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Current Insights into the Pathogenesis of Rheumatoid Arthritis

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Abstract: ***Rheumatoid arthritis (RA) is a common autoimmune disease that is associated with progressive disability, systemic complications, decreased quality of life, higher mortality rate than healthy individuals and socioeconomic costs. The cause of rheumatoid arthritis is unknown, and the prognosis is guarded. However, advances in understanding the disease pathogenesis have fostered the development of new therapeutics, which have improved disease outcomes. RA is a chronic, progressive disease characterized by synovial inflammation and hyperplasia, autoantibody production (rheumatoid factor, anti–citrullinated protein antibody [ACPA] and others), cartilage and bone destruction, and systemic features, including cardiovascular, pulmonary, psychological, and skeletal disorders. RA affects approximately 0.5-1% of European and North-American adults, with considerable regional differences. Prevalence estimates for Southern European countries are lower than for Northern Europe and highest rates are found in North America. Although some patients have mild self-limited disease, many experience joint destruction, severe physical disability and multiple co-morbidities. Mortality rates are more than twice as high in patients with RA as in the general population and this gap appears to be widening. Although much is known about the etiology and pathogenesis of rheumatoid arthritis (RA), understanding of all involved immune pathways remains incomplete and is due to a complex interplay among genotype, environmental triggers, and chance. This article reviews some of the most important risk factors and elements considered significant in RA pathogenesis, which are: genetic susceptibility (association with class II major histocompatability (MHC) antigens, specifically the shared epitope found in HLA-DR4 which predisposes to the development of ACPA-positive RA), environmental factors (smoking, periodontal disease, gastrointestinal pathogens etc), humoral factors (predominance of female gender, age of menarche) immunological factors responsible for initiating and maintaining inflammation (role of T lymphocytes, B Lymphocytes, inflammatory cytokines, fibroblasts and osteoclasts).***

Keywords: Disease mechanisms; rheumatology; immunology; biologics therapy

1. Introduction

Rheumatoid arthritis (RA) is a chronic systemic inflammatory disease, which affects mainly synovial joints, reducing quality of life and life expectancy[1]. Although joint involvement is the dominant feature of the disease with the typical symmetric swelling and tenderness of the proximal interphalangeal (PIP) joints, metacarpophalangeal (MCP) joints, wrists and metatarso-phalangeal (MTP) joints, extra-articular manifestations, which reflect systemic involvement are not uncommon and include manifestations on the skin, eye, heart, lung, renal, nervous and gastrointestinal systems.

The etiopathogenesis of the disease is multi-factorial and results from the complex interaction between genes, environment and stochastic factors (Ig and TCR repertoire), which lead to the breakdown of immune tolerance and creation of synovial inflammation in a characteristic symmetric pattern. Different mechanisms and pathways regulate inflammation and matrix destruction, including damage to bone and cartilage and generate a highly variable pathology whose severity ranges from mild cases with non-erosive, even sometimes spontaneously remitting disease, to severe, rapidly progressive and destructive RA.

The prevalence of RA is 0.5–1.0% in most populations. Prevalence estimates for Southern European countries (median of 3.3 cases per 10^3) are lower than for Northern Europe (5.0 per 10^3), and highest rates are found in North America (10.7 per 10^3)[2]. The concordance rate in monozygotic twins is 12–30% and the sibling recurrence rate for RA lies between 5 and 10%.The heritability of RA is estimated to be 65%.[4–6]. Annual incidence rates are estimated to be 16.5 cases (per 10^5) in Southern Europe, 29 cases in Northern Europe and 38 cases in North America. Women are more frequently affected than men, with the overall gender ratio being 0.3[2-3].

Some of the most important risk factors and elements which play a role in the generation of this autoimmune inflammatory disease, including : genetic susceptibility (association with class II major histocompatability (MHC) antigens), environmental factors (smoking, periodontal disease, gastrointestinal pathogens etc), humoral factors (predominance of female gender, age of menarche) immunological factors responsible for initiating and maintaining joint inflamation (role of lymphocyte T, Lymphocyte B, dentritic neutrofil and mast cells, inflammatory citokynes, fibroblasts and osteoclasts), are reviewed in this article.

2. Autoimmunity and immune dynamics in RA

The immune system can eliminate a wide variety of pathogens through its powerful effector mechanisms. Autoimmune responses resemble normal immune responses to pathogens - they are activated by antigens, in this case self antigens or autoantigens, and give rise to autoreactive effector cells and to antibodies, called autoantibodies against the self antigen. When reactions to self tissues do occur and are then improperly regulated, chronic inflammatory syndromes develop[7]. The immune system has multiple tolerance mechanisms to prevent autoimmunity, each of which is partly effective in preventing anti-self responses.

Central tolerance occurs in the thymus and bone marrow. T lymphocytes derived from common lymphocyte progenitor in the bone marrow migrate and seed the thymus where they mature and become ready to be transported in the periphery. This process comprises several stages starting with the most immature double negative ($CD4^-CD8^-$) thymocytes, which undergo several differential phases during which they commit to the T-cell lineage and begin to rearrange their T-cell receptor (TCR) gene loci. Those that successfully rearrange their TCRβ chain proliferate, initiate rearrangement of their TCRα chains, and become double positive ($CD4^+CD8^+$) [85], which after consecutive "positive and negative selection" become single positive mature cells of the CD4+ and CD8+ lineage. Positive selection ensures the successful differentiation of only those CD4+CD8+ thymocytes that are eventually able to recognize self-MHC molecules on thymic cortical epithelial cells. T cells that react too strongly with self antigens are deleted in the thymus, (negative selection) and this process is driven most efficiently by bone marrow derived antigen-presenting cells in the medullary region of the thymus[7]. Those cells that survive thymic selection leave the thymus and form the peripheral T-cell repertoire. However, clonal deletion in the thymus is not foolproof, nor should it be since a repertoire completely devoid of reactivity to self might well be devoid or reactivity to anything[8]. Central tolerance of B cells takes place in the bone marrow during preB to immature B cell transition as they express rearranged immunoglobulin (Ig) genes on their surface[9]. It appears that the dominant mechanism for B cells with high affinity to membrane-bound self-antigen is receptor editing (replacement of L-chain) and, to a lesser extent, deletion, while soluble self-antigens induce both receptor editing and anergy[10-11]. B cells are also controlled in the periphery.

Some peripheral tolerance mechanisms include: Insufficient self-antigen or co-stimulation, sequestration, crypticity; anergy (functional unresponsiveness- based on the two-signal paradigm T cell activation requires both TCR engagement and a co-stimulatory signal usually provided by CD28. Because the two ligands for CD28, CD80 and CD86, are primarily expressed at high levels on activated professional antigen-presenting cells, presentation of self-antigen by quiescent antigen-presenting cells would lead to tolerance. Indeed, immature DCs promote tolerance in this manner by constitutively presenting low doses of self-antigen on MHC, resulting in cell death or anergy of the corresponding T cells[12].); suppression by regulatory T cells (Tregs); induction of regulatory T-cell development instead of effector T-cell development (functional deviation), deletion of lymphocytes from the repertoire due to activation-induced cell death [7]. Innate immune system also plays a role in maintaining tolerance. Failure of natural tolerance mechanisms in the presence of genetic susceptibility and environmental triggers leads to the creation of an autoimmune syndrome as seen in Rheumatoid arthritis.

RA disease mechanism involves activation of autoreactive CD4 T cells by dendritic cells and by inflammatory cytokines produced by macrophages. Once activated, the autoreactive T cells provide help to B cells to differentiate into plasma cells producing arthritogenic antibodies[7]. A variety of self-antigens are implicated in the development and progression of RA, including joint-derived proteins, such as type II collagen and human cartilage derived glycoprotein HC gp39, and ubiquitous antigens, such as the endoplasmic reticulum stress protein BiP and other stress proteins. Post-translationally modified proteins, including citrullinated and carbamylated self-antigens, and glycosylated IgG Fc, are major RA autoantigens. Autoantigenic citrullinated self-proteins include vimentin, fibrinogen, type II collagen, and α-enolase[13]. The activated T cells produce cytokines, which in turn stimulate monocytes/macrophages, endothelial cells, and fibroblasts to produce more pro-inflammatory cytokines such as TNF-a, IL-l and IFN-y, or chemokines (CXCL8, CCL2), and finally matrix metalloproteinases, which destroy tissue[14-15].

RA synovial tissue is the inflammatory effector site, it differs substantially from the normal one consisting of a lining layer of macrophages and type B synovicytes from one to three cells thick, associated with a loose, vascularized, connective tissue sublining layer. It is characterized by proliferation of synovial lining cells (4-10 cells deep), increased vascularization (angiogenesis and high endothelial venule development), and infiltration of the sublining by inflammatory cells, including lymphocytes, plasma cells, mast cells, DCs and fibroblasts and monocytes/macrophages [16-18]. Fibroblast like synoviocytes, T lymphocytes (mainly) and other cells activate osteocytes through the receptor activator of nuclear kappa B/receptor activator of NFκB ligand system (RANK/RANKL) which drives osteoclastogenesis[39-91]. The end process is creation of an aggressive pannus tissue, which contains osteoclasts.

3. Genetic Risk Factors

Autoimunity is closely intertwined with genetic predisposition. The long-established association with the human leukocyte antigen (HLA)–DRB1 locus has been confirmed in patients who are positive for rheumatoid factor or ACPA and the shared epitope confers particular susceptibility[19].

The human major histocompatibility complex is a region of ~ 3.6Mbp on chromosome 6p21.31, comprising 4200 genetic loci including the human leukocyte antigens (HLA). The main gene products of this region that are relevant to immunity are designated class I molecules (HLA-A, C and B) and class II molecules (HLA-DR, DQ and DP). The former consists of a type I transmembrane protein of 45 kD (the heavy chain) and a soluble protein of 12 kD (β2-microglobulin, the light chain). They bind and present intracellular antigens to the T-cell receptors of CD8+ T cells. In contrast, the latter consists of two type I transmembrane proteins (the 32 kD α chain and 28 kD β chain) and present both intra- and extracellular antigens to the T-cell receptors of CD4+ T cells. The HLA-A, C and B genes encode HLA-A, C and B heavy chains and the B2M gene, located elsewhere (on chromosome 15) encodes the β2-microglobulin light chain. The DRA1, DRB1 genes (and additionally DRB3, DRB4 and DRB5 genes in some haplotypes), and the DQA1, DQB1, DPA1 and DPB1 genes

encode the α and β chains of HLA-DR, DQ and DP molecules respectively [20]. HLA is hyperpolymorphic.

Several HLA-DRB1 molecules (*0101, *0102, *0401, *0404, *0405, *0408, *1001 and *1402) share a common amino acid sequence at position 70-74 in the third hypervariable region of the DRβ1-chain. This sequence consisting of glutamine-leucine-arginine-alanine-alanine (QKRAA), QRRAA or RRRAA, has, therefore, been termed the "shared epitope"[21] (SE). Studies show that the presence of *HLA-DRB1* shared epitope alleles influence the development of anti-CCP positive RA[22-23].

Even though the exact mechanism of SE is not fully elucidated, it could play a role and influence the following processes: (1) thresholds for T-cell activation (based on avidity between the T-cell receptor, MHC, and peptide, especially in the context of post-translational modification events important in RA pathogenesis;(2) thymic selection of high-affinity, self-reactive T cells (based on the T-cell synovial repertoire); and (3) creation of molecular mimicry of microbial antigens[24].

Irrespective on the exact mode of action the presence of the SE is associated with development of anti-CCP positive RA and of a more severe phenotype.

Some other identified genetic risk factors include: 1- The single nucleotide polymorphism (SNP) in tyrosine phosphatase PTPN22 at position 1858 (C->T) which leads to a missense mutation is associated with several autoimmune diseases and has been found to be an HLA-independent risk factor for RF- and ACPA-positive (but not ACPA-negative) RA. As PTPN22 is important in the inhibition of T- and B-cell receptor signaling, this mutation is thought to lower the threshold for T- and B-cell activation, facilitating in this way the development of autoreactive T- and B-lymphocytes [25].

2 - The peptidyl arginine deiminase type IV (PADI4) gene is a risk allele that encodes an enzyme involved in the conversion of arginine to citrulline, which is postulated to play a role in the development of antibodies to citrullinated antigens. PADI4 polimorphism has been only associated with RA in Asian populations. 3- Other SNPs associated with RA have been found in the genes for signal transducer and activator of transcription 4 (STAT4), CD244 (natural killer cell receptor 2B4), Fc receptor-like3 (FCLR3), tumor necrosis factor (TNF) alpha-induced protein 3(TNF-AIP3), and TNF receptor-associated factor 1 (TRAF1). These genes encode proteins with various roles in B and T cell signaling[26].

4. Environmental Risk Factors

Environmental risk factors might influence RA pathogenesis directly through increase of protein citrulination or indirecty through initiation of a general inflammatory response.

Several pathogens have been proposed and studied as potential candidates in triggering the initial immune response necessary for RA-development in a genetically susceptible host. Some of these pathogens are: mycobacteria, Ebstein-Barr Virus, Parvovirus B19, porphyromonas gingivalis (which expresses PADI4 and is capable of promoting citrullination of mammalian proteins [27]) and filamentous bacterial species in small intestine, lamina propria, or large intestine for whom studies have shown links with rapid emergence of IL-17-expressing effector T cells in the context of autoimmunity, including inflammatory arthritis[28]. A recent study in patients with RA has highlighted Prevotella species, a Gram-negative anaerobe of the Bacteroides genus, as being enriched in the gut of patients with early but not established RA [29].

Until now however, no pathogen-derived antigen has been clearly linked to RA-pathogenesis and no relevant cross-reactivity, of self-antigen-specific T- or B-cells with pathogen-derived peptides ("molecular mimicry"), is detected.

Smoking and other forms of bronchial stress (e.g., exposure to silica) increase the risk of rheumatoid arthritis among persons with susceptibility HLA–DR4 alleles [30]. Cigarette smoke could be involved in the induction of protein-citrullination. Supporting this hypothesis, citrullinated proteins have been detected in bronchoalveolar lavage fluid from smokers but not from non-smokers [31].

Obesity is another proposed risk factor, pertaining to the immune-modulatory properties of adipocytes, which are an abundant source of pro- and anti-inflammatory cytokines.

RA is more common in women, the ratio of female-to-male patients is 2 : 1 to 3 : 1. This suggests a role for sex hormones in RA pathogenesis. Data supports the concept that estrogens modulate immune function [32].For example, autoantibody producing B cells exposed to estradiol are more resistant to apoptosis and could escape tolerance. Estrogen receptors are expressed on fibroblast-like synoviocytes (FLS) and increase production of metalloproteinases. Several cohort studies showed that women who had an older menarche age had increased risk for developing RA. The same is true for nulliparity.

5. Autoantibodies

Rheumatoid factors are autoantibodies directed against antigenic determinants of the Fc fragment of the immunoglobulin G (Ig G) molecules. They present mostly as IgM-RF, but IgG-G and IgA-RF have also been detected in subgroups of patients[33]. IgE-RF is observed in RA patients with extrarticular symptoms.

The role of RF and other autoantibodies in the pathogenesis of RA has been known from observed evidence - patients with a positive test result for RF in blood have more severe clinical disease and complications than seronegative patients including increased cardiovascular complications [34] RFs can activate complement but they may be also positive in healthy controls (1% in younger individuals, up to 5% in individuals older than 70 years) and in patients with numerous other non-RA diseases (including other rheumatic diseases such as Sjögren's syndrome and cryoglobulinemia), as well as chronic infections[35-38]. The RFs produced in RA differ from those produced by healthy individuals or from patients with paraproteins. The avidity of RF for the Fc

portion of IgG is much greater in RA than in other diseasess. Many RFs expressed by abnormal B cells (such as Waldenström's macroglobulinemia) and by normal B cells in human tonsils are derived from the germline. In contrast, RFs in RA are derived through rearrangements and somatic mutations of the germline genes. Production of RF could happen many years before onset of RA. IgM RF represents about 7% of the total IgM and 3% of the total IgG produced by synovial cell cultures [39]. Sensitivity and specificity of RF are, depending on the population studied, 60-70% and 50-90%, respectively.

RF is a very important rheumatological index, it is one of 7 diagnostic criteria for RA put forward by the American College of Rheumatology in 1987 [15, 40-45] and part of the 2010 American College of Rheumatology/European League Against Rheumatism Rheumatoid arthritis classification criteria[46].

Citrullination arises through post-translational modification of arginine residues to the non-naturally occurring amino acid citrulline by a process known as de-imination, catalyzed by a group of enzymes called peptidyl-arginyl-deiminases (PADs). Citrullination is a common manifestation of inflammatory responses, especially those associated with significant cell death [47]. It produces changes in the protein structure and reduces their net charge. What appears to be specific to RA is the host immune response to citrullinated protein antigens (anti-CCP), a response that may be linked to RA associated MHC class II molecules[48].

The PADs, which are deiminating enzymes that hydrolyze guanidinium side chains in peptidylarginine to yield peptidylcitrulline and ammonia, belong to a larger group of guanidino-modifying enzymes called the amidinotransferase (AT) superfamily)[49,50]. Five human PAD homologs have been identified and the PADI genes are located at a single cluster on chromosome1p36, [52, 51] and are named PAD1-4 and PAD6. PAD1 is primarily expressed in uterus and skin. PAD2 is more widely expressed in muscle, skin, brain, spleen, rheumatoid synovium, and secretory glands, PAD3 is expressed in skin. PAD4 is expressed in hematopoietic cells, and PAD6 is found in germ cells and peripheral blood leukocytes [52,53,51,54,55-57]. PAD2 and PAD4 are thought to generate citrullization of self antigens in RA.

AKA/APF were the first ACPAs to be characterized, and they recognized citrullinated forms of filaggrin or its precursor profilaggrin[58,59], but their role today is historical only. Among the citrullinated candidate antigens present in the joint, the best characterized clinically and pathogenically relevant to RA are fibrinogen, vimentin and α-enolase ones. Citrullinated fibrinogen antibodies are among the most frequent ACPAs found in patients with RA (≈60% to 66%)[60-62]. In total, 54 of the 81 arginines (66%) in human fibrinogen have been found to be susceptible to citrullination [63-65]. Vimentin citrullinated antibodies are highly specific for RA (≈95%), with a sensitivity that varies with the stage of the disease tested, ranging from 20% to 25% in early RA cohorts and 47% in patients with more established disease [66-68]. ACPAs are present in the serum of 80% to 90% of RA patients. In some studies, they are also more specific for RA than RF, with specificity approaching 90%.

Studies indicate direct involvement of ACPAs in RA pathogenesis : A type T cells are primed with native protein, while exogenous peptide gives rise to priming of both type A and B type T cells. In the thymus, T cells recognizing the type A conformation for a peptide are deleted, whereas type B T cells escape thymic negative selection[69]. Immunization of non autoimmune mice with unmodified hen egg lysozyme (HEL) showed that T cells specifically reactive to citrullinated epitopes of HEL were present among the responding repertoire to immunization. The citrullinated HEL epitopes were processed and loaded onto MHC class II complexes and were presented to T cells by DC. Hence naturally occurring self-reactive T cells, which have evaded thymic negative selection, may recognize citrullinated peptides loaded as flexible type B conformers onto MHC class II molecules and subsequently provide help for B cell–mediated antibody responses against citrullinated proteins [70] their targets are present and enriched in the synovium and fluid of RA patients. ACPA positive patients have a more severe disease and ACPA positivity is linked to HLA DR shared epitope alleles.

ACPAs could be detected years before onset of RA, epitope spreding is also observed before disease onset and they are an important serologic markers of the ACR/Eular 2010 RA classification criteria [46].

Other autoantibodies have been found in a minority of patients with RA, but they also occur in the setting of other types of arthritis. They bind to a diverse array of autoantigens, including type II collagen, human cartilage gp-39, aggrecan, calpastatin, BiP (immunoglobulin binding protein), and glucose-6-phosphate isomerase, carbamylated proteins, heat shock proteins etc.

6. Immune effector cells, synovial patterns and the inflammatory milieu

After breakdown of the immunological tolerance, the thin synovial layer gets infiltrated by T- and B-lymphocytes, neutrophils, monocytes, mast cells, which proliferate and produce inflammatory cytokines and chemokines. Cytokines, chemokines and vascular growth factors promote neovascularization and vascular leakage, facilitate cellular migration and result in the recruitment to the joint of other effector cells. Over time, this process creates a cytokine rich enviroment in the joint that activates synovial fibroblasts and osteoclasts, which in turn degrade cartilage and destroy bone. Distinct patterns of synovitis have been identified, suggesting topographical organization of lymphoid microarchitecture at the cellular level[71-73].

Patterns of synovitis include : diffuse inflammatory infiltrates of T cells, B cells, macrophages, and dendritic cells (present in about 50% of patients); clusters of lymphoid follicular aggregates comprising T cells, B cells, and dendritic cells (about 20%);structures with germinal center (GC)-like reactions (about 25%); and, rarely, granulomatous lesions (<5%; which have features similar to those of rheumatoid nodules). Of most interest are those tissues comprising secondary follicles (called ectopic lymphocyte like structures = ELS) and GCs; they are characterized by the presence of follicular dendritic cells, which are critical

components in follicle formation in the synovium and for perpetuation of adaptive immunity and autoimmunity[73,74].

The creation of these structures (ELS) that direct various lymphocyte responses (including the induction of effector functions, antibody generation, affinity maturation, class switching and clonal expansion), and occur in some patients with chronic inflammation, autoimmunity and cancer is governed by mechanisms which are poorly defined and require further study. Lymphoid neogenesis and propagation of ELS within inflamed tissue is driven by communication between local stromal cells, tissue specific resident mononuclear cells and infiltrating immune cells [75,76]. Although the actual cells that are responsible for the positioning of these structures within the inflamed tissue are not concisely defined, several new candidates have recently been proposed. These include lymphoid tissue inducer cells (LTi cells), IL-17-secreting CD4+ T cell populations and T follicular helper cells. As ELSs mature, dendritic cells within these structures continue to support the efficient priming of T cell responses through antigen presentation and they contribute to class-switch recombination, antibody generation and the formation of new lymphatic vessels[77–79]. Patients with this synovial phenotype on average had higher clinical disease activity and a more severe disease.

In RA the majority of T lymphocytes are primed within the central lymphoid organs. In addition, repeated activation of innate immunity can directly lead to chronic inflammation and possibly antigen presentation in the synovium. Although local antigen-specific expansion can occur, it is probably responsible for a relatively small percentage of the T cell infiltrate[39].
RA synovium contains large numbers of activated T cells, which are mainly CD4+ memory T cells (CD45RO+). Naïve T cells develop into effector T cells, upon antigen recognition, while lineage specificity depends on cytokine milieu; Recognition of antigen by naïve T cells in the context of MHC-molecules, however, requires additional costimulatory signals for efficient T cell activation. Without costimulatory signals, T cells become anergic. Interaction of CD28 and CD80/86 generates one of the most important costimulatory signals, which is controlled by expression of the natural inhibitor CTLA-4. This mechanism is exploited therapeutically by an engineered CTLA4-Ig fusion protein (Abatacept), which efficiently inhibits disease activity in RA. Next to cytokine production, CD4+ effector T cells have been attributed additional functions in RA synovium. By means of direct cell-to-cell contact they provide crucial help to B cells for antibody production, and by the expression of RANKL they can directly induce osteoclastogenesis [81].
After activation CD4+T cells differentiate along the Th1, Th2 and Th17 pathways. RA is considered a Th1/Th17 disease.

Th1 lymphocites produce cytokines such as IL-2 and IFN-γ and express the CXCR3 and CCR5 chemokine receptors. IL-12 is crucial for Th1 development. Th2 cytokine levels are very low in RA. Th17 cells produce IL-17, IFN-γ or IL-10 and can also produce TNF-α, IL-6, IL-22 and GM-CSF [82].
Tregs produce TGF-β and IL-10, they are not able to suppress inflammation in RA and are downregulated by TNF- α. Data has showed that treatment with TNF-antagonists restored Treg cell function.

RA is often considered to be a macrophage-driven disease because this cell type is the predominant source of proinflammatory cytokines inside the joint. Key proinflammatory cytokines released by synovial macrophages include TNF-alfa, IL-1, IL-6, IL-12, IL-15, IL-18, and IL-23. Synovial fibroblasts, the other major cell type in this microenvironment, produce the cytokines IL-1 and IL-6 as well as TNF-α. Fibroblasts secrete matrix metalloproteinases (MMPs) as well as other proteases that are chiefly responsible for the breakdown of articular cartilage. Osteoclast activation at the site of the pannus is closely tied to the presence of focal bone erosion. Receptor activator of nuclear factor κB ligand (RANKL) is expressed by stromal cells, synovial fibroblasts, and T cells. Upon binding to its receptor RANK on osteoclast progenitors, RANKL stimulates osteoclast differentiation and bone resorption. RANKL activity is regulated by osteoprotegerin (OPG), a decoy receptor of RANKL that blocks osteoclast formation[83].

7. Some cytokines involved in RA pathogenesis

TNF-α : Member of the Tumor Necrosis Family (TNF) family of cytokines which regulates the development, effector function, and homeostasis of cells participating in the skeletal, neuronal, and immune systems, among others[85]. TNF- afla activates leukocytes, endothelial cells, and synovial fibroblasts, inducing production of cytokines, chemokines, adhesion molecules, and matrix enzymes; suppression of regulatory T-cell function; activation of osteoclasts; and resorption of cartilage and bone; mediates metabolic and cognitive dysfunction[84].

Interleukin-1α and 1β : Part of the IL-1 family comprising IL-1α, IL-1 β, IL-1Ra, IL-18, IL-33 [85], activate leukocytes, endothelial cells, and synovial fibroblasts; mononuclear cells, induce matrix-enzyme production by chondrocytes; activate osteoclasts; mediate fever; enhance glucose metabolism; and reduce cognitive function[84].

Interleukin-6 : activates leukocytes and osteoclasts; induces immunoglobulin synthesis in B cell lines, is involved in the differentiation of cytotoxic T lymphocytes, and is a major factor in the regulation of acute-phase response proteins like C-reactive protein by the liver [39], lipidemia and anemia of chronic disease; it is implicated in hypothalamic–pituitary–adrenal axis dysfunction and fatigue[84]. In RA, IL-6 is produced by FLSs and by chondrocytes, but the major local source is probably macrophages [86].

Interleukin-7 and 15 : promote and maintain T-cell and natural killer–cell activation and T-cell memory, block apoptosis, and maintain T-cell–macrophage cognate interactions[84].

Interleukin-17A and 17F: act synergistically to enhance activation of synovial fibroblasts, chondrocytes,and osteoclasts[84].

BLyS and APRIL : Activate B cells and have a role in the maturation of B cells and enhancement of autoantibody production
GM-CSF and M-CSF : Enhance differentiation of granulocyte and myeloid-lineage cells in the bone marrow and synovium
RANKL : is produced by T, FLS, osteoblasts[86] and promotes maturation and activation of osteoclasts

JAK : the JAK proteins are key proteins that transduce signals from a wide variety of cytokine and growth factor receptors.Four JAKs have been identified (JAK1, JAK2, JAK3, and TYK1), and they can form heterodimers and homodimers. The JAK proteins phosphorylate the signal transducers and activators of transcription (STATs). The STATS can then translocate to the nuclei, where they can alter gene transcription. STATs have been implicated in the expression of many proinflammatory genes. IFNs signal through STAT1, IL-6 signals through STAT3, and IL-12 signals through STAT4. Multiple STATs are expressed in rheumatoid synovium [39].

8. Biologics agents in Rheumatoid Arthritis

TNF- α blockers are the largest groups of biologics now available to treat RA and include: etanercept (Enbrel): a soluble TNF- α receptor fusion protein; adalimumab (Humira): recombinant human IgG1 monoclonal antibody; infliximab (Remicade): a chimeric IgG1 anti-TNF-α antibody; certolizumab (Cimzia): a recombinant humanized fragment of TNF-antibody coupled with polyethylene glycol and golimumab (Simpon): a recombinant human IgG1 monoclonal antibody specific for TNF-α.

Toclizumab (Actemra) is a recombinant humanized IL-6 receptor monoclonal antibody, Rituximab (Rituxan) is a B-cell inhibitor; Abatacept (Orencia), an immunoglobulin fused to the extracellular domain of cytoxic T-cell antigen, Anakinra (Kineret) is a IL-1 inhibitor, and, the newest biologic in RA Tofacitinib (Xeljanz) is a JAK 1 and 3 inhibitor. Other clinical trials on new treatment modalities (trial for IL-17A, LTα/β IL-12, Baff and others) are currently being conducted [87-90].

9. Conclusion

The pathogenesis of RA remains a complex puzzle although the level of understanding has progressed considerably in recent years. The pathogenetic mechanisms and hypothesis summarized in this review have paralleled the introduction of new, effective therapies and remarkable improvement in clinical outcomes. Anyhow, much remains to be resolved, we still do not know what specifically leads to the breakdown of tolerance, new mechanisms need to be elucidated, in order to reach the ultimate goal of developing curative and preventive therapeutics that will transform and improve management of Rheumatoid Arthritis, which, at the present is still a chronic debilitating disease that decreases patients' quality, and length of life.

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