

# In-Silico Identification and Characterization of Wheat 4AL - *Triticum militinae* Introgression



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## EXECUTIVE SUMMARY

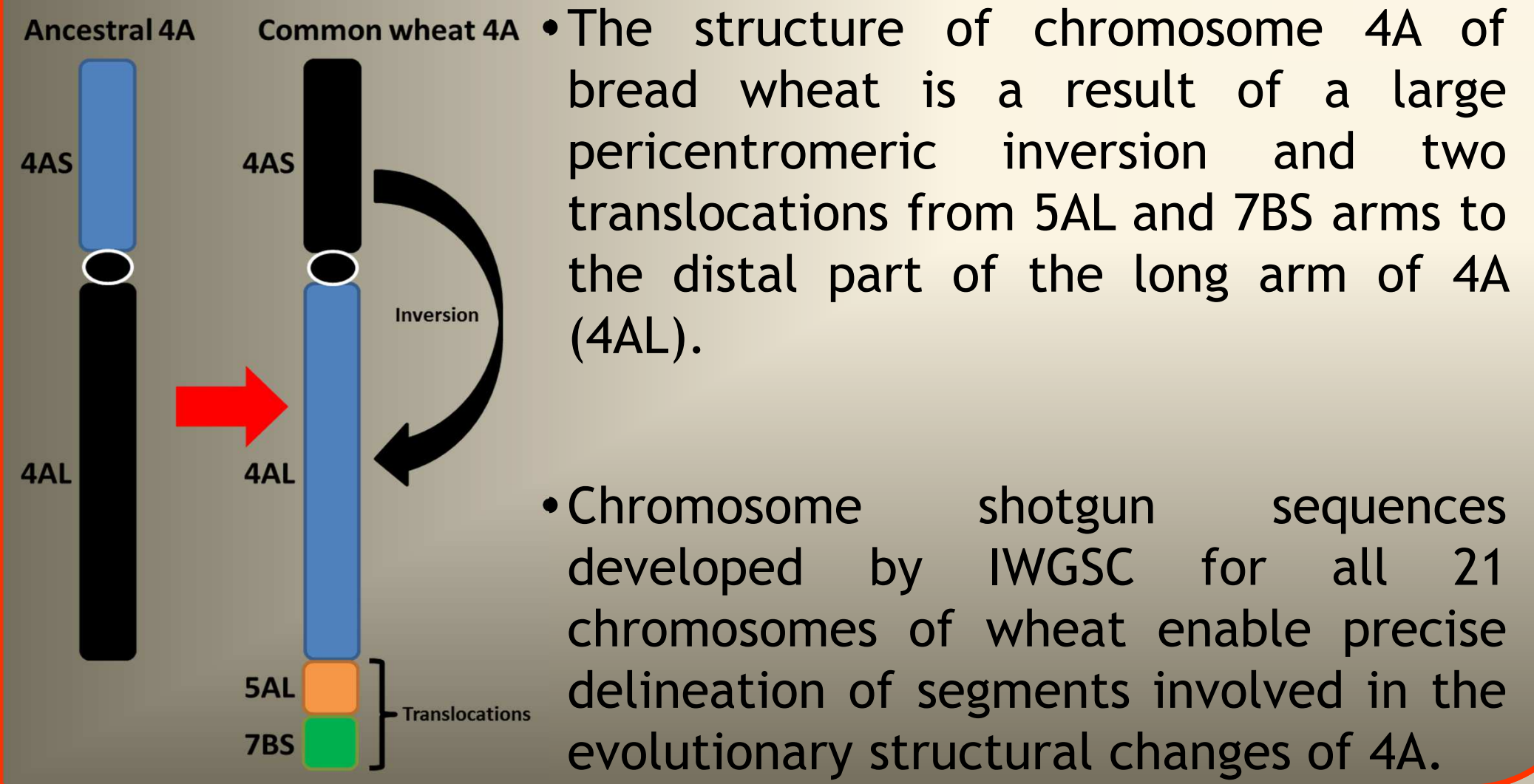
Common wheat (*Triticum aestivum*) has adapted to a wide range of environmental conditions due to the plasticity of its hexaploid genome. The ability of wheat to withstand abiotic and biotic stress can be increased by introducing new genes and alleles from wild relatives. However, the lack of appropriate molecular tools impedes rapid development of useful introgression lines, development of markers linked to the introduced traits, and cloning genes of interest.

The introgression of a segment of *T. militinae* (TM) genome to the long arm of wheat chromosome 4A (4AL) (Jakobson et al., 2006) makes this arm a suitable model to verify a possibility of identifying alien chromatin using chromosome next generation sequencing.

In addition to the recent introgression, 4AL arm carries a large pericentromeric inversion and two ancestral translocations from chromosome arms 5AL and 7BS (Hernandez et al., 2012).

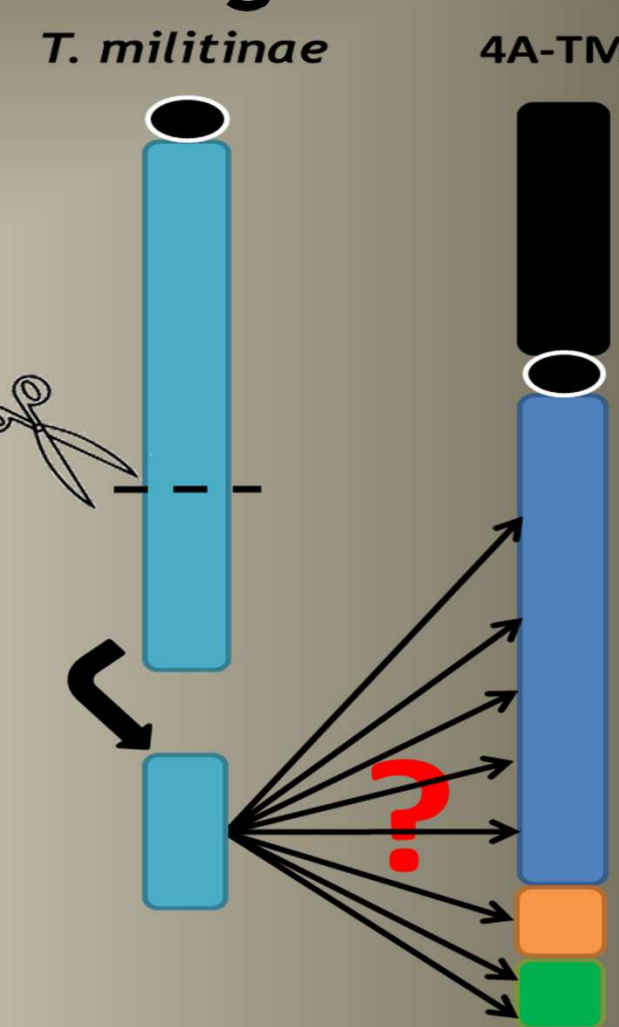
We have flow-sorted 4AL arm from cv. Chinese Spring (4AL-CS) and the same arm with a translocation from *T. militinae* (4AL-TM), and sequenced their DNA by Illumina. We then constructed virtual gene order map using Genome Zipper (Mayer et al., 2009) and 624 DArT markers available for chromosome 4A-CS. The virtual chromosome comprises 2395 ordered genes (653 and 1742 for 4AS and 4AL, respectively) that were used for comparison with 4AL-TM. This approach permitted characterization of the ancestral translocations from 5AL and 7BS and identified the segment introgressed from *T. militinae*.

### Aim 1: Identification of translocations in 4A

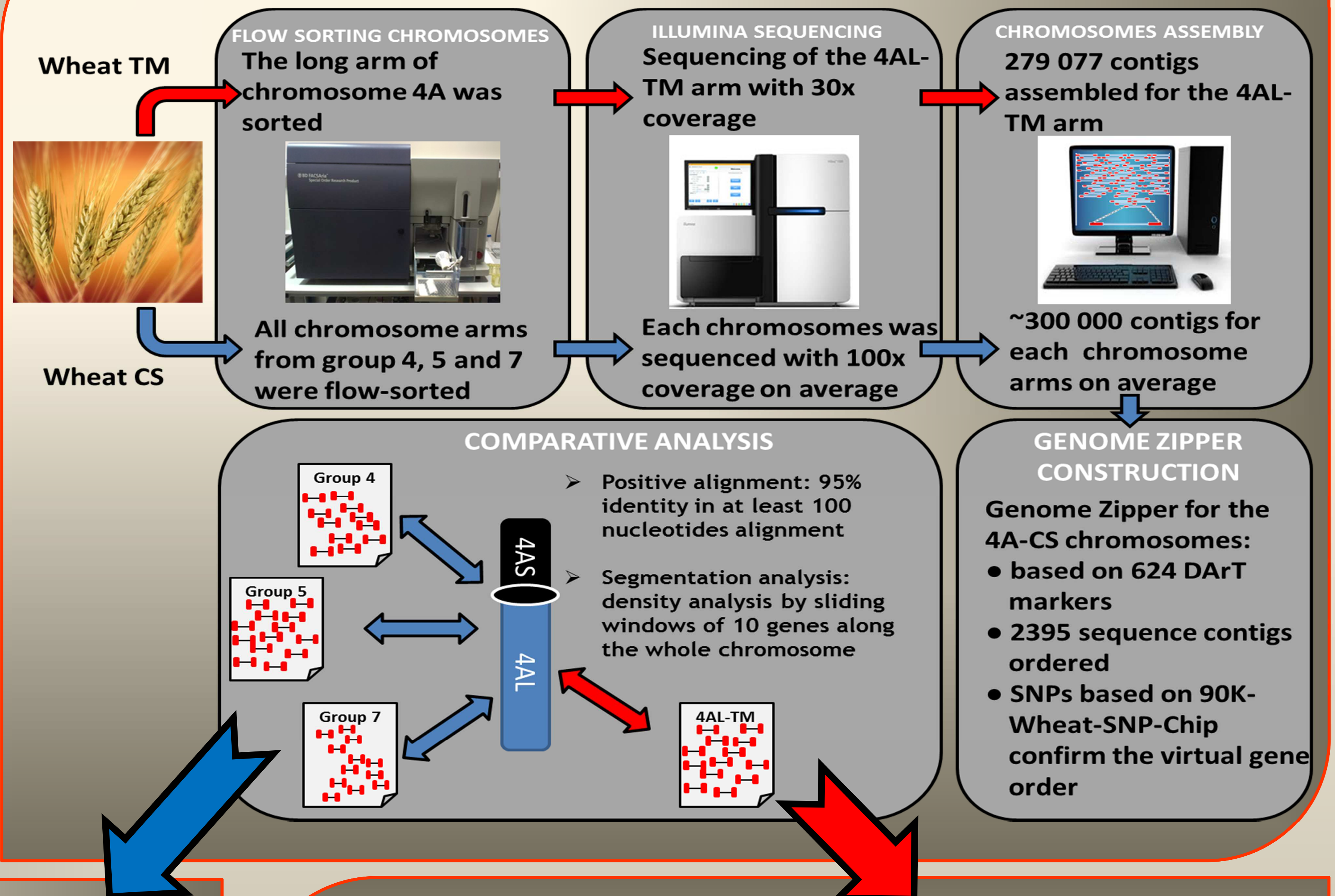


### Aim 2: Identification of alien introgression

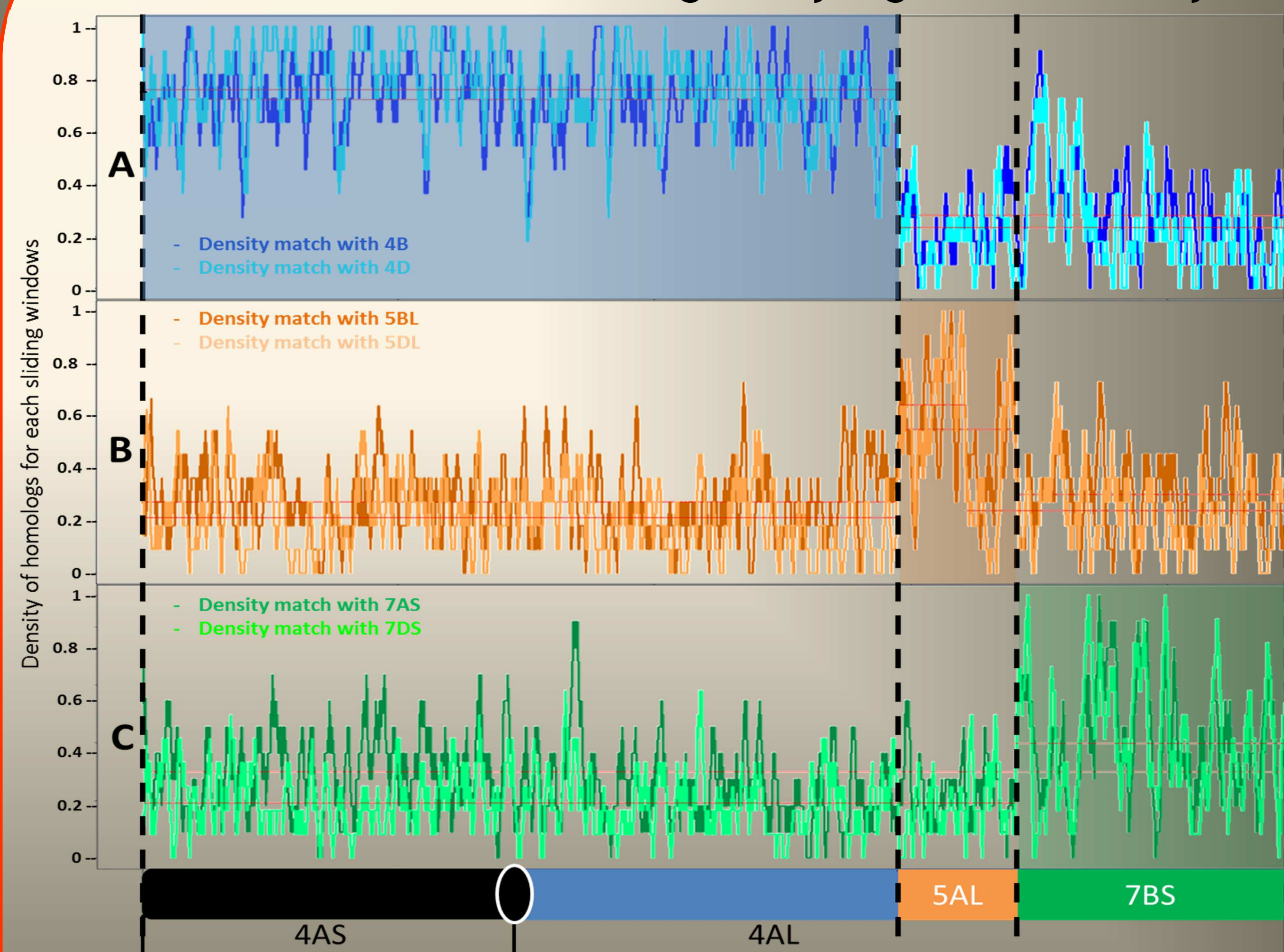
- A bread wheat - *T. militinae* introgressive line (Jakobson et al., 2006) confers resistance to powdery mildew. In order to clone the resistance gene on 4AL, ditelosomic (Dt) 4AL-TM line was developed.
- The availability of Dt4AL-TM line permitted flow sorting the 4AL-TM arm and its shotgun sequencing.



### In-silico identification of chromosome structural changes

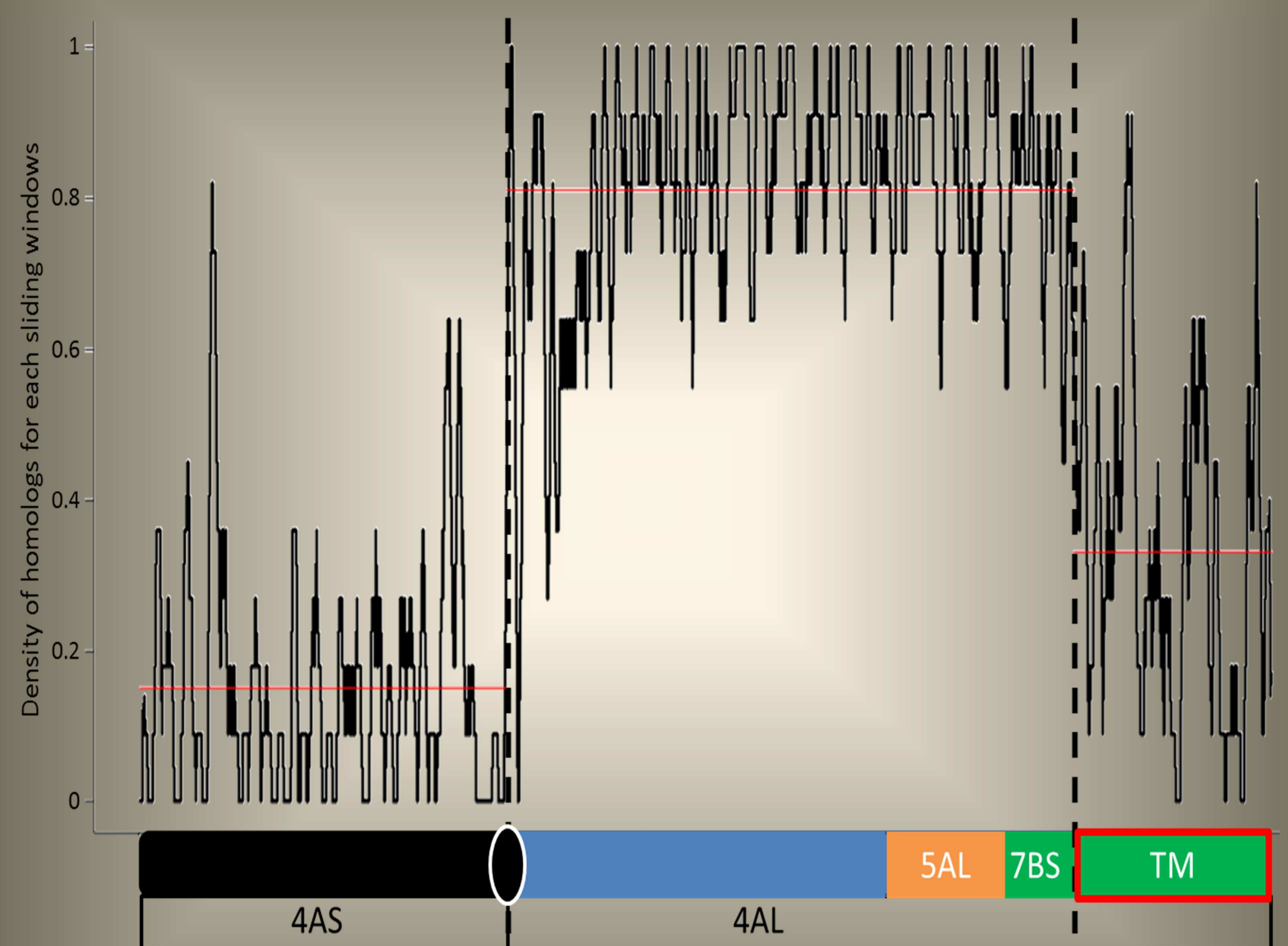


### Identification of translocated regions by segmentation analysis



Comparison of homeologous gene density within 4A-CS and chromosomes 4B and 4D (A), 5BL and 5DL (B), and 7AS and 7DS (C). Please note that homeologous region shows higher similarity - horizontal red lines. Surprisingly, 7BS translocated region shows significantly lower similarity with group 7 chromosomes compared to chromosomes of group 4 and 5.

### Identification of introgressed region by segmentation analysis



Comparison of homeologous gene density within 4A-CS and chromosome arm 4AL-TM. Introgressed region was delimited as the distal part of the 7BS translocated region, which is characterized by a significantly lower density of homeologous genes.

## CONCLUSIONS

- Coupling chromosome sorting and Next Generation Sequencing provides a powerful tool for structural genomics
- Decrease of density of homeologous genes along chromosome can detect translocated and/or introgressed regions even for two closely related species

## FUTURE GOALS

- Develop specific molecular markers to confirm the introgression from *T. militinae* to chromosome 4A of bread wheat
- Test the present method for *in silico* delineation of introgressions of different size and from different species

## REFERENCE

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## ACKNOWLEDGEMENTS

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