ExRNA

The role of host miRNAs on *Mycobacterium tuberculosis*

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Abstract

MicroRNAs are non-coding RNAs, playing an important role in regulating many biological pathways, such as innate immune response against various infections. Different studies confirm that many miRNAs act as important regulators in developing a strategy for the survival of `Mycobacterium tuberculosis` in the host cell. On the other hand, an innate immune response is one of the important aspects of host defense against *Mycobacterium*. Considering the importance of miRNAs during tuberculosis infection, we focused on studies that performed on the role of various miRNAs related to pathogenic bacteria, *M. tuberculosis* in the host. Also, we have introduced important miRNAs that can be used as a biomarker for the detection of Mycobacterium.

Keywords: *Mycobacterium tuberculosis*, miRNA, Biomarker

Introduction

Nowadays, the broadness of infections caused by *Mycobacterium tuberculosis (M. tuberculosis)*, and the mechanism of contracting tuberculosis (TB) is not well understood. It is estimated that 2 billion people worldwide are infected with *M. tuberculosis,* among those, 10% are active *M. tuberculosis* carriers, which can be the cause of 1.4 million annual deaths. Approximately, 5–10% of people infected with TB, are active carriers through their lifetime [1]. Most of the people are asymptomatic, known as Latent tuberculosis infection (LTBI), which is detectable only by shreds of evidence of immunological test to mycobacteria proteins, such as pure protein, Mtb and purified protein derivative (PPD), and they lack clinical signs and symptoms of active disease [2]. The World Health Organization (WHO) estimates that nearly a third of the world's the population are positive for the PPD test [3]. This vast reservoir consists of people with LTBI infection as a source of illness that can lead to re-activation of the disease, especially in developing countries with high rates of tuberculosis infection. The risk of TB re-activation, among those with LTBI, is estimated in 10 % of immunocompromised patients.

Immunity weakness due to infections like HIV increases the risk of the disease up to 10% per year, and 50% throughout the lifetime [4, 5]. This latency may depend on Mtb strain and host immune response [6]. The use of immune inhibitors, for example, using anti-TNF-α in rheumatoid arthritis patients or people with Acquired Immune Deficiency Syndrome (HIV) may lead latent bacteria to reactivate [7]. Currently, the attenuated strain of *Mycobacterium bovis (M. bovis), Calmette–Guérin* (BCG) is used as a vaccine against TB, which is extremely ineffective [8]. Nowadays, the prevalence of this illness has declined through serious human efforts in research and medical care, although the occurrence of multi-drug-resistant (MDR) and extensive drug resistance (XDR) strain is increasing, and reports on the emergence of totally drug-resistant strains (TDR) have been documented [9]. The initial diagnosis of TB infection is required to control the spreading of TB and antimicrobial therapy against mycobacterial infections. The standard method involves the growth of microorganisms in a selective medium that normally requires a period of 3 to 12 weeks [10]. The preparation of smear from sputum has low sensitivity, and although evaluations based on PCR and immunological tests are rapid diagnostic methods [11–15], the existence of false positive and negative results make it unreliable. Therefore, there is an increasing need for new biomarkers or new diagnostic

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methods for TB diagnosis. Recently, microRNAs (miRNA) have been introduced as new diagnostic biomarkers that are widely involved in several cases such as cancer, heart disease, pregnancy, diabetes, psoriasis and many infectious diseases [16, 17]. Determining the physiological properties of miRNAs in immunity leads to the development of miRNA-based tests and treatments. Twenty-four years after the discovery of the first miRNA, the medical applications of mRNAs in infectious diseases has begun [18]. On the other hand, the importance of epigenetic changes as part of the pathogenesis of infectious disease increases our understanding of this matter [19]. Many microorganisms, including *M. tuberculosis*, induce epigenetic changes during infection [20]. Changes in histone post-translational modification (PTM), DNA methylation and miRNAs, all play an important role in response to an infection. The discovery of sequences of 22-nucleotide RNA, as an inhibitor for the expression of protein-coding genes, was made by Ambros et al. [21], and it was first discovered in nematodes, and then hundreds of RNA molecules in size of 20–24 nucleotides were discovered in viruses, plants, animals, and humans in the next decades. These small single-strand transcripts RNA molecules can regulate gene expression, and known as microRNAs, and have led to a change in our understanding of the regulation of gene expression. miRNA binds to complementary sequences in 3′ untranslated region of messenger transcripts (mRNA) and prevent the translation process [22]. Each miRNA may be an inhibitor for several genes, and an mRNA can be targeted by several miRNAs [23–25]. Although studies on miRNAs are still relatively elementary, it has been shown that miRNAs are the key interfaces of gene expression, there are about 2558 human miRNAs, and these miRNAs are regulated for expression of 60% of protein-coding genes [26]. MiRNAs are the main regulator of cell differentiation and cell functions, as well as modulators in most cellular functions, including innate and acquired immune systems [27, 28]. For example, acquired immune responses, B cell differentiation, antibody production, T cell development, and function are controlled by miRNAs [29], and many studies describe the role of mammalian miRNAs in response to bacterial infections [30]. *M. tuberculosis* is an intracellular pathogen and can survive within the host macrophages. Macrophages are one of the most important cells in innate immune responses that can produce antimicrobial responses, such as antimicrobial peptides, hydrolases, toxic reactive oxygen, and nitro-intermediates [31]. Survival possibility of Mtb in such antimicrobial environments is very significant, and many studies have suggested that Mtb can modulate cellular function [32]. On the other hand, many studies reported that several cellular processes are

regulated by eukaryotic miRNAs [22, 33]. Now, it has been determined that these processes are one of the important strategies of pathogenic bacteria for intracellular survival [34]. Pathogens exploit hosted miRNAs to eliminate immune responses [35–43]. In this article, we briefly reviewed the expression and role of various miRNAs, during infection with *M. tuberculosis*. Nowadays, due to the importance of miRNA role in the pathogenesis of tuberculosis, many types of research have focused on its practical aspects, although several researchers have explored new dimensions of miRNA effects in pathogenesis, to identify a biomarker for diagnosis of tuberculosis. Given the importance of this issue and the importance of gaining much more information from the researches on the subject of miRNA and its relation with facilitate survive of tuberculosis, the reading of such articles could have an intense involvement in performing prospective investigations.

The role of miRNAs in TB infection

M. tuberculosis is an ancient organism that has been coordinated with its human host, so it has been adapted to macrophage in the host cell for survival [44]. To date, little is known about how the macrophage immune response changes during tuberculosis infection by host miRNAs, which is the first phagocyte immune response in pulmonary microenvironments relative to *M. tuberculosis* [44]. To ensure survival and proliferation, pathogenic bacteria manipulate a wide range of host cellular pathways and functions [45]. The regulation of miRNA expression by infection due to bacterial pathogens, as soon as infection occurs, is as an essential part of the host response to infection, as well as a new molecular strategy for regulating host cell pathways by bacteria. While macrophages are target cells for Mycobacterium infection but are not affected by miRNAs, during infection. The critical point of the inherent and acquired immune responses is dendritic cells that can activate and polarize the topical T cell responses, regulated by miRNAs [8]. miRNAs play an essential role in regulating the primary function of macrophages, dendritic cells, and Natural Killer Cells (NKCs) [46, 47]. Many studies indicate a change in the gene expression in macrophage and NKC, due to latent and active TB, and also in healthy individuals, compared to those with TB [48–50]. miRNAs regulate the gene expression changes and variation in cellular compositions. Several miRNAs regulate T cell differentiation and their function [43, 51]. Bin et al. showed that the activation pathway of the intrinsic macrophages could change regulation, through several miRNAs (Fig. 1).

Further, they showed that *M. tuberculosis* modifies miR-26a, miR132 and other host miRNAs, attenuating immune responses to ensure survival. They also showed that miR-132 and miR-29a typically act as negative
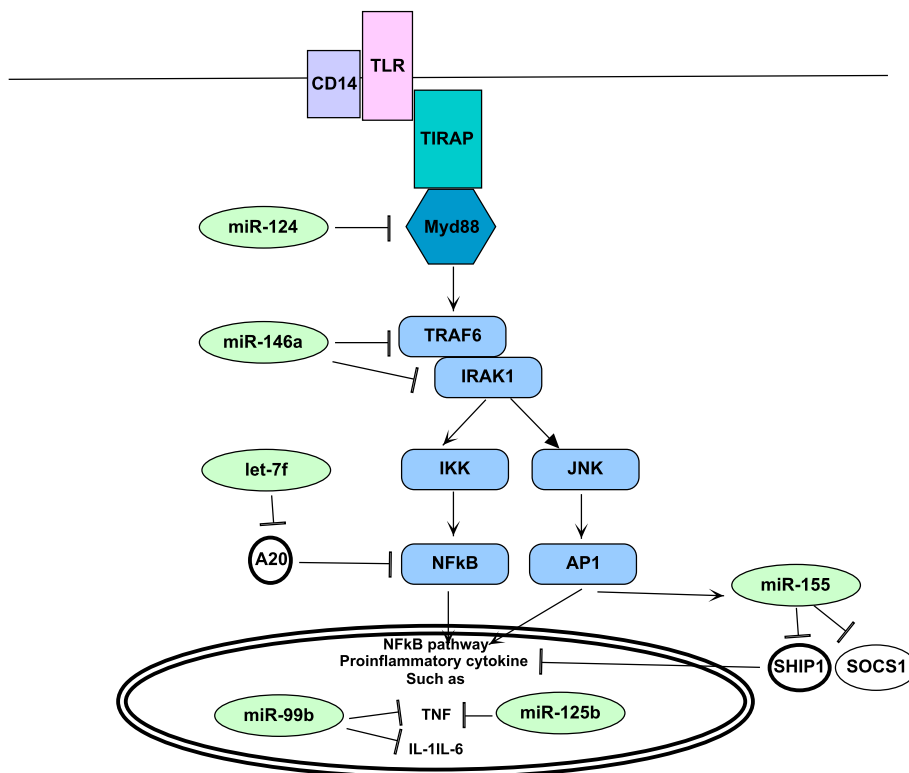
Fig. 1 MiRNAs involved in activation of the immune response and macrophage defense, during *M. tuberculosis* infection

regulators for macrophage function via interferon gamma. In the case of pulmonary TB, the induction of these two miRNAs in alveolar macrophages limits the immune response and degenerates the alveolar space [52]. On the other hand, previous studies have shown that miR-361-5p is relatively similar to the amount of bleomycin-induced fibrosis in the mouse lung, and may be involved in understanding the mechanisms of lung injury and fibrosis [53]. Yuhua et al. showed for the first time that high levels of miR-361 were expressed in the serum of patients with TB, compared with the healthy individuals, and it can be speculated that this reflects lung injury due to TB infection, although the associated mechanism is unclear (Table 1) [54].

miRNA-29, miRNA-21 and miRNA-26a

It has been observed that the miR-29 expression increases after infection with *Mycobacterium* virulent species [54–56]. Similar to what has been found for listeria infection, the miR-29 expression is downregulated, in interferon gamma-producing NKC cells, as soon as the *M. bovis* infection occurs [57]. It should be noted that expressing and regulating miRNA is cellular context-dependent [58]. The miR-29 knockdown in mice, results in more resistance to *M. bovis* infection and *M. tuberculosis* [57], suggesting that miR-29 induction in the T cell during

the infection, facilitates bacterial virulence. Another study showed that upregulation of miR-29 inhibits the interferon-gamma expression [57]. miR-29 causes inhibition of interferon-gamma and excessive miR-29 may change latent TB to an active TB [57]. In a study, miR-29 was found to be increased in T cells of TB patients, compared with LTBI and negative control [59].

In contrast, kleinsteuber et al. showed a decrease in miR-29 in CD4 T cell of TB patients, compared to LTBI

Table 1 MiRNAs and its regulatory effects on genes involved in immunity against *M. tuberculosis*

MiRNAs	The role of miRNA
MiR-29	Inhibition of interferon gamma (IFNγ)
MiR-21	Reduced expression of IL-1B IL-12P3 inhibition IL-10 increase inhibitory cytokine
MiR-99b	Target TNF-α mRNA transcript
MiR-125	Target TNF-α mRNA transcript Reduce inflammatory responses
MiR-155	Target TNF-α mRNA transcript Inhibition of interferon gamma
MiR-144	Inhibition of interferon gamma and TNF-α
MiR-223	Inhibition of Interleukin 6
MiR-26a	Polarization induction of anti-inflammatory M2 macrophage phenotype.

(but not in the negative control group) [60]. Fu et al. also investigated the expression of 1223 miRNAs on pooled serum samples, from TB patients. Meanwhile, an increase in miR-29 expression was observed in sputum specimens [54]. A similar group examined the pattern of miRNA expression in sputum and confirmed the difference in reported appearance [56]. Wu and colleagues showed that *Mycobacterium* is an inducer of miR-21 expression, leading to weakening of the activation of the macrophages and Th1-dependent immunity [61]. Although the exact mechanism for regulating the Bcl2 expression by miR-21 is unknown, inhibition of miR-21 induces IL-12 production and induces anti-mycobacterial responses, and miR-21 may be considered as an effective strategy for mycobacterium to escape the host immune responses and establish chronic infection [62]. Bin et al. showed that TB is an inducer of miR-26a, and inducing this miRNA leads to a reduction in the P300 expression, which in turn leads to a reduction in the transcription of interferon-gamma-inducing genes and the response of macrophages to this crucial cytokine. Interferon-gamma repression in CD4, CD8 T cells by tuberculosis may be a survival strategy in the host cell [52].

miRNA-125b and miR-155

In a study, Rajaram et al. showed a link between virulence of *Mycobacterium* species and TNF-α production, and a difference in expression between miR-155 and miR-125b [63]. miR-125b directly targets mRNA of TNF-α and results in associated destabilization. Murugesan et al. showed that miR-125b is attached to 3′-UTR of TNF-α transcript and caused downregulation [64]. On the other hand, the enhancer of sustainability is KB2-Ras2 that is NFkB signaling inhibitor in human macrophages, thus reduces the inflammatory responses [65]. miR-55 can be the inducer of TNF-α synthesis by targeting SHIP-1, which is a negative regulator of the P13K / AKT route. Munigesan et al. found that *Mycobacterium smegmatis* (SmegLM) is an inducer of the miR-155 expression in macrophages, which reduces the SHIP1 expression and thus increases TNF mRNA stability and TNF production. Their studies showed that miRNAs were essential regulators for the production of TNF during mycobacterial infection [57]. Interestingly, the induction of cells with lipomannan, components of the bacterial cell wall, caused by virulent TB strain or *M. smegmatis* non-virulent strain, also lead to opposite effects on the synthesis of TNF-α, in a way that the lipomannan produced by TB is an inhibitor of TNF-α synthesis, while the lipomannan provided by *M. smegmatis* is the inducer of TNF-α expression. This phenomenon is related to the balance between the expression of miR-155 and miR-125b [63, 66]. In another study, transfection of murine macrophages with miR-155 resulted in a decrease

in mycobacterium intracellular survival [67]. It is possible that the miR-155 varies the antimicrobial activity by regulating two processes, including the macrophage apoptosis [68] and autophagy [69] for immunity. Another study, by Wang et al., showed that miR-155 upregulation could determine TB infection in mouse macrophages by activating the autophagy pathway [69], and inducing autophagy through inhibition of the negative regulator Rheb and other components of the signaling pathway mTOR [69, 70]. Another study reported that *M. tuberculosis* causes high levels of miR-155 and lower levels of miR-125b, while *M. smegmatis* is an inducer of low levels of miR-155 and high levels of miR-125b. The induction of miR-155 expression in active or harmful TB infection is still uncertain. Kumar et al. showed that in mouse macrophages, *M. tuberculosis* could modulate the cell's environment in its favor, and that act is due to the miR-155 expression through the EAST-6 protein, which correlates with the virulence of bacteria [67]. The mutant strain of ESAT-6 TB has a lower induction of miR-155 in macrophages than wild type [67]. Upregulation of miR-155 can activate the AKT pathway, involve for the survival of *M. tuberculosis* in macrophages, and it is the inhibitor of the cytokine-induced pro-inflammatory IL-6 [67]. Given the increase in the synthesis of TNF-α through SHIP1 pathway [63], and considering some negative effects, miR-155 function in the survival of mycobacteria within the host cell remains unclear. Despite these issues, it has been shown that mycobacteria have a mechanism to deal with the negative effects of miR-155, which help *mycobacterium* to survival in the host, for example, the lipomannan from the cell wall of TBF-α is an inhibitor of TNF-α synthesis and is contrast with the effect of upregulation of miR-155 [63]. On the other hand, TB is an inducer of miR-125b, which directly targets mRNA of TNF. Therefor miR125-b can also reduce TNF synthesis and balance the effects of upregulation of miR-155.

miRNA-144 and miRNA-146a

Overexpression of miR-144 has been observed in patients with active TB [71]. Cheng et al. showed that miR-144 is significantly altered in PBMC of patients with active TB [72]. Yuhua et al. showed that miRNAs, mostly upregulated in the serum of patients with TB, while only seven miRNAs are downregulated, although the miR-144 expression in this group did not confirm by q-PCR [54]. miR-144 can target Janus / kinase (JAK) signal transducer genes, MAPK and TLR signaling pathways, and Cyto-Cyto receptor interactions. miR-144 is also the inhibitor of the production of TNF-α and interferon gamma, both playing an important role in protecting immunity. Different findings have been reported about miR-144 expression, Wang et al, indicate the increase of the miR-144 expression in TB patients (only in

comparison with the negative control group) [7], while no expression difference was found in miR-144 by others [73]. Since miR-144 is an important factor in T cells in TB patients, such diverse and confusing results can be due to heterogeneity in PBMC samples [74]. In addition, miRNA array shows a reduction in the miR144 expression in CD4 T cell in TB patients, compared to LTBI, but the results from the analysis of pooled samples with q-PCR did not confirm this result [62]. miR-223 acts like miR-146a, which modulates the IKK-α subunit of NFkB and regulates inflammatory responses in phagocytic monocytes. miR-223 is significantly upregulated in the blood and lung of patients with TB [75]. Also, upregulated miR-223 is the inhibitor of CCl3, CXCL2, and IL-6, and it has recently been reported that miR-223 deletion causes hypersensitivity to TB infection [76].

Mycobacterial infections in macrophages significantly induce the miR-146a expression; that expression level is a dose-dependent [77]. This miRNA involves two critical factors in the TLR / NFkB signaling pathway, including IRAK1 and TRAF6; the increase in the expression of this miRNA during the infection, affects the TLR / NF-kB pathways, and subsequently reduces the cytokines TNF-α, IL-1b, IL-6, and chemokine MCP-1. In particular, *M. tuberculosis* seems to use mannose receptors to escape the bactericidal effects of superoxide [78].

Other miRNAs

The ability of *M. tuberculosis* to survive and develop a disease is associated with the escape from the host defense and immune mechanisms. In particular, tuberculosis has a significant potential for survival within the hostile environments of macrophage. *M. tuberculosis* has expanded many pathways to inhibit macrophage antimicrobial effects for intracellular survival [32]. One of these strategies is the ability to prevent phagosome maturation and other measures to avoid autophagy and to escape from the phagosome environment [79–81]. Autophagy has recently been introduced as a mechanism for killing pathogens. Autophagy is an intracellular process involved in self-digestion or self-eating, in which cytoplasmic components are transmitted to the lysosome and are ultimately degenerated [82]. The pathways associated with autophagy are challenging to regulate at post-transcriptional levels and are well described, but the involvement of miRNAs inactivating or inhibiting autophagy during TB infection is largely unknown [30]. Some reports show the induction of the miR-33 expression in THP-1 and HEK-293 cells, leading to inhibition of pathways involved in autophagy, and also resulting in reprogramming of host lipid metabolism for intracellular survival and TB stability [30]. Recent studies have also shown that miR-33 leads to inhibition of autophagia through inhibition of possible autophagy factors, such as

ATG5, ATG12, LC3B, and transcription factors, such as FOXO3 and TFEB (as an important regulatory factor in regulating transcription of the genes associated with autophagy) [75].
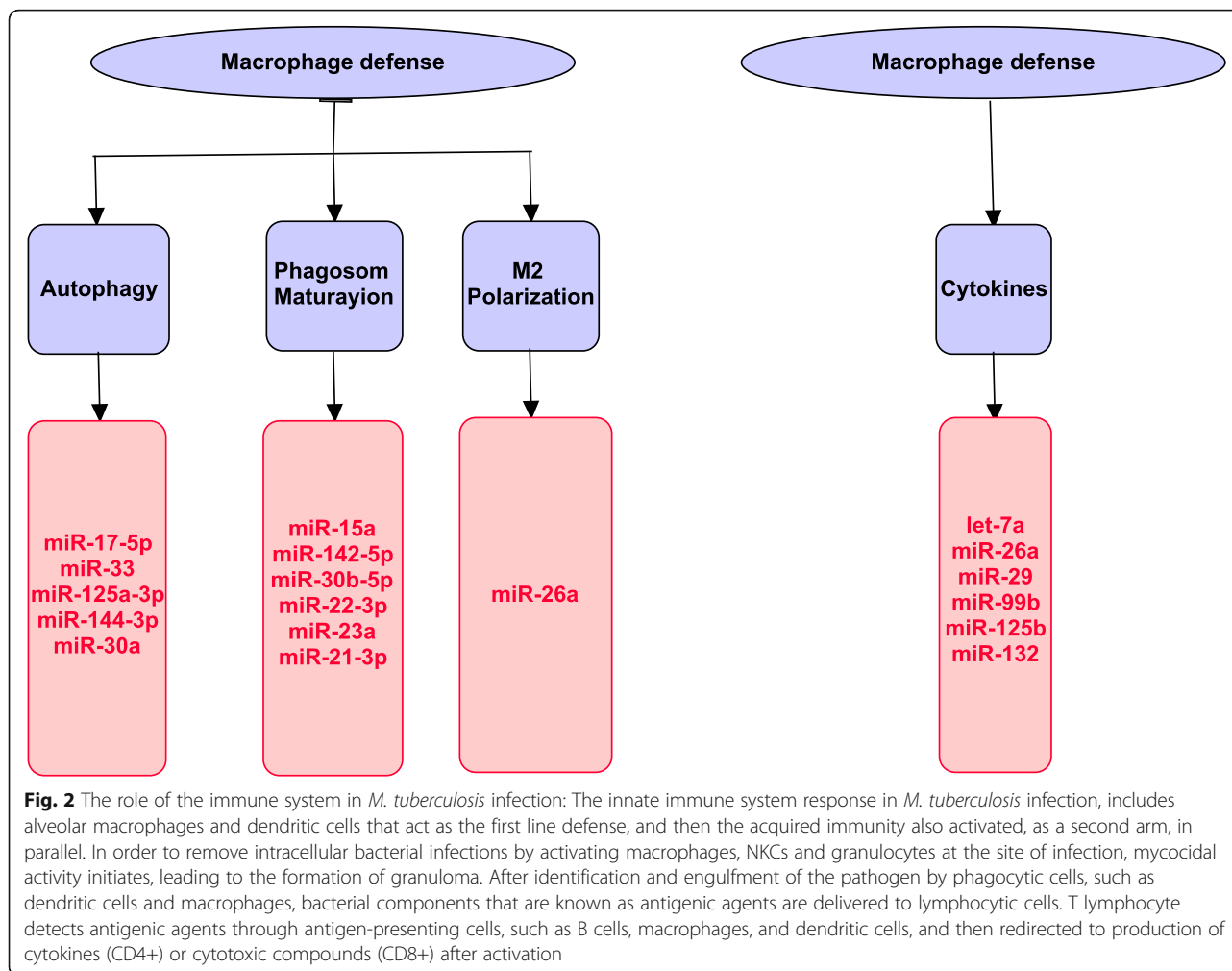
Kim et al. [75] stated that miR-125a-3p was upregulated in macrophages infected with TB, which is related to the inhibition of autophagy by targeting UVRAG. Guo et al. [83] also showed an increase in the miR-144-3p expression that is an inducer of the ATG4a gene (a gene involved in autophagy inhibition). Another study suggested that overexpression of miR-23a-5p inhibited autophobic activity [9]. Another study demonstrated the downregulation of miR-3619-5p by BCG, leading to up-regulation of cathepsin S (CTSS) (Lysosomal Cysteine Protease), and the inhibition of the CTSS expression can enhance autophagy. Chen et al., showed that the miR-30a is a negative regulator of autophagy that was up-regulated in macrophages infected with TB, although they believed that increase in the miR-30a expression alone could not be the leading cause of inhibition of autophagy, speculating that this miRNA is part of a complex mechanism that is regulated by many molecules, associated with autophagy (Fig. 2) [84].

miRNAs as a biomarker

miRNAs are widely considered as non-invasive prognosis and prognostic markers. Many of studies have used miR-NAs, as diagnostic biomarkers for early detection of many cancers, such as breast cancer [85], lung carcinoma [86, 87], and colorectal cancer. Considering the new findings, concerning miRNAs, and also the fact that miRNAs are stable in the serum [88]. Therefore, they can be considered as a good biomarker [89, 90].

Recently, the role of miRNAs in host-pathogen responses has been considered. Human miRNAs may play an essential role in viral proliferation, limitation of anti-viral responses, inhibition of apoptosis and induction of cell growth [91]. Also, miRNAs play a significant role in immune response and inflammatory responses in bacterial infections [57, 92]. Diagnosis of TB infection is severe, compared to many other bacterial infections [44]. One of the effective methods for controlling the spread of TB is the early diagnosis of the disease. Nowadays, many diagnostic tests do not distinguish between active TB and LTBI, and thus miRNAs can be reliable, as potential diagnostic biomarkers [93]. Although the suitable biomarker has not yet been identified, [94], recently, several types of miRNA as a biomarker have been investigated in the diagnosis of TB [72, 95, 96], using PBMC and serum [72] of patients with TB.

Interestingly, an active link between the expression of miRNA and gene expression has been found [30]. Wang et al. [97] showed that miR-31 is significantly reduced in patients with TB, compared to the healthy children, and

Fig. 2 The role of the immune system in *M. tuberculosis* infection: The innate immune system response in *M. tuberculosis* infection, includes alveolar macrophages and dendritic cells that act as the first line defense, and then the acquired immunity also activated, as a second arm, in parallel. In order to remove intracellular bacterial infections by activating macrophages, NKCs and granulocytes at the site of infection, mycocidal activity initiates, leading to the formation of granuloma. After identification and engulfment of the pathogen by phagocytic cells, such as dendritic cells and macrophages, bacterial components that are known as antigenic agents are delivered to lymphocytic cells. T lymphocyte detects antigenic agents through antigen-presenting cells, such as B cells, macrophages, and dendritic cells, and then redirected to production of cytokines (CD4+) or cytotoxic compounds (CD8+) after activation

besides, this study indicates that expression of this miRNA has a negative correlation with the levels of IL-6, TNF-α, and IFN. They also argued that the expression profile of miRNAs varies, among many individuals and it is not gendered specific or clinical phenotype-dependent, although they were able to distinguish the expression of an active TB group from the latent TB group, using the 17miRNA predicted by the SVM method, most (12 out of 17) upregulated in patients with active TB [7]. Barry et al. (2015) also showed that miR-93 as miRNA is suitable for normalizing miRNA levels in TB patients [98]. Latorre et al. also introduced nine miRNAs with different expressions, in patients with active TB, compared to healthy individuals or people with LTBI.

MiR-361-5p, miR-889, and miR-576-3p also showed good ability to detect TB infection from other microbial infections. Information gathered from these three miRNAs showed a significant difference between TB infections and three groups of microbial infections [53]. Miotto et al. also distinguished a cluster of

15miRNA, among children with TB and healthy controls, and introduced miR-192 as the only candidate, showing significant differences in adults and children [92]. On the other hand, some studies suggest that miRNA may also be useful in the development of TB-resistant strains, for example, Ren et al. (2015) [99] showed 142 different miRNAs are expressed in individuals with MDR TB, not seen in sensitive strains.

All of these studies have significantly contributed to the presentation of various miRNAs as biomarker candidates for TB diagnosis, but so far no miRNA has been included as a biomarker, and many factors are relevant in this regard, including the heterogeneity of data. For example, data from Zhou and colleagues revealed a lot of inconsistencies with previous studies; for instance, they showed that miR-155 is downregulated in people with TB [100]. While Wu et al. [96] showed miR-155 in the PBMC of patients with active TB was upregulated. On the other hand, Zhou et al. showed that miR-141, miR-32, miR-29b were overexpressed in the TB group,

while the expression level of miR-144 varied in previous studies, for example, Wang et al., showed upregulation of miR-144 in TB patients. [7] While Wu and colleagues report the downregulation [96], Zhou et al. [100] did not see expression changes and this controversy in the results is due to different conditions and the use of different protocols. Although, Ueberberg et al. [101] reported that miR-22, miR-25, miR-19, miR-365, miR-4835p, miR-590 and miR-885-5p are suitable biomarkers, because of being validated in two different studies. Other studies that led to introducing this factor as an appropriate biomarker were lacking a statistical significance, as well as using a small group size, which requires further studies to validate the potential diagnostic marker.

Conclusion

TB is one of the deadliest diseases in the world, which is very difficult to eradicate because of its ability to survive in macrophages. Intracellular bacteria, such as *M. tuberculosis* can survive and multiply in phagocytic cells and generally

can regulate the host defense system to survive and replicate through various pathways. One of these pathways is the change in the miRNA expression, to change the immune response and ultimately facilitate the establishment of infection in the host cell. In recent years, the role of miRNAs as regulatory factors in the inherent and acquired immune responses to TB infection has been widely considered. MicroRNAs have been studied extensively and have an important ability to regulate gene expression. miRNAs affect many important processes and are important regulators of the immune system (Fig. 3).

On the other hand, many studies have confirmed the different expressions of miRNAs in people with active TB and those with latent infection, and these findings provide new insights for the use of miRNAs as diagnostic biomarkers. Although there are some limitations in this regard, including the fact that miRNAs are not entirely gene-specific, many of their characteristics have made them suitable biomarker candidates. One of the important properties that make them more suitable
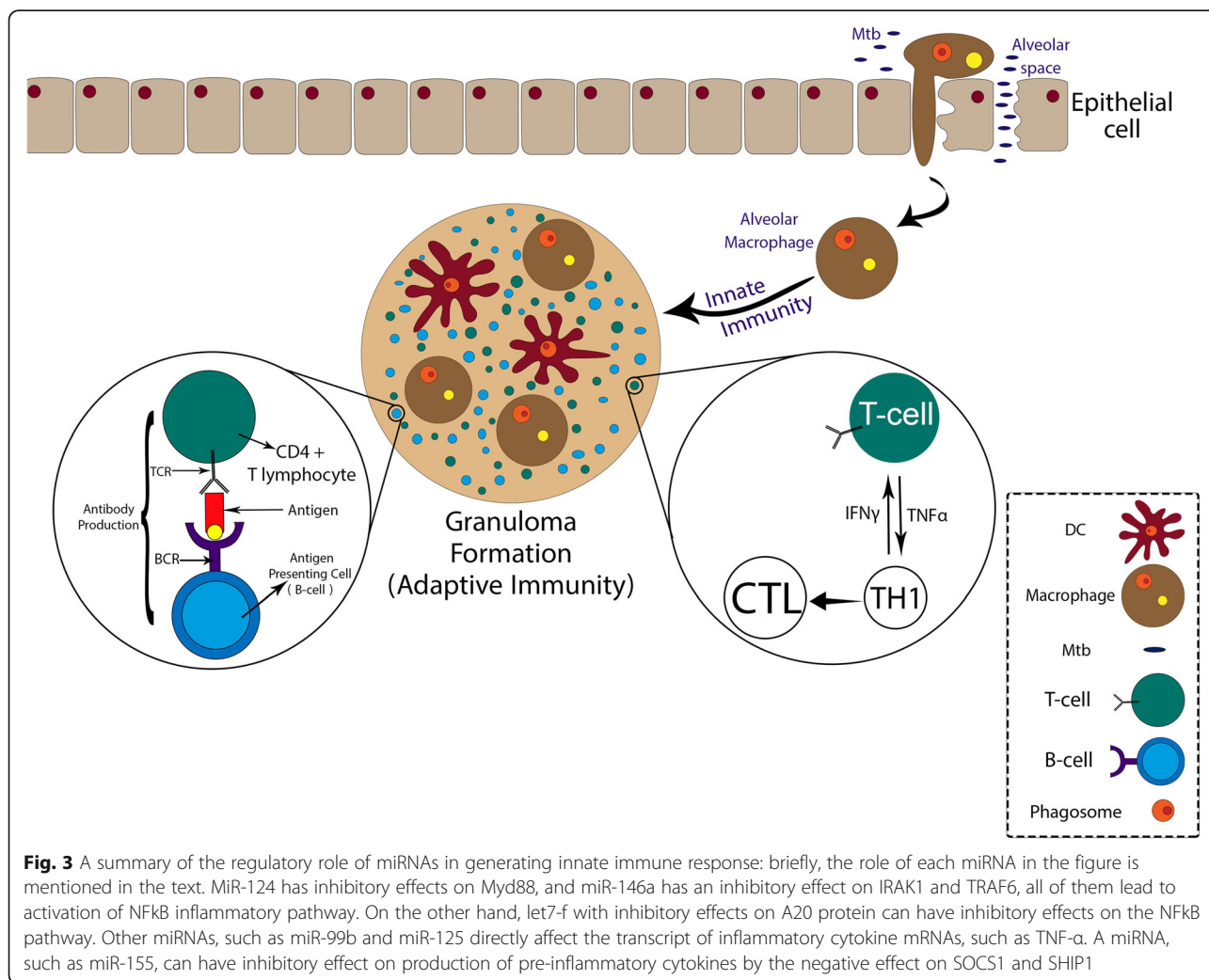

Fig. 3 A summary of the regulatory role of miRNAs in generating innate immune response: briefly, the role of each miRNA in the figure is mentioned in the text. MiR-124 has inhibitory effects on Myd88, and miR-146a has an inhibitory effect on IRAK1 and TRAF6, all of them lead to activation of NFkB inflammatory pathway. On the other hand, let7-f with inhibitory effects on A20 protein can have inhibitory effects on the NFkB pathway. Other miRNAs, such as miR-99b and miR-125 directly affect the transcript of inflammatory cytokine mRNAs, such as TNF-α. A miRNA, such as miR-155, can have inhibitory effect on production of pre-inflammatory cytokines by the negative effect on SOCS1 and SHIP1

candidates is their high stability in body fluids and their relationship to many diseases that can be used as bio-markers for the classification of infectious diseases, as well as for therapeutic purposes. On the other hand, the involvement of miRNAs in autophagy processes has opened a new window to scientists. All of these findings can provide valuable information on the diagnosis, treatment, and design of appropriate vaccines against infections caused by *M. tuberculosis*. Ultimately, the potential for the use of miRNAs as biomarkers in the treatment of TB requires further extensive studies in this field.

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