Breeding blackcurrant varieties – new tools and directions

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James Hutton Institute

Plan

- Where we started…
- Breeding programme structure and timescales
- Gall mite resistance
- Anthocyanin content
- Environmental resilience
- Tools and resources
- Future thoughts

Back to the past…

- 30 years ago
  - 2-3 commercial vars.
    - Ben Lomond, Baldwin
  - No commercially-funded breeding
  - Objectives almost entirely agronomic
    - Frost resistance
    - Yield
  - No fresh market for blackcurrants
  - Limited underpinning science
    - Mechanisms of frost resistance
  - Biggest UK problems – frost damage at flowering time and gall mite

JHI Group blackcurrant breeding

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- Breeding at SCRI from 1960s
- Commercially funded since 1990

SmithKline Beecham   GlaxoSmithKline Consumer Healthcare   LUCOZADE RIBENA SUNTORY

- First release – Ben Lomond (1974)
    - Frost tolerance was the main objective initially
- Subsequent releases increasingly successful commercially
- Very wide *potential* genetic base – cv. and species collection
- Extensive genomic resources developed at JHI

    - Linkage maps, outline genome sequence

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Breeding Objectives

Fruit quality

- High Brix/acid ratio
- Low total acidity
- Anthocyanins
- Sensory traits
- Vitamin C
- Berry size

- Berry size (2 g +)
- Green strigs preferred
- Higher Brix/acid ratio

Processing

95% of fruit used for processing

Processing quality requirements

Agronomical suitability

Interest in nutritional aspects of the fruit

Fresh market

Agronomic

- Environmental resilience
- Winter chill levels
- Pest resistance
- Acceptable crop yield
- Juice yield also quantified

Often different cultural practices

- Hand harvesting
- Often grown with supports

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Breeding timescales

Pre-breeding → Selection of appropriate parents

x

Year 0 → Hybridisations made in insect-proof greenhouse

Year 1 → Seed germination, seedlings raised (up to 20k)

Year 2 → **Stage I** selections (vegetative characters only)

Year 3-5 → **Stage II** selections (fruiting characters), single plants ← Marker deployment

Years 6-8 → **Stage III** selections (fruiting characters), 5-plant units

Years 9-12 → Commercial trials in England

Years 13 onwards → Propagation of virus-tested plants for release to industry

Gall mite resistance 1

- Resistant backcross material based on *Ce* gene from gooseberry
  - Single gene, complex introgression into blackcurrant

- Up to $BC_{9\text{-}10}$ to restore processing quality and agronomic performance
  - Emerging linkage between *Ce* and berry size, also low anthocyanins and frost susceptibility

- Selection for resistance up to 2010 based on infestation plot data

Gall mite resistance 2

- Molecular marker linked to *Ce* resistance gene developed in 2010

- Whole progenies screened for resistance using marker

  - crosses made with *Ce*-containing parent

  - usually combined with high quality large fruited type

  - prioritisation of objectives

- Field plantings of resistant material

  - entire plots of resistant material now available at JHI

  - resistant seedlings in commercial trials

  - Ben Finlay

- Single gene resistance….

  - New sources required

New trial lines

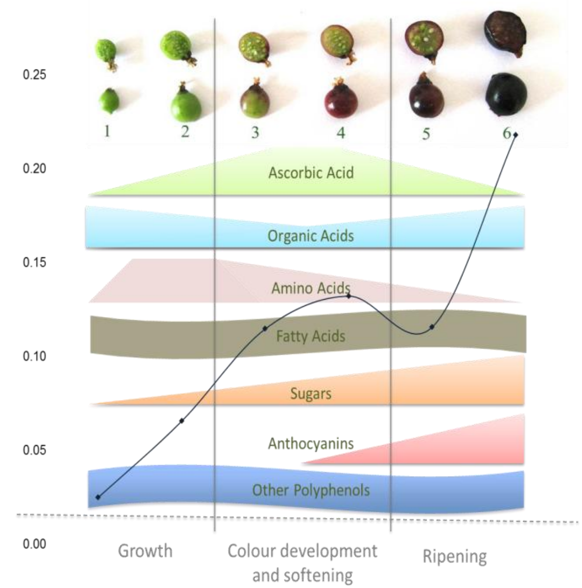
- **JHL 9918-2**
  [(Ben Tirran x EM B1834-164) x Ben Hope]
  - Gall mite resistant
  - Late season
  - Tall upright habit
  - Medium Brix, AsA > 200 mg/100ml
  - 2015 yield 8.4 t/ha

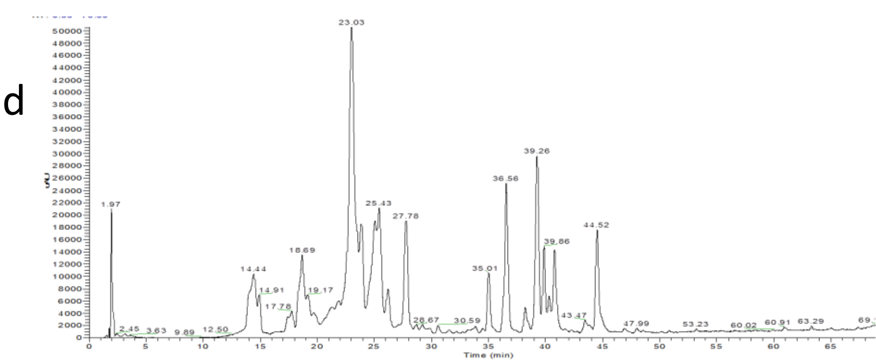
- **JHL 91129-1** [Ben Dorain x EM B1836-120]
  - Gall mite resistant
  - Mid-season
  - Compact growth
  - Medium Brix, AsA > 240 mg/100ml

- **JHL 00-54-21**
  [Ben Hope x (Ben Alder x F6/3/39)]
  - Mid-season
  - Compact habit, slightly spreading
  - Medium-large fruit (*ca*. 1g)

Anthocyanin content

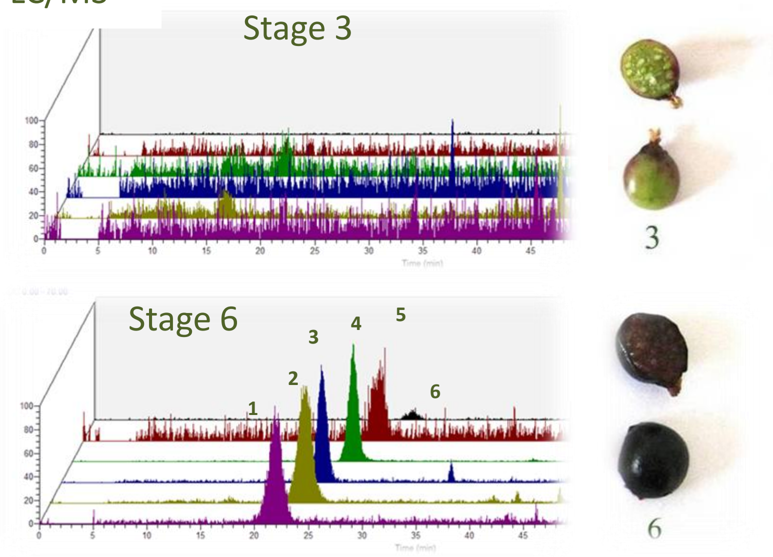
- *Ca*. 400 seedlings p.a. from JHI breeding programme assessed for anthocyanin content
  - HPLC measurements for total and individual anthocyanins
  - Other quality traits eg AsA
- Preferential selection for high delphinidins vs. cyanidins
  - Initially for stability of colour, now potentially for health-related attributes
- High levels of anthocyanins in Ben Alder and offspring
  - Strong maternal effects in heritability
  - New material emerging with enhanced anthocyanin profiles



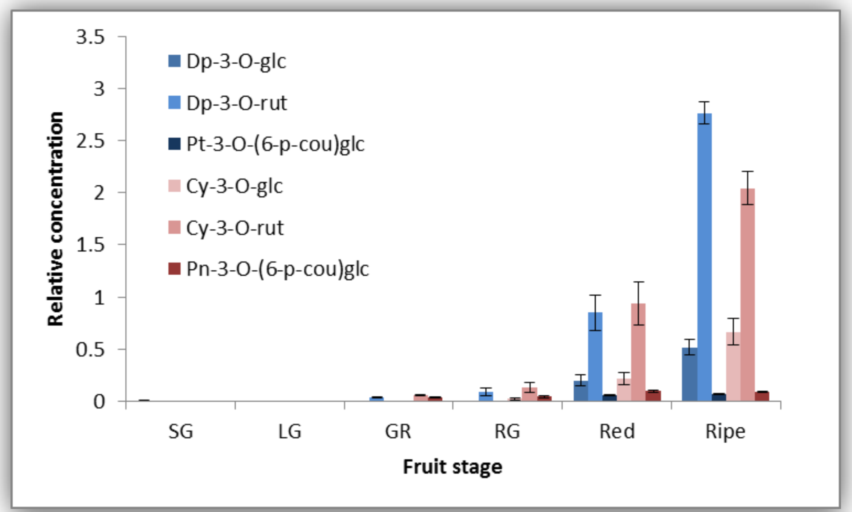


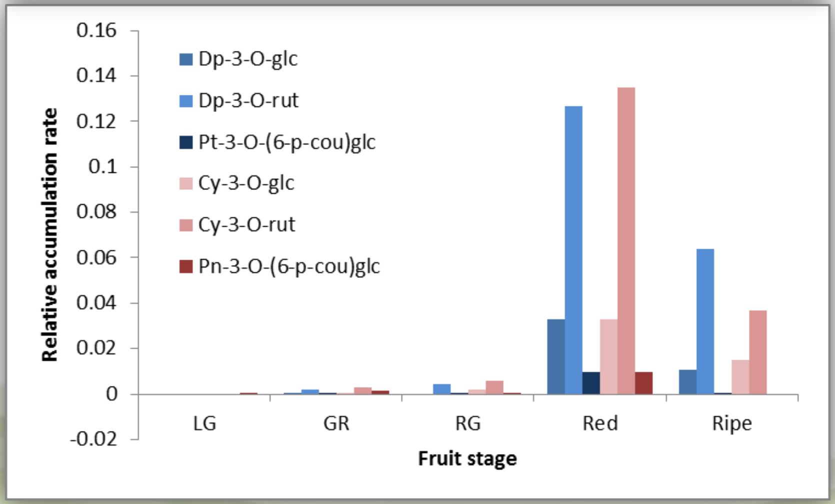
Anthocyanin accumulation

LC/MS



peak(s)	MS (*m*/ *z*)	MS-MS (*m*/ *z*)	tentative identification
			Anthocyanins
1	465	303	delphinidin 3-*O*- glucoside
2	611	303, 465	delphinidin 3-*O* -rutinoside
3	449	287	cyanidin 3-*O* -glucoside
4	595	449	cyanidin 3-*O* -rutinoside
5	625	317	petunidin -3-(6''-coumaroyl)glc
6	609	301	peonidin-3-(6''-coumaroyl)glc





Candidate genes for anthocyanin accumulation in blackcurrant fruit

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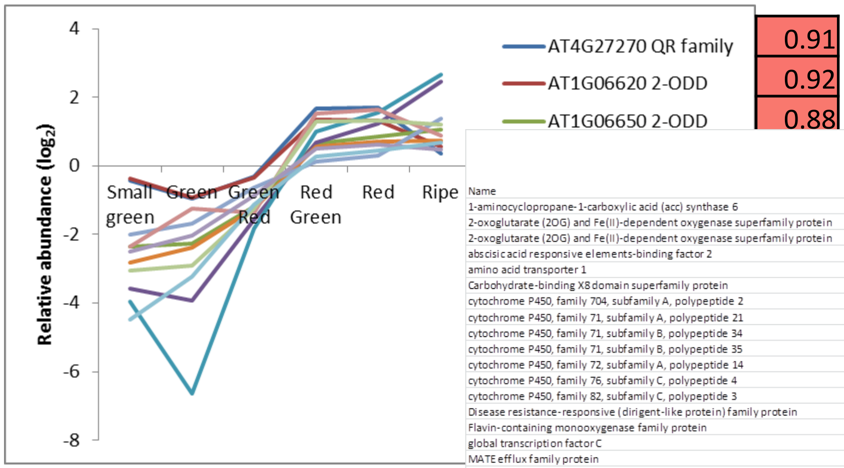
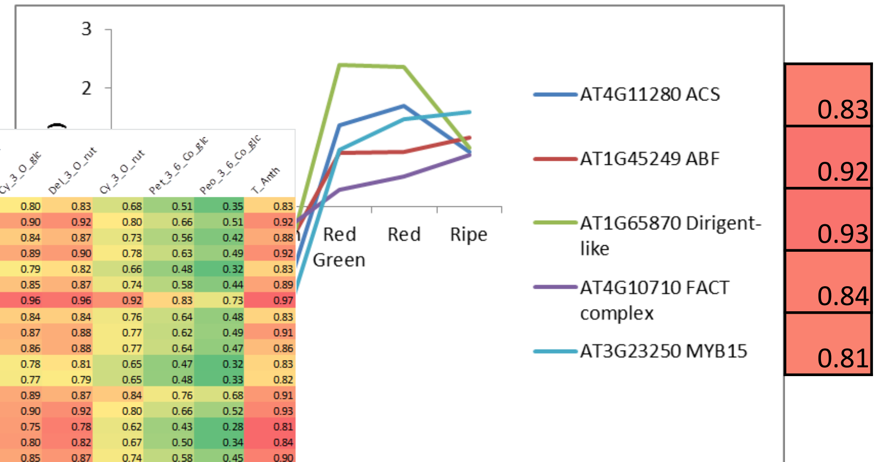
Structural genes

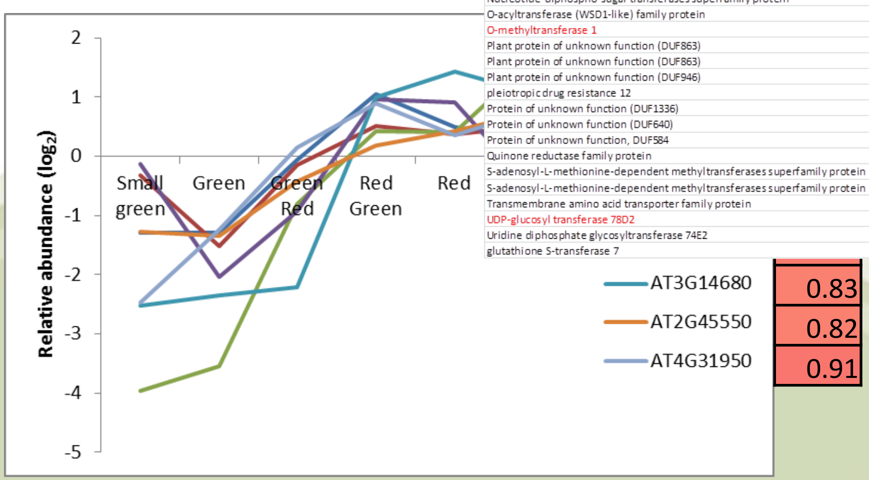


Regulatory genes



Name	Del_3_O_glc_	Cy_3_O_glc	Del_3_O_rut	Cy_3_O_rut	Pet_3_6_Co_glc	Peo_3_6_Co_glc	T_Anth
1-aminocyclopropane-1-carboxylic acid (acc) synthase 6	0.73	0.80	0.83	0.68	0.51	0.35	0.83
2-oxoglutarate (2OG) and Fe(II)-dependent oxygenase superfamily protein	0.84	0.90	0.92	0.80	0.66	0.51	0.92
2-oxoglutarate (2OG) and Fe(II)-dependent oxygenase superfamily protein	0.77	0.84	0.87	0.73	0.56	0.42	0.88
abscisic acid responsive elements-binding factor 2	0.82	0.89	0.90	0.78	0.63	0.49	0.92
amino acid transporter 1	0.71	0.79	0.82	0.66	0.48	0.32	0.83
Carbohydrate-binding X8 domain superfamily protein	0.78	0.85	0.87	0.74	0.58	0.44	0.89
cytochrome P450, family 704, subfamily A, polypeptide 2	0.93	0.96	0.96	0.92	0.83	0.73	0.97
cytochrome P450, family 71, subfamily A, polypeptide 21	0.78	0.84	0.84	0.76	0.64	0.48	0.83
cytochrome P450, family 71, subfamily B, polypeptide 34	0.80	0.87	0.88	0.77	0.62	0.49	0.91
cytochrome P450, family 71, subfamily B, polypeptide 35	0.81	0.86	0.88	0.77	0.64	0.47	0.86
cytochrome P450, family 72, subfamily A, polypeptide 14	0.71	0.78	0.81	0.65	0.47	0.32	0.83
cytochrome P450, family 76, subfamily C, polypeptide 4	0.69	0.77	0.79	0.65	0.48	0.33	0.82
cytochrome P450, family 82, subfamily C, polypeptide 3	0.85	0.89	0.87	0.84	0.76	0.68	0.91
Disease resistance-responsive (dirigent-like protein) family protein	0.84	0.90	0.92	0.80	0.66	0.52	0.93
Flavin-containing monooxygenase family protein	0.67	0.75	0.78	0.62	0.43	0.28	0.81
global transcription factor C	0.72	0.80	0.82	0.67	0.50	0.34	0.84
MATE efflux family protein	0.78	0.85	0.87	0.74	0.58	0.45	0.90
MATE efflux family protein	0.77	0.84	0.86	0.73	0.57	0.41	0.88
multidrug resistance-associated protein 14	0.83	0.89	0.90	0.79	0.65	0.51	0.92
multidrug resistance-associated protein 14	0.67	0.76	0.78	0.63	0.46	0.32	0.82
multidrug resistance-associated protein 2	0.82	0.88	0.90	0.78	0.63	0.47	0.90
myb domain protein 15	0.68	0.75	0.79	0.62	0.44	0.30	0.81
Nucleotide-diphospho-sugar transferases superfamily protein	0.72	0.80	0.82	0.68	0.52	0.38	0.85
O-acyltransferase (WSD1-like) family protein	0.76	0.84	0.86	0.73	0.56	0.43	0.88
O-methyltransferase 1	0.81	0.87	0.89	0.76	0.61	0.47	0.91
Plant protein of unknown function (DUF863)	0.94	0.94	0.93	0.94	0.91	0.81	0.89
Plant protein of unknown function (DUF863)	0.67	0.76	0.78	0.63	0.46	0.30	0.80
Plant protein of unknown function (DUF946)	0.72	0.75	0.71	0.74	0.70	0.65	0.74
pleiotropic drug resistance 12	0.74	0.81	0.84	0.69	0.52	0.37	0.86
Protein of unknown function (DUF1336)	0.77	0.80	0.82	0.73	0.64	0.47	0.77
Protein of unknown function (DUF640)	0.88	0.91	0.89	0.89	0.83	0.74	0.90
Protein of unknown function, DUF584	0.82	0.88	0.90	0.79	0.64	0.49	0.90
Quinone reductase family protein	0.82	0.88	0.90	0.78	0.63	0.48	0.91
S-adenosyl-L-methionine-dependent methyltransferases superfamily protein	0.82	0.88	0.91	0.78	0.63	0.49	0.92
S-adenosyl-L-methionine-dependent methyltransferases superfamily protein	0.78	0.85	0.87	0.74	0.58	0.44	0.90
Transmembrane amino acid transporter family protein	0.86	0.91	0.91	0.84	0.73	0.61	0.93
UDP-glucosyl transferase 78D2	0.67	0.75	0.78	0.62	0.43	0.27	0.80
Uridine diphosphate glycosyltransferase 74E2	0.78	0.85	0.87	0.74	0.57	0.43	0.89
glutathione S-transferase 7	0.84	0.90	0.92	0.80	0.66	0.52	0.93

Cytochrome



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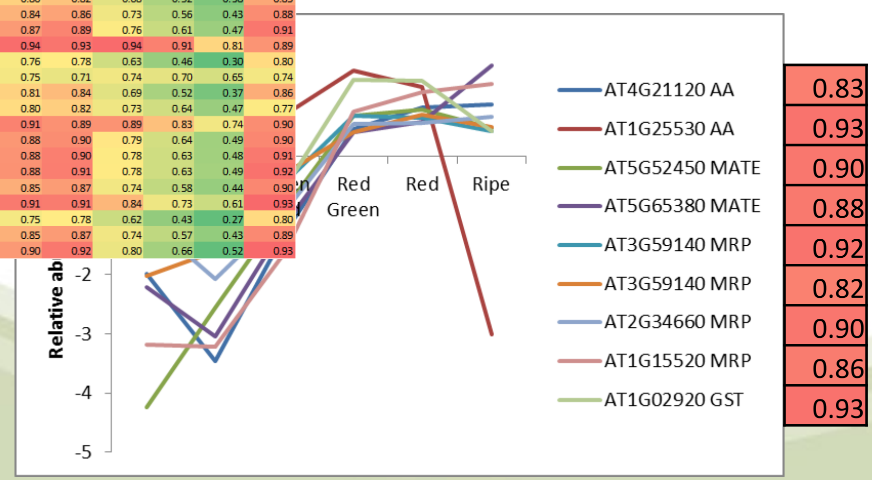

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Pathway to new varieties with enhanced anthocyanin content

- Genes controlling anthocyanin content are being identified

- Genes/alleles specifically linked to high anthocyanins (delphinidins)

- Markers developed for selection of new high-anthocyanin varieties (2018?)

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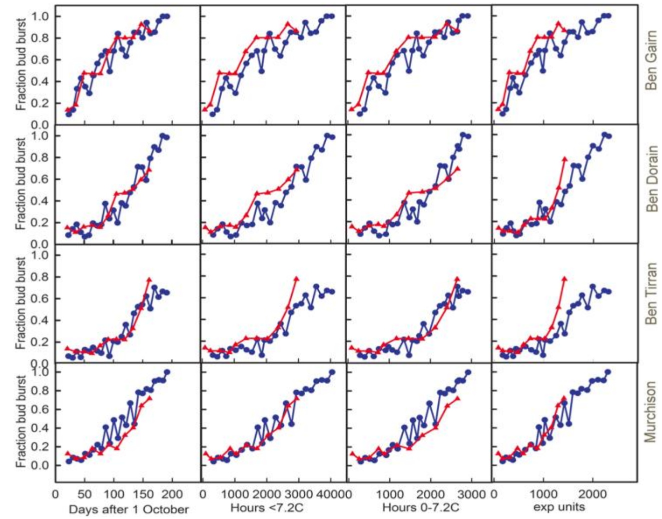
Environmental resilience

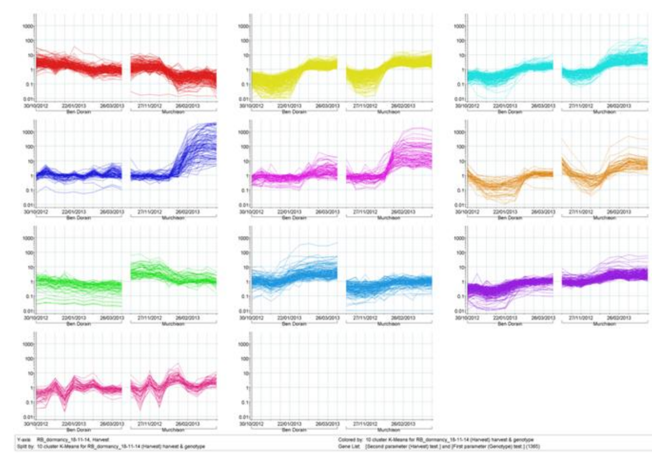
- Response to warmer winters in Europe
  - 2006-7, 2013-14, 2014-15, 2015-16
  - Economically significant effects
- Screening breeding lines for chill requirement
  - Cuttings taken throughout winter placed into a forcing environment
  - Differing response between varieties
- Chill modelling based on budbreak data for diverse varieties in progress
  - Models based simply on temperature thresholds show only limited alignment with field observations

UK Trade & Investment

Innovate UK
Technology Strategy Board

- NZ collaboration
  - Joint mapping population
  - Mapping of traits



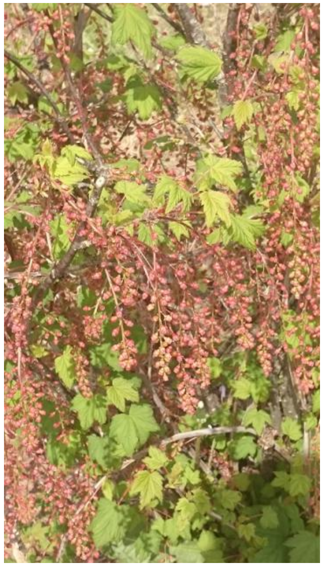


Tools and resources 1 - Germplasm

- Diverse range of breeding populations, varieties and species at JHI
    - > 13k individual genotypes

- Links to other germplasm collections within Europe and elsewhere

- *Ribes* accessions from the National Fruit Collections are being transferred to JHI



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Tools and resources 2 - Genomics

- Linkage maps developed
  - First map in 2008
  - Latest GbS map 2014
  - Further GbS mapping of new mapping populations incl. NZ collaboration
- Trait locations
  - > 50 key metabolites incl. anthocyanins, AsA etc. mapped onto latest GbS map
- Markers for breeding
  - Gall mite marker 2010
  - Berry size markers under validation
  - Quality and developmental trait markers in progress

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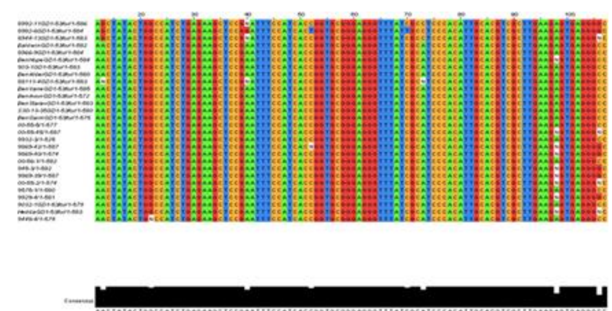
Blackcurrant Genome

- Genome sequencing of *Ribes nigrum* cv. Ben Finlay
  - Whole genome shotgun sequencing (WGS)
  - Mate-pair (MP) scaffolding sequencing
  - **First reported genome sequence for blackcurrant**

- Specialised browser developed at JHI for identifying genes linked to fruit quality and climate effects
  - Will be made available to other groups after publication



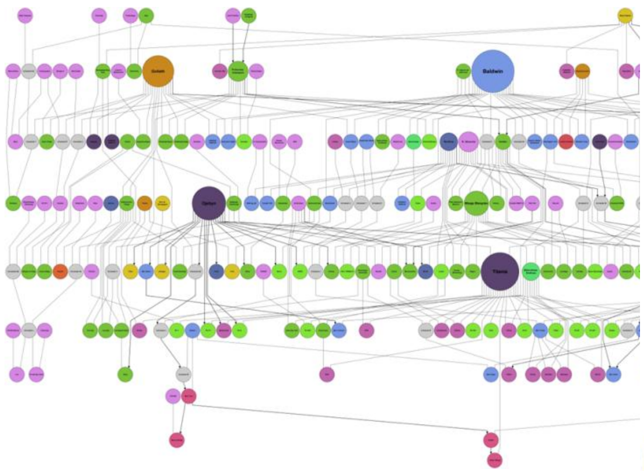
Changing positions

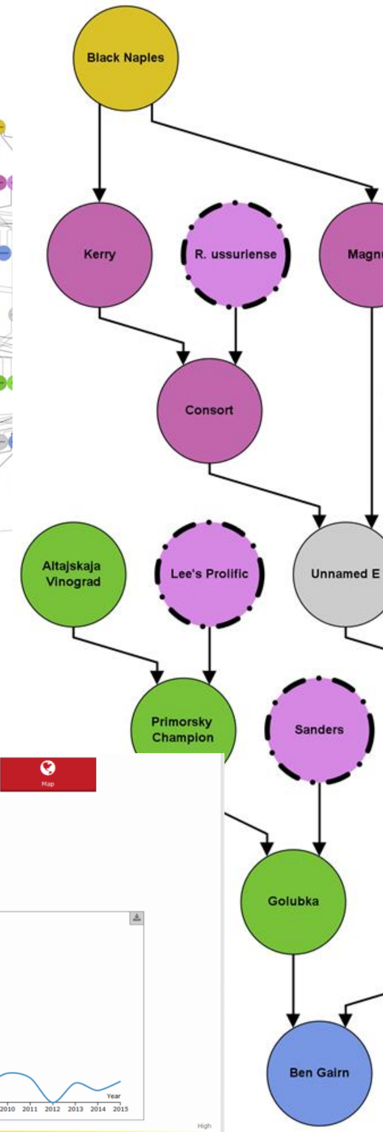
- 30 years ago
  - 2-3 commercial vars.
    - Ben Lomond, Baldwin
  - No commercially-funded breeding
  - Objectives almost entirely agronomic
    - Frost resistance
    - Yield
  - No fresh market for blackcurrants
  - Limited underpinning science
    - Mechanisms of frost resistance
    - No computers
  - Biggest UK problems – frost damage at flowering time and gall mite

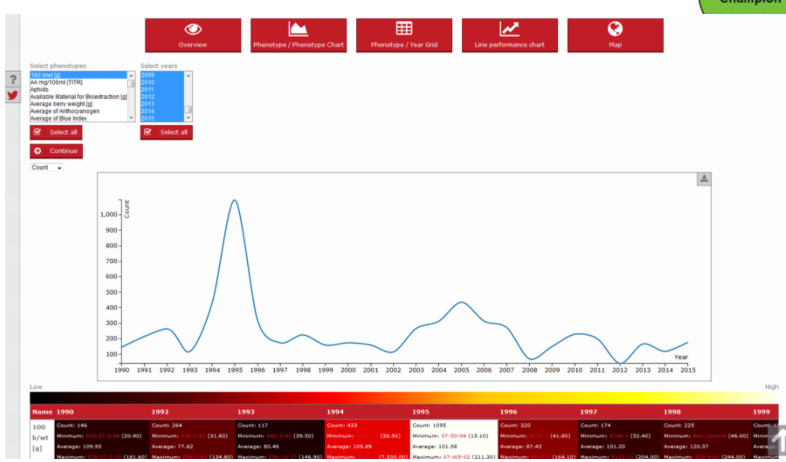
- 2016
  - 10 commercial vars.
  - Breeding funded commercially for over 25 years
  - Objectives related to quality plus agronomy
  - Growing fresh market for blackcurrants
  - Unrivalled scientific base underlying the breeding programme
    - Genomics and genetic analysis
    - Computer-based analyses
  - Biggest UK problems – lack of winter chilling, gall mite
    - (frost damage increasing)

`Germinate' database

- Quality data from breeding programme
  - *ca.* 35 years
- Crossing and pedigree details
  - *ca.* 40 years
- Trials data (agronomic and quality)
  - *ca.* 25 years
- New database under construction
  - Access will be public for some areas, restricted for others
  - `Germinate'
  - Pedigree details - `Helium'

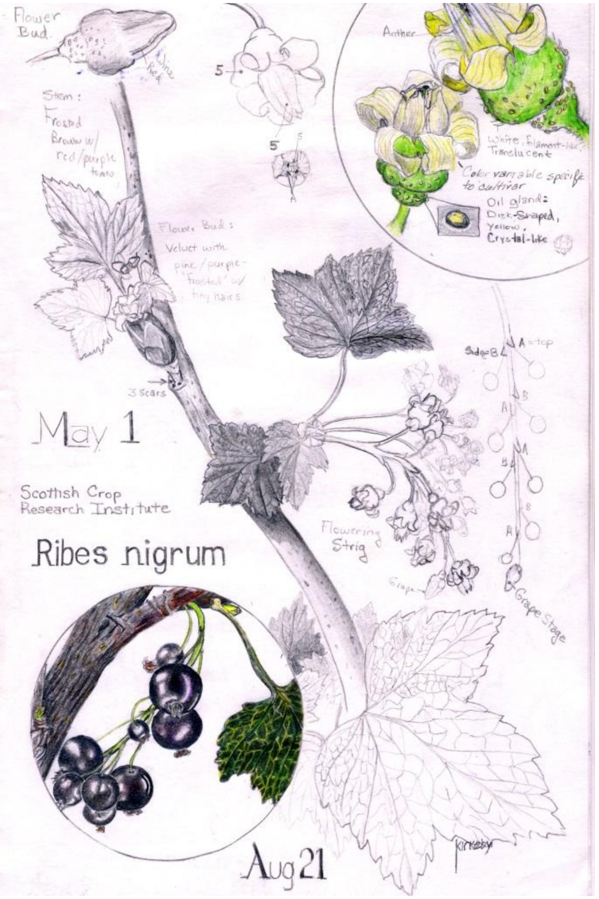






Future directions

- Expansion of genetic base within breeding programmes
- Varieties linked to new products
  - Selection of seedlings with specific attributes linked to NPD
  - Specialised supply chain management to ensure quality
- Integrated crop management
  - Better resistance to pests and diseases
  - Reduced inputs
- Full exploitation of genomics platforms
  - Gene-based markers for quality and environmental traits
  - Improved breeding efficiency
  - Faster delivery of new varieties



Acknowledgements

James Hutton Institute

Sandra Gordon
Dorota Jarret

Linzi Jorgensen
Joanne Russell
Pete Hedley
Linda Milne
Rob Hancock
Paul Shaw

James Hutton Limited

Jonathan Snape
Lesley Beaton

BioSS
Chris Hackett
Katharine Preedy

University of Dundee
Lyn Jones

University of Reading
Matt Ordidge
Nick Battey

New Zealand
Alastair Currie
Kathryn Wright

Poland
Stan Pluta
Bogusia Badek
Margaret Korbin

UK Growers

Funding

Breeding
LR Suntory (2015-)
James Wickham
Harriet Roberts
Rob Saunders
Jonathan Gilkes
Paul Dockerty *et al*. (Thatchers)

GlaxoSmithKline (1990-2015)

Winterwood Farms
Steve Taylor
Alan Reeves

Research
Scottish Government
Innovate
EU