nature portfolio

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Reporting Summary

Nature Portfolio wishes to improve the reproducibility of the work that we publish. This form provides structure for consistency and transparency in reporting. For further information on Nature Portfolio policies, see our <u>Editorial Policies</u> and the <u>Editorial Policy Checklist</u>.

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For	all statistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.
n/a	Confirmed
	\square The exact sample size (n) for each experimental group/condition, given as a discrete number and unit of measurement
	🔀 A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
	The statistical test(s) used AND whether they are one- or two-sided Only common tests should be described solely by name; describe more complex techniques in the Methods section.
\times	A description of all covariates tested
\boxtimes	A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
	A full description of the statistical parameters including central tendency (e.g. means) or other basic estimates (e.g. regression coefficient) AND variation (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals)
	For null hypothesis testing, the test statistic (e.g. <i>F</i> , <i>t</i> , <i>r</i>) with confidence intervals, effect sizes, degrees of freedom and <i>P</i> value noted <i>Give P values as exact values whenever suitable.</i>
\boxtimes	For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
\boxtimes	For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
\boxtimes	Estimates of effect sizes (e.g. Cohen's <i>d</i> , Pearson's <i>r</i>), indicating how they were calculated
	Our way collection an statistics for highering articles on many of the points above

Software and code

Policy information about availability of computer code

Data collection

Long and short reads were generated on ONT, PacBio Sequel II and Illumina platform.

Data analysis

Nextdenovo v2.2.0, NextPolish v1.0, Minimap2 v2.17, BWA v0.7.17,LACHESIS, Platanus v1.2.4, Hifiasm v0.14, RepeatMasker v.4.0.7, RepeatModeler v.1.0.11, Augustus v3.2.3, TBLASTN v2.6.0, GENEWISE v2.4.1, PASA v2.3.3, EvidenceModeler v1.1.1, BUSCO v3.0, Maxent 3.4.3, ArcGIS 10.8, OrthoFinder v2.3.11, RAxML v8.2.11, CAFE v5.0, WGDI, BLASTP v2.7.1, ASTRAL v.5.6.2, PAML v4.9e, LTRharvest, LTRdigest, MUSCLE v3.8.31, HISAT2 v2.1.0, StringTie v1.3.3b, R v4.0.3, Bowtie2 v2.4.1, Bismark v0.22.3, MAFFT v7.313, AVID v2.1, VISTA (https://genome.lbl.gov/vista/mvista/submit.shtml)

For manuscripts utilizing custom algorithms or software that are central to the research but not yet described in published literature, software must be made available to editors and reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Portfolio guidelines for submitting code & software for further information.

Data

Policy information about availability of data

All manuscripts must include a data availability statement. This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A description of any restrictions on data availability
- For clinical datasets or third party data, please ensure that the statement adheres to our policy

All raw sequence data, genome assembly and annotation information have been deposited in the National Genome Data Center (NGDC; https://bigd.big.ac.cn/bioproject) under BioProject accession number PRJCA022976.

Research involving human participants, their data, or biological material

Policy information about studies with humar	<u>n participants or human</u>	<u>ı data</u> . See also policy	information a	bout <u>sex, g</u>	ender (ide	<u>:ntity/pr</u>	<u>esentation)</u>	,
and sexual orientation and race, ethnicity ar	d racism.							

Reporting on sex and gender	No human participants or human data used in this study	
Reporting on race, ethnicity, or other socially relevant groupings	No human participants or human data used in this study	
Population characteristics	No human participants or human data used in this study	
Recruitment	No human participants or human data used in this study	
Ethics oversight	No human participants or human data used in this study	

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Field-specific reporting

Please select the one belo	w that is the best fit for your research.	If you are not sure, read the appropriate sections before making your selection. $ \\$
Life sciences	Behavioural & social sciences	Ecological, evolutionary & environmental sciences

 $For a \ reference \ copy \ of \ the \ document \ with \ all \ sections, see \ \underline{nature.com/documents/nr-reporting-summary-flat.pdf}$

Ecological, evolutionary & environmental sciences study design

All studies must disclose on these points even when the disclosure is negative.

Study description

We sequenced and assembled the genomes of 13 species from different genera in the Salicaceae, and used multi-omics data along with molecular methods to study the genome evolutionary history of Salicaceae. This study revealed the types and characteristics of polyploidization events in the Salicaceae, and further explored how different genomic evolutionary patterns following allopolyploidy have driven trait innovation and subsequent adaptive radiation in species.

Research sample

Fresh leaf, flower and fruit samples used in this study were collected: Plant material of C. decandra, D. caffra, S. chinensis, X. longifolia and F. jangomas were collected in XiShuangBanNa Tropical Botanical Garden (Mengla, China), I. orientalis, I. polycarpa and P. deltoides were collected in Chengdu, S. rehderiana was collected in Minya Konka of China, P. euphratica and P. alba var. pyramidalis were collected in Lanzhou, Gansu, S. chaenomeloides was collected in Hanzhong, Shanxi, and Dianyuea turbinata was collected from Dehong, Yunnan.

Sampling strategy

Our sampling was based on the reported phylogenetic relationships within the Salicaceae family. Fresh leaf, flower, and fruit tissues were immediately placed on dry ice or in liquid nitrogen for subsequent sequencing.

Data collection

DNA was extracted from fresh leaf tissues and sequenced on the ONT, PacBio Sequel II and Illumina platforms, generating long and short reads for genome assembly. ATAC-seq libraries and whole genome bisulfite sequencing were performed on the Illumina platform to obtain epigenetic data. RNA was extracted from leaves, flowers, and fruits at different developmental stages, and transcriptome data were obtained through sequencing on the Illumina platform.

Timing and spatial scale

Mature leaf samples of different species were collected for genome sequencing from 2019 to 2023. Flower and fruit samples from I. orientalis, I. polycarpa and P. deltoides were collected at different developmental stages from March to November, and leaves of I. orientalis and I. polycarpa were collected from November to April.

Data exclusions

No data were excluded.

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Reproducibility	Two to three replicates were collected for each tissue type of RNA-seq, ATAC-seq, and WGBS. The bootstrapping for phylogenetic analyses based on orthologs in all genomes and different subgenomes was replicated 100 times. The dual-luciferase assay was repeated three to six times to detect the enhancer activity of conserved noncoding elements (CNEs). All attempts at replication were successful.
Randomization	Since this is a genome sequencing study and the data were from a single individual, and leaf, flower and fruit tissues at different developmental stages were collected for transcriptome sequencing, no randomization was applied in this manuscript.
Blinding	Group allocation was not relevant to this study, so blinding was not required for our work.
Did the study involve f	
Reporting t	or specific materials, systems and methods
	m authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each materia elevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.
Materials & experin	nental systems Methods
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Antibodies	ChIP-seq
Eukaryotic cell lir	
Palaeontology ar	
Animals and other	er organisms
Clinical data Dual use research	o of concern
Plants	To Concern
Plants	
Seed stocks Novel plant genotypes	Plant material of C. decandra, D. caffra, S. chinensis, X. longifolia and F. jangomas were collected in XiShuangBanNa Tropical Botanica Garden (Mengla, China), I. orientalis, I. polycarpa and P. deltoides were collected in Chengdu, S. rehderiana was collected in Minya Konka of China, P. euphratica and P. alba var. pyramidalis were collected in Lanzhou, Gansu, S. chaenomeloides was collected in Hanzhong, Shanxi, and Dianyuea turbinata was collected from Dehong, Yunnan Fresh leaf, flower and fruit tissues were collected and placed on dry ice or liquid nitrogen for subsequent sequencing.

No novel plant genotypes were produced in this study.

Authentication

No seed stocks were used and no novel plant genotypes were generated in this study.