

# MagaBio FFPE Tissue Genomic DNA Extraction Kit

## Kit Components

Cat#	BSC31S1E	BSC31S1	BSC31M1
<b>Components</b>	<b>32 Tests</b>	<b>50 Tests</b>	<b>100 Tests</b>
PK Solution	640 $\mu$ L	1 mL	2 mL
Deparaffinization Solution	32 mL	50 mL	50 mL $\times$ 2
Lysis Buffer	6.4 mL	10 mL	20 mL
Binding Buffer	96 Well pre-packed plate 2 pieces	11mL (add 13.75mL ethanol before use)	22mL (add 27.5mL ethanol before use)
PW Buffer		13mL (add 19.5mL ethanol before use)	26mL (add 39mL ethanol before use)
Wash Buffer		12mL $\times$ 2 (add 18mL ethanol before use)	24mL $\times$ 2 (add 36mL ethanol before use)
Elution Buffer		25 mL	50 mL
MagaBio Reagent		0.5 mL	1 mL
Handbook		1 copy	1copy

## Storage

1. The kit can be transported at room temperature.
2. All reagents are stored at 2-8°C.
3. All reagents, when stored properly, are stable for 12 months from the time of delivery.

## Introduction

This kit used to extract high—purity DNA from FFPE tissue sections, with non-toxic deparaffinization solution , high-performance lysis buffer to release DNA from FFPE efficiently .MagaBio sample processing is based on proprietary magnetizable particles--MagaBio. The pure DNA can be applied extensively in PCR, sequencing, Southern hybridization, mutant analysis, SNP and the others.

According to the special interaction, use MagaBio nucleic acid separation system with a general protocol---sample processing, MagaBio adsorption, washing and elution, and can go high-throughput.

## Principle and Advantage

DNA in the sample is released by PK Solution and Lysis Buffer. Released DNA is bound exclusively and specifically to the magnetic beads in presence of a Binding Buffer. The DNA bound to magnetic particles is captured by magnetic material; contaminants will be removed by washing with Wash Buffer once or more. The DNA is then eluted from the particles with Elution Buffer or molecular grade water.

## Apparatus and materials to be prepared by the user

1. Magnetic Rack or Bioer NPA-32P purification instrument
2. Water bath or Dry bath
3. Vortex mixer
4. Absolute alcohol (For BSC31S1 and BSC31M1)

## Protocol

### 1. Sample processing

- a) FFPE sections with a thickness of up to 10  $\mu$ m. Up to 8 sections, each with a thickness of up to 10  $\mu$ m and a surface area of up to 5 $\times$ 5 mm, placed in a 1.5mL microcentrifuge tube.
- b) Paraffin-embedded blocks: Use a sterile scalpel to cut off the paraffin surface in contact with air, scrape up to

30mg of sample, Avoid paraffin as possible, placed in a 1.5mL microcentrifuge tube.

c) Formalin fixed tissue samples: Cut the sample to small pieces, placed in a 1.5mL microcentrifuge tube. Add 1mL 10mM pH7.0~7.4 PBS or physiological saline, mix with vortex, centrifuge at full speed for 1 min, Remove the supernatant by pipetting. Repeat this step again, and then start step 5. 2. Add 1mL Deparaffinization Solution to the 1.5mL tube, close the tube and vortex 10s, place the tube in heating block or water bath at 56°C for 3 min.

3. Centrifuge at 14,000g for 2 min, And remove the supernatant by pipetting.

4. Add 1m ethanol to the tube, and mix 10s by vortexing. Centrifuge at 14,000g for 2 min, Remove the supernatant by pipetting. Open the tube and incubate at room temperature or up to 37 °C. Incubate for 10 min or until all residual ethanol has evaporated.

5. Add 200µL Lysis Buffer I, 20µL PK Solution to the tube, mix 10s by vortexing, Briefly centrifuge the 1.5 mL tube. Then Incubate at 56 °C for 1 h or until the sample has been completely lysed (may overnight).

6. Incubate at 90 °C for 1 h.

7. Optional: If RNA free is required, add 2 µL RNase A (100 mg/mL) mix completely and incubate for 2 min at room temperature, then start next step.

8. Centrifuge at 10,000g for 1min. Add 450 µL of the Binding Buffer followed by 10µL of the well-mixed (particles are uniformly suspended) MagaBio Reagent.

9. Mix the tube gently and incubate for 10 minutes at room temperature, while mixing.

Note: using an end-over-end rotator or manual mixing every 2-3 minutes.

10. Sediment the MagaBio DNA bound particles using a magnetic rack. Discard the supernate, remove the tube from the magnetic rack and wash the particles as described in below.

11. Add 500 µL of PW Buffer to the tube from the above. Mix well by inverting the tube several times to ensure the particles are completely dispersed. Sediment the particles on the magnetic rack and aspirate the supernate.

12. Add 500 µL of Wash Buffer to the tube from the above. Mix well by inverting the tube several times to ensure the particles are completely dispersed. Sediment the particles on the magnetic rack and aspirate the supernate.

13. Remove the tube from the magnetic rack and repeat washing once more following the above step.

14. Add 100~200µL of Elution Buffer and mix for 10 minutes.

**Note:** Vortex gently every 2-3 minutes.

Sediment the particles on the magnetic rack and carefully transfer the supernatant containing the isolated DNA into a clean tube. The material is ready for further analysis. If the isolated DNA sample is not going to be tested on the same day, freeze at -20°C until the time of analysis.

### Analysis Nucleic Acid

Get some DNA, diluted in a advisable factor with elution buffer.

Survey the OD260, OD280 and OD320.

Concentration (µg/mL) = 50 × OD260 × dilution fact

$2.1 \geq \text{OD}260\text{-}320 / \text{OD}280\text{-}320 \geq 1.7$

Notice:  $1.0 \geq \text{OD}260 \geq 0.1$ , the result of ratio is much reliable.

### The automation purification

With semi-automation machine, the kit is deeply suitable for several samples, which supply a really platform of automation or streamLine protocol and achieve high-throughput and high-speed but effective purification. An example for applying the kit on our product NPA-32+:

1. Reagent prepare

#### 1) For BSC31S1 and BSC31M1

Join 450 µL Binding Buffer to the 2.2mL 96 Deep Well column 1、7, 500µL PW Buffer to the 96 Deep Well column 2、8, 500µL Wash Buffer to the 96 Deep Well column 3、9 and to the 96 Deep Well column 4、10, 100µL Elution Buffer to the 96 Deep Well column 5、11, 190µL pure water and 10µL MagaBio Reagent to the 96 Deep Well column 6、12.

#### 2) For BSC31S1E

Shake 96-well plate upside down for three times after placing under room temperature, then rip off plastic film, centrifuge in 96-well centrifuge for a couple of seconds(or swing by hand) to avoid adhered liquid. Rip off aluminum foil film of 96 well plates; identify the direction of the plate (magnetic beads in column 6th&12th).

2. The sample lysis processing of samples are the same with the manual extraction (**Protocol:1-7**) .

3. Join the lysis product to the 96 Deep Well column 1、 7.

4. Place 96 Deep Well to the instrument, then plug in 8-strip Tip and run the program.

Step	Well	Name	Waiting Time (min : ss)	Mixing Time (min : ss)	Magnet Time (min : ss)	Adsorption	Speed	Volume (μL)
1	1	Mixing	0 : 0	0 : 30	0 : 0		F	650
2	6	Beads	0 : 0	0 : 15	0 : 30		M	200
3	1	Binding	0 : 0	10 : 0	0 : 35	Strong	F	650
4	2	Wash 1	0 : 0	3 : 0	0 : 30	Strong	F	500
5	3	Wash 2	0 : 0	2 : 0	0 : 30	Strong	F	500
6	4	Wash 3	0 : 0	2 : 0	0 : 30	Strong	F	500
7	5	Elution	2 : 0	5 : 0	0 : 30		M	100
8	6	Discard	0 : 0	0 : 30	0 : 0		M	200

**Elution temperature: 60°C, Elution start heating step: 7th.**

#### Important Notes

1. Please add absolute ethanol to Wash Buffer and mix thoroughly before the first use.
2. Typically, >85% of the DNA is recovered in the first elution. If desired, more DNA can be recovered by applying a second elution.
3. The procedure above is just suitable for the NPA-32P nucleic acid purification instrument .Adjust the running programs according to the different instrument.
4. All reagents should be stored at 2-8°C.

#### Company Information

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