Short insert size WGS libraries preparation for assembly of the Lateolabrax maculatus genome

Chang Li

Abstract

This protocol is used to clarity the process of the short insert size WGS libraries preparation for the L. maculatus.

Citation: Chang Li Short insert size WGS libraries preparation for assembly of the Lateolabrax maculatus genome. **protocols.io**
dx.doi.org/10.17504/protocols.io.sszeef6
Published: 23 Aug 2018

Protocol

Genomic DNA interruption

Step 1.

The extracted DNA was sheared into fragments between 50 bp and 800 bp in size using a Covaris E220 ultrasonicator (Covaris, Brighton, UK). Treat time 20s, Acoustic Duty Factor 25%, Peak Incident Power 500W, Cycles Per Burst 500, 24 cycles.

DURATION
00:00:20 :

Fragment selection

Step 2.

Fragments between 150 bp and 250 bp or 200 to 500 bp were selected using AMPure XP beads (Agencourt, Beverly, the U.S.).

REAGENTS
AMPure XP beads by Beckman Coulter

End-repair

Step 3.

Repaired using T4 DNA polymerase, (ENZYMATICS, Beverly, the U.S.) 30 min. at 20 ℃ to obtain blunt ends which were then 3’-adenlyated to create sticky ends.

TEMPERATURE
20 °C :

REAGENTS
T4 DNA polymerase by Enzymatics

DURATION

00:30:00 :

Add adapter

Step 4.

These DNA fragments were ligated at both ends to T-tailed adapters and amplified.

PCR amplification

Step 5.

The temperature profile was 3 min. at 95 ℃ followed by 8 cycles of 20 sec. at 98 ℃, 15 sec. at 60 ℃, 30 sec. at 72 ℃, and more 10 min. at 72 ℃ for further elongation.

Library purification

Step 6.

AMPure XP beads (Agencourt, Beverly, the U.S.) was used to purify the PCR production.