

Research Use Only

Preloaded DNA/RNA Extraction Set

User Manual

Manufacturer:

GeneReach Biotechnology Corporation

TEL: 886-4-2463-9869 Email: sales@genereach.com

No. 19, Keyuan 2nd Road., Central Taiwan Science Park, Taichung City 407, Taiwan

Website: www.tacomag.com

2015/07

Content

Symbols1
Reagent Set Components2
Shipping and Storage2
Materials and Equipment Required, but Not Provided3
Introduction4
Intended Use4
Important Notes5
Product Limitations5
Nucleic Acid Extraction Procedure6
Troubleshooting8
Appendix I—Sample Preparation10
Appendix II—Purity Analysis and Quantification of Nucleic
Acid12

tacoTM Preloaded DNA/RNA Extraction Set

Symbols



Date of manufacturing



Manufacturer



Lot number

Reagent Set Components

taco [™] Preloaded DNA/RNA Extraction Set						
Cat. No.: atc-pd/rna						
Number of reaction: 48 reactions						
Item	Specification	Quantity				
Preloaded 48-Well Extraction Plate	8 rxns/plate	6 plates				
Mixing Comb	N/A	6 pieces				
User manual	N/A	1 copy				

Note: Treat all reagents as potential irritants.

Note: Do not reuse the Plate & Comb.

Shipping and Storage

All preloaded plates should be stored and transported at room temperature ($16\sim30^{\circ}$ C), and store the reagent set in a cool and dry place. The expiration date of the reagent set is stated on the exterior package. Do not use the reagent set beyond the expiration date, as it could affect the accuracy of subsequent nucleic acid test result.

Materials and Equipment Required, but Not Provided

- taco[™] Nucleic Acid Automatic Extraction System: taco[™] 24 or taco[™] mini
- Disposable gloves
- Micro-centrifuge tubes
- Micropipette and filter tips (p1000, p200)
- Phosphate buffered saline (PBS)
- Laminar air-flow (optional)

Introduction

The $taco^{TM}$ Preloaded DNA/RNA Extraction Set is specially designed for $taco^{TM}$ 24 and $taco^{TM}$ mini Automatic Nucleic Acid Extraction System. Based on the magnetic separation technology, nucleic acids are captured by silica-coated magnetic beads after sample lysis. Washing Buffer is then applied to remove impurities, followed by Eluting Buffer to recover nucleic acids from magnetic beads. This reagent set can extract viral DNA and RNA from shrimp muscle for research use purpose only. Other sample types must be validated by users.

Intended Use

This reagent set can extract viral DNA and RNA from various sample types such as shrimp tissue. The **tacoTM** Preloaded DNA/RNA Extraction Set has to be used with the **tacoTM** Automatic Nucleic Acid Extraction System.

This product is intended to be used by professional users such as well-trained laboratory technicians who are familiar with molecular biology techniques.

Important Notes

- After receiving the product, check the components for any damage. Contact GeneReach or your local distributor if the components are damaged. Do not use damaged items, as that could affect the reagent performance.
- All plastic consumables are for one-time use only. Repeated usage may lead to cross-contamination.
- When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles.
- To minimize the risks from contacting potentially infectious materials, we recommend working under a laminar hood until the samples are lysed.
- This reagent set should only be used by trained personnel.
- Disposal of waste must be compliant with local laws.

Product Limitations

The system performance has been validated with virus-infected shrimp muscle for viral nucleic acid isolation. The user is responsible for validating the performance of the **taco[™]** Preloaded DNA/RNA Extraction Set for other particular uses.

The reagent set and plastic parts are not intended for any therapeutic or diagnostic purposes for animals or humans.

Nucleic Acid Extraction Procedure

Note: Do not reuse Plate & Comb.

Note: The following protocol is for fresh and frozen shrimp samples. For other sample types, please refer to <u>www.tacomag.com</u> for instructions.

Note: Perform extraction at room temperature.

- **a.** Slowly peel the cover film off Preloaded 48-Well Extraction Plate.
- **b.** Transfer **200** μ **l sample** to well #1 of the preloaded plate (see illustration below).



Note: For sample preparation please see Appendix I.

 c. Open the door of the instrument, insert Mixing Comb and Preloaded 48-Well Extraction Plate with sample (please refer to the user manual of taco[™] instrument).

- d. Close the door of the instrument and start extraction program (please refer to the user manual of taco[™] instrument).
- e. After the extraction program is finished, take out the 48-Well Extraction Plate and Mixing Comb.
- f. Transfer the nucleic acids from well #6 to new micro-centrifuge tubes (see "Purity Analysis and Quantification of Nucleic Acid", Appendix II, for basic rules of nucleic acid storage and analysis). For subsequent applications in the iiPCR POCKIT[™] platform, please refer to the user manual of each kit.
- g. It is strongly recommended to use freshly extracted nucleic acids for downstream applications, such as amplification. Keep the extracted nucleic acids at -80°C for long-term storage (See "Storage of Nucleic Acid", Appendix II).

Troubleshooting

Low DNA/RNA yield	
(a) Poor sample quality	Poor sample quality may influence final
	nucleic acid quality. Use fresh samples for
	extraction if possible. Avoid repeated
	freeze-thaw cycles of samples.
(b) Incorrect sample	Reagent set performance is affected by
volume	sample volume. Optimize sample volume
	when dealing with different sample types.
(c) Mixing Comb was not	Contact your local distributor or GeneReach
installed properly	Biotechnology Corporation for assistance.
(d) Inappropriate operation	Operating temperature affects the recovery
environment	rate of the reagent set. Ensure the ambient
	temperature is within the range of 16-30°C.
(e) Use non-recommended	The performance of taco™ Preloaded
extraction instrument	DNA/RNA Extraction Set in instruments
	not recommended is not guaranteed. We
	strongly recommend users to apply $taco^{TM}$
	Preloaded DNA/RNA Extraction Set only
	on taco™ 24 or taco™ mini system.

Comments and suggestions

Comments and suggestions

Poor DNA/RNA performance in downstream applications Note: A spectrophotometer is required for the following analysis.

(a)	Insufficient DNA/RNA	Quantify extracted	DNA	A/RNA	with	a
	is used in downstream	spectrophotometer	to	measur	e t	he
	application	absorbance at 260 nr	n (see	"Quanti	ficati	on
		of Nucleic Acid", Appendix II).				

(b) Excess DNA/RNA used Excess DNA/RNA can inhibit some enzymatic reactions. Quantify extracted application DNA/RNA with a spectrophotometer to measure the absorbance at 260 nm (see "Quantification of Nucleic Acid", Appendix II).

Low A₂₆₀/A₂₈₀ ratio

(a) Absorbance reading at
320 nm was not
subtracted from the
absorbance readings at
260 nm and 280 nm

To correct for the presence of magnetic bead residues in the eluent absorbance readings obtained at 320 nm should be taken and subtracted from the absorbance readings at 260 nm and 280 nm.

Appendix I—Sample Preparation

A. Shrimp Tissue: fresh or frozen shrimp muscle

- i. Grind the tissue (40 mg) with 450 μl PBS in a 1.5 ml micro-centrifuge tube with a disposable grinder.
- ii. Centrifuge at 12,000 x g for 5 minutes to spin down the debris.
- iii. Use 200 μl supernatant for extraction (see Nucleic Acid Extraction Procedure).

B. Shrimp Tissue: ethanol preserved shrimp muscle

- Grind the tissue (40 mg) with 500~1000 µl PBS in a 1.5 ml micro-centrifuge tube with an automatic homogenizer, such as taco[™] Prep Bead Beater.
- ii. Centrifuge at 12,000 x g for 5 minutes to spin down the debris.
- iii. Use 200 μl supernatant for extraction (see Nucleic Acid Extraction Procedure).

C. Swab sample (for cotton tips of diameter of ≤ 0.2 inches (0.5 cm))

- i. Use the cotton swab to collect swab samples from host.
- ii. Cut off the swab tip and place it into a 1.5 ml or 2 ml micro-centrifuge tube with 1 ml PBS or saline.

- iii. Mix for 10 seconds.
- iv. Spin for 1 minute in a centrifuge.
- v. Transfer 200 µl supernatant into a new micro-centrifuge tube (for subsequent nucleic acid extraction, follow the manufacturer's instructions of the extraction system.

D. Swab sample (for cotton tips of diameter of > 0.2 inches (0.5 cm))

- i. Use the cotton swab to collect swab samples from host.
- ii. Place it into a 1.5 ml or 2 ml micro-centrifuge tube with 1 ml PBS or saline.
- iii. Swirl the swab in PBS or saline for 30 seconds.
- iv. Discard swab and spin the tube for 1 minute in a centrifuge.
- v. Transfer 200 µl supernatant into a new micro-centrifuge tube (for subsequent nucleic acid extraction, follow the manufacturer's instructions of the extraction system.

Appendix II—Purity Analysis and Quantification of

Nucleic Acid

A. Storage of Nucleic Acid

Extracted nucleic acids should be stored at -80°C.

B. Quantification of Nucleic Acid

Note: A spectrophotometer is required for following check-up.

Concentration is determined by calculating the absorbance at 260 nm with a background correction at 320 nm, i.e., $(A_{260}-A_{320})$. A subtraction by A_{320} reading is to correct for signals from residual magnetic particles in the eluent. Residual magnetic particles may affect the A_{260} reading, but should not affect the performance of nucleic acids in most downstream applications.

Use distilled water or TE buffer as the blank to zero the spectrophotometer.

C. Purity Analysis of Nucleic Acid

Purity is determined by calculating the ratio of A_{260} to A_{280} with a background correction with A_{320} , i.e., $(A_{260}-A_{320}) / (A_{280}-A_{320})$. A subtraction with A_{320} reading is to correct for the presence of residual magnetic particles in the eluent solution. An A_{260} / A_{280} ratio of 1.6~2.0 is indicative of high nucleic acid purity.

© 2015 GeneReach Biotechnology Corporation. All rights reserved. For research use only. Not intended for animal or human therapeutic or diagnostic uses.