NucleoSpin® miRNA

RNA Purification Products from MACHEREY-NAGEL

RNA Mini spin kit for small and large RNA species

Are you also looking for small RNA species?

NucleoSpin® miRNA

Superior selectivity – purify your RNA fractionated by size

Small RNA (18 – 200 bases) ***or***

Large RNA (> 200 bases) ***or***

Total RNA (small and large RNA in one fraction)**,**

and isolate your total protein fraction

... without phenol/chloroform

NucleoSpin® miRNA: Patent pending

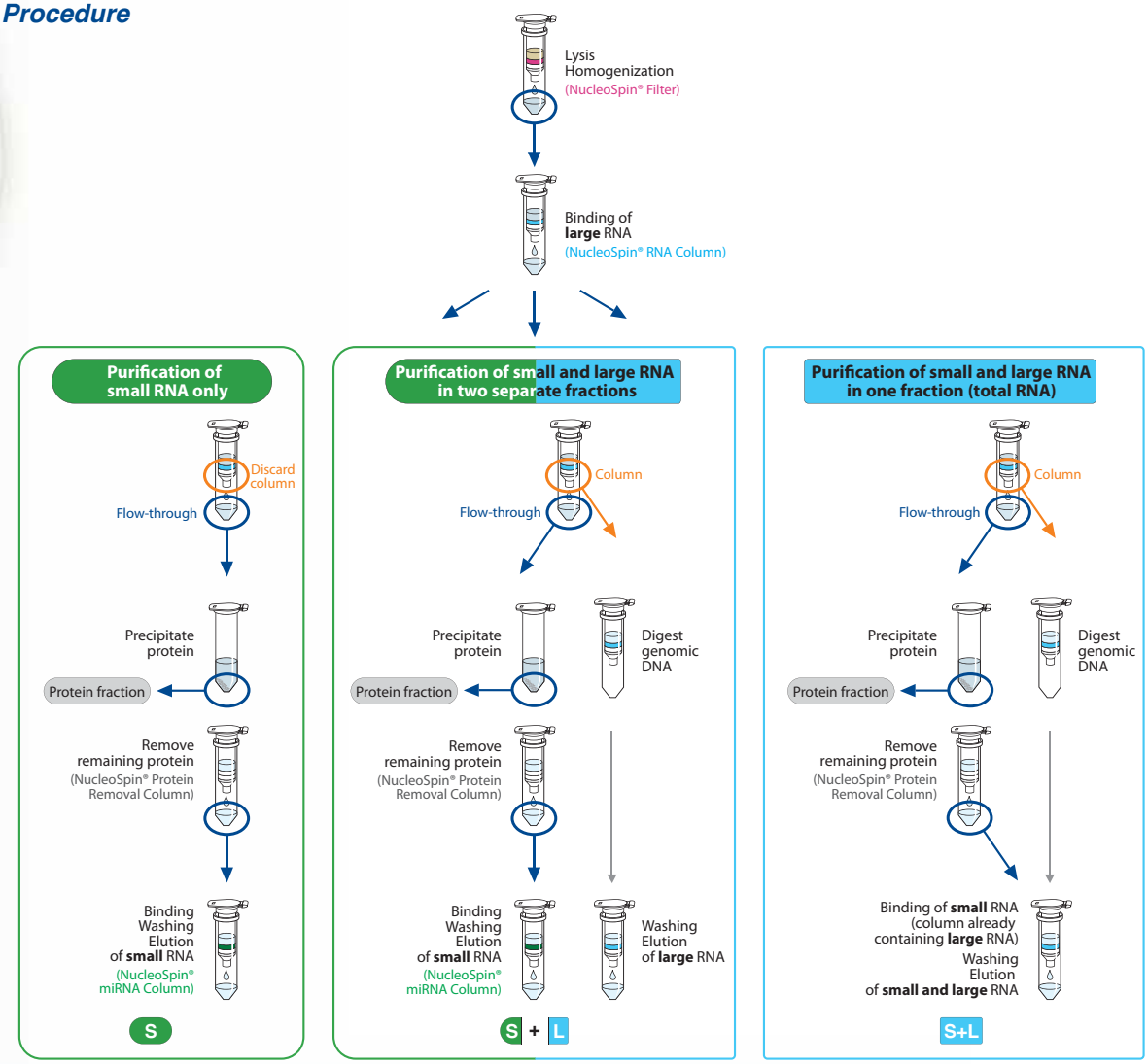
NucleoSpin® miRNA

NucleoSpin® miRNA

Parallel isolation of small RNA, large RNA, and protein

- ***Superior selectivity – purify your RNA fractionated by size*** *... highest flexibility!*
  - Isolation of small RNA only (18 – 200 bases) ***OR***
  - Isolation of small (18 – 200 bases) and large (>200 bases) RNA in two fractions ***OR***
  - Isolation of total RNA (small and large RNA in one fraction)
- ***Additional isolation of total protein*** *... confirm RNA results on protein level!*
  - Denatured protein ready-to-use for analysis in SDS-PAGE and Western Blot
- ***Excellent RNA recovery – even without phenol/chloroform*** *... less hazardous!*
  - Lysis with chaotropic salt
  - Spin-column based procedure
- ***Convenient handling – complete equipment provided*** *... ease of use!*
  - NucleoSpin® Filters → efficient sample homogenization
  - NucleoSpin® Protein Removal Columns → highest purity of small RNA
  - rDNase → highly efficient on-column digestion of genomic DNA at RT

Procedure



Product at-a-glance

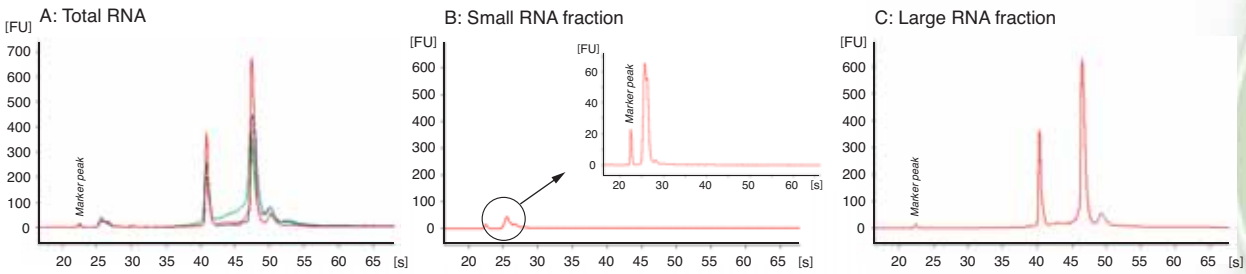
Technology	Silica-membrane technology
Format	Mini spin columns
Sample size	< 10^7 cultured cells, < 30 mg human/animal tissue < 50 mg plant tissue, < 150 µl reaction mixture
Fragment size small RNA	18 – 200 bases
Fragment size large RNA	> 200 bases
Typical yield	10 µg small RNA, 95 µg large RNA from 10^7 HeLa cells
Binding capacity	200 µg
Elution volume	30 – 100 µl
Preparation time	< 45 min (6 preps human/animal tissue, small and large RNA) < 35 min (6 preps human/animal tissue, small RNA only)

Application data

Very convenient RNA fractionation with highest selectivity

Total RNA was isolated from 10^7 HeLa cells using NucleoSpin® miRNA (—) and two competitor kits based on phenol/chloroform lysis and extraction (—) or phenol/chloroform extraction (—). Equal amounts of total RNA fractions were analyzed on an Agilent Bioanalyzer (A).

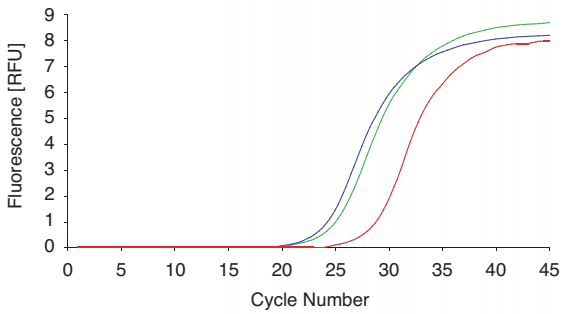
NucleoSpin® miRNA allows isolation of small (B) and large (C) RNA in separate fractions in addition to the total RNA fraction (A) with very high recovery without the need of phenol/chloroform.



Highly efficient removal of genomic DNA by on-column DNase digestion

Total RNA from 10^7 HeLa cells was purified with NucleoSpin® miRNA (—) and two competitor kits Q (—) and A (—). The RNA was assayed for residual traces of DNA by amplifying a 200 bp fragment of the ATPase 6 gene.

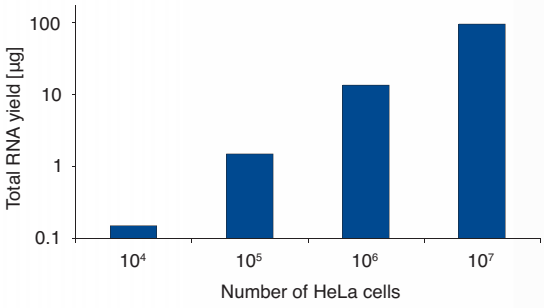
The ΔCts of 3.5 and 4.3 between NucleoSpin® miRNA and competitor A or Q, respectively, indicate a more than **tenfold increase in DNA removal** by the NucleoSpin® miRNA on-column DNase digestion compared to standard phenol/chloroform extractions.



Direct linear correlation of input cells to RNA yield shown by quantitative RT-PCR

Total RNA was purified from 10^4, 10^5, 10^6, and 10^7 HeLa cells using NucleoSpin® miRNA. MiR-16 was amplified in a qRT-PCR reaction using a Roche LightCycler™.

The graph shows a perfect linear correlation of input cells and RNA yield. The data table additionally shows corresponding linear decrease of Ct values with increasing RNA input.



Number of HeLa cells	10^4	10^5	10^6	10^7
Total RNA yield [µg]	0.15	1.49	13.4	94.5
qRT-PCR [Ct]	30.2	25.9	21.3	18.2

Application data (cont.)

Excellent yields for all types of sample materials

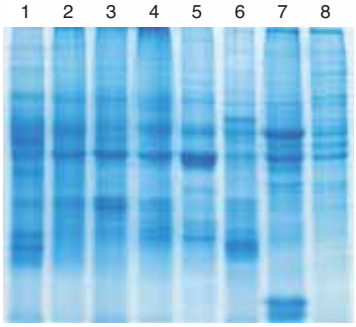
Total RNA and fractionated RNA from maximum amounts of different sample materials were purified with NucleoSpin® miRNA according to the individual protocols. Note that RNA yields as well as the ratio of small to large RNA vary due to species, developmental stage, etc.

Sample material	Amount	Protocol used	Yield total RNA [µg]	Yield fractionated RNA [µg] large RNA	small RNA
Mouse liver	30 mg	Tissue	100	105	19
Mouse kidney	30 mg	Tissue	35	31	9
Mouse spleen	30 mg	Tissue	48	36	22
Mouse lung	30 mg	Tissue	27	21	9
Mouse heart	30 mg	TRIzol®	24	19	4
Porcine liver	30 mg	Tissue	80	70	13
Human brain	30 mg	Tissue	11	10	3
Human brain	30 mg	TRIzol®	17	14	3
HeLa cells	10^7 cells	Cells	100	100	10

Cultured cells or soft tissue like liver, kidney, lung, etc. can easily be processed with the phenol-free standard procedures. For lipid tissue like brain or very hard-to-lyse, fibrous tissue like heart tissue it might be advantageous to use the protocol for RNA purification in combination with phenol/chloroform (e.g., TRIzol®) lysis to obtain optimal yields.

SDS-PAGE of precipitated protein fraction

Total protein from various tissues (see table left) was isolated during the NucleoSpin® miRNA procedure. The protein precipitate was dissolved in Laemmli-like protein solubilization buffer PSB and 40 µg were run on a 12% SDS polyacrylamide gel (100 V, 45 min).



1: Mouse liver
2: Mouse kidney
3: Mouse spleen
4: Mouse lung
5: Mouse heart
6: Porcine liver
7: Human brain
8: HeLa cells

Ordering information

Product	Preps	Cat. No.
Mini spin columns ***NucleoSpin® miRNA*** Mini spin kit for parallel isolation of small and large RNA.	10/50/250	740971.10/.50/.250
Related products		
Mini spin columns ***NucleoSpin® RNA II*** Mini spin kit for isolation of total RNA. Including rDNase and shredders.	10/50/250	740955.10/.50/.250
NucleoSpin® RNA/Protein Mini spin kit for simultaneous isolation of total RNA and protein from unsplit samples. Including rDNase and shredders.	10/50/250	740933.10/.50/.250
NucleoSpin® TriPrep* Mini spin kit for simultaneous isolation of total RNA, genomic DNA, and total protein from a wide variety of unsplit samples.	10/50/250	740966.10/.50/.250
Mini spin columns – XS design ***NucleoSpin® RNA XS*** Mini spin kit for isolation of highly concentrated total RNA from extremely small amounts of starting material. Elution volume down to 5 µl.	10/50/250	740902.10/.50/.250
NucleoSpin® FFPE RNA Mini spin kit for isolation of total RNA from formalin fixed, paraffin embedded samples. Including Paraffin Dissolver, Decrosslinking Buffer, and rDNase.	10/50/250	740969.10/.50/.250
Buffer and reagents sets ***Protein Quantification Assay*** Buffers and reagents for quantification of proteins. Compatible with detergents and reducing agents. Reference BSA included.	50/250	740967.50/.250

Visit ***www.mn-net.com/bioanalysis*** for detailed information

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NucleoSpin® miRNA

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