



# T2T genome, pan-genome analysis, and heat stress response genes in *Rhododendron* species

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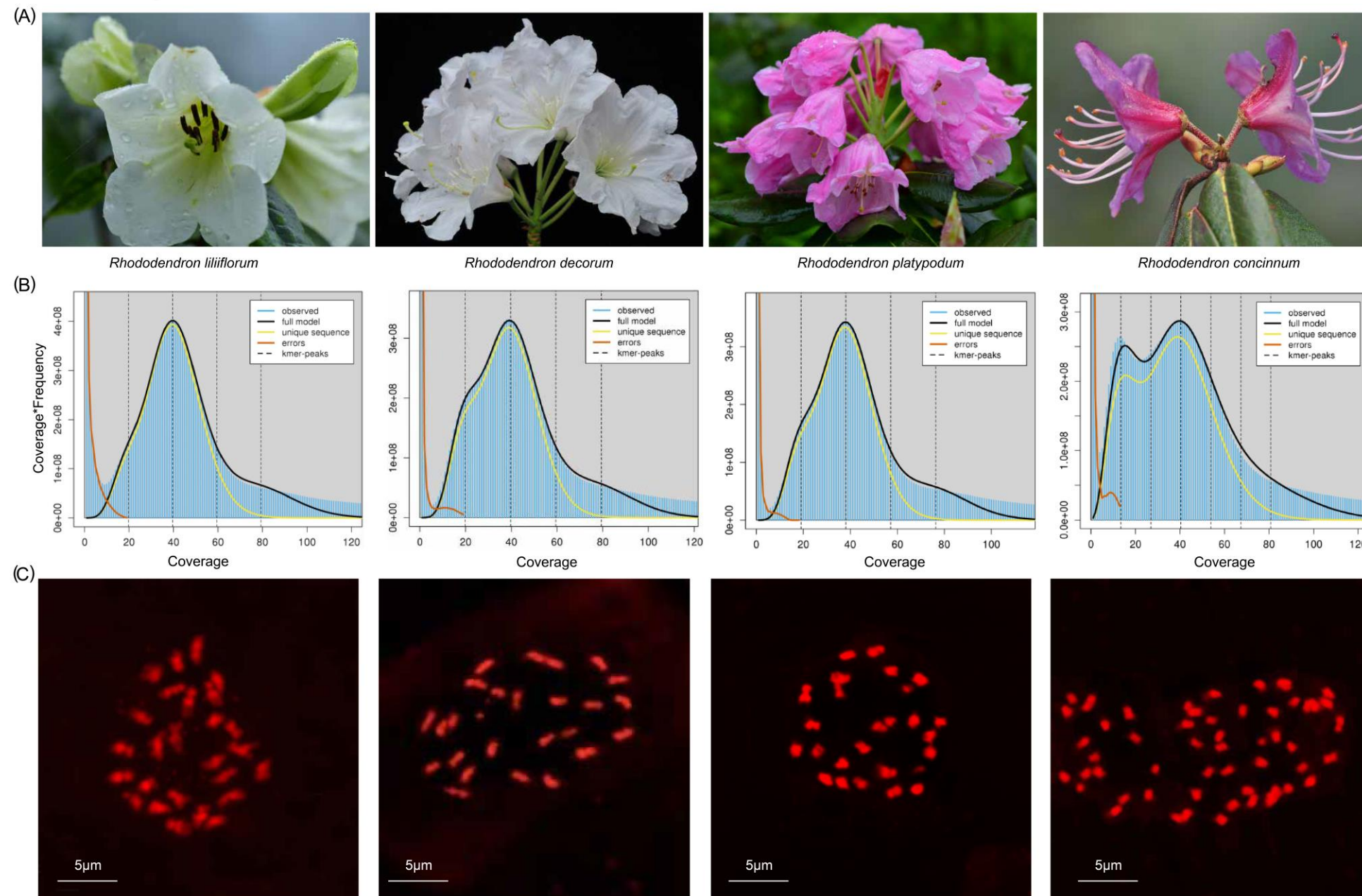
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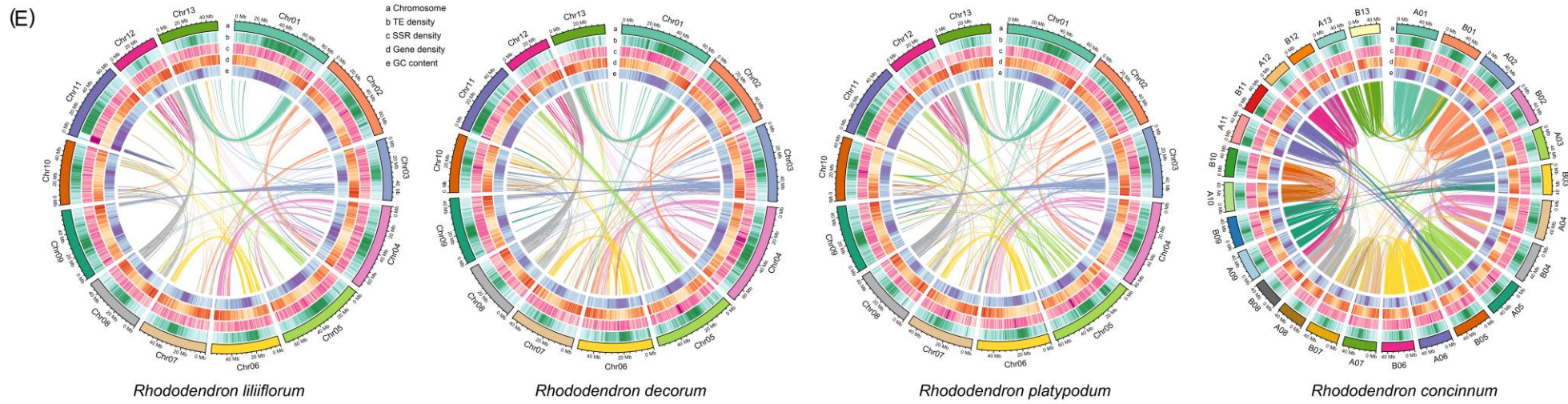
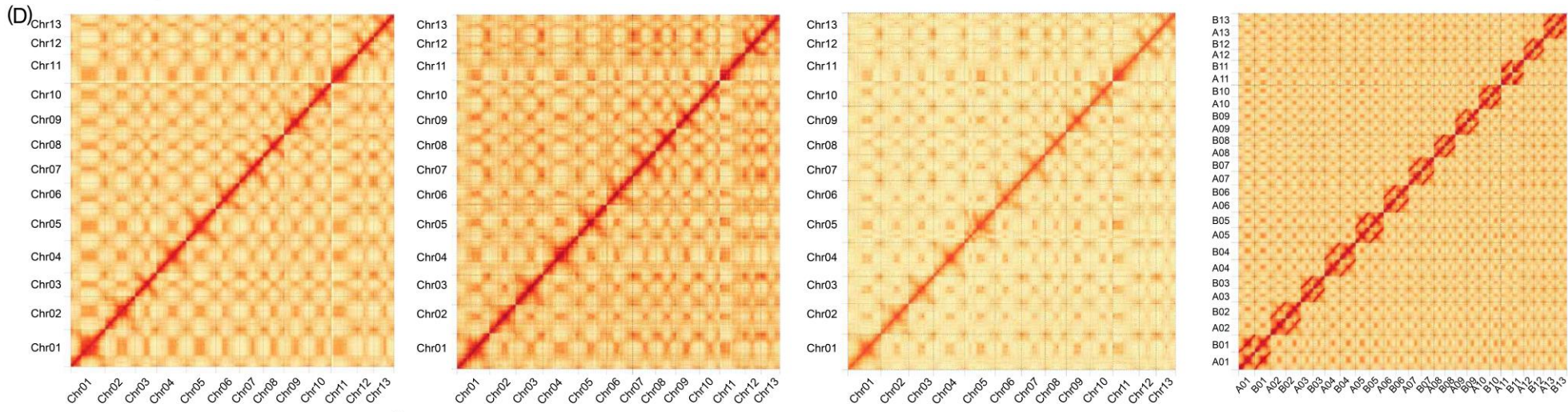
# Genome sequencing and assembly of Rhododendron

- Here, we perform the de novo genome sequencing of four Rhododendron plants by PacBio HiFi, Oxford Nanopore Technology (ONT), Illumina, and Hi-C technology





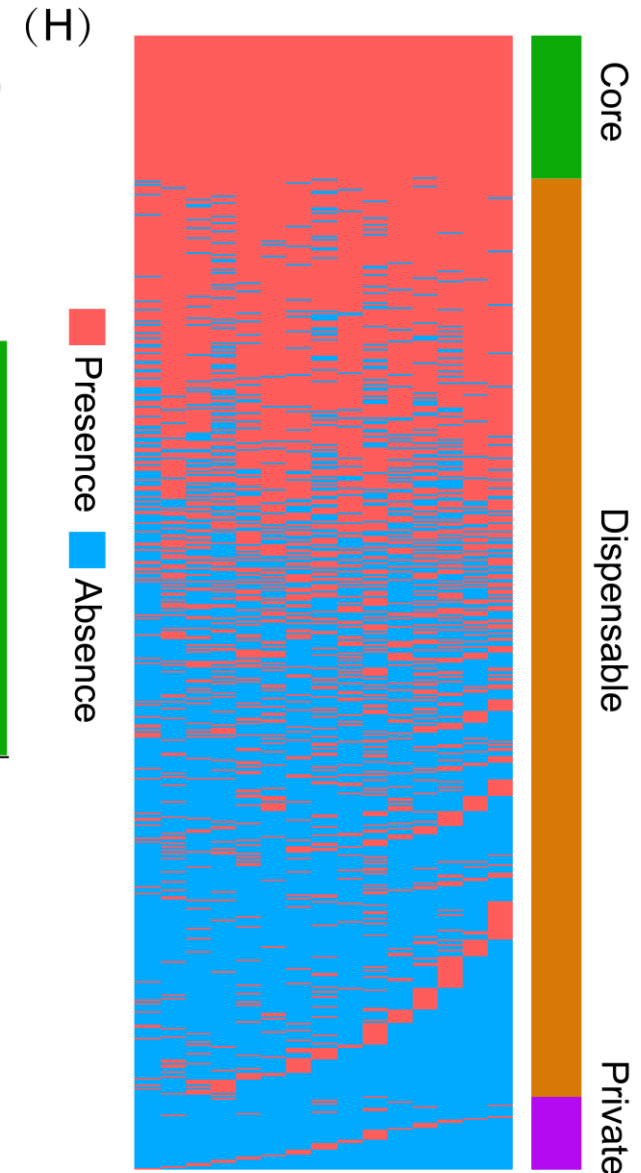
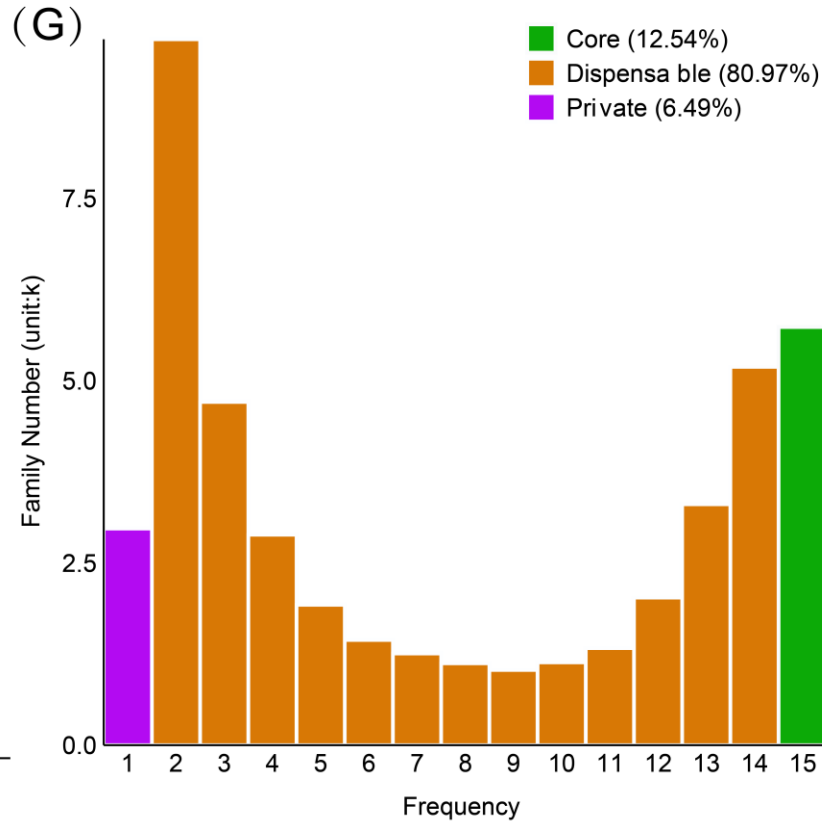
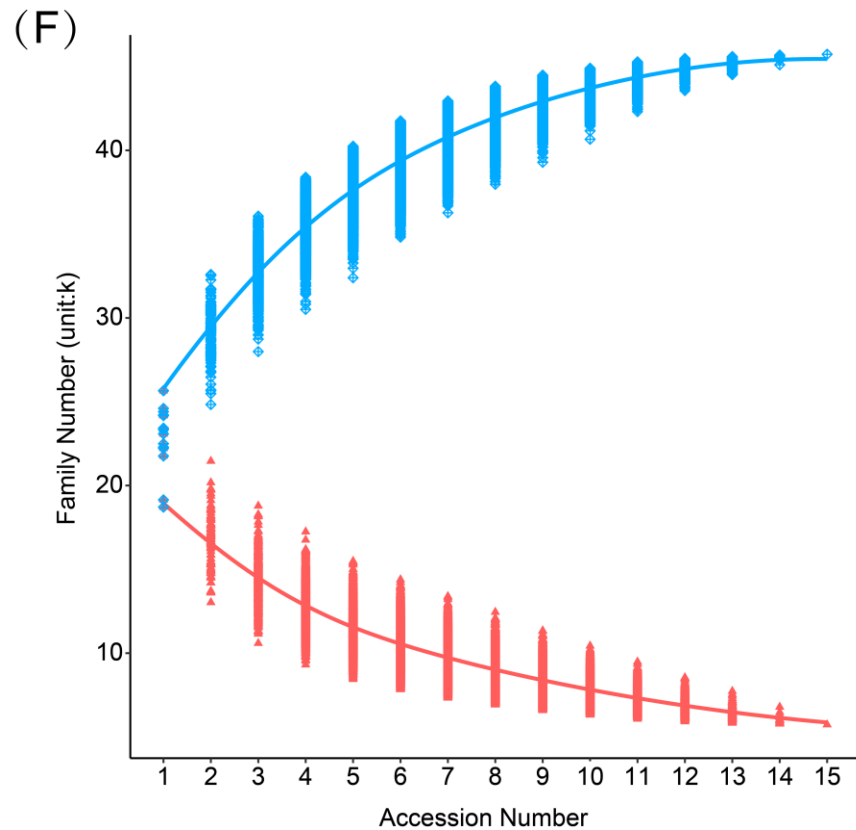
# Genome sequencing and assembly of Rhododendron



- The chromosomal anchored ratio was over 97.90% among four species by Hi-C
- Repetitive sequences accounted for over 49.10 % of four genomes, and most repetitive sequences were long-terminal repeats (LTRs)



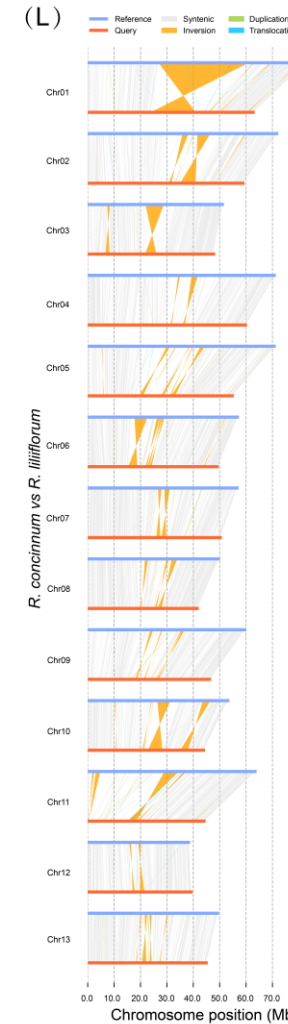
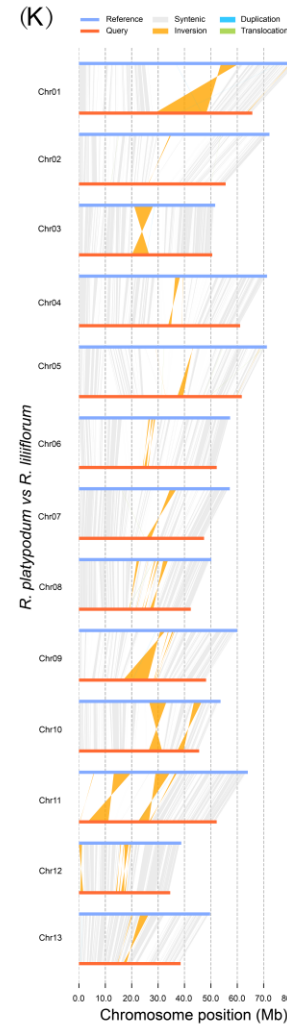
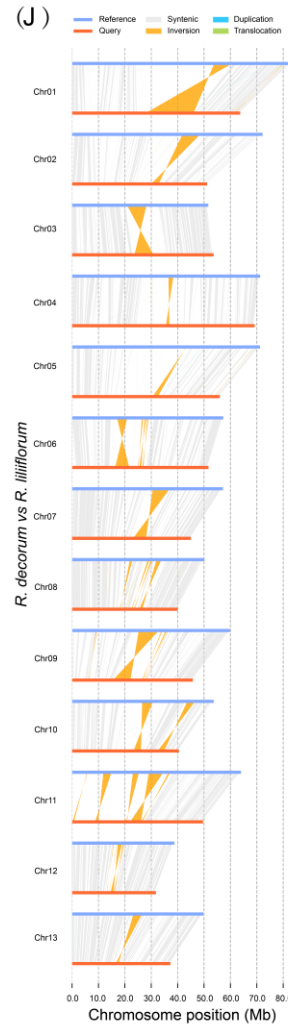
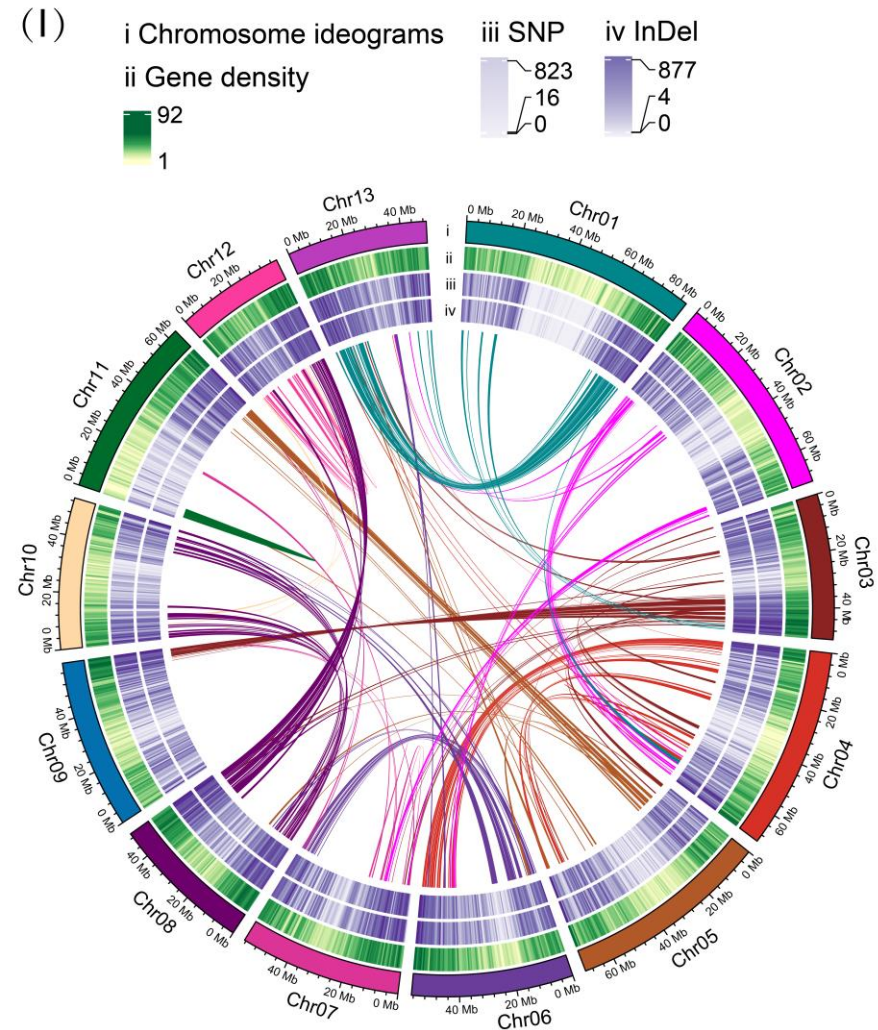
# Pan-genome analysis of Rhododendron



- Trend chart of core (red) and non-core (blue) gene family quantities
- The number of gene families across 15 species is 45,731, including 5734 core gene families, 37,027 dispensable gene families, and 2970 private gene families
- we constructed a distribution map of presence and absence of gene families based on clustering analysis



# Variant analysis of 15 *Rhododendron* genomes

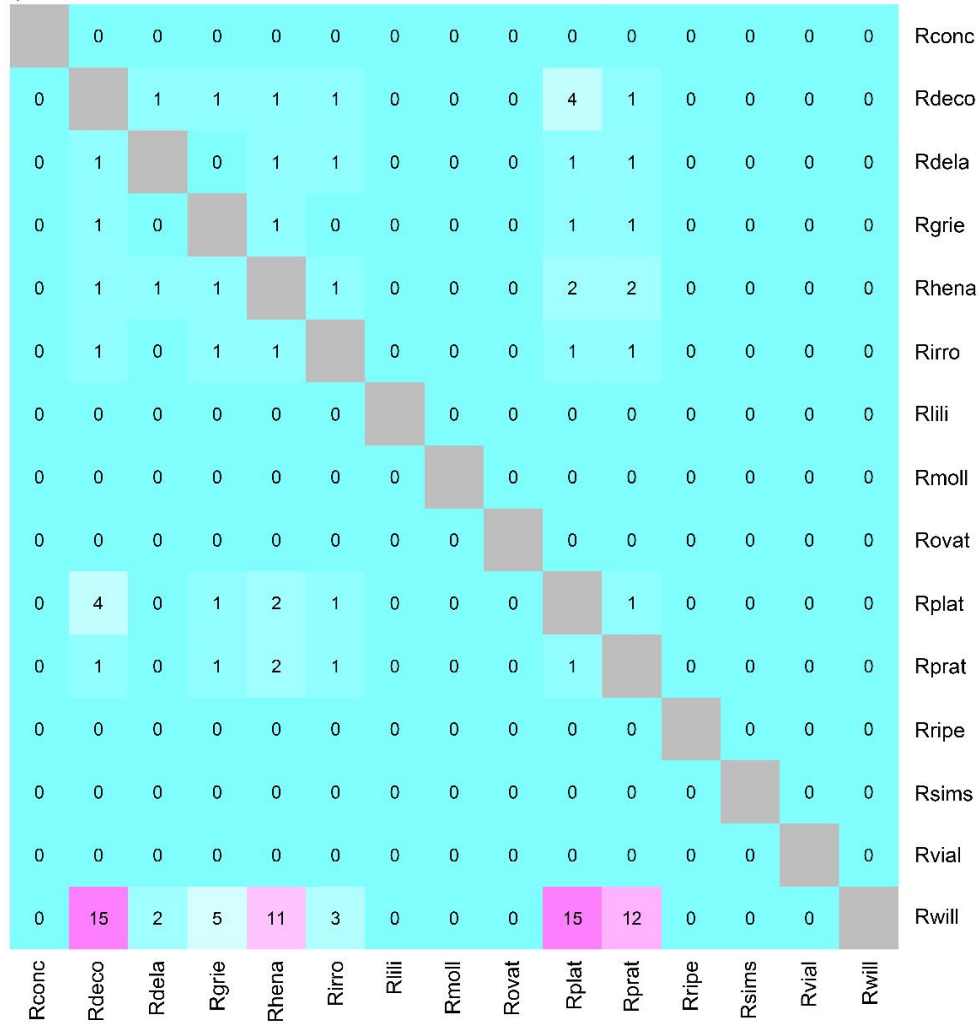


- We perform a comprehensive identification of variations such as single nucleotide polymorphisms (SNPs), insertions and deletions (InDels), and structural variations (SVs) in *Rhododendron* based on pan-genome analysis using T2T genome(*Rhododendron liliflorum*) as reference

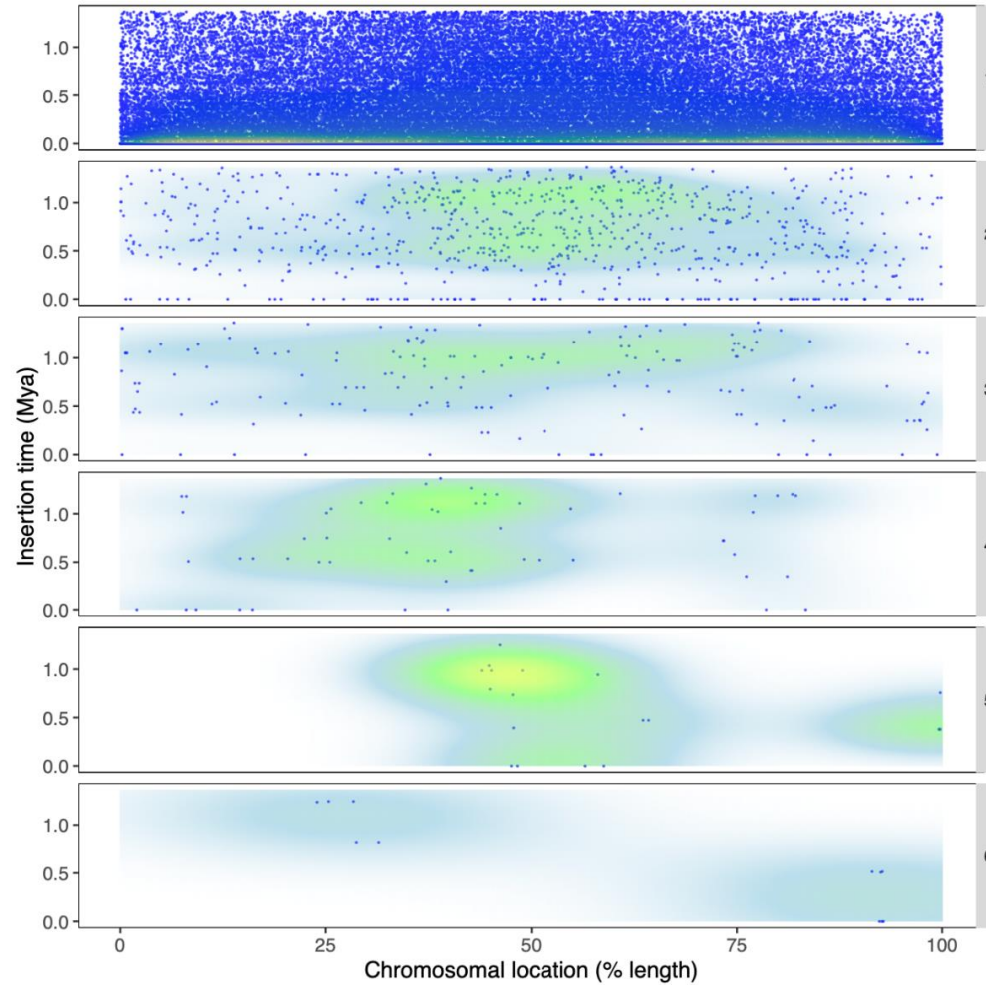


# LTR analysis of 15 Rhododendron genomes

(M)



(N)

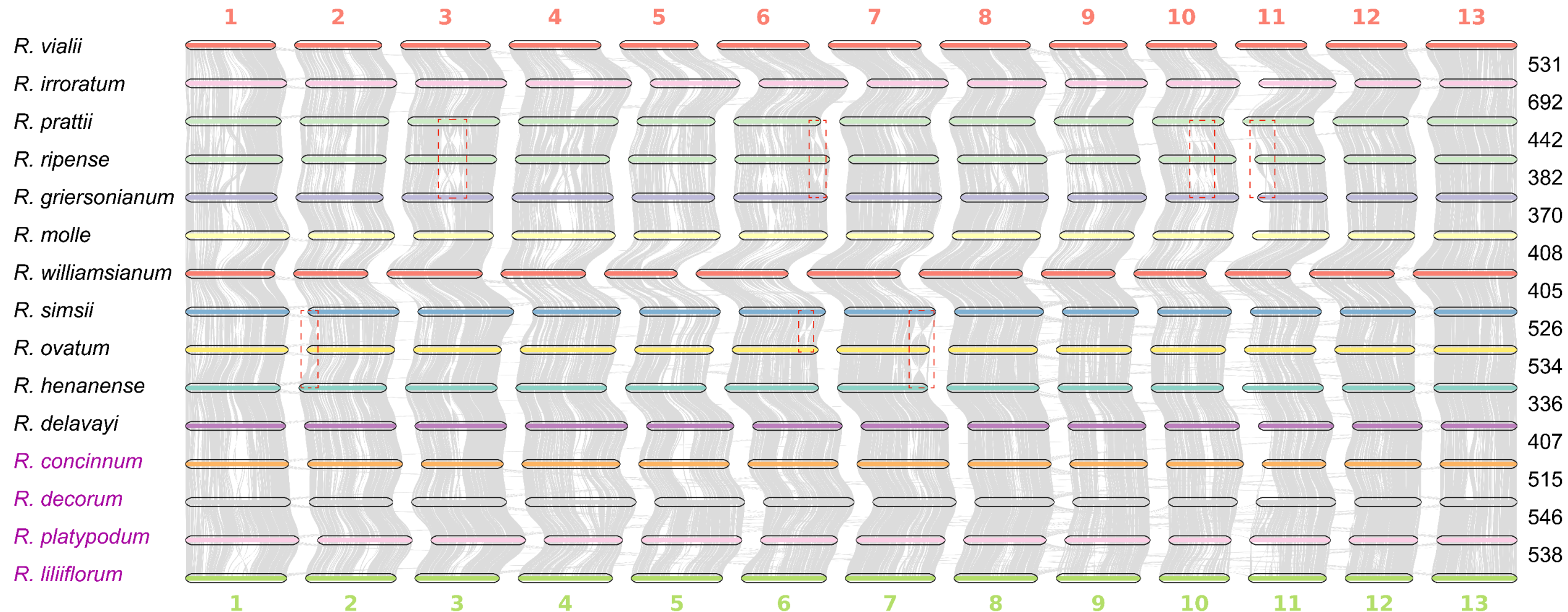


- We performed clustering on LTRs of 15 species to obtain the shared LTRs within each cluster
- Furthermore, the distribution density of LTRs in the middle of chromosomes was greater than two ends

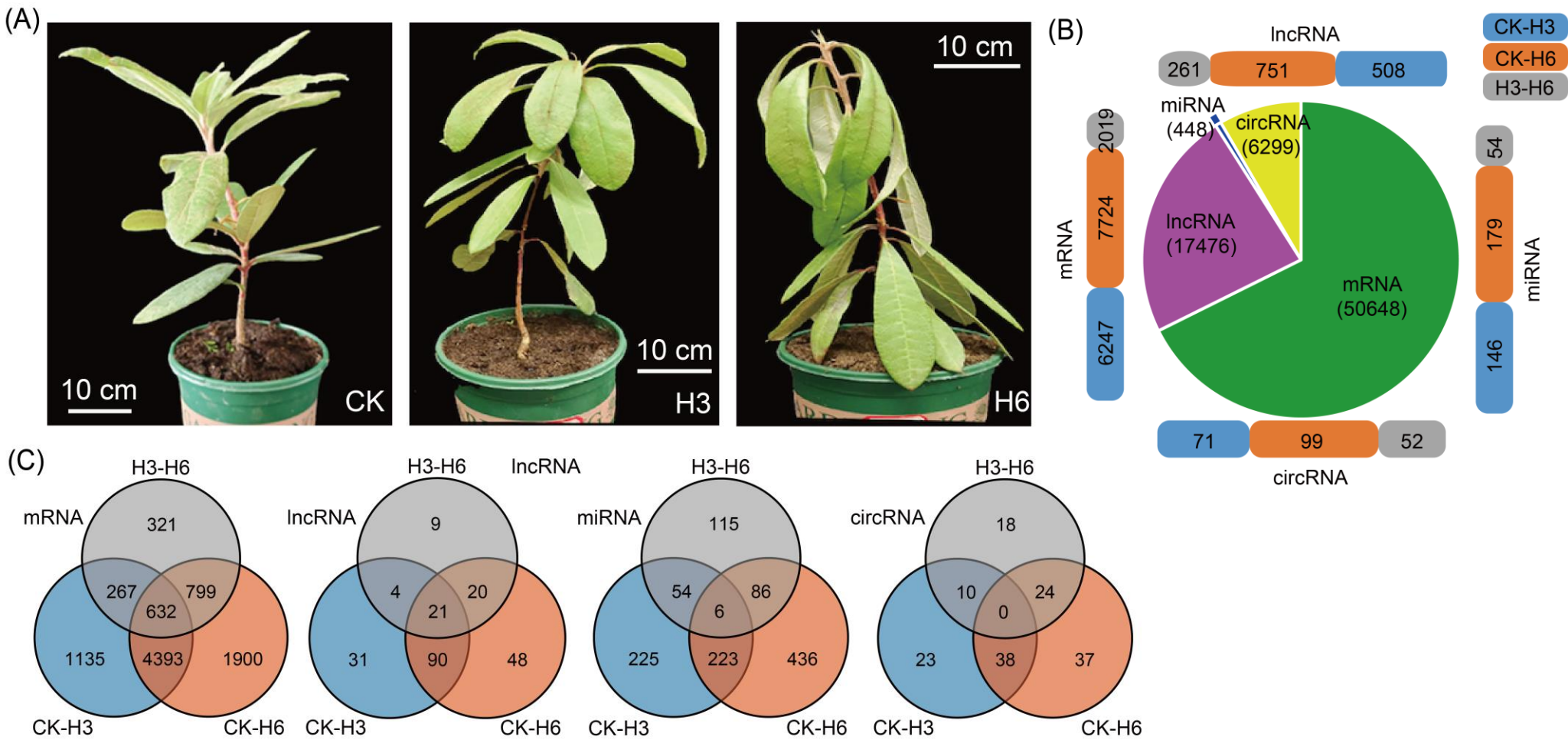


# Collinearity analysis of 15 *Rhododendron* genomes

(O)



# Whole transcriptome sequencing and detection of heat responsive genes

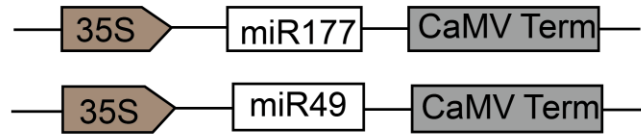


- To explore heat-resistant genes and regulatory mechanisms of *Rhododendrons*, we conducted whole transcriptome sequencing under heat treatment of CK, heat treatment of 3 days (H3) and 6 days (H6)

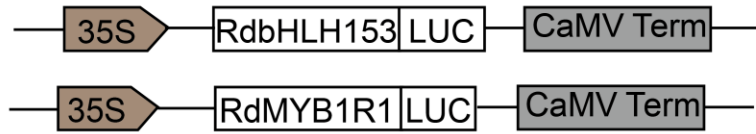


# Functional validation of candidate genes

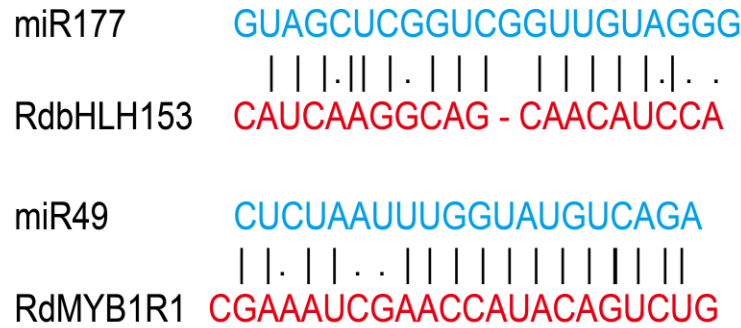
(D) Effector



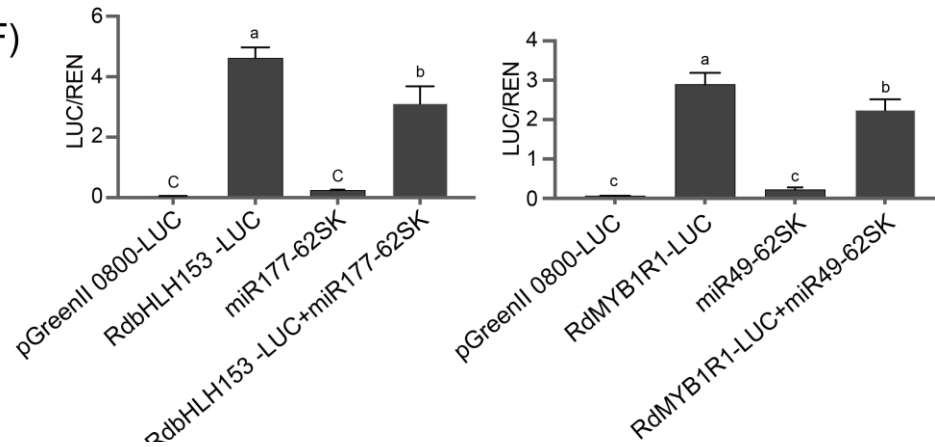
Reporter



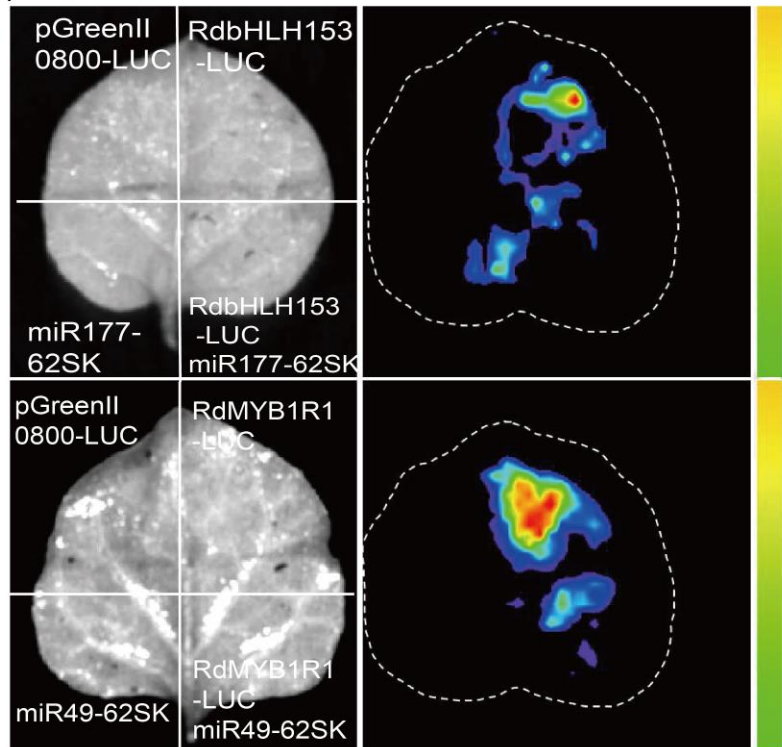
(E)



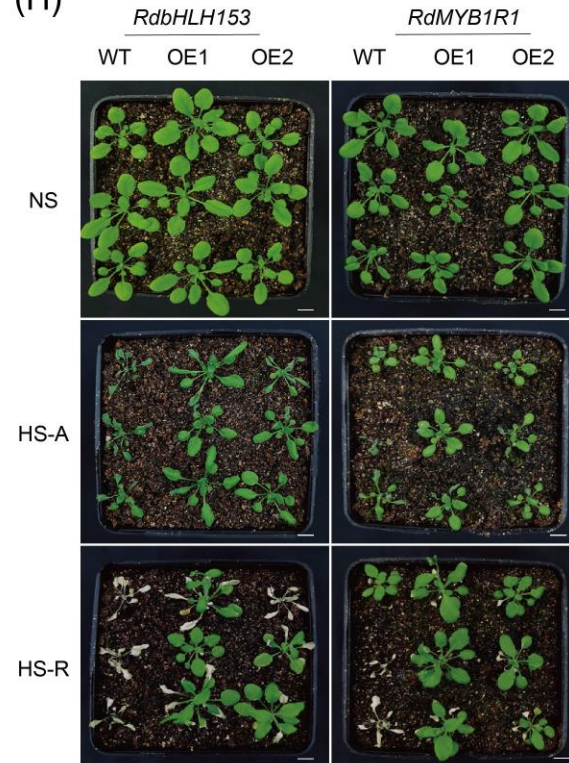
(F)



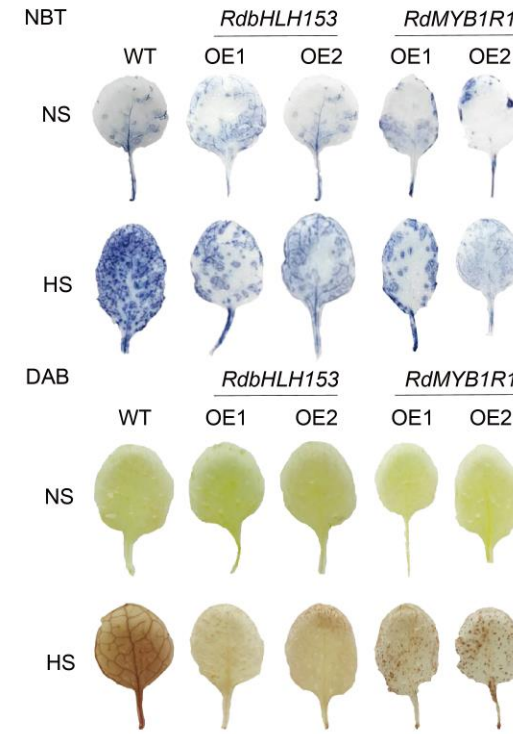
(G)



(H)



(I)



● We selected two representative pairs of miRNAs and related target genes for functional validation



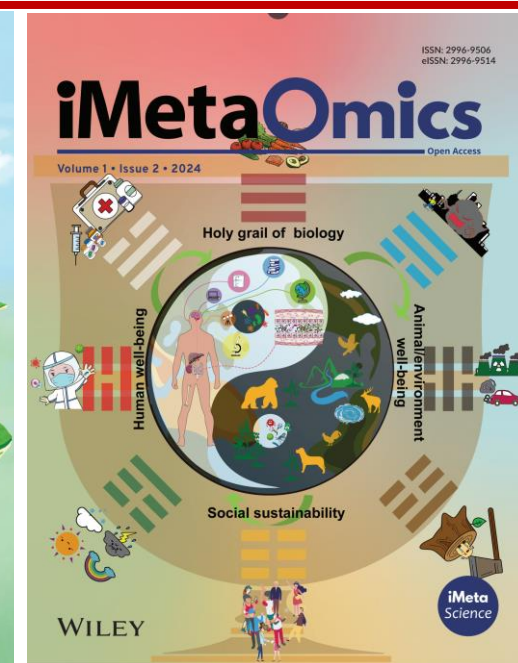
# Summarize

## T2T genome, pan-genome analysis, and heat stress response genes in *Rhododendron* species

- This study reports for the first time the high-quality telomere to telomere (T2T) genome of *Rhododendron* lilies with 13 chromosomes
- Based on the genomes of 15 *Rhododendron* plants, a pan genomic analysis was conducted on *Rhododendron* plants
- By combining genome sequencing and whole transcriptome sequencing, several key genes and miRNAs related to heat stress were identified, providing abundant resources for comparative genomics and functional genomics research of *Rhododendron* plants


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