Use of ground based hyperspectral imaging for high throughput field phenotyping



**Dominic Williams** 



### Why Image?

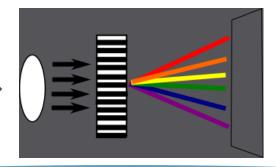


- Non invasive/ destructive measurements
- Disease detection and diagnosis
- Monitor progression of symptoms across plant both spatially and temporally
- Less labour intensive
- Can combine with computer vision techniques for automated decision making

### Hyperspectral imaging

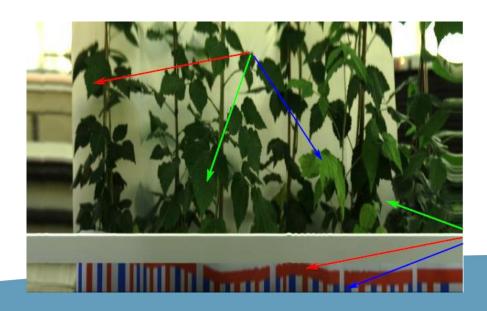


- Image where spectra at every point in the image is recorded
- Visible and near infrared (VNIR) 400-1000nm
- Short wave infrared (SWIR) 900-2500nm
- Line scanner



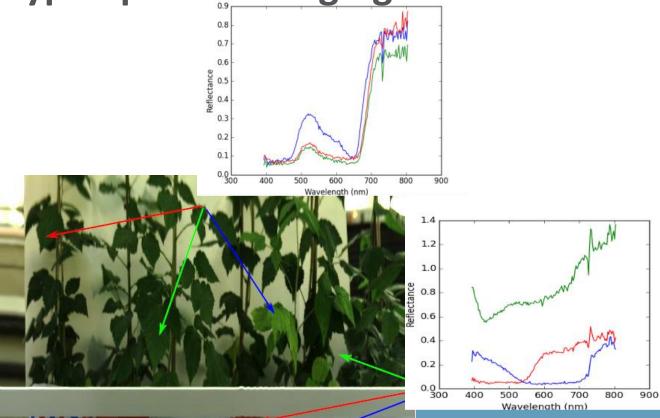
# **VNIR** Hyperspectral imaging





VNIR Hyperspectral imaging





### Field data acquisition

The James

- Imaged Raspberry Moy x Latham mapping population with 8 different stress treatments
- Different stress treatments spatially separated
- Imaged every 4 weeks in growing season over 3 years



# **Image Analysis**



Initial image from field. True colour representation

- Want to identify plants and white reference
- Need to split into individual plants and match these plants to field plan

### **Image Analysis**



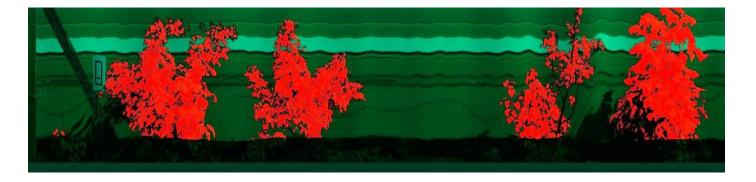


Image from field plants identified

- Identify plants using NDVI
- Need to remove grass from bottom of image.
   Use gradient to find bottom of board

## Match to field plan



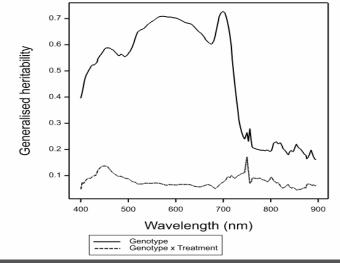


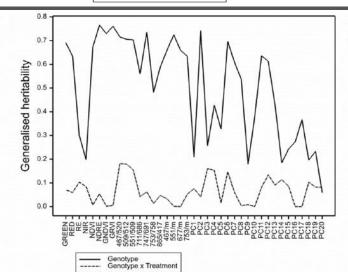
- Identify posts using yellow tape
- Find gaps between plant and split into known number of plants
- Manual correction applied at this stage to check for any errors

### **Extract plant spectrum**



- Final stage is to normalise against white reference tile
- Mean spectrum of each plant is then taken
- This data is used in downstream analysis

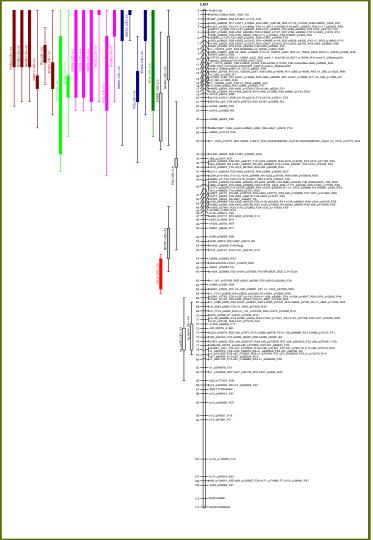




# QTL Mapping - all treatments



- Reflectance showed higher heritability for genotype
- Same pattern seen for a series of ratios and principal components
- Decided to focus on genotype effects first
- Mapped a series of ratio of principal components



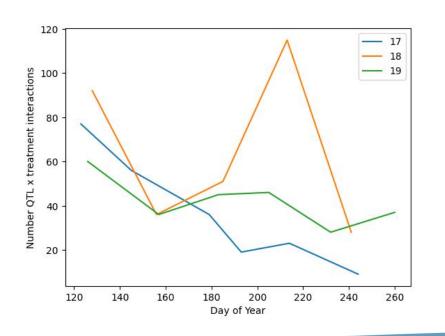
# **QTL Mapping**



- Successfully mapped series of ratios
- QTLs seen at multiple timepoints through the year
- Indicates traits have biological significance

# QTL Mapping – look for treatment interactions



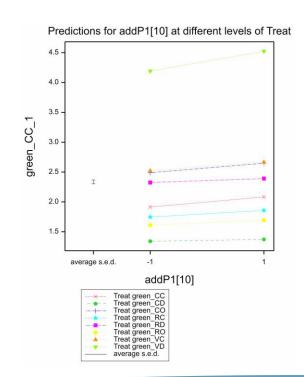


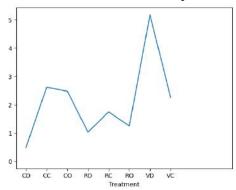
- Use Genstat to find QTL by treatment interactions
- Found interactions across all dates

### **Look at interactions**

The James

- Aim to see which interactions are same pattern
- Have plot for each interaction from genstat (794 of these!)
- Used standard error to normalise then responses to same scale

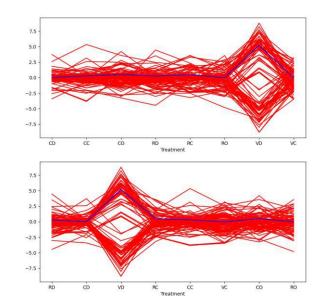




### **Look at interactions**



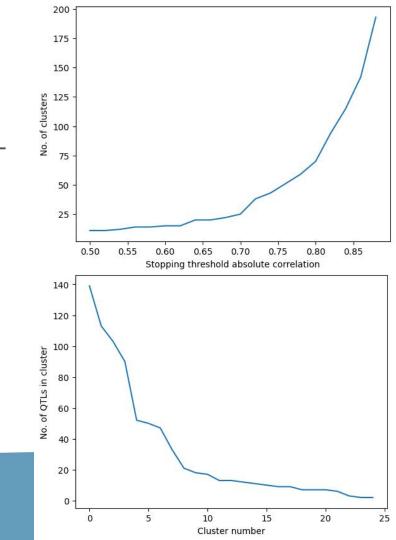
- Now want to form groups where the same
- Used hierarchical clustering
- Used absolute correlation as the similarity metric



# Hierarchical clustering

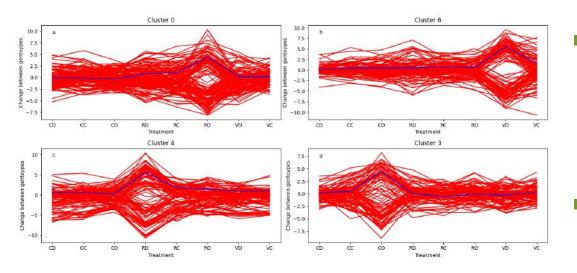
- Start with equal number of clusters and QTL interactions (794)
- Merge most similar cluster
- Continue until get reach stopping criteria
- Used absolute correlation as metric
- Stopped merging cluster at threshold

 Used threshold of 0.7 as big reduction then (gives 25 clusters but some very small)



## Hierarchical clustering

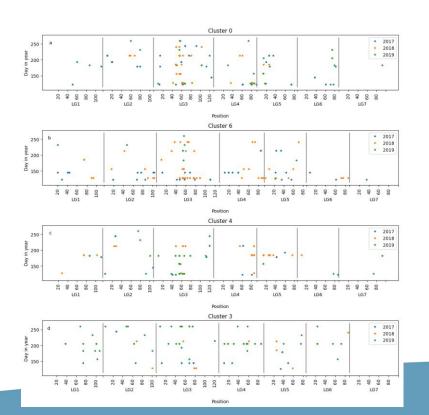




- Four largest clusters mainly driven by single treatment response
- These are not all located on same place on genome

### Hierarchical clustering





- Location of QTLs is distributed across the genome
- A few patterns can be seen
- See a lot on LG3 which always sees more QTLs

## Relationship to root rot QTLs



2017

9

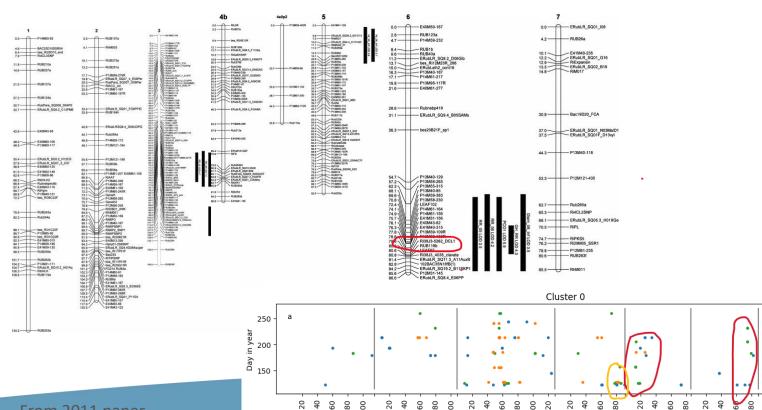
LG7

LG5

Position

2018

2019



LG1

### Relationship to Physical Fruit QTLs

Plantier EniteR SOAF CEEDAD Responded Rype

46 P13M58-288

454 COORF EMB1644

- ERIGHT SQLS.3 BIRTS

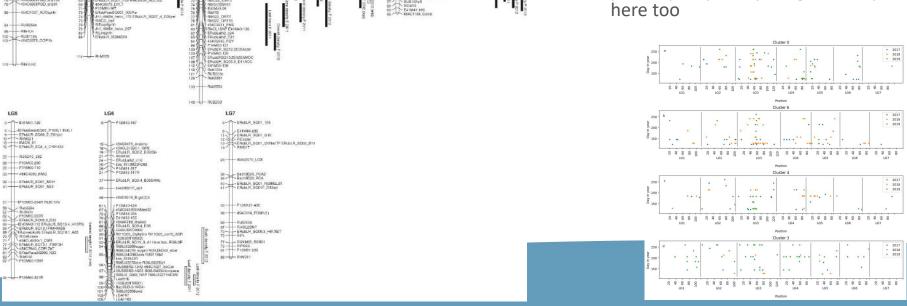
| RISSONS AST\_LIR | UPLACE R. SEGOZ. AGROCII | RIBBITOS | SEGOZENSE | ASSOCIATE R. SEGOZ. AGROCIATE | EPILACE R. SEGOZ. AGROCIATE | EPILACE, SEGOZ. FOSABA | 4540237 E. SEGOZ. FOSABA

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Many architectural traits in LG3 – possibly why everything else maps here too



6 EMBLD 184 10 ERUEZTIPE 12 ERUEZTIPE 14 266 prot 16 Rental

15 Harf4 17 ERIALP, 80071 E10TF 18 Phate, 8013320 12PP 19 RabnebH15 20 FRUTEBONT POWER-112

- ERV&LR\_8019.2E00Exp

46 454CBSB ISPH 454CBTO BPH 454CLB146C1 ZFP9 51 NUONES

- 454C1913\_PG F2 -154C1913\_PG F2

E41M31-147 FRUETER 65-4C0016\_F ERubLPcom74PME-

A 10001-100

P-I2M121-100

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16-454c1717Embde

26 ---- ER40P3195Q008004 26 ---- ER40LR\_SQR 2\_C12PE

46 ERUBLE SQ5.2 H12Cell

-tise\_Rist 022R -4540638EPGD\_snp155

454C8398PGO gruph

22 RUB 124w

41 -- E40M68-108

PLANSIAN

63 — E40M43-110 67 — bes\_RiM023F 70 — RiCelti

12 - IRipy E40Mso-126

- ERUELR 805.3 E08608

NPG215 RUB4s (\*bes\_Ri29G13 PR25D10\_295

- ERABLES DA AGOSANDAS

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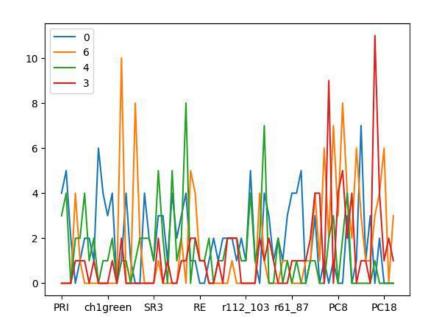
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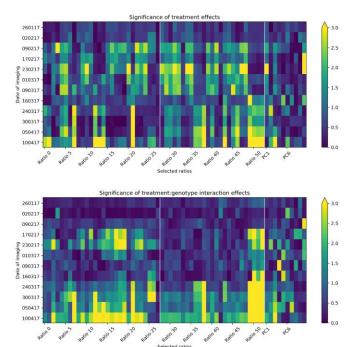
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# **Spectral traits**







#### **Conclusions**



- We have mapped plant spectral responses
- Interactions between mapped QTLs and treatment response have been found
- These partially collocate with previously known QTLs
- Technique could be used to find genetic control of novel stress
- No evidence to linking a particular spectral response to a particular stress







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