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Hi-C library construction from young Maize leaves

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Yikun Zhao[1], Yuancong Wang[1], De Ma[2], Guang Feng[3], Yongxue Huo[1], Zhihao Liu[1], Ling Zhou[4], Yunlong Zhang[1], Liwen Xu[1], Liang Wang[3], Han Zhao[4], Jiuran Zhao[1], Fengge Wang[1]

[1]Maize Research Center, Beijing Academy of Agricultural and Forest Sciences (BAAFS)/Beijing Key Laboratory of Maize DNA Fingerprinting and Molecular Breeding;
[2]Novogene Bioinformatics Institute; [3]Dandong Academy of Agricultural Sciences;
[4]Provincial Key Laboratory of Agrobiology, Institute of Crop Germplasm and Biotechnology, Jiangsu Academy of Agricultural Sciences

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Hongling Zhou
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We use this protocol and it's working

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Abstract

In a recent study we constructed a high-quality chromosome-level reference genome for the maize cultivar Dan340 by combining PacBio long HiFi sequencing reads, Illumina short reads and chromosomal conformational capture (Hi-C) sequencing reads.

A Hi-C library was constructed using young leaves following previously published procedures with slight modifications. See the source paper and the following protocol.

CITATION

Belton JM, McCord RP, Gibcus JH, Naumova N, Zhan Y, Dekker J (2012). Hi-C: a comprehensive technique to capture the conformation of genomes.. Methods (San Diego, Calif.).
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1 Approximately 5 g leaf samples from seedling were cut into minute pieces and cross-linked by 4% formaldehyde solution at room temperature in a vacuum for 00:30:00 . 30m

2 Each sample was mixed with an excess of [M] 2.5 Molarity (M) glycine to quench the crosslinking reaction for 00:05:00 and then placed on ice for 00:15:00 . 20m

3 The cross-linked DNA was extracted and then digested for 12:00:00 with 20 units of DpnII restriction enzyme (NEB) at 37 °C , and the resuspended mixture was incubated at 65 °C for 00:20:00 to inactivate the restriction enzyme. 12h 20m

4 The sticky ends of the digested fragments were biotinylated and proximity ligated to form enriched ligation junctions and then ultrasonically sheared to a size of 300 - 600 bp.

5 The biotin-labelled DNA fragments were pulled down and ligated with Illumina paired-end adapters, and then amplified by PCR to produce the Hi-C sequencing library.

6 The library was sequenced using an Illumina HiSeq X Ten platform with 2 × 150 bp paired-end reads (Illumina, San Diego, CA, USA).

7 After removing low-quality sequences and trimming adapter sequences, we had 304.37 Gb (approximately 130×) of clean data generated. This is then used for genome assembly.

Citations

Belton JM, McCord RP, Gibcus JH, Naumova N, Zhan Y, Dekker J. Hi-C: a comprehensive technique to capture the conformation of genomes.
https://doi.org/10.1016/j.ymeth.2012.05.001