Microbial Infection and Rheumatoid Arthritis

Song Li¹, Yangsheng Yu¹, Yinshi Yue¹, Zhixin Zhang¹,² and Kaihong Su¹,²*,³

¹Department of Pathology and Microbiology, University of Nebraska Medical Center, Omaha, NE 68198, USA
²The Epilepsy Institute for Research in Cancer and Allied Diseases, University of Nebraska Medical Center, Omaha, NE 68198, USA
³Department of Internal Medicine, University of Nebraska Medical Center, Omaha, NE 68198, USA

Abstract

Rheumatoid arthritis (RA) is a complex autoimmune disease affecting 1-2% of general worldwide population. The etiopathogenesis of RA involves the interplay of multiple genetic risk factors and environmental triggers. Microbial infections are believed to play an important role in the initiation and perpetuation of RA. Recent clinical studies have shown the association of microbial infections with RA. Accumulated studies using animal models have also found that microbial infections can induce and/or exaggerate the symptoms of experimental arthritis. In this review, we have identified the most common microbial infections associated with RA in the literature and summarized the current evidence supporting their pathogenic role in RA. We also discussed the potential mechanisms whereby infection may promote the development of RA, such as generation of neo-autoantigens, induction of loss of tolerance by molecular mimicry, and bystander activation of the immune system.

Keywords: Rheumatoid arthritis; Infection; Microbes; Etiopathogenesis

Introduction

Rheumatoid arthritis (RA) is one of the most common inflammatory autoimmune diseases. It is characterized by persistent synovitis, systemic inflammation and production of autoantibodies [1]. The molecular mechanisms of RA pathogenesis are not fully understood. It is believed that approximately half of the risk factors for RA are attributed to genetic factors such as the human leukocyte antigen (HLA) alleles while the other half of the risks are environmental factors including infection and smoking [2]. Clinical and animal model studies have suggested that infections by many microorganisms, such as Porphyromonas gingivalis (P. gingivalis), Proteus mirabilis (P. mirabilis), Epstein–Barr virus (EBV), and mycoplasma contribute to the etiopathogenesis of RA (Table 1).

For this review, we first identified the most common microbial infections associated to RA in the literature [3-5] and then performed a key word search using “arthritis” and “name of the microorganism” for original publications in English in the databases including PubMed/Medline, Embase, EBSCO, SCOPUS, and Cochrane Library till November, 2013. The candidate microorganisms included in our search were P. gingivalis, P. mirabilis, EBV, cytomegalovirus (CMV), human immunodeficiency virus (HIV), parvovirus, hepatitis virus, herpes virus, human T-lymphotropic virus 1 (HTLV-I), mycoplasma, Streptococcus pyogenes (S. pyogenes), Salmonella, mycobacterium, and enterobacterium. Thus, this review will discuss studies regarding to those microorganisms with RA and emphasize on P. gingivalis which shows the strongest association with RA. Our discussion is organized in three sections, namely, clinical association of infection with RA, induction of arthritis by infection in animal models, and the pathogenic mechanisms of infection in RA.

Clinical Association of Infection with RA

Clinical co-existence of infection and RA

Periodontal disease (PD) is the most commonly associated RA disease. The association between the two has been considered since the early 1820s. PD is caused by chronic infection of approximately twenty different bacterial species, of which P. gingivalis, Prevotella intermedia, Tannerella forsythia, and Aggregatibacter actinomycetemcomitans are the most common ones. PD can progress from gingivitis to periodontitis and cause bone degeneration in the jaw. Clinical association studies consistently show that the prevalence of periodontitis is increased about two-fold in RA patients than non-RA patients. In a large study involving 4461 participants aged 60 or older in the US population, subjects with RA were more likely to have periodontitis (odds ratio (OR)=1.82) or complete tooth loss (edentulism, OR=2.27), compared to non-RA subjects after adjusting for age, gender, race/ethnicity, and smoking [6]. Another study reported that moderate to severe periodontitis was more prevalent in RA patients (51%) than age and gender matched osteoarthritis patients (26%) [7]. A recent study in the Dutch population confirmed the higher prevalence of severe periodontitis in RA patients [8]. They also reported that RA patients with severe periodontitis had higher DAS28 scores than RA patients with no or moderate periodontitis, suggesting that the severity of periodontitis is related to the severity of RA [8].

However, it is less clear that whether subjects with PD have increased incidence of RA. In a large prospective study involving 81,132 American women in the Nurses’ Health Study cohort, there is no increased risk of later-onset RA in subjects with a history of periodontal surgery and/or tooth loss compared to subjects with healthy periodontal conditions [9]. In another large prospective study using the National Health and Nutrition Examination Survey cohort, subjects with PD experienced higher odds of prevalent/incident RA, but most odd ratios were not statistically significant [10]. It is also important to keep in mind that both studies are not specifically designed to examine the relationship between PD and RA. It is quite possible that there are missing data about PD and RA status in these cohorts and differential RA and PD ascertainment bias may also complicate the interpretation of data.

*Corresponding author: Kaihong Su, Ph.D., Associate Professor, Department of Pathology and Microbiology, University of Nebraska Medical Center, LTC 11724, 987660 Nebraska Medical Center, Omaha, NE 68198-7660, USA, Tel: 402-559-7612; Fax: 402-559-7716; E-mail: ksu@unmc.edu

Received October 14, 2013; Accepted November 27, 2013; Published December 03, 2013

Citation: Li S, Yu Y, Yue Y, Zhang Z, Su K (2013) Microbial Infection and Rheumatoid Arthritis. J Clin Cell Immunol 4: 174. doi:10.4172/2155-9899.1000174

Copyright: © 2013 Li S, et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.
Another common infection associated with RA is Proteus-caused urinary tract infection. Patients with RA had significantly increased incidence of urinary tract infection and subclinical/asymptomatic bacteriuria compared to non-RA subjects [11]. P. mirabilis bacteria were isolated at a higher rate from urine samples of both female (63%) and male (50%) patients with RA than from healthy female (32-35%) and male (7-11%) subjects and patients with other autoimmune diseases including osteoarthritis, fibromyalgia, and psoriasis [12]. These studies implicate a plausible role of Proteus microorganisms in the development of RA.

Furthermore, it has been well documented that infections by a range of bacteria and viruses frequently manifest rheumatic diseases, including reactive arthritis. Gastrointestinal or genitourinary infections with Salmonella, Shigella, Campylobacter, Yersinia, and Chlamydia trachomatis may cause inflammatory oligoarticular or polyarticular sterile arthritis, usually starting within four weeks of infection [13]. Viruses including HIV, parvovirus, hepatitis viruses B and C, alpha viruses like Chikungunya can cause acute or chronic forms of arthritis, and in some cases, mimic RA [13]. The precedence of infection over clinical arthritis suggests a causal relationship of the two events.

In summary, the clinical association studies suggest that infection is a risk factor for the development of RA. In addition, antibiotics such as sulphasalazine, minocycline, and rifampicin have been reported to be beneficial for the treatment of RA [14,15]. Reversely, periodontal treatments (oral hygiene and supra gingival scaling) decreased the DAS28-CRP scores in RA patients [16], further implicating the pathogenic role of microbial infection in RA.

Presence of microbial contents in RA tissues

Besides the disease association, the presence of microbial contents in RA tissues provides additional evidence for the correlation between infection and RA. Molecular techniques such as PCR and DNA/RNA-in situ hybridization have been widely used to detect bacterial or viral infections. P. gingivalis [17], mycoplasma [18,19], parvovirus [20], EBV [21,22], and cytomegalovirus (CMV) [22,23] have been identified in the synovial fluid, synovial membranes or serum samples from RA patients. Herpes viruses were detected in salivary cells and circulating lymphocytes from RA patients [24,25]. Besides nucleic acids, other microbial components can be used in the detection of infections in RA patients. Bacterial fatty acids, peptidoglycan, and muramic acid quantified by techniques such as gas-liquid chromatography (GLC), enzyme-linked immunosorbent assays (ELISA) and mass spectrometry have been used to detect the presence of microbes in RA samples [26-29]. For example, in a cohort of patients with early RA before any specific treatment, analyses of bacterial cellular fatty acids by GLC revealed the abundance of anaerobic bacteria in the intestinal flora of RA patients, implicating a possible role of intestinal anaerobic bacteria in the development of RA [26-29].

Immune response to microbes in RA patients

Another strategy to detect previous and ongoing infections is to measure the immune responses to microbial components in patients. Indeed, antibodies against infectious microbes were detected in the sera of early RA patients and the levels of these antibodies correlated with the disease activity of RA. For example, elevated levels of IgM and IgA antibodies to P. mirabilis were found in rheumatoid factor (RF)-positive early RA patients [30]. The levels of anti-P. mirabilis antibodies in RA patients went down after one year of treatment and this decrease was significantly correlated with the decrease in a modified Stoke disease activity index in RA patients [31]. The specific antigens from P. mirabilis were later identified as haemolysin and urease [32,33].

Another prominent example is that increased antibody responses to P. gingivalis, one of the common bacteria causing PD, were detected in RA patient sera and synovial fluid. Furthermore, the anti-P. gingivalis antibody levels were correlated with the titers of anti-cyclic citrullinated peptide (CCP) antibodies (the recently added RA diagnosis criteria) in RA patients [34,35]. Interestingly, a recent study showed that anti-P. gingivalis antibodies were significantly associated with the presence of RA-related autoantibodies (anti-CCP and rheumatoid factor) in individuals at high risk of RA [36]. This result supports the hypothesis that infection by P. gingivalis may play a central role in the early loss of self-tolerance that occurs in the pathogenesis of RA. Increased antibody responses to other infectious agents, such as EBV [37], B19 parovirus [38], and mycoplasma [39,40], have also been reported in RA patients. Furthermore, T cell responses to EBV [41-43] and CMV [44] were detected in the inflamed joints from RA patients.

In summary, clinical studies using human materials revealed a possible causative link between microbial infection and RA. However, more definitive studies in well characterized cohorts are necessary before we can conclude that microbial infection plays a crucial role in the initiation and perpetuation of RA. Moreover, the “chicken or egg” relationship between infection and RA may be hard to address.
in human studies. First, the RA-susceptible genetic and environmental factors, such as predisposed genes and living habits, may cause increased risks to infection even before or in the early stage of RA. Second, the RA-associated abnormal immune response and immunosuppressive medicine may contribute to decreased host defense to infection [45]. In this case, studies using animal models are very useful as experimental animals normally have homogeneous genetic and environmental backgrounds.

Induction of Arthritis by Infections in Animal Models

Animal studies can directly address the causal relationship between infection and RA in accordant to Koch’s postulates, although there is no perfect animal model for human RA yet. Infections by *P. gingivalis* or mycoplasma induced or aggravated experimental arthritis in mice or rats [46-50]. Interestingly, a very recent study showed that *P. gingivalis* facilitated the development and progression of destructive arthritis in CIA mice through its unique bacterial peptidylarginine deiminase (PPAD) [51]. PPAD can lead to the generation of RA related citrullinated autoantigens by converting protein arginine residues to citrulline. This result suggests that *P. gingivalis* infection may play an important role in the loss of tolerance to citrullinated proteins in RA, implicating the causative link between infection and RA. In another study, experimental arthritis was strongly attenuated in the K/BxN mouse model under germ-free (GF) conditions, featured with reduced Th₁ cells. Furthermore, introduction of segmented filamentous bacteria into GF mice reinstated the production of autoantibodies and arthritis symptoms [52]. This study shows that a single commensal microbe can drive the autoimmune arthritis possibly via its ability to promote Th₁ cells.

Components derived from infectious pathogens can also induce or potentiate arthritis in animal models. For example, bacterial cell wall extracts can induce chronic arthritis in certain susceptible rat strains; and bacterial lipopolysaccharide (LPS) potentiates type II collagen-induced arthritis in mice [53,54]. Interestingly, EBV does not infect mice; and in a humanized mouse model, EBV induced erosive arthritis with many features resembling those of RA [55,56]. Human T-lymphotropic virus 1 (HTLV-1) transgenic mice also developed inflammatory arthropathy, resembling RA [57].

In summary, the animal studies provide direct evidence for the causal relationship between infection and RA in experimental arthritis. Combined with the clinical association between infection and RA in human patients, it is convincing that microbial infection contributes to the etiopathogenesis of RA. However, the translation of findings from animals to humans is still arbitrary to some extent. First, adjuvant is used in the immunization protocol in these arthritis animal models. Second, the methods of delivering microbes in animal studies may cause increased arthritis symptoms in animals only resemble the joint inflammation in human patients. Nevertheless, these problems are common for animal models of human diseases, and arthritis animal studies certainly provide valuable information for our understanding of RA pathogenesis.

Pathogenic Mechanisms of Infection in RA

Generation of neo-autoantigens

Autoantibodies play a crucial role in the development of RA. In fact, serum RF and anti-citrullinated protein antibodies (ACPAs) are included in the 2010 ACR/EULAR diagnostic criteria of RA [58,59]. ACPAs are highly specific for RA and appear years earlier than the clinical diagnosis of RA [60]. Protein citrullination is a post-translational modification catalyzed by the enzyme peptidylarginine deiminase (PAD). *P. gingivalis* is the only prokaryotic organism that contains PAD. Recent studies showed that *P. gingivalis*-mediated citrullination of bacterial and host proteins provided an important mechanism for generating neo-antigens that drive the ACPA responses in RA [61]. Endogenous citrullinated proteins such as citrullinated α-enolase were abundant in *P. gingivalis*; and *P. gingivalis* PAD can citrullinate human proteins including common RA antigens fibrinogen and α-enolase [62]. Interestingly, a recent animal study showed that *P. gingivalis* facilitated the development and progression of destructive arthritis through its bacterial PAD enzymatic activities [51]. Microbial infections may also facilitate the citrullination process by activating host monocytes and neutrophils which express high levels of PAD [63-65].

Neutrophil extracellular trap (NET) is a structure released by activated neutrophils. It is composed of decondensed chromatin and granular molecules which can enhance the killing of extracellular microbes. It has been shown that bacterial LPS and some inflammatory cytokines can strongly induce NET. NET provides a source of autoantigens for several autoimmune diseases, such as vasculitis, systemic lupus erythematosus, and RA [66,67]. NET contains citrullinated RA autoantigens including α-enolase and vimentin. Furthermore, netting neutrophils have been found in synovial tissues, rheumatoid nodules, and skin from RA patients [67], implicating a role of NET in the pathogenesis of RA.

Collectively, microbial infections directly or indirectly induce the generation of citrullinated neo-autoantigens which may trigger the aberrant immune responses in RA [68].

Loss of tolerance by molecular mimicry

Molecular mimicry plays an important role in the loss of tolerance in autoimmunity. Microbes may have elements that are similar in amino acid sequences or structure to self-proteins thus trigger autoantibody production through epitope spreading. For example, the *P. gingivalis* enolase and human α-enolase share 82% homology at the 17-amino acid immunodominant regions. Therefore, antibodies against bacteria enolase can recognize the homologous human α-enolase and promote the production of anti-human α-enolase autoantibodies. Indeed, the levels of anti-citrullinated human α-enolase antibodies were tightly correlated with the levels of antibodies to bacterial α-enolase in RA patients [69]. In addition, the affinity-purified antibodies to the human α-enolase peptide displayed cross-reactivity with the *P. gingivalis* enolase peptide [69]. Other examples include antibodies against EBV peptide p107 cross-react with the denatured human collagen and keratin [70]. Molecular mimicry also promotes autoreactive T cell activation and proliferation. The mimicry peptides for T cell activation are generally shorter and more linear compared to those of B cells. *E. coli* heat shock protein DnaJ contains a QKRAA motif that is also present in the HLA-DRB1 shared epitopes. DnaJ strongly activated RA synovial T cells which had passed the positive selection in the thymus through weak binding with the corresponding HLA epitopes [71,72]. Mycobacterial 65 kD heat shock protein (HSP65) shares homology with human HSPs. Clonal expansion of mycobacterial HSP65-reactive T lymphocytes was found in the synovial fluids and blood samples of RA patients. In addition, mycobacterial HSP65 can induce the proliferative response of mononuclear cells derived from RA synovial fluids [73,74]. These studies support the hypothesis that microbial molecular mimicry
plays an important role in priming autoimmunity in patients with RA.

**Bystander activation of the immune system**

Bystander activation is a process by which microbial products non-specifically activate lymphocytes and immune effector cells. It has been shown that bystander activation also plays a role in driving the autoimmunity and tissue injury in RA. The pathogen-associated molecular patterns (PAMPs) can bind to the pattern recognition receptors (PRRs) and lead to both innate and adaptive immune cell activation [75]. *P. gingivalis* and *E. coli* LPS induced monocyte activation and the production of RA-associated cytokines interleukin (IL)-1 and IL-33 through the TLR pathways [76,77]. Peptidoglycan, a bacterial cell wall component, is a potent arthritogen. It can activate lymphocytes and induce production of cytokines and polyclonal autoantibodies including RF in vivo using animal models and *in vitro* using cell culture systems [78,79].

**Microbial superantigenis**

Superantigens have long been suggested to play a role in pathogenesis of autoimmune diseases. The frequency of Vβ14+ T cells in the synovial fluid of affected joints are significantly higher than that in the peripheral blood of RA patients, implicating that the etiology of RA may involve initial activation of Vβ14+ T cells by a Vβ14-specific superantigen [80]. The skewed accumulation of Vβ14+ T cells in RA synovial joints was confirmed by another study [81]. EBV infection of human lymphocytes can cause *in vitro* expansion of non-specific B cells and CD8+ T cells, leading to polyclonal antibody production and cytotoxic T cell activation [43,82,83]. In animal models, several superantigens, such as mycoplasma arthritis mitogen and toxic shock syndrome toxin, were able to exacerbate arthritis [50,84].

**Direct effects on joint tissues**

Microbial infection can have direct activating or damaging effects on the joint tissues. For example, *Streptococcus pyogenes* infection resulted in the increased expression of receptor activator of NF-κB ligand (RANKL) in mouse osteoblasts in cell culture [85,86]. In another study, Salmonella infection led to RANKL upregulation in synovial fibroblasts derived from mice [87]. Furthermore, co-cultures of Salmonella-infected synovial fibroblasts with osteoclast precursors resulted in the differentiation of multinucleated bone-resorbing, osteoclast-like cells and the formation of bone-resorbing pits [87]. This study provided evidence that Salmonella infection can mediate osteoclast differentiation and activation, which may contribute to bone destruction in infected joints. Recently, it was reported that *P. gingivalis* directly promotes early and later stages of apoptosis of human chondrocytes, which may contribute to the cartilage loss in RA patients [88].

**Conclusion**

RA is a complex autoimmune inflammatory disease. The etiopathogenesis of RA involves the interplay of multiple genetic risk factors and environmental triggers. Numerous studies have shown the clinical association of microbial infection with RA. Infection is often detected in early RA and can precede the occurrence of clinical arthritis. These observations suggest that infection contributes to the initiation and exaggeration of RA, arguing against the theory that the RA-associated infection is simply a sequela of immunosuppressive treatments. The pathogenic role of infection in RA is also suggested by studies using arthritis animal models. Among the RA associated microbes, *P. gingivalis* shows the greatest promise as a significant contributor to RA etiology. *P. gingivalis* is the only known prokaryotic organism that contains enzyme peptidylarginine deiminase (PAD) which is essential for the generation of citrullinated autoantigens. Human studies have shown the association of *P. gingivalis* infection with RA patients and individuals at high risk for RA. Animal studies also demonstrated that *P. gingivalis* infection facilitated the development and progression of destructive arthritis. And more interestingly, this effect is dependent on *P. gingivalis* PAD. Future prospective studies examining *P. gingivalis* infection in patients before and at the early-onset of RA using serial collections of patient sera are necessary to confirm the etiopathogenetic role of *P. gingivalis* in RA. Multivariate analyses stratified by RA related factors such as susceptible gene alleles and smoking are also required to pinpoint the role of *P. gingivalis* infection in RA. In addition, studies that elucidate the arthritogenic pathways of *P. gingivalis* infection hold great promise to provide therapeutic targets for the prevention and treatment of RA, a disease affecting 1-2% of the general worldwide population.

**Acknowledgements**

Research reported in this publication was supported in part by National Institutes of Health Grants AR059251 (to KS), AR048592, AI073174, AI074948, and AI076475 (to ZZ) and by a Research Support Fund grant from the Nebraska Medical Center and the University of Nebraska Medical Center (to KS). The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health.

**References**

1. Scott DL, Wolfe F, Huizinga TW (2010) Rheumatoid arthritis. Lancet 376: 1094-1108.
2. McInnes IB, Schett G (2011) The pathogenesis of rheumatoid arthritis. N Engl J Med 365: 2205-2219.
3. Hitchon CA, El-Gabalawy HS (2011) Infection and rheumatoid arthritis: still an open question. Curr Opin Rheumatol 23: 352-357.
4. Carty SM, Snowden N, Silman AJ (2004) Should infection still be considered as the most likely triggering factor for rheumatoid arthritis? Ann Rheum Dis 63: i46-i49.
5. Silman AJ (2009) Microbes in the pathogenesis of rheumatoid arthritis. In: Silman AJ, Smolen JS, Weinblatt ME, Weissman MH (eds.) Rheumatoid arthritis. (1st edn), Mosby Publishers, USA.
6. de Pablo P, Dietrich T, McAlindon TE (2008) Association of periodontal disease and tooth loss with rheumatoid arthritis in the US population. J Rheumatol 35: 70-76.
7. Dissick A, Redman RS, Jones M, Rangan BV, Reimold A, et al. (2010) Association of periodontitis with rheumatoid arthritis: a pilot study. J Periodontol 81: 223-230.
8. Smit MD, Westra J, Visserink A, Doorens-van der Meer B, Brouwer E, et al. (2012) Periodontitis in established rheumatoid arthritis patients: a cross-sectional clinical, microbiological and serological study. Arthritis Res Ther 14: R222.
9. Arkema EV, Karlsen EW, Costenbader KH (2010) A prospective study of periodontal disease and risk of rheumatoid arthritis. J Rheumatol 37: 1800-1804.
10. Demmer RT, Mollitor JA, Jacobs DR, Jr, Michalowicz BS (2011) Periodontal disease, tooth loss and incident rheumatoid arthritis: results from the First National Health and Nutrition Examination Survey and its epidemiological follow-up study. J Clin Periodontol 38: 998-1006.
11. Puntis D, Malik S, Saravanan V, Ryne K, Heycock C, et al. (2013) Urinary tract infections in patients with rheumatoid arthritis. Clin Rheumatol 32: 355-360.
12. Senior BW, Anderson GA, Morley KD, Kerr MA (1999) Evidence that patients with rheumatoid arthritis have asymptomatic ‘non-significant’ Proteus mirabilis bacteriuria more frequently than healthy controls. J Infect 38: 99-106.
13. Becker J, Winthrop KL (2010) Update on rheumatic manifestations of infectious diseases. Curr Opin Rheumatol 22: 72-77.
14. O’Dell JR, Blakely KW, Mallie JA, Eckhoff PJ, Leff RD, et al. (2001) Treatment of early seropositive rheumatoid arthritis: a two-year, double-blind comparison of minocycline and hydroxychloroquine. Arthritis Rheum 44: 2235-2244.

15. Pillemer S, Gulkos P, Ligier S, Yarboro C, Gourley M, et al. (2003) Pilot clinical trial of intravenous doxycycline versus placebo for rheumatoid arthritis. J Rheumatol 30: 41-43.

16. Okada M, Kobayashi T, Ito S, Yokoyama T, Abe A, et al. (2013) Periodontal Treatment Decreases Levels of Antibodies to Porphyromonas Gingivalis and Citrulline in Patients with Rheumatoid Arthritis and Periodontitis. J Periodontol.

17. Martínez-Martínez RE, Abud-Mendoza C, Patiño-Marín N, Rizo-Rodríguez JC, Little JW, et al. (2009) Detection of periodontal bacterial DNA in serum and synovial fluid in refractory rheumatoid arthritis patients. J Clin Periodontol 36: 1004-1010.

18. Schaeveerbeke T, Renaudin H, Clerc M, Lequen L, Vrenhes JP, et al. (1997) Systematic detection of mycoplasmas by culture and polymerase chain reaction (PCR) procedures in 209 synovial fluid samples. Br J Rheumatol 36: 310-314.

19. Hoffmann RW, O’Sullivan FX, Schafermeyer KR, Moore TL, Roussel F, et al. (1997) Mycoplasma infection and rheumatoid arthritis: analysis of their relationship using immunoblotting and an ultrasensitive polymerase chain reaction detection method. Arthritis Rheum 40: 1219-1228.

20. Saal JG, Steidele M, Einsele H, Müller CA, Fritz P, et al. (1992) Persistence of B19 parvovirus in synovial membranes of patients with rheumatoid arthritis. Rheumatol Int 12: 147-151.

21. Takeda M, Mizugaki Y, Matsubara L, Imai S, Koike T, et al. (2000) Lytic Epstein-Barr virus infection in the synovial tissue of patients with rheumatoid arthritis. Arthritis Rheum 43: 1218-1225.

22. Mehræin Y, Lennzer C, Enlund L, Remmberger K, Ojaj A, et al. (2004) Latent Epstein-Barr virus (EBV) infection and cytomegalovirus (CMV) infection in synovial tissue of autoimmune chronic arthritis determined by RNA- and DNA-in situ hybridization. Mod Pathol 17: 781-789.

23. Tamm A, Ziegler T, Lautenschlager I, Nikkari S, Möttönen T, et al. (1993) Detection of cytomegalovirus DNA in cells from synovial fluid and peripheral blood of patients with early rheumatoid arthritis. J Rheumatol 20: 1489-1493.

24. Newkirk MM, Watanabe Duffy KN, Leclerc J, Lambert N, Shiroky JB (1994) Detection of cytomegalovirus, Epstein-Barr virus and herpes virus-6 in patients with rheumatoid arthritis with or without Sjögren’s syndrome. Br J Rheumatol 33: 317-322.

25. Zhang L, Nikkari S, Skurnik M, Ziegler T, Luukkainen R, et al. (1993) Detection of herpesviruses by polymerase chain reaction in lymphocytes from patients with rheumatoid arthritis. Arthritis Rheum 36: 1080-1086.

26. van der Heijden IM, Wilbrink B, Tchetverikov I, Schrijver IA, Schouls LM, et al. (2000) Presence of bacterial DNA and bacterial peptidoglycans in joints of patients with rheumatoid arthritis and other arthritides. Arthritis Rheum 43: 593-598.

27. Chen T, Rimplainen M, Luukkainen R, Mottonen T, Yli-Jama T, et al. (2003) Bacterial components in the synovial tissue of patients with advanced rheumatoid arthritis or osteoarthritis: analysis with gas chromatography-mass spectrometry and pan-bacterial polymerase chain reaction. Arthritis Rheum 49: 328-334.

28. Eerola E, Möttönen T, Hannonen P, Luukkainen R, Kantola I, et al. (1994) Intestinal flora in early rheumatoid arthritis. Br J Rheumatol 33: 1030-1038.

29. Kawahito Y, Ichinose S, Sano H, Tsuouchi Y, Kohno M, et al. (2008) Mycoplasma fermentans glycolipid-antigen as a pathogen of rheumatoid arthritis. Biochem Biophys Res Commun 369: 561-566.

30. Newkirk MM, Goldbach-Mansky R, Senior BW, Klippe J, Schumacher HR Jr, et al. (2005) Elevated levels of IgM and IgA antibodies to Proteus mirabilis and IgM antibodies to Escherichia coli are associated with early rheumatoid factor (RF)-positive rheumatoid arthritis. Rheumatology (Oxford) 44: 1433-1441.

31. Kjeldsen-Kragh J, Rasmid T, Dybwad A, Sioud M, Haugen M, et al. (1995) Decrease in anti-Proteus mirabilis but not anti-Escherichia coli antibody levels in rheumatoid arthritis patients treated with fasting and a one year vegetarian diet. Ann Rheum Dis 54: 221-224.

32. Tiwana H, Wilson C, Alvarez A, Abuknesha R, Bansal S, et al. (1999) Cross-reactivity between the rheumatoid arthritis-associated motif EQKRAA and structurally related sequences found in Proteus mirabilis. Infect Immun 67: 2769-2775.
segmented filamentous bacteria drive autoimmune arthritis via T helper 17 cells. Immunity 32: 815-827.

53. Simelyte E, Rimpiläinen M, Zhang X, Toivanen P (2003) Role of peptidoglycan subtypes in the pathogenesis of bacterial cell wall arthritis. Ann Rheum Dis 62: 976-982.

54. Caccese RG, Zimmerman JL, Carlson RP (1992) Bacterial lipopolysaccharide potentiates type II collagen-induced arthritis in mice. Mediators Inflamm 1: 273-279.

55. Kuwana Y, Takei M, Yajima M, Imadome K, Inomata H, et al. (2011) Epstein-Barr virus induces erosive arthritis in humanized mice. PLoS One 6: e26630.

56. Warde N (2011) Experimental arthritis: EBV induces arthritis in mice. Nat Rev Rheumatol 7: 683.

57. Ikawara Y, Tsou M, Yoshida E, Takiguchi M, Sato K, et al. (1991) Induction of inflammatory arthropathy resembling rheumatoid arthritis in mice transgenic for HTLV-I. Science 253: 1026-1028.

58. Aletaha D, Neogi T, Silman AJ, Funovits J, Felson DT, et al. (2010) 2010 Rheumatoid arthritis classification criteria: an American College of Rheumatology/European League Against Rheumatism collaborative initiative. Arthritis Rheum. 62: 2569-2581.

59. Aletaha D, Neogi T, Silman AJ, Funovits J, Felson DT, et al. (2010) 2010 rheumatoid arthritis classification criteria: an American College of Rheumatology/European League Against Rheumatism collaborative initiative. Ann Rheum Dis. 69: 1580-1588.

60. van de Stadt LA, de Koning MH, van de Stadt RJ, Woblink G, Dijkmans BA, et al. (2011) Development of the anti-citrullinated protein antibody repertoire prior to the onset of rheumatoid arthritis. Arthritis Rheum 63: 3226-3233.

61. Wegner N, Lundberg K, Kinloch A, Fisher B, Malmström V, et al. (2010) Autoimmunity to specific citrullinated proteins gives the first clues to the etiology of rheumatoid arthritis. Immun Rev 233: 34-54.

62. Wegner N, Wait R, Sroka A, Eick S, Nguyen KA, et al. (2010) Peptidylarginine deaminase from Porphyromonas gingivalis citrullinates human fibrinogen and alpha-enolase: implications for autoimmunity in rheumatoid arthritis. Arthritis Rheum. 62: 2662-2672.

63. Vossenaar ER, Radstake TR, van der Heijden A, van Mansum MA, Dieteren C, et al. (2004) Expression and activity of citrullinating peptidylarginine deiminase enzymes in monocytes and macrophages. Ann Rheum Dis 63: 373-381.

64. Fouquier C, Sebbag M, Clavel C, Chapuy-Regaud S, Al Badine R, et al. (2008) Antibodies to citrullinated proteins have increased immunogenicity and inflammatory arthropathy resembling rheumatoid arthritis in mice transgenic for HTLV-I. Science 253: 325-329.

65. Huang J, Jörgensen PF, Ellingsen EA, Chen G, Huber BT (2006) Cutting edge: Epstein-Barr virus transactivates the HERV-K18 superantigen by docking to the human complement receptor 2 (CD21) on primary B cells. J Immunol 177: 2056-2060.

66. Schwab JH, Brown RR, Randerfer SK, Schlievert PM (1993) Superantigen can reactivate bacterial cell wall-induced arthritis. J Immunol 150: 4151-4159.

67. Okahashi N, Sakurai A, Nakagawa I, Fujiwara T, Kawabata S, et al. (2003) Peptidylglycan primers for LPS-induced release of proinflammatory cytokines in whole human blood. Shock 18: 178-182.

68. Palaid X, West SG, Lafferty JA, Kappler JW, et al. (1991) Evidence for the effects of a superantigen in rheumatoid arthritis. Science 253: 325-329.

69. Howell MD, Dively JP, Lundeen KA, Eky A, Winters ST, et al. (1991) Limited T-cell receptor beta-chain heterogeneity among interleukin 2 receptor-positive synovial T cells suggests a role for superantigen in rheumatoid arthritis. Proc Natl AcadSci U S A 88: 10921-10925.

70. Rickinson AB, Moss DJ (1997) Human cytotoxic T lymphocyte responses to Epstein-Barr virus infection. Annu Rev Immunol 15: 405-431.

71. Hsiao FC, Lin M, Tai A, Chen G, Huber BT (2006) Cutting edge: Epstein-Barr virus transactivates the HERV-K18 superantigen by docking to the human complement receptor 2 (CD21) on primary B cells. J Immunol 177: 2056-2060.

72. Schwab JH, Brown RR, Randerfer SK, Schlievert PM (1993) Superantigen can reactivate bacterial cell wall-induced arthritis. J Immunol 150: 4151-4159.

73. Kogure A, Miyata M, Nishimaki T, Kasukawa K (1994) Proliferative response to citrullinated alpha-enolase peptide 1 are specific for rheumatoid arthritis and severity. Arthritis Res Ther 7: R458-467.

74. Schwab JH, Brown RR, Randerfer SK, Schlievert PM (1993) Superantigen can reactivate bacterial cell wall-induced arthritis. J Immunol 150: 4151-4159.

75. Okahashi N, Sakurai A, Nakagawa I, Fujiwara T, Kawabata S, et al. (2003) Infection by Streptococcus pyogenes induces the receptor activator of NF-kappaB ligand expression in mouse osteoblastic cells. Infect Immun 71: 948-955.

76. Takahashi D, Udagawa N, Mogi M, Yano K, et al. (2001) Activated lymphokine-activated杀免 cells. Immunity 32: 815-827.

77. Albani S, Keystone EC, Nelson JL, Ollier WE, La Cava A, et al. (1995) Positive selection in autoimmunity: abnormal immune responses to a bacterial DNAJ protein in the pathogenesis of rheumatoid arthritis. Clin Immunol Immunopathol 54: 14-25.

78. Dziarski R (1982) Preferential induction of autoantibody secretion in polyclonal human T cells. J Exp Med 155: 1180-1188.

79. Wang JE, Jørgensen PF, Ellingsen EA, Almiöf M, Thiemermann C, et al. (2006) Epstein-Barr virus infection. Annu Rev Immunol 15: 405-431.

80. Albani S, Keystone EC, Nelson JL, Ollier WE, La Cava A, et al. (1995) Positive selection in autoimmunity: abnormal immune responses to a bacterial DNAJ protein in the pathogenesis of rheumatoid arthritis. Clin Immunol Immunopathol 54: 14-25.

81. Albani S, Keystone EC, Nelson JL, Ollier WE, La Cava A, et al. (1995) Positive selection in autoimmunity: abnormal immune responses to a bacterial DNAJ protein in the pathogenesis of rheumatoid arthritis. Clin Immunol Immunopathol 54: 14-25.

82. Albani S, Keystone EC, Nelson JL, Ollier WE, La Cava A, et al. (1995) Positive selection in autoimmunity: abnormal immune responses to a bacterial DNAJ protein in the pathogenesis of rheumatoid arthritis. Clin Immunol Immunopathol 54: 14-25.