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综 述

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MicroRNA 对胚胎干细胞的多能性网络调控

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摘要: 胚胎干细胞(Embryonic stem cells, ESCs)是一类能够无限增殖和诱导分化为多种类型细胞的干细胞。MicroRNA(miRNA)是一类内源性具有调控基因表达功能的非编码 RNA, 在 ESCs 增殖和分化过程中起重要作用。MiRNA 可以通过对 ESCs 多能性网络中的转录因子、细胞周期、表观遗传学、信号转导等方面调控, 促使 ESCs 维持多能性状态。文章重点综述了 miRNA 的生成过程、调控 ESCs 多能性的主要 miRNA 家族以及 miRNA 对 ESCs 多能性网络调控作用等内容。

关键词: ESCs; miRNA; 多能性; 调控

The effect of microRNAs on the regulatory network of pluripotency in embryonic stem cells

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Abstract: Embryonic stem cells (ESCs) are pluripotent stem cells characterized by their ability to self-renew and their pluripotency to differentiate into all cell types. MicroRNA (miRNA) is a small non-coding RNA molecule which can regulate transcriptional and post-transcriptional gene expression, and may also play significant roles in regulating proliferation and differentiation of ESCs. The maintenance of pluripotency in ESCs may involve a regulatory network of many factors and pathways regulated by miRNA, which includes ESCs transcription factors, cell cycle regulation, epigenetic modifications as well as intracelluar signal transduction. This review mainly elaborates the biogenesis of miRNA, the miRNA families regulating the pluripotency of ESCs, and the effect of miRNA on the regulatory network of pluripotency in ESCs.

Keywords: ESCs; miRNA; pluripotency; regulation

1981 年, Evans和Kaufman[1]首次成功分离得到小鼠 ESCs(Mouse embryonic stem cells, mESCs), 1998 年Thomson等[2]成功建立了人的ESCs(Human embryonic stem cells, hESCs)细胞系, 从此干细胞得到快速的发展。随着对ESCs调控机制的深入研究, 也促使了诱导多功能干细胞(Induced pluripotent stem cells, iPSCs)的诞生[3,4], 在临床医学上具有潜在的应用价值, 成为近几年来干细胞领域研究的热

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点[5]。研究发现, microRNA(miRNA)在ESCs和iPSCs增殖、分化过程中起着重要的调控作用。

miRNA是一类在动植物中广泛表达的内源性的单链非编码的RNA片段, 长度一般为21～24个核苷酸[6]。miRNA通过与mRNA的3′端非翻译区(UTRs)完全或不完全配对, 来阻断mRNA的正常翻译, 实现对靶基因转录后调控[7]。miRNA参与调控干细胞自我更新、定向分化、信号转导、细胞周期、表观遗传修饰和代谢凋亡等一系列的生理活动, 同时肿瘤的发生也往往是由于miRNA的异常突变、缺失或者表达而产生的[8,9]。Dicer和Dgcr8是参与miRNA生物合成的两个主要蛋白。缺失Dicer或Dgcr8的mESCs表现出细胞周期和表观遗传学功能异常的现象, 丧失了自我更新和定向分化的能力[10~12]。研究发现, miRNA-302家族的表达能促使体细胞重编程为iPSCs[13]; miR-9/9*和miR-124促使人的成纤维细胞转化为神经细胞[14]。此外, miR-302d、miR-372、miR-200c和miR-367等miRNAs调控hESCs的58个靶基因, 约有一半的基因参与转录调控[15]。可见, miRNA对细胞的生理调控具有重要作用。

1 miRNA 的生成过程

以哺乳动物miRNA生成过程为例：首先, miRNA是由RNA聚合酶Ⅱ从基因组转录生成单顺反子或者多顺反子的转录初级产物(Primary transcripts, pri-miRNA), 但不能翻译成蛋白质[16]; 其次, pri-miRNA在细胞核内被Drosha酶和RNA结合蛋白Dgcb8组装成的复合体加工, 形成长度约为70个核苷酸并具有茎环结构的miRNA前体(Precursor miRNA, pre-miRNA)[17]; 接着, 在转运蛋白Exportin 5的协助下, pre-miRNA从细胞核转运到细胞质中[18], 被Dicer酶切割成长度为20~24个核苷酸的miRNA双体[19]; 然后, miRNA双体中一条miRNA链进入由Ago2蛋白组成的RNA诱导基因沉默复合物(RNA-induced silencing complex, RISC)中, 形成非对称RISC复合物, 另外一条链被RNA酶降解[20]; 最后, 该复合物通过完全或不完全配对结合到mRNA的3′UTR上, 最终引起mRNA的降解, 下调了靶基因的表达水平。

2 调控 ESCs 的主要 miRNA 家族

2013年3月, miRbase数据库(http://www.mirbase.org/)公布人类和小鼠的细胞中分别有2 237和1 410个成熟的miRNA存在。人类基因图谱显示, 人类有3万多个基因, 预测其中约有1 000个miRNA基因存在, 大约1/3的人类基因受到miRNA调控[21]。miRNA家族一般在染色体上成簇排列, 受一个启动子调控而产生多顺反子初级转录产物[22]。目前, 调控hESCs多能性的miRNA家族主要包括人(*Homo sapiens*, hsa)miR-302家族、hsa-miR-371~373家族、hsa-miR-17家族、hsa-miR-200家族和hsa-miR-520家族[23](表1)。在mESCs中, 小鼠(*Mus musculus*, mmu) miR-290家族替代了hsa-miR-371~373家族而存在[24], 并且尚未有mmu-miR-520家族的报道。其中miR-17家族又分为miR-17~92家族、miR-106a~363家族和miR-106b~25家族, 分别位于3条不同的染色体上[25]; hESCs和mESCs的miR-200家族由5个miRNA组成, 位于两条染色体上, 它们种子序列完全一样, 但不同的是has-miR-200家族位于1和12号染色体上, 而mmu-miR-200家族位于4和6号染色体上[26]; hsa-miR-520家族是包含21个miRNA家族成员的大家族[27](表1)。这些调控ESCs细胞周期的miRNA家族具有类似的种子序列, 被通称为ESCs特有细胞周期调控家族(ESC-specific cell-cycle regulating, ESCC)。ESCC实际上不仅对ESCs的细胞周期有重要的调控作用, 而且还是ESCs增殖和维持多能性的主要miRNA家族, 广泛参与ESCs的转录因子、表观遗传学和信号转导等方面的调控[28]; 同时在体细胞重编程过程中, 也显著地提高重编程的效率[29,30]。

3 ESCs 多能性网络调控

ESCs多能性网络调控主要是通过核心转录因子(Oct4、Sox2和Nanog)及其信号通路, 激活ESCs的多能性基因, 抑制分化基因的表达, 对ESCs的细胞周期和表观遗传学进行调控, 最终促进ESCs维持多能性[31~33]。miRNA可以快速准确地调控基因的表达水平, 是完成转录因子对ESCs自我更新和分化调控的直接参与者[34]。

miRNA对ESCs多能性网络调控可归纳为4个

表 1 调控 hESCs 和 mESCs 多能性的主要 miRNA 家族种子序列表

家族	miRNA 名称	染色体	种子序列	家族	miRNA 名称	染色体	种子序列
hsa-miR-302	miR-302b	4	aagugcu▲	hsa-miR-17	miR-17-5p	13	aaagugc▲
	miR-302b*	4	cuuuaac●		miR-18a	13	aaggugc▲
	miR-302c	4	uuaacau●		miR-19a	13	gugcaaa–
	miR-302c*	4	aagugcu▲		miR-20a	13	aaagugc▲
	miR-302a	4	aagugcu▲		miR-19b-1	13	gugcaaa–
	miR-302a*	4	cuuaaac●		miR-92a-1	13	auugcac■
	miR-302d	4	aagugcu▲		miR-106a	X	aaagugc▲
	miR-367	4	auugcac■		miR-18b	X	aaggugc▲
					miR-20b	X	aaagugc▲
hsa-miR-371–373	miR-371	19	cucaaac●		miR-19b-2	X	gugcaaa–
	miR-372	19	aagugcu▲		miR-92a-2	X	auugcac■
	miR-373*	19	cucuaaa●		miR-363	X	auugcac■
	miR-373	19	aagugcu▲		miR-106b	7	aaagugc▲
					miR-93	7	aaagugc▲
hsa-miR-520	miR-512-5p	19	acucagc●		miR-25	7	auugcac■
	miR-498	19	uucaagc●				
	miR-515-3p	19	agugccu▲	mmu-miR-302	miR-302b	3	aagugcu▲
	miR-519e*	19	ucuccaa●		miR-302b*	3	cuuuaac●
	miR-520f	19	agugcuu▲		miR-302c	3	agugcuu▲
	miR-519c	19	ucuagag♦		miR-302c*	3	cuuuaac●
	miR-520a-3p	19	aagugcu▲		miR-302a	3	aagugcu▲
	miR-526b	19	ucuugag♦		miR-302a*	3	cuuaaac●
	miR-519b	19	ucuagag♦		miR-302d	3	cuuuaac●
	miR-525	19	uccagag♦		miR-367	3	auugcac■
	miR-523	19	aacgcgc▼				
	miR-518f	19	aaagcgc▼	mmu-miR-290	miR-290-3p	7	aagugcc▲
	miR-518f*	19	ucuagag♦		miR-290-5p	7	cucaaac●
	miR-520b	19	aagugcu▲		miR-291a-3p	7	aagugcu▲
	miR-518b	19	aaagcgc▲		miR-291b-3p	7	aagugca▲
	miR-526a	19	ucuagag♦		miR-292-3p	7	aagugcc▲
	miR-520c	19	ucuagag♦		miR-293-5p	7	cucaaac●
	miR-524-3p	19	aaggcgc▼		miR-294-3p	7	aagugcu▲
	miR-517a	19	ucgugca–		miR-295-3p	7	aagugcu▲
	miR-519d	19	aaagugc▲				
	miR-521	19	acgcacu●	mmu-miR-17	miR-17-5p	14	aaagugc▲
	miR-520d	19	uacaaag►		miR-18a	14	aaggugc▲
	miR-517b	19	cgugcau–		miR-19a	14	gugcaaa–
	miR-520g	19	caaagug▲		miR-20a	14	aaagugc▲
	miR-518e	19	aagcgcu▲		miR-19b-1	14	gugcaaa–

续表 1

家族	miRNA 名称	染色体	种子序列	家族	miRNA 名称	染色体	种子序列
	miR-518a	19	ugcaaag▶		miR-92a-1	14	auugcac■
	miR-517c	19	ucgugca▬		miR-106a	X	aaagugc▲
	miR-527	19	ugcaaag▶		miR-18b	X	aaggugc▲
	miR-516a-5p	19	ucucgag◆		miR-20b	X	aaagugc▲
					miR-19b-2	X	gugcaaa▬
hsa-miR-200	miR-200a	1	aucuuac◀		miR-92a-2	X	auugcac■
	miR-200b	1	aucuuac◀		miR-363	X	auugcac■
	miR-429	1	aauacug		miR-106b	5	aaagugc▲
	miR-200c	12	gucuuac◀		miR-93	5	aaagugc▲
	miR-141	12	aucuucc◀		miR-25	5	auugcac■

注：表中同一右上角标符号的种子序列为相近或者相同的序列。

部分：核心转录因子(Oct4、Sox2 和Nanog)的调控、细胞周期的调控、表观遗传学修饰以及信号转导的调控[35]，如图 1 所示。

3.1 miRNA 对 ESCs 核心转录因子的调控

转录因子对ESCs多能性网络调控起着核心作用[36]。研究证实Oct4、Sox2 和Nanog是ESCs多能性调控的核心转录因子，同时也是体细胞重编程过程中的重要因子[3,4]，它们共同参与调控数百个基因的启动子[37, 38]，促进更多的多能性转录因子的表达和抑制分化基因的表达来阻止ESCs的分化[39]。最初Ivanova等[40]证明了Oct4、Sox2、Nanog、Esrrb、Tbx3和Tcl1 是ESCs体外维持自我更新必需的因子；随后Chen等[41]在mESCs中鉴定出 13 个多能性转录因子Oct4、Sox2、Nanog、Klf4、c-Myc、n-Myc、 Stat3、Smad1、Zfx、Esrrb、Tcfcp2l1、E2f1 和Ctcf；同时Orkin等[42]也报道了mESCs的 9 个多能性转录因子Oct4、Sox2、Nanog、Klf4、c-Myc、Dax1、Rex1、Nac1 和Zfp281。这些研究表明，ESCs多能性调控的转录因子网络的复杂性。此外，还发现维持ESCs多能性的其他转录因子包括Sall4、Zfx、Ronin、Prdm14、foxp1、Foxo1、Zic3、Wdr5、Cdx2、Cbx7 和Chd7等[43~53]，它们也是维持ESCs多能性网络的成员，共同参与对ESCs多能性网络调控。Marson等[54]报道了ESCs特异表达的miRNA基因启动子受转录因子Oct4、Sox2、Nanog和Tcf3 共同调控，而一些miRNA又可以直接或间接地调控这些转录因子的表达。因此，ESCs多能性网络调控方式可能是通过核心转录因子与miRNA的相互作用来实现。

ESCC miRNA家族和Let-7 miRNA家族是调控ESCs增殖和分化的对立家族[55]。在hESCs和mESCs中，Oct4、Sox2 和Nanog分别促进ESCC的miR-302家族和miR-290 家族的表达[54]，来实施对ESCs多能性网络的调控，促进ESCs的增殖。miRNA和核心转录因子对ESCs多能性网络调控涉及 3 个重要的调控环路：(1) NR2F2、Oct4 和miR-302 家族间的调控环路；(2) c-Myc、E2F1 和miR-17~92 家族的调控环路；(3) Lin28 与Let-7 miRNA家族的调控环路[56]。它们是控制ESCs增殖和分化的关键点，如图 2 所示。

(1)NR2F2、Oct4 和miR-302 家族组成的调节环路：在维持hESCs自我更新时，高表达的Oct4 水平能促进miR-302 家族的转录，同时miR-302 家族又能下调Oct4 抑制因子NR2F2 的表达水平，促进Oct4 的表达，从而抑制hESCs分化。相反，NR2F2 表达量升高，可以直接下调Oct4 的表达，促进hESCs分化[57]。因此，转录因子Oct4 通过miR-302 家族抑制其抑制因子NR2F2 的表达，来保持自身高表达水平，最终促进维持ESCs的多能性。

(2)c-Myc、E2F1 和miR-17~92 家族的调控环路：c-Myc和n-Myc可以促进miR-17~92 家族[58]、miR-290家族[41]和miR-200 家族以及细胞周期转录因子E2F1的转录[59]，同时可以抑制促进ESCs分化的miR-15、miR-21、miR-29a、miR-143 和Let 7 miRNA家族的表达[29]，它们是ESCs维持多能性和体细胞重编程过程中的重要调节蛋白。c-Myc、E2F1 和miR-17~92 家族间的调控是促进ESCs细胞周期

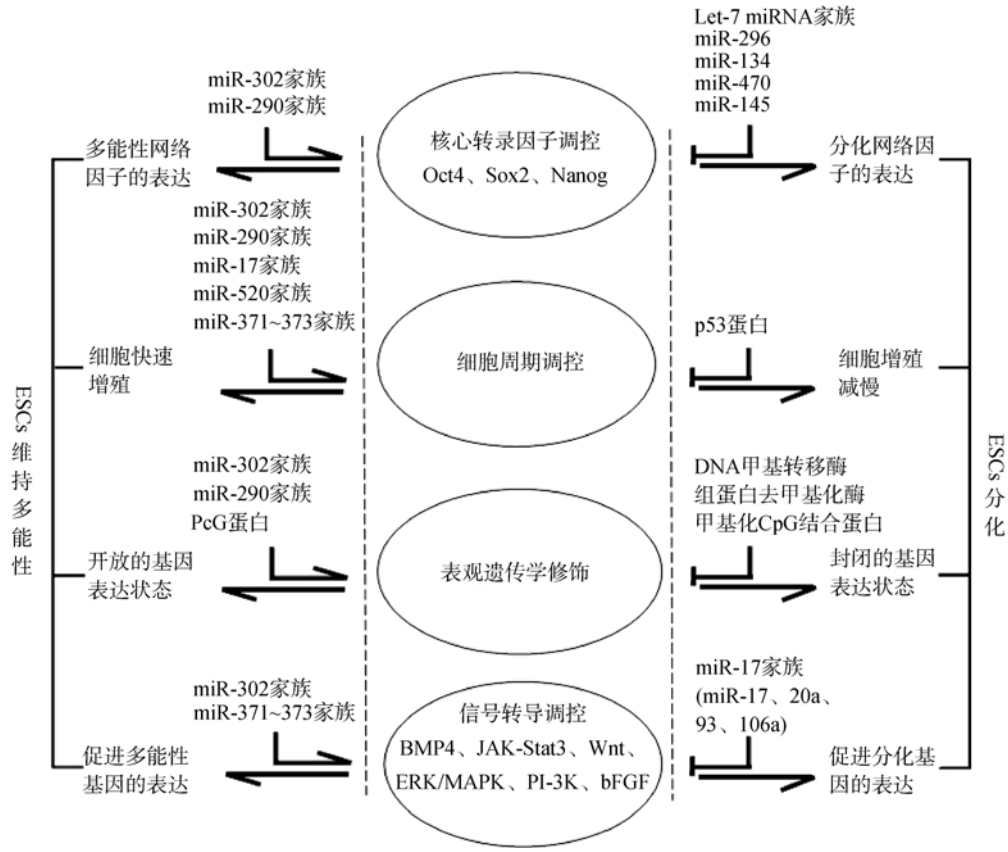

图 1　miRNA 对 ESCs 多能性网络的调控

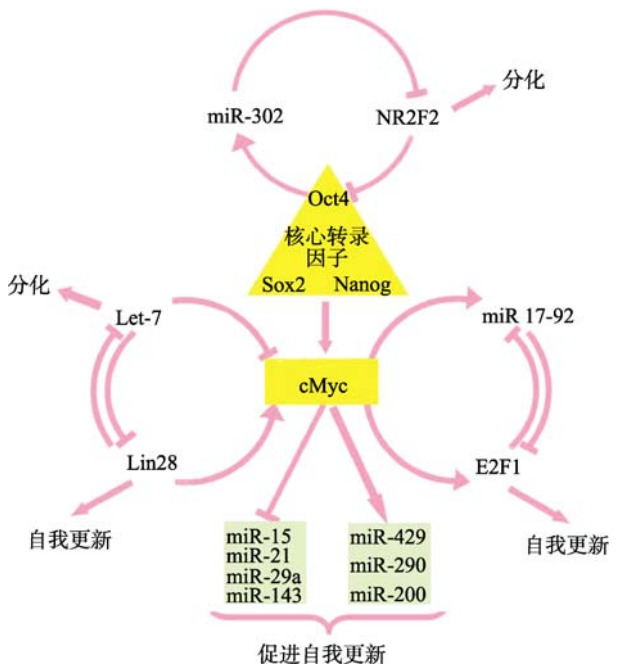

图 2　miRNA 和核心转录因子对 ESCs 多能性和分化的调控

从G_1/S期转换的重要开关[60]。c-Myc能促进miR-17~92 家族和E2F1 的转录，且E2F1 又能结合在miR-17~92 家族的启动子上，促进其转录。然而miR-17~92家族中的miR-17-5p和miR-20a却抑制了E2F1 的表达，形成严密控制细胞增殖的调控环路[61]。

(3)在ESCs分化过程中，Let-7 miRNA家族可以直接下调Lin28、c-Myc、n-Myc和Sall4 的表达，同时也导致ESCs多能性因子中的Oct4、Sox2、Nanog和Tcf3 的下调，促使ESCs快速分化[55]。然而在未分化的ESCs中，Lin28 不仅可以抑制Let-7 miRNA家族成熟，还可以抑制原生质网相关基因的表达[62]，是调控ESCs多能性的一个重要基因[63]。Lin28 和Let-7 miRNA家族相互作用成为ESCs增殖与分化的一个重要开关[64]。

可见，控制 ESCs 维持多能性的关键点是多能的转录因子通过 miRNA 下调其抑制因子或抑制分化的 miRNA 表达，从而阻止 ESCs 分化，促进了自我更新。

3.2　miRNA 对 ESCs 细胞周期的调控

细胞周期分为间期(G_1期、S期、G_2期)和分裂期(M期)两个阶段。ESCs的细胞周期中G_1期特别短，促

使其在体外能快速增殖[65]。ESCs能快速地从G_1期进入S期，这和miRNA调控是分不开的[66]，如图 3 所示。

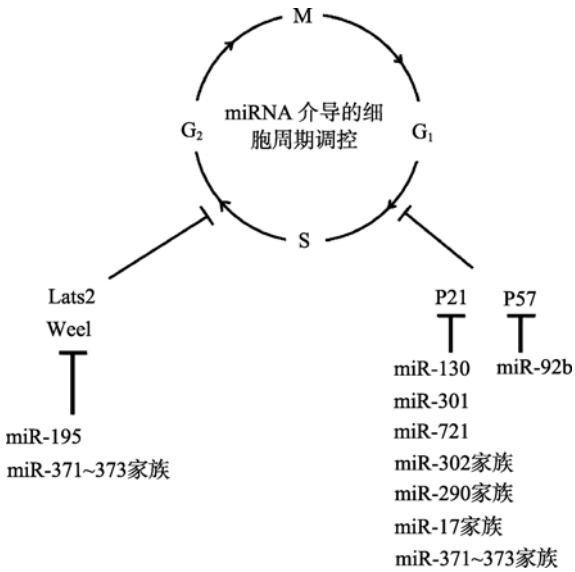

图 3 ESCs 细胞周期的 miRNA 调控

调控G_1/S期过渡的细胞周期复合体主要包括Cyclin D/Cdk4、Cyclin D/Cdk6 和Cyclin E/Cdk2[67]，但在mESCs中只发现Cyclin E/Cdk2 复合体存在[68]。ESCC miRNA家族通过下调CyclinE/Cdk2 复合体的抑制剂，促进了ESCs G_1/S期的快速转换[12]。CyclinE/Cdk2 复合体的抑制剂主要包括Cdkn1a(p21)、Cdkn1b(p27)、Rb1、Rbl1、Rbl2 和Lats2 等[69]。也有报道指出p21 蛋白的表达水平升高会导致ESCs分化[70]。

在hESCs中，miR-371~373 家族和miR-17 家族(miR-17、25 和 106)能直接下调p21 蛋白的表达水平; miR-92b可以下调p57 蛋白的表达水平; miR-17-5p和miR-20a又减少E2F1 的累积，促进细胞周期G_1期向S期过渡，促使ESCs的增殖[71~73]; miR-130、miR-301和miR-721 通过下调p16 和p21 激活Meox2 的表达水平，促进了ESCs的增殖[74]。另外, miR-371~373 家族和miR-195 分别下调Lats2 和Wee1 表达水平，促使细胞周期G_2/M期过渡，加快了ESCs的自我更新[75,76]。mESCs中miR-290 家族可以直接抑制p21、Lats2、Rbl2、Wee1 和Fbxl5 的表达，促进mESCs的增殖[12]。

在 hESCs 和 iPSCs 中，miR-302 家族对多能性调控起着非常重要的作用，但它可以下调Cyclin D1/D2、Cdk2 和Bmi1 的表达，抑制细胞恶性增殖，增加细胞的成活比例，促进体细胞的重编程，从而获得更加安全的iPSCs [77,78]。

miR-290 家族、miR-302 家族和miR-371~373 家族有一个共同种子序列“AAGUGCU”，miR-17 家族和miR-520 家族也有类似的种子序列，它们都能调控ESCs的细胞周期，促进ESCs的增殖[72]。因此，不同的miRNA只要有类似“AAGUGCU”种子序列都可能具有调控细胞周期的功能。

3.3 miRNA 对 ESCs 表观遗传学的调控

表观遗传学修饰是通过DNA甲基化、组蛋白修饰以及染色质重塑等对基因的表达与沉默进行调控[79]。ESCs保持着较为“开放”的染色质构象，这使它具有转变为任何类型细胞的潜能[80]。表观遗传学修饰在ESCs增殖与分化过程中发挥着重要作用，而miRNA参与调控该过程。

多梳家族蛋白(Polycomb group proteins, PcG蛋白)是一组通过染色质修饰调控靶基因转录的抑制子，可以沉默调控发育相关的基因，对维持ESCs的多能性具有积极作用[81,82]。在mESCs中, PcG蛋白的两个核心蛋白PRC1 和PRC2 共同调控 512 个靶基因，其中大部分是 3 个胚层以及胚外组织分化的重要转录因子[83]，同时，这些基因绝大部分也受到Oct4、Sox2 和Nanog的调控。这说明维持ESCs多能性抑制分化基因的表达不仅是通过PcG蛋白在表观遗传学上进行调控，同时还受到Oct4、Sox2 和Nanog在转录水平上的调控。

ESCC miRNA家族在表观遗传学调控上也同样起着重要的作用。miRNA通过调节表观遗传修饰的关键酶来实现对ESCs表观遗传学的调控。例如，miR-290 家族通过下调DNA甲基转移酶(Dnmt1、Dnmt3a和Dnmt3b)的激活因子Rbl2 的表达水平，促进ESCs的全基因组保持低甲基化的状态[84,85]，保证了多能性因子Oct4 等的转录，进而维持ESCs的多能性[86,87]。miR-302 家族在重编程过程中，下调了组蛋白去甲基化酶(Aof1 和Aof2)和甲基化CpG结合蛋白(Mecp1-p66 和Mecp2)以及甲基化相关蛋白的表达水平[88]，促进整个基因组的去甲基化，从而完成体细胞的重编程过程[89]。

3.4　miRNA 对 ESCs 信号通路的调控

ESCs维持多能性的关键信号通路主要包括JAK-Stat3(LIF)、TGFβ(BMP4)、ERK/MAPK、PI-3K和Wnt等通路[90]。mESCs和hESCs信号转导通路存在显著差异，JAK-Stat3(LIF)和TGFβ(BMP4)通路是mESCs维持多能性的关键通路，而hESCs维持多能性依赖bFGF和Activin/Nodal介导的信号通路[91]。这些信号通路通过对转录因子进行调控，最终激活多能性基因，促进ESCs的自我更新。miRNA对维持ESCs多能性的信号通路具有重要的调控作用。miR-302家族下调BMP的抑制剂Tob2、DaZAp2和Slain1，激活BMP信号通路促进hESCs的自我更新[92]。Wnt通路促进miR-371~373家族的表达，同时miR-371~373家族又可以激活Wnt通路，促进Cyclin-D1、c-Myc和c-Jun等上调表达，促使hESCs的自我更新[93]。miR-17家族成员(miR-17、20a、93和106a)具有相同的种子序列，它们的一个靶基因是Stat3，Stat3调控c-Myc的表达，而c-Myc又能促进miR-17家族转录[58]。这表明Stat3/miR-17家族成员之间的相互作用可以调控ESCs分化[94]。因此，同一个家族里面的miRNA成员或者同一个miRNA可能起着不同的调控作用。

4　miRNA 对 ESCs 分化和体细胞重编程的调控

ESCs分化和体细胞重编程是两个相反的过程。在体细胞重编程过程中，通过表达ESCC miRNA家族的一些成员，显著提高了重编程效率[30]，而一些ESCC miRNA家族也能调控体细胞获得多能性[13]。因此，促进ESCs维持多能性的核心因子或者miRNA家族都是提高体细胞重编程的重要因子，而直接或间接抑制核心转录因子或阻碍细胞周期运转以及促进表观遗传学封闭的蛋白或者miRNA都是体细胞重编程的障碍，它们在调控ESCs分化时起了重要的作用。如图 4 所示。

4.1　miRNA 对体细胞重编程过程调控

研究证实，多能性转录因子可以改变体细胞的命运，使其重编程为干细胞。小鼠和人的成纤维细胞分别转入转录因子Oct4、Sox2、c-Myc、Klf4或Oct4、Sox2、Nanog、Lin28，获得了iPSCs[3,4]。由于iPSCs和ESCs具有相同的性质和功能[96]，这为临床医学带来了新的曙光。

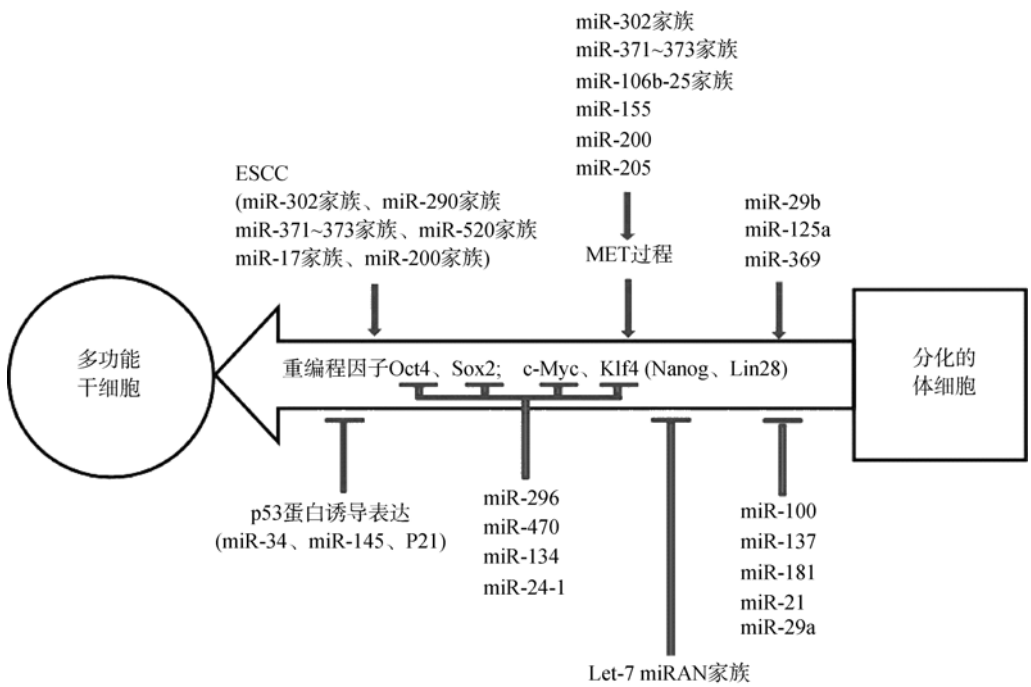

图 4　miRNA在体细胞重编程过程中的作用[95]

研究证实miRNA可以调控体细胞重编程[13]。p53 蛋白介导的细胞信号转导在调节细胞正常生命活动中起着重要作用，但它却是体细胞重编程过程的一个主要障碍[97,98]。p53 蛋白促进miR-34[99]、miR-145[100]、miR-192、miR-215[101]和p21 的表达，抑制细胞增殖，促进凋亡。其中miR-34 表达可显著降低iPS的效率[99]。miR-21 和miR-29a可以激活p53 和ERK1/2 信号通路，也明显降低了iPS效率[29]。而miR-125a下调p53 表达水平，提高了重编程效率[102]。受Sox2 调控表达，miR-29b可以下调Dnmt3a和Dnmt3b表达水平，促进体细胞重编程[103]。miR-302家族、miR-106b~25 家族、miR-371~373 家族和miR-155 等可以直接下调TGFβR2 表达水平促进间质-上皮细胞转化(Mesenchymal-to-epithelial transition, MET)过程，加速体细胞重编程[104,105]。而miR-200 家族和miR-205 通过下调钙粘蛋白(E-cadherin)的抑制剂Zeb1/2 的表达，也促进了MET过程，提高了体细胞重编程效率[106,107]。miR-24-1 可以直接下调c-Myc、E2F2 和Smad3 的表达[108]，在抑制miR-24-1 条件下可以不转入c-Myc和Klf4，就能获得iPSCs[109]；miR200c、miR302 和miR369 可以有效促进人和小鼠体细胞重编程[110]。可见，miRNA对ESCs多能性网络调控的重要作用。

4.2 miRNA 对 ESCs 分化的调控

ESCs的分化过程和多能性的维持过程是两个对立的调控过程。ESCs分化时要抑制多能性基因的表达，促进 3 个胚层基因和组织特异的基因以及相关miRNA家族的表达[111]。

Let-7 miRNA家族是由 12 个(7a-1、7a-2、7a-3、7b、7c、7d、7e、7f-1、7f-2、7g、7i和miR-98)成员组成，位于 8 条染色体上[112]。它们广泛表达于分化的组织[112]，是促进ESCs分化的重要miRNA家族[113,114]，它可以直接下调Oct4、Lin28、c-Myc、Hmga2、K-Ras和Cyclin D1 等多能相关因子的表达，促进ESCs分化。例如let-7e调控Wnt信号通路促进mESCs分化[115]以及let-7 和miR-18 家族共同调控早期胚胎生殖层形成[116]等。有研究报道miR-134、miR-296、miR-470、miR-145、miR-21、miR-200c、miR-203 和miR-183 等在mESCs分化过程中表达上调，它们的靶基因主要是Oct4、Sox2、Nanog或Klf4[117~120]，从而促进ESCs分化。miR-34a、miR-100和miR-137 直接下调了Sirt1、Smarca5 和Jarid1b的表达，促进ESCs的分化[121]。miR-125 和miR-181 通过调控多梳蛋白同源基因Cbx 7 的表达，促进mESCs分化[52]。因此，ESCs进入分化过程中，分化因子和分化相关的miRNA首先下调核心多能性因子的表达水平，关闭ESCs的多能性调控网络，进入细胞分化的调控网路，促进细胞分化。

5 结 语

ESCs 的细胞周期和表观遗传学是调控 ESCs 维持多能性的主要方面。调控细胞周期，可以促进ESCs 自我更新；表观遗传学调控，促使 ESCs 保持一个更为开放的状态，具有多向分化的潜能，从而维持 ESCs 多能性。miRNA 参与生命活动的调控，掌控着细胞生理状态，是后基因组学研究的主要内容，也是 ESCs 调控网络中的主要部分。ESCs 多能性调控网络中的转录因子网络、功能基因组数据网络和miRNA 调控网络的建立和完善，促进了干细胞技术的发展；ESCs 分化网络的建立丰富了发育生物学和再生医学等学科，为细胞定向分化和转分化技术提供理论支持，也为癌症发病机制和防治等提供理论依据，同时为临床医学提供治疗疾病的方法。ESCs内有很多成熟的 miRNA 存在，而且每个 miRNA 都有很多的靶基因，同一家族 miRNA 或者某一 miRNA 调控功能也可能存在相反的现象等问题。因此，未来还需要继续深入研究 miRNA 对 ESCs 的调控作用。

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•综合信息•

2013 年 35 卷第 10 期《遗传》封面说明

2 型糖尿病(Type 2 diabetes mellitus, T2DM)是由于遗传与环境因素共同作用而引起葡萄糖代谢紊乱的疾病。环境因素对 T2DM 的致病机理极有可能是通过影响表观遗传修饰, 从而导致 T2DM 的发生及发展。表观遗传学修饰是不涉及基因组的碱基序列改变, 却能导致可遗传性的表型变异。其中 DNA 甲基化是表观遗传修饰一种主要的修饰方式。目前 DNA 甲基化修饰的研究已发现环境因素可以通过影响 DNA 甲基化修饰, 继而显著地增加 T2DM 的患病风险。T2DM 的多种环境因素与 DNA 甲基化修饰有显著关联。这些环境因素包括饮食、肥胖、体育运动、工作压力、年龄的增加以及性别差异等。另外, T2DM 环境因素相关基因的 DNA 甲基化有明显的组织特异性。目前, T2DM 环境相关基因的 DNA 甲基化修饰研究已在人及动物的不同组织中取得进展, 这些主要是胰岛素作用的靶器官如胰岛、骨骼肌、肝脏、脂肪以及外周血等组织。详见本期第 1143~1152 页汤琳琳, 刘琼, 步世忠, 徐雷艇, 王钦文, 麦一峰, 段世伟的文章“2 型糖尿病环境因素与 DNA 甲基化的研究进展”一文。

(汤琳琳, 段世伟)