



Soluble triggering receptor expressed on myeloid cells-1 as a new therapeutic molecule in rheumatoid arthritis

Tae-Hwan Kim^a, Sung Jae Choi^b, Young Ho Lee^b, Gwan Gyu Song^b, Jong Dae Ji^{b,*}

^aThe Hospital for Rheumatic Diseases, Hanyang University, Seoul, Republic of Korea

^bDivision of Rheumatology, College of Medicine, Korea University, Seoul, Republic of Korea

ARTICLE INFO

Article history:

Received 17 June 2011

Accepted 27 October 2011

ABSTRACT

Triggering receptor expressed on myeloid cells-1 (TREM-1) is a recently identified cell surface receptor that is expressed mainly on monocytes and neutrophils, and plays an important role as an amplifier of inflammatory response in acute and chronic inflammatory conditions. Recent studies suggested that TREM-1 contributes to the pathogenesis of rheumatoid arthritis (RA) and therefore TREM-1 could be a new therapeutic target in RA. In addition to its membrane-bound form, a soluble form of TREM-1 (sTREM-1) exists that is liberated by the proteolytic cleavage of membrane-bound form. This soluble form works as decoy receptor to prevent the binding of its ligand to membrane-bound TREM-1 and to inhibit the effect of TREM-1 activation. Proteolytic cleavage of TNF receptor (TNFR) has been reported and soluble TNFR are capable of binding and neutralizing TNF, thus working as natural TNF antagonist. Currently, etanercept, a soluble TNF-receptor fusion protein has been widely used to treat RA. In this report, we suggest that sTREM-1 can be used as a new therapeutic molecule in RA.

© 2011 Elsevier Ltd. All rights reserved.

Introduction

Rheumatoid arthritis (RA) is a systemic autoimmune disease characterized by chronic inflammation of synovial joints, which leads to progressive destruction of cartilage and bone. Although the cause of this disease remains obscure, the extensive research on RA has resulted in markedly improved understanding of its pathogenesis. It is now clear that a large number of inflammatory cytokines such as TNF α , IL-1 β , IL-6, IL-12, IL-23 and IL-17A play an important role in the process that cause persistent inflammation and progressive destruction of cartilage and bone [1]. Biologic agents designed to suppress action of these inflammatory cytokines involved in the pathogenesis of RA have been shown to inhibit the progression of inflammation and joint destruction in RA patients [2]. Among these biologic agents, TNF α inhibitors have been shown to improve greatly the signs, symptoms and quality of life in RA patients and inhibit definitively the radiographic bone destruction of RA. Although the use of TNF inhibitors has revolutionized the treatment of RA, substantial proportion (up to a third) of RA patients treated with TNF inhibitors fail to achieve complete remission [3]. Also serious bacterial infection and reactivation of latent tuberculosis have been observed in RA patients treated with

TNF inhibitors [4]. Therefore, new therapeutic molecule needs to be developed which reduces excessive inflammatory responses but allows sufficient control of infection.

Triggering receptor expressed on myeloid cells-1 (TREM-1) is a recently identified cell surface receptor that is expressed mainly on monocytes and neutrophils, and plays an important role as an amplifier of inflammatory response in acute and chronic inflammatory conditions [5–7]. Human TREM-1 consists of a single extracellular immunoglobulin (Ig)-like domain, a transmembrane region with a positively charged lysine residue, and a short cytoplasmic region lacking any signaling motifs [8]. Its natural ligand has yet to be identified, so signaling and function of TREM-1 were studied using an agonistic antibody, which induces receptor cross-linking. TREM-1 associates with the adapter protein DNAX activation protein 12 (DAP12) via a positively charged lysine residue of its transmembrane region. Engagement of TREM-1 triggers a signaling pathway including tyrosine phosphorylation of DAP12 that leads to the phosphorylation of phospholipase C γ (PLC γ) and extracellular signal-related kinase (ERK) 1/2, an increase in intracellular calcium and activation of NF- κ B [9]. The activation of these pathways eventually leads to the production of proinflammatory cytokines and chemokines such as MCP-1, MCP-3, IL-8, MIP-1 α and TNF α [10,11]. In addition to induction of cytokine production by TREM-1 engagement alone, TREM-1 engagement and Toll-like receptor (TLR) stimulation synergize in the production of proinflammatory cytokines and chemokines [10–12]. In contrast, TLR-induced production of IL-10 is reduced by simultaneous TREM-1 engagement.

* Corresponding author. Address: Division of Rheumatology, College of Medicine, Korea University, 126-1, Anam-Dong 5-Ga, Sungbuk-Ku, Seoul 136-705, Republic of Korea. Tel.: +82 2 920 5489; fax: +82 2 920 5974.

E-mail address: jjdjmesy@korea.ac.kr (J.D. Ji).

TREM-1 modulates the TLR function by increasing the availability of TLR signaling molecules such as MyD88, CD14 and I κ B α , and TREM-1 expression is upregulated by TLR stimulation [10,11,13].

Biological significance of TREM-1 as an amplifier of inflammatory response has been confirmed in studies using mouse models and human disease samples of acute and chronic inflammatory diseases. TREM-1 is highly expressed in acute infectious lesions caused by bacteria and fungi [12]. In addition to increased expression of TREM-1 in acute infectious diseases, TREM-1 expression is significantly increased in the non-infectious inflammatory diseases such as acute pancreatitis, inflammatory bowel diseases, gout and rheumatoid arthritis [14–19]. The *in vivo* role of TREM-1 in inflammatory conditions has been well documented using agents that inhibit or activate TREM-1 signaling. Engagement of TREM-1 by an agonistic TREM-1 antibody induced a significant increase of mortality rate in LPS-induced septic shock [20]. TREM-1 blockade using a fusion protein containing murine TREM-1 extracellular domain and human immunoglobulin- γ (IgG1) Fc portion (mTREM-1/IgG1) was reported to reduce serum TNF α and IL-1 β to sublethal levels, preventing shock and death in mouse models of microbial peritonitis and sepsis [12]. Administration of LP17, an antagonistic TREM-1 peptide mimicking a short, highly conserved domain of sTREM-1, also protected septic mice from death by attenuating systemic inflammatory responses [20–22]. Interestingly, although TREM-1 blockade using mTREM-1/IgG1 or LP17 was shown to reduce systemic inflammatory responses, the capacity of immune systems to fight bacterial infection was not affected by blocking of TREM-1 signaling [12,20–22]. Besides a beneficial effect of TREM-1 blockade in infectious diseases, administration of antagonistic TREM-1 peptide inhibited disease activity of chronic inflammatory diseases [16,18]. It was demonstrated that TREM-1 expression is upregulated on intestinal macrophages of patients with Crohn's disease and ulcerative colitis and that engagement of TREM-1 by an agonistic TREM-1 antibody results in increased secretion of inflammatory cytokines and chemokines in intestinal macrophages isolated from inflammatory bowel disease patients [18]. In this study, blocking TREM-1 by the administration of an antagonistic peptide substantially attenuated intestinal inflammation in animal model of colitis. Also blockade of TREM-1 using a recombinant adenovirus containing a gene for the extracellular domain of TREM-1 fused with IgG-Fc (TREM-1 Ig) or antagonistic peptide significantly suppressed collagen-induced arthritis without affecting adaptive immune responses [16]. In this study, an increase in serum concentrations of TREM-1 Ig was observed in mice injected with a recombinant adenovirus containing a gene for the extracellular domain of TREM-1. These findings suggest that modulation of TREM-1 activation might be a new therapeutic strategy for the treatment of chronic inflammatory diseases including RA.

In addition to membrane-bound form of TREM-1, a soluble form of TREM-1 (sTREM-1) exists that is liberated by the proteolytic cleavage of membrane-bound form [23]. This soluble form works as decoy receptor to prevent the binding of its ligand to membrane-bound TREM-1 and to inhibit the effect of TREM-1 activation. Actually, TREM-1-dependent enhancement of respiratory burst and IL-8 production of polymorphonuclear leukocyte (PMN) by platelets was significantly suppressed by recombinant soluble TREM-1 (rsTREM-1) [24].

Hypothesis

Because TREM-1 blockade suppressed inflammatory responses but did not affect the capacity of immune systems to fight bacterial infection, modulation of TREM-1 signaling might be effective and safe therapeutic strategy not only in infectious diseases but also in chronic inflammatory diseases such as RA. TREM-1 signaling

was blocked in recent studies by a recombinant adenovirus containing a gene for the extracellular domain of TREM-1 fused with IgG-Fc (TREM-1 Ig), antagonistic TREM-1 peptide and sTREM-1 [16]. Gene therapy using adenovirus is not easily used in clinical setting. Peptides as drug candidate have little resistance to serum and tissue proteases in body, therefore have too short half-life. Many previous studies demonstrated that soluble receptor act as a specific, high affinity antagonist of inflammatory molecule such as IL-1 β , IL-15 and TNF α . Despite significant advantages, soluble receptors have a potential limitation as therapeutic agents for chronic inflammatory diseases. Because soluble receptors are generally unstable, the strategy to make these proteins stable was needed for their clinical application. To overcome this problem, soluble receptors have been conjugated with the Fc region of immunoglobulin or polyethylene glycol (PEG) [25]. Currently, etanercept, a soluble TNF-receptor (sTNFR) fusion protein has been widely used to treat RA. Etanercept is a hybrid protein consisting of the Fc region of human IgG1 and dimers of human sTNFR. Also sTNFR conjugated with PEG was reported to have improved biostability [26].

In this article, we present a hypothesis that sTREM-1 can be used as effective therapeutic strategy for treating RA, via attenuating inflammatory response and not increasing the risk of infection.

Conflict of interest statement

The authors declare no conflicts of interest.

References

- [1] Brennan FM, McInnes IB. Evidence that cytokines play a role in rheumatoid arthritis. *J Clin Invest* 2008;118:3537–45.
- [2] Smolen JS, Aletaha D, Koeller M, Weisman MH, Emery P. New therapies for treatment of rheumatoid arthritis. *Lancet* 2007;370:1861–74.
- [3] Buch MH. Sequential use of biologic therapy in rheumatoid arthritis. *Curr Opin Rheumatol* 2010;22:321–9.
- [4] Caporali R, Pallavicini FB, Filippini M, Gorla R, Marchesoni A, Favalli EG, et al. Treatment of rheumatoid arthritis with anti-TNF-alpha agents: a reappraisal. *Autoimmun Rev* 2009;8:274–80.
- [5] Derive M, Massin F, Gibot S. Triggering receptor expressed on myeloid cells-1 as a new therapeutic target during inflammatory diseases. *Self Nonself* 2010;1:225–30.
- [6] Klesney-Tait J, Turnbull IR, Colonna M. The TREM receptor family and signal integration. *Nat Immunol* 2006;7:1266–73.
- [7] Tessarz AS, Cerwenka A. The TREM-1/DAP12 pathway. *Immunol Lett* 2008;116:111–6.
- [8] Colonna M. TREMs in the immune system and beyond. *Nat Rev Immunol* 2003;3:445–53.
- [9] Sharif O, Knapp S. From expression to signaling: roles of TREM-1 and TREM-2 in innate immunity and bacterial infection. *Immunobiology* 2008;213:701–13.
- [10] Bleharski JR, Kiessler V, Buonsanti C, Sieling PA, Stenger S, Colonna M, et al. A role for triggering receptor expressed on myeloid cells-1 in host defense during the early-induced and adaptive phases of the immune response. *J Immunol* 2003;170:3812–8.
- [11] Bouchon A, Dietrich J, Colonna M. Cutting edge: inflammatory responses can be triggered by TREM-1, a novel receptor expressed on neutrophils and monocytes. *J Immunol* 2000;164:4991–5.
- [12] Bouchon A, Facchetti F, Weigand MA, Colonna M. TREM-1 amplifies inflammation and is a crucial mediator of septic shock. *Nature* 2001;410:1103–7.
- [13] Ornatowska M, Azim AC, Wang X, Christman JW, Xiao L, Joo M, et al. Functional genomics of silencing TREM-1 on TLR4 signaling in macrophages. *Am J Physiol Lung Cell Mol Physiol* 2007;293:L1377–1384.
- [14] Collins CE, La DT, Yang HT, Massin F, Gibot S, Faure G, et al. Elevated synovial expression of triggering receptor expressed on myeloid cells 1 in patients with septic arthritis or rheumatoid arthritis. *Ann Rheum Dis* 2009;68:1768–74.
- [15] Kuai J, Gregory B, Hill A, Pittman DD, Feldman JL, Brown T, et al. TREM-1 expression is increased in the synovium of rheumatoid arthritis patients and induces the expression of pro-inflammatory cytokines. *Rheumatology (Oxford)* 2009;48:1352–8.
- [16] Murakami Y, Akahoshi T, Aoki N, Toyomoto M, Miyasaka N, Kohsaka H. Intervention of an inflammation amplifier, triggering receptor expressed on myeloid cells 1, for treatment of autoimmune arthritis. *Arthritis Rheum* 2009;60:1615–23.
- [17] Murakami Y, Akahoshi T, Hayashi I, Endo H, Kawai S, Inoue M, et al. Induction of triggering receptor expressed on myeloid cells 1 in murine resident peritoneal macrophages by monosodium urate monohydrate crystals. *Arthritis Rheum* 2006;54:455–62.

- [18] Schenk M, Bouchon A, Seibold F, Mueller C. TREM-1-expressing intestinal macrophages crucially amplify chronic inflammation in experimental colitis and inflammatory bowel diseases. *J Clin Invest* 2007;117:3097–106.
- [19] Wang DY, Qin RY, Liu ZR, Gupta MK, Chang Q. Expression of TREM-1 mRNA in acute pancreatitis. *World J Gastroenterol* 2004;10:2744–6.
- [20] Gibot S, Kolopp-Sarda MN, Bene MC, Bollaert PE, Lozniewski A, Mory F, et al. A soluble form of the triggering receptor expressed on myeloid cells-1 modulates the inflammatory response in murine sepsis. *J Exp Med* 2004;200:1419–26.
- [21] Gibot S, Alauzet C, Massin F, Sennoune N, Faure GC, Bene MC, et al. Modulation of the triggering receptor expressed on myeloid cells-1 pathway during pneumonia in rats. *J Infect Dis* 2006;194:975–83.
- [22] Wiersinga WJ, Veer CT, Wieland CW, Gibot S, Hooibrink B, Day NP, et al. Expression profile and function of triggering receptor expressed on myeloid cells-1 during melioidosis. *J Infect Dis* 2007;196:1707–16.
- [23] Gomez-Pina V, Soares-Schanoski A, Rodriguez-Rojas A, Del Fresno C, Garcia F, Vallejo-Cremades MT, et al. Metalloproteinases shed TREM-1 ectodomain from lipopolysaccharide-stimulated human monocytes. *J Immunol* 2007;179:4065–73.
- [24] Haselmayer P, Grosse-Hovest L, von Landenberg P, Schild H, Radsak MP. TREM-1 ligand expression on platelets enhances neutrophil activation. *Blood* 2007;110:1029–35.
- [25] Fernandez-Botran R, Crespo FA, Sun X. Soluble cytokine receptors in biological therapy. *Expert Opin Biol Ther* 2002;2:585–605.
- [26] Edwards 3rd CK. PEGylated recombinant human soluble tumour necrosis factor receptor type I (r-Hu-sTNF-RI): novel high affinity TNF receptor designed for chronic inflammatory diseases. *Ann Rheum Dis* 1999;58:173–81.