IVD

WizMag™ Cell/Tissue DNA

User Manual

Ver 2.0

REF W7040 | W7041 | W7042 | W7043

For *in vitro* diagnostic use

wizbiosolutions
THE INNOVATION OF GENOMICS

INTENDED USE

The WizMag™ Cell/Tissue DNA kit is designed to be used on the CLEO™ AP16 Nucleic Acid Extractor System for simple and easy purification of DNA from various biological sources including animal cultured cells, bacterial cells and animal tissues. Purified DNA is free of enzyme inhibitors and other contaminants and highly suited for downstream applications such as PCR-based or enzyme-based reactions.

KIT CONTENTS

Contents	W7040	W7041	W7042	W7043	Storage
No. of preparation	64	192	32	96	Room Temperature (15-25°C)
Pre-packed 96-well Plate	4 ea	12 ea	-	-	
Pre-packed 6-well Strip	-	-	32 ea	96 ea	
Plunger	8 ea	24 ea	8 ea	24 ea	
Buffer TDL	20 mL	55 mL	10 mL	30 mL	
Buffer PKR	1.5 mL	6 mL	1 mL	3 mL	
Proteinase K *	14 mg x 2	42 mg x 2	14 mg	42 mg	
RNase A solution	270 µL	780 µL	140 µL	400 µL	
Blank solution A	500 µL	500 µL	500 µL	500 µL	

This kit is delivered under ambient conditions. When being used immediately on arrival, all the components can be stored at room temperature (15 - 25 °C). But if the kit is going to be stocked for a long time, Proteinase K should be stored at 2 - 8°C for optimal conservation. Long exposure to heat sources can deteriorate the performance of the kit significantly.

* After dissolving, store proteinase K solution at 2 - 8°C for optimal conservations.

QUALITY CONTROL ANALYSIS

In accordance with Wizbiosolutions Inc. ISO 13485-certified Quality Management System, each lot WizMag™ Cell/Tissue DNA kit is tested against predetermined specifications to ensure consistent product quality.

RECONSTITUTION OF PROTEINASE K

Before the first experiment, dissolve completely Proteinase K with Buffer PKR, as indicated on the product label. Do not vortex when dissolving. Store the reconstituted Proteinase K solution at 2 - 8°C.

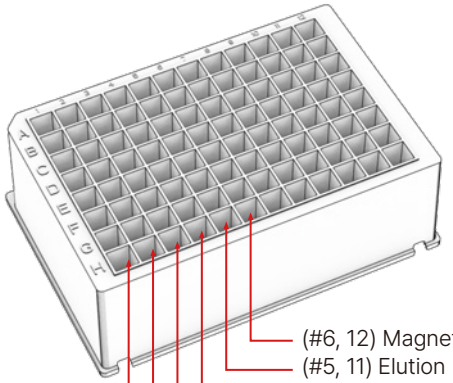
PRECAUTIONS

- This product is reserved exclusively for ***in vitro*** diagnostic purposes.
- Intended for single use only. Do not reuse.
- Check the expiration date on the box. Do not use it after the expiration date.
- Wear protective clothing, and use disposable gloves, goggles, and a mask.
- Do not eat, drink or smoke in areas where samples or test reagents are being used. Once you finish the test wash your hands.

- Specimens must be treated as potentially infectious as well as all reagents and materials that have been exposed to the samples and handled in the same manner as an infectious agent.
- Regular decontamination of commonly used equipment is recommended, especially micropipettes and work surfaces.
- This product contains irritants that are harmful when in contact with skin or eyes, or inhaled or swallowed. Care should be taken when handling this product.
- Some of the reagents in the 96-well Plate contain chaotropic which can form highly reactive compounds when combined with bleach. Do NOT add bleach or acidic solutions directly to the sample-preparation waste.
- Any significant incidents related to the product should be notified to the competent authorities and manufacturers.
- Do not use it if the package is damaged.

COMPOSITION OF THE PRE-PACKED 96-WELL PLATE (W7040 | W7041)

A total of 16 samples can be simultaneously processed per plate.



Columns 7 - 12 in the right half of a 96-well plate have the same composition as columns 1 - 6 in the left half.

COMPOSITION OF THE PRE-PACKED 6-WELL STRIP (W7042 | W7043)

(#1) Sample
(#2,3,4) Washing
(#5) Elution
(#6) Magnetic bead

PROTOCOL

A. Setup of program (For CLEO™ AP16 & AP48 devices, preset program can be used.)

Edit and run the experiment program as follows:

No.	1	2	3	4	5	6	7
Well #	6	1	2	3	4	5	6
Step	Beads	Lysis	Wash	Wash	Wash	Elute	Discard
Wait time	-	-	-	-	-	03:00	-
Mix time	00:20	10:00	02:00	02:00	02:00	05:00	00:20
Collect time	00:25	00:30	00:30	00:30	00:30	00:45	-
Volume(µL)	200	800	750	750	750	100	200
Mixing speed	Medium	Fast	Fast	Fast	Fast	Bottom	Medium
Collect speed	Strong	Strong	Strong	Strong	Strong	Strong	Normal
Lysis temp.		70°C					
Elute temp.						65°C	

B. Sample Preparation

1. **Cultured cells**
   a. Harvest the cells by centrifugation as below conditions :

Sample	Capacity(max.)	Centrifuge (xg)	Time (min.)
Bacterial cells	1 x 10^9	10,000 - 13,000	1 - 2
Animal cells or mycoplasma-infected cells	2 x 10^6	200 - 1,000	5 - 10

* Cultured mycoplasma cells can be harvested by centrifugation at 15,000 xg for 30 minutes.

   b. Resuspend thoroughly the cell pellet in 200 µL of Buffer TDL.
   c. (Optional) If RNA should be removed, add 4 µL of RNase A solution to the sample tube, vortex to mix and incubate for 2 minutes at room temperature.
   d. Add 20 µL of Proteinase K solution into the tube and mix briefly by vortexing.
   e. Incubate the tube at 56°C for 10 minutes.
      - For efficient lysis, vortex occasionally during incubation. The use of a specialized instrument such as a thermo-mixer will accelerate the lysis.
   f. Use the mixture as a sample.

2. **Animal tissues**
   a. Place up to 20 mg of finely ground tissue or minced tissue into a 1.5 mL tube.
      - A mortar and pestle is a good conventional method for grinding, but other methods like a bead-beating instrument or a rotor-stator homogenizer can be good alternatives.
      - Generally, the finer ground sample may lead to less lysis time and better results.
   b. Apply 200 µL of Buffer TDL and 20 µL of Proteinase K solution (20 mg/mL) into the tube and mix completely by vortexing or pipetting.
      - It is essential for good results to mix the components thoroughly to make a homogenate.
   c. Incubate the tube at 56°C for 10 minutes - 3 hrs until the tissue has been lysed clearly.
      - For efficient lysis, vortex occasionally during incubation. The use of a specialized instrument such as a thermo-mixer will accelerate the lysis.
      - Lysis time can vary depending on the type and amount of starting sample. Samples can be further incubated for complete lysis and longer incubation will not affect the recovery yield.
      - After complete lysis, the lysate will turn clear or translucent from turbid, but it may

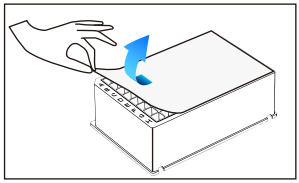
still have some remaining debris originating from the sample, such as bone or exoskeleton.

d. (Optional) If RNA should be removed, add 4 µL of RNase A solution to the sample tube, vortex to mix, and incubate for 2 minutes at room temperature.

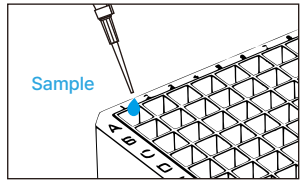
e. Use the mixture as a sample.

- If some debris remains in the lysate, spin down briefly to pellet the debris, and use 200 µL of the cleared supernatant as a sample.

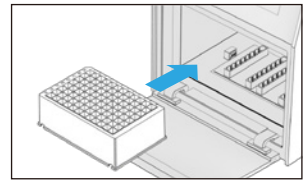
C-1. DNA extraction procedure (W7040, W7041)



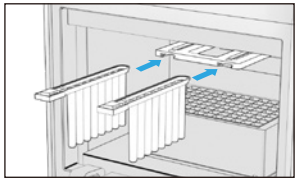
1. Carefully peel off the film of the 96-well Plate not to cross-contaminate.



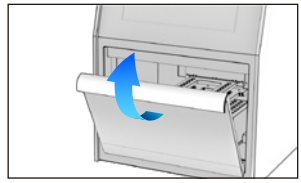
2. Apply all the sample mixture into the each first well (#1,7)



3. Mount the 96-well Plate on the CLEO™ AP16 carefully.

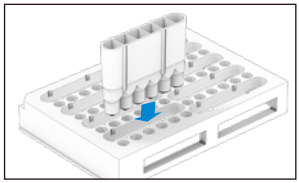


4. Insert a Plunger all the way into the socket above the 96-well Plate.

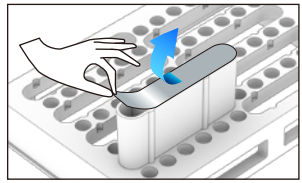


5. Close the front door of the instrument.

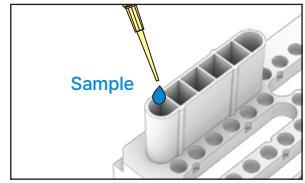
C-2. DNA extraction procedure (W7042, W7043)



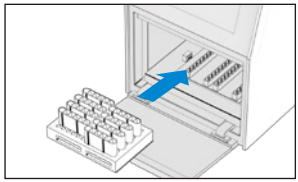
1. Mount the 6-well Strip onto the Strip Adapter Plate.



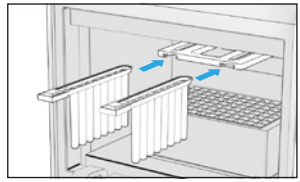
2. Carefully peel off the film of the 6-well Strip not to cross-contaminate.



3. Apply all the sample mixture into the each first well (#1)



4. Mount the 6-well Strip Adapter Plate on the CLEO™ AP16 carefully.



5. Insert a Plunger all the way into the socket above the 96-well Plate.

6. Close the front door of the instrument.
7. Select **MENU** ▶ **DNA** ▶ **Cell/Tissue DNA** on the screen.

8. Press **'RUN'** button on the screen.
9. After the alarm finishes, open the door and carefully remove the Plunger.
10. Detach the 96-well plate (or the Strip Adapter Plate) from the machine carefully.
11. Transfer the 60 - 80 µL eluate of each fifth well (#5,11) into a new 1.5 mL centrifuge tube.
    NOTE : The volume of eluate can be decreased slightly during the process.
12. Dispose of 96-well Plates (or 6-well Strip) and Plunger used in the test according to local or national waste disposal methods.

TROUBLE SHOOTING GUIDE

Problem	Possible causes	Recommendations
Low or no recovery	Low cells in the starting sample	Some samples may have a low population of cells. Use more samples. Otherwise, an addition of carrier RNA (5~10 ug/sample) into the sample lysate can be helpful for the recovery of DNA when the cellular mass in the sample is very low.
	Too many cells in the starting sample	The sample amount over the maximum capacity will lead to poor lysis of cells, resulting in significantly low recovery. Reduce the amount of starting sample.
Low purity	Too much sample amount used	Do not overload the sample. Keep the volume and the cell number of sample as procedure.
Degraded DNA	Starting sample is too old or improperly stored	Too old or improperly stored samples may have degraded DNA. Use a fresh sample.
Inconsisten recovery of DNA	Contamination between reagent wells	The reagent in the well may evaporate and form a deposit on the film during storage, which may cause contamina-tion between wells when the film is removed. Prepacked plate or tube always should be stored at proper condition. Before removing the film of the plate or the tube, it is recommended to shake off the deposit on the film with holding the plate or the tube tightly.

SYMBOL GLOSSARY

REF	Catalogue number		Manufacturer		Use-by date
LOT	Batch code		Do not re-use		Temperature limitation
IVD	in-vitro diagnostic use		Instructions for use		Keep away from sunlight
	Contents sufficient for <n> tests		Caution		Keep dry
	Do not use if package is damaged	UDI	Unique Device Identification		

ORDERING INFORMATION

Product	Cat No.	Package	Note
WizMag™ Cell/Tissue DNA	W7040	64 Prep	16 prep/run
	W7041	192 Prep	
	W7042	32 Prep	Single prep
	W7043	96 Prep	
CLEO™ AP16 Nucleic acid Extractor	CL2016	1 system	1-16 sample
CLEO™ AP48 Nucleic acid Extractor	CL2048	1 system	1-48 sample

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