

DNA/RNA Co-Isolation Technology

Introduction

The purification techniques of DNA and RNA mainly include phenol chloroform extraction, ion exchange, salting out, glass milk method, and silica gel column method. However, these methods often only extract one type of nucleic acid and waste the other. When the sample is limited, we often need to extract both DNA and RNA from the same sample separately. Karlinsey J (1989) first used phenol extraction and LiCl method to simultaneously extract DNA and RNA from eukaryotic cells, but this method was cumbersome to operate and required the use of ultracentrifuge, which was not widely used; Chomczynski (1993) improved the one-step RNA extraction method using the Trizol reagent, which can purify DNA, RNA, and protein separately using a single solution extraction. However, the DNA obtained using this method has low purity and is extremely difficult to dissolve (need to be dissolved in 8mM NaOH solution), and the DNA fragments are severely fragmented, only about 10Kb, which is difficult to meet downstream experimental requirements.

Magen's Co-isolation series products use silica gel column purification technology, which can quickly and efficiently separate DNA, total RNA, and proteins from a biological sample simultaneously. HiPure DNA/RNA Kits are suitable for simultaneously extracting DNA and RNA from tissue, cultured cell and plant samples. During the entire process, there is no need to use phenol chloroform extraction or time-consuming ethanol or isopropanol precipitation. The DNA and RNA extraction of several samples can be completed in just 30 minutes. Purified DNA and RNA can be directly used for various downstream applications.

This product series includes:

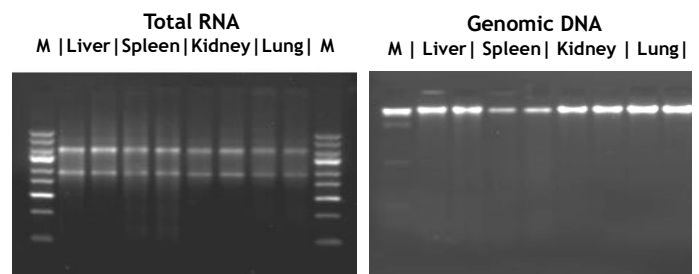
Name	Sample type
HiPure DNA/RNA Kit	soft tissue, cell, plant
HiPure Fibrous DNA/RNA Kit	hard-to-lyse sample (skin, muscle, tissue, cell, plant)
HiPure FFPE DNA/RNA Kit	Paraffin embedded tissue
HiPure DNA/RNA/Protein Kit	soft tissue, cell, plant

Compared to the performance of Trizol:

	Trizol Reagent	HiPure Kits
RNA extraction time	1 hour	20 minutes
DNA extraction time	1 hour	15 minutes
	dissolve overnight	immediately use
Protein	40 minutes	30 minutes
Time spend	3 hours	50 minutes
Toxicity	Phenol chloroform extraction, highly toxic	Safe and non-toxic
RNA purity	High, A260/280>1.9	High, A260/280>1.9
DNA purity	Low, A260/280<1.6	High, A260/280=1.8
DNA integrity	Poor, <10kb	Good, 20-60kb
DNA yield	Low	Medium

1. Extract DNA/ RNA from conventional samples simultaneously

Take 10mg conventional tissue samples (chicken liver/kidney/spleen/lung) and extract them with HiPure DNA/RNA Kit. After extraction, take 1µg total RNA and analyze by 1.0% agarose gel electrophoresis. Take 2% genomic DNA, analyze by 0.8% agarose gel electrophoresis. From the electrophoresis chart, it can be seen that the RNA obtained using the reagent kit does not degrade, and the obtained DNA fragments are intact without tailing phenomenon. The Lambda DNA/Hind III Marker indicates that the obtained DNA fragments are above 23KB.

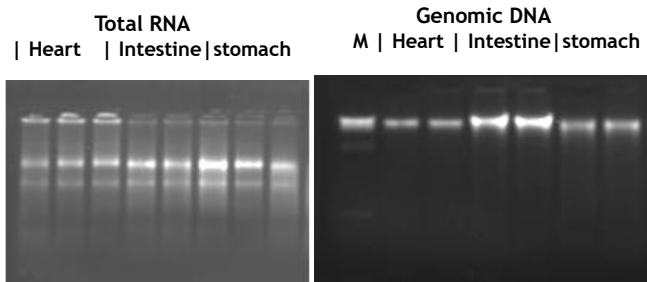


Take purified DNA and RNA, analyze using a Nanodrop 2000 (Thermo Fisher) UV spectrophotometer. Results show that the RNA and DNA obtained had high purity, of which OD260/OD280 and OD260/230 are within the ideal range. According to the yield, the total RNA content of 10mg liver reach 43µg, and the DNA content is as high as 14µg. The repeatability between the two repeated samples is good, and the yield is uniform.

Sample	Conc. µg/µl	260/280	260/230	Yield µg
Total RNA Isolation				
Chicken liver	0.219	2.1	1.98	43.8
	0.205	2.11	1.72	41
Chicken kidney	0.0634	2.16	2.41	12.68
	0.0629	2.15	2.47	12.58
Chicken spleen	0.0698	2.11	1.27	13.96
	0.0654	2.09	2.18	13.08
Chicken lung	0.0294	2.15	1.85	5.88
	0.029	2.13	1.74	5.8
Genomic DNA Isolation				
Chicken liver	0.1493	1.96	0.55	14.93
	0.137	1.96	0.44	13.7
Chicken kidney	0.5159	1.94	1.44	51.59
	0.4116	1.94	1.78	41.16
Chicken spleen	0.1934	1.9	2.3	19.34
	0.2502	2.13	2.3	25.02
Chicken lung	0.1208	1.89	2.76	12.08
	0.1279	2.1	2.44	12.79

2. Extract DNA/ RNA from Fibrous samples simultaneously

Take 10mg difficult-to-lyse animal tissue samples (chicken heart/intestine/stomach) and extract them using the HiPure Fibrous DNA/RNA Kit. After extraction, 1 µg of total RNA is taken for electrophoretic analysis on 1.0% agarose gel, and 5% of genomic DNA is taken for electrophoretic analysis on 0.8% agarose gel. From the electrophoresis chart, it can be seen that the RNA obtained using the kit does not degrade, and the obtained DNA fragments are intact without tailing phenomenon. The Lambda DNA/Hind III Marker indicates that the DNA fragment obtained is around 23KB.



Take purified DNA and RNA and analyze using a Nanodrop 2000 (Thermo Fisher) UV spectrophotometer. From the electrophoresis chart, it can be seen that the RNA and DNA obtained are intact. Data analysis results show that the DNA and RNA obtained have high purity and can be suitable for various downstream applications.

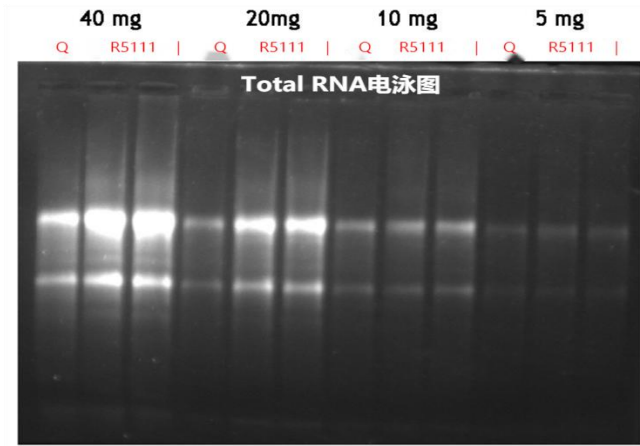
Sample	Conc. µg/µl	260/280	260/230	Yield µg
Total RNA Isolation				
heart	0.1062	2.13	1.95	10.62
	0.0935	2.13	1.72	9.35
intestine	0.3947	2.14	2.25	39.47
	0.3485	2.14	2.28	34.85
stomach	0.2885	2.14	2.23	28.85
	0.2896	2.14	2.26	28.96
Genomic DNA Isolation				
heart	0.0291	1.96	1.74	2.91
	0.0103	2.28	5.51	2.06
intestine	0.1299	1.91	2.07	12.99
	0.1229	1.91	2.12	12.29
stomach	0.0685	1.87	2.22	6.85
	0.0625	1.87	0.55	6.25

3. Comparison between R5111 and Qiagen AllPure DNA RNA Kit

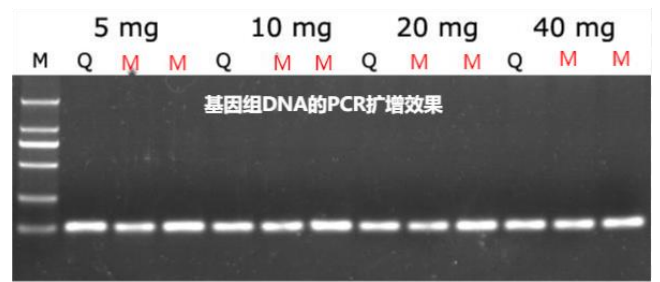
- Take different chicken liver samples, extract DNA/RNA using Magen R5111 (HiPure DNA/RNA Kit) and Qiagen AllPure DNA/RNA Kit. After extraction, elute with 100µl RNase Free Water (RNA component) and 100µl Elution Buffer (DNA component), measure OD value and perform electrophoresis analysis. Results are as follows:

RNA						
Sample	Conc.	Unit	260/280	260/230	Yield	Company
40mg Liver	1.4701	µg/µl	2.11	2.02	147.01	Qiagen
	1.5608	µg/µl	2.14	2.01	156.08	Magen R5111
	1.4703	µg/µl	2.15	2.1	147.03	
20 mg Liver	0.8395	µg/µl	2.13	2.19	83.95	Qiagen
	0.9847	µg/µl	2.11	2.14	98.47	Magen R5111
	0.947	µg/µl	2.11	2.27	94.7	
10 mg Liver	0.369	µg/µl	2.11	2.08	36.9	Qiagen
	0.4129	µg/µl	2.13	2.24	41.29	Magen R5111
	0.4658	µg/µl	2.13	2.07	46.58	
5 mg Liver	0.1946	µg/µl	2.09	2.05	19.46	Qiagen
	0.2104	µg/µl	2.12	2.09	21.04	Magen R5111
	0.1864	µg/µl	2.11	2.28	18.64	
DNA						
Sample ID	Conc.	Unit	260/280	260/230	Yield	Company
40mg Liver	0.477	µg/µl	1.90	1.12	47.68	Qiagen
	0.527	µg/µl	1.90	1.12	52.68	Magen R5111
	0.532	µg/µl	1.90	1.12	53.18	
20 mg Liver	0.256	µg/µl	1.89	1.54	25.61	Qiagen
	0.256	µg/µl	1.90	0.60	25.62	Magen R5111
	0.244	µg/µl	1.89	1.97	24.37	
10 mg Liver	0.165	µg/µl	1.91	2.21	16.51	Qiagen
	0.170	µg/µl	1.91	1.38	16.97	Magen R5111
	0.168	µg/µl	1.92	2.12	16.81	
5 mg Liver	0.073	µg/µl	1.90	2.38	7.29	Qiagen
	0.070	µg/µl	1.92	2.38	6.99	Magen R5111
	0.083	µg/µl	1.88	2.44	8.29	

Total RNA electrophoresis



Genomic DNA PCR amplification



Genomic DNA electrophoresis

