7040 Virginia Manor Road
Beltsville, MD 20705, USA
Web: www.abpbio.com
Email: info@abpbio.com

EasyMag DNA Purification Kit from animal tissue

Catalog Number: D114-1, D114-2

Table 1. Kit Components and Storage

Kit Component	D114-1 (50 preps)	D114-2 (200 preps)	Storage	Stability
Buffer ATL	15 mL	60 mL	RT	The product is stable for one year when stored as directed.
Buffer DL	15 mL	60 mL	RT	
Buffer AW1*	15 mL	53 mL	RT	
Buffer AW2*	15 mL	2×25 mL	RT	
Buffer AE	15 mL	30 mL	RT	
Proteinase K (20 mg/mL)	1 mL	4×1 mL	-20℃	
MagBinding Beads	2.5 mL	10 mL	2-8 °C	

***** Prior to use, add absolute ethanol to **Buffer AW1, Buffer AW2** according to the bottle label.

Product Description

EasyMag DNA Purification Kit from animal tissue provides a rapid and reliable isolation of high-quality genomic DNA from fresh or frozen animal tissue samples. This kit can be processed manually or on an automated platform. This system combines the reversible nucleic acid binding properties of paramagnetic particles with the time-proven efficiency of ABP's buffer chemistries to provide a fast and convenient method to isolate DNA from a variety of samples. Phenol/chloroform extraction, and time-consuming steps such as precipitation with isopropanol or ethanol have been eliminated. Utilizing paramagnetic particles provides high-quality DNA that is suitable for direct use in most downstream applications such as Next Generation Sequencing, qPCR, PCR, and microarrays.

Features

- ❖ Fast – DNA purification process in less than 20 min post-lysis.
- ❖ Safe – No Phenol/chloroform extractions.
- ❖ High-quality – DNA is suitable for a variety of downstream applications.

Purification Protocol

1. Weigh ~25 mg tissue (or <10 mg liver, lung, or spleen tissue), and cut the tissue into small pieces, then transfer it into a 1.5 mL microcentrifuge tube.

   Note: Using too much tissue sample may reduce yield and purity. Spleen, liver, and kidney tissues are rich in DNA, and should be used no more than 10 mg. Muscle and skin tissues can be used at 30 mg. Liquid nitrogen grinding, mechanical homogenizer, or glass homogenizer can be used to homogenize tissue sample to reduce digestion time.

2. Add 230 µL Buffer ATL and 20 µL Proteinase K solution. Mix thoroughly by vortexing.
3. Incubate at 55°C for 1-3 hr or overnight. Vortex briefly every 20-30 min during incubation, or incubate with shaking.

Note: Lysis time depends on the amount and type of tissue used. The average time is 0.5-3 hr, and mouse tails may take 6-8 hr. Lysis can be proceed overnight.

4. (*Optional*) If RNA-free genomic DNA is required, add 20 µL RNase A (10 mg/mL). Mix by vortexing, and incubate at RT for 15 min.
5. Centrifuge at 12,000 x g for 3 min. Transfer the supernatant to a new 1.5 mL microcentrifuge tube.
6. Add 250 µL Buffer DL. Mix thoroughly by vortexing. Incubate at 70°C for 10 min.

   Note: Buffer DL may be precipitated during storage, if happen, heat it at 50^{o}C to dissolve. A precipitate may form upon the addition of Buffer DL. This precipitate does not interfere with DNA recovery.
7. Add 250 µL 100% isopropanol and 50 µL MagBinding Beads. Mix thoroughly by vortexing.

   Note: A precipitate may form upon the addition of isopropanol when processing liver or spleen tissue. Pipette up and down 5-10 times to break up the sediment.
8. Incubate with shaking at RT for 5 min.
9. Transfer the tube to a magnetic rack for an additional 2 min or until beads pellet and supernatant is cleared. With the tube on the magnetic rack remove the supernatant and discard.
10. Add 600 µL of Buffer AW1 to the tube. Resuspend the beads by pipetting up and down or vortexing at 1,500 rpm for 30 seconds. Replace the tube on the magnetic rack for 2 min or until beads pellet. Remove and discard the supernatant.

    Note: Buffer AW1 must be diluted with absolute ethanol according to the bottle label before use.
11. Add 600 µL of Buffer AW2 to the tube. Resuspend the beads by pipetting up and down or vortexing at 1,500 rpm for 30 seconds. Replace the tube on the magnetic rack for 2 min or until beads pellet. Remove and discard the supernatant.

    Note: Buffer AW2 must be diluted with absolute ethanol according to the bottle label before use.
12. Repeat step 11 for a second wash step.
13. Leave the tube on the magnetic rack, open the cap and air dry the beads at RT for 10-15 min.

    Note: This step is critical for removing of trace ethanol that may interfere with downstream applications.
14. Add 100 µL Buffer AE to the tube, resuspend the beads by vortexing. Incubate at 55ºC for 5 min, vortex for 15 seconds every 1-2 min for 5 min.
15. Replace the tube on the magnetic rack for 2 min or until beads pellet. Transfer the cleared supernatant to a clean tube.
16. Store the DNA at -20ºC.

Troubleshooting

Problem	Possible cause and suggestions
Low yield	• Incomplete homogenization: Completely homogenize sample. • Improper washing: Buffer AW1, buffer AW2 must be diluted with absolute ethanol before use. • Sample has low DNA content: Increase starting material and volume of all reagents proportionally. • Incomplete resuspension of MagBinding Beads: Resuspend MagBinding Beads by vortexing virgously before use. • Loss of MagBinding Beads during operation: Avoid disturbing MagBinding Beads during aspiration. • DNA remains bound to MagBinding Beads: Increase elution volume

	and incubation time.
Poor performance in downstream applications	• Salt contamination: Remove any residual liquid with a pipettor after each wash step. • Ethanol contamination: Ensure the beads are completely dried before elution.
RNA contamination	• Sample is rich in RNA: Increase RNase digestion time to 60 min.