Proteinase K Solution (20mg/ml)

Product Name	Product Code	Kit Packing
Proteinase K Solution (20mg/ml)	DS0013-1ML	1 ml
	DS0013-5ML	5 ml

Intended Use

Recommended for isolation of DNA from human blood, plasma, tissue and serum samples.

Introduction: Proteinase K Solution (20mg/ml) is an enzyme used for digestion of proteins. It is used for removal of endogenous nucleases during the preparation of DNA and RNA.

Application: Proteinase K Solution (20mg/ml) is used for isolating DNA from blood cells, animal cells, tissues and bacterial cells.

Properties:

Appearance: Colorless solution
Clarity: Clear and free of particles
DNase & RNase: None detected
Suitability Test: This reagent has been tested and is suitable for isolation of DNA from blood cells, animal cells, tissues and bacterial cells.

Storage conditions: Proteinase K Solution (20mg/ml) has to be stored on receipt at - 20°C. The shelf- life of this solution in 36 months.

Specimen Handling and Collection

Collect whole blood in an anticoagulant tube (an EDTA tube is preferred) under sterile conditions (if to be used for future) and tissues in a sterile container. Store the samples at 2-8°C for short term storage or -20°C for long term storage. Ensure that the blood sample/ tissues are at room temperature (15-25° C) before beginning the protocol. After use, contaminated material must be sterilized by autoclaving before discarding.

Types of Specimen

Clinical samples: Whole blood, Plasma, Serum and tissues.

Procedure

1. **Collect Blood**

Collect whole blood in an anticoagulant tube (an EDTA tube is preferred) under sterile conditions (if to be used for future). Ensure that the blood sample is at room temperature before beginning the protocol.

For Frozen blood: To 200 µl of frozen blood pellet (kept on ice), add 200 µl of Lysis Solution (C1) (DS0010). Thaw the pellet with continuous pipetting and proceed with step 2 for Proteinase K and RNase A treatment (optional). Incubate at 55°C for 10 minutes and then proceed to step 4 of the protocol.

ISO 9001-2015 CERTIFIED
WHO GMP CERTIFIED
ISO 13485-2016 CERTIFIED
Registered Office
HiMedia Laboratories Pvt Ltd.
Plot No. C-40, Road No. 21Y, MIDC, Wagle Industrial Area,
Thane, (West) 400604, Maharashtra, INDIA.
Customer Care No.: 00-91-22-6116 9797
Tel : 00-91-22-6147 1919, 6903 4800
Fax : 6147 1920
Web : www.himedialabs.com
Email : info@himedialabs.com
mb@himedialabs.com

The information contained herein is believed to be accurate and complete. However no warranty or guarantee whatsoever is made or is to be implied with respect to such information or with respect to any product, method or apparatus referred to herein

NOTE: When extracting DNA from frozen blood, it is very important that the blood should be kept on ice and directly mixed with Lysis Solution (C1). Do not allow the frozen blood pellet to thaw except when it is directly mixed with Lysis Solution (C1). This prevents release of apoptotic enzymes that can decrease the DNA yield drastically.

NOTE: If the sample is less than 200 µl, add the Resuspension solution (ML116) to bring the volume upto 200 µl.

Procedure

Add 20 µl of the Proteinase K solution (20 mg/ml) (DS0013) into 2.0 ml capped collection tube containing 200 µl of the whole blood. Vortex for 10-15 seconds to ensure thorough mixing.

Optional RNase A treatment
If RNA-free genomic DNA is required, add 20 µl of RNase A solution (20 mg/ml) (DS0003). Vortex for 10-15 seconds and incubate for 2 minutes at room temperature (15-25°C); continue with step 3.

1. **Lysis reaction**
   Add 200 µl of the Lysis Solution (C1) (DS0010) to the sample, vortex thoroughly for a few seconds to obtain a homogenous mixture. Incubate at 55°C for 10 minutes.

   NOTE: If cell clumps are visible, the sample can be mixed gently by pipetting to obtain a homogenous mixture.

2. **Prepare for Binding**
   Add 200 µl of ethanol (96-100%) to the lysate obtained from the above step for preparation of lysate for binding to the spin column. Mix thoroughly by gentle pipetting.

   NOTE: A homogenous solution is essential.

3. **Load lysate in HiElute Miniprep Spin Column (Capped)** [**DBCA03]**
   Transfer the lysate obtained from step 3 into the spin column provided. Centrifuge at ≥6,500 x g (≈10,000 rpm) for 1 minute. Discard the flow-through liquid and place the column in a same 2.0 ml collection tube.

   NOTE: Use a wide bore pipette tip to reduce shearing of the DNA when transferring contents into the column.

4. **Prewash**
   Add 500 µl of Prewash Solution diluted to the column and centrifuge at ≥6,500 x g (≈10,000 rpm) for 1 minute. Discard the flow-through liquid and re-use the same collection tube with the column.

5. **Wash**
   Add 500 µl of Wash Solution diluted to the column and centrifuge at 12,000-16,000 x g (≈13,000-16,000 rpm) for 3 minutes to dry the column. Discard the flow-through liquid and spin the empty column for another minute at the same speed if residual ethanol is observed. Discard the collection tube containing the flow through liquid and place the column in a new uncapped 2.0 ml collection tube.

   NOTE: The column must be free of ethanol before eluting the DNA. The tube can be emptied and re-used for this additional centrifugation step.

6. **DNA Elution**
   Pipette 100 µl of the Elution Buffer (ET) (DS0040) directly onto the column without spilling to the sides. Incubate for 1 minute at room temperature (15-25°C). Centrifuge at ≥6,500 x g (≈10,000 rpm) for 1 minute to elute the DNA. Repeat the step again with another 100 µl of Elution Buffer (ET) for high yield of DNA.

   NOTE: DNA elution can also be performed in single step by the addition of 200 µl of Elution Buffer (ET) at a time (DNA yield would be low). Storing DNA in water may cause acid hydrolysis. To increase the elution efficiency, incubate for 5 minutes at room temperature (15-25^{0}C) after adding the Elution Buffer (ET), then centrifuge. Elution with volume less than 200 µl increases the final DNA concentration in the eluate significantly, but slightly reduces the overall DNA yield.

7. Transfer the eluate to a fresh capped 2ml collection tube for longer DNA storage.

Warning and Precautions

Not for Medicinal Use. Read the procedure carefully before beginning the protocol. Wear protective gloves/protective clothing/eye protection/face protection. Follow good clinical laboratory practices while handling clinical samples. Standard precautions should be followed as per established guidelines. Safety guidelines may be referred in safety data sheets of the product.

Limitations

1. The yield of DNA depends upon the type and the volume of starting material used.

Performance and Evaluation

Performance of the enzyme is expected when the enzyme is used as per the protocol mentioned in the product insert within the expiry period when stored at recommended temperature.

Quality Control

Type of Sample	DNA Yield	DNA Purity
200 µl whole blood	4-12 µg	1.6-1.9

Safety Information

Proteinase K Solution (20mg/ml) contains irritants. Take appropriate laboratory safety measures and wear gloves when handling. Not compatible with disinfecting agents containing bleach. Please refer the Safety Data Sheet (SDS) for information regarding hazards and safe handling practices.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed off in accordance with current laboratory techniques.

Technical Assistance

At HiMedia, we pride ourselves on the quality and availability of our technical support. For any kind of technical assistance, mail to mb@himedialabs.com.

Symbol

	Manufacturer		Do not use if package is damaged
LOT	Batch code		Temperature limit
	Date of manufacture (YYYY-MM)		Consult instructions for use
	Use-by date (YYYY-MM)	REF	Catalogue number

Identification No.: PIDS0013
Rev. No.: 03
Date of Issue: 2026-03

Disclaimer :

User must ensure suitability of the product(s) in their application prior to use. Products conform solely to the information contained in this and other related HiMedia™ publications. The information contained in this publication is based on our research and development work and is to the best of our knowledge true and accurate. HiMedia™ Laboratories Pvt Ltd reserves the right to make changes to specifications and information related to the products at any time. Products are not intended for human or animal or therapeutic use but for laboratory,diagnostic, research or further manufacturing use only, unless otherwise specified. Statements contained herein should not be considered as a warranty of any kind, expressed or implied, and no liability is accepted for infringement of any patents.

HiMedia Laboratories Pvt. Ltd. Reg.office : Plot No. C-40, Road No. 21Y, MIDC, Wagle Industrial Estate, Thane, (West) 400604, Maharashtra, INDIA.
Customer Care No.: 00-91-22-6116 9797 Tel: 00-91-22-6147 1919, 6903 4800 Email: techhelp@himedialabs.com Website: www.himedialabs.com