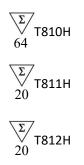
Viral DNA and RNA Extraction Kit (For Environmental Detection Use)

User Guide



Version 2.0

For use with Automatic nucleic acid extractor compatible with Viral DNA and RNA Extraction Kit (For Environmental Detection Use)



T810H T811H T812H



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Kit Version	2.0		
Changes	Address of Manufacturer Chapter "Content of the Kit " Chapter "Warnings and Precautions" Chapter "Materials Required but not Provided" Chapter "Limitations of Test Methods" Chapter "Safety Symbols and Signs" Small lexical corrections.	Additions	/

Intended Use

The **Viral DNA and RNA Extraction Kit** is designed to rapidly extract viral DNA and RNA from environmental samples, including environmental water samples, aerosol samples, solid surface wipe samples, stool samples and swab samples. The extracted viral DNA and RNA are of high purity and stability and can be used in a variety of routine operations, including enzyme digestion, Polymerase Chain Reaction, DNA library constructions, and Southern hybridization and blotting.

The *Viral DNA and RNA Extraction Kit* is intended to be used by professionals, such as biotechnologists, microbiologists, clinical technicians, and physicians who are trained in molecular and biological techniques.

Product Performance Indicators

The extraction kit can extract more than 200 copies/mL viral DNA nucleic acid, and more than 200 copies/mL viral RNA nucleic acid. Both the intra and inter-batch variations of the kit are less than or equal to 5%.

Special Notes

The *Viral DNA and RNA Extraction Kit* is worked with TIANLONG[®] automatic nucleic acid extractor (GeneRotex 96) that has been disinfected by UV light before use. After an experiment, wipe the inside of the extractor with 75% ethanol and disinfect it with UV light for 15 min. An automatic nucleic acid extractor automates the entire purification process and can process 1-96 samples in a single run.

The *Viral DNA and RNA Extraction Kit* is used to extract viral DNA and RNA targets. To avoid RNA degradation by RNase during operation, use exclusive-use utensils and sample injectors, and use disposable centrifuge tubes and tips preprocessed by autoclave before using. The operator should wear powder-free gloves, a mask and a protective coverall.

The kit has magnetic beads with a unique separation function and buffer system to extract, separate and purify high-quality nucleic acids from environmental water samples, aerosol samples, solid surface wipe samples, stool samples and swab samples.

Magnetic beads enable the purification of high-quality nucleic acids that are free of proteins, nuclease, and other impurities. Purified nucleic acids can be widely used in a variety of routine operations, including experiments such as enzyme digestion, Polymerase Chain Reaction, DNA library construction, and Southern hybridization and blotting.

Please carefully read the manual of instructions before attempting to install or use the product for the first time. To consider all possible consequences of incorrect operation or non-recommended functions, pay special attention to the possible consequences.

Testing Principle

The **Viral DNA and RNA Extraction Kit** is worked with TIANLONG[®] automatic nucleic acid extractor (GeneRotex 96). During the nucleic acid extraction process, magnetic beads are adsorbed, transferred and released by special magnetic rods based on the principle of magnetic bead adsorption. The extraction process enables the conduction of nucleic acid extraction and the final adsorption of highly pure nucleic acids with the transfer of magnetic beads and nucleic acids.

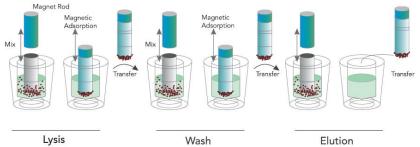


Figure 1. Schematic Diagram of Automatic Nucleic Acid Extractor

An automatic nucleic acid extractor performs the following steps on sample containing magnetic particles:

A magnetic rod protected by the mixing sleeve inserts into a well which contains samples. The mixing sleeve stirs rapidly and repeatedly in the liquid to ensure complete mixing of the liquid and magnetic beads. After cell lysis, nucleic acid adsorption, washing and elution, highly pure nucleic acid is obtained.

GeneRotex 96 is equipped with an array of 96n magnetic rods which allow it to process up to 96n samples simultaneously.

Content of the Kit

Short Code Name of Component		Т810Н	T811H	Т812Н
	Size	64 T/Box	20 T/Box	20 T/Box
	Component	Pre-filled	Pre-filled	Pre-filled
REAG1		96-deep well plate	96-deep well plate	6 strip tube
	Quantity	4	4	20
	Specification	16 Tests	5 Tests	1 Test
REAG2	Specification	1.28 mL	0.4 mL	0.4 mL
	Quantity	1	1	1
Instructions for Use		1 Сору	1 Сору	1 Сору

Materials Required but Not Provided

When working in a laboratory, make sure to wear a proper lab coat, disposable powder-free gloves and protective goggles. For more information, please consult the appropriate Safety Data Sheets (SDSs) which are available from the product supplier.

- Pipettor: 20 μL, 1000 μL
- Tip: 20 μL, 1000 μL
- Vortex mixer
- High-speed centrifuge
- Sample holder
- 75% ethanol
- Single kit docking (matched with T812H (6 strip tube), can be purchased from Tianlong)
- Extractor

Warnings and Precautions

Please be sure to read the precautions before using the kit.

The Viral DNA and RNA Extraction Kit is particularly used to extract viral DNA and RNA targets. To avoid RNA degradation by RNase during operation, use exclusive-use utensils and sample injectors, and use Viral DNA and RNA Extraction Kit (T810H/T811H/T812H) – User Guide Page 2 of 9

disposable centrifuge tubes and tips preprocessed by autoclave before using. The operator (researcher or clinical expert) should wear powder-free gloves and a mask.

Please read the manual carefully before using the kit, and strictly follow the manual throughout operation. The clinical samples should be collected on a clean bench or in a bio-safety cabin.

Before using TIANLONG[®] automatic nucleic acid extractor (GeneRotex 96), it must be disinfected by UV light. After an experiment, wipe the inside of the extractor with 75% ethanol and disinfect it with UV light for 15 min.

Due to the possibility of residual magnetic beads in the eluate following extraction, every possible effort should be made to avoid suctioning of any magnetic beads during eluate absorption.

Do not mix reagents from different batches, use the kit within expiry date.

Dispose all used samples and reagent materials after experiments, clean and disinfect the experimental workbench thoroughly.

The Viral DNA and RNA Extraction Kit is intended for the field of public health and scientific research samples. When using the kit, always wear a proper lab coat, disposable gloves and protective goggles. For more information, please consult the appropriate Material Safety Data Sheets (MSDSs). These documents convenient are available online and compact PDF format in at а https://www.medtl.net/resources/download/catalogue-all/catalogue ,, where the operator can find, view and print the appropriate MSDSs.

A Caution: Do not add any bleach or acidic solution directly to the pre-filled reagent.

The pre-filled reagent contains guanidinium salts, which, when combined with bleach can form highly reactive compounds. If any of buffers are spilled, clean immediately with a suitable laboratory detergent and water. If the spilled liquid contains potentially infectious agents, clean the affected area with laboratory detergent and water first. Then clean with sodium hypochlorite at a concentration of 1% (v/v). The **Viral DNA and RNA Extraction Kit** comes with the following warnings and precautions.

Name of Component		Hazard pictograms (CLP)	Classification under CLP:	H- and P-statements
REAG 1	Lysis Buffer Washing Buffer A Washing Buffer B		Acute toxicity (oral), Category 4 Skin corrosion/irritation, Category 2 Serious eye damage/eye irritation, Category 2	 Hazard statements (CLP) H302: Harmful if swallowed. H315: Causes skin irritation. H319: Causes serious eye irritation. Precautionary statements (CLP) P264: Wash hands, forearms and face thoroughly after handling. P280: Wear protective gloves/protective clothing/eye protection/face protection. P321: Specific treatment (see supplemental first aid instruction on this label). P337+P313: If eye irritation persists: Get medical advice/attention. P501: Dispose of contents/container to hazardous or special waste collection point, in accordance with local, regional, national and/or international regulation.

	Magnetic Beads Dilution Buffer Washing Buffer C Elution Buffer	None	None	None
REAG 2	Proteinase K Solution	None	None	None

Please see MSDS for more details.

Precautions for Safe Handling

Do not dispose of the preparations or the packaging waste in drains leading to the sewage system or in the drainage system for waste not produced by industrial processing/analysis waste.

Any material in contact with reagents should be treated as a biological contaminant and treated in accordance with relevant local regulations.

Reagent Storage and Handling

The *Viral DNA and RNA Extraction Kit* should be stored at room temperature in a cool, dry and well-ventilated area. All components of the kit can be adequately stored for up to 12 months.

The kit should be used in a well-ventilated area, keep away from the source of heat, sparks, open flames, and smoking.

To avoid evaporation, the pre-filled reagent should be used immediately after opening and should not be placed for a long period of time.

Avoid exposure to UV light (e.g., for decontamination), which may result in accelerated aging.

Sample Handling and Storage

Avoid foam inside or on the samples. Depending on the starting material, sample pretreatment may be required. Samples should be stored at room temperature (10~25°C) before starting the experiment.

Samples should be used immediately after collection to extract nucleic acid or stored at 2~8°C for further experiment within 24 hours. For long-term storage, the samples should be placed at -20°C.

For detail information on sample pretreatment, please refer to 2.1.3.

Operation Guide

1. Automated Extraction Process

Automatic nucleic acid extractor (GeneRotex 96) enables nucleic acid extraction by magnetic beads. It uses magnetic rods to move the beads adsorbed with nucleic acid into different reagent wells. Magnetic rod protected by the mixing sleeve which stirs rapidly and repeatedly in the liquid to ensure complete mixing of the liquid and magnetic beads. After cell lysis, nucleic acid adsorption, washing, and elution, the highly pure nucleic acids are obtained. Automatic nucleic acid extractors are characterized by high automation, rapid extraction speed, stable results, and ease of operation.

The user needs to load samples and magnetic bead nucleic acid extraction reagents into the reaction consumables, the nucleic acid extractors are going to perform all nucleic acid extraction operations according to the experimental procedures. Please refer to the user manual provided with the respective instruments for operating instruction and start-up of tests.

2. Operation Steps of Automated Extraction

2.1 Automatic Nucleic Acid Extractor (model: GeneRotex 96)

2.1.1 Edit Experiment Program

The extraction procedure of GeneRotex 96 Automic Nucleic Acid Extractor is as follows:

Step	Name	Well	Stir (min:s)	Magnetic (min:s)	Wait (min:s)	Speed (rpm)	Volume (µL)	T Control (°C)
1	Remove Bead	2	00:10	00:10	00:00	1600	625	0
2	Lysis	1	05:00	00:45	00:00	2000	850	120
3	Washing 1	3	01:00	00:00	00:00	2000	650	90
4	Washing 1	3	01:00	01:00	00:00	3000	650	90
5	Washing 2	4	00:30	00:00	00:00	2000	750	90
6	Washing 2	4	00:30	00:30	00:00	3000	750	90
7	Washing 3	5	00:30	00:00	00:00	2000	850	120
8	Washing 3	5	00:30	00:30	01:00	3000	850	120
9	Elution	6	05:00	00:35	00:00	2500	80	120

2.1.2 Reagent Preparation

96-deep well plate:

Open the kit and take out the REAG1 from the plastic package, slowly invert it several times to resuspend the magnetic beads. Gently shake the 96-well plate so that the reagent and magnetic beads are concentrated on the bottom of the 96-well plate (A 96-well plate horizontal centrifuge can also be used for centrifugation at 500 rpm for 1 min). Carefully tear off the aluminum foil sealing film before use to avoid liquid splashing.

6 strip tube:

Open the kit and take out the REAG1 from the plastic package, slowly invert it several times to resuspend the magnetic beads. Gently shake the 6 strip tube so that the reagent and magnetic beads are concentrated on the bottom of the tube. Put the reagent on the docking (Note the direction and make sure that the tube is placed at the lowest level), carefully tear off the aluminum foil sealing film before use to avoid splashing, which is shown in Figure 2.



Figure 2. Put the 6 strip tube on the singe kit docking

2.1.3 Adding Samples to the Reagent

Sample pretreatment:

a. Environmental Water Sample

1) Method 1: 50 mL sewage sample is centrifuged at 2500 $\,\times\,g$ for 30 min at 4°C, then 400 $\,\mu\,L$ supernatant is aspirated for extraction.

Caution: This method is suitable for water sample with high viral content and does not require enrichment and concentration.

2) Method 2: Virus concentrate is obtained by polyethylene glycol precipitation. The concentrate is centrifuged at 12000 rpm for 3 min, then 400 μ L of supernatant is sucked for extraction.

b. Aerosol Sample

Samples are collected according to the specific requirements of different types of bioaerosol samplers. At the end of the collection, 400 μ L of the collection solution is aspirated for extraction.

c. Solid Surface Wipe Sample

One swab is thoroughly infiltrated with 3 mL of virus preservation solution and then repeated on a 10 cm square surface. The swab is placed back into the sampling tube for infiltration, and after being removed, the swab is smear-sampled again, repeated three times and then sealed back to the testing site. The swab samples are vortexed and mixed for 30 s before suction of 400 μ L of preservation solution for extraction.

Caution: Samples with obvious dirt on the surface of the object are vortexed and mixed, centrifuged at 12000 rpm for 3 min, and 400 μL of supernatant is sucked for extraction.

d. Stool Sample

0.05 g of fecal sample is weighed and placed in a 1.5 mL centrifuge tube, 1 mL of normal saline is added, thoroughly mixed, centrifuged at 12000 rpm for 3 min, then 400 μ L of supernatants is aspirated for extraction.

96-deep well plate: Add 20 μ L REAG2 and 400 μ L sample to column 1 or column 7 of the pre-filled reagent respectively. (Be aware of the column No. is for effective wells).

6 strip tube: Add 20 μ L REAG2 and 400 μ L sample to column 1 of the 6 strip tube respectively.

Caution: When pipetting the sample, avoid having substance other than liquid adhere to the tip of the sample injector; do not add the sample too quickly to avoid contaminating the upper portion of the well wall; and do not splash air bubbles to avoid contaminating adjacent wells.

(ID) Note: The following points should be taken into consideration when determining whether a sample is suitable for the *Viral DNA and RNA Extraction Kit*.

Type of sample: As stated in the intended use.

Short-term storage: Samples can be used immediately after collection for nucleic acid extraction or stored at 2~8°C for testing with a maximum storage period of 24 hours.

Long-term storage: If the user does not operate the sample temporarily, it should be kept sealed in a refrigerator at -20°C.

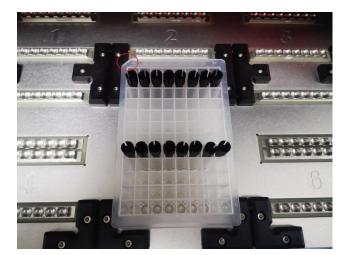
2.1.4 Loading in Deep Well Plate

Place the 96-deep well plate or 6 strip tube in the Automatic Nucleic Acid Extractor and ensure the marked notch of the plate faces front.

Insert the mixing sleeve into the mixing sleeve holder and close the cabin door.

• Note: As shown in Figure 3 and Figure 4, the user should ensure that the 96-deep well plate and the single kit docking is properly positioned with the notch facing outward.

• Note: Insert the 96-deep well plate or the single kit docking into the experiment cabin and push the magnetic rod covers into the right position. Check the position of the magnetic rod covers. Otherwise, instrument dysfunction or malfunction may occur and affect the experiment results.



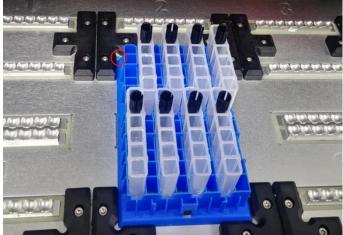


Figure 3. 96-deep well plate

Figure 4. Put the single kit docking into the instrument

2.1.5 Experimental Procedure Run

For special operations please see 2.1.1. After the procedure is completed, the instrument will notify the user of the experiment completion. Transfer the extracted product from column 6 and column 12 to a clean centrifuge tube which is free of nuclease.

•Note: If the user does not analyze the extracted product for the immediate use, please seal and store it in a refrigerator at -20°C.

Caution: Any used deep well plate and mixing sleeve should be considered as biological contaminants and disposed of in accordance with relevant regulations.

Caution: Using expired reagents or those that are not compatible with this instrument does not guarantee the expected results.

2.1.6 Cleaning and Maintenance of the Instrument

Follow the Cleaning and Maintenance of the Instrument section in accordance with the instructions in the user manual provided with the equipment. Ensure that the experimental cabin is cleaned regularly to minimize the risk of cross-contamination.

Troubleshooting Guide

This troubleshooting guide should assist you in resolving any problems that arise during the experimental process. For more information, please visit our Technical Support Centre and Frequently Asked Questions, page at <u>http://www.medtl.net</u>. The scientists in our Tianlong company's Technical Services Department are always available to answer any questions you may have about the information and protocols contained in the manual, as well as sample and assay technologies (Contact information is included on the back cover or at <u>http://www.medtl.net</u>).

When an exception or error occurs during the experiment, the current run step is terminated/stopped. After resolving the error or exception, restart the run from the beginning. The troubleshooting guide is shown in the following table.

No.	Fault Symptom	Fault Cause	Handling Method
1	The well plate vibrates and the liquid splashes when tearing off the aluminium foil sealing film.	When tearing the film, please press the well plate to prevent it from rocking.	The reagent for this plate shall be scrapped, and re-extraction shall be performed.
2	Add the sample to unexpected wells.	Please read this manual carefully before adding samples.	The reagent for this plate shall be scrapped, and re-extraction shall be performed.

3	The amount of liquid in the reagent wells is insufficient	/	Contact the after-sales service of Tianlong.
4	Reuse of pre-filled components	Please read the precautions in this manual before using the kit.	Perform re-extraction of nucleic acid.
F	Abnormal noise from the	The 96-deep well plate may be placed correctly.	Conduct reposition of the deep well plate.
5 instrument during extraction	The mixing sleeve may not be inserted in place.	Reinsert the mixing sleeve.	
		Please follow the operation requirements in the manual.	Contact the Sales Support team of Tianlong.
6	Poor extraction performance	The temperature control components of the instrument may be abnormal.	Contact the Sales Support team of Tianlong.
		Others	Contact the Sales Support team of Tianlong.

* Ensure that the reagents have been preserved and used according to the manufacturer's instructions.

Quality Control

In accordance with Tianlong Company's ISO-certified Quality Management, each lot of the *Viral DNA and RNA Extraction Kit* has been tested against predetermined specifications to ensure consistent product quality.

Limitations of Test Methods

The system performance has been established through performance evaluation studies using environmental water samples, aerosol samples, aerosol samples, solid surface wipe samples, stool samples and swab samples to purify viral DNA and RNA.

It is the user's responsibility to validate system performance for any procedures used in their laboratory that are not covered by the performance evaluation studies of Xi'an Tianlong Science and Technology Co., Ltd.

Although the kit is intended for use in public health and scientific research, the purity and quality of extraction results are also affected by the testing instruments and personnel. Moreover, the kit uses a specially formulated eluent that can affect the absorbance value, so it is not recommended to use a UV-spectrophotometer to measure the extraction effect directly.

The extraction kit is intended for use with clinical diagnostic samples, forensic materials, and scientific research samples. The instrument and operator have an effect on the concentration and purity of the extracted product. Any generated diagnostic results must be interpreted in conjunction with the other clinical or laboratory findings.

Safety Symbols and Signs

No.	Symbol	Implication
1	REF	Catalogue number
2	LOT	Batch code

3	∑ <n></n>	Contains sufficient for <n> tests</n>
4	Σ	Use by date
5	\land	Caution
6	X	Temperature limit
7	(!)	Reminder
8		Manufacturer
9	\otimes	Do not re-use
10	CONT	Content of the kit
11	REAG1	Pre-filled 96-deep well plate/6 strip tube
12	REAG2	Proteinase K Solution
13		Warning
14	PAP PAP	PAP21: Not-corrugated cardboard

Contact Information

For technical assistance and more information, please contact our Technical Support Center at +86-2982682132(Tel), +86-29-82216680 (Fax), inquiry@medtl.com or contact your local distributor.

For up-to-date licensing information or product-specific disclaimers, please see the respective User Guide. Tianlong User Guide is available at <u>www.medtl.net</u> or can be requested from Tianlong Technical Services or the local distributor.

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