QlAsymphony[®] PowerFecal[®] Pro DNA Kit Handbook

For semi-automated purification of microbial genomic DNA from stool and all soil types using QIAsymphony SP



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Kit Contents

QIAsymphony PowerFecal Pro DNA Kit	(192)
Catalog no.	938036
Number of preps	192
PowerBead Pro Tubes	200
Solution CD1	4x 40 ml
Solution CD2	60 ml
Reagent Rack Virus/Bacteria Midi (48)	2
QIApack Piercing Lid	2
Reuse Seal Set	2
Microcentrifuge Tubes 2ml	200
Quick-Start Protocol	1

Intended Use

The QIAsymphony PowerFecal Pro DNA Kit is intended for the automated extraction of microbial genomic DNA using the QIAsymphony SP instrument.

The QlAsymphony PowerFecal Pro DNA Kit is intended for molecular biology applications. This product is not intended for the diagnosis, prevention, or treatment of a disease.

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.

Summary and Explanation

The QIAsymphony PowerFecal Pro DNA Kit comprises a novel and proprietary method for automated mid-throughput isolation of both microbial and host genomic DNA from stool and gut samples using the second generation of QIAGEN's patented Inhibitor Removal Technology® (IRT). This kit is intended for use with samples containing inhibitory substances commonly found in stool, such as polysaccharides, heme compounds, and bile salts. Improved IRT combined with more efficient bead-beating and lysis chemistry in an automated format results in high-quality DNA that can be used immediately in downstream application including PCR, qPCR, and NGS applications (16s and Whole Genome Sequencing).

Principle of the Procedure

The QlAsymphony PowerFecal Pro DNA Kit is effective at removing PCR inhibitors from even the most difficult stool types. Samples are added to the PowerBead Pro Tube for rapid and thorough homogenization. Cell lysis occurs by mechanical and chemical methods. Total genomic DNA is captured on silica magnetic beads on the QlAsymphony instrument. DNA is then automatically washed and eluted from the magnetic beads and is ready for NGS, PCR, and other downstream applications.

QIAsymphony technology combines the speed and efficiency of silica-based nucleic acid purification with the convenient handling of magnetic particles (Figure 1). The purification procedure is designed to ensure safe and reproducible handling of potentially infectious samples, and comprises 3 steps: bind, wash, and elute (see Workflow, page 7).

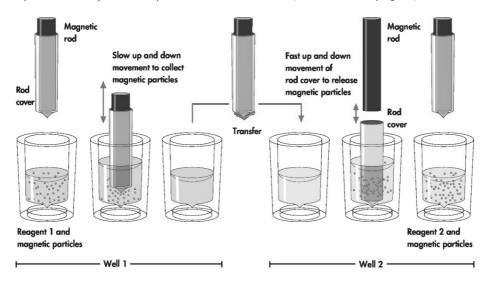


Figure 1. Schematic of the QIAsymphony SP principle. The QIAsymphony SP processes a sample containing magnetic particles as follows: A magnetic rod protected by a rod cover enters a well containing the sample and attracts the magnetic particles. The magnetic rod cover is positioned above another well and the magnetic particles are released. The QIAsymphony SP uses a magnetic head containing an array of 24 magnetic rods, and can therefore process up to 24 samples simultaneously. Steps 1 and 2 are repeated several times during sample processing.

Starting Material

The intended primary use of the protocol is for the extraction of microbial gDNA from stool samples. 50 to 100 mg is the recommended amount of starting material for these sample types, but up to 200 mg can be processed with the protocol.

Stool samples typically contain high contents of inhibitors that interfere with downstream enzymatic reactions and compounds that can degrade DNA. The QIAsymphony PowerFecal Pro DNA protocol removes these substances and extracts microbial gDNA that is free of proteins, nucleases, and other contaminants or inhibitors.

Microbial gDNA that is purified using the QIAsymphony PowerFecal Pro DNA protocol is ready for use in enzymatic reactions, such as PCR, or storage at -30 to -15° C or -90 to -65° C.

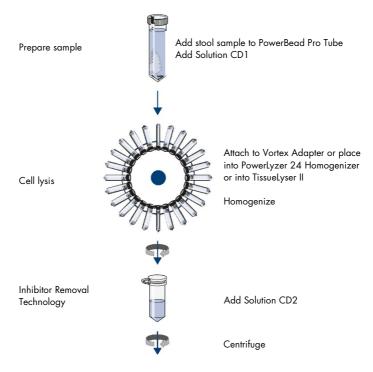
Bead beating

The QlAsymphony PowerFecal Pro DNA Kit does not require homogenization using a high-velocity bead beater. However, if the microorganism of interest requires stronger homogenization than provided by a vortex with vortex adapter for 1.5–2 ml tubes (cat. no. 13000-V1-24), or if using a bead beater is desired, the QlAsymphony PowerFecal Pro DNA Kit contains bead tubes suitable for high-powered bead beating and may be used in conjunction with the PowerLyzer® 24 Homogenizer (110/220V) (cat. no. 13155) or the TissueLyser II (cat. no. 85300) using a 2 ml Tube Holder Set (cat no. 11993).

Use of the PowerLyzer 24 instrument (cat.no. 13155) allows the simultaneous disruption of up to 24 samples.

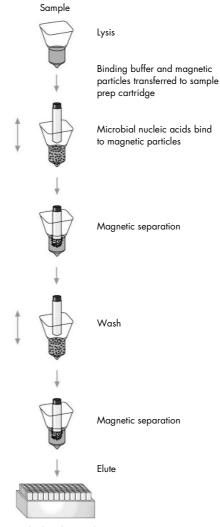
For convenient high-throughput 96-well homogenization, we offer the TissueLyser II (cat. no. 85300) and Plate Adapter Set (cat.no.11990). In conjunction with Power Bead Pro Plates (cat. no. 19301), the instrument provides high-throughput processing for simultaneous, rapid, and effective disruption of up to 2 x 96 samples in only a few minutes.

Workflow



Transfer supernatant to fresh 2 ml micro tube (Sarstedt® cat. no. 72.694)
Transfer micro tubes containing sample to QIAsymphony instrument

QIAsymphony PowerFecal Pro DNA Procedure



Pure, microbial nucleic acids

Figure 2. QIAsymphony PowerFecal Pro DNA Kit Procedure. Sample lysis, inhibitor removal, and DNA purification on QIAsymphony SP.

Materials Required but Not Provided

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.

- Pipettes and disposable pipette tips with aerosol barriers (20–1000 µl)
- Disposable gloves
- Microcentrifuge (up to $15000 \times g$) or Centrifuge with Plate Rotor 2×96 (up to $4500 \times g$)
- Vortex-Genie[®] 2
- Vortex adapter for 24 (1.5–2 ml) tubes (cat. no. 13000-V1-24)
 - **Optional, for high velocity bead beating with PowerBead Pro Tubes**: PowerLyzer 24. (cat.no. 13155) or TissueLyser II (cat. No. 85300) with adapter sets for use with the PowerBead Pro Tubes (TissueLyser Adapter Set 2 x 24 cat. no. 69982, 2ml Tube Holder cat. No. 11993 in conjunction with Plate Adapter Set cat.no.119909)
- For disruption in 96-well format: PowerBead Pro Plates (cat. no. 19301), TissueLyser II (cat. no. 85300), and adapter sets for use with the PowerBead Pro Plates (Plate Adapter Set cat.no.119909)
- Sample Prep Cartridges, 8-well (cat. no. 997002)
- 8-Rod Covers (cat. no. 997004)
- Filter-Tips, 200 μl and 1500 μl (cat. nos. 990332 and 997024)
- Sample tubes: Recommended 2 ml micro tube (Sarstedt® cat. no. 72.694)

Warnings and Precautions

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

Reagent Storage and Handling

Solution CD2 should be stored at 2–8°C upon arrival. All other components of the QIAsymphony PowerFecal Pro DNA Kit can be stored at room temperature (15–25°C) until the expiration date printed on the box label. The magnetic particles in the reagent cartridges (RC) remain active when stored at this temperature. Do not store RCs at temperatures below 15°C.

Partially used RCs can be stored for a maximum of 4 weeks, enabling cost-efficient reuse of reagents and more flexible sample processing. If an RC is partially used, replace the cover of the trough containing the magnetic particles and seal the RC with the provided Reuse Seal Strips immediately after the end of the protocol run to avoid evaporation.

Running batches with low sample numbers (<24) will potentially reduce the total number of sample preparations possible per cartridge.

To avoid reagent evaporation, the RC should be open for a maximum of 15 hours (including run times) at a maximum environmental temperature of 30°C.

Avoid exposure of the RC to UV light (e.g., used for decontamination), as exposure may cause accelerated aging of the RCs and buffers.

Note: The label on the QIAsymphony PowerFecal Pro DNA Kit displays the expiration date of the kit. The result file documents the expiration dates for only the Reagent Rack Virus/Bacteria Midi (if required).

Note: The reagent cartridge used with the QIAsymphony PowerFecal Pro DNA Kit is called Reagent Rack Virus/Bacteria Midi. This is the name that will appear in the result file generated after running the protocol in the instrument.

Quality Control

In accordance with QIAGEN's ISO-certified Quality Management System, each lot of QIAsymphony PowerFecal Pro DNA Kit is tested against predetermined specifications to ensure consistent product quality.

Procedure

Automated purification on QIAsymphony SP

The QIAsymphony SP makes automated sample preparation easy and convenient. Samples, reagents and consumables, and eluates are separated in different drawers. Simply load samples, reagents provided in special cartridges, and pre-racked consumables in the appropriate drawer before a run. Start the protocol and remove purified DNA from the "Eluate" drawer after processing. Refer to the user manuals supplied with your instrument for operating instructions.

Note: Optional maintenance is not mandatory for instrument function, but is highly recommended to reduce risk of contamination.

Loading RCs into the "Reagents and Consumables" drawer

Reagents for purification of DNA are contained in an innovative RC (Figure 3). Each trough of the RC contains a particular reagent, such as magnetic particles, binding buffer, wash buffer, or elution buffer. Partially used RCs can be reclosed with Reuse Seal Strips (RSS) for later reuse, which avoids generation of waste due to leftover reagents at the end of the purification procedure.

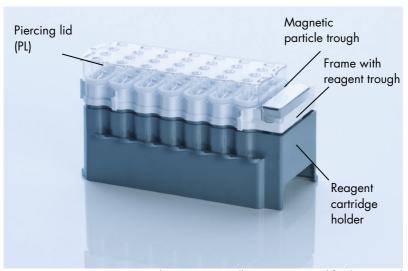


Figure 3. QIAsymphony reagent cartridge (RC). The RC contains all reagents required for the protocol run.

Before starting the procedure, make sure that the magnetic particles are fully resuspended. Remove the magnetic-particle trough from the reagent cartridge frame, vortex it vigorously for at least 3 minutes then replace it in the reagent cartridge frame before the first use.

Note: Magnetic particles can change color. This has no influence on performance.

Place the RC into the reagent cartridge holder. Before using an RC for the first time, place the piercing lid (PL) on top of the RC (Figure 3).

Note: The PL is sharp. Take care when placing it onto the RC. Make sure to place the PL onto the RC in the correct orientation.

After the magnetic-particle trough cover is removed, the RC is subsequently loaded into the "Reagents and Consumables" drawer.

Partially used RCs can be stored until needed again (see "Reagent Storage and Handling" page 10).

Note: Make sure that RCs and magnetic-particle troughs bottles are not interchanged between different kit lots

Loading plastic ware into the "Reagents and Consumables" drawer

Sample prep cartridges, 8-Rod Covers (both pre-racked in unit boxes), and disposable filtertips (200 µl tips provided in blue racks, 1500 µl tips provided in black racks) are loaded into the "Reagents and Consumables" drawer.

Note: Make sure that the covers of the unit boxes are removed before loading the unit boxes into the "Reagents and Consumables" drawer.

Note: Tips have filters to help prevent cross-contamination.

Tip rack slots on the QIAsymphony SP worktable can be filled with either type of tip rack. The QIAsymphony SP will identify the type of tips loaded during the inventory scan.

Note: Do not refill tip racks or unit boxes for sample prep cartridges or 8-Rod Covers before starting another protocol run. The QIAsymphony SP can use partially used tip racks and unit boxes.

For the consumables required, see the relevant protocol sheet, which can be found under the resource tab of the product page on **www.qiagen.com**. For plasticware Ordering Information, see page 25.

Loading the "Waste" drawer

Sample prep cartridges and 8-Rod Covers used during a run are re-racked in empty unit boxes in the "Waste" drawer. Make sure that the "Waste" drawer contains sufficient empty unit boxes for plastic waste generated during the protocol run.

Note: Make sure that the covers of the unit boxes are removed before loading the unit boxes into the "Waste" drawer. If you are using 8-Rod Cover boxes for collecting used sample prep cartridges and 8-Rod Covers, ensure that the box spacer has been removed.

A bag for used filter-tips must be attached to the front side of the "Waste" drawer.

Note: The presence of a tip disposal bag is not checked by the system. Make sure that the tip disposal bag is properly attached before starting a protocol run. For more information, see the user manuals provided with your instrument. Empty the tip bag after a maximum of 96 samples have been processed to avoid a tip jam.

A waste container collects liquid waste generated during the purification procedure. The "Waste" drawer can only be closed if the waste container is in place. Dispose of the liquid waste according to your local safety and environment regulations. Do not autoclave the filled waste bottle. Empty the waste bottle after a maximum of 96 samples have been processed.

Loading the "Eluate" drawer

Load the required elution rack into the "Eluate" drawer. As long-term storage of eluates in the "Eluate" drawer may lead to evaporation or condensation, the cooling position must be used. Only use "Elution slot 1" with the corresponding cooling adapter.

Inventory scan

Before starting a run, the instrument checks that sufficient consumables for the queued batch(es) have been loaded into the corresponding drawers.

Preparation of sample material

Stool and soil samples require a sample pretreatment step. Samples are added to a bead-beating tube for rapid and thorough homogenization. Cell lysis occurs by mechanical and chemical methods. Total genomic DNA is captured on a silica surface of magnetic particles automated on QIAsymphony SP. DNA is then automatically washed and eluted from the magnetic-particles and ready for NGS, PCR, and other downstream applications.

Prevent formation of foam in or on the samples before placing them on the QIAsymphony SP instrument. Foam on samples can lead to pipetting of wrong sample volume. Samples should be equilibrated to room temperature (15–25°C) before starting the run.

For more information about the automated procedure (including information about sample tubes that can be used with specific protocols), see the relevant protocol sheet which can be found under the resource tab of the product page on **www.qiagen.com**.

Storing DNA

After sample preparation, eluates must be stored at -30 to -15° C or -90 to -65° C to prevent degradation Frozen eluates must not be thawed more than three times.

Important points before starting

- Ensure that the PowerBead Pro Tubes rotate freely in the centrifuge without rubbing.
- Perform all centrifugation steps at room Temperature (15–25°C).
 Optional: Set a thermomixer or shaker–incubator to 56°C for use in step 5 of sample pre-treatment procedure.
- Make sure that you are familiar with operating the QIAsymphony SP. Refer to the user manuals supplied with your instrument for operating instructions.
- Optional maintenance is not mandatory for instrument function, but is highly recommended to reduce risk of contamination.
- \bullet $\,$ Before beginning the procedure, read "Principle of the Procedure", page 5.
- Make sure that you are familiar with the protocol sheet corresponding to the procedure you want to use. (Protocol sheets can be found under the resource tab of the product page on www.qiagen.com).
- Avoid vigorous shaking of the RC, otherwise foam may be generated, which can lead to liquid-level detection problems.

Things to do before starting

- Before starting the procedure, make sure that the magnetic particles are fully resuspended. Vortex the trough containing the magnetic particles vigorously for at least 3 minutes before first use.
- Make sure that the PL is placed on the RC and the lid of the magnetic-particle trough has been removed or, if using a partially used RC, make sure the Reuse Seal Strips have been removed.
- For information about sample tubes compatible with a certain protocol, see the
 corresponding labware list which can be found under the resource tab of the product
 page on www.qiagen.com.

Sample Pretreatment using PowerBead Pro Tubes:

- 1. Spin the PowerBead Pro Tube briefly to ensure that the beads have settled at the bottom.
- 2. Add 50–100 mg of stool or up to 250 mg of soil and 800 μ l of Solution CD1. Vortex briefly to mix.
- 3. Secure the PowerBead Pro Tube horizontally on a vortex adapter for 1.5–2 ml tubes (cat. no. 13000-V1-24). Vortex at maximum speed for 10 min.

Note: If using the vortex adapter for more than 12 preps simultaneously, increase the vortex time by 5–10 min. Avoid using tape, which can become loose and result in reduced homogenization efficiency, inconsistent results, and reduced yields.

Important: This step is critical for complete homogenization and cell lysis. Cells are lysed by a combination of chemical agents from step 2 and mechanical shaking introduced at this step. By randomly shaking the beads in the presence of disruption agents, collision of the beads with microbial cells will cause the cells to break open.

- 4. Centrifuge the PowerBead Pro Tube at $15,000 \times g$ for 1 min.
- 5. Transfer the supernatant to a clean 2 ml microcentrifuge tube (provided).

Note: Expect a volume of $500-600 \, \mu l$. The supernatant may still contain some stool or soil particles.

Optional: Perform the optional steps below, if RNA-free DNA is required.

- 5a. Add 4 µl RNaseA and vortex shortly. Spin it down and incubate the mixture for 5 min at room temperature.
- 5b. Add 30 μl Proteinase K and vortex shortly. Spin it down and incubate the mixture for 15 min at 56°C.
- 6. Add 300 µl Solution CD2 and vortex for 5 s.

Note: Solution CD2 contains Inhibitor Removal Technology (IRT), and can precipitate non-DNA organic and inorganic material, including polysaccharides, heme compounds, bile salts, humic substances, cell debris, and proteins. It is important to remove contaminating organic and inorganic matter that may reduce DNA purity and inhibit downstream DNA applications.

- 7. Centrifuge at $15,000 \times g$ for 1 min at room temperature.
- 8. Avoiding the pellet, transfer $600 \mu l$ of supernatant to a clean 2 ml micro tube (Sarstedt cat. no. 72.694) (not provided).

Note: The pellet at this point contains non-DNA organic and inorganic material including polysaccharides, heme compounds, bile salts, humic substances, cell debris, and proteins. For best DNA yields and quality, avoid transferring any of the pellet.

Note: After manual transfer, proceed with the QIAsymphony DNASoilStool_600_V1 Protocol on the QIAsymphony SP instrument. Transfer the 2 ml micro tubes containing the sample to the QIAsymphony instrument.

Sample Pretreatment using PowerBead Pro Plate:

Materials required but not provided

- PowerBead Pro Plate (cat. no. 19311)
- TissueLyser II (cat. no. 85300)
- Plate Adapters (cat. no. 11990)
- Collection Microtubes (CMTR) (cat. no. 19560)
- S-Blocks (cat. no. 19585)
- 1. Spin the PowerBead Pro Plate briefly to ensure that the beads have settled at the bottom.
- 2. Remove Square Well Mat from a PowerBead Pro Plate (cat. no. 19311). Add 50 to 150 mg of stool or up to 250 mg of soil sample.

Note: Avoid cross contamination between sample wells.

- 3. Add 800 µl of Solution CD1 to the wells of the PowerBead Pro Plate.
- 4. Make sure to remove any residual liquid on top of the plate.

Note: Liquid on top of the plate will prevent a tight sealing of the plate with the sealing film. This may result in leakage during disruption in the Tissuelyser II (cat. no. 85300).

5. Secure the sealing film tightly onto the bead plate. Use a tool such as a scraper or plate roller to make the sealing film adhere firmly onto the plate.

Note: A strong seal is essential to prevent leakage during disruption in the TissueLyser II.

6. Put the silicone compression mat on top of the bead plate that is sealed with the sealing film.

Note: Two silicone compression mats are provided with the PowerBead Plates (cat. no. 19311) so that 2 plates can be processed in parallel in the Tissuelyser II. The mats are reused for the remaining plates.

7. Place this entire assembly (from steps 1 to 6) between 2 Plate Adapters for disruption in the TissueLyser II.

Important: When using this assembly, do not exceed the recommended disruption time and setting of 2×5 min at 25 Hz, because doing so might lead to leakage.

8. Shake at speed 25 Hz for 5 min. Re-orient plates so that the side that was closest to the machine body is now furthest from it and shake again at speed 25 Hz for 5 min.

Note: This protocol uses a combination of mechanical and chemical lysis. Mechanical lysis is introduced at this step. By randomly shaking the beads, they collide with one another and with microbial cells causing them to break open.

9. Centrifuge at room temperature for 6 min at 4500 x g.

Note: Particulates, including polysaccharides, heme compounds, bile salts, humic substances, cell debris, and proteins, will form a pellet at this point. DNA is in the supernatant.

10. Discard Sealing Film. Transfer the supernatant to the Collection Microtubes (CMTR).

Note: Expect 500-600 µl. The supernatant may still contain some stool or soil particles.

11.Add 300 µl of Solution CD2. Seal Collection Microtubes with the caps provided and vortex.

Note: Solution CD2 contains Inhibitor Removal Technology (IRT), and can precipitate non-DNA organic and inorganic material including polysaccharides, heme compounds, bile salts, humic substances, cell debris, and proteins. It is important to remove contaminating organic and inorganic matter that may reduce DNA purity and inhibit downstream DNA applications.

- 12. Centrifuge the plate at room temperature for 6 min at $4500 \times g$.
- 13. Transfer up to 600 µl of supernatant to an S-Block (cat. no. 19585).

Note: The pellet at this point contains non-DNA organic and inorganic material including polysaccharides, heme compounds, bile salts, humic substances, cell debris, and proteins. For best DNA yields and quality, avoid transferring any of the pellet.

QIAsymphony SP Procedure

- 1. Close all drawers and the hood.
- 2. Switch on the QIAsymphony SP and wait until the "Sample Preparation" screen appears and the initialization procedure has finished.
- 3. Log in to the instrument.
- 4. Ensure the "Waste" drawer is prepared properly and perform an inventory scan of the "Waste" drawer, including the tip chute and liquid waste. Replace the tip disposable bag if necessary.
- 5. Load the required elution rack into the "Eluate" drawer.

We recommend using slot 1 (cooling position) with Elution Microtubes CL (cat. no. 19588) or Sarstedt 2 ml micro tubes (cat.no. 72.693 or 72.694). When using the Elution Microtubes CL rack, remove the bottom by twisting the rack until the bottom comes off

Do not load a 96-well plate onto "Elution slot 4".

When using a 96-well plate, make sure that the plate is in the correct orientation, as incorrect placement may cause sample mix-up in downstream analysis.

- 6. Load the required RCs and consumables into the "Reagents and Consumables" drawer.
- 7. Perform an inventory scan of the "Reagents and Consumables" drawer.
- 8. Place the samples into the appropriate sample carrier and load them into the "Sample" drawer.
- 9. Using the touchscreen, enter the required information for each batch of samples to be processed.
 - 9a. Sample information depending on sample racks used (e.g., Sarstedt 2 ml micro tube with cap (cat. no. 72.694) with tube carrier or QIAGEN S-Block, (cat. no. 19585) with plate carrier)
 - 9b. Protocol to be run (DNASoilStool_600_V1)

- 9c. Elution volume and output position
- After information about the batch has been entered, the status changes from **LOADED** to **QUEUED**. As soon as one batch is queued the **Run** button appears.
- 10. Press the "Run" button to start the purification procedure.
 - All processing steps are fully automated. At the end of the protocol run, the status of the batch changes from **RUNNING** to **COMPLETED**.
- 11. Retrieve the elution rack containing the purified nucleic acids from the "Eluate" drawer.
- 12. The DNA is ready to use or can be stored at 2-8°C, -20°C, or -80°C.

We recommend removing the eluate plate from the "Eluate" drawer immediately after the run has finished. Depending on temperature and humidity, elution plates left in the QIAsymphony SP after the run is completed may experience condensation or evaporation.

In general, magnetic particles are not carried over into eluates. If carryover does occur, magnetic particles in eluates will not affect most downstream applications.

Result files are generated for each elution plate.

13. If an RC is only partially used, seal it with the provided Reuse Seal Strips after the end of the protocol run to avoid evaporation.

Note: For more information about storage of partially used RCs, see "Reagent Storage and Handling", page 10.

- 14. Discard used sample tubes and waste according to your local safety regulations.

 See "Warnings and Precautions", page 10, for safety information.
- 15. Clean the QIAsymphony SP.

Follow the maintenance instructions in the user manuals supplied with your instrument. Make sure to clean the tip guards regularly to minimize the risk of cross-contamination.

16. Close the instrument drawers and power OFF the QIAsymphony SP.

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 at our Technical Support Center: www.qiagen.com/FAQ/FAQList.aspx.

For contact information, see back cover or visit www.qiagen.com.

Comments and suggestions

		Comments and suggestions	
Instru	ment issues		
a)	Error message displayed on the touchscreen	If an error message is displayed during a protocol, refer to the user manuals supplied with your instrument.	
Prec	ipitate in reagent trough of	opened cartridge of the QIAsymphony kit	
a)	Buffer evaporation	Excessive evaporation may lead to increased salt concentration in buffers. Discard RC. Make sure to seal buffer troughs of a partially used RC with Reuse Seal Strips when not being used for purification.	
b)	Storage of RC	Storage of RC below 15°C may lead to formation of precipitates.	
Low	Low yield of DNA		
a)	Magnetic particles were not completely resuspended	Before starting the procedure, ensure that the magnetic particles are fully resuspended. Vortex for at least 3 min before use.	
b)	Clogging of pipette tip due to insoluble material	Insoluble material was not removed from the sample prior to starting the QIAsymphony purification procedure.	
Stool	Processing		
a)	Amount of stool to process	The QlAsymphony PowerFecal Pro DNA Kit is designed to process up to 0.2 grams of stool or up to 0.25 grams of soil. For inquiries regarding the use of larger sample amounts, please contact Technical Support for suggestions.	

Comments and suggestions

		Comments and soggestions
b)	Stool/soil sample is high in water content	Remove contents from the PowerBead Pro Tube (beads) and transfer into another sterile microcentrifuge tube (not provided). Add stool/soil sample to PowerBead Pro Tube and centrifuge at room temperature for 30 seconds at $10,000 \times g$. Remove as much liquid as possible with a pipette tip. Add beads back to PowerBead Pro Tube.
DNA		
a)	DNA does not amplify	Make sure to check DNA yields by gel electrophoresis or spectrophotometer reading. An excess amount of DNA will inhibit a PCR reaction.
		Diluting the template DNA should not be necessary with DNA isolated using the QIAsymphony PowerFecal Pro DNA Kit, however, it should still be attempted.
		If DNA will still not amplify after trying the steps above, then PCR optimization may be needed.
b)	Eluted DNA is brown	If you observe coloration in your samples, please contact Technical Support for suggestions.
c)	Storing DNA	DNA is eluted in Buffer AVE and must be stored at -30 to -15°C or -90 to -65°C to prevent degradation.
Alterr	native lysis methods	
a)	Cells are difficult to lyse	After adding Solution CD1 and prior to the bead-beating step, incubate at 65°C for 10 min. Resume protocol from step 2.
b)	Reduction of shearing of DNA	After adding Solution CD1, vortex for 3-4 seconds, then heat to 70°C for 5 min. Repeat once. This alternative procedure will reduce shearing but may also reduce yield.

Ordering Information

Product	Contents	Cat. no.
QIAsymphony PowerFecal Pro DNA Kit	Includes 2 reagent cartridges and enzyme racks and accessories	938036
Related Products		
PowerBead Pro Tubes (50)	For disruption of stool and all soil samples in 2ml format	19301
PowerBead Pro Plates (4)	For high-throughput disruption of stool samples and all soil types in 96 well format	19311
S-Blocks (24)	24 x 96-well blocks with 2.2 ml wells	19585
Collection Microtubes (racked, 10 x 96)	Nonsterile polypropylene tubes (1.2 ml), 960 in racks of 96	19560
Collection Microtube Caps (120 x 8)	Nonsterile polypropylene caps for collection microtubes (1.2 ml) and round-well blocks, 960 in strips of 8	19566
TissueLyser II	Bead mill (100–120/220–240 V, 50/60 Hz) for medium- to high-throughput sample disruption for molecular analysis; requires the TissueLyser Adapter Set 2 x 24 or 2ml Tube Holder Set or TissueLyser Adapter Set 2 x 96*	85300
TissueLyser Adapter Set 2 x 24	2 sets of adapter plates and 2 racks for use with 2 ml microcentrifuge tubes on the TissueLyser II	69982
TissueLyser Adapter Set 2 x 96	2 sets of adapter plates for use with Collection Microtubes (racked) on the TissueLyser II	69984

^{*} The TissueLyser II must be used in combination with the TissueLyser Adapter Set 2 x 24 or TissueLyser Adapter Set 2 x 96.

Product	Contents	Cat. no.
2ml Tube Holder Set	For sample homogenization in 2 ml bead tubes on the TissueLyser II	11993
Plate Adapter Set	Set of four adapters required to assemble two 96 well plates onto the 96 Well Plate Shaker	11990
TissueLyser LT	Compact bead mill, 100-240 V AC, 50–60 Hz; requires the TissueLyser LT Adapter, 12-Tube*	85600
TissueLyser LT Adapter, 12-Tube	Adapter for disruption of up to 12 samples in 2 ml microcentrifuge tubes on the TissueLyser LT	69980
Vortex Adapter for 24 (1.5–2.0 ml) tubes	For vortexing 1.5 ml or 2 ml tubes using the Vortex-Genie 2 Vortex	13000- V1-24
PowerLyzer 24 Homogenizer (110/220 V)	For complete lysis and homogenization of any biological sample	13155
QIAGEN Proteinase K (2 ml)	2 ml (>600 mAU/ml, solution)	19131
QIAGEN Proteinase K (10 ml)	10 ml (>600 mAU/ml, solution)	19133
RNase A (17,500 U)	2.5 ml (100 mg/ml; 7000 units/ml, solution) Unit definition: That amount of enzyme causing the hydrolysis of RNA at a rate such that k (velocity constant) equals unity (Kunitz units) at 25°C and pH 5.0.	19101
Sample Prep Cartridges, 8-well (336)	8-well sample prep cartridges for use with the QIAsymphony SP	997002
8-Rod Covers (144)	8-Rod Covers for use with the QIAsymphony SP	997004

 $[\]star$ The Tissuelyser LT must be used in combination with the Tissuelyser LT Adapter, 12-Tube.

Product	Contents	Cat. no.
Reagent Cartridge Holder (2)	Reagent cartridge holder for use with the QIAsymphony SP	997008
Accessory Trough (10)	Accessory troughs for use with the QIAsymphony SP	997012
Tip Disposal Bags (15)	Tip disposal bags for use with the QIAsymphony SP/AS instruments	9013395
Sample Carrier, plate, Qsym	Plate carrier for sample input; for use with the QIAsymphony SP	9017660
Cooling Adapter, EMT, v2, Qsym	Cooling adapter for EMT racks. For use with the QIAsymphony SP/AS instruments	9020730
Cooling Adapter, 2 ml, v2, Qsym	Cooling adapter for 2 ml screw-cap tubes. For use with the QIAsymphony SP/AS instruments	9020674
Insert, 2 ml, v2 sample carr. (24), Qsym	Secondary tube adapter (for 2 ml screw-cap tubes) for use with the QIAsymphony tube carrier	9242083
Adapter, tubes, 2 ml, v2, Qsym	Non-cooling adapter for 2 ml screw-cap tubes. For use in the QIAsymphony "Eluate" drawer	9021670
Filter-Tips, 200 µl (1024)	Disposable Filter-Tips, racked; (8 x 128). For use with the QIAcube® and the QIAsymphony SP	990332
Filter-Tips, 1500 µl (1024)	Disposable Filter-Tips, racked; (8 x 128). For use with the QIAsymphony SP/AS instruments	997024
Reuse Seal Set (20)	Reuse seal sets for sealing partly used QIAsymphony reagent cartridges	997006
Elution Microtubes CL (24 x 96)	Nonsterile polypropylene tubes (0.85 ml maximum capacity, less than 0.7 ml storage capacity, 0.4 ml elution capacity); 2304 in racks of 96; includes cap strips	19588

Product	Contents	Cat. no.
QIAsymphony SP	QlAsymphony sample prep module, 1-year warranty on parts and labor	9001297
UCP Multiplex PCR Kit (100)	For 100 reactions: For highly specific and sensitive multiplex PCR with minimized background using nucleic acid-depleted reagents	206742
UCP Multiplex PCR Kit (500)	For 500 reactions: For highly specific and sensitive multiplex PCR with minimized background using nucleic acid-depleted reagents	206744
UCP SYBR Green® 16S Quant Kit (100)	1x1ml UCP SYBR Green PCR MasterMix (2x); 1x Microbial DNA Standard, 1x 16S DNA SYBR Green Assay; 1x200 µl UCP Yellow Template Dil. Buffer; 1x250 µl UCP ROX Dye; 1x1.9 ml UCP PCR Water	208082
UCP Probe 16S/18S Quant Kit (100)	1x1ml UCP Probe PCR MasterMix (2x); 1x Microbial DNA Standard, 1x 16S/18S DNA Probe Assay; 1x100 µl Internal Control DNA; 1x200 µl UCP Yellow Template Dil. Buffer; 1x250 µl UCP ROX Dye; 1x1.9 ml UCP PCR Water	208282

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.

Notes

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Handbook Revision History

Date	Changes
10/2019	Initial release

Limited License Agreement for QIAsymphony PowerFecal Pro DNA Kit

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