

# WGIN

## Resource Development Update

Simon Griffiths  
John Innes Centre

# Paragon Mutants Plant Morphology



waxless – 11/6500



monoculm – 15/6500



awned – 8/6500

# Types of resources

- **Germplasm collections**
  - AE Watkins
  - Gediflux
- **Segregating populations**
  - Avalon x Cadenza
  - Paragon x Garcia
  - Paragon x Chinese Spring
  - Paragon x Synthetic
- **Near Isogenic Lines**
  - Avalon x Cadenza (Yield, height, heading, grain shape\*, stem characters\*)
  - Malacca x Hereward
  - Lr19, 3N\*
- **Mutant populations**
  - Paragon EMS
  - Paragon gamma
- **Molecular markers**
  - Conserved orthologous set (COS)

# Types of resources

- **Germplasm collections**
  - AE Watkins
  - Gediflux
- **Segregating populations**
  - Avalon x Cadenza
  - Paragon x Garcia F2-F3 seed- so F4 seed in August, F5 Feb 13, F6 Aug 13, Multiply from a 2013 drilling- John F measure anything?
  - Paragon x Chinese Spring
  - Paragon x Synthetic
- **Near Isogenic Lines**
  - Avalon x Cadenza (Yield, height, heading, grain shape\*, stem characters\*) email spreadsheet, grain shape is at BC3 going to BC4, so multiplied homozygotes Feb 2013
  - Malacca x Hereward- homozygous BC3F2 in ear now (Malacca background) Hereward vern problems- now in outside glasshouses- likely to lose 1 generation.
  - Lr19, 3N\*- Lr19 Paragon at BC4, select to self in June, select homozygotes in December, multiplies homozygotes for drilling in 2013. Problem germinating Alchemy (BC3), seem to get male sterility in BC2 plants
  - 3N at BC2
- **Mutant populations**
  - Paragon EMS
  - Paragon gamma- Reextracting DNA for X 1000
- **Molecular markers**
  - Conserved orthologous set (COS)

# Delving Back

## The A. E. Watkins Collection

- Foresight for collection in the 1930s through the London Board of Trade to 'capture' diversity
- Currently 1200 accessions from around the world
- 32 Countries



# Germplasm Collections

- AE Watkins previously discussed- 800?? Lines expanded to 10?? Accessions as they segregated.
- Focus today on Gediflux Collection

Country	Varieties	Years of release	examples
Austria	40	40-90	Tassilo (50s) Hubertos (90s)
Belgium	24	50-90	Norda (60s) Escorial (80s)
Germany	18	80-90	Calif (80s) Pegassos (90s)
E Germany	30	40-80	Mahndorf (50s) Kanzler (80s)
W Germany	19	50-90	Muck (50s) Borenos (90s)
Denmark	5	80-90	Anja (80s) Pepital (90s)
France	34	40-90	Vague d'epis (40s) Isengrain (90s)
UK	66	40-90	Holdfast (40s) Equinox (90s)
Netherlands	19	40-80	Lovink (40s) Nautica (80s)
Sweden	26	25-90	Jarl (20s) Meridien (90s)
UK NL	229		

# Gediflux genotyping

- 42 SSR markers Trait Genetics as part of original Framework 6 work programme
- Used to identify core sets and structure within population
- WGIN committed to DArT on Gediflux, changed milestone to equivalent expenditure on KASPar



# Measures of Genetic Diversity

- allele richness
- average allele number
- number of rare alleles ( $< 2\%$ )
- Nei's Gene Diversity Index

$$H = 1 - \sum_i^n (p_i)^2$$

- Polymorphic Information Content

$$PIC = 1 - \sum_i^n (p_i)^2 - \sum_i^n \sum_{j=i+1}^n 2p_i^2 p_j^2$$

$p_i$  is the frequency of the  $i$ th allele.

# Comparison GEDIFLUX vs Watkins

	Gediflux	Watkins
markers (loci)	42	45
accessions	511	1059
allele richness	443	1051
average allele no	10.5	18.4
range	2-26	8-62
avg. rare allele no	5.5	11.8
range	0-16	3-27
PIC	0.649	0.738

# Definition of Population Structure

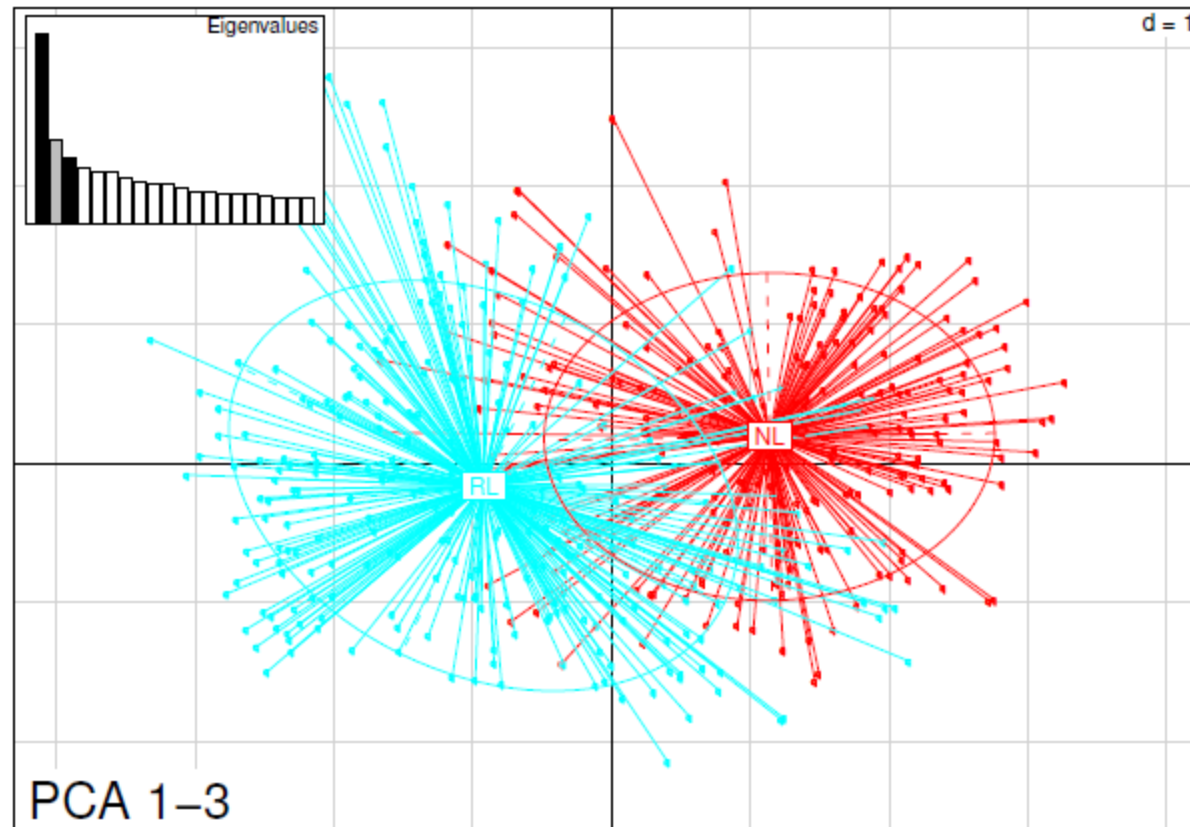
Population Structure means, that a **population is subdivided** in some way.

It is important to understand the population structure in order to mine the genetic diversity.

Strategies to understand the hidden structure

- Using Geography - clustering by countries
- Discriminate Analysis (DAPC) - multivariate analysis
- STRUCTURE - model based clustering

# Discriminant Analysis (DA) on Gediflux



Gediflux: genetic diversity of modern European winter wheat:

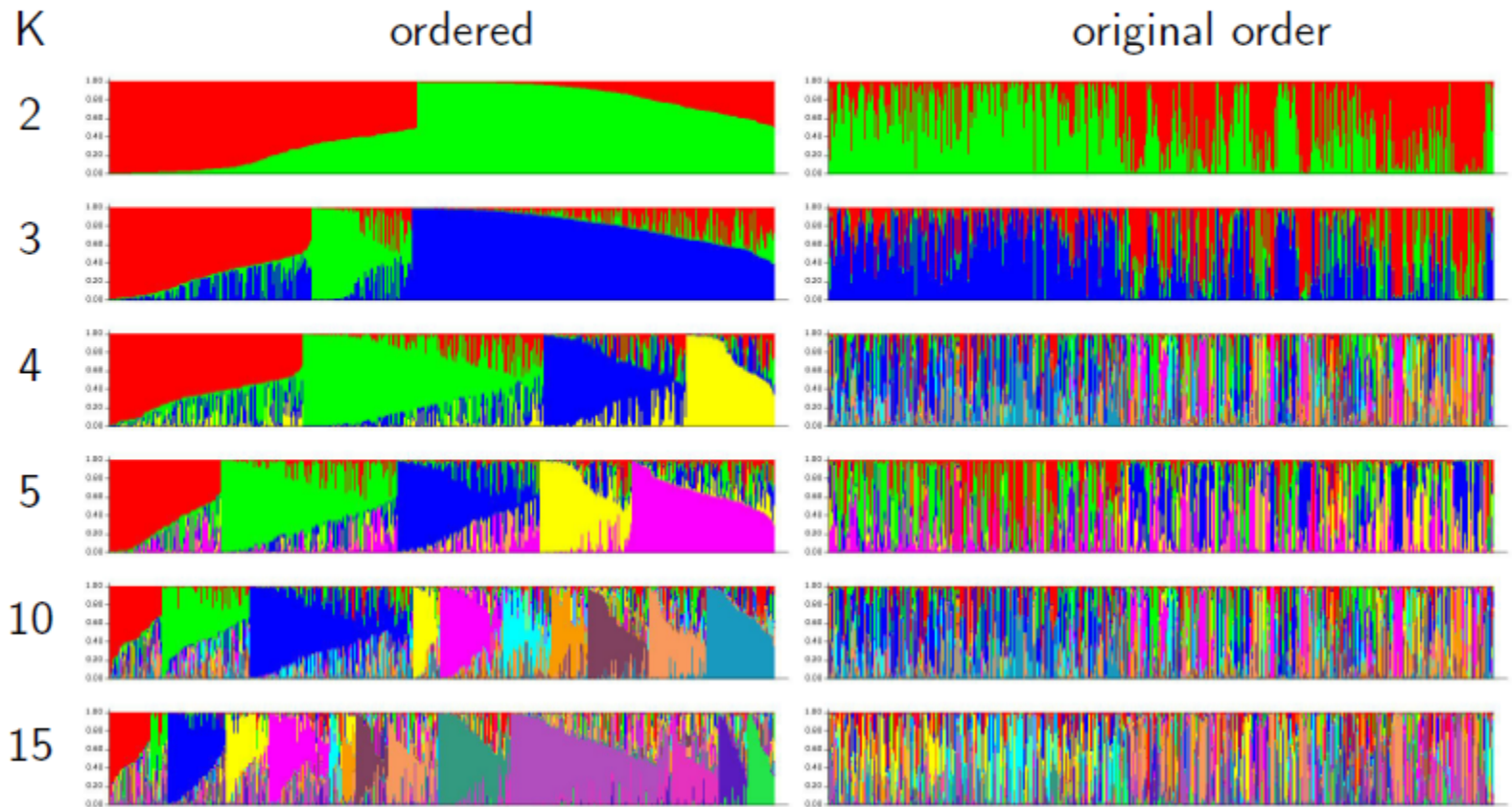
NL: UK National List (late 1990s), RL: EU-recommended List (1945-2000)

Principal Component Analysis, PC 1+3

DAPC: Jombart, BMC Genetics 2010

# STRUCTURE: Inference on population structure

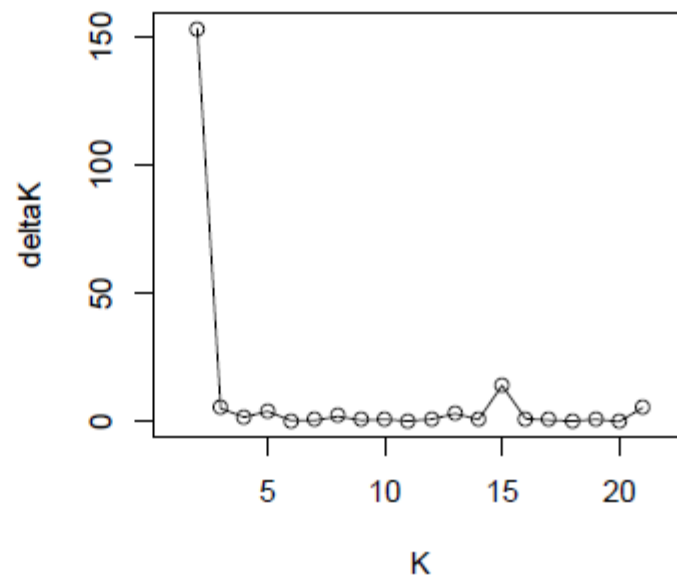
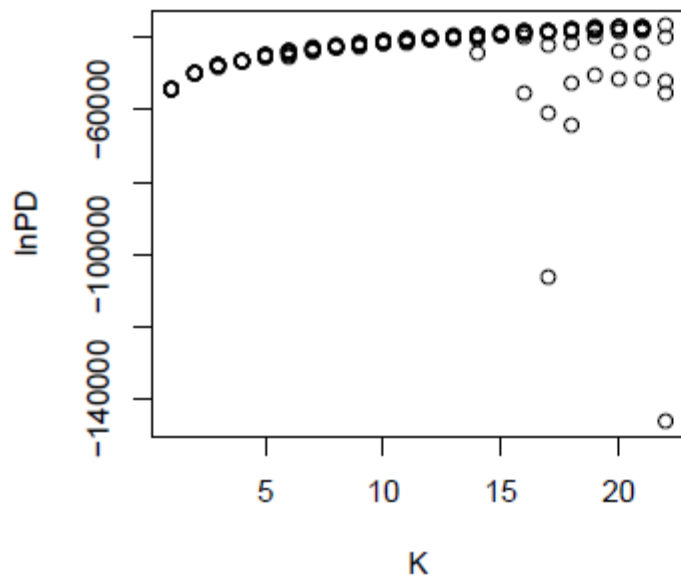
Model-based clustering to assign individuals to ancestral populations.



STRUCTURE: Pritchard, Genetics 2000

# STRUCTURE analysis: How many Ks?

Finding the number of ancestral populations: Evanno's delta K:



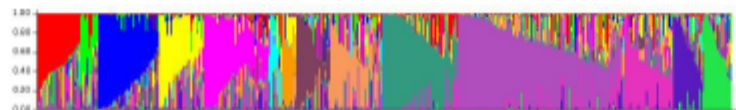
Evanno,  
Mol. Ecology  
2005

STRUCTURE finds 2 ancestral groups (K) + some some support for K=15.

K=2



K=15



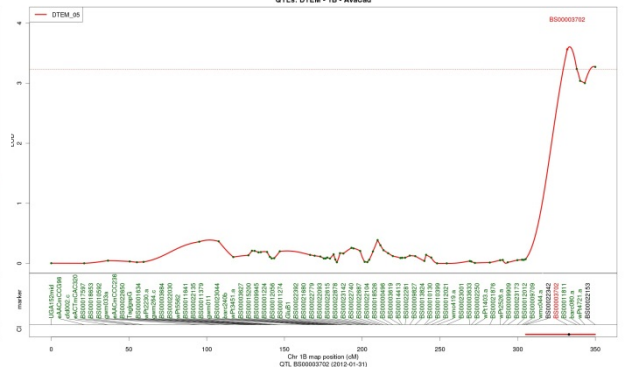
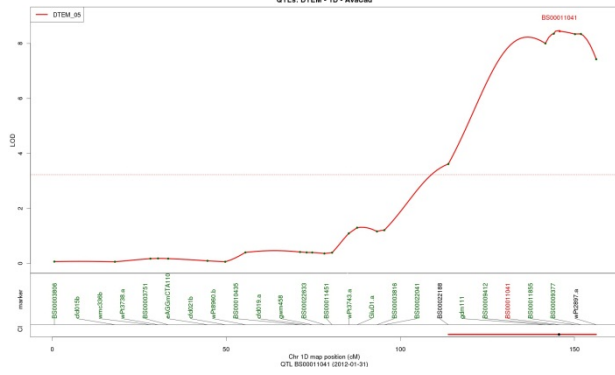
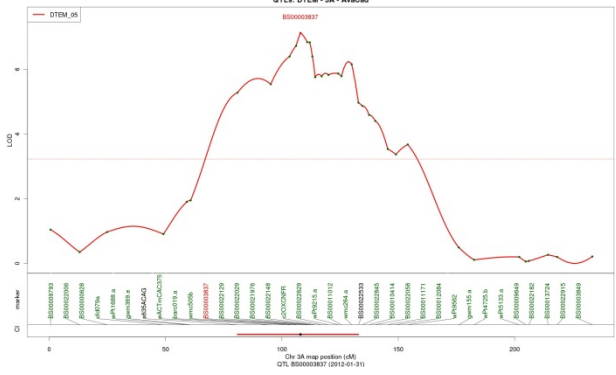
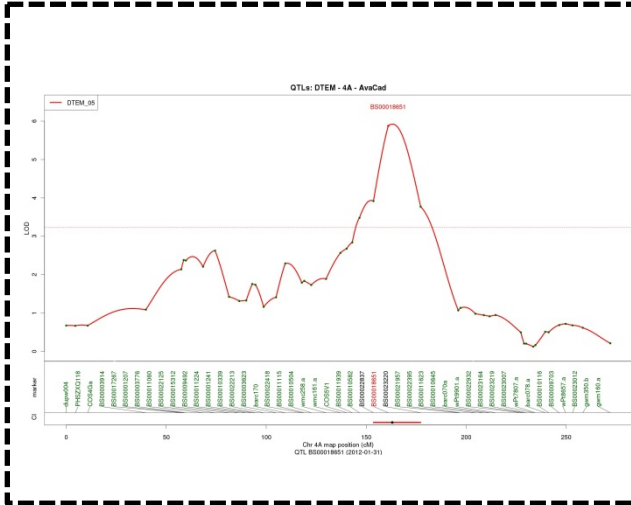
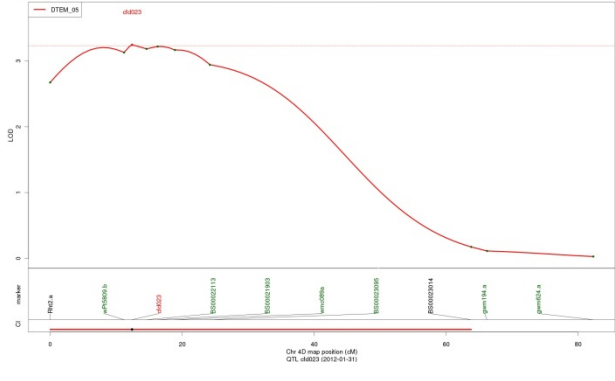
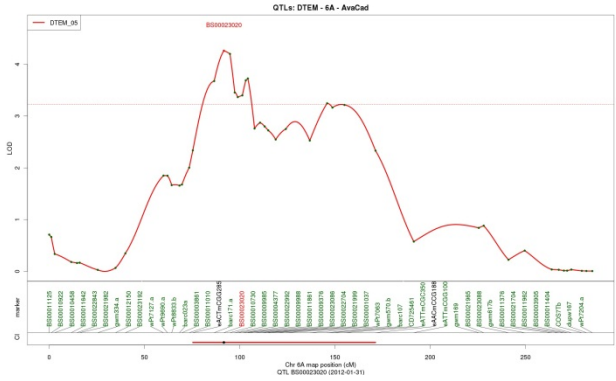
# Gediflux phenotyping

- 3 replicates of 1m<sup>2</sup> plots at JIC in '10-'11 season
- Field measures
  - Height
  - Ear emergence
- Post harvest on grab samples
  - Spikelet number
  - Non/Fertile floret number
  - Ear weight and length
  - Thousand grain weight + grain length and width
  - Internode length and number

Trait	Marker	Chromosome	F	p
Height	RHT2	4D	43.50056	1.17E-10

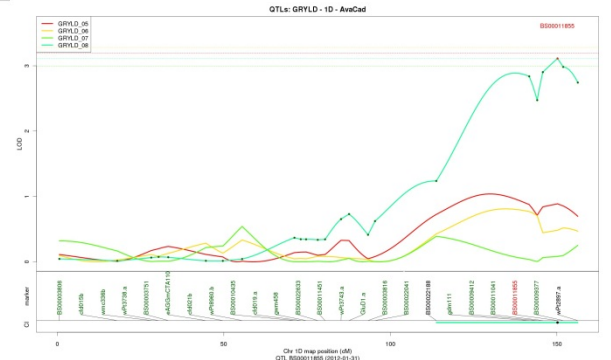
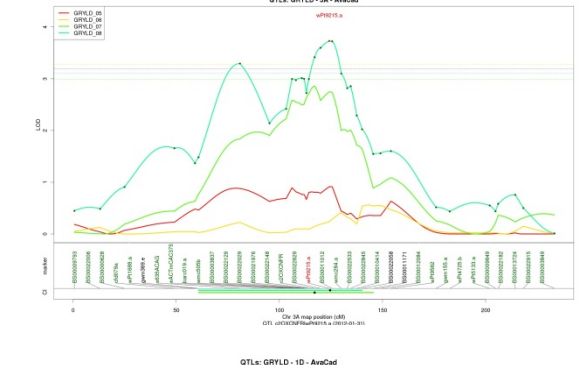
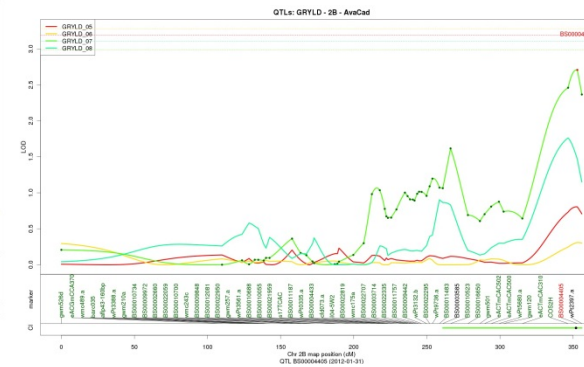
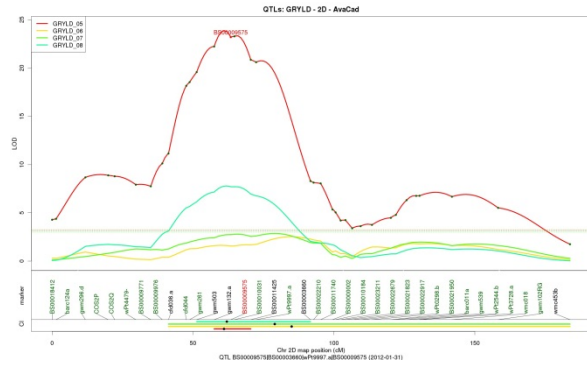
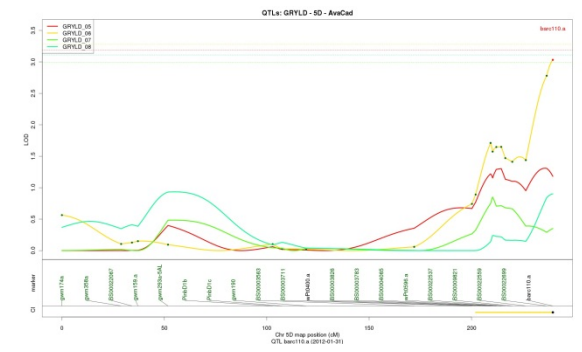
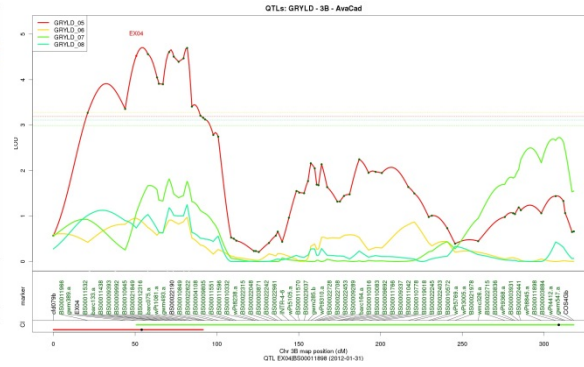
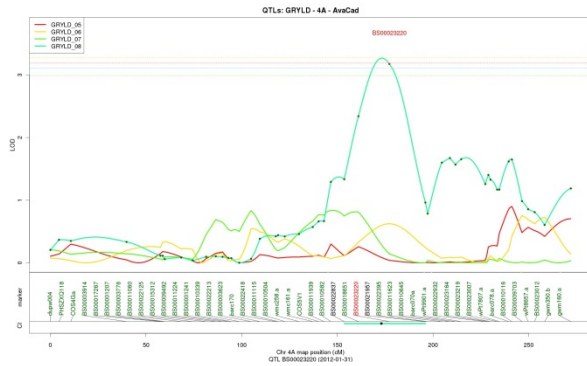


# Avalon x Cadenza QTL summary- ear emergence

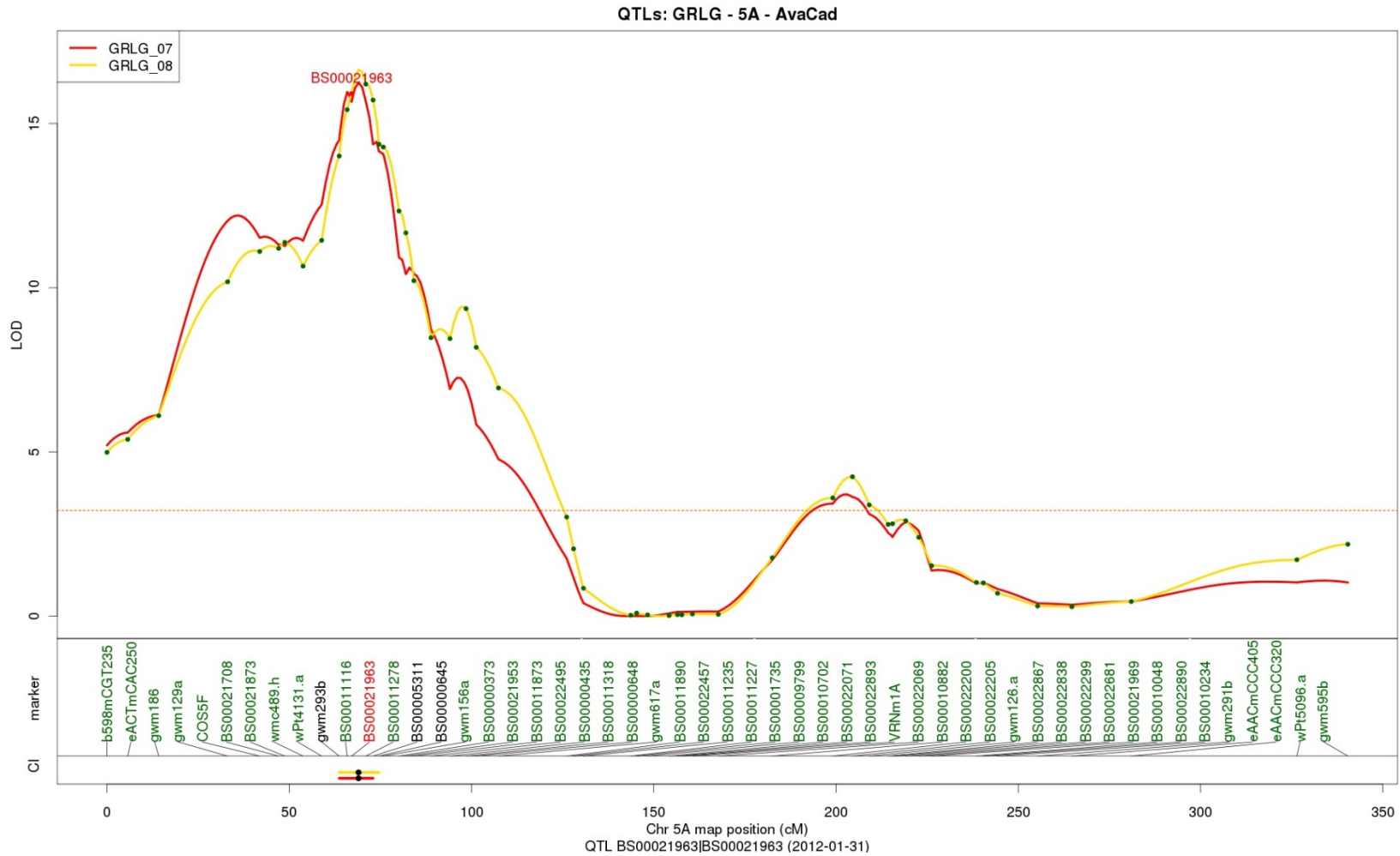




# Avalon x Cadenza QTL summary- grain yield



# New traits for WGIN eg grain length



# Stem solidness and its relationship to water-soluble carbohydrates: association with wheat yield under water deficit

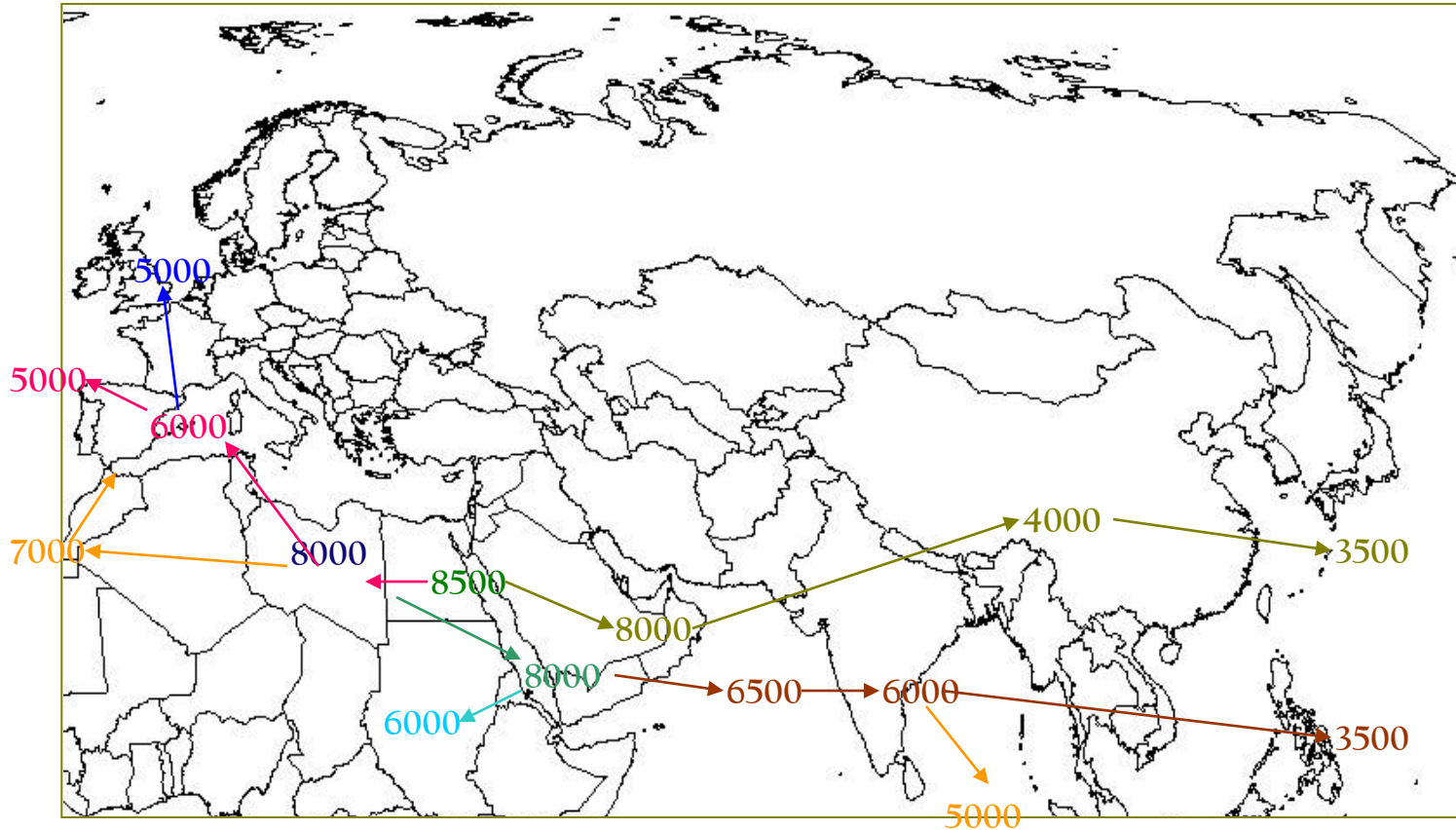
Author(s): [Saint Pierre, C](#) (Saint Pierre, Carolina)<sup>1</sup>; [Trethowan, R](#) (Trethowan, Richard)<sup>2</sup>; [Reynolds, M](#) (Reynolds, Matthew)<sup>1</sup>

**Source:** FUNCTIONAL PLANT BIOLOGY Volume: 37 Issue: 2 Pages: 166-174 DOI: 10.1071/FP09174 Published: 2010

Times Cited: [4](#) (from Web of Science)

Cited References: [41](#) [ [view related records](#) ] [Citation Map](#)

**Abstract:** A study of 36 wheat (*Triticum aestivum* L.) genotypes with different levels of stem solidness was conducted to assess the heritability and relationship among stem morphological properties, stem water-soluble carbohydrates (WSC) storage capacity and grain yield. The total amount of pith-fill in the upper stem internode (VOL) was highly correlated with the total content of WSC per stem under both water deficit (DEF) ( $r = 0.56$ ) and well irrigated conditions (IRR) ( $r = 0.49$ ). A positive correlation was also found between VOL and grain yield under DEF ( $r = 0.49$ ), which was explained by the positive contribution of WSC to grain yield. A closer association of grain yield and morphological traits was identified under DEF than under IRR. The closer associations found among estimations of % WSC and WSC-area and grain yield under DEF indicate that these variables may be adaptive rather than constitutive traits. High heritability values (0.77-0.84) observed for stem morphological traits reinforce their potential use in breeding for high WSC and ultimately, higher grain yield under water-limited environments. Stem length, diameter and solidness could be combined in an ideal plant ideotype to maximise WSC reserves as a strategy to improve yield under water-limited conditions.



Map to show the migration of wheat from the Fertile Crescent (years BC)

Adaptations: Now from Argentina to Canada to New Zealand

# Early Development

- Eight Lines - derived from India (1190034), China (1190141), Egypt (1190209), Cyprus (1190292), Yugoslavia (1190352), Afghanistan (1190468), Poland (1190481), Iran (1190729)
- Initially selected on height and heading time extremes from single row trial 2006
- F1s produced 2008 – early initiative (F6)





# NIL field trials

- 1m<sup>2</sup> plots grown in 10-11 season for Cadenza background
- Cadenza and Avalon background sown Autumn '11
- 225 lines in Avalon background representing height, heading, and yield QTL in non replicated 1m<sup>2</sup> plots
- 125 lines in Cadenza background grown in three randomised blocks of three hundred and seventy five 1m<sup>2</sup> plots
- Sow replicated 6m<sup>2</sup> plots this year

# Aknowledgements

- Sue Freeman
- Cathy Mumford
- Luzie Wingen
- Simon Orford
- Michelle Leverington



## Objective 8 – Nitrogen update

M J Hawkesford

WGIN Management Meeting

21<sup>st</sup> February 2012

# Why nitrogen?

- Required for yield
- Required for protein
- Costs – financial/environmental
- Low efficiency on worldwide scale but higher in UK
- Management and genetic components



# WGIN NUE Objectives

- Define important NUE traits
- Identify and evaluate diversity
- Determine mechanisms
- Provide leads for markers and genes
- Examine trait stability over multiple years
- Trials and basic datasets



# Approaches

- Modern commercial germplasm (Diversity) and mapping population (Avalon x Cadenza) trials
- Assessment of trial diversity in the field
- Identify QTL



# WGIN 2009-2013

- Deeper phenotyping
  - Partitioning
  - Canopy longevity
  - Roots and uptake
- Stability data
- NUE – WUE – Take-all interactions
- Encourage spin off projects



# Diversity trial history

Trial	Year	Varieties (core of 9)	N-levels	kg N/ha
1	2004	32	4	0,50,200,350
2	2005	20	2	0,200
3	2006	24	3	0,100,200
4	2007	24	4	0,100,200,350
5	2008	24	4	0,100,200,350
6	2009	24 (include 6 x A x Cs)	4	0,100,200,350
7	2010	25 (include 6 x A x Cs)	4	0,100,200,350
8	2011	25 (include 4 x A x Cs)	4	0,100,200,350
9	2012	25 (include WUE/take-all lines)	4	0,100,200,350
10	2013	25 (include WUE/take-all lines)	4	0,100,200,350



# Diversity Trial 2011



WGIN management 21st February 2012

# 2012 varieties list (part 1)

Wheat varieties for WGIN-NUE 2010/11

W=WGIN data, D=desk study

Variety	Source	Code	Nabim	Rationale	inclusion in trial requested by	Previous years of trials (harvest year)
1. Avalon	Av	AV	1	WGIN DH parent; Low NupE & NutE (D)	PB, RG, MJH	05-10
2. Cadenza	Ca	CA	2	WGIN DH parent; Best NupE (W)	PB, RG, MJH	04-10
3. Chablis NEW 09/10	KWS		2	SPRING variety (previous grown in 2004 trial) as very N-MH responsive variety		only in 04 and 10
4. Claire NEW 2005	Nick	CL	3	Biggest area on RL; WGIN DH parent; <b>Good second wheat</b>	PB,PS	05-10
<b>5. Conqueror</b>	<b>KWS</b>	<b>CN</b>	<b>4</b>	<b>New Grp 4, very high yielding</b>	<b>MH</b>	<b>new</b>
6. Cordiale NEW 2006	KWS	CO	2	<b>Good second wheat. BBSRC Quality project</b>	RG	06-10
7. Crusoe NEW 10/11	Nick	CR	2	Carries dicoccoides. Shows the 'stay green' character		
8. Gallant NEW 09/10	Syn	GA	1	new claimed high yield and high protein type	MH	
<b>9. Hereford</b>	<b>Syn</b>	<b>HF</b>	<b>4</b>	<b>Feed (not on RL), high yield, brown rust susceptible, KHK/RG possible low take-all build-up and good resistance.</b>		<b>new</b>
10. Hereward	RAGT	HE	1	Best protein on RL; benchmark bread variety. <b>BBSRC Quality project</b>	PB,PS	04-10
11. Istabraq NEW 2005	Nick	IS	4	Best yield on RL; Distilling cultivar; In LINK 'GREENgrain'; <b>Good second wheat. BBSRC Quality project. WUE trial</b>	PB,PS	05-10
12. Malacca	KWS	MA	1	Biggest Group 1 area; DH choice; Low NupE, high NutEPS (W). <b>BBSRC Quality project</b>		04-10
13. Marksman	RAGT	MK	2	new for 2009, PRS request for <b>BBSRC Quality project</b>		only 09 and 10

# 2012 varieties list (part 2)

W=WGIN data, D=desk study

Variety	Source	Code	Nabim	Rationale	inclusion in trial requested by	Previous years of trials (harvest year)
14. Maris Widgeon		MW	1	Tall (rht), old cultivar	PB, AM	04-10
15. Mercia		ME	1	Low NupE & NutE (desk); Low Canopy N requirement; In IGF micro-array. <a href="#">WUE trial</a> . RHT series	RG	04 and 06-10
16. Paragon	RAGT	PA	1	Spring variety; WGIN mutagenesis population; High NupE (W)	PB	04-10
17. Riband	RAGT	RI	3	WGIN DH parent; Distilling cultivar; In LINK 'GREENgrain'; High NutERG (W)		04-10
18. Robigus NEW 2005	KWS	RO	3	Best Group 3 yield; Best NUE, high NupE & NutE (D); <a href="#">Good second wheat. WUE trial</a>	PB, AM	05-10
19. Stigg NEW 10/11	Nick	ST	?4	Carries dicoccoides. High disease resistance. Shows the 'stay green' character		
20 Soissons	Elsoms	SS	2	WGIN DH parent; Early maturing; High NupE, low NutE (W)	PB, RG, AM	04-10
21. Solstice	Nick	SL	2	Biggest Group 2 area; DH choice; Worst NupE (W)	RG	04-10
22. Xi19	Nick	XI	1	Best Group 1 yield; High NUE, NupE, NutE (D); Low NupE (W). <a href="#">BBSRC Quality project. WUE trial</a>	PB, PS	04-10
<b>23. Zebedee</b>	<b>LIM</b>	<b>ZE</b>	<b>3</b>	<b>High WUE, grp 3</b>	<b>JFoulkes</b>	<b>new</b>
24. AxC line 127		D5		new in 2009 - good early export from leaves	MJH	09/10/
25. AxC line 82		D6		new in 2009 - slow early export from leaves	MJH	09/10/

Removed from trial: 2 A x C lines and Oakley

# Example results

- Trait performance rankings
- Stability
- Yield plateauing
- qtls

Variety Performance at 200 kg-N/ha (2004-08)

Variety	Code	Nabim	Years	Yield	%N	Uptake	Utilisation
Avalon	AV	1	5				
Flanders	FL	1	1				
Hereward	HE	1	5				
Hurley	HU	1	5				
Malacca	MA	1	5				
Mercia	ME	1	4				
Maris Widgeon	MW	1	5				
Shamrock	SH	1	4				
Solstice	SL	1	5				
Spark	SP	1	1				
Xi 19	XI	1	5				
Cadenza	CA	2	5				
Cordiale	CO	2	3				
Einstein	EI	2	1				
Lynx	LY	2	5				
Rialto	RL	2	1				
Scorpion	SC	2	1				
Soissons	SS	2	5				
Beaver	BE	3	4				
Claire	CL	3	4				
Riband	RI	3	5				
Robigus	RO	3	4				
Istabraq	IS	4	4				
Napier	NA	4	3				
Savannah	SA	4	4				
Paragon (spring)	PA	1	5				
Chablis (spring)	CH	2	1				
Arche	AR	F	1				
Batis	BA	G	5				
Caphorn	CP	F	1				
Cappelle Desprez	CD	F	1				
Enorm	EN	G	1				
Isengrain	IG	F	1				
Monopol	MO	G	5				
Opus	OP	G	1				
PBis	PB	G	1				
Petrus	PE	G	1				
Sokrates	SK	G	5				
Zyta	ZY	P	1				

Upper-Q  
Inter-Q  
Inter-Q  
Lower-Q



Summary of variety performance (quartile rankings) based on 2004-07 WGIN datasets

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Contents lists available at ScienceDirect

**European Journal of Agronomy**

journal homepage: [www.elsevier.com/locate/eja](http://www.elsevier.com/locate/eja)

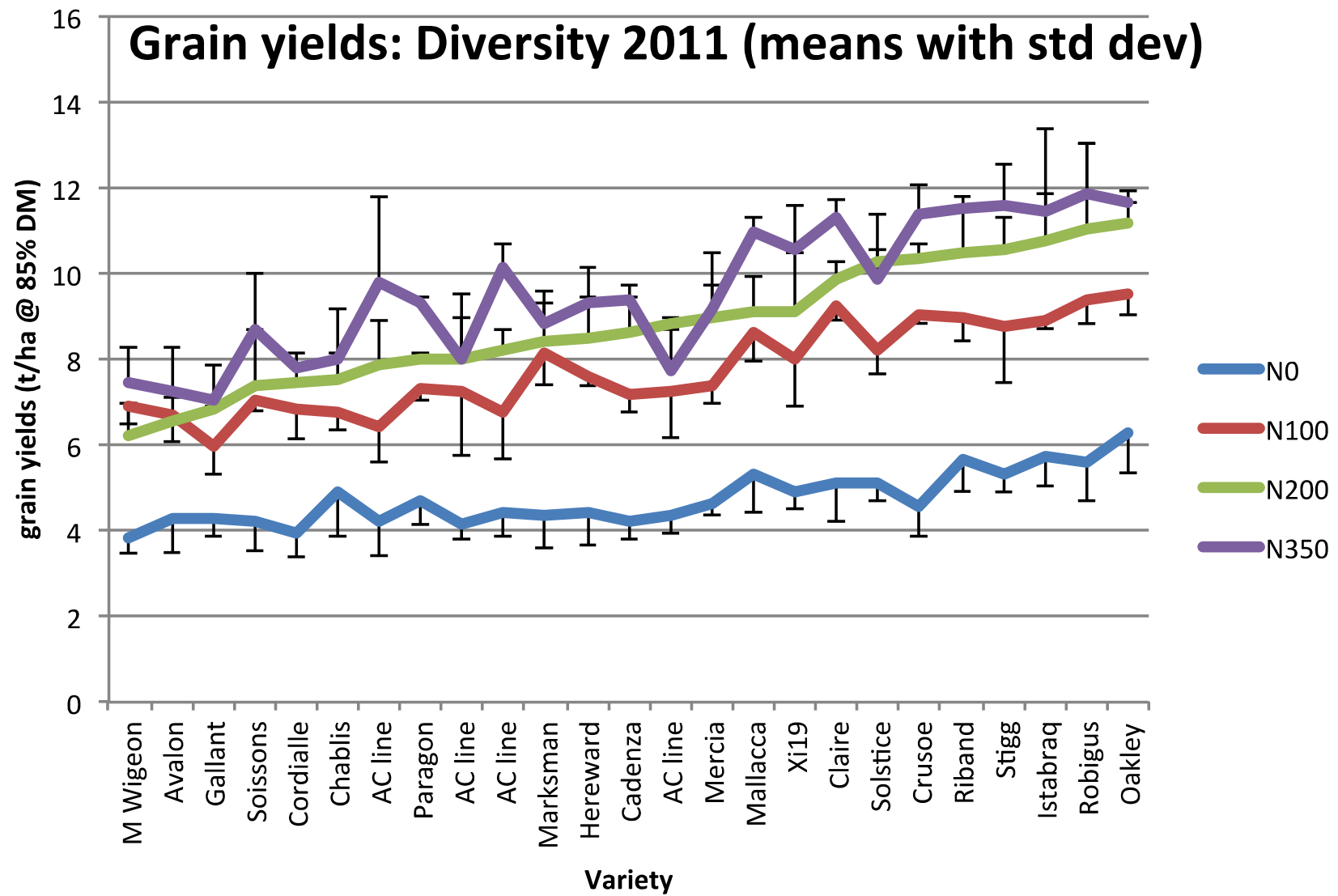



**Nitrogen efficiency of wheat: Genotypic and environmental variation and prospects for improvement**

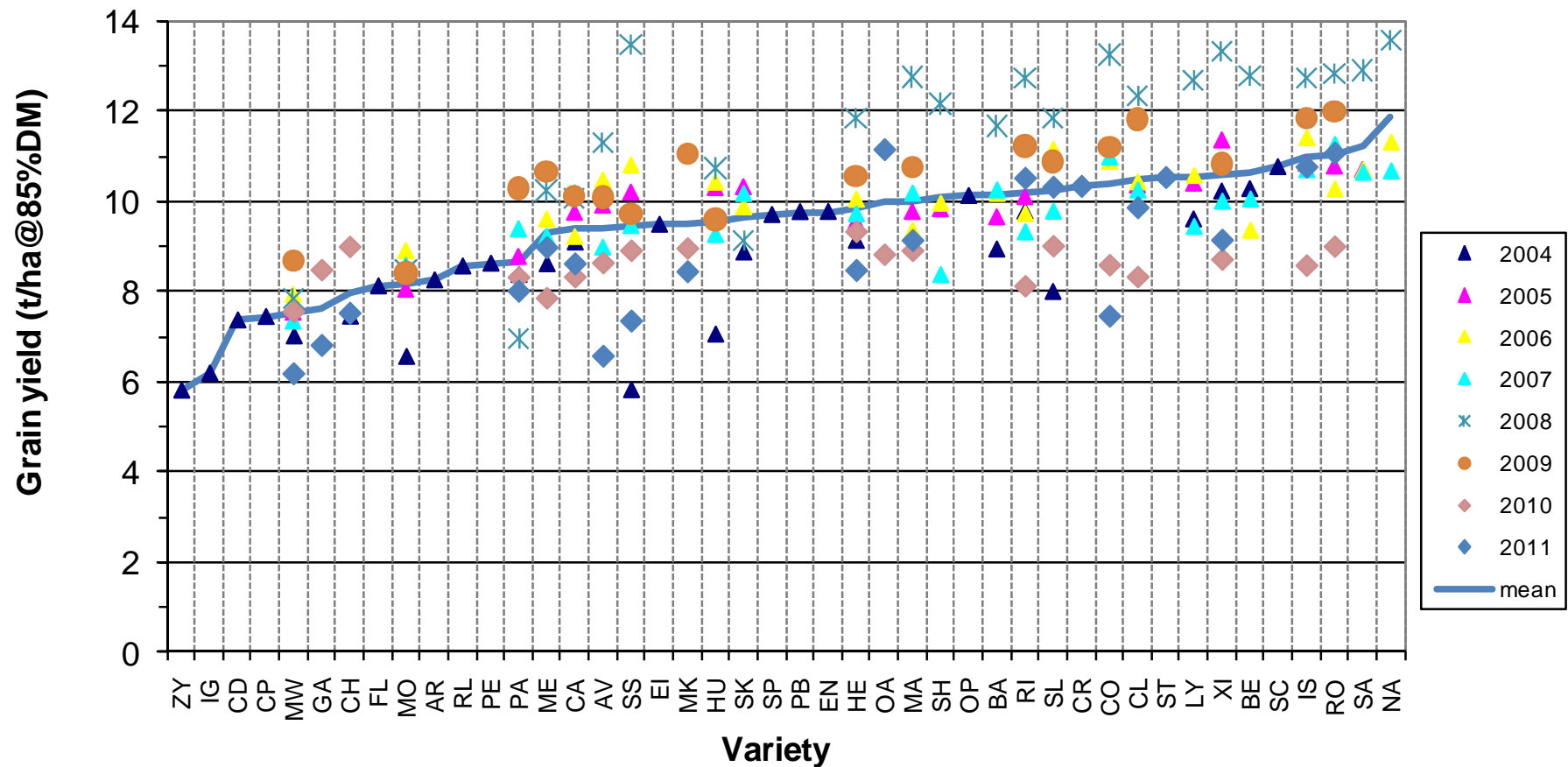
Peter B. Barraclough<sup>a,\*</sup>, Jonathan R. Howarth<sup>a</sup>, Janina Jones<sup>a</sup>, Rafael Lopez-Bellido<sup>b</sup>, Saroj Parmar<sup>a</sup>, Caroline E. Shepherd<sup>a</sup>, Malcolm J. Hawkesford<sup>a</sup>

EJA (2010) 33, 1-11

February 2012



## Rothamsted WGIN-N200      Combine Grain Yield (2004-11)



44 varieties

# Avalon x Cadenza traits

- Yield (at high and low N availability)
- Height, flowering time
- N uptake/partitioning → NUpE, NUtE, NHI
- Canopy senescence
- Roots
- Micronutrients and other macro nutrients
- Take all





# Avalon x Cadenza Trials

Trial	Harvest year	Sites	Notes	kg N/ha
1	2007	Blackhorse		200
2, 3	2008	Bones and Woburn	Woburn trial split	Both 100
4	2009	Fosters/Summardells	Split over 2 fields (wet)	100
5	2010	Blackhorse	(very dry)	200
6	2011	Great Harpenden	(very dry)	200
7*	2012	Bones		Tbc: 100?
8*	2013?	?		200?

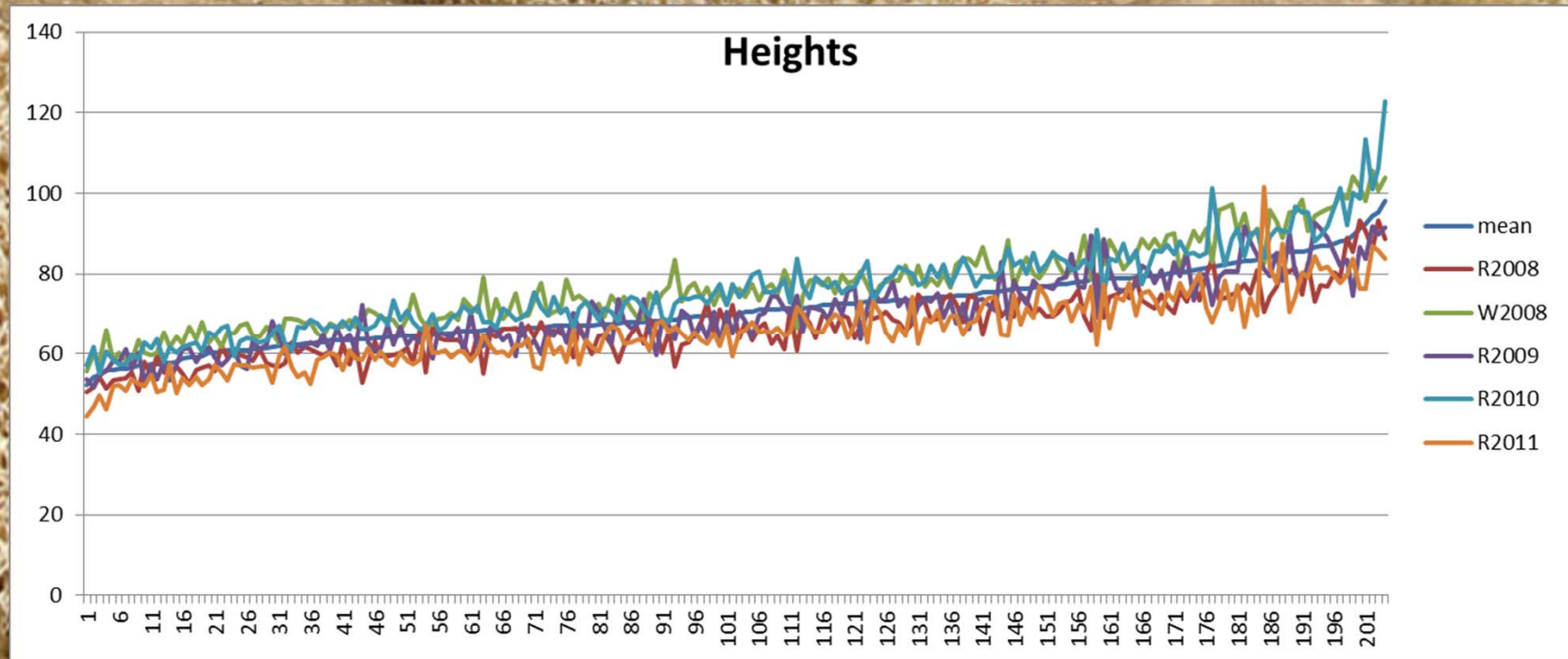
\*= not WGIN funded

# Avalon x Cadenza 2011

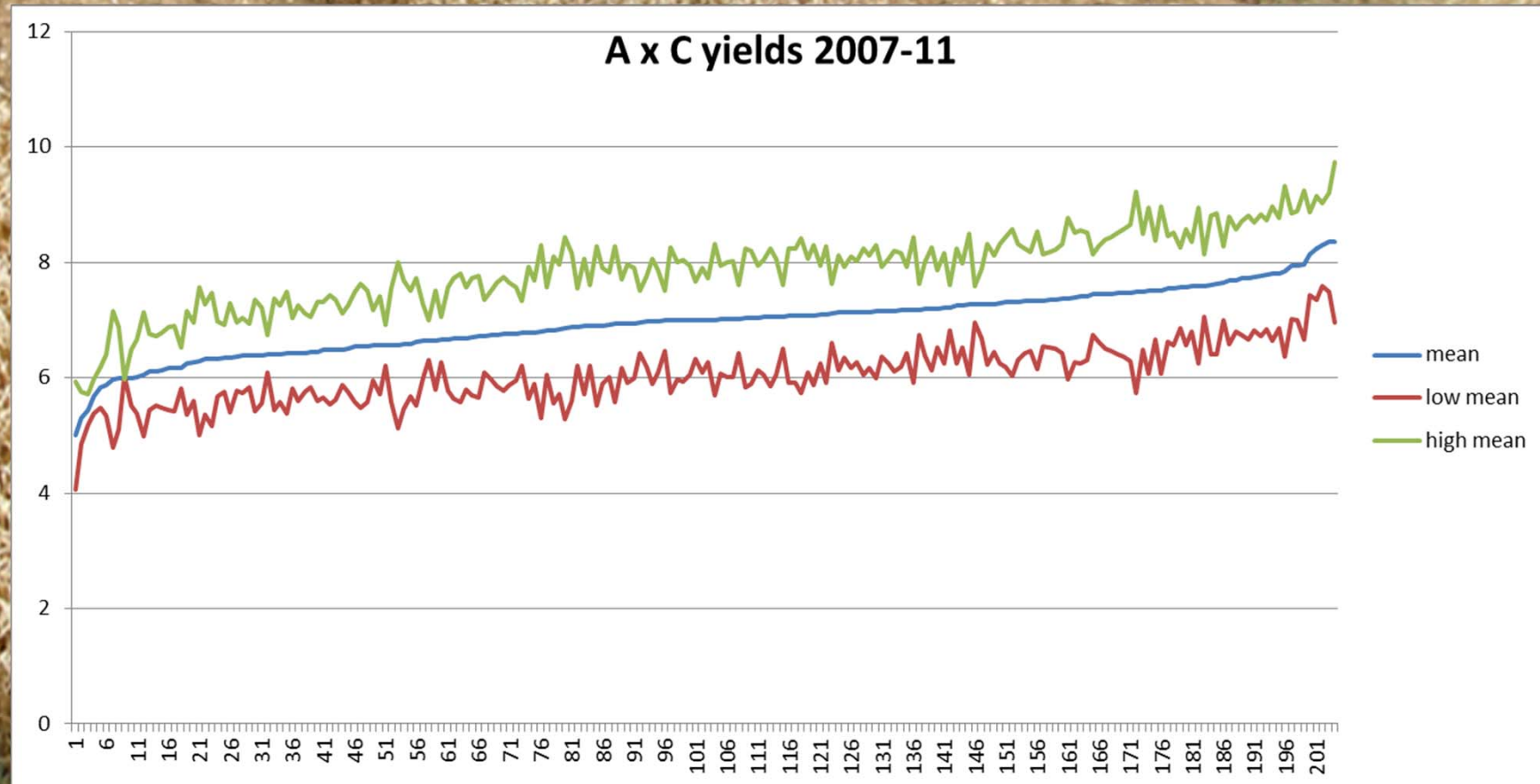


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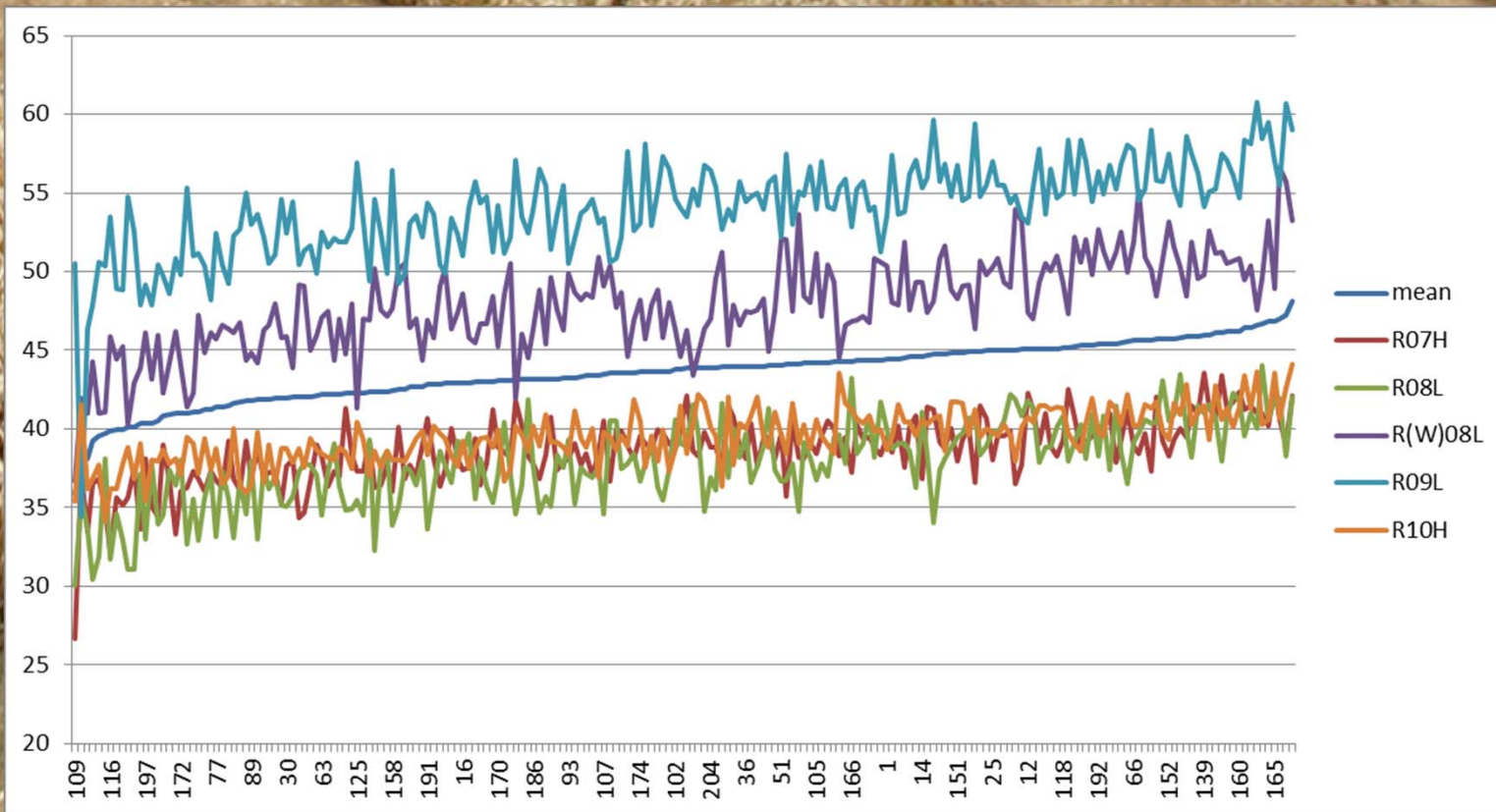
# Height data – all trials



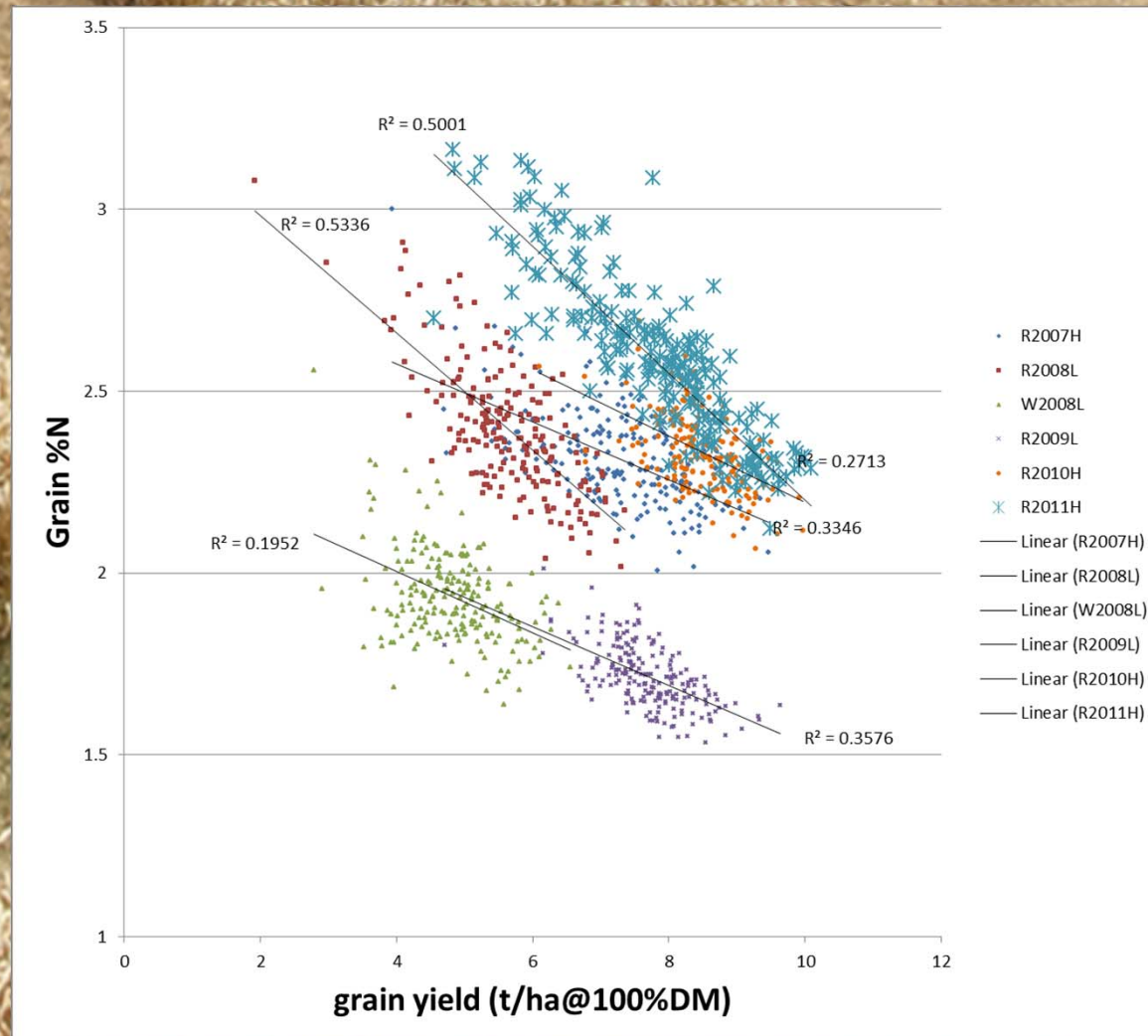
# Yield data – means



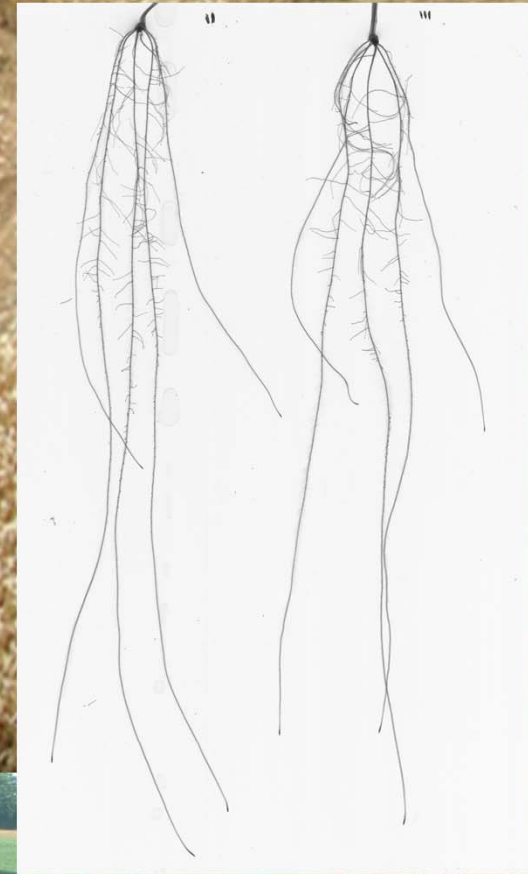
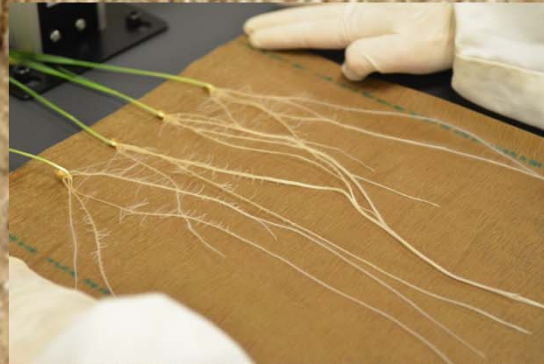
# NUtE (2007-10)



# Grain %N versus yield – all trials



# Variation in root traits: mapping QTLs



# Forward plans....

- 2 years Diversity –varieties now fixed
- NUE/RUE/Take-all interactions to be evaluated
- Addition field trial(s) for Avalon x Cadenza
- Greenhouse N uptake trials for Avalon x Cadenza (one completed)







# Thanks



- RRes Farm staff
- WGIN team
- Group and field team: Peter Barraclough, Caihong Bai, Peter Buchner, Andrew Riche, Yongfan Wan, Saroj Parmar, Janina Jones, Adinda Derkx, + many summer students



WGIN management 21st February 2012

# Drought tolerance

WGIN-2 SG meeting  
Rothamsted Research 21 Feb 2012



## Activity 9, Drought tolerance (2009-14)

- Obj 1. Identify traits for WUE and drought tolerance (DT) in elite winter wheat varieties. (Yrs 1-2)**
- Obj 2. Identify QTLs for WUE and DT traits using one DH pop in an elite background. (Yrs 2-3)**
- Obj 3. Develop one new DH pop for drought research. (Yrs 2-4)**
- Obj 4. Identify novel genes and alleles for WUE and DT using the AE Watkins and Gediflux collections. (Yrs 2-4)**
- Obj 5. Collate a diverse germplasm (cvs, advanced lines) for future association genetics studies. (Yrs 4 -5)**

# WGIN 2 (9.1 Trait Identification)

## WUE trial 2009-10 & 2010-11

Split plot design (3 reps): plot size 1.6 x 12 m

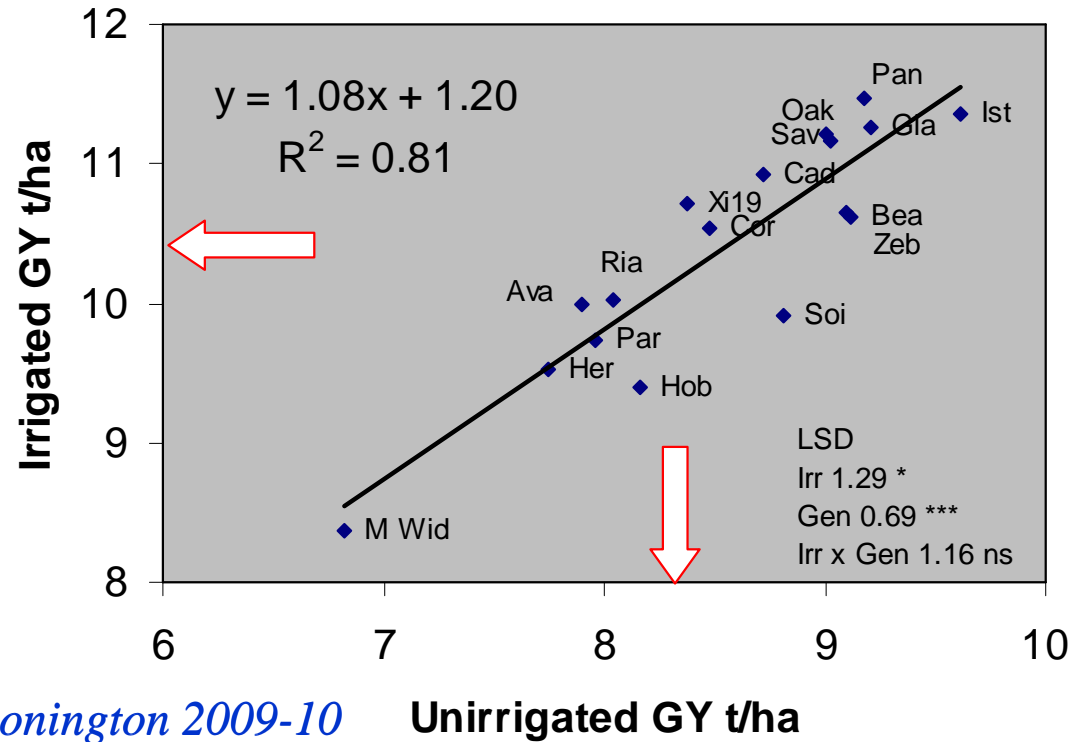
Main plot:           Fully irrigated (trickle irrigation)  
                          Unirrigated

Split plot (variety):

- |                              |                  |
|------------------------------|------------------|
| 1. Avalon *                  | 10. M. Widgeon * |
| 2. Beaver                    | 11. Oakley *     |
| 3. Cadenza *                 | 12. Panorama     |
| 4. Cappelle Desprez/Sterling | 13. Paragon *    |
| 5. Cordiale                  | 14. Rialto       |
| 6. Glasgow                   | 15. Savannah     |
| 7. Hereward *                | 16. Soissons     |
| 8. Hobbit                    | 17. Xi 19 *      |
| 9. Istabraq                  | 18. Zebedee      |

\* Common with NUE trial

## Grain yield responses to irrigation



*Sutton Bonington 2009-10*

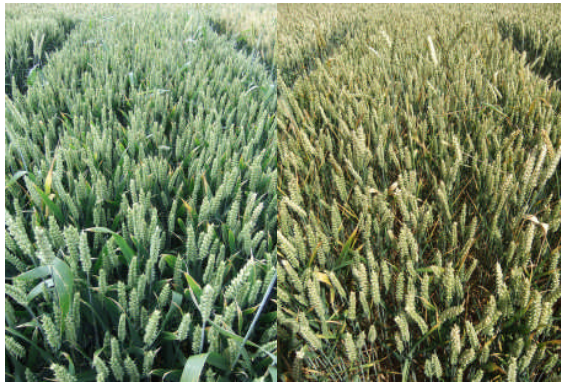
Unirrigated GY t/ha

	Rainfall (mm)	
	2010 LTM 75-09	
Jan	33	54.1
Feb	41.6	43.4
Mar	36	45.7
Apr	24	44.4
May	18.2	45.6
Jun	69.2	58.7
Jul	42.6	49.8

Glasgow Irrigated vs Unirrigated 19 July



# Drought effects 11 July 2011



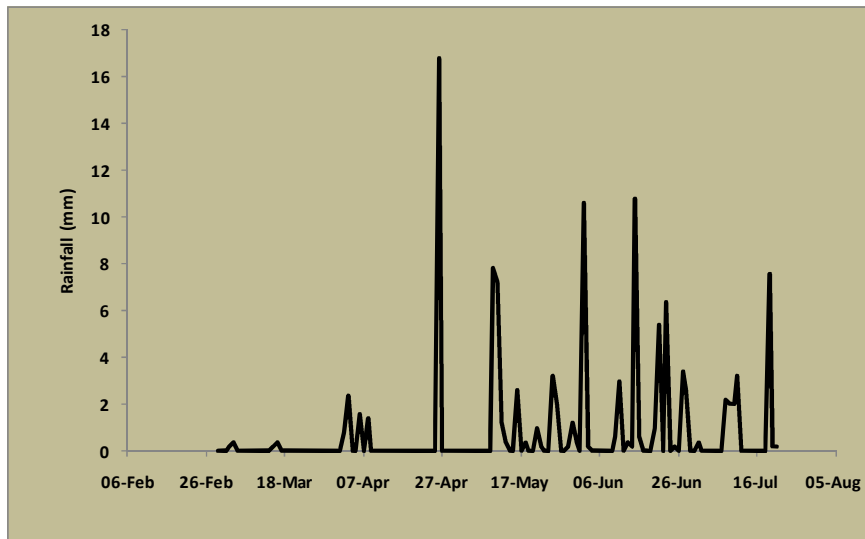
Panorama



Cadenza

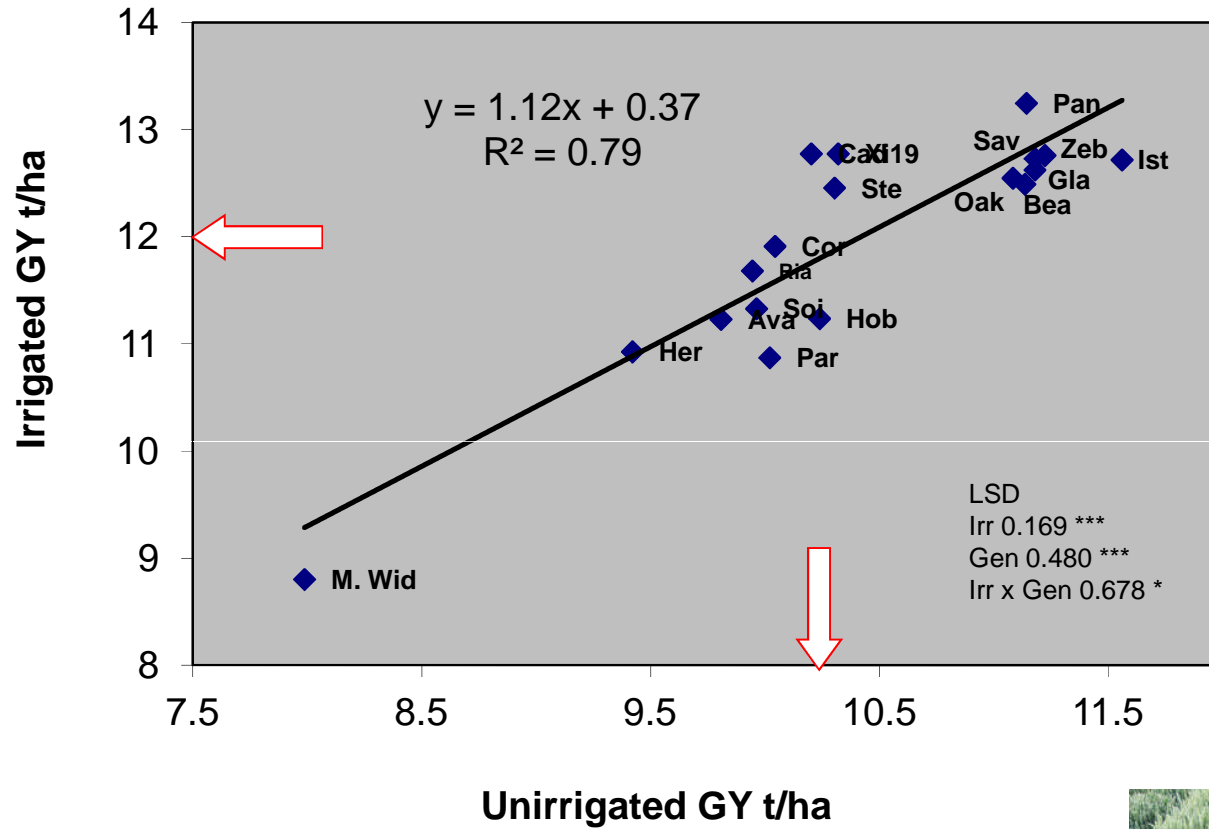


Istabraq



	Rainfal (mm)	LTM
	<b>2011</b>	<b>75-09</b>
<b>March</b>	<b>1.2</b>	<b>54.1</b>
<b>April</b>	<b>23</b>	<b>43.4</b>
<b>May</b>	<b>27.8</b>	<b>45.7</b>
<b>June</b>	<b>45.4</b>	<b>45.6</b>
<b>July</b>	<b>17.8</b>	<b>49.8</b>

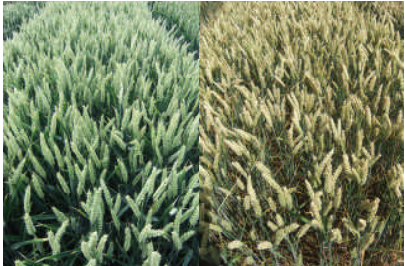
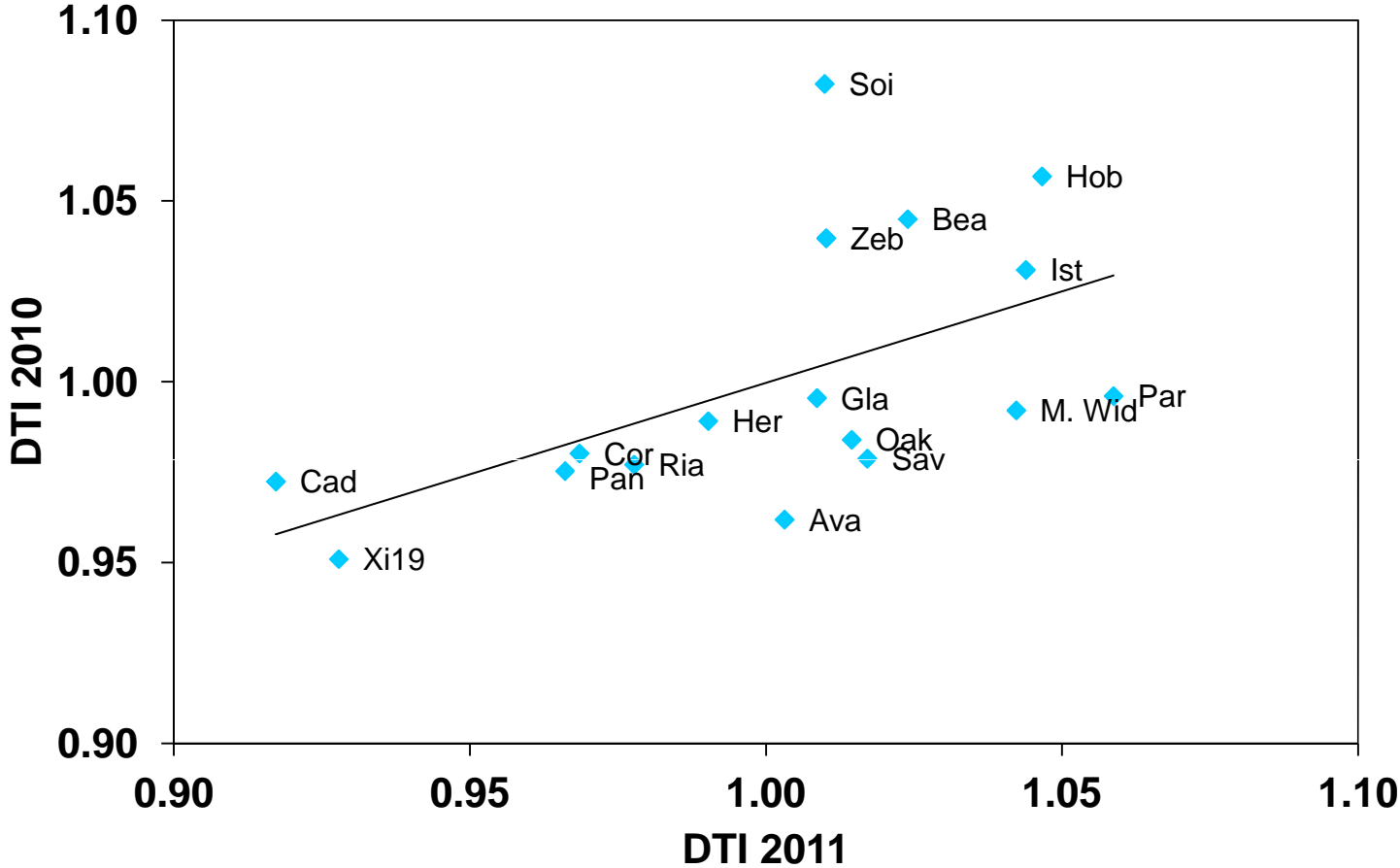
## Grain yield responses to irrigation



*Sutton Bonington 2010-11*



Drought tolerance index:  $(Y_{Dr}/Y_{Irr}) / (\text{mean } Y_{Dr} / \text{mean } Y_{Irr})$

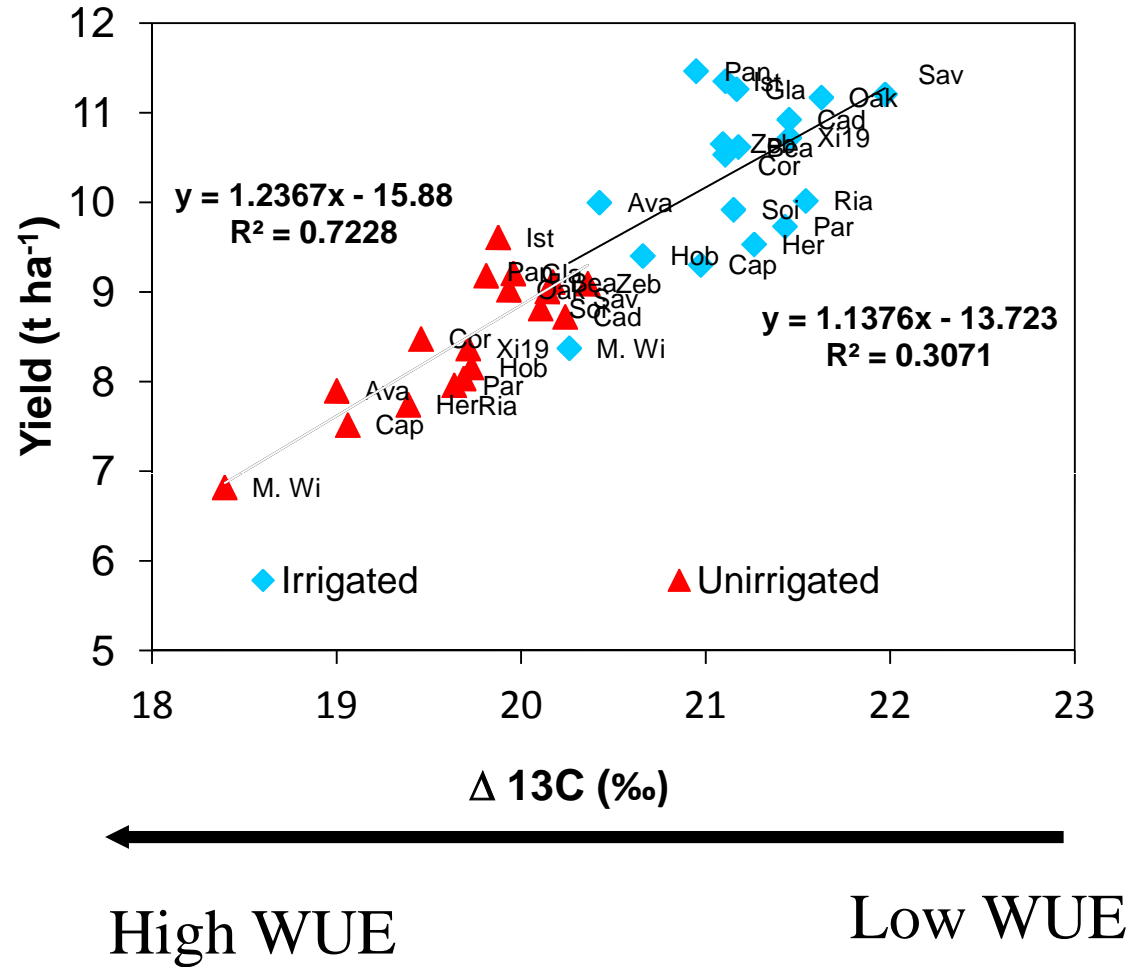




# Water use efficiency: definition and measurement

- **Water-use efficiency (WUE) is the ratio of above-ground dry matter production to evapotranspiration.**
- **$^{13}\text{C}/^{12}\text{C}$  isotope ratio of fixed  $\text{CO}_2$  can be used as an indicator of WUE**
- **Low discrimination against  $^{13}\text{CO}_2 \rightarrow$  high WUE**

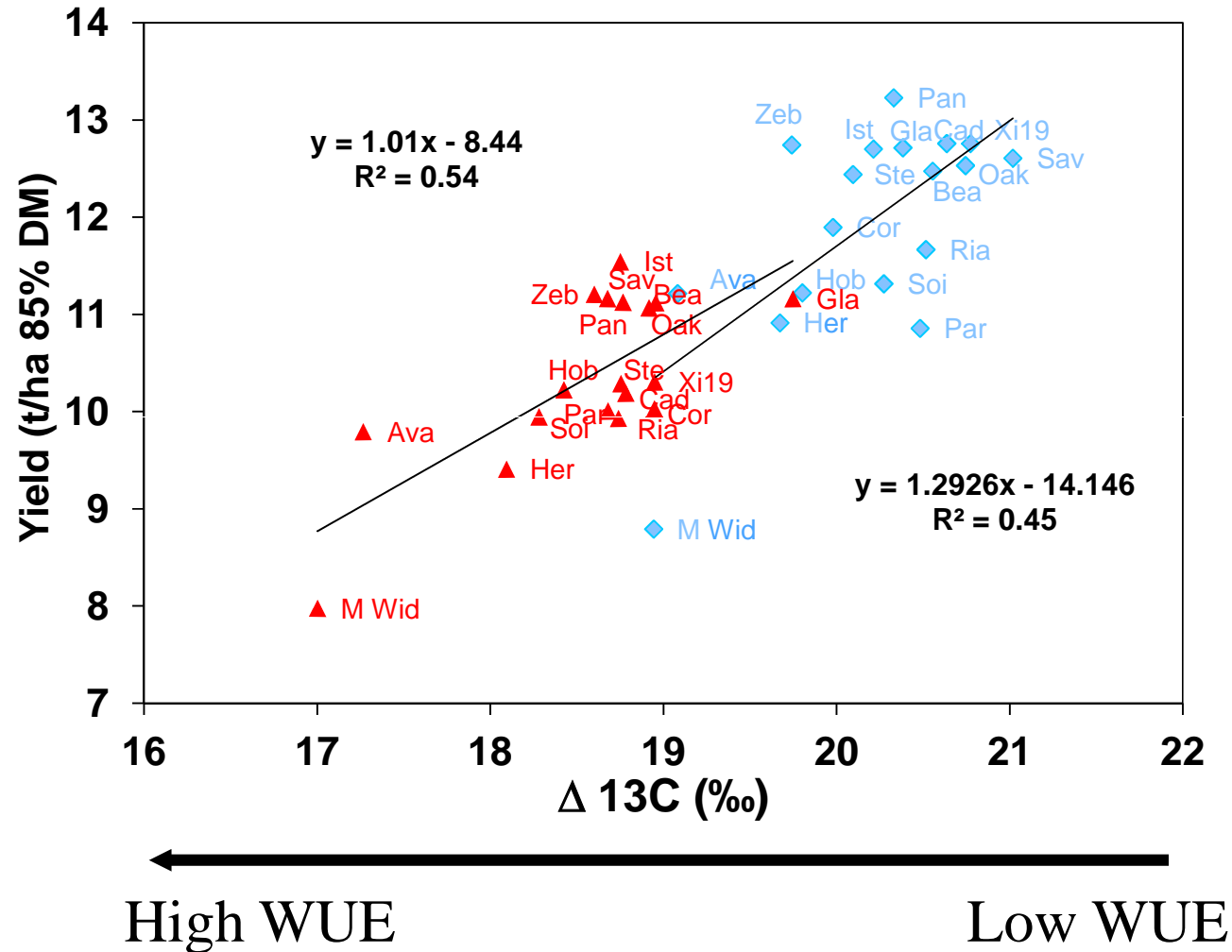
# $\Delta^{13}\text{C}$ vs grain yield in 18 wheat cultivars



*Sutton Bonington 2009-10*



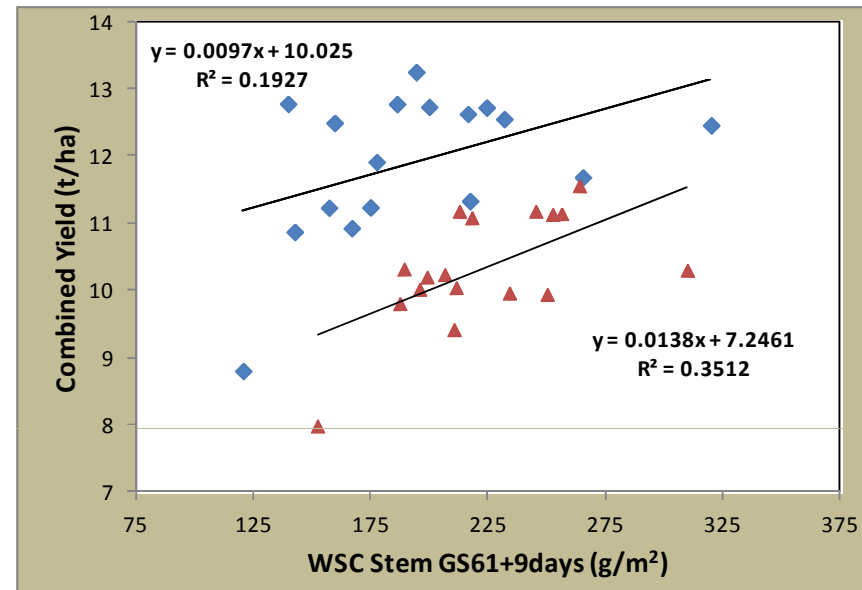
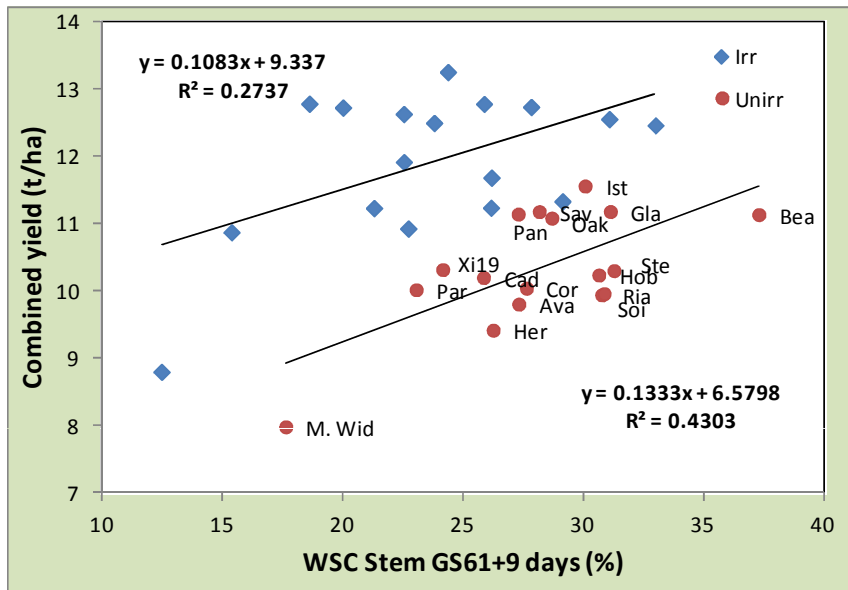
# $\Delta^{13}\text{C}$ vs grain yield in 18 wheat cultivars



# Other traits correlations: 2010-11

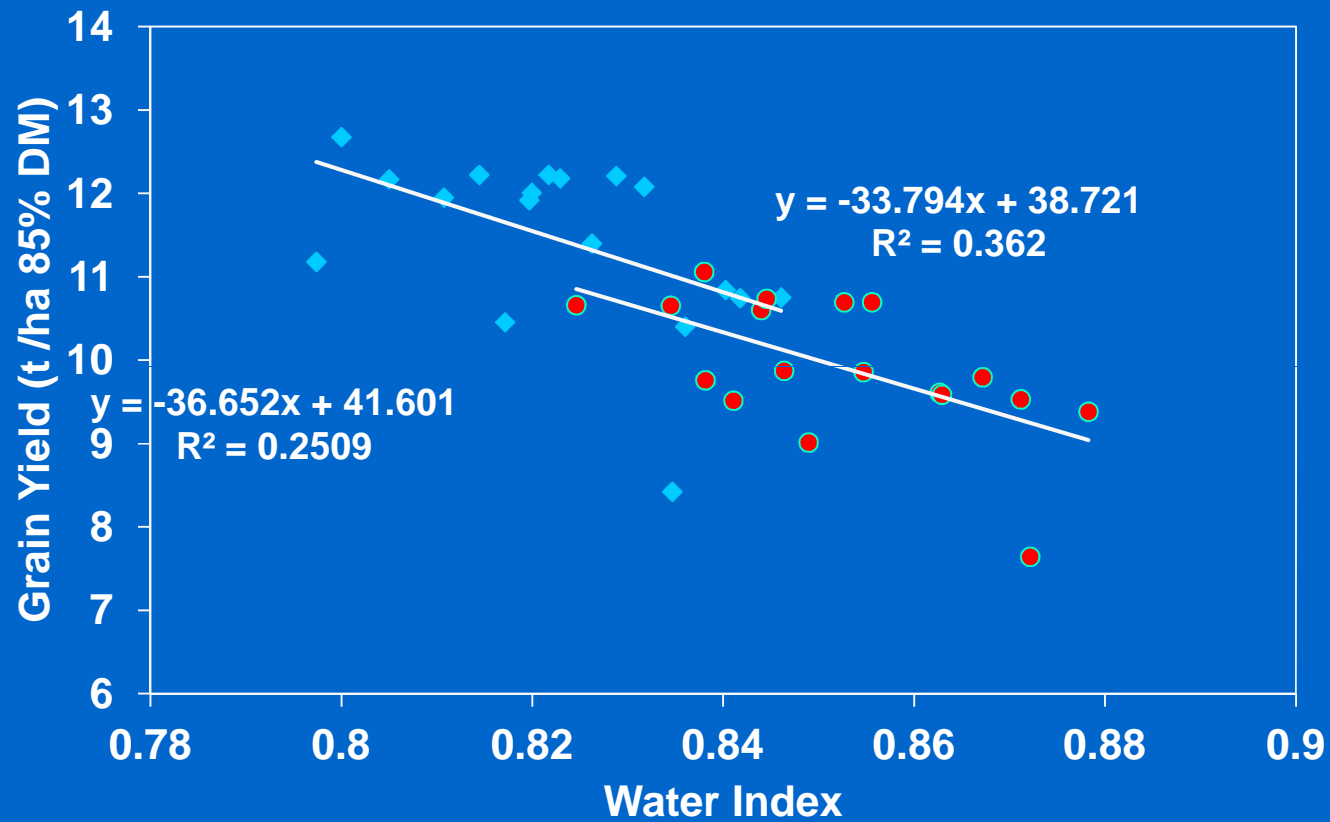


## Grain yield versus stem WSC reserves



*Sutton Bonington 2010-11*

# Spectral Reflectance Index ( $R_{970}/R_{990}$ ): 27 June



# WGIN 2 (9.2 QTL Detection)

2010-11 and 2011-12 expts

- Rialto x Savannah DH population for phenotyping for yield physiological traits (94 lines and 2 parents)
- 2 sites: Nottingham - irrigated & unirrigated; JIC - unirrigated
- Target traits
  - $^{13}\text{C}$   $\Delta$  grain
  - senescence kinetics
  - canopy temperature
  - stem WSC



# Measurements on DH pop

- Combine grain yield, yield components
- % stem WSC at GS61+10d (unirrigated)
- Leaf senescence kinetics for flag-leaf, L2 and L3.
- Canopy temperature
- grain  $\Delta$  13C (unirrigated)
- NDVI



Drought effects 11 July 2011



L2

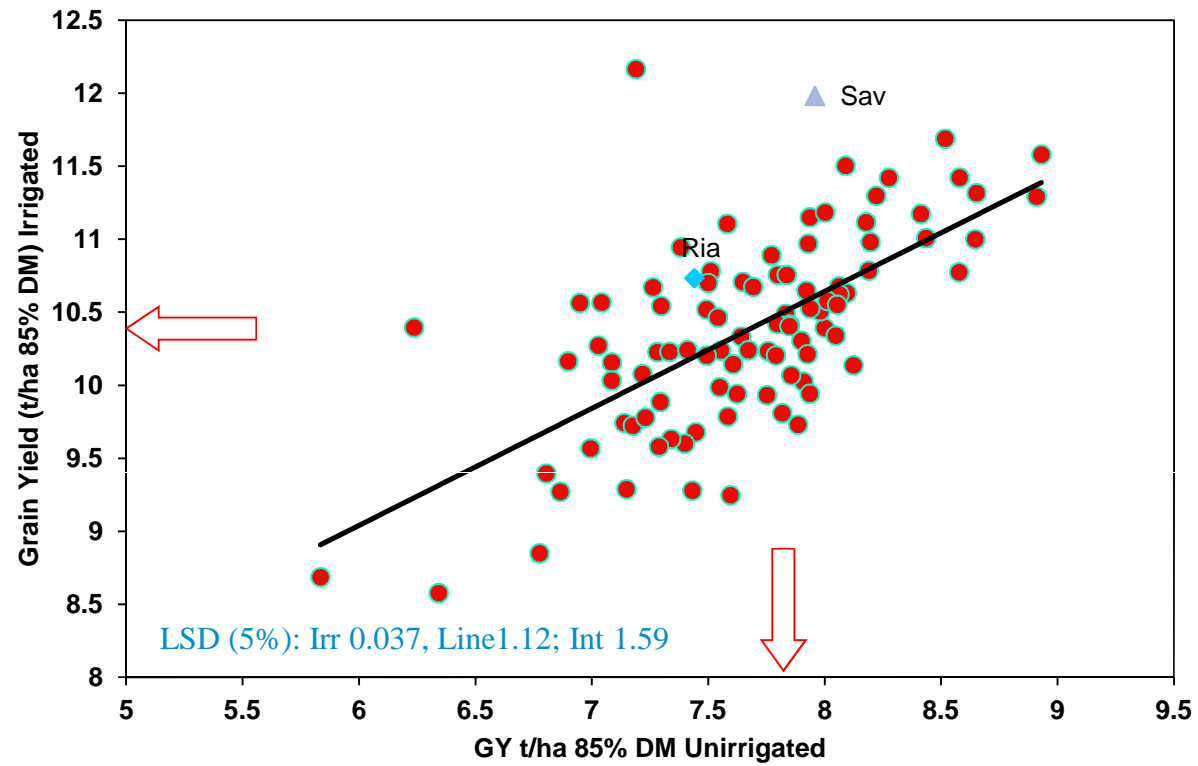


L39



L47

# Grain yield responses to irrigation

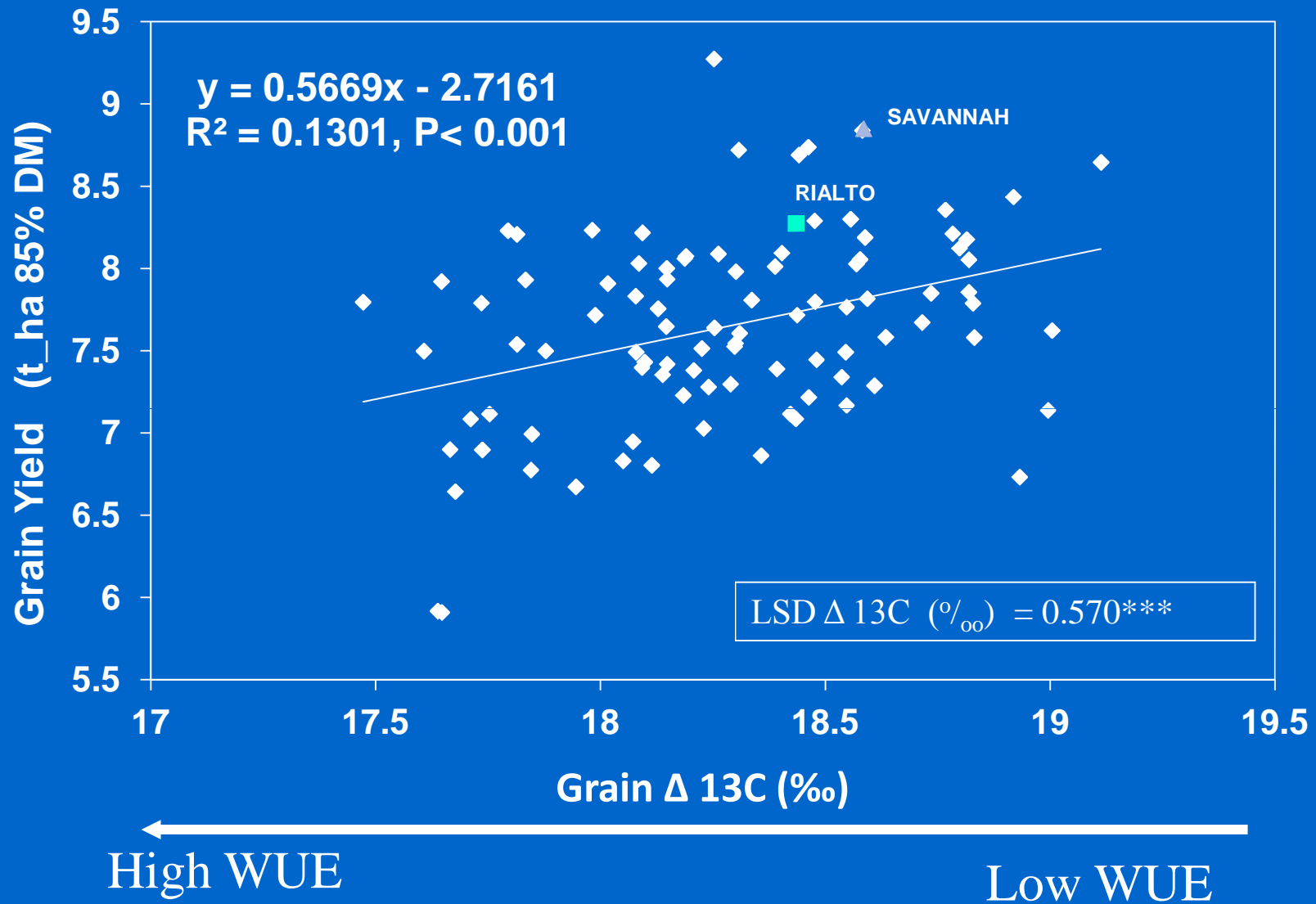


*Sutton Bonington 2010-11*





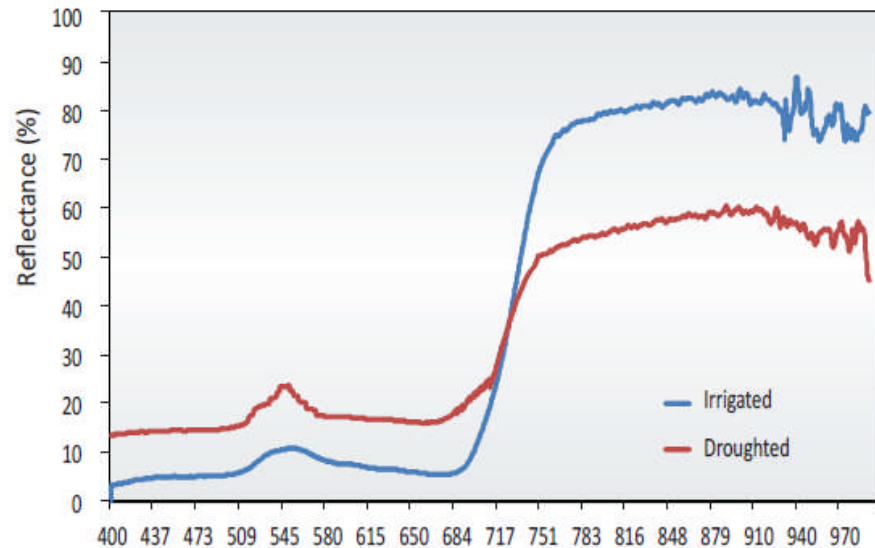
# $\Delta^{13}\text{C}$ vs grain yield: R x S DH (94 lines) 2010-11



*SB2010-11 Unirrigated*

# Spectral reflectance (NDVI)

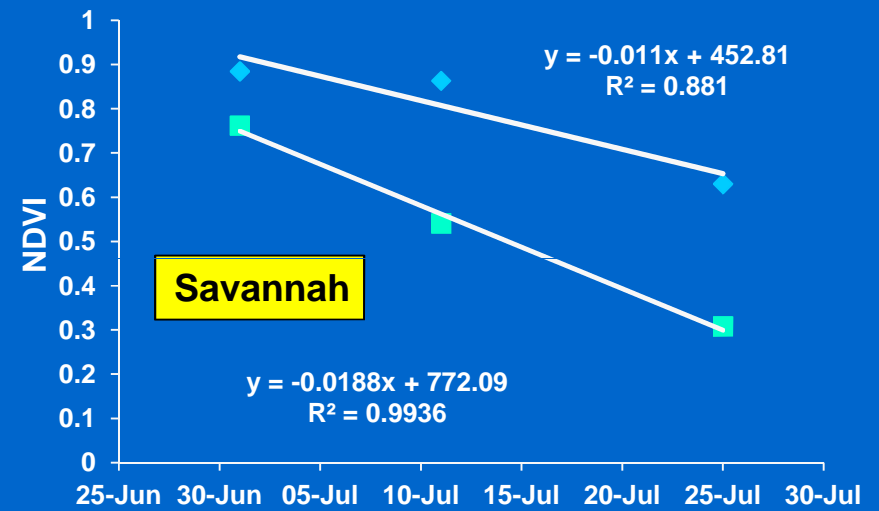
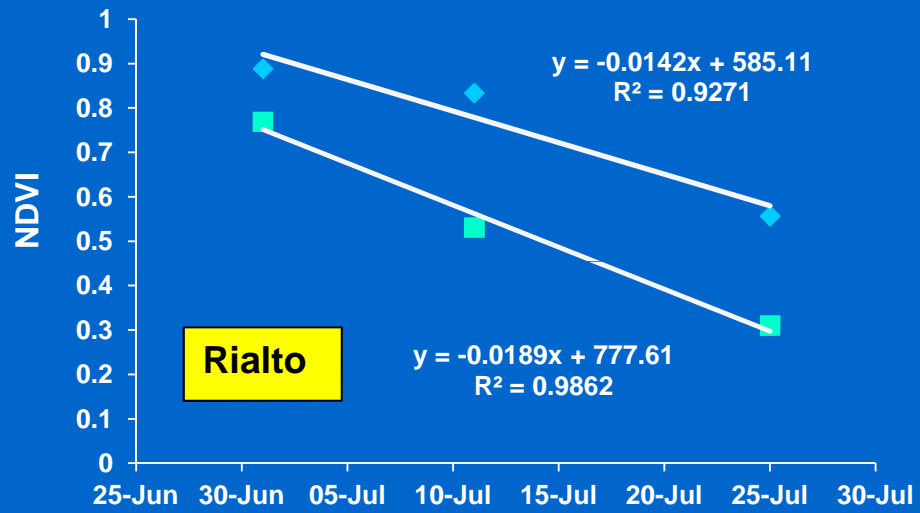
- Shows promise for biomass in some crops
- May be used to estimate chemical components. e.g. water soluble



**Table 7.1. Commonly used spectral reflectance indices (SRI) for wheat canopy analysis where index types are: VI – vegetation index; PI – pigment related index; WI – water index.**

Index	Name	Physiological process	Type	Calculation
NDVI	Normalized difference vegetation index	Green area, photosynthetic capacity, N status	VI	$[R_{900}-R_{680}]/[R_{900}+R_{680}]$
R-NDVI	Red normalized difference vegetation index	Green area, photosynthetic capacity, N status	VI	$[R_{780}-R_{670}]/[R_{780}+R_{670}]$
G-NDVI	Green normalized difference vegetation index	Green area, photosynthetic capacity, N status	VI	$[R_{780}-R_{550}]/[R_{780}+R_{550}]$
SRa	Simple Ratio	Green biomass	VI	$[R_{800}/R_{680}]$ and $[R_{900}/R_{680}]$
RARS <sub>a</sub>	Ratio analysis of reflectance spectra chlorophyll a	Chlorophyll a content	PI	$[R_{675}/R_{700}]$
RARS <sub>b</sub>	Ratio analysis of reflectance spectra chlorophyll b	Chlorophyll b content	PI	$R_{675}/[R_{650} * R_{700}]$
RARS <sub>c</sub>	Ratio analysis of reflectance spectra carotenoid	Carotenoid content	PI	$[R_{760}/R_{500}]$
NPQI	Normalized pheophytinization index	Normal chlorophyll degradation; can be used to estimate phenology, pest and diseases	PI	$[R_{415}-R_{435}]/[R_{415}+R_{435}]$
SIPI	Structural independent pigment index	Senescence related to stress	PI	$[R_{800}-R_{435}]/[R_{415}+R_{435}]$
PRI	Photochemical reflectance index	Dissipation of excess radiation	PI	$[R_{531}-R_{570}]/[R_{531}+R_{570}]$
WI	Water index	Plant water status	WI	$[R_{970}/R_{900}]$
NWI-1	Normalized water index 1	Plant water status	WI	$[R_{970}-R_{900}]/[R_{970}+R_{900}]$
NWI-2	Normalized water index 2	Plant water status	WI	$[R_{970}-R_{850}]/[R_{970}+R_{850}]$
NWI-3	Normalized water index 3	Plant water status	WI	$[R_{970}-R_{880}]/[R_{970}+R_{880}]$
NWI-4	Normalized water index 4	Plant water status	WI	$[R_{970}-R_{920}]/[R_{970}+R_{920}]$

# Post-anthesis NDVI



# Spectral Reflectance Indices vs Grain Yield

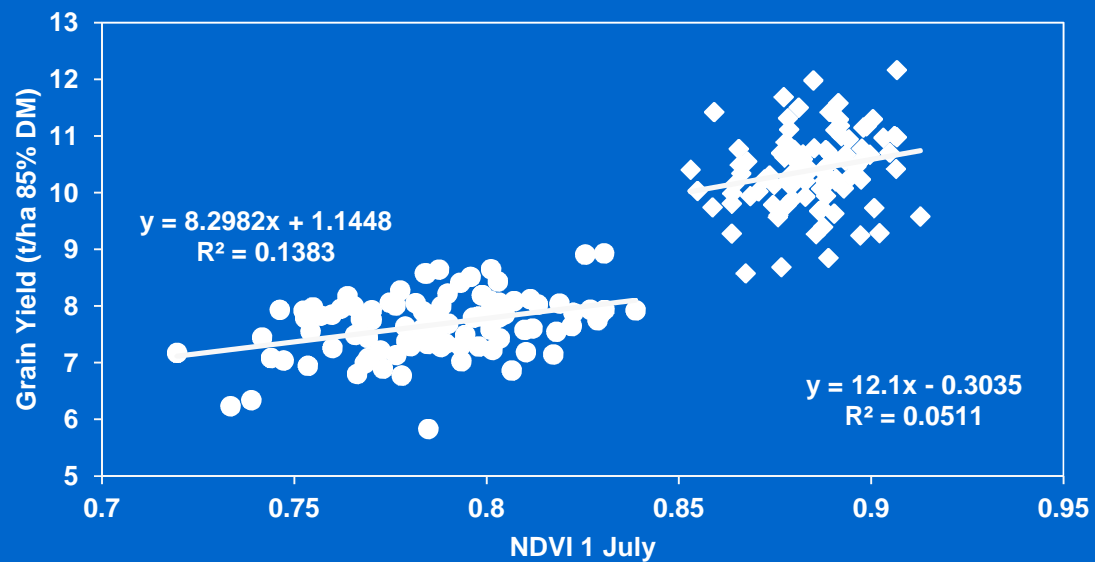
## NDVI

LSD:

Irr 0.074 \*\*\*;

Line 0.061 ns

Int 0.042 ns



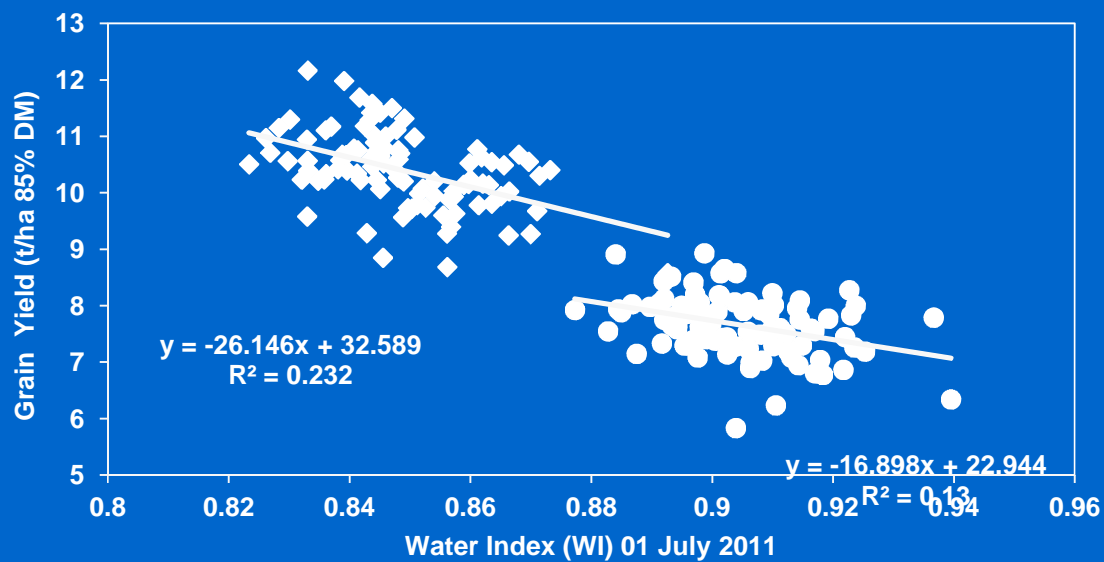
## Water Index

LSD:

Irr 0.003 \*\*\*

Line 0.002 \*\*

Int 0.004 ns



# Preliminary Conclusions

- **Consistent differences in Drought Tolerance Index identified amongst panel of 18 cultivars**
- **Ability to access water appears to be a key driver for productivity under UK drought.**
- **High  $^{13}\text{C}$   $\Delta$  correlated with grain yield under drought. Physiological basis ~ increased stomatal conductance, deeper roots?**
- **Measurement of stable isotopes in plant dry matter may a useful phenotypic tool for speeding up breeding**
  - Grain  $^{13}\text{C}$   $\Delta$
  - Flag leaf  $\Delta$   $^{18}\text{O}$
- **Work is ongoing to:**
  - identify opportunities to break linkage between WU and WUE
  - develop high-throughput screens for breeding
  - Understand the genetic basis of drought tolerance and WUE traits (QTL detection)

# WGIN 2 (Drought tolerance, 9.2)

	Project Month	Milestone
30/11/2012	48	Act 9 Obj2: QTL analysis to identify genome locations associated with WUE and drought tolerance traits completed.

## 2011-12 expts

Rialto x Savannah DH population for phenotyping for yield physiological traits (94 lines and 2 parents)

~ 2 sites: Nottingham - irrigated and unirrigated

JIC - unirrigated

~ extend molecular map as necessary with Dart and SSR

# WGIN 2 (9.3 Develop new SSD pop)

	Project Month	Milestone
31/03/2012	40	Act 9 Obj3. Complete development of one new SSD population in an elite modern background segregating for drought-tolerance traits.

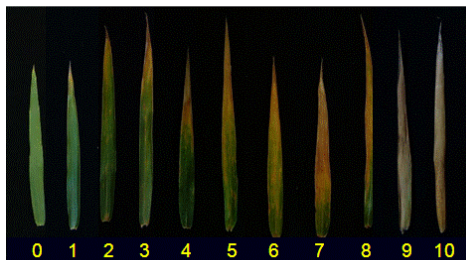
- Candidate F1(s) made at JIC informed by data analysis from LK0986 project
- Population segregating for *Ppd1a*:
  - use WGIN resource to select against *PpdD1a* - ie make the pop photoperiod sensitive.
  - keep the pop large, so we can have flowering time strata and perform analysis within them
- F2 sown in October 2011, population is in excess of 350 lines



# WGIN 2 (9.4 Association Genetics)

	Project Month	Milestone
31/03/2012	40	Act 9 Obj3. Complete development of one new DH population in an elite modern background segregating for drought-tolerance traits.

**120 Watkins lines assessed for leaf Ps rate (quantum yield, fluorpen) and visual senescence scores (weekly post anthesis) in 2010-11**



# WGIN 2 (Activity 9, Drought tolerance)

	Project Month	Milestone
30/11//2011	36	Act 9 Obj1: Complete phenotyping and data analysis for drought tolerance traits in elite winter wheat varieties in 2009/10 &10/11.
30/11/2012	48	Act 9 Obj2: QTL analysis to identify genome locations associated with WUE and drought tolerance traits completed.
31/03/2012	40	Act 9 Obj3. Complete development of one new DH population in an elite modern background segregating for drought-tolerance traits.
28/02/2013	51	Act 9 Obj4: Association genetics analysis of drought tolerance traits using AE Watkins & Gediflux collections completed.
28/02/2013	51	Act 9 Obj5: Collation of diverse germplasm collection (cultivars, advanced lines) from worldwide drought-tolerance wheat breeding programmes completed.

# WGIN Sub-Contract Update

## *Chemical analysis in WGIN drought tolerance trial in 2009/10 & 2010/11*

Samples (per year) on 18 cultivars x 2 Irr trts x 3 reps  
(=108 plots)

	Chemical analysis			Total
No. samples	13C	18O	Ash%	
Flag leaf @ GS61	108	108		216
Grain @ harvest	108		108	216

***Core funding***

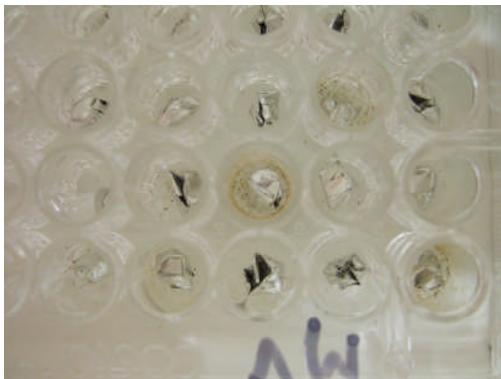
***Sub-contract funding***

# Isotope analysis, sample submission: :Weighing and encapsulate of milled samples samples for $^{13}\text{C}$ and $\text{O}^{18}$ isotope analysis

## Encapsulation of 2 mg sample



## Submit samples to Mylnefield Isotope lab at JHI



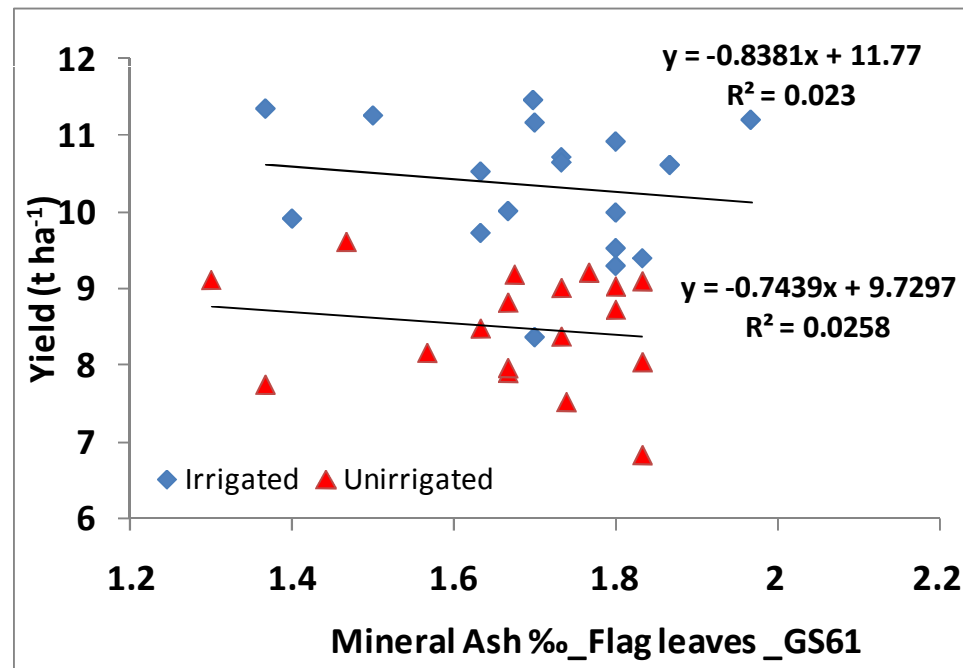
All 2009/10 samples submitted 25 Jan 2011 (data returned Mar 2011)

2010/11: 18 cv grain  $^{13}\text{C}$  and S x R DH grain  $^{13}\text{C}$  submitted to lab on 5 Jan 2012 (data returned Feb 2012)

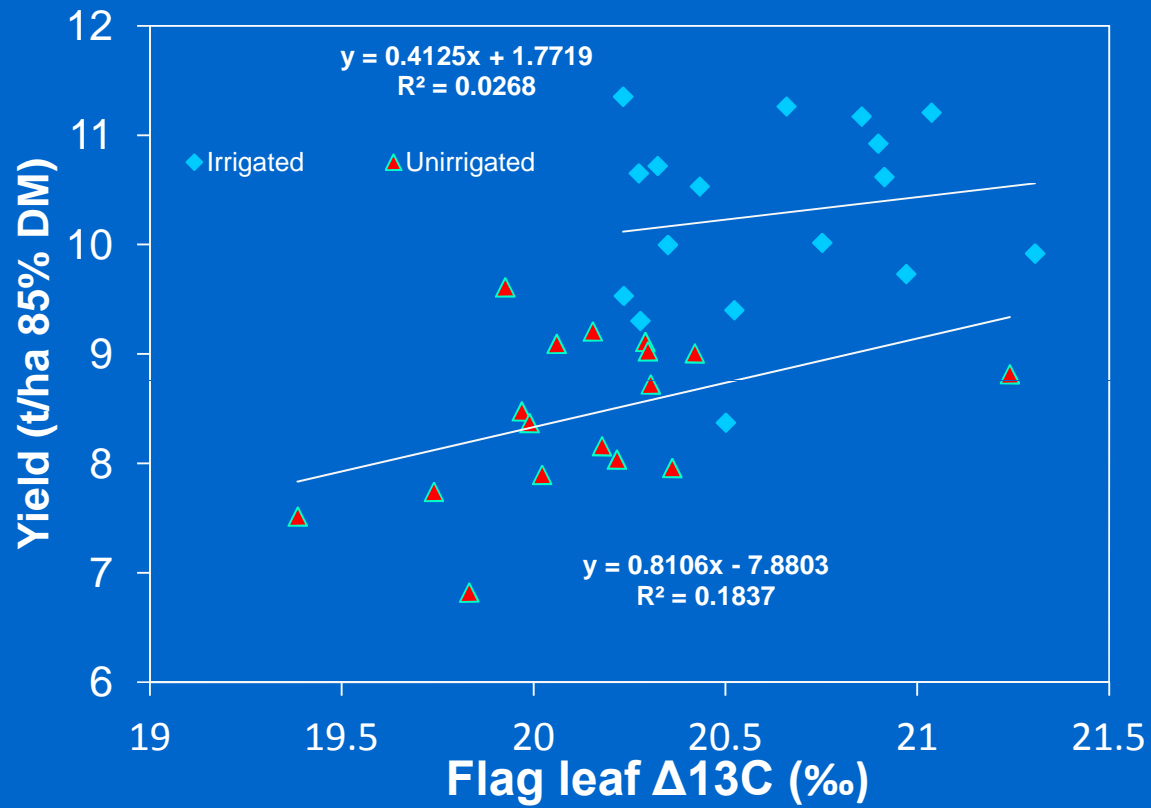
2010/11: 18 cv FL  $^{13}\text{C}$  and  $^{18}\text{O}$  submitted on 15 Feb 2012

## Mineral ash content ~ water use

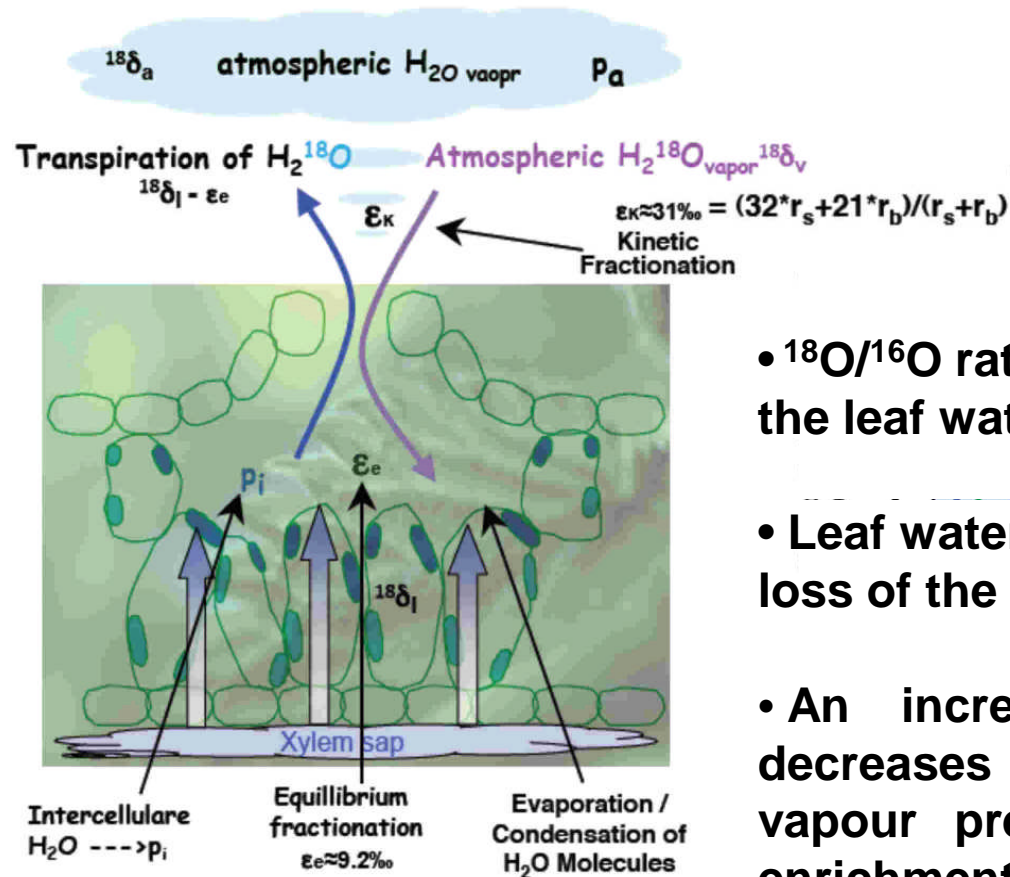
- Total leaf ash content of plant tissues is suggested as a useful tool to predict yield under drought.
- The mechanism of mineral accumulation in plant tissues appears to be explained through the passive transport of minerals via xylem driven by transpiration.
- Ash content measured provides an indicator of the total water transpired.
- Analysis of mineral ash content is less expensive than  $\Delta^{13}\text{C}$  or  $\Delta^{18}\text{O}$ .



# Flag leaf $\Delta^{13}\text{C}$ vs Grain Yield 2009-10



# Techniques: Oxygen isotope ratio ~ leaf transpiration



- $^{18}O/^{16}O$  ratio determined by enrichment in the leaf water due to transpiration.
- Leaf water enriched due to the preferential loss of the lighter  $H_2^{16}O$  during evaporation.
- An increase in stomatal conductance decreases leaf  $T^\circ C$  (hence intercellular vapour pressure) resulting in less  $H_2^{18}O$  enrichment at the evaporating site.

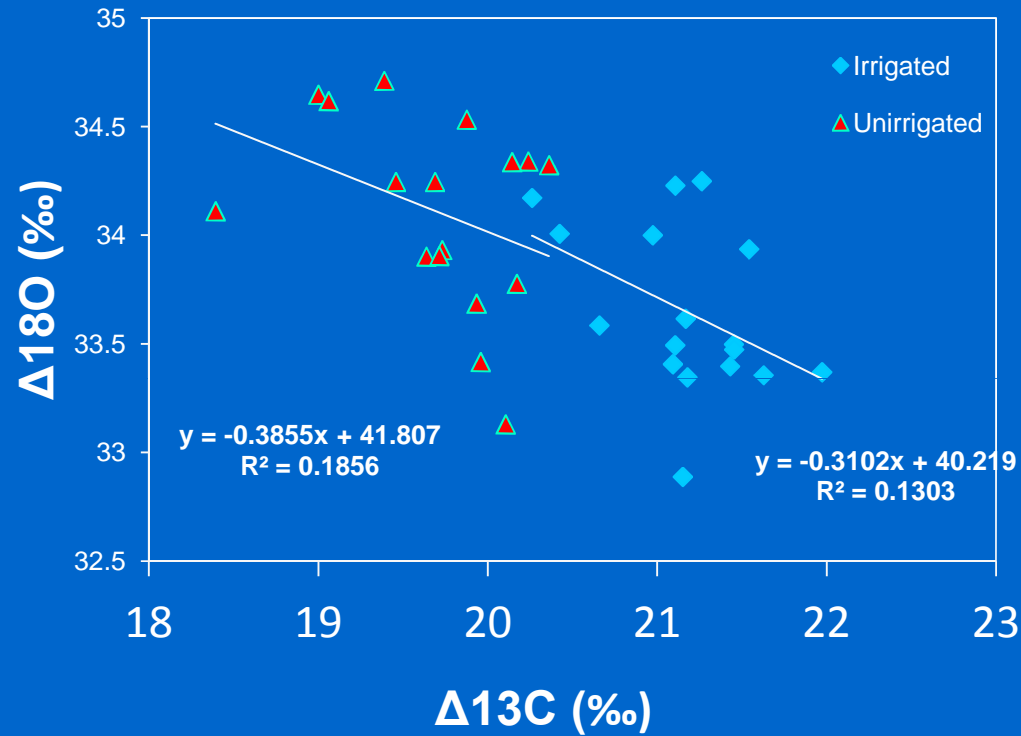


# Flag leaf $\Delta^{13}\text{C}$ vs Flag leaf $\Delta^{18}\text{O}$ 2009-10

Low Transp.



High Transp.



High WUE

Low WUE

# Acknowledgments:



## Nottingham

J. DeSilva

J. Alcock

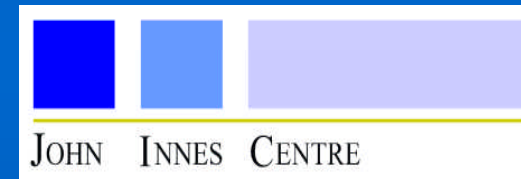
M. Tovey

## JIC

S. Griffiths

S. Orford

L. Wingen



# Resistance to Take-all Resistance to Septoria

**Richard Gutteridge**  
**Kostya Kanyuka**  
**Kim Hammond-Kosack**



**WGIN MM@RRes**  
**21<sup>st</sup> Feb 2012**

# Take-all disease – soil-borne fungus

In 1<sup>st</sup> wheats - no disease problem



A major problem  
for 2<sup>nd</sup> / 3<sup>rd</sup> wheat crops

**2<sup>nd</sup> wheat syndrome**



Typical take-all patch showing stunting  
and premature ripening of the crop  
- in dry years even lower disease levels  
can causes patches

The risk of take-all is largely dependent on the amount of fungal inoculum in the soil at the time of sowing

# Objective 10 - Talk Outline

---

- **Hexaploid wheat – Watkins / Gediflux collections**  
**Diploid wheat**
  - field evaluation to identify potentially resistance genotypes
- **Diploid wheat**
  - mapping populations
  - introgression to *T. aestivum*
- **Hexaploid wheat – inoculum build-up in 1<sup>st</sup> wheats – 2010, 2011**
  - results so far / current activities

# Watkins / Gediflux Hexaploid wheat collections

## Take-all assessments

---

3<sup>rd</sup> wheat situation

**2008 and 2009 field season – one plot / genotype –**  
established from 45 seeds

**823 lines** Watkins + **60 lines** Gediflux collection

**Method - Discarded all lines more susceptible than Hereward**

**2010 field season - one plot / genotype**

**504 Watkins lines** and **36 Gediflux lines**

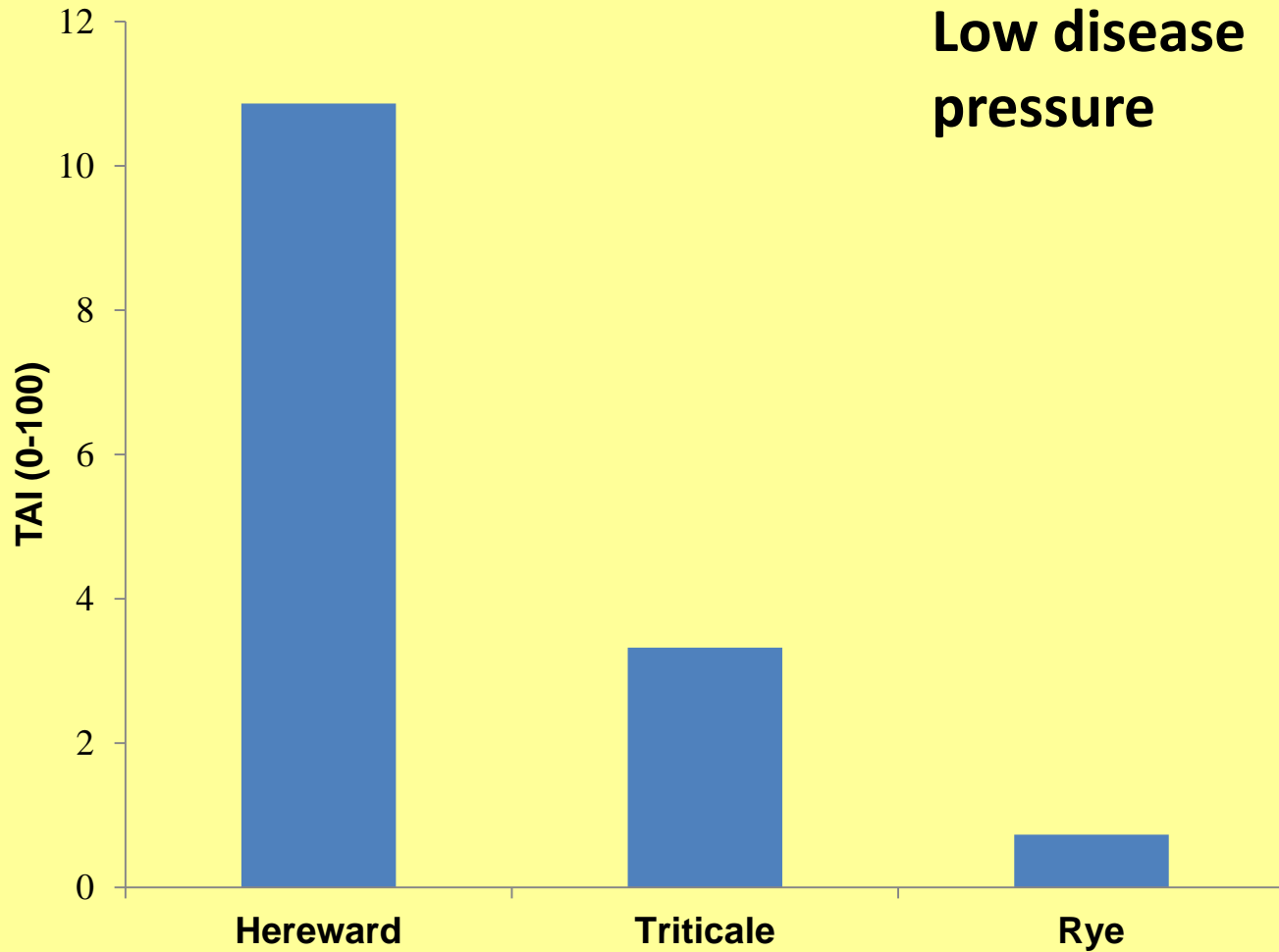
5 blocks of 8 controls + 20 Hereward control plots.

**2011 field season - one plot / genotype**

**490 Watkins lines** and **36 Gediflux lines**

**All roots assessed**

# Watkins field experiment 2011 – Control plots



# Watkins / Gediflux Hexaploid wheat collections

## Take-all assessments

---

3<sup>rd</sup> wheat situation

**Field experiment 2012: one plot / genotype**

**104 Watkins lines, 36 Gediflux lines**

**5 blocks of 8 controls, 20 Hereward plots.**

**Sown October 2011.**

**Plus the most resistant 144 lines Watkins lines have gone into the wheat pre-breeding LoLa field trials - 5 reps / genotype**

**30% of Watkins lines (248 lines) remaining after 4 years of trialling**

**Protocol change**

**2011 - hand sowing 60 seeds per plot (3 rows x 50cm)**

**2012 - combined drilling 200 seeds (4 rows x 1m) - WORKED WELL**

**Feb 2012 - All plots have full plant cover**

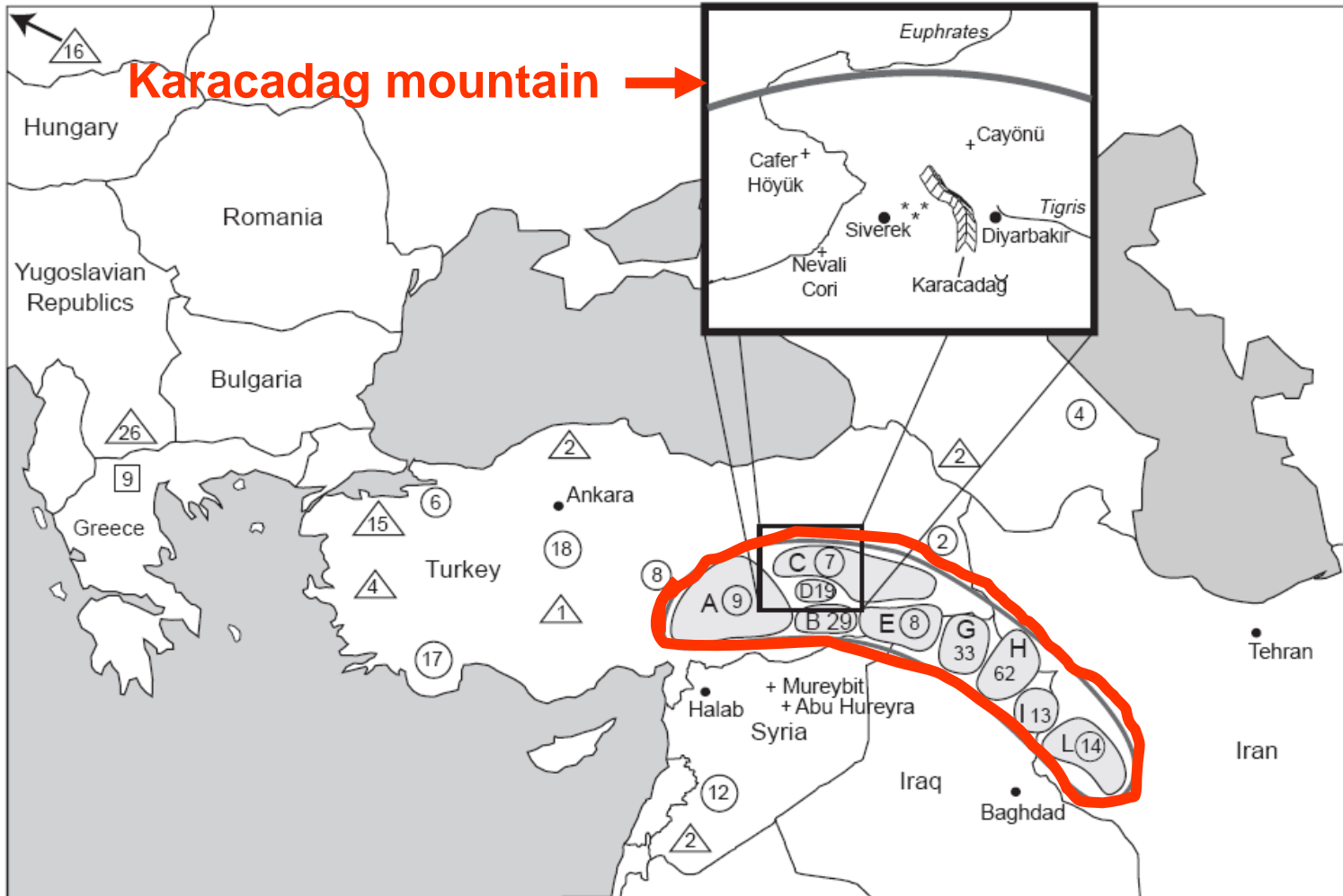


# Objective 10 - Talk Outline

---

- Hexaploid wheat – Watkins / Gediflux collections  
Diploid wheat
  - field evaluation to identify potentially resistance genotypes
- Diploid wheat – *T. monococcum*
  - mapping populations
  - introgression to *T. aestivum*
- Hexaploid wheat – inoculum build-up in 1<sup>st</sup> wheats – 2010, 2011
  - results so far / current activities

# *T. monococcum* (AA genome) domesticated from *T. boeoticum*



**—** Limits of Fertile Crescent

\* Sampling of Karacadağ lines

+ Archeological site

○ *T. m. boeoticum*

△ *T. m. monococcum*

□ *T. m. aegilopoides*

(with number of samples)

A–L: areas of wild *T. m. boeoticum* sampling in the Fertile Crescent

# Diploid wheat field experiment 2008

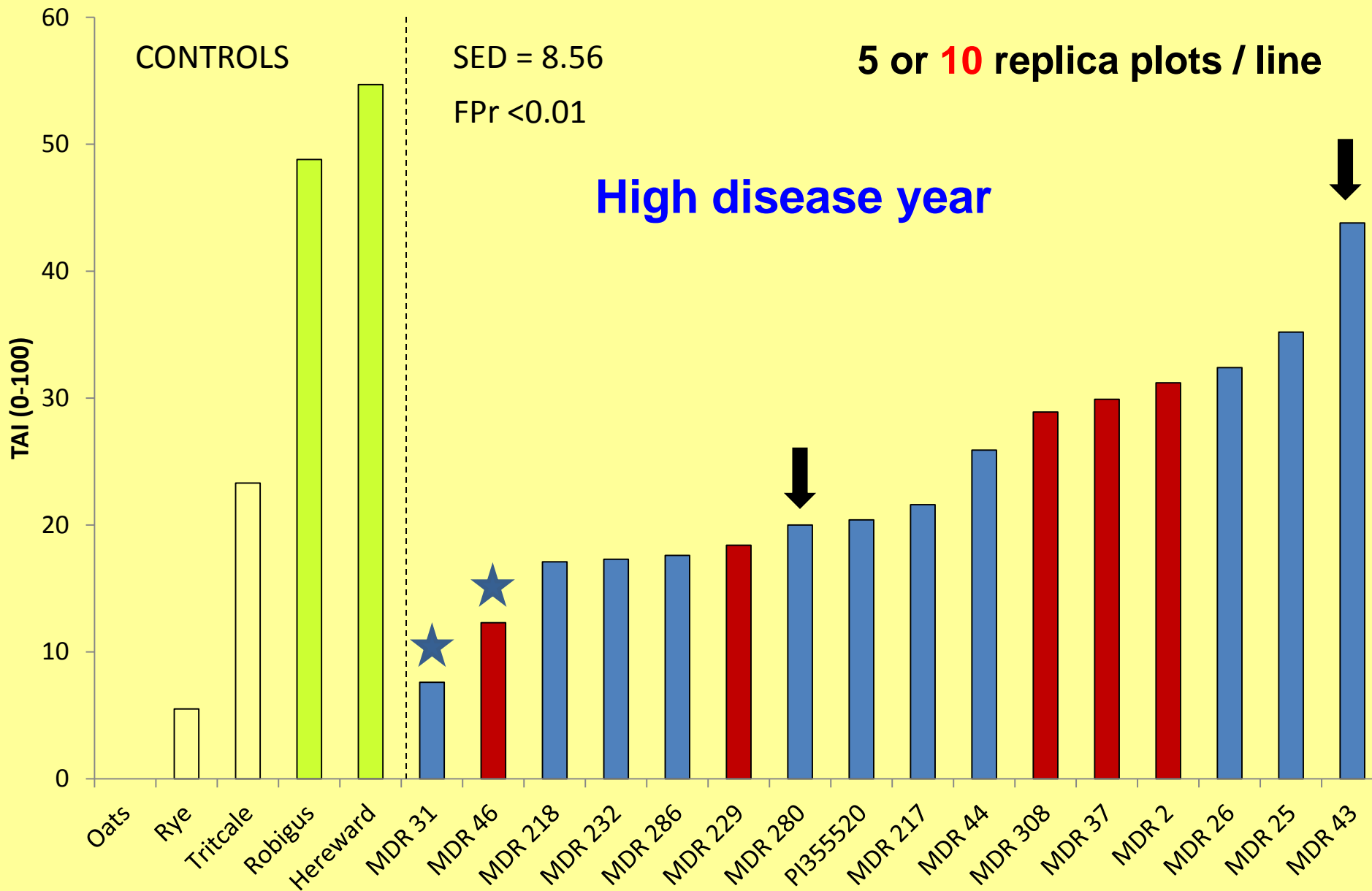
CONTROLS

SED = 8.56

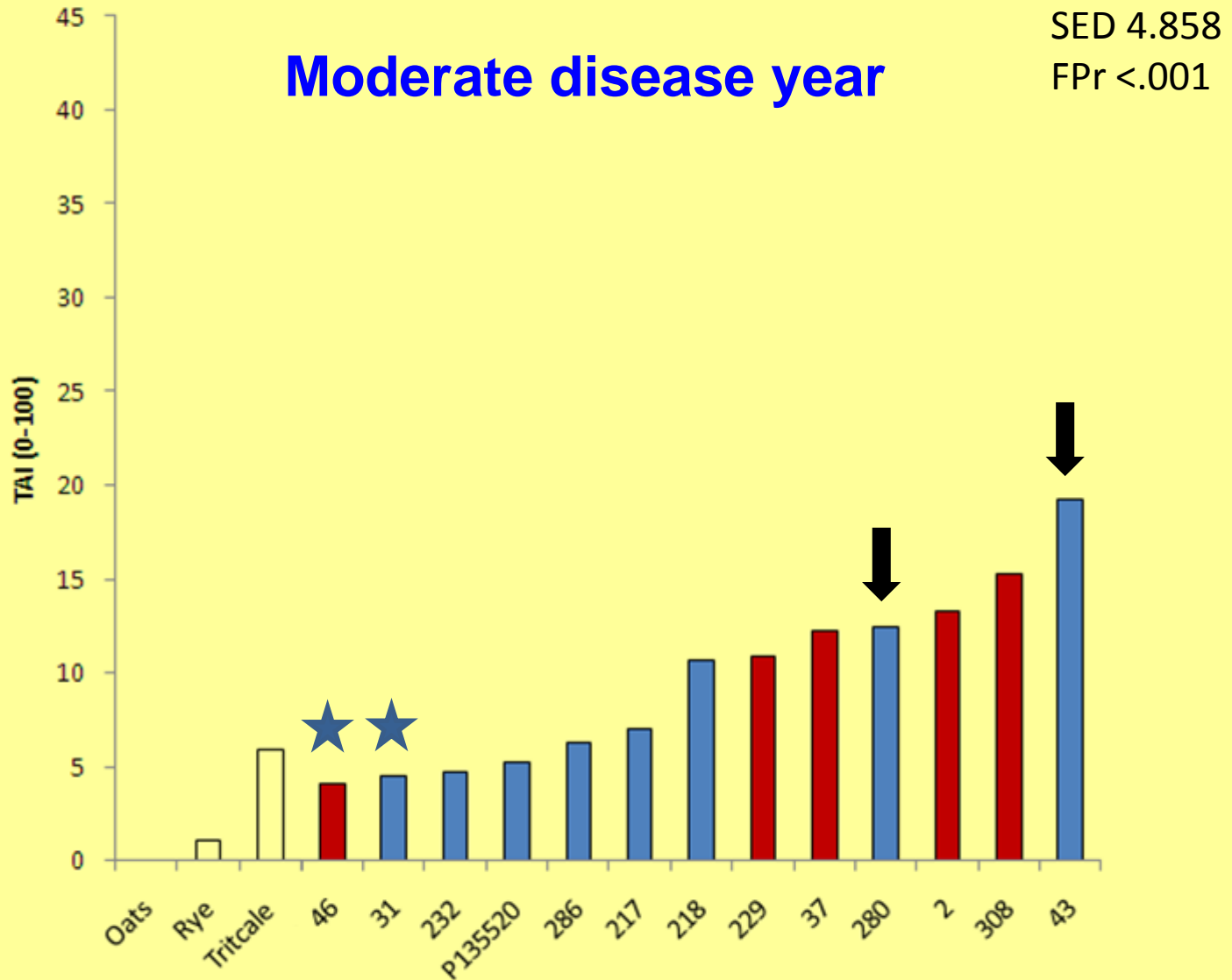
FPr < 0.01

5 or 10 replica plots / line

High disease year

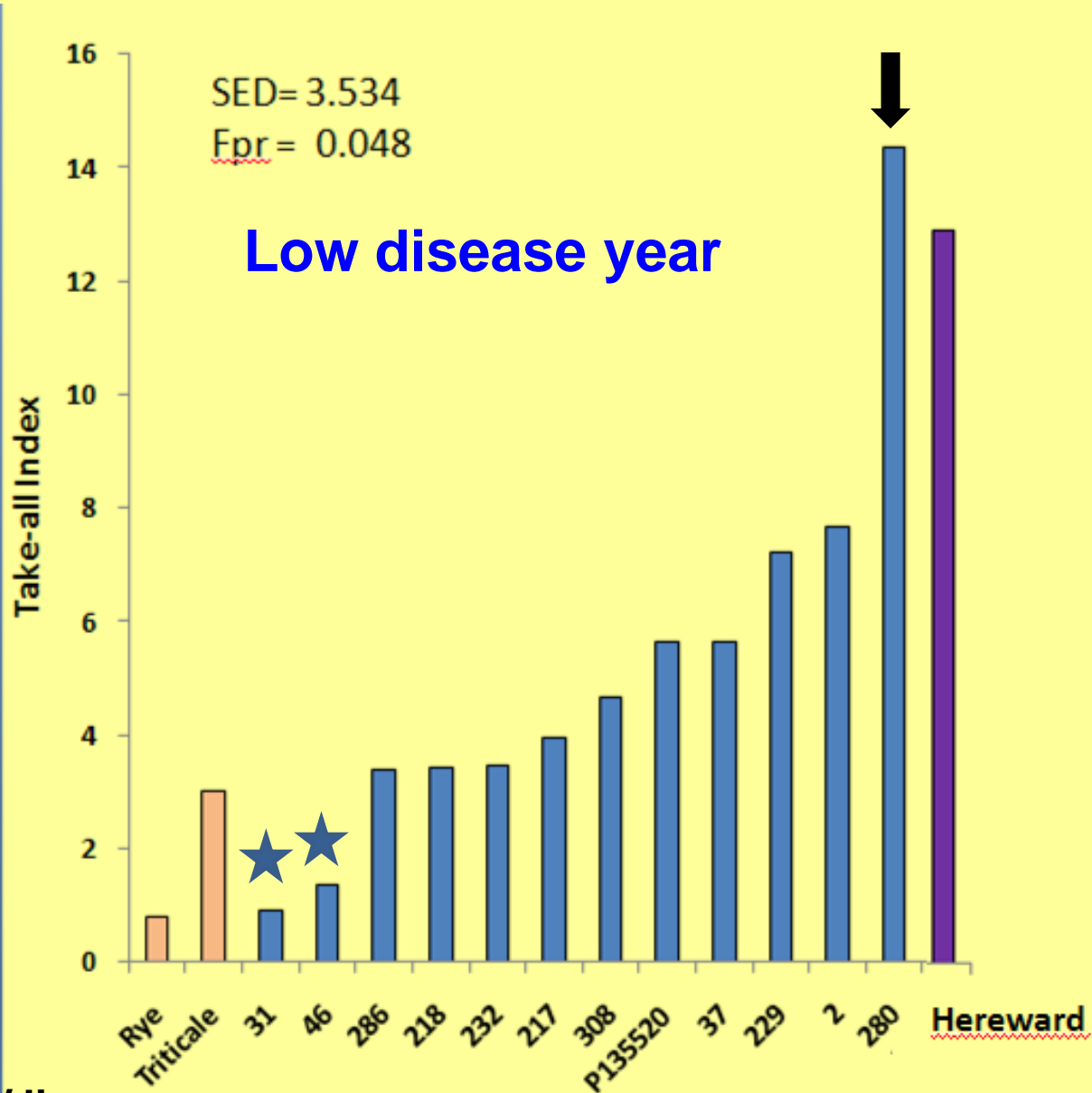


# Diploid wheat field experiment 2010



10 replica plots / line

# Diploid wheat field experiment 2011



10 replica plots / line

# ***T. monococcum* introgression to *T. aestivum***

---

## **New strategy**

### **1. Graham Moore ( JIC)**

#### **New *T. aestivum* sources**

**Paragon** - *ph1* mutant and wild-type  
**Chinese Spring** - *ph1* mutant and wild-type

**Received 10 seed from JIC in July 2011  
and multiplying up the stock**

**To sow in batches starting late Feb to ensure good  
*Tm* pollen production**

# Objective 10 - Talk Outline

---

- Hexaploid wheat – Watkins / Gediflux collections  
Diploid wheat
  - field evaluation to identify potentially resistance genotypes
- Diploid wheat – *T. monococcum*
  - mapping populations
  - introgression to *T. aestivum*
- Hexaploid wheat – inoculum build-up in 1<sup>st</sup> wheats – 2010, 2011

# A key discovery in WGIN 1

The genotype of the 1<sup>st</sup> wheat influence the amount of take-all inoculum build-up in the bulk soil



Diversity trial – 200 kg N / plots

8 soil cores / plot – just post harvest

Pot bioassay to assess Take-all Infectivity

Take-all inoculum build up ranking by varieties:  
1 = Low – 9 = High

Variety	2004	2006	2007	2008	Overall ranking
→ Cadenza	1	2	1	1	1.25
Xi19	7	1	2	2	3
→ Riband	5	4	4	3	4
Mercia	3	5	3	6	4.25
Monopol	6	7	5	4	5.5
→ Avalon	nd	6	8	5	6.33
Soissons	8	3	6	9	6.5
Malacca	4	9	7	7	6.75
→ Hereward	9	8	9	8	8.5



# Hexaploid wheat – inoculum build-up in 1<sup>st</sup> wheats – 2010 , 2011

---

**Avalon x Cadenza mapping population = 203 lines  
3 REPLICA BLOCKS**

**2009 / 2010 : no inoculum build up due to the very dry winter / spring**

**2010 / 2011 – cores taken from all plots  
+ entire trial over-sown with cv Oakley**

**BLOCK 1 – just out of the pot bioassay - more promising levels of disease**

# Objective 11

***T. monococcum*** – Introgression of the *TmStb1* locus conferring resistance to *Septoria tritici* blotch into hexaploid wheat

# 5 years of field assessment of resistance

---

Hexaploid wheat



*Triticum monococcum*  
- diploid wheat AA



No lesions !!!





# Many thanks to



## RRes Farm staff

Richard Gutteridge

Vanessa McMillan (HGCA-BBSRC)

Kostya Kanyuka

Steve Freeman (WGIN casual)

Gail Canning (BBSRC-Pre-breeding LoLa)

## Summer students 2011

- root sampling – seed harvest

Martha Jones (Pre-breeding LoLa)

David Franklin (Pre-breeding LoLa)

Leanne Freeman (WGIN)



# Screening of WGIN EMPP (Paragon mutants)

**Richard Weightman**

**WGIN management meeting 21<sup>st</sup> Feb 2012**



# Acknowledgements

- **JIC**            **Simon Orford, Joe McNeice**  
**(scanning EMPP samples)**
  
- **Aunir**        **Chris Piotrowski**  
**(development of NIR calibrations)**
  
- **JHI**            **Stuart Swanston**  
**(provision of calibration dataset)**

# Background and Aims

- **EMS mutated Paragon population is a major resource but trait data sparse**
- **Image analyzers and Near Infra Red (NIR) spectroscopy can provide rapid and non-destructive grain quality assessments**
- **GG LINK project developed NIR calibrations for alcohol yield and gliadin content**
  - **Hypothesis that reduced Gli would be a valuable trait for reduced N storage requirements and better N use efficiency**
- **Aim: to transfer GG NIR calibrations to JIC and to scan the EMPP to identify potentially interesting lines**

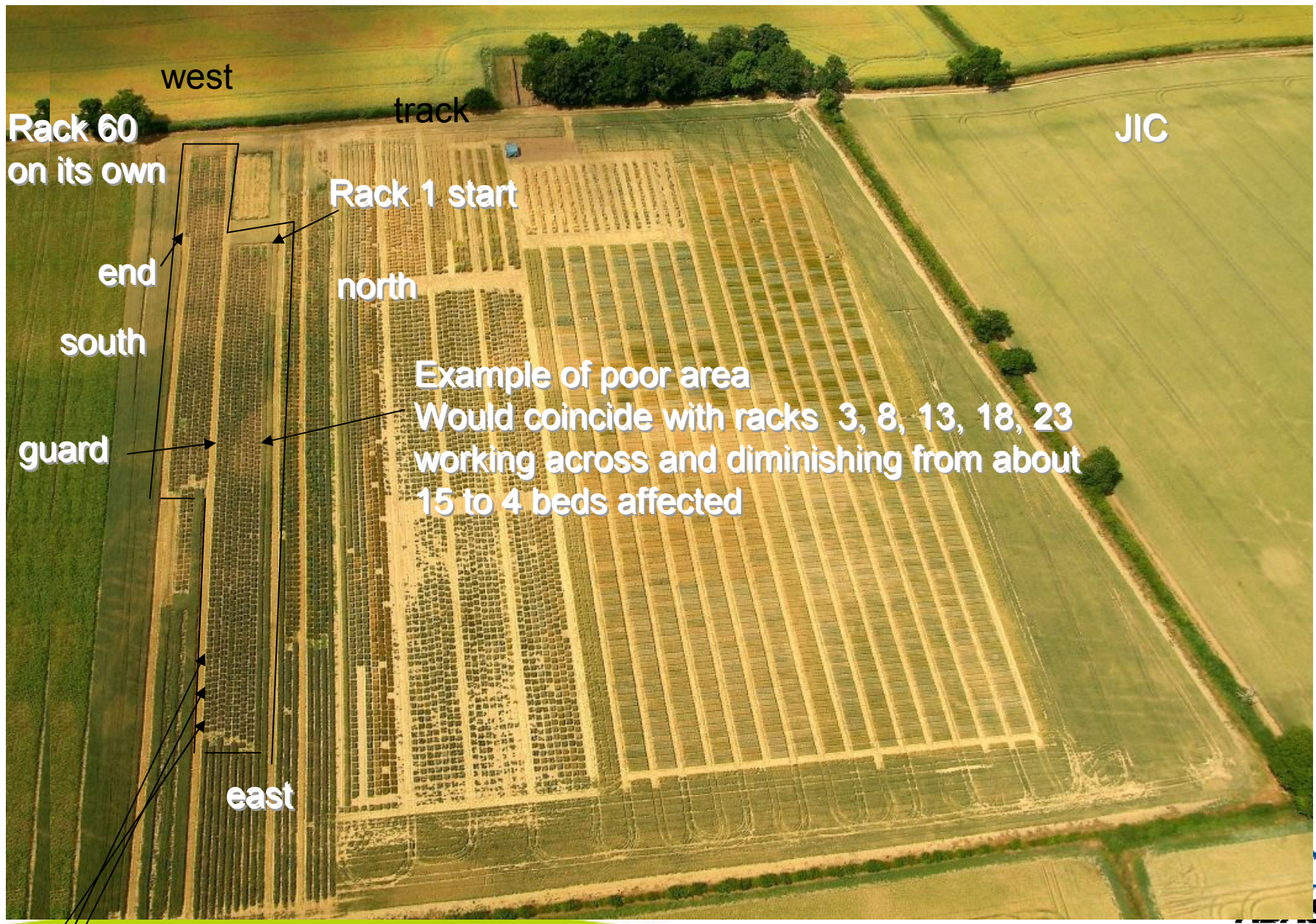


# Approach

1. Grain samples transferred from JHI and reference data from ADAS to Aunir (n=552)
2. Reference samples scanned on FOSS 6500 NIR at Aunir,
3. Calibrations developed for grain protein fractions and alcohol yield (AY)
4. NIR calibrations transferred to FOSS 6500 NIR to JIC (Norwich) and train staff to use with small sample cell
5. 4,737 samples from EMPP scanned with FOSS 6500 and Marvin analysers (inc. 14 control samples)
6. Data transferred to ADAS for analysis



Crop sown March 2006 (JIC)



Rack 60  
on its own

west

track

JIC

Rack 1 start

end

north

south

Example of poor area  
Would coincide with racks 3, 8, 13, 18, 23  
working across and diminishing from about  
15 to 4 beds affected

guard

east

Controls 141, 142, 143 would be about here

# Key traits for NIR calibration

- Alcohol yield (litres/tonne)
- Residue viscosity (mPa.s)
- Gliadin content (% DM)
- LMW Glutenin content (%DM)
- HMW Glutenin content (%DM)
- Albumin & Globulin content (%DM)
- Gliadin fraction (proportion of protein)
- LMW fraction (proportion of protein)
- HMW fraction (proportion of protein)
- Albumin & Globulin fraction (proportion of protein)

# Plus

## Standard FOSS calibrations for:

- Protein
- Moisture
- Starch

## And by Marvin image analyzer:

- Grain dimensions (length, width, L:W ratio)
- Thousand grain weight

# Database in Excel spreadsheet

Microsoft Excel - JIC NIR&Marvin data\_Final.xls

File Edit View Insert Format Tools Data Window Help

Type a question for help

B7

1 **These data were generated by ADAS, Aunir, the John Innes Centre and James Hutton Institute through a subcontract with the Wheat Genetic Improvement Network (WGIN)**

2 This database relates to samples from the WGIN EMPP (Paragon mutants) population grown in 2006,

3 and must be reviewed in conjunction with the report dated 6th October 2011

4

5 **Abbreviations**

6 GG = Green Grain (LINK) project - for further information see HGCA Project report No 468, June 2010.

7 JIC = John Innes Centre

8 AY = Alcohol Yield

9

10 **Description of traits in worksheet 'Data Paragon population'**

11 Columns with yellow blocked heading represent NIR predictions

12 Columns with green blocked heading represent physical measurements (TGW and grain size by Marvin)

13 Columns with blue blocked heading are derived from primary data


Column	Heading	Units	Description of calibration (for RPD values see 'Cal performance' worksheet)
A	Position	n	Sequential sample number as processed at JIC
B	Ref	n	Unique line ID as scanned (to be included with id)
C	id	text	a or b population from which line was taken
D	GH	n	Measure of scan quality (higher value, poorer quality; rejects already removed)
19 NIR data (with the exception of standard FOSS calibrations, must be viewed in conjunction with worksheet for cal performance)			
E	FOSS_Moisture	g/100gFW	Standard grain network cal
F	FOSS_Protein	g/100gDM	Standard grain network cal for total protein
G	FOSS_Starch	g/100gFW	Standard grain network cal; see column BD for DW basis
H	NIR_ExtractProt	Protein g/100g DM	Developed from ADAS HPLC method - protein solubilised and quantified compared to a gliadin standard
I	NIR_GLDDMHPLC	g/100gDM	Developed from ADAS HPLC method - Proportion of fraction x total protein
J	NIR_GLDDMProt	g/100gDM	Developed from ADAS HPLC method - Proportion of fraction x extracted protein
K	NIR_LMWDMProt	g/100gDM	ibid
L	NIR_ResViscosy	mPa.s	Measurement at SWRI - wheat cook method
M	NIR_AlcYielddryt	Litres/tonne DM	Measurement at SWRI - wheat cook method

Macro1 \ Notes \ Data Paragon population \ Cal performance \ Parental ref \ Field trial layout \ Plot posn for spatial

Draw AutoShapes

Ready NUM

start Microsoft ... The James H... Nitrate in Roc... WGIN EMPP s... Microsoft Exc... EN 11:47



Microsoft Excel - JIC NIR&Marvin data\_Final.xls

File Edit View Insert Format Tools Data Window Help

Type a question for help

10 B I U

Reply with Changes... End Review...

A4450 1665

	A	B	C	D	E	F	G	H	I	J	K
1	Position	Ref	id	GH	NIR_Moisture	NIR_Protein	NIR_Starch	NIR_ExtractProt	NIR_GLDDMHPLC	NIR_GLDDMProt	NIR_LMWDMPro
2	Position	Ref	id	GH	FOSS_Moisture	FOSS_Protein	FOSS_Starch	NIR_ExtractProt	NIR_GLDDMHPLC	NIR_GLDDMProt	NIR_LMWDMPro
4450	1665	1702	b	3.317757	10.75	15.4	63.4	16.92	7.75	5.99	3.2
4451	1666	1703	b	1.841727	10.29	13.4	60.7	16.02	7.07	5.34	2.8
4452	1667	1704	b	1.364391	10.18	13.6	60.0	16.97	7.60	5.66	2.9
4453	1668	1705	b	3.02377	10.90	15.3	61.3	16.84	7.77	5.91	2.9
4454	1669	1706	b	1.750733	10.26	13.6	60.7	16.27	7.19	5.40	2.8
4455	1670	1707	b	2.552548	10.34	15.3	61.5	17.61	8.03	6.13	3.3
4456	1671	1708	b	1.291938	10.32	14.2	59.7	17.63	7.97	6.18	3.0
4457	1672	1710	b	1.795759	10.17	12.8	59.3	16.21	7.04	5.24	2.7
4458	1673	1711	b	1.751353	10.52	14.5	58.4	17.48	7.83	6.22	3.2
4459	1674	1712	b	3.72037	10.80	17.3	63.6	18.29	8.81	7.08	3.4
4460	1675	1715	b	1.465668	10.22	14.2	58.4	17.71	7.87	6.12	3.1
4461	1676	1716	b	1.840056	9.98	14.3	62.8	16.87	7.68	5.85	3.0
4462	1677	1717	b	2.582787	9.90	16.3	66.8	17.20	8.15	6.30	3.3
4463	1678	1718	b	2.575664	10.33	14.5	61.7	16.90	7.63	5.76	2.9
4464	1679	1719	b	1.759618	10.49	13.8	58.5	17.09	7.64	5.86	2.9
4465	1680	1720	b	1.518924	10.16	13.9	61.2	16.41	7.41	5.71	2.9
4466	1681	1721	b	2.68768	10.55	16.0	63.3	17.62	8.35	6.50	3.1
4467	1682	1722	b	2.078019	10.63	15.2	59.7	17.90	8.09	6.31	3.3
4468	1683	1724	b	3.292013	11.01	15.6	63.9	16.50	7.88	6.23	3.0
4469	1684	1725	b	3.011532	10.27	17.6	63.6	19.28	9.15	7.17	3.7
4470	1685	1726	b	1.904771	10.13	13.5	62.3	15.42	6.96	5.46	2.6
4471	1686	1727	b	1.791851	10.00	15.3	61.2	17.79	8.24	6.23	3.1
4472	1687	1728	b	2.183258	10.66	14.9	61.9	16.51	7.65	6.16	3.1
4473	1688	1729	b	2.147755	9.76	13.7	63.6	15.87	7.24	5.39	2.6
4474	1689	1730	b	2.302089	10.27	17.5	61.4	19.39	9.21	7.40	3.6
4475	2292	18	bM5	2.036504	9.82	15.0	62.9	16.08	7.73	5.96	2.5
4476	1353	1562	bM5	1.025257	9.99	13.8	61.0	16.32	7.27	5.58	2.6
4477	1588	1608	bM5	2.464269	10.90	16.0	60.4	17.72	8.17	6.42	3.3
4478											

Macro1 / Notes / Data Paragon population / Cal performance / Parental ref / Field trial layout / Plot posn for spatial

Draw AutoShapes

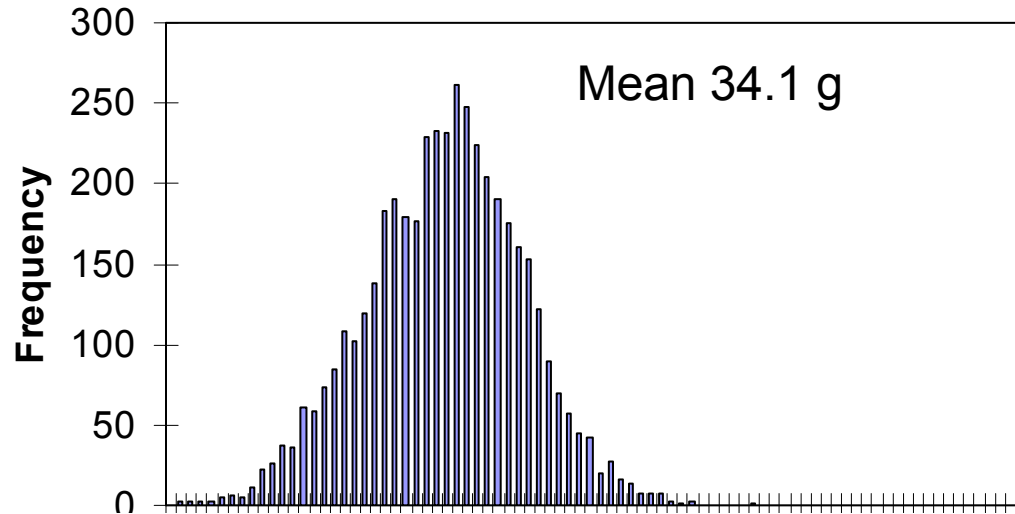
Ready NUM



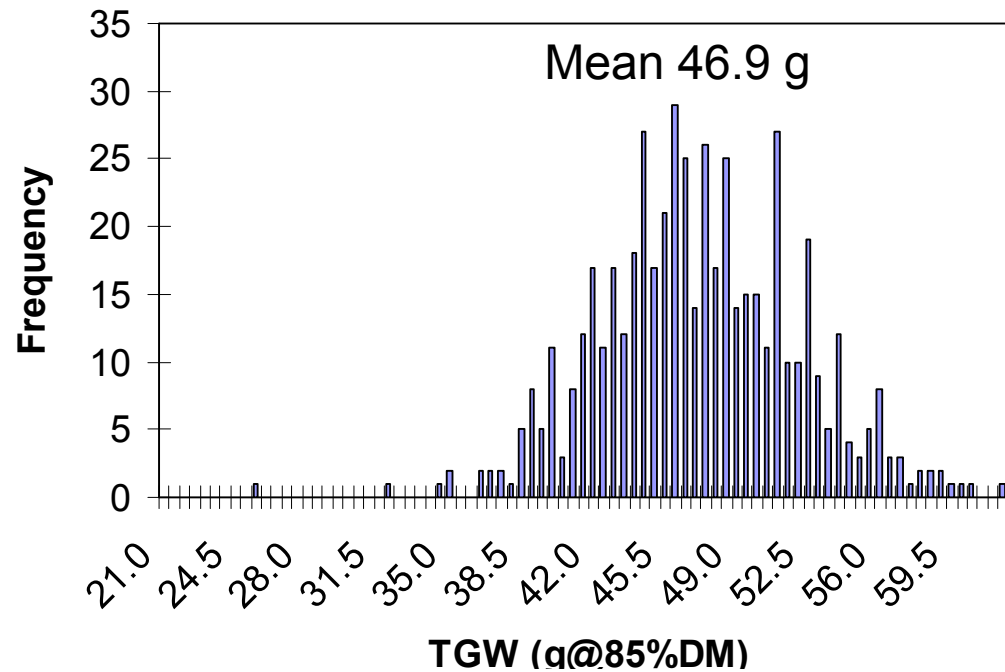
# Results

Thousand grain weight

### Histogram - TGW (EMPP)



### Histogram - TGW (GREEN Grain)

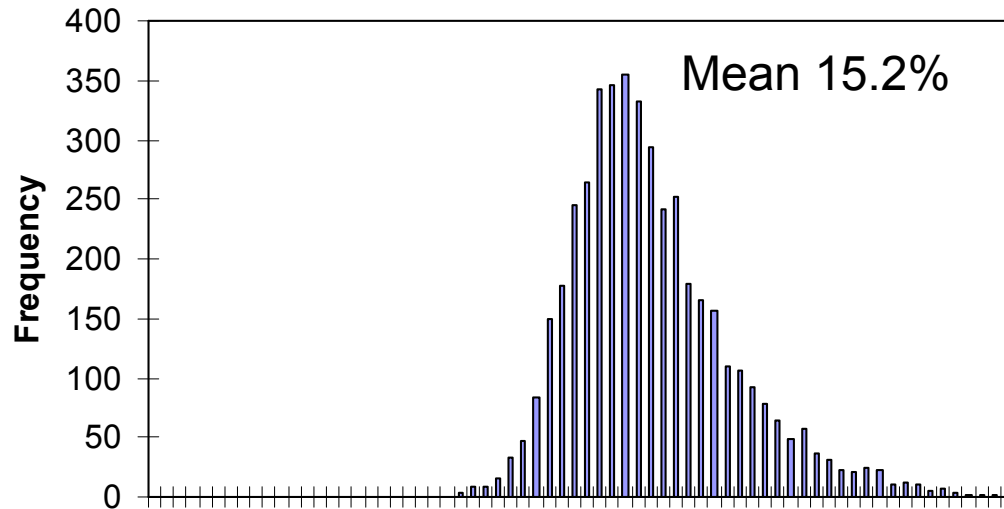




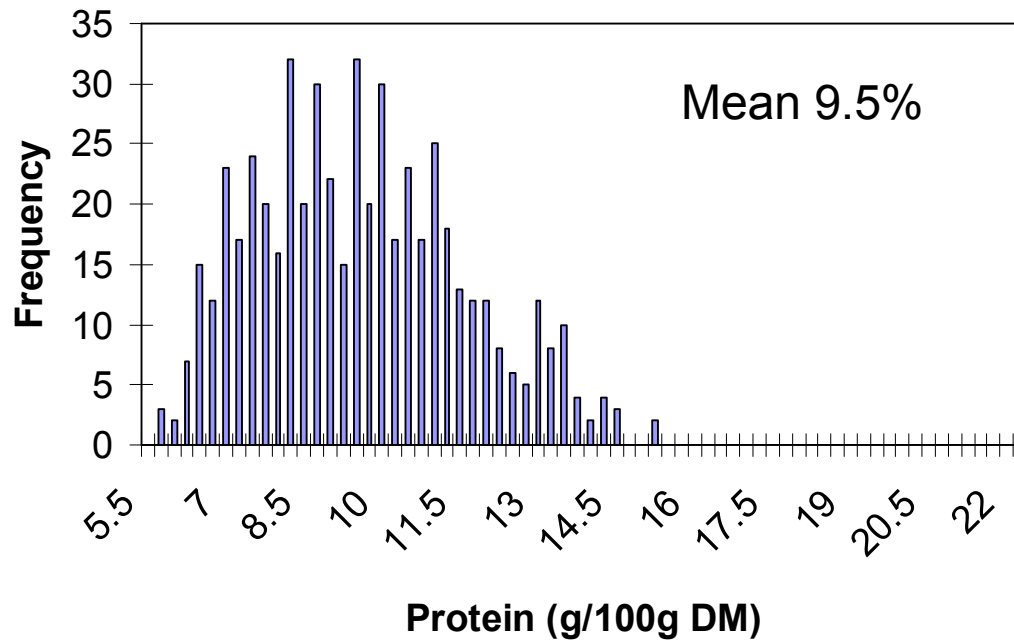
# Results

Grain protein

### Histogram - protein EMPP



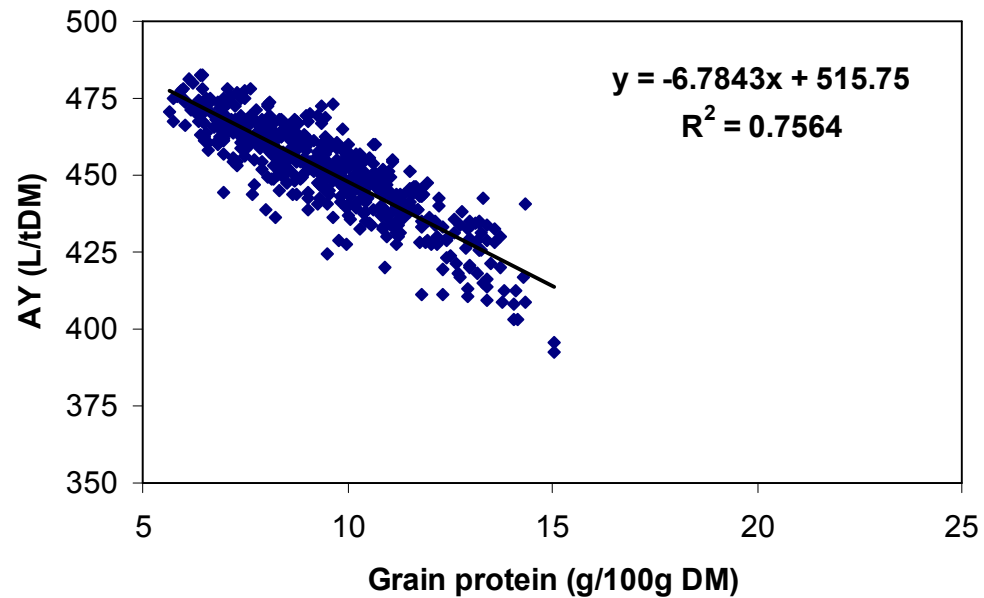
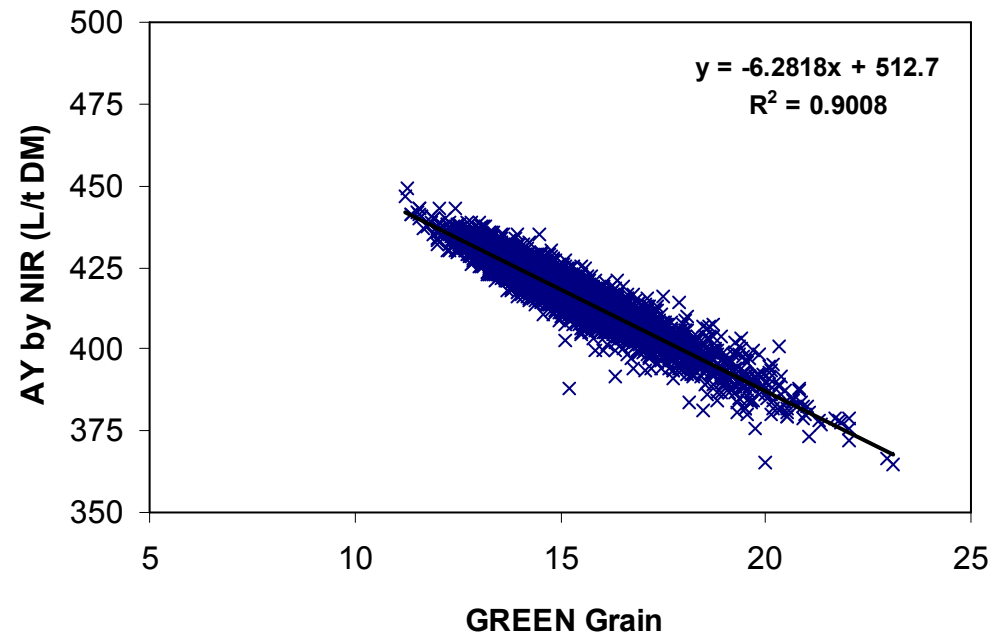
### Histogram - Protein (GREEN grain)



# Results

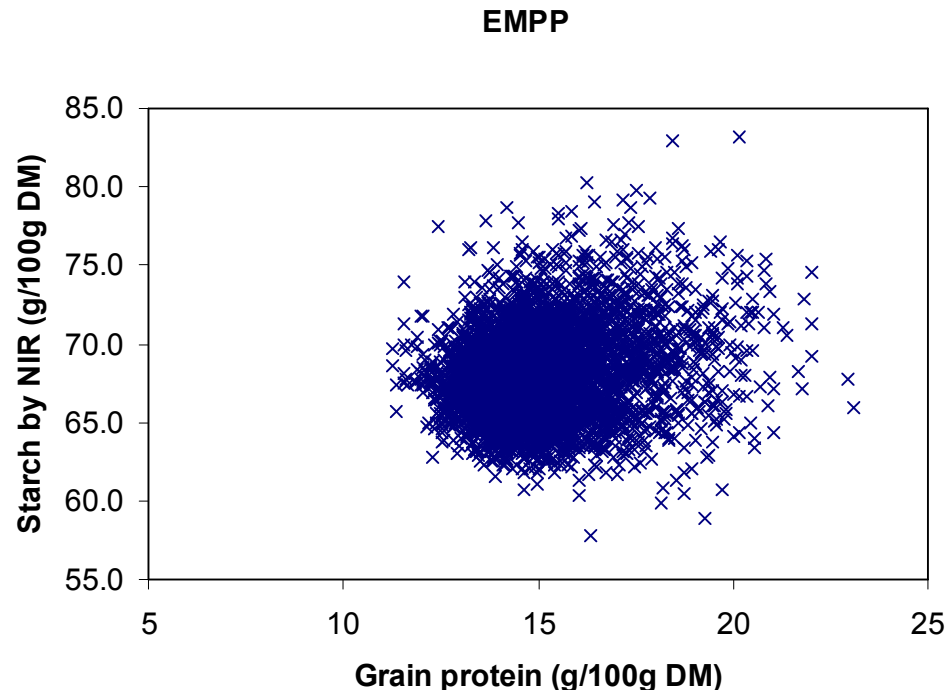
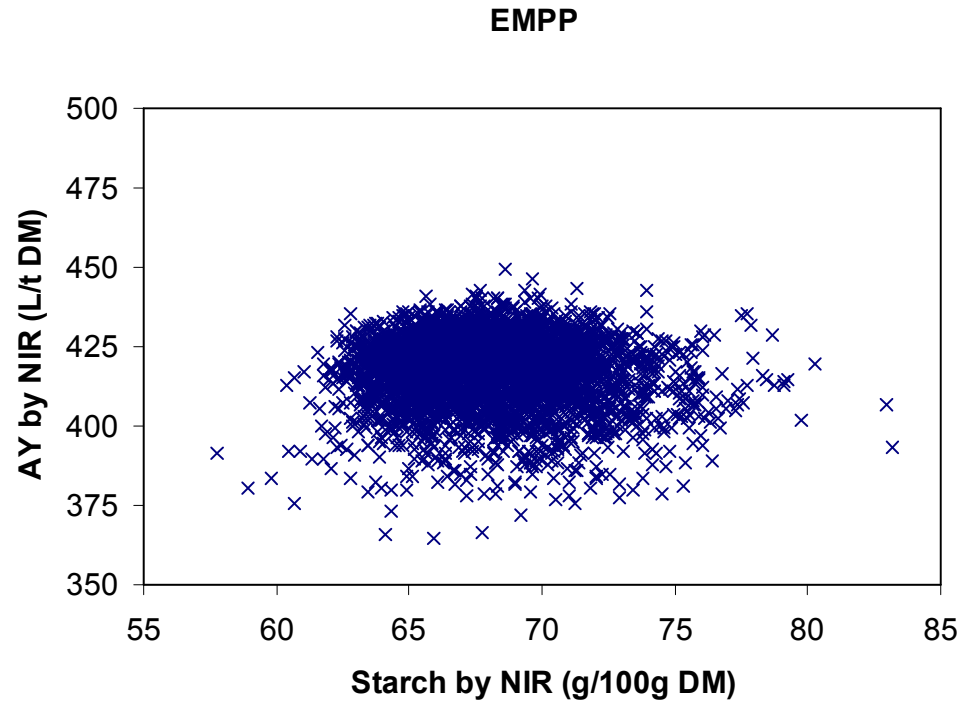
Relationship between AY and protein

EMPP



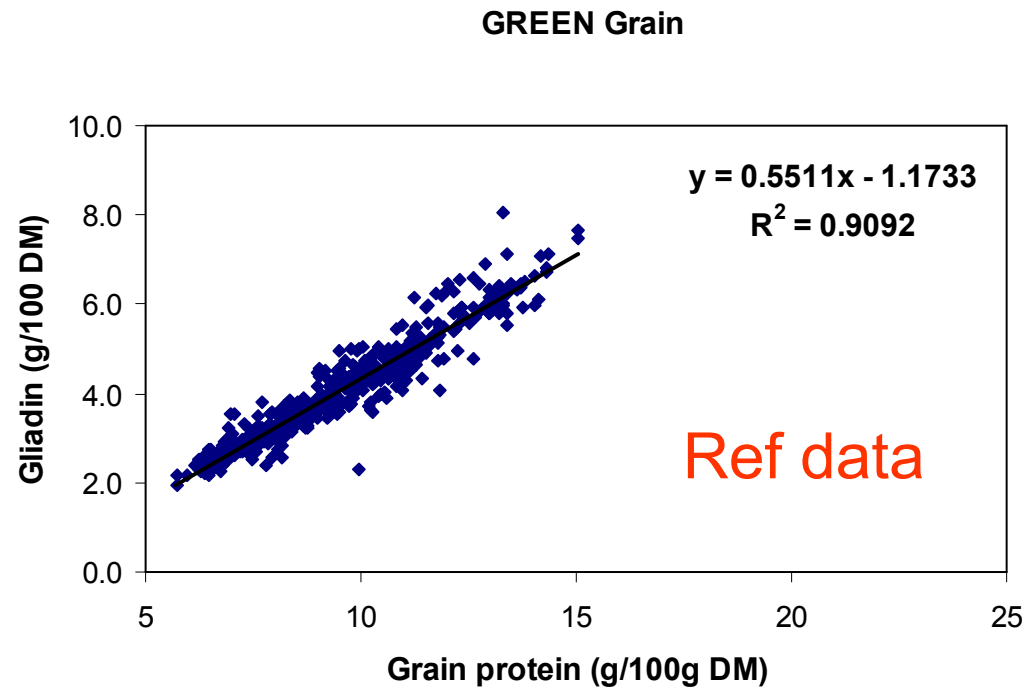
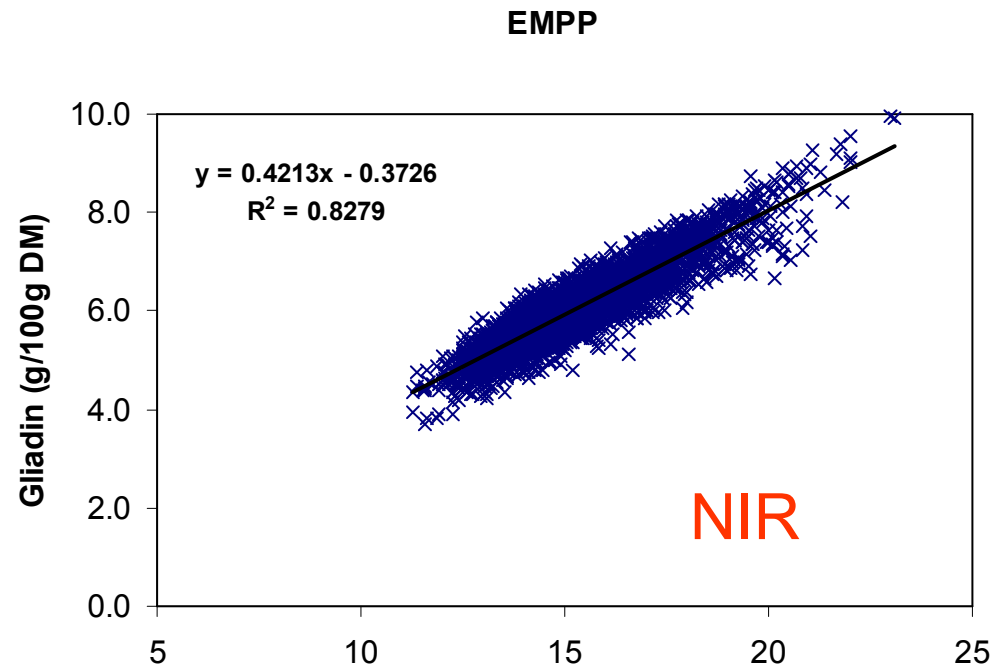
# Results

FOSS starch calibration of dubious value ?

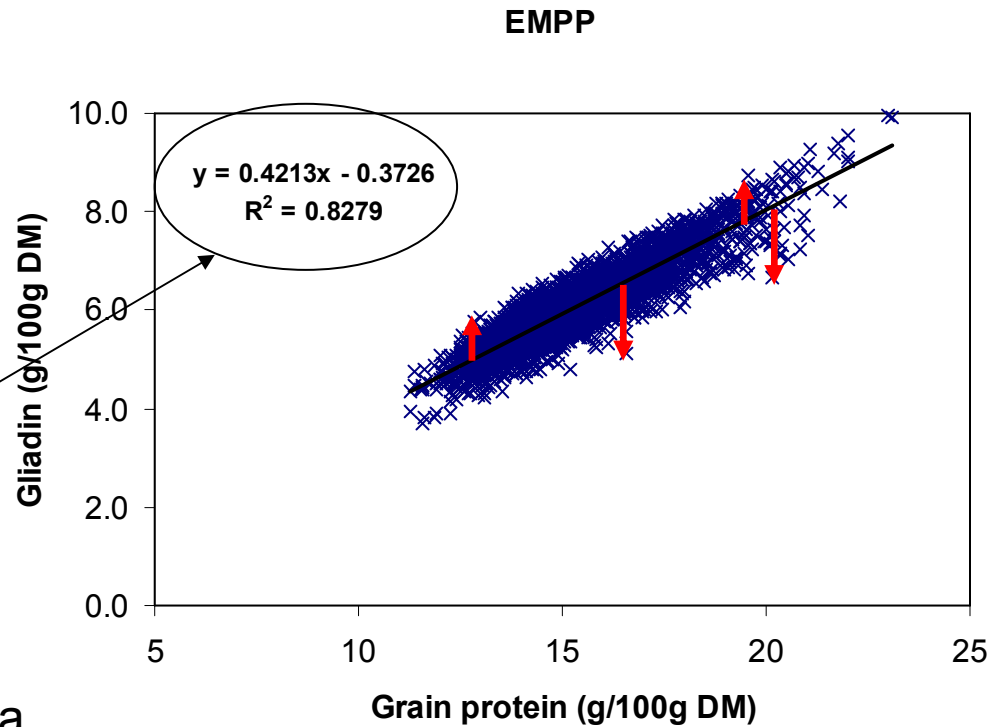


# Results

Smaller gliadin response to increasing GP in EMPP ?



# Identification of high/low gliadin lines



Use slope equation to look for deviations from the expected gliadin at a given GP

# Identification of high and low gliadin lines

## Low gliadin % deviation

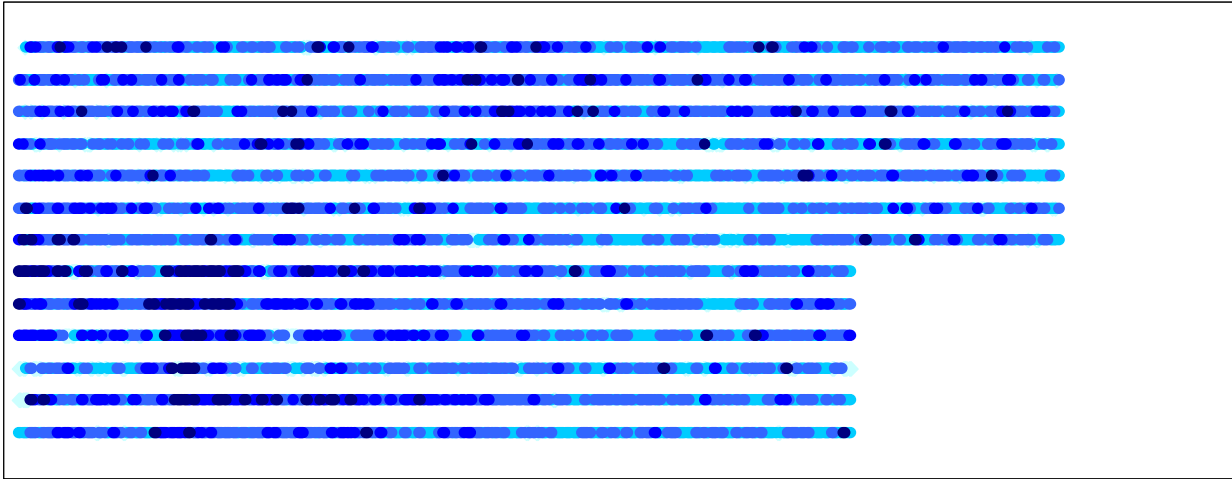
Ref	id	Grain protein (g/100g DM)	Gliadin (g/100gDM)	Gli % meas	Gli % pred	Gli % dev (obs-pred)
1077	a	16.6	5.13	31.0	40.2	-9.2
1094	a	15.2	4.78	31.4	40.0	-8.5
1159	a	12.3	3.90	31.8	39.5	-7.7
1990	a	20.1	6.65	33.0	40.5	-7.5
1865	a	13.5	4.36	32.3	39.7	-7.4

# Identification of low and high gliadin lines

## High gliadin %

Ref	id	GP (g/100g DM)	Gliadin (g/100gDM)	Gli % meas	Gli % pred	Gli % dev (obs-pred)
1513	b	14.3	6.43	45.0	39.8	+5.1
1239	b	12.9	5.77	44.9	39.6	+5.3
1275	b	14.5	6.55	45.2	39.9	+5.4
1493	b	13.0	5.85	45.0	39.6	+5.4

# Spatial variation in grain protein



- **Spatial variation assessed by ANOVA**
  - appears to be a relatively small part of the total variance,
  - but care should be taken to check the exact position of lines before selection for further analysis



# Summary

- **The EMPP & Paragon parent appear to be relatively low gliadin for a given protein content, compared to the elite lines seen previously**
- **GG ref data set were:**
  - **winter types**
  - **grains of soft endosperm texture**
  - **low grain protein samples (<15%) due to N deficiency, and**
  - **predominantly varieties other than Paragon**

# Questions

- **How relevant is Paragon as a hard/spring wheat**
- **Is the apparent, low gliadin content significant (requires wet chem to confirm)**
- **Given that the trial was droughted (low TGW/high protein) how useful are these samples**
- **Identification strategy**

# Further work

- **Multiplication of EMPP followed by reference chemistry on grain**
- **Repeat screening using material from less droughted/lower protein experiment(s)**
  - **Paragon deletion mutant set ?**
- **Develop other populations**
  - **Winter types ?**

# Thank you

**Richard Weightman**

**ADAS Boxworth**

**[richard.weightman@adas.co.uk](mailto:richard.weightman@adas.co.uk)**



# **BBSRC funded BBR project - Phytopath (Jan 2011 - Dec 2013)**

## **2 partners**

**European Bioinformatics Institute (EBI)**

**– Genome Campus, Hinxton, Cambs**

**PIs : Paul Kersey + Ewan Berney**

**Rothamsted Research**

**PIs : Kim Hammond-Kosack + Mansoor Saqi**

**Topic : Full sequenced plant pathogen genomes  
(35 - 50 Mb in size)**

**Linking individual pathogen genes / gene variants  
to disease phenotypes**

**Focus : Fungi and protists which cause disease on key  
Agricultural Crops**

## Welcome to PhytoPath

PhytoPath is a new bioinformatics resource that integrates genome-scale data from important plant pathogen species with literature-curated information about the phenotypes of host infection. Using the [Ensembl Genomes](#) browser, it provides access to complete genome assembly and gene models of priority crop and model-fungal and oomycete phytopathogens. PhytoPath also links genes to disease progression using data from the curated [PHI-base](#) resource.

## Last added genomes

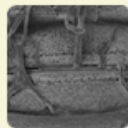
### *Phaeosphaeria nodorum*



*Phaeosphaeria nodorum* is a major fungal pathogen of wheat (*Triticum aestivum*) and a member of the Dothideomycetes, a large fungal taxon that includes many important plant pathogens affecting all major crop plant families. The Dothideomycetes is a newly recognised major class of filamentous fungi that replaces the long-recognized loculoascomycetes. It includes the causal organisms of many economically important plant diseases. *Phaeosphaeria nodorum* is an experimentally tractable organism, which is easily handled in defined media. It was one of the first fungal pathogens to be genetically manipulated. *Phaeosphaeria nodorum* has been a model for fungicide development and emerged as a model for dothideomycete pathology.

Browse this genome at the [Ensembl Genomes website](#).

### *Mycosphaerella graminicola*



*Mycosphaerella graminicola* causes septoria tritici blotch in wheat, the second most important disease in the United States after rust. This disease is heavily targeted by the fungicide companies worldwide being nevertheless very difficult to control. The latest version of the *Mycosphaerella graminicola* genome assembly was released in September 2008. The genome comprises 21 chromosomes with a total size of 39.7 Mb and encodes about 10952 protein-coding genes.

Browse this genome at the [Ensembl Genomes website](#).

## This release

The current release of PhytoPath is built from the twelfth release (Dec 2011) of Ensembl Genomes and version 3.3 of PHI-base and was released on January 2012. For all species, available data includes genome sequence and gene models, functional annotation, and protein-based comparative analysis with other fungal/oomycete species (provided by Ensembl Genomes), together with expertly curated molecular and biological information on genes proven to affect the outcome of pathogen-host interactions (provided by PHI-base). In addition, the following specific data sets are also available:

- RNA-Seq alignments for *Puccinia Triticina*
- Variation data for *Gibberella zeae*
- EST alignments for all the species except *Puccinia graminis f. sp. tritici* and *Phytophthora ramorum*
- DNA alignments in the *Phytophthora* group and for the *Hypocreales* order.

## Featured Gene

### Tomatinase

Saponin detoxification enzymes from pathogenic fungi are involved in the infection process of their host plants. *Fusarium oxysporum f. sp. lycopersici*, a tomato pathogen, produces the tomatinase enzyme **Tom1**, which degrades alpha-tomatine to less toxic derivatives. A curated interaction for **Tom1** in *Septoria lycopersici* is described on PHI-base.

# A portal to the genomes of phytopathogenic fungi and protists which infect key crop species



## Fungi



*Fusarium oxysporum f. sp. lycopersi*



*Gibberella moniliformis*



*Gibberella zeae* ★



*Mycosphaerella graminicola* ★



*Nectria haematococca*



*Phaeosphaeria nodorum* ★



*Puccinia graminis f. sp. tritici* ★



*Puccinia triticina* ★



*Ustilago maydis*

## Protists



*Phytophthora infestans*



*Phytophthora ramorum*



*Phytophthora sojae*



*Pythium ultimum*

★ = wheat infecting

The PhytoPath project is funded by the BBSRC and includes data from:



# @ RRes is linked to EBI PhytopathDB

- **PHI-base**: is a database of interactions between pathogens and their hosts
- The current version contains **1335 interactions** associated with 1065 genes from 102 pathogenic species
- PHI-base contains **highly curated phenotypic data** linked to gene sequences, checked by the international community

## Key point

**Gene sequence within the genome browser in Phytopath are directly linked to the phenotype and the peer reviewed literature in PHI-base**

**25% cereal infecting fungi (15% wheat)**

**[www.PHI-base.org.uk](http://www.PHI-base.org.uk)**



# Geospatial Resource for Agricultural Species and Pests (GRASP Wheat) Workshop

The University of Nottingham  
Dr Rumiana Ray

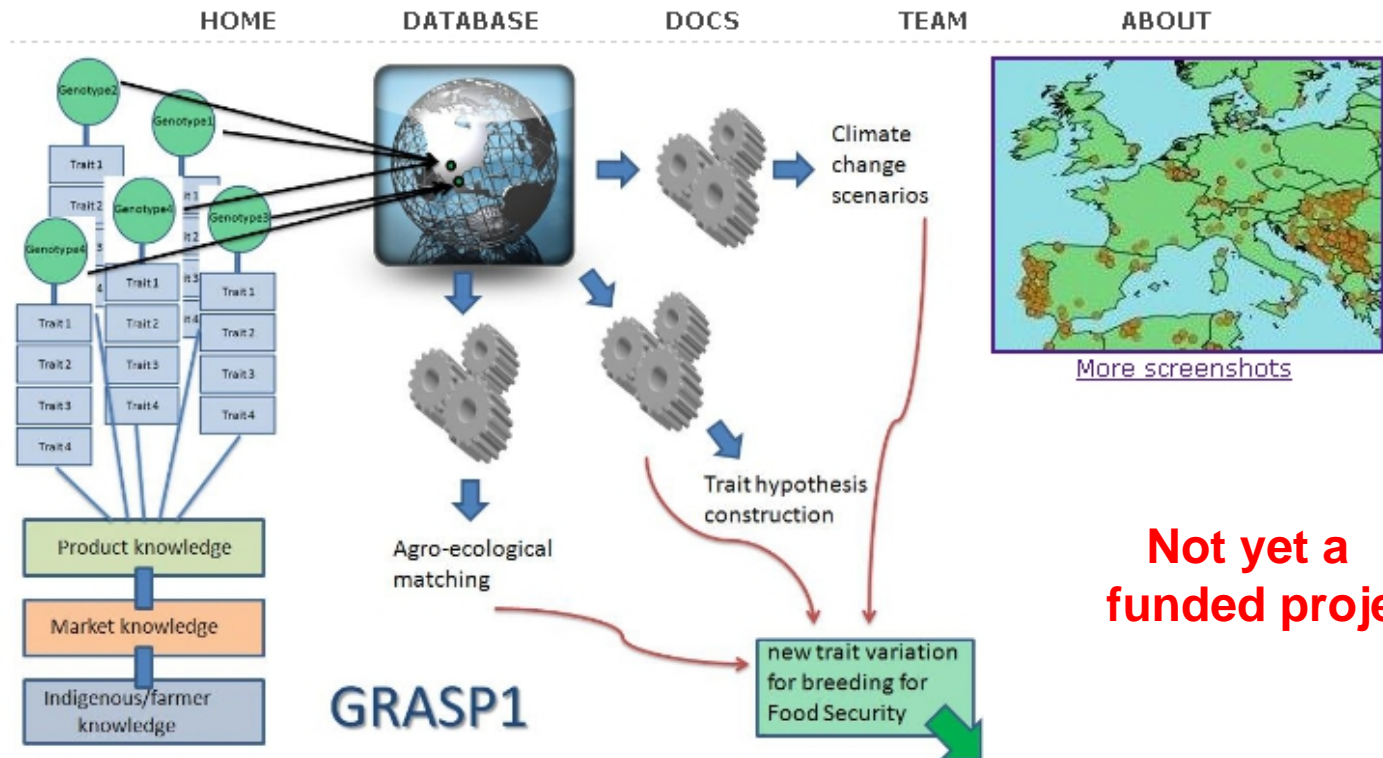
Rothamsted Research  
Dr Toby Bruce

The University of Reading  
Dr Alastair Culham

Background -GRASP Prototype at The University of Nottingham

## GRASP

Geospatial Resource for Agricultural Species Pests



GRASP1

GRASP Overview (CPIB, 2011).

**Not yet a  
funded project**

**Aim:** to develop a source/standards-based framework for geospatial data capture to exploit genetic trait diversity in crop breeding integrating environment, pest and pathogen data

**How: One day International workshop**

**Objectives:**

- Use cross-institutional and multi-disciplinary expertise to generate common framework and standards – ontologies and phenotyping
- Integrate existing database resources – reference database from Breeders, PHI-base, Plantwise, RRES insect survey, Crop Monitor, i4life into a geospatial prototype for **Wheat UK as proof of concept**
- Develop consortium for future collaborative proposal on GRASP Wheat EU/Global

**Who:** Pathologists, Physiologists, Entomologists, Breeders, Geospatial scientists, Integrative Biologists, Bioinformaticians, Geneticists working on **Wheat traits for Food Security**

**When: Provisionally Autumn 2012**

**Where:** TBC

**Contact : [Rumiana.Ray@nottingham.ac.uk](mailto:Rumiana.Ray@nottingham.ac.uk)**