DNA extraction using the ammonium acetate technique

Version 2

Juan Carlos Illera

Abstract

A simple protocol to extract DNA

Citation: Juan Carlos Illera DNA extraction using the ammonium acetate technique. **protocols.io** dx.doi.org/10.17504/protocols.io.knycvfw
Published: 07 Nov 2017

Before start

If blood is preserved in ethanol, it has to be dried before starting.

Protocol

Step 1.

Add 50 µl of blood, 200 µl of a DNA extraction solution (which includes tris-HCL 30 mM ph 8, EDTA 10 mM, and 0,4% SDS), and 3 µl proteinase K.

AMOUNT
50 µl Additional info: Blood
AMOUNT
200 µl Additional info: DNA extraction solution
AMOUNT
3 µl Additional info: Proteinase K

Step 2.

Vortex and overnight at 56°.

TEMPERATURE
56 °C Additional info:

Step 3.

Also incubate 70°C around three hours shaking frequently.

TEMPERATURE
70 °C Additional info:

Step 4.

Add 200 µl AcNH4 4 M. Vortex and incubate 30 minutes. Shake every 10 minutes.

AMOUNT
200 µl Additional info: AcNH4 4 M

Step 5.

Centrifuge 15 minutes at 13000 rpm.

Step 6.

Move the supernatant to new tubes.

Step 7.

Add 800 µl of cold EtOH 100%.

AMOUNT
800 µl Additional info: EtOH

Step 8.

Centrifuge 15 minutes at 13000 rpm.

Step 9.

Remove the supernatant.

Step 10.

Wash the pellet with 800 µl of EtOH 70%.

AMOUNT
800 µl Additional info: EtOH 70%

Step 11.

Centrifuge 5 minutes at 13000 rpm.

Step 12.

Remove the supernatant and dry.

Step 13.

Add 200 µl TE Buffer and leave 2-3 hours in the oven at 37°C

AMOUNT
200 µl Additional info: TE Buffer
TEMPERATURE
37 °C Additional info: