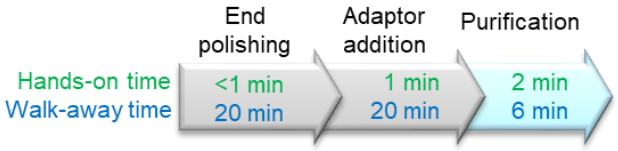
NGS DNA Library Prep Customization Kit (illumina platform)

Catalog No. 30001S: 24 reactions
Catalog No. 30001L: 48 reactions

Description

The **NGS DNA Library Prep Customization Kit** was developed for construction of high quality libraries for next generation sequencing (illumina platform). The kit adds 3'-dT-tailed library adapters to both ends of DNA fragments efficiently. The kit needs double strand DNA fragments (blunt and/or sticky) as input DNA for NGS library construction, and is compatible with DNA fragments generated from both enzymatic methods and mechanical methods.

To facilitate the customization of library prep, the kit made it possible for scientists to use their own adaptors, PCR primers and PCR reagents.



Features

- Flexibility: Customized adaptors, PCR primers and PCR reagents allowed
- Fast
  - Total time: <1 hr
  - Hands-on time: 4 min
- Simple work flow: Less reaction steps
- Less magnetic beads required: Reduced more than 50%
- Guaranteed quality: Higher library conversion efficiency
- Input DNA amount: From 100 ng to 1 ug

Component

	Cat.# 30001S	Cat.# 30001L
• DL1 Buffer	96 ul	192 ul
• DL1 Enzyme	96 ul	192 ul
• DL2A Buffer	576 ul	1152 ul
• DL2 Enzyme	48 ul	96 ul
• Sodium Chloride (2 M)	960 ul	1920 ul

Storage Condition

- Store kit at -20°C, stable up to 12 months.

Reagent & Equipment Needed (not provided in this kit)

- 3'-end T-overhang adaptor
- PCR primers and PCR reagent (optional)
- Magnetic particle concentrator
- PCR thermal cycler
- 96-well PCR plate
- 80% ethanol (prepare before use)
- Sodium Chloride (2 M)*
- Magnetic Beads (BioDynami Cat.# 40051) or equivalent

*Sometimes the tube of Sodium Chloride (2 M) may crack during dry ice shipping. Customers need to prepare Sodium Chloride (2 M) in this case.

Protocol

Step 1: End polishing

1) Add the following to one well of a 96-well PCR plate:

Sheared DNA	42 ul (100 ng~1 ug)
DL1 Buffer	4 ul
DL1 Enzyme	4 ul
Total	50 ul

2) Mix by pipetting ten times.
3) Incubate at 20°C for 15 min, 70°C for 5 min. Proceed immediately to step 2.

Step 2: Adaptor addition

1) Add the following to Step 1 reaction mixture. Slow pipetting of the viscous DL2 Buffer is needed for precise aliquot.

Adaptor (20 uM)*	4 ul
DL2A Buffer	24 ul
DL2 Enzyme	2 ul
Total	30 ul

* Not included in the kit for customization purpose.

2) Mix by pipetting ten times.
3) Incubate at 20°C for 20 min.
4) Add Sodium Chloride (2 M) 40 ul to the reaction mixture. Proceed immediately to step 3.

Step 3: Purification

1) Resuspend Magnetic Beads and transfer 40 ul to the above reaction mixture, mix by pipetting and incubate for 3 min.
2) Load the plate on a magnet, incubate for 2 min, and discard the supernatant carefully. Remove all residual supernatant without disturbing the beads.
3) Add 180 ul of 80% ethanol, incubate for 30 sec, and discard the supernatant carefully. Remove all residual ethanol without disturbing the beads.
4) Remove the plate from the magnet, resuspend the beads in 22 ul of water.
5) Load the plate on the magnet, incubate for 1 min, and transfer 20 ul supernatant (containing library) to a new tube without disturbing the beads.

Step 4: Library amplification when needed (PCR reagent is not provided)

Quality Control

Kit components passed stringent functional quality test.

Product Use Limitation

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BioDynami

601 Genome Way, Huntsville, Alabama 35806, USA
https://BioDynami.com
support@BioDynami.com

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