Catalog # R05220 (1X)
Catalog # R05221 (10X)

Enzymatic DNA/RNA Extraction Buffer

For research use only
Store at -20 ˚C long-term

01 INTRODUCTION

The **Enzymatic DNA/RNA Extraction Buffer** provides a robust DNA and RNA extraction method for high efficiency, high throughput sample processing for complex biological and environmental samples. The buffer is stable at 4 ˚C for short term and -20 ˚C for long term storage, and includes chemical and enzymatic properties to prevent degradation of the extracted nucleic acid and improve the accessibility of DNA and RNA for downstream applications such as PCR, qPCR and RT-qPCR.

The Enzymatic DNA/RNA Extraction Buffer includes specialized enzymes that are active at ambient temperatures and inactivated at 98 ˚C. The enzymatic composition improves the extraction yield by digesting contaminating proteins including nucleases present in a sample that may otherwise impede extraction, degrade extracted DNA/RNA, or inhibit PCR. No spin column purification, toxic phenol-chloroform extraction, or addition of other organic solvents or proteins is necessary for sample processing. The enzymatic extraction is ideal for difficult-to-lyse samples that may contain RNase, DNase, or other antimicrobial proteins such as those found in upper and lower respiratory specimens. The buffer also enhances the stability of the lysate, with extracted DNA/RNA shown to be stable for up to 72 hours at room temperature. For long-term storage, DNA may be stored at -20 ˚C, and RNA at -80 ˚C.

To process a sample, add Enzymatic DNA/RNA Extraction Buffer, briefly homogenize, and incubate at room temperature for 15 minutes followed by incubation at 98 ˚C for at least 5 minutes. Optional processing steps may be performed to improve sample yield and performance in downstream applications. DNase treatment may be performed after extraction to degrade gDNA for RT-PCR. The solution is DNase and RNase-Free, non-inhibitory to PCR, and allows high percentages of lysate to be added directly to PCR reactions for increased sensitivity. Extracted DNA or RNA can be used for direct PCR, qPCR and RT-qPCR-based analysis and screening.

Enzymatic DNA/RNA Extraction Buffer is available at 1X (Cat #R05220) and 10X (Cat #R05221) concentrations and supports different samples including viruses, bacteria, protozoa, animal tissue-culture cells, environmental samples and more. The 1X concentration is ideal for swabs, membrane filters, bacterial culture pellets, and cell pellets while the 10X concentration may be used for liquid samples such as saliva, water, biofluid and transport media. Enzymatic DNA/RNA Extraction Buffer is provided at 50 mL and 250 mL volumes for the 1X concentration, 5 mL and 50 mL volumes for the 10X concentration, and is available license-free for OEM use.

02 PRODUCT SIZING

Concentration	Size	Catalog Number	Volume
1X	1 x 50 mL	R05220S	50 mL
	5 x 50 mL	R05220M	250 mL
	5 x 250 mL	R05220L	1.25 L
	25 x 250 mL	R05220XL	6.25 L
10X	1 x 5 mL	R05221S	5 mL
	5 x 5 mL	R05221M	25 mL
	5 x 50 mL	R05221L	250 mL
	25 x 50 mL	R05221XL	1.25 L

03 PRODUCT SPECIFICATIONS

Buffer Storage

Store Enzymatic DNA/RNA Extraction Buffer 1X at -20 °C. Minimize the number of freeze/ thaw cycles. The solution can be stored at 4 °C for a week or refrozen in small aliquots.

Store Enzymatic DNA/RNA Extraction Buffer 10X at -20 °C. Minimize the number of freeze/ thaw cycles. The solution can be stored at 4 °C for a month or refrozen in small aliquots.

Lysate Storage and Stability

Lysate made with Enzymatic DNA/RNA Extraction Buffer is highly stable for both DNA and RNA. Stability may vary depending on the sample type. Perform validation testing for individual sample types.

Lysate Storage Conditions

	Up to 72 hours	Short-term	Long-term
DNA	4 – 25 ˚C	4 ˚C	-20 ˚C
RNA	4 – 25 ˚C	-20 ˚C	-70 ˚C or below

04 PROTOCOL

A. Enzymatic DNA/RNA Extraction Buffer 1X

1. Add Enzymatic DNA/RNA Extraction Buffer 1X to the sample. 200 µL - 500 µL is generally sufficient for swabs, membrane filters, and cell pellets. The required buffer volume may vary by sample type and size. Mix completely by vortexing for 15 seconds, then incubate the sample at room temperature for a minimum of 15 minutes.

Optional: Vortex at least once during the incubation period.

2. Incubate the sample at 98 ˚C for 5 minutes.

Optional: If extracted RNA will be used for PCR in conditions where primers are not designed to avoid DNA amplification, perform in-tube DNase treatment after sample incubation to remove genomic DNA (gDNA).

3. Proceed with PCR analysis or store the sample lysate at the recommended storage temperature. Large volumes of the lysate may be used in PCR reactions; up to 30% volume of the lysate has been tested in conjunction with Chai **Sahara One-Step RT-qPCR Master Mix with UNG** with no inhibition.

Note: *For solid or viscous samples such as saliva, centrifuge the lysate for 30 seconds to pellet cellular debris and utilize the supernatant for PCR analysis.*

B. Enzymatic DNA/RNA Extraction Buffer 10X

1. Add Enzymatic DNA/RNA Extraction Buffer 10X at 1:10 v/v of buffer to sample. For a 180 uL liquid sample, for example, add 20 uL of 10X buffer. The required buffer volume may vary by sample type and size. Mix completely by vortexing for 15 seconds, then incubate the sample at room temperature for a minimum of 15 minutes.

Optional: Vortex at least once during the incubation period.

2. Incubate the sample at 98 ˚C for 5 minutes.

Optional: If extracted RNA will be used for PCR in conditions where primers are not designed to avoid DNA amplification, perform in-tube DNase treatment after sample incubation to remove genomic DNA (gDNA).

3. Proceed with PCR analysis or store the sample lysate at the recommended storage temperature.

Note: For solid or viscous samples such as saliva, centrifuge the lysate for 30 seconds to pellet cellular debris and utilize the supernatant for PCR analysis.

05 TROUBLESHOOTING

1. Poor PCR amplification may result from incomplete inactivation of the enzyme included in the Enzymatic DNA/RNA Extraction Buffer. To prevent degradation of PCR reagents, incubate the sample at 98 ˚C for a minimum of 5 minutes prior to PCR.

2. If the DNA/RNA yield is low, increase the amount of initial sample, the duration of sample lysis at room temperature, and/or intensity of homogenization. Avoid harsh vortexing by allowing breaks in between pulses and store sample lysate according to recommendations to prevent DNA/RNA degradation.

3. Overloading the extraction system may result in poor DNA/RNA yield and/or PCR inhibition in downstream applications. Reduce the initial sample volume or dilute the crude extract if necessary.

4. If the sample is inhibited during PCR, centrifuge the lysate after heat incubation to pellet cell debris. Use only the supernatant for downstream applications.

06 CONTACT

For additional assistance on extraction protocol, sample stability, and PCR analysis, contact support@chaibio.com or your local distributor.

CHAI

990 Richard Ave, Suite 110 | Santa Clara, CA 95050
www.chaibio.com

sales@chaibio.com
support@chaibio.com

Toll-free +1 (800) 642-4002
International +1 (650) 779-5577

002403 Rev A