HigherPurity™ Blood & Cell culture DNA Isolation Kit

Cat. No: AN0044 (50 reactions)
Cat. No: AN0045 (100 reactions)

Description
HigherPurity™ Blood & Cell culture DNA Isolation Kit is a simple and rapid method for high-quality genomic DNA purification from various sources, including: whole blood, plasma, serum, buffy coat and cell culture.
The kit is based in DNA ability to bind silica in the presence of high concentrations of chaotropic salts.

Features
- **Safe**: no phenol-chloroform extraction.
- **Efficient**: 3-6 µg of genomic DNA from a 200 µl blood sample or 15–20 µg from Cultured Cells (5 x 10^6).
- **Ready to use** genomic DNA, for all molecular biology applications.

Applications
- gDNA purified with the kit is suitable for direct use in all common molecular biology applications: PCR, cloning, DNA sequencing, Southern blot analysis, etc.

Quality Certifications
HigherPurity™ Blood & Cell culture DNA Isolation Kit is tested on a lot-to-lot basis by isolating total DNA from 200µl of whole human blood. DNA purified is analysed by:
- Spectrophotometer: Ratio 260/ 280 (1.6-1.8)
- Agarose gel electrophoresis.

Kit Components

<table>
<thead>
<tr>
<th>Item</th>
<th>AN0044</th>
<th>AN0045</th>
</tr>
</thead>
<tbody>
<tr>
<td>Minispin columns</td>
<td>50</td>
<td>100</td>
</tr>
<tr>
<td>Collection tubes (2 mL)</td>
<td>50</td>
<td>100</td>
</tr>
<tr>
<td>BLY Buffer</td>
<td>20 ml</td>
<td>40 ml</td>
</tr>
<tr>
<td>Proteinase K*</td>
<td>15 mg</td>
<td>30 mg</td>
</tr>
<tr>
<td>WB1 Buffer</td>
<td>30 ml</td>
<td>60 ml</td>
</tr>
<tr>
<td>WB2 Buffer**</td>
<td>6 ml</td>
<td>2X6 ml</td>
</tr>
<tr>
<td>EB Buffer</td>
<td>10 ml</td>
<td>10 ml</td>
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</tbody>
</table>

Kit Storage:
Store the kit at room temperature. If any kit reagent forms a precipitate, warm at 55–65 °C until the precipitate dissolves, and allow to cool to room temperature before use.

*Dissolve Proteinase K in water (1.5 ml) to obtain a 20 mg/mL stock solution. The Proteinase K solution can be stored for several days at 2–8 °C. For longer-term storage, the unused portion of the solution may be stored in aliquots at –20 °C until needed. This product as supplied is stable at room temperature.

**Add the volume ethanol (96%-100%) specified [Not included] to WB2 Buffer prior to initial use (see the label on the bottle for a volume indication). After ethanol has been added, mark the bottle to indicate that this step has been completed.

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PROTOCOL A: DNA Purification from Blood

This protocol is for purification of total DNA from whole blood, plasma, serum or buffy coat.

1. Transfer **15µL proteinase K** into the bottom of a 1.5 ml microcentrifuge tube (not provided).
2. Add **200 µL** of sample.
   
   *Use up to 200 µl whole blood, plasma, serum or buffy coat. If the sample volume is less than 200 µl, add the appropriate volume of PBS.*
3. [Optional Step] RNA Degradation:
   
   If RNA-free gDNA is required, add **4 µl of RNase A (100 mg/ml)** [not provided].
4. Add **200 µL** of buffer BLY and mix by vortexing (it is important to observe a homogeneous solution).
5. Incubate in a water bath at 55 °C for 10 minutes.
6. Add **200 µl** of ethanol (96–100%) and mix by vortexing vigorously.
7. Transfer the mix to the minispin column by pipetting and centrifuge at 8000 rpm for 1 minute. Discard the flow-through solution.
8. Place the minispin column in a collection tube and add **500 µL** of WB1 buffer. Centrifuge at 8000 rpm for 1 minute. Discard the flow-through solution.
9. Place the minispin column in a collection tube and add **500 µL** of WB2 buffer. Centrifuge at 8000 rpm for 3 minutes. Discard the flow-through solution.
10. Centrifuge at full speed for 1 minute to dry the minispin column.
   
   *This step will avoid the residual liquid to inhibit subsequent enzymatic reactions.*
11. Place the minispin column into a new, labelled, 1.5 microcentrifuge tube (not provided) and pipet **50-100µL EB Buffer** or **pre-warm water** directly into the membrane. Close the tube and incubate for 1 minute at room temperature.
12. Centrifuge at full speed for 1 minute to elute the DNA
13. The purified genomic DNA can be stored at 2-8°C for a few days. For longer term storage, -20°C is recommended.

PROTOCOL B: DNA Purification from Cultured Cells (5 x 10⁶ cells)

1. Transfer the appropriate number of cells (5 x 10⁶ cells) to a 1.5 ml microcentrifuge tube.
2. Centrifuge for 5 minutes at 3000 rpm to pellet the cells.
3. Discard the supernatant.
4. Resuspend cell pellet in **PBS** to a final volume of **200 µl**.
5. Add **15 µl** proteinase K
6. Continue with step 3 of “Protocol A: DNA Purification from Blood”

PRODUCT USE LIMITATION

This product is developed, designed and sold exclusively for research purposes and in vitro use only. The product was not tested for use in diagnostics or for drug development, nor is it suitable for administration to humans or animals. Please refer to [www.canvaxbiotech.com](http://www.canvaxbiotech.com) for Material Safety Data Sheet of the product.