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Product Datasheet

Version：1.0, Issue Date：2026-10-27

Japanese Datasheet https://www.biodynamics.co.jp/products/
English Datasheet https://bdl-biodynamics.com/products/

Product Name：Extrap™ Bone/Teeth DNA Kit <DNA Extraction Kit>
Product Code：212-010
Size：1 kit (20 preps)

Product Name：Solution A
Product Code：212-011
Size：200 mL

Product Name：Solution B
Product Code：212-012
Size：14 mL

Product Name：Solution C
Product Code：212-013
Size：10 mL

For Research Use Only

Description

This kit is designed for the extraction of genomic DNA from hard tissues such as bones and teeth.

This kit can be extracted from highly degraded forensic specimens, including samples buried in soil for extended periods and human remains recovered following major disasters, such as fires, floods, hurricanes, earthquakes, and aircraft accidents. The extracted DNA can be used for mitochondrial DNA (mtDNA) sequencing and short tandem repeat (STR) profiling.
This kit enables DNA extraction directly from whole teeth or small bone fragments without the need for pulverization.

NOTE：Depending on the condition and size of the specimen, fragmentation may be required prior to DNA extraction. For large teeth or bone samples, use an electric cutter or similar tool to cut the sample into pieces approximately $1^{L} \times 0.5^{W} \times 0.5^{H}$ cm in size before extraction.

Kit Components and Storage Condition

Kit Components	Size	Storage Condition
① Solution A	200 mL	Room Temperature
② Solution B	14 mL	Room Temperature
③ Solution C	10 mL	Room Temperature
④ PhazeKeep, Light	20 tubes	Room Temperature

Important Notice

Solution A, B and C may cause irritation upon contact with the eyes or skin. Avoid direct contact with the solution. Wear appropriate personal protective equipment, including protective gloves and eye protection, when handling this solution.

Product Shelf Life

1 year after the date of receipt

Additional Materials Required

1. Manufacturer-Specified Reagents

The DNA purification procedure using this product requires certain QIAGEN products listed below. Please ensure that these reagents are available to fully utilize the performance of this product.

Product Name	Catalog Code	Supplier
QIAamp DNA Blood Mini Kit (50)	51104	QIAGEN

The catalog numbers for the recommended reagents listed above are current as of August 24, 2026.

2. Other Reagents

☐ Proteinase K
☐ TE-saturated phenol
☐ Sterile distilled water
☐ 100%Ethanol (Absolute ethanol, ≥99.5%)
☐ Sodium hypochlorite solution for laboratory work surface cleaning

3. Consumables

☐ Sterile plastic tray
☐ Sterile disposable 15 mL tubes
☐ Sterile disposable 1.5 mL tubes
☐ Sterile disposable 10 mL serological pipettes
☐ Sterile disposable pipette tips
☐ Laboratory gloves
☐ Laboratory coat
☐ Surgical mask
☐ Disposable hair cap
☐ Laboratory paper towels
☐ Laboratory tissues
☐ Neutral detergent for specimen cleaning (dishwashing detergent)
☐ Toothbrushes for specimen cleaning
☐ Microscope slide holders
☐ Dental probes

4. Equipment

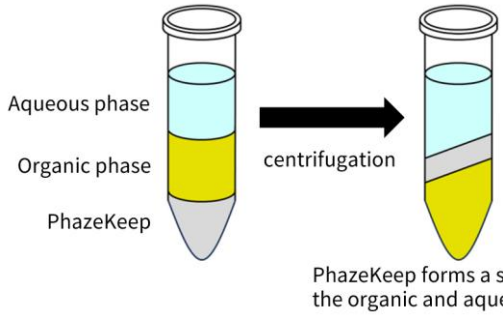
☐ Hair dryer for air-drying
☐ Incubator (settable to 23°C, 37°C, and 56°C)
☐ Thermometer

☐ Electronic pipette controller
☐ Centrifuge
☐ Micropipettes
☐ Vortex mixer

Introduction

1. Overview of the DNA Extraction Workflow

Preparation of the Laboratory Environment
↓
Sample Cleaning
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Decalcification
↓
Protein Digestion
↓
DNA Extraction using PhazeKeep Light
↓
DNA Purification



PhazeKeep Light forms a seal between the organic and aqueous phase during phenol/chloroform extraction, enabling easier handling and improving nucleic acid recovery efficiency. It reduces the carryover of proteins such as collagen from bone samples. PhazeKeep does not interfere with standard nucleic acid restriction enzyme reactions and does not adsorb to or degrade DNA when present during heat inactivation at 65°C for 30 minutes.

2. Preparation of the Laboratory Environment

（1）Physical Separation

It is recommended that the DNA extraction area and PCR amplification area be physically separated. To minimize the risk of cross-contamination, personnel should avoid entering the DNA extraction area after performing PCR amplification.
In addition, incubators and centrifuges should be placed near the appropriate workbench to minimize the need to transport samples between work areas.

（2）Personnel Preparation

During DNA extraction, contamination with DNA from sources other than the samples may result in the unintended DNA being co-amplified during PCR. In addition, contamination with substances can inhibit PCR reaction. Therefore, personnel should take appropriate precautions to prevent contamination, including wearing a laboratory coat, washing hands, and using disposable gloves, hair caps, masks, and other appropriate personal protective equipment (PPE). Disposable gloves should be changed frequently and replaced with a fresh pair as needed.

（3）Laboratory Work Surface Cleaning

Remove any unnecessary items from the work surface to ensure sufficient space for the procedure. Apply sodium hypochlorite solution to laboratory paper towels and wipe down the work surface thoroughly. Then wipe the surface with pure water to remove any residual sodium hypochlorite solution.

（4）Preparation of Laboratory Equipment and Instruments

Wipe pipettes, centrifuges, incubators, and other laboratory equipment with sodium hypochlorite solution, followed by pure water, in the same manner as the work surface. Wash sample cleansing tools, such as toothbrush and dental explorer, with sterilized distilled water. Tubes and pipette tips must be disposable and should not be reused.

（5）General Precautions

Please follow the rules of your institution or laboratory. Dispose of samples and reagents in accordance with local regulations.

Protocol

1. Sample Cleaning

（1） Preparation of Neutral Detergent

Dilute the neutral detergent prepared for sample cleaning (dishwashing detergent) to approximately 0.2% with sterile distilled water. Transfer the required amount of the diluted detergent solution into a beaker for use.

（2） Cleaning with a Neutral Detergent

Clean the surface of the teeth or bone using a toothbrush moistened with the diluted neutral detergent. Remove dirt from small or hard-to-reach areas by gently scraping it with a dental explorer or similar instrument.

（3） Washing with Sterile Distilled Water

Rinse the sample with sterile distilled water at least five times. If any residual detergent remains, continue rinsing with sterile distilled water until all the detergent has been removed.

（4） Washing with 100% Ethanol

Rinse the sample with 100% ethanol and place it on a sterile plastic tray.

（5） Air-Drying with a Hair Dryer

Use a hair dryer to remove moisture from both the surface and interior of the sample.

（6） Sample Fragmentation*

Using an electric cutter or similar tool, cut the sample into pieces approximately $1^{L} \times 0.5^{W} \times 0.5^{H}$ cm in size, or approximately 0.5 g in weight so that the sample can be fully immersed in the respective solutions during the decalcification and protein digestion steps.

＊Depending on the size and condition of the sample used for extraction, DNA extraction may not be successful. In such cases, it is recommended to fragment the sample before proceeding with the subsequent extraction steps.

Workflow Branch
If DNA extraction is to be performed immediately, proceed to “**2. Decalcification step.**”
If DNA extraction will not be performed immediately, proceed to the following “**(7) Storage step**”.

（7） Storage

If DNA extraction will not be performed immediately, place the air-dried sample in an empty sterile tube or equivalent container and store it at 23°C in the dark.

2. Decalcification

（1） Set the Incubator Temperature

Set the incubator temperature to 23°C.

（2） Addition of Solution A

Place the sample in a 15 mL tube. Add 10 mL of Solution A and mix by gently inverting the tube.

（3） Incubation

Place the 15 mL tube containing the sample and Solution A in a tube rack and incubate at 23°C for 16 hours. Mix once by gently inverting the tube approximately 15 hours after the start of incubation.

（4）Set the Incubator Temperature
After incubating for 16 hours in Step (3), set the incubator temperature to 37°C.

（5）Addition of Solution B
Confirm that the incubator has reached 37°C.
(Approximately 15 minutes; the time required may vary depending on the equipment in your lab. Always verify the temperature with a thermometer before proceeding.)
Add 0.6 mL of Solution B, cap the 15 mL tube, and mix by gently inverting the tube three times. Place the tube in a tube rack and incubate at 37°C for 2 hours. During incubation, mix the tube by gently inverting it 3-4 times.

NOTE：
Confirm that the incubator has reached 37°C before adding Solution B.
If precipitate is observed in Solution B, place the solution in a 37°C incubator and use it only after confirming that the precipitate has completely dissolved.

（6） Set the Incubator Temperature
After removing the sample from the incubator following Step (5), set the incubator temperature to 56°C.

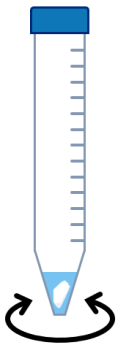
（7） Removal of Solutions A and B
Remove the solutions (Solutions A and B) using a sterile disposable 10 mL serological pipette. After briefly centrifuging the tube, remove any remaining solution using a sterile disposable pipette tip.
Then centrifuge the tube at 2,000 rpm for 2 minutes and completely remove the remaining solution using a sterile disposable pipette tip.

NOTE：
Thoroughly remove any solution trapped within the cavities of the teeth or bone by centrifugation at this step to avoid increasing the total volume after adding Solution C in Step 3-(1).
Completely remove Solutions A and B, as any residual solution may interfere with subsequent steps.

3．Protein Digestion

（1） Addition of Solution C
Add 400 μL of Solution C to the 15 mL tube from Step 2-(7). Avoid allowing Solution C to contact the inner wall of the tube.

（2） Addition of Proteinase K
Add 50 μL of Proteinase K (20 mg/mL) to the tube. Place the tube in a tube rack and incubate at 56°C for 3 hours. During incubation, gently mix the lower portion of the tube 3-4 times, as shown in the image on the right.
The extent of sample dissolution varies depending on the sample. See Item 2 in the “Troubleshooting” section on the final page.



4．DNA Extraction

（1） Preparation PhazeKeep Light

Confirm that PhazeKeep Light (gel) is at the bottom of the tube before using it. If the gel is not at the bottom, briefly spin down the tube (3,000 × g for approximately 30 seconds).

（2） Addition of TE-Saturated Phenol

Transfer **400 μL** of the reaction mixture from Step 3-(2) into the tube containing PhazeKeep Light. Add 400 μL of sterile distilled water and **800 μL** of TE-saturated phenol.
Gently mix by inverting the tube 15 times.

Caution：

・Do not add more than 400 μL of the reaction mixture from Step 3-(2) to the tube containing PhazeKeep Light.

・TE-saturated phenol is a hazardous and corrosive substance. Contact may cause injury to the skin and eyes. It may also be absorbed through the skin and cause poisoning and may have a depressant effect on the central nervous system. Always wear laboratory gloves and handle with care.

（3） Centrifugation

Centrifuge at 13,000 rpm for 15 minutes.
（Alternatively, centrifuge at 15,000 rpm for approximately 10 minutes or at 10,000 rpm for approximately 30 minutes.）

NOTE：

・When using a centrifuge with a fixed-angle rotor, orient the tubes in the same direction as when centrifuging in Step 4-(1), “Preparation of PhazeKeep Light.”

・Although a barrier will form, a small amount of PhazeKeep Light may remain at the bottom of the tube. If this occurs, proceed directly to the next step.

（4） Recovery of the Aqueous Phase (DNA Layer)

Using a pipette, transfer approximately 800 μL of the aqueous phase (DNA layer) to a 15 mL tube.

5．DNA Purification

The DNA purification procedure uses QIAGEN QIAamp DNA Blood Mini Kit. For precautions and safety information concerning QIAGEN products, please refer to the QIAamp DNA Blood Mini Kit Protocol.

Important Notice：

The QIAamp DNA Blood Mini Kit is sold as a 50-reaction kit; however, because the protocol below requires a relatively large volume of Buffer AL the kit provides sufficient reagents for approximately 15 reactions when used according to this protocol. If the supplied Buffer AL is insufficient, please purchase additional Buffer AL separately (Catalog Code 19075).

（1） Add an equal volume (approximately 800 μL) of Buffer AL (supplied with the QIAamp DNA Blood Mini Kit) to the solution from Step 4-(4). Vortex for 15 seconds, then briefly spin down the tube.

（2） Add an equal volume (approximately 800 μL) of 100% ethanol to the solution from Step 4-(4). Vortex for 15 seconds, then briefly spin down the tube.

（3） Carefully transfer 600 μL of the solution from Step 5-(2) to a Mini spin column (supplied with the QIAamp DNA Blood Mini Kit), taking care not to wet the rim of the column. Close the cap and

centrifuge at 8,000 rpm for 1 minute.

（4） After centrifugation, remove the flow-through from the collection tube using a sterile disposable pipette tip.

（5） Transfer 600 μL of the remaining solution from Step 5-(2) (approximately 1,800 μL) to the same Mini spin column and centrifuge at 8,000 rpm for 1 minute.

（6） After centrifugation, remove the flow-through from the collection tube using a sterile disposable pipette tip.

（7） Transfer 600 μL of the remaining solution from Step 5-(2) (approximately 1,200 μL) to the same Mini spin column and centrifuge at 8,000 rpm for 1 minute.

（8） After centrifugation, remove the flow-through from the collection tube using a sterile disposable pipette tip.

（9） Transfer the remaining solution from Step 5-(2) (approximately 600 μL) to the same Mini spin column and centrifuge at 8,000 rpm for 1 minute.

（10） After centrifugation, discard the collection tube containing the flow-through and transfer the Mini spin column to a new collection tube supplied with the QIAamp DNA Blood Mini Kit.

（11） Carefully open the cap of the Mini spin column. Add 500 μL of Buffer AW1 (supplied with the QIAamp DNA Blood Mini Kit), taking care not to wet the rim of the column. Close the cap and centrifuge at 8,000 rpm for 1 minute.

（12） After centrifugation, discard the collection tube containing the flow-through and transfer the Mini spin column to a new collection tube supplied with the QIAamp DNA Blood Mini Kit.

（13） Carefully open the cap of the Mini spin column. Add 500 μL of Buffer AW2 (supplied with the QIAamp DNA Blood Mini Kit), taking care not to wet the rim of the column. Close the cap and centrifuge at 8,000 rpm for 1 minute.

（14） Centrifuge at 13,000 rpm for an additional 1 minute.

（15） After centrifugation, discard the collection tube containing the flow-through and transfer the Mini spin column to a new 1.5 mL disposable tube.

（16） Add 50 μL of Buffer AE (supplied with the QIAamp DNA Blood Mini Kit) directly to the column membrane. Incubate at room temperature (15-20°C) for 1 minute.

（17） Centrifuge at 8,000 rpm for 1 minute.

（18） Use the resulting eluate as the DNA extract for PCR-based applications, including mitochondrial DNA sequencing and STR profiling.

Troubleshooting

【1】	Precipitate is observed in Solution B used in the decalcification step.	Incubate at 37 °C until the precipitate has completely dissolved.
【2】	The sample does not completely dissolve during the protein digestion step.	Complete dissolution of the specimen is generally not expected. However, to maximize DNA recovery, it is important to fragment large specimens into pieces approximately $1^{L} \times 0.5^{W} \times 0.5^{H}$ cm in size and to perform the washing and decalcification steps according to the protocol.
【3】	PhazeKeep Light does not separate properly.	・ Increase the centrifugation speed or extend the centrifugation time. ・Store PhazeKeep Light at room temperature. Do not freeze. ・Briefly spin down the tube before use to bring PhazeKeep Light to the bottom of the tube.