**The Chromosome Region Capture Sequencing**

Strains: B10.S/SgMcdJ, C3H/HeSnJ (C3H/HeN), CAST/EiJ, CBA/CaJ (CBA/N), MOLF/EiJ, SJL/J

**Fragment DNA**

For the chromosome region capture sequencing, 3 µg of genomic DNA was sheared into fragments of approximately 200 bp with the Covaris E220 system (Covaris, USA), and purification was performed with 1.4-fold volume of AMPure XP Beads (Beckman, USA).

**DNA Library Construction**

After the purification of the sheared DNA, the library was constructed with SureSelect Library Prep Kit (Agilent, USA). End-repair was performed with the volume of 10× End Repair Buffer, dNTP Mix, T4 DNA Polymerase, Klenow DNA Polymerase and T4 Polynucleotide Kinase, reacted at 20° C for 30 min, and 1.8× the volume of AMPure XP Beads was added for purification; adding A at the 3' end was performed with the volume of 10×Klenow Polymerase Buffer, dATP and Exo(-) Klenow, reacted at 37° C for 30 min. T4 DNA Ligase Buffer, SureSelect Adaptor Oligo Mix and T4 DNA Ligase were added and reacted at 20° C for 15 min. Adaptor-ligated library was amplified through added in SureSelect Primer, SureSelect ILM Indexing Pre Capture PCR Reverse Primer, 5×Herculase II Rxn Buffer, 100 mM dNTP Mix and Herculase II Fusion DNA Polymerase, and the reaction procedure is: 98° C pre-denaturation 2 min, 98° C denaturation 30 sec, 65° C annealing 30 sec, 72° C extension 30 sec, amplified for 4 rounds. Purification was performed with 1.8× Agencourt AMPure XP beads after each enzymatic reaction. The final adaptor-ligated library was 225-275 bp in size.

**Hybridization capture**

Hybridization capture was performed on the prepared library with the SureSelect Target Enrichment Kit (Agilent, USA). The prepared library was reacted with SureSelect Block Mix in 95° C 5 min, followed by maintaining at 65° C. Hybridization Buffer and capture library mix were added and incubated at 65°C for 24 hrs. Streptavidin Dynabeads M-280 (Life Technologies, USA) were used for the enrichment of the Captured DNA library ([Gnirke et al., 2009](#_ENREF_4); [Mamanova et al., 2010](#_ENREF_7)).

**Index amplification**

5× Herculase II Rxn Buffer, 100 mM dNTP Mix, SureSelect ILM Indexing Post Capture Forward PCR Primer and Herculase II Fusion DNA Polymerase were added to the enriched captured DNA library for index amplification. The reaction procedure consisted of 98° C pre-denaturation 2 min, 98° C denaturation 30 sec, 57° C annealing 30 sec, 72° C extension 30 sec, amplification for 12 rounds, followed by the purification using 1.8× the volume of AMPure XP Beads. A sequencing library of 250-350 bp range was obtained ([Gnirke et al., 2009](#_ENREF_4)).

**Sequencing**

A 10 ng library was used for cluster generation in cBot with the TruSeq PE Cluster Kit (Illumina, USA) followed by bidirectional 2× 150 bp sequencing on Illumina Hiseq 2500.

**Whole-genome sequencing**

Strains: BPL/1J, BPN/3J, CASA/RkJ, CZECHII/EiJ, JF1/MsJ, MOLD/EiJ, MRL/MpJ, MSM/MsJ, NU/J, SF/CamEiJ, SKIVE/EiJ, PWD/PhJ, and RF/J

**DNA Library Construction**

For whole-genome sequencing, DNA libraries were constructed according to Illumina recommended protocols. Briefly, 3 µg of genomic DNA was sheared into fragments of approximately 300-400 bp with the Covaris E220 system, followed by end-repair, A-tailing, and ligation of the Illumina multiplexing PE adaptors. Purification was performed with 1.8× Agencourt AMPure XP beads after enzymatic reactions. An agarose gel electrophoresis with a concentration of 2% to separate DNA products was performed, and DNA fragments with a length between 300-400 bp were excised and purified using the Qiagen Gel Extraction Kit. A PCR enrichment experiment was performed to ensure the presence of sufficient DNA products for sequencing.

**Library inspection**

After construction of the library, preliminary quantification was performed using Qubit 2.0, the library was diluted to 1 ng/ul, and then the insert size of the library was detected using Agilent 2100. If the insert size was as expected, qPCR was performed to quantify the effective concentration of the library (library effective concentration >10 nM) to ensure library quality.

**Sequencing**

DNA fragments were subjected to the Illumina Hiseq 2000 platform for paired-end sequencing (2× 150bp). The raw image data files obtained by high-throughput sequencing (Illumina) were converted into Sequenced Reads by CASAVA, and the results were stored in FASTQ format.

**Data processing and analysis**

To ensure the quality of subsequent information analysis, the original sequences of the all strains were filtered by SolexaQA software to get high quality Clean Reads ([Cox et al., 2010](#_ENREF_2)). Efficient high-quality sequencing data was mapped to the reference genome mm10 by BWA software ([Li and Durbin, 2009](#_ENREF_5)). SAMtools ([Li et al., 2009](#_ENREF_6)) was used for sorting, picard tools was used for duplication, and GATK was used for Indel Realignment and Base Recalibration ([McKenna et al., 2010](#_ENREF_8)). Finally, HaplotypeCaller of GATK was used for variant detection. The VCF format file was filtered with VCFtools ([Danecek et al., 2011](#_ENREF_3)). The SNP filtered results of each sample are annotated by ANNOVAR software ([Wang et al., 2010](#_ENREF_10)), which included three aspects: annotation based on gene, genomic region, and function. GATK software was also used to detect InDel, and pindel was used to detect SV ([Ye et al., 2009](#_ENREF_11)), which was classified into four types: deletions (>5bp), insertions (>5bp), inversions and tandem duplication. The VCF format files were converted to Plink files with VCFtools. .

**Reference**

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