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# Whole genome sequencing and assembly data of *Moricandia moricandioides* and *M. arvensis*



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## ABSTRACT

*Moricandia* is a genus belonging to the family Brassicaceae.  $C_3$  and  $C_3$ - $C_4$  photosynthesis *Moricandia* species exist in a close phylogenetic proximity, as well as to Brassica crops. Here, we performed PacBio genome sequencing on *M. moricandioides* and *M. arvensis*. The genomes were assembled using Flye assembler, then polished with Illumina reads and reduced duplication with Purge Haplotigs. The total length of genome assemblies of *M. moricandioides* and *M. arvensis* was 498 Mbp and 759 Mbp, respectively. These data will be useful for studies of the genetic control of  $C_3$ - $C_4$  characteristics, therefore gaining new insights into the early evolutionary steps of  $C_4$  photosynthesis. Further, it can be integrated into Brassica crop breeding. The data can be accessed at ENA under the project number PRJEB39764.

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## Specifications Table

Subject	(Plant) Biology
Specific subject area	Genomics
Type of data	Table, figure, genomic assembly data in FASTA format
How data were acquired	Whole genome sequencing was performed using the PacBio RSII platform
Data format	Raw and analyzed
Parameters for data collection	gDNA was extracted from mature leaf tissues of <i>M. moricandioides</i> and <i>M. arvensis</i>
Description of data collection	Sequencing reads were generated on the PacBio RS II platform. The reads were then assembled <i>de novo</i> using Flye assembler.
Data source location	Düsseldorf, Germany
Data accessibility	Accessible as a project on European Nucleotide Archive (PRJEB39764)
	https://www.ebi.ac.uk/ena/submit/sra/#studies
	Link for direct download of raw data:
	<https: browser="" ena="" prjeb39764="" view="" www.ebi.ac.uk=""></https:>
	Link for direct download of FASTA-format assemblies:
	<https: browser="" ena="" gca_905132765.1="" view="" www.ebi.ac.uk=""></https:>
	<https: browser="" ena="" gca_905132885.1="" view="" www.ebi.ac.uk=""></https:>

## Value of the Data

- *M. moricandioides* and *M. arvensis* are closely related sister species within the crucifers (Brassicaceae)
- *M. moricandioides* performs C<sub>3</sub> photosynthesis whereas *M. arvensis* performs C<sub>3</sub>-C<sub>4</sub> intermediate photosynthesis
- These are the first whole genome assemblies of M. moricandioides and M. arvensis
- The genome assemblies provide the basis for identification of genetic factors underpinning  $C_{3-}C_4$  photosynthesis, through comparative genomics
- $C_{3-}C_4$  photosynthesis is an intermediary step on the evolutionary trajectory from  $C_3$  to  $C_4$  photosynthesis; hence the data will provide novel insights into the early evolutionary steps into the direction of  $C_4$  photosynthesis.
- The genome data serve as a valuable resource for engineering of the photosynthetic efficiency of C<sub>3</sub> plants.

## 1. Data Description

We present here the genome sequencing and assembly of  $C_3$  *M. moricandioides* and  $C_{3-}C_4$  *M. arvensis*. A total output of 43 and 71.9 Gb was generated by PacBio sequencing for *M. moricandioides* and *M. arvensis*, respectively. The *de novo* assembly was performed using Flye assembler, followed by reducing the duplication with Illumina reads through Purge Haplotigs. The resulting draft genome of *M. moricandioides* was 498,312,484 bp in size with a GC content of 36.02% with N50 contig length of 114.3 kb. And the assembled genome size of *M. arvensis* was 758,710,152 bp with a GC content of 36.75% with N50 contig length of 82.1 kb. Main *de novo* assembly statistics of the *Moricandia* genome assemblies are shown in Table 1. We evaluated the assembled genomes by Benchmarking Universal Single-Copy Orthologs (BUSCO) (Fig. 1). The majority (96.4% and 95.7% of *M. moricandioides* and *M. arvensis*, respectively) was complete BUSCO genes. For the rest of the genomes, the amount of fragmented genes was 1.3% and 1.2% and the missing genes presented 2.3% and 3.1% for *M. moricandioides* and *M. arvensis*, respectively. The genome assembly of *M. moricandioides* comprised more complete and single copy BUSCOS (83.4% of the complete BUSCOS) than *M. arvensis* genome assembly (71.3% of the complete BUSCOS).

BUSCO completeness assessments of *M. moricandioides* (Mm) and *M. arvensis* (Ma) genome assembly. The proportions of complete (C, sum of light and dark blue bars), complete single-copy (S, light blue), complete duplicated (D, dark blue), fragmented (F, yellow), and missing (M, red) BUSCOs are shown in the bar chart.

#### Table 1

Main de novo assembly statistics.

	M. moricandioides	M. arvensis
Yield	43 Gb	71.9 Gb
Mean read length	7.5 kb	10.2 kb
SMRT cells	50	56
Yield/SMRT cell	0.859 Gb	1.284 Gb
Genome size estimated (Gb)	0.66 Gb	0.737 Gb
Coverage (est. genome size)	65	98
Coverage (assembled genome size)	86	95
Corrected bases (Gb)	21.2	40.8
Trimmed bases (Gb)	21.2	40.2
Bases assembled (bp)	498,312,484	758,710,152
No. contigs	9638	15,876
N50 (kb)	114.3	82.1
GC content	36.02%	36.75%

## **BUSCO Assessment Results**



Fig. 1. Genome assembly assessment by BUSCO.

## 2. Experimental Design, Materials and Methods

## 2.1. Sampling and DNA extraction

For PacBio sequencing, plants of *M. moricandioides* (Botanical Garden Osnabrück: 04–0393– 10–00) and *M. arvensis* (IPK Gatersleben: MOR1) were grown in soil under controlled conditions in the CLF climate chamber under conditions of 12 h day (23 °C, ca. 200 µmol m-2 s-1)/12 h night (20 °C) in December 2015; plants of *M. arvensis* (IPK Gatersleben: MOR1) were grown under the same conditions in June 2016. For Illumina sequencing, plants of *M. moricandioides* and *M. arvensis* were grown under the same conditions in April/May 2019. The whole plant shoot samples were harvested ca. five weeks after germination and stored at –80 °C. For preparation of gDNA, nuclei were extracted first after the method described by [1]. Nuclei were treated with RNAse A (10 mg/ml) for 30 min at 37 °C followed by incubation with protease K (0.8 mg/ml) for 2 h at 50 °C. The DNA samples were purified using Qiagen G20 tips (Qiagen, Germany) following manufacturer's instructions. DNA was resuspended in TE (10 mM Tris, pH 8.0, 1 mM EDTA).

## 2.2. Library preparation and sequencing

The preparation of the *M. moricandioides* sequencing library was made by the SMRTbell Express Template Prep Kit 3.0 and whole genome sequencing was performed on PacBio RS II platform (Pacific Biosciences, Menlo Park, CA, USA) by the Max Planck-Genome-centre Cologne, Germany (https://mpgc.mpipz.mpg.de/home/). The *M. arvensis* gDNA was constructed into a library using the SMRTbell Express Template Prep Kit 2.0 and was sequenced on PacBio RS II platform by KeyGene, N. V., Wageningen, Netherlands (https://keygene.com). In total, the output of 43 and 71.9 Gb was generated for *M. moricandioides* and *M. arvensis*, respectively. These yielded at least 86 × coverage of each genome.

The gDNA of *M. moricandioides* and *M. arvensis* was subjected to a paired-end library preparation for genome sequencing on the Illumina HiSeq 3000 platform (Illumina, San Diego, USA) by the Biologisch-Medizinisches Forschungszentrum (BMFZ) of the Heinrich-Heine University (Düsseldorf, Germany). Prior to library preparation 650 ng of gDNA were sheared with Covaris ME220 (Covaris, Inc.) to a mean fragment size of 350 bp. Library preparation was performed according to the manufacturer's protocol by using the VAHTS Universal DNA Library Prep Kit for Illumina (Vazyme Biotech Co.; Ltd) without any amplification step and an additional size selection step after adaptor ligation to get rid of smaller fragments. Library was quantified via qPCR by using KAPA library quantification kit (Roche Diagnostics Corporation) on a QuantStudio 3 (Thermo Fisher Scientific Inc.) and subsequently sequenced on a HiSeq3000 system (Illumina Inc) with a read setup of  $2 \times 151$  bp. 114 and 167 million reads per library were obtained from *M. moricandioides* and *M. arvensis*, respectively.

## 2.3. Quality control, genome assembly, and genome assessment

*De novo* assembly of the PacBio reads was performed by Flye [2] with the estimated genome size set to 1Gbp for both species. The PacBio reads were mapped back to the assembly with BLASR [3] via pbalign (https://github.com/PacificBiosciences/pbalign), and used to generate a polished consensus with Arrow (https://github.com/PacificBiosciences/GenomicConsensus). The Illumina reads were quality controlled using FASTQC, mapped using BWA [4], and used to polish the genome assembly through Pilon [5]. Finally the PacBio reads were mapped to the twice-polished assembly using Minimap2 with PacBio preset settings [6], and then used for the reduction of duplication with Purge Haplotigs [7]. Coverage thresholds of 6, 42, 140 (*M. moricandioides*) and 4, 32, 140 (*M. arvensis*) for Purge Haplotigs' low, middle, and high coverage, respectively. The genome assembly evaluation was conducted by BUSCO v3 [8] and QUAST [9].

## **CRediT Author Statement**

**Nils Koppers:** Conceptualization, Methodology, and Software; **Meng-Ying Lin:** Data curation, Writing - Original draft preparation, and Visualization; **Alisandra Denton:** Data curation and Validation, Writing - Reviewing and Editing; **Urte Schlüter:** Sample preparation, Reviewing and Editing; **Andreas P.M. Weber:** Supervision, Reviewing and Editing.

## **Declaration of Competing Interest**

The authors declare that they have no known competing financial interests or personal relationships which have, or could be perceived to have, influenced the work reported in this article.

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