

Quick-DNA/RNA™ HT

High-throughput, total nucleic acid purification system

Highlights

- High-throughput, magnetic-bead based purification of total nucleic acid (DNA/RNA) from any biological or clinical sample (e.g., host, plasma, serum, urine, cell culture media, blood, saliva, cellular suspensions, swab, fecal and biopsies).
- High-quality DNA/RNA is ready for Next-Gen sequencing, RT/qPCR, hybridization, etc.

Catalog Numbers: R2150, R2151



Scan with your smart-phone camera to view the online protocol/video.







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Product Contents

<i>Quick</i> -DNA/RNA™ HT	R2150 (250 preps)	R2151 (1000 preps)
DNA/RNA Buffer HT	100 ml	400 ml
Proteinase K (lyophilized)	20 mg	60 mg
Proteinase K Storage Buffer	1.2 ml (x3)	10 ml
HT MagBinding Beads	3 ml	12 ml
MagBead DNA/RNA Wash 1 (concentrate)	30 ml (x2)	120 ml (x2)
MagBead DNA/RNA Wash 2 (concentrate)	20 ml (x2)	80 ml (x2)
DNase/RNase-Free Water	30 ml	100 ml
Instruction Manual	1 pc	1 pc

Materials/Equipment Needed (user provided)

- ✓ Beta-mercaptoethanol (β-Me)
- √ Isopropanol (99.5-100%)
- ✓ Ethanol (95-100%)
- ✓ Nuclease-free water
- ✓ Vortex Mixer
- ✓ Magnetic stand or separator
- Liquid handler or robotic sample processor

Materials Available Separately (not provided)

- DNA/RNA Shield[™] (2X concentrate) (R1200-25, 25 ml)
- o ZR-96 MagStand (P1005)
- Collection Plate (C2002; capacity 1.2 ml/well)
- o 96-Well Block (P1001-2; capacity 2 ml/well)
- Elution Plate (C2003; capacity 0.35 ml/well)
- Cover Foil (C2007-4; 4 pack)
- ZR BashingBead Lysis Tubes (S6003-50; 2.0 mm beads), (S6012-50; 0.1 & 0.5 mm beads), (S6014-50; 0.1 & 2.0 mm beads)
- DNase I Set (E1010; 250 U DNase I (lyophilized) supplied with DNA Digestion Buffer, 4 ml)
- DNA/RNA Prep Buffer (D7010-2-50, 50 ml)

Specifications

Sample Sources

Any biological sample (e.g., swabs, liquids, cells, tissue, etc.), and samples collected/stored in media (e.g., UTM, VTM, saline, PBS, DNA/RNA Shield[™], PAXgene, RNAlater, RNAProtect, etc.)

Binding Capacity

5 μg DNA/RNA per 10 μl HT MagBinding Beads

Elution Volume

≥ 15 µl DNase/RNase-Free Water

Purity

DNA/RNA is ready for Next-Gen Sequencing, RT/qPCR, etc.

Storage Temperature and Stability

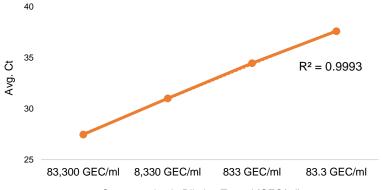
- ✓ Store all components (i.e., buffers/reagents, columns) at room temperature (15-30°C).
- Expiration dates for each of the unopened components are indicated on the individual component labels. These storage conditions apply to both opened and unopened components.
- ✓ Eluted DNA/RNA can be used immediately or stored frozen (-20/-80°C).

Product Description

The *Quick*-DNA/RNA™ HT kit is intended for rapid, high-throughput nucleic acid extraction (automated or manual) from any biological or clinical sample (e.g., swabs – nasal/nasopharyngeal, oropharyngeal, etc.; biological liquids – blood, plasma, serum, saliva, sputum, cells in suspension, etc.; tissue – needle biopsies, LCM, etc.) and/or samples stored in most collection matrices and devices (e.g., UTM, VTM, DNA/RNA Shield™, RNAlater, RNAProtect, etc.).

The kit is compatible with robotic-type sample processors (i.e, bead movers) in combination with sensitive downstream molecular amplification assays. High-quality DNA/RNA extracted with the *Quick*-DNA/RNA™ HT kit can be used for Next-Gen sequencing, RT/gPCR and more.

Limit of Detection Preliminary Assay from Biological Specimens using Automated Extraction



Concentration in Dilution Tested (GEC/ml)

Concentration	83,300 GEC/ml	8,330 GEC/ml	833 GEC/ml	83.3 GEC/ml	8.33 GEC/ml
in Dilution	(5,000 GEC/rxn)	(500 GEC/rxn)	(50 GEC/rxn)	(5 GEC/rxn)	(0.5 GEC/rxn)
Avg. Ct	27.5	31.0	34.5	37.6	Not detected
Positive, n=5	5/5	5/5	5/5	5/5	0/5

Automated extraction of whole genome viral RNA (i.e., SARS-CoV-2) spiked in sputum/swab samples collected in DNA/RNA Shield™ was performed with the *Quick*-DNA/RNA™ HT kit, followed by quantification by RT-qPCR. Preliminary limit of detection (LoD) assay determined the lowest concentration for which 5/5 independent replicates tested positive.

Protocol

The protocol consists of: (I) Buffer Preparation, (II) Sample Preparation and (III) DNA/RNA Purification.

(I) Buffer Preparation

- ✓ Add 500 µl beta-mercaptoethanol (user supplied) per 100 ml DNA/RNA Buffer HT, (final 0.5% (v/v)).
- Add 20 ml (R2150) or 80 ml (R2151) of isopropanol to the MagBead DNA/RNA Wash 1 concentrate.
- ✓ Add 30 ml (R2150) or 120 ml (R2151) of isopropanol to the MagBead DNA/RNA Wash 2 concentrate.
- Reconstitute lyophilized Proteinase K at 20 mg/ml with Proteinase K Storage Buffer and mix by vortexing. Use immediately or store frozen aliquots:
 - #R2150 (**20 mg**), add 1.04 ml **buffer** #R2151 (**60 mg**), add 3.12 ml **buffer**
- ✓ Optional: To prepare DNA/RNA Shield™ (1X)¹, dilute the 2X concentrate with an equal volume of nuclease-free water (user provided) (1:1) and mix well. Reagent is not provided (available separately, see page 10).

¹ DNA/RNA Shield™ is a sample collection medium for storage and preservation of nucleic acids. It also inactivates pathogens and prevents viral infectivity. Specimens stored and transported in DNA/RNA Shield™ can be processed directly without reagent removal, using standard laboratory operating procedures, for the detection of nucleic acids with molecular amplification assays.

(II) Sample Preparation

✓ Perform all steps at room temperature (15-30°C).

Sample Input – Depending on the sample type, up to 200 µl can be processed per prep (see examples below).

	Swabs	
Nasal/Nasopharyngeal	Oropharyngeal	Buccal/cheek
Vaginal	Fecal	

Stored in UTM, VTM, saline, PBS, DNA/RNA Shield™. etc.

Optional - To inactivate, store and preserve samples at room temperature, add 100 µl DNA/RNA Shield™ (2X concentrate)¹ to 100 µl sample (1:1). Mix well².

	Liquids (I)	
Plasma	Serum	CSF
Cells in suspension		
	and preserve samples at room	

DNA/RNA Shield™ (2X concentrate)¹ to 100 µl sample (1:1). Mix well².

	Liquids (II)	
Whole blood	Saliva	Sputum
Urine		

Recommended: To inactivate, lyse and preserve samples at room temperature, add 100 µl DNA/RNA Shield™ (2X concentrate)¹ to 100 µl sample (1:1). Mix well², centrifuge debris and process the cleared supernatant.

Tissue

Tissue (< 5 mg) Feces (< 20 mg)

LCM, needle biopsy or samples stored in media/device.

Recommended: To inactivate, lyse and preserve samples at room temperature, add 200 μ l DNA/RNA Shield (1X)^{1,3} and homogenize⁴. Mix well, centrifuge debris and process the cleared supernatant.

¹ At this point, samples in DNA/RNA Shield™ can be stored at ambient temperature (4-30°C) for a month, 7 days at 35°C, or long-term (> 1 year) at -20°C or below.

² For all buffer additions and incubation steps, **mix well** for ≥1 minute by pipetting the beads up and down and/or by shaking (vortexing) at ~1,300 rpm. Optimization may be required.

³ To prepare DNA/RNA Shield™ (1X), dilute the **2X concentrate** with an equal volume of nuclease-free water (user provided) (1:1) and mix well.

⁴ For efficient homogenization of tough-to-lyse tissue samples, bead-beat with ZR BashingBead Lysis Tubes (S6003, S6012, S6014), sold separately.

(II) Sample Preparation (continued)

Proteinase K Treatment (optional)

- ✓ At this point, protein-rich samples e.g., plasma, serum, saliva, sputum, tissue, can be treated.
- Add 1% Proteinase K (v/v) at 20 mg/ml¹ directly to a liquid sample. Mix well².
- 2. Incubate at room temperature for 15 minutes.

Note: Up to 5% Proteinase K can be added to protein-rich samples (e.g., tissue). For example: Add 2-10 μ l Proteinase K (20 mg/ml) to each 200 μ l sample.

Transfer up to **200 µI** of the prepared sample into a new tube/well and proceed to **DNA/RNA Purification, page 7**.

¹ For automation platforms with "dead volume" liquid handler dispensing, the lyophilized Proteinase K can be reconstituted to a working concentration of 4-20 mg/ml.

² For all buffer additions and incubation steps, **mix well** for ≥1 minute, by pipetting the beads up and down and/or by shaking (vortexing) at ~1,300 rpm. Optimization may be required.

(III) DNA/RNA Purification

- ✓ Perform all steps at room temperature (15-30°C).
- Add 400 μl DNA/RNA Buffer HT to each 200 μl sample¹ (2:1) and mix well².
- 2. Add 10 µl HT MagBinding Beads and mix well² for 10 minutes.
- 3. Transfer the plate/tube to a magnetic stand³ until beads have pelleted, then aspirate⁴ and discard the cleared supernatant.
- 4. Add 250 μl **MagBead DNA/RNA Wash 1** and mix well². Pellet the beads^{3,4} and discard the supernatant.
- 5. Add 250 µl **MagBead DNA/RNA Wash 2** and mix well². Pellet the beads^{3,4} and discard the supernatant.
- 6. Add 250 μ l ethanol (95-100%) and mix well². Pellet the beads³,4 and discard the supernatant.
- 7. Add 250 µl of ethanol (95-100%) and mix well². Then transfer the sample (beads and liquid) to a new plate/tube (not provided). Pellet the beads^{3,4} and discard the supernatant.
 - Optional: At this point, DNase I treatment can be performed (see Appendices, page 8).
- 8. Dry the beads for 10 minutes.
- To elute DNA/RNA from the beads, add 30 µl DNase/RNase-Free Water and mix well².
 - Alternatively, for highly concentrated DNA/RNA use ≥ 15 µl volume.
- 10. Transfer the plate/tube to a magnetic stand³ until beads have pelleted, then aspirate⁴ and dispense the eluted DNA/RNA to a new plate/tube.
 - The eluted ${\rm DNA/RNA^5}$ can be used immediately or stored frozen.

¹ Up to 200 µl sample (including the volume of DNA/RNA Shield™, if added) can be processed per prep.

² For all buffer additions and incubation steps, **mix well** for ≥1 minute, by pipetting the beads up and down and/or by shaking (vortexing) at ~1,300 rpm. Optimization may be required.

³ Use a strong-field magnetic stand or separator (e.g., ZR-96 MagStand, P1005; sold separately) until beads have pelleted.

⁴ Some beads will adhere to the sides of the well. When removing the supernatant, aspirate slowly to allow these beads to be pulled to the magnet as the liquid level is lowered.

⁵ It is recommended to titrate the DNA/RNA eluate for downstream applications (i.e., RT/qPCR, etc.).

Appendices

Automation Scripts

The **Quick-DNA/RNA™ HT** (R2150, R2151) is compatible with automated platforms*. For automation scripts and related technical support, email <u>automation@zymoresearch.com</u>. In the subject line, please include "Automation Scripts", instrument used and the product catalog number.

DNase I Treatment

✓ For DNA-free RNA, DNase I treatment can be performed using DNase I Set (E1010; 50 reactions) and DNA/RNA Prep Buffer (D7010-2-50), materials sold separately.

For each sample to be treated, prepare **DNase I Reaction Mix** in an RNase-free tube (not provided) and mix by gentle inversion:

DNase I Reaction Mix

Nuclease-free water (user provided)	40 µl
DNA Digestion Buffer	5 µl
DNase I (reconstituted; 1 U/µI) ¹	5 μl

- Add 50 µl DNase I Reaction Mix to the beads and mix gently for 10 minutes.
- 2. Add 500 µl **DNA/RNA Prep Buffer** and mix well² for 10 minutes. Pellet the beads^{3,4} and discard the supernatant.
- 3. Add 250 µl ethanol (95-100%) and mix well². Pellet the beads^{3,4} and discard the supernatant.
- Add 250 μl of ethanol (95-100%) and mix well². Then transfer the sample (beads and liquid) to a new plate/tube (not provided). Pellet the beads^{3,4} and discard the supernatant.
- 5. Proceed with step 8 in DNA/RNA Purification, page 7.

^{*}For automation platforms with "dead volume" liquid handler dispensing, additional reagents are available separately.

¹ Prior to use, reconstitute lyophilized 250 U DNase I (E1009-A) to 1U/μI (final concentration) with 275 μI nuclease-free water (user provided), mix by gentle inversion and store frozen aliquots.

² For all buffer additions and incubation steps, **mix well** for ≥1 minute, by pipetting the beads up and down and/or by shaking (vortexing) at ~1,300 rpm. Optimization may be required.

³ Use a strong-field magnetic stand or separator (e.g., ZR-96 MagStand, P1005; sold separately) until beads have pelleted.

⁴ Some beads will adhere to the sides of the well. When removing the supernatant, aspirate slowly to allow these beads to be pulled to the magnet as the liquid level is lowered.

Troubleshooting Guide

Problem	Possible Causes and Suggested Solutions		
RNA degradation	To prevent RNA degradation:		
	Immediately collect and lyse fresh sample into a stabilization reagent (i.e., DNA/RNA Shield™) to ensure nucleic acid stability. Homogenized samples in DNA/RNA Shield™ can be stored frozen for later processing.		
Low nucleic acid content and/or low	Incomplete deproteinization due to high-protein content in the sample (blood, plasma/serum, tissue etc.):		
sensitivity in downstream	- Increase the volume of DNA/RNA Shield™ to the sample.		
application	- Perform Proteinase K treatment (see Sample Preparation, page 6).		
	Increase binding time:		
	At all binding steps, increase binding time for an additional ≥10 minutes (e.g., 30 minutes). Depending on the amount of biomass, more time may be required to allow nucleic acids to be sufficiently bound to beads.		
	Inefficient washing of beads:		
	-Shaking/Mixing: Mix well for ≥1 minute, by pipetting up and down and/or by shaking (vortexing) at high speed. Make sure that the beads are resuspended throughout the bind, wash and elution steps. Optimization may be required.		
	Increase eluate input:		
	- Titrate the DNA/RNA eluate for downstream applications (i.e., RT/qPCR).		
DNA contamination	To remove DNA:		
	- Perform DNase I treatment during the purification (page 7) or perform DNase I treatment post-purification (#R1082), then clean-up the treated sample.		

For technical assistance, please contact 1-888-882-9682 or email tech@zymoresearch.com

Ordering Information

Product Description	Catalog No.	Size
<i>Quick</i> -DNA/RNA [™] HT	R2150	250 preps
<i>Quick</i> -DNA/RNA [™] HT	R2151	1000 preps

Individual Kit Component	Catalog No.	Amount
DNA/RNA Buffer HT	R2150-1-100	100 ml
Proteinase K Set supplied w/ Storage Buffer	D3001-2-20 D3001-2-60	20 mg 60 mg
MagBead DNA/RNA Wash 1 (concentrate)	R2130-1-30 R2130-1-120	30 ml 120 ml
MagBead DNA/RNA Wash 2 (concentrate)	R2130-2-20 R2130-2-80	20 ml 80 ml
HT MagBinding Beads	R2150-2-3 R2150-2-12	3 ml 12 ml
DNase/RNase-Free Water	W1001-30 W1001-100	30 ml 100 ml
DNA/RNA Shield™ (2X concentrate)	R1200-25 R1200-125	25 ml 125 ml

Notes

Notes



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