

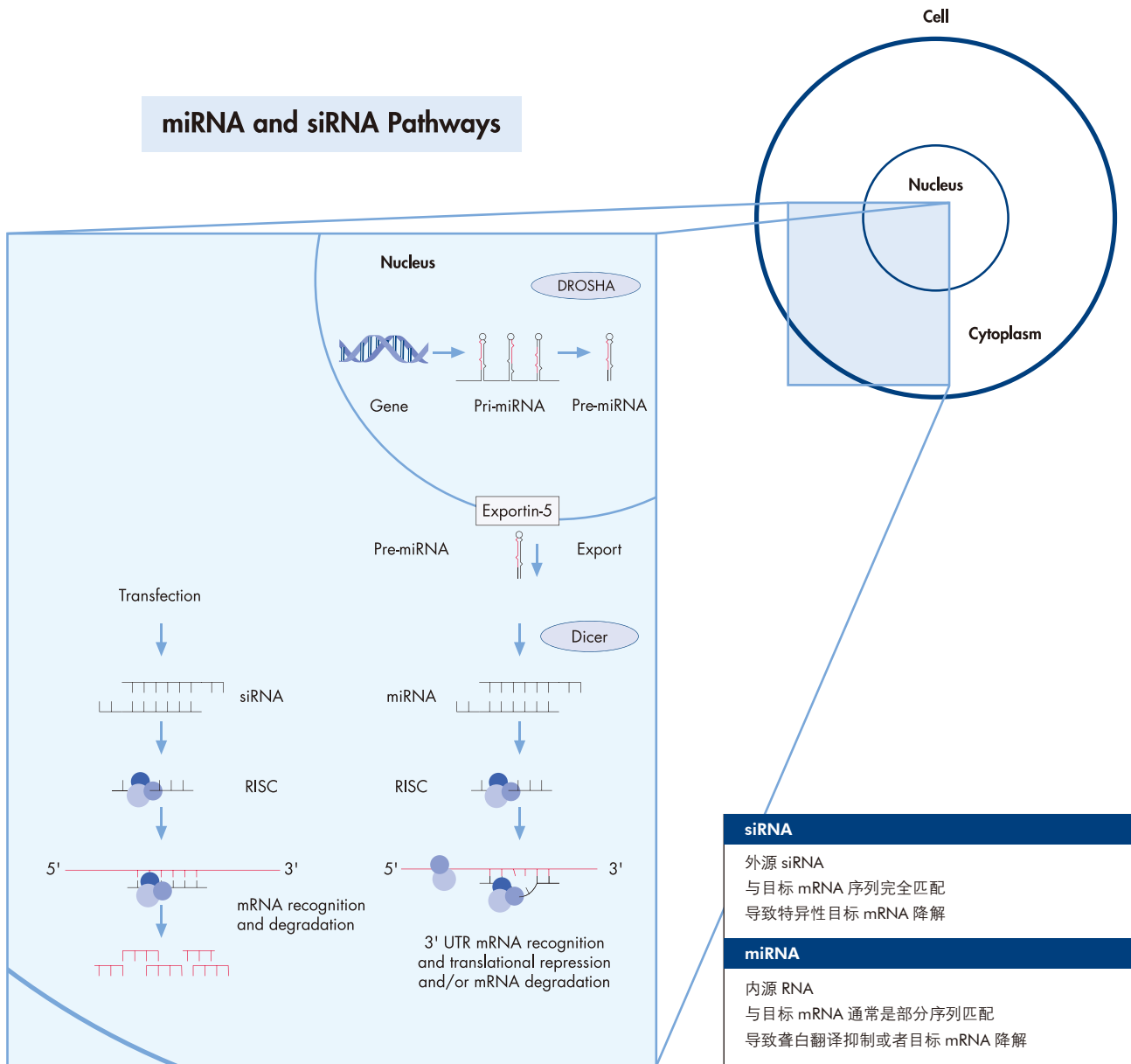
# microRNA 研究完整解决方案

NEW



Sample & Assay Technologies

## miRNA and siRNA Pathways



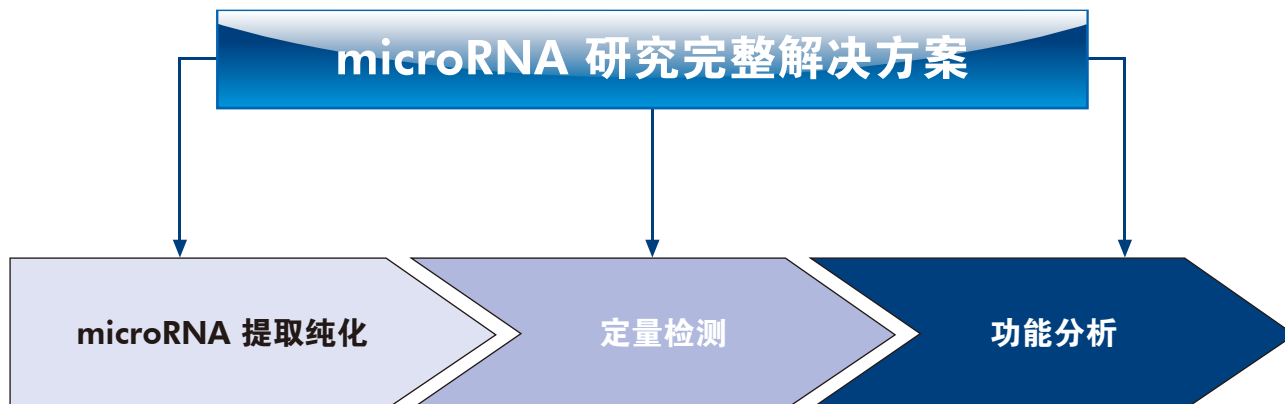
miRNAs的形成首先是在细胞核内转录得到片段较长的初级miRNA(Pri-miRNA)，然后在核内由dsRNA特异性核酸酶Drosha加工成前体miRNA(Pre-miRNA)。Pre-miRNA通过Exportin-5由核内运送到细胞质中，再由Dicer酶复合体进一步加工成为~22 nt的成熟miRNAs，随即被整合到RNA沉默复合体RISC中，当RISC复合体识别互补或部分互补的目标mRNA分子后，就会阻止蛋白质的翻译或者降解mRNA，从而抑制基因的表达。

### miRNA 研究相关资源

请登陆 [www.qiagen.com/miRNA](http://www.qiagen.com/miRNA) 查询:

- 实验操作方法
- 应用实例
- 参考文献
- 背景信息
- miRNA 数据库链接

## microRNA 研究完整解决方案



miRNeasy Mini Kit	miScript Reverse Transcription Kit	HiPerFect Transfection Reagent
miRNeasy 96 Kit	miScript SYBR <sup>®</sup> Green PCR Kit	miRNA Inhibitors
miRNeasy FFPE Kit	miScript Primer Assays	miRNA Mimics

### 本手册的内容包括：

- 从细胞、组织等样品中提取纯化miRNA ..... P3
- 针对miRNA的简单一步法cDNA合成 ..... P4
- 基于SYBR Green法real-time PCR定量检测miRNA ..... P5
- miRNA特异性引物数据库 ..... P5
- miRNA功能研究概述 ..... P6
- 用于miRNA功能研究的miRNA模拟物 ..... P7
- 用于miRNA功能研究的miRNA抑制剂 ..... P8
- 模拟物和抑制剂的转染 ..... P9

## miRNeasy Mini Kit & miRNeasy 96 Kit

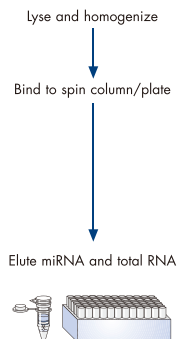
### 高效提取纯化含 miRNA 的总 RNA 或单独富集 miRNA

- 纯化 >18nt 至 200 nt 的小 RNA，可单独富集 miRNA 组分，这种富集可能有利于芯片分析及低丰度 miRNA 检测
- miRNA 回收效率高，利于提高检测灵敏度
- 纯度高，可用于 northern、real-time RT-PCR、microarray 等分析
- 选择灵活，提供单个样品和 96 孔板纯化方式

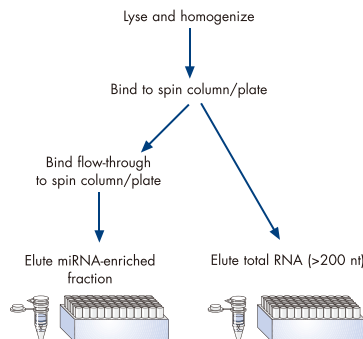
miRNA 在 Chaotropic 盐溶液环境下和硅胶膜柱吸附，通过简单的裂解、上柱、洗涤、洗脱步骤得到 18nt 至 200nt 的小 RNA 或含小 RNA 的总 RNA 组分，整个过程约 30 分钟。适合各种细胞和动物组织 (FFPE 组织使用 miRNeasy FFPE Kit)，处理其他样品 (如细菌、植物、血液等) 请咨询 QIAGEN 技术支持。

### miRNA 纯化操作流程简单快速

#### Copurification of miRNA and total RNA



#### Separate purification of miRNA-enriched fraction and total RNA\*



\* Separate purification using the miRNeasy Mini Kit requires additionally the RNeasy MinElute Cleanup Kit. For separate purification using the miRNeasy 96 Kit, an additional RNeasy 96 plate is required. For economical purchase of an additional RNeasy 96 plate, we recommend ordering an RNeasy 96 Kit (订购信息见第10页)。

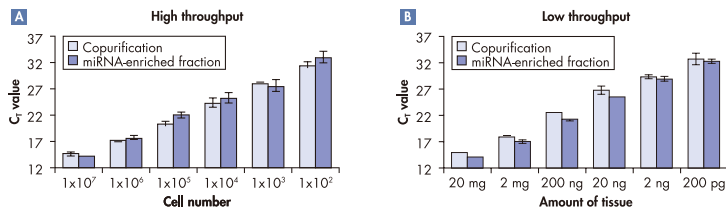


图1. 从不同起始量的样品中高效纯化miRNA。

Total RNA was purified from A) 10<sup>2</sup>-10<sup>7</sup> Jurkat cells using the miRNeasy 96 Kit or B) a dilution series of rat lung tissue homogenate from 20 mg to 200 pg tissue, using the miRNeasy Mini Kit. miRNA-enriched fractions (<200 nt) were also isolated from the same samples. Purified RNA was used as a template in quantitative, real-time RT-PCR assays for the miRNA miR-16.

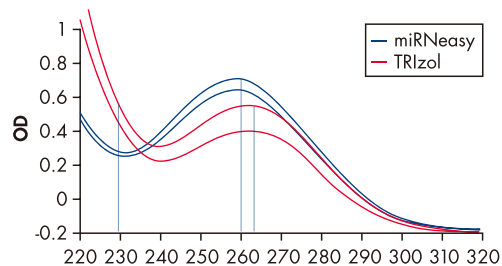


图2. 高纯度RNA，无苯酚干扰。

Total RNA including miRNA was purified from 1x10<sup>6</sup> Jurkat cells using the miRNeasy Mini Kit or precipitation from TRIzol Reagent. OD260 was lower when using TRIzol, indicating phenol carryover. In addition, the OD230 measurement was higher in the TRIzol prepared RNA, indicating salt carryover from the TRIzol Reagent.

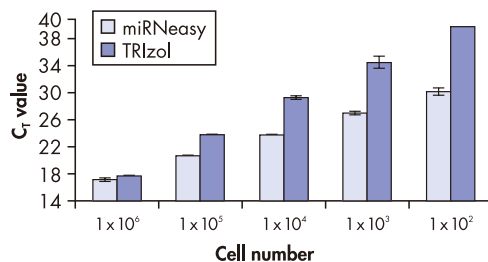


图3. miRNeasy Kit纯化的结果优于TRIzol。

Real-time RT-PCR assays for the miRNA miR-16. Results showed that CT values were lower after purification using the miRNeasy Kit, indicating that higher amounts of miRNA were purified than when using TRIzol. miRNA was effectively purified from as little as 1x10<sup>2</sup> cells using the miRNeasy Kit. In contrast, no miRNA was detected after 40 PCR cycles from 1x10<sup>2</sup> cells when TRIzol was used for purification.

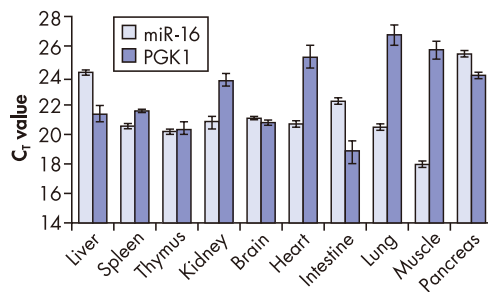


图4. 从不同组织中同时纯化miRNA和mRNA。

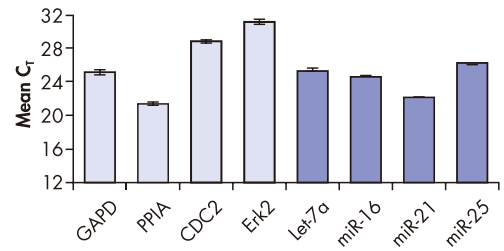
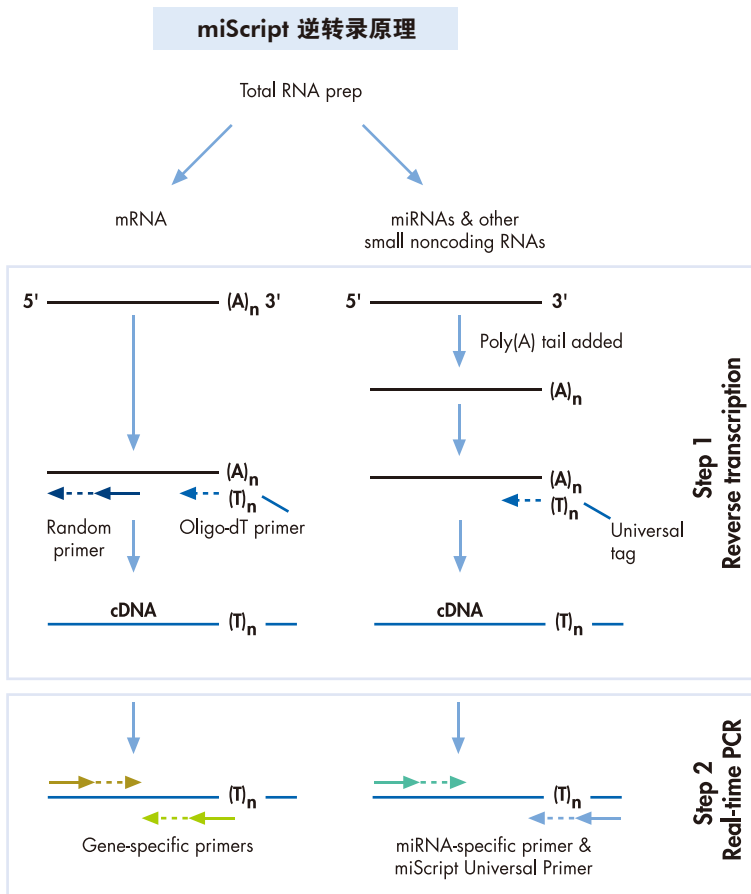
Total RNA including miRNA was purified from 25 mg of a range of RNAlater® stabilized rat tissues using the miRNeasy 96 Kit. Purified RNA was used as a template in quantitative, real-time RT-PCR assays for the miRNA miR-16 and for the larger mRNA of the PGK1 gene. Results showed successful detection of both PGK1 mRNA (large RNA) and miR-16 (small RNA) from the same eluates.

## miScript Reverse Transcription Kit

miRNA 逆转录试剂，以单独 miRNA 组分或含 miRNA 的总 RNA 为起始模板

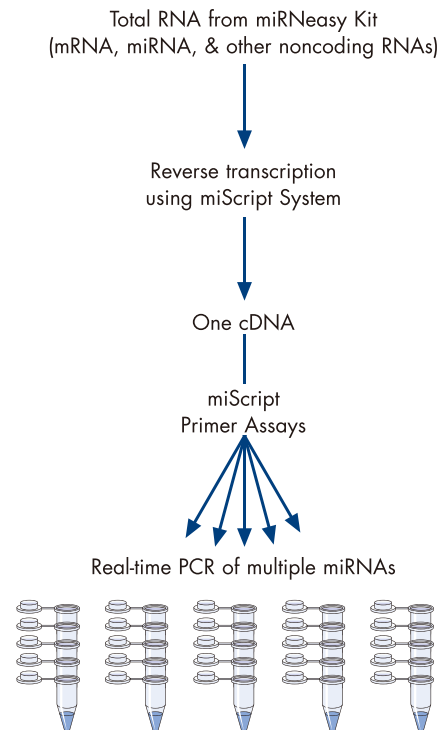
- 单次 cDNA 合成反应可用于检测多种 miRNAs 和 mRNAs
- miRNA、其它小 RNA 和 mRNA 同时逆转录得到 cDNA
- 线性范围广，起始总 RNA 从 10 pg 到 1 μg
- 操作简单，只需要两步孵育即可完成

由于 miRNA 无 poly(A) 尾巴，在逆转录步骤中需要通过 poly(A) 聚合酶先在其末端加 A，然后通过 5' 端带通用 tag 的 oligo-dT、随机引物将 miRNA 和 mRNA 同时转录得到 cDNA。此试剂盒包含逆转录步骤所需全套试剂。



**图5. 单次cDNA合成反应可用于检测多种miRNAs和mRNAs**  
Total RNA was prepared from HeLa S3 cells using the miRNeasy Mini Kit. The miScript System was used for real-time PCR analysis of 4 miRNAs(Let-7a, miR-16, miR-21, and miR-25). QuantiTect Primer Assays were used for real-time PCR analysis of 4 mRNAs (GAPDH, PPIA, CDC2, and Erk2).

### 单次 cDNA 合成反应可用于检测多种 miRNAs



## miScript System — miScript SYBR Green PCR Kit

### 使用 SYBR Green real-time PCR 方法定量检测 miRNA

- 可用于检测 miRNA、小 RNA 和 mRNA，只需加入各自的引物即可
- 灵敏度高，至 10 pg 含 miRNA 的总 RNA
- 特异性高（如 Let-7 isoforms 也可以被区分）
- 操作简单、快速，以 master mix 预混液形式提供

用于 miRNA 差异表达分析，含反向通用引物，只需加入 miRNA 特异性引物 miScript Primer Assay 和 cDNA 模板即可进行检测，实验条件已经优化过，参照说明书操作即可。

表 1. miScript Primer Assay 的高特异性

cDNA used in PCR	Relative detection (as % of perfect match)							
	miRNA primer assay used							
	Let-7a	Let-7b	Let-7c	Let-7d	Let-7e	Let-7f	Let-7g	Let-7i
Let-7a	100.00	0.00	0.29	0.33	2.44	0.01	0.00	0.00
Let-7b	0.00	100.00	1.68	0.00	0.00	0.01	0.00	0.00
Let-7c	0.27	0.14	100.00	0.00	0.00	0.00	0.00	0.00
Let-7d	4.11	0.00	0.03	100.00	0.01	0.00	0.00	0.00
Let-7e	1.23	0.00	0.01	0.01	100.00	0.00	0.00	0.00
Let-7f	5.77	0.00	0.00	0.00	0.00	100.00	0.00	0.00
Let-7g	0.01	0.00	0.00	0.00	0.00	0.00	100.00	0.00
Let-7i	0.00	0.00	0.00	0.00	0.00	0.00	0.00	100.00

Synthetic miRNAs of each Let-7 isoform were used in cDNA synthesis reactions performed with the miScript Reverse Transcription Kit. An aliquot of the resultant cDNA was used as a template in real-time PCR reactions with a miScript Primer Assay for each isoform and the miScript SYBR Green PCR Kit. The % relative detection was calculated using the differences between the CT values achieved from the mismatching miScript Primer Assays and those from the perfectly matching miScript Primer Assays (% relative detection =  $2^{-\Delta CT} \times 100$ ).

miRNA sequence	
Let-7a	UGAGGUAGUAGGUUGUAUAGUU
Let-7b	UGAGGUAGUAGGUUGUGUGUU
Let-7c	UGAGGUAGUAGGUUGUAUGUU
Let-7d	<u>A</u> GAGGUAGUAGGUUG <u>C</u> AUAGU•
Let-7e	UGAGGUAG <u>G</u> AGGUUGUAUAGU•
Let-7f	UGAGGUAGUAG <u>A</u> UUGUAUAGUU
Let-7g	UGAGGUAGUAG <u>U</u> UUGUA <u>C</u> AGU•
Let-7i	UGAGGUAGUAG <u>U</u> UUGU <u>G</u> CUGU•

表 2. 人 Let-7 家族异构体

These sequences show the Let-7 isoforms. Base changes are red and underlined. Changes in length are indicated by a red dot.

## miScript Primer Assay

### 预设计的 miRNA 特异性引物

- 选择灵活，提供单管装（供200次25ul检测体系）或96孔板文库
- 目前提供的miRNA引物文库有人的（714种）、小鼠（561种）及大鼠的（346种），以96孔板或384孔板提供，并且随着miRBase数据库不断更新
- 通过[www.qiagen.com/GeneGlobe](http://www.qiagen.com/GeneGlobe) 检索引物，方便快捷

提供现有miRNA数据库miRBase中所有的人、小鼠、大鼠、病毒miRNA对应的引物，其中人的引物已经通过实验验证。

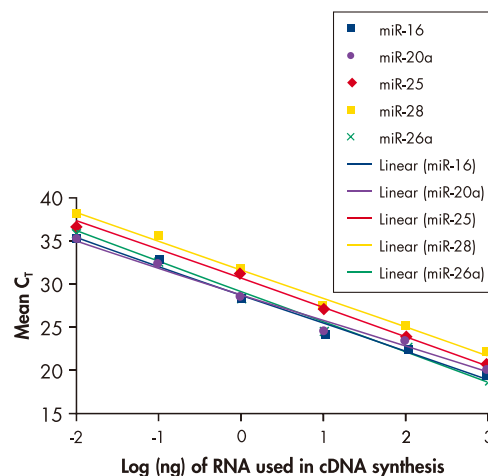


图6. 高度线性的cDNA合成及real-time PCR检测。

RNA was purified from HeLa S3 cells using the miRNeasy Mini Kit. A range of amounts of RNA from 10 pg to 1 μg were used in cDNA synthesis reactions using the miScript Reverse Transcription Kit. CDNA was used as a template in quantitative, real-time PCR assays for 5 miRNAs (miR-16, miR-20a, miR-25, miR-28, and miR-26a).



[www.qiagen.com/GeneGlobe](http://www.qiagen.com/GeneGlobe)

**Tips**

GeneGlobe™ 是全球最大的基因特异性产品的数据库。覆盖了人、小鼠、大鼠全基因组及其他物种的部分基因，通过网页界面您可以很方便的查找到针对特定基因的经验证的 microRNA 引物、real-time PCR 引物、siRNA 产品、蛋白检测产品以及相关的试剂，只需要输入基因名称或 ID 即可搜索到对应的产品。

## miScript miRNA Mimics and miScript miRNA Inhibitors

### 用于miRNA功能研究和基因调控分析

miScript miRNA Mimics是化学合成的双链RNA分子，转染至细胞后模拟内生miRNA发挥作用；miScript miRNA Inhibitors是化学合成的单链RNA分子，转染至细胞后特异性抑制miRNA的功能。因此，转染mimics或inhibitors可以研究误调控miRNA所引起的生物学效应，并通过下游的基因表达分析或表型分析，可确定特定miRNA的靶标基因及其功能。如果转染mimic引起某基因表达下调或转染inhibitor引起某基因表达上调，说明此miRNA参与那个基因的调控（图7）。典型的实验设计包括转染mimic或inhibitor，或者还可以共转染带有miRNA结合位点的报告基因载体系统，下游的检测包括报告系统分析、real-time PCR、芯片分析、蛋白分析。

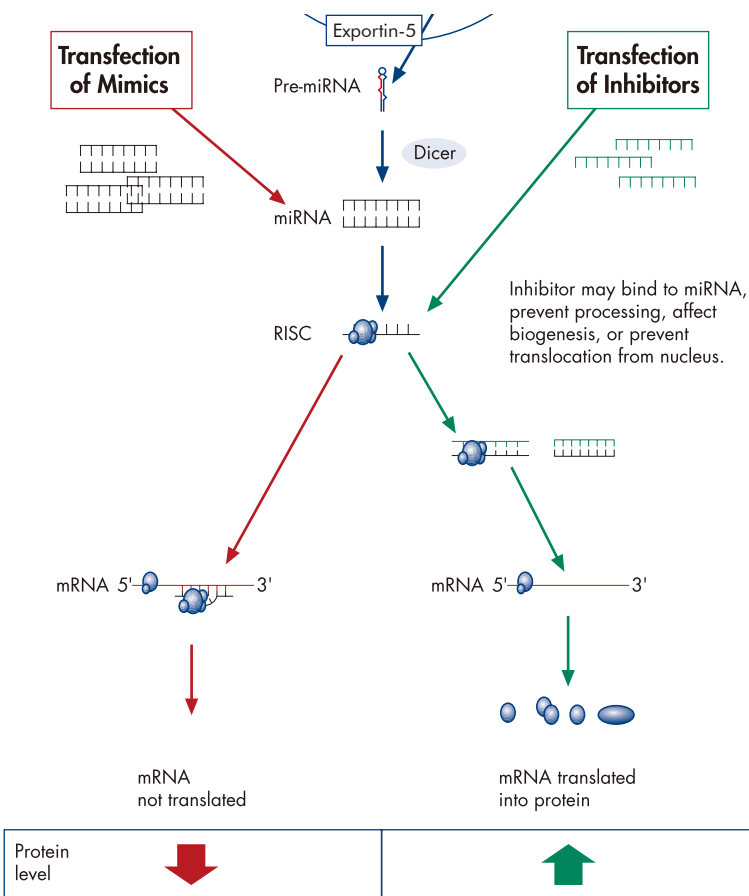


图 7. Mimics 下调蛋白表达水平，inhibitors 上调蛋白表达水平。

**miRNA 的产生：**核内初级 miRNAs ( pri-miRNAs ) 经 Drosha 加工成前体 miRNAs ( pre-miRNAs )，然后 Pre-miRNAs 经 Exportin-5 转运到胞浆内，经 Dicer 加工成成熟 miRNAs 并整合到 RISC 中，RISC 识别靶 mRNA 并抑制其翻译，引起蛋白质水平下调，miRNAs 也可能通过去腺苷酸和脱帽引起 mRNA 降解。

**miRNA mimic 的功能：**miRNA mimics 模拟内生 miRNA 起作用，因此转染 miRNA mimic 可能引起蛋白质水平进一步下降。

**miRNA inhibitor 的功能：**转染 miRNA inhibitor 导致蛋白水平上升，miRNA inhibitor 通过下面几个可能的机制抑制 miRNA 的功能，包括结合到成熟的 microRNA 上、干扰 miRNA 的生成、阻止 miRNA 前体的加工、或者阻止其从核内转运到胞浆。

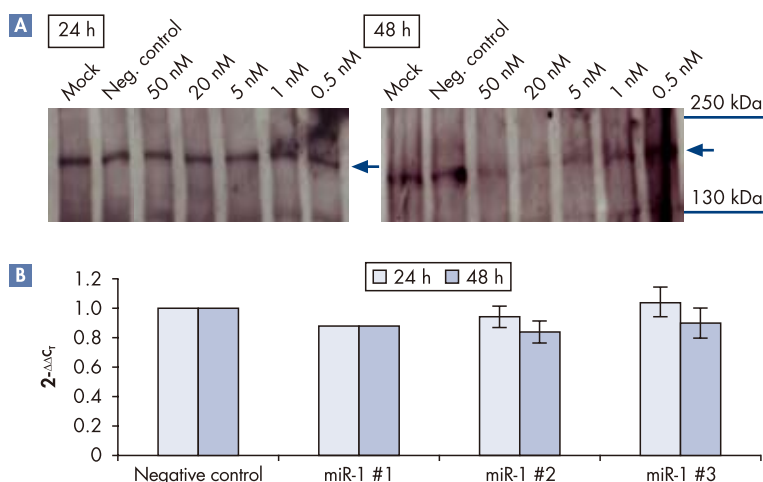
## miScript miRNA Mimics

### 用于 miRNA 功能研究的 miRNA 模拟物

- 即用型化学合成的双链 RNA 分子，模拟内生成熟的 miRNAs
- 提供现有 miRBase 数据库中所有人、小鼠、大鼠、病毒 miRNA 的 Mimics
- 提供 Mimics 客户定制合成服务
- 包装灵活，纯度分为细胞转染级和动物体内实验级

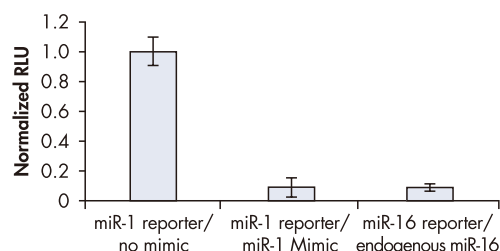
### 应用实例

miScript miRNA Mimics 模拟内生成熟的 miRNAs 发挥作用。miScript miRNA Mimics 引起基因表达下调，类似于内生 miRNAs 引起的现象（图 8）。所观察到的基因表达的下调发生在翻译水平，在转录本水平未检测到改变（图 9）。miScript miRNA Mimics 在转染后有效抑制基因表达长达 72 小时。



**图9. 转染miScript miR-1 Mimic后蛋白表达水平下调。**

HDAC4 is predicted to be a target of miR-1 and has 2 miR-1 binding sites in the 3' UTR (see Ref. 2). HeLa S3 cells ( $8 \times 10^4$  cells/well) were transfected using HiPerFect Transfection Reagent and a range of concentrations of miR-1 Mimic. After 24 and 48 hours, **A** HDAC4 protein level was analyzed by western blotting (indicated with arrows). Tubulin was also analyzed as an internal control (not shown). No effect was observed after 24 hours. After 48 hours, protein level was significantly decreased. **B** HDAC4 transcript level was analyzed by real-time PCR using the miScript System after transfection using 50 nM miR-1 Mimic. The negative control was AllStars Negative Control siRNA. miR-1 #1, miR-1 #2, and miR-1 #3 are different batches of miR-1 Mimic. Results were normalized to GAPDH and to the negative control and are presented as  $2^{-\Delta\Delta CT}$ . Transcript levels did not change significantly at either the 24-hour or 48-hour time point.



**图8. 内生miRNA和miScript miRNA Mimic引起的基因表达下调结果具有可比性。**

miR-1 is not endogenously expressed in HeLa S3 cells. miR-16 is endogenously expressed at high levels in HeLa S3 cells (see Ref. 1). Using HiPerFect Transfection Reagent, HeLa S3 cells ( $6 \times 10^4$  cells/well) were transfected with a luciferase reporter construct with a binding site for miR-1 in the 3' UTR; cotransfected with a luciferase reporter construct with a binding site for miR-1 in the 3' UTR and with an miR-1 Mimic; or transfected with a luciferase reporter construct with a binding site for miR-16 in the 3' UTR. Twenty-four hours later, luciferase activity was measured. miR-1 Mimic resulted in a similar decrease in luciferase level as endogenous miR-16.

#### Tips

#### Mimic 查找:

登陆 GeneGlobe 数据库 [www.qiagen.com/GeneGlobe](http://www.qiagen.com/GeneGlobe), 输入 miRNA 名称, 即可搜索到对应的 Mimic。细胞转染级提供 1nmol (够 24 孔板约 400 次实验)、5nmol 包装, 体内实验级提供 20nmol 包装。可以 96 孔板或 384 孔板提供。

#### 实验对照设置:

阳性对照: 使用 Syn-hsa-miR-1 miScript miRNA Mimic (cat. no. MSY0000416), Syn-hsa-miR-1 是仅在人肌肉细胞中表达的 miRNA, 转染对应的 Mimic 后可检测 miRNA 的靶基因 HDAC4 表达水平是否下降, 或者共转入带 hsa-miR-1 结合位点的报告载体用于分析。

阴性对照: 使用 AllStars Negative Control siRNA (cat. no. 1027280), 此 RNA 序列经过最严格的实验验证与已知哺乳动物基因无同源性, 如果使用报告载体用于分析, 与载体一起转染。



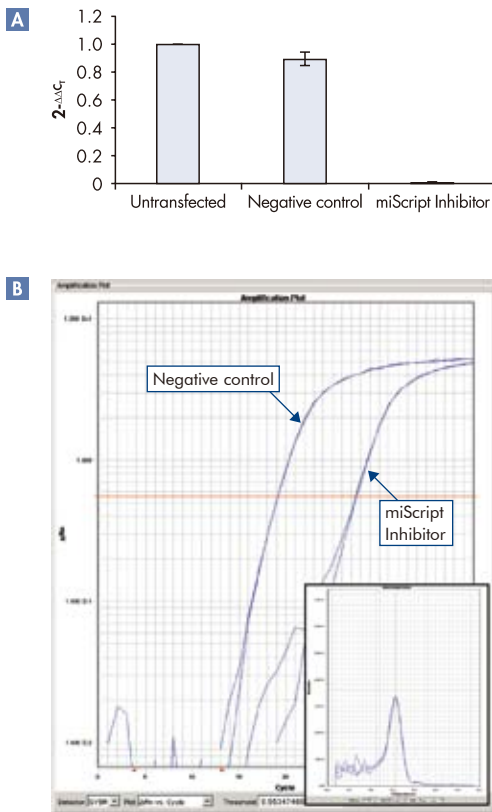
## miScript miRNA Inhibitors

### 用于miRNA功能研究的miRNA抑制剂

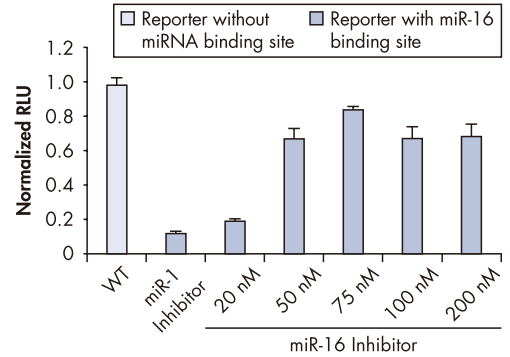
- 即用型2-OMe化学修饰的单链RNA分子，抑制内生miRNAs功能
- 提供现有miRBase数据库中所有人、小鼠、大鼠、病毒 miRNA 的Inhibitors
- 提供Inhibitors客户定制合成服务
- 包装灵活，纯度分为细胞转染级和动物体内实验级

### 应用实例

miScript miRNA Inhibitors 通过设计和修饰以达到有效地抑制内生 miRNAs 的作用 (图 10)。转染 Inhibitors 后 real-time PCR 检测到 miRNA 水平显著下降 (图 11)。分时检测实验表明转染后 Inhibitors 的作用可长达 96 小时。



**图11. 转染miScript miRNA Inhibitor后下调了内生miRNA的表达水平。** HeLa S3 cells ( $6 \times 10^4$  cells/well) were transfected using HiPerFect Transfection Reagent with miScript miR-16 Inhibitor or a negative control. Endogenous miR-16 levels were quantified by real-time PCR using the miScript System. U6B was used as the reference RNA for data normalization and miR-16 levels were expressed as  $2^{-\Delta\Delta CT}$  values relative to the miR-16 levels in untransfected cells. **A** Transfection of miScript miR-16 Inhibitor strongly reduced the amount of detectable endogenous miR-16. **B** The amplification plots indicated a significant decrease in the level of detectable miR-16 as a result of miR-16 Inhibitor transfection. Dissociation curve analysis (inset) reflects the specificity of the real-time PCR detection using the miScript System.



**图10. miScript miRNA Inhibitor 抵消了miRNA引起的靶基因沉默。**

HeLa S3 cells express miR-16 at high levels and do not express miR-1. In a cotransfection experiment using HiPerFect Transfection Reagent, HeLa S3 cells ( $6 \times 10^4$  cells/well) were cotransfected with a luciferase reporter construct with an miR-16 binding site in the 3' UTR together with miR-16 Inhibitor. miR-16 Inhibitor was used at varying concentrations in the experiment to evaluate the optimal inhibitor concentration required to see the inhibitory effect. Alternatively, cells were transfected with miR-1 Inhibitor alone as a control. A luciferase construct without an miRNA binding site (WT) was transfected as a positive control. An increase in luciferase expression following transfection of the miR-16 Inhibitor indicated that it prevented endogenous miR-16 from downregulating the reporter gene.

#### Tips

#### Inhibitors 查找:

登陆 GeneGlobe 数据库 [www.qiagen.com/GeneGlobe](http://www.qiagen.com/GeneGlobe), 输入 miRNA 名称, 即可搜索到对应的 Inhibitors。细胞转染级提供 1nmol (够 24 孔板约 40 次实验)、5nmol 包装, 体内实验级提供 20nmol 包装。可以 96 孔板或 384 孔板提供。

#### 实验对照设置:

**阳性对照:** 由于靶基因可能会受多个 miRNA 调控, Inhibitor 的效果可能会较复杂。建议同时转染 hsa-miR-1 inhibitor (cat. no. MIN0000416) 和 hsa-miR-1 mimic (cat. no. MSY0000416), 另外再单独转染 hsa-miR-1 mimic, 前者的靶基因表达应该高于后者。如使用了报告载体, 则将三者共同转染的数据对比共转染载体与 mimic 的数据。

**阴性对照:** 使用 miScript Inhibitor Negative Control, 此单链 RNA 序列经过最严格的实验验证与已知哺乳动物基因无同源性, 如果使用报告载体用于分析, 与载体一起转染。

## HiPerFect Transfection Reagent

### 经优化的用于miRNA Mimics和miRNA Inhibitors的转染试剂

HiPerFect试剂采用独特的正电和中性脂质分子的混合物，确保转染复合物在细胞内有效的释放出RNA分子，达到高效转染，在转染siRNA的实验中，已经被广泛用于各种帖壁真核细胞，甚至是难转染的原代细胞、悬浮细胞。经过优化及实验测试，用于miRNA Mimics和miRNA Inhibitors的转染条件见下表，推荐使用5nM ( Mimics )、50nM ( Inhibitors ) 及3 $\mu$ l转染试剂作为初始值。

**表3. 优化24孔板miRNA Mimics/miRNA Inhibitors转染**

Amount (conc.) of miRNA mimic	(10 nM)75 ng	(10 nM)75 ng	(10 nM)75 ng
Amount (conc.) of miRNA inhibitor	(100 nM)750 ng	(100 nM)750 ng	(100 nM)750 ng
Volume of HiPerFect Reagent	1.5 $\mu$ l	3 $\mu$ l	4.5 $\mu$ l
Amount (conc.) of miRNA mimic	(5 nM)37.5 ng	<b>(5 nM)37.5 ng</b>	(5 nM)37.5 ng
Amount (conc.) of miRNA inhibitor	(50 nM)375 ng	<b>(50 nM)375 ng</b>	(50 nM)375 ng
Volume of HiPerFect Reagent	1.5 $\mu$ l	<b>3 <math>\mu</math>l</b>	4.5 $\mu$ l
Amount (conc.) of miRNA mimic	(1 nM)7.5 ng	(1 nM)7.5 ng	(1 nM)7.5 ng
Amount (conc.) of miRNA inhibitor	(25 nM)187.5 ng	(25 nM)187.5 ng	(25 nM)187.5 ng
Volume of HiPerFect Reagent	1.5 $\mu$ l	3 $\mu$ l	4.5 $\mu$ l

\* 以上的量是以24孔板每孔计算

## Attractene Transfection Reagent **New**

### 新一代脂质技术，用于共转染报告载体和miRNA Mimics和/或miRNA Inhibitors

使用的报告载体DNA的量可能需要优化，推荐使用0.3 $\mu$ g报告载体DNA和1.5 $\mu$ l转染试剂作为初始值，Mimics初始值为5nM，Inhibitors初始值为50nM。DNA优化可按照下表进行。

**表4. 优化24孔板DNA转染**

Amount of DNA	0.2 $\mu$ g	0.2 $\mu$ g	0.2 $\mu$ g
Volume of Attractene Reagent	0.5 $\mu$ l	0.75 $\mu$ l	1.5 $\mu$ l
Amount of DNA	0.4 $\mu$ g	<b>0.4 <math>\mu</math>g</b>	0.4 $\mu$ g
Volume of Attractene Reagent	1 $\mu$ l	<b>1.5 <math>\mu</math>l</b>	3 $\mu$ l
Amount of DNA	0.6 $\mu$ g	0.6 $\mu$ g	0.6 $\mu$ g
Volume of Attractene Reagent	1.5 $\mu$ l	2.25 $\mu$ l	4.5 $\mu$ l

\* 以上的量是以24孔板每孔计算

以上转染试剂操作非常方便、快速，无需转染前一天铺细胞，转染当天铺细胞即可，加入转染复合物到细胞中孵育后，无需更换培养液，可在有血清及抗生素条件下进行，6-72小时后进行结果检测，最佳检测时间与使用的细胞类型及研究的miRNA分子有关，建议进行time-point分时检测。

## 订购信息

产品	规格	货号
miRNeasy Mini Kit (50)	50 RNeasy Mini Spin Columns, Collection Tubes (1.5 ml and 2 ml), QIAzol Lysis Reagent, RNase-Free Reagents and Buffers	217004
miRNeasy FFPE Kit (50)	50 RNeasy MinElute Spin Columns, 50 gDNA Eliminator Spin Columns, Collection Tubes, Proteinase K, RNase-Free Reagents and Buffers	217404
miRNeasy 96 Kit (4)	4 RNeasy 96 Plates, Collection Microtubes (racked), Elution Microtubes CL, Caps, S-Blocks, AirPore Tape Sheets, QIAzol Lysis Reagent, RNase-Free Reagents and Buffers	217061
miScript Reverse Transcription Kit (10)	For 10 reactions: miScript Reverse Transcriptase Mix, miScript RT Buffer, RNase-Free Water	218060
miScript Reverse Transcription Kit (50)	For 50 reactions: miScript Reverse Transcriptase Mix, miScript RT Buffer, RNase-Free Water	218061
miScript Primer Assay (100)	10x miScript Primer Assay (contains one miRNA-specific primer)	Varies*
miScript SYBR Green PCR Kit (200)	For 200 reactions: QuantiTect SYBR Green PCR Master Mix, miScript Universal Primer	218073
miScript SYBR Green PCR Kit (1000)	For 1000 reactions: QuantiTect SYBR Green PCR Master Mix, miScript Universal Primer	218075
miScript miRNA Mimic (1 nmol)	1 nmol (for approx. 400 transfections in 24-well plates)* cell-culture-grade miRNA mimic	Varies*
miScript miRNA Mimic (5 nmol)	5 nmol (for approx. 2000 transfections in 24-well plates)* cell-culture-grade miRNA mimic	Varies*
miScript miRNA Inhibitor (1 nmol)	1 nmol (for approx. 40 transfections in 24-well plates)* cell-culture-grade miRNA inhibitor	Varies*
miScript miRNA Inhibitor (5 nmol)	5 nmol (for approx. 200 transfections in 24-well plates)* cell-culture-grade miRNA inhibitor	Varies*
<b>单独纯化miRNA组分所额外需要的试剂</b> RNeasy® MinElute® Cleanup Kit (50)	50 RNeasy MinElute Spin Columns, Collection Tubes (1.5 ml and 2 ml), RNase-Free Reagents and Buffers	74204
RNeasy 96 Kit (4) <sup>†</sup>	4 RNeasy 96 Plates, Elution Microtubes CL, Caps, S-Blocks, AirPore Tape Sheets, RNase-Free Reagents and Buffers	74181
<b>经优化的用于miRNA Mimics和miRNA Inhibitors的转染试剂</b> HiPerFect Transfection Reagent (1 ml)	HiPerFect Transfection Reagent for up to 333 transfections in 24-well plates or up to 1333 transfections in 96-well plates	301705
Attractene Transfection Reagent (1 ml)	Attractene Transfection Reagent for up to 660 transfections in 24-well plates	301005
<b>相关产品</b> QuantiTect Primer Assay(200)	10x QuantiTect Primer Assay(contains a mix of forward and reverse primers for a specific target)	Varies*
QuantiTect SYBR Green PCR Kit(200) <sup>†</sup>	For 200 X 50 µl reactions: 3 x 1.7ml 2x QuantiTect SYBR Green PCR Master Mix, 2 x 2 ml RNase-Free Water	204143

\*请登陆[www.qiagen.com/GeneGlobe](http://www.qiagen.com/GeneGlobe)查询所需miRNA引物。†更大包装规格及‡实验操作手册，请咨询QIAGEN

## microRNA研究完整解决方案

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