Note: vortex per 10s will facilitate lysis.

4. Centrifuge 3min at 12,000g. Carefully transfer the supernatant into a new tube.

Add 4µL RNase A (Cat. # BSA40S1). Incubate at room temperature for 5min.

- 5. Add 200µL Lysis B buffer, mix thoroughly.
- 6. Add 200µL absolute ethanol, mix thoroughly.

### **■** Blood spots

- 1. Take 2-5 pieces spots  $(3\times3mm)$  into a tube.
- 2. Add 300μL TES Buffer and 10μL of PK solution into the tube. Mix intensively for 10s. Put it in 56°C thermostat oscillator metal bath, vortex for 60min at 900rpm.
- 3. Centrifuge for 3min at 12,000g. Carefully transfer the supernatant into a new tube.

Add 4µL RNase A (Cat. # BSA40S1). Incubate at room temperature for 5min.

- 4. Add 300μL Lysis B buffer, mix thoroughly.
- 5. Add 150µL absolute ethanol, mix thoroughly.

#### > DNA Purification

- 1. Transfer all the mixture ( $\leq$ 750 $\mu$ L) to spin column. Centrifuge the mixture at 12,000×g for 1 minute. Discard flow-through.
- 2. Add  $500\mu L$  PW buffer to the Spin column. Centrifuge the spin column at  $12,000\times g$  for 1 minute. Discard flow-through.
- 3. Add  $500\mu L$  Wash buffer to the spin column. Centrifuge at  $12,000 \times g$  for 1 minute. Discard flow-through. Take the spin column back to the tube.
- 4. Repeat step 3
- 5. Centrifuge the spin column at  $12,000 \times g$  for 2 minute.
- 6. Place the spin column in a new 1.5 or 2.0mL micro centrifuge tube. Add  $30\text{-}100\mu\text{L}$  of the Elution Buffer. Incubate at room temperature for 2 minutes. Centrifuge the mixture at  $10,000\times\text{g}$  for 1 minute. The DNA in the collection tube is ready for further analysis. If the isolated DNA sample is not going to be tested on the same day, freeze it at -20°C.

# 【Company Information】

Manufacturer: Hangzhou Bioer Technology Co., Ltd

Address: No.1192 Bin'An Rd, Binjiang District, Hangzhou, Zhejiang Province, China

Tel: 0571-87774567 Fax: 0571-87774553

Web: www.bioer.com.cn

Zip Code: 310053

Aftersales Service Provider: Hangzhou Bioer Technology Co.,Ltd

# Biospin Blood/Cell/Tissue Genomic DNA Extraction Kit

#### TECHNICAL SUPPORT:

For technical support, please dial phone number : 0086-571-87774567-5215 or 5211, or fax to 0086-571-87774553 Email to reagent@bioer.com.cn.

Website: www.bioer.com.cn

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### Kit Components

Cat#	BSC47S1	BSC47M1
Components	50 Tests	100 Tests
TES buffer	15mL	30mL
LysisB buffer	25mL	50mL
PK solution	0.5mL	1mL
PW Buffer	12mL (add 18mL absolute ethanol)	24mL (add 36mL absolute ethanol)
Wash Buffer	24mL (add 36mL absolute ethanol)	48mL (add 72mL absolute ethanol)
Elution Buffer	5mL	10mL
Spin Columns	50	100
Handbook	1	1

### **Storage & shipping conditions**

- 1. Store Protease K at 2~8°C, store all other components at room temperature.
- 2. All components are stable for 18 months from date of receipt under proper storage condition.

### [Introduction]

The kit provides a simple, fast and cost effective method that can isolate high quality DNA. Using one simple protocol, high yield purified DNA can be isolated from various samples including animal tissue, whole blood, buffy coat, leukocytes, and cultured cells. The kit utilizes silica-based membrane techniques within a convenient spin column, no need of expensive resins, toxic phenol-chloroform extractions, or time-consuming alcohol precipitation. The whole procedure takes less than 30 minutes including lysis steps, and more than 20 kb DNA could be purified by this kit. Isolated DNA can be directly applied to PCR, Southern blotting and other enzymatic reactions.

# 【Typical genomic DNA yields from various samples】

Source	Quantity	Yield (μg)
Mammalian tissue	25mg	5-30
Mammalian blood	200μL	2-10
Poultry blood / Amphibian	5-20μL	5-40
Cell	≤ 2×10 <sup>6</sup>	5-30
Blood spots	2-5 pieces	0.1-1

## 【Additional apparatus and materials required but not supplied】

## [Important notes]

Please add absolute ethanol to PW Buffer and Wash Buffer, mix thoroughly before use.

#### [Protocol]

**➤** Sample lysis

#### ■ Cell and blood

1. Sample pre-processing

A. **Culture cells**: adherent cells shall be digested with trypsin, no more than 107cells. Spin for 5min in 1500rpm, discard the supernate. Add  $200\mu L$  PBS buffer into the tube, re-suspend cells.

**B. Leukocytes**: if blood volume is more than  $200\mu L$ , use Red Blood Cell Lysis Buffer (Cat. # BSA06M1 or Cat. # BSA07M1) first to obtain white blood cells, add  $200\mu L$  PBS buffer into the tube, re-suspend cells.

**C. Mammalian Whole blood**: Add  $200\mu L$  well mixed blood into 1.5 mL tube (if the blood is less than  $200\mu L$ , add to  $200\mu L$  with PBS buffer).

**D. Birds, amphibians, anticoagulant whole blood:** Add 5-20 $\mu$ L well mixed blood into 1.5mL tube, add to 200 $\mu$ L with PBS buffer.

2. Add 10μL PK solution, mix thoroughly for 15s.

Add  $4\mu L$  RNase A (Cat. # BSA40S1). Incubate at room temperature for 5min.

- 3. Add 200 $\mu$ L Lysis B buffer, mix thoroughly. Incubate at 56°C for 10min.
- 4. Add 200μL absolute ethanol, mix thoroughly.

#### ■ Animal tissue

- 1. Add 200μL TES Buffer and 10μL PK solution into tube.
- 2. Grind tissue to powder (no more than 25mg) with liquid nitrogen. Add it to the tube and mix thoroughly.

**Note:** for spleen, no more than 10mg, samples must be submerged within solution.

3. Incubate for 1-4 hours at 56°C. For samples hard to lysis, prolong lysis time or even overnight.

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