SHANKARANARAYANA LIFE SCIENCES
A NEW GENERATION COMAPANY

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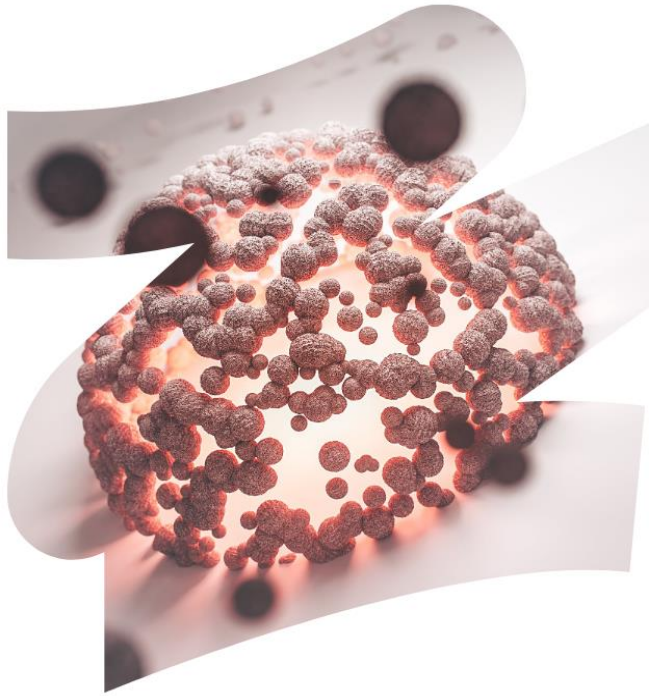
SnLs
A NEW GENERATION COMPANY



SN Blood DNA extraction Kit

Column Based

Innovation Speaks

SN BLOOD DNA EXTRACTION KIT
(Column Based)

DESCRIPTION:

SN Blood Genomic DNA Extraction kit provides a cost- effective system designed for the fast and easy isolation of the DNA fragments from whole blood samples like Mammalian etc. The preparation is based on a silica-membrane technology, for binding DNA in high-salt and elution in low-salt buffer. The kit provides a simple and efficient way to purify DNA in a size range between 100 bp to 10 kb.

CONTENTS OF KIT:

Sl. No	Components	Volume 5 Rxn	50 Rxn	100 Rxn
1	LS Buffer	2ml	20ml	40ml
2	BB Buffer	2ml	20ml	40ml
3	W1 Buffer	1.5ml	15ml	30ml
4	Elution Buffer	250µl	2.5ml	5ml
5	SN Spin Column & Collection tube	5 No.	50 No.	100 No.

NOTE: Preparation for first use after receiving the kit (Add 100% Ethanol to Wash Buffer) (5Rxn: 3.4ml, 50Rxn: 34ml, and 100Rxn: 68ml) mix well and store the buffer at room temperature.

- If **Proteinase K** shipped in lyophilized form, resuspend with Nuclease free water. **(5Rxn:100µl,50Rxn:1ml,100Rxn:2ml)**
- If **RNase A** shipped in lyophilized form, upon receiving resuspend with Nuclease free water **(5Rxn:50µl,50Rxn:500µl,100Rxn:1ml)**
- **Proteinase K / RNase A store at -20°C either in Liquid form or Lyophilized form**

REQUIRED MATERIALS NOT PROVIDED

- ✓ 100% Ethanol
- ✓ Dry bath
- ✓ 1.5ml Centrifuge tubes
- ✓ micro centrifuge
- ✓ Vortex

STORAGE / SHIPPING:

- Shipped at: Ambient Temperature.
- Storage: All Buffers are stored at Room temperature.

SPECIFICATIONS:

Principle	Spin Column
Recommended input amount : 200µI of whole blood	
Binding Capacity : ~30-40µg genomic DNA	
Elution Volume: 50µl	
Purity: A260/280 - 1.8±0.1, A260/230 - 2.0±0.1	
Compatible Downstream Applications : Endpoint PCR, qPCR, Sequencing, etc	
Expected Yield : Up to 10 µg (depending upon the type. Quality & quantity of the starting material used)	

PROCEDURE:

1) Take 200µl of blood sample in a 1.5ml centrifuge tube.
2) Add 300µl of LS Buffer, 20µl Proteinase K and 10µl of RNAse A , resuspend by gentle pipetting and vortex for 30secs.
3) Incubate at 56˚C for 15mins. Transfer entire lysate to SN Spin Column and leave at room temperature for 1min.
4) Centrifuge for 3-5mins at 15,000rpm and discard the flow through .Then add 400µl of BB Buffer.
5) Centrifuge for 2mins at 15,000rpm and discard the flow through.
6) Add 500µl of W1 buffer and centrifuge for 2mins at 15,000rpm and discard flow through.

Note : (W1 buffer concentration per reaction: W1 buffer -300µl: 100% ETOH -200µl)

1) Repeat step (6).
2) Then centrifuge at 15,000rpm for an additional 1min to dry the column.
3) Place the SN Spin Column in a new 1.5ml centrifuge tube and incubate for 10mins at 56˚C. (with cap open). This step is very important to remove the residual of ethanol.

NOTE: Elution should be pre-heated for 10mins before adding into the SN Spin Column.

10) Add 50µl of pre-heated Elution to the membrane center of SN Spin Column and incubate for 4mins at 56˚C (with cap close).

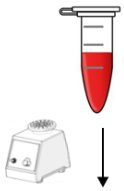
Important step: For effective elution, make sure that the elution buffer is dispensed onto the membrane center and is absorbed completely.

11) Centrifuge the tube at 15,000rpm for 1 min to Elute the DNA and store at -20°C.

FLOW CHART:

- Take 200µl Blood sample in a centrifuge tube along with 300µl LS Buffer , 20µl Proteinase K and 10µl of RNase A

- Mix well by gentle pipetting and vortex for 30 secs.
- Incubate at 56˚C for 15mins



- Transfer entire lysate to SN Spin Column and leave at room temperature for 1min

- Centrifuge for 3-5mins at 15,000 rpm
- Discard flow through

- Add 400µl BB Buffer
- Centrifuge for 2mins at 15,000rpm
- Discard flow through

- Add 500µl W1 buffer

- Centrifuge for 2mins at 15,00rp and discard flow through

- Add 500µl W1 buffer again and centrifuge for 2mins at 15,000rpm

- Discard flow through
- Centrifuge for additional 1min at 15,000rpm

- Place the SN Spin Column in a new 1.5ml Eppendorf tube

- Incubate at 56˚C for 10mins (with cap open)

- Add 50µl of pre-heated Elution to the membrane center of SN Spin Column (with close cap)

- Incubate at 56˚C for 4mins Centrifuge for 1min at 15,000rpm

- Elute Pure DNA (store at -20°C)

TROUBLE SHOOTING:

Problems	Possible reasons	Solutions
Low or none recovery of DNA fragment	Apply more than 200µl Blood cells	If product is more than 200µl, separate it into multiple tubes.
	Elution of DNA fragment is not efficient	Make sure the pH of Elution Buffer 8.0.
		Make sure that the elution solution has been completely absorbed by the column membrane before centrifugation.
		Preheat the elution solution to 56°C before use.
Poor Performance in the downstre am applications	Salt residue remains in eluted DNA	Wash the column twice with W1 Buffer.
	Ethanol residue remains in eluted DNA	Do discard the flow-through after washing with W1 Buffer and centrifuge for an additional 1min.

IMPORTANT NOTES:

a) Buffer provided in this kit contains irritants. Wear gloves and lab coat when handling these buffers.

b) Add required volume of ethanol (96-100%) to wash Buffer before use.

c) Centrifugation steps are done by a micro centrifuge capable of the speed at 11,000 ~ 15,000 rpm.

d) Fresh electrophoresis buffer is recommended to acquire highly efficient recovery.

e) Fresh TBE electrophoresis buffer is recommended on electrophoresis for experiments requiring high purity.

f) Then excising the agarose gel, make sure to shorten the time of exposure to ultraviolet irradiation.