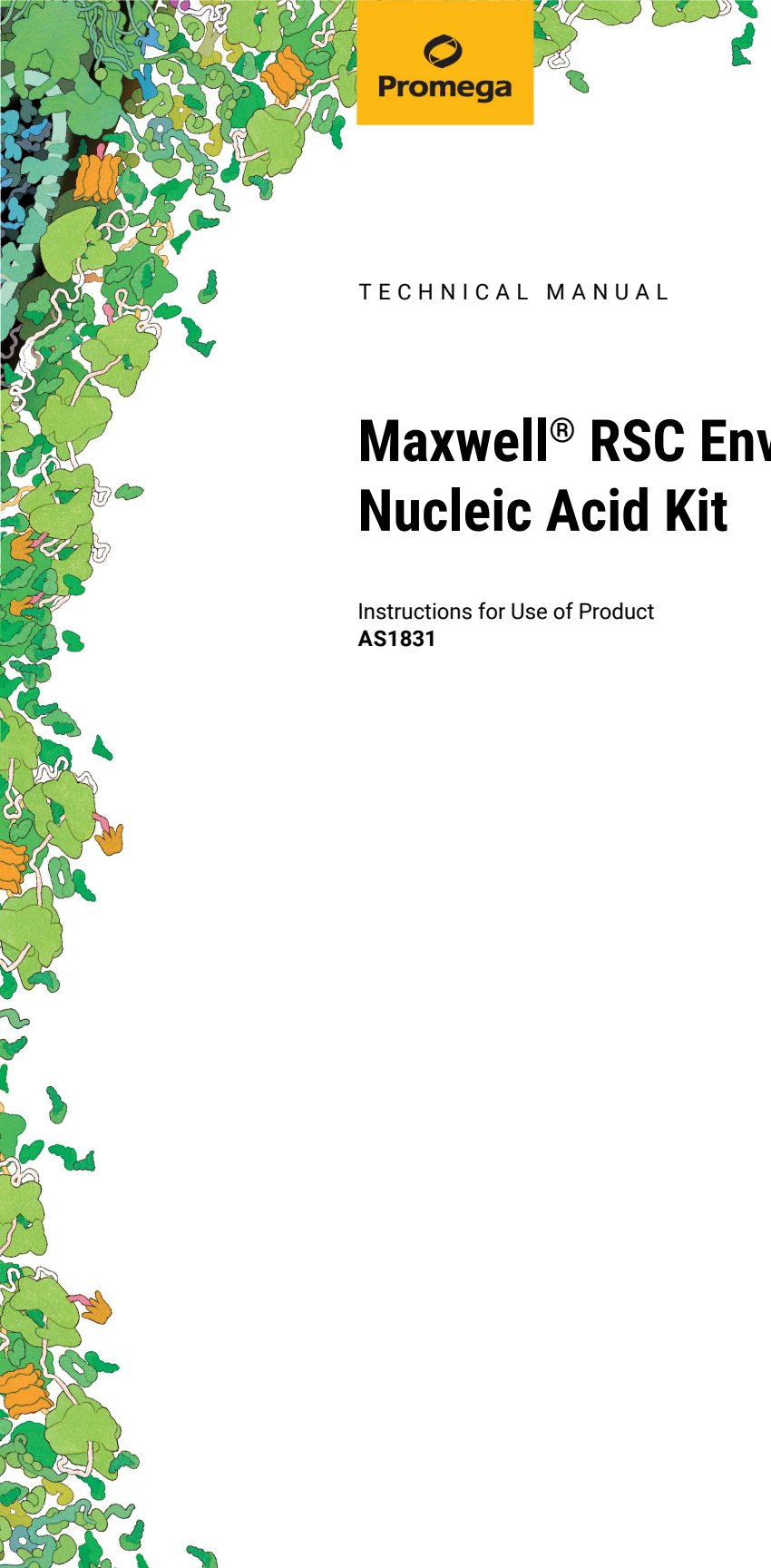


TECHNICAL MANUAL

Maxwell® RSC Enviro Total Nucleic Acid Kit

Instructions for Use of Product
AS1831

Revised 1/23
TM663

Maxwell® RSC Enviro Total Nucleic Acid Kit

All technical literature is available at: www.promega.com/protocols/
Visit the web site to verify that you are using the most current version of this Technical Manual.
E-mail Promega Technical Services if you have questions on use of this system: techserv@promega.com

1. Description

The process of detecting genetic signatures in wastewater samples involves collecting wastewater, either as a grab sample or as a 24-hour composite sample. This is followed by an optional pasteurization and sample concentration. Viral or other microbial matter and/or its genetic signature may be present at a low concentration in water samples, making sample concentration a prerequisite for sensitive detection. Concentrating microbial matter can be performed using a variety of methods such as charged membrane filtration, centrifugal ultrafiltration and flocculation/precipitation using skim milk or polyethylene glycol (PEG)/NaCl. Most of the concentration methods were originally developed to concentrate live matter with the objective of culturing for detection of viruses or bacteria, though concentrated samples have also been used for PCR-based detection. These methods have proven to be inconsistent, labor intensive and time consuming.

To address these issues, we have developed a convenient method to directly capture and concentrate total nucleic acid (TNA) from a large volume of water using PureYield™ columns. The method uses a short protocol that minimizes the need for specialized laboratory equipment. In a first step total nucleic acid from a large volume sample (e.g., 40ml of wastewater) is captured on a PureYield™ Binding Column and then eluted in 0.5ml. In a second step the material is purified and concentrated using the Maxwell® RSC System for walk-away-automation with fast, reliable total nucleic acid purification. This method achieves high yields, consistent recovery and a significant reduction in PCR inhibitors (1).

The total nucleic acid extracted using the Maxwell® RSC Enviro Total Nucleic Acid Kit[a] (Cat.# AS1831) can be analyzed for SARS-CoV-2 targets using a SARS-CoV-2 RT-qPCR kit for wastewater. Please visit the Promega web site for more information on these products:
www.promega.com/applications/infectious-diseases/covid19-wastewater-sars-cov-2-detection/

Table 1. Supported Instruments. The following instruments can be used with the Maxwell® RSC Enviro Total Nucleic Acid Kit.

Instrument	Cat.#	Technical Manual #
Maxwell® RSC	AS4500	TM411
Maxwell® RSC 48	AS8500	TM510
Maxwell® CSC 48 RUO Mode	AS8000	TM628
Maxwell® FSC	AS4600	TM462

2. Product Components and Storage Conditions

PRODUCT	SIZE	CAT.#
Maxwell® RSC Enviro Total Nucleic Acid Kit	**48 preps**	**AS1831**

Contains sufficient materials and reagents for 48 samples. Includes:

- 2 × 320ml Binding Buffer 1 (BBD)
- 2 × 30ml Binding Buffer 2 (BBE)
- 1 × 30ml Protease Solution
- 2 × 85.3ml Column Wash 1(CWE)
- 2 × 206ml Column Wash 2 (RWA)
- 1 × 150ml Nuclease-Free Water
- 12 × 4 each PureYield™ Binding Columns
- 1 × 48/pack RSC Plungers
- 48 × 1 each Maxwell® Cartridge (RSCJ)
- 1 × 50/pack Elution Tubes (0.5ml)
- 2 × 25 each Reservoir Extension Funnel

Storage Conditions: Store all components at +15°C to +30°C.

3. Before You Begin

Materials to Be Supplied by the User

- isopropanol
- ethanol, 95%
- tabletop centrifuge (capable of 3,000 × *g*)
- swinging bucket rotor (that accommodates 50ml tubes)
- 50ml disposable plastic snap-cap tubes (e.g., Eppendorf SnapTec® 50, Cat.# 0030118677) or screw-cap tubes (e.g., Corning® or Falcon® brand)
- 1.5ml microcentrifuge tubes
- heat block (capable of reaching 60°C)
- vacuum manifold (e.g., Vac-Man® Laboratory Vacuum Manifold, Cat.# A7231)
- Eluator™ Vacuum Elution Device (Cat.# A1071)
- vacuum pump, single- or double-stage, producing pressure of approximately 650mm Hg (25.6 inches Hg, 12.57psi, 86.7kPa).
- Maxwell® Instrument (e.g., Cat.# AS4500 or as noted in Table 1)

Note: Start-up bundles include hardware (Eluator™ Vacuum Elution Device, Vac-Man® Laboratory Vacuum Manifold, Vac-Man® Jr Laboratory Vac Manifold #10, Polypropylene Vacuum Flask, and vacuum-grade tubing) and a vacuum pump. The vacuum pump is available by region. See Section 8.C, Related Products.

3. Before You Begin (continued)

Prepare the following solutions prior to beginning nucleic acid extractions in Section 4.

Column Wash 1 (CWE): Add 57ml of isopropanol to the Column Wash 1 (CWE) bottle and mark on the bottle "plus isopropanol". The reagent is stable at +15°C to +30°C when tightly capped.

Column Wash 2 (RWA): Add 350ml of 95% ethanol to each Column Wash 2 (RWA) bottle and mark the bottle "plus ethanol". The reagent is stable at +15°C to +30°C when tightly capped.

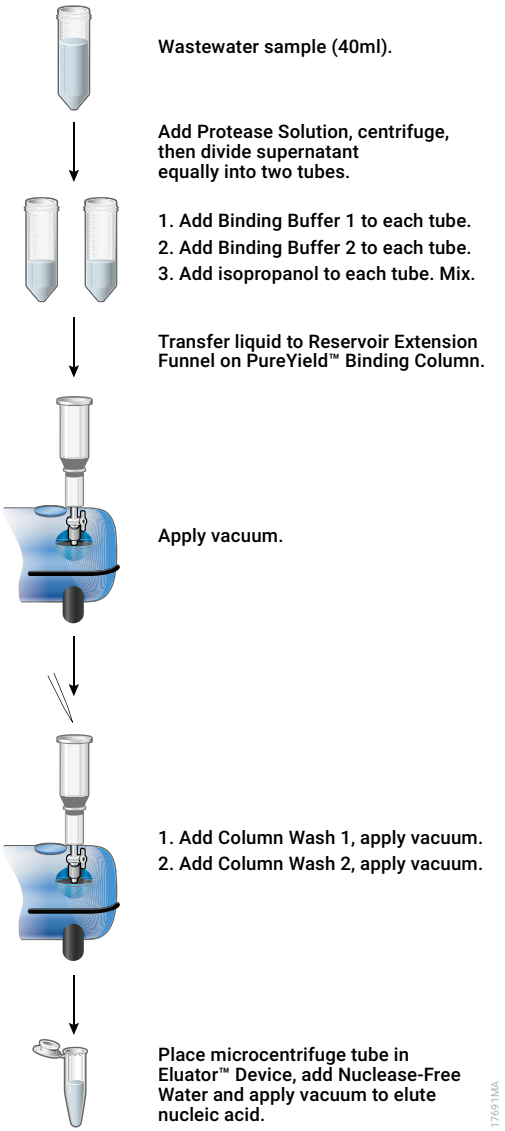


Figure 1. Schematic for direct capture of nucleic acid from wastewater using a Reservoir Extension Funnel on a PureYield™ Binding Column.

4. Standard Protocol for Capture, Concentration and Clean-Up

4.A. Capture and Concentration

1. Dispense 40ml of wastewater into a 50ml conical tube.

Note: Either pasteurized or unpasteurized wastewater samples can be processed in this protocol. To pasteurize wastewater, incubate at 60°C for 1 hour. Please follow your institution's biosafety guidelines.

2. Preheat 1.2ml of Nuclease-Free Water, per sample, to 60°C for 2–5 minutes.
3. Add 0.5ml of Protease Solution. Mix well by inversion and incubate for 30 minutes at ambient temperature.
4. Clarify sample by centrifuging at 3,000 × *g* for 10 minutes.

   Note: It is important to remove solids to avoid clogging the PureYield™ Binding Column.

5. Carefully decant 20ml of the supernatant into each of two clean 50ml conical tubes. Discard the 50ml conical tube containing the pellet into an appropriate biohazard waste container.

   Note: If you wish to process the pelleted solids to collect additional total nucleic acid, see Section 6.A.

6. To each tube containing 20ml of the clarified supernatant, add 6ml of Binding Buffer 1 (BBD) followed by 0.5ml of Binding Buffer 2 (BBE).
7. Mix well by inversion.
8. Add 24ml of isopropanol to each tube.
9. Mix well by inversion.
10. Prepare the vacuum manifold assembly. Please refer to the Q*uick Start Guide for Assembly of a Vacuum Apparatus with the Welch® Vacuum Pump* #TB355 for details. Note: For optimal results, we recommend confirming that the vacuum pressure is ≥60 kPa. Lower vacuum pressure may result in slow sample flowthrough.

11. Remove the vacuum port cap. Attach a Reservoir Extension Funnel to the PureYield™ Binding Column, then connect the column to the vacuum manifold by pressing the nozzle gently into the vacuum port (Figure 2). Using the Reservoir Extension Funnel allows up to 100ml of sample mixture to be added to the PureYield™ Binding Column at one time.

Figure 2. The Reservoir Extension Funnel and PureYield™ Binding Column attached to a Vac-Man® Vacuum Manifold port.

12. Pour the mixture from each tube from Step 8 into the Reservoir Extension Funnel on the PureYield™ Binding Column (combine both tubes of the same sample if applicable), turn on the pump and apply vacuum to capture TNA on the column.

    Notes:

    a. To ensure even vacuum pressure is applied to samples, close all Luer-Lok® Stopcocks at unused positions on the vacuum manifold before turning on the vacuum pump.

    b. Empty the liquid waste collected in the blue Vac-Man® Laboratory Vacuum Manifold (Cat.# A7231). Dispose of the alcohol-containing waste following your institutional policies.

    c. If the manifold flow rate is <2ml/minute or the pressure reading on the vacuum pump is lower than 10 inches (250mm) of Hg or >60kPa, check that all of the valves on the unused ports of the vacuum manifold are closed. If flow rate is still lower than recommended (or desired), there may be a leak in the tubing connections, the Luer-Lok® Stopcocks or the vacuum manifold.

13. Add 5ml of Column Wash 1 (CWE) and apply a vacuum to pull the liquid through the PureYield™ Binding Column.

14. Add 20ml of Column Wash 2 (RWA) and apply a vacuum to pull the liquid through the PureYield™ Binding Column. Continue the vacuum for an additional 30 seconds after all liquid has passed through the membrane.

15. Release the vacuum by turning off the vacuum pump and opening ports at unused positions or unseating the stopper of the sidearm flask. Remove the column from the vacuum manifold.

4.A. Capture and Concentration (continued)

16. Assemble the elution device by placing a 1.5ml microcentrifuge tube into the base of the Eluator™ Vacuum Elution Device (Cat.# A1071) and securing the tube cap in the open position, as shown (Figure 3, Panel A). Insert the PureYield™ Binding Column into the top of the Eluator Device, making sure the column is fully seated on the collar as shown in Figure 3, Panel B.

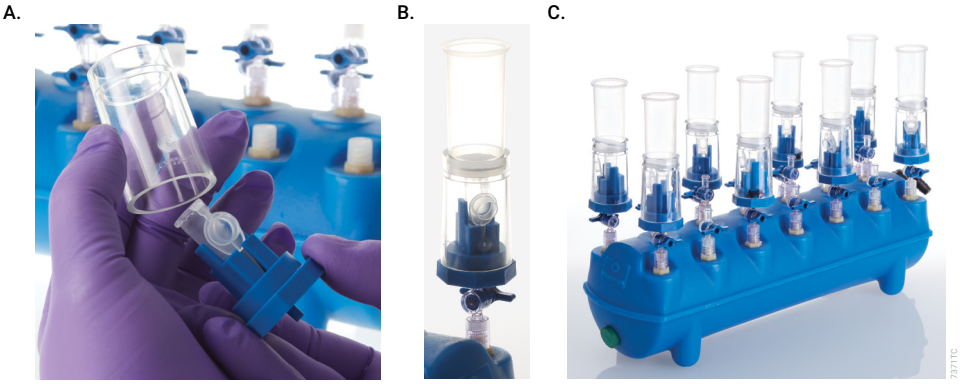


Figure 3. Elution by vacuum. Panel A. A 1.5ml microcentrifuge tube is placed in the base of the Eluator™ Vacuum Elution Device with the microcentrifuge tube cap is locked as shown. **Panel B.** The final Eluator™ Vacuum Elution Device assembly, including the binding column, ready for use on a vacuum manifold**. Panel C.** Example of eight Eluator™ Vacuum Elution Devices with binding columns assembled onto a Vac-Man® Laboratory Vacuum Manifold.

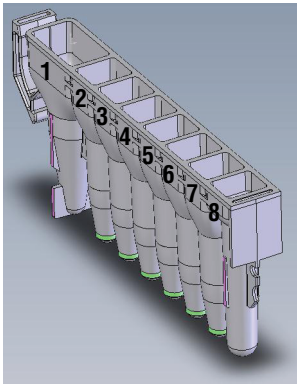
17. Place the Eluator™ Device assembly onto a vacuum manifold (Figure 3, Panel B). Add 250µl of preheated (60°C) Nuclease-Free Water to the PureYield™ Binding Column. Apply maximum vacuum for 1 minute or until all liquid has passed through the column. Repeat the process by adding another 250µl of preheated Nuclease-Free Water to the PureYield™ Binding Column to elute a total of 0.5ml of TNA solution.

    Note: You can reuse the Eluator™ Vacuum Elution Device after cleaning with 70% ethanol or other standard laboratory disinfectants.

5. Total Nucleic Acid Extraction and Clean-Up on the Maxwell® Instrument

5.A. Cartridge Preparation

1. Place each cartridge to be used in the deck tray(s) with well #1 (the largest well) facing away from the elution tube (Figure 4).



User adds to wells:

1. Sample concentrate in well #1.
2. Plunger in well #8.

Figure 4. Maxwell® RSC Cartridge.

2. Press down on the cartridge to snap it into position. Carefully peel back the seal so that all plastic comes off the top of the cartridge. Ensure that all sealing tape and any residual adhesive are removed before placing cartridges in the instrument.
3. Place a plunger in well #8 of each cartridge. Well #8 is the well closest to the elution tube.
4. Place an empty elution tube in the elution tube position for each cartridge. Add 80µl of Nuclease-Free Water to the bottom of each elution tube.

   Note: Use only the Elution Tubes (0.5ml) provided in the kit; other tubes may be incompatible with supported Maxwell® Instruments.

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5.B. Sample Processing

1. Add 150µl of Binding Buffer 1 and 50µl of Binding Buffer 2 to the 0.5ml of liquid eluted in Section 4.A, Step 15.
2. Load the entire volume of sample concentrate mixture to well #1 (the largest well).

5.C. Maxwell® Instrument Setup and Run

For detailed information, refer to the technical manual specific to your Maxwell® Instrument. See Table 1 for a list of Maxwell® Instruments and technical manuals.

1. Turn on the Maxwell® Instrument and Tablet PC. Log in to the Tablet PC and start the Maxwell® software by double-touching the icon on the desktop. The instrument will power up, proceed through a self-check and home all moving parts.
2. Touch **Start** to access the 'Methods' screen.
3. On the extraction 'Methods' screen, select a method using one of these two options:
   a. Touch the 'Enviro Total Nucleic Acid' method (option available on Maxwell® RSC Instruments only).
   b. For the Maxwell® CSC 48, use a bar code reader to scan the 2D bar code on the kit box to automatically select the appropriate method. This method is optional for the Maxwell® RSC and Maxwell® RSC 48 Instruments.
4. Verify that the 'Enviro Total Nucleic Acid' method is selected, and press the **Proceed** button. If requested by the software, enter any kit lot and expiration information that has been required by the administrator.
5. On the 'Cartridge Setup' screen (if shown), touch the cartridge positions to select or deselect the positions to be used for this extraction run. Enter any required sample tracking information, and touch the **Proceed** button to continue.

   Note: When using a 48-position Maxwell® Instrument, press the **Front** and **Back** buttons to select or deselect cartridge positions on each deck tray.

4. After the door has been opened, confirm that all Extraction Checklist steps have been performed. Verify that cartridges are loaded on the instrument, preprocessed samples are added to well #1 of the cartridges, uncapped elution tubes are present with 80µl of Nuclease-Free Water and plungers are present in well #8. Transfer the deck tray(s) containing the prepared cartridges onto the Maxwell® Instrument platform.
5. **Inserting the Maxwell® deck tray(s):** Hold the deck tray by the sides to avoid dislodging cartridges from the deck tray. Ensure that the deck tray is placed in the Maxwell® Instrument with the elution tubes closest to the door. Angle the back of the deck tray downward and place into the instrument so that the back of the deck tray is against the back of the instrument platform. Press down on the front of the deck tray to firmly seat the deck tray on the instrument platform. If you have difficulty fitting the deck tray on the platform, check that the deck tray is in the correct orientation. Ensure the deck tray is level on the instrument platform and fully seated.

   Note: Check the identifier on the 24-position Maxwell® deck trays to determine whether they should be placed in the front or back of the instrument. Deck trays are keyed and will only fit in their intended positions.

6. Touch **Start** to begin the extraction run. The platform will retract, and the door will close.

7. The Maxwell® Instrument will immediately begin the purification run. The screen will display information including who started the run, the current method step being performed and the approximate time remaining in the run.

   Notes:

   a. When using a 48-position Maxwell® Instrument, if the Vision System has been enabled, the deck trays will be scanned as the door retracts. Any errors in deck tray setup (e.g., plungers not in well #8, elution tubes not present and open) will cause the software to return to the 'Cartridge Setup' screen and problem positions will be marked with an exclamation point in a red circle. Resolve all error states, and press the **Start** button again to repeat deck tray scanning and begin the extraction run.

   b. Pressing the **Abort** button will abandon the run. The samples will be lost for all aborted runs.

   c. If the run is abandoned before completion, you will be prompted to check whether plungers are still loaded on the plunger bar. If plungers are present on the plunger bar, perform Clean Up when requested. If plungers are not present on the plunger bar, you can choose to skip Clean Up when requested. The samples will be lost for all abandoned runs.

8. Follow the on-screen instructions at the end of the method to open the door. Verify that the plungers are located in well #8 of the cartridge at the end of the run. If plungers are not removed from the plunger bar, follow the instructions in the Technical Manual appropriate to your Maxwell® Instrument (see Table 1) to perform a Clean Up process to attempt to unload the plungers. Remove the Elution Tubes containing nucleic acid and cap the tubes. Store at –30°C to –10°C. Avoid multiple freeze-thaw cycles. TNA purified using this method can be directly used for RT-qPCR.

9. After the run has been completed, the extraction run report will be displayed. From the 'Report View' screen, you can print and/or export this report.

   Note: Following the automated purification procedure, the deck tray(s) will be warm but not too hot to touch. To remove the deck tray from the instrument platform, hold onto the sides of the deck tray.

10. Remove the cartridges and plungers from the deck tray(s). Discard the cartridges and plungers as hazardous waste following your institution's recommended guidelines. Do not reuse reagent cartridges, plungers or elution tubes. Ensure samples are removed before performing any required UV light treatment to avoid damage to the nucleic acid.

6. Supplemental Protocols

Wastewater samples may contain a range of targets and solids. In some cases, processing either the solid fraction or a variable amount of water may be desirable. In the capture protocol described in Section 4, suspended solids are removed by centrifugation prior to filtration. In other cases, sludge is allowed to precipitate by gravity and then analyzed. Larger or smaller water volumes are also sometimes prescribed by local sampling protocols. The following sections describe how to extract total nucleic acids by processing either solids or a variable amount of wastewater.

6.A. Extracting Nucleic Acid from Pelleted Solids

The pelleted solids collected in Section 4.A can be processed to recover total nucleic acid as described here.

1. Add 5ml of Nuclease-Free Water to the pellet from Section 4.A, Step 4. This is the pellet recovered after treatment with the Protease Solution and after decanting the supernatant that contains the soluble suspension into a separate tube.
2. Add 1.5ml of Binding Buffer 1 (BBD) and 125µl of Binding Buffer 2 (BBE).
3. Add 6ml of isopropanol.
4. Mix well by inversion.
5. Centrifuge the mixture at 3,000 × *g* for 10 minutes.
6. The supernatant will contain nucleic acid from the solids. Add the supernatant to the Reservoir Extension Funnel on the PureYield™ Binding Column for capture of nucleic acid using the Vac-Man® Vacuum Manifold. Turn on the pump and apply vacuum to capture TNA on the column.

   Notes:

   a. Empty liquid waste collected in the blue Vac-Man® Laboratory Vacuum Manifold (Cat.# A7231). Dispose of the alcohol-containing waste following your institutional policies.

   b. If the manifold flow rate is <2ml/minute or the pressure reading on the vacuum pump is lower than 10 inches (250mm) of Hg, close the valves on the unused ports of the vacuum manifold. If this does not improve flow rate, there may be a leak in the tubing connections, the Luer-Lok® Stopcocks or the vacuum manifold.
7. Add 5ml of Column Wash 1 (CWE) and apply a vacuum to pull the liquid through the PureYield™ Binding Column.
8. Add 20ml of Column Wash 2 (RWA) and apply a vacuum to pull the liquid through the PureYield™ Binding Column. Continue the vacuum for an additional 30 seconds after all fluid has passed through the membrane.
9. Elute the captured nucleic acid using the Eluator™ Vacuum Elution Device (Figure 3) by eluting in 250µl of Nuclease-Free Water, twice, for a total elution volume of 0.5ml.
10. Proceed to Section 5.

6.B. Extracting Nucleic Acid from Sludge and Solids

An additional method for processing waste samples is to extract nucleic acid from settled solids from large volumes of wastewater or from sludge. Processing settled material can increase the sensitivity of assaying for pathogens by allowing material from larger samples to be efficiently processed.

1. To 2ml of solid material (sludge or settled solids) add 8ml of Nuclease-Free Water resulting in a 10ml final volume.
2. Add 200µl of Protease Solution, mix well and incubate for 30 minutes.
3. Add 3ml of Binding Buffer 1 (BBD) and 250µl of Binding Buffer 2 (BBE).
4. Add 12ml of isopropanol.
5. Mix well by inversion.
6. Centrifuge the mixture at 3,000 × *g* for 10 minutes.
7. The supernatant will contain nucleic acid from the solids. Add the supernatant to the Reservoir Extension Funnel on the PureYield™ Binding Column for capture of nucleic acid using the Vac-Man® Vacuum Manifold. Turn on the pump and apply vacuum to capture TNA on the column.

   Notes:

   a. Empty liquid waste collected in the blue Vac-Man® Laboratory Vacuum Manifold (Cat.# A7231). Dispose of the alcohol-containing waste following your institutional policies.

   b. If the manifold flow rate is <2ml/minute or the pressure reading on the vacuum pump is lower than 10 inches (250mm) of Hg, close the valves on the unused ports of the vacuum manifold. If this does not improve flow rate, there may be a leak in the tubing connections, the Luer-Lok® Stopcocks or the vacuum manifold.
8. Add 5ml of Column Wash 1 (CWE) and apply a vacuum to pull the liquid through the PureYield™ Binding Column.
9. Add 20ml of Column Wash 2 (RWA) and apply a vacuum to pull the liquid through the PureYield™ Binding Column. Continue the vacuum for an additional 30 seconds after all fluid has passed through the membrane.
10. Elute the captured nucleic acid using the Eluator™ Vacuum Elution Device (Figure 3) by eluting in 250µl of Nuclease-Free Water, twice, for a total elution volume of 0.5ml.
11. Proceed to Section 5.

6.C. Scaling Sample Volume

If the volume of wastewater sample needs to be adjusted, a proportional adjustment to the reagents can be made using Table 2 as a guide. Use appropriate containers for accommodating the final mixture. We recommend not using wastewater samples of less than 20ml as this may impact RT-qPCR assay sensitivity. When viral load is high, RT-qPCR signal can be obtained with as little as 5ml of sample. However, to obtain sufficient sensitivity and consistency, we recommend a sample size of 40ml.

Note: The amount of buffer and columns provided with this system is sufficient for a 40ml sample volume. Protease Solution, Binding Buffer 1 and Binding Buffer 2 are available separately (see Section 8.C).

Table 2. Proportional Sample and Reagents Volumes.

Sample volume (ml)	Protease Solution (ml)	Binding Buffer 1 (ml)	Binding Buffer 2 (ml)	Isopropanol (ml)
5	0.1	1.5	0.125	6
10	0.125	3	0.25	12
20	0.25	6	0.5	24
40	0.5	12	1	48
80	1	24	2	96

7. Considerations for Processing and Analysis

7.A. Quantitating Genome Units in the Sample

The nucleic acid purified using this method can be used for various downstream analysis and detection techniques, such as qPCR, RT-qPCR, ddPCR or sequencing. To quantitate genome units for a particular bacterial or viral target, qPCR or RT-qPCR is a commonly used technique.

7.B. Calculating Viral Genetic Material

Use the following equation to determine the viral genome units/liter in the water sample:

$$\text{Viral genome (copies/liter)} = \frac{\text{Copies in RT-qPCR} \times 1{,}000}{\text{Volume of nucleic acid extract used in RT-qPCR (ml)*} \quad \times \text{Concentration factor}}$$

*If 5µl of nucleic acid extract is used in RT-qPCR, the value in ml is 0.005.

$$\text{Concentration factor} = \frac{\text{Water sample volume used (ml)}}{\text{Volume of nucleic acid extracted (ml)}}$$

Notes:

- If a sample volume of 40ml is used and total nucleic acid is eluted in 40µl after the extraction and clean-up step, the concentration factor = 1,000.
- If a sludge sample volume of 2ml is used and total nucleic acid is eluted in 100µl of Nuclease-Free Water after the extraction and clean-up step, the concentration factor = 20.
- If a sample volume of 40ml is used and total nucleic acid is eluted in 80µl of Nuclease-Free Water after the extraction and clean-up step, the concentration factor = 500.

8. Appendix

8.A. Troubleshooting

For questions not addressed here, please contact your local Promega Branch Office or Distributor. Contact information available at: **www.promega.com**. E-mail: **techserv@promega.com**

Symptoms	Causes and Comments
Lower than expected A_{260} (yield)	Use more starting material.
	Sample is relatively low in DNA content or degraded.
	Insufficient lysis.
	Check wastewater storage conditions. We recommended processing wastewater within 48 hours of sample collection. Store wastewater at +4°C to +10°C for use within a week.
	Store wastewater samples at –20°C for long-term storage. Frozen samples should not be thawed more than once.
	Check that the correct amount of Binding Buffers 1 and 2 was used to ensure sufficient lysis and binding.
Inhibitors present	Reduce the amount of starting material used per sample.
Sample takes a long time to flow through the PureYield™ Column	Reduce the amount of starting material used per sample.
	Check the pressure in the vacuum pump and vacuum system.
	Consider replacing the stopcocks, tubing or vacuum manifold.

8.B. Frequently Asked Questions (FAQ)

1. I forgot to preheat the Nuclease-Free Water prior to extraction. Is this ok?

   Answer: Total nucleic acid will still be eluted if the Nuclease-Free Water is not preheated.

2. Can nucleic acid extracted from the first step be stored for later use?

   Answer: Store the extracted nucleic acid at –20°C for short-term storage (<6 months) and at –80°C for long-term storage (>6 months).

   Consider eluting total nucleic acid in a TE buffer such as Elution Buffer (Cat.# A8281) for long-term storage. Consider the appropriate downstream application when using a TE buffer for eution. TE Buffer (10mM Tris [pH 8], 0.1mM EDTA).

3. Can a laboratory vacuum line used for the vacuum process?

   Answer: The laboratory in-line vacuum can be used with the Vac-Man® Manifold (Cat.# A7231), if the pressure is appropriate. However, house vacuum systems can be variable and we recommend a dedicated vacuum source for best results.

8.C. Reference

1. Mondal, S. *et al*. (2021) A direct capture method for purification and detection of viral nucleic acid enables epidemiological surveillance of SARS-CoV-2. *Sci Total Environ*. **795**, 148834.

8.D. Related Products

Buffers and Solutions

Product	Size	Cat.#
Binding Buffer 1 (BBD)	320ml	A2981
Binding Buffer 2 (BBE)	30ml	MC1501
Protease Solution	30ml	A1442
Elution Buffer	50ml	A8281
RQ1 RNase-Free DNase	1,000 units	M6101

Start Up Bundle (includes Kit, Pump, Vacuum Trap, Vac-Man® Manifold, Eluator™ Device, tubing)

Product	Size	Cat.#
Maxwell® RSC Enviro Total Nucleic Acid Start up Kit (NA)	1 each	A3070
Maxwell® RSC Enviro Total Nucleic Acid Start up Kit (EU)	1 each	A3080

Nucleic Acid Purification and Detection

Product	Size	Cat.#
Eluator™ Vacuum Elution Device	4 each	A1071
ReliaPrep™ RNA Miniprep System*	50 preps (cells)	Z6010
	50 preps (tissues)	Z6110
ReliaPrep™ RNA Clean-Up and Concentration System*	10 preps	Z1071
Maxwell® RSC Instrument	1 each	AS4500
Maxwell® RSC 48 Instrument	1 each	AS8500
Vac-Man® Laboratory Vacuum Manifold	1 each	A7231
Vac-Man® Jr. Laboratory Vacuum Manifold	1 each	A7660
Luer-Lok® Stopcocks	10 each	A7261

*Additional kit sizes are available.

9. Summary of Changes

The following changes were made to the 1/23 version of this document:

1. Revised Sections 1, 3, 4 and 8.D.
2. Added new Section 8.C.
3. Updated document font.

[a]U.S. Pat. No. 7,329,488 and S. Korean Pat. No. 100483684.

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