

MagicPure® Size Selection DNA Beads

Please read the manual carefully before use.

Cat. No. EC401

Version No. Version 2.0

Storage: at 2-8°C for one year. Avoid freezing.

Description

MagicPure® Size Selection DNA beads is based on the technology of solid phase reverse immobilization. The beads are suitable for purify, concentrate, and size selection of DNA when constructing NGS library. MagicPure® Size Selection DNA beads can be used together with any library preparation kits.

Kit Contents

Component	EC401-02	EC401-03	EC401-04
Magnetic DNA Beads	5 ml	60 ml	450 ml

Procedures

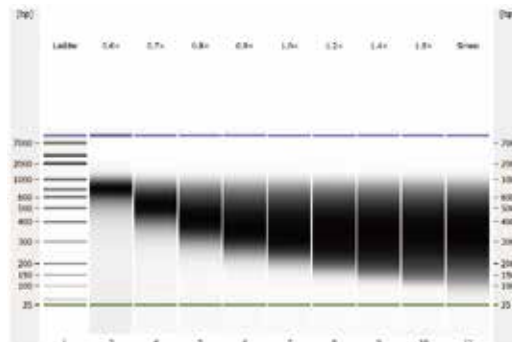
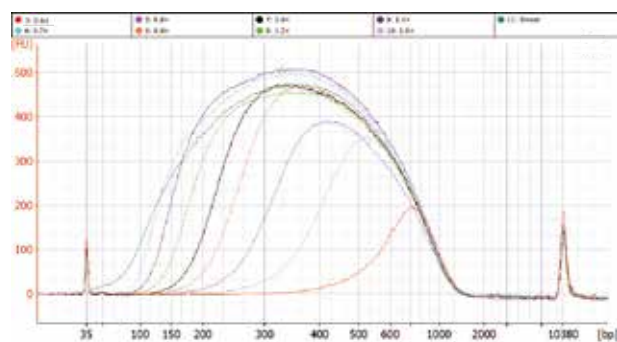
When preparing WGS (Whole genome sequencing) library, you can use the DNA beads according the conditions bellow directly prior to end repair of fragmented DNA or after library amplification. If you want to size select DNA after adapter ligation, you would better purify your sample by 1.0× DNA beads first before size select DNA according the conditions bellow.

Reagents supplied by users: freshly prepared 80% ethanol

Operate at room temperature.

Purification of DNA

Choose the ratio of DNA beads before purification according to the following figure.



Agilent high sensitivity DNA chip electropherogram

Smear - Input control DNA sample in water; 0.6×~1.8× - purified DNA samples with given ratio of DNA beads volume to sample volume.

1. Take out the from 2-8°C refrigerator. Equilibrate the beads at room temperature for 30 minutes before use.
2. Add the DNA sample into a new 1.5 ml tube.
3. Vortex the tube to resuspend DNA beads. Add appropriate volume of beads to the sample according to the volume of samples.

Volume of beads to add = volume of samples × ratio chosen

Example:

50 µl DNA sample is purified by 1.8× DNA beads,

90 µl (beads to add) = 50 µl × 1.8



4. Mix by pipetting up and down. Incubate at room temperature for 5 minutes.
Note: Insufficient mixing will affect the results significantly.
5. Place the tube on an appropriate magnetic stand to separate beads from the supernatant at room temperature. When the solution is clear (about 5 minutes), discard the supernatant.
Note: Spin down briefly before put on magnetic stand if there is liquid on the wall. Make sure that DNA beads are settled to the magnet completely and not to be disturbed when discarding the supernatant. Discarding beads will result in reduced yield.
6. Keep the tube still on the magnetic stand, add 200 μ l of freshly prepared 80% ethanol and incubate at room temperature for 30 seconds without pipetting up and down. Carefully discard the supernatant.
Note: Use freshly-prepared 80% ethanol; otherwise it may affect the result.
7. Repeat step 6 one time.
8. Air dry the beads at room temperature until the magnetic beads just appear dry (about 5 minutes) while the tube is on the magnetic stand.
Note: Be sure to dry the magnetic beads, otherwise it will affect subsequent experiments. Do not heat dry, otherwise it will affect the final yield.
9. Remove the tube from the magnetic stand and elute DNA with ≥ 20 μ l Nuclease-free Water or TE buffer. Mix by pipetting up and down or vortexing. Then incubate at room temperature for 2 minutes.
10. Place the tube on the magnetic stand. Incubate at room temperature until the solution is clear (about 2 minutes). Make sure the beads settle to the magnet completely.
Note: Spin down briefly before put on magnetic stand if there is liquid on the wall. Prolong incubation to 5 minutes if necessary to make sure that RNA beads are settled to the magnet completely.
11. Transfer the supernatant to a new tube. The DNA product can be stored at -20°C .

Size selection of DNA

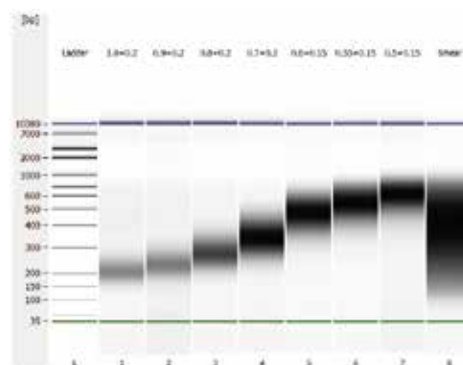
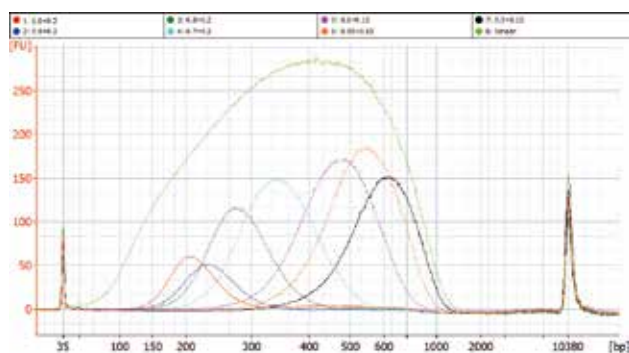
1. Take out the from $2-8^{\circ}\text{C}$ refrigerator. Equilibrate the beads at room temperature for 30 minutes before use.
2. Put the DNA sample to a new 1.5 ml tube.
3. Vortex the tube to resuspend DNA beads. Add appropriate volume of beads to the sample. Add appropriate volume of beads to the sample according the table below (**Reference conditions for size selection**).
Volume of beads to add = volume of sample \times 1st beads ratio chosen
Example:
 $30\ \mu\text{l (beads to add)} = 50\ \mu\text{l sample} \times 0.6$
4. Mix by pipetting up and down or vortexing. Incubate at room temperature for 5 minutes.
Note: Insufficient mixing will affect the results significantly.
5. Place the 1.5 ml tube on an appropriate magnetic stand to separate beads from the supernatant at room temperature until the solution is clear (about 5 minutes).
Note: Spin down briefly before put on magnetic stand if there is liquid on the wall. Make sure that the DNA beads are settled to the magnet completely.
6. Keep the tube still on the magnetic stand, transfer the supernatant to a new 1.5 ml tube. **Discard the beads.**
7. Add appropriate volume of beads to the supernatant according the following table (**Reference conditions for size selection**).
Volume of beads to add = volume of sample \times 2nd beads ratio chosen
Example: 50 μ l of sample, $0.15 \times 2^{\text{nd}}$ beads ratio chosen according to the table
 $7.5\ \mu\text{l (beads to add)} = 50\ \mu\text{l} \times 0.15$
8. Mix by pipetting up and down. Incubate at room temperature for 5 minutes.
Note: Insufficient mixing will affect the results significantly.
9. Place the tube on an appropriate magnetic stand to separate beads from the supernatant at room temperature. When the solution is clear (about 5 minutes), discard the supernatant.
Note: Spin down briefly before put on magnetic stand if there is liquid on the wall. Make sure that DNA beads are settled to the magnet completely and not be disturbed when discarding the supernatant. Beads loss will result in reduced yield.



10. Keep the tube still on the magnetic stand, add 200 μ l of freshly prepared 80% ethanol and incubate at room temperature for 30 seconds without pipetting up and down. Carefully discard the supernatant.
Note: Use freshly-prepared 80% ethanol; otherwise it may affect the result.
11. Repeat step 10 one time.
12. Air dry the beads at room temperature until the magnetic beads just appear dry (about 5 minutes) while the tube is on the magnetic stand.
Note: Be sure to dry the magnetic beads, otherwise it will affect subsequent experiments. Do not heat dry, otherwise it will affect the final yield.
13. Remove the tube from the magnetic stand and elute DNA with ≥ 20 μ l Nuclease-free Water or TE buffer. Mix by pipetting up and down or vortexing. Then incubate at room temperature for 2 minutes.
14. Place the tube on the magnetic stand. Incubate at room temperature until the solution is clear (about 2 minutes). Make sure the beads settle to the magnet completely.
Note: Spin down briefly before put on magnetic stand if there is liquid on the wall. Prolong incubation to 5 minutes if necessary to make sure that RNA beads are settled to the magnet completely.
15. Transfer the DNA Product to a new tube. The DNA product can be stored at -20°C .

Reference conditions for size selection

The average size of selected (bp)	190~220	220~250	250~300	300~400	400~500	500~600	600~750
1 st beads ratio(DNA Beads : DNA)	1.0×	0.90×	0.80×	0.70×	0.60×	0.55×	0.50×
2 nd beads ratio(DNA Beads : DNA)	0.20×	0.20×	0.20×	0.20×	0.15×	0.15×	0.15×



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Smear - Input control DNA sample in water; 1.0+0.2~0.5+0.15 – size selected DNA samples with given conditions for size selection.



Notes

- Avoid freezing the beads.
- Take out the magnetic beads from 2-8°C and incubate for 30 minutes at room temperature before use.
- Beads should be mixed thoroughly before use.
- 80% ethanol should be freshly prepared.
- When transferring DNA product, we recommend leaving 2-3 μ l of supernatant to avoid the beads being disturbed during the process.
- If ligation product is being selected directly. PEG and some other compound in the ligation system may affect the result of the selection. User may need to adjust the selection system according to depending on the circumstances.

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