

High Pure PCR Template Preparation Kit

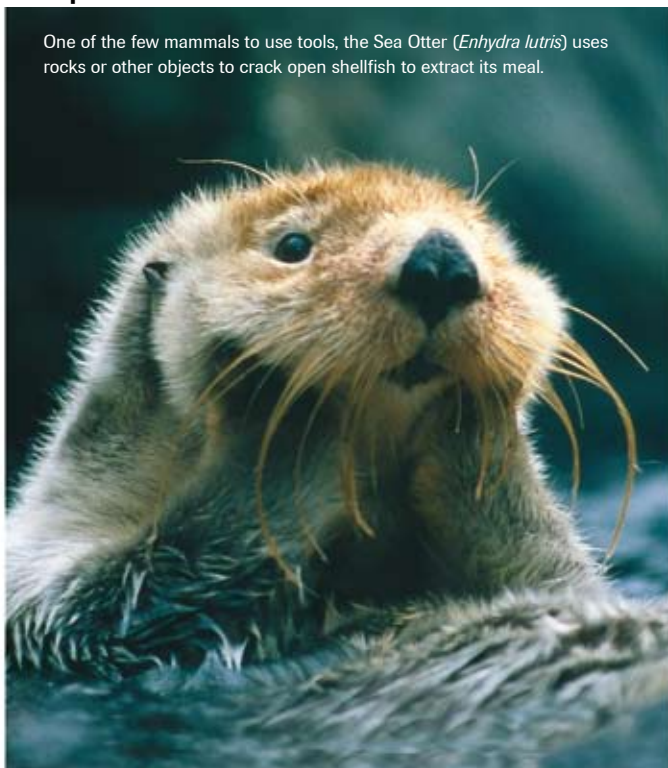
Rapidly purify genomic DNA for diverse applications

Choose the versatile **High Pure PCR Template Preparation Kit** to rapidly and easily isolate genomic DNA from a wide variety of research sample materials, including: Whole blood, buffy coat, cultured cells, clinical research samples (e.g., sputum, feces, bronchoalveolar lavage [BAL], urine, and swabs), animal tissue, mouse tail, yeast, formalin-fixed, paraffin-embedded (FFPE) tissue sections, and environmental water samples.

Produce multiple PCR templates in minutes using efficient High Pure spin columns. Generate high-purity DNA that improves reproducibility and reliability in applications such as standard or long template PCR, qPCR, SNP detection, microarray analysis, and Southern blotting.

Experts at Extraction

One of the few mammals to use tools, the Sea Otter (*Enhydra lutris*) uses rocks or other objects to crack open shellfish to extract its meal.



■ Maximize flexibility.

Use a single versatile kit to purify DNA from a broad range of sample materials.

■ Facilitate long template applications.

Efficiently purify high molecular weight DNA (30-50 kb) (Figure 1).

■ Improve assay results.

Ensure optimal performance by efficiently removing contaminants that can interfere with PCR or other downstream applications.

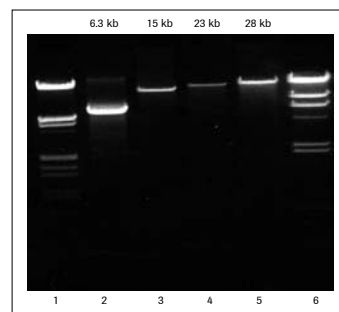


Figure 1: PCR amplification of different template sizes from nucleic acid samples prepared using the High Pure PCR Template Preparation Kit.

Nucleic acids were prepared from a human whole blood research sample or cultured human K-562 cells. A portion (250 ng) of each preparation was amplified using a tissue plasminogen activator (tPA)-specific primer and the Expand

Long Template PCR System or the Expand 20 kb^{PLUS} PCR System following pack insert instructions. The amplicons obtained were:

- Lane 2:** 6.3 kb: obtained from blood and amplified using Expand Long Template PCR Buffer 1
 - Lane 3:** 15 kb: obtained from blood and amplified using Expand Long Template PCR Buffer 3
 - Lane 4:** 23 kb: obtained from blood and amplified using Expand Long Template PCR Buffer 3
 - Lane 5:** 28 kb: obtained from K-562 cells and amplified using the Expand 20 kb^{PLUS} PCR System
- Result:** All samples yielded a distinct, specific band of the expected size, even as large as 28 kb, from genomic DNA prepared with the High Pure PCR Template Preparation Kit.

Efficiently purify DNA

Procedure

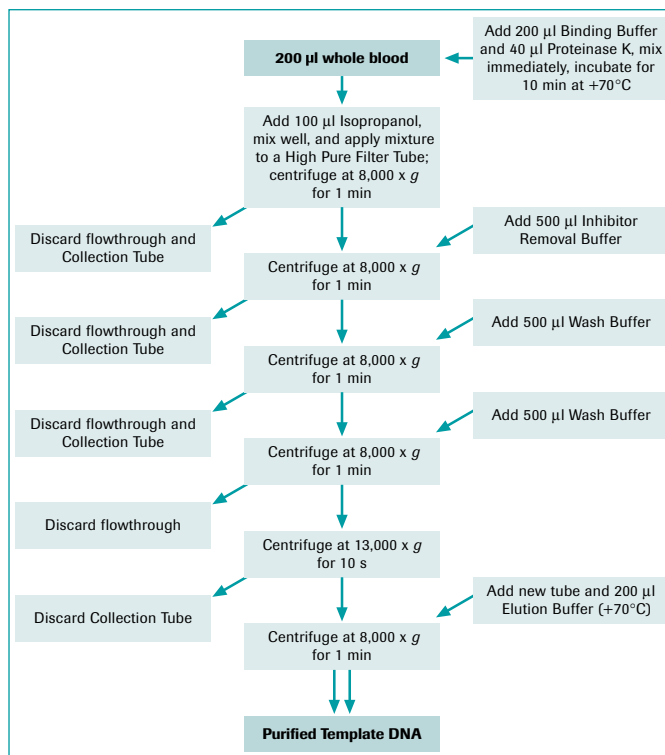
Protocol for isolating DNA from 200 µl mammalian blood, buffy coat, or cultured mammalian cells.

View detailed procedures for other sample materials in the pack insert at www.roche-applied-science.com

Note: Before starting the purification, warm the Elution Buffer to +70°C.

- 1 To a nuclease-free 1.5 ml microcentrifuge tube:
 - Add 200 µl of sample material.
 - Add 200 µl Binding Buffer.
 - Add 40 µl Proteinase K (reconstituted).
 - Mix immediately and incubate at +70°C for 10 min.
- 2 Add 100 µl Isopropanol and mix well.
- 3
 - Insert one High Pure Filter Tube into one Collection Tube.
 - Pipet the sample into the upper buffer reservoir of the Filter Tube.
 - Insert the entire High Pure Filter Tube assembly into a standard table-top centrifuge.
 - Centrifuge 1 min at 8,000 × *g*.
- 4 After centrifugation:
 - Remove the Filter Tube from the Collection Tube; discard the flowthrough and the Collection Tube.
 - Combine the Filter Tube with a new Collection Tube.
 - Add 500 µl Inhibitor Removal Buffer to the upper reservoir of the Filter Tube.
 - Centrifuge 1 min at 8,000 × *g*.
- 5
 - Remove the Filter Tube from the Collection Tube; discard the flowthrough and the Collection Tube.
 - Combine the Filter Tube with a new Collection Tube.
 - Add 500 µl Wash Buffer to the upper reservoir of the Filter Tube.
 - Centrifuge 1 min at 8,000 × *g* and discard the flowthrough.
- 6
 - Remove the Filter Tube from the Collection Tube; discard the flowthrough and the Collection Tube.
 - Combine the Filter Tube with a new Collection Tube.
 - Add 500 µl Wash Buffer to the upper reservoir of the Filter Tube.
 - Centrifuge 1 min at 8,000 × *g* and discard the flowthrough.
- 7 After discarding the flowthrough:
 - Centrifuge the entire High Pure assembly for an additional 10 s at full speed.
 - ⚠ The extra centrifugation time ensures removal of residual Wash Buffer.
 - Discard the Collection Tube.
- 8 To elute the DNA:
 - Insert the Filter Tube into a clean, sterile 1.5 ml microcentrifuge tube.
 - Add 200 µl prewarmed Elution Buffer to the upper reservoir of the Filter Tube.
 - Centrifuge the tube assembly for 1 min at 8,000 × *g*.
- 9 The microcentrifuge tube contains the eluted, purified DNA, which can be used directly or stored at +2 to +8°C or +15 to +25°C for later analysis. For details on adding an optional RNase digestion, see related procedures in the pack insert.

High Pure PCR Template Preparation Kit workflow



Ordering information

Product	Cat. No.	Pack Size
High Pure PCR Template Preparation Kit	11 796 828 001	Up to 100 isolations

For more information about the **High Pure PCR Template Preparation Kit** and other products for nucleic acid isolation and purification, visit

www.roche-applied-science.com/napure

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