MGIEasy Magnetic Beads Blood Genomic DNA Extraction Kit User Manual

Manual Version：1.0　　Model: BDT-48，BDT-864

【Product Name】

MGIEasy Magnetic Beads Blood Genomic DNA Extraction Kit

【Package】

Cat. No.	Model	Specification
1000026451	BDT-48	48 preps
1000019634	BDT-864	864 preps

【Intended Use】

Used for nucleic acid extraction, enrichment, purification.

【Inspection principle】

In this product, the high salt lysate can release DNA from the sample. The released nucleic acid is captured by the superparamagnetic nano magnetic beads with high binding force. The impurities bound on the surface of nucleic acid are washed away by the washing effect of the washing solution. Finally, the nucleic acid on the magnetic beads is eluted to obtain high-quality genomic DNA. The extracted genomic DNA can be used in a variety of routine operations, including enzyme digestion, PCR, fluorescent quantitative PCR, library construction, microarray hybridization, high-throughput sequencing and so on.

【Kit Components】

Table 1　Main Components and specification

	Reagent	Package and amount	
		(48 **Preps**)	(864 **Preps**)
Box1	Buffer LYS	15 mL×1 bottle	260 mL×1 bottle
	Buffer WB1	14 mL×1 bottle	240 mL×1 bottle
	Buffer W3	32 mL×1 bottle	650 mL×1 bottle
	Buffer EB	9.6 mL×1 bottle	180 mL×1 bottle
	Proteinase K	1.5 mL×1 tube	18 mL×1 bottle

	Magnetic Beads H	1.5 mL×1 tube	18 mL×1 bottle

【Storage Conditions】

Different reagents in this kit have different storage conditions. Please store them respectively according to the following conditions:

Table 2 Reagents storage conditions and validity period

Reagent	Storage Conditions	Validity Period
Proteinase K	2°C to 8°C	18 months
Magnetic Beads H	2°C to 8°C	18 months
Others	0°C to 30°C	18 months

Note:

1. ***Proteinase K and magnetic beads can be transported at 0~30°C. For long-term storage, please store the kit at 2-8°C***
2. ***The Buffer LYS May have some precipitation which will not affect the function. If the precipitation occurs, please heat the reagent bottle in a 37°C water bath properly for around 10 min until the precipitation disappear, then mix thoroughly for use.***

【Applicable Automation Instrument】

Applicable automation instrument:

High-throughput automated sample preparation system, Model: MGISP-960;

Automated Nucleic Acid Extraction and Purification System, Model: MGISP-NE384;

ThermoFisher Sample Purification System: KingFisher Flex.

【Sample Conditions】

1. This kit is suitable to extract DNA from Blood, saliva and buffy coat.
2. The samples are recommended to be extracted within 24 h at 4°C after collection; If can't be extracted within 24 h, the samples should be stored at -70°C or below. Avoid repeated freezing and thawing; Frozen samples need to be thawed and mixed before use.
3. Sample transportation: use dry ice for transportation. Don't transport the samples exceeding 7 days. Avoid repeated freezing and thawing during transportation.
4. Sample Safety: All samples are regarded as potentially infectious items. The operations

shall be performed in accordance with relevant national standards.

【 Experimental Workflow 】

Please follow the workflow as below:

A. Required Materials Not Supplied

a) Required Materials for Manual Workflow:

Table 3 Required Materials for Manual Extraction

Type	Item Name	Note
Instrument	Table top centrifuge	Rotation speed not lower than 10,000 rpm/min
	Vortex	/
	Metal heater	Or instead by water bath
	1.5 mL tube magnets	/
	Pipette	1 mL、200 μL、20 μL
Reagent	Absolute ethanol	AR
Consumable	1.5 mL centrifuge tube	Nonstick, DNase-free, RNase-free
	Tips	1 mL、200 μL、20 μL

b) Required Materials for MGISP-960 Automatic Workflow:

Table 4 Required Materials for Automatic Extraction

Type	Name	Brand	item	48 Preps	864 Preps
Instrument	Vortexer	/	/	1	1
	Plate centrifuge	/	/	1	1
	Pipette	/	/	1 set	1 set
Reagent	Absolute ethanol	/	/	/	/
	Isopropanol	/	/	/	/
Consumable	Tips	/	/	/	/
	250 μL automated filter tips	MGI	1000000723	4	18×4
	1.3 mL U-bottom deep-well plate	MGI	1000004644	5	18×5
	Hard-shell thin-wall 96-well skirted PCR	MGI	1000012059	1	18×1

	plates, white shell/clear well				

Table 5 Required Materials for MGISP-NE384

Type	Name	Brand	item
Instrument	Vortexer	/	/
	Plate centrifuge	/	/
	Pipette	/	/
Reagent	Absolute ethanol	/	/
	Isopropanol	/	/
Consumable	Tips	/	/
	96-well tips comb	MGI	1000025661
	2.2 mL V-bottom deep-well plate	MGI	1000008088
	96-well PCR plate	/	/

Table 6 Required Materials for KingFisher Flex

Type	Name	Brand	item
Instrument	Vortexer	/	/
	Plate centrifuge	/	/
	Pipette	/	/
Reagent	Absolute ethanol	/	/
	Isopropanol	/	/
Consumable	Tips	/	/
	96-well tips comb	/	/
	96-well V-bottom deep-well plate	/	/

B. Read before use

1. This product is for scientific research purposes only and is not intended for clinical diagnosis.
2. Avoid repeatedly freezing and thawing samples, which may result in low DNA quality.
3. If Buffer LYS and Buffer WB1 has a precipitate, it can be re-dissolved in a 37 °C water bath. Shake and mix well before use.
4. All reagents and samples need to equilibrate to room temperature (10°C ~30°C) before use.
5. Before use, please make sure to add absolute (100%) ethanol into Buffer WB1 according to

the amount indicated on the reagent bottle label. And please prepare 75% ethanol labeled as Buffer W2. Isopropanol alcohol need to prepared by customer.

6. Please use the recommended consumables for automated or manual operations.

7. Please read the manual carefully before the experiment.

8. Buffer EB is divided into 10 mM Tris-HCl（pH8.0）and 0.5 mM EDTA（pH8.0），if there is a special need to provide their own elution buffer.

C. Manual Extraction Standard Workflow

1. Please prepare the mixture as following according to different sample types:

Table 7 The amount of reagent added according to different sample types

Sample types	Sample	Buffer LYS	Isopropanol
Buffy coat, Freeze blood without plasma	200 μL	300 μL	210 μL
Blood，Saliva，Freeze blood	300 μL	300 μL	350 μL
Saliva(with Salivary preservation)	300 μL	100 μL	250 μL

2. Add 20 μL Proteinase K, vortex once during this period.

3. Add Buffer LYS according to Table 7，vortex once during this period，Place the centrifuge tube on the Metal heater，Incubate at 65℃，1000 rpm，15 min.（***Buffy coat, Freeze blood without plasma*** and ***Frozen blood*** for more than three years should extended to 30 min）

4. Add Isopropanol according to Table 7，vortex once during this period.

5. Add 20 μL Magnetic Beads H，vortex once during this period, incubate at room temperature for 5 min，mix once or twice during the process.

6. Centrifuge instantaneously and place it on the magnetic stand for 2 min. After the liquid clears, carefully discard the supernatant liquid.

7. Remove the tube from the magnetic stand. Add 500 μL Buffer WB1 (ensure that absolute ethanol has been added), and mix thoroughly for 5-10 s, incubate at room temperature for 1 min.

Note：After adding Buffer WB1, please mix thoroughlyotherwise the purity of nucleic acid extracted will be affected.

8. Centrifuge instantaneously and place it on the magnetic stand for 1 min. After the liquid

clears, carefully discard the supernatant liquid.

9. Remove the tube from the magnetic stand. Add 600 μL Buffer W2（75% ethanol）, and mix thoroughly for 5-10 s, incubate at room temperature for 1 min.
10. Centrifuge instantaneously and place it on the magnetic stand for 1 min. After the liquid clears, carefully discard the supernatant liquid.
11. Remove the tube from the magnetic stand. Add 600 μL Buffer W2（75% ethanol）, and mix thoroughly for 5-10 s, incubate at room temperature for 1 min.(Repeat Step 9)
12. Centrifuge instantaneously and place it on the magnetic stand for 1 min. After the liquid clears, carefully discard the supernatant liquid. (Repeat Step 10)
13. Open the tube, and dry at room temperature for 5-10 min to ensure that the ethanol is completely evaporated.
14. Remove the tube from the magnetic stand. Add 60 μL -100μL Buffer EB, mix by vortex and place it on a metal heater. Incubate at 56°C, 1000 rpm for 5 min.
15. Centrifuge instantaneously and place the centrifuge tube on the magnetic stand. After the liquid is completely clear, carefully transfer 45 μL nucleic acid solution to a new 1.5 mL tube. Label and store at -20°C.

✔ **Stopping point: The extracted samples can be stored in the -20°C refrigerator.**

D. MGISP-960 Automated Extraction Standard Workflow

D1. MGISP-960 Automated Extraction Preparation

1. Instrument Setup

1) Before first use, install application scripts according to ***MGISP-100 & MGISP-960 Application Script Installation Instructions.***

2) Perform a pre-clean after powering on the device and before experiment according to ***MGISP-100 & MGISP-960 Cleaning Instructions***.

2. Preparing Consumables

Take out the consumables required for one workflow at room temperature for further use, as listed in the table 8:

Table 8 Material required but not provided

Consumables	Brand	Cat. No.	Quantity
250 μL automated filter tips	MGI	1000000723	8 Boxes
1.3 mL U-bottom deep-well plate	MGI	1000004644	7 Plates
Hard-shell thin-wall 96-well skirted PCR plates, white shell/clear well	MGI	1000012059	1 Plate

3. Preparing Samples

The script of MGISP-960 automation system is suitable for 96 sample.

According to the type of sample, the samples need to be prepared before running on MGISP-960. Take sample to a deep-well plate (MGI, 1000004644) according to table 9. And make sure that there are no air bubbles at the bottom and no hanging liquid on the side walls. Keep on ice for later use.

Table 9 The Sample added according to different sample types

sample types	volume（μL）
Blood	200
Saliva	300
Buffy coat、Freeze blood without plasma	165

4. Preparing Reagents

1) Preparation of Buffer WB1：Absolute ethanol needs to be added according to the label.

2) Preparation of Buffer W2： prepare 75% ethanol labeled as Buffer W2.

3) Isopropanol needed to prepared by customer

4) Take out 6 U-bottom deep-well plate（MGI，1000004644），label the plate and add the reagents according to the table 10.

Table 10 The reagent added according to different sample types

Reagent	Volume（μL）			Plate
	Blood	Saliva	Buffy coat,Freeze blood without plasma	
Proteinase K	20	20	20	Sample
Buffer LYS	200	100	320	Sample
Buffer WB1	400	400	400	W1
75% ethanol	800	800	800	W2
Buffer EB	110	80	160	EB
Magnetic Beads H	20	20	20	Beads
Isopropanol	300	300	220	Isopropanol

Note： Mix Magnetic Beads H and Proteinase K thoroughly before use.；Make sure there is no bubble at the bottom and no hanging fluid on the side wall.

D2. MGISP-960 Operation

1) Double-click the icon of MGISP-960 on the desktop. The mode selection interface is displayed, as shown in following figure 1. Select “Real” and click “Create”.

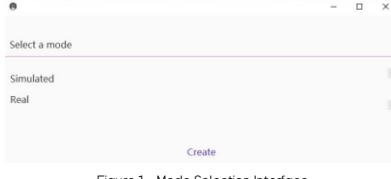

Figure 1 Mode Selection Interface

2) In the Authentication interface, click "User Entry" to enter the initialization interface.

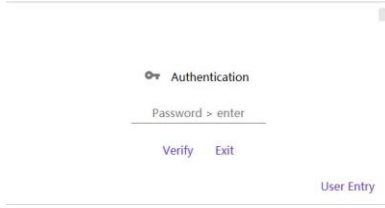

Figure 2 Mode Selection Interface

3) The initialization interface is displayed, as shown in following figure 3.

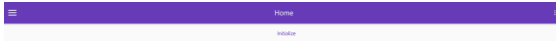

Figure 3 Initialization Interface

4) Click "Initialize". The initialization takes about 2 min. If Initialized successfully is displayed (as shown in following figure 4, the device is connected successfully, and you can go to the next step.

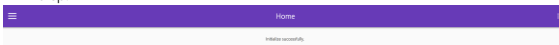

Figure 4 Initialization Successful Interface

Note: If the initialization fails, check whether the power switch is turned on, and whether more than one software program is running. Try to restart the software. If the problem persists, contact MGI technical support.

5) Click the menu button and select "Run Wizard" in the menu. In the Run Wizard interface, click "Solution", and select【JB-A09-027 MGIEasy Beads Blood Genomic DNA Extraction Kit】, click "Script", to select【Genomic DNA Extraction for Blood.py】or【Genomic DNA Extraction for Saliva.py】or【Genomic DNA Extraction for Buffy Coat.py】, operation deck arrangement of the first phase is displayed, as shown in following figure 6 and table 11. Follow the on-screen instructions to place the consumables, samples, and reagents, as shown in the figure 6. Confirm the placement and close the door.

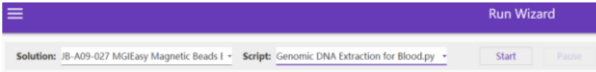

Figure 5 Run Wizard Interface

Figure 6 First Phase Operation Deck Arrangement

Table 11 First Phase Operation Deck Arrangement

Name	Position
250 μL automated filter tips	Pos1-Pos8
Hard-shell thin-wall 96-well skirted PCR plates, white shell/clear well	Pos12
Sample	Pos20
Buffer EB	Pos13
Buffer WB1	Pos14
Isopropanol	Pos16
75% Ethanol	Pos14
Beads	Pos18
Waste	Pos23

6) Click "Run" to start extraction workflow.

7) It is expected to run 1 h 15 min-2 h. After the process is finished, the product at Pos12 can be taken out.

8) Perform the next testing operation.

9) Dispose of the used deep-well plates, PCR plates, and waste bag to the designated waste area. Perform a post-clean before powering off the device according to MGISP-100 & MGISP-960 Cleaning Instructions.

✔ **Stopping point：The extracted samples can be stored in the -20 °C.**

E. MGISP-NE384 Automated Extraction Standard Workflow

E1. MGISP- NE384 Automated Extraction Preparation

1. 1. Preparing Device

Before first use, please confirm that the application script has been imported into the location of MGISP-NE384.

Before starting each round of experiment, please make sure that the machine has finished [clean].

2. Preparing Consumable

Take out the consumables required for one workflow for 384 samples, as listed in the table below:

Table 12 Materials required but not provided

Consumables	Brand	Cat. No.	Quantity
96 well tips comb	MGI	1000025661	4 pieces
2.2 mL V-bottom deep-well plate	MGI	1000008088	20 plates

3. Preparing Samples

1) The Automated Nucleic Acid Extractor can process 1-384 samples at one time.

2) Pretreat the sample to be extracted and place the samples on ice for later use.

4. Preparing Reagents

According to the number of samples, transfer the extraction reagents into new 2.2 mL V-bottom deep-well plate according to the Table 14：

Table13 The reagent added according to different sample types

Reagent	Volume（μL）			Adding Stage
	Blood	Saliva	Buffy coat, Freeze blood without plasma	
Proteinase K	20	20	20	Sample Preparation
Buffer LYS	300	100	330	Sample Preparation
Sample	200	300	165	Sample Preparation
Isopropanol	300	250	210	Pause After Lysis

Table 14 Input volume of each set of reagents

Item	Consumables	Volume/well
Sample	2.2 mL V-bottom deep-well plate	According to table13
75% Ethanol	2.2 mL V-bottom deep-well plate	500 μL
Buffer W3	2.2 mL V-bottom deep-well plate	500 μL
Buffer EB	2.2 mL V-bottom deep-well plate	60-150 μL
Buffer WB1	2.2 mL V-bottom deep-well plate	Buffer WB1: 500 μL Magnetic Beads H：20μL
Isopropanol	2.2 mL V-bottom deep-well plate	According to table13

E2. MGISP-NE384 Operation

Instrument Operation

1. Double-click the icon of MGISP-NE384 on the desktop. The authentication interface will be displayed. Select "User", enter password: "123456", click "login".
2. The initialization interface will be displayed.
3. Click "Initialize". The initialization will take approximate 1 minutes. If Initialized successfully displayed, means the device connected successfully, and you can go to the next step.

Note: If the initialization fails, check whether the power switch is turned on, and whether more than one software program is running. If yes, please restart the software. If the problem unsettled, please contact MGI technical support

4. Select the "Clean" option, emptying the console, wiping the console and tray with a dust-free paper soaked with 75% alcohol and closing the window. click "Start", and the instrument will open the fan filter unit and UV lamp to clean the internal environment of the instrument. The default cleaning time is 20 minutes. You can also modify the cleaning time accordingly.
5. After "Clean", return to the main interface select "Workflow".
6. In the Workflow interface, click "Script", select "MGIEasy Magnetic Beads Blood Genomic DNA Extraction Kit". Follow the on-screen instructions to place the consumables and reagents (Table15). Install the tips comb.

Table 15 Operation Deck layout

Reagents	Position
Buffer LYS+ Sample + Proteinase K	LaneA、LaneB、LaneC、LaneD：Pos1
Buffer WB1	LaneA、LaneB、LaneC、LaneD：Pos2
75% Ethanol	LaneA、LaneB、LaneC、LaneD：Pos3
Buffer W3	LaneA、LaneB、LaneC、LaneD：Pos4
Buffer EB	LaneA、LaneB、LaneC、LaneD：Pos6

7. Confirming the consumables and reagents are placed correctly, close the instrument window. Click "Run". Check the corresponding test channel according to the number of samples and check the tips comb are placed correctly. Click the "Confirm".
8. The whole run will take approximate 80 minutes, please arrange the following work properly.
9. After the run ended, please take out the extraction product of pos6 immediately. It can be used directly for subsequent experiments or stored at -20℃.
10. Dispose the used deep-well plates and tips comb. Select the "Clean" option, emptying the console, wiping the console and tray with a dust-free paper soaked with 75% alcohol and closing the window. click "Start", and the instrument will open the fan filter unit and UV lamp to clean the internal environment of the instrument. The default cleaning time is 20 minutes. You can also modify the cleaning time as needed.

Note: After the experiment, please take out the extracted product immediately. It is forbidden to leave the product at pos6 for a long time, otherwise it will affect the quality of the product.

F. KingFisher Flex Operation

F1. KingFisher Flex Automated Extraction Preparation

1） Refer to table 17, add buffer WB1 (make sure absolute ethanol has been added)、Buffer W2（75% ethanol）、Buffer W3 and Buffer EB to the 96-well plates.

2） Vortex the Magnetic Beads H and add 20 μL Magnetic Beads H to Buffer WB1 plate，then put the tips comb into the buffer WB1 plate.

3） Add 20 μL Proteinase K to sample plate，then add Sample and Buffer LYS according to table 16 and table 17.

Table 16 The reagent added according to different sample types

Reagent	Volume（μL）			Adding Stage
	Blood	Saliva	Buffy coat, Freeze blood without plasma	
Proteinase K	20	20	20	Sample Preparation
Buffer LYS	300	100	330	Sample Preparation
Sample	200	300	165	Sample Preparation
Isopropanol	300	250	210	Pause After Lysis

Table 17 Input volume of each set of reagents

Item	Consumables	Volume/well
Sample	deep-well plate	According to table 16
75% Ethanol	deep-well plate	500 μL
Buffer W3	deep-well plate	500 μL
Buffer EB	deep-well plate	60-150 μL
Buffer WB1	deep-well plate	Buffer WB1: 500 μL Magnetic Beads H：20μL
Isopropanol	deep-well plate	According to table 16

F2. KingFisher Flex Operation

1. Run 【Kingfisher Bindit】and import【MGIEasy Blood Genomic DNA 864 KF】 program.
2. Put the prepared Sample plate, Buffer WB1 plate, Buffer W2 (75%Ethanol) plate, Buffer W3 plate and Buffer EB plate into the corresponding slot of the instrument.

3. Start the program，it will pause after 30 minutes，add 210-350μL isopropanol (refer to table 16) into sample plate，put the sample plate back and continue the program.
4. After about 60 minutes, the program ended.
5. After the run ended, please take out the extraction product of "Buffer EB plate" immediately. It can be used directly for subsequent experiments or stored at -20℃.

【Precautions】

1. This product is only used for scientific research, not for clinical diagnosis, please read this instruction carefully before use;
2. Please familiarize the operation and precautions of various instruments to be used before testing;
3. When all the reagents are taken out from the specified storage environment, please use them according to the requirements. The reagents should be shaken and mixed before use;
4. The micro- Pipette should be used for sample addition;
5. All samples and reagents should be avoided to directly contact with skin and eyes; do not swallow, once happen, immediately rinse with plenty of water and go to the hospital for treatment in time;
6. All samples and various wastes should be treated in accordance with relevant regulations.

【Production Company Information】

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