& Magen

[Product Name] MagPure Stool DNA Kit

[Product specification] 400 Preps/Kit

[Intended Use]

This product is specially designed for high throughput DNA extraction from stool samples or other sample. DNA can be directly used for downstream applications such as PCR, Viral DNA testing, bacterial DNA testing, ect.

[Principle]

This product is based on the purification method of high binding magnetic particles. The sample is lysed and digested under the action of lysate and Protease. DNA/RNA is released into the lysate. After adding magnetic particles and binding solution, DNA/RNA will be adsorbed on the surface of magnetic particles, and impurities such as proteins will be removed without adsorption. The adsorbed particles were washed with washing solution to remove proteins and impurities, washed with ethanol to remove salts, and finally DNA/RNA was eluted by Nuclease Free Water.

[Main Composition]

Cat.No	D6364	Contents
2ml Bead Tubes	400	PP Column
MagPure Particles N	14 ml	Magnetic Particles
PVP-10	6 g	PVP-10000
Buffer PCI	300 ml	Phenol/Chloroform
RNase A	75 mg	Ribonuclease A
Proteinase K	180 mg	Proteinase K
Protease Dissolve Buffer	20 ml	Protease
Buffer ATL	300 ml	Tris/EDTA/SDS
Buffer MLE	180 ml	Tris/EDTA/Guanidine Salt
Buffer GW1*	132 ml	Guanidine Salt
Elution Buffer	60 ml	10mm Tris,pH8.4

[Storage conditions and validity]

This kit is shipped and stored at room temperature and is valid for 12 months.

[Applicable Instrument]

CE

Nucleic Acid Extraction Machine such as KingFisher Flex, Kingfisher Duo or similar. Automatic Nucleic acid workstation such as Tican, HAMILTON, Aurora, BGI or similar.

[Preparation before Use]

- Add 9ml ml Protease Dissolve Buffer to the Proteinase K, and store at -20~8 °C after dissolve
- Add 5m Protease Dissolve Buffer to the RNase A, and store at -20~8°C after dissolve
- Dilute Buffer GW1 with 138ml 100% ethanol and store at room temperature
- Dilute Buffer MLE with 120 ml Isopropanol and store at room temperature
- Magnetic Particles N should be shake violently for 1~2 minutes to be homogeneous.

[Protocol of stool DNA]

- 1. Transfer 100~150mg stool sample to 2ml Bead tube. For liquid samples, pipette ~0.15ml samples. Cut the end of pipet tip to make pipetting easier.
- 2. Add 0.6ml Buffer ATL/PV-10 and 0.6ml Buffer PCI into the sapmle. Place on a bead beater machine or vortex at maximum speed for 10 min.

Note: Add PVP-10 powder into Buffer ATL before use, dissolve completely by up side down.We recommend to use FastPrep-24, Tissue Lyser, ect. Processing time depends on sample input and bead beater.

- 3. Incubate sample at 65°C for 20 minutes. This step makes bacteria lyse more completely.
- 4. Centrifuge at 13,000 x g for 5 minutes and follow below purification process.
- 5. Transfer 400µl supernatant to a new 2ml centrifuge tube.
- 6. Add 10µl RNase A and mix thoroughly, sit at room temperature for 5~10 minutes.
- 7. Add 30µl MagPure Particles N, 20 µl Proteinase K and 600 µl Buffer MLE to the samples. Mix thoroughly by vortex for 10 minutes. Place the tube to the magnetic stand for ~2 minutes until the beads have formed a tight pellet. Then remove the supernatant.
- 8. Add 600µl Buffer GW1 and vortex for 15 seconds to re-suspend beads. Place the tube to the magnetic stand for 1 minute until the beads have form a tight pellet. Then remove the supernatant.
- Add 600µl 75% ethanol, and vortex for 15 seconds to re-suspend beads. Place the tube to the magnetic stand for 1 minute until the beads have form a tight pellet. Then remove the supernatant.

10. Repeat step 9.

- 11. Centrifuge shortly to collect liquid on the tube. Place the tube to the magnetic stand and remove all the liquid carefully. Dry on air for 10 minutes.
- Add 50~100µl Elution Buffer to the sample, re-suspend the beads by vortex. Incubate at 55°C for 10 minutes by shaking. If there is no shaking device, vortex 2~3 times to mix DNA with magnetic particles.
- Place the tube to the magnetic rack for 2 minutes. Transfer the supernatant containing the purified DNA to a clean 1.5ml centrifuge tube.

【Automatic protocol of KingFisher Flex】

- 1. Prepare the Sample superatant by follow Manual protocol step 1~5.
- 2. Add the Reagents/sample to the well of f the deep well plate according to the table below.

Name	Pre-loaded reagents		
	1: 10µl RNase A and 400µl Supernatant (Manual protocol step 5).		
Sample plate	2: Incubate at room temperature for 5-10min.		
	3: 20µl Proteinase K and 600µl Buffer MLE.		
Wash Plate 1	600µl Buffer GW1, Put in 96 magnetic Tip		
	30µl MagPure Particle MPN		
Wash Plate 2	h Plate 2 600µl 75% ethanol		
Wash Plate 3	600µl 75% ethanol		
Elution plate	100µl Elution Buffer		

- 3. Turn on the machine, start the corresponding program(D6364_F96_CE). Place the 96-well plate into the instrument as prompted.
- 4. Finish the operation after ~30minutes.
- 5. Remove the 96-well plate and magnetic jacket. Store the Elute product at -20~8°C.

[Product performance]

- 1. Appearance inspection: The kit should be completely composed, the appearance of the package should be clean, no leakage, and no damage; the signs and labels should be clear.
- Nucleic acid purity: Extract 5 mg liver homogenate (PBS, 200µl) according to the instructions. The OD260 / 280 value is 1.7-2.0, A260 / 230 value is 1.2-1.8.
- 3. Nucleic acid yield: Extract 5 mg liver homogenate (PBS, 200µl) according to the instructions, the

yield is 2ug.

4. Nucleic acid integrity: 1 mg liver homogenate was extracted according to the instructions. There was no obvious degradation of DNA during electrophoresis of the product.

5.

[Basic Information]

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[Explanation of Marks]

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IVD	The product is used in vitro, please don't swallow	2	Please don't reuse it
R	Validity	[]i	Please read the instruction book carefully before using
Δ	Warning, please refer to the instructions in the annex	***	Manufacturer
2°C 1 8°C	Temperature scope within which the product is reserve	LOT	Batch number
EC REP	European union authorization representativ		Keep dry
	Avoid overexposure to the sun		Don't use the product when the package is damaged
CE	The product meets the basic requirements of European in vitro diagnostic medical devices directive 98/79/EC		