



WAGENINGEN UNIVERSITY
PLANT SCIENCES



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A Map-Based Cloning Approach to Unravel Genes for Basal Resistance to Biotrophic Fungi in Barley

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Basal Resistance to Biotrophic Fungi

“What makes the plant less than extremely susceptible, non-hypersensitive resistance”

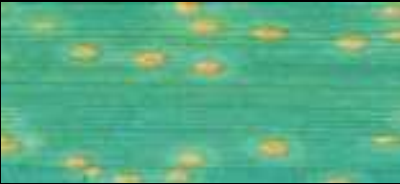



Working hypotheses

- Basal resistance determines the host status of barley to rusts and powdery mildews



Basal Resistance to Biotrophic Fungi

Host status - "Barley is a host to *Puccinia hordei*, but is a non-host to leaf rusts of rye, wheat and grasses..."

	<i>P. hordei</i>	<i>P. tritricina</i>
Barley		
Wheat		

Basal Resistance to Biotrophic Fungi

“What makes the plant less than extremely susceptible, non-hypersensitive resistance”

Quantitative → host partial resistance

- latency period
- infection frequency
- severity scale
- number of pustules
- lesion length

QTL mapping



Basal Resistance to Biotrophic Fungi

“What makes the plant less than extremely susceptible, non-hypersensitive resistance”

Quantitative → host partial resistance

Qualitative → non-host resistance



Basal resistance

Development of hyper-susceptible experimental lines...

P. triticina

most barley accessions,
like Vada



some barley accessions,
like L94



SusPtrit (barley)



Basal resistance

Development of hyper-susceptible experimental lines...

<i>P. triticina</i>	<i>P. recondita</i>
<i>P. hordei-bulbosi</i>	<i>P. holci</i>
<i>P. hordei-murini</i>	<i>P. bromi</i>
<i>P. gram. lolii</i>	<i>... etc</i>
<i>P. persistens</i>	

Vada		immune	immune
SusPtrit		susceptible	immune
Cebada Capa		immune	immune

Basal resistance

Development of hyper-susceptible experimental lines...

B. graminis f.sp. *tritici*

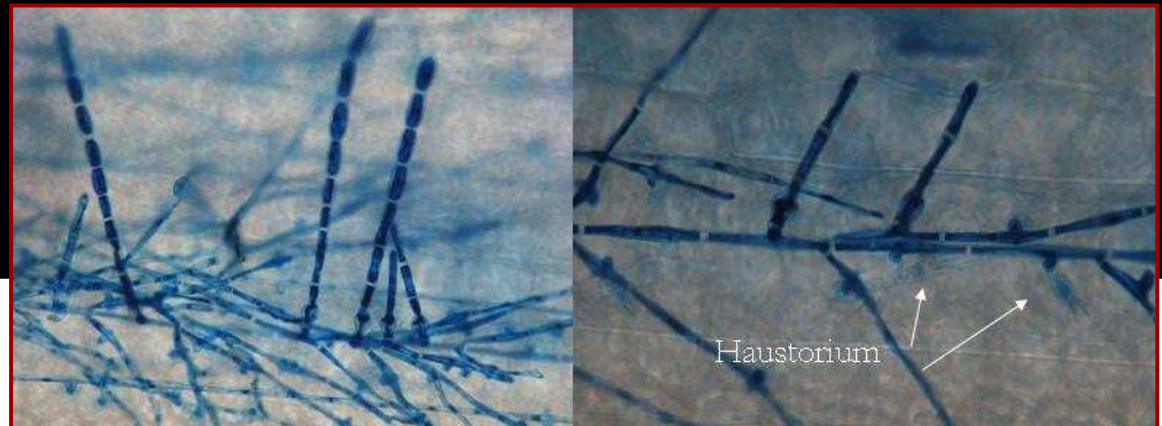
most barley accessions,
like Vada



susceptible wheat



SusBgt (barley)



Basal Resistance to Biotrophic Fungi

“What makes the plant less than extremely susceptible, non-hypersensitive resistance”

Quantitative → host partial resistance

Qualitative → non-host resistance

Working hypotheses

- Basal resistance determines the host status of barley to rusts and powdery mildews
- Partial and non-host resistances belong to the same system



"Barley, Integrated, Marcel 2009" (GG2)

L94/Vada

Vada/SusPtrit

C. Capa/SusPtrit

Dom/Rec (OWBs)

Steptoe/Morex

Nure/Tremois

Igri/Franka

Integration of the available markers from 7 mapping populations using JoinMap 4



"Barley, Integrated, Marcel 2009" (GG2)

L94/Vada

Vada/SusPtrit

C. Capa/SusPtrit

Dom/Rec (OWBs)

Steptoe/Morex

Nure/Tremois

Igri/Franka

6990 molecular markers

- RFLP, AFLP, SSR, DArT, TDM...

3561 from Steptoe/Morex

- Wenzl *et al.* 2006 (DArT)

- Stein *et al.* 2007 (EST)

- Potokina *et al.* 2008 (TDM)

2846 EST-based markers

- functional markers

- candidate genes

- synteny among cereals

217 BINs of about 5cM

- Kleinhofs *et al.* 2001



Distribution of QTLs for basal resistance across the barley genome



BIN markers

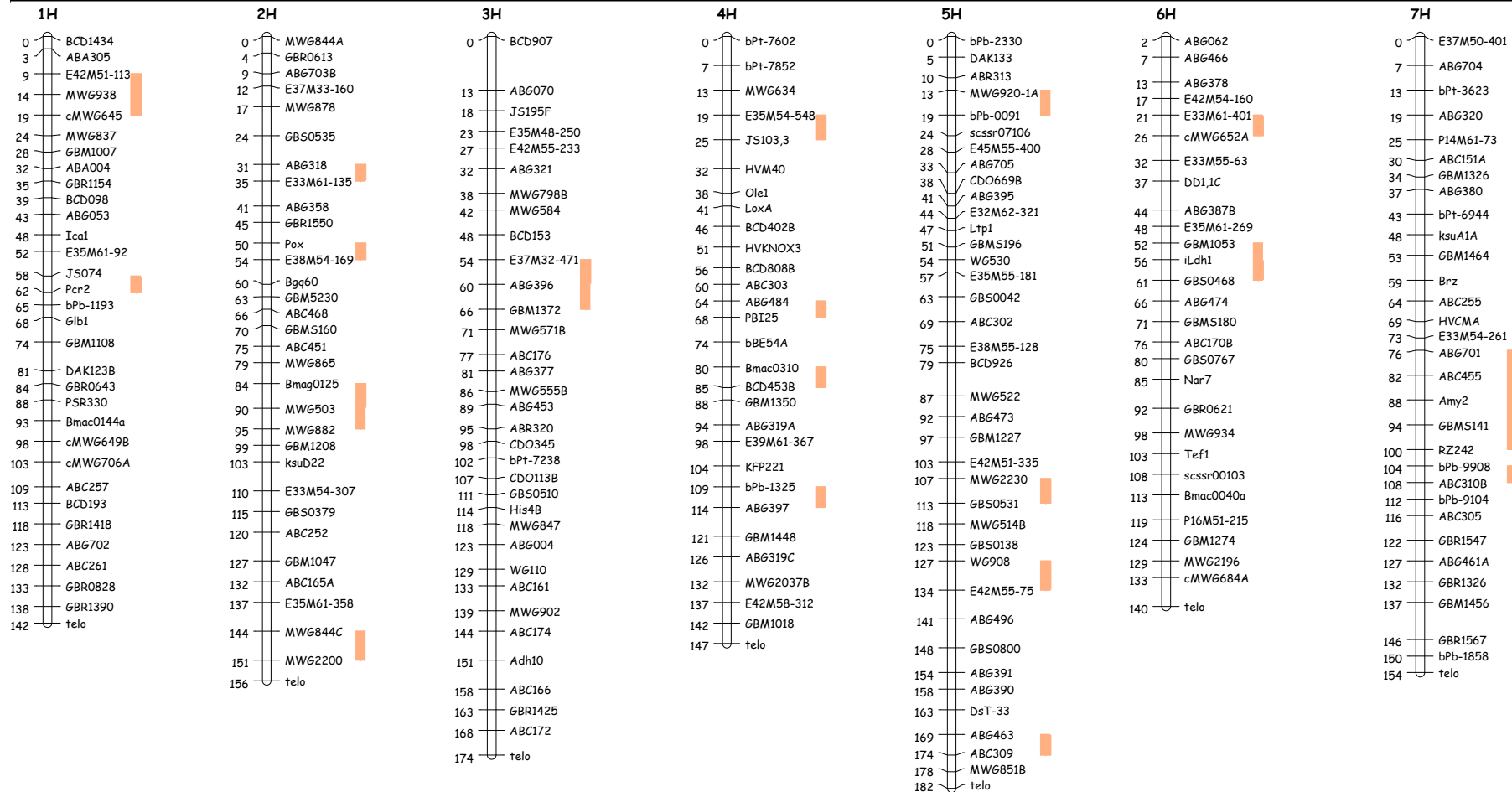


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Marcel *et al.* (2007), TAG 114

Aghnoum *et al.*, submitted

Distribution of QTLs for basal resistance across the barley genome



Ph

BIN markers

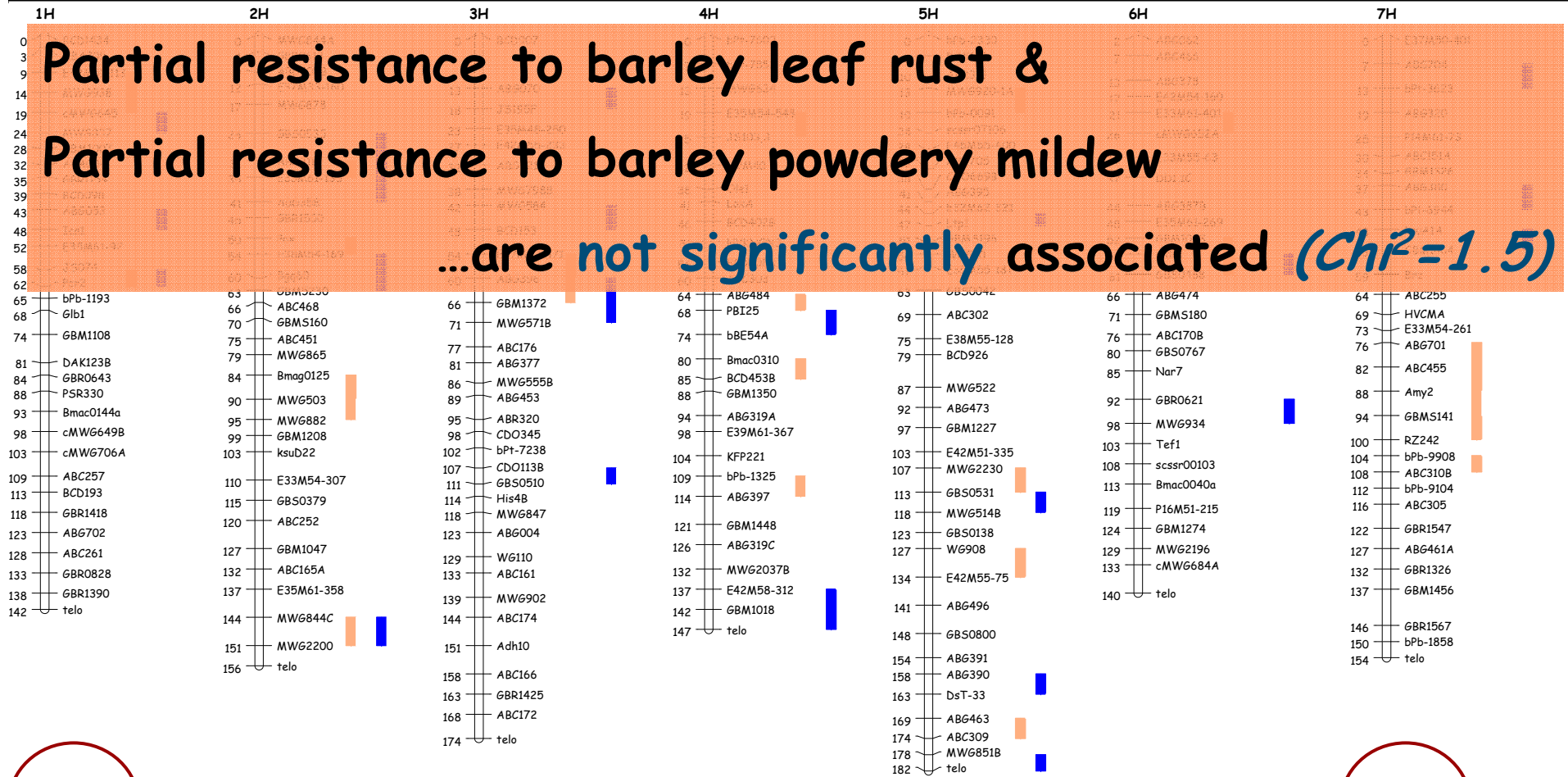


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Distribution of QTLs for basal resistance across the barley genome

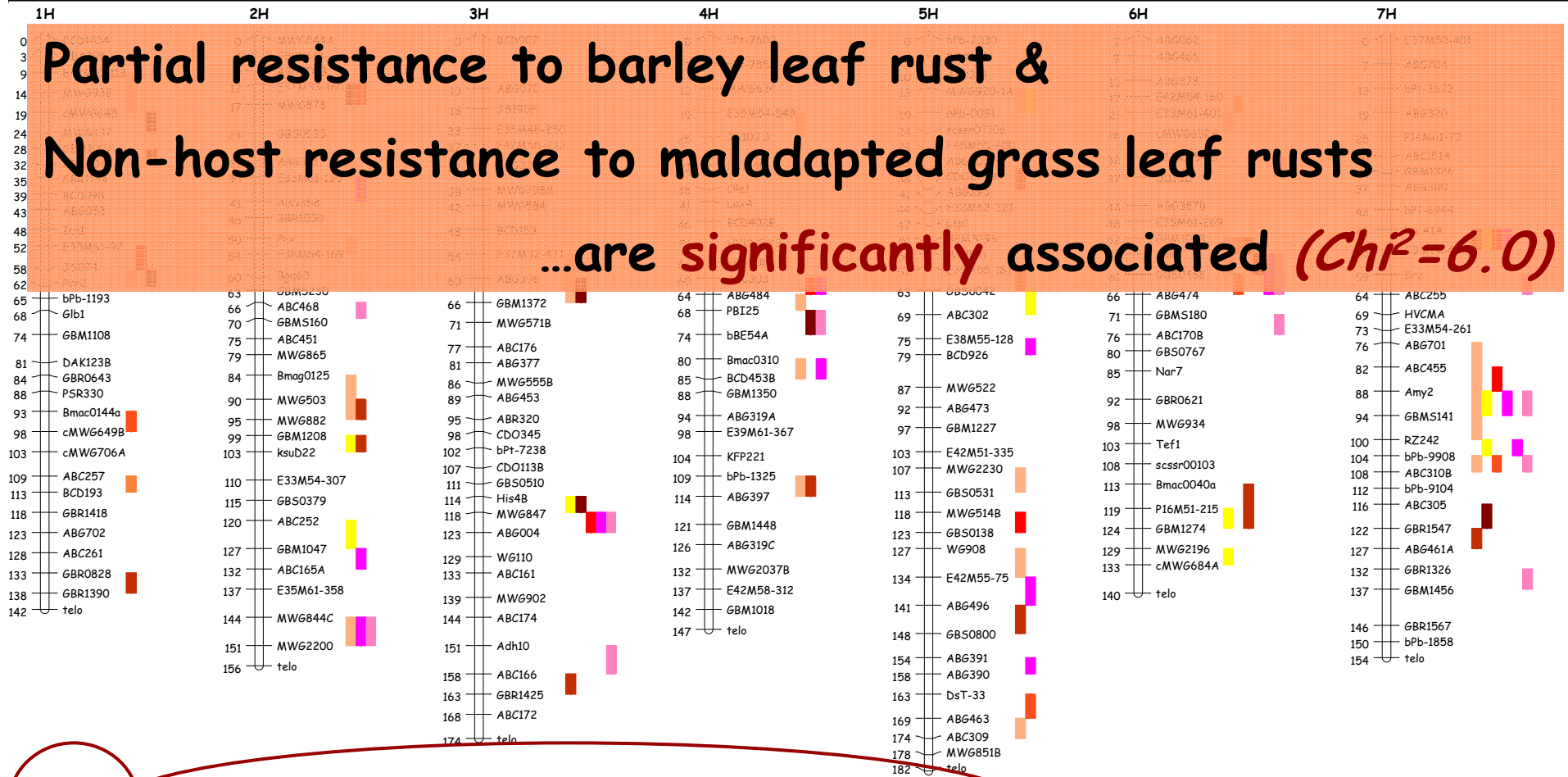


Ph

Bgh

BIN markers

Distribution of QTLs for basal resistance across the barley genome



Ph
Pca
Pgl
Pgt
Phb
Phm
Phs
Pp
Pt

BIN markers

Peroxidase Profiling in barley

1H

2H

3H

4H

5H

6H

7H

Peroxidase Profiling in barley

- *PRX* genes have conserved motives that can be used for mapping
- About 15% polymorphism in the two barley populations tested
- 168 PERO markers predicted to represent about half of the 124 known *PRX* genes in barley (PeroxiBase, February 2009)
- *PRX* genes are organized in clusters in barley
- About 40 *PRX* clusters predicted over the barley genome
- *PRX*-based markers are **significantly** associated with:
 - partial resistance to barley leaf rust ($Chi^2=6.3$)
 - partial resistance to barley powdery mildew ($Chi^2=6.3$)
 - non-host resistance to other grass leaf rusts ($Chi^2=5.5$)

BIN markers

EST-based peroxidase markers

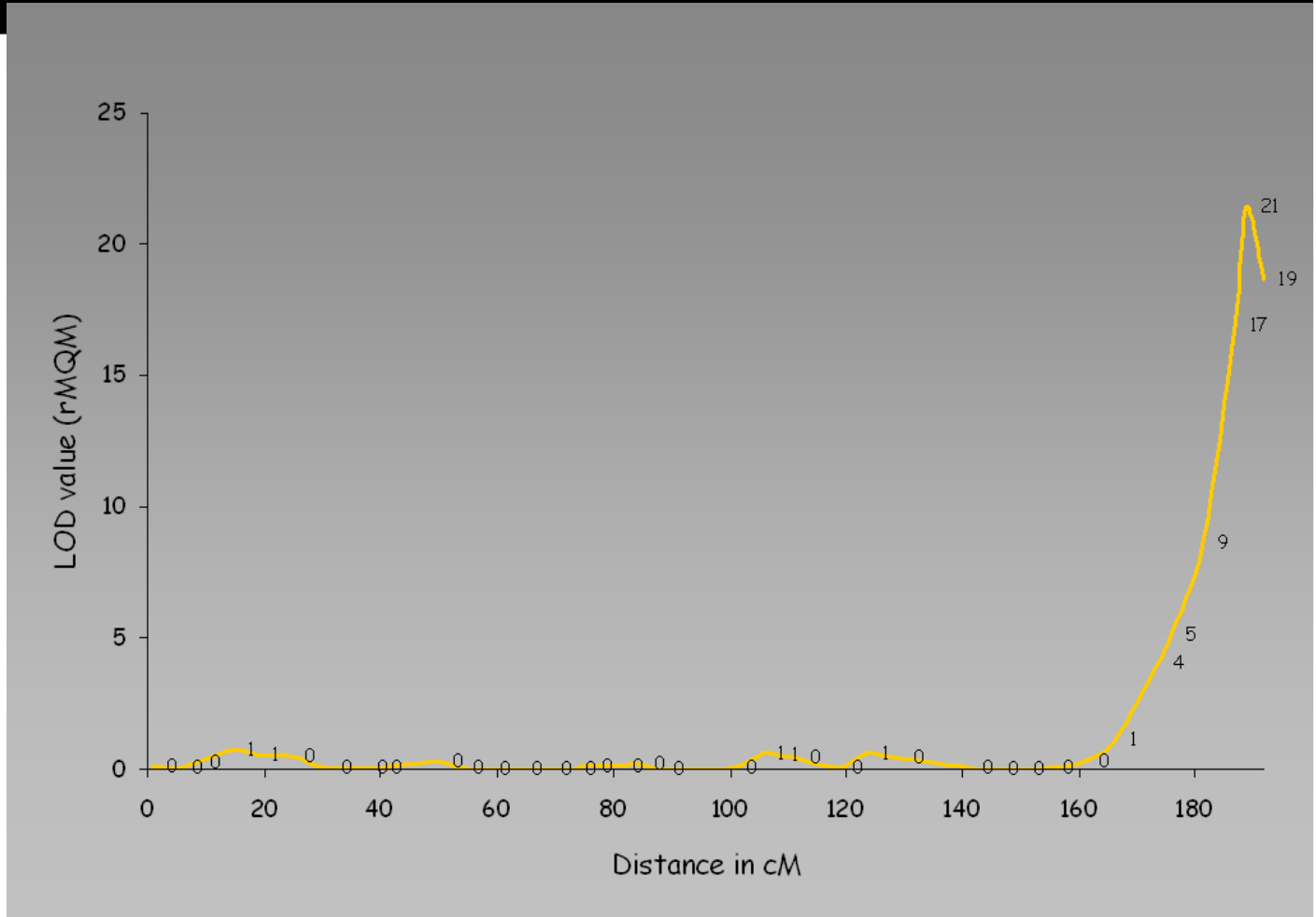
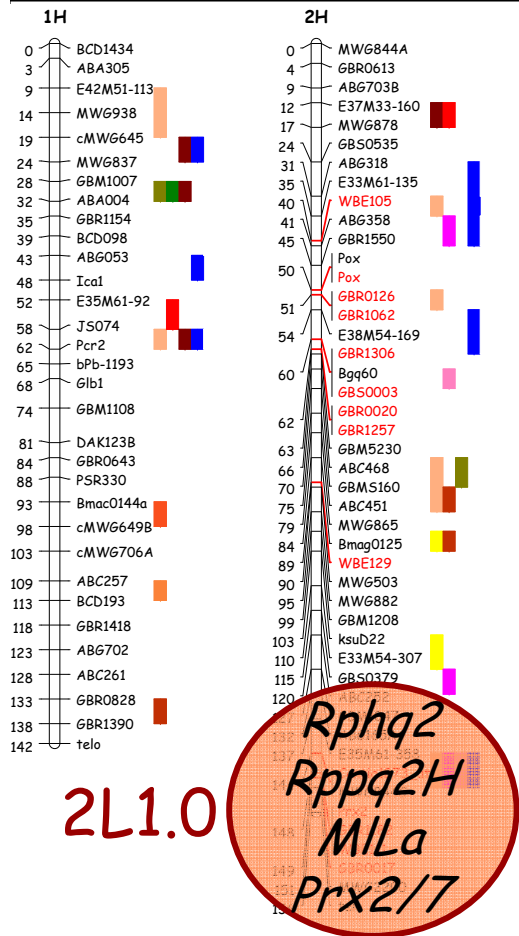
Peroxidase profiling markers



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Poster 047 (session 3), González *et al.*

Distribution of QTLs for basal resistance across the barley genome



BIN markers

EST-based peroxidase markers

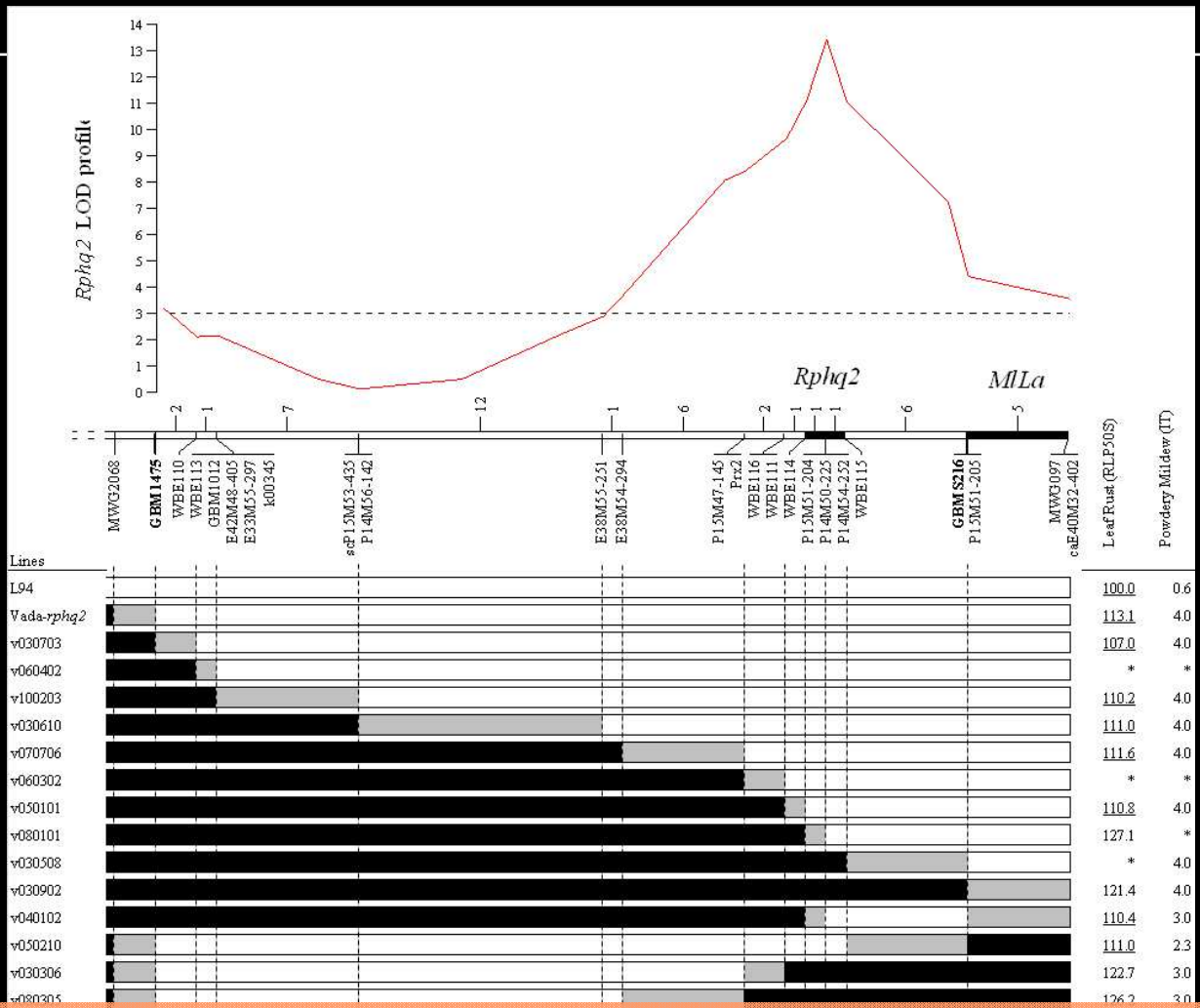


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Jafary *et al.* (2008), *Genetics* 178

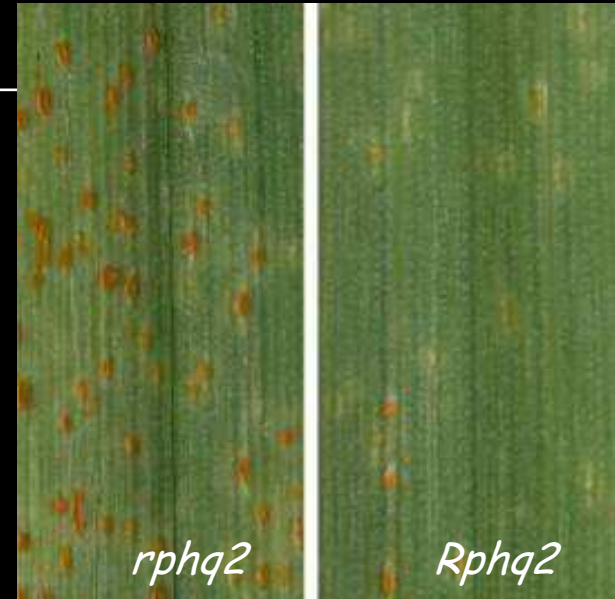
Aghnoum *et al.*, submitted

Substitution mapping of Rphq2



- 38 sub-NILs identified within 1,846 gametes
- Rphq2 fine-mapped at 0.11 cM
- M/La is about 2 cM distal from Rphq2

PR - Puccinia hordei



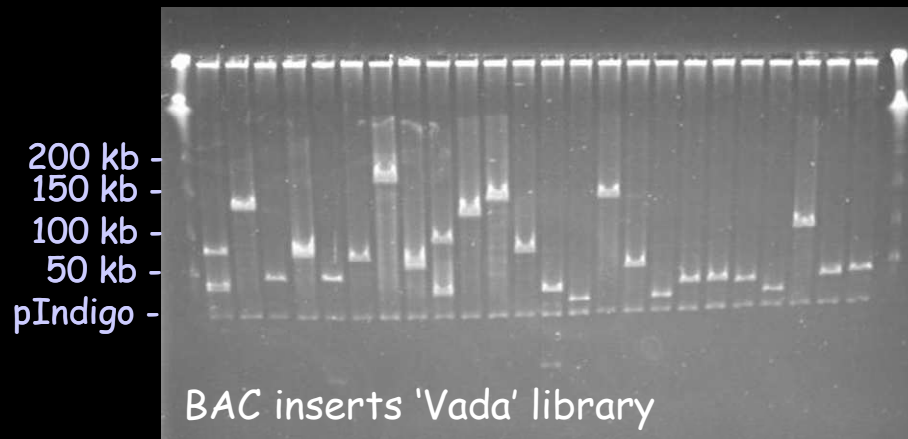
HR - Erysiphe graminis



Construction of two barley BAC libraries

Vada pooled library

- 161,000 clones
- 81 Kbp per clone
- 2.9x genome coverage

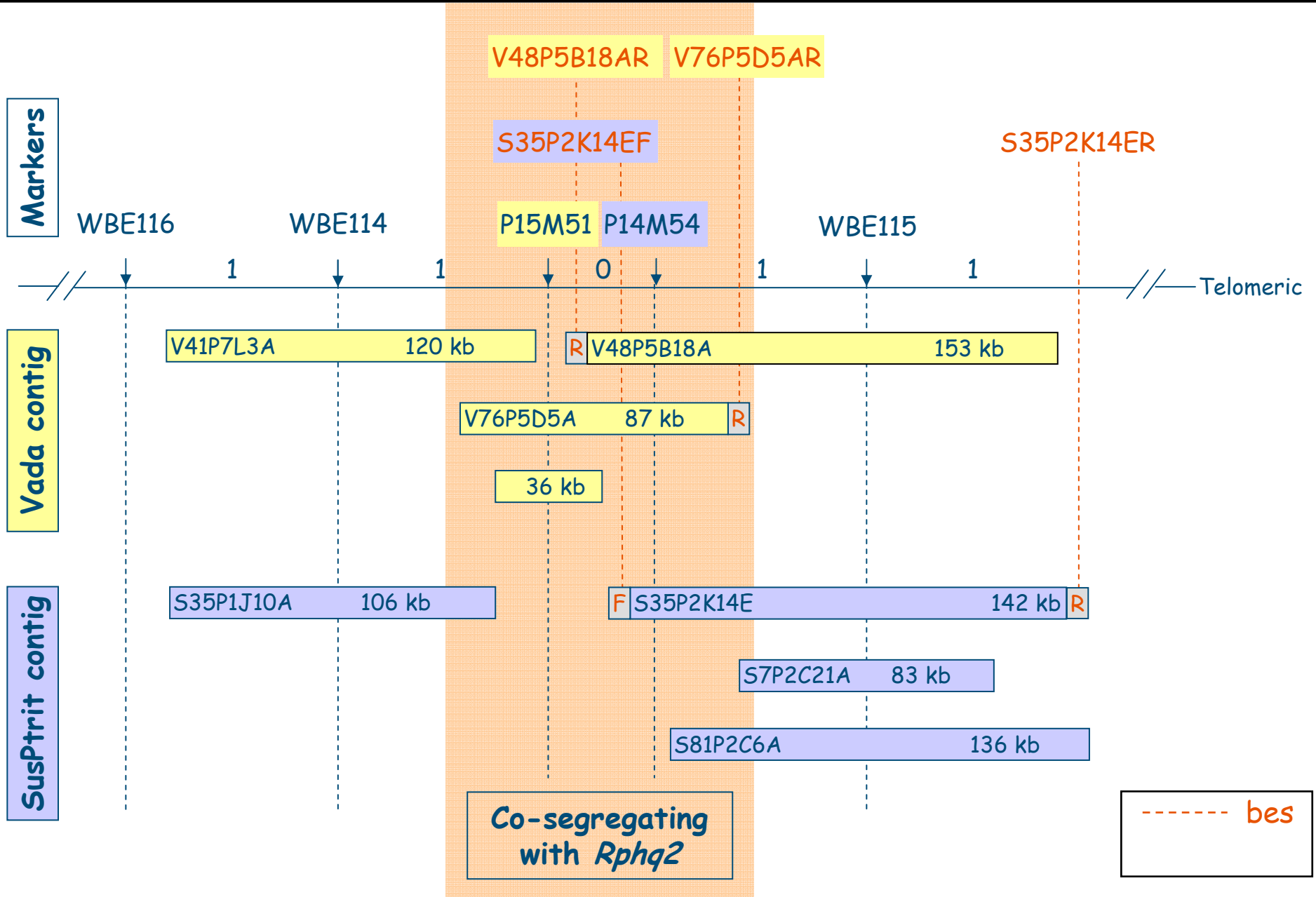


SusPtrit pooled library

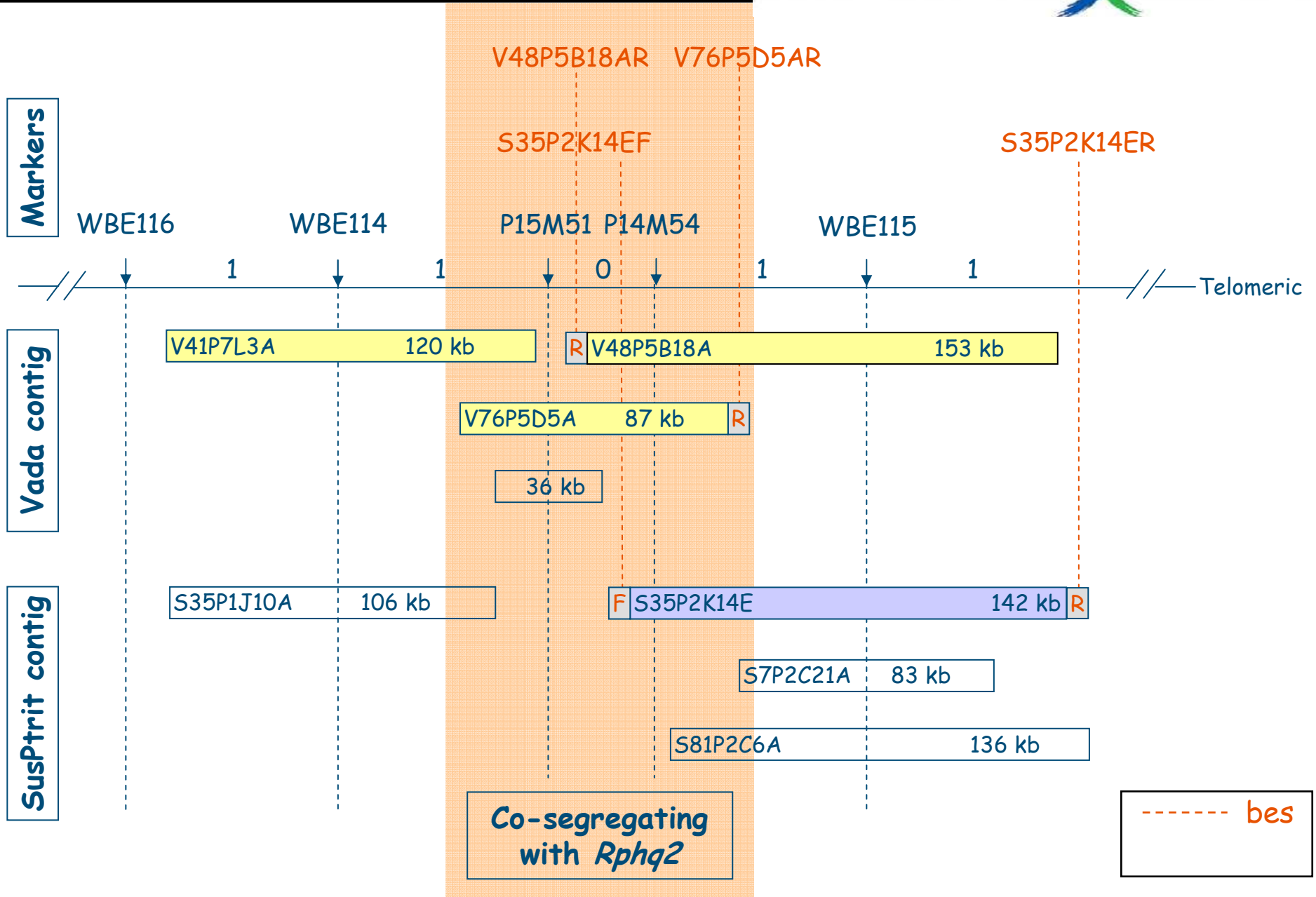
- 173,000 clones
- 108 Kbp per clone
- 4.1x genome coverage



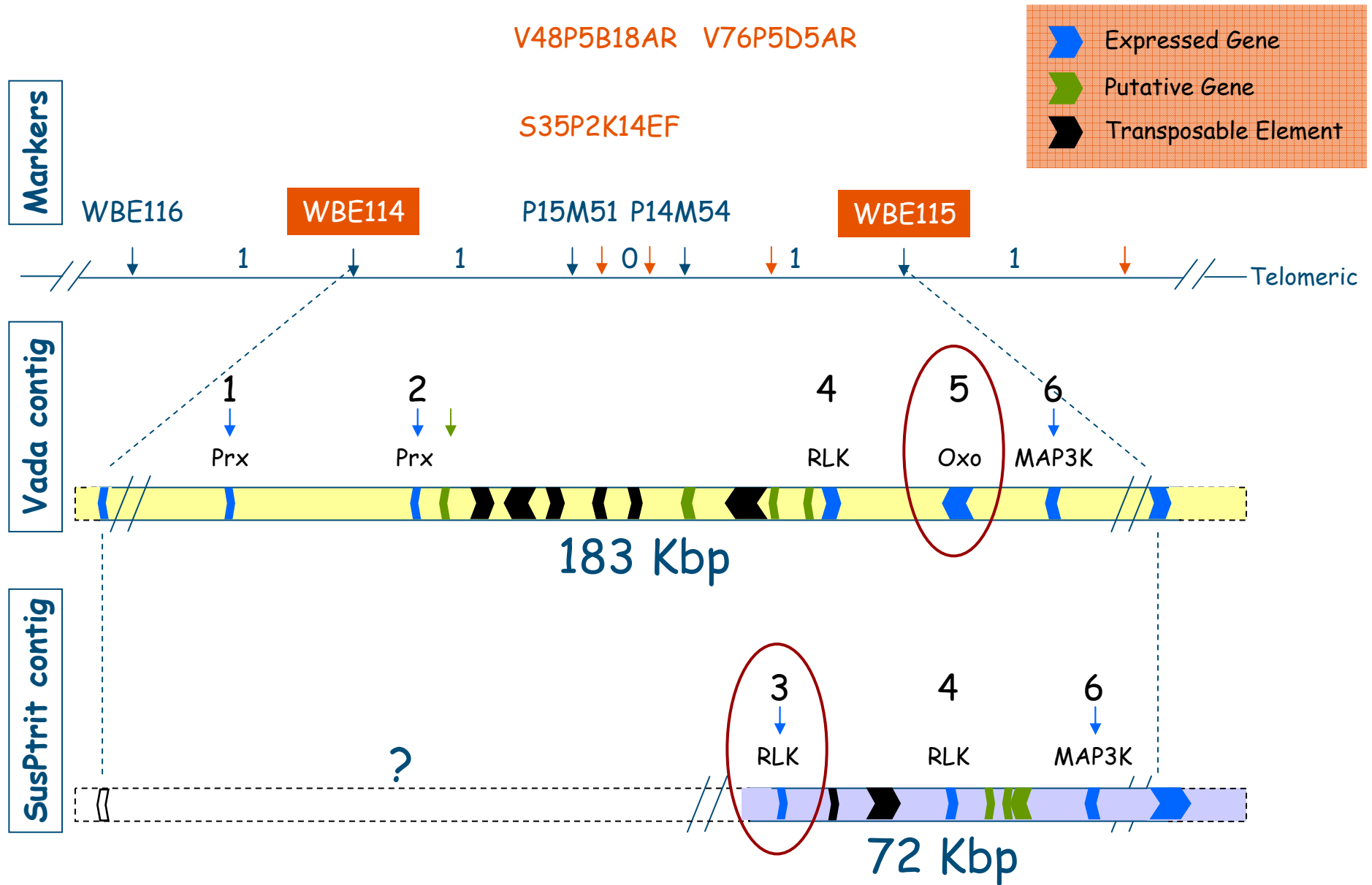
Physical map construction



Physical map construction



Physical mapping at Rphq2



Conclusions

Tools to unravel the genes for basal resistance in barley

- Hyper-susceptible barley experimental lines to maladapted forms of leaf rusts and powdery mildews, SusPtrit & SusBgt
- Several immortal barley mapping populations segregating for basal resistance to several rusts and powdery mildews
- Integrated linkage map of barley with 6,990 molecular markers
- Motif Directed Profiling of peroxidase genes
- Two barley BAC libraries from genotypes Vada & SusPtrit

Take-home messages

- QTLs for host partial & non-host resistances significantly overlap
- Different PRX genes may explain a substantial part of the QTLs for basal resistance to leaf rusts and powdery mildew
- PRXs, RLKs and MAP3K are prime candidates to explain *Rphq2*



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Thank you for your attention...



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