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Immune cell diversity contributes to the pathogenesis of myocarditis

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Abstract

Myocarditis (MCD) is a type of inflammatory disease in which inflammatory cells infiltrate the myocardium, leading to cardiac dysfunction, myocardial necrosis, and fibrosis. Although it has been reported that MCD is mediated by T cells, the immune system is complex and includes many types of immune cells that interact with one another. Through investigations of the inflammatory responses in MCD including myocardial necrosis, fibrosis, and arrhythmia, we have gained further insight into the pathogenesis of MCD. This article aims to discuss the diversity and the roles of immune cells involved in the pathogenesis of MCD. Moreover, immunotherapy for the treatment of MCD remains controversial, and further investigation is required to identify accurate immunotherapies for special cell types.

Keywords Myocarditis · Immune cell diversity · Interaction · Pathogenesis · Immunotherapy

Introduction

Myocarditis (MCD) refers to the clinical and histological manifestations of a broad range of pathological immune processes in the heart, in which alterations in the number and function of lymphocyte subsets and macrophages and antibody-mediated injuries are present [1]. In most cases, MCD resolves spontaneously, but in susceptible individuals it may progress to a chronic stage, which leads to pathological cardiac remodeling that includes tissue fibrosis, hypertrophy, and apoptosis of cardiomyocytes. This remodeling results in a phenotype of dilated heart chambers with impaired cardiac function, in particular impaired contractility (inflammatory cardiomyopathy) [2]. Endocardial biopsy is the gold standard for diagnosing MCD, despite a lack of sensitivity, mainly due to sampling error [3, 4].

Many pathogenic factors contribute to the occurrence of MCD, including infectious factors (viruses, bacteria, and protozoa) and noninfectious factors (toxins, vaccines, and several drugs), as well as systemic autoimmune diseases that can also trigger heart-specific autoimmunity and inflammatory responses [5]. Inflammatory responses are mainly mediated by immune cells and cytokines. In the resting or a diseased state, there are many types of immune cells that make up the whole cardiac cell library [6].

In the resting heart, the exact percentage of immune cells present is unknown, but presumed to be low, while a large number of immune cells infiltrate the space around cardiomyocytes in disease conditions, especially MCD [7, 8]. As has been previously demonstrated, a diverse set of immune cells contribute to inflammatory response processes, including innate and adaptive immune cells, so it is essential to determine the roles of specific immune cells in the pathogenesis of MCD. This review aims to summarize all the immune cells reported to be involved in MCD so far and to determine the related mechanisms in the process of MCD.

Immune anatomy in the resting heart

Innate and adaptive immune cells make up the cells of the immune system (Fig. 1). Like most tissues, the primary immune cells that reside in the heart are macrophages, which localize near endothelial cells or within the interstitial space, with very few monocytes found within cardiac tissue [9, 10]. Scarce numbers of dendritic cells (DCs) have been found within cardiac tissue and cardiac valves [11]. Mast cells, which are also found in resting cardiac tissue, are thought to be important early triggers of immune responses [12]. Neutrophils, which mostly take part in the pathophysiology of bacterial infection, are typically not found within noninfectious cardiac tissue [13–15]. As for adaptive immune cells, a

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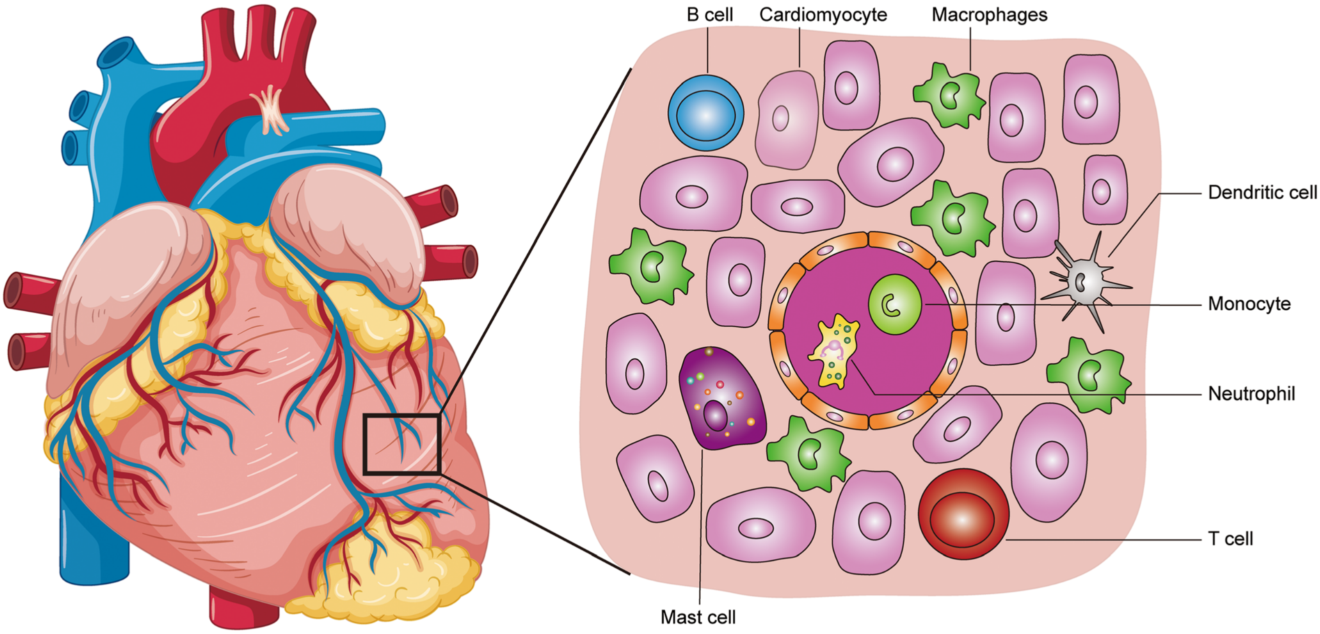

Fig. 1 Immune anatomy in the resting heart. Macrophages make up the majority of the immune cell population, which localize around the endothelial cells and myocardial interstitium. The mast cells and dendritic cells (DCs) can also be found in the resting heart. As has been shown previously, neutrophils and monocytes are not found in the resting heart, except in the blood vessels. T cells and B cells were seldom found in normal condition

small number of regulatory T cell subsets and B cells are present in cardiac tissue during resting conditions [16, 17]. During resting conditions, innate immune cells make up a major proportion of all the immune cells present. Regardless of the inflammatory trigger, cardiac tissue initiates a dynamic cellular cascade that initially activates resident immune cells and over time evolves in a coordinated manner, leading to the recruitment of diverse leukocyte populations into the inflamed tissue, especially MCD (Table 1).

Macrophages protect against MCD by alleviating myocardial inflammation

Macrophages, which have characteristic CD68 and CD163 surface markers, make up a large portion of the cardiac immune cell population [18]. Recent studies using genetic fate mapping and adoptive transfer techniques have demonstrated that resident cardiac macrophage populations are defined by parabiosis. Rather than being a homogenous population, three discrete subsets with different origins and functions make up the resident cardiac macrophage population [19]. These three macrophage subsets can be well defined based on the expression of MHC class II and CC chemokine receptor 2 (CCR2). MHC class II^{hi} and MHC class II^{low} cardiac macrophages are both CCR2- and are numerically the dominant subsets in the heart [19]. These two subsets are primarily derived from embryonic progenitors, with a substantial number arising from embryonic yolk sac precursors. They renew through in situ proliferation rather than through monocyte input. The third cardiac macrophage subset is made up of CCR2+ macrophages, which are derived from, and slowly replenished by, circulating blood monocytes. Previously, dichotomous expression of two classical myeloid cell markers (F4/80 and CD11b) was used to discern embryonic versus adult monocyte origin [43]. However, genetic fate mapping remains the most accurate method for lineage discrimination of cardiac macrophage because these cell surface markers alone failed to distinguish the different cardiac macrophage lineages [19, 44].

Interestingly, constructing a cardiac tissue injury model is the way to define the role of recruited monocytes and monocyte-derived macrophages. Nonselective depletion strategies have shown that in the absence of both monocytes and macrophages scar formation is impaired after cardiac ischemic injury, with decreased collagen production, decreased angiogenesis, and increased mortality due to myocardial rupture [10, 45]. These data suggest that recruited monocytes have a pathological role in the setting of sterile cardiac injury.

Macrophages play an important role in the pathogenesis of MCD. In CVB3-induced MCD when macrophages were depleted by liposome-encapsulated clodronate treatment, the mice presented higher viral titers but reduced acute myocarditis and chronic fibrosis [20]. In further studies, interleukin (IL)-33 upregulation significantly alleviated the severity of viral MCD with an increased cardiac contractive function and higher survival rate. Mechanistic

Table 1 Summary of immune characteristics in MCD

Type of immune cells	Markers	Functional roles	Association with MCD	Reference
Innate immune cells				
Macrophage (M)	CD68/CD163	Antigen presenting, innate immune response, electrical conduction facilitation	Macrophage M2 polarization protects mice from viral MCD by alleviating inflammation	[18]
MHC II^{hi} M	MHC II^{hi}/CCR2^{-}			[19–21]
MHC II^{low} M	MHC II^{low}/CCR2^{-}			
CCR2^{+} M	MHC II^{-}/CCR2^{+}			
Dendritic cell (DCs)				[22]
Plasmacytoid DCs (pDCs)	CD123 CD303	Type 1 interferon production activating CTL	Limiting MCD	[23, 24]
Conventional or classical DCs (cDCs)	CD209, CD103 and CD141 MHC II	Antigen presenting		[24–26]
Natural killer cells (NK)	CD56 and CD94 (NKG2A)	Killing cells and regulating cardiac inflammation	Protect against MCD	[27–29]
Mast cells	Tryptases, chymases, and carboxypeptidase A3	The first line of defense and the release of granules to regulate the inflammatory response	The promotion of MCD and fibrosis need more investigation	[30, 31]
Adaptive immune cells				
CD4^{+} T cells	CD3, CD4			[2, 32]
Th1 cells	IL-12, IFN-γ, TNF-α	Regulating the inflammatory response	Limiting MCD	[33, 34]
Th2 cells	IL-4, IL-5, IL-10, IL-23		Promoting MCD	[33, 35]
Th17 cells	IL-17, IL-6, IL-21, IL-22		Promoting the MCD transition to DCM	[36, 37]
Tregs	CD4, CD25, FOXP3	Immunosuppressive activity against a broad and diverse array of antigens within different microenvironments by produce inhibitory cytokines	Limiting MCD	[32, 38]
CD8^{+} T cells	CD8	Cytotoxic T lymphocytes (CTLs), memory T cells and regulatory T cells	Promoting MCD, but this needs to be further investigated	[39–41]
B cells	CD19, CD20	Antibody production	Promoting MCD	[16, 42]

studies demonstrated that IL-33 could stimulate ST2L+ F4/80 + macrophages and ST2L+ CD4 + T cells in cardiac tissue to express IL-4, which was a potent inducer of macrophageM2 polarization by promoting the activation of JAK/STAT6, CEBP-β, PI3K, and other signaling pathways to increase the M2 gene expression of arginase-1, IL-10, macrophage mannose receptor (MMR), and macrophage galactose type C-type lectin (MGL). Additionally, M2 polarization resulted in the release of IL-10, which was an inducer of regulatory T cell-mediated immune responses (Fig. 2). Mice with adoptively transferred M2 macrophages exhibited less cardiac inflammation and attenuated myocarditis, suggesting a protective role for M2 macrophage in viral myocarditis [21]. Furthermore, male and female mice had different susceptibilities to MCD due to differences in M2 macrophage polarization and strikingly, the transfer of M2 macrophages into susceptible male mice remarkably alleviated myocardial inflammation by modulating local cytokine profiles and promoting peripheral regulatory T cell differentiation [46]. Taken together, these data indicate that M2 macrophage polarization can protect mice from MCD by alleviating myocardial inflammation.

Macrophages play an essential role in not only the inflammatory response, but they also facilitate electrical conduction through the distal atrioventricular node where conducting cells are densely interspersed with elongated macrophages expressing connexin (CX)-43 [47]. Studies showed that when coupled to spontaneously beating cardiomyocytes via CX-43-containing gap junctions, cardiac macrophages have a negative resting membrane potential and depolarize in synchrony with cardiomyocytes. Conversely, macrophages render the resting membrane potential of cardiomyocytes more positively charged and accelerate their repolarization, according to computational modeling. Mechanistic studies suggested that the photostimulation of channelrhodopsin-2-expressing macrophages improves atrioventricular conduction, whereas conditional deletion of CX-43 in macrophages and congenital lack of macrophages delay atrioventricular conduction [47]. Meanwhile, some reports have indicated that a portion of

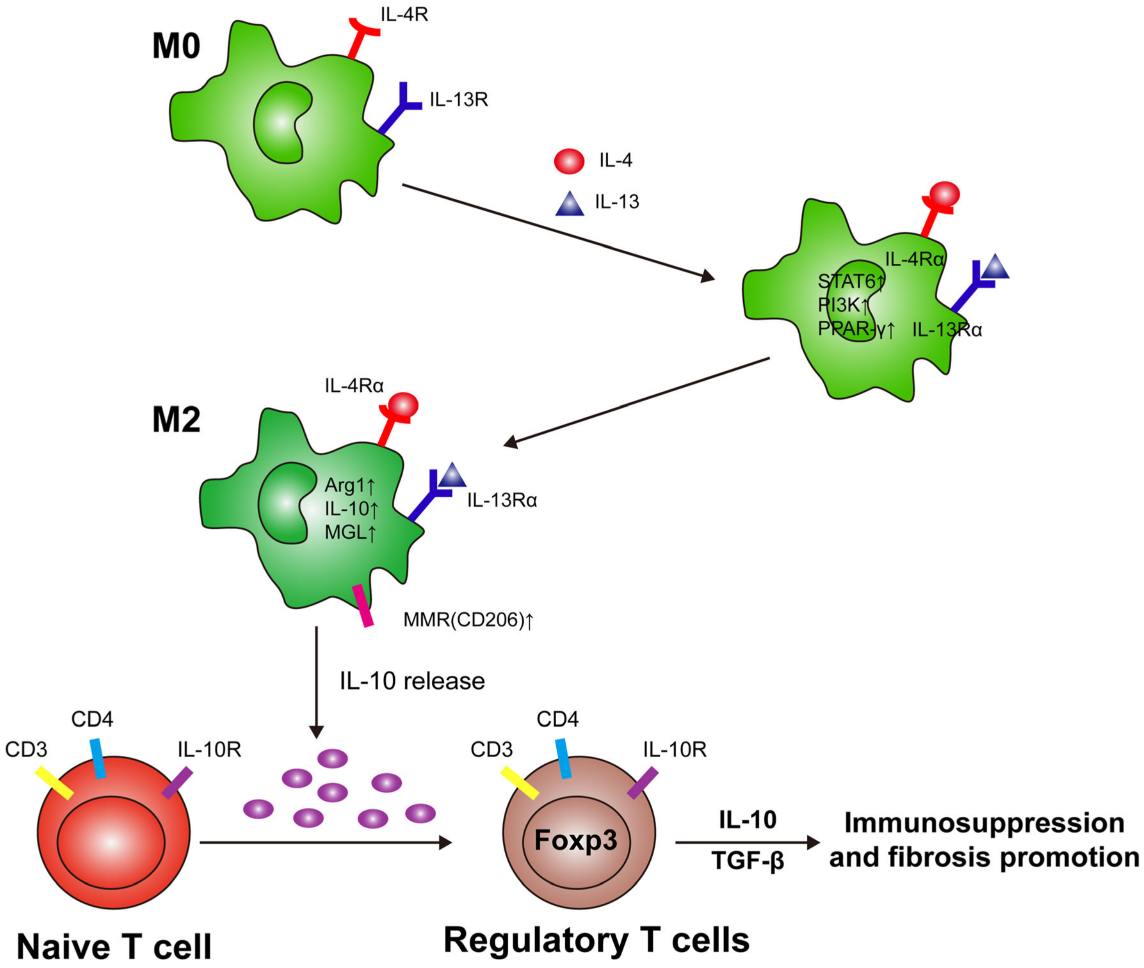

Fig. 2 M2 polarization function in MCD. IL-4/13 induces the transformation from M0 to M2 by activating the JAK/STAT6, PI3K, CEBP/β, and other pathways to increase the expression of M2 genes. Furthermore, M2 cells release IL-10, which can help activate Treg, which in turn release IL-10 and TGF-β to induce inflammation and promote fibrosis

MCD patients have arrhythmias; for example, Uemura et al. reported that 3 of 50 (6%) patients with unexplained atrioventricular heart block had myocarditis [48]. However, whether macrophages contribute to atrioventricular heart block in MCD needs to be further investigated.

Dendritic cells induce autoimmune myocarditis by increasing Th1 polarization

Ralph Steinman, who discovered DCs and their role in initiating the adaptive immune response, was awarded as the 2011 Nobel Prize in Physiology or Medicine. DCs are specialized sentinel cells that act as antigen presenting cells (APCs) and bridge the innate and adaptive immune systems without directly engaging in effectors activities, such as pathogen killing [22]. Through pattern recognition receptors, such as toll-like receptors (TLRs), DCs recognize pathogens and migrate to the T cell area of lymphoid organs to present pathogen-derived antigens to antigen-specific T cells.

DCs comprise two major classes: plasmacytoid DCs (pDCs) and conventional or classical DCs (cDCs). The pDCs are characterized by the surface markers CD123 and CD303 [23], while cDCs are characterized by the surface markers CD209, CD103, and CD141 [25, 26]. The pDCs rapidly produce type 1 interferon (IFN) following activation through nucleic acid-sensing TLRs, such as TLR7 and TLR9. cDCs are dedicated APCs that have a characteristic dendritic morphology and express high levels of MHC class II molecules. DCs play an important role in many inflammatory processes such as immune tolerance and autoimmunity. It has been reported that adoptive transfer of DCs loaded with cardiac antigen can induce the infiltration of $CD4^+$ T cells into cardiac tissue and the development of experimental autoimmune myocarditis (EAM), which indicates that DCs are sufficient to induce MCD [24]. CD40 is essential in triggering self-peptide-loaded DCs, and TLR simulation together with antigen-specific DCs has been shown to be required for disease induction [24]. Recently, Kania et al. found that IFN-γ, TLR, and nitric oxide (NO) signaling cooperated together to limit disease severity in EAM [49]. These data suggested that NO–mediated negative regulation of autoreactive T cells by tumor necrosis factor-α (TNF-α) and inducible NO synthase producing dendritic cells (TipDCs) occurs during autoimmune myocarditis. Together, factors-related infection or injury exposes the self-peptide and TLRs, which can then be taken up by and activate cDCs. Activated cDCs can induce Th1 polarization by releasing IL-12 to induce MCD (Fig. 3).Together, DCs play a role not only in antigen presentation, but also in the regulation of MCD. However, whether DCs are strictly necessary for the development of autoimmune myocarditis or just have a role in antigen mimicry-associated cardiac inflammation in MCD remains to be determined.

Natural killer cells can protect against myocarditis

Natural killer (NK) cells are a type of innate immune cell that characterized by the surface markers CD56 and CD94 (also called NKG2A) [27, 28].NK cells are a versatile subpopulation of lymphocytes that make essential contributions to human innate and adaptive immune responses to infection, especially viral infection [27]. In addition to their role as killer cells, NK cells can also regulate the cardiac inflammatory environment by inhibiting viral replication, autoreactive T cells, and ILC-derived Th2 cytokines, promoting eosinophil apoptosis and monocyte maturation, and inhibiting activated cardiac fibroblasts (Fig. 4).

In viral MCD, NK cells and macrophages are plentiful in the early infiltration of the heart after CVB3 infection [51]. They efficiently clear the virus by releasing perforin to kill infected cells and by stimulating adaptive immunity by producing IFN-γ [29]. Thus, NK cells were thought to suppress viral MCD by limiting viral replication by killing the virally infected cardiomyocytes; however, this process also results in the unavoidable release of autoantigens, which induce autoimmunity.

NK cells can also play an important role in regulating the inflammatory response by releasing cytokines to regulate other immune cells [52]. For example, NK cells can combine the cells surface receptors and cytokines to boost the maturation and activation of DCs, macrophages, and T cells. Conversely, NK cells can also kill immature DCs, activated CD4$^+$T cells, and hyperactivated macrophages to decrease the inflammatory response. These regulatory functions of NK cells are kept in check by the recognition of constitutively expressed self-molecules by means of inhibitory receptors. It is still unknown whether there is any difference between NK cell killing of cardiomyocytes and NK cell regulation of other types of immune cells.

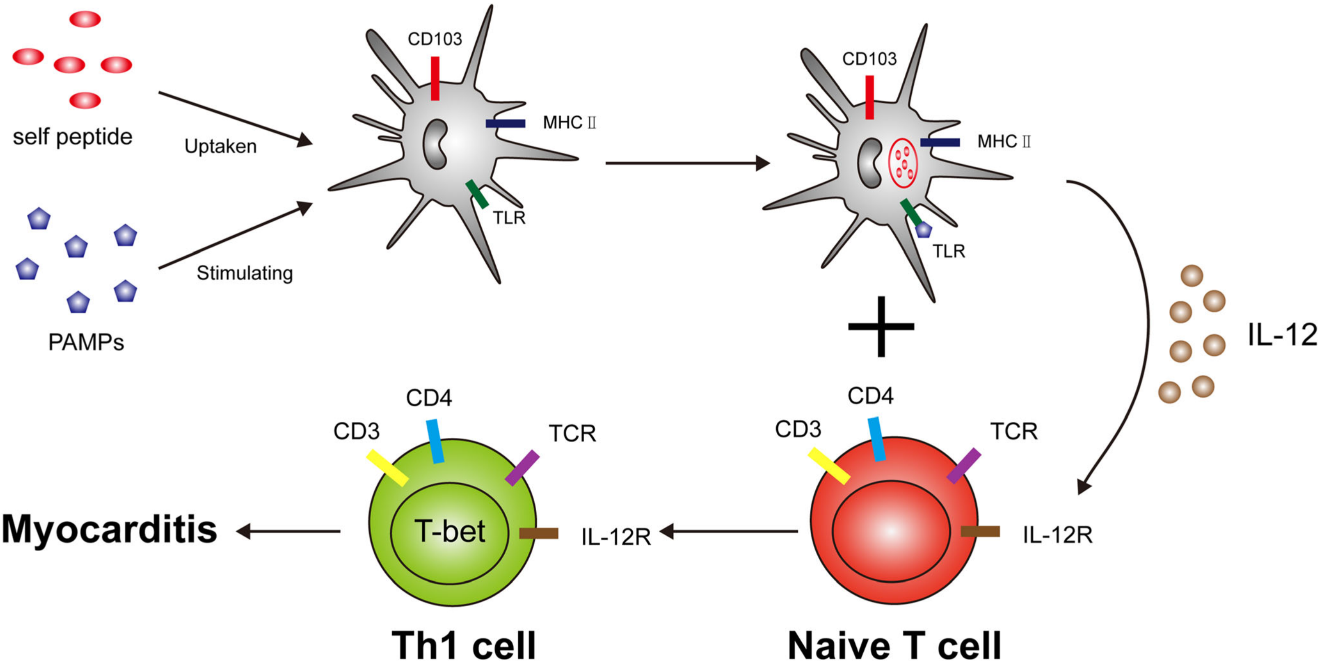

Fig. 3 cDCs can induce autoimmune myocarditis by inducing Th1 polarization. Self-peptide and PAMPs/DAMPs can induce the maturation of cDCs, which can combine with naïve T cells through MHC II and TCR and can induce Th1 cells with the help of IL-12 released by cDCs. Th1 cells can induce myocarditis [50]

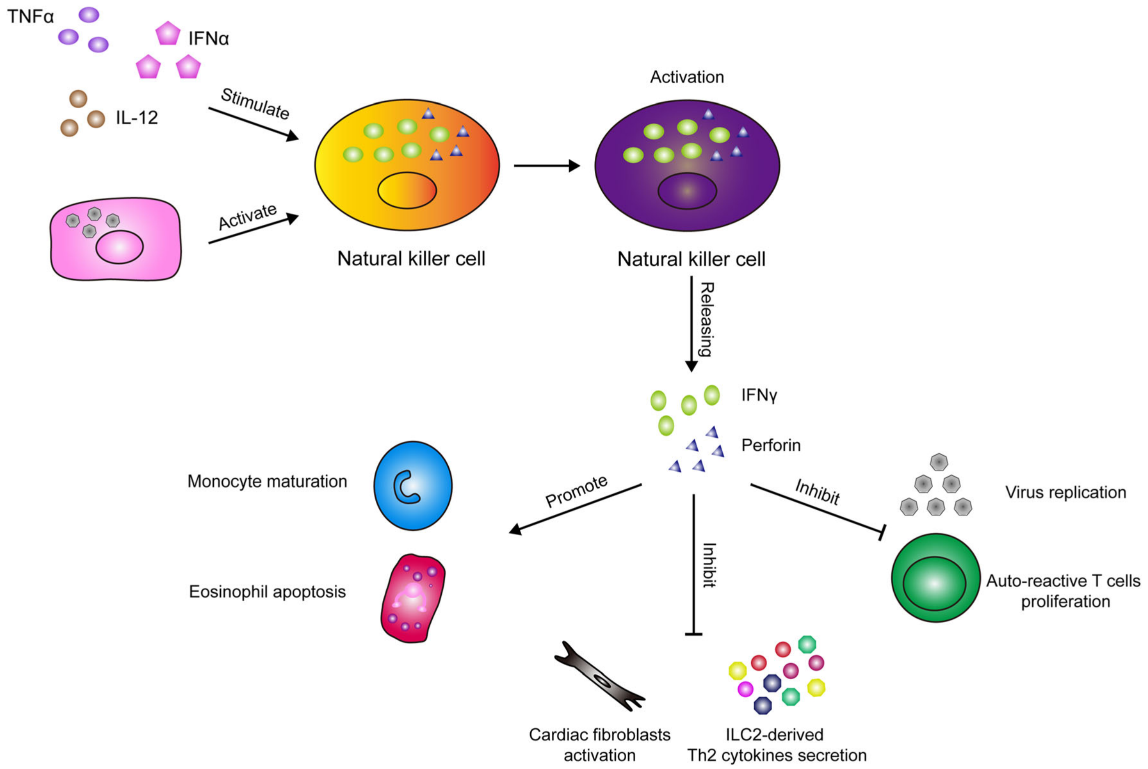

Fig. 4 Natural killer (NK) cells take part in MCD by releasing perforin and IFN-γ. NK cells can be activated by the TNF-α, IL-12, or IFN-α cytokines and infectious cells. Then, perforin and IFN-γ are released by activated NK cells to promote or inhibit several inflammatory responses

Mast cells play a critical role in the pathogenesis of myocarditis as inflammatory regulator

Mast cells are hematopoietic cells that arise from pluripotent precursors in the bone marrow. The distinguishing morphological feature of mast cells is their high content of electron-dense lysosome-like secretory granules (also known as secretory lysosomes) that occupy a major proportion of the cytoplasm of mature mast cells [30]. Mature mast cells are found in most tissues of the body and they are typically most abundant at sites close to host-environment interfaces, such as the skin and various other mucosal tissues. On account of the specific anatomical location, mast cells are thought as the first line of defense against external pathogens and attack by other environmental substances [53, 54]. Mast cells are characterized by proteases, including tryptases, chymases, and carboxypeptidase A3, which are different in humans and mice [30].

To investigate the role of mast cells in MCD (Fig. 5), Higuchiet al. used two strains of mast cell-deficient mice to verify that mast cells participate in the acute inflammatory reaction and the onset of ventricular remodeling associated with acute viral MCD [31]. These studies indicated that after constructing CVB3-induced MCD, the survival rate of mast cell-deficient mice was significantly higher than that of their control littermates (77% versus 31%; $p = 0.03$; $n = 13$). Additionally, myocardial necrosis and cellular infiltration were significantly less pronounced in mast cell-deficient mice than in their control littermates. Furthermore, the expression levels of mast cell proteases were upregulated in the acute phase of viral MCD and rose further in the subacute phase of heart failure. The activation coincided with the development of myocardial necrosis and fibrosis and correlated with the upregulation of the expression of matrix metalloproteinase (MMP)-9 [31]. Thus, granule compounds released by mast cells can affect processes that are relevant to extracellular matrix (ECM) remodeling by activating ECM-remodeling enzymes.

Mast cells can also contribute to the immune regulation of Th2 polarization. In recent studies, these results showed that after adoptive transfer of immune cells, the mast cell-deficient mice showed stronger Th2 responses in the intestine than control mice. Mechanistic studies showed that mast cell-derived mouse mast cell protease-6 increased the expression of Bcl-6 in Th2 cells. Bcl-6 inhibited the expression of GATA-3 in Th2 cells, and subsequently, forkhead box P3 (Foxp3) was increased and Th2 cytokines were reduced. Thus, the cells showed immune regulatory properties similar to regulatory T cells [55]. Based on this research, we can speculate that mast cells may contribute to the pathogenesis of MCD in conditions in which MCD is characterized by a type 2 immune response

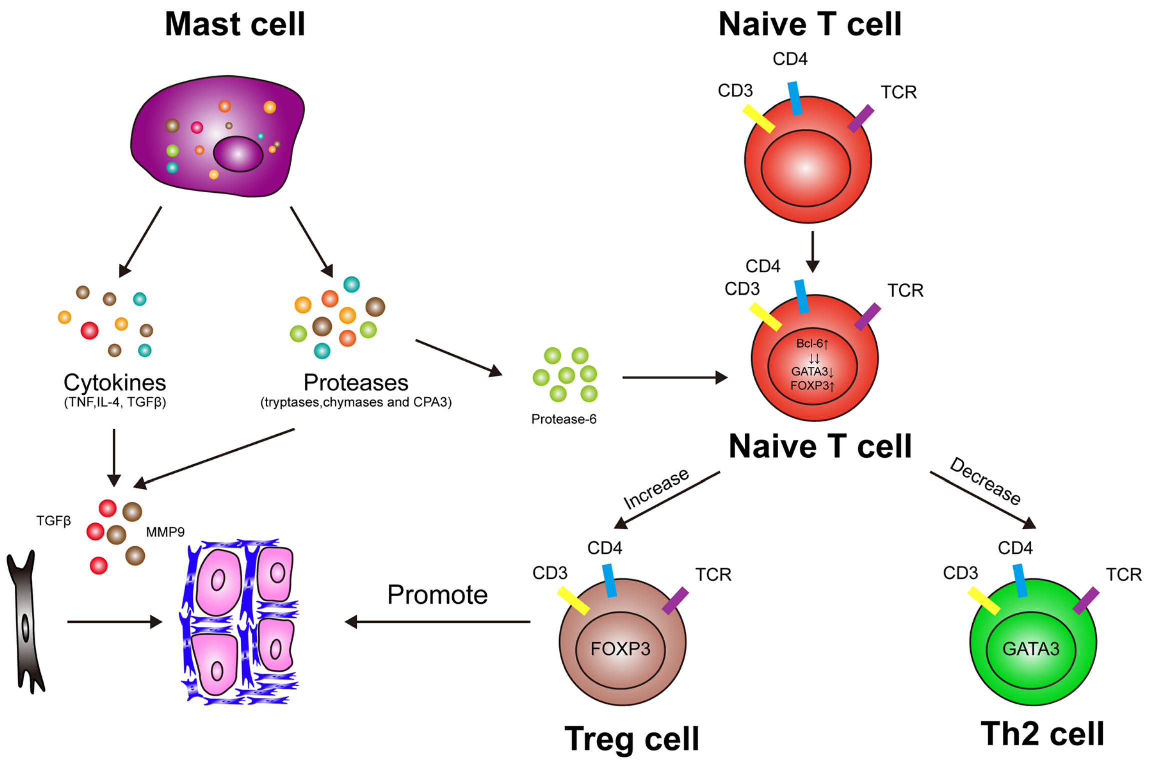

Fig. 5 Mast cells contribute to the process of EAM in fibrosis and reduce Th2 polarization. Mast cells are characterized by granule constituents including mast cell-specific proteases and cytokines. MMP9 and TGF-β contribute to the progression of fibrosis. Protease-6 can change the transformation of naïve T cells to Treg cells than to Th2 cells. Treg cells further promote fibrosis development

[33, 56]. However, this hypothesis needs to be investigated further. For the role of mast cells in ECM remodeling leading to fibrosis, which is the most characteristic feature of the process from MCD to cardiomyopathy [57, 58], it is necessary to further investigate the role of mast cells in the chronic myopathy phase.

Role of CD4$^+$ T cells in myocarditis as major immune cell effectors

As many studies have reported, autoreactive and most often infection-triggered CD4$^+$ T cells are confirmed to be critical for MCD induction [2, 59]. According to the surface markers on T cells, there are two major subsets of T cells, CD4$^+$T cells, and CD8$^+$T cells (Fig. 6) [59]. Most of the T cells that are positive for the CD4$^+$surface marker are defined as the main drivers of heart-specific autoimmunity in MCD [60–62]. Expansion of heart-specific CD4$^+$T cell effectors is facilitated in human and mice due to a high frequency of circulating naïve α-MyHC-specific CD4$^+$T cells. CD4$^+$T cells are also known as T helper (Th) cells and include Th1, Th2, Th17, and other T helper cells, aside from the CD4$^+$ T cell subsets that include regulatory T cells (Tregs) [63]. Many studies, most of which are based on mouse models, indicate an exclusive role for CD4$^+$T cells in MCD development and progression. Unfortunately, the regulatory roles of the different CD4$^+$T cell subtypes in MCD depend highly on the stage of disease and on a complex and not yet understood interaction between different inflammatory heart infiltrating and heart resident cell types.

With regard to Th cells, MCD is considered type 2 immunity so Th2 cells contribute to the pathogenesis of MCD [33, 35]. Song et al. found that besides tissue damage, a lower expression of VDR, a high frequency of Th2 cells, and an increase in Th2 cytokines were characteristics of MCD patient hearts during the end stages of heart failure. Spontaneous Th2-biased inflammation was observed in the hearts of VDR$^{-/-}$ mice. CD4$^+$ T cells from VDR$^{-/-}$ mouse hearts were in a highly activated state. The naïve VDR$^{-/-}$ CD4$^+$ T cells and naïve CD4$^+$ T cells from human hearts with MCD were prone to differentiate into Th2 cells. VDR formed complexes with GATA3, the IL-4 transcription factor, to suppressIL-4 gene transcription. Transplantation with VDR$^{-/-}$ CD4$^+$T cells induced Th2-biased inflammation in the hearts of Rag2$^{-/-}$ mice. Reconstitution of VDR in CD4$^+$ T cells inhibited Th2-biased inflammation in the heart.

MCD is a potentially lethal inflammatory heart disease that frequently leads to dilated cardiomyopathy (DCM); however, it is unclear which type of Th cells contributes to this

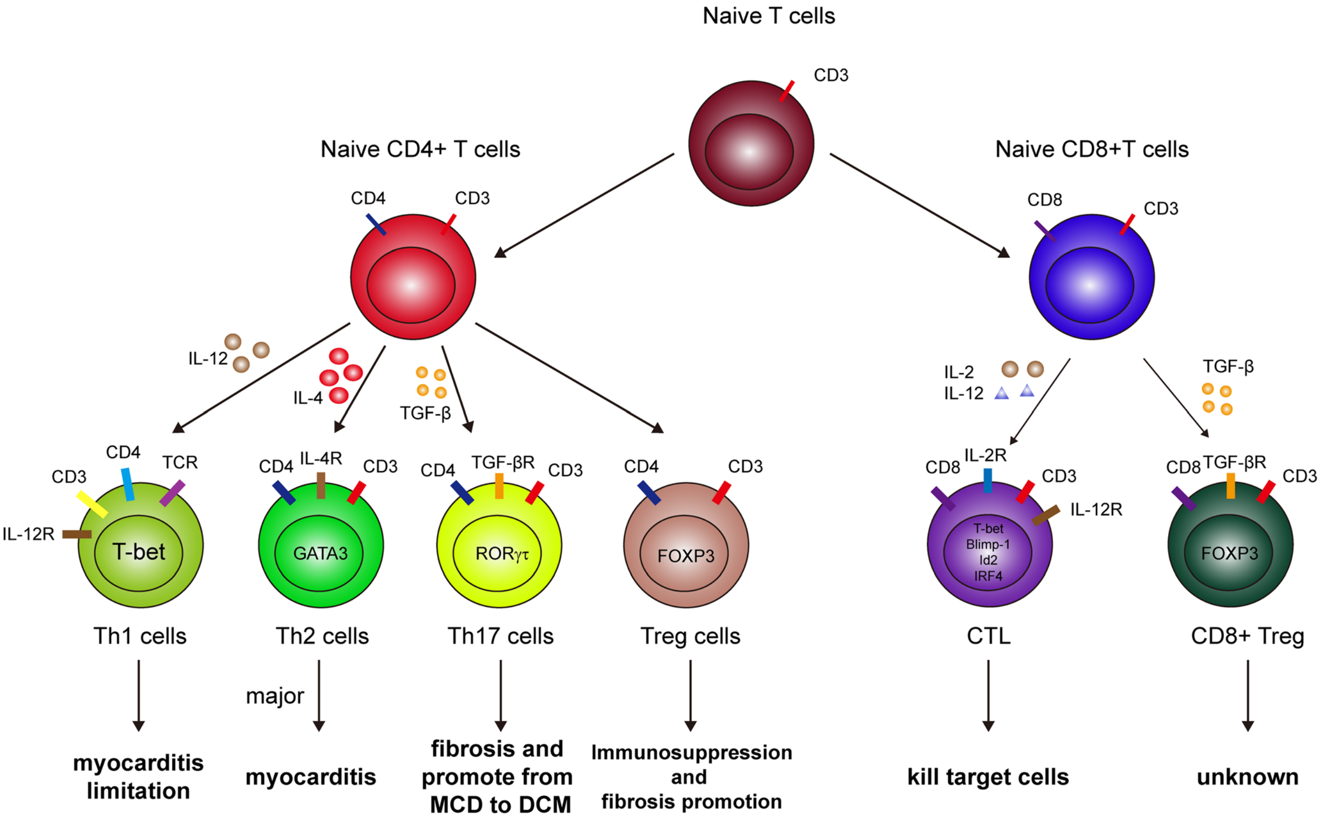

Fig. 6 Development of differentiated T cells and their roles in MCD. T cells are divided into two major types according to their surface markers (CD4 or CD8). Naïve CD4$^+$ T cells can be divided into four types of cells during the accumulation of different cytokines from the innate cells, especially antigen presenting cells (APC). Naïve CD8$^+$ T cells can also be induced by cytokines from APC

progression. Veronika Nindl et al. found that cooperation of Th1 and Th17 cells determines the transition from autoimmune MCD to DCM [34], that IFN-γ was the major effector cytokine driving the initial inflammatory process, and that the cooperation of IFN-γ and IL-17A was essential for the development of progressive disease. Nevertheless, some contradictory findings have also been reported. For example, after immunizing IFN-γ- and IFN-γR-deficient mice with α-MyHC/CFA, the mice developed more severe and persistent MCD, suggesting a protective regulatory role for IFN-γ in this disease model [36, 37].However, which type of Th cells contributes to the troublesome progression from MCD to DCM still needs to be investigated.

Treg cells are a specific subpopulation of T cells that comprise 5–10% of all peripheral CD4$^+$T cells [32]. Treg cells are characterized by the surface markers CD4$^+$, CD25$^+$, and FOXP3$^+$. FOXP3 is a specific marker of Treg cells that is essential for their maturation and function, so a loss of FOXP3 can result in disabled Treg cell function [2]. Treg cells can execute their immunosuppressive activity against a broad and diverse array of antigens within different microenvironments by producing inhibitory cytokines. Masahiro Ono et al.

Table 2 The methods of immunomodulation in myocarditis

Immunomodulatory drugs	Functional mechanism	Reference
High-dose intravenous immunoglobulin (IVIG)	Neutralization of autoantibodies; neutralization of chemokines and/or cytokines; Saturation and modulation of the expression of Fcγ receptors	[67, 68]
Peptide-ligands	Specify epitope-derived peptides as antibody-scavengers; direct targeting/suppression of aab-producing B cells and/or plasma-cells	[69, 70]
Immunoadsorption	IA of disease-causing aabs	[71–73]
Immunosuppression (azathioprine, steroids, cyclosporine A)	Widespread inhibitory effects on the immune system and action through a variety of signaling pathways; immunotoxicity	[74, 75]

found that GITR^{high} and FOXP3-expressing natural Tregs, made up of both CD25^{+} and CD25^{-}cell populations, contributed to the prevention of a variety of autoimmune/inflammatory diseases and that depletion of these cells allowed the activation of even weaker and rarer autoreactive T cells, causing widespread severe autoimmune disease [38]. However, diseases induced in this way include many which have been suspected of an autoimmune etiology in humans without much evidence.

Role of CD8^{+} T cells in myocarditis as cytotoxic lymphocytes

CD8 + T cells are referred to as cytotoxic T lymphocytes (CTLs), which were first reported in the 1960s to show specific cytotoxicity in vitro using mostly allogeneic combinations of cells [39]. Mechanistic studies suggest that CTLs most often kill target cells by inducing signaling through the same apoptotic pathways that are used for developmental cell death, including the perforin–granzyme pathway and the FAS pathway.

Additionally, various subgroups of CD8 + T lymphocytes demonstrate not only cytotoxic effects but also serve memory and regulatory roles in the body's immune response [40]. Massilamany et al. found that the CD8 + T cell response induced by Myhc338–348 was antigen-specific and transferred the disease to naïve animals, as evaluated by MHC class I/H-2Dd dextramer staining. Furthermore, the antigen-sensitized T cells predominantly produced interferon-gamma, the critical cytokine of effector cytotoxic T lymphocytes in EAM [41]. However, the role of regulatory CD8+ cells in MCD was unknown (Fig. 6).

B cells contribute to the autoimmunity process of myocarditis through autoantibody production

As mentioned intensively, autoimmunity contributes to the pathogenesis of MCD. In this condition, autoantibodies are key factors produced by B cells, which are characterized by CD19/CD20 [16]. Yoh Matsumoto et al. examined whether the cardiogenic epitopes that reside in the cardiac C-protein fragment2 (CC2) could be successful in inducing MCD and the development of DCM, compared with cardiac C-protein fragment 2 peptide 12 (CC2P12). They found the CC2-immunized rats exhibited marked B cell epitope spreading 4 weeks after immunization, whereas CC2P12-immunized rats raised antibodies against CC2P12 and CC2, but there was no difference in CC2- andCC2P12-immunized rat groups [42]. Then, they performed transfer experiments and demonstrated that both activation of T cells and anti-peptide antibody elevation were required for the initiation and subsequent progression of the disease. These findings strongly suggest that B cell epitope spreading is an essential step for the switch from MCD to DCM. The mechanism of how B cells identify the self-antigen to releasing autoantibody still needs further investigation to decrease the production of autoantibody.

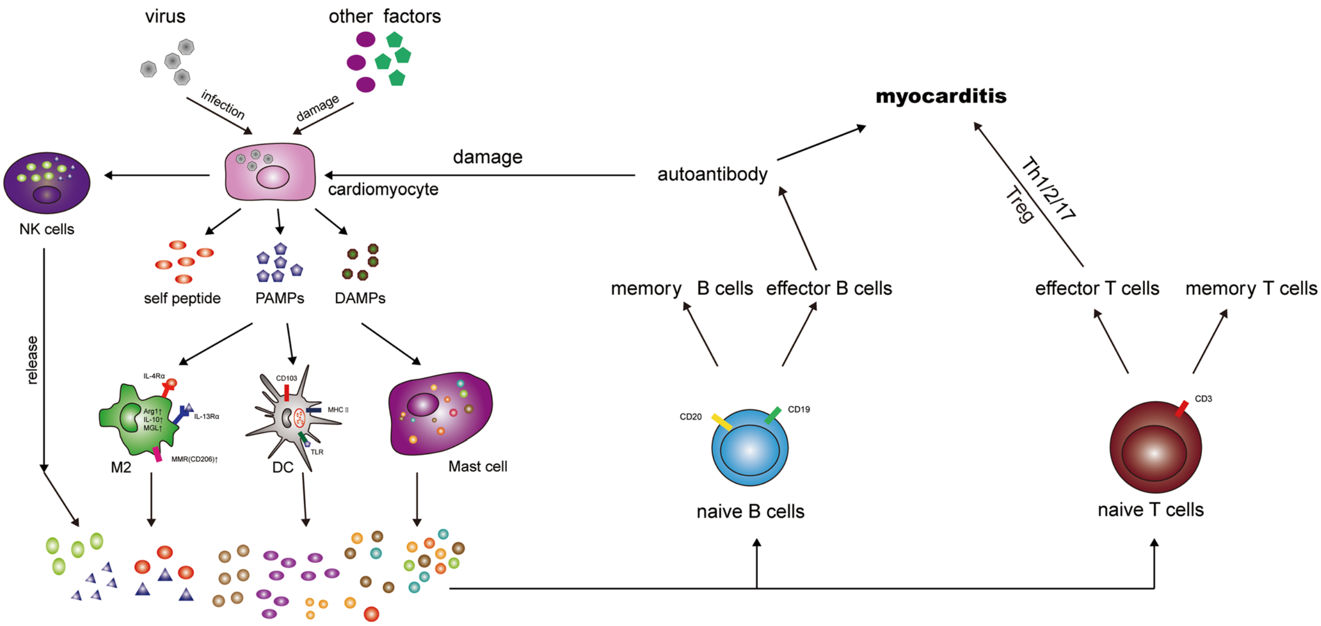

Fig. 7 The immune network in the pathogenesis of MCD

The prospects of immunotherapy in myocarditis

Immunotherapy has been advanced in cancer therapy and this has brought much hope for the patients who are suffering from cancer. In the field of cancer immunotherapy, adopting such methods regulates the immune system to inhibit the proliferation of tumors, such as enhancing T cell ability, because $CD8^+$ T cells can clear up tumor cells [64]. Currently, almost all of the therapeutic strategies focus on promoting the ability of CTLs within a tumor, the type of $CD8^+$ T cells, help in priming tumor-specific CTLs in the lymphoid organs and establishing efficient and durable antitumor immunity. With the advance of immunotherapy in tumors, immune checkpoint inhibition, such as the PD-1 antibody, can enhance the activity of antitumor T cells (majority CTLs) to clear up the tumor cells as therapy for cancer [65]. However, MCD is different from the cancer in the immune system, in that cancer may be the result of the exhaustion of T cells, while on the contrary, MCD may result from the abnormal activation of T cells to impair cardiomyocytes. Thus, the immunotherapy strategies used in cancer, which would have an opposite effect on MCD, are not appropriate for MCD. It has been reported the immune checkpoint inhibitors can induce MCD unexpectedly [66]. As there is abnormal activation of immune cells in MCD, some immunomodulation methods can be used for MCD (Table 2). For example, immunosuppressants have been applied to treat MCD with good outcomes in virus-negative cases without a known specificity of immune cells [76]. Additionally, it has been reported that TCR-based immunotherapy, which could be the future direction of MCD therapy, can be applicable to MCD, not only in animal but also in human MCD whose pathogenic mechanism is poorly understood [77]. As PD-1 can protect against inflammation and cardiomyocyte damage in T cell-mediated MCD [78], maybe increasing the expression of PD-1 induced by drugs or biosynthesis could help to treat MCD in the future.

Conclusions

Myocarditis is a type of inflammation of the heart involving innate and adaptive immune cells as shown in Table 1; it can lead to dilated cardiomyopathy. Different types of immune cells contribute to the development of MCD, and there is a complex network of inflammation (Fig. 7). Understanding the specific roles of these innate and adaptive cell populations at different stages of disease progression might provide the key to our insight into the pathogenesis of MCD as a base for developing potential therapeutic strategies. Immunotherapy has a huge potential role in the treatment of MCD, and further investigation is needed to accurately utilize immunotherapy on special cell types.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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