SmartLid™
Viral DNA/RNA Extraction Starter Kit

Research Use Only. Not for use in diagnostic procedures.

Catalogue Number: 100125 **Quantity: 50 Extractions**

Definition of symbols used	
	Instructions for use
REF	Reference number or Catalogue Number
	Manufacturer
	Storage temperature range
LOT	LOT Number
	Expiry Date
	Corrosive
	Harmful
	Serious health hazard

Resources and Training Videos

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Contents

1. Introduction

Product Description

The SmartLid Viral DNA/RNA Extraction Kit has been designed for the rapid extraction of viral DNA and RNA from a variety of sample storage media and cell-free (or nearly cell-free) liquid samples. For example, the kit is compatible with biological fluids such as saliva, urine, plasma, serum, as well as water and inactivating or non-inactivating viral transport media. The kit can also process dry swabs.

Purified extractions are suitable for a variety of downstream molecular applications, such as amplification reactions (RT-PCR, RT-LAMP, etc.) and further analytical procedures.

The SmartLid Viral DNA/RNA Extraction Kit leverages a novel (patent pending) sample preparation method for nucleic acid extraction, centring around a proprietary magnetic lid, called SmartLid, to transfer nucleic acids through three simple steps: Lysis, Wash, and Elution. SmartLid requires no electricity and enables ultra-fast (less than 10 minutes), user-friendly, and economical nucleic acid extractions. The procedure is based on magnetic separation and utilizes the fastest collecting superparamagnetic beads on the market.

Advantages

Key Features	
High Yield	Ultra-pure DNA/RNA for sensitive downstream applications in molecular biology
Rapid	From sample to eluted DNA/RNA in less than 10 minutes
Easy to use	Simple process that does not require any powered equipment
Accessories	SmartLid Rack and SmartLid Shaker provide simultaneous processing of up to 12 samples
Environment	Designed to minimise plastic waste Carton and paper inserts made with recycled content and are fully recyclable Magnetic keys are reusable, reducing rare-earth waste

2. Product Specifications

Key Specifications	
Applications	PCR, RT-PCR, qPCR, RT-qPCR, LAMP, RT-LAMP, dPCR, RT-dPCR (and other amplification chemistries)
Technology	Superparamagnetic beads (silica)
Main Sample Types	Cell-free, or nearly cell-free, liquid media (VTM, eNAT, PBS, water, saliva, serum, plasma, urine, etc.)
Isolated Molecules	DNA, RNA
Sample Input Volume	200 µl
Elution Volume	30-100 µL
Percentage Yield	>88% recovery
Processing	Manual, power-free
Protocol time	Less than 10 minutes
Storage conditions	Room temperature (15-25°C)

3. Safety precautions

The techniques of “good laboratory practice” should be employed when using the kit. If such practices are used, the reagents constitute a very low potential risk to health. Wear protective gloves/protective clothing/eye protection/face protection. IF ON SKIN: Wash with plenty of water. IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Take off contaminated clothing and wash it before reuse. Dispose of contents/container in accordance with national regulations. It is important to be aware of the allergic, toxic, or infectious potential of analytical samples. In all cases of doubt, or if experiencing symptoms following exposure to any ingredients, seek medical advice.

The following components of the ProtonDx SmartLid Viral DNA/RNA extraction kit contain hazardous contents. For comprehensive information on these hazards and proper handling instructions, please refer to the Material Safety Data Sheet (MSDS) available at protondx.com. Alternatively, you can request a copy of the MSDS document by emailing info@protondx.com.

Item	GHS symbol	Hazard phrases	Supplemental Information
Tube A1		Harmful if swallowed, in contact with skin or if inhaled. Causes severe skin burns and eye damage. Harmful to aquatic life with long lasting effects.	Corrosive to the respiratory tract. Contact with acids liberates very toxic gas.
Tube A2		May cause an allergic skin reaction. May cause allergy or asthma symptoms or breathing difficulties. Suspected of causing genetic defects. May cause cancer. May damage fertility. Suspected of damaging the unborn child. Harmful to aquatic life with long lasting effects.	Restricted to professional users

Safety Information	
Caution	All chemicals and biological material should be considered potentially hazardous.
	Lysis Binding bottle contains guanidine-thiocyanate. Ensure tubes are sealed prior to disposal, as when combined with bleach, guanidine-thiocyanate can react to produce a highly toxic gas.
	When working with this kit always use appropriate PPE and avoid any skin contact.
	After use, components should be disposed of using appropriate routes, in compliance with local regulations.
	Aerosol-barrier pipette tips are recommended for pipetting the sample elution.
	This kit contains magnets and magnetic materials. Please handle with care to avoid injury and damage to nearby objects. Magnetic fields may interfere with pacemakers and other medical devices. Consult with a healthcare professional before use if you have a medical implant.
Important	Ensure all reaction tubes are not damaged or cracked prior to use. If the bottles are damaged, wear gloves and protective goggles when discarding the bottles.
	Handle and discard liquid waste according to local health and safety guidelines.
	Do not add bleach or acidic components to the waste after sample preparation.

4. Storage Information

The SmartLid Viral DNA/RNA Extraction Kit should be stored dry at room temperature (15–25°C) and is stable for at least 6 months from production under these conditions. If any kit components show signs of leakage, dispose of appropriately and contact customer support.

Before every use make sure that all components are at room temperature. If any precipitate is observed within the provided solutions, dissolve by gentle warming. Note: kit components are not suitable for freezing. Storage at 2-8°C is possible, but significantly increases the chances of buffer precipitation.

5. Kit Contents

Materials Included		
Qty.	**Item**	**Description**
1.1 mL	Magnetic Beads	Silica based magnetic beads (50 mg/mL)
40 mL	Lysis Binding Buffer	Lysis and binding buffer (contains guanidine thiocyanate*)
6 mL	Wash Buffer	Wash buffer (concentrated)
5 mL	Elution Buffer	High-purity elution buffer
50 pcs	SmartLid	Proprietary transfer lids compatible with provided flip-cap tubes
150 pcs	Flip-cap tube (2 mL)	Nuclease-free flip-cap tubes (2 mL) compatible with SmartLid
12 pcs	Green magnetic key	Reusable Magnetic Keys to use in conjunction with SmartLid

* Guanidine-thiocyanate is an irritant. Not compatible with disinfecting reagents that contain bleach, refer to Section 3.

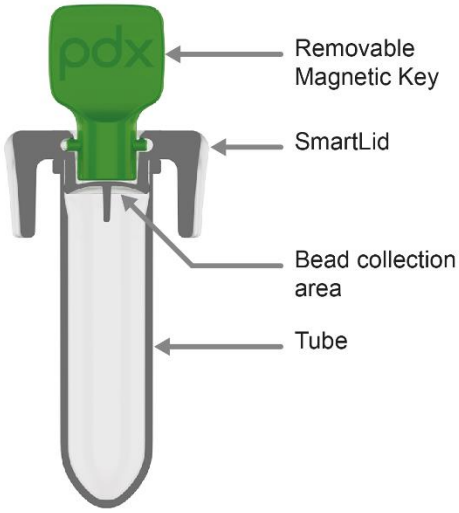
Materials not included (REQUIRED)	
Item	**Purpose**
Molecular biology grade isopropanol (>99%)	To add during the lysis binding step
Molecular biology grade ethanol (>99%)	To add to the Wash Buffer (concentrate) bottle before kit use

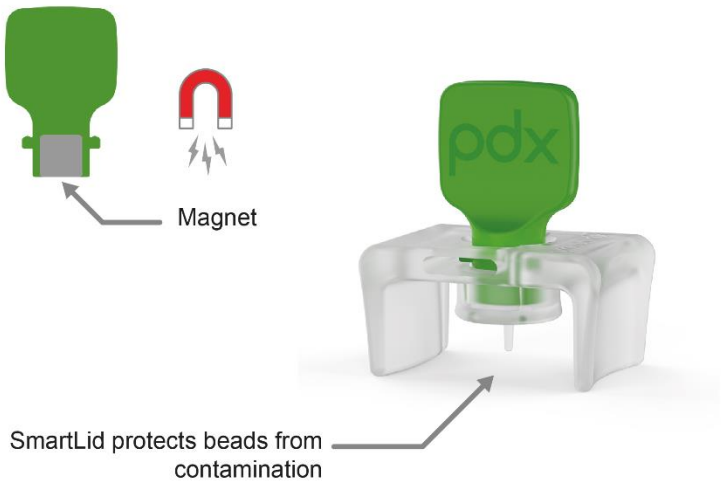
Equipment not included (can be purchased separately)		
Catalogue No.	**Item**	**Description**
100175	SmartLid Rack	Preparation rack to enable easy multi-sample processing
100173	SmartLid Shaker	Multi-tube shaking device to enable high-throughput mixing

6. Visual Guide

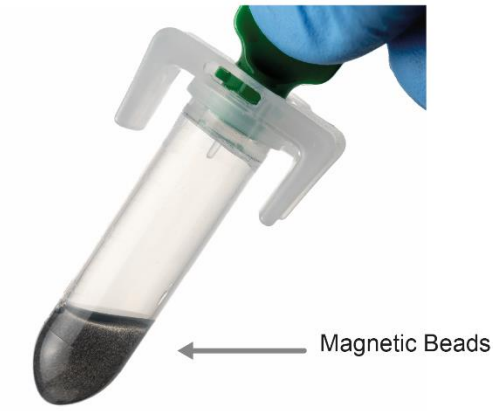
Resources and Training

What is SmartLid?





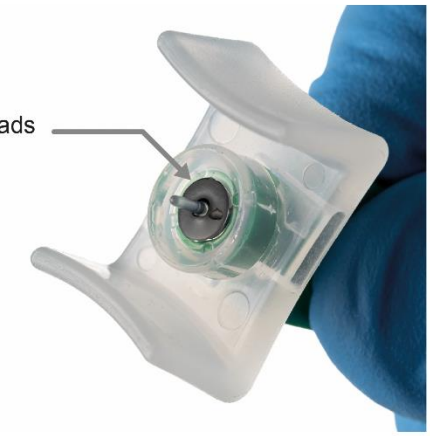
SmartLid in use



1 Magnetic Beads capture nucleic acids for transfer between steps.

2 Inverting the tube with the Magnetic Key inserted collects the Magnetic Beads onto SmartLid.

3 Within seconds the liquid is clear and collection is complete.

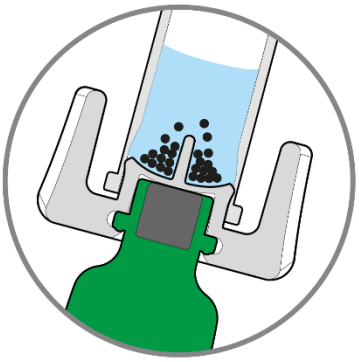


4 Magnetic Beads are safely attached to SmartLid and ready for transfer into the next tube.

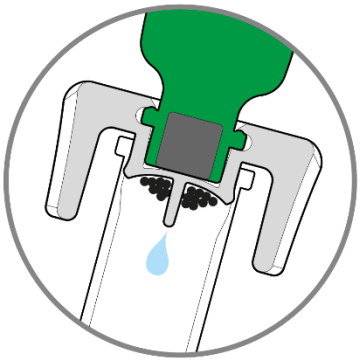
How to transfer Magnetic Beads with SmartLid

SmartLid is designed to easily transfer Magnetic Beads through a series of sample extraction steps.

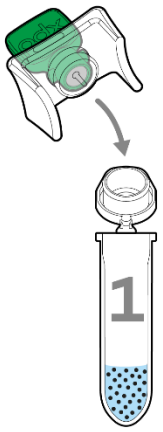
The SmartLid protocol repeats the following simple transfer process several times.



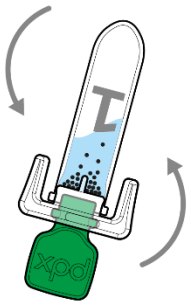
A powerful removable Magnetic Key is utilised to either capture or release the Beads.



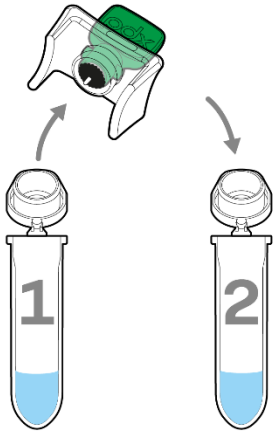
Residual buffer efficiently wicks away from the Beads limiting carry-over.



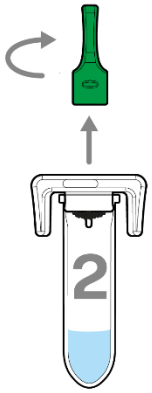
1 **Capture**: Insert SmartLid with Magnetic Key into tube 1.



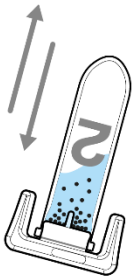
2 Invert the tube several times to collect the Beads until liquid is clear.



3 **Transfer**: Remove the SmartLid with collected Beads from tube 1 and place into tube 2.



4 **Release**: Remove the Magnetic Key to release the Beads.



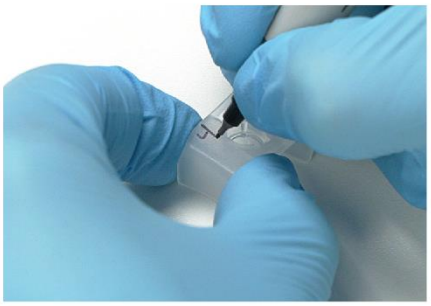
5 Resuspend the Beads through gentle mixing.

6 Beads with attached nucleic acids are successfully transferred to tube 2.

Multi-sample processing

Multiple samples can be easily processed simultaneously using the optional SmartLid Rack and SmartLid Shaker accessories.

Setup: Set out and prefill the required number of SmartLids and tubes. Labelling each SmartLid provides easy sample tracking.



Mixing: The SmartLid Shaker provides a convenient solution for simultaneously mixing up to 12 samples at a time.

Bead collection: Inverting the SmartLid Shaker, with Magnetic Keys inserted, enables rapid and efficient Magnetic Bead collection.

7. Protocols for Extraction of Viral DNA/RNA

IMPORTANT Before Starting

When opening a new SmartLid Viral DNA/RNA Extraction Kit, **add 24 mL of molecular biology grade ethanol (>99%) to the Wash Buffer (concentrate)** as indicated on the bottle. There is a box on the bottle label to check once this is completed.

Always ensure all buffers are liquid and homogeneous prior to use. In the case of visible precipitate, redissolve by warming and gentle mixing.

Resources and Training Videos

If using the SmartLid Viral DNA/RNA Extraction Kit for the first time, we recommend watching our instructional videos by visiting www.protondx.com or scanning the QR code above.

SETUP for Multiple Extractions

1. Set up the required number of flip-cap tubes in a rack (Recommended: SmartLid Rack 100175), with different rows for **Lysis** (Row A), **Wash** (Row B), and **Elution** (Row C). For example, 6 extractions will require a total of 18 tubes, split in 3 rows of 6 tubes.

2. If extracting from a **liquid sample**, we recommend creating a master mix of Lysis Binding Buffer and Magnetic Beads to ensure more consistent extractions. Prepare this master mix according to ratios in the table below, and multiply by the number of extractions:

Component	Volume per tube [1]	Volume of Master Mix for n extractions
Lysis Binding Buffer	700 µL	700 µL × **n**
Magnetic Beads	20 µL [2]	20 µL × **n** [2]
Total volume	720 µL [2]	720 µL × **n** [2]

[1] Use 10% overage calculation when making a master mix for use with multiple samples.

[2] Up to 40 µL of Magnetic Beads per sample can be used to improve yield if necessary, resulting in a master mix volume per extraction of 740 µL.

Note: If extracting directly from dry swabs without a liquid transport medium, skip this step (step 2) and follow the second half of the **Lysis Binding** section below ("Extracting from dry swabs without transport medium").

3. Prefill all tubes according to the table below:

Tube/Step	Components and Volumes	
A: Lysis Binding	***From Liquid Media:*** **720 µL** [1] Lysis Binding Master Mix	***From Dry Swab:*** **700 µL** Lysis Binding Buffer **ONLY*** **(*Magnetic Beads will be added later.)**
B: Wash	**300 µL** Wash Buffer [2]	
C: Elution	**50 µL** [3] Elution Buffer	

[1] See note above regarding Magnetic Bead concentration/volume.

[2] Ensure that 24 mL of molecular biology grade ethanol (>99%) has been added to the Wash Buffer (concentrate) as indicated on the bottle.

[3] As little as **30 µL** can be used. While this will result in a more concentrated purified sample, final recovery may be less than 30 µL. *DO NOT centrifuge tubes with SmartLids inserted in order to recover more elution from the lid.*

LYSIS BINDING

Extraction from liquid sample:

1. Add up to **200 µL** of sample to the lysis tube (prefilled with Lysis Binding Buffer and Magnetic Beads master mix).

2. Close with a SmartLid (without a Magnetic Key inserted) and briefly shake the tube (**1-2 times**) to ensure the sample and Lysis Binding Buffer is sufficiently mixed.

3. Wait **2-5 minutes** for complete lysis of the sample.

   Note: For any sample stored and preserved in lysing/inactivating media (e.g., Copan eNat® Buffer or similar), skip this step and proceed directly to Step 4.

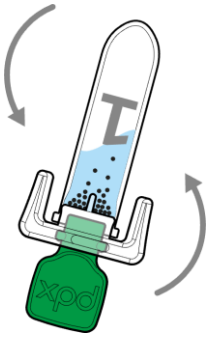
4. Temporarily remove the SmartLid and add **500 µL** of IPA (>99%) to the lysed sample.

5. Return the SmartLid (still without the Magnetic Key) to the tube and mix/shake for **60 seconds**.

   Note: For this mixing/shaking step, and all to follow, the SmartLid Shaker (100173) can be used to conveniently enable simultaneous mixing of up to 12 samples at a time. Always ensure that the retainer arm of the SmartLid Shaker is latched securely before shaking. (See instructional video for more information at www.protondx.com or the QR code above.)

6. Insert a Magnetic Key into the SmartLid, turning it 90 degrees clockwise to lock, and invert the tubes several times to collect all Magnetic Beads onto the SmartLid.

   Note: This collection step is the longest of the entire protocol, due to the large volume and high viscosity of the Lysis Binding Buffer. We recommend allowing the tube to remain upside down for **~30 seconds** after the first inversion, followed by multiple quicker (**~2-3 seconds**) inversions until the liquid is clear and all Magnetic Beads are collected.



Extraction from dry swabs without transport medium:

1. Carefully place the tip of the swab into the lysis tube (with **700 µL** of Lysis Binding Buffer), swirl for **30 seconds,** then remove.

2. Close with a SmartLid and briefly shake the tube (**1-2 times**) to ensure the sample and Lysis/Binding Buffer is sufficiently mixed.

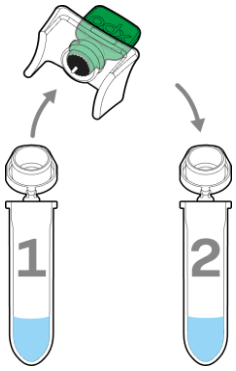
3. Wait **2-5 minutes** for complete lysis of the sample.

4. Add **500 µL** of IPA (>99%) and **20-40 µL** of Magnetic Beads.

5. Follow steps 5-6 from above to complete the lysis/binding process.

Protocol continues on the next page →

WASH



1. Transfer the SmartLid (with Magnetic Key INSERTED) from the lysis tube into the corresponding wash tube, ensuring it's firmly inserted in the wash tube.

   Note: If the Magnetic Key is not inserted during transfer of the SmartLid, the Magnetic Beads can fall off, and the captured nucleic acids will be lost.

2. Remove the Magnetic Key and mix/shake the tube for **60 seconds**, ensuring that all Magnetic Beads are fully resuspended.

3. Insert/lock the Magnetic Key into the SmartLid and invert the tube several times to collect all Magnetic Beads onto the SmartLid. Ensure the liquid is clear before proceeding.

   Note 1: It can help to initially shake the tube with the Magnetic Key inserted to first resuspend all Magnetic Beads, before gently inverting to collect the beads onto the SmartLid.

   Note 2: There is enough Wash Buffer provided to enable a second washing step. If a second wash is desired, remove the SmartLid (setting it down on any clean surface), discard the old wash buffer appropriately, refill the tube with **300 µL** of wash buffer, and repeat steps 1-3 above.

4. Once all washing steps are complete, and all Magnetic Beads are collected onto the SmartLid, remove the SmartLid and set it down (Magnetic Beads facing down) on a clean surface for **60 seconds** to **allow all EtOH to evaporate.**

ELUTION

1. Once the evaporation step is complete, insert the SmartLid (with Magnetic Key still INSERTED) into the corresponding elution tube.

2. Remove the Magnetic Key and mix/shake the tube for **60 seconds**, ensuring that all Magnetic Beads are fully resuspended.

   Note: Even though the liquid volume is significantly smaller than in previous steps, there is no need to mix/shake any differently, even when working with <50 µL.

3. Insert/lock the Magnetic Key into the SmartLid. This time, due to the small liquid volume for the elution step, after gently inverting 1-2 times, **shake the tube to fully collect all Magnetic Beads.**

4. Flick down the tube (do not centrifuge) to collect as much elution volume as possible, discard the SmartLid and attached Magnetic Beads (all nucleic acids are now released into solution), and place the closed elution tube in a cold block or on ice until further use.

   Note: Retain the Magnetic Keys for future extractions. They can be reused indefinitely to limit disposable waste.

The purified and eluted nucleic acids are now ready for immediate use in downstream applications.

8. Troubleshooting

Problem	Possible Cause	Suggested Solution
Low nucleic acid yield or quality	Quantity of Magnetic Beads	Increase the volume of Magnetic Beads per sample to as much as 40 µL. Keep other reagents in the same ratios and volumes.
	Incomplete elution or insufficient elution volume	Increase elution volume (note: this will dilute the sample). We do not recommend eluting in less than 30uL.
		Heating the elution volume (prior to adding and mixing with the Magnetic Beads) may increase final yield.
	Inconsistent resuspension of Magnetic Beads during dispenses	The included Magnetic Beads settle extremely quickly. Before dispensing from storage bottle, mix well by pipetting several times (or vortexing bottle) to make sure the beads are resuspended. Also ensure that beads are always resuspended through the binding, washing and elution steps.
		Keep a constant ratio between each buffer to match kit specification and ensure buffer amounts are correct.
	RNA/DNA is degraded	Maintain a sterile, nuclease-free environment while working to avoid any contamination.
		Make sure the elution is immediately processed or stored at -80°C after extraction is complete.
		Avoid repeated freeze/thaw cycles to preserve the RNA/DNA.
Low elution volume	When working with small volumes, liquid may adhere to the SmartLid due to surface tension	Flick down the tube to collect more elution volume from the SmartLid. Do not centrifuge tubes while SmartLid is inserted.
Magnetic Beads clumping	Dirty or viscous sample	Mix more vigorously and/or for longer periods of time. A vortex mixer can be used if necessary.
		Increase wash buffer volume to dilute contaminants or introduce a secondary wash step.
Low purity	Buffer carry-over or contamination	Introduce a secondary wash step (identical volume) or increase wash buffer volume to dilute contaminants.

9. Performance

Consistently High Performance

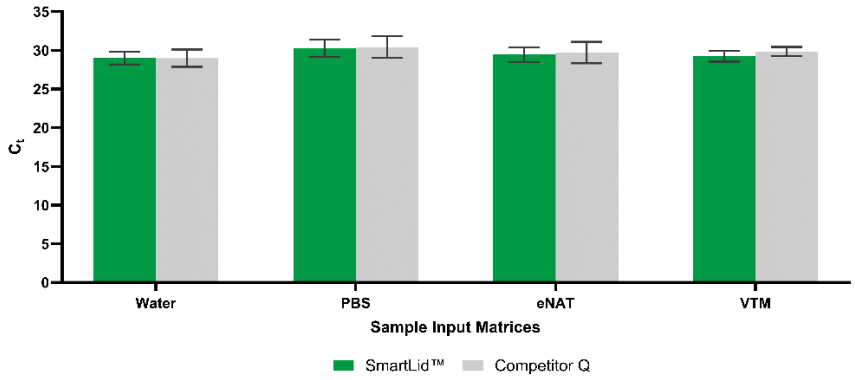


Figure 1. Extraction performance of spiked samples (200 µL of 5×10^3 copies/mL inactivated SARS-CoV-2 viral particles) with analysis in RT-qPCR versus Competitor Q.

Exceptional Recovery Rate

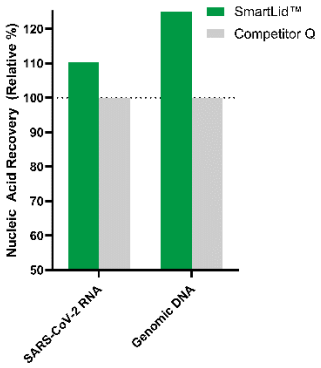


Figure 2. Relative percentage recovery of spiked SARS-CoV-2 RNA (left) and Genomic DNA (right) vs Competitor Q.

Rapid & Simple Protocol

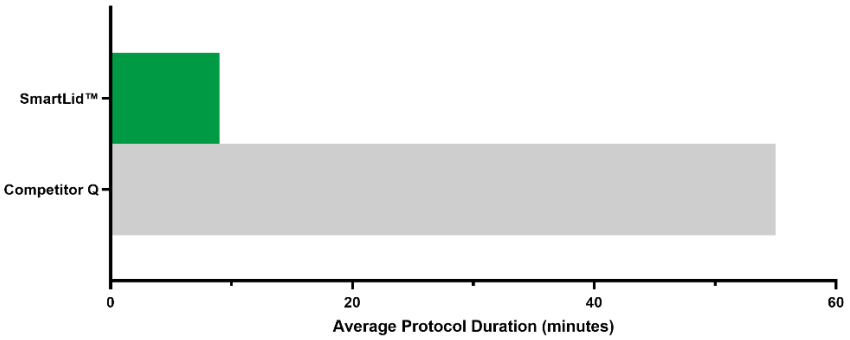


Figure 3. Comparison of average protocol time between the power-free SmartLid process and Competitor Q.

Contamination-free Workflow

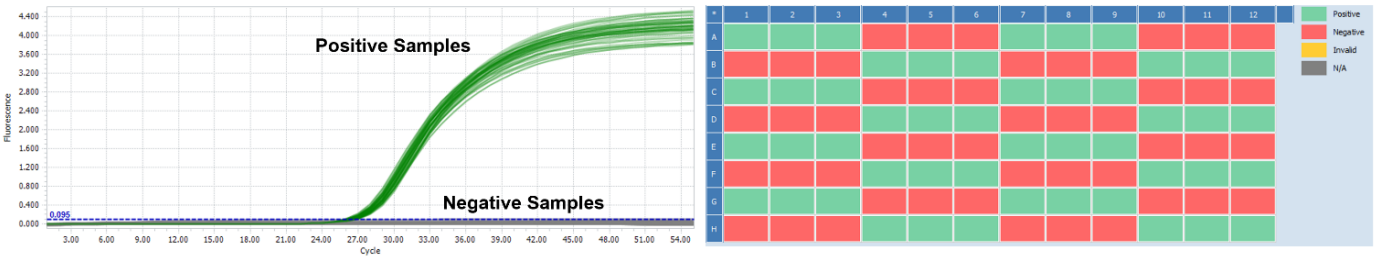


Figure 4. Alternating high-positive samples (spiked with 200 µL of 1×10^6 pfu/mL inactivated SARS-CoV-2 viral particles) and negative control samples were extracted in close proximity in a single batch. As shown by the raw RT-qPCR readout (LightCycler® LC96), all positive samples amplified while all negative samples did not amplify.

10. Ordering information

Catalogue No.	Item Name	Contents
100125	SmartLid Viral DNA/RNA Extraction Starter Kit	Everything needed to perform 50 viral extractions
100179	SmartLid Viral DNA/RNA Extraction Refill Kit	Same as starter kit, excluding reusable Magnetic Keys
100175	SmartLid Rack	Accessory rack for processing 12 samples simultaneously
100173	SmartLid Shaker	Accessory shaker for mixing 12 samples simultaneously

11. Technical Support

For any questions or to report any issues, please contact support@protondx.com.

Answers to workflow questions may be found by watching the videos and reviewing the documentation linked to the QR Code below:

Resources and Training Videos