


**Lettuce Crop Improvement
Research
at Warwick HRI**

Dr. Paul Hand

THE UNIVERSITY OF
WARWICK


General Strategy

- Identify source(s) of variation in the trait
- Develop mapping populations & genetic maps
- Obtain **robust** phenotypic data
- Identify QTL & linked markers
- Determine marker assisted selection strategies
- Fine mapping & identification of genes

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Genetic Resources

- Lettuce diversity set
- Mapping populations
- Genetic maps
- TILLING population

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'Warwick HRI' lettuce diversity set

Identifying trait variation



24 accessions including mapping parents

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WHRI lettuce mapping populations



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WHRI lettuce mapping populations

- **Saladin x Iceberg recombinant inbred population**
 - >300 F₆ recombinant inbred lines (RIL).
 - Map construction & phenotypic analysis for QTL discovery
 - ~120 most informative lines (MIL) selected for F₇ & F₈
 - F₇ map under construction
- **Iceberg x *L. serriola* interspecific F₂ population**
 - Parents from WHRI & U.C. Davis (Michelmore) maps
 - Map anchorage: access to EST & SNP markers
 - Access wider trait variation

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Trait examples

Peach-potato aphid

Downy mildew

Discolouration

Bolting

Nitrate content

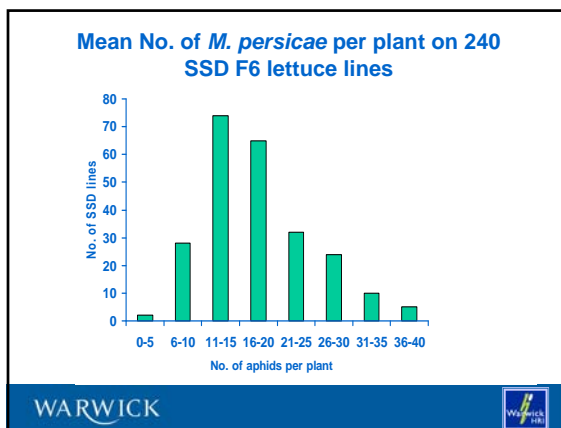
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Peach-Potato Aphid

Myzus persicae

- Wide host range
- Virus vector
- No resistant lettuce varieties
- Some partial resistance

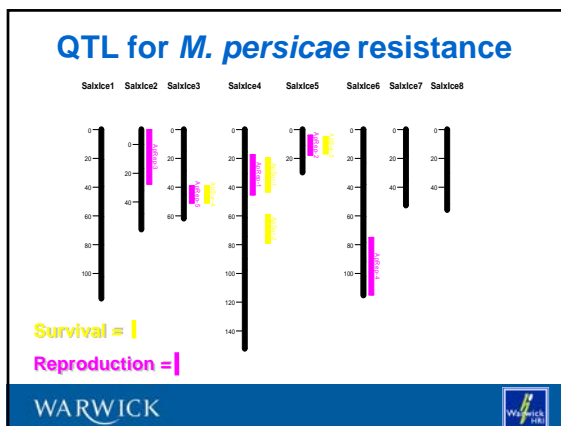
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Mean no. of aphids per plant on the most resistant & most susceptible lettuce F₆ RILs

Most resistant	± s.e.	Most susceptible	± s.e.
1.75	0.92	33.5	6.59
3.5	1.76	31.5	6.46
5.25	1.44	24.25	3.42
5.25	1.89	22.5	5.62
7.0 (Iceberg)	3.19	20.25 (Saladin)	4.96

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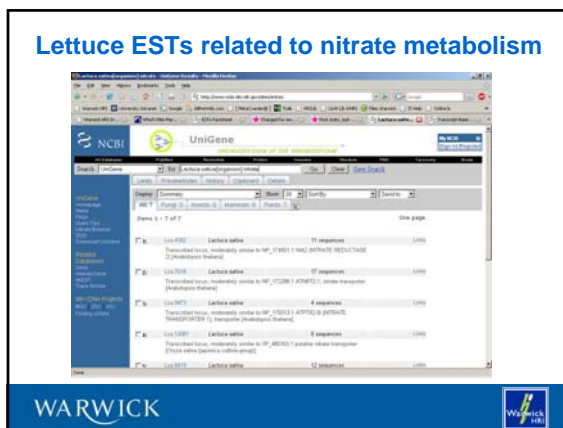
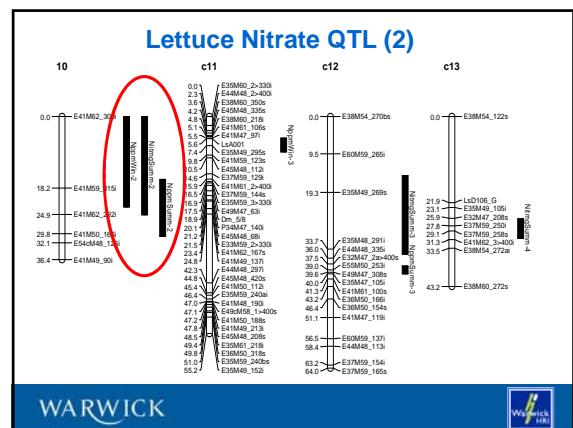
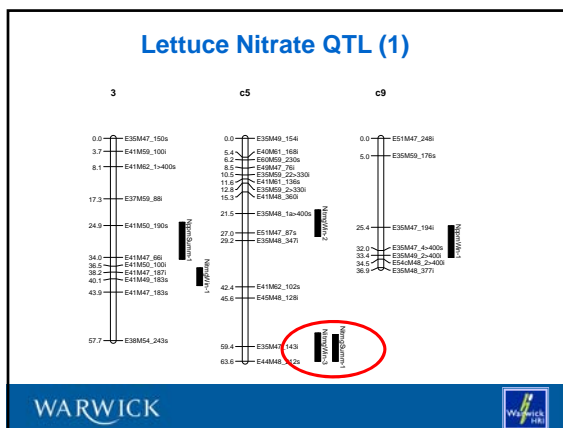
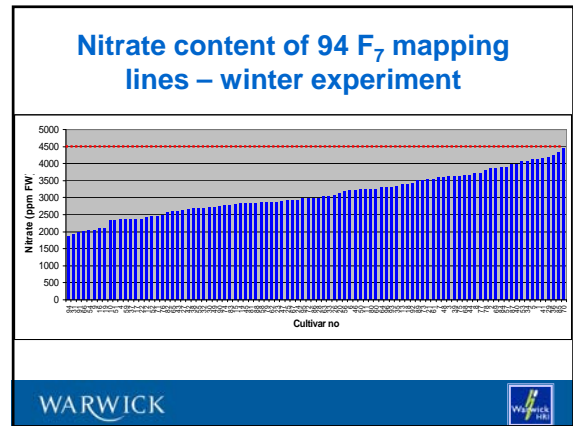
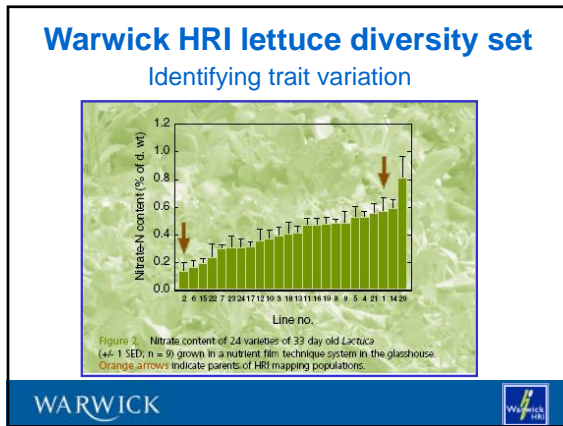


Lettuce nitrate content

Background

- Concern over high nitrate levels in the diet
- EU places limits on nitrate in lettuce
 - Lower light levels in Northern Europe limit assimilation
 - Temporary derogations – until Dec. 2008
- Cultural practices can minimise accumulation
- Can nitrate levels be reduced through breeding?

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VeGIN Lettuce Programme

Objectives

- Develop lettuce genetic resources
- Further understand key traits for sustainable production

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Develop Lettuce Genetic Resources

- Diversity set
 - Increase to 96 accessions (domestic & wild)
 - Extract & amplify DNA from single plants
- Mapping population
 - Produce F8 seed by single seed descent in 2009

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Develop Lettuce Genetic Resources

• Map improvement & marker discovery

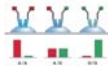
- Current F7 Saladin x Iceberg RIL map
 - 140 AFLPs
 - 57 SNPs
 - 20 ESTs
 - 1 cos marker
 - 1 gene (seed coat colour: *w*)
 - **Total: 219 markers**
- 15 linkage groups covering all 9 chromosomes
- Map length of 723.2 cM, covers >80% genome
- All groups link with the integrated lettuce map by Truco *et al.*, 2007.

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Develop Lettuce Genetic Resources High-throughput genotyping & sequencing

- Polymorphic ESTs
 - Illumina GoldenGate genotyping of F₇ RILs (2 x 384)
- SNP discovery
 - Lettuce 6.6 million feature Genechip® (35,000 genes)
- Genome & transcriptome sequencing
 - Illumina Genome analyser
- Targets
 - 1500 polymorphic SNPs for mapping
 - High-density lettuce map anchored to Michelmore integrated map
 - Extensive lettuce genome sequence

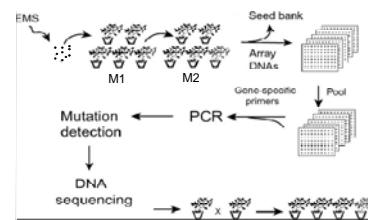


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A lettuce TILLING population

Targeting Induced Local Lesions IN Genomes

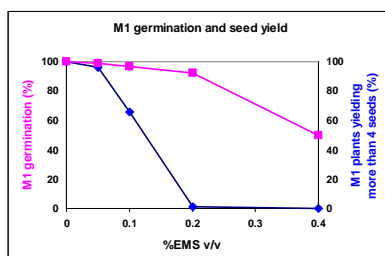


From: Henikoff, S. & Comai, L. (2003) Ann. Rev. Plant Biol. 54: 375-401

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Effect of EMS on lettuce seed



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Effects of EMS on M₁ leaf development & pigmentation

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Lettuce TILLING Populations

Larissa (65 lines) **Saladin (1,000 lines)**

0.1% EMS
18hrs

M0 seed → M1 seed → M1 plants → M2 seed → M2 plants → M3 seed

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Lettuce TILLING population 2008/09

- Approx 4000 M2 plants sown
- Sampled for DNA extraction & pooling
- Seed collected from 3257 plants
- Some phenotypes recorded
- Screening for bolting/flowering mutants

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Traits for Sustainable Production

Resistance to *Nasnovia ribis-nigri*

- Resistance already deployed **BUT**
- New resistance-breaking biotype of *N. ribis-nigri*
- Assay diversity set for resistance

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Traits for Sustainable Production

Postharvest Discolouration

- Fine map main effect QTL
– Linked markers
- Map ESTs of genes in discolouration pathways
– Identify potential target genes through links to QTL

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Other linked projects

- Attachment of human pathogens to salad leaves – BBSRC
- Control of bolting & flowering – BBSRC & Industry

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