

Insight into the Role of the Immune System in Atherosclerosis and Related Immune Therapy Strategies

Yiyang Li *

Harrow International School Beijing, Beijing, China

* Corresponding Author Email: quli@harrowbeijing.cn

Abstract. Atherosclerosis is identified as the most common reason of numerous cardiovascular diseases (CVD), which is a chronic disease caused by the low-density lipoprotein (LDL) accumulate together and form a plaque. One of the factors contributing to that is the inflammatory response which is related to various signaling pathways. Both innate immune cells and adaptive immune cells participate in inflammatory responses, which lead to atherosclerosis. Their specific roles in the disease are explained in detail respectively in this project. In order to mitigate overactivated inflammatory responses in atherosclerosis, lots of research and studies have been made to find the potential treatment. Various antagonists which target interleukin 6 (IL-6) or interleukin-1 (IL-1) have been evaluated and applied in clinical treatment for atherosclerosis. In addition, monoclonal antibodies are also developed to target oxidized LDL. Despite the progress on the current therapy for atherosclerosis, more research on the field is needed due to the limit efficiency and adverse effects.

Keywords: Atherosclerosis, Inflammation, Immune System.

1. Introduction

The buildup of cholesterol, especially LDL, in blood vessels leads to atherosclerosis, a prevalent chronic illness [1]. The cholesterol deposits are highly associated with the development of plaque, which could lead in the narrowing of the blood vessel and obstructed blood flow. As a result, some CVD occurs, such as coronary artery disease, angina, myocardial infarction and aneurysms. Additionally, the inflammatory response is a significant factor that could result in atherosclerosis, which directly related to the multiple over-activated immune cells. The damaged endothelium of blood vessel could secrete various pro-inflammatory cytokines in order to recruit immune cells, leading to inflammation. Then, the fatty deposit could be accumulated in the injured lining. After that, more fibrous tissue and calcium salts build up the blood clot to block the wound and then causes more erythrocytes and leukocytes to attach to the blood clot. This process led to the cholesterol accumulation as well, which caused the plaque and finally cause the atherosclerosis.

The pathogenesis involves both innate immune cells and adaptive immune cells. The cells that first defend the body are the non-specific immune cells, such as dendritic cells (DCs), macrophages, and neutrophils [2]. They directly react to the pathogen that entered the human body and kill them through phagocytosis. Lymphocytes play regulatory role in the atherosclerosis as well.

T cells could both secrete different cytokines and directly kill the pathogens when B cells are mainly responsible to producing antibodies. Due to the critical role of inflammation in atherosclerosis, several cytokines are widely studied as the potential therapeutic target.

In this project, functions of innate immune cells and lymphocytes in atherosclerosis will be illustrated in details. According to the mechanisms, several pro-inflammatory cytokines antagonists will also be described in the following sections.

2. Innate immune system and atherosclerosis

2.1. Macrophage

Innate immune system serves as cellular sentinel and is the first line of human body. However, the overactivation of innate immune system also could result in multiple diseases. Recent studies

highlighted the vital role of innate immune cells in vascular inflammation and further atherosclerosis. Endothelial dysfunction and LDLs clustering is the most causes of the beginning of atherosclerosis. Furthermore, the change of endothelial permeability and the upregulated cell adhesion molecules result in the immune cells influx. Macrophages, DCs and neutrophils attend this process as the critical immune mediators. In this section, the role of three innate immune cells in atherosclerosis will be described respectively. The significant causing factor of atherosclerosis is the plaque that is made by the accumulation of cholesterol in human body. At the same time, the innate immunocytes such as monocytes and macrophages would infiltrate into artery wall and the plaque. Selectin, intercellular adhesion molecule-1 (ICAM-1), and vascular cell adhesion molecule-1 (VCAM-1) are a few of the cell adhesion molecules that are overexpressed and are responsible for this inflow. Cytokines and chemokines are also involved in the immunocytes regulation. C-C chemokine receptor 2 (CCR2) and C-C motif chemokine ligand 2 play critical role in monocyte emigration from circulation to plaques. The recruited monocyte next differentiates to macrophages and further regulate T cells. As pro-inflammatory phenotype, M1 macrophage is the cell that assemble a lot to cause the inflammatory response in human blood vessel to further prompt the atherosclerosis around endothelium wound [3]. Additionally, the foam cell, the macrophages that contain cholesterol, is also an important factor to cause atherosclerosis [4]. It can be seen in Figure 1, the LDLs are easy to be oxidized and modified, then taken by the scavenger receptors of macrophages to form foam cells. The accumulation of these foam cells can lead to the production of plaques. In addition to the scavenger receptors, many studies also demonstrate that the toll-like receptors (TLRs) also are involved in atherosclerosis. Moreover, activated macrophage can secrete pro-inflammatory mediator to present to other immune cells to prompt the immune response in human blood vessel.

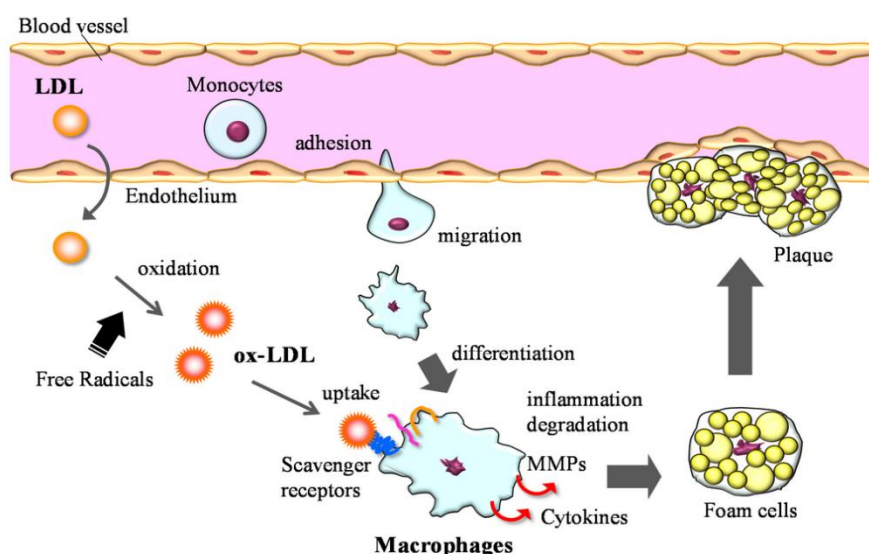


Figure 1. The schematic of macrophages causing atherosclerosis [5].

2.2. DCs

The usual antigen-presenting cells are dendritic cells (APCs). To identify the roles of macrophages and DCs in atherosclerosis, researchers conducted extensive research. DCs' most important function is to present antigens to the leukocytes' receptors, leading to the activation of antigen-specific T cell in lymph nodes, while macrophages are impressed by phagocytic ability. Unlike macrophages, DCs exist in healthy blood vessel, but are upregulated in atherosclerotic lesions. It is remarkable that mostly DCs are locally involved in atherosclerosis in the plaques. Naïve DCs are resident in the vessel before exposure for antigen. The natural and modified LDLs are the main mediator to induce mature DCs which then migrate to regulate T cells. The activated DCs normally are found to cluster with recruited T cells. Since now, the mechanism of DCs to attend atherosclerosis is still not clear, and their specific function need to be further investigated [6].

2.3. Neutrophils

Neutrophils play a critical role in cause inflammatory response to kill invading pathogens in human body. They are non-specific and able to initiate the inflammatory response by secrete a huge amount of pro-inflammatory mediator [7]. These pro-inflammatory cytokines next can present to other immune cells and further cause inflammatory response. In the past, the neutrophils were considered as atherosclerosis irrelevant factor. However, more and more studies revealed that neutrophils are also involved in the progress as a key regulator. The cytokines from neutrophils could directly relate to the endothelium injury. Neutrophils can also be found in the lesion areas associated with hyper-inflammatory response. Except from similar function to monocytes, neutrophils also produce granule proteins to prompt atherosclerosis and upregulate macrophage and foam cells. In a word, understanding the role of neutrophils in atherosclerosis is vital to develop potential therapy method.

3. Adaptive immune system and atherosclerosis

3.1. T cell

The adaptive immune system is usually activated later than the innate immune system. However, the specific adaptive immune system can target to the specific pathogen or infected cells to eliminate the infection. Lymphocytes are the main members in adaptive immune system. Meanwhile, the over-activated B and T lymphocytes can connect to the cause of autoimmune diseases, including atherosclerosis. B cells and T cells are primary lymphocytes, which highly associated with atherosclerosis through different mechanism. One of the important factors causing atherosclerosis is the T lymphocytes, which are highlight in the pathogenesis of atherosclerosis by multiple studies. T cells can be recruited into the lesion, which would be related to the production of the pro-atherogenic mediators. The T cells related immune responses are involved in the injury and lesion expansion and are further related to the disease deterioration. An abundance of T cells can be found in the plaque in atherosclerosis patients' artery walls, indicating the regulating role of T cells in this disease through complex inflammatory responses [8]. This phenomenon also provides evidence that T cells have infiltrated into the blood vessel wall to prompt immune progress in the plaque. In addition, seral studies demonstrated that T cells have the ability to mediate this disease through binding to peptide epitope.

After naïve T cell activation, they can upregulate the expression of different biomarkers and are divided into seral subpopulations which have specific functions. CD4⁺ T cells are recognized as the predominate lymphocyte type in the development of atherosclerosis when CD8⁺ T cells are linked to the pathophysiology by promoting cell death. One of the most prominent subsets of CD4⁺ cells are T helper (Th) cells [9]. Th cells can further differentiate into Th1, Th2 and Th17 phenotypes. Th1 cells are critical in the pro-inflammatory responses while Th2 cells are highlighted in the anti-inflammatory progress. The imbalanced Th1 and Th2 proportion is crucial risk factor of autoimmune diseases including atherosclerosis. Secreting pro-inflammatory cytokines including interferon- γ (IFN- γ), interleukin-2 (IL-2) and tumor necrosis factor (TNF) are the mechanism of Th1 cells regulating immunity in atherosclerosis. Inflammatory M1 macrophage could be further stimulated by these cytokines and enhance the pro-inflammatory pathways in atherosclerosis. In contrast, the mechanism of Th2 regulating atherosclerosis is still remain uncertain [9]. However, studies have been demonstrated that Th2 can secret anti-inflammatory cytokines to modulate the hyperinflammation in the autoimmune disease, such as interleukin-4 (IL-4) and interleukin-10 (IL-10). Additionally, Th17 cells are also an important subtype of Th cells, which are mainly served to secreting interleukin-17 (IL-17) to prompt immune responses in atherosclerosis.

In addition to Th cells, T regulatory (Treg) cells are also highly involved in the modulating of immunity in atherosclerosis. IL-10 and transforming growth factor beta (TGF- β) are the main cytokines produced by Treg cells. Treg cells could cause the deactivation of specific innate immune cells to mitigate the disease.

3.2. B cells and antibodies

Previous studies identified multifunction of B cells in the immunity in atherosclerosis through genome wide association and transcriptome sequencing. As a critical modulator in antigen-specific and anti-atherogenic humoral immunity, by increasing the expression of major histocompatibility complex class II (MHC II), B cells can act as APCs and promote CD4⁺ T cell development [10]. Also, B cells under activated status are significant risk factors for cardiovascular disease (CVD). However, recent research suggests that B cells act as a protective role in atherosclerosis by secreting multiple antibodies to neutralize oxidized LDLs, when the major causes of inflammatory response is neo epitopes on LDL, and the dead cells which can be recognized by natural antibodies. Moreover, oxidation-specific epitopes are very significant to atherosclerosis and it is the target for both natural and adaptive antibodies.

4. Potential immunotherapy strategies for atherosclerosis

In the therapy of atherosclerosis, there are three main aims which are known as anti-inflammatory response, immunization against antigens, and preventing the synthesis of cholesterol. Immunotherapy for atherosclerosis is being developed as a result of recent studies on the innate immune system and the adaptive immune system.

Multiply immune cells are responsible for secreting IL-6, including Th2 cells, monocytes and macrophages. Some other cells like endothelial and fibroblasts are also involved in the IL-6 signal pathways to regulate the immune responses in patients' bodies. The downstream inflammation is normally associated with the upregulated IL-6. IL-6 signaling pathways play a vital role in the pathogenesis of atherosclerosis. As a result, IL-6 is considered as a candidate of the target for immunotherapies to atherosclerosis. Tocilizumab is a humanized monoclonal antibody and tocilizumab-related medicines has been approved by the U.S. Food and Drug Administration (FDA) to treat atherosclerosis patients through blocking IL-6 [3]. However, the IL-6 antagonists are still need to be widely studied due to the limited efficiency. Some studies also indicate that direct inhibition of IL-6 could lead to adverse effects, such as raising the LDL level. Increasing LDL in the blood could neutralize the presumed athero-protective effect. Furthermore, some research shows that injecting low-dose methotrexate may be a therapy approach as it decreases several inflammatory biomarkers including IL-6, C-reactive protein and TNF- α . And it has been demonstrated that that methotrexate will not affect lipid levels or platelet function in the human blood. Additionally, IL-1 is an apical pro-inflammatory mediator. IL-1 can significantly encourage the development of atherosclerosis. Therefore, IL-1 β have been identified as a potential therapeutic target. Studies show that several agents that could inhibit IL-1 have been studied and available, including the IL-1 receptor antagonist, anakinra, and a type of IL-1 trap which named rilonacept, as well as an antibody of IL-1 which is known as canakinumab [7]. Apart from the pro-inflammatory cytokine's antagonists, specific antibodies are also applied to neutralize pathogenic antigens in order to mitigate atherosclerosis. Oxidized LDL is highlighted in the pathogenesis of atherosclerosis as an autoantigen. A specific immunoglobulin M antibody has been produced to neutralize the oxidized LDL in mice with atherosclerosis [11]. In this way, plaque accumulation is largely mitigated, which contributes to a high cardioprotective efficiency.

In addition, tolerogenic vaccination has been studied as a potential therapy to prevent the progress of immunity in atherosclerosis over the past 5 decades (Figure 2). The LDL-specific Treg cells could be loaded in the vaccines to suppress the inflammation in mice body.

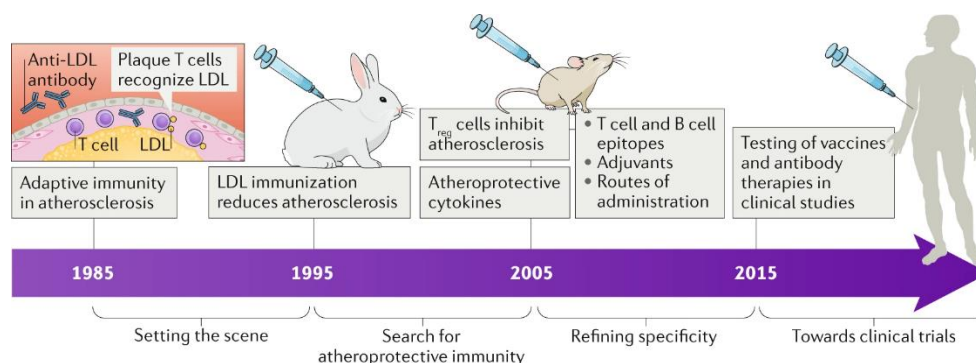


Figure 2. The timeline describes the development of the tolerogenic vaccination for atherosclerosis in the past 5 decades [12].

5. Conclusion

Atherosclerosis is identified as an autoimmune disease. The accumulation of LDL is mainly contributed to the disease. Atherosclerosis is highly associated with multiple CVD, which is the main cause of heart failure. As a result, finding an effective treatment for atherosclerosis is energy for human. Atherosclerosis has a very close connection to the immune system. Inflammation is the main factor contribute to atherosclerosis. In healthy human body, immune cells are responsible to protect the host from pathogens. Neutrophils play a critical role in causing inflammatory responses due to its ability to kill invading pathogens. Activated macrophages also are able to clear pathogens through phagocytosis. However, overactivated immune responses could be caused by the endothelium wound and the accumulated cholesterol there. As a result, the blood vessel could be blocked by the formation of plaque. Besides, LDLs migrate into the intimal section on blood vessel and be oxidized to form oxidized LDL, which is the main autoantigen to cause the immune responses. The degenerative development of atherosclerosis primarily confines both innate immune cells and adaptive immune cells. T cells, DCs, and microphages all contribute significantly to the inflammation.

Based on the understanding of immune cells in atherosclerosis, several immunotherapy approaches have been widely studied to treat atherosclerosis. IL-6 and IL-1 are pro-inflammatory cytokines and critical to induce downstream inflammation which is highlighted in the atherosclerosis. Given that, multiple antagonists target them has been learned a lot. For example, tocilizumab is classified as a monoclonal antibody to block IL-6 signal and has been approved by FDA, although with some unwanted effect. In addition, tolerogenic vaccination also were discussed as a potential therapy to prevent the progress of immunity in atherosclerosis. Despitess the development of immunotherapy of atherosclerosis, researchers also face a few difficulties. The huge gap between the animal models and clinical application is the biggest one.

References

- [1] Ley K. Role of the adaptive immune system in atherosclerosis[J]. *Biochemical Society Transactions*, 2020, 48(5): 2273-2281.
- [2] Hermansson A, Johansson D K, Ketelhuth D F J, et al. Immunotherapy with tolerogenic apolipoprotein B-100-loaded dendritic cells attenuates atherosclerosis in hypercholesterolemic mice[J]. *Circulation*, 2011, 123(10): 1083-1091.
- [3] Chávez-Sánchez L, Espinosa-Luna J E, Chávez-Rueda K, et al. Innate immune system cells in atherosclerosis[J]. *Archives of medical research*, 2014, 45(1): 1-14.
- [4] Sage A P, Tsiantoulas D, Binder C J, et al. The role of B cells in atherosclerosis[J]. *Nature Reviews Cardiology*, 2019, 16(3): 180-196.
- [5] Kishimoto Y, Yoshida H, Kondo K. Potential anti-atherosclerotic properties of astaxanthin[J]. *Marine Drugs*, 2016, 14(2): 35.

- [6] Roy P, Orecchioni M, Ley K. How the immune system shapes atherosclerosis: roles of innate and adaptive immunity[J]. *Nature reviews Immunology*, 2022, 22(4): 251-265
- [7] Moriya J. Critical roles of inflammation in atherosclerosis[J]. *Journal of cardiology*, 2019, 73(1): 22-27.
- [8] Hansson G K, Hermansson A. The immune system in atherosclerosis[J]. *Nature immunology*, 2011, 12(3): 204-212.
- [9] Saigusa R, Winkels H, Ley K. T cell subsets and functions in atherosclerosis[J]. *Nature Reviews Cardiology*, 2020, 17(7): 387-401.
- [10] Legein B, Temmerman L, Biessen E A L, et al. Inflammation and immune system interactions in atherosclerosis[J]. *Cellular and Molecular Life Sciences*, 2013, 70(20): 3847-3869.
- [11] Binder C J, Silverman G J. Natural antibodies and the autoimmunity of atherosclerosis[C]. *Springer seminars in immunopathology*. Springer-Verlag, 2005, 26(4): 385-404.
- [12] Hansson G K, Nilsson J. Developing a vaccine against atherosclerosis[J]. *Nature Reviews Cardiology*, 2020, 17(8): 451-452.