

Before You Begin

- 1) Prepare absolute isopropanol and absolute ethanol (not provided in the kit).
- 2) Gels containing \leq 1% agarose for purification are required.
- Ensure to add 29 ml of absolute isopropanol (not provided) to PB Buffer before the purification.
- Add correct amount of absolute ethanol (not provided) to FWMB1 Buffer and WB2 Buffer, respectively (see bottle label).
- 5) Pre-heat EA Buffer at 60°C before use.

Fragment DNA purification from PCR product sample

- 1) Add **5 volumes** of **PB Buffer** to the PCR product and mix thoroughly by vortexing. (e.g., For 20 µl of PCR product, add 100 µl of PB Buffer in 1.5 ml tube.)
- 2) Add 100 µl of Magnetic Nano Beads Solution and mix thoroughly by vortexing. (Note) Please vortex the Magnetic Nano Beads Solution before use.
- 3) Place the tube in *MagListo*[™]-2 Magnetic Separation Rack. The *MagListo*[™] stand is attached with the magnet plate. Then gently invert the tube 3-4 times.
- 4) Without removing the tubes from the rack, carefully discard the supernatant and remove the remaining supernatant using the paper towel by blotting.
- 5) Detach the magnet plate from the stand. Add **700 µl** of **WB2 Buffer** to the tube and mix thoroughly by vortexing.
- 6) Attach the magnet plate to the stand and gently invert the tube 3-4 times until the beads are tightly bound to the magnet.
- 7) Remove the supernatants without removing the tubes from the rack.
- 8) Detach the magnet plate from the stand. Add 700 µl of absolute ethanol (not provided), and mix thoroughly by vortexing, then repeat the above steps 6-7.
- 9) Detach the magnet plate from the stand. Completely dry the beads for 10 min with the tube opened at 60°C using the Heating Block. Remove the remaining supernatant using the pipette.
- 9) Add $30-50~\mu l$ of EA Buffer and mix thoroughly by vortexing. Incubate the tube at $60^{\circ}C$ for more than 1 min.
- 10) Attach the magnet plate to the stand and carefully transfer the supernatant containing the nucleic acid to a sterile microcentrifuge tube.



- (III) Fragment DNA purification from Agarose gel
- 1) Visualize the band in agarose gel stained with any nucleic acid staining chemicals and cut the gel around the DNA band of interest using a scalpel blade.
- 2) The maximum amount of gel slice in each sample is **approximately 400 mg**. Add **3 volumes** of **FB Buffer** to the gel slice (e.g., For 200 mg gel slice, add 600 µl of FB Buffer).
 - (Note) Ensure that the FB Buffer color remains yellow.
- 3) Incubate the sample at 60°C for 10 min and invert the tube every 2-3 min during the incubation to mix.
 - (Note) The gel slice should be completely dissolved. If it has not melted completely, extend the incubation time.
- 4) Add 100 µl of Magnetic Nano Beads Solution and mix thoroughly by vortexing. (Note) Please vortex the Magnetic Nano Beads Solution before use.
- 5) Place the tube in **the rack** in which the stand is attached with the magnet plate and gently invert the tube 3-4 times.
- 6) Without removing the tubes from the rack, carefully discard the supernatant out and remove the remaining supernatant using the paper towel by blotting.
- 7) Detach the magnet plate from the stand. Add **700 µl** of **FWMB1 Buffer** to the tube, and mix thoroughly by vortexing.
- 8) Attach the magnet plate to the stand and invert the tube 3-4 times gently, until the beads are tightly bound to the magnet.
- 9) Remove the supernatant without removing the tubes from the rack.
- 10) Detach the magnet plate from the stand. Add 700 μl of WB2 Buffer to the tube and mix thoroughly by vortexing. Repeat the above steps 8-9.
- 11) Detach the magnet plate from the stand. Add **700 µl** of **absolute ethanol (not provided)** to the tube, mix thoroughly by vortexing, and repeat the above steps 8-9.
- 12) Detach the magnet plate from the stand. Completely dry the beads for 10 min with the tube opened at 60°C using the Heating Block. Remove the remaining supernatant using the pipette.
- 13) Add 30-50 μI of EA Buffer and mix thoroughly by vortexing. Incubate the tube at 60°C for more than 1 min.
- 14) Attach the magnet plate to the stand and carefully transfer the supernatant containing the nucleic acid to a sterile microcentrifuge tube.

For more information, please visit www.bioneer.com and refer to the User Guide of this kit.

Step	lmage	Description
Binding		Add PB Buffer to the PCR product and mix. - Add 5 volumes of PB buffer to the PCR product. (ex. For 20 µl of PCR product, add 100 µl of PB buffer)
	No.	Add 100 μl of Magnetic Nano Bead and mix
	1. 2. 3.	Follow these 3 Magnet Attach/Detach Steps 1. Attach the magnet 2. Discard the supernatant 3. Detach the magnet
1# Wash	E	Add 700 μl of WB2 Buffer and mix
		Repeat the above "magnet attach/detach step" (step 1,2 and 3)
2 nd Wash		Add 700 µl of Absolute ethanol and mix
		Repeat the above "magnet attach/detach step" (step 1,2 and 3)
Drying	Drying Bead	Dry the beads with the tube opened at 60°C for 10 min
Elution		Add 30-50 µl of EA buffer and mix
		Use the 60°C Heating block for more than 1 min

BQ-042-101-15 Revision: 0 (2020-05-28)

Step	lmage	Description
Gel solublization		Add FB Buffer to the gel slice - Gel sample: approx. 400 mg - Add 3 volumes of FB buffer to the gel slice (e.g., for 200 mg of gel slice, add 600 µl of FB buffer)
		Use the 60°C Heating block for 10 min and invert the tube every 2-3 min during incubation
Binding		Add 100 μl of Magnetic Nano Bead and mix
	1. 2. 3.	Follow these 3 Magnet Attach/Detach Steps 1. Attach the magnet 2. Discard the supernatant 3. Detach the magnet
1st Wash	E Service Serv	Add 700 µl of FWMB1 Buffer and mix
		Repeat the above "magnet attach/detach step" (step 1,2 and 3)
2 nd Wash	No.	Add 700 µl of WB2 Buffer and mix
		Repeat the above "magnet attach/detach step" (step 1,2 and 3)
3 rd Wash		Add 700 µl of Absolute ethanol and mix
		Repeat the above "magnet attach/detach step" (step 1,2 and 3)
Drying	Drying Bead	Dry the beads with the tube opened at 60°C for 10 min
Elution	E	Add 30-50 µl of EA buffer and mix
		Use the 60°C Heating block for more than 1 min

BQ-042-101-15 Revision: 0 (2020-05-28)