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June 2017

# RNeasy<sup>®</sup> PowerSoil<sup>®</sup> Total RNA Kit Handbook

For the isolation of high-quality total RNA  
from all soil types

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# Kit Contents

<b>RNeasy PowerSoil Total RNA Kit</b>	<b>(25)</b>
<b>Catalog no.</b>	<b>12866-25</b>
<b>Number of preps</b>	<b>25</b>
PowerBead Tubes, Carbide	25
PowerBead Solution	2 x 42 ml
Solution SR1	7 ml
Solution IRS	2 x 15 ml
Solution SR3	42 ml
Solution SR4	6 x 27.5 ml
Solution SR5	110 ml
Solution SR6	28 ml
Solution SR7	2 x 1.5 ml
JetStar 2.0 Mini Columns	25
Collection Tubes (2.2 ml)	25
Collection Tubes (15 ml)	2 x 50
Quick Start Protocol	1

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## Storage

The RNeasy PowerSoil Total RNA Kit reagents and components should be stored at room temperature (15–25°C).

## Intended Use

All RNeasy products are intended for molecular biology applications. These products are 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.

# 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](http://www.qiagen.com/safety) where you can find, view and print the SDS for each QIAGEN kit and kit component.

**WARNING:** Solution SR4 is flammable.

**WARNING:** Phenol/chloroform/isoamyl alcohol is a caustic organic solution. Do not inhale vapors. Phenol is highly corrosive and can cause severe burns. Chloroform is a carcinogen. Follow local ordinances for disposal of phenol waste. In the event of an accident, seek medical attention immediately.

<p><b>CAUTION</b></p> 	<p><b>DO NOT add bleach or acidic solutions to directly to the sample preparation waste</b></p>
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PowerBead Solution contains guanidine salts, which can form highly reactive compounds when combined with bleach. If liquid containing these buffers is spilt, clean with a suitable laboratory detergent and water. If the spilt liquid contains potentially infectious agents, clean the affected area first with laboratory detergent and water, and then with 1% (v/v) sodium hypochlorite.

## Quality Control

In accordance with QIAGEN's ISO-certified Quality Management System, each lot of RNeasy PowerSoil Total RNA Kits is tested against predetermined specifications to ensure consistent product quality.

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# Introduction

The QIAGEN RNeasy PowerSoil Total RNA Kit is designed to isolate total RNA from organisms found in soil and reliably provide RNA for experiments requiring qualitative and quantitative RT-PCR analysis. This kit permits consistent removal of humic substances, fulvic acids and other RT-PCR inhibitors from RNA purified from soil. Diverse soil types, including compost, manure, estuary sediment and other soil types high in organic content have successfully provided intact and RT-PCR-amplifiable RNA using this kit.

The LifeGuard Soil Preservation Solution is a specially formulated treatment for the collection and transportation of soils for RNA extraction. The solution protects the viability of bacteria in soil while keeping them in stasis. RNase and DNase activity is prevented, allowing accurate, full-length cDNA synthesis and 16S rRNA profiling on samples collected in the field. Microbial community profiles are maintained in soils stabilized with LifeGuard Solution for up to 30 days at  $-20^{\circ}\text{C}$ , 1 week at  $4^{\circ}\text{C}$  and 3 days at room temperature.

We do not recommend the use of RNeasy Protect and RNeasy Protect for preservation of nucleic acids in soils.

For up to 2 grams of soil, homogenization occurs in a 15 ml tube containing silica carbide beads, lysis buffers, phenol/chloroform/isoamyl alcohol (pH 6.5–8) and Solution IRS to ensure complete lysis of all microorganisms and neutralization of RNases. Cleared lysates are precipitated to concentrate total nucleic acids, and pellets are resuspended in a buffer optimized for binding to anion-exchange gravity flow columns. RNA is eluted using a high-salt buffer and precipitated to obtain the final pure RNA resuspended in RNase-free water. DNA may be purified separately from the RNA using an alternative high-salt buffer provided the RNeasy PowerSoil DNA Elution Kit (cat. no. 12867-25). Isolated nucleic acids are ready for use in enzymatic applications.

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## 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.

- Centrifuge capable of centrifuging 15 ml tubes (2500 x *g* minimum)
- Microcentrifuge (13,000 x *g*)
- Pipettors (20 µl–1000 µl)
- Serological pipettes (1 ml and 10 ml)
- Vortex-Genie® 2 Vortex
- Vortex Adapter for 4 (15 ml) tubes (cat. no. 13000-V1-15)
- RNase-free gloves (cat. nos. 1556-S, 1556-M and 1556-L)
- Lab cleaner for RNase removal (cat. no. 12095-500)
- Phenol/chloroform/isoamyl alcohol solution
- Heat block set at 45°C (optional)

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# Protocol: Experienced User

## Important points before starting

- If Solution IRS has precipitated, heat at 60°C until precipitate dissolves.
- Perform all centrifugation steps at room temperature (15–25°C).
- Wear RNase-free gloves at all times and remove RNase from the work area.

## Procedure

1. Add up to 2 g of soil to the 15 ml PowerBead Tube (provided). Please refer to the Troubleshooting Guide for information regarding the amount of soil to process.
2. Add 2.5 ml of PowerBead Solution, 0.25 ml of Solution SR1 and 0.8 ml of Solution IRS.
3. Add 3.5 ml of phenol/chloroform/isoamyl alcohol (pH 6.5–8.0, [User supplied]). Cap and vortex the PowerBead Tube to mix until the biphasic layer disappears.
4. Place the PowerBead Tube on a Vortex Adapter (cat. no. 13000-V1-15) and vortex at maximum speed for 15 min.
5. Remove the PowerBead Tube and centrifuge at 2500 x g for 10 min.
6. Transfer the upper aqueous phase (avoid the interphase and lower phenol layer) to a clean 15 ml Collection Tube (provided). Discard the phenol/chloroform/isoamyl alcohol.  
**Note:** The biphasic layer will be thick and firm in soils high in organic matter and may need to be pierced to remove the bottom phenol layer.
7. Add 1.5 ml of Solution SR3 to the aqueous phase and vortex to mix. Incubate at 2–8°C for 10 min and then centrifuge at 2,500 x g for 10 min at room temperature.
8. Transfer the supernatant, without disturbing the pellet (if there is one), to a new 15 ml Collection Tube (provided).
9. Add 5 ml of Solution SR4 to the supernatant in the Collection Tube and invert or vortex to mix. Incubate at room temperature for 30 min.  
**Note:** Previous protocol instructions were to incubate at –20°C. If you have achieved good results for your soil type using the previous protocol, you may continue to follow it.
10. Centrifuge at 2500 x g for 30 min.



11. Decant the supernatant and invert the 15 ml Collection Tube on a paper towel for 5 min.
12. Shake Solution SR5 to mix and add 1 ml to the 15 ml Collection Tube. Resuspend the pellet completely by repeatedly pipetting or vortexing.  
**Note:** If the pellet is difficult to resuspend, place the tube in a heat block or water bath at 45°C for 10 min, followed by vortexing. Repeat until the pellet is resuspended.
13. Prepare one JetStar Mini Column (provided) for each RNA isolation sample:
  - 13a. Remove the cap of a 15 ml Collection Tube (provided) and place the JetStar Mini Column inside it. The column will hang in the Collection Tube.
  - 13b. Add 2 ml of Solution SR5 to the JetStar Mini Column. Allow it to completely gravity flow through the column and collect in the 15 ml Collection Tube.  
**Note:** Do not allow the column to dry out before loading the RNA isolation sample.
14. Add the RNA isolation sample from Step 12 onto the JetStar Mini Column and allow it to gravity flow through the column into the 15 ml Collection Tube.
15. Add 1 ml of Solution SR5 to the JetStar Mini Column and allow it to completely gravity flow into the 15 ml Collection Tube.
16. Transfer the JetStar Mini Column to a new 15 ml Collection Tube (provided). Shake Solution SR6 to mix and then add 1 ml to the JetStar Mini Column to elute the bound RNA. Allow Solution SR6 to gravity flow into the 15 ml Collection Tube.
17. Transfer the eluted RNA to a 2.2 ml Collection Tube (provided). Add 1 ml of Solution SR4. Invert at least once to mix and incubate at –15°C to –30°C for a minimum of 10 min.
18. Centrifuge the 2.2 ml Collection Tube at 13,000 x g for 15 min to pellet the RNA.
19. Decant the supernatant and invert the 2.2 ml Collection Tube onto a paper towel for 10 min to air dry the pellet.
20. Resuspend the RNA pellet in 100 µl of Solution SR7.

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# Protocol: Detailed

## Important points before starting

- If Solution IRS has precipitated, heat at 60°C until precipitate dissolves.
- Perform all centrifugation steps at room temperature (15–25°C).
- Wear RNase-free gloves at all times and remove RNase from the work area.

## Procedure

1. Add up to 2 g of soil to the 15 ml PowerBead Tube (provided). Please refer to the Troubleshooting Guide for information regarding the amount of soil to process.
2. Add 2.5 ml of PowerBead Solution, 0.25 ml of Solution SR1 and 0.8 ml of Solution IRS.  
**Note:** The PowerBead Solution is a buffer that disperses cells and soil particles. Solution SR1 contains SDS and other disruption agents which aid in complete cell lysis. In addition to aiding in cell lysis, SDS is an anionic detergent that breaks down fatty acids and lipids associated with the cell membrane of several organisms. Solution IRS is a precipitation reagent that removes non-DNA organic and inorganic material including cell debris and proteins.  
**Note:** If it gets cold, Solution SR1 will form a white precipitate. Heating to 60°C will dissolve the SDS and will not harm the other disruption agents.
3. Add 3.5 ml of phenol/chloroform/isoamyl alcohol (pH 6.5–8.0, [User supplied]). Cap and vortex the PowerBead Tube to mix until the biphasic layer disappears.  
**Note:** Phenol/chloroform/isoamyl alcohol maximizes lysing efficiency and yield. Lysed cell components are trapped in the solvent and proteins are denatured leaving the nucleic acid in solution.
4. Place the PowerBead Tube on a Vortex Adapter (cat. no. 13000-V1-15) and vortex at maximum speed for 15 min.  
**Note:** Cells are lysed by a combination of chemical agents from steps 1-3 and mechanical shaking introduced by vortexing. Use of the vortex adapter will maximize homogenization, which can lead to higher DNA yields. Using tape to attach tubes is not recommended.

5. Remove the PowerBead Tube and centrifuge at 2500 x g for 10 min.  
**Note:** Centrifugation results in phase separation of the sample mixture. Three phases will be visible after centrifugation: the lower organic phase containing proteins and cellular debris, the interphase containing humics and other organic and non-organic material, and the upper aqueous phase containing total nucleic acid.  
**Note:** The thickness of the interphase will depend on the sample type. Samples high in organic content will have a thicker interphase.
6. Transfer the upper aqueous phase (avoid the interphase and lower phenol layer) to a clean 15 ml Collection Tube (provided). Discard the phenol/chloroform/isoamyl alcohol.  
**Note:** The biphasic layer will be thick and firm in soils high in organic matter and may need to be pierced to remove the bottom phenol layer. The upper aqueous phase containing total nucleic acids from the sample is transferred to a new tube. Cellular debris, proteins and organic material are left behind. Take care not to transfer material from the lower phase or the interphase.
7. Add 1.5 ml of Solution SR3 to the aqueous phase and vortex to mix. Incubate at 2–8°C for 10 min and then centrifuge at 2,500 x g for 10 min at room temperature.  
**Note:** Solution SR3 is a secondary precipitation step to further remove proteins and cellular debris.
8. Transfer the supernatant, without disturbing the pellet (if there is one), to a new 15 ml Collection Tube (provided).  
**Note:** The supernatant containing nucleic acids are transferred to a new 15 ml tube. Non-nucleic acid material is left behind.
9. Add 5 ml of Solution SR4 to the supernatant in the Collection Tube and invert or vortex to mix. Incubate at room temperature for 30 min.  
**Note:** Previous protocol instructions were to incubate at –20°C. If you have achieved good results for your soil type using the previous protocol, you may continue to follow it.
10. Centrifuge at 2500 x g for 30 min.
11. Decant the supernatant and invert the 15 ml Collection Tube on a paper towel for 5 min.  
**Note:** Solution SR4 is 100% isopropanol. Nucleic acid is precipitated and the isopropanol is discarded.

12. Shake Solution SR5 to mix and add 1 ml to the 15 ml Collection Tube. Resuspend the pellet completely by repeatedly pipetting or vortexing.  
**Note:** If the pellet is difficult to resuspend, place the tube in a heat block or water bath at 45°C for 10 min, followed by vortexing. Repeat until the pellet is resuspended. Solution SR5 is a proprietary salt solution used to resuspend the precipitated nucleic acids from Step 11. It is also used to equilibrate the JetStar Mini Column in Step 13 and to wash and prep the column for RNA elution in Step 15.
13. Prepare one JetStar Mini Column (provided) for each RNA isolation sample:
  - 13a. Remove the cap of a 15 ml Collection Tube (provided) and place the JetStar Mini Column inside it. The column will hang in the Collection Tube.
  - 13b. Add 2 ml of Solution SR5 to the JetStar Mini Column. Allow it to completely gravity flow through the column and collect in the 15 ml Collection Tube.  
**Note:** Do not allow the column to dry out before loading the RNA isolation sample.
14. Add the RNA isolation sample from Step 12 onto the JetStar Mini Column and allow it to gravity flow through the column into the 15 ml Collection Tube.
15. Add 1 ml of Solution SR5 to the JetStar Mini Column and allow it to completely gravity flow into the 15 ml Collection Tube.  
**Note:** The sample is added to the JetStar Mini Column and the nucleic acids are bound to the column matrix. The Capture Column is then washed with a second volume of Solution SR5 to ensure unbound contaminants are removed from the sample and column prior to the elution of RNA.
16. Transfer the JetStar Mini Column to a new 15 ml Collection Tube (provided). Shake Solution SR6 to mix and then add 1 ml to the JetStar Mini Column to elute the bound RNA. Allow Solution SR6 to gravity flow into the 15 ml Collection Tube.  
**Note:** The Solution SR6 is a proprietary salt solution that allows for the preferential release of RNA from the JetStar Mini Column, leaving DNA, residual debris and inhibiting substances in the column.
17. Transfer the eluted RNA to a 2.2 ml Collection Tube (provided). Add 1 ml of Solution SR4. Invert at least once to mix and incubate at -15°C to -30°C for a minimum of 10 min.
18. Centrifuge the 2.2 ml Collection Tube at 13,000 x g for 15 min to pellet the RNA.

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19. Decant the supernatant and invert the 2.2 ml Collection Tube onto a paper towel for 10 min to air dry the pellet.

**Note:** Solution SR4 is 100% isopropanol. Eluted RNA from the Capture Column is precipitated, centrifuged and allowed to air dry before it is resuspended and concentrated.

20. Resuspend the RNA pellet in 100  $\mu$ l of Solution SR7. The RNA is now ready for downstream applications.

**Note:** Solution SR7 is RNase/DNase-free water used to resuspend the pelleted RNA. Solution SR7 contains no EDTA. For long term storage of samples, 10 mM Tris (pH 8.0) or TE buffer may be used to resuspend the pelleted RNA.

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# Protocol: LifeGuard Soil Preservation Solution

## Procedure

1. Add 2–2.5 volumes of LifeGuard Soil Preservation Solution per 1 g of soil. If collecting the soil directly in an RNeasy PowerSoil 15 ml PowerBead Tube, add 5 ml of LifeGuard Soil Preservation Solution to 2 g of soil.

**Note:** If you are working with sediment samples, use 3 volumes of LifeGuard Soil Preservation Solution per gram of wet sample weight.

2. Vortex or hand-mix the solution and soil until the entire sample is saturated with solution. Excess LifeGuard Solution should be on top of the soil sample.

3. Store at room temperature, at 2–8°C or at –20°C. When ready to process the RNA, centrifuge sample at 2500 x g for 5 min to collect the soil. Remove the LifeGuard Solution from the tube.

4. If you collected soil directly in an RNeasy PowerSoil 15 ml PowerBead Tube, you can remove the LifeGuard Solution after centrifugation and continue from Step 2 of the RNeasy PowerSoil Total RNA Isolation Kit protocol.

5. If you stabilized soil in another vessel, weigh the amount of soil you need and transfer to an RNeasy PowerSoil 15 ml PowerBead Tube, or a conical tube, and then centrifuge to remove the LifeGuard Solution.

**Note:** Several different soils have been tested using LifeGuard, and each soil differs in its biomass and its saturation with water. Sediment samples require a higher volume of LifeGuard (3:1) due to dilution of the reagent.

**Note:** Biomass content may play a role in the stabilization of soils at different temperatures and the ability of LifeGuard to freeze the community profile. For a soil with unknown microbial content or where temperatures may exceed room temperature (22–25°C), it may be preferable to store the soil at –20°C or 2–8°C to ensure the original community profile is maintained, and to use more than 2.5 ml of LifeGuard per gram of soil. For long term storage and transport (>30 days), store stabilized soil at –15°C to –30°C.

# 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](http://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 and/or protocols in this handbook or sample and assay technologies (for contact information, visit [www.qiagen.com](http://www.qiagen.com)).

## Comments and suggestions

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### Soil processing

- a) Amount of soil to process      It is possible to use the RNeasy PowerSoil Total RNA Kit to process up to 2 grams of most soil types. For inquiries regarding the use of larger sample amounts, please contact Technical Support for suggestions. For soils with high organic content, 1 gram of soil typically gives an adequate amount of RNA while reducing the potential for DNA carryover during purification.
- For sediment samples, more soil may be used. We have used up to 5 grams of sediment with RNeasy PowerSoil Total RNA Kit. You may increase the volume of Solution SR4 at step 9 to equal the volume of lysate.
- b) Processing different types of soil      The yield and purity of RNA will depend on the soil type processed. The RNeasy PowerSoil Total RNA Kit has been validated with diverse soil types that represent a wide range of physical, chemical and biological characteristics.
- Some soils and sediments may contain excess salinity. This is visualized after step 10 by a large white or yellow salt pellet. This type of pellet may result in reduced nucleic acid yields. If you have a salt-enriched soil or sediment, to overcome the precipitation of salt in the pellet after adding Solution SR4, incubate the samples at room temperature for 30 minutes instead of at  $-20^{\circ}\text{C}$ . Proceed as directed for the rest of the protocol.

### Procedure

- a) Phase separation of phenol/chloroform/isoamyl alcohol      To ensure effective phenol/chloroform/isoamyl alcohol and aqueous phase separation, centrifuge the 15 ml tubes at  $2500 \times g$  at room temperature for at least 10 minutes (step 5). Following centrifugation, the interphase thickness between the phenol/chloroform/isoamyl alcohol and aqueous layers will vary depending on the organic content of the soil. When removing the upper aqueous phase, be careful to avoid the lower phenol phase and interphase,

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## Comments and suggestions

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- which contains proteins and lipids. If a portion of the phenol layer or the interphase is removed, re-centrifuge the transferred aqueous phase containing tube to obtain a phase separation that will permit removing only the aqueous phase. Alternatively, an equal volume (2 ml) of chloroform may be added to the phenol or interphase-contaminated aqueous phase, inverted or vortexed to mix, and the tubes centrifuged at  $2500 \times g$  for 10 minutes at room temperature. Remove the upper aqueous phase and discard the lower chloroform/phenol interphase. Then combine with the rest of the aqueous phase and continue with the protocol.
- b) Pellet resuspension in Solution SR5  
Soil types with high organic content may yield RNA pellets that are difficult to resuspend. Heating the RNA pellet in Solution SR5 at  $45^{\circ}\text{C}$  will aid in the resuspension process. Disrupting the RNA pellet with a pipette tip and vortexing vigorously will also aid in RNA resuspension. It is important to resuspend the pellet completely before applying it to the column in step 14. Failure to completely resuspend the RNA pellet will result in RNA loss through reduced column binding and will result in reduced column flow rate.
- c) Column flow  
The JetStar Mini Columns are rated for gravity flow and should not be used with centrifugal or vacuum force. Additional positive pressure can be applied to the column using a 5 cc syringe barrel. Hold the 5 cc syringe barrel flush against the top of the JetStar Mini Column and gently push the syringe inside the 5 cc barrel to force air through the JetStar Mini Column. Flow rate should not exceed 1 drop per second.
- d) DNA contamination of RNA  
DNA carryover typically does not occur with most soil types. However, certain soils high in organic matter may present unique carryover situations. The purified RNA may be tested for potential DNA carryover by agarose gel analysis or by performing PCR with qualified primers on the isolated RNA without performing prior reverse transcription amplification. The absence of a detectable amplification fragment by agarose electrophoresis indicates the absence of detectable carryover DNA. If DNA is detected, DNase treatment of the isolated RNA is recommended.  
  
To remove genomic DNA from RNA preps, we recommend the DNase Max<sup>®</sup> Kit (cat. no. 15200-50). The DNase Max Kit uses a high-velocity DNase enzyme, which is efficiently removed post digestion using a resin that inactivates and binds to the enzyme. RNA is purified from DNA without the use of inhibitors (EDTA) or heat and is ready to use.
- e) Multiple elutions from the same column  
Multiple elutions beyond those called for in the protocol are not recommended when using the JetStar Mini Columns. Although a small amount of additional RNA or DNA (if using the RNeasy PowerSoil DNA Elution Kit; cat. no. 12867-25) may come off the column with multiple elutions, the inhibitors associated with the starting material will also begin to wash off the column and could disrupt downstream applications.



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# Appendix A: Using Phenol/Chloroform/Isoamyl Alcohol

Phenol and chloroform are toxic and potentially dangerous to your health and safety. Please see all manufacturers' warnings and precautions before working with these compounds.

Phenol and phenol/chloroform/isoamyl alcohol are subject to oxidation reactions that cause them to become yellow or pink, which serves as an indicator that the phenol is **NOT** useable for RNA extraction. Using colored phenol or colored phenol/chloroform/isoamyl alcohol will compromise the quality of the extracted RNA. Prior to each use, a sample of the phenol/chloroform/isoamyl alcohol should be placed in a clear container to determine its clarity and color. When not in use, securely cap and store the phenol/chloroform/isoamyl alcohol at 2°–8°C in the dark. Avoid any prolonged exposure to light.

## Preparing Phenol/Chloroform/Isoamyl Alcohol Solution

Mix 25 parts phenol, 24 parts chloroform, and one part isoamyl alcohol. This solution can be stored under TE buffer (10mM Tris, 1mM EDTA, pH 8.0) or 0.1M Tris, pH 8.0, for periods up to 3 months at 2°–8°C. Store in an amber bottle to protect from light. If storing under TE Buffer, we recommend you add a small volume of Tris buffer.

**Note:** Chloroform is mixed with phenol to increase the efficiency of nucleic acid extractions by reducing losses of DNA and RNA at the phenol/aqueous interphase. Chloroform denatures proteins and aids in the removal of lipids, while isoamyl alcohol reduces foaming during the extraction and facilitates the separation of the aqueous and organic phases.

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## Appendix B: Co-isolating DNA from the JetStar Mini Columns

After RNA is eluted from the JetStar Mini Columns, DNA may also be eluted from the same columns using the RNeasy PowerSoil DNA Elution Kit (cat. no. 12867-25). Everything required for DNA isolation is provided in the kit.

### Important points before starting

- Wear RNase- and DNase-free gloves at all times.
- Remove RNases and DNases from work surfaces before starting.

### Procedure

1. Transfer the JetStar Mini Column from step 16 of the RNeasy PowerSoil Total RNA Kit to a 15 ml Collection Tube (provided).
2. Add 1 ml of Solution SR8 to the JetStar Mini Column to elute the bound DNA into the 15 ml Collection Tube. Allow Solution SR8 to gravity flow into the Collection Tube.
3. Transfer the eluted DNA to a 2.2 ml Collection Tube (provided) and add 1 ml of Solution SR4. Invert at least once to mix and incubate at  $-15^{\circ}\text{C}$  to  $-30^{\circ}\text{C}$  for 10 min.
4. Centrifuge the 2.2 ml Collection Tube at  $13,000 \times g$  for 15 min at room temperature to pellet the DNA.
5. Decant the supernatant and invert the 2.2 ml Collection Tube onto a paper towel for 10 min to air dry the DNA pellet.
6. Resuspend the DNA pellet in 100  $\mu\text{l}$  of Solution SR7.

**Note:** Although RNA carryover does not occur with most soil types, certain soils high in organic matter may present unique carryover situations. When the absence of RNA contamination is critical, an RNase treatment of the isolated DNA is recommended; please refer to the Troubleshooting Guide in the Handbook for instructions.

# Ordering Information

<b>Product</b>	<b>Contents</b>	<b>Cat. no.</b>
RNeasy PowerSoil Total RNA Kit (25)	For 25 preps: Isolate high quality total RNA from all soil types	12866-25
RNeasy PowerSoil DNA Elution Kit (25)	For 25 preps: Co-isolate DNA and RNA from soil using the RNeasy PowerSoil Total RNA Kit	12867-25
LifeGuard Soil Preservation (100 ml)	Stabilize ambient temperature of microbial RNA in soil	12868-100
LifeGuard Soil Preservation (1000 ml)	Stabilize ambient temperature of microbial RNA in soil	12868-1000
DNeasy PowerSoil Kit (50)	For 50 preps: Isolate microbial genomic DNA from all soil types	12888-50
DNeasy PowerSoil Kit (100)	For 100 preps: Isolate microbial genomic DNA from all soil types	12888-100
DNeasy PowerLyzer PowerSoil Kit (50)	For 50 preps: Isolate DNA from tough soil microbes, optimized for use with bead-based homogenizers	12855-50
DNeasy PowerLyzer PowerSoil Kit (100)	For 100 preps: Isolate DNA from tough soil microbes, optimized for use with bead-based homogenizers	12855-100
RNeasy PowerMicrobiome Kit (50)	For 50 preps: Isolate total RNA from stool and gut material	26000-50
RNeasy PowerWater® Kit (50)	For 50 preps: Isolate total RNA from filtered water samples	14700-50-NF

Product	Contents	Cat. no.
RNeasy PowerPlant® Kit (50)	For 50 preps: Isolate total RNA from plant and seed samples, including those high in polyphenols and polysaccharides.	1350-500
DNase Max Kit	For 50 preps: Remove genomic DNA contamination in RNA preparations using a high-activity DNase I enzyme and a highly specific DNase removal resin	15200-50

For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit handbook or user manual. QIAGEN kit handbooks and user manuals are available at [www.qiagen.com](http://www.qiagen.com) or can be requested from QIAGEN Technical Services or your local distributor.

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## Notes

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## Notes

Trademarks: QIAGEN<sup>®</sup>, Sample to Insight<sup>®</sup>, DNeasy<sup>®</sup>, RNeasy<sup>®</sup>, DNase Max<sup>®</sup>, LifeGuard<sup>®</sup>, PowerSoil<sup>®</sup>, PowerLyzer<sup>®</sup>, PowerPlant<sup>®</sup>, PowerWater<sup>®</sup>, QIAvac<sup>®</sup>, (QIAGEN Group), Vortex-Genie<sup>®</sup> (Scientific Industries). Registered names, trademarks, etc. used in this document, even when not specifically marked as such, are not to be considered unprotected by law.

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Use of this product signifies the agreement of any purchaser or user of the product to the following terms:

1. The product may be used solely in accordance with the protocols provided with the product and this handbook and for use with components contained in the kit only. QIAGEN grants no license under any of its intellectual property to use or incorporate the enclosed components of this kit with any components not included within this kit except as described in the protocols provided with the product, this handbook, and additional protocols available at [www.qiagen.com](http://www.qiagen.com). Some of these additional protocols have been provided by QIAGEN users for QIAGEN users. These protocols have not been thoroughly tested or optimized by QIAGEN. QIAGEN neither guarantees them nor warrants that they do not infringe the rights of third-parties.
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