

# Extraction of DNA From Bone or Teeth Using the EZ1<sup>®</sup> DNA Investigator<sup>®</sup> Kit

This protocol is designed to enable efficient recovery of inhibitor-free DNA from bone and teeth samples using the EZ1 Advanced or EZ1 Advanced XL with the EZ1 DNA Investigator Kit. The EZ1 Advanced and EZ1 Advanced XL are intended to be used only in combination with QIAGEN kits indicated for use with these instruments for the applications described in the kit handbooks.

## Equipment and reagents to be supplied by user

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, consult the appropriate safety data sheets (SDSs), available from the product supplier.

### For all users:

- EZ1 DNA Investigator Kit (cat. no. 952034)
- 0.5 M EDTA, pH 8.0
- 3 M sodium acetate (NaOAc), pH 5.0
- Buffer MTL (cat. no. 1023430)
- Pipet tips  
**Note:** We recommend pipet tips with aerosol barriers to prevent cross-contamination
- Centrifuge
- Thermal mixer or orbital incubator

### For EZ1 Advanced users:

- EZ1 Advanced instrument
- EZ1 Advanced DNA Investigator Card (cat. no. 9018302)

### For EZ1 Advanced XL users:

- EZ1 Advanced XL instrument
- EZ1 Advanced XL DNA Investigator Card (cat. no. 9018699)

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## Important points before starting

- If using the EZ1 DNA Investigator Kit for the first time, read “Important Notes” in the *EZ1 DNA Investigator Handbook*.
- Sodium acetate (NaOAc) is required for this protocol but is not included in the EZ1 DNA Investigator Kit. NaOAc should be purchased separately.
- EDTA is required for this protocol, but is not included in the EZ1 DNA Investigator Kit. EDTA should be purchased separately.
- Buffer MTL is required for this protocol and for the Large-Volume EZ1 Protocol used to complete sample processing. Buffer MTL is not included in the EZ1 DNA Investigator Kit and should be purchased separately (cat. no. 1023430).

## Things to do before starting

Heat a thermal mixer or orbital incubator to 56°C for the proteinase K digest in step 7.

## Procedure

1. Remove and discard the bone or teeth surfaces. Grind the remaining bone or tooth root to a fine powder.  
**Note:** Grind using a metal blender half-filled with liquid nitrogen. Alternatively, grind using the TissueLyser II and the Grinding Jar Set, S. Steel. The TissueLyser II instrument is not included in the EZ1 DNA Investigator Kit and should be purchased separately (cat. no. 85300).  
**Note:** When using the TissueLyser II, transfer the bone or tooth sample and the ball into the grinding jar. Pour liquid nitrogen into the grinding jar over the ball and bone or tooth fragments. Allow the temperature to equilibrate (i.e., until the liquid nitrogen stops boiling). Decant the excess liquid nitrogen, then close the grinding jar with the lid and transfer it to the TissueLyser II. Grind the bone at 30 Hz for 1 min or until the sample is pulverized (grinding times depend on type, condition and size of sample).
2. Place up to 150 mg of powdered bone into a 2 ml sample tube. Do not exceed the amount of bone powder. If higher overall yields are required, we recommend processing additional extractions and to pool and concentrate the eluates.
3. Add 225 µl Buffer G2.
4. Add 25 µl proteinase K.
5. Add 250 µl 0.5 M EDTA, pH 8.0.
6. Mix by inverting the 2 ml tube several times.

7. Place the tube into the thermal mixer or heated orbital incubator, and incubate with constant motion at 56°C for 24 hours.
8. Centrifuge at 6000 rpm for 4 min to pellet any remaining debris. Transfer the supernatant to an EZ1 sample tube.
9. Add 400 µl Buffer MTL to each 2 ml EZ1 sample tube.
10. Add 50 µl 3 M NaOAc, pH 5.0 to each 2 ml EZ1 sample tube.
11. Add 1 µl carrier RNA to each 2 ml EZ1 sample tube.

**Note:** A master mix of Buffer MTL, NaOAc and carrier RNA can be prepared and used within the same day.

12. Continue with Protocol: DNA Purification (Large-Volume Protocol) in the *EZ1 DNA Investigator Handbook*.

For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit handbook or user manual. QIAGEN kit handbooks and user manuals are available at [www.qiagen.com](http://www.qiagen.com) or can be requested from QIAGEN Technical Services or your local distributor. The EZ1 DNA Investigator Kit is intended for molecular biology applications. This product is not intended for the diagnosis, prevention, or treatment of a disease.