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DNA extraction from plants

Forked from [DNA extraction from plants](#)Remco Stam¹¹Technical University of Munich*In Development* This protocol is published without a DOI.

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ABSTRACT

Widely used protocol to extract DNA from plant leaves.
Many versions circulate on the web, this is the version as we use it.
It works well on maize, tomato and probably many other plants.

PROTOCOL CITATION

Remco Stam 2020. DNA extraction from plants. **protocols.io**
<https://protocols.io/view/dna-extraction-from-plants-bqzwm7e>

FORK NOTE

FORK FROM

Forked from [DNA extraction from plants](#), Remco Stam

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45846

MATERIALS TEXT

MATERIALS

☒ RNase A Contributed by users☒ Isopropanol Contributed by users☒ Chloroform Contributed by users☒ Liquid nitrogen Contributed by users☒ Ethanol Contributed by users☒ Isoamylalcohol Contributed by users☒ Beta-mercaptoethanol Contributed by users☒ Cetrimonium bromide Contributed by users☒ Tris Contributed by users☒ EDTA Contributed by users

SAFETY WARNINGS

When working with Beta-Mercaptoethanol or Chloroform, always work in a fume hood.

DISCLAIMER:

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BEFORE STARTING

Prepare all reagents and materials.

TE Buffer, pH 8.0

For 250 ml:

2.5 ml 1 M Tris HCl, (10 mM)

0.5 ml 0.5 M EDTA, (1 mM)

Add H₂O to 250 ml

CTAB Extraction Buffer

For 500 ml:

10 g CTAB (1.5% w/v) (Cetyltrimethylammonium bromide or Certridium Bromide: C₁₉H₄₂BrN)

50 ml 1 M Tris HCl, pH 8.0 (75 mM)

20 ml 0.05 M EDTA, pH 8.0 (100 mM)

140 ml NaCl (5M)

290 ml H₂O

In a 2 ml tube, add 800 µL of 1.5x CTAB and 1 µL of Beta-mercaptoethanol to the ground leaf material

Incubate 1 hour at 60-65 degrees (C)

Cool at Room Temp

Add 1 volume of Chloroform/Isoamylalcohol (24:1) mixture

Mix on overhead shaker for 10 minutes

Centrifuge at 3000 rpm for 25 minutes

Transfer supernatant to a new tube

Use wide pipette tips (cut point of tips if needed)

Add 10 µL RNase (10 mg/ml)

incubate at 37 degrees (C) for 30 minutes

Add one volume 100% Isoopropanol (pre-chilled at -20)

Allow sample to preprecipitate for up to 20 minutes at -20

Centrifuge: full speed, 10 minutes
Remove supernatant

Add 1 ml 70% Ethanol
Centrifuge: full speed, 10 minutes
Remove Supernatant

Repeat once

Dry pellet in air or approx 20 minutes in vacuum centrifuge

Dissolve pellet in 1x TE (10 mM Tris, pH8, 1 mM EDTA)