CTAB Extraction Solution

For genomic DNA extraction

(Cat. # 786-564, 786-565)

INTRODUCTION

CTAB Extraction Solution is a widely-used reagent used to isolate DNA from tissues, particularly plants. Polysaccharides and polyphenols are problematic contaminants associated with DNA isolated from plants and when supplemented with polyvinylpyrrolidone CTAB Extraction Solutioneffectively eliminates polysaccharides and polyphenols by employing the cationic detergent CTAB (hexadecyltrimethylammonium bromide or cetyltrimethylammounium bromide), and the polyphenol binding agent, Polyvinylpyrrolidone.

Traditional CTAB protocols typically require the homogenization of plant samples in CTAB Extraction Buffer prior to centrifugation to pellet debris and polysaccharides. The supernatant is then extracted using chloroform, and DNA is precipitated with alcohol. Isolated DNA is typically very clean.

ITEM(S) SUPPLIED

Cat. #	Description	Size
786-564	CTAB Extraction Solution	60ml
786-565	CTAB Extraction Solution	125ml

STORAGE CONDITIONS

Shipped at ambient temperature. Upon arrival, store at room temperature.

ADDITIONAL ITEMS REQUIRED

- PVP (Polyvinylpyrrolidone) (Cat. # RC-085, RC-086)

PREPARATION BEFORE USE

For plant samples, dissolve 1% (w/v) of PVP into the required volume of CTAB Extraction Solution (0.6g in 60ml).

PROTOCOL

1. Pulverize 100 mg of sample using a liquid nitrogen chilled mortar and pestle. Once processed, mix 100 mg of frozen powdered sample with 500 µl of CTAB Extraction Solution.
   NOTE: *Samples can be ground with pestle and mortar with suitable grinding resin (Molecular Grinding Resin, 786-138PR), in lieu of liquid nitrogen.*

2. Place the homogenate into a 60°C bath for 30 min.

3. Centrifuge the homogenate for 10 minutes at 10,000 x g.

4. Transfer the supernatant into a clean tube and add 5 µl of RNase (10 mg/ml in water) to the lysate. Incubate at room temperature for 15 minutes.

5. Centrifuge for 5 minutes at 10,000 x g.

6. Extract the lysate with equal volume of chloroform: isoamyl alcohol (24:1). Vortex for 5 seconds then centrifuge for a minute at 10,000 x g to separate the phases.

7. Transfer the upper phase to a clean tube.

8. Repeat step 7 until upper layer is clear. Transfer upper phase to a new tube.

9. Add 0.7 volumes of isopropanol. Mix and incubate at -20°C for 15 minutes.

10. Centrifuge for 10 minutes at 10,000 x g. Decant and wash the pellet with 70% ethanol.

11. Decant without disturbing the pellet. Dry the pellet briefly in a speed vac or at room temperatuer. Do not over-dry the DNA.

12. Resuspend the DNA in 50 µl of TE buffer.

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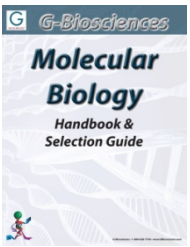

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