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## Extraction method F (CR)



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## Abstract

Gen-IALFirst All-tissue DNA extraction kit -This protocol provides an efficient DNA extraction and purification of historic sample (tissue material)

## Materials

### MATERIALS

 Proteinase K

 Isopropanol

 Ethanol

## Before start

Ancient DNA lab

## Extraction

- 1 Cut tissue (25-50 mg) into small pieces.
- 2 Add 500  $\mu$ L Lyse 1.  
 500  $\mu$ L Lyse 1
- 3 Add 50  $\mu$ L Lyse 2.  
 50  $\mu$ L Lyse 2
- 4 Add 10  $\mu$ L Proteinase K.  
 10  $\mu$ L Proteinase K
- 5 Incubate in a thermomixer at 65°C for 12 hours.  
 12:00:00 Incubation at 65°C
- 6 Centrifuge for 10 min at 13000rpm.  
 00:10:00 Centrifugation
- 7 Discard supernatant without disturbing the pellet.
- 8 Add 375  $\mu$ L Lyse 3  
 375  $\mu$ L Lyse 3
- 9 Vortex for 20 sec.  
 00:00:20 Vortex
- 10 Leave it for 5 min in the freezer (-20°C)  
 00:05:00 Freezer
- 11 Centrifuge 20 min at 13000rpm.  
 00:20:00 Centrifugation
- 12 Transfer the supernatant in a new tube.
- 13 Add 640  $\mu$ L isopropanol.



 640 µL Isopropanol

14 Mix by inversion.

15 Centrifuge for 15 min at 13000 rpm.

 00:15:00 Centrifugation

16 Remove the supernatant (don't touch the pellet).

17 Add 150 µl ethanol.

 150 µL Ethanol

18 Centrifuge for 5 min at 13000 rpm.

 00:05:00 Centrifugation

19 Remove the ethanol with a pipette (don't touch the pellet).

20 Dry the pellet (37°C open lid max 5 min).

 00:05:00 Dry

21 Re-suspend the pellet in 100 µl Buffer AE.

 100 µL Buffee AE

22 Measure DNA concentration on Nanodrop (Thermo Fischer Scientific, Darmstadt, DE).