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EZ1&2® DNA Investigator® Kit Handbook

For automated purification of DNA from forensic and human ID samples using EZ1® instruments

Sample to Insight

QIAGEN

Contents

Kit Contents

EZ1&2 DNA Investigator Kit	**(48)**
Catalog no.	**952034**
No. of preps	**48**
Reagent Cartridge, DNA Investigator*	48
Disposable Tip Holders	50
Disposable Filter-Tips	50
Sample Tubes (2 ml)	50
Elution Tubes (1.5 ml)	50
Buffer G2	2 x 11 ml
Proteinase K	1 x 1.2 ml
Carrier RNA	1 x 310 µg
Q-Card‡	1
Quick-Start Protocol	1

* Contains a guanidine salt. Not compatible with disinfectants containing bleach. See page 6 for Safety Information.

‡ The information encoded in the bar code on the Q-Card is needed for reagent data tracking using the EZ1 Advanced, EZI Advanced XL, or EZ2 Connect Fx instruments.

Additional filter-tips and tip holders are available separately. Additional Buffer G2, required for some protocols, is available separately. See page 46 for Ordering Information.

Shipping and Storage

The EZ1&2 DNA Investigator Kit is shipped at ambient temperature. All buffers and reagents can be stored at room temperature (15–25°C). Do not freeze the reagent cartridges. When stored properly, the reagent cartridges are stable until the expiration date on the Q-Card. Lyophilized carrier RNA is stable until the expiration date on the Q-Card when stored at room temperature.

The ready-to-use Proteinase K solution is stable for up to 1 year after delivery when stored at room temperature.

Intended Use

The EZ1&2 DNA Investigator Kit is intended for molecular biology applications in forensic, human identity, and paternity testing. The EZ1&2 DNA Investigator Kit is intended to be used with EZ1 Advanced, BioRobot® EZ1, EZ1 Advanced XL, and EZ2 Connect Fx instruments. This product is neither intended for the diagnosis, prevention, or treatment of a disease, nor has it been validated for such use either alone or in combination with other products. All due care and attention should be exercised in the handling of the products. We recommend all users of QIAGEN® products to adhere to the NIH guidelines that have been developed for recombinant DNA experiments, or to other applicable guidelines.

Safety Information

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, please consult the appropriate safety data sheets (SDSs). These are available online in convenient and compact PDF format at **www.qiagen.com/safety**, where you can find, view, and print the SDS for each QIAGEN kit and kit component.

CAUTION 	DO NOT add bleach or acidic solutions directly to the sample-preparation waste.

Quality Control

In accordance with QIAGEN's ISO-certified Quality Management System, each lot of EZ1&2 DNA Investigator Kits is tested against predetermined specifications to ensure consistent product quality. Functional QC testing ensures that the EZ1&2 DNA Investigator Kit meets the high standards required by forensic scientists. The EZ1&2 DNA Investigator Kit meets ISO 18385 requirements.

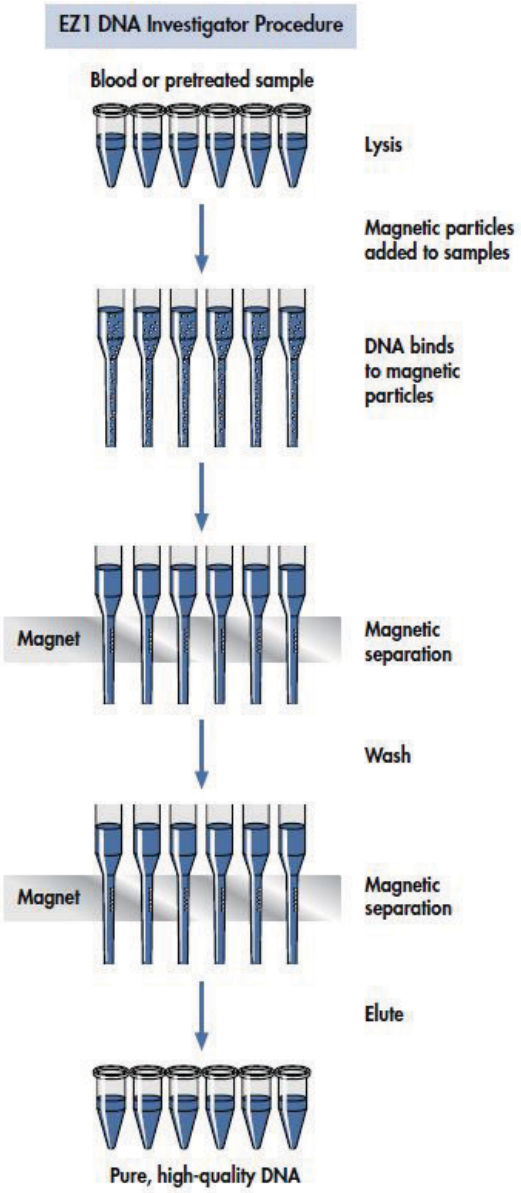
Introduction

EZ1 instruments and the EZ1&2 DNA Investigator Kit reproducibly automate purification of genomic DNA from 1–6 samples (EZ1 Advanced and BioRobot EZ1) or 1–14 samples (EZ1 Advanced XL) encountered in forensic, human identity, and biosecurity applications. Purification is efficient and purified DNA performs well in downstream analyses, such as quantitative PCR and STR analysis, with high signal-to-noise ratios. Magnetic-particle technology provides high-quality DNA that is suitable for direct use in downstream applications, such as STR analysis or other enzymatic reactions. EZ1 instruments perform all steps of the sample preparation procedure, and the user can choose sample input volumes of 200 µl or 500 µl, allowing purification from varying amounts of starting material. Up to 6 samples (BioRobot EZ1, EZ1 Advanced) or up to 14 samples (EZ1 Advanced XL) can be processed in a single run.

The EZ1&2 DNA Investigator Kit is also compatible with EZ2 Connect Fx instrument. Please refer to the corresponding handbook: "*EZ1&2 DNA Investigator Kit Handbook — For automated purification of DNA from forensic and human ID samples using the EZ2 Connect Fx*".

Principle and procedure

Magnetic-particle technology combines the speed and efficiency of silica-based DNA purification with the convenient handling of magnetic particles (see flowchart in the next page). DNA is isolated from lysates in one step through its binding to the silica surface of the particles in the presence of a chaotropic salt. The particles are separated from the lysates using a magnet. The DNA is then efficiently washed and eluted in the user's choice of either water or TE buffer. The user can choose elution volumes of 40 µl (EZ1 Advanced XL only), 50 µl, 100 µl, or 200 µl.


EZ1 DNA Investigator Procedure
Blood or pretreated sample
Lysis
Magnetic particles added to samples
DNA binds to magnetic particles
Magnet
Magnetic separation
Wash
Magnet
Magnetic separation
Elute
Pure, high-quality DNA

Description of protocols

This handbook contains 2 types of protocols (Table 1).

- Pretreatment protocols detail the preliminary steps, such as Proteinase K digestion, prior to processing on the EZ1 instrument.
- DNA purification protocols describe setting up the EZ1 instrument and starting a fully automated run.

Pretreatment protocols

Since the type of samples that can be processed using the EZ1&2 DNA Investigator Kit can vary greatly, there is also a variety of different pretreatments, optimized for specific sample types.

DNA purification protocols

There are 3 DNA purification protocols, which can be used in conjunction with the pretreatment protocols. Within each protocol, the user can specify elution in water or TE buffer, with elution volumes of 40 µl (EZ1 Advanced XL only), 50 µl, 100 µl, or 200 µl. The standard Protocol: DNA Purification (Trace Protocol) can be used with all sample types (page 31).

In the Protocol: DNA Purification ("Tip Dance" Protocol), page 34, the filter-tip moves back-and-forth relative to the worktable platform while pipetting. This enables processing of solid materials, such as swabs, fabrics, blood discs, or cigarette butts, directly in the sample tube. There is generally no need for prior centrifugation to remove solid materials that could clog the tip. However, when processing fluffy sample material such as cotton wool, we recommend removing solid material if you cannot process a replicate sample or if the sample material is precious.

The Protocol: DNA Purification (Large-Volume Protocol), page 37, enables fully automated processing of starting volumes up to 500 µl. This allows efficient DNA purification from dilute

samples with low concentrations of DNA, such as diffuse stains, as well as purification from samples that require larger volumes for thorough lysis. The ability to process larger sample volumes, with the same elution volume as the standard Trace Protocol, enables higher yields of more concentrated DNA for greater sensitivity in downstream applications.

Table 1. Protocol information for different sample types

Sample type	Pre-treatment protocols	Purification protocols	Sample amount	Buffer G2	Proteinase K	DTT 1 mM
Blood/saliva	Page 23	Trace	Up to 50 µl	140–190 µl	10 µl	no
FTA® cards	Page 23	Trace or Tip Dance	4 x 3 mm punches	290 µl	10 µl	no
Surface swabs	Page 23	Trace or Tip Dance	1 swab	290 µl	10 µl	no
Chewing gum	Page 23	Trace or Tip Dance	Up to 40 mg	190 µl	10 µl	no
Cigarette butts	Page 23	Trace or Tip Dance	1 cm^2	190 µl	10 µl	no
Paper/similar materials	Page 23	Trace or Tip Dance	0.5–2.5 cm^2	190 µl	10 µl	no
Nail scrapings	Page 23	Trace	Up to 40 mg	190 µl	10 µl	no
Nail clippings	Page 23	Trace	1	160 µl	20 µl	20 µl
Hair	Page 23	Trace	0.5–1 cm	160 µl	20 µl	20 µl
Tissues	Page 23	Trace	Up to 10 mg	190 µl	10 µl	no
Blood or saliva stains	Page 23	Trace or Tip Dance	0.5 cm^2	290 µl	10 µl	no
Semen stains	Page 23	Trace or Tip Dance	0.5 cm^2	270 µl	10 µl	20 µl
Large volume	Page 23	Large-Volume	Varies	475 µl	25 µl	no
Large volume semen	Page 23	Large-Volume	Varies	455 µl	25 µl	20 µl
Sexual assault samples	Page 25	Trace	Varies	Up to 2.5 ml*	20 µl	40 µl
Bones or teeth	Page 27	Large-Volume	150–200 mg	0.5 M EDTA	25 µl	no
Soil	Page 29	Trace or Tip Dance	Up to 0.5 g	100 µl	no	no

* Depends on number of sperm pellet washes.

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.

All protocols

- Thermomixer, heating block, or water bath
- Vortexer
- Pipets and pipet tips (to prevent cross-contamination, we strongly recommend the use of pipet tips with aerosol barriers)

For BioRobot EZ1 users

- EZ1 DNA Investigator Card (cat. no. 9016387)

For EZ1 Advanced users

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

For EZ1 Advanced XL users

- EZ1 Advanced XL DNA Investigator Card (cat. no. 9018699) or EZ1 Advanced XL DNA Investigator Flip-Cap Card (cat. no. 9022763)
- Optional: EZ1 Advanced XL Flip-Cap Tube Rack (cat. no. 9022818)

For EZ1 Advanced and EZ1 Advanced XL users

For documentation purposes, one of the following is required:

- EZ1 Advanced Communicator Software (supplied with the EZ1 Advanced and EZ1 Advanced XL instruments), PC (can be connected with up to 4 EZ1 Advanced and EZ1 Advanced XL instruments), and monitor (cat. no. for PC and monitor 9016643)

- EZ1 Advanced Communicator Software (supplied with the EZ1 Advanced and EZ1 Advanced XL instruments) and your own PC and monitor (connection with up to 4 EZ1 Advanced and EZ1 Advanced XL instruments not recommended)

For purification of DNA from epithelial cells mixed with sperm cells

- Buffer G2 (cat. no. 1014636)
- 1 M dithiothreitol (DTT), forensic grade quality (cat. no. 1117316)
- Microcentrifuge

For purification of DNA from hair

- 1 M dithiothreitol (DTT), forensic grade quality

For purification of DNA from bones or teeth

- 0.5 M EDTA, pH 8.3
- Liquid nitrogen
- 3 M sodium acetate (NaOAc), pH 5.0
- Buffer MTL (cat. no. 19112)
- Pipette tips (with aerosol barriers recommended)
- Centrifuge
- Thermal mixer or orbital incubator
- TissueLyser II (cat. no. 85300), with the Grinding Jar Set, S. Steel (cat. no. 69985), or an equivalent bead mill

For DNA purification, Large-Volume Protocol

- Buffer MTL (54 ml) (cat. no.19112)

Important Notes

Starting material

The amount of starting material for use in EZ1&2 DNA Investigator procedures can vary greatly, depending on the amount of DNA in the sample. Specific guidance for starting amounts is given in the individual protocols and Table 1. EZ1 instruments can process 200 µl pretreated samples using the Trace Protocol (page 31) or the "Tip Dance" Protocol (page 34) for DNA purification. With the Large-Volume Protocol (page 37), up to 500 µl pretreated samples can be processed.

Purification of low amounts of DNA

For purification of DNA from very small amounts of sample, such as low volumes of blood (<10 µl) or forensic casework samples, we recommend adding carrier RNA. For samples containing larger amounts of DNA, addition of carrier RNA is optional. Add 310 µl TE buffer or water to the tube containing 310 µg lyophilized carrier RNA to obtain a solution of 1 µg/µl. Dissolve the carrier RNA thoroughly, divide in to conveniently sized aliquots, and store at –30 to –15°C. Do not freeze-thaw the aliquot of carrier RNA more than 3 times. Carrier RNA should be added to the sample after the lysis is completed to avoid degradation. When using the Large-Volume Protocol, carrier RNA can be added to buffer MTL prior to setting up the instrument run. Use the mixture of MTL and carrier RNA within the same day.

Working with EZ1 Instruments

The main features of the EZ1 instruments include:

- Purification of high-quality nucleic acids from 1–6 or 1–14 samples per run
- Small footprint to save laboratory space
- Preprogrammed EZ1 Cards containing ready-to-use protocols for nucleic acid purification
- Prefilled, sealed reagent cartridges for easy, safe, and fast setup of EZ1 instruments

- Complete automation of nucleic acid purification, from opening of reagent cartridges to elution of nucleic acids, with no manual centrifugation steps

Additional features of the EZ1 Advanced and EZ1 Advanced XL include:

- Bar code reading and sample tracking
- Kit data tracking with the Q-Card provided in the kit
- UV lamp to help eliminate sample carryover from run-to-run and to allow pathogen decontamination on the worktable surfaces

  Note: UV decontamination helps to reduce possible pathogen contamination of the EZ1 Advanced and EZ1 Advanced XL worktable surfaces. The efficiency of inactivation has to be determined for each specific organism and depends, for example, on layer thickness and sample type. QIAGEN cannot guarantee complete eradication of specific pathogens.

EZ1 Cards, EZ1 Advanced Cards, and EZ1 Advanced XL Cards

Protocols for nucleic acid purification are stored on preprogrammed EZ1 Cards (integrated circuit cards). The user simply inserts an EZ1 Advanced XL Card into the EZ1 Advanced XL; an EZ1 Advanced Card into the EZ1 Advanced; or an EZ1 Card into the BioRobot EZ1, and the instrument is then ready to run a protocol (see Figure 1). The availability of various protocols increases the flexibility of EZ1 instruments. Note that the Flip-Cap Tube Rack can only be used with the EZ1 DNA Investigator Flip-Cap card on an EZ1 ADV XL.

Figure 1. Ease of protocol setup using EZ1 Cards. Inserting an EZ1 Card, containing a protocol, into an EZ1instrument. The instrument should only be switched on after an EZ1 Card is inserted. EZ1 Cards should not be exchanged while the instrument is switched on.

The EZ1&2 DNA Investigator Kit requires use of the EZ1 Advanced XL DNA Investigator Card with the EZ1 Advanced XL, use of the EZ1 Advanced DNA Investigator Card with the EZ1 Advanced, or use of the EZ1 DNA Investigator Card with the BioRobot EZ1. These EZ1 Cards contain protocols for purification of DNA from forensic and human identity samples.

EZ1 instruments should only be switched on after an EZ1 Card is inserted. Make sure that the EZ1 Card is completely inserted (see Figure 2); otherwise, essential instrument data could be lost, leading to a memory error. EZ1 Cards should not be exchanged while the instrument is switched on.

Figure 2. Complete insertion of EZ1 Card. The EZ1 Card must be completely inserted before the EZ1 instrument is switched on.

Reagent cartridges

Reagents for the purification of nucleic acids from a single sample are contained in a single reagent cartridge (see Figure 3). Each well of the cartridge contains a particular reagent, such as magnetic particles, lysis buffer, wash buffer, or elution buffer. Since each well contains only the required amount of reagent, generation of waste due to leftover reagent at the end of the purification procedure is avoided.

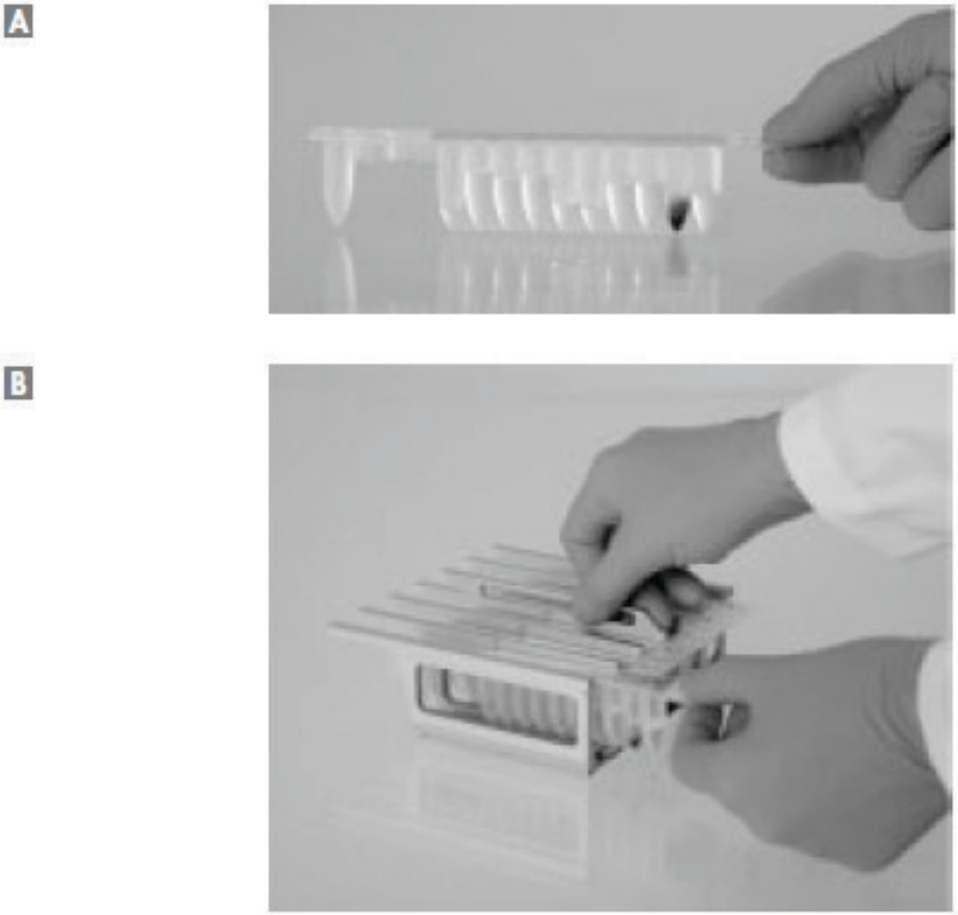


Figure 3. Ease of setup using reagent cartridges. (A) A sealed, prefilled reagent cartridge. Fill levels vary, depending on the type of reagent cartridge. (B) Loading reagent cartridges into the cartridge rack. The cartridge rack itself is labeled with an arrow to indicate the direction in which reagent cartridges must be loaded.

Worktable

The worktable of EZ1 instruments is where the user loads samples and the components of the EZ1&2 DNA Investigator Kit (see Figure 4). For the EZ1 Advanced XL, 2 sample racks are available: The standard rack for use with the screw cap sample and elution tubes provided

with the EZ1&2 DNA Investigator Kit, and a Flip-Cap Tube Rack that can be used with both screw cap and Flip-Cap Tubes. Please note that the Flip-Cap Tube Rack can only be used with the EZ1 DNA Investigator Flip-Cap Card. Details on worktable setup are provided in the protocols in this handbook and the display also shows protocol status during the automated purification procedure.

Figure 4. Typical EZ1 worktable.

1. First row: Elution tubes (1.5 ml) are loaded here.
2. Second row: Tip holders containing filter-tips are loaded here.
3. Third row: Tip holders containing filter-tips are loaded here. (In some protocols, this row is empty or loaded with 2 ml Sarstedt tubes).
4. Fourth row: Sample tubes (2 ml) are loaded here.
5. Reagent cartridges are loaded into the cartridge rack.
6. Heating block with 2 ml tubes in the reagent cartridges for lysis.

Data tracking with the EZ1 Advanced and EZ1 Advanced XL

The EZ1 Advanced and EZ1 Advanced XL enable complete tracking of a variety of data for increased process control and reliability. The EZ1&2 Kit lot number and expiration date are entered at the start of the protocol using the Q-Card bar code. A user ID and the Q-Card bar code can be entered manually via the keypad or by scanning bar codes using the handheld bar code reader. Sample and assay information can also be optionally entered at the start of the protocol. At the end of the protocol run, a report file is automatically generated. The EZ1 Advanced and EZ1 Advanced XL can store up to 10 result files, and the data can be transferred to a PC or directly printed on a printer (for ordering information, see "Equipment and Reagents to Be Supplied by User" on page 11).

To receive report files on a PC, the EZ1 Advanced Communicator software needs to be installed. The software receives the report file and stores it in a folder that you define. After the PC has received the report file, you can use and process the file with LIMS (Laboratory Information Management System) or other programs. An example of a report file is shown in Appendix A (page 43). In report files, the 6 pipetting channels of the EZ1 Advanced are named, from left to right, channels A to F; or the 14 pipetting channels of the EZ1 Advanced XL are named, from left to right, channels 1–14. When scanning a user ID or Q-Card bar code with the bar code reader, a beep confirms data input. After the information is displayed for 2 seconds, it is automatically stored, and the next display message is shown. When scanning sample ID, assay kit ID, or notes, a beep confirms data input, the information is displayed, and a message prompts you to enter the next item of information. After scanning sample ID, assay kit ID, and notes, press "ENT" once to confirm that the information entered is correct. If, for example, a wrong bar code was scanned for one of the samples, press "ESC" and then re-scan all sample bar codes according to the on-screen instructions. For user ID and notes, you can enter the numbers using the keypad, or you can easily generate your own bar codes to encode these numbers. For details about data tracking and using EZ1 Advanced Communicator software, see the *EZ1 Advanced User Manual* or the *EZ1 Advanced XL User Manual*.

Workflow of EZ1 operation

Insert EZ1 Card into the EZ1 Card slot

↓

Switch on the EZ1 instrument

↓

Follow on-screen messages for data tracking*

↓

Follow on-screen messages for worktable setup

↓

Start the protocol

↓

Collect purified nucleic acids

↓

UV decontamination*

* EZ1 Advanced and EZ1 Advanced XL only.

Yield of Purified DNA

DNA yields depend on the sample type, number of nucleated cells in the sample, and the protocol used for DNA purification. Table 2 shows typical yields for some common reference sample types.

Table 2. DNA yields from common reference sample types using EZ1 DNA Investigator procedures

Sample type	Sample amount	Protocol	DNA yield
Blood*	10–200 µl	Trace or Tip Dance	150 ng – 2 µg
Dried blood	4 x 3 mm disc	Tip Dance	0.2–0.5 µg
Buccal cells	1 swab	Tip Dance	100 ng – 2 µg

* Whole blood with 3–7 x 10^6 white blood cells/ml; elution volume 200 µl.

Precipitate in Reagent Cartridge

The buffer in well 1 of the reagent cartridge (the well that is nearest to the front of the EZ1 instrument when the reagent cartridge is loaded) may form a precipitate upon storage. If necessary, redissolve by mild agitation at 37°C and then place at room temperature (15–25°C).

Equilibrating Reagent Cartridges

If reagent cartridges have been stored at 2–8°C, they must be equilibrated to operating temperature before use. Place the reagent cartridge into a shaker–incubator and incubate at 30–40°C with mild agitation for at least 2 hours before use. If precipitates are visible at the bottom of the wells, redissolve by incubating at 30–40°C with mild agitation for a further 2 hours. Do not use the reagent cartridges if the precipitates do not redissolve.

Lysis with Proteinase K

The EZ1&2 DNA Investigator Kit contains Proteinase K, which is the enzyme of choice for lysis buffers used in EZ1 DNA Investigator protocols. Proteinase K is a recombinant protein expressed in Pichia pastoris and is particularly suitable for short digestion times. It possesses a high specific activity and remains stable over a wide range of temperatures and pH values, with substantially increased activity at higher temperatures. The activity of the Proteinase K solution is 600 mAU/ml solution (or 40 mAU/mg protein). This activity provides optimal results in EZ1 DNA Investigator protocols.

Quantification of DNA

Degraded, inhibited, or mixed DNA samples are common in forensic casework and other human identity testing applications. Such samples can create challenges in STR analysis. Prior quantification of the purified DNA using real-time PCR is recommended and reduces the need to repeat downstream analyses. This greatly reduces costs and time and improves the statistical relevance of results. Investigator Quantiplex™ Kits, use quantitative real-time PCR to quantify human total and human male DNA in a sample. These kits also detect if the sample contains inhibitors that may interfere with downstream applications, or if the DNA is degraded.

Protocol: Pretreatment for Various Casework and Reference Samples

This protocol is designed for isolation of total DNA (genomic and mitochondrial) from various types of casework and reference samples. The protocol describes the preliminary lysis using Proteinase K.

Important points before starting

- Before beginning the procedure, read “Important Notes”, page 13.
- We recommend using the Investigator Lyse&Spin Basket Kit (cat. no. 19597 or 19598) when solid sample materials have to be removed from the lysate. If using this kit, please follow the Pretreatment Protocol described in the corresponding handbook and the Large-Volume Protocol for DNA purification. The Lyse&Spin Basket Kit collection tubes can be used as sample tubes for the EZ1 Advanced XL, using the Flip-Cap Tube Rack (cat. no. 9022818) and Flip-Cap Card (cat. no. 9022763).

Things to do before starting

- Heat a thermomixer, heating block, or water bath to 56°C for the Proteinase K digest in step 3.

Procedure

1. Place the sample in a 2 ml sample tube.
2. Set up the Proteinase K digest according to information given in Table 1, page 10. Mix sample thoroughly by vortexing for 10 s.
3. Incubate at 56°C for 15 min to overnight in a thermomixer shaking at 900 rpm.

   15 min may be sufficient to recover adequate DNA for STR typing from samples containing abundant DNA. More than 1 h is recommended where a low amount of DNA is expected.

4. If necessary, flick the tube to remove drops from inside the lid.

   Optional: Add 1 µg carrier RNA (see "Important Notes", page 13)

5. Continue with "Protocol: DNA Purification," using one of the following options:

   5a. Trace Protocol, page 31.
   For samples that do not contain solid materials. The lysate volume should be approximately 200 µl.

   5b. "Tip Dance" Protocol, page 34.
   When using the "Tip Dance" Protocol, there is generally no need to remove solid material from the tube. However, when processing fluffy sample material such as cotton wool, we recommend removing solid material if you cannot process a replicate sample or the sample material is precious. Note that the "Tip Dance" Protocol will not recover the lysate absorbed by the sample substrate (e.g. swab, piece of fabric), therefore, a slightly reduced sensitivity has to be expected compared to methods that fully recover the lysate. We recommend using the Investigator Lyse&Spin Basket Kit for maximum sensitivity.

   5c. Large-Volume Protocol, page 37.
   The Large-Volume Protocol purifies DNA from 500 µl lysate.

Protocol: Pretreatment for Epithelial Cells Mixed with Sperm Cells

This protocol is designed for purification of total (genomic and mitochondrial) DNA from epithelial cells mixed with sperm cells. The protocol describes the preliminary lysis of samples using Proteinase K and dithiothreitol (DTT).

Important points before starting

- Before beginning the procedure, read “Important Notes”, page 13.
- As some sample types (e.g., fabrics) tend to be very absorbent, it may be necessary to add a greater volume of digestion buffer to the sample in step 2.

Things to do before starting

- Heat a thermomixer, heating block, or water bath for the Proteinase K digest to 56°C in steps 4 and 70°C in step 12.

Procedure

1. Place the forensic sample in a 1.5 ml or 2 ml sample tube.
2. Add 190 µl Buffer G2 to the sample.
3. Add 10 µl Proteinase K, and mix thoroughly by vortexing for 10 s.
4. Incubate at 56°C for 1–2 h. Do not exceed 2 h.

   Vortex the tube once or twice during the incubation, or place in a thermomixer.
5. Centrifuge the tube briefly to remove drops from inside the lid.
6. Remove any solid material from the tube.
7. Centrifuge the tube at 15,000 x *g* for 5 min. Carefully transfer the supernatant to a new tube without disturbing the sperm cell pellet.

DNA from epithelial cells can be purified from the tube containing the supernatant following Protocol: DNA Purification (Trace Protocol), page 31, or if the epithelial cell fraction is very dilute, Protocol: DNA Purification (Large-Volume Protocol), page 37.

Note: The cell pellet may not be visible

8. Wash the sperm cell pellet by resuspending the pellet in 500 µl Buffer G2. Centrifuge the tube at 15,000 x *g* for 5 min and discard the supernatant.
9. Repeat step 8 two or three times.
10. Add 160 µl Buffer G2 to the pellet and resuspend the pellet.
11. Add 10 µl Proteinase K and 40 µl 1 M DTT, and mix thoroughly by vortexing for 10 s.
12. Incubate at 70°C for 10 min at 850 rpm in a shaker–incubator or thermomixer.

    For maximum recovery, place samples in an Ultrasonicator for 10 min. Alternatively, vortex vigorously for 10 s.
13. Centrifuge the tube briefly to remove drops from inside the lid. DNA from sperm cells can now be purified from this tube.
14. Continue with Protocol: DNA Purification (Trace Protocol), page 31.

    The two tubes in which the epithelial and sperm cells have been separated are now ready for DNA purification.

Protocol: Extraction of DNA From Bone or Teeth

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&2 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.

Important points before starting

- If using the EZ1&2 DNA Investigator Kit for the first time, read "Important Notes".
- Sodium acetate (NaOAc) is required for this protocol but is not included in the EZ1&2 DNA Investigator Kit. NaOAc should be purchased separately.
- EDTA is required for this protocol, but is not included in the EZ1&2 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&2 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&2 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., liquid nitrogen stops boiling). Decant the excess liquid nitrogen, 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 bone 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), page 37.

Protocol: Pretreatment for Soil

This protocol is designed for isolation of total (genomic and mitochondrial) DNA from soil. The protocol describes the preliminary lysis of soil samples and adsorption of inhibitors using InhibitEX® tablets (contact QIAGEN Technical Services, see back cover).

Starting material

Up to 0.5 g of soil can be used, depending on the type of soil. With flocculent soil samples, less starting material should be used.

Important points before starting

- Before beginning the procedure, read "Important Notes", page 13.
- Proteinase K is not required in this protocol.
- This protocol requires InhibitEX tablets (contact QIAGEN Technical Services, see back cover).

Things to do before starting

- Heat a thermomixer, heating block, or water bath to 95°C for use in step 2.

Procedure

1. Place the soil sample in a 2 ml sample tube.
2. Add 900 µl distilled water. Resuspend the soil by vortexing, and incubate at 95°C for 10 min.
3. Centrifuge the tube at 4000 x *g* for 10 min. Transfer the supernatant to another 2 ml sample tube and add 190 µl Buffer G2. Mix by vortexing.
4. Add 1 InhibitEX tablet and incubate at room temperature (15–25°C) for 1 min.

5. Mix by vortexing and centrifuge at 10,000 x *g* for 2 min. Transfer 200 µl of the supernatant to an EZ1 sample tube if proceeding with Protocol: DNA Purification (Trace Protocol) or transfer 500 µl of the supernatant to an EZ1 sample tube if proceeding with Protocol: DNA Purification (Large-Volume Protocol).
6. Continue with Protocol: DNA Purification (Trace Protocol), page 31, or Protocol: DNA Purification (Large-Volume Protocol), page 37.

Protocol: DNA Purification (Trace Protocol)

This protocol is designed for isolation of total (genomic and mitochondrial) DNA from forensic samples that have been pretreated as described in the relevant protocols in this handbook (pages 23–29). The protocol describes the simple procedure for setting up the EZ1 instrument and starting a run.

Important points before starting

- If using the EZ1&2 DNA Investigator Kit for the first time, read "Important Notes" (page 13).
- The reagent cartridges contain guanidine salts and are therefore not compatible with disinfecting reagents containing bleach. See page 6 for Safety Information.
- Perform all steps of the protocol at room temperature (15–25°C). During the setup procedure, work quickly.
- In some steps of the procedure, one of 2 choices can be made. Choose ▲ if using the EZ1 Advanced or the EZ1 Advanced XL; choose ● if using the BioRobot EZ1.

Things to do before starting

- If reagent cartridges have been stored at 2–8°C, equilibrate to operating temperature before use. See "Equilibrating Reagent Cartridges", page 21.
- Remove any solid material from the sample tube.
- The lysis buffer in the reagent cartridge may form a precipitate during storage. If necessary, redissolve by warming at 37°C, and then place at room temperature (15–25°C).

Procedure

1. Insert ▲ the EZ1 Advanced DNA Investigator Card completely into the EZ1 Advanced Card slot of the EZ1 Advanced, or the EZ1 Advanced XL DNA Investigator Card

completely into the EZ1 Advanced XL Card slot of the EZ1 Advanced XL, or ● the EZ1 DNA Investigator Card completely into the EZ1 Card slot of the BioRobot EZ1.

2. Switch on the EZ1 instrument.
3. Press "START" to start protocol setup. ▲ Follow the on-screen instructions for data tracking.
4. Press "1" (for Trace Protocol).
5. Choose the elution buffer and volume: press "1" to elute in water or "2" to elute in TE buffer. Then press "1", "2", or "3" (or "4", EZ1 Advanced XL only) to select the elution volume.
6. Press any key to proceed through the text shown on the display and start worktable setup.

   The text summarizes the following steps which describe loading of the worktable. Wear gloves when loading the required items on the worktable.
7. Open the instrument door.
8. Invert reagent cartridges twice to mix the magnetic particles. Then tap the cartridges to deposit the reagents at the bottom of their wells. Check that the magnetic particles are completely resuspended.
9. Load the reagent cartridges into the cartridge rack.

   Note: After sliding a reagent cartridge into the cartridge rack, ensure that you press down on the cartridge until it clicks into place.
10. Load opened elution tubes into the first row of the tip rack.
11. Load tip holders containing filter-tips into the second row of the tip rack.
12. Load opened sample tubes containing digested samples into the back row of the tip rack.

    Pretreat the samples following the individual protocols in this handbook.

    Note: When using the data tracking option, ensure that the sample ID follows the same order as the samples on the worktable to avoid data mixup.
13. Close the instrument door.
14. Press "START" to start the purification procedure.

    The automated purification procedure takes 15–20 min.

15. When the protocol ends, the display shows “Protocol finished”. ▲ Press “ENT” to generate the report file.

    The EZ1 Advanced and the EZ1 Advanced XL can store up to 10 report files. Report files can be printed directly on a connected printer or transferred to a computer.

16. Open the instrument door.

17. Retrieve the elution tubes containing the purified DNA. The DNA is ready to use, or can be stored at 2–8°C for 24 h or at –30 to –15°C for longer periods. Discard the sample-preparation waste.*

    If the purified DNA is to be analyzed by real-time PCR, tubes containing eluate should first be applied to a suitable magnetic separator and the eluate transferred to a clean tube in order to minimize the risk of magnetic-particle carryover.

18. ▲ Optional: Follow the on-screen instructions to perform UV decontamination of the worktable surfaces.

19. To run another protocol, press “ESC”, prepare samples as described in the relevant protocol, and follow the procedure from step 4 onward. Otherwise, press “STOP” twice to return to the first screen of the display, close the instrument door, and switch off the EZ1 instrument.

20. Clean the EZ1 instrument.

    Follow the maintenance instructions in the user manual supplied with your EZ1 instrument.

* Sample waste contains guanidine salts and is therefore not compatible with bleach. See page 6 for Safety Information.

Protocol: DNA Purification ("Tip Dance" Protocol)

This protocol is designed for isolation of total (genomic and mitochondrial) DNA from forensic samples that have been pretreated as described in the relevant protocols in this handbook (pages 23–29). This protocol describes the simple procedure for setting up the EZ1 instrument and starting a run. In the "Tip Dance" Protocol, the filter-tip moves back-and-forth relative to the worktable platform while pipetting. This enables processing of solid materials, such as swabs, fabrics, blood discs, or cigarette butts, directly in the sample tube. There is generally no need for prior centrifugation to remove solid materials that could clog the tip. However, when processing fluffy sample material such as cotton wool, we recommend removing solid material if you cannot process a replicate sample or the sample material is precious.

Important points before starting

- If using the EZ1&2 DNA Investigator Kit for the first time, read "Important Notes" (page 13).
- The reagent cartridges contain guanidine salts and are therefore not compatible with disinfecting reagents containing bleach. See page 6 for Safety Information.
- Perform all steps of the protocol at room temperature (15–25°C). During the setup procedure, work quickly.
- In some steps of the procedure, one of 2 choices can be made. Choose ▲ if using the EZ1 Advanced or the EZ1 Advanced XL; choose ● if using the BioRobot EZ1.

Things to do before starting

- If reagent cartridges have been stored at 2–8°C, equilibrate to operating temperature before use. See "Equilibrating Reagent Cartridges", page 21.
- The lysis buffer in the reagent cartridge may form a precipitate during storage. If necessary, redissolve by warming at 37°C, and then place at room temperature (15–25°C).

Procedure

1. Insert ▲ the EZ1 Advanced DNA Investigator Card completely into the EZ1 Advanced Card slot of the EZ1 Advanced, or the EZ1 Advanced XL DNA Investigator Card completely into the EZ1 Advanced XL Card slot of the EZ1 Advanced XL, or ● the EZ1 DNA Investigator Card completely into the EZ1 Card slot of the BioRobot EZ1.
2. Switch on the EZ1 instrument.
3. Press "START" to start protocol setup. ▲ Follow the on-screen instructions for data tracking.
4. Press "2" (for Trace TD protocol).
5. Choose the elution buffer and volume: press "1" to elute in water or "2" to elute in TE. Then press "1", "2", or "3" (or "4", EZ1 Advanced XL only) to select the elution volume.
6. Press any key to proceed through the text shown on the display and start worktable setup.

   The text summarizes the following steps which describe loading of the worktable. Wear gloves when loading the required items on the worktable.
7. Open the instrument door.
8. Invert reagent cartridges twice to mix the magnetic particles. Then tap the cartridges to deposit the reagents at the bottom of their wells. Check that the magnetic particles are completely resuspended.
9. Load the reagent cartridges into the cartridge rack.

   Note: After sliding a reagent cartridge into the cartridge rack, ensure that you press down on the cartridge until it clicks into place.
10. Load opened elution tubes into the first row of the tip rack.
11. Load tip holders containing filter-tips into the second row of the tip rack.
12. Load opened sample tubes containing digested samples into the back row of the tip rack.

    Pretreat the samples following the individual protocols in this handbook.

    Note: When using the data tracking option, ensure that the sample ID follows the same order as the samples on the worktable to avoid data mixup.
13. Close the instrument door.

14. Press “START” to start the purification procedure.

    The automated purification procedure takes 15–20 min.

15. When the protocol ends, the display shows “Protocol finished”. ▲ Press “ENT” to generate the report file.

    The EZ1 Advanced and the EZ1 Advanced XL can store up to 10 report files. Report files can be printed directly on a connected printer or transferred to a computer.

16. Open the instrument door.

17. Retrieve the elution tubes containing the purified DNA. The DNA is ready to use, or can be stored at 2–8°C for 24 h or at –30 to –15°C for longer periods. Discard the sample-preparation waste.

    If the purified DNA is to be analyzed by real-time PCR, tubes containing eluate should first be applied to a suitable magnetic separator and the eluate transferred to a clean tube in order to minimize the risk of magnetic-particle carryover.

18. ▲ Optional: Follow the on-screen instructions to perform UV decontamination of the worktable surfaces.

19. To run another protocol, press “ESC”, prepare samples as described in the relevant protocol, and follow the procedure from step 4 onward. Otherwise, press “STOP” twice to return to the first screen of the display, close the instrument door, and switch off the EZ1 instrument.

20. Clean the EZ1 instrument.

    Follow the maintenance instructions in the user manual supplied with your EZ1 instrument.

Protocol: DNA Purification (Large-Volume Protocol)

This protocol is designed for isolation of total (genomic and mitochondrial) DNA from forensic samples that have been pretreated as described in the relevant protocols in this handbook (pages 23–29). This protocol describes the simple procedure for setting up the EZ1 instrument and starting a run.

Starting material

Using this protocol, up to 500 µl of pretreated sample can be processed. This not only allows efficient DNA purification from dilute samples with low concentrations of DNA, such as diffuse stains, but also enables purification from samples that require larger volumes for thorough lysis. The ability to process larger sample volumes, with the same elution volume as the standard Trace Protocol, enables higher yields of more concentrated DNA for greater sensitivity in downstream applications.

Important points before starting

- If using the EZ1&2 DNA Investigator Kit for the first time, read "Important Notes" (page 13).
- The reagent cartridges contain guanidine salts and are therefore not compatible with disinfecting reagents containing bleach. See page 6 for Safety Information.
- Perform all steps of the protocol at room temperature (15–25°C). During the setup procedure, work quickly.
- This protocol requires extra Buffer MTL (cat. no. 19112).
- In some steps of the procedure, one of 2 choices can be made. Choose ▲ if using the EZ1 Advanced or the EZ1 Advanced XL; choose ● if using the BioRobot EZ1.

Things to do before starting

- If reagent cartridges have been stored at 2–8°C, equilibrate to operating temperature before use. See "Equilibrating Reagent Cartridges", page 21.
- Remove any solid material from the sample tube.
- The lysis buffer in the reagent cartridge may form a precipitate during storage. If necessary, redissolve by warming at 37°C, and then place at room temperature (15–25°C).

Procedure

1. Insert ▲ the EZ1 Advanced DNA Investigator Card completely into the EZ1 Advanced Card slot of the EZ1 Advanced or the EZ1 Advanced XL DNA Investigator Card completely into the EZ1 Advanced XL Card slot of the EZ1 Advanced XL or ● the EZ1 DNA Investigator Card completely into the EZ1 Card slot of the BioRobot EZ1.
2. Switch on the EZ1 instrument.
3. Press "START" to start protocol setup. ▲ Follow the on-screen instructions for data tracking.
4. Press "3" (for Large-Volume Protocol).
5. Choose the elution buffer and volume: press "1" to elute in water or "2" to elute in TE buffer. Then press "1", "2", or "3" (or "4", EZ1 Advanced XL only) to select the elution volume.
6. Press any key to proceed through the text shown on the display and start worktable setup.

   The text summarizes the following steps which describe loading of the worktable. Wear gloves when loading the required items on the worktable.
7. Open the instrument door.
8. Invert reagent cartridges twice to mix the magnetic particles. Then tap the cartridges to deposit the reagents at the bottom of their wells. Check that the magnetic particles are completely resuspended.

9. Load the reagent cartridges into the cartridge rack.

   Note: After sliding a reagent cartridge into the cartridge rack, ensure that you press down on the cartridge until it clicks into place.

10. Load opened elution tubes into the first row of the tip rack.
11. Load tip holders containing filter-tips into the second row of the tip rack.
12. Add 400 µl Buffer MTL to each sample tube containing digested samples. Load opened sample tubes containing Buffer MTL and digested samples into the back row of the tip rack.

    Pretreat the samples following the individual protocols in this handbook.

    Note: When using the data tracking option, ensure that the sample ID follows the same order as the samples on the worktable to avoid data mixup.

13. Close the instrument door.
14. Press "START" to start the purification procedure.

    The automated purification procedure takes 15–20 min.

15. When the protocol ends, the display shows "Protocol finished". ▲ Press "ENT" to generate the report file.

    The EZ1 Advanced and the EZ1 Advanced XL can store up to 10 report files. Report files can be printed directly on a connected printer or transferred to a computer.

16. Open the instrument door.
17. Retrieve the elution tubes containing the purified DNA. The DNA is ready to use, or can be stored at 2–8°C for 24 h or at –30 to –15°C for longer periods. Discard the sample-preparation waste.*

    If the purified DNA is to be analyzed by real-time PCR, tubes containing eluate should first be applied to a suitable magnetic separator and the eluate transferred to a clean tube in order to minimize the risk of magnetic-particle carryover.

* Sample waste contains guanidine salts and is therefore not compatible with bleach. See page 6 for Safety Information.

18. ▲ Optional: Follow the on-screen instructions to perform UV decontamination of the worktable surfaces.
19. To run another protocol, press “ESC”, prepare samples as described in the relevant protocol, and follow the procedure from step 4 onward. Otherwise, press “STOP” twice to return to the first screen of the display, close the instrument door, and switch off the EZ1 instrument.
20. Clean the EZ1 instrument.

    Follow the maintenance instructions in the user manual supplied with your EZ1instrument.

Troubleshooting Guide

This troubleshooting guide may be helpful in solving any problems that may arise. For more information, see also the Frequently Asked Questions page in our Technical Support Center: **www.qiagen.com/FAQ/FAQList.aspx**. The scientists in QIAGEN Technical Services are always happy to answer any questions you may have about either the information or protocols in this handbook (for contact information, visit **support.qiagen.com**).

	Comments and suggestions
Statement of the problem	
a) Error message in instrument display	Refer to the user manual supplied with your EZ1 instrument.
b) Report file not printed	Check whether the printer is connected to the EZ1 Advanced or EZ1 Advanced XL via "PC/Printer" serial port. Check whether the serial port is set for use with a printer.
c) Report file not sent to the PC	Check whether the printer is connected to the EZ1 Advanced or EZ1 Advanced XL via "PC/Printer" serial port. Check whether the serial port is set for use with a printer.
d) Wrong Q-Card ID entered	If the wrong ID was entered instead of the Q-Card ID, the EZ1 Advanced/EZ1 Advanced XL will not accept the ID and will prompt for the Q-Card ID until the correct ID is entered. Press "STOP" twice to go to the main menu.
Statement of another problem	
a) Magnetic particles not completely resuspended	Ensure that you invert the reagent cartridges several times to resuspend the magnetic particles.
b) Insufficient reagent aspirated	After inverting the reagent cartridges to resuspend the magnetic particles, ensure that you tap the cartridges to deposit the reagents at the bottom of the wells.
c) Purified DNA stored in water	Elute in TE buffer instead of water. Elution in TE buffer gives comparable performance and provides increased stability for long-term storage of small amounts of purified DNA.

		Comments and suggestions
d)	Varying pipetting volumes	To ensure pipetting accuracy, it is important that buffer volumes in the reagent cartridges are correct and that the filter tips fit optimally to the tip adapter. Ensure that samples are thoroughly mixed and that reagent cartridges have not passed their expiry date. Perform regular maintenance as described in the instrument user manual. Check the fit of the filter tips regularly as described in the user manual.
Statement of another problem		
a)	Insufficient DNA used in downstream applications	If possible, repeat the downstream application using more eluate.
b)	Excess DNA used in downstream applications	Excess DNA can inhibit some enzymatic reactions. Dilute the eluate or use less in the downstream application. Quantify the purified DNA by measurement of the absorbance using an appropriate method.

Appendix A: Example of an EZ1 Advanced Report File

This appendix shows a typical report file generated on the EZ1 Advanced. The values for each parameter will differ from the report file generated on your EZ1 Advanced. Please note that "User ID" is allowed a maximum of 9 characters, and that "Assay kit ID" and "Note" are allowed a maximum of 14 characters. The EZ1 Advanced XL generates a similar report file containing instrument and protocol information relevant to the EZ1 Advanced XL and information for channels 1–14.

REPORT - FILE EZ1 Advanced:

_ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _

Serial No. EZ1 Advanced: _ _ _ _ _ _ _ _ 0301F0172

User ID: _ 4121

Firmware version: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ V 1.0.0

Installation date of instr.: _ _ _ _ _ _ _ _ Jan 05, 2008

Weekly maintenance done on: _ _ _ _ _ Apr 15, 2008

Yearly maintenance done on: _ _ _ _ _ _ Mar 10, 2008

Date of last UV-run: _ _ _ _ _ _ _ _ _ _ _ Apr 20, 2008

Start of last UV-run: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 16:06

End of last UV-run: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 16:26

Status UV-run: _o.k.

Protocol name: _ _ _ _ _ _ _ _ _ _ _ _ DNA Investigator

_ Trace

Date of run: _ _ _ _ _ _ _ _ _ _ _ _ _ _ Apr 21, 2008

Start of run: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 12:57

End of run: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 13:31

Status run: _ o.k.

Error Code: _

Sample input Vol [ul]: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 200
Elution volume [ul]: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 100
Channel A:
Sample ID: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 123456789
Reagent Kit number: _ _ _ _ _ _ _ _ _ _ _ _ _ 9801301
Reagent Lot number: _ _ _ _ _ _ _ _ _ _ _ _ 23456789
Reagent Expiry date: _ _ _ _ _ _ _ _ _ _ _ _ _ _ 1208
Assay kit ID: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 848373922
Note: _ 2000
Channel B:
Sample ID: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 234567890
Reagent Kit number: _ _ _ _ _ _ _ _ _ _ _ _ _ 9801301
Reagent Lot number: _ _ _ _ _ _ _ _ _ _ _ _ 23456789
Reagent Expiry date: _ _ _ _ _ _ _ _ _ _ _ _ _ _ 1208
Assay kit ID: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 836266738
Note: _
Channel C:
Sample ID: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 345678901
Reagent Kit number: _ _ _ _ _ _ _ _ _ _ _ _ _ 9801301
Reagent Lot number: _ _ _ _ _ _ _ _ _ _ _ _ 23456789
Reagent Expiry date: _ _ _ _ _ _ _ _ _ _ _ _ _ _ 1208
Assay kit ID: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 883727832
Notes: _1000
Channel D:
Sample ID: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 456789012
Reagent Kit number: _ _ _ _ _ _ _ _ _ _ _ _ _ 9801301
Reagent Lot number: _ _ _ _ _ _ _ _ _ _ _ _ 23456789
Reagent Expiry date: _ _ _ _ _ _ _ _ _ _ _ _ _ _ 1208
Assay kit ID: _ _ _ __ _ _ _ _ _ _ _ _ _ 763684837
Note: _

Channel E:
Sample ID: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 567890123
Reagent Kit number: _ _ _ _ _ _ _ _ _ _ _ _ _ 9801301
Reagent Lot number: _ _ _ _ _ _ _ _ _ _ _ _ 23456789
Reagent Expiry date: _ _ _ _ _ _ _ _ _ _ _ _ _ _ 1208
Assay kit ID: _ _ _ _ _ _ _ _ _ _ _ _ _ _ 4387728002
Note: _
Channel F:
Sample ID: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 678901234
Reagent Kit number: _ _ _ _ _ _ _ _ _ _ _ _ _ 9801301
Reagent Lot number: _ _ _ _ _ _ _ _ _ _ _ _ 23456789
Reagent Expiry date: _ _ _ _ _ _ _ _ _ _ _ _ _ _ 1208
Assay kit ID: _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ _ 509389403
Note: _ 50

Ordering Information

Product	Contents	Cat. no.
EZ1&2 DNA Investigator Kit (48)	For 48 preps: Reagent Cartridges, Disposable Tip Holders, Disposable Filter-Tips, Sample Tubes, Elution Tubes, Buffers and Reagents; includes Certificate of Analysis	952034
EZ1 Advanced XL	Robotic instrument for automated purification of nucleic acids from up to 14 samples using EZ1 Kits,1-year warranty on parts and labor*	9001492
EZ1 Advanced XL Flip-Cap Tube Rack	Tube rack for the use of Flip-Cap tubes on EZ1 Advanced XL	9022818
EZ1 Advanced XL DNA Investigator Flip-Cap Card	Preprogrammed card for purification of DNA using the EZ1 Advanced XL with an EZ1 Advanced XL Flip-Cap Tube Rack	9022763
EZ1 Advanced XL DNA Investigator Card	Preprogrammed card for EZ1 Advanced XL DNA Investigator protocols on the EZ1 Advanced XL	9018699
EZ1 Advanced DNA Investigator Card	Preprogrammed card for EZ1 Advanced DNA Investigator protocols	9018302
EZ1 DNA Investigator Card	Preprogrammed card for BioRobot EZ1 DNA Investigator protocols	9016387

* Warranty PLUS 2 (cat. no. 9237720) recommended: 3-year warranty, 1 preventive maintenance visit per year, 48-hour priority response, all labor, travel, and repair parts.

Product	Contents	Cat. no.
EZ2 Fx Connect	Robotic instrument for automated purification of nucleic acids from up to 24 samples, 1-year warranty on parts and labor	9003220
Accessories		
Filter-Tips and Holders, EZ1 (50)	50 Disposable Filter-Tips, 50 Disposable Tip Holders; additional tips and holders for use with EZ1 Kits	994900
12-Tube Magnet	Magnet for separating magnetic particles in 12 x 1.5 ml or 2 ml tubes	36912
Buffer G2 (260 ml)	Lysis buffer for EZ1 DNA procedures	1014636
DTT (1 ml)	1M DTT, forensic grade quality; for sperm cell lysis	1117316
Buffer MTL (54 ml)		19112
QIAGEN Proteinase K (1.2 ml)	1.2 ml (>600 mAU/ml, solution)	1014023
TissueLyser II	Universal laboratory mixer-mill disruptor	85300
Grinding Jar Set, S. Steel (2 x 10 ml)	2 Grinding Jars (10 ml), 2 Stainless Steel Grinding Balls (20 mm)	69985
PC and TFT Monitor, 17"	PC capable of connection with up to 4 EZ1 Advanced or EZ1 Advanced XL instruments; Monitor for use with PC	9016643
HID related products		
Investigator Lyse&Spin Basket Kit (50)	50 pouches containing 50 baskets and 100 collection tubes	19597
Investigator Lyse&Spin Basket Kit (250)	10 pouches containing 5 x 50 baskets and 5 x 50 collection tubes	19598

Product	Contents	Cat. no.
Investigator Quantiplex Pro Kit (200)	For use on Applied Biosystems 7500 Real-Time Systems: Quantiplex Pro Reaction Mix, Quantiplex Pro Primer Mix, Quantiplex Pro Control DNA M1, QuantiTect Nucleic Acid Dilution Buffer	387216
Investigator Quantiplex Pro RGQ Kit (200)	For use on QIAGEN Rotor-Gene Q Real-Time Systems: Quantiplex Pro RGQ Reaction Mix, Quantiplex Pro RGQ Primer Mix, Male Control DNA M1, QuantiTect Nucleic Acid Dilution Buffer	387316
Investigator 24plex QS Kit (100)*	Primer Mix, Fast Reaction Mix 2.0, Control DNA, allelic ladder, DNA Size Standard, and nuclease-free water	382415
Investigator 26plex QS Kit (100)*	Primer Mix, Fast Reaction Mix 3.0, Control DNA, allelic ladder 24plex, and nuclease-free water	382615
Investigator Argus X-12 QS Kit (25)*	Primer Mix, Fast Reaction Mix 2.0, Control DNA, Allelic Ladder, DNA Size Standard, nuclease-free water	383223
Investigator Argus Y-28 QS Kit (100)*	Primer Mix, Fast Reaction Mix 3.0, Control DNA, Allelic Ladder, DNA Size Standard, nuclease-free water	383625

* Larger sizes are available. Please inquire.

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** or can be requested from QIAGEN Technical Services or your local distributor.

Document Revision History

Date	Changes
09/2021	Rebranded "EZ1 DNA Investigator Kit" to "EZ1&2 DNA Investigator Kit". Incorporated the supplementary protocol for extraction of DNA from bone or teeth in the handbook. Updated the Ordering Information section. Editorial and layout changes.

Notes

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