·文献综述·

微小 RNA 网络控制及其在骨关节炎中的分子机制研究进展

王欢[1] 王庆甫[2△] 张栋[1] 王伟利[1] 丁昊彬[1]
殷岳杉[2] 郭玉茹[1] 甘稳[1] 徐铭康[1] 杜汪洋[1]

[关键词] 骨关节炎;微小 RNA;toll 样受体;机制
[中图分类号] R684.3 [文献标志码] A [文章编号]1005-0205(2017)03-0077-06

骨关节炎(Osteoarthritis,OA)是最常见的退行性关节疾病,伴随着疼痛、肿胀和功能障碍。OA 以其高发病率与高致残率特征,成为当前世界各国共同关注的公共健康问题。作为一种严重危害着中老年人健康并有逐年年轻化趋势的疾病,面对我国逐步进入老龄化社会的国情,使对其预防和治疗的需求显得更加的迫切。尽管如此,由于缺乏大规模系统的分子学研究,限制了对 OA 病理生理学机制的理解和识别药物靶点的开发,临床治疗也往往不能令人满意。最近,使用基因网络、表观遗传及微小 RNA(microRNA,miRNA,miR)等探寻 OA 病理生理学新靶点的方法被越来越广泛的发现和应用,miRNA 作为一种新兴的靶点在调节细胞功能中的重要性也被越来越清楚的揭示。研究表明,OA 是一种受 miRNA 异常调控的代谢性疾病[1],miRNA 在其诊断中的重要性和治疗中的潜力,很可能为 OA 的诊疗提供新的思路和手段。本篇综述总结了 miRNA 的生物学合成和它们在转录后调控中的作用机制及其在决定 OA 复杂的基因表达模式中发挥重要作用的现有证据。

1 miRNA 概述及生物学特征

最新的研究表明,对非编码 RNA 基因学改变的研究有助于推进对人类疾病发病机制的探寻[2]。近年来,越来越多的实验证据表明,一类叫做 miRNA 的新型的内源性非编码小分子核糖核酸在细胞基因调控网络中发挥着重要的作用。这种长约 20～25 个核苷酸(nt)的 RNA 分子能识别靶 mRNA 上的 3'非翻译区(Untranslated Regions,UTR)并对其部分互补的位点进行结合绑定,通过对 mRNA 的降解或阻碍翻译,调控靶基因的表达,从而发挥其生物功能[3,4]。作为一种重要的基因调节因子,随着第一个 miRNA 于 1993 年被 Ambros[5,6]在哈佛大学发现之后,众多研究陆续发现了各种 miRNA,其专属基因序列数据库"miRBase"迄今为止已收录了超过 15000 余种预测的 miRNA 广泛存在于包括植物、动物和病毒等几乎所有的多细胞生物中,其中在人类表达的 miRNA 也有超 900 种之多[7]。生物信息学预测,人体内多达三分之一编码蛋白质的 mRNA 可能受 miRNA 的调控[8-11],它们主要参与如脂质代谢、器官发育等细胞增殖、分化和凋亡的过程,对维持人体正常机能起着至关重要的调控作用[12]。miRNA 的调节失衡将会严重影响细胞靶基因的表达而导致疾病,几乎所有人类疾病的病理过程都可以见到 miRNA 的身影[13-15]。

2 miRNA 的加工和转录后调控

具有基因沉默功能的成熟 miRNA 的生成是一个多步骤的过程,从细胞核中开始,在细胞质中完成。首先,miRNA 在细胞核内通过 RNA 聚合酶Ⅱ(RNA Polymerase Ⅱ)、RNA 聚合酶Ⅲ(RNA Polymerase Ⅲ)的作用转录形成一种称为初级 miRNA(pri-miRNA)的长的 RNA 前体,它含有一个或多个带着部分互补序列发夹外观的茎环结构,孕育着至少一个未来的功能性的 miRNA[16]。随后,pri-miRNA 在细胞核内经 miRNA 处理器———一种由 Drosha(高度保守的核糖核苷酸酶Ⅲ)联合 DGCR8(迪格奥尔格综合征临界区基因 8)组成的蛋白质络合物切除其侧翼序列,形成一个 70 nt 左右长度的较短的发夹/茎环结构的前体 miRNA(pre-miRNA)[17]。

除上述 miRNA 转录的经典途径之外,也有报道称部分 miRNA 还可通过其他的途径合成:以 miR-140 为代表的 miRNA 不经独立基因而由 mRNA 的内含子转录[18]、[19];致癌基因和有效的发育调节器 miR-

基金项目:国家自然科学基金面上项目(81373662)
北京中医药大学自主选题项目(2015-JYBZZ-XS-202)
[1] 北京中医药大学
[2] 北京中医药大学第三附属医院
△通信作者 E-mail:qingpu-wang@sohu.com

17-92 集群则被发现存在于编码不只一个 miRNA 基因的多顺反子性单元[20,21]；还有小部分 miRNA 直接通过剪接体由 pri-miRNA 加工的 mirtrons 途径合成，而不是依赖于含 Drosha 酶的 miRNA 处理器也被报道[22,23]。

经过上述在细胞核中发生的转录过程，pre-miRNA 被一种有转运作用的输出蛋白 5（Exportin-5）识别并从细胞核转运至细胞质中[24]。随后，另一种叫做 Dicer 酶的核糖核酸酶Ⅲ将其进行第二次剪切，形成长度 22～25 nt 左右的不完全匹配的双链 miRNA 二聚体，其中包含一条可继续参与反应的成熟的引导链（Guide Strand）和一条与之相对应的随后可降解的过客链（Passenger Strand）[25]。此时由于 RNA 解旋酶的参与，上述两条 miRNA 链被解聚，打开的引导 miRNA 链由于带有热力学效应的不稳定的磷酸化 5'UTR 和碱基凸出的 3'UTR，可载入 RNA 诱导的沉默复合物（RNA-induced Silencing Complex，RISC），并与其主要组件蛋白 Argonaute（Ago）结合，形成 miRNA-RISC 复合体[26,27]。它能识别并结合靶 mRNA 上的 3'UTR 完全或不完全配对结合位点，引导 RISC 降解 mRNA 或阻碍其翻译过程，介导转录后的基因沉默，调节目标转录物的蛋白质生成，进而达到调节基因表达的目的[28]。其结合的特异性主要由引导链上的第 2～8 nt 的种子序列（Seed Sequence）决定[29,30]，不同组合的 miRNA 表达在不同的细胞类型中，能协同调节细胞特异性靶基因，但 miRNA 详细的协同调节机制至今尚未被完全阐明[31]。

3 miRNA 与 OA 的相关性研究

无论是哪一种加工方法和转录后的调控途径，一系列现代研究表明，在不同类群中显示出相当保守而独立进化的 miRNA 确实是通过沉默靶 mRNA 在细胞基因调控网络中发挥着重要的作用[32,33]，其显示出的组织特异性和时序特异性表达模式与一些人类疾病有关[34,35]，其中，OA 是一种受 miRNA 异常影响的复杂的多因素疾病[36,37]，然而目前，人们对 miRNA 在 OA 中的表达和功能还知之甚少。

3.1 miRNA 在软骨中的调控作用

以往的研究普遍将原发性 OA 的发病机理主要归咎于关节软骨的生物力学改变[38-40]。近年来，越来越多进行在软骨细胞、细胞外基质和趋化因子等与 OA 关节软骨相关的研究证明了 miRNA 在软骨功能中的重要性[41]。早在 2008 年，Kobayshi 等[42]就通过对 miRNA 合成过程中的关键剪切酶 Dicer 敲除小鼠展开的工作，发现该基因缺失小鼠的软骨细胞增殖与分化缓慢，表现出软骨生长发育的严重缺陷，证明 miRNA 在正常软骨细胞发育和功能中不可或缺的作用。2009 年，Miyaki 等[43]利用高通量的微阵列基因芯片技术发现 miRNA 在人类软骨中的高特异性表达，并用中通量的实时定量 PCR 法对其检测结果进行了验证。随后的一年，他们利用 miR-140 基因敲除小鼠成功模拟出 OA 退变的特征[44]，进一步证实了 miR-140 在维持关节软骨正常代谢中发挥的保护性作用，并且，此种作用很可能通过对聚蛋白多糖酶（A Disintegrin and Metalloproteinase with Thrombospondin Motifs 5，ADAMTS-5）的调控而发挥[45,46]。除此之外，miRNA 还可以调控关节软骨基质金属蛋白酶（Matrix-metallo Proteinase，MMP）等基因的表达[47]，如 miR-27 就是一种典型的通过绑定 MMP-13 的 mRNA 序列上 3'UTR 从而抑制 MMP-13 表达的 miRNA[48]。还有研究数据显示，特定的 miRNA 和与其相匹配的靶向蛋白质的表达密切相关，如 miR-22 可以调节 PPARA 和 BMP7 的表达，抑制性阻断了软骨细胞炎症和分解代谢的变化[49]；miR-18 靶向干预结缔组织生长因子（Connective Tissue Growth Factor，CTGF/CCN2）的 mRNA 序列上 3′utr 影响软骨细胞的分化[50]。

3.2 miRNA 在滑膜炎症中的调控作用

然而，OA 综合征的病变部位并不仅仅局限于关节软骨，还要考虑多组织的共同作用[51,52]。分子学与临床数据整合表明，OA 是一种影响关节滑膜的最常见的结缔组织疾病[53]。滑膜作为控制分子出入关节间隙的交通要道，维持着滑液成分，是分解代谢物的主要来源，在 OA 的发病和进程中扮演着重要的角色[54,55]。聚集在 OA 滑膜组织巨噬细胞、B 细胞和 T 细胞等单核细胞，使诱发软骨保护性因子浓度的下调和基质降解因子产量的增加的滑膜炎症成为软骨退化最可能的驱动因素[56]。各种 miRNA 在滑膜组织炎症反应中的差异性表达和生物学作用已有部分记载。如在滑膜炎症反应中，OA 滑膜成纤维细胞中的 miR-146a 有明显的表达[57]，分析 miR-146a 可能响应一种微生物成分和促炎性细胞因子的感应模式[58,59]。的确，有研究从人类单核细胞 200 个 miRNA 的表达中发现除 miR-146a 以外，miR-132，miR-155 以及 miR-9，miR-21 和 miR-147 也是致炎因子脂多糖（Lipopolysaccharide，LPS）诱导的体外炎症反应的应答基因，表现为迅速、暂时性上调的 miRNA 表达[60-62]。而 miR-124 在 OA 滑膜成纤维细胞中的表达则为下调，且研究表明，过表达的 miR-124 对滑膜成纤维细胞的增殖起到了明显抑制作用[63]。

3.3 miRNA 在 Toll 样受体信号通路中的调控作用

以上研究表明，miRNA 参与了主要涉及软骨破坏及滑膜炎症等 OA 发病过程中的病理变化。最近的

报告称，如关节肿胀或滑膜积液等炎症过程的临床症状是原发性 OA 最常见临床表现[64]，致使一些 miRNA 参与调控的炎症通路受到了极大的关注[65-67]。其中，由于哺乳动物固有免疫应答在激发 OA 滑膜炎症反应中起到了关键的作用[68,69]，其激活受到复杂机制的严格调控，而 Toll 样受体（Toll-Like Receptors，TLRs）信号通路正是在体内维持机体正常免疫功能的重要通道[70,71]。随着对 TLRs 信号通路调节机制研究的逐渐深入，科研人员逐渐发现 miRNA 对固有免疫调控失调可以诱导或加速相关炎症疾病进程，将研究焦点越来越多的放在 miRNA 与 TLRs 信号通路的相互作用及机理的探索上[72-78]。miRNA 能在几乎所有水平上对 TLRs 信号通路进行调控：有些 miRNA 可直接调节 TLRs，如 miR-26a 在降植烷诱导的大鼠关节炎巨噬细胞中可以调控 TLR3 的表达，而 miR-21，miR-105，miR-143，miR-146a 均可以调控 TLR2 的表达[79,80]；miRNA 还可以调节 TLRs 信号通路上的相关蛋白，如 miR-146 家族中的 miR-146a 以通路上的 IRAK-1 和 TRAF-6 基因为靶点，而 miR-146b 则还能调节 MyD88、TRAF6 等基因[81-84]；研究表明，部分 miRNA 还能调节通路中的转录因子（Transcription Factors），如 miR-9，miR-210 和 miR-329 均可对 NF-kB——一种重要的核转录因子进行调控，以抑制细胞的炎症反应[85,86]。除直接干预 TLRs 及其信号通路上的相关蛋白和转录因子的基因表达之外，miRNAs 还可通过调节通路相关分子来完成对 TLRs 通路的监管，研究指出，miR-132 可以调节乙酰胆碱酯酶（Acetylcholine Esterase，AchE）基因的表达，从而对 TLRs 通路起到抑制作用[87-89]；miR-21 则通过对抑癌基因程序性细胞死亡蛋白 4（Programmed Cell Death Protein 4，PDCD4）的调控，减轻与 TLRs 通路相关的炎症反应[90-92]。

4 miRNA 网络控制及其在 OA 诊疗中的潜力

多种 miRNA 广泛参与到调节 OA 的各个过程中去，连同相关蛋白等基因在不同作用层次联合构成了转录后水平的复杂调控网络，共同维持着软骨与滑膜等的各项机能[93]，研究这个 miRNA 与蛋白质组学、生物信息学和临床数据等综合形成的包含着彼此潜在关联的网络系统，有助于增进对 OA 这种多因子疾病病理机制的理解，进而推动新治疗方法的发展[94-96]。国内外研究提示，基因网络方法为阐明如 OA 等受 miRNA 异常影响的疾病提供了新的见解，差异表达的 miRNA 既可作为 OA 诊断的生物标志物，还可作为治疗 OA 的作用靶点[97]。Dimitrios Iliopoulos 等[98]在 OA 患者的软骨与正常软骨相比较综合分析的研究中，鉴定出 16 个差异性表达的 miRNA，并用反相蛋白阵列的方法检测到上述组织中差异表达的 76 个相关蛋白。琼斯和他的同事们也做了大量关于差异性表达的 miRNA 在人类 OA 软骨、骨和正常组织间的对比研究[99]。越来越多的证据表明，对 miRNA 的管制可以有效调控 OA，一些研究试图通过使用 miRNA 的模拟剂 mimics 和抑制剂 inhibitors 调节 miRNA 的表达和活动以确定其在具体的分解代谢途径中的作用[100,101]。相对于临床现有的应用非甾体抗炎药（Non-steroidal Anti-inflammatory Drugs，NSAIDs）、环氧化酶-2（cyclo-oxygenase-2，COX-2）抑制剂、类固醇等药物缓解 OA 症状的程度有限，也不能有效的扭转关节软骨等的损耗，而且其引起的包括胃肠道反应、心血管疾病等副作用等问题[102-104]，对 miRNA 相关制剂的研究显示出了良好的应用前景。

5 展望

OA 是一种全身性的疾病，诸多因素与它的发病和进程一一对应的存在。在过去的十年里，随着基因网络、表观遗传等方法的利用，miRNA 作为 OA 治疗干预的新的潜在性靶点已经引起了广泛的关注，它们参与到 OA 病理生理学，调节与 OA 相关的通路、蛋白等的水平，进而控制关节组织的破坏并刺激修复。因此，结合现代医学研究，进一步深化对 miRNA 参与 OA 发生发展的分子机制的理解，寻找特异性生物标志物，探究 OA 可能的发病机制和潜在的分子靶点是探寻 OA 诊疗途径的新方法，也是维护国民健康与提高生活质量，减轻我国社会经济发展负担的迫切要求。

参考文献

[1] Akhtar N, Rasheed Z, Ramamurthy S, et al. MicroRNA-27b regulates the expression of matrix metalloproteinase 13 in human osteoarthritis chondrocytes[J]. Arthritis & Rheum, 2010, 62(5): 1361-1371.

[2] Zhang ZJ. Artificial trans-acting small interfering RNA: a tool for plant biology study and crop improvements[J]. Planta, 2014, 239(6): 1139-1146.

[3] Smalheiser NR. Regulation of mammalian microRNA processing and function by cellular signaling and subcellular localization[J]. Biochim Biophys Acta, 2008, 1779(11): 678-681.

[4] Figliuzzi M, Marinari E, Demartino A. MicroRNAs as a Selective Channel of Communication between Competing RNAs: a Steady-State Theory[J]. Biophys J, 2013, 104(5): 1203-1213.

[5] Lee RC, Feinbaum RL, Ambros V. The C. elegans heterochronic gene lin-4 encodes small RNAs with antisense complementarity to lin-14[J]. Cell, 1993, 75(5): 843 - 854.

[6] Wightman B, Ha I, Ruvkun G. Posttranscriptional regulation of the heterochronic gene lin-14 by lin-4 mediates temporal pattern formation in C. elegans[J]. Cell, 1993, 75(5): 855 - 862.

[7] Schmitz U, Vearasilp K. MiRBase[M]. New York: Springer, 2013.
[8] Lewis BP, Burge CB, Bartel DP. Conserved seed pairing, often flanked by adenosines, indicates that thousands of human genes are microRNA targets[J]. Cell, 2005, 120(1): 15-20.
[9] Jayaswal V, Lutherborrow M, Ma DD, et al. Identification of microRNA-mRNA modules using microarry data[J]. BMC Genomics, 2011, 12: 138.
[10] Kozomara A, Griffiths-Jones S. miRBase: integrating microRNA annotation and deep- sequencing date[J]. Nacleic Acids Res, 2011, 39: 152-157.
[11] Olena AF, Patton JG. Genomic organization of microRNAs[J]. J Cell Physiol, 2010, 3: 540 -545.
[12] Zheng Y, Zhang J, Gong D Q. Research Progress on miRNA of Poultry[J]. China Animal Husbandry & Veterinary Medicine, 2014, 41(1): 76-79.
[13] Mendell JT, Olson EN. MicroRNAs in stress signaling and human disease[J]. Cell 2012, 148(6): 1172-1187.
[14] Esteller M. Non-coding RNAs in human disease[J]. Net Rev Genet, 2011, 12: 861-874.
[15] Nojima M, Matsui T, Tamori A, et al. Global, cancer-specific microRNA cluster hypomethylation was functionally associated with the development of non-B non-C hepatocellular carcinoma[J]. Molecular Cancer, 2016, 15(1): 31.
[16] Voglova K, Bezakova J, Herichova I. Progress in micro RNA focused research in endocrinology[J]. Endocrine Regulations, 2016, 50(2): 83-105.
[17] Juanchich A, Bardou P, Rué O, et al. Characterization of an extensive rainbow trout miRNA transcriptome by next generation sequencing[J]. BMC Genomics, 2016, 17(1): 1-12.
[18] Voglova K, Bezakova J, Herichova I. Progress in micro RNA focused research in endocrinology[J]. Endocrine Regulations, 2016, 50(2): 83-105.
[19] Luo J W, Wang X, Yang Y, et al. Role of micro-RNA (miRNA) in pathogenesis of glioblastoma. [J]. Eur Rev Med Pharmacol Sci, 2015, 19(9): 1630-1639.
[20] Concepcion CP, Bonetti C, Ventura A. The microRNA-17-92 family of microRNA clusters in development and disease. [J]. Cancer Journal, 2011, 18(18): 262-267.
[21] Piubelli C, Meraviglia V, Pompilio G, et al. microRNAs and Cardiac Cell Fate. [J]. Cells, 2013, 3(3): 802-823.
[22] Lin SL, Ying SY. Gene Silencing In Vitro and In Vivo Using Intronic MicroRNAs[J]. Methods Mol Biol, 2015, 342: 295-312.
[23] Curtis HJ, Sibley CR, Wood MJA. Mirtrons, an emerging class of atypical miRNA[J]. Wiley Interdisciplinary Reviews-rna, 2012, 3(5): 617-32.
[24] Pisarello MJL, Howard BN, Trussoni CE, et al. Abstract 192: Exportin-5 downregulation induces miRNA dysregulation and HDAC6 overexpression in cholangiocarcinoma [J]. Cancer Research, 2015, 75(15 Supplement): 192-192.
[25] Srivastava S, Yun Z, Kudapa H, et al. High throughput sequencing of small RNA component of leaves and inflorescence revealed conserved and novel miRNAs as well as phasiRNA loci in chickpea[J]. Plant Sci, 2015, 235: 46-57.
[26] Roberts TC. The microRNA Machinery[J]. Advances in Experimental Medicine & Biology, 2015, 887: 15-30.
[27] Yoon JH, Kim J, Gorospe M. Long noncoding RNA turnover[J]. Biochimie, 2015, 117: 15-21.
[28] Djuranovic S, Nahvi A, Green R. A parsimonious model for gene by miRNAs[J]. Science, 2011, 331(6017): 550-553.
[29] Green D, Fraser WD, Dalmay T. Transfer RNA-derived small RNAs in the cancer transcriptome[J]. Pflügers Arch, 2016, 468(6): 1-7.
[30] He J, Hua J, Ding N, et al. Modulation of microRNAs by ionizing radiation in human gastric cancer. [J]. Oncology Reports, 2014, 32(2): 318-327.
[31] Bosia C, Pagnani A, Zecchina R. Modelling Competing Endogenous RNA Networks[J]. Plos One, 2013, 8(6): e66609.
[32] Valinezhad Orang A, Safaralizadeh R, Kazemzadeh-Bavili M. Mechanisms of miRNA-Mediated Gene Regulation from Common Downregulation to mRNA-Specific Upregulation[J]. Int J Genomics, 2014, 970607.
[33] Harrison A, Shanahan H. An Overview of Gene Regulation[M]// Approaches in Integrative Bioinformatics. Springer Berlin Heidelberg, 2014: 21-69.
[34] Ludwig N, Leidinger P, Becker K, et al. Distribution of miRNA expression across human tissues. [J]. Nucleic Acids Research, 2016, 44(8): 3865-3877.
[35] Rager JE, Moeller BC, Miller SK, et al. Formaldehyde-associated changes in microRNAs: tissue and temporal specificity in the rat nose, white blood cells, and bone marrow. [J]. Toxicol Sci, 2014, 138(1): 36-46.
[36] Yu XM, Meng HY, Yuan XL, et al. MicroRNAs' Involvement in Osteoarthritis and the Prospects for Treatments. [J]. Evidbased Complement Alternat Med, 2015, 236179.
[37] Barter MJ, Young DA. Epigenetic Mechanisms and Non-coding RNAs in Osteoarthritis[J]. Curr Rheumatol Rep, 2013, 15(9): 1-9.
[38] Jackson MT, Smith MM, Smith SM, et al. Activation of cartilage matrix metalloproteinases by activated protein C [J]. Arthritis Rheum, 2009, 60(3): 780 - 791.
[39] Lippiello L, Hall D, Mankin HJ. Collagen synthesis in normal and osteoarthritic human cartilage[J]. Clin Invest, 1977, 59(4): 593 - 600.
[40] Eyre DR, McDevitt CA, Billingham ME, et al. Biosynthesis of collagen and other matrix proteins by articular cartilage in experimental osteoarthrosis[J]. Biochem, 1980, 188(3): 823 - 837.
[41] Mirzamohammadi F, Papaioannou G, Kobayashi T. microRNAs in Cartilage Development, Homeostasis, and Disease[J]. Current Osteoporosis Reports, 2014, 12(4): 410-419.
[42] Kobayashi T, Lu J, Cobb BS, et al. Dicer-dependent pathways regulate chondrocyte proliferation and differentiation[J]. Proc Natl Acad Sci USA, 2008, 105: 1949-1954.
[43] Miyaki S, Nakasa T, Otsuki S, et al. MicroRNA-140 is expressed in differentiated human articular chondrocytes and modulates interleukin-1 responses[J]. Arthritis Rheum,

2009,60(9):2723-2730.
[44] Miyaki S,Sato T,Inoue A,et al. MicroRNA-140 plays dual roles in both cartilage development and homeostasis [J]. Gene Dev,2010,21:1173-1185.
[45] Plaas A,Osborn B,Yoshihara Y,et al. Aggrecanolysis in human osteoarthritis: confocal localization and biochemical characterization of ADAMTS5 - hyaluronan complexes in articular cartilages[J]. Osteoarthritis & Cartilage,2007,15(7):719-734.
[46] Le LTT,Swingler TE,Crowe N,et al. The microRNA-29 family in cartilage homeostasis and osteoarthritis [J]. Journal of Molecular Medicine,2015,94:1-14.
[47] Liang ZJ,Zhuang H,Wang GX,et al. MiRNA-140 is a negative feedback regulator of MMP-13 in IL-1β-stimulated human articular chondrocyte C28/12 cells[J]. Inflamm Res,2012,61(5):503-509.
[48] Akhtar N,Rasheed Z,Ramamurthy S,et al. MicroRNA-27b regulates the expression of matrix metalloproteinase 13 in human osteoarthritis chondrocytes[J]. Arthritis & Rheumatism,2010,62(5):1361 - 1371.
[49] Min Z,Zhang R,Yao J,et al. MicroRNAs associated with osteoarthritis differently expressed in bone matrix gelatin (BMG) rat model. [J]. Int J Clin Exp Med,2015,8(1):1009-1017.
[50] Ohgawara T,Kubota S,Kawaki H,et al. Regulation of chondrocytic phenotype by micro RNA 18a: Involvement of Ccn2/Ctgf,as a major target gene[J]. FEBS Letters,2009,583(6):1006-1010.
[51] Dieppe P. Developments in osteoarthritis. [J]. Rheumatology,2011,50(50):245-247.
[52] Poulet B,Staines KA. New developments in osteoarthritis and cartilage biology[J]. Curr Opin Pharmacol,2016,28:8-13.
[53] Eckstein F,Graverand M P H L. Plain Radiography or Magnetic Resonance Imaging (MRI) - Which are Better in Assessing Outcome in Summary of a Debate Held at the World Congress of Osteoarthritis 2014[J]. Semin Arthritis Rheum,2015,45(3):251-256.
[54] Suzuki S,Muneta T,Tsuji K. et al. Properties and usefulness of aggregates of synovial mesenchymal stem cells as a source for cartilage regeneration[J]. Arthritis Res Ther,2012,14(3):R136.
[55] Yu M,Ye WB,Zhong ZM,et al. Effect of Advanced Oxidation Protein Products on Articular Cartilage and Synovium in a Rabbit Osteoarthritis Model[J]. Orthop Surg,2015,7(2):161-167.
[56] Karczewski J,Dobrowolska A,Rychlewskahańczewska A,et al. New insights into the role of T cells in pathogenesis of psoriasis and psoriatic arthritis. [J]. Autoimmunity,2016,49(7):435-450.
[57] Takada S,Asahara H. Current strategies for microRNA research[J]. Modern Rheumatology,2012,22(5):645-653.
[58] Chan EKL,Ceribelli A,Satoh M. MicroRNA-146a in autoimmunity and innate immune responses. [J]. Ann Rheum Dis,2013,72(2):90-95.
[59] Ordas A,Kanwal Z,Lindenberg V,et al. MicroRNA-146 function in the innate immune transcriptome response of zebrafish embryos to Salmonella typhimurium,infection [J]. BMCC Genomics,2013,14:496.
[60] Contreras J,Rao DS. MicroRNAs in inflammation and immune responses[J]. Leukemia Official Journal of the Leukemia Society of America Leukemia Research Fund U K,2012,26(3):404-413.
[61] Anglicheau D,Muthukumar T,Suthanthiran M. MicroRNAs: Small RNAs with Big Effects[J]. Transplantation,2010,90(2):105-112.
[62] Li M,Shao H,Zhang X,et al. Hesperidin Alleviates Lipopolysaccharide-Induced Neuroinflammation in Mice by Promoting the miRNA-132 Pathway. [J]. Inflammation,2016,39(5):1681-1689.
[63] Nakamachi Y,Kawano S,Takenokuchi M,et al. MicroRNA-124a is a key regulator of proliferation and monocyte chemoattractant protein 1 secretion in fibroblastlike synoviocytes from patients with rheumatoid arthritis[J]. Arthritis Rheum,2009,60(5):1294-1304.
[64] Pan X,Huang L,Chen J,et al. Analysis of synovial fluid in knee joint of osteoarthritis:5 proteome patterns of joint inflammation based on matrix-assisted laser desorption/ionization time-of-flight mass spectrometry[J]. Int Orthop,2012,36(1):57-64.
[65] Zhang C,Shu L,Kong AT. MicroRNAs: new Players in Cancer Prevention Targeting Nrf2,Oxidative Stress and Inflammatory Pathways[J]. Curr Pharmacol Rep,2015,1(1):21-30.
[66] Sharma AR,Sharma G,Lee SS,et al. miRNA-Regulated Key Components of Cytokine Signaling Pathways and Inflammation in Rheumatoid Arthritis[J]. Med Res Rev,2016,36(3):425-439.
[67] Singh RP,Massachi I,Manickavel S,et al. The role of miRNA in inflammation and autoimmunity[J]. Autoimmunity Reviews,2013,12(12):1160-1165.
[68] Fahy N,Farrell E,Ritter T,et al. Immune modulation to improve tissue engineering outcomes for cartilage repair in the osteoarthritic joint[J]. Tissue Engineering Part B Reviews,2014,21(1):55-66.
[69] Orlowsky EW,Kraus VB. The Role of Innate Immunity in Osteoarthritis: When Our First Line of Defense Goes On the Offensive [J]. J Rheumatol,2015,42(3):363-371.
[70] He X,Jia H,Jing Z,et al. Recognition of pathogen-associated nucleic acids by endosomal nucleic acid-sensing toll-like receptors [J]. Acta Biochim Biophys Sin,2013,45(4):240-258.
[71] Kondo T,Kawai T,Akira S. Dissecting negative regulation of Toll-like receptor signaling[J]. Trends Immunol,2012,33(9):449-458.
[72] Li Y,Shi X. MicroRNAs in the regulation of TLR and RIG-I pathways[J]. Cell Mol Immunol,2012,10(1):65-71.
[73] Olivieri F,Rippo MR,Prattichizzo F,et al. Toll like receptor signaling in "inflammaging": microRNA as new players[J]. Immunity Ageing,2013,10(1):65-71.
[74] O'Neill LA,Sheedy FJ,McCoy CE. MicroRNAs: the fine-tuners of Toll-like receptor signalling[J]. Nature Reviews. Immunology,2011,11(3):163-175.

[75] Nahid MA, Satoh M, Chan EK. MicroRNA in TLR signaling and endotoxin tolerance. [J]. Cell Mol Immunol, 2011, 8(5): 388-403.

[76] Xiaodong Ma, Lindsey E, Becker Buscaglia, et al. MicroRNAs in NF- B signaling[J]. Journal of Molecular Cell Biology, 2011, 3(3): 159-166.

[77] Zhou R, O'Hara SP, Chen XM. MicroRNA regulation of innate immune responses in epithelial cells[J]. Cell Mol Immunol, 2011, 8(5): 371-379.

[78] Coll RC, O'Neill LA. New insights into the regulation of signalling by toll-like receptors and nod-like receptors[J]. J Innate Immun, 2010, 2(5): 406-421.

[79] Benakanakere MR, Li Q, Eskan MA, et al. Modulation of TLR2 protein expression by miR-105 in human oral keratinocytes[J]. J Biol Chem, 2009, 284(34): 23107-23115.

[80] Guo H, Chen Y, Hu X, et al. The regulation of Toll-like receptor 2 by miR-143 suppresses the invasion and migration of a subset of human colorectal carcinoma cells[J]. Molecular cancer, 2013, 12: 77.

[81] Curtale G, Mirolo M, Renzi TA, et al. Negative regulation of Toll-like receptor 4 signaling by IL-10-dependent microRNA-146b[J]. Proc Nat Acad U S A, 2013, 110(28): 11449-11504.

[82] Li X, Gibson G, Kim JS, et al. MicroRNA-146a is linked to pain-related pathophysiology of osteoarthritis [J]. Gene, 2011, 480(2): 34-41.

[83] Zilahi E, Tarr T, Papp G, et al. Increased microRNA-146a/b, TRAF6 gene and decreased IRAK1 gene expressions in the peripheral mononuclear cells of patients with Sjögren's syndrome[J]. Immunol Lett, 2012, 141(2): 165-168.

[84] Park H, Huang X, Lu C, et al. MicroRNA-146a and MicroRNA-146b Regulate Human Dendritic Cell Apoptosis and Cytokine Production by Targeting TRAF6 and IRAK1 Proteins [J]. J Biol Chem, 2015, 290(5): 2831-2841.

[85] Bazzoni F, Rossato M, Fabbri M, et al. Induction and regulatory function of miR-9 in human monocytes and neutrophils exposed to proinflammatory signals[J]. Proc Nat Acad Sci U S A, 2009, 106(13): 5282-5287.

[86] Qi J, Qiao Y, Wang P, et al. microRNA-210 negatively regulates LPS-induced production of proinflammatory cytokines by targeting NF-kappaB1 in murine macrophages [J]. FEBS Lett, 2012, 586(8): 1201-1207.

[87] Shaked I, Meerson A, Wolf Y, et al. MicroRNA-132 Potentiates Cholinergic Anti-Inflammatory Signaling by Targeting Acetylcholinesterase[J]. Immunity, 2009, 31(6): 965-973.

[88] de Gonzalo-calvo D, Dávalos A, Montero A, et al. Circulating inflammatory miRNA signature in response to different doses of aerobic exercise. [J]. J Appl Physiol, 2015, 119(2): 124-134.

[89] Liu F, Li Y, Jiang R, et al. miR-132 inhibits lipopolysaccharide-induced inflammation in alveolar macrophages by the cholinergic anti-inflammatory pathway. [J]. Exp Lung Res, 2015, 41(5): 261-269.

[90] Sheedy FJ, Palsson-McDermott E, Hennessy EJ, et al. Negative regulation of TLR4 via targeting of the proinflammatory tumor suppressor PDCD4 by the microRNA miR-21[J]. Nat Immunol, 2010, 11(2): 141-147.

[91] Squadrito ML, Etzrodt M, Palma MD, et al. MicroRNA-mediated control of macrophages and its implications for cancer[J]. Trends Immunol, 2013, 34(7): 350-359.

[92] Peacock O, Lee AC, Cameron F, et al. Inflammation and MiR-21 pathways functionally interact to downregulate PDCD4 in colorectal cancer. [J]. Plos One, 2014, 9(10): e110267.

[93] Li YH, Tavallaee G, Tokar T, et al. Identification of synovial fluid MIRNA signature in knee osteoarthritis: differentiating early and late knee osteoarthritis[J]. Osteoarthritis & Cartilage, 2016, 24(9): S347-S348.

[94] Pio G, Ceci M, Malerba D, et al. ComiRNet: a web-based system for the analysis of miRNA-gene regulatory networks[J]. BMC Bioinformatics, 2015, 16(Suppl 9): S7.

[95] Zhu W, Chen YP. Computational developments in microRNA-regulated protein-protein interactions[J]. BMC Systems Biol, 2014, 8(1): 14.

[96] Prasadam I, Batra J, Perry S, et al. Systematic Identification, Characterization and Target Gene Analysis of microRNAs Involved in Osteoarthritis Subchondral Bone Pathogenesis[J]. Calcif Tissue Int, 2016, 99(1): 43-55.

[97] Murata K, Yoshitomi H, Tanida S, et al. Plasma and synovial fluid microRNAs as potential biomarkers of rheumatoid arthritis and osteoarthritis[J]. Arthritis Res Ther, 2010, 12(3): R86.

[98] Iliopoulos D, Malizos KN, Oikonomou P, et al. Integrative MicroRNA and Proteomic Approaches Identify Novel Osteoarthritis Genes and Their Collaborative Metabolic and Inflammatory Networks [J]. Plos One, 2008, 3(11): e3740.

[99] Jones SW, Watkins G, Le Good N, et al. The identification of differentially expressed microRNA in osteoarthritic tissue that modulate the production of TNF-alpha and MMP13[J]. Osteoarthritis Cartilage, 2009, 17(4): 464-472.

[100] Jin L, Zhao J, Jing W, et al. Role of miR-146a in human chondrocyte apoptosis in response to mechanical pressure injury in vitro. [J]. Int J Med, 2014, 34(2): 451-463.

[101] Akhtar N, Rasheed Z, Ramamurthy S, et al. MicroRNA-27b regulates the expression of matrix metalloproteinase 13 in human osteoarthritis chondrocytes [J]. Arthritis Rheum, 2010, 62(5): 1361-1371.

[102] Shamoon M, Hochberg MC. Treatment of osteoarthritis with acetaminophen: efficacy, safety, and comparison with nonsteroidal anti-inflammatory drugs. [J]. Curr Rheumatol Rep, 2000, 2(6): 454-458.

[103] Balmaceda CM. Evolving guidelines in the use of topical nonsteroidal anti-inflammatory drugs in the treatment of osteoarthritis[J]. BMC Musculoskeletal Disord, 2014, 15(1): 1-5.

[104] Lanas A, Garcia-Tell G, Armada B, et al. Prescription patterns and appropriateness of NSAID therapy according to gastrointestinal risk and cardiovascular history in patients with diagnoses of osteoarthritis[J]. BMC Medicine, 2011, 9(15): 38.

(收稿日期:2016-09-07)