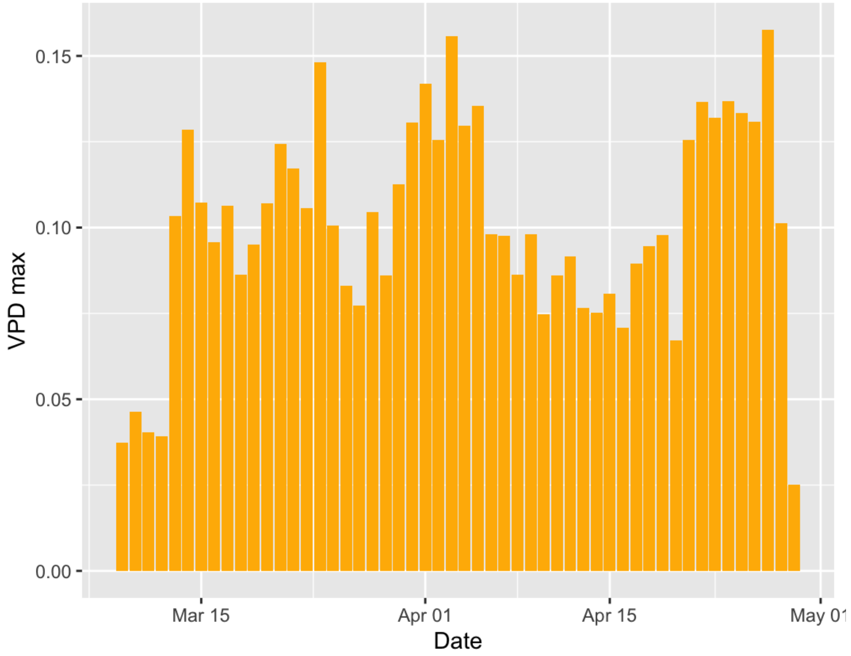
***P. parvifolius* G40264**

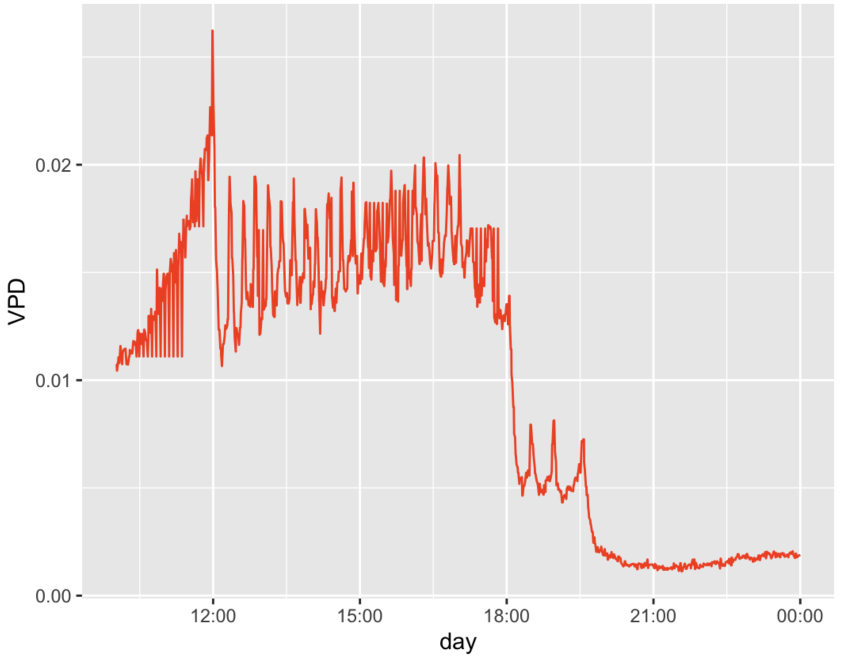
***P. vulgaris* Ica Pijao**

***Hybrid* G52443**

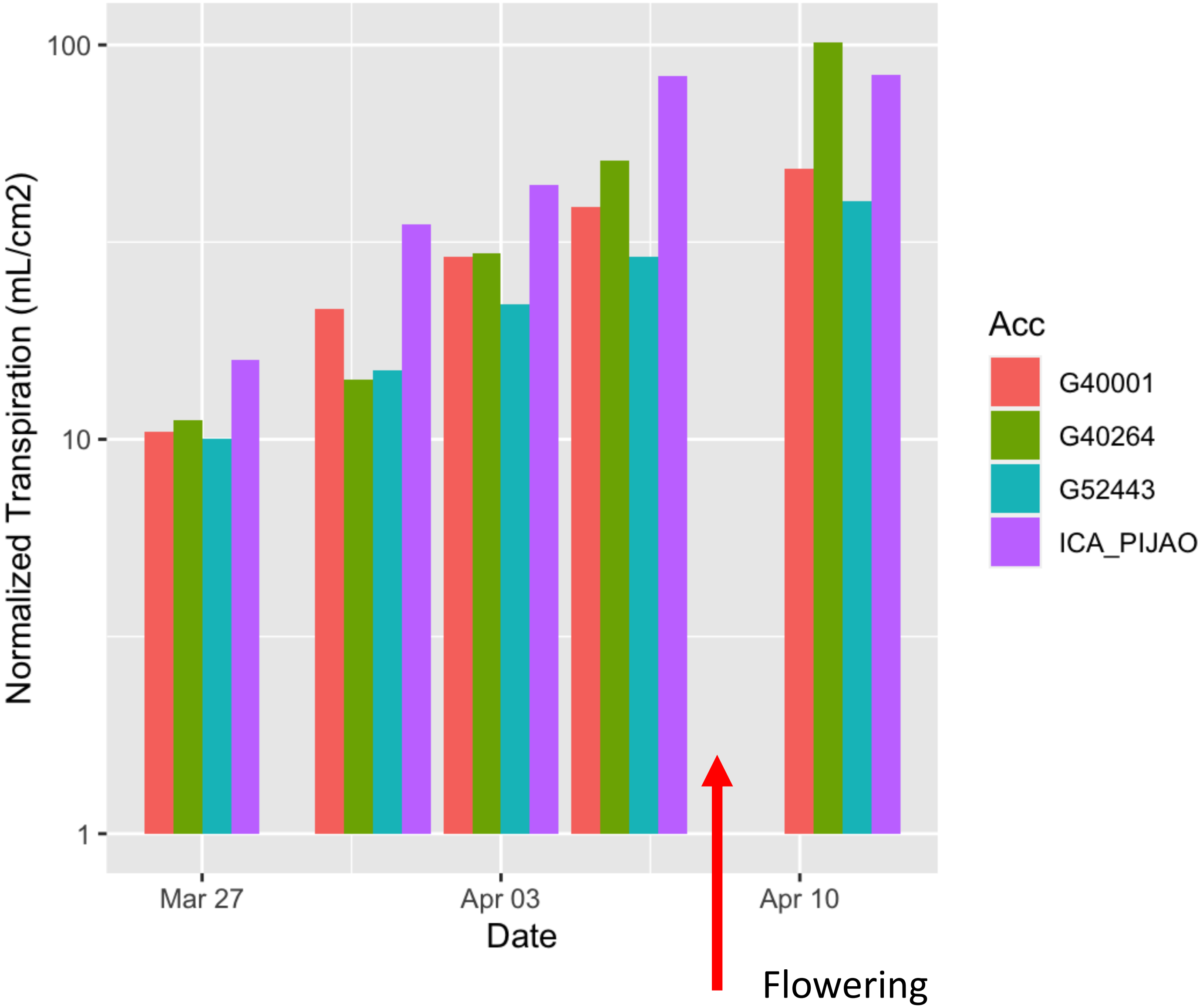
Transpiration and vapor pressure déficit variations

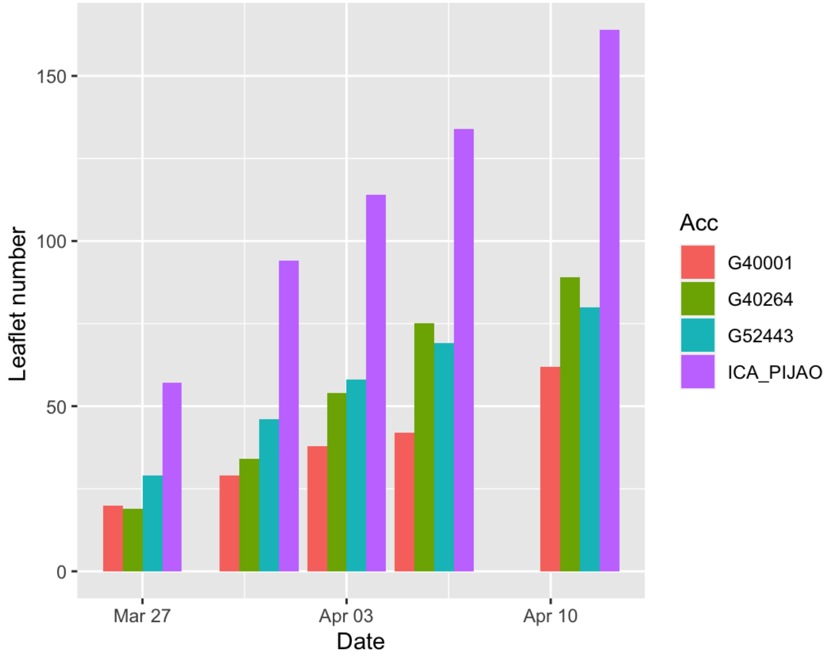






Relationship of transpiration and leaf area in parental and its interspecific hybrids





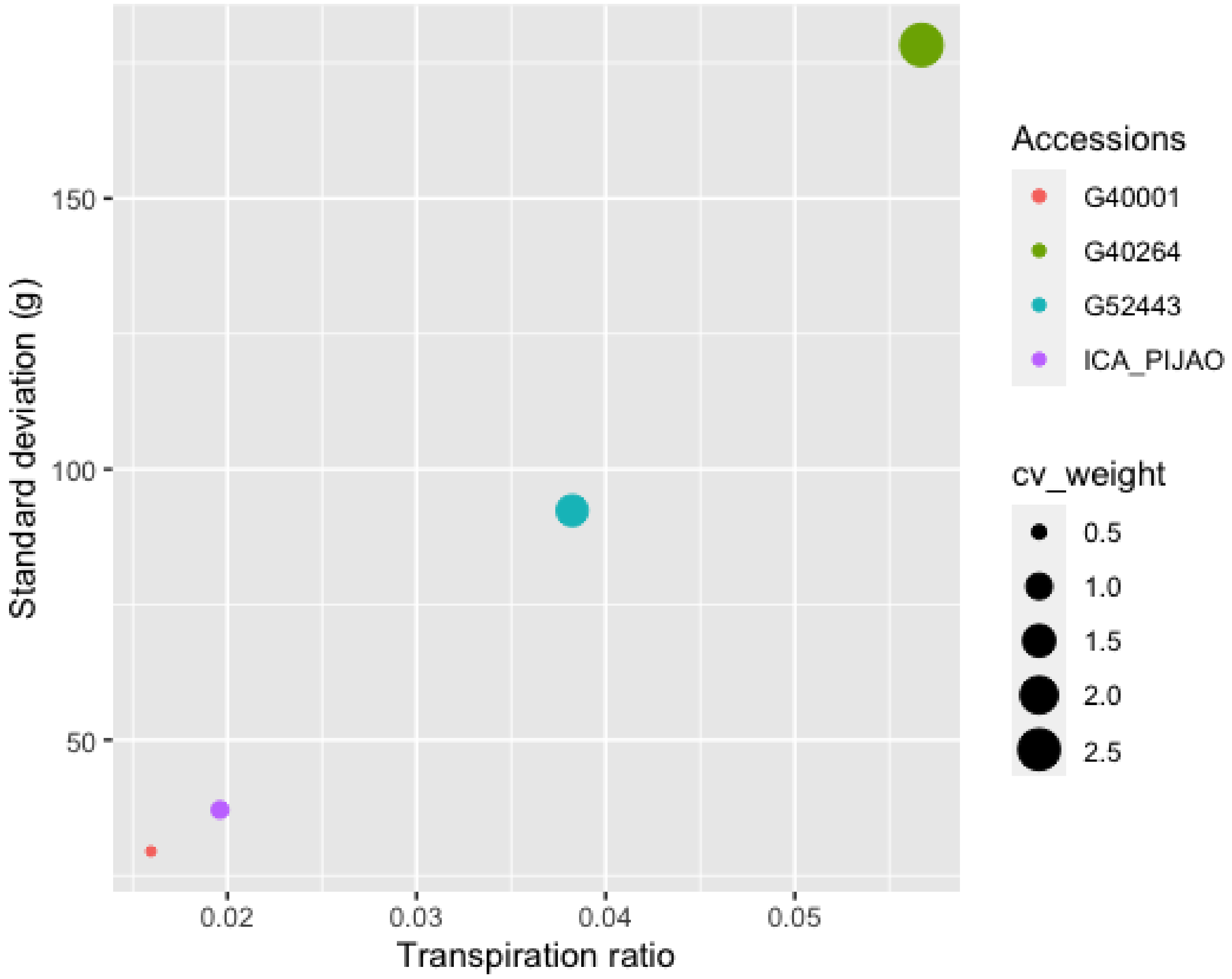
$$\textbf{\textit{Leaf area cm2}} = CLL + CLW + (CLL * CLL) + (CLW * CLW)$$

CLL = Central leaflet length

CLW = Central leaflet width

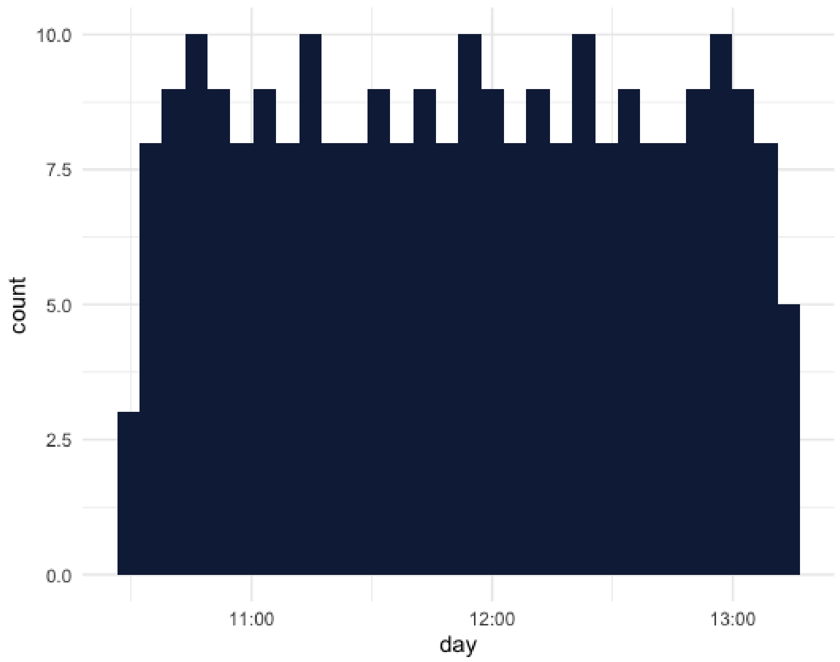
Salazar et al (2022)

Transpiration at maximum VPD of parental and interspecific hybrids during flowering



Transpiration ratio

$$\frac{maximum_{pot\_weight} - minimum_{pot\_weight}}{maximum_{pot\_weight}}$$



Maximun VPD

Effect of increase of 4°C referring Palmira ambient temperature on embryo formation

G40141	INB 604	BFS 81	SER 16	IJR
P. acutifolius	*P. vulgaris x P. acutifolius*	*P. Vulgaris - Meso*	*P. Vulgaris - Meso*	*P. Vulgaris – Andean*
Final seed: 5	*Final seed: 4*	*Final seed: 3*	*Final seed: 3*	*Final seed: 2*

Increase of 4°C = 24°C minimum temperature

Lines with genes from acutifolius / parvifolius
9 seeds / pod

Resume

- THERE IS AN EXCITING UNIVERSE OF WILDS/SPECIES OUT THERE!
- There is no ONE type of drought or heat
- NO silver bullet
- Seed quality
- TIMING
- **Every trait can serve to higher drought resistance** (the right scenario)
  - More roots ≠ higher tolerance! In all soils more important than root biomass is **root spatial distribution and its conductivity (timing of WU and C cost)**

Common bean ideotype for better adaptation to heat

LOW GY potential!

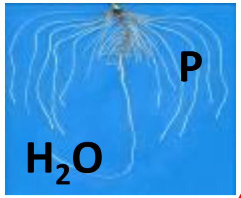
Uniform senescence, Rapid seed filling; higher GY

Tissue damage, photosynthetic limitations

Effective photosynthate translocation from veg to pods; higher transfer into seeds, higher number of seeds/pod, PHI

Trade-offs: dwarfed, too compacted plants

Clever biomass (LAI, senescence, Dynamic LTD, low SLA – thick, small); architecture; more flowers (F2); termal dissipation, pollen+stigma



Higher MUE (Rhizobia, AMF…)

+ redistribution

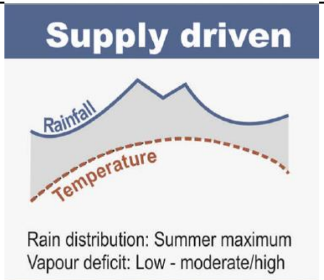
Plant water deficit to evaporative demands

Plastic Root System – conductivity, root hairs, Dynamic to VPD, sensitive to phenology

Suggestions based on published/own information

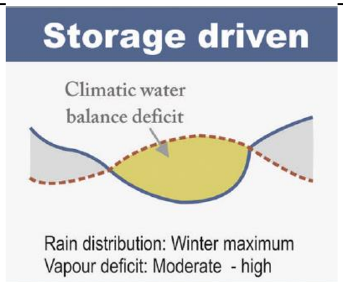
Supply driven (intermittent)

- WATER SPENDING = DYNAMIC
- Efficient + optimized root WU (thick diameters)
- Shallow root system
- ROOT/STEM conductivity (AQP)
- Non limited GS, high T
- EUW (max soil moisture for Tr)
- Less sensitive Pn + growth
- No ovule/seed abortion
- Mycorrhiza + Rhizobia
- Use high-yield potential cvs.



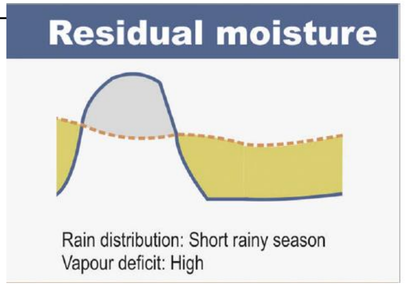
Storage driven (terminal)

- DROUGHT ESCAPE
- Synchrony of fast growth to meet water supplies = develop. plasticity
- Early vigor/flowering/maturity
- Earlier high EUW, later high WUE
- Quick stomatal responses to high VPD
- Slow/sensitive leaf growth
- Deep rooting + conductivity in deep soils
- Root + Stem reserves (N_2 in leaves)
- Check nutrient availability in depth
- Stable + very efficient photosynthetic apparatus = STAYGREEN, root comp.
- Use early materials with high PHI and already high seed nutritional profile



Residual moisture (dry, post-monsoon)

- WATER SAVING+ESCAPE = CONSERVATIVE
- Red GS, red T, high WUE (=low biomass)
- Synchrony of flowering/early pod filling
- Transpiration to VPD break
- Deep rooting in deep soils (fine roots)
- Plant architecture + pod distribution
- Slow/sensitive biomass growth (R/S)
- Source to sink remobilization
- Sensitive grain abortion
- Cuticle, wax, trichomes, rapid leaf movements
- OA, LEA proteins, solutes, enzymes
- Delayed senescence + recovery growth?
- Check nutrient availability in depth



The resume of **Physiological Breeding,** with special focus on using wilds/new species

What is needed

- Multi-location testing
- Hypothesis-driven physiological breeding
- New models of crop processes and on new ideotypes
- HTP
- New allelic diversity into existing genepools

Challenges

- LORAwan, min design, Mr.Bean
- Complex traits in realistic conditions
- Lacking basic knowledge, new generation of models
- Quantitative vs Qualitative + accept digital descriptors
- Only few are interested to risk

Future perspectives/collaboration: where alleles variability is important

- **PAR:** Bean leaves as a vegetable
  - minerals
  - Type 2 - Diabetes – antihyperglycemic agents (Carb Blocker)
- **PAR:** Beans as a fodder
- **PAR:** Perennial beans or new species
- Q: nunas, cooking time, biofortification, flatulence!
- T: Bean-(Rice) rotation
- T: Rhizobia (TE, drought)
- T: Mycorrhiza; GPB
- H: Leaf Variegation, CNGlcs, trypsin,