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REVIEW ARTICLE

Research progress on the role of sialic acid-binding immunoglobulin-like lectin 9 in various diseases

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ABSTRACT

Sialic acid-binding immunoglobulin-like lectin 9 (Siglec-9) is a receptor that expresses on the surface of immune cells. It plays an important role in the body's immune response. Increased expression of Siglec-9 has been reported in infectious diseases, autoimmune diseases and cancer. Pathogenic microorganism and tumor cells can inhibit the recognition and killing of immune cells by upregulating their own specific sialic acid and binding with Siglec-9 on the surface of host immune cells, and suppress the release of pro-inflammatory cytokines and promote the release of anti-inflammatory cytokines, eventually leading to immunosuppression, tumor immune escape and the like. However, the immunosuppressive function of Siglec-9 may be advantageous for diseases such as neutrophil asthma and autoimmune diseases. Therefore, further research on the mechanism of action of Siglec-9 is of great significance.

Keywords: Siglce-9; Immune Response; Immunosuppression; Sialic Acid; sSiglec-9

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1. Introduction

Sialic acid-binding immunoglobulin-like lectins (Siglecs) family is a kind of transmembrane proteins that can recognize sialic acid ligands. It is a major subgroup of type I lectins[1]. Siglecs found in mammals can be divided into two categories based on evolutionary conservation and sequence similarity. The first group is composed of Sn (sialoadesin, Siglec-1) and CD22 (Siglec-2), MAG (myelin associated glycoprotein, Siglec-4) and Siglec-15; the second group is CD33 related sialic acid-binding immunoglobulin-like lectins (CD33-related Siglecs, CD33rSiglecs), including 10 of human (Siglec-3, -5, -6, -7, -8, -9, -10, -11, -14, -16) and 5 of rodents (Siglec-3, -E, -F, -G, -H). They have 50%–80% sequence similarity[1,2]. Human Siglec-9 and mouse Siglec-E have high homology, while the structural sequences of Siglec-9 and Siglec-7 in human are highly similar. Most members of the Siglec family are inhibitory receptors, including Siglec-9. At present, many studies have shown that Siglec-9may play an important role in the pathogenesis of many diseases, including infectious diseases, autoimmune diseases and cancer, but its mechanism of regulating immune response is not yet clear. Further research on the mechanism of Siglec-9 in diseases is of great significance for clinical diagnosis and treatment. This paper summarizes the current research on the mechanism of Siglec-9 in diseases.

2. Biological characteristics of Siglec-9

Siglec-9, also known as CD329, contains 463 amino acids and its

gene is located on the long arm of chromosome 19[3,4]. Siglec-9 is widely expressed on the surface of immune cells, such as neutrophils, monocytes, macrophages, dendritic cells and natural killer (NK) cells[1]. All CD33rSiglecs members have similar sequences and are type I transmembrane proteins. Siglec-9 contains an immunoglobulin like N-terminal, a V-region domain and two C2 region domains, in which the V region is the site of sialic acid binding; the cytoplasmic tail includes an immunoreceptor tyrosine-based inhibitor motif (ITIM) and an ITIM like sequence[5]. Phosphorylation of tyrosine residues on the ITIM motif near the membrane end of Siglec-9 can recruit protein tyrosine phosphatases SHP-1 and SHP-2 (SRC homology 2 domain containing protein tyrosine phosphatase 1/2), and activate tyrosinase phosphorylation, so as to down regulate or inhibit the downstream intracellular activation signal[6–8]. Therefore, Siglec-9 is an inhibitory receptor on the cell surface.

The ligands of Siglecs are mainly sialic acid containing glycoproteins or glycolipids. Siglecs bind to ligands in roughly the same way, which can be divided into two categories: one is cis interactions, that is, they bind to sialic acid ligands (cis ligands) on the same cell membrane; the second is trans interactions, which combines with sialic acid ligands (trans ligands) on the surface of other cell membranes[6,9]. A high concentration of sialic acid formed locally on the same cell membrane can achieve cis interactions with Siglec, directly act on or mask the sialic acid binding site of Siglec, so as to weaken trans interactions. Siglec-9 on immune cells preferentially interacts with sialic acids ligands whose terminal are linked in the form of α(2,3)- and α(2,6)-[10]; the immune response between cells and between cells and pathogenic microorganisms can inhibit the activity of immune cells, facilitating the survival of pathogenic microorganisms and the immune escape of tumor cells[11–15]. In addition, vascular adhesion protein-1 (VAP-1) is an important molecule regulating leukocyte migration to inflammatory sites. Siglec-9, as a leukocyte ligand, can be used for positron emission tomography (PET) imaging to assist inflammation and cancer diagnosis[16].

Soluble Siglec-9 (sSleclec-9) is the extracellular part of Siglec-9, which has antibacterial effect by competitively inhibiting the binding between Siglec-9 and its ligands, and preventing down-regulation of host immune response, and can play an anti-tumor role by inhibiting downstream signal transduction mediated by tumors related mucin1 (MUC1)[17,18].

3. Mechanism of Siglec-9 in various diseases

3.1 Pulmonary diseases

Siglec-9 ligands are widely distributed in human's lung tissues (submucosal glands, epithelial cells and connective tissue) and are consistent with the distribution of neutrophil inflammation, which may be related to Siglecs-9's involvement in regulating neutrophil function[19]. And inflammation leads the up regulation of Siglec-9 ligands' expression in lung tissues, which may help to control lung inflammation[20].

Zeng *et al.* found that Siglec-9 expression on neutrophils increased in alveolar and peripheral blood of patients with the chronic obstructive pulmonary disease (COPD)[21]. At the same time, cigarette extract (CSE), lipopolysaccharide (LPS), some cytokines and dexamethasone (DEX) can up regulate the expression of Siglec-9. And they further found that DEX may have anti-inflammatory effect on neutrophils by up regulating the expression of Siglec-9. The expression of Siglec-9 also increased in COPD patients, and increased the oxidative burst of neutrophils and the chemotaxis of interleukin (IL)-8 by competitively inhibiting the binding of Siglec-9 to its ligands. Other studies have shown that through the analysis of Siglec-9 genotype and clinical characteristics in patients with COPD, it is found that some variant Siglec-9 weakens the response to inflammation. The inhibition of immune cell activation may be a risk factor for the development of emphysema[22].

In the mouse model of acute pulmonary inflammation, neutrophil recruitment in the lungs of Siglec-E-deficient mice increased[23]. In addition, mouse Siglec-E inhibits the recruitment of neutrophils to the lung by promoting the activation of NADPH (nicotinamide adenine dinucleotide phos-

phate) oxidase[24]. Therefore, it is speculated that human Siglec-9, which is homologous to it, may also regulate the function of neutrophils and inhibit lung inflammation in a similar way. Therefore, Siglec-9 may become a new target for immunosuppressive therapy of neutrophilic pulmonary inflammation (including some COPDs, and severe asthma).

3.2 Tumors

Tumor cells can achieve immune escape by up regulating inhibitory molecules to inhibit the host immune system, including PD-L1 (programmed death-ligand 1) and Siglec-9 ligands[25–28]. In addition, the change of sialylation on the surface of tumor cells is of great significance for tumor progression, especially in combination with Siglecs to regulate immunity cell function[29,30]. Monocytes expressing Siglec-9 have been found to increase in a variety of tumor tissues, including the non–small cell lung cancer[28].

MUC1 is a kind of transmembrane glycoprotein containing sialoglycan, which is abnormally expressed in a variety of tumor tissues, and can directly bind to Siglec-9 as Siglec-9 ligands. MUC1 expressed by tumor cells is combined with Siglec-9 on the surface of immune cells[31–33]. On the one hand, it directly inhibits antitumor immunity; on the other hand, it triggers MUC1 mediated immune response signal transduction, inducing recruitment β-catenin to enter into the nucleus, which leads to the growth of tumor cells. In sSiglec-9 transgenic mice, the proliferation of breast tumor cells expressing MUC1 was inhibited, and the expression of MUC1 tended to decrease[18]. It was found that Siglec-9 inhibits tumor proliferation by inhibiting MUC1 mediated downstream signal transduction, but this antitumor effect does not exist in cell research in vitro[31]. The difference of this result may depend on many factors. For example, the influence of experimental environment in vivo is more complex than that in vitro.

On the other hand, Siglec-9 or Siglec-E expressed on the surface of macrophages may inhibit the differentiation into M2 macrophages that promote tumors[1,28]. Heinz L äubi *et al.* found that there were more M2 macrophages and faster tumor growth in Siglec-E-deficient mice, indicating that Siglec-E can limit tumor growth; moreover, Siglec-9 has polymorphism, and the binding of K131Q Siglec-9 to ligands is reduced, which can improve the early survival rate of patients with non–small cell lung cancers (NSCLC)[28,34]. Overall, the effect of Siglec-9 on tumors is complex and may be different at different stages of tumor cell growth.

3.3 Autoimmune diseases

Siglec-9 is also up-regulated in rheumatoid arthritis (RA). It inhibits collagen induced arthritis by promoting the differentiation of anti-inflammatory regulatory T cells (Treg) and inhibiting the differentiation of pro-inflammatory helper T cells 17 (Th17) in a certain dependent manner[35]. In the serum and synovial fluid of RA patients, the level of Siglec-9 was also increased and correlated with the severity of RA disease. On the contrary, the results of Takuya Matsumoto *et al.* may be significantly different due to different animals and conditions used in the experiment[36], that sSiglec-9 obviously inhibits arthritis incidence rate and severity, and through in vitro experiments, it has been found that the anti-inflammatory role is achieved by inhibiting nuclear factor-kappaB (NF-κB) pathway to reduce the activity of M1 macrophages. In addition, Siglec-9 can specifically bind to VAP-1, label Siglec-9 peptide with radionuclide 68Ga, and detect it by PET imaging VAP-1 in blood vessels, clearly showing synovitis and contributing to the early diagnosis of RA[37].

However, in systemic lupus erythematosus, the expression of Siglec-9 on monocytes and neutrophils did not change significantly, while the expression of Siglec-14, another member of Siglecs family, increased[38].

3.4 Sepsis

Although the immune response is the body's protective response in the early stage of sepsis, the subsequent sustained strong immune response will lead to organ dysfunction. Therefore, timely and appropriate intervention of immune response is beneficial to alleviate sepsis.

Macrophage polarization and cytokines play an important role in the development of sepsis[39].

Siglec-9 can regulate the polarization of macrophages and promote the differentiation into M2 type that inhibits inflammation. On the other hand, the interaction between Siglec-9/Siglec-E and toll-like receptor 4 (TLR4) on immune cells is also involved in the pathogenesis of sepsis; Siglec-9/Siglec-E mediates the endocytosis of TLR4, which may regulate its activity by affecting its signal transduction and degradation, and then change the expression of downstream cytokines[40–42]. The specific mechanism needs to be further studied. In vitro experiments have shown that Siglec-9 activation can promote the production of anti-inflammatory cytokine IL-10 in macrophages and inhibit pro-inflamatory cytokine IL-6 and tumor necrosis factor by inhibiting LPS induced TLR4 Signal transduction-α (tumor necrosis factor-α, TNF-α)[43,44]. Therefore, Siglec-9, as an immunosuppressive receptor, may become a new strategy for anti-inflammatory treatment of sepsis.

3.5 Others

Changes in sialylation on the bacterial surface can enhance the virulence of bacteria and contribute to the survival of bacteria in the host[45]. For example, group B streptococcus (GBS) binds to Siglec-9 on neutrophils, resulting in oxidative burst and damage to the formation of neutrophil extracellular traps (NETs), which inhibits the host immune response and is conducive to the survival of pathogenic microorganisms[13]. In transgenic mice, sSiglec-9 can inhibit GBS infection by restoring the immune response of neutrophils[17].

Human immunodeficiency virus (HIV)-1 can interact with Siglec-1, -3, -9 through surface acidulated glycoprotein 120 (gp120), which promotes the sensitivity of macrophages[46]. In chronic hepatitis B, it was found that the expression of Siglec-9 ligands increased in HBV infected liver tissue; the expression of Siglec-9 positive NK cells decreased and was negatively correlated with the DNA titer of serum hepatitis e antigen and hepatitis B virus (HBV); the expression of Siglec-9 on NK cells of patients with sustained viral response returned to the normal level[47]. In addition, blocking Siglec-9 can reverse the inhibition of NK cells. Overall, Siglec-9, as an inhibitory receptor on the surface of NK cells, participates in the regulation of immunity and is related to the persistence of HBV in the host, but the specific mechanism is not clear.

4. Conclusions

According to many studies, Siglec-9, as an inhibitory receptor on immune cells, is involved in the pathogenesis of many diseases, mainly inhibiting the immune response; on the other hand, Siglec-9 may play different roles in different diseases or different periods of the same disease, having both advantages and disadvantages for the development of the disease. However, the specific mechanism of Siglec-9 is not completely clear and needs to be further studied. sSiglce-9 may weaken the function of Siglec-9 and positively regulate immune function through competitive inhibition, but there are still disputes in the current experimental results in vivo and in vitro.

In addition, Siglec-9 may be used as a potential biological marker to help the diagnosis, staging and treatment of some diseases. Using the immunosuppressive effect of activating Siglec-9 as a treatment strategy can inhibit the sustainable development of inflammation and provide a new choice for autoimmune diseases and allergic diseases. On the other hand, inhibiting or blocking the activity of Siglec-9 can enhance immune response and provide new therapeutic targets for tumors and infectious diseases. In conclusion, in-depth study of the mechanism of Siglec-9 has far-reaching significance for disease diagnosis and treatment.

Conflict of interest

The authors declare no potential conflicts of interest.

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