High-Q™-Spin-Column Tissue Genomic DNA Purification Kit

Ordering info

TBK0162. 3 reactions (sample) TBK0163. 50 reactions TBK0164. 200 reactions

Description

High-Q™-Spin-Column Tissue Genomic DNA Purification Kit is an easy silica-membrane-based system for genomic DNA purification from a wide variety of tissue samples. An optimized lysis buffer guarantees a good yield while the use of High-Q™ Spin Columns allow a good quality DNA, suitable for downstream applications.

Features

- Starting material up to 100 mg of tissue sample.
- Typical yields are 0.5- 50 µg of DNA depending on the material tissue used.
- No organic extraction, no ethanol precipitation.
- High DNA purity, the isolated DNA is ready to use for downstream molecular biology applications.
- Easy and Fast protocol.

Applications

- Purification of DNA from animal and human tissue.
- Purification of DNA tissue using different starting materials: frozen, fresh or stabilized with Tiaris™ Tissue Protect (TBB0431).
- DNA obtained is suitable for downstream molecular biology applications such as PCR, enzymatic digestion for cloning or Southern, genotyping, etc.

Quality Control

DNA purified is checked by: integrity (agarose gel electrophoresis), quantity and quality (A260/280= 1.9 ± 0.2).

Kit Components

Components	TBK0163	TBK0164
High-Q™ Spin Column with Collection Tubes	50	200
Tissue-1 Buffer	30 mL	110 mL
Tissue-2 Buffer	12 mL[a]	40 mL[b]
WB2 Buffer	12 mL[c]	45 mL[d]
Elution Buffer	15 mL	25 mL
Proteinase K	2x 30 mg [e]	6x 30 mg [e]
Proteinase K Resuspension Buffer	2x 1.5 mL	6x 1.5 mL
RNase A	15 mg [f]	3 x 15 mg [f]
RNase A Resuspension Buffer	1 x 1.5 mL	3 x 1.5 mL

Order Info Kit Components: *High-Q™ Spin Column with Collection Tubes (TBM0010) | Tissue-1 Buffer (TBB0579) | Tissue-2 Buffer (TBB0580) | WB2 Buffer (TBB0512) | Elution Buffer (TBB0510) | Proteinase K (TBZ0303) | Proteinase K Resuspension Buffer (TBB0546) | RNase A (TBZ0318) | RNase A Resuspension Buffer (TBB0309).*

¡Components for samples are ready to use!

Before its use:

[a] Add 18 mL isopropanol and mix well.

[b] Add 60 mL isopropanol and mix well.

[c] Add 48 mL absolute ethanol and mix well

[d] Add 180 mL absolute ethanol and mix well.

[e] Add 1.5 mL Proteinase K Resuspension Buffer and mix well. Store at -20°C.

[f] Add 1.5 mL RNase A Resuspension Buffer and mix well. Store at -20°C.

Storage

Store the kit at 25°C.

Store Proteinase K and RNase solutions at -20°C.

Material required (not supplied)

- 1.5 mL Tubes.
- Ethanol (CAS 64-17-5).
- Isopropanol (CAS 67-63-0).

PROTOCOL

1. Grind up to 50-100 mg of tissue material in liquid nitrogen using a mortar and a pestle. With a freeze spatula, collect the powder into a 1.5 mL tube. Commercially available equipment for homogenization also can be used.
2. Add **500 µL Tisuue-1 Buffer** and mix by vortex briefly.
3. Add **40 µL Proteinase K (20 mg/mL)**. Mix well.
4. Incubate at 65°C until the tissue is completely lysed. Lysis is usually complete in 1-3 h. Some samples as rodent tail and ears should be lysed overnight without affect the procedure results. Mix 2-3 times by inversion during incubation.
5. Centrifuge at 13,000 g for 5 minutes. Transfer the supernatant to a fresh tube.
6. *[Optional] Add **20 µL RNase-A 20 µg/ µL** and incubate for 5 minutes at room temperature.*
7. Add **400 µL Tissue-2 Buffer** and mix by inversion.

   Check isopropanol has been added to Tissue-2 Buffer (✓).
8. Transfer up **700 µL mixture** to a High-Q™ Spin Column placed into a Collection Tube.
9. Centrifuge at 10,000 g for 1 minute. Remove the flow-through and place back the High-Q™ Spin Column into a Collection Tube. Repeat steps 8 and 9 with the remaining mixture.
10. Add **500 µL WB2 Buffer**.

    Check absolute ethanol has been added to WB2 Buffer (✓).
11. Centrifuge at 10,000 g for 30 seconds. Discard the flow-through and place back the High-Q™ Spin Column into a Collection Tube. Repeat steps 10 and 11 one more time.
12. To dry High-Q™ Spin Column and eliminate residual ethanol, centrifuge again at 13,000 g for 1 minute.
13. Place the High-Q™ Spin Column into a clean 1.5 mL Tube. Add **50-100 µL prewarmed Elution Buffer** or **Water (Molecular Biology Grade)**.

    Prewarm Elution Buffer or Water at 70°C.
14. Incubate at room temperature, 2 minutes. Centrifuge at 13,000 g for 1 minute to elute purified DNA.
15. Check DNA quantity by spectrophotometry and quality on agarose electrophoresis gel.
16. Store DNA at -20°C.