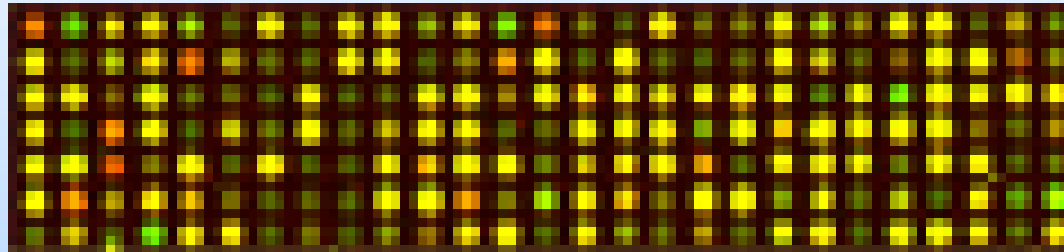


JIC/JCVI/Cogenics *Brassica* microarray

12th January 2008

Ian Bancroft
John Innes Centre



Array design

Assembly statistics:

810,254 raw ESTs

803,326 ESTs after clean-up

Assembly conducted at 95% cut-off

Paralogues within C genome 86.5 +/- 3.6% conserved in coding regions, so should resolve

Homoeologues between A and C genomes 96.8 +/- 1.2% conserved in coding regions, so should co-assemble

94,558 unique sequences

42,642 assemblies

51,946 singletons

72,148 sequences have UniRef100 annotation

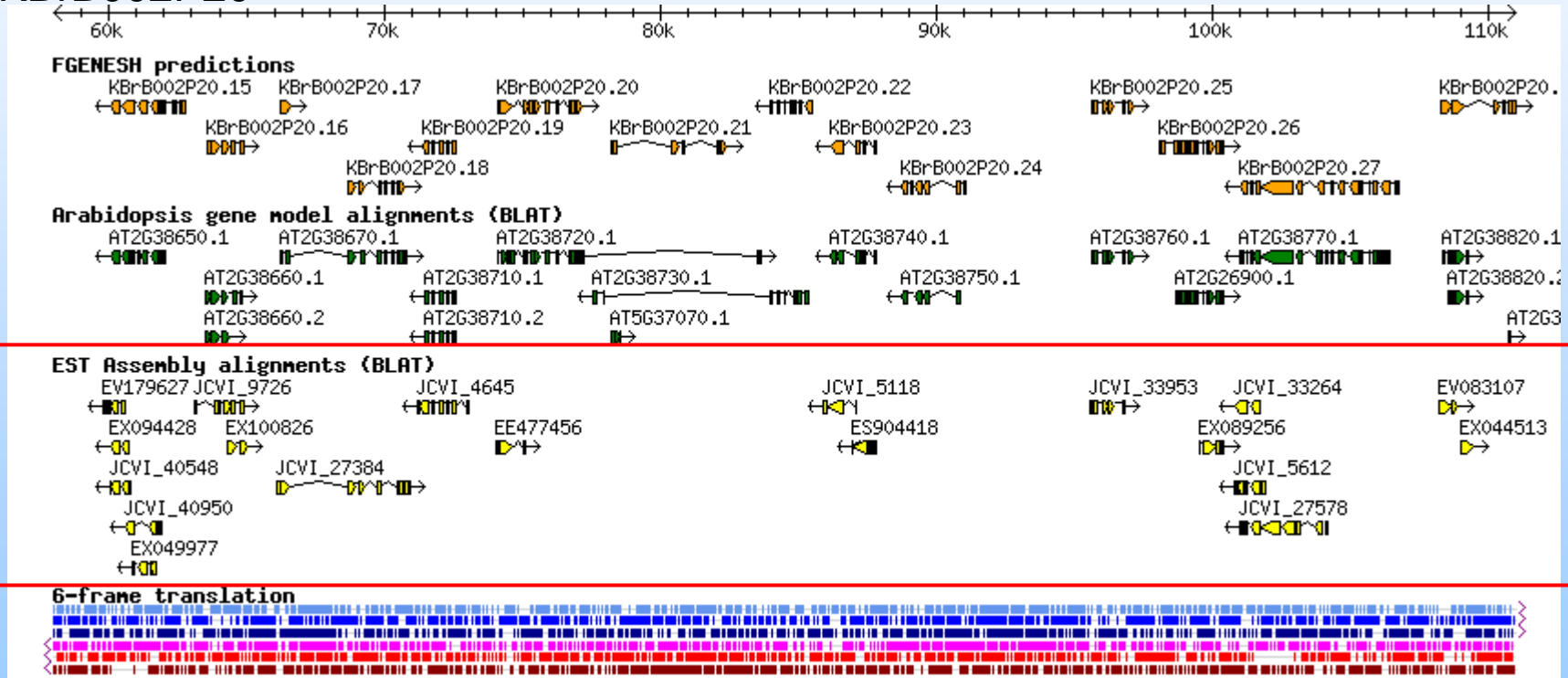
Assemblies available from:

<ftp://149.155.100.41/pub/brassica/>

Array design

Assemblies (and singletons) incorporated into the JIC *Brassica* genome sequence annotation (accessible via *Brassica* Genome Gateway site: <http://brassica.bbsrc.ac.uk>)

KBrB002P20



Array design

Array statistics:

Agilent 2 x 105k format (<http://www.chem.agilent.com>)

60-mer oligos

91,854 good probes designed

81,388 probes with no cross-hybridization potential

10,466 probes have cross-hybridization potential

Access to array:

Freely available, but as it is an Agilent custom array, access needs to be authorised. Contact:

Jim Lovgren (Cogenics; USA): jlovgren@cogenics.com

JIC Genome Lab: enquiries@jicgenomelab.co.uk

Commercial service provider: Cogenics

Array validation

Aim:

As most ESTs in the assemblies were from *B. napus*, we aimed to confirm that homoeologues in the “diploid” species hybridize efficiently to the probes.

We anticipate no problems, as homology in coding regions is very high

B. oleracea - *B. napus* C genome: 99.85 +/- 0.14% conserved

B. rapa - *B. napus* A genome: 98.63 +/- 0.80% conserved

Array validation

Experiment:

Analyse leaf transcriptome in *B. oleracea* (Alboglabra A12DH), *B. rapa* (Trilocularis RO18) and reciprocal resynthesised *B. napus* produced from these lines



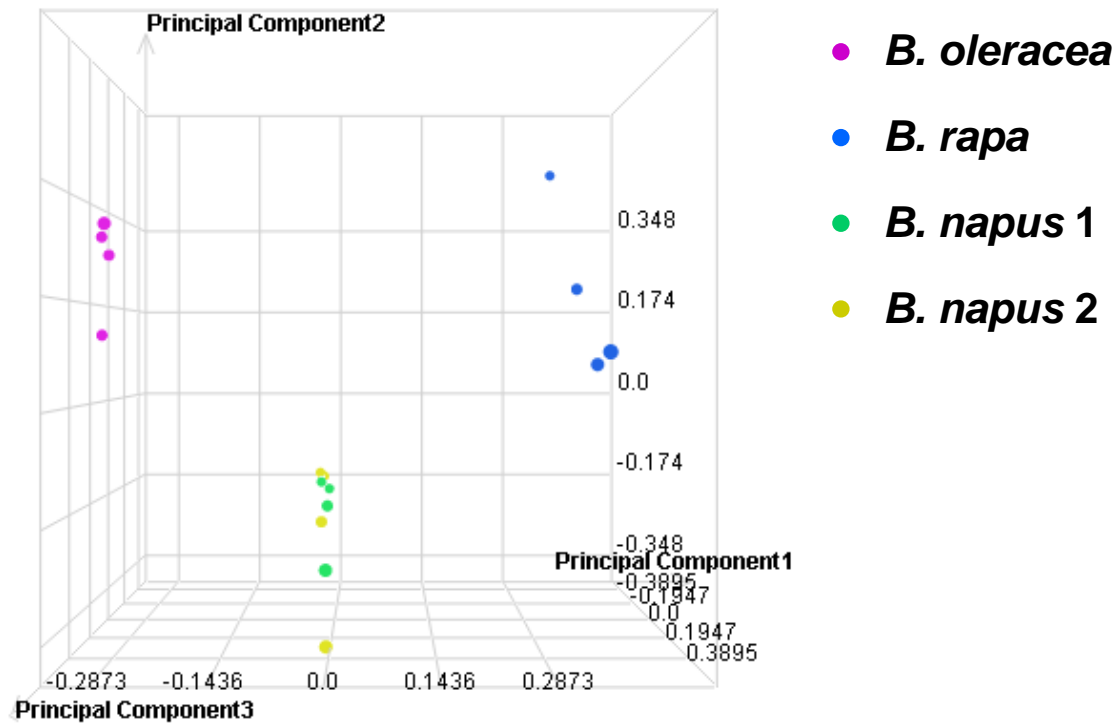
Validation results

Quantitative differences in expression can be detected between *B. rapa* and *B. oleracea* ($P < 0.005$)

Quantitative differences in expression can be detected between the reciprocal resynthesised *B. napus* ($P < 0.005$)

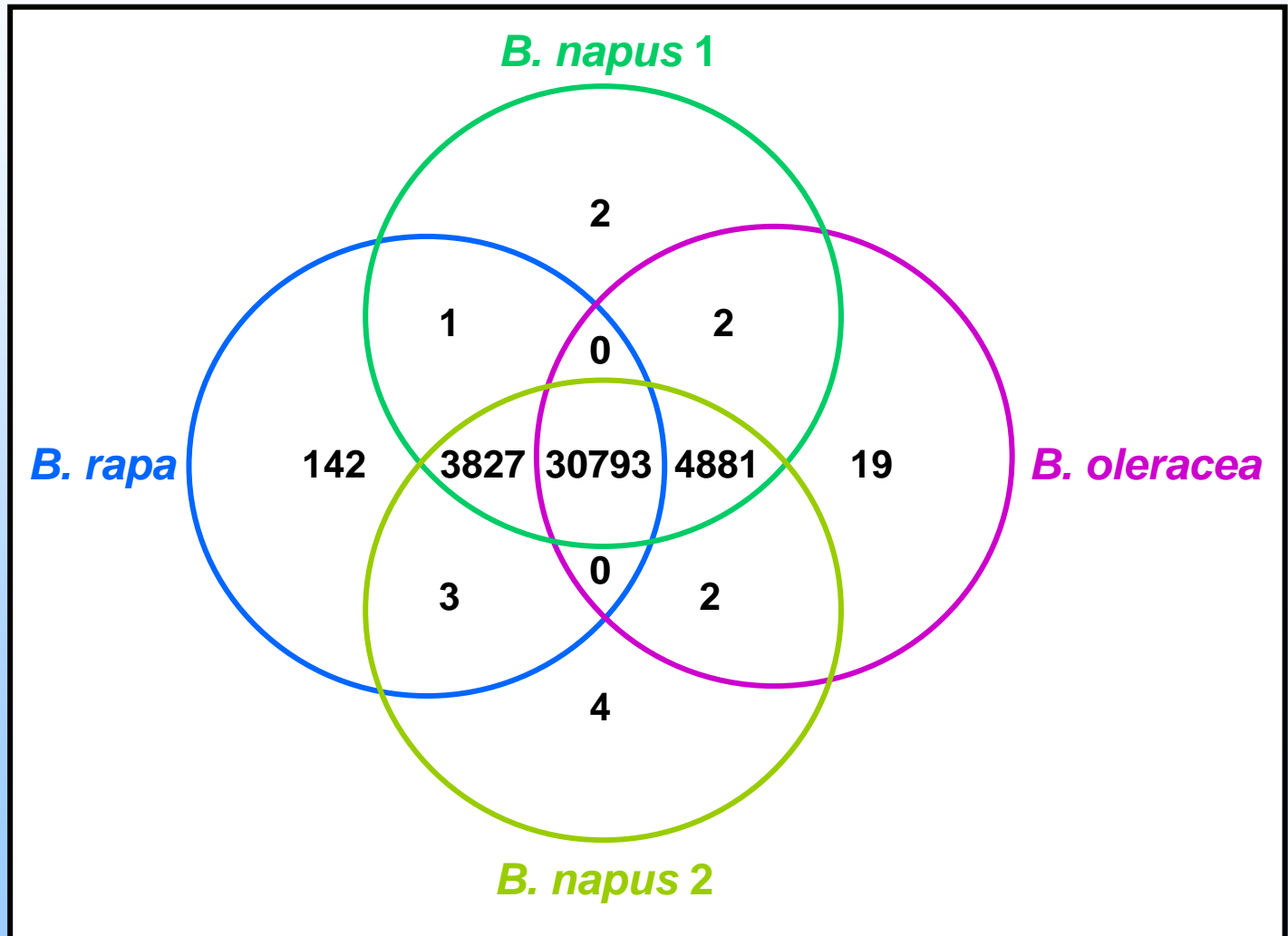
Validation results

Principal component analysis resolves the genotypes



Validation results

Genome-specific expression identified by qualitative analysis of ~34,000 unigenes



Number of probes yielding significant signals in all 4 replicates

Acknowledgements

ESTs - The *Brassica* genomics community

Assemblies

JCVI: Foo Cheung, Chris Town

Annotation

JIC: Nizar Drou, Martin Trick

Probe and array design

Cogenics: Ed Lobenhofer

Resynthesised *B. napus*

IAPB, Gottingen: Stefan Abel

Biological materials for validation experiment

JIC: Fiona Fraser

Data analysis for validation experiment

JIC, Cogenics: Andreas Magusin, Ed Lobenhofer

For further information

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