Brassica C-genome Chromosomes, Molecular markers and Linkage Groups

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Brassica Triangle of U (1935)

B. juncea
BB

B. carinata
2n=34
BBCC

B. oleracea
2n=18
CC

B. nigra
2n=16
BB

B. napus
2n=38
AACC

B. rapa
2n=20
AA

B. juncea
2n=36
AABB

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Brassica oleracea (CC, 2n =18)

Cytological mapping

Genetic linkage mapping

Sequence mapping

Physical mapping
Cytological mapping
Heneen et al. (1995)

*B. rapa*  
*B. oleracea*
Mitotic karyotypes (Cheng et al. 1995)

**B. rapa (AA, 2n=20)**

**B. oleracea (CC, 2n=18)**

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Genetic linkage mapping: Markers

Isozymes

Known genes

RFLP
AFLP
RAPD
SCAR, SRAP and CAPS
SSR
SNP
In/Del

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Slocum et al. (1990)

CC

258 RFLP markers
**Sharpe et al. (1995)**

**Parkin et al. (1995)**

**CC in AACC**

**RFLP**

**117 polymorphic loci**
Gao et al. (2007)  
CC  
1257 markers (mainly SRAP)
Wang et al. (2012)

CC

602 SSR
625 SNP
1227

186 C3
99 C9

1197.9 cM
0.98 cM
503.3 kbp

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230 single-locus in AACC when tested on CC:
125 single-locus
47 two loci
24 three or more loci
34 no amplification

Li et al. (2013)

SSR
Physical mapping: Markers

FISH
   rDNA
   Repetitive DNA
   DNA sequences
   cDNA
   SSR

GISH

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Partial physical mapping

Armstrong et al. (1998)
Correspondence between physical and genetic linkage maps

Howell et al. (2002) [MBGP: Parkin et al. (1995), Sharpe et al. (1995)]

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Complete physical mapping

Xiong and Pires (2011)

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Xiong and Pires (2011)
Armstrong et al. (1998)

Howell et al. (2002)

Xiong and Pires (2011)

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Mitotic karyotypes (Cheng et al. 1995)

B. rapa (AA, 2n=20)  

B. oleracea (CC, 2n=18)

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Development of Monosomic alien addition lines (MAALs)

Background genome
AA
2n=20
*Brassica rapa*

Dissected genome
CC
2n=18
*B. oleracea*

MAALs
AA + C₁

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AA + C₉
2n=21

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Origin of MAALs

AA + 1 C Chromosome

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Why MAALs

Relating genes and linkage groups to specific chromosomes

Physical mapping of genes and chromosome-specific DNA markers

Monitoring homoeologous chromosome pairing, intergenomic gene transfer, and phylogenetic relationships

Providing bridging materials for plant breeding

Suitable for studies on chromosome organization and gene expression

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Background genome

*Brassica rapa*

- **AA**
- 2n=20

Dissected genome

*B. oleracea ssp. alboglabra*

- **CC**
- 2n=18

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**Plant morphology**

**Cytology: Chromosome morphology**

**Molecular markers (RAPD, SSR)**

**Isozymes**

**Glucosinolates**
Monosomic Addition Line
AA + C4 Chromosome

Cheng et al. (1995)
Diakinesis: Chromatin condensation

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C6

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Meiotic chromosomes of addition line AA + chromosome C4
Meiotic chromosomes of addition line AA + chromosome C4
Diakinesis
Univalents, bivalents and multivalents

Chromosome homoeology

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Frequencies of uni-, bi-, tri- and pentavalents

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Mitotic Chromosomes of Addition Line AA + Chromosome C₉

Fluorescence in situ hybridization 25S and 5S rDNA

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Multi-target FISH

Method described by Xiong and Pires (2011)

5S rDNA
25S rDNA
BAC KBrB072L17
BAC KBrH092N24

CentBr1
CentBr2
BAC BNIH 123L05

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Simple Sequence Repeats (SSRs)

- Defining SSR markers specific to the C-genome

- Defining markers specific to each of the C-chromosomes which was decisive for developing the MAALs

- Elucidating the correspondence between the cytological nomenclature and physical maps/linkage group designations
SSRs

185 tested

88 C-genome specific

64 C-chromosome specific

2-10 specific to individual chromosomes

17 specific to 2-5 chromosomes

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Availability of the MAALs

Seeds available from the gene bank NordGen
www.nordgen.org

C1, C2, C3d, C4, C4d, C5, C6, C7, C8 and C9

Parental species AA and CC

Resynthesized AACC
Prospects: MAALs

- Physical mapping and chromosome painting.
FISH: Use of BAC clones. Multicolour pachytene chromosome painting in Arabidopsis thaliana (Lysak and Lexer 2006)
Xiong et al. (2010) A7 B. napus

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Prospects: MAALs

- Physical mapping and chromosome painting.

- Painting of heterobivalents and multivalents, and defining homoeologous A and C chromosomes.

- Expanding the list of C-chromosome specific SSRs, and physical and genetic mapping of SSRs (Cuadrado and Jouve 2010).

- Detailed studies on introgression in AA progeny plants and in AA and C chromosomes of mono- and disomic addition lines.

- Further studies on the control of seed colour exerted by seven C chromosomes. Understanding how the deletion in C4 affects the expression of the seed colour gene on this chromosome.

- Contributing to marker-assisted breeding of desirable characters.

- Comparing with *B. oleracea* – *B. rapa* MAALs (Li *et al*. 2013).

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Summary and conclusions

- Karyotype, genetic linkage groups and physical mapping of *B. oleracea* highlighted, and correspondence between designations indicated.

- All nine possible *Brassica rapa* – *B. oleracea* MAALs are now available (NordGene Bank, Alnarp, Sweden).

- Number of cotyledons, flower colour, leaf appearance and seed colour are useful characters for differentiating C-chromosome carriers. Genes on seven C-chromosomes maternally or embryonically control seed colour in a major or minor way.

- Meiotic homoeological pairing between C- and A-chromosomes infers intergenomic gene transfer.

- GISH and FISH with rDNA, repetitive sequences and BAC probes are instrumental for differentiating the alien C-chromosomes.

- SSRs specific for, and shared by, alien chromosomes are defined.

- Need of further physical mapping and chromosome painting emphasized.

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Our sincere thanks to many colleagues

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Thanks very much!