TIANcombi DNA Lyse & Det PCR Kit

Fastest DNA Extraction from Plant, Bacteria and Tissue Samples Followed by PCR

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Cat. No. 4992527/4992528

Kit Contents

Contents	4992527 50 preps	4992528 200 preps
Buffer B1	6 ml	24 ml
Buffer B2	6 ml	24 ml
2× Det PCR MasterMix	500 µl	2× 1 ml
Grinding Pestles	10	20
Handbook	1	1

Compatible device

TGrinder (Cat. No. OSE-Y50)

Storage

Buffer B1 and Buffer B2 can be stored at room temperature (15-30°C) for up to 12 months without showing any reduction in performance and quality. 2× Det PCR MasterMix could be stored at -30~-15°C for 12 months, and repeated freezing-thawing will not affect its activity.

Introduction

TIANcombi DNA Lyse & Det PCR Kit uses special buffer system for fast one-step DNA extraction and PCR from a wide range of starting materials, including plant tissues, seeds, animal tissues, blood, yeast and bacteria. The kit contains both the solution for DNA extraction and the PCR amplification reagent. The whole extraction process will not include protein, RNA and secondary metabolite removal step, as well as phenol extraction and ethanol precipitation. Grinding in liquid nitrogen is also not required.

The 2× Det PCR MasterMix could amplify raw samples with high compatibility, high efficiency and specificity without removing impurities such as proteins. This reagent contains *Taq* DNA polymerase, dNTPs, $MgCl_2$, buffer, PCR enhancers and stabilizers, which is fast and simple to use, and especially qualified for high-throughput screening.

Features

Simple and fast: DNA from different tissues can be extracted in 5 min without the need for liquid nitrogen grinding.

Wide applications: Applicable for plant leaves, seeds, animal tissues, blood samples (e.g. fresh blood, anti-coagulation blood, blood clots, dried blood spots), yeast and bacteria, etc.

Good compatibility: The PCR reagents in this kit are suitable for the amplification of DNA extracted from various sample sources.

Gene detection: Especially suitable for large-scale gene detection.

Important Notes Before Starting

1. Avoid repeated freezing and thawing of the sample, otherwise the extracted DNA fragments will be smaller and the extraction yield will be decreased.
2. For phenols-rich samples like cotton leaves, the samples amount should not be over 0.4 mg, or it will reduce PCR reaction efficiency.
3. Buffer B1 and Buffer B2 should be stored at room temperature (15-30°C). If a precipitate has formed in Buffer, please place the buffer at room temperature or warm at 37°C for 10 min to dissolve the precipitate.
4. 2× Det PCR MasterMix provided in this kit is a 2× stock solution. Template, primers and sterilized water should be added to make up to a 1× solution before use.

Protocol

1. For first use, please check if there is precipitate formed in the two bottles of buffer. If there is, please put the buffer at room temperature or 37°C water bath until the precipitation is dissolved. The dissolved buffer should be stored at room temperature.
2. Put small amount (take table 1 for reference) of samples to 1.5 ml centrifuge tube, add 100 µl Buffer B1, and make sure that Buffer B1 could completely cover the sample.
3. Grind samples with a grinding pestle.

   Note: For blood or bacteria samples which are difficult to discriminate whether have been fully grinded, please grind for 30 sec with grinding pestle. And bacteria samples should be centrifuged to collect the cell pellet. For plant seeds, skin, connective tissue and etc., please grind it to be muddy with grinding pestle (TGrinder (Cat. no. OSE-Y50) will be more convenient to use).
4. Add 100 µl Buffer B2, vortex to mix, then centrifuge at 12,000 rpm (~13,400 x g) for 2 min.
5. Pipet 100 µl supernatant to a clean 1.5 ml centrifuge tube to be used as template.
6. Set up the PCR amplification reaction.

Reaction system

Components	Volume
2× Det PCR MasterMix	10.0 µl
Forward Primer (10µM)	0.5 µl
Reverse Primer (10µM)	0.5 µl
Template DNA	1.0 µl
ddH_2O	up to 20 µl

PCR cycle set-up:

94°C 3 min
94°C 30 sec
55°C 30 sec } 35 cycles
72°C 1 min
72°C 5 min

Result Detection

Take 5-10 μl reaction products for agarose electrophoresis detection.

Note: the example provided is for your reference. As the template and primers differs, the real reaction condition should be modified accordingly.

Appendix Table 1

Samples	Volume
Plant leaves	1-5 mg
Plant seeds	1-5 mg (without seed peels)
Animal tissues	1-5 mg
Bacteria	Cell pellet collected from 0.2-0.5 ml bacteria liquid
Yeast	Cell pellet collected from 0.2-0.5 ml yeast liquid
Blood	20 μl