

Product Application

Viral Nucleic Acid Isolation from Whole Blood

To isolate total nucleic acids from viruses in whole blood using ReliaPrep™ Blood gDNA Miniprep System.

Kit: ReliaPrep[™] Blood gDNA Miniprep System

Analyses: qPCR or RT-qPCR

Sample Type(s): Whole blood

Input: 200μl of Sample

Materials Required:

Heat Block capable of 56°C

Protocol:

1. Add 20µl proteinase K to 200µl of whole blood sample and vortex.

2. Add 200µl CLD to sample. Mix by vortexing for at least 10 seconds.

3. Incubate 56°C for 10 minutes.

4. Add 250µl of BBA buffer and vortex to mix.

5. Load lysate to binding column/collection tube assembly.

6. Centrifuge at max speed for 1 minute.

7. Add 500µl CWD and centrifuge at max speed for 3 minutes. Discard flow through.

8. Repeat step 7.

9. Place column in a clean Elution Tube. Add 50µl of Nuclease-Free Water to the column,

10. Centrifuge at max speed for 1 minute. Store eluted RNA at -20 or -70°C.

This protocol was developed by Promega Applications Scientists and is intended for research use only.

Users are responsible for determining suitability of the protocol for their application.

Further information can be found in Technical Manual #TM330, available at: www.promega.com/protocols or for further information, please contact techserv@promega.com



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Results: Zika Virus RNA Isolation from whole blood

Zika from Blood	
Rep.	Ave Ct
1	28.1
2	28.4
3	27.9
Ave	28.1

Cq values from RT-qPCR for Zika Virus RNA with nucleic acids isolated whole blood. Zika Virus obtained from Zeptometrix was used for testing (Zeptometrix, Cat. # NATZIKV-ERCM). Zika Virus standard was spiked into human whole blood at a ratio of 1:10 v/v. Viral recovery was tested by amplification with GoTaq® 1-step RT-qPCR System (A6020) using Zika virus specific primers. n=3.