Quick Protocol

DNSol Clotted Blood Kit

DNA isolation based on solution

- MiniPrep

For DNA Isolation from

Clotted Blood

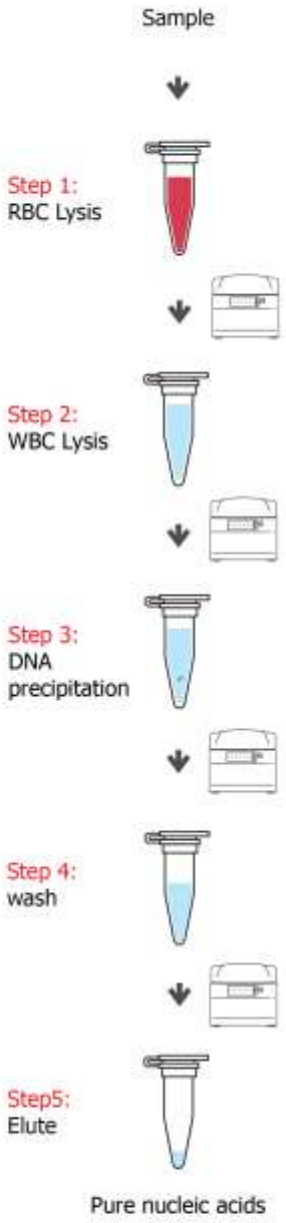
Kit Content

Component	50 preps
RBC Lysis Buffer, DNSol Edition	100ml
ROS	25ml
RRB	5ml
Prime-RNase A	250µl
RJ-Protease	1ml

Before Start

- If ROS or RBC Lysis Buffer or PPS forms precipitate, please warm it to 56°C until the precipitate has fully dissolved. This is due to storage condition and won't influence the efficiency of buffer.

Procedure of solution-based DNA isolation in quick look



Protocol

Isolation of Genomic DNA (based on solution)

Sample type: Clotted Blood

Some tips to know:

- All centrifugation steps are carried out at room temperature (15–25°C).
- If any buffer (ROS or RBC Lysis Buffer) forms precipitate, please warm it to 56°C until the precipitate has fully dissolved.

Process

- Add 1ml RBC lysis buffer to 400µl clotted blood in microtube, invert 5 times, vortex 10s at high speed and centrifuge at 13000 rpm for 3min.
- Discard supernatant and add 1ml RBC lysis buffer to the pellet, invert 5 times, vortex for 10s at high speed and centrifuge at 10000 rpm for 2min.
- Discard supernatant. Add 500µl ROS and then 20µl RJ-Protease. Mix thoroughly by pulse vortexing for 30s, then incubate at 56°C for 30-60 min until the sample is completely lysed. Pulse vortex every 10min during incubation to intersperse the sample, or place it in a thermomixer or shaking water bath.
- Add 200µl phenol %50 and then 200µl chloroform. Vigorously shake it for 30s. Then pulse vortex for 15s and centrifuge at 13000 rpm for 10min.
- Transfer the supernatant to a new tube. Add 5µl Prime-RNase A to the isolated aqueous phase. Then pulse vortex for 5s and incubate for 10min at room temperature, 25°C.
- Add equal volume of isopropanol to the microcentrifuge tube. Invert 10-15 times rapidly. Centrifuge at 12000 rpm for 1min.

  Note: The DNA should be visible as a small white pellet.

- Discard supernatant, aspirate the pellet. Add 600µl ethanol 70% to the pellet; centrifuge at 10000 rpm for 2min.
- Discard supernatant and aspirate the pellet. Then, add 50µl RRB. Mix by pipetting until the pellet is dissolved completely. Alternatively, you can vortex for 10 s after adding the RRB, then incubate at 37°C for 10min (or 20min at room temperature, 25°C); afterward vortex for 10s, to dissolve the DNA. The DNA is ready for further applications; you can use 2-5µl of it for PCR reaction.

  Note: Do not dry the pellet and add RRB immediately.