## **PureBind Viral Nucleic Acid Isolation Kit**

For isolation and purification of viral DNA/RNA from plasma or serum samples.

Version	210201
Catalog	K1622-SMP
	K1622-100



7964 Arjons Dr., Ste G San Diego, CA 92128 Tel.: (858) 689-8808 Website: www.oceannanotech.com Email: <u>info@oceannanotech.com</u>

PureBind Viral Nucleic Acid Isolation Kit

## **Table of Contents**

Product description
Contents and storage
Required materials not supplied4
Principle4
Procedural guidelines5
Before you begin
Prepare working buffers5
Prepare proteinase K solution5
Protocol: Isolate viral DNA/RNA from plasma or serum samples6
Lyse sample and bind PureBind Beads to genomic DNA
Wash PureBind Beads with wash buffers6
Elute the cfDNA7
Document History
Warranties and Disclaimers12

## **Product description**

The PureBind Virus Nucleic Acid Isolation Kit is designed to isolate viral DNA and RNA from enveloped viruses (e.g. HBV, HCV, and HIV) in plasma and serum. The magnetic technology enables efficient, consistent, and scalable extraction of virus nucleic acid from plasma or serum. The obtained nucleic acid is of high yield and purity and is suitable for direct downstream applications such as PCR, qPCR, and NGS. Furthermore, the magnetic particles feature uniformed size and slow sedimentation rate, which makes it excellent choice for high throughput sample preparation.

### **Contents and storage**

Reagents have a one (1) year expiration date from date of purchase when stored properly.

PureBind Blood DNA Isolation Kit Catalog no.	4 preps <sup>[3]</sup> K1622-SMP	100 preps <sup>[3]</sup> K1622-100	Storage
PureBind Beads <sup>[1]</sup>	100 ul	3 ml	2-8°C
Carrier RNA [1]	500 ul	500 ul	15-30°C
Lysis/Binding Buffer <sup>[1]</sup>	2 ml	40 ml	15-30°C
Wash Buffer 1 <sup>[1, 2]</sup> Customer adds Isopropanol	6 ml	2 x 60 ml	15-30°C
Wash Buffer 2 <sup>[1, 2]</sup> Customer adds Ethanol	3 ml	2 x 30 ml	15-30°C
Elution Buffer <sup>[1]</sup>	500 ul	5 ml	15-30°C

**Note**: Some reagents in each kit are provided in excess of the amount required.

<sup>[1]</sup> PureBind Beads are also available as Catalog no. A1602 and Lysis/Binding Buffer is available as Catalog no. R1247 and Wash Buffer 1 is available as Catalog no. R2340 and Wash Buffer 2 is available as R2355, and Elution Buffer is available as R0510.

<sup>[2]</sup> Prior to first time use, add the required amount of alcohol as indicated on the label of each bottle, then check the box and mix well.

<sup>[3]</sup> The number of preps or isolations are based on a 200 ul plasma or serum sample.

## **Required materials not supplied**

Item	Source	Catalog no.
Ethanol, absolute (100%)	Fisher Scientific	BP291914
Isopropanol (100%)	Sigma Aldrich	I9516
Proteinase K, lyophilized	Gold Biotech	P-480-100
Micro- and conical tubes (1-15 ml)	Various	Not applicable
Magnetic separator for micro-tubes	Ocean NanoTech	MMS
Vortex mixer	Various	Not applicable
Heat block for tubes	Various	Not applicable
Rotator (End-Over-End Mixer)	Thermo Fisher	HulaMixer
Pipettors and tips	Various	Not applicable
Buccal cell collection swabs	Puritan	25-3406-H

## Principle

The magnetic nanoparticle technology and buffers in this kit are used in a simple separation protocol to isolate virus nucleic acid from human serum or plasma. Lysis Buffer, Proteinase K, and Carrier RNA is first added to the sample containing virus particles, followed by incubation with beads and isopropanol. The beads bind to virus nucleic acid in the sample and is easily separated using a magnet. The magnet separation facilitates simple washing and eluting of the isolated virus nucleic acid.



**Figure**. Illustration showing magnetic separation of viral nucleic acid from a plasma sample using the PureBind Viral Nucleic Acid Isolation Kit.

## **Procedural guidelines**

- Read the USER GUIDE and make sure all the directions are followed and all recommended volumes are used as indicated.
- Prior to use, bring all kit components to room temperature.
- All vortex steps should be performed at maximum speed to ensure mixing.
- Do not add proteinase K directly to the Lysis/Binding Buffer.
- PureBind Beads should be resuspended to a homogenous suspension prior to use.

## **Before you begin**

#### **Control and Prevention Measures**

- Clean and disinfect laboratory working areas and equipment with a nuclease decontamination solution.
- Wear a laboratory coat and gloves when handling all components related to this kit. A laboratory coat and gloves will protect you from the reagents and kit components, as well as nucleic acid from nucleases that are present on human skin.
- Use DNase-free pipette tips to handle all components of the kit and avoid putting "used" tips into the reagent bottles.

#### Prepare working buffers

- Prior to first time use, add isopropanol (100%) as indicated on the label of each bottle(s) of Wash Buffer 1 to obtain a working solution. Check the box on the label and mix well by inverting 10 times.
- Prior to first time use, add ethanol (100%) as indicated on the label of each bottle(s) of Wash Buffer 2 to obtain a working solution. Check the box on the label and mix well by inverting 10 times.

#### Prepare proteinase K solution

Prepare enough proteinase K solution at 20 mg/ml (not provided) by dissolving lyophilized proteinase K powder in 50 mM Tris (pH 8.0). Then divide the stock solution into small aliquots and store at -20°C. Alternatively, a 20 mg/ml proteinase K solution can be purchased from Gold Bio (Catalog no. P-480-SL2) or a different supplier.

# **Protocol: Isolate viral DNA/RNA from plasma or serum samples**

The following protocol is for viral DNA/RNA isolation from 200 ul plasma or serum sample. For questions regarding other input ranges contact customer support at (858) 689-8808 or <u>info@oceannanotech.com</u>.

#### Lyse sample and bind PureBind Beads to genomic DNA

- 1. Add 200 ul plasma or serum sample to a micro tube.
- Add 25 ul Proteinase K (20 mg/ml) (not provided in kit) and 5 ul Carrier RNA. Vortex for 10 seconds to mix. Then incubate for 2 minutes at room temperature.
- Add 400 ul Lysis/Binding Buffer. Vortex for 10 seconds to mix. Then incubate on a rotator (end-over-end) for 10 minutes at room temperature.
- Add 10 ul PureBind Beads and 400 ul Isopropanol. Vortex for 10 seconds to mix. Then place the mixture in a rotator (end-over-end) for 10 minutes to bind DNA to the beads, followed by a final vortex for 10 seconds.
- Briefly centrifuge to collect sample to the bottom of the tube. Place the tube on a magnetic separator for 2 minutes or until beads are pelleted against the magnet. Then carefully discard the supernatant without disturbing the beads.

#### Wash PureBind Beads with wash buffers

- 6. Remove the tube from magnetic separator. Add 1 mL of Wash Buffer 1 and vortex for 10 seconds.
- Place the tube on a magnetic separator for 2 minutes or until beads are pelleted against the magnet. Then carefully discard the supernatant without disturbing the beads.
- 8. Repeat Steps 6-7 for a second wash with the Wash Buffer 1.
- 9. Remove the tube from magnetic separator, add 1 mL of Wash Buffer 2 and vortex for 10 seconds.
- 10. Place the tube on a magnetic separator for 2 minutes or until beads are pelleted against the magnet. Then carefully discard the supernatant without disturbing the beads.
- 11. Repeat Steps 9-10 for a second wash with Wash Buffer 2.

PureBind Viral Nucleic Acid Isolation Kit

12. Keep the tube on the magnetic separator with the lid open. Air dry the beads for 5 minutes. Remove any visible supernatant without disturbing the beads.

Caution. Do not over dry beads as it may result in loss of cfDNA.

#### Elute the cfDNA

- 13. Remove the tube from magnetic separator and add 50 ul of Elution Buffer. Completely resuspend beads by pipetting 10 times. Then incubate for 2 minutes at room temperature.
- 14. Place the tube on the magnetic separator for 2 minutes or until the solution clears and beads are pelleted.
- 15. Carefully transfer the supernatant containing the purified genomic DNA without disturbing the pellet to a clean, labeled microcentrifuge tube.

Note. The purified DNA is ready for immediate use. Alternatively, you can store the DNA at 4°C for up to 24 hours or -20°C for long-term storage.

## **Document History**

Version	Person	Description of Change
150725	Luis Moreno	Initial draft
180718	Luis Moreno	Consultation draft – to Alice Bu
190422	Luis Moreno	Final version – approved by Luis Moreno

This page is intentionally left blank.

This page is intentionally left blank.

This page is intentionally left blank.

## **Warranties and Disclaimers**

The Ocean NanoTech products ("Product") is warranted to operate or perform in conformance with published Product specifications at the time of sale, as set forth in the Product documentation, specifications and/or accompanying package inserts ("Documentation") and to be free from defects in material and workmanship. Unless otherwise expressly authorized in writing, Products are supplied for research use only. No claim of suitability for use in applications regulated by FDA is made. The warranty provided herein is valid only when used by professionally trained individuals. Unless otherwise stated in the Documentation, this warranty is limited to one year from date of shipment when the Product is subjected to normal, proper, and intended usage. This warranty does not extend to anyone other than the original purchaser of the Product ("Buyer").

No other warranties, expressed or implied, are granted, including without limitation, implied warranties of merchantability, fitness for any purpose, or non-infringement. Buyer's exclusive remedy for non-conforming Products during the warranty period is limited to replacement of or refund for the non-conforming Product(s).

There is no obligation to replace Products as the result of accident, disaster or event of force majeure, misuse, fault or negligence of or by Buyer, or use of the Products in a manner for which they were not designed, or improper storage and handling of the Products.

© 2018 Ocean NanoTech, LLC. All rights reserved. Unless otherwise indicated, all trademarks are property of Ocean Nano Tech, LLC. Printed in the USA.