Rheumatoid Arthritis: Epigenetic Drug Targets

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Autoimmune diseases, such as rheumatoid arthritis (RA) arise as a result of a breakdown in immune tolerance, for reasons that are as yet unknown. RA affects up to 1% of the population worldwide and has a lifetime risk of 4% for women and 2% for men. While a number of successful immunotherapies, such as anti-TNF, CTLA-4Ig and anti-CD20, have revolutionized the treatment of RA in recent years, many patients still fail to respond sufficiently to treatment. Moreover, these costly therapies are not curative, so treatment must be continued for many years to manage disease. Although the etiology and pathogenesis of RA remains elusive, studies have revealed a genetic predisposition, with variants of several genes, including human leukocyte antigen DRB1 (HLA-DRB1), IRF5, PTPN22, CD28 and CTLA-4, having strong association with the condition. Despite this extensive genetic predisposition, the concordance of RA in monozygotic twins is low (12–20%), suggesting that environmental and epigenetic factors are important in disease development[1]. The environmental factor most consistently associated with RA is cigarette smoke, with alcohol consumption seeming to have a mild protective effect.

RA Pathology

RA is a chronic destructive inflammatory disease that is characterized by severe swelling, pain and stiffness of the joints. In healthy individuals the synovial membrane, which lines the joint capsule, acts to lubricate the joint by producing synovial fluid. The synovial membrane contains two main immune cell types, macrophages and synovial fibroblasts (synoviocytes) that lie within the connective tissue. In RA, however, there is an influx of T cells, NK cells and B cells, in addition to other chronically inflamed cell types; the synoviocytes become activated and invade the surrounding cartilage, resulting in cartilage degradation and bone erosion.

RA Inflammation

Antigen-activated T cells produce pro-inflammatory cytokines, such as IL-17, that stimulate monocytes, macrophages and synovial fibroblasts to produce other pro-inflammatory cytokines, IL-1, IL-6 and TNF-α. This leads to recruitment of other immune cells, such as NK cells and B cells, which in turn secrete IFN-γ and immunoglobulins such as rheumatoid factor, respectively. Overall, the combination of these proinflammatory cytokines contribute to the articular inflammation. The activation of macrophages, lymphocytes and fibroblasts can also induce angiogenesis, which may explain the increased vascularity found in the synovium of RA patients. Moreover, the endothelial cells in these newly formed vessels express high levels of adhesion markers leading to increased inflammatory cell recruitment and infiltration. Activated $CD4^+$ T cells express osteoprotegerin ligands that stimulate osteoclastogenesis; similarly NK cells from the RA synovium express increased RANKL, which also stimulates osteoclastogenesis, leading to bone erosion.

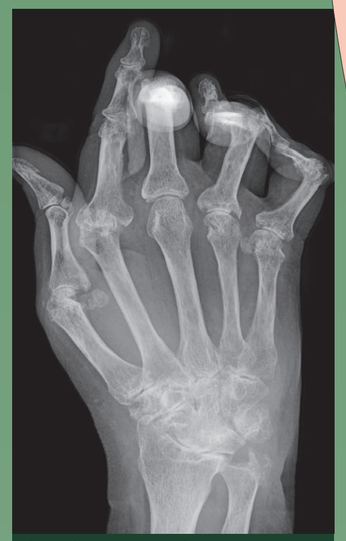
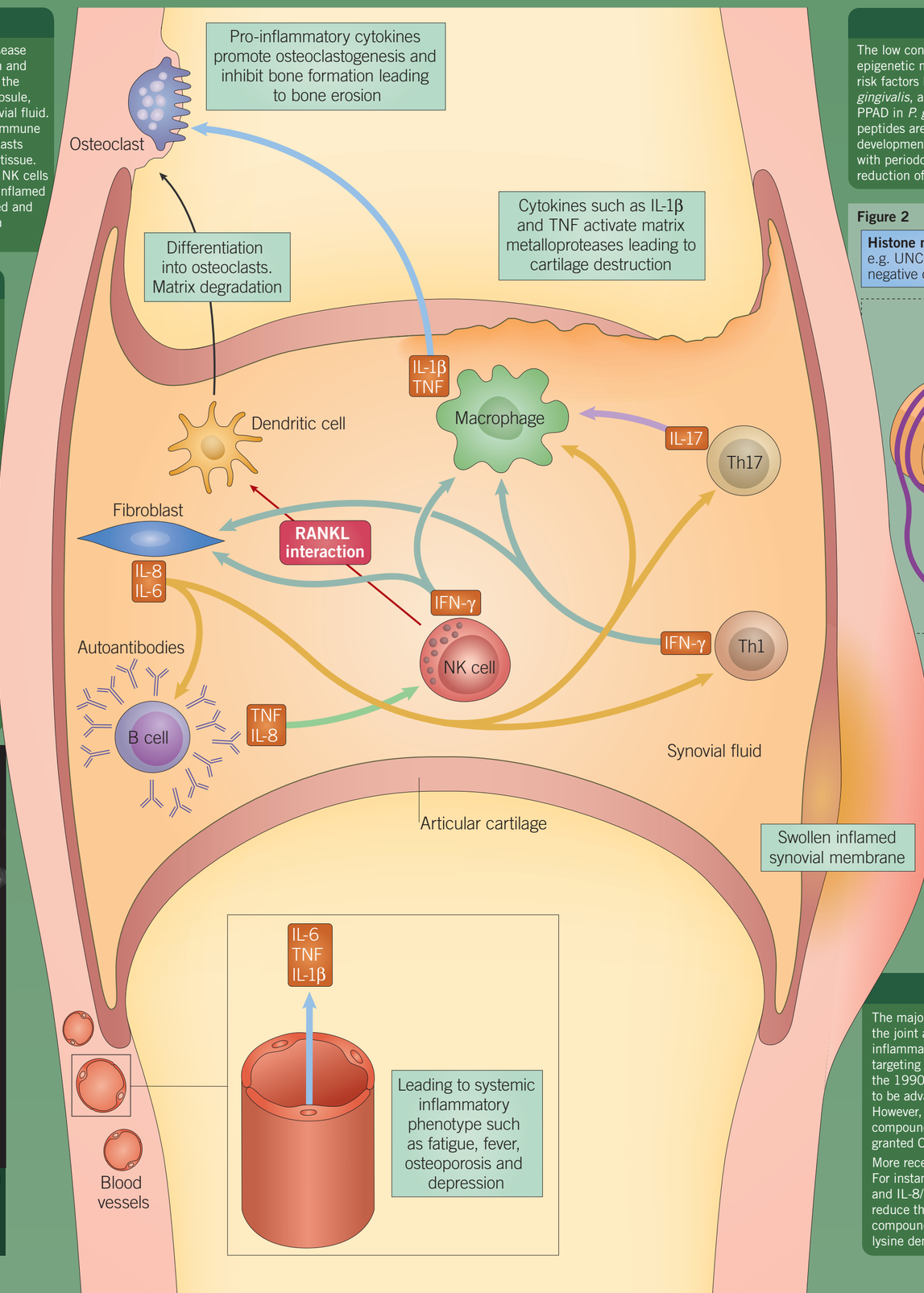


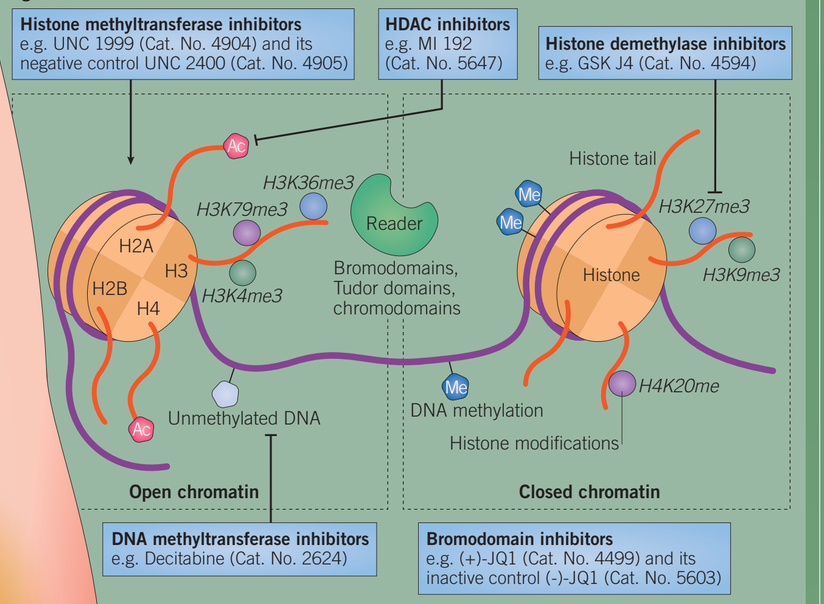
Figure 1 Typical X-ray of hand with advanced rheumatoid arthritis. Bone erosion, cartilage degradation and bone displacement are classic features of this disease (from Bernd Brägelmann via Wikimedia Commons)



Epigenetic and Environmental Evidence in RA

The low concordance rate of RA between monozygotic twins and the late onset of RA provide evidence that epigenetic modifications may be important for development of the disease. A number of potential environmental risk factors have been implicated, one of these being periodontitis. It has been shown that *Porphyromonas gingivalis*, a cause of periodontitis, expresses an enzyme with peptidylarginine deiminase (PAD) activity (called PPAD in *P. gingivalis*), which leads to the citrullination of peptide residues. Autoantibodies against citrullinated peptides are present in the majority of RA patients and it has been suggested that PPAD activity may drive the development of autoantibodies in RA[2]. Another important risk factor is cigarette smoking, which is also associated with periodontitis, and can heighten RA severity. One mechanism implicated in cigarette smoking is through the reduction of histone deacetylase (HDAC) SIRT1 and SIRT2 expression in macrophages within the lungs[3].

Figure 2



Epigenetic Modifications in RA

A number of epigenetic modifications have been identified as having pathogenic importance in RA. Synovial fibroblasts in RA display a DNA methylome pattern distinct from healthy fibroblasts. Similarly, $CD4^+$ T cell hypomethylation in RA patients has been shown to result in dysregulated cell function. In T_{regs}, DNA methylation of promoter and enhancers of key genes can also inhibit the ability of these cells to suppress the ongoing inflammation[4].

RA patients display a significantly increased expression of HDAC activity in PBMCs, suggesting that this pathway is dysregulated, while studies in synovial fibroblasts have also shown an increase in HDAC activity[5]. Investigations of histone methylation in the context of RA are rare, although the histone methyltransferase EZH2 has been shown to be overexpressed in RA fibroblasts, suggesting that H3K27me3 may be altered in RA.

Figure 2 shows some of the epigenetic modifications that may be involved in rheumatoid arthritis.

Epigenetic Pathways for Potential Intervention in RA

The major goal for RA drug discovery is to identify a compound that stops the inflammatory cascade within the joint and reverses the damage associated with RA. Current medicines, while successful at preventing inflammation, are not curative and are required for the lifetime of the patient. One potential advantage of targeting an epigenetic pathway in RA could be the ability to induce longer-term remission. Investigations in the 1990s using the DNA methylation inhibitor 5-azacytidine showed that targeting this pathway was unlikely to be advantageous since the drug led to self-reactive $CD4^+$ T cells and the induction of a lupus-like disease. However, HDAC inhibitors are effective for reducing several pro-inflammatory cytokines and one such compound, givinostat, has been evaluated in clinical trials to treat juvenile idiopathic arthritis and been granted Orphan Drug designation for this indication in the European Union.

More recently it has been demonstrated that targeting other epigenetic pathways may be beneficial for RA. For instance, BET and CBP/p300 bromodomain inhibition reduces the production of IL-17A from Th17 T cells and IL-8/IL-6 from fibroblasts[6]. Similarly, inhibition of the JMJD3/UTX jumonji histone demethylases can reduce the expression of IL-17A in Th17 cells[7]. Currently, drug discovery efforts are focused on identifying compounds that can target other epigenetic readers, writers and erasers of the histone code, such as kinases, lysine demethylases and histone methyltransferases.

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Cytokines
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DMARDs
Methotrexate

Indoleamine 2,3-dioxygenase
1-Methyl-D-tryptophan

Matrix Metalloprotease
Batimastat, GI 254023X, GM 6001, Marimastat

Nitric Oxide Signaling
1400W, BYK 191023, L-NAME, SNAP

p38
AMG 548, SB 202190, SB 203580, SB 239063, VX 745

Abbreviations
Ac, acetylation
DMARD, disease-modifying antirheumatic drug
IFN, interferon
IL, interleukin
NK, natural killer
PBMC, peripheral blood mononuclear cell
RANKL, receptor activator of nuclear factor κ-B ligand
Th, T helper
TNF, tumor necrosis factor
T_{regs}, regulatory T cells

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