【Product Name】 MagPure Tissue HW DNA Kit

【Product specifications】 48 Preps, 96 Preps, 480 Preps

【Intended Use】

This product provides a simple and fast solution for extracting high molecular weight DNA from complex animal tissues (aquatic or marine organisms, lower organisms, etc), cultivated cells, exfoliated cells and other biological samples. The obtained DNA can be directly used for down stream applications such as PCR, chip analysis, virus DNA detection, NGS, Nanopore sequencing etc.

【Main Composition】

Cat.No.	D638201	D638202	D638203
Purification Times	48 Preps	96 Preps	480 Preps
MagPure Particles G2	1.5 ml	3.0 ml	16 ml
Buffer ATL Minus	40 ml	90 ml	350 ml
Buffer SDS	1.8 ml	5 ml	20 ml
Buffer CXP	30 ml	50 ml	220 ml
RNase A	15 mg	30 mg	150 mg
Proteinase K	30 mg	60 mg	300 mg
Protease Dissolve Buffer	3 ml	5 ml	30 ml
Buffer MWX1	35 ml	70 ml	350 ml
Buffer GW1*	22 ml	44 ml	220 ml
Buffer GW2*	20 ml	50 ml	2 x 100 ml
Buffer BW3	50 ml	90 ml	450 ml
Elution Buffer	10 ml	30 ml	120 ml

【Storage conditions and Validity】

Proteinase K, RNase A and MagPure Particles G2 should be stored at 2–8°C upon arrival. However, short-term storage (up to 24 weeks) at room temperature (15–25°C) does not affect their performance. The remaining kit components can be stored at room temperature (15–25°C) and are stable for at least 18 months under these conditions.

【Prefilled Plate/Single Strip Component】

Components		D6382-TL-06	D6382-S-48
Proteinase K		60 mg	30 mg
RNase A		30 mg	15 mg
Buffer ATL Minus		90 ml	40 ml
Buffer SDS		5 ml	1.8 ml
Protease Dissolve Buffer		5 ml	3 ml
DA-Tip		12	24
V bottom plate/ Reagent strip	Row 1/7: 400µl Buffer CXP	6 plates	48 strips
	Row 2/8: 600µl Buffer MWX1		
	Row 3/9: 600µl Buffer GW1		
	Row 4/10: 600µl Buffer GW2 25µl MPG2		
	Row 5/11: 600µl Buffer BW3		
	Row 6/12: 100µl Elution Buffer		

【Preparation before Use】

- Add 0.6ml (48Preps), 1.2ml (96 Preps) or 6.0ml (480 Preps), Protease Dissolve Buffer to the Rnase A, and store at -20~8℃ after dissolve.
- Add1.5ml (48Preps), 3.0ml (96 Preps) or 15ml (480 Preps), Protease Dissolve Buffer to the Proteinase K, and store at -20~8℃ after dissolve
- Dilute Buffer GW1with 28ml (48 Preps), 56ml (96 Preps) or 280ml (480 Preps) 100% ethanol and store at room temperature
- Dilute Buffer GW2 with 80ml (48 Preps), 200ml (96 Preps) or 2 x 400ml (480 Preps) 100% ethanol and store at room temperature

【 Protocol Part 1: Small amount extraction of tissues and cells】

1. Sample preparation

- **Animal tissue:** Take 10~50mg tissue samples into a glass homogenizer, add 0.6ml Buffer ATL Minus, slowly squeeze up and down for 5 times. Transfer 0.5ml homogenized solution to a 1.5ml centrifuge tube, add 30μl Proteinase K Solution and 30μl Buffer SDS, mix by inverting for several times, incubate at 55℃ for 30-60 minutes or until the sample is completely digested. If there exist obvious undigested impurities, centrifuge at 10,000 x g for 3 minutes.
- **Cultivated cells (<1x10^7 cells):** Take an appropriate amount of culture medium, urine, amniotic fluid, ascites or other liquid samples into a centrifuge tube, centrifuge at 2,000 x g for 10 minutes to collect cells, and remove the supernatant. Add 500μl Buffer ATL Minus, vortex to resuspend cells, add 30μl Protease K Solution and 30μl Buffer SDS, mix by inverting for several times, incubate at 55℃ for 15~30 minutes.

2. **Transfer 500μl solution to a 2.0 ml centrifuge tube, add 10μl RNase Solution, mix well, and place at room temperature for 10~30 minutes.**

   Samples rich in RNA such as liver, kidney or cultivated cells are recommended to be placed at room temperature for 30 minutes.

3. **Add 400μl Buffer CXP, mix by inverting for 6~8 times, incubate at 50℃ for 3~5 minutes until the precipitation disappear.**

4. **Add 25μl MagPure Particles G2, mix by inverting for 10~15 times, place at room temperature for 5 minutes, during which invert and mix for several times.** Place at magnetic stand for 1 minute, and remove the supernatant. Vortex for an instant and aspirate the supernatant again.

5. **Add 600μl Buffer MWX1** and vortex for 10 seconds. Place the tube to the magnetic stand for 1 minute. Then remove the supernatant.

6. **Add 600μl Buffer GW1** and vortex for 10 seconds. Place the tube to the magnetic stand for 1 minute. Then remove the supernatant.

7. **Add 600μl Buffer GW2** and vortex for 10 seconds. Place the tube to the magnetic stand for 1 minute. Then remove the supernatant.

8. **Repeat Step 7 once**.

9. Do not remove the centrifuge tube from the magnetic stand, **add 750μl Buffer BW3 slowly,** do not disperse the magnetic beads, place for 60 seconds, and be careful to aspirate the supernatant.

10. **Add 100~150μl Elution Buffer,** gently tap to drop the magnetic beads from wall and resuspend in Elution Buffer. Incubate with shaking (600~800rpm) at 55℃ for 10 minutes. Place the tube to the magnetic stand for 2 minutes. Transfer the supernatant containing the purified DNA to a new centrifuge tube.

【 Part 2: Medium amount extraction】

1. Sample preparation

- **Animal tissue:** Take 50~120mg tissue samples into a glass homogenizer, add 1.2ml Buffer ATL Minus, slowly squeeze up and down for 5 times. Transfer 1.0ml homogenized solution to a 2.0ml centrifuge tube, add 60μl Proteinase K Solution and 60μl Buffer SDS, mix by inverting for several times, incubate at 55℃ for 30-60 minutes or until the sample is completely digested. If there exist obvious undigested impurities, centrifuge at 10,000 x g for 3 minutes.
- **Cultivated cells (<1x10^7 cells):** Take an appropriate amount of culture medium, urine, amniotic fluid, ascites or other liquid samples into a centrifuge tube, centrifuge at 2,000 x g for 10 minutes to collect cells, and remove the supernatant. Add 1,000μl Buffer ATL Minus, vortex to resuspend cells, add 60μl Protease K Solution and 60μl Buffer SDS, mix by inverting for several times, incubate at 55℃ for 15~30 minutes.

2. **Transfer 1,000μl solution to a 5.0 ml centrifuge tube, add 20μl RNase Solution, mix well, and place at room temperature for 10~30 minutes.**

   Samples rich in RNA such as liver, kidney or cultivated cells are recommended to be placed at room temperature for 30 minutes.

3. **Add 800μl Buffer CXP, mix by inverting for 6~8 times, incubate at 50℃ for 3~5 minutes until the precipitation disappear.**

4. **Add 50μl MagPure Particles G2, mix by inverting for 10~15 times, place at room temperature for 5 minutes, during which invert and mix for several times.** Place at magnetic stand for 1~2 minutes, and remove the supernatant. Vortex for an instant and aspirate the supernatant again.

5. **Add 1,200μl Buffer MWX1** and vortex for 10 seconds. Place the tube to the magnetic stand for 1 minute. Then remove the supernatant.

6. **Add 1,200μl Buffer GW1** and vortex for 10 seconds. Place the tube to the magnetic stand for 1 minute. Then remove the supernatant.

7. **Add 1,200μl Buffer GW2** and vortex for 10 seconds. Place the tube to the magnetic stand for 1 minute. Then remove the supernatant.

8. **Add 1,200μl Buffer GW2** and vortex for 10 seconds. Place the tube to the magnetic stand for 1 minute. Then remove the supernatant.

9. Do not remove the centrifuge tube from the magnetic stand, add 1,200μl Buffer BW3 slowly, do not disperse the magnetic beads, place for 60 seconds, and be careful to aspirate the supernatant.
10. Add 100~300μl Elution Buffer, gently tap to drop the magnetic beads from wall and resuspend in Elution Buffer. Incubate with shaking (600~800rpm) at 55℃ for 10 minutes. Place the tube to the magnetic stand for 2 minutes. Transfer the supernatant containing the purified DNA to a new centrifuge tube.

【 Part 3: Auto Purify by 32/48 channel nucleic acid extractor 】

1. Bottled reagents: add the reagents to the 96 well plate following the above table of prefilled kit contents.

   Prefilled reagents: invert the 96 well plate to suspend the magnetic beads completely. Pat the plate to make reagents fall back to the bottom of plate. Stay the plate at table for 1 minute, remove the sealing pack and sealing film.
2. Add 500μl of mixture (from Part 1 step 2) to each well of row 1/7.
3. Insert the magnetic tip (DA-Tip) and 96-well plate in to the machine (hole A1 is placed at the left inner corner). Turn on the machine and start the program.
4. About 30 minutes, extraction finish.
5. Take out the 96 well plate and magnetic tip comb.

Transfer DNA into a 1.5ml centrifuge tube and store at -20~8℃.

【 Program recommendation for Magen MagMix 32/48 extractor 】

No.	Name	Well	Volume	Mix		Wait		Magnet			Magnet	Heat	
				Time	Speed	Time	Position	Up/Down	Surface	Bottom		Plate	Temp
1	Mix	1	900	120s	6	0	0	0	0	0	Auto	1	50
2	Wash	2	750	10s	7	0	0	0	0	0	Auto	1	50
3	Magnet	4	750	20s	7	0	0	90s	0	0	Auto	1	50
4	Bind	1	900	250s	7	0	0	120s	0	0	Auto	/	/
5	Wash1	2	750	90s	7	0	0	90s	0	0	Auto	/	/
6	Wash2	3	750	90s	7	0	0	90s	0	0	Auto	/	/
7	Wash3	4	750	60s	8	0	0	60s	0	0	Auto	/	/
8	Wash4	5	750	0	8	0	0	60s	0	0	Auto	/	/
9	Elute1	6	100	180	7	0	0	0	0	0	Auto	6	55
10	Elute2	6	100	300s	6	0	0	90s	0	40	Auto	6	55
11	Remove	3	500	30s	8	0	0	0	0	0	Auto	/	/