nature portfolio

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Last updated by author(s):	05-16-2023

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 Editorial Policies and the Editorial Policy Checklist.

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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
	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.
\boxtimes	A description of all covariates tested
	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>
X	For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
X	For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
\times	Estimates of effect sizes (e.g. Cohen's <i>d</i> , Pearson's <i>r</i>), indicating how they were calculated
	Our web collection on statistics for biologists contains articles on many of the points above.

Software and code

Policy information about availability of computer code

Data collection

No software was used to collect the phenotype data

Data analysis

Here are software packages used in the study. RNA-seq data analysis, EMTA, and mapping: Trimmomatic version 0.32; STAR v2.7.10a; IGV version 2.8.9; Freebayes v1.3.6; BCFtools v1.14; Spades version 3.14.1; Samtools v1.8; Genome-specific primers: Primer 3 v0.4.0; Phylogenetic analysis: NCBI (ttps://www.ncbi.nlm.nih.gov); Reference genomes (https://wheat.pw.usda.gov/blast/); MEGA v7.0; iTOL v5.0 (https:// itol.embl.de/); Structure prediction: AlphaFold v2.0.1; Evaluation of agronomic and quality traits: Wanshen SC-G; Instron 5848 Microtester; Model SpectraStar 2600 XT-R; TA-XT-plus Texture Analyzer; Other softwares used for data analysis: ESPript 3.0 (https://espript.ibcp.fr/ESPript/ $cgi-bin/ESPript.cgi); cNLS \ Mapper \ (https://nls-mapper.iab.keio.ac.jp/cgi-bin/NLS_Mapper_form.cgi); Pfam \ (http://pfam-legacy.xfam.org/lineary.cgi); pfam \ (http://pfam-legacy.xfam.org/linea$ search/sequence)

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

Data supporting the findings of this work are available within the paper and its supplementary information files. All the raw sequencing data and genome assembly generated for this project are archived at the National Genomics Data Center under BioProject accession number CRA011051 that are publicly accessible at https:// ngdc.cncb.ac.cn/search/?dbId=gsa&q=CRA011051. The sequence of Lr47 is available in the GenBank database as accession number OQ919262.

Research involving human participants, their data, or biological material

Policy information about studies with <u>human participants or human data</u> . See also policy information about <u>sex, gender (identity/presentation),</u> and sexual orientation and <u>race, ethnicity and racism</u> .			
Reporting on sex and gender	N/A		
Reporting on race, ethnicity, or other socially relevant groupings	N/A		
Population characteristics	N/A		
Recruitment	N/A		
Ethics oversight	N/A		
Note that full information on the approval of the study protocol must also be provided in the manuscript.			
ield-specific reporting			
Please select the one below that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection.			
X Life sciences ☐ Be	ehavioural & social sciences		

Life sciences study design

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

For a reference copy of the document with all sections, see <u>nature.com/documents/nr-reporting-summary-flat.pdf</u>

Sample size

The sample size was listed in the specific experiments. In our mapping experiments, three F2 populations including 2654, 1141, and 128 plants were used to map the Lr47 gene within a small interval. A collection of 118 accessions of Ae. speltoides, 24 of Triticum monococcum, 78 of T. turgidum, and 144 of T. aestivum was used to determine the value of the diagnostic marker for Lr47. The population sizes are bigger than most reported population sizes in similar studies.

Data exclusions No data were excluded

Replication Mutants and transgenic phenotypes were repeated twice with similar outcomes

Inoculated plants and qRT-PCR experiments were allocated randomly among groups. The phenotyping experiment was done in a controlled Randomization environment where all of the exact same methods were used and scored by experienced researchers.

Blinding was performed when phenotyping plants (the person doing the phenotyping did not have access the genotyping data). Blinding

Reporting for specific materials, systems and methods

We require information from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, system or method listed is relevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.

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Materials & experimental systems		ntal systems Me	ethods	
n/a	Involved in the study	n/a	Involved in the study	
	Antibodies	\boxtimes	ChIP-seq	
\boxtimes	Eukaryotic cell lines	\boxtimes	Flow cytometry	
\boxtimes	Palaeontology and a	rchaeology	MRI-based neuroimaging	
\boxtimes	Animals and other organisms			
\boxtimes	Clinical data			
\boxtimes	Dual use research of concern			
Plants				
,				
Antibodies				
An	tibodies used	Commercial antibody: Anti-GFP (green fluorescent protein; Abcam, ab290)	
Va	idation	Validation of commercial antibodies is provided on the manufacturer's website: Abcam, ab290, https://www.abcam.com/products/primary-antibodies/gfp-antibody-ab290.html		