One step direct PCR lysis buffer

Cat No	Pack size
LB0020	20 ml
LB0100	100 ml

Description

We develop a reagent that would allow a wide variety of biological samples to be used directly in PCR, without further neutralization or DNA isolation. It offers a considerable simplification of present direct PCR approaches and it will be particularly useful for screening transgenic mutants and trace amounts of precious materials.

STABILITY:

One step direct PCR lysis reagent is stable at 4℃ for at least one year . During storage, keep the bottle tightly closed.

Protocol

1. Take 1-10 µl or 1-10 mg of sample (contain bacteria or cell pellets) into 50 µl of One step direct PCR lysis buffer and mix well..
2. Stand at room temperature for 15-30 minutes for cell lysis. Then, the samples were kept on ice until used for direct-PCR or stored at 4℃ for future use.
3. Vortex the sample lysate and take 2~5 µl aliquot directly into 20~50 µl of PCR premix for PCR reaction.

SPECIFIC APPLICATIONS

Plant and animal tissues were cut several pieces as possible.

Animal/Plant tissues.
Place 10-40 mg of tissue in 50ul to 200 ul of One step direct PCR lysis buffer by incubating at RT or 80℃for 15 minutes. After incubation, vortex the lysate and use 1 - 5 for 50 µl final volume of PCR mixture

Whole blood.
Mix 200 µl of fluid sample with 1ml of 1x RBC.lysis buffer and incubate for 15 minutes at RT. Then spin down the lysate by centrifuge at 500-1000xg for 3-5 min. Discard supernatant complicate as possible. Add 50ul of One step direct RCR lysis buffer into the pellet and mix well.

NOTES

1. The lysate volume should not exceed 10% of the total volume of the PCR mix.

2. For optimal amplification, use enough sample lysate to PCR mix. Typically, the minimal amount of DNA required for 35~40 cycles of PCR is 0.1 - 1 ng.

3. Incubation of samples at 80 - 90℃ for 5 - 15 minutes improves release of DNA from samples. Alternatively, improve release of DNA by incubating samples overnight at room temperature.

4. This excess of DNA and other cellular material can inhibit PCR.. Increasing the sample-to-reagent ratio is necessary.

5.increase the amount of tissue 2 - 3 times per volume of One step direct PCR reagent.

6. After lysis in One step direct PCR lysis reagent, use samples for PCR immediately or store them at 4℃ for future use.

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