

MPure Blood DNA Extraction Kit 200

For extraction and purification of DNA from mammalian whole blood or a suspension of mammalian blood cells using the MPure-12 Nucleic Acid Purification System.

Cat. No. 117022100

Size: 48 tests

Storage: 15-25 °C

Revision Date: 2022-08





INTRODUCTION

MPure Blood DNA Extraction Kit 200 is used with the MPure-12 Nucleic Acid Purification System for the extraction and purification of DNA from 100-400 μ L mammalian whole blood or from a suspension of mammalian blood cells. Nucleic acids extracted from MPure Blood DNA Extraction Kit 200 can be used in a number of downstream applications, including PCR, qPCR, Sequencing (NGS), Microarray, RFLP, and Southern Blot Analysis.

DESCRIPTION OF SYMBOLS USED

The following are graphical symbols used in or found on MP Biomedicals' products and packaging. They are explained in more detail in the European Standard BS EN ISO 15223-1:2012.







KIT COMPONENTS

Component Name	Quantity	
Reagent Cartridge	48 pcs (4 x 6 x 2)	
Reaction Chamber	48 pcs (4 x 6 x 2)	
Tip Holder	48 pcs (24 x 2)	
Filtered Tip	50 pcs (25 x 2)	
Piercing Pin	50 pcs (25 x 2)	
Sample Tube (2 mL)	50 pcs (25 x 2)	
Elution Tube (1.5 mL)	50 pcs (25 x 2)	
Instructions for Use	1 pc	

STORAGE

Store at room temperature (15–25 $^{\circ}$ C). Do not freeze the reagent cartridges. The kits are stable for 12 months under these conditions.

After extraction, store the purified nucleic acids at 4 °C (short-term) or aliquot and store at -70 °C (long-term) before performing downstream analysis. Repeated freeze-thawing is not recommended.

REAGENT CARTRIDGE CONTENT



Well 1: Proteinase K Solution (40 μL)

Well 2: Lysis Buffer 2 (1000 μL)

Well 3: Binding Buffer 1 (600 μ L)

Well 4: Magnetic Bead Solution (800 μL)

Well 5: Washing Buffer 1 (1000 μL)

Well 6: Washing Buffer 2 (1000 μ L)

Well 7: Washing Buffer 3 (1000 μ L)

Well 8: Elution Buffer 1 (1000 μ L)

Well 9: Elution Buffer 2 (1000 μL)

Well 10: Empty

STARTING MATERIAL

- If the sample volume is less than 100 μ L, add the appropriate volume of PBS.
- MPure Blood DNA Extraction Kit has been tested with fresh or frozen blood samples collected in tubes containing common anticoagulants, such as EDTA, heparin* and citrate.
 - *EDTA is recommended as an anticoagulation agent, since heparin has inhibitory effects on nucleic acid amplification.
- We recommend using fresh whole blood samples (within one week) for extraction. The total nucleic acid yield and quality will likely decrease beyond one week.
- MPure Cultured Cell DNA Extraction Kit is recommended when using concentrated buffy coat (purified and free of blood cells).
- If the whole blood sample is granulocyte-rich (white blood cell number is more than 2×10^5 cells/ μ L), dilute the blood sample or extract the DNA using the MPure Blood DNA Extraction Kit 1200.
- For short-term storage (up to 10 days), store tubes at 2–8 °C. However, for applications
 requiring maximum fragment size, such as Southern blotting, store purified DNA at 2–8 °C for
 up to 3 days. This will minimize DNA degradation.
- For long-term storage, store tubes at -70 °C.
- The extracted product contains total nucleic acid (DNA and RNA); RNA is not the major product (about 10%) and will degrade quickly. If an RNA-free product is needed, treat the eluted product with RNase.

Sample Type	Whole blood, Buffy coat, Leukocyte concentrate*
Target Nucleic Acid	Total DNA (Genomic DNA, mitochondrial and/or viral DNA)**
	100-400 μ L whole blood (WBC count less than 2 x 10 ⁴ cells/ μ L); 100-400 μ L *leukocyte concentrate (no more than 5 x 10 ⁶ cells); 100-400 μ L buffy coat**
Sample volume	NOTE: *For those samples which have low leukocyte count (less than 1×10^3 cells/ μ L), concentrate the blood cells by centrifugation at 3000 rpm for 15 min at 4 °C. We recommend measuring the leukocyte concentration.
	**If the WBCs count in the blood sample is more than 2 x 10 ⁴ cells/µL, we recommend using the MPure Blood DNA Extraction Kit 1200 or diluting the blood sample with PBS. (e.g. the whole blood sample from lymphoma/myeloma patient or other granulocyte-rich blood sample/buffy coat)
Controls/ Internal control	Add controls/internal control in the extraction procedure if required in the downstream analysis
Elution volume	50-300 μL

RESULTS

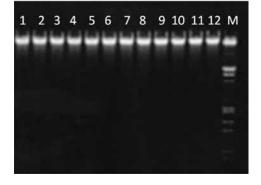
Expected Purity and Yield

DNA was purified using the MPure Blood DNA Extraction Kit 200 and the MPure-12 Nucleic Acid Purification System and five different blood samples. The DNA concentration was measured using a NanoDrop^T 2000 spectrophotometer. The range of DNA yield is 2–18 mg (from a sample with WBC counts of 2–20 x 10 3 cells/mL).

Sample Material	Volume/Amount	DNA Yield	Purity
Whole Blood (WBC # is 1.8 x 10³/mL)	100 μL 200 μL 300 μL 400 μL	1.0-1.2 μg 2.0-2.1 μg 2.8-3.1 μg 4.0-4.3 μg	
Whole Blood (WBC # is 4 x 10³/μL)	100 μL 200 μL 300 μL 400 μL	1.6-2.1 μg 3.8-3.9 μg 5.1-5.2 μg 8.5-8.8 μg	
Whole Blood (WBC # is 6.9 x 10³/μL)	100 μL 200 μL 300 μL 400 μL	2.9-3.1 µg 5.8-6.2 µg 8.2-8.8 µg 11.9-12.5 µg	$OD_{260}/OD_{280} \ge 1.7$ $OD_{260}/OD_{230} \ge 1.5$
Whole Blood (WBC # is 10.9 x 10³/μL)	100 μL 200 μL 300 μL 400 μL	4.3-4.6 μg 8.7-9.1 μg 11.5-12.2 μg 16.6-17.5 μg	
K562 cells	6 x 10 ⁵ cells 2 x 10 ⁶ cells	9.0-9.6 μg 22.0-25.0 μg	

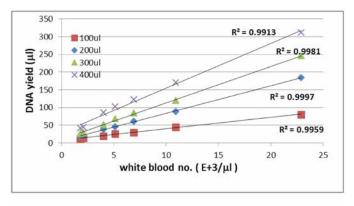
Integrity

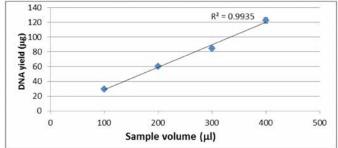
The DNA was isolated in replicates of 12 from 200 μ L human whole blood collected from two different donors. Elution volume was set to 100 μ L. Integrity of DNA was demonstrated by subjecting each eluted product to TAE agarose gel electrophoresis together with a suitable molecular weight marker. All samples show a single band with molecular weight at least 20 kb with no smear.



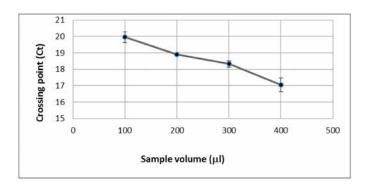
Scalability

DNA was isolated from different whole blood samples (WBCs count range $1.8-22 \times 10^3$ cells/ μ L). The DNA yield (measured by Nanodrop 2000 UV-Vis spectrophotometer) shows excellent scalability in different extraction volumes (100, 200, 300 and 400 μ L) and WBC counts of the samples.



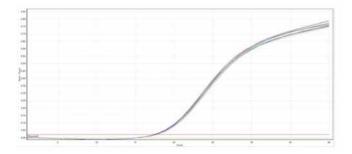


DNA isolated from different amounts of whole blood samples was amplified by real-time qPCR using β -globin gene specific primers. The resulting crossing points confirm the scalability of extraction.



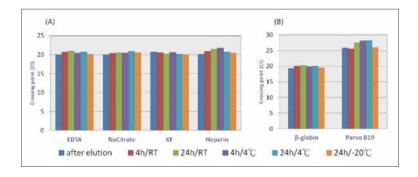
Reproducibility

DNA was isolated from twenty whole blood samples using the MPure Blood DNA Extraction Kit 200. The β -globin gene was detected by real-time qPCR. This data shows ultra-high stability and reproducibility of MPure-12 Automated Nucleic Acid Purification System.



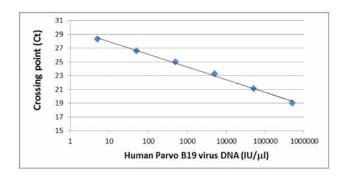
Stability

DNA was extracted from whole blood using the MPure Blood DNA Extraction Kit 200. Real-time qPCR detection of the β -globin (A) and spiked parvovirus B-19 DNA (A, B) was performed. No significant influences on samples with different anticoagulants and storage conditions were detected.



Sensitivity

Whole blood (in EDTA collection tube) spiked with serial-diluted human Parvo B19 Virus (in range of 25–2,500,000 IU/mL) was used for this analysis. 200 μ L sample was extracted and eluted in 100 μ L. 10 μ L of the eluate was used for real-time PCR using RealStar® Parvovirus B19 PCR kit 1.0. As little as 5 IU spiked (about 1 IU in PCR reaction) sample can be detected, proving the excellent sensitivity and linearity of the isolation procedure.



MPURE-12 PURIFICATION PROTOCOL

- 1 Turn on the MPure-12 instrument. Open the instrument door and remove the sample rack.
- 2 Load the Reagent Cartridge, then insert the Reaction Chamber, Tip Holder, Piercing Pins, and Filtered Tips into the instrument.
- 3 Insert the Sample tubes and Elution tubes into the Sample Rack.
- 4 Load the appropriate volume of sample into the sample tube, and place the sample rack back into the instrument.
- Scan the protocol barcodes provided on the following page for the purification protocol, and the sample and elution volumes when prompted. Double check the selected options on the display screen to verify the appropriate program settings.
- 6 Press [ENTER] to start the program. At the end of the run, instrument will alarm briefly.
- Open the instrument door and collect the elution tubes with the purified, ready-to-use nucleic acid. For short term, store at 4 °C. For long-term, store at -70 °C.
- 8 Discard all used cartridges and consumables into appropriate disposal.
- 9 Return the sample rack and close the door of the instrument.
- Place in sleep mode by holding the "Start" button for 2 seconds or switch off the power to the instrument.

BARCODES FOR PURIFICATION PROTOCOL. **SAMPLE VOLUME & ELUTE VOLUME SELECTION**

NOTE Follow the instruction guide shown on MPure system's LCD screen to scan the barcodes.

1. Select Protocol

MPure Blood DNA **Extraction Kit 200**



2. Select Sample Volume









Input sample volume by user via instrument control pad

3. Select Elute Volume







Input elute volume by user via instrument control pad

User

QUALITY CONTROL

In accordance with MP Biomedicals' ISO-certified Quality Management System, each lot of MPure Blood DNA Extraction Kit 200 is tested against predetermined specifications to ensure consistent product quality.

LIMITED EXPRESSED WARRANTY DISCLAIMER

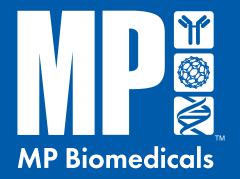
The manufacturer makes no expressed warranty other than that the test kit will function as a Research Use Only assay within the specifications and limitations described in the Instructions for Use and used in accordance with the instructions provided in the kit. The manufacturer disclaims any expressed or implied warranties with respect to merchantability, fitness for use or implied utility for any other purpose. The manufacturer's liability is limited.

TECHNICAL PROBLEMS

Should there be any technical problems, please do the following:

- Note the kit lot number and the expiry date.
- Retain the kits and the results that were obtained.
- Contact the nearest MP Biomedicals office or your local distributor.

NOTES



MP BIOMEDICALS

AMERICAS: 800.854.0530 | custserv.na@mpbio.com **EUROPE:** 00800.7777.9999 | custserv.eur@mpbio.com **APAC:** +65 6775.0008 | custserv.ap@mpbio.com

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