

PHENOMICS: CLOSING THE GENOTYPE-PHENOTYPE GAP

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Robert Louis Stevenson, 1881:

“.....for to travel hopefully is a better thing than to arrive, and the true success is to labour.”

What is Phenomics?

Phenomics is the equivalent analysis of **phenotypes** as **genomics** is to **genotype**.

Just as we also now have :

Transcriptomics

Metabolomics

Proteomics

Etc

The term “omics” refers to the comprehensive analysis of biological systems

Phenomics

One definition is:

Phenomics is: “-....acquisition of high-dimensional phenotypic data on an organism-wide scale”

Nature Reviews Genetics 11, 855- 866 (December 2010)

Phenomics

- ▣ The need to **assess/evaluate phenotypes** has long been recognised
- ▣ Genetics is a modern science because it was only in about 1900 what was displayed as the phenotype was able to be associated with its underlying determination in the genes **i.e. Mendel**
- ▣ So relating the phenotype of plants is key to the identification and characterisation of genes underlying important traits

Phenomics

A single gene with just two alleles (A–a)

Parents AA x aa
 ▽

F₁ Aa
 SELF ▽

F₂ AA 2Aa aa

So we have 3 genotypes and either 2 or 3 phenotypes
(depending on presence of dominance)
that we can identify and link genotype to phenotype

Phenomics

Two loci both affecting the same character say A-a and B-b

Parents	AAbb	x	aaBB
		▽	

F_1 $AaBb$
 ∇

4 types of gamete

giving 9 possible different genotypes

F₂

AABB, AABb, AaBB, AaBb, AAbb, Aabb, aaBB, aaBb, aabb

Phenomics

No. of genes

No. of possible
F₂ genotypes

1

3

2

9

3

27

4

81

5

243

10

59,000

15

14 Million

21

10.4 Billion

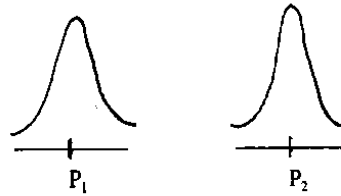
n

3^n

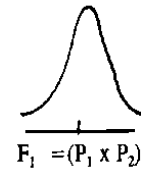
Phenomics

Frequency distributions for the phenotypes of families derived from two true-breeding parents for a character showing quantitative variation

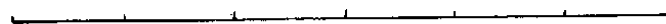
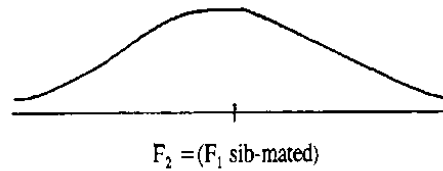
No genetic
segregation
only environmental
variation



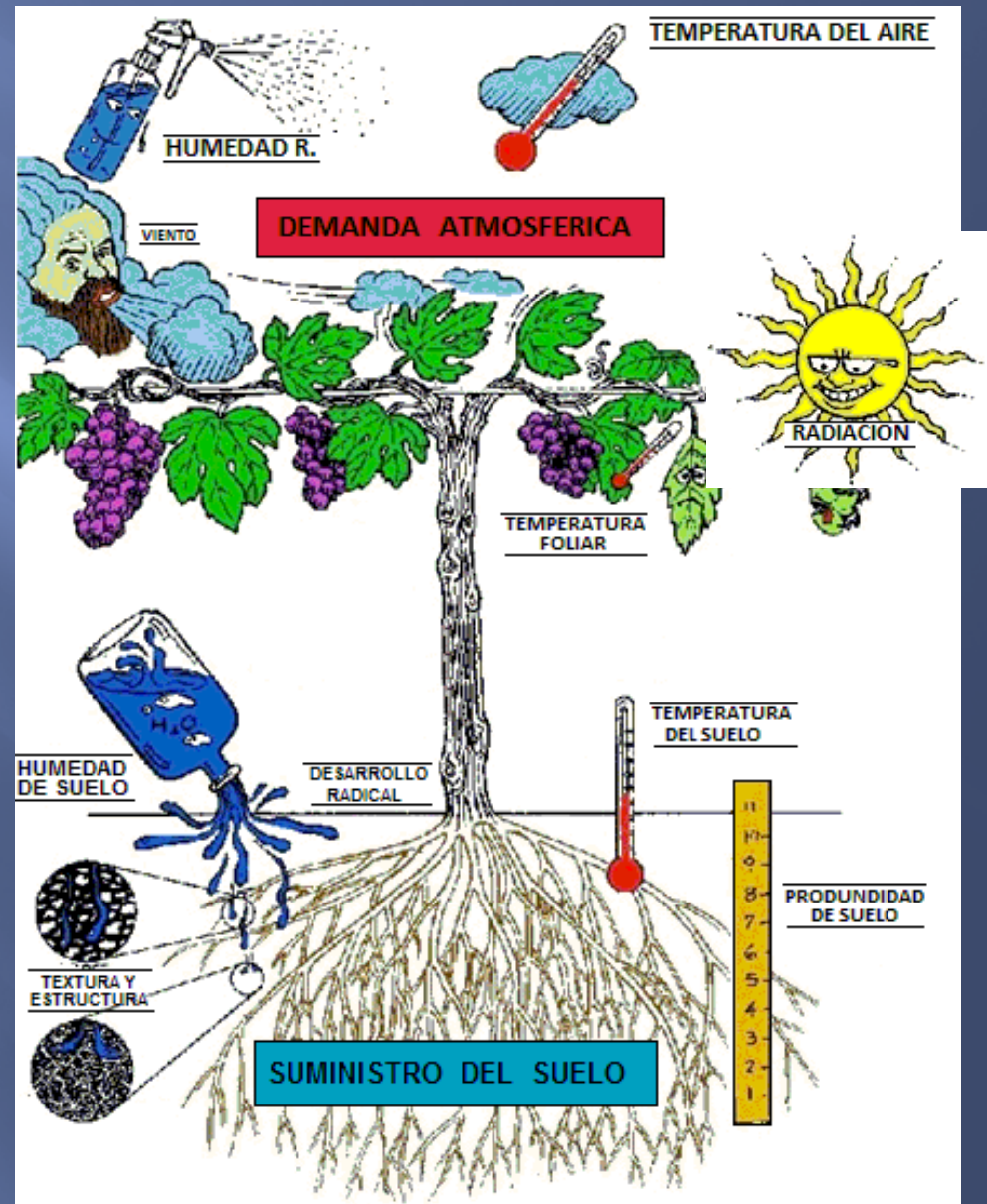
No genetic
segregation
only environmental
variation



Genetic
segregation
as well as environmental
variation

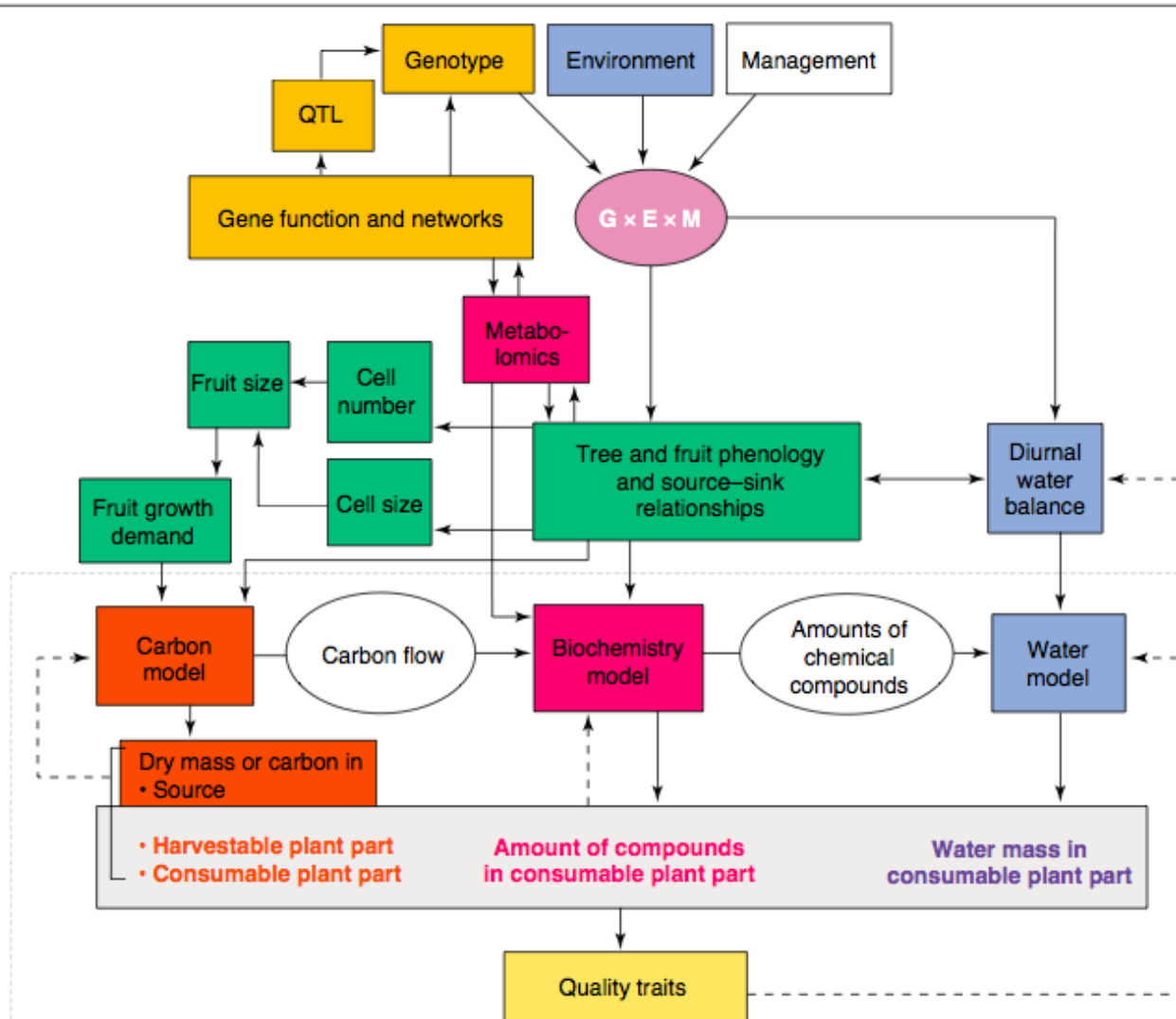


Environmental effects – including climate



But we need add:

- ▣ Other organisms
 - Pests
 - Diseases
 - Neighbours
 - Weeds
- ▣ Management practices
 - Fertilisers
 - Spacing
 - Pruning
- ▣ etc



Phenomics

Elucidating the relationship between plant genotypes and the resultant phenotypes in complex, non-constant environments is one of the foremost challenges in plant biology. (NRC, 2008).

Genotype to Phenotype (iPG2P)

- ▣ So bottlenecks currently exists, relating:
 - **phenotypes back to genotypes or genes** - so finding the genes or gene complexes determining the traits we are interested in.
 - **genes into complete phenotypes** and so determining what are the direct and indirect effects of any particular gene or set of genes that we change or want to modify.

Phenomics: the next challenge

David Houle Diddahally R. Govindaraju & Stig Omholt. *Nature Reviews Genetics* **11**, 855-866 (December 2010)

A key goal of biology is to understand phenotypic characteristics,

Phenotypic variation is produced through a complex web of interactions between genotype and environment, and such a 'genotype-phenotype' map is inaccessible without the detailed phenotypic data that allow these interactions to be studied.

Phenomics

.....Despite this need, our ability to characterize phenomes - the full set of phenotypes of an individual - lags behind our ability to characterize genomes.

[illegible]

Phenomics

So we need plant phenomics.

“Borrowing imaging techniques from medicine, phenomics offers plant scientists new windows into the inner workings of living plants

“Phenomics will give plant scientists the tools to unlock the information coded in genomes”

(Mark Tester, Australian Plant Phenomics Facility)

Why in my view is it important to develop more plant phenomics?

- By 2050, there will be 9 billion people in the World
- We need to produce enough food for this increase – estimated 70 per cent more food needed, under more difficult climate conditions.
- Crop yields need to therefore increase by about 2.4% per year while in reality they are increasing by about 1.2%
- Improve crop yields.

In addition:

- Most cultivars of current crop plants have been selected under conditions of high resource input
- However, the pressure is to reduce levels of inputs and therefore it is essential that these inputs are utilized efficiently.
- **And** there is a consensus that environmental constraints for crop production will increase worldwide with GCC

Phenomics

What is the role of phenomics in crop improvement?

Phenomics

- ▣ Phenomics can speed up the evaluation of phenotypes by using automated, high-tech imaging systems and computing power.
- ▣ It can cut evaluation time down from weeks to minutes, or even seconds.

Phenomics

- ▣ So phenomics:
 - Decreases the time for evaluating,
 - Increases the scale on which it can be done,
 - Quantifies characteristics,
 - Increases accuracy

Phenomics

How is this achieved?

Some phenomics techniques are:

- 3D imaging.
- thermal imaging.
- infrared and near-infrared imaging.
- fluorescence imaging.
- magnetic resonance imaging.
- spectral reflectance.

Phenomics

There are two basically different ways of approaching crop plant phenomics

One way is to build very **sophisticated controlled environments** with plants in pots on conveyor belts which are automatically moved passed fixed evaluation equipment (imager/detector).

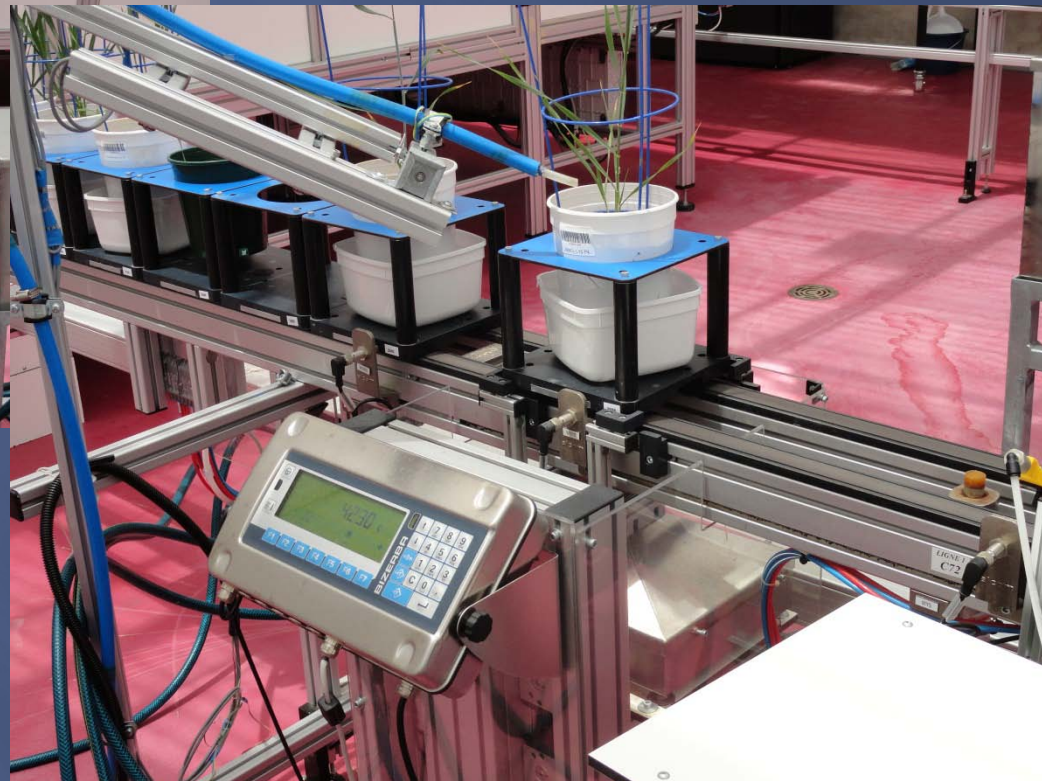
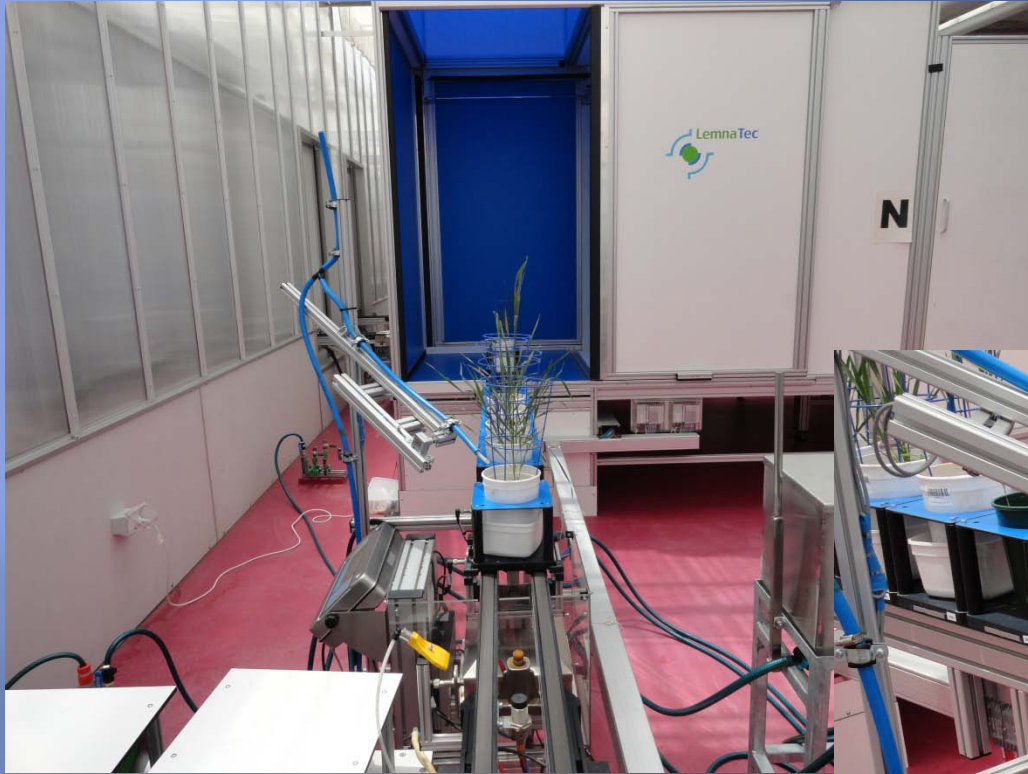
The other is to **use portable equipment** which can be taken to **the field** and screen material in trials grown in a traditional way and with replication and randomisation.

Phenomics

Plant accelerator (Univ. of Adelaide)



Phenomics



Three-dimensional (3D) imaging

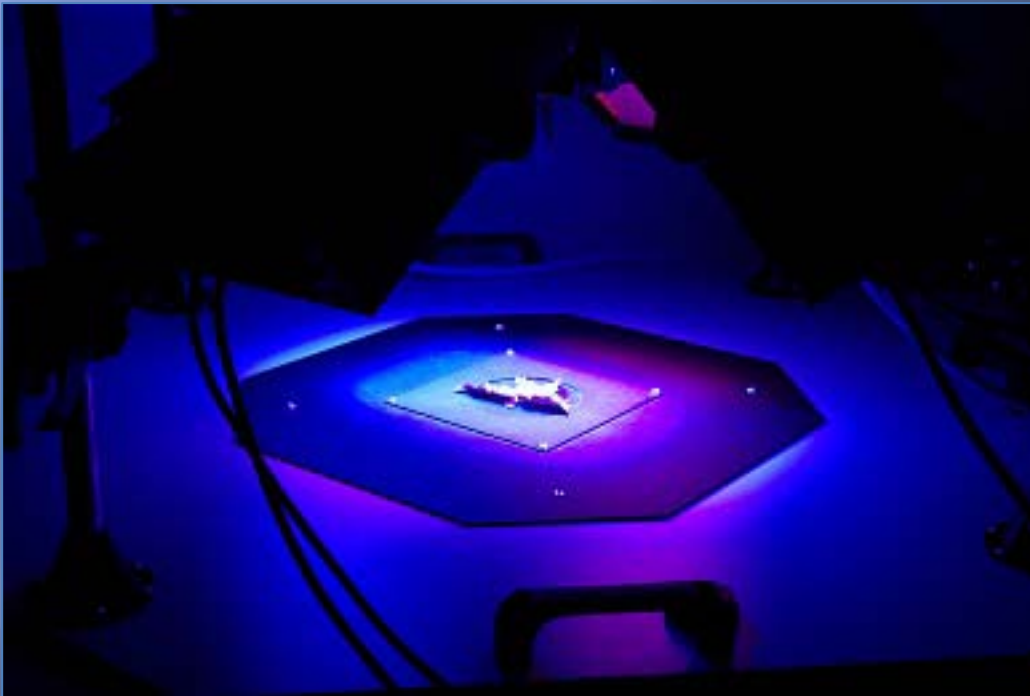
Digital photos of the top and sides of plants are combined into a 3D image.

Measurements that can be taken using a 3D image include:

- shoot mass
- leaf number, shape and angle
- leaf colour
- leaf health.



Fluorescence imaging is used to study plant health and photosynthesis.



Chlorophyll fluorescence to study the effect of different genes or environmental conditions on photosynthesis.

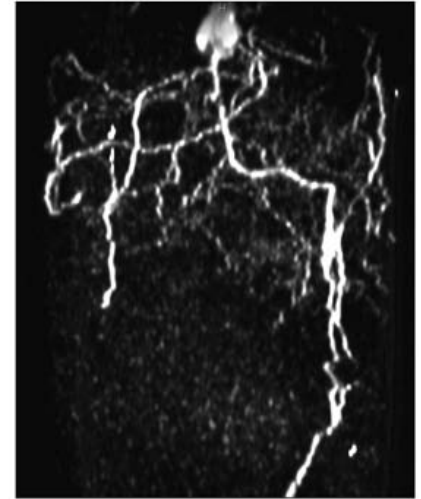
Magnetic resonance imaging (MRI) to study plant roots.

- MRI uses a magnetic field and radio waves to take images of roots in the same way as for imaging body organs in medicine.

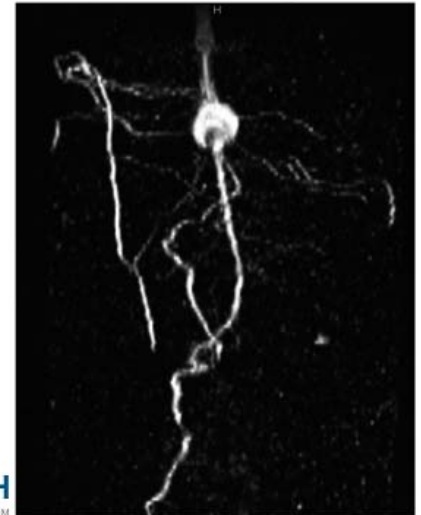
Root
Temperature

MRI

24°C

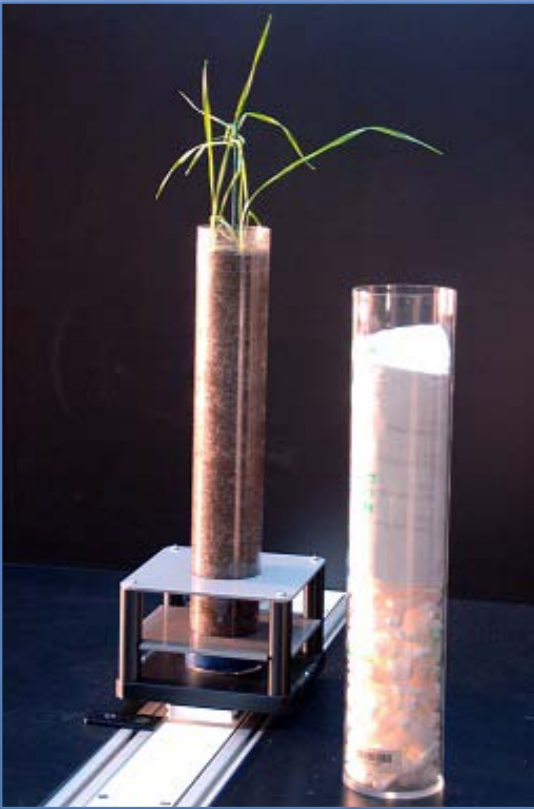


14°C



Near-infrared (NIR) cameras study water content and movement in leaves and soil.

- Light in the NIR region of the spectrum (900–1550 μm)



Plants in clear pots so roots can be photographed while the plant is growing.

Soil NIR measurements are used to calculate:

- how much water the roots remove from the soil
- where and how much water the plant is using.

Phenomics

Use equipment which can be taken to the field and screen material in trials grown in a traditional way and with replication and randomisation.

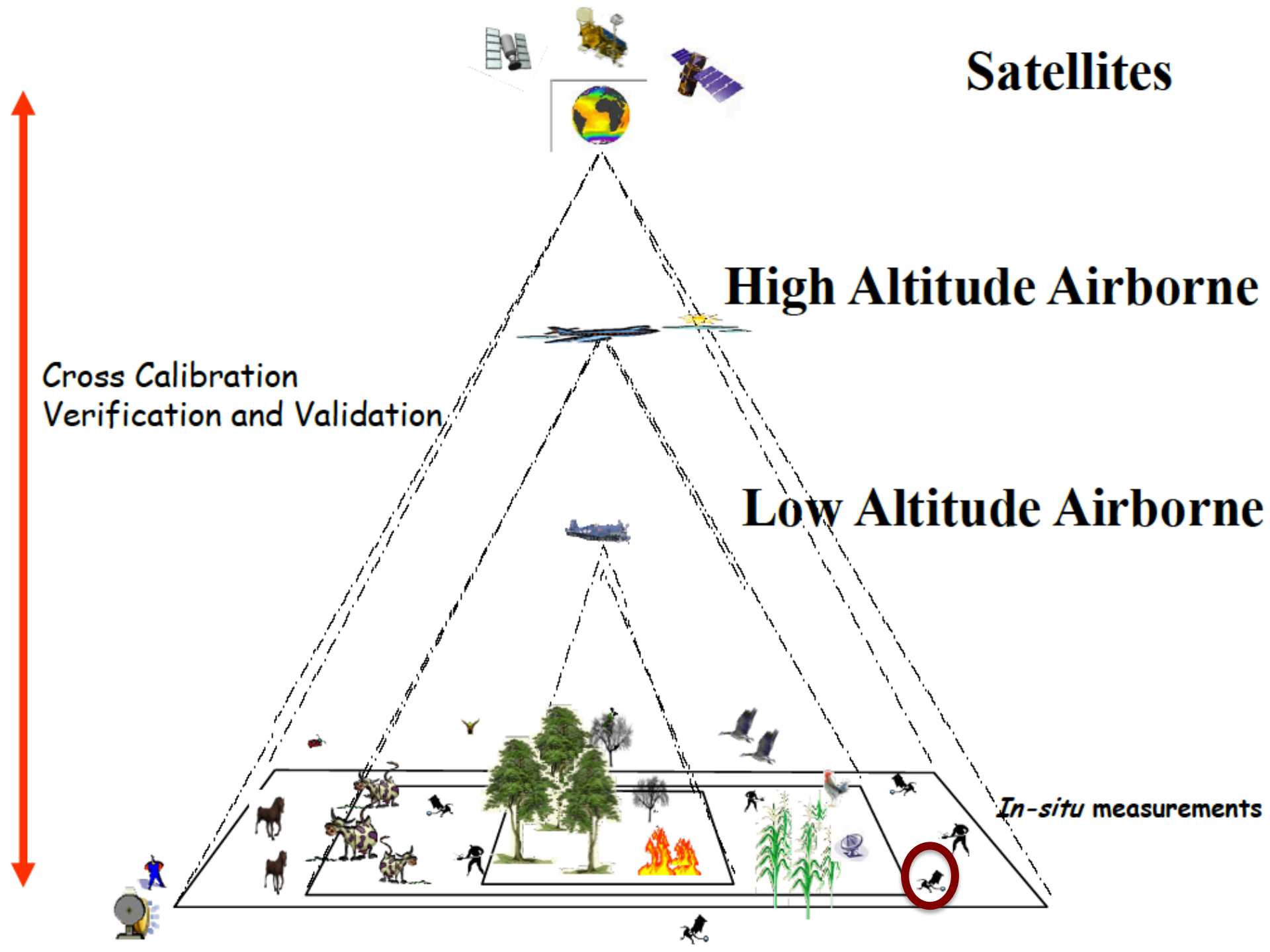
Satellites

High Altitude Airborne

Low Altitude Airborne

Cross Calibration
Verification and Validation

In-situ measurements



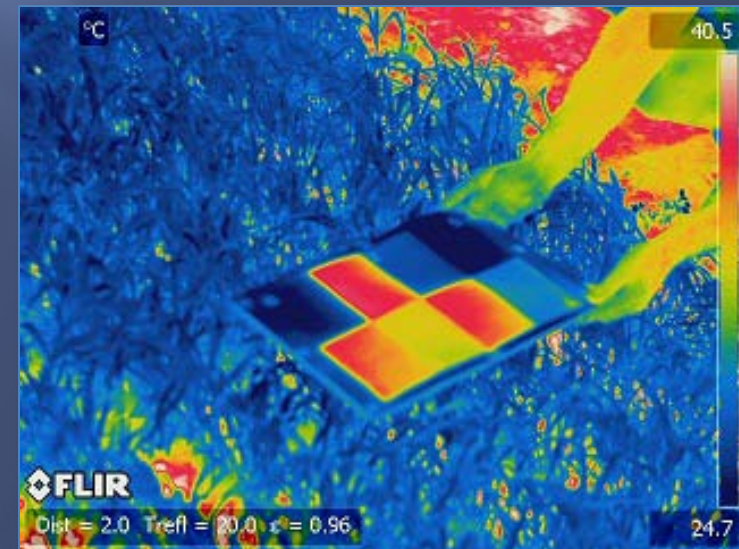
Far infrared (FIR) imaging 1

FIR cameras are used to study temperature.

- ▣ They use light in the FIR region of the spectrum (15–1000 μm).

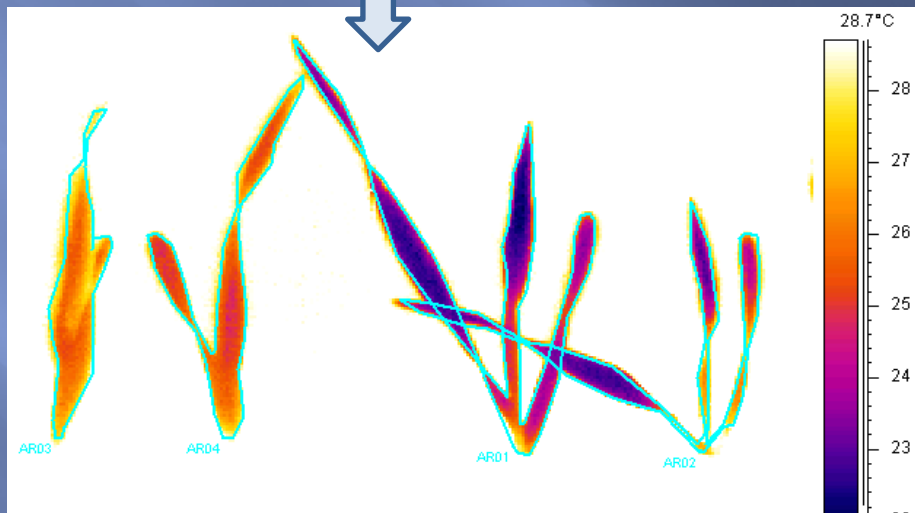
Temperature differences can be used to study:

- ▣ salinity tolerance
- ▣ water usage
- ▣ photosynthetic efficiency.



Far infrared (FIR) imaging 2

Cooler plants have better root systems and take up more water.



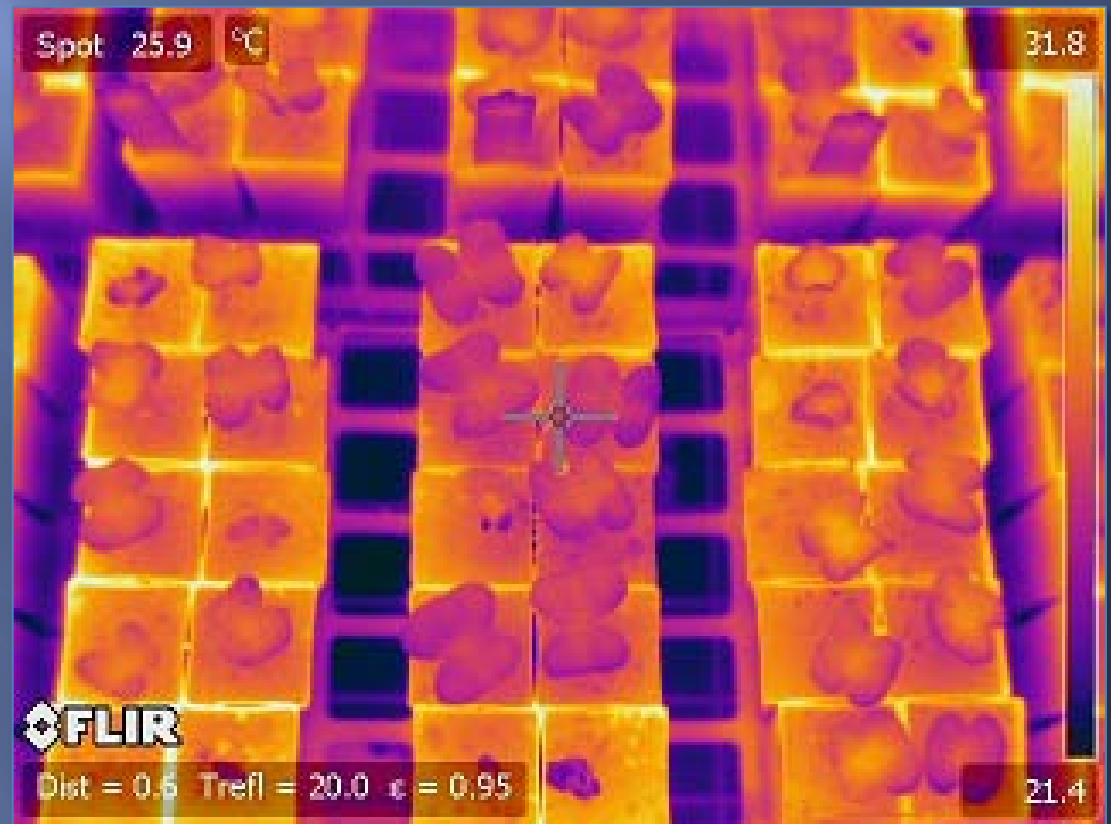
FIR imaging can be used for individual plants or for whole crops.



Spectral reflectance

Spectral reflectance is the fraction of light reflected by a non-transparent surface.

Variation in spectral reflectance when a plant is stressed even before it can be seen by eye.



Field Scanning

World's first Field Scanalyzer at Rothamsted Research

13th July 2015

A world first for automated measuring of crop growth and health in the field was installed for Rothamsted Research in 2015 by LemnaTec GmbH. This is the world's largest and most sophisticated facility built today and will revolutionise the way that crop health and growth are monitored in the field. The development of the facility has been supported by Rothamsted Research and BBSRC.

Phenomics

Some current work
of colleagues and some of my own

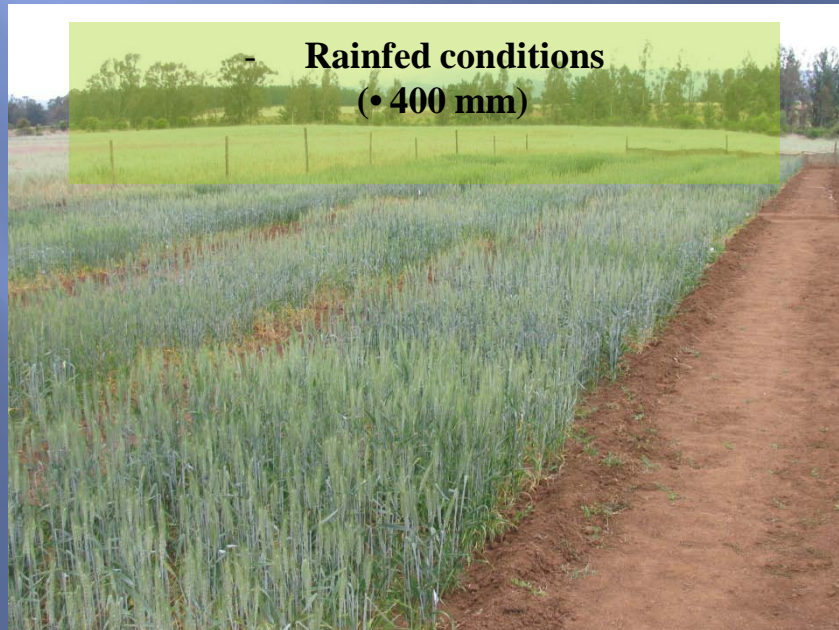
Phenotyping 384 lines of spring wheat 1

Alejandro del Pozo

A set of 384 advanced lines and cultivars from Chile, Uruguay and CIMMYT, with good agronomic characteristics and disease tolerance were evaluated in two Mediterranean environments:

Cauquenes (35°58' S, 72°17' W)

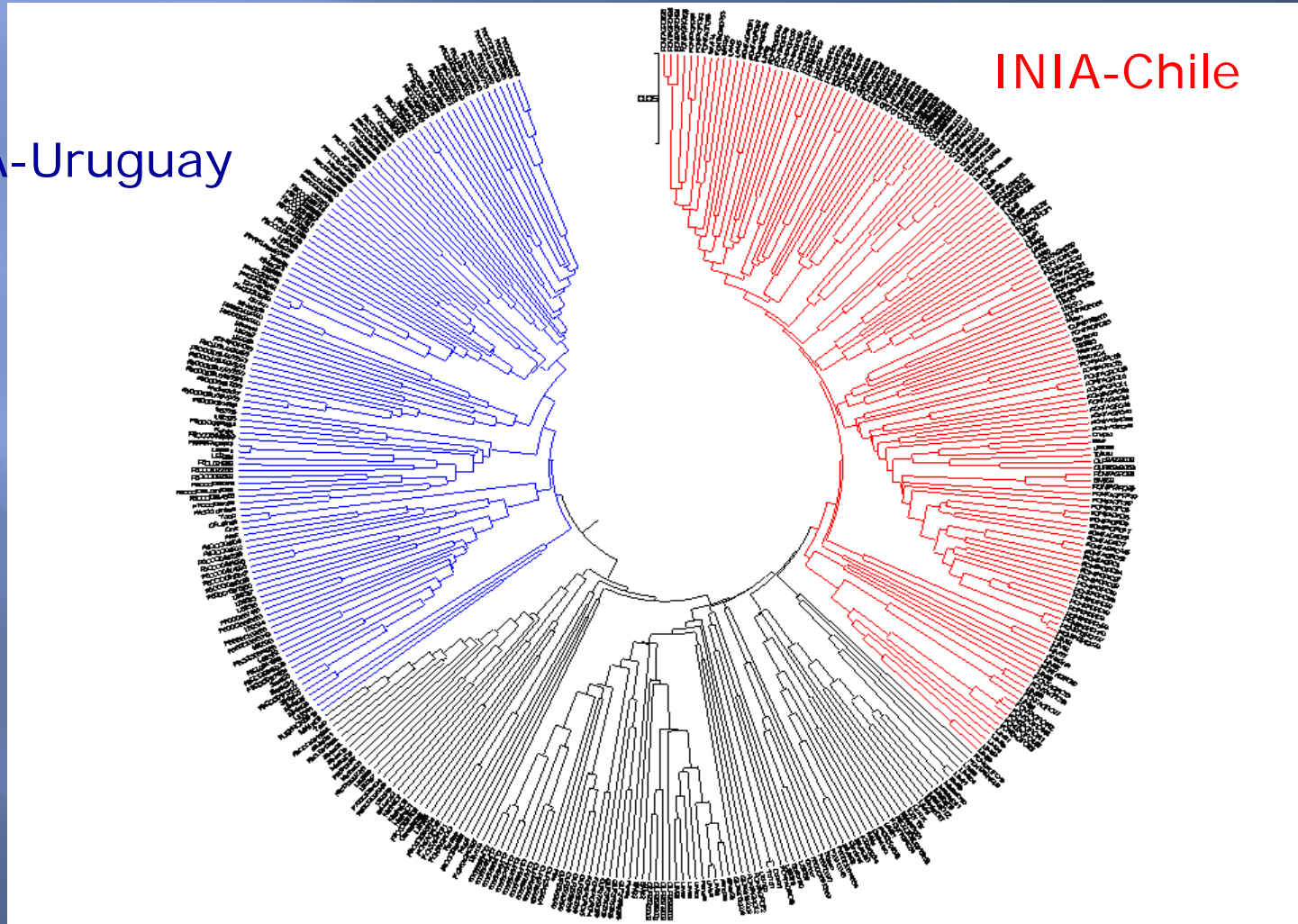
Santa Rosa (36°32' S, 71°55' W)



The 384 genotypes have been genetically characterized using 28,000 SNPs (genotyping by sequence)

INIA-Uruguay

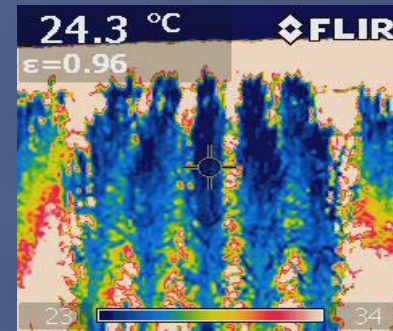
INIA-Chile



Phenotyping 384 lines of spring wheat 2



Multispectral reflectance
(FieldSpec, 350-2500 nm)



Canopy temperature
(Flir i-40)

- **Other physiological traits:**

- Relative Water Content
- PAR interception (ceptometer)
- Stem water soluble CHO
- Carbon discrimination

- **Agronomic traits:**

- Plant height
- N° ears per m²
- N° grain per ear
- Thousand kernel weight
- Grain yield

Testing tolerance to drought and heat stress of blueberry germplasm

- Genotypes of different origin and genetic background are being tested in the greenhouse under 4 different conditions of heat and water level to judge their response to conditions likely to occur in the future under GCC.





Research Paper

Classification of blueberry fruit and leaves based on spectral signatures

Ce Yang ^a, Won Suk Lee ^{a,*}, Jeffrey G. Williamson ^b

^a Agricultural and Biological Engineering Department, University of Florida, Gainesville, FL 32611, USA ^b Horticultural Sciences Department, University of Florida, Gainesville, FL 32611, USA

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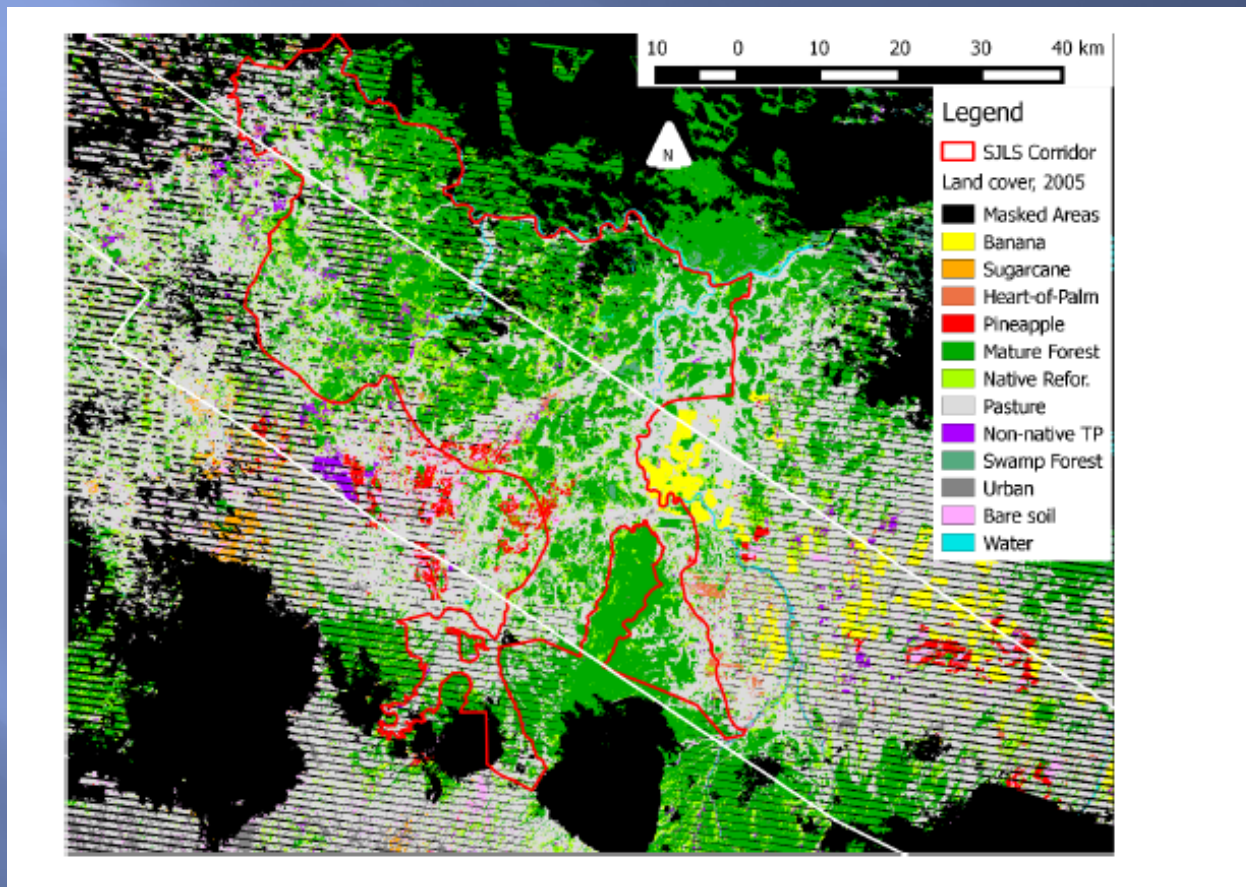
Blueberry spectral analysis can provide necessary wavelengths, for use in multispectral imaging that could be applied in blueberry yield estimation system. Samples of fruit and leaves were obtained from a commercial blueberry field in Waldo, Florida and an experimental field in Citra, Florida, USA in 2011. Samples were also collected in 2010 in Waldo. Seven representative southern highbush varieties were chosen for the experiment. Spectral reflectance was measured in the 200–2500 nm with an increment of 1 nm. Samples were divided into leaf, mature fruit, near-mature fruit, near-young fruit and young fruit. Normalised indices were used as the candidate variables for classification. Each index was composed of the two wavelengths that had the greatest difference in reflectance between two classes. Classification tree, principal component analysis (PCA) and multinomial logistic regression (MNR) were conducted to develop classification models. An MNR model with six wavelengths (233, 551, 554, 691, 699 and 1373 nm) performed the best for the 2011 dataset, with a prediction accuracy of 100% for leaf and mature fruit, 97.8% for young fruit, 97.9% for near-young fruit and 94.6% for near-mature fruit. Four wavelengths (553, 688, 698 and 1373 nm) were used in the classification models of two years' data with four classes (mature fruit, intermediate fruit, young fruit and leaf), and accuracies of 100%, 100%, 99%, and 98.5% were obtained for the classification of leaf, mature fruit, intermediate fruit and young fruit, respectively. An easy-to-use and low cost blueberry fruit detector could thus be developed using multispectral imaging. (

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Mapping tree species



Matthew E. Fagan^{1*}, Ruth S. DeFries², Steven E. Sesnie³, J. Pablo Arroyo-Mora⁴,
Carlomagno Soto⁴, Aditya Singh⁵, Philip A. Townsend⁵ and Robin L. Chazdon⁶

**Mapping Species Composition of Forests and Tree Plantations
in Northeastern Costa Rica with an Integration of
Hyperspectral and Multitemporal Landsat Imagery**

Remote Sens. **2015**, *7*, 5660–5696; doi:10.3390/rs70505660

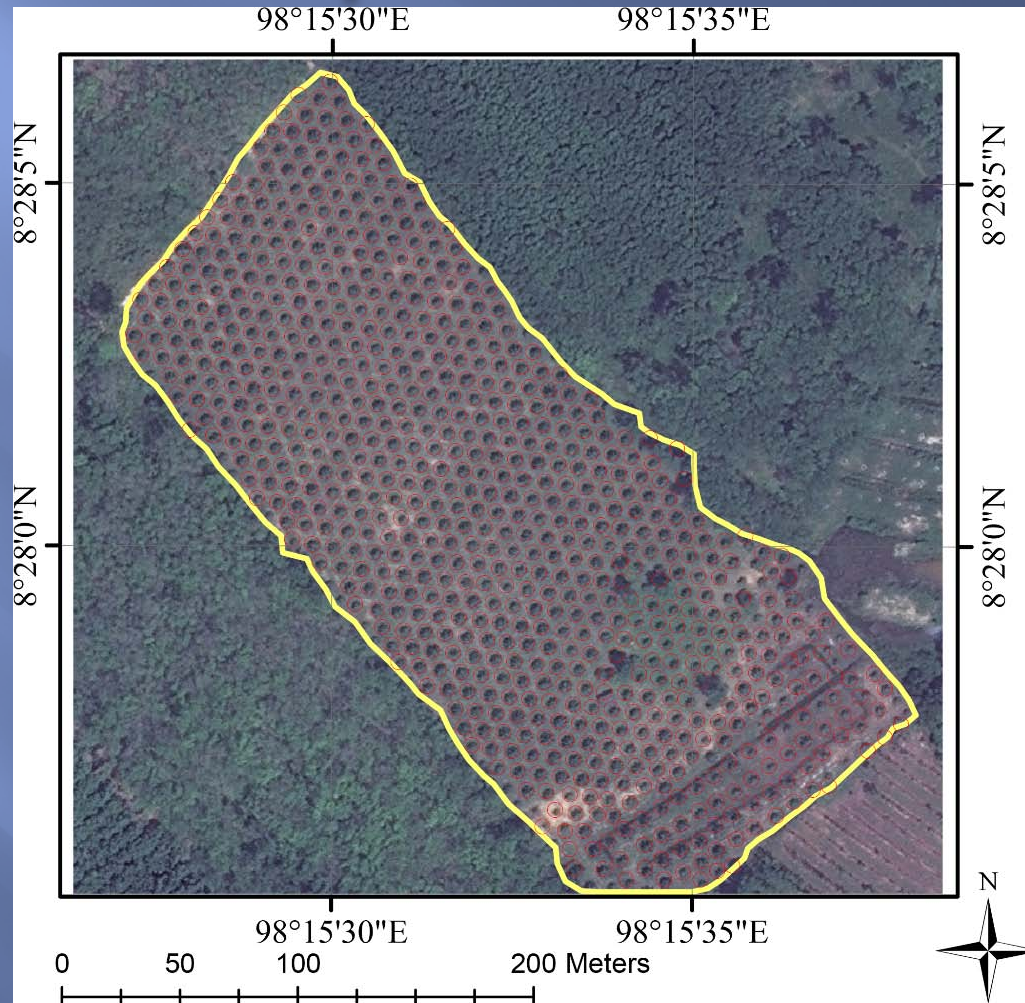
Oil palm



Elaeis guineensis

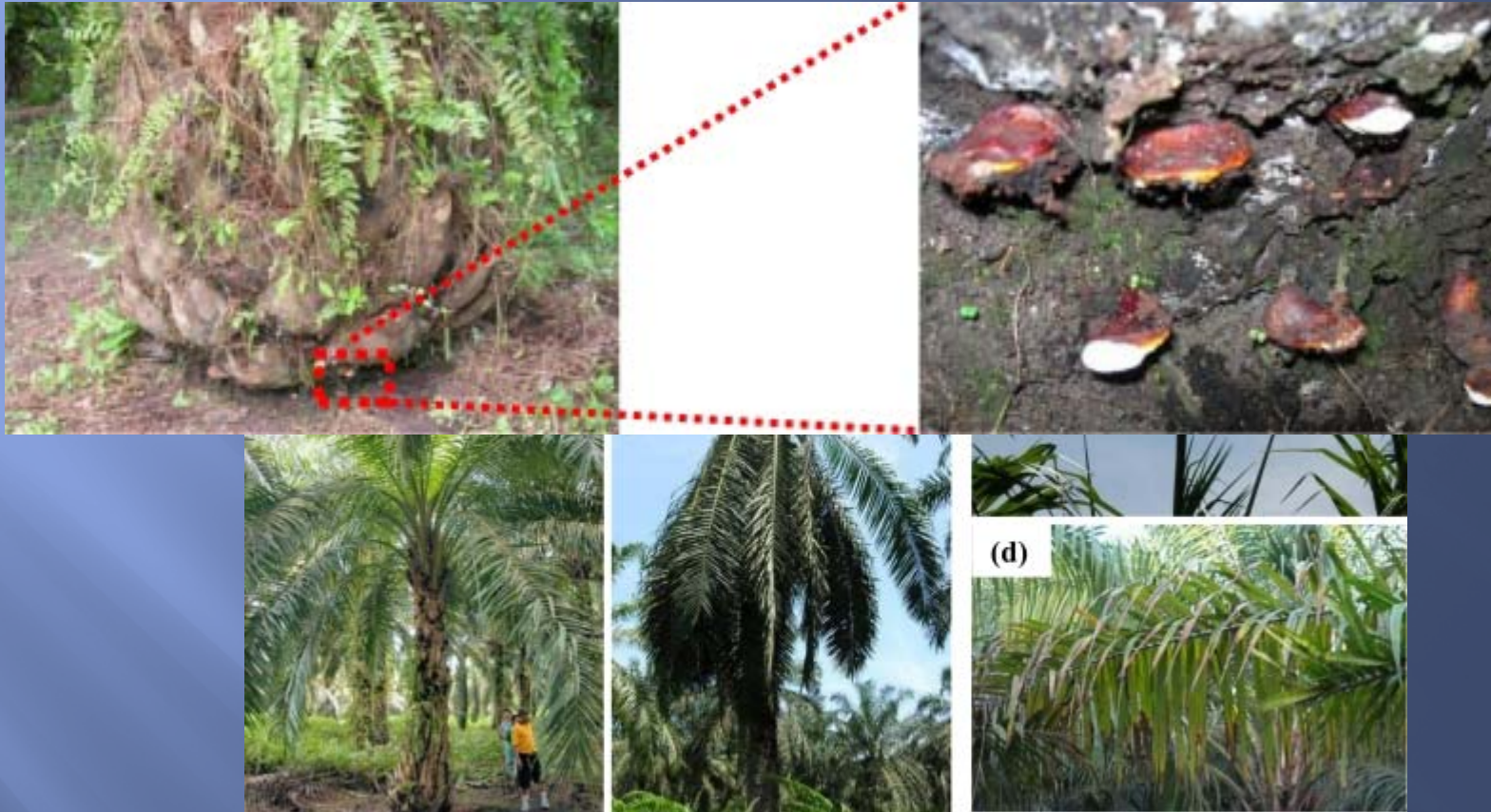


Oil Palm – plantation numbers



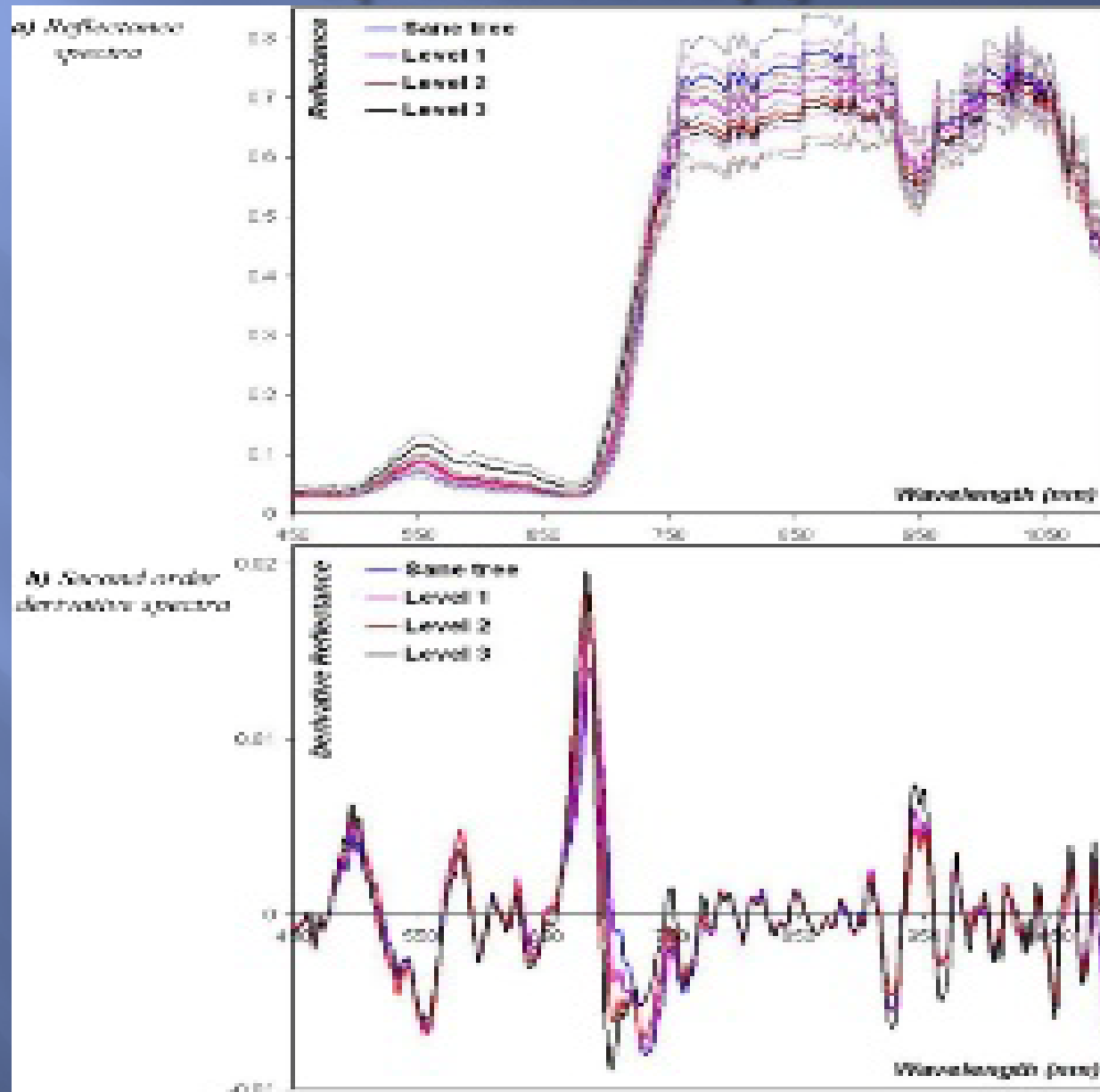
Panu Srestasathiern and **Preesan** Rakwatin
Oil Palm Tree Detection with High Resolution Multi-Spectral
Satellite Imagery. *Remote Sensing* **2014**, 6(10), 9749-9774;

Oil palm – Ganoderma disease



Ganoderma boninense

Hyperspectral reflectance spectroscopy



Phenomics

General Conclusions

- ❖ A lot of work needed to relate phenomic measurements to plant characteristics
- ❖ How far this can be taken we still have to discover
- ❖ Whether it can be developed to give robust predictions of truly polygenic characters such as yield is still to be proven.
- ❖ But it can certainly provide massive automated determination of certain traits
- ❖ It can be developed also on a commercial basis for assessing at least certain characteristics of food and food products

Thank you