

【Product Name】 MagPure Circulating DNA Rich Mini Kit

【Product specifications】 50 Preps/Kit, 200 Preps/Kit

【Intended Use】

MagPure Circulating DNA Rich Mini Kit designed for purification of high quality circulating DNA (cfDNA) from cell-free body fluids (such as plasma, serum). The purified DNA is suitable for direct use in downstream applications such as PCR, real-time PCR, Biochip analysis and NGS.

【Principle】

This product is based on the purification method of high binding magnetic particles. The sample is lysed and digested under the action of lysate and Protease. DNA is released into the lysate. After adding magnetic particles and binding solution, Large DNA(>500bp) will be adsorbed on the surface of magBind particles. After removal of large size, small size of DNA(<500bp) will be adsorbed on the surface of MagPure Particle F and impurities such as proteins will be removed without adsorption. The adsorbed particles were washed with washing solution to remove proteins and impurities, washed with ethanol to remove salts, and finally DNA was eluted by Elution Buffer.

【Main Composition】

Cat.No.	1291750	12917200	Composition
Purification Times	50 Preps	200 Preps	-
MagPure Particles F	1.2 ml	4.5 ml	Magnetic Particles
MagBind Particles	1.2 ml	4.5 ml	Poly A
Proteinase K	24 mg	90 mg	Protease
Protease Dissolve Buffer	1.8 ml	10 ml	Glycorel/Tris/CaCl ₂
Buffer MLB	30 ml	120 ml	Guanidine Salt
Buffer GDP	15 ml	60 ml	
Buffer MAW1	40 ml	250 ml	Guanidine Salt
Buffer MW2*	20 ml	50 ml	Tris/NaCl
Elution Buffer	15 ml	60 ml	Tris

【Storage conditions and Validity】

Proteinase K, MagBind Particles and MagPure Particles F should be stored at 2–8°C upon arrival. However, short-term storage (up to 12 weeks) at room temperature (15–25°C) does not affect their performance. The remaining kit components can be stored dry at room temperature (15 - 25° C) and are stable for at least 18 months under these conditions. The entire kit can be stored at 2 - 8° C, but in this case buffers should be redissolved before use. Make sure that all buffers are at room temperature when used.

【Preparation before Use】

- Add 1.2/4.5 ml Protease Dissolve Buffer to the Proteinase K, and store at -20~8°C after dissolve.
- Add 80ml (50 Preps) or 200ml (200 Preps) 100% ethanol to Buffer MW2 and store at room temperature.

【Manual Protocol for 300ul】

1. Add 20µl Proteinase K and 0.3ml plasma/serum to a sterile 1.5ml centrifuge tube.
2. **Add 0.45ml Buffer MLB and 20µl MagBind Particles to the sample, vortex for 1 minutes. Incubate at room temperature for 10min with occasional mix by inverting.** MagBind Particles adsorb large fragments in this step, discard the MagBind Particles in this step.
3. Place the tube to the magnetic stand for 5-10 minutes until the beads have formed a tight pellet.
4. Transfer the supernatant to a new 1.5ml centrifuge tube. **Add 0.15ml Buffer GDP and 20µl MagPure Particles F to sample,** Inverting to mix for 10 minutes.
5. Place the tube to the magnetic stand for 1 minutes until the beads have formed a tight pellet. Then remove the supernatant.
6. **Add 600µl Buffer MAW1** and vortex for 15 seconds to re-suspend beads. Place the tube to the magnetic stand for 1 minute until the beads have form a tight pellet. Then remove the supernatant.
7. **Add 600µl Buffer MW2,** and vortex for 15 seconds to re-suspend beads. Place the tube to the magnetic stand for 1 minute until the beads have form a tight pellet. Then remove the supernatant.
8. **Add 600µl Buffer MW2,** and vortex for 15 seconds to re-suspend beads. Place the tube to the magnetic stand for 1 minute until the beads have form a tight pellet. Then remove the supernatant.
9. Centrifuge shortly to collect liquid on the tube. Place the tube to the magnetic stand and remove all the liquid carefully.
10. Air dry for 10 minutes.
11. Add 30~50µl Elution Buffer/Low TE/Sterile Water and re-suspend the beads by vortex. Sit at room temperature for 5 minutes. Shake 1~2 times to dissolve DNA from magnetic particles more efficiently. Place the tube to the magnetic stand for 3 minutes.

12. Transfer the supernatant containing the purified DNA to a clean 1.5ml centrifuge tube.

【Manual Protocol for 600ul】

1. Add 40µl Proteinase K and 0.6ml plasma/serum to a sterile 2.0ml centrifuge tube.
2. **Add 0.9ml Buffer MLB and 40µl MagBind Particles to the sample, vortex for 1 minutes. Incubate at room temperature for 10 min with occasional mix by inverting.** MagBind Particles adsorb large fragments in this step, discard the MagBind Particles in this step.
3. Place the tube to the magnetic stand for 10 minutes until the beads have formed a tight pellet.
4. Transfer the supernatant to a new 2.0ml centrifuge tube. **Add 0.3ml Buffer GDP and 40µl MagPure Particles F to sample,** Inverting to Mix for 10 minutes.
5. Place the tube to the magnetic stand for 1 minutes until the beads have formed a tight pellet. Then remove the supernatant.
6. **Add 600µl Buffer MAW1** and vortex for 15 seconds to re-suspend beads. Place the tube to the magnetic stand for 1 minute until the beads have form a tight pellet. Then remove the supernatant.
7. **Add 600µl Buffer MW2,** and vortex for 15 seconds to re-suspend beads. Place the tube to the magnetic stand for 1 minute until the beads have form a tight pellet. Then remove the supernatant.
8. **Add 600µl Buffer MW2,** and vortex for 15 seconds to re-suspend beads. Place the tube to the magnetic stand for 1 minute until the beads have form a tight pellet. Then remove the supernatant.
9. Centrifuge shortly to collect liquid on the tube. Place the tube to the magnetic stand and remove all the liquid carefully.
10. Air dry for 10 minutes.
11. Add 30~50µl Elution Buffer/Low TE/Sterile Water and re-suspend the beads by vortex. Sit at room temperature for 5 minutes. Shake 1~2 times to dissolve DNA from magnetic particles more efficiently. Place the tube to the magnetic stand for 3 minutes.
12. Transfer the supernatant containing the purified DNA to a clean 1.5ml centrifuge tube.

Auto Purify by KingFisher Flex for 300µl Sample

1. Add the Reagents/sample to the wells of the deep well plate according to the table below.

Name of the Plate	Pre-loaded reagents	Addition before use
Sample Plate A	450µl Buffer MLB	300µl Plasma 20µl Proteinase K 20µl MagBind Partilces
Wash Plate 1	600µl Buffer MAW1, Put in 96 magnetic Tip 20µl MagPure Particle F	
Wash Plate 2	600µl Buffer MW2	
Wash Plate 3	600µl Buffer MW2	
Large Elute	50µl Elution Buffer	
Elution plate	50µl Elution Buffer	

2. Turn on the machine, start the corresponding program.
3. **Add 150µl Buffer GDP to the Sample plate during the dispense step.**
4. Place the sample plate back into the instrument and press Start.
5. After the run is completed, remove the plates and store the purified total DNA.