RoomTemp Sample Lysis Kit

P073

Version 22.1

Product Description

RoomTemp Sample Lysis Kit is a simple and rapid blood room temperature lysis kit. This kit contains two components, Lysis Buffer and Stabilizing Buffer. Lysis Buffer contains special components that can rapidly destroy cell membrane proteins and membrane structures, which can fully release genomic DNA in cells. Stabilizing Buffer contains protective proteins and stabilizing factors, which can eliminate the inhibitory effect of inhibitors in lysed samples on downstream qPCR and PCR. And the DNA solution can be stored stably for a long time. Applicable blood sample types include fresh blood, frozen blood and conventional anticoagulant blood (EDTA, citrate, heparin sodium, etc.). Genomic DNA can be released from whole blood samples after 3 min lysis at room temperature. The DNA solution can be directly used as templates for SNP detection by Taqman probe method, quantification by qPCR probe method, PCR amplification, etc. It can achieve the same effect as traditional genome extraction methods without complicated extraction operations. In addition to blood samples, this kit is also compatible with FTA blood cards, oral swabs, plant tissues and other samples.

Components

Components	P073-01 (250 rxns)	P073-02 (1,000 rxns)	P073-03 (5,000 rxns)
Lysis Buffer	5 ml	20 ml	100 ml
Stabilizing Buffer	5 ml	20 ml	100 ml

Storage

Store at 2 ~ 8℃. Adjust the shipping method according to different destinations.

Applications

It is applicable for blood sample types include fresh blood, frozen blood and conventional anticoagulant blood (EDTA, citrate, heparin sodium, etc.).

Notes

For research use only. Not for use in diagnostic procedures.

Mechanism & Workflow

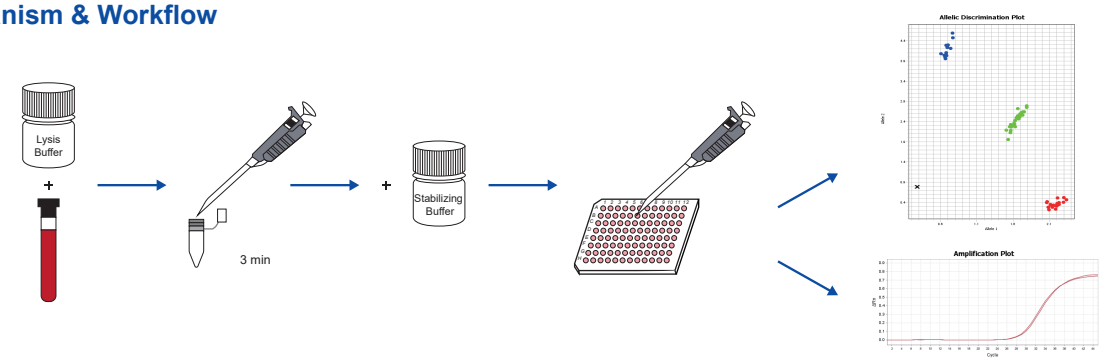

Fig 1. Workflow of RoomTemp Sample Lysis Kit

Experiment Process

1. Mix Lysis Buffer and Stabilizing Buffer by invert slightly before use. And avoid generating a lot of bubbles.
2. Put the sample into a centrifuge tube, add an appropriate amount of Lysis Buffer, vortex to mix, and collect it to the bottom of the centrifuge tube by low-speed brief centrifugation (refer to Table 1 for the recommended adding volume of the sample and Lysis Buffer)[a].
3. Incubate at room temperature or 95℃ for 3 min (please refer to Table 1 for the incubation conditions of different types of samples).
4. Add Stabilizing Buffer equal to the volume of Lysis Buffer, vortex to mix, and collect by low-speed brief centrifugation to the bottom of the centrifuge tube to complete the preparation of DNA solution (refer to Table 1 for the recommended adding volume of Stabilizing Buffer)[b].

5. For a 20 μl amplification system, it is recommended to take 1 - 4 μl of the lysed DNA solution as a template for downstream detection experiments:

(1) It is recommended to use ChamQ Geno-SNP Probe Master Mix (Vazyme #Q811) for Taqman probe SNP detection.

(2) It is recommended to use AceQ Universal Probe Master Mix V2 (Vazyme #Q513-EN) for quantitative detection by qPCR probe method.

(3) It is recommended to use 2 × Rapid Taq Master Mix (Vazyme #P222), 2 × Phanta Max Master Mix (Vazyme #P515), etc. for PCR amplification detection.

a. If the sample volume is large, the added volume of Lysis Buffer and Stabilizing Buffer can be scaled up proportionally for lysis. For example, 5 μl of blood can be lysed by adding 50 μl of Lysis Buffer. After incubating at room temperature for 3 min, add 50 μl of Stabilizing Buffer and mix by inversion.

b. The obtained DNA solution can be stored at 4℃ for 1 month. For long-term storage, freeze the obtained DNA solution at -20℃ and mix by inversion before use.

Table 1. Lysis conditions for different types of samples

	Sample amount	Lysis Buffer volume	Incubation conditions[a]	Stabilizing Buffer volume
Fresh Blood, EDTA/Citrate/ Heparin sodium anticoagulant blood[b]	2 μl	20 μl	Room temperature, 3 min	20 μl
Whatman 903 and FTA cards	3 mm	50 μl	95℃, 3 min	50 μl
Oral swabs[c]	1	400 μl	95℃, 3 min	400 μl
Cell suspension	2 μl	20 μl	Room temperature, 3 min	20 μl
Tissues homogenate	5 μl	50 μl	95℃, 3 min	50 μl
Mice tails	1 - 2 mm	50 μl	95℃, 3 min	50 μl
Hair (with follicles)	2 - 3	50 μl	95℃, 3 min	50 μl
Leaves	2 - 3 mm	50 μl	95℃, 3 min	50 μl

a. Room temperature: 20 ~ 25℃. For samples that are difficult to lyse, the processing time can be extended appropriately.

b. This kit has good resistance to impurities and is compatible with hyperlipidemia whole blood, hyperbilirubin whole blood and hemolysis samples.

c. Oral swab: There are two options for lysing the oral swab. Option 1 is to directly place the oral swab that has collected oral cells in 400 μl Lysis Buffer, rotate it 5 times, squeeze out the absorbed content and discard the swab. After incubating at 95℃ for 3 min, add 400 μl Stabilizing Buffer and mix by inverting. Option 2 is to elute the collected oral swabs with a solution such as physiological saline to form a cell suspension, take 2 μl of the cell suspension into 20 μl of Lysis Buffer, incubate at 95℃ for 3 minutes, add 20 μl of Stabilizing Buffer, and invert to mix.

FAQ & Troubleshooting

◇ Low qPCR amplification platform or low PCR yield?

May be due to low sample size used for lysis, or low expression of the gene being tested. You can try to increase the amount of sample input for lysis and the input amount of template in the amplification system. Taking blood as an example, the amount of lysate recommended in Table 1 can lyse 2 - 10 μl sample, and the 20 μl amplification system is compatible with 1 - 10 μl template, and has no effect on subsequent qPCR and PCR.

◇ Does the blood sample appear blood red after lysing?

Under normal circumstances, the solution after blood lysing is brown, but individual blood samples appear blood red, which is related to blood samples. This is a normal phenomenon which has no effect on subsequent experiments.

◇ Is it possible to use only Lysis Buffer for lysis and subsequent amplification reactions?

Protective proteins and stabilizing factors are added to the Stabilizing Buffer, which can eliminate the inhibitory effect of inhibitors in the lysed sample on downstream qPCR and PCR, so that the lysed DNA solution can be stored stably for a long time. If only Lysis Buffer is used for sample lysis, the recommended template input volume is 1 - 2 μl in the 20 μl downstream amplification system, and the lysed DNA solution can be stored at -20℃ for 1 week. In other cases, it is not recommended to only add Lysis Buffer for lysis.