Epigenetic regulation mediated by microRNAs in the susceptibility and pathogenesis of rheumatoid arthritis

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Abstract

MicroRNAs (miRNAs) play crucial roles in the regulation of the transcriptome and development of diseases including cancer and autoimmune diseases, such as rheumatoid arthritis (RA). Currently, a comprehensive map, illustrating how miRNAs regulate transcripts, pathways, immune system differentiation, and their interaction with terminal cells, such as T cells, fibroblast-like synoviocytes (FLS), osteoblasts, and osteoclasts, is still missing. In this review, we provide a thorough summary of the roles of miRNAs in the susceptibility to pathogenesis, diagnosis, therapeutic intervention, and prognosis of RA. Numerous miRNAs are abnormally expressed in cells involved in RA, and regulate target genes and pathways including the NF-κB, Fas-FasL, JAK-STAT, IRE1-RIDD, and mTOR pathways. By regulating gene expression, miRNAs affect T cell differentiation to diverse cell types, including Th17 and T-reg cells, and thus constitute promising gene therapy targets to modulate the immune system in RA. We summarize the diagnostic and prognostic potential of blood-circulating and cell-free miRNAs, highlighting the novel opportunities to combine these with rheumatoid factor (RF) and anti-cyclic citrullinated peptide (anti-CCP) to provide accurate diagnosis and prognosis, especially for seronegative patients. Furthermore, we outline how functional genetic variants of *miR-499* and *miR-146a* partly explain the unmet susceptibility to RA. Additionally, we review the evidence implicating miRNAs as promising biomarkers of efficiency, response, and resistance to disease-modifying anti-rheumatic drugs (DMRDs) and immunotherapy. Finally, we discuss the autotherapeutic effect of miRNA intervention as a step toward the development of miRNA-based anti-RA drugs. Collectively, the current evidence supports miRNAs as interesting targets to better understand the pathogenetic mechanisms of RA and design more efficient therapeutic interventions.

Introduction

Rheumatoid arthritis (RA) is an autoimmune disease characterized by chronic joint inflammation and structural damage, accompanied by extra-articular manifestations such as rheumatoid nodules, interstitial pneumonia, vasculitis, and systemic complications. RA is usually progressive and insidious, and the incidence rate is 0.5-1% in Europe and North America (Ibanez-Cabellos et al., 2019). However, the precise mechanisms underlying pathogenesis, disease activity, and severity of RA as well as the causes of different response to treatment are not fully understood. In view of the current therapy strategies and treatment frames, early accurate diagnosis, effective and personalized treatment, and precision medicine have become increasingly urgent for RA patients. A comprehensive understanding of RA is hence required, from the angles of both genetics (HLA and non-HLA variants) (Okada et al., 2014) epigenetics (DNA methylation (Guo et al., 2017; Chen et al., 2019; Guo et al., 2020b), microRNA (Furer et al., 2010; Mizoguchi and Kohsaka, 2012), lncRNA (Bi et al., 2019; Guo et al., 2019), and histone modifications (Angiolilli et al., 2017)).

MicroRNAs (miRNAs) are small endogenous non-coding single-stranded RNAs, with a length of about 22 nucleotides, which are involved in the post-transcriptional regulation of

gene expression. In recent years, accumulating studies have demonstrated that miRNAs play a key role in various cancers (He et al., 2011; Lin et al., 2013; Zhang et al., 2015; Ding et al., 2016; Fan et al., 2017) as well as autoimmune diseases, including RA, systemic lupus erythematosus (SLE) (Xie and Xu, 2018; Senousy et al., 2019), sjogren's syndrome (SS) (Jang et al., 2019), and systemic sclerosis (Iwamoto et al., 2016). In this review, we systematically summarize the recent advances on the role of miRNAs in RA, with special emphasis on how the genetic variants and expression variations correlate to the susceptibility to and pathogenesis of RA, based on different inflammation-related cells, inflammatory cytokines, and inflammatory signaling pathways (**Figure 1**).

Genetic variations in miRNAs explained missing susceptibility of rheumatoid arthritis.

Genome-wide association studies have identified >100 genetic factors for RA. However, the reported genetic variants only explain <40% of the overall heritability of RA, leaving the majority of the heritability unaccounted for, thus suggesting the need for more studies that employ different approaches and populations in order to identify the missing causes. Association studies to miRNA loci provided the opportunity to identify RA-associated functional or causal variants within different populations, such as Chinese (Yang et al., 2011; Yang et al., 2012), Egyptian (El-Shal et al., 2013; Ayeldeen et al., 2018; Shaker et al., 2018), Polish (Bogunia-Kubik et al., 2016), Mexican (Aleman-Avila et al., 2017), and Iranian (Hashemi et al., 2013). The rs3746444 (20q11.22, A>G) polymorphism of *miR-499*, which is encoded by an intron of *MYH7B*, is significantly linked to RA risk, disease activity, and methotrexate (MTX) toxicity (Toraih et al., 2016); interestingly, the AA genotype shows higher disease activity and MTX toxicity than AG/GG genotypes (Fattah et al., 2018). Gene expression and genetic polymorphisms of *miR-146a* and *miR-499* showed diagnostic potential for RA (Ayeldeen et al., 2018).In addition, *miR-146a* rs2910164 is associated with RA susceptibility in the Egyptian population, in which the C allele is protective (Ayeldeen et al., 2018; Fattah et al., 2018). Consistently, the polymorphism rs3027898 in *IRAK1*, the target gene of *miR-146a*, is linked to RA in the Greek population (Chatzikyriakidou et al., 2010). However, follow-up studies showed inconsistent results in Polish (Bogunia-Kubik et al., 2016), Mexican (Aleman-Avila et al., 2017), and Chinese (Yang et al., 2011; Yang et al., 2012; Zhou et al., 2015b; Yang et al., 2017) populations. SNPs in other miRNAs were also tested in some studies, but their association with RA in other populations was not significant. For example, *miR-196a-2* rs11614913 C/T and *miR-499* rs3746444 A/G are not significantly associated with RA in Mexicans (Aleman-Avila et al., 2017), while *miR-146a* rs2910164 (Yang et al., 2011; Zhou et al., 2015b) and *miR-499* rs3746444 (Yang et al., 2011; Yang et al., 2017) do not significantly correlate with RA in Chinese. In our recent study, we demonstrated that meta-analysis could identify more significant SNPs in a large sample size, and found that the interaction between HLA alleles and miRNA SNPs (such as rs5997893 in *miR-3928* and rs4947332 in *HLA-DRB1*) should also be considered to explain unmet susceptibility (Guo et al., 2020a). In summary, genetic variations in miRNAs aids in explaining the missed susceptibility of rheumatoid arthritis.

Genes and signaling pathways regulated by miRNA in the development of rheumatoid arthritis

The cells involved in the pathogenesis of RA include CD4^{+} T cells, such as Th1, T-reg, and Th17 cells, as well as fibroblast-like synoviocyte (FLS), osteoclasts, and macrophages. The current research has mainly focused on understanding miRNA-mediated

transcriptional regulation of *FAF1* (Li et al., 2010), *TNF-α* (Trenkmann et al., 2013; Gao et al., 2018), *STAT1* (Zhou et al., 2015a), *STAT3* (Liu et al., 2017), *TLR4* (Li et al., 2019a) and *mTOR* (Tang et al., 2019; Zhu et al., 2020). miRNAs regulate inflammation, immune response, proliferation, differentiation, and influence of micro-environment within synovial joints by targeting these genes and their related pathways, including Fas-FasL (Li et al., 2010) and NF-κB (Stanczyk et al., 2011; Trenkmann et al., 2013; Wang et al., 2019a) pathway. In this section, we summarize the regulatory roles of miRNAs in the main RA-associated cell entities, focusing on T cells, FLS, and osteoclasts, to highlight the importance of miRNAs in the pathogenesis of RA.

miRNA-mediated innate and adaptive immune cell differentiation in rheumatoid arthritis

The balance of T-reg/Th17 cells plays a crucial role in RA. IL-17 released by Th17 up-regulates expression of receptor activator of nuclear factor-κB ligand (RANKL) on synovial fibroblasts and stimulates production of inflammatory cytokines such as TNF-α, IL-6, and IL-1 by immune cells (van Hamburg and Tas, 2018). The regulatory roles of *miR-146a* have been widely studied in T cells. The expression of *miR-146a* is significantly increased in CD4$^+$ T cells, peripheral blood mononuclear cells (PBMCs), and Jurkat T cells, promoting T cell differentiation and inhibiting apoptosis (Li et al., 2010; Niimoto et al., 2010). Interestingly, the expression of *miR-146a* decreases in T-reg cells during high RA activity, leading to a proinflammatory phenotype in these cells caused by concomitant up-regulation of its target *STAT1* (Zhou et al., 2015a). High levels of *miR-99b-5p* (Zhu et al., 2020), *miR-361-5p* (Romo-Garcia et al., 2019), and *miR-17* (Wang et al., 2018a) contribute to enhanced T cell proliferation and differentiation, and inhibit apoptosis. In PBMCs, *miR-99b-5p* down-regulates *mTOR* and *RASSF4* genes, thereby inhibiting T cell apoptosis, and promoting T cell proliferation and inflammatory response (Zhu et al., 2020). Up-regulation of *miR-17* in RA exosomes inhibits the differentiation of T-reg by suppressing the expression of *TGFB2* (Wang et al., 2018a). Interestingly, *miR-21* level is decreased in circulating PBMCs (Dong et al., 2014) but increased in Vγ9Vδ2T cells (Guggino et al., 2018) in RA patients, which could be associated with the imbalance of Th17/T-reg cells. Although *miR-120*-mediated negative regulation of *HIF-1* also affects the dynamic equilibrium of Th17/T-reg, no association has been found between this miRNA and RA (Huang et al., 2018).

MiRNA-induced DNA methylation, in addition to T cell differentiation, is also critical for the pathogenesis of RA. For example, *miR-126* inhibits methylation of the promoter region in CD70 and CD11a, thereby promoting their expression (Yang et al., 2015). In macrophages, binding of *miR-6089* and *lncRNA-HIX003209* enhances the expression of *TLR4* and exacerbates inflammation through the TLR4/NF-κB pathway (Yan et al., 2019). In addition, *miR-30a* can increases inflammation, by aggravating autophagy and reducing apoptosis (Xu et al., 2013). Over-expression of *miR-192-5p* ameliorates the inflammatory response in RA by targeting the up-regulation of ras-related c3 botulinum toxin substrate 2 (*RAC2*) (Zheng et al., 2020). Increasing the expression of *miR-20* and *miR-211* down-regulates their target *ATF2*, thereby reducing inflammation in RA model cells SW982 (Xie et al., 2018). The decreased expression of *miR-671* and *miR-96* in PBMCs of patients with RA may correlate with expression of *mTOR* (Tang et al., 2019) and endoplasmic reticulum stress induced by inositol-requiring enzyme 1 alpha (IRE1)/endoplasmic reticulum stress (RIDD) pathway (Ahmadiany et al., 2019). In PBMCs, *miR-29b* enhances the anti-apoptotic effect by inhibiting HMG-box transcription factor 1 (HBP1) pathway (Ren et al., 2019). Evidence shows that *miR-198*, *miR-4647*, and *miR-7167-5p* are also connected to

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T cell signaling, apoptosis, and immune response (Raj Christian et al., 2019). Numerous other miRNAs are associated with the T-reg subpopulations, such as *miR-21* and *miR-155* that are related to the memory phenotype, or *miR-92a* that is related to the naive phenotype (Smigielska-Czepiel et al., 2014). The expression of *miR-223* is high in naive CD4+ T cell, but hardly expressed in Th17 (Fulci et al., 2010). Overexpression of *miR-361-5p* in early RA is associated with T cell activation and inflammatory response (Romo-Garcia et al., 2019). Overall, miRNAs cooperate with other non-coding RNAs (ncRNAs) to alter the DNA methylation and/or expression of their targets, thus regulating innate and adaptive immune cell differentiation and apoptosis, and ultimately influencing the inflammatory and autoimmune response in RA.

miRNA-mediated cell differentiation of fibroblast-like synoviocytes in rheumatoid arthritis

Synovial fibroblasts are key regulators of inflammation and bone destruction in RA. In addition to producing RANKL, fibroblast-like synoviocytes in RA (RA-FLS) also activate osteoclast differentiation by releasing inflammatory cytokines, chemokines, and matrix metalloproteinases (MMPs) (Rana et al., 2018). Ample evidence shows that down-regulation of *miR-22* (Lin et al., 2014), *miR-29c-3p* (Tseng et al., 2019), *miR-124a* (Nakamachi et al., 2009), and *miR-4701-5p* (Bi et al., 2019), and up-regulation of miR-143 (Hong et al., 2017), miR-145 (Hong et al., 2017), and *miR-191* (Yu et al., 2019) enhances the proliferation, migration, and invasion of RA-FLS. In contrast, down-regulation of *miR-132-3p* (Tseng et al., 2019) and *miR-29a* (Liu et al., 2017), and up-regulation of *miR-31-5p* (Tseng et al., 2019) and *miR-124a* (Meng et al., 2020) inhibits these processes (Tseng et al., 2019). Besides, *miR-199a-3p* (Wangyang et al., 2018), *miR-449a* (Cai et al., 2019), *miR-506* (Li et al., 2019a)*, and miR-126* (Gao et al., 2018), whose expression is decreased in RA, inhibit RA-FLS proliferation and induce apoptosis by targeting *RB1*, high-mobility group box protein 1 (*HMGB1*), *TLR4*, and *IL-23R*, respectively. Proliferation and invasion of RA-FLS is also correlated with *MMP* (Tolboom et al., 2002). In RA-FLS, up-regulation of *miR-145-5p* (Wang et al., 2019a), *miR-18a* (Trenkmann et al., 2013), *miR-155* (Stanczyk et al., 2008; Long et al., 2013), and *miR-203* (Stanczyk et al., 2011), and down-regulation of *miR-27a* (Shi et al., 2016) contribute to *MMP* expression by targeting the NF-κB (Stanczyk et al., 2011; Trenkmann et al., 2013; Wang et al., 2019a), TLR4 (Oka et al., 2017), and Follistatin like-1 (FSTL1) (Shi et al., 2016) pathway. What's more, miRNAs can also regulate specific genes related to RA-FLS phenotypic differentiation. For example, *miR-625* is down-regulated in RA-FLS, which negatively impacts on expression of *CTSC*, *KLF8*, *EBF3*; instead, *miR-551b* is up-regulated in RA-FLS, inhibiting expression of *ITGBL1* (de la Rica et al., 2013). In RA, bone loss is mainly due to overabsorption of bone by osteoclasts and weakened osteoblast bone formation (Okamoto et al., 2017).In vitro, overexpression of *miR-221-3p* inhibits osteoblast differentiation (Maeda et al., 2017). Instead, *miR-218* overexpression promotes osteogenic differentiation of RA-FLS by suppressing the Roundabout-1 (ROBO1)/Dickkopf-1 (DKK1) axis (Iwamoto et al., 2018). In summary, miRNAs are widely involved in cell differentiation of fibroblast-like synoviocytes and osteoblast and therefore may be promising targets for drug development and enhance our understanding to the pathogenesis of rheumatoid arthritis.

Blood and serum-circulating miRNAs provide novel opportunities for the diagnosis of rheumatoid arthritis

Emerging evidence points out the potential of blood-circulating miRNAs associated with RA as potential biomarkers for diagnosis, prognosis, as well as disease activity. The levels

of *miR-371b*, *miR-483*, and *miR-642b* are significantly up-regulated while *miR-25* and *miR-378d* are down-regulated in PBMCs in individuals that eventually develop RA from early undifferentiated arthritis (Kurowska et al., 2018). Meanwhile, *miR-22* (Ouboussad et al., 2017), *miR-361-5p* (Romo-Garcia et al., 2019), and *miR-223-3p* (Romo-Garcia et al., 2019) are significantly up-regulated in high-risk or CCP-positive populations. All these miRNAs could therefore be used as biomarkers for early diagnosis of RA. Expression of *miR-103a-3p* is significantly increased in autoantibody-positive, symptomatic first-degree relatives (FDR) and RA patients, suggesting it as a potential biomarker for predicting imminent disease in individuals at risk for developing RA (Anaparti et al., 2017). Additionally, higher levels of *miR-143-3p*, *miR-145-5p*, and *miR-99b-5p* are found in the plasma of early RA patients with bone erosion, indicating that they could be monitored for bone erosion surveillance in RA patients. Furthermore, *miR-99b-5p* is demonstrated to be an independent predictor of bone erosion progression in early RA (Yue et al., 2019).

In addition to playing a role in early recognition of RA, the expression of some miRNAs aids to diagnose RA with higher accuracy (Evangelatos et al., 2019). Expression of *miR-146a* and *miR-155* are significantly increased in RA PBMCs, and shows a similar trend in whole blood (Mookherjee and El-Gabalawy, 2013). The levels of *miR-24* and *miR-125a* are significantly higher in the serum of RA patients regardless of CCP status (Murata et al., 2013). Interestingly, analysis of *miR-24-3p*, *miR-26a-5p*, and *miR-125a-5p* levels in combination constitutes a better diagnostic tool for RA, even though these miRNAs are not related to disease activity (Ormseth et al., 2015). What's more, *miR-122-3p*, *miR-3925-3p*, *miR-342-3p*, and *miR-4764-5p* show differential expression not only between healthy individuals and RA patients, but also between RA patients and osteoarthritis (OA), SLE, or Graves patients (Wang et al., 2015). Other differentially expressed miRNAs in RA patients serum include *miR-4634*, *miR-181d*, *miR-3926*, *miR-3926*, *miR-9-5p*, *miR-219-2-3p6*, *miR-221*, *miR-222*, *miR-532*, *miR-106a*, and *miR-987*, which highlights their potential as RA-specific diagnostic markers (Wang et al., 2015; Khalifa et al., 2016).

The serum levels of *miR-146a* (Abou-Zeid et al., 2011), *miR-22-3p* (Ormseth et al., 2020a), *miR-5571-3p* (Liu et al., 2019a), and *miR-135b-5p* (Liu et al., 2019a) are significantly higher in RA patients than in healthy controls and OA patients. Additionally, in RA patients, the expression of *miR-451* in T cells is significantly increased, which is positively correlated with the levels of disease activity score 28 (DAS28), erythrocyte sedimentation rate (ESR) and serum IL-6 (Smigielska-Czepiel et al., 2014). The level of *miR-146a* is positively correlates with the level of ESR and DAS28 (Abou-Zeid et al., 2011), while *miR-5571-3p* (Liu et al., 2019a) correlates with the level of ESR and C-reactive protein (CRP), and *miR-135b-5p* only with CRP (Liu et al., 2019a). These miRNAs could therefore be suitable markers of disease activity in RA patients.

Increase in serum *miR-194-5p* levels is associated with disease recurrence (Fernandez-Ruiz et al., 2018). Serum expression of *miR-23b*, which positively correlates with ESR, CRP, and DAS28, is significantly up-regulated after appropriate treatment, indicating that *miR-23b* is a dual marker of disease activity and prognosis (Liu et al., 2019b). Similarly, *miR-96-5p*, *miR-134-5p*, *miR-140-3p*, and *miR-627-5p* are not only diagnostic markers for RA, but also reflect disease activity (Ormseth et al., 2020b). In summary, the changes in miRNA levels in the serum of RA patients provide a promising opportunity for early diagnosis, as well as indication of disease activity and prediction of RA outcomes.

miRNAs as potential biomarkers for pharmacogenetics, therapeutic outcome, and treatment response prediction

Common and widely used anti-rheumatic drugs include cDMARD (MTX, sulfasalazine, and hydroxychloroquine), bDMARD (TNF inhibitors, Rituximab, and Tocilizumab), and tsDMARD (Tofacitinib, Barretinib, and Filgotinib). Several studies have explored the relationship between serum miRNA levels and the response to DMARD. Evidence shows that high serum level of *miR-10* in RA patients correlate with good response to MTX (Hong et al., 2018). After 3 months of adalimumab (ADA)/MTX combined treatment, serum level of *miR-27a-3p* significantly decrease and the clinical symptoms of RA show remission (Sode et al., 2018). Serum level of *miR-5196* also significantly decrease in RA and ankylosing spondylitis (AS) patients after anti-TNF-α therapy, and indicate lower DAS28 (Ciechomska et al., 2018). Meanwhile, serum levels of *miR-146a* diminish in RA patients who respond well to anti-TNF therapy, and can interestingly be considered as predictors of the response to anti-TNF-α therapy together with CRP (Castro-Villegas et al., 2015; Bogunia-Kubik et al., 2016; Liu et al., 2019c). By contrast, serum levels of *miR-23* and *miR-223* are increased in RA patients who respond well to anti-TNF-α/DMARD combination therapy, but correlate negatively to the response to anti-TNF drugs (Castro-Villegas et al., 2015). High serum level of *miR-125b* is potential indicator of good clinical response to Rituximab therapy (Duroux-Richard et al., 2014). Notably, *miR-432-5p* is significantly down-regulated in RA patients who are responsive to Tofacitinib therapy, but up-regulated in RA relapse patients (Fernandez-Ruiz et al., 2018). In RA, treatment with Rituximab increases the levels of *miR-16-5p* and *miR-23a-3p* in peripheral blood (Perez-Sanchez et al., 2019). The levels of *miR-425-5p*, *miR-21-5p*, and *miR-212-3p* significantly decrease in RA patients treated with glucocorticoids, although no clinical response studies have been conducted (Balzano et al., 2017).

In addition to DMARD treatment, alternative and complementary medicine preparations and mesenchymal stem cell (MSC) treatments are also used in the clinical treatment of RA. The expression of *miR-550b-2-5p*, *miR-4797-5p*, *miR-6509-5p*, *miR-378g*, *miR-4720-5p*, *miR-374b-5p*, and *miR-185-3p* are found differently between individuals who have good and poor response to treatment with tripterysium glycosides (TG) (Zhang et al., 2018; Wang et al., 2019b). Furthermore*, miR-26b-5p*, *miR-487b-3p*, and *miR-495-3p* are significantly up-regulated in the responders to adipose-derived mesenchymal stem cell treatment (Mallinson et al., 2017). Finally, *miR-124a* level in FLS increase following geniposide treatment; however, the relevance of this finding has not been assessed in clinical response studies (Wang et al., 2018b).

The auto-therapeutic effect of miRNAs has been demonstrated in mouse models of RA synovial fibroblasts (RASF) and autoimmune arthritis. For example*, miR-506* mimics reduce the proliferation of RA-FLS and production of pro-inflammatory cytokines, while also promoting the apoptosis of RA-FLS (Li et al., 2019a), *miR-449a* mimics also inhibit proliferation, migration, and IL-6 production of RA-FLS by regulating *HMGB1* and *YY1* expression (Cai et al., 2019). In a rat model of collagen-induced arthritis (CIA), *miR-708-5p* mimic improved the pathological changes by inhibiting inflammatory cell infiltration, synovial hyperplasia, and cartilage destruction (Wu et al., 2018), *miR-126* agonist inhibits the expression of IL-23R, TNF-α, and IFN-γ in the FLS (Gao et al., 2018). MSC-derived *miR-124a* exosomes inhibit proliferation and migration and promote apoptosis of FLS cell lines (Meng et al., 2020). In addition, exo-*miR-150* has been shown to inhibit RA-FLS proliferation and angiogenesis and reduce RA joint destruction by targeting *MMP14* and *VEGF* in rat RASF and CIA models (Chen et al., 2018). In conclusion, miRNAs have important and demonstrated roles in the treatment of RA, and could represent not only promising outcome biomarkers, but also novel drug targets to decrease the severity of the disease.

miRNA research in rheumatoid arthritis: remaining challenges and future opportunities

MiRNAs play multiple roles in the development of RA, from susceptibility to pathogenesis. Blood and serum-circulating miRNAs have been explored as important biomarkers for the early diagnosis, prognosis, as well as drug response prediction. Furthermore, miRNAs have been proposed for autotherapeutic approaches and as novel drug targets for the treatment of RA. Genetic variants in specific miRNAs can increase or decrease the risk and disease activity of RA in different ethnicities, and are associated with methotrexate toxicity and responses to other treatments. Moreover, the changes in miRNAs in various cells are related to the pathogenesis of and pathological changes occurring in RA, such as proliferation and differentiation of immune cells, proliferation and apoptosis of synovial cells, and synovial inflammation and cartilage destruction. Research has remarkably progressed towards the development of miRNAs as biomarkers for the diagnosis, prognosis, disease activity, and response to therapeutic drugs with RA, providing a direction for early diagnosis and accurate treatment of RA, to ultimately achieve better treatment efficiency and precision medicine in the near future. Numerous miRNAs have been shown to act as therapeutic targets in RA-FLS and CIA rat models. MiRNAs also showed the promising ability to identify subtypes of RA: for example, the expression levels of *miR-7* and *miR-214-5p* are significantly increased in the serum of RA associated-interstitial lung disease (RA-ILD) patients (Oka et al., 2017), while *miR-9-5p* targets REST/*miR-132* pathway to protect Schwann cells from inflammatory damage in RA-induced peripheral neuropathy (Li et al., 2019b). Although we have reached exciting milestones in the research on the multiple roles of miRNA in RA, more relevant studies should be implemented to understand and transfer the available knowledge into the clinical application, but also solve the current inconsistent results among different studies employing different methods or populations. For example, studies on *miR-99*, *miR-143*, and *miR-197* as the landmark miRNAs to predict the response to anti-TNF-α therapy have failed to yield results consistent with those previously reported (Cuppen et al., 2016). Finally, we expect the future development of miRNA-based baseline RA polygenetic risk score models, especially in conjunction with HLA. Meanwhile, miRNA-based early diagnosis, prognosis, and drug response prediction models could be applied in future clinical applications. Hopefully, with the identification of more miRNA-based drug targets in clinical research, miRNA-based autotherapeutic treatments could show more promising results.

Conflict of Interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Author contributions

SG and DH conceived the content. SG, CC, and LX wrote the manuscript, RZ, YJ, and MX edited the manuscript. All authors read and approved the final manuscript.

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Figure Legends

Figure 1. miRNA based regulatory network in rheumatoid arthritis. We extract all the regulatory network from the included studies and constructed the regulator network based on Cytoscape. We can find numerous studies are focusing on TNF-a and cytokines while some other studies focusing on epigenetic regulation and inflammatory pathways