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
\boxtimes	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
\boxtimes	A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
\boxtimes	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)
\boxtimes	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	\square Estimates of effect sizes (e.g. Cohen's d , Pearson's r), indicating how they were calculated
	Our web collection on <u>statistics for biologists</u> contains articles on many of the points above.

Software and code

Policy information about <u>availability of computer code</u>

Data collection

PacBio Sequel II

Data analysis

 $\label{lem:canuv1.8} \ \ \text{Canu v1.8, seqkit v0.11.0, pbmm2 v1.2.1, minimap2 v2.14, IGV browser v2.8.2, Quast v5.0.2, PROKKA v1.14.6, Interproscan v5.46-81.0, Kofamscan v1.3.0, Augustus v. 3.4.0, CD-HIT v4.8.1, SMRTLink v8.0.0, MAFFT v7.475, RaxML v8.2.12, FigTree v1.4.4, GTDB-tk v1.1.1, BLASTN, BLASTP, checkM v1.1.2, nucmer v4.0.0beta2, Plasflow v1.1, MOB-suite v3.0.0, Virsorter2 v2.2.2, CheckV v0.7.0, gggenes (R v3.6.0), BlastKOALA (https://www.kegg.jp/blastkoala/), AAI calculator (http://enve-omics.ce.gatech.edu/aai/)$

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

The data is available in NCBI database. The accession number is PRJNA929667 (BioProject), SAMN32580422 (BioSample), SRX19223815 and SRX19223816 (SRA).

Human research participants		
Policy information about studies involving human research participants and Sex and Gender in Research.		
Reporting on sex and gen	der N/A	
Population characteristics	S N/A	
Recruitment	N/A	
Ethics oversight	N/A	
Note that full information on the	ne approval of the study protocol must also be provided in the manuscript.	
Field-specific	creporting	
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.	
Life sciences	Behavioural & social sciences	
For a reference copy of the docume	ent with all sections, see <u>nature.com/documents/nr-reporting-summary-flat.pdf</u>	
Ecological, e	volutionary & environmental sciences study design	
All studies must disclose on	these points even when the disclosure is negative.	
Study description	a metagenomic analysis using long-read sequences to reveal the genetic makeup of rice leaf microbiome	
Research sample	Rice plants (Oryza sativa cultivar 'Koshihikari')	
Sampling strategy	Rice plants were collected from an experimental paddy field since we obtain the required amount of gDNA for PacBio Sequel II sequencing. The sample size was estimated by previous reports.	
Data collection	The rice roots and aerial parts were separated and stored at -80 °C before use. The aerial parts were grounded, and plant-associated microbes were enriched using a bacterial cell enrichment method. Genomic DNA was extracted from the enriched microbes using enzymatic lysis. The library for sequencing was constructed and sequenced within a week after DNA extraction. SMRTbell libraries for sequencing were constructed according to the manufactural protocol without shearing. The libraries were cut off at 20 kbp using the BluePippin size selection system and sequenced on two SMRT Cells 8M.	
Timing and spatial scale	The rice plants were sampled before heading on August 6th, 2018	
Data exclusions	No data were excluded	
Reproducibility	N/A	
Randomization	Rice plants were randomly sampled from the paddy field on the same day	
Blinding	No blinding was performed	
Did the study involve field	d work? X Yes No	
Field work, collection and transport		
Field conditions	The information is available at the Institute for Sustainable Agro ecosystem Services, Graduate School of Agricultural and Life Sciences, The University of Tokyo when researches inquire (https://www.isas.a.u-tokyo.ac.jp/en/page/about/sample)	

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Location

An experimental paddy field at the Institute for Sustainable Agro ecosystem Services, Graduate School of Agricultural and Life Sciences, The University of Tokyo (35°74'N, 139°54'E)

Access & import/export

Sampling site access (https://www.isas.a.u-tokyo.ac.jp/en/)

Disturbance

No disturbance

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.

Materials & experimental systems		Methods		
n/a	Involved in the study	n/a	Involved in the study	
\boxtimes	Antibodies	\boxtimes	ChIP-seq	
\times	Eukaryotic cell lines	\boxtimes	Flow cytometry	
\times	Palaeontology and archaeology	\boxtimes	MRI-based neuroimaging	
\boxtimes	Animals and other organisms			
\boxtimes	Clinical data			
\boxtimes	Dual use research of concern			