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Instruction Manual

viRNAtrap™ Extraction Kit

Catalogue Number: 3315-100, 3315-500

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DESCRIPTION AND CONTENTS

Intended Use

The viRNAtrap™ Extraction Kit enables isolation of total RNA from biological material (i.e. nasopharyngeal, oropharyngeal or buccal smears) in sampling/transport medium.

Principle of the Extraction Kit

Total RNA is isolated from sampling and transport medium with the use of magnetic beads in lysis buffer. The RNA isolation can be performed either manually or by automated methods. The isolated RNA is also suitable for use in other analytical methods, e.g. RNA sequencing, RT-qPCR, transcription profiling, hybridization etc.

Parameters

The lysis buffer inhibits the activity of RNases and inactivates viruses and other infectious agents. The binding capacity of the magnetic beads is up to 10 µl of RNA when using 300 µl of viRNAtrap™ Lysis Buffer. The volume of eluted sample is approximately 50 µl.

Contents of the Kit

Component	Catalogue Number	
	3315-100	3315-500
viRNAtrap™ Lysis Buffer	30 ml	150 ml
Wash Buffer 1	12 ml	60 ml
Wash Buffer 2	3 ml	15 ml
Elution Buffer	15 ml	75 ml
Manual_CZ_093_viRNAtrap Extraction	1 pc	1 pc
Manual_EN_093_viRNAtrap Extraction	1 pc	1 pc

Additional Reagents and Equipment

- 100% isopropanol (85 ml per 100 of isolations, 425 ml per 500 isolations)
- microtubes for lysis and elution steps (recommended volume at least 1.5 ml)
- adjustable automated pipettes (10–1000 µl)
- disposable pipette filter tips
- magnetic stand
- personal protective equipment (lab wear, gloves, goggles)
- vortex
- centrifuge suitable for 1.5 ml microtubes


WARNINGS AND MEASURES

Storage and Shelflife

- All reagents contained in the kit are stable until expiry date (see the label on the box) when stored at room temperature.
- Always mix the viRNAtrap™ Lysis Buffer with magnetic beads properly prior the use.

Safety Precautions

- Prior the use read the enclosed instruction manual carefully. Always use the update version of the instruction manual. Make sure that you have understood all the given information.
- WARNING: The viRNAtrap™ Lysis Buffer contains dangerous substance. This solution MUST NOT come into direct contact with bleach.
- Keep the following safety handling instructions - **H and P phrases** - that apply to the viRNAtrap™ solution:

Component	GHS symbol	H phrases	P phrases
viRNAtrap™ Lysis Buffer	 Warning	302+332, 315, 319	P280, P301 + P330, P304 + P340, P302 + P352, P305 + P351 + P338, P337 + P313, P312

H phrases

- H302 + H332: Harmful if swallowed or inhaled.
- H315: Causes skin irritation.
- H319: Causes serious eye irritation.

P phrases

- P280: Wear protective gloves/protective clothing/eye protection/face protection.
- P301 + P330: IF SWALLOWED: Rinse mouth.
- P304 + P340: IF INHALED: Remove person to fresh air and keep comfortable per breathing.
- P302 + P352: IF ON SKIN: Wash with plenty of water.
- P305 + P351 + P338: IF IN EYES: Rinse with water cautiously for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
- P337 + P313: If eye irritation persists: Get medical advice/attention.
- P312: Call a POISON CENTER/doctor if you feel unwell.





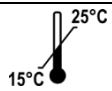


Technical Recommendations

- Keep the expiry date stated on the label of the kit. Do not use the reagents after the expiry date.
- Do not use damaged components. Keep the damaged component safe for a complaint and contact the manufacturer immediately.
- Use only calibrated pipettes and instruments.

Contamination Precautions

- Always wear a clean lab coat and use protective gloves.
- Change gloves immediately on suspicion of contamination.
- Use only automated pipettes and filter tips.
- Decontaminate lab surfaces, instruments and equipment regularly.
- Negative control is recommended in each isolation procedure.

Symbols on Labels

	LOT number
	Expiry date
	Manufacturer
	Contents
	Store at recommended temperature
	Catalogue number
	In vitro diagnostics medical device

WORKING PROCEDURE

Preparation of Working Solutions for the Kit of 100 rxn (3315-100)

Prior working with the extraction kit it is necessary to prepare the following reagents:

- **Wash Buffer 1:** Prior the first use, add 18 ml of pure isopropanol into Wash Buffer 1 (concentrate). Mix thoroughly and note adding isopropanol on the vial label.
- **Wash Buffer 2:** Prior the first use, add 27 ml of pure isopropanol into Wash Buffer 2 (concentrate). Mix thoroughly and note adding isopropanol on the vial label.

Preparation of Working Solutions for the Kit of 500 rxn (3315-500)

Prior working with the extraction kit it is necessary to prepare the following reagents:

- **Wash Buffer 1:** Prior the first use, add 90 ml of pure isopropanol into Wash Buffer 1 (concentrate). Mix thoroughly and note adding isopropanol on the vial label.
- **Wash Buffer 2:** Prior the first use, add 135 ml of pure isopropanol into Wash Buffer 2 (concentrate). Mix thoroughly and note adding isopropanol on the vial label.

Solutions are stable at least 1 year at room temperature.

MANUAL ISOLATION PROCEDURE

1. Pipette 100 µl of transport medium with sample into 1.5 ml microtube.
2. Add 300 µl of viRNAtrap™ Lysis Buffer.
Caution: Always shake the viRNAtrap™ Lysis Buffer thoroughly.
3. Add 400 µl of pure isopropanol and mix thoroughly by pipetting or vortex.
Caution: Add internal control in the volume according to the PCR kit instructions into each tube.
4. Incubate 3 minutes at room temperature.
5. Place the microtube with sample on the magnetic stand and wait approx. 2 min until the solution clears up.
6. Pipette off the supernatant.
7. Leave the microtube with sample on the magnetic stand and add 250 µl of Wash Buffer 1.
8. Let it take effect for 20 sec and pipette Wash Buffer 1 off.
9. Add 250 µl of Wash Buffer 2, let it take effect for 20 sec and pipette it off.
10. Centrifuge the microtube with sample shortly, place it back on the magnetic stand and pipette off the supernatant.
11. Let the beads dry approx. 2 min.
12. Take the microtube with sample from the magnetic stand, add 50 µl of Elution Buffer and mix thoroughly.
13. Incubate for 2 min, then place the microtube with sample back on the magnetic stand and wait for the solution to clear up (approx. 2 min).
14. Pipette the supernatant containing RNA into a new microtube.

The eluted RNA can be used immediately or stored at -70 °C.

ISOLATION OF VIRAL RNA USING THE KINGFISHER FLEX SYSTEM

Preparation of 96-well plates for isolation:

1. Prepare a Sample Plate. Pipette 300 µl of viRNAtrap lysis solution with magnetic beads, 100 µl of sample, 400 µl of pure isopropanol. and internal control according to the PCR kit instructions into each well.

Caution: Add internal control in the volume according to the PCR kit instructions into each well. Samples can be placed in a maximum of 94 wells, the remaining two wells are necessary for the positive and negative controls.

2. Prepare Wash Plate 1. Add 250 µl of Wash Buffer 1 into each well.
3. Prepare Wash Plate 2. Add 250 µl of Wash Buffer 2 into each well.
4. Prepare Elution Plate. Add 50 - 100 µl of Elution Buffer into each well.


Used plates:

- DeepWell 96-well plate - Sample Plate, Wash Plate 1, Wash Plate 2
- 96-well Elution Plate

The KingFisher Flex System Isolation Protocol is available at tech_support@genespector.com.

AUTOMATED ISOLATION PROCEDURE

The working procedure of isolation with the viRNAtrap™ Extraction Kit is compatible with various robotic pipetting stations. Contact the manufacturer for further information.

GENERI BIOTECH s.r.o.
Machkova 587/42
 CZ-500 11, Hradec Králové 11 – Třebeš
CZECH REPUBLIC

www.generi-biotech.com
tel.: +420 495 056 314
e-mail: info@generi-biotech.com

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