

Reporting Summary

Nature Research 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 Research policies, see our [Editorial Policies](#) and the [Editorial Policy Checklist](#).

Statistics

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.
- 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. F , t , r) with confidence intervals, effect sizes, degrees of freedom and P value noted
Give P values as exact values whenever suitable.
- For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
- For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
- Estimates of effect sizes (e.g. Cohen's d , Pearson's r), 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 were used for data collection

Data analysis The manuscript describes the hifiasm assembly algorithm, which is free available at: <https://github.com/chhylp123/hifiasm>. We used hifiasm (v0.12), Falcon-Unzip (v1.8.1 for Falcon-kit & v1.3.7 for Falcon-unzip-kit), HiCanu (v2.1), Peregrine (v0.1.6.1) to generate different assemblies for comparison. Purge_dups (v1.2.3) was used to post-process assemblies of HiCanu. Mminimap2/paftools (r974), minigraph (r361), BUSCO (v3.0.2), dipcall (v0.1), QUAST (v5.0.2), dna-nn (r60), winnowmap (v1.11), Merqury (v1.1), htsbox (r345) and yak (r55) were used to evaluate all assemblies. BAC resolution of each assembly was measured by: <https://github.com/skoren/bacValidation>. The collapsed misassemblies for inbred samples were measured by the scripts at: <https://github.com/lh3/CHM-eval/blob/master/misc/clustreg.js>, and <https://github.com/lh3/CHM-eval/blob/master/misc/select-collapse-het.js>. All tools are open source and available at their corresponding repositories.

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 Research [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 list of figures that have associated raw data
- A description of any restrictions on data availability

All HiFi data were obtained from NCBI Sequence Read Archive (SRA): SRR11606869 for Z. mays, SRR11606870 for M. musculus, SRR11606867 for F. x ananassa,

SRR11606868 and SRR12048570 for *R. muscosa*, SRP251156 for *S. sempervirens*, SRR11292120 through SRR11292123 for CHM13, ERX3831682 for HG00733, and four runs (SRR10382244, SRR10382245, SRR10382248 and SRR10382249) for HG002. For trio binning and computing QV, short reads were also downloaded: SRR7782677 for HG00733, ERR3241754 for HG00731 (father), ERR3241755 for HG00732 (mother) and SRX1082031 for CHM13. GIAB's "Homogeneity Run01" short-read runs were used for the HG002 trio. These HG002 reads were downsampled to 30-fold coverage. The BAC libraries of CHM13 and HG00733 can be found at <https://www.ncbi.nlm.nih.gov/nuccore/?term=VMRC59+and+complete> and <https://www.ncbi.nlm.nih.gov/nuccore/?term=VMRC62+and+complete>, respectively. The HG002 MHC reference sequences can be found at <https://github.com/NCBI-Hackathons/TheHumanPangenome/tree/master/MHC/assembly/MHCv1.1>. For BUSCO, the embryophyta dataset, the tetrapoda dataset and the mammalia dataset are available at https://busco-data.ezlab.org/v4/data/lineages/embryophyta_odb10.2020-09-10.tar.gz, http://busco.ezlab.org/v2/datasets/tetrapoda_odb9.tar.gz and http://busco.ezlab.org/v2/datasets/mammalia_odb9.tar.gz, respectively. The CHM13 reference (v0.9) generated by the T2T consortium can be found at https://s3.amazonaws.com/nanopore-human-wgs/chm13/assemblies/chm13.draft_v0.9.fasta.gz. The hifiasm assemblies produced in this work are available at <ftp://ftp.dcfi.harvard.edu/pub/hli/hifiasm/submission/>.

Field-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.

Life sciences Behavioural & social sciences Ecological, evolutionary & environmental sciences

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

Life sciences study design

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

Sample size	<input type="text" value="Not applicable since no statistical tests or power analyses were performed"/>
Data exclusions	<input type="text" value="No data were excluded from analysis"/>
Replication	<input type="text" value="Not applicable since this study only describes deterministic algorithms without statistic analysis"/>
Randomization	<input type="text" value="Not applicable since this study introduces a method and does not include biological hypotheses analysis. All assembly algorithms are deterministic."/>
Blinding	<input type="text" value="Not applicable since this study does not involve statistic analysis and data acquisition"/>

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

n/a	Involved in the study
<input checked="" type="checkbox"/>	<input type="checkbox"/> Antibodies
<input checked="" type="checkbox"/>	<input type="checkbox"/> Eukaryotic cell lines
<input checked="" type="checkbox"/>	<input type="checkbox"/> Palaeontology and archaeology
<input checked="" type="checkbox"/>	<input type="checkbox"/> Animals and other organisms
<input checked="" type="checkbox"/>	<input type="checkbox"/> Human research participants
<input checked="" type="checkbox"/>	<input type="checkbox"/> Clinical data
<input checked="" type="checkbox"/>	<input type="checkbox"/> Dual use research of concern

Methods

n/a	Involved in the study
<input checked="" type="checkbox"/>	<input type="checkbox"/> ChIP-seq
<input checked="" type="checkbox"/>	<input type="checkbox"/> Flow cytometry
<input checked="" type="checkbox"/>	<input type="checkbox"/> MRI-based neuroimaging