



New insights into genome assembly at the chromosome-level of *Prunus tomentosa* in evolution and cold tolerance

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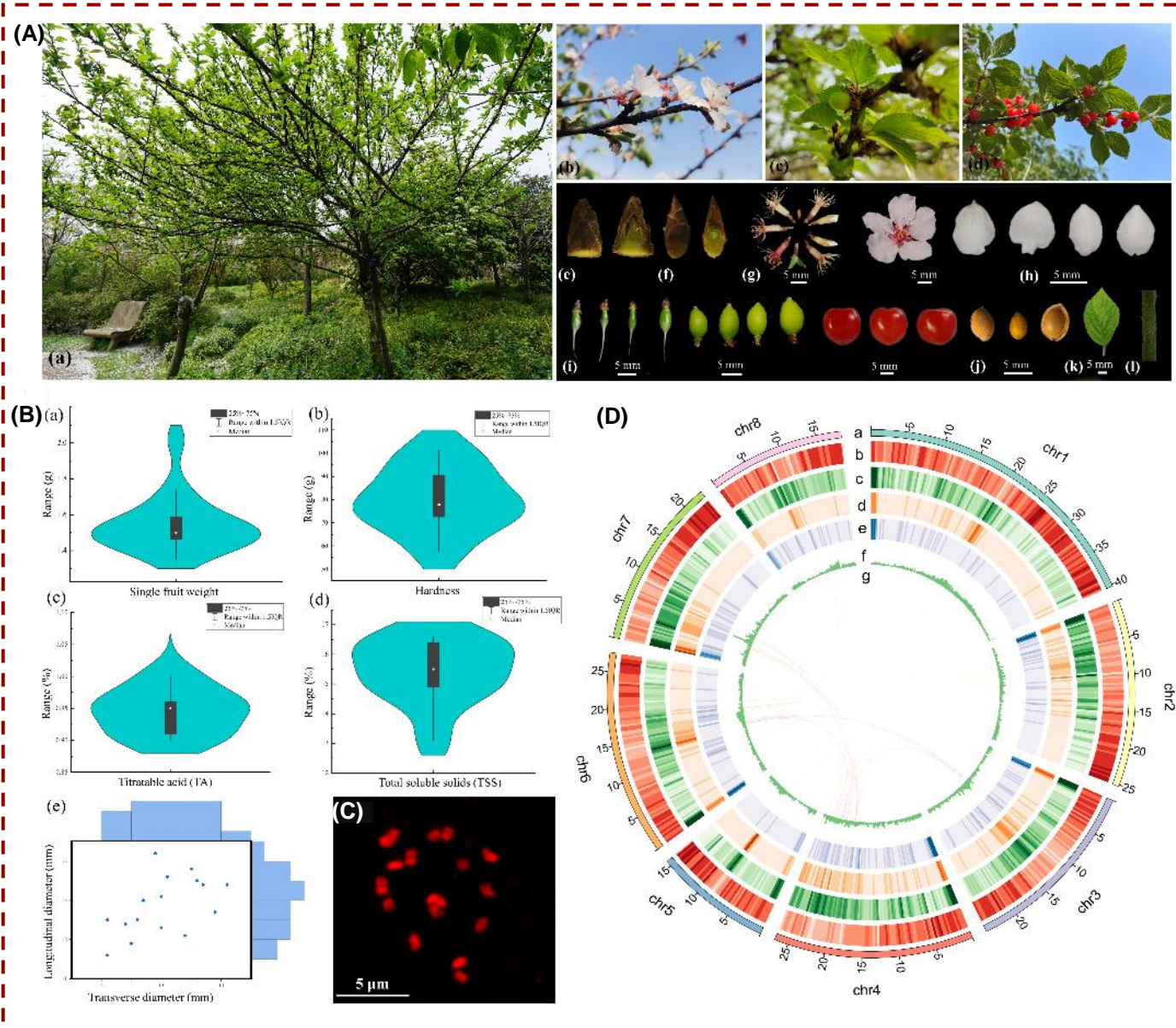
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Introduction



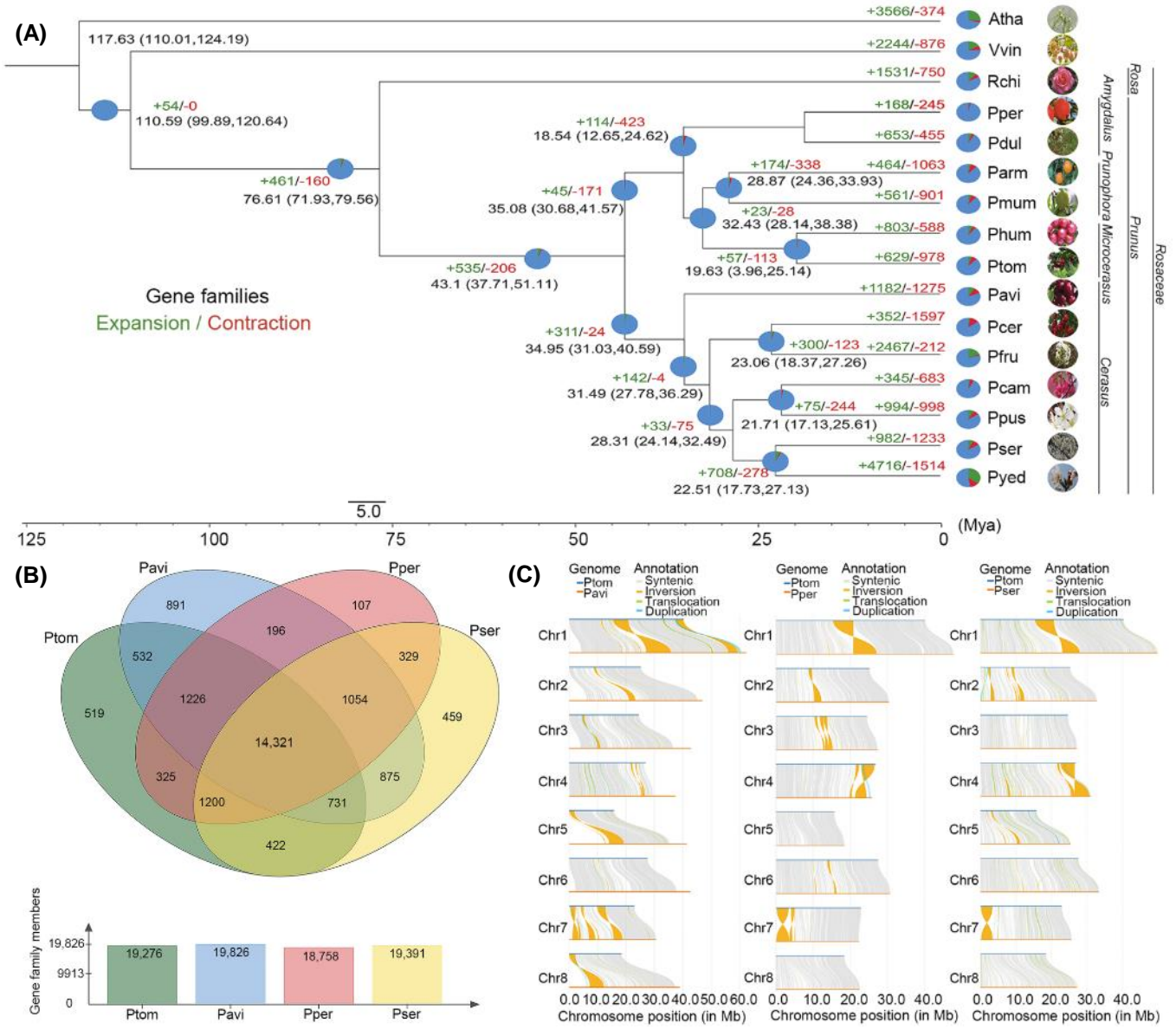
□ *Prunus tomentosa* has been a component of the Chinese diet for over 2000 years, noted for its rich content of vitamins and other antioxidant compounds. It has long been classified within the subgenus *Microcerasus*. However, studies on high-quality chromosome-scale genomes and phylogenetic analyses of *P. tomentosa* are lacking, rendering its genetic background relatively unknown.

□ *P. tomentosa* exhibits superior performance in cold tolerance, withstanding extreme temperatures down to -40°C , widely distributed in northern China and other frigid regions. However, the molecular regulation mechanisms underlying its cold tolerance remain unclear.

□ We assembled the high-quality reference genome covering 211.55 Mb, with scaffold N50 of 25.22 Mb and contig N50 of 3.40 Mb.



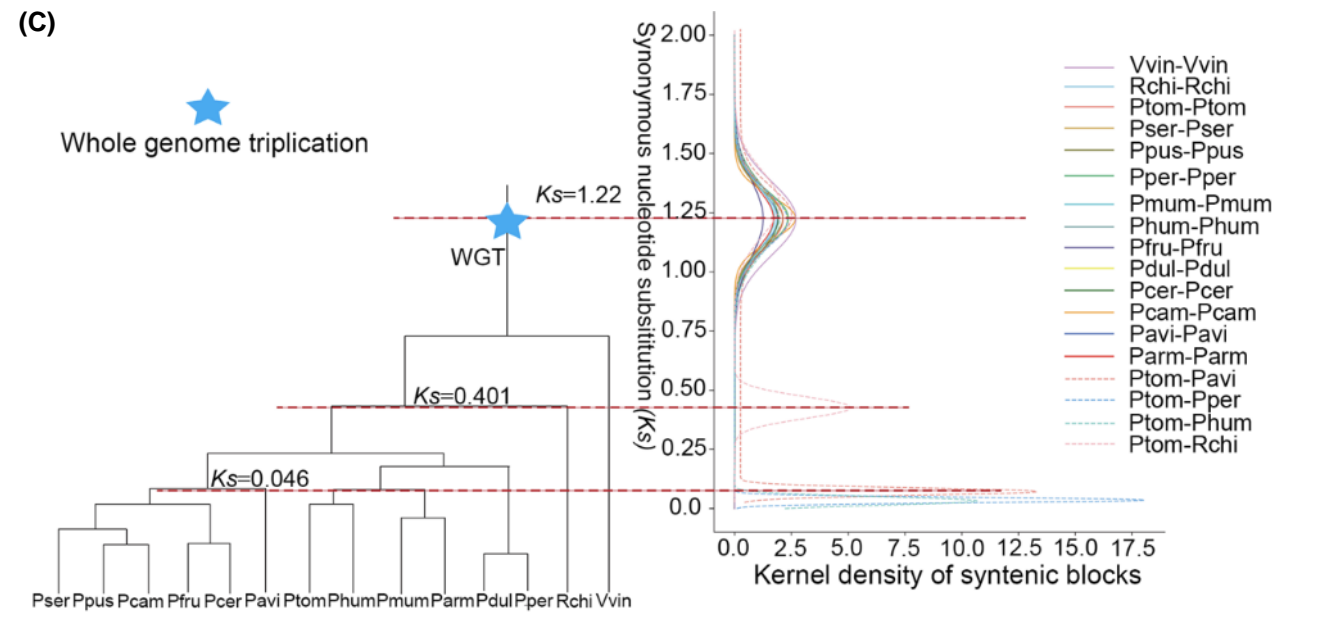
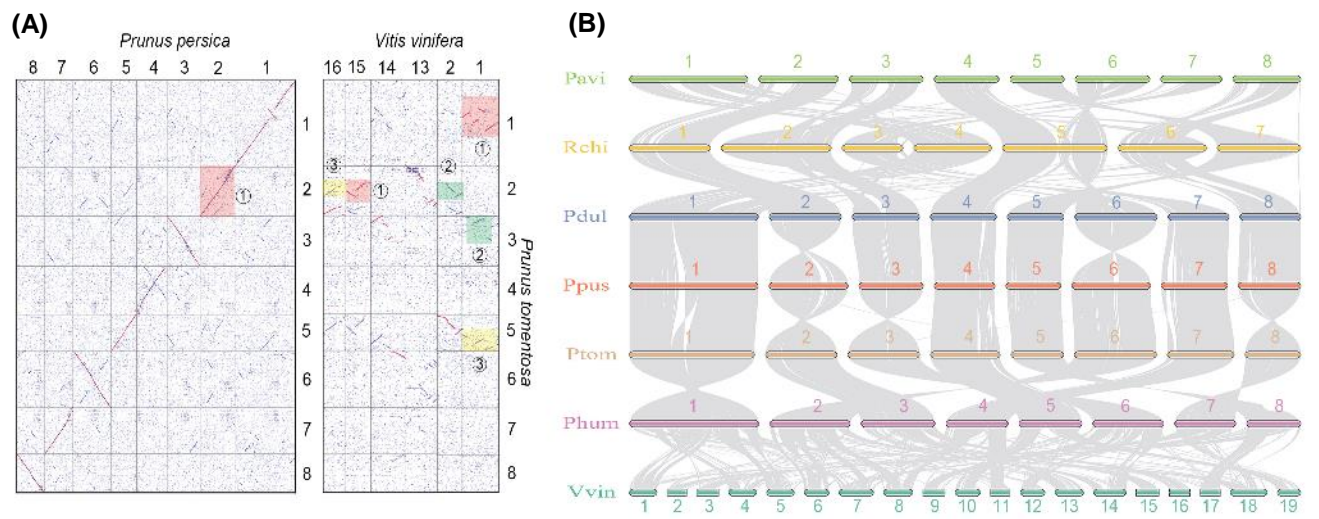
Results



- ❑ *P. tomentosa* is genetically closer to subgenus *Prunophora* rather than subgenus *Cerasus* (cherries), challenging previous taxonomy classification.
- ❑ *P. tomentosa* diverged from its common ancestor with *P. humilis* approximately 19.63 Mya; their common ancestor and two *Prunophora* species (*P. mume* and *P. armeniaca*) diverged at 32.43 Mya.
- ❑ *P. tomentosa* shares 14,321 gene families with other *Prunus* species but contains 519 unique gene families, more than *P. serrulata* (459) and *P. persica* (107).
- ❑ *P. tomentosa* exhibited 140.71 Mb of syntenic blocks with *P. avium*, 129.14 Mb with *P. persica*, and 123.49 Mb with *P. serrulata*, indicating their strong evolutionary conservation.



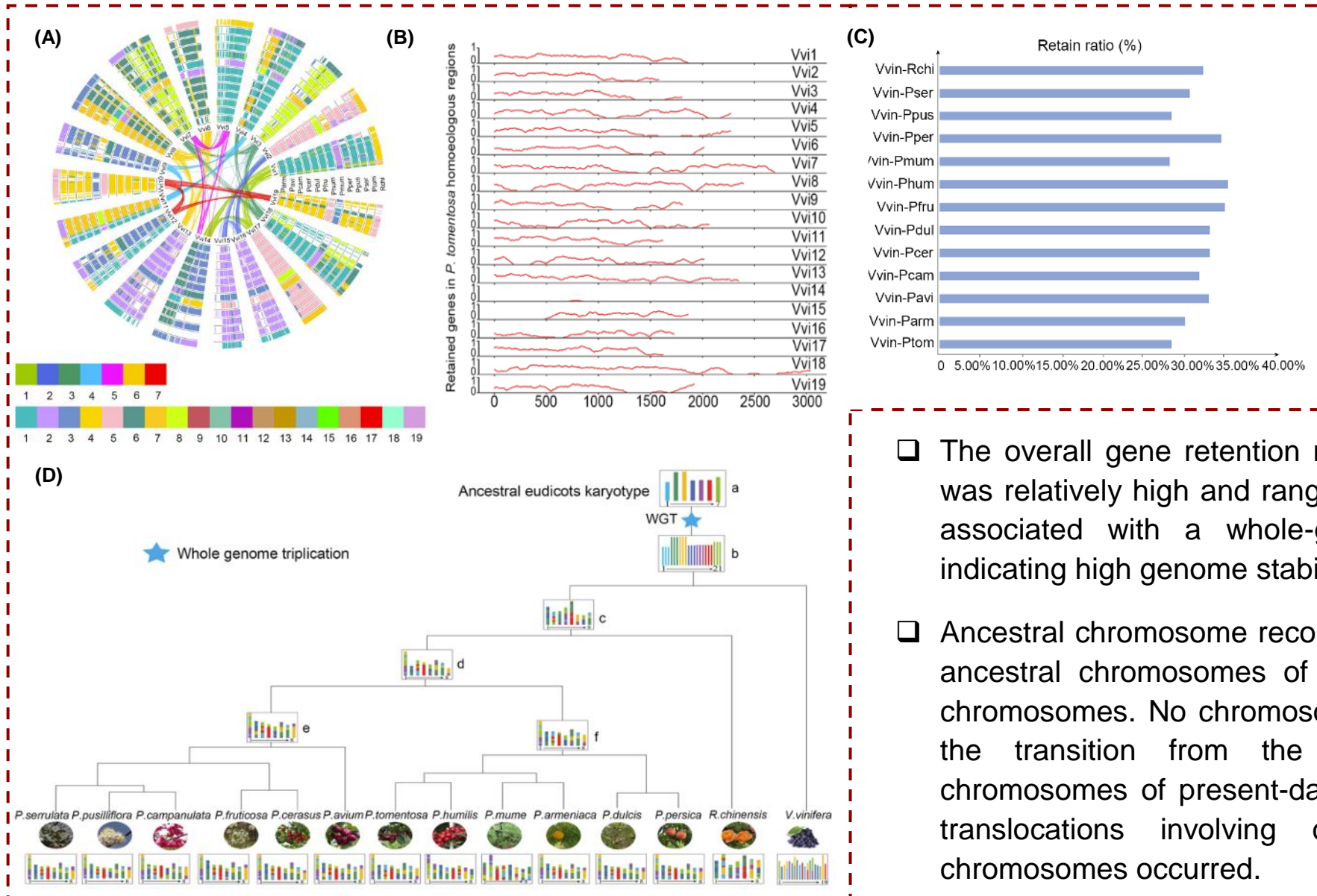
Results



- ❑ *P. tomentosa* underwent a single whole-genome triplication (γ event) using homologous dot plots between *V. vinifera*/*P. persica* and *P. tomentosa*.
- ❑ The presence of extensive syntenic blocks across multiple *Prunus* species suggested strong genomic conservation.
- ❑ K_s analysis indicated that γ event occurred 115–130 million years ago, with a peak at K_s value of 1.22, confirming its evolutionary significance within the *Prunus* lineage.



Results



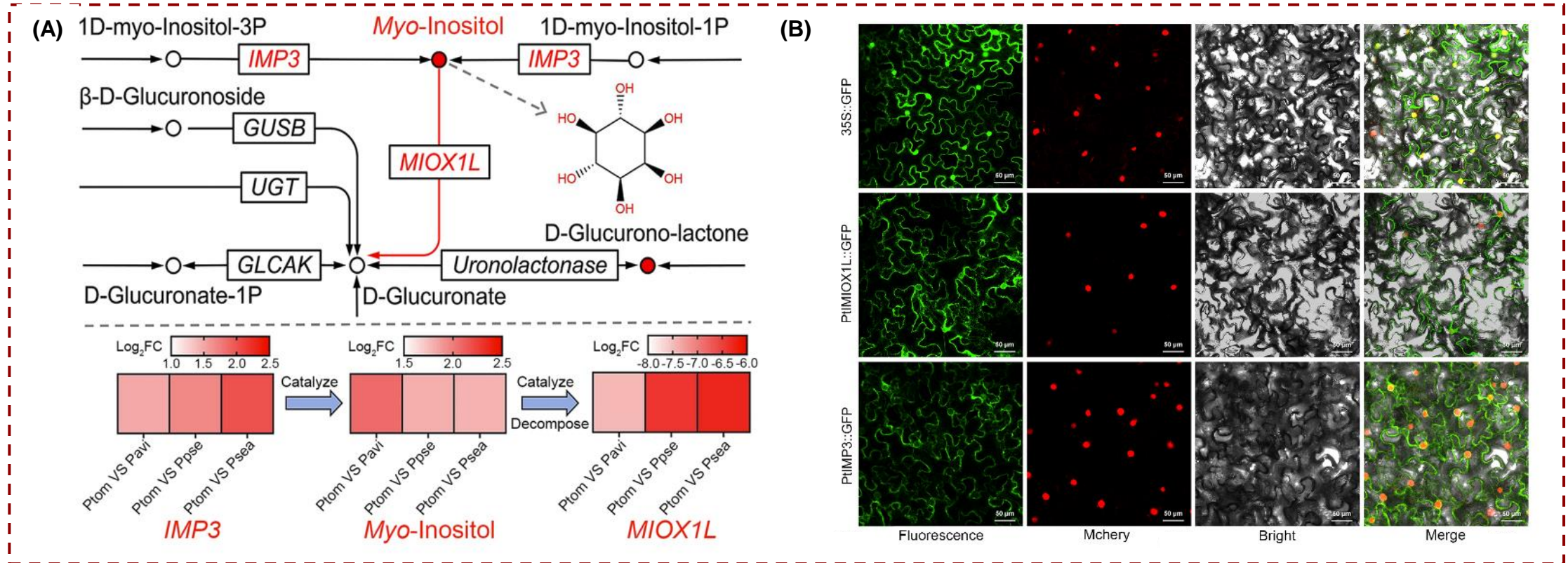
Analysis of collinear regions between grape and *Prunus* species revealed a 1:1 ratio, indicating the occurrence of only ancient gamma (γ) events.

The overall gene retention rate for the *Prunus* species was relatively high and ranged from 28% to 36%, likely associated with a whole-genome triplication event, indicating high genome stability.

Ancestral chromosome reconstruction suggests that the ancestral chromosomes of *Prunus* consisted of eight chromosomes. No chromosome fusion occurred during the transition from the ancestral state to the chromosomes of present-day *Prunus* species. Instead, translocations involving cross-exchanges between chromosomes occurred.



Results

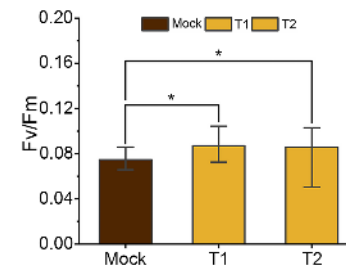
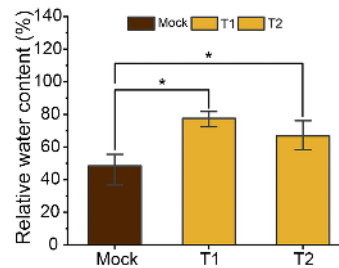
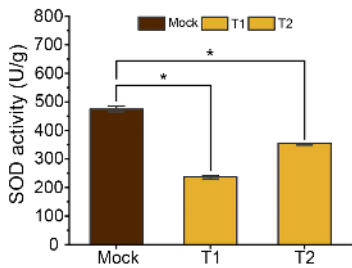
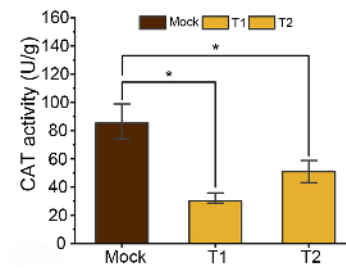
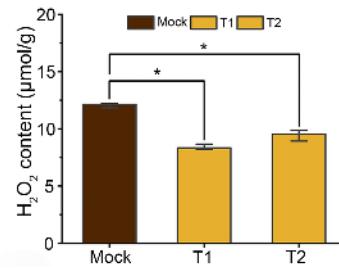
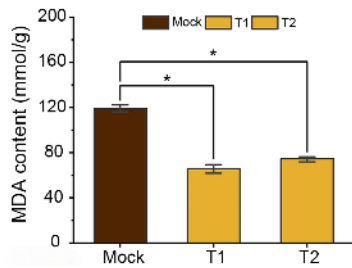
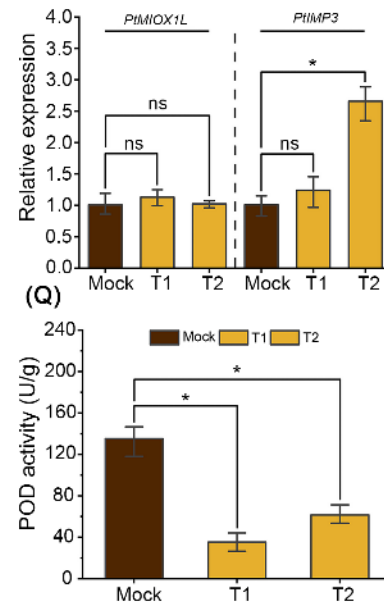
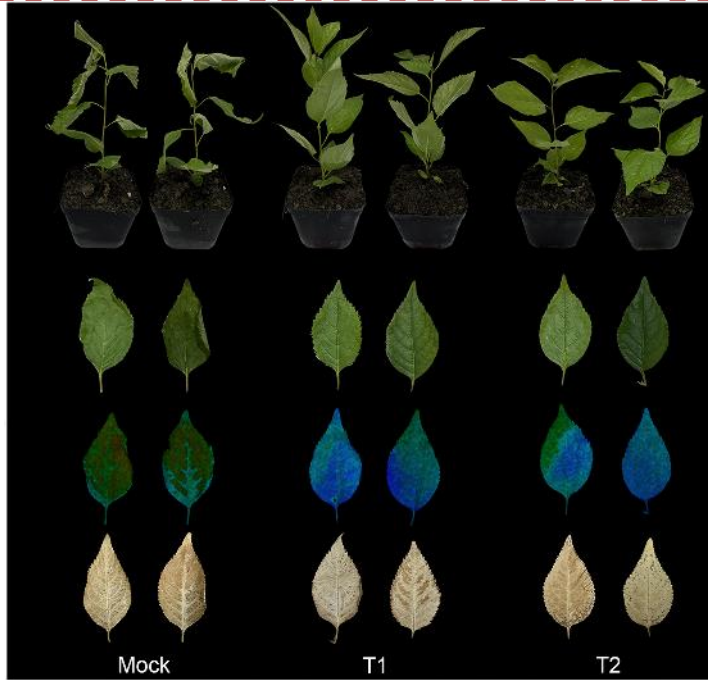


- ❑ Multi-omics integration revealed *PtIMP3* and *PtMIOX1L* as key factors in cold tolerance of *P. tomentosa*.
- ❑ The high expression abundance of *PtIMP3* and low expression abundance of *PtMIOX1L* resulted in high endogenous inositol levels in *P. tomentosa*, potentially conferring superior cold tolerance.
- ❑ The subcellular localization showed that *PtIMP3* and *PtMIOX1L* were expressed mainly in the cytoplasm.



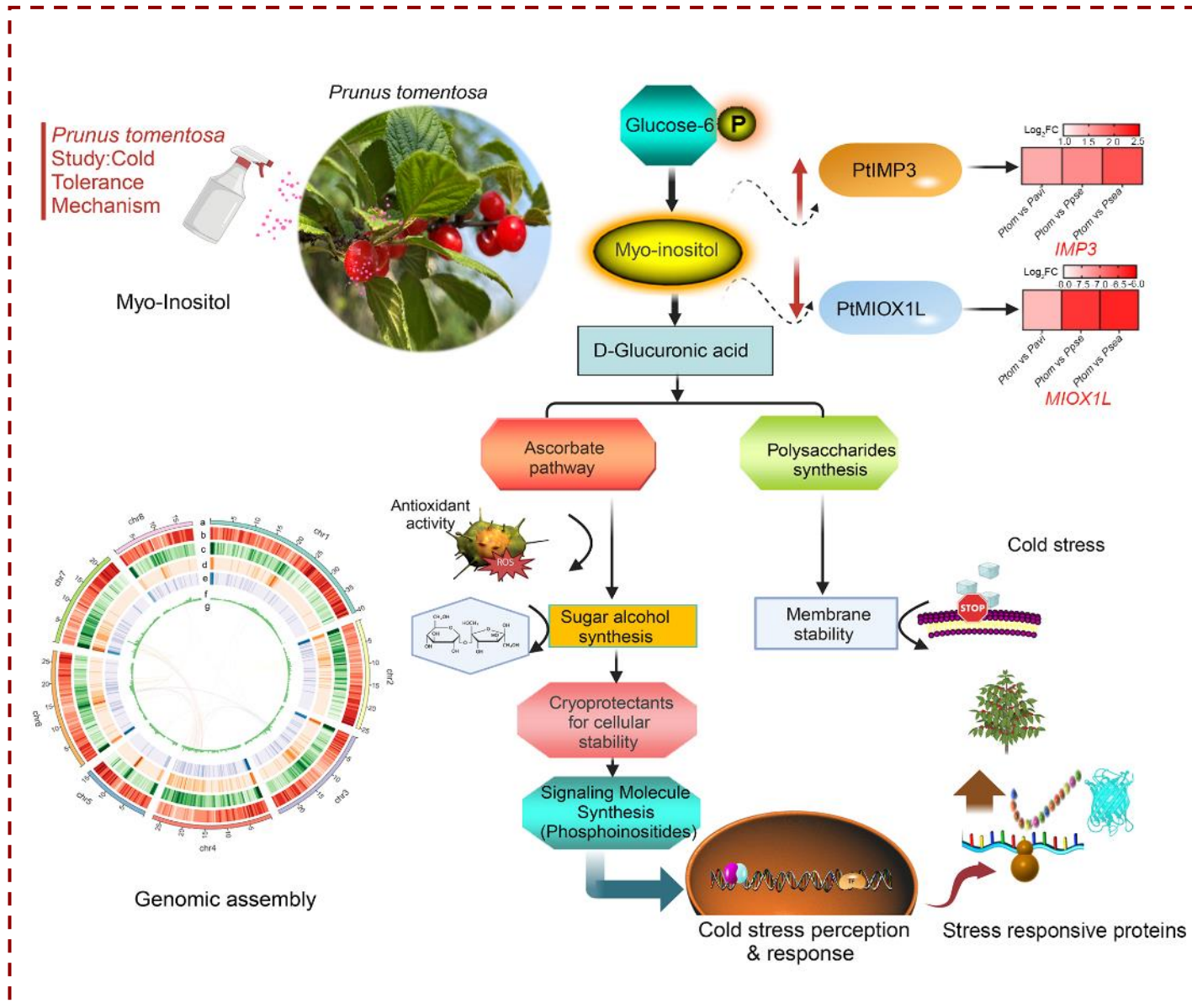
Results

- ❑ Exogenous *myo*-inositol enhances cold resistance in cherry rootstocks.
- ❑ The cherry rootstocks with spraying of *myo*-inositol showed lower POD, MDA, CAT, SOD activities, and H₂O₂ levels, and higher RWC and photosynthetic efficiency.
- ❑ These results suggest *myo*-inositol acts as a protective solute, mitigating ROS damage and stabilizing cellular water balance under cold stress.

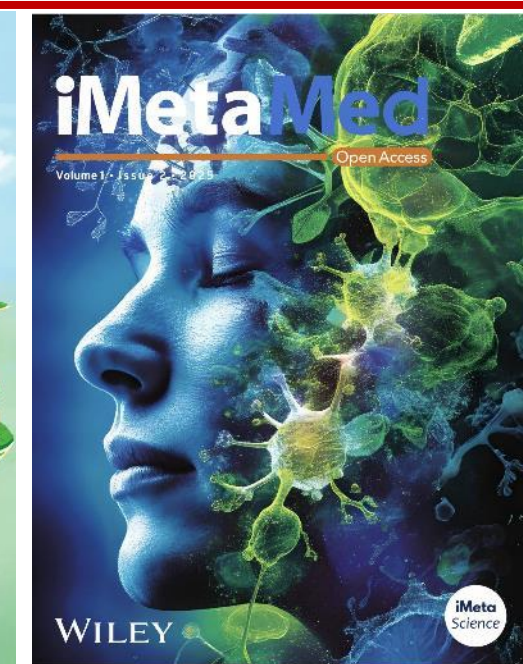
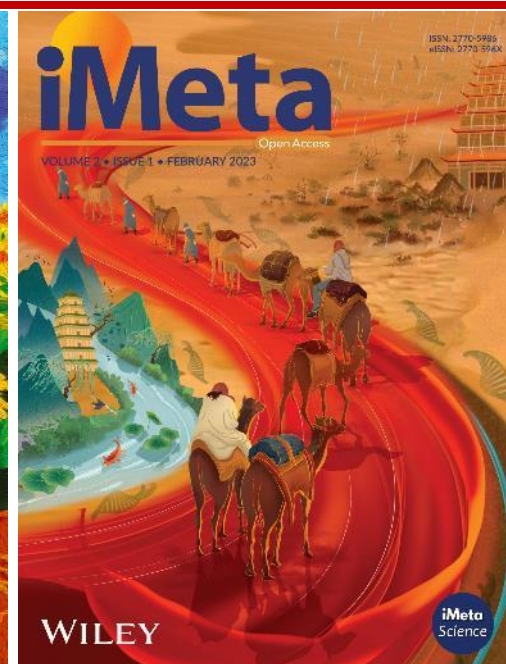




Summary



- ❑ In this study, we assembled a high-quality chromosome-level genome of *P. tomentosa* and clarified its genetic diversity, structural variation, phylogenetic relationships with other genus *Prunus* species.
- ❑ We identified *PtIMP3* and *PtMIOX1L* as key factors in cold tolerance; they help maintain higher inositol contents in *P. tomentosa*, potentially conferring its superior cold tolerance.
- ❑ The application of *myo*-inositol enhanced the cold tolerance of cherry rootstocks by modulating reactive oxygen species (ROS) concentrations and maintaining a stable relative water content (RWC).



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