New Targets in Rheumatoid Arthritis: SYK, JAKs, and BTK

Written by Maria Vinall

Iain B. McInnes, PhD, FRCP, University of Glasgow, Glasgow, United Kingdom, discussed the mechanisms of kinase inhibitors, on the targeted new therapies for rheumatoid arthritis (RA). The presentation was designed to inform clinicians about this new class of compounds and summarize key early studies to help guide clinical decision-making in the future.

The key concepts in RA pathogenesis and therapeutics are derived from the knowledge that RA is a genetic and epigenetic process that interacts with sequential, varied environmental factors that bring about alterations in the structure of cell proteins that in turn promote a breach of immunological tolerance (the inability of the body to tolerate its own proteins). This process can last from 5 to 10 years before the transition to overt synovitis. This early breach of tolerance, involving adaptive immunity, leads to a further cascade of events that promotes tissue remodeling, inflammation, and perturbations in innate immunity, driving joint damage and dysfunction. Cytokines regulate a broad range of inflammatory processes that are involved in the pathogenesis of RA, and an imbalance between pro- and anti-inflammatory cytokines favors the induction of autoimmunity, chronic inflammation, and subsequent joint damage.

Prof. McInnes stressed the need to understand intracellular signaling pathways in order to understand the new RA treatment paradigm that will support targeting these pathways. Signaling pathways transmit their signals via cascades of protein kinase-dependent interactions, mediated usually via phosphorylation (kinases) of tyrosine, threonine, or serine residues, from the cell membrane to the nucleus. There can be multiple members within a pathway and multiple pathways for one extracellular molecule. The purpose is to allow the cell to sense its extracellular environment by integrating varied external stimuli while enabling crossregulation of signals within the cell. Imagine the signal pathway as a complex circuit with redundancy and checkpoints built in.

Though numerous kinase inhibitors exist, they are not highly selective. The inhibitors that have been approved by the United States Food and Drug Administration still do not function as a one molecule inhibitor to one kinase inhibitor, which may be the ultimate goal. The new kinase inhibitors of interest for RA treatment include spleen tyrosine kinase (SYK), Janus kinase (JAK), and Bruton tyrosine kinase (BTK) inhibitors. When using kinases as targets in RA, the clinician should question whether or not the signaling pathway is present in relevant tissues (joint, lymph node, vasculature, etc), if the pathway actually mediates a relevant biological effect, and what happens upon manipulation of that pathway *in vitro* and *in vivo*.

SYK is a key regulator of cell signaling that is induced by B cell receptor or Fc receptor engagement and is widely expressed in RA synovium. It is involved in activation of synoviocytes, B cells, and macrophages. SYK is also implicated in osteoclast function. In one study, Western blot analysis demonstrated significantly greater amounts of phospho-SYK expression in RA synovial tissue compared with osteoarthritis synovial tissue. The kinase was expressed and functionally activated by TNF-α in fibroblast-like synoviocytes and was blocked by R406, the active ingredient of fostamatinib (an oral inhibitor of SYK) [Cha HS et al. *J Pharmacol Exp Ther* 2006]. Fostamatinib blocks SYK-dependent FcR-mediated activation of monocytes/macrophages and neutrophils and BCR-mediated activation of B lymphocytes [Braselmann S et al. *J Pharmacol Exp Ther* 2006]. Clinically, inhibition of SYK-kinase produced significant ACR 20, 50, and 70 response rates at 6 months in patients with active RA, despite methotrexate therapy [Weinblatt ME et al. *Arthritis Rheum* 2008; Weinblatt ME et al. *N Eng J Med* 2010]. Early reductions (1 week) in serum interleukin-6

Independent
Peer-Reviewed
Highlights from the

American College of Rheumatology 75th Annual Scientific Meeting

and matrix metalloproteinase 3 levels were also noted. Major adverse effects were gastrointestinal side effects (predominantly diarrhea) and neutropenia, both of which were dose-related, and hypertension. In contrast, no difference in ACR 20 response was observed in a 3-month double-blind, placebo-controlled trial of fostamatinib in patients who previously had an inadequate response to biologics [Genovese M et al. *Arthritis Rheum* 2010].

Janus kinases (JAKs) are a family of structurally distinct receptor-associated tyrosine kinases (JAK1, JAK2, JAK3, TYK2) that phosphorylate themselves, cytokine receptors, and STATS. They are associated with common γ-chain cytokine receptors. JAK1/JAK3 pathway signaling is important for immune cell development, survival, proliferation, and differentiation. JAK's actions on T, B, and NK cell development are important processes that are associated with RA. When used in murine collagen-induced and rat adjuvant-induced models of RA, CP-690,550 (tofacitinib), a small-molecule inhibitor of JAK3, dose-dependently decreased endpoints of disease in both models, with greater than 90% reduction observed at the highest administered dose. The compound also reduced inflammatory cell influx and joint damage, as measured histologically [Milici AJ et al. *Arthritis Res Ther* 2008]. In RA patients, the JAK3 inhibitor tofacitinib, at doses ≥3 mg BID, showed significantly higher ACR 20 responses ($p \leq 0.05$) through Week 24 when added to methotrexate compared with methotrexate alone among those with active disease and an inadequate response to methotrexate. Infections were the most common adverse and serious adverse events [Kremer JM et al. *Arthritis Rheum* 2011]. On-target and off-target toxicity concerns with JAK inhibition include immune suppression, increases in transaminases and serum creatinine, diarrhea, and dyslipidemia (elevated LDL-C and TG).

The BTK pathway is involved with B cell activation, maturation, and immunoglobulin production. It is also implicated in macrophage cytokine production and is involved with neutrophil/mast cell degranulation via immune complexes and osteoclastogenesis. The majority of BTK inhibitors that are in development are generally selective, irreversible, and of a small molecular size. PCI-32765, a BTK-selective inhibitor, is efficacious in collagen-induced arthritis and in immune-complex models that do not depend upon autoantibody production from B cells. Thus, PCI-32765 targets not only B lymphocytes but also monocytes, macrophages, and mast cells, which are important BTK-expressing effector cells in arthritis. PCI-32765 has been shown to reduce inflammation, and following stimulation of cultured human mast cells, PCI-32765 inhibits the release of histamine, PGD2, TNF-α, IL-8, and MCP-1 [Chang BY et al. *Arthritis Res Ther* 2011]. Another BTK inhibitor, CGI1746, was shown to block B cell receptor-dependent B cell proliferation and, in prophylactic regimens, reduced autoantibody levels in collagen-induced arthritis. CGI1746 also abolishes FcγRIII-induced TNFα, IL-1β, and IL-6 production in macrophages; decreases cytokine levels within joints; and ameliorates disease in myeloid- and FcγR-dependent autoantibody-induced arthritis models [Di Paolo JA et al. *Nat Chem Biol* 2011].

Prof. McInnes concluded by emphasizing that kinases are implicated in each phase of the pathogenesis of RA and therefore provide a plausible and enticing therapeutic target. Recent success in early clinical trials, as shown above, indicate the beginning of a new era of therapeutic agents with the ease of oral administration and therapeutic efficacy that may approach those of antibody-based therapy. As with all emerging treatments, the long-term safety and durability of response of kinase inhibitors will require ongoing study.