

# COMPENDIOUS REVIEW ON ANIMAL MODELS AND SIGNALING PATHWAYS INVOLVED IN THE PATHOGENESIS OF RHEUMATOID ARTHRITIS

<sup>1</sup>S. Selvakumar, <sup>2</sup>Dr. P. Amudha, <sup>3</sup>Esther Rani.E

Department of Pharmacology  
C. L. Baid Metha College of Pharmacy  
Thoraipakkam, Chennai-600 097.

**Abstract-** Rheumatoid arthritis is an autoimmune disease that can lead to joint discomfort, swelling, and stiffness. It occurs when the immune system malfunctions and targets the synovium, the tissue that lines the joints. As a part of the innate immune response, macrophages produce TNF, IL-6, IL-1, GM-CSF, IL-15, IL-18, IL-32, and chemokines that induce tissue inflammation. The inflammatory synovium thickens, causing the joint area to feel sensitive and painful, appear red and swollen, and become more difficult to move. Occasionally, RA can also result in complications for the heart, circulatory system, eyes, and/or lungs. Rheumatoid arthritis (RA) experimental models have made a significant contribution to our knowledge of the etiology and management of this inflammatory disease. Furthermore, a range of transgenic and genetically engineered knockout mice are available to analyze intricate disease processes. This is a big step forward for arthritis translational research. The current review will be focusing on improving knowledge of the experimental arthritis models in rodents and pathophysiology of RA based on the signaling pathways involved.

**Keywords:** RA, Chemokines, GM-CSF, transgenic model, signaling pathways.

## 1. INTRODUCTION:

Rheumatoid arthritis (RA) is a chronic inflammatory autoimmune disease that causes joint deterioration and inflammation, resulting in a shorter life expectancy, an increase in mortality rates, and increasing impairment. [1-4] In addition to causing lesions in the joints, RA can also cause anemia, osteoporosis, or weakness in the muscles. [1,5] These systemic symptoms can hinder the functioning of the urinary, neurological, and cardiovascular systems. [6,7] This condition often manifests between the ages of 35 and 60, with periods of remission and aggravation. It can also affect young children, even younger than 16 years old; this is known as juvenile RA (JRA). [5,8-10] Rheumatoid nodules under the skin, tiredness, fever, weight loss, and morning stiffness of the afflicted joints lasting more than thirty minutes are typical signs of RA. [8-10]

The fact that RA is more common in females than in males is most likely due to the strong immunity that females possess. [5] According to certain research, female hormones such as prolactin and estrogen may promote the development of B cell autoantibodies, which may contribute significantly to an individual's susceptibility to disease. [5,8,10]

Specifically, it has been observed that inflammatory mediators are important in the development of RA-related inflammation and joint degeneration. [13-15] Consequently, several RA therapeutic strategies seek to inhibit specific cytokines, including tumor necrosis factor (TNF) and interleukin-1 (IL-1), which have been shown to be crucial for the onset of RA. [13,16-19] However, in addition to their unclear long-term hazards and benefits, their extreme side effects, including potentially fatal infections and an elevated risk of cancer, restrict their usage in many communities. [20,21] Patients with RA can avoid joint deterioration and maintain their productivity at work by receiving early diagnosis and therapy. [22,23] The diagnosis of RA cannot be made with a single test. As a stand-in for diagnosis, classification criteria created by the European League Against Rheumatism (ACR/EULAR 2010) and the American College of Rheumatology (ACR 1987) are frequently utilized. [22,24,25]

Over the last few decades, numerous plant extracts and chemicals have been employed in basic and clinical studies regarding the management of RA, and a few of these have provided a reliable adjuvant therapeutic strategy. [20,21] In 2012, tofacitinib became the first JAK inhibitor to be licensed in the US for treating moderate to severe RA. [26-28] The EU and Japan thereafter followed suit with approvals. [26,29] The more recent JAK inhibitors are filgotinib (authorized in 2020 in Japan and the EU) and Upadacitinib (approved for RA in the US and the EU in 2019). [26,28,30] Peficitinib was approved in the Asian countries in 2019. Several regulatory bodies have now approved JAK inhibitors for use in various contexts. Any elevated risk of opportunistic hazards with JAK inhibitors is particularly relevant to their safe use in clinical practice, given their extensive use in the therapy of RA. [28,31]

Creating an animal model of a disease is a productive technique to investigate the course of the illness, its routes, medication treatment, behavioral characteristics, and associated pain. Animal models of RA have been developed, according to several research, although they are only useful for figuring out the pathogenesis and new treatment approaches. [32-35] On the other hand, little information is currently available about how behaviors contribute to the etiology of rheumatoid arthritis. [32,33,35] A crucial component of many research projects is behavioral analysis, which offers information on the behavioral expressions of molecular changes that take place as a disease progresses. The methodological technique known as behavioral phenotyping helps to clarify the physiological alterations, behavioral patterns, and underlying mechanisms that contribute to the development of disease. [32]

Collagen-induced arthritis (CIA) resembles human RA in many morphological aspects, including pannus development, articular cartilage and bone degradation, and synovitis. Similar biological components, namely the cytokines found in the cartilage and synovium, are shared by CIA and RA. Consequently, CIA has been commonly employed as the animal model in RA research since its development in 1977. [13,36]

## **2. SIGNALLING PATHWAYS INVOLVED IN THE PATHOGENESIS OF RA:**

### **2.1 JAK/STAT SIGNALING PATHWAY IN RA:**

The JAK-STAT signaling system is critical for cytokine signaling and has a role in inflammation, cell proliferation, differentiation, apoptosis, and immunological control. [6,37]

The JAK kinase family has four members: JAK1, JAK2, JAK3, and TYK2. Signal transduction is performed by different cytokine receptor families via unique JAK isoforms. Research indicates that JAK has a significant impact on RA. [6,37,38]

The STAT family includes STAT1-4, STAT5A, STAT5B, and STAT6. STATs are cytoplasmic proteins that activate transcription and transmit signals. The SH2 domain enables STAT to connect to an active receptor. STAT interacts with JAK to produce a dimer that enters the nucleus and regulates gene expression. [37-40]

#### **THE ROLE OF JAK/STAT IN THE PATHOGENESIS OF RA:**

Recently, aberrant JAK-STAT pathway activity has been linked to the onset and progression of RA. The interplay of pro-inflammatory cytokines phosphorylates JAK and activates STAT. STAT gene expression is linked to chronic inflammation and severity of RA joint damage. Inflammatory reactions in RA synovium include activation of adhesion molecule genes and cytokines, which are linked to specific signaling pathways through transcription factors. [6,37-40]

### **2.2 MAPK SIGNALING PATHWAY IN RA:**

The MAPK pathway includes protein kinases RAS, RAF, MEK, and ERK. Mitogen-activated protein kinase (MAP3K) triggers MAP2K, which then activates MAPK. RA can result from abnormal MAPK signaling pathway activation. Various external stimuli, such as neurotransmitters, hormones, inflammation, stress, viruses, and growth factors, can activate the MAPK pathway. Overactivation is linked to articular cartilage damage and synovial tissue inflammation. ERK1 and ERK2 play critical roles in cell differentiation, proliferation, and survival. Oxidative stress, ischemia, and neurotransmitter activation activate the ERK1/2 signaling pathway. [6,37,41-43]

In rheumatoid arthritis, P38 is a key member of the MAPK family that contributes to inflammation. In RA synovial tissue, p38 is activated and strongly expressed by MKK3 and MKK6,207. Commonly used p38 MAPK inhibitors suppress pro-inflammatory cytokines in neutrophils, macrophages/monocytes, and T cells. [6,37,46]

#### **THE ROLE OF MAPKS IN THE PATHOGENESIS OF RA:**

MAPKs have a crucial function in regulating pro-inflammatory cytokines and signaling downstream of IL-1, IL-17, and TNF- $\alpha$  receptors in damaged joint tissue. JNK MAPKs primarily contribute to the pathophysiology of RA by destroying cartilage through matrix metalloproteinase (MMP) activity. IL-1 $\beta$  and TNF- $\alpha$  activate the JNK signaling pathway, which promotes extracellular matrix breakdown by modulating MMP production in chondrocytes and FLS. [6,37,41-46]

### **2.3 THE PI3K/AKT SIGNALING PATHWAY:**

The PI3K family of kinases catalyzes phosphatidylinositol. PIP2 can be phosphorylated to PIP3 by PI3K, while PIP3 can be dephosphorylated to PIP2 by phosphatases like PTEN. This cycle ultimately ends PI3K signaling. PI3K primarily regulates PIP3. AKT, a serine/threonine kinase, plays a critical role in the PI3K pathway, influencing several cellular processes via multiple downstream effectors. [6,47-49]

The PI3K/AKT signaling pathway facilitates growth factor signaling in various cellular activities, including glucose homeostasis, lipid metabolism, protein synthesis, and cell survival. The PI3K-AKT pathway involves the phosphate group at position 3 of PIP3 recruiting PDK1 and AKT proteins to the plasma membrane. This causes PDK1 to phosphorylate threonine at position 308 (T308) of the AKT protein. This stimulates AKT, which triggers downstream regulatory pathways. [6, 47,48]

#### **THE ROLE OF PI3K/AKT IN THE PATHOGENESIS OF RA:**

The PI3K/AKT signaling pathway has been linked to RA onset and progression. RA patients have elevated PI3K expression in their synovial tissue, which may regulate synovial fibroblasts and cause inflammatory erosive arthritis. Inflammatory substances such as IL-1 $\beta$ , IL-6, IL-17, IL-21, IL-22, and TNF- $\alpha$  play a significant role in the development

of RA clinical alterations. The PI3K/AKT signaling pathway affects both FLS cell proliferation and synovial inflammation, as well as osteoclast development and production. [6,47-50]

### 3. ANIMAL MODELS OF RHEUMATOID ARTHRITIS:

There are several animal models of arthritis that have been used to evaluate the substances that may be used as new therapies for RA. Generally, animal RA models are used to assess the effectiveness of treatment medications. The immunologic features of these animal models help identify the similarities and differences between various animal models and humans. As a result, careful thought must go into choosing the right animal model for a variety of in vivo investigations.

#### 3.1 COLLAGEN-INDUCED ARTHRITIS:

Considering the striking similarities between real RA and collagen-induced arthritis (CIA), including synovitis, symmetric joint involvement, and cartilage and bone degradation, CIA is the most widely utilized animal model of human RA. Because of two characteristics such as the induction of autoantibodies against collagen and the self and the shattering of tolerance, the CIA model is the gold standard in vivo paradigm for studies on RA. In susceptible strains of rats and mice, type II collagen immunization in incomplete Freund's adjuvant (IFA) or complete Freund's adjuvant (CFA) induces arthritis, respectively. Though Th17 cells appear to play a major pathogenic role, both Th1 and Th17 responses are elevated in CIA. It is confined to the peripheral joints and primarily affects the fore and hind paws and rarely affects the knees. Notable characteristics of the consequent polyarthritis include moderate to severe synovitis, bone resorption, periosteal bone growth, and significant cartilage degeneration brought on by immune complex deposition on articular surfaces. At the time of the clinical beginning of arthritis, pro- and anti-inflammatory mediators, such as TNF- $\alpha$ , IL-1 $\beta$ , and IL-6, as well as transforming growth factor-beta isoforms, such as TGF $\beta$ 1, TGF- $\beta$ 2, and TGF- $\beta$ 3 can be seen in CIA joints. [1,36,51-54]

#### 3.2 ADJUVANT-INDUCED ARTHRITIS:

The adjuvant-induced arthritis model, often known as Freund's adjuvant arthritis, was developed in the 1950s by the bacteriologist Freund. Later, In 1956, Pearson discovered that rats inoculated with mineral oil containing heat-killed Mycobacterium tuberculosis (MT) particles, known as Complete Freund's Adjuvant (CFA), developed polyarthritis. Complete Freud's adjuvant (CFA) was injected subcutaneously once at the base of the tail to cause arthritis in rats. Adjuvant induction of RA can cause persistent joint deformities, including ankylosis, depending on how severe it is. Early phases of inflammation in the joint are characterized by the expression of cytokines that stimulate macrophages, such as TNF- $\alpha$ , IFN, and IL-17. Elevated cytokine levels are observed in the joint, as inflammation worsens. It is appropriate for researching the molecular relationships between T cells and subpopulations. The pathogenic alterations in this model, however, are self-limiting, and it does not have the chronic, progressive nature of RA. [52, 54-58]

#### 3.3 PRISTANE-INDUCED ARTHRITIS:

Pristane-induced arthritis (PIA) mimics the characteristics of human RA. Synovial hyperplasia, bone and cartilage erosions, inflammatory cell infiltration, and pannus-like development are the principal histological characteristics. Although the exact mechanism causing PIA is yet unknown, researchers have found that pristane stimulates the immune system to produce autoimmune responses when it encounters antigens on common environmental microorganisms. To develop arthritis, 150  $\mu$ l of pristane (2,6,10,14 tetramethylpentadecane) can be injected in mice, intradermally at the base of the tail, or 0.5 ml can be injected intraperitoneally. If the trial is intended to last 180 days (about 6 months), an equivalent booster injection is administered seven weeks later. After receiving a pristane injection, the rats are typically examined one to four times a week for 28 days (about 4 weeks). [59,60]

A macroscopic scoring system for the four limbs is commonly used to monitor arthritis development. The scoring scheme that is employed is as follows:

0.5 = Extremely mild ankle/wrist swelling

1= Mild swelling of paw

2= moderate swelling of paw

3= Marked swelling of paw

4= Significant swelling of paw

Thus, the total score for each animal will be 16.

#### 3.4 PROTEOGLYCAN-INDUCED ARTHRITIS:

PGIA can only be induced in the BALB/c and C3H mouse lines. Previously, CFA was employed as an adjuvant. However, DDA is now commonly used in trials due to its early onset and increased severity of arthritis. If Freund's adjuvants are utilized for immunization, females should be employed due to the aggressive behavior and high mortality rates of males following repeated intraperitoneal injections with FCA. Using DDA as an adjuvant reduces susceptibility and mortality among BALB/c colonies but causes more acute and dramatic disease compared to Freund's adjuvants.

On day 0, 21, and 42, human cartilage proteoglycans emulsified with dimethyl-dioctadecyl ammonium bromide (DDA) or Complete Freund's Adjuvant (CFA) are administered intraperitoneally. After the third inoculation, mice's paws,

including their ankle and wrist joints, were examined twice a week for signs of arthritis, such as swelling and redness. The degree of arthritis was determined visually on a scale of 0 to 4 for each paw, as described in pristane-induced arthritis.

Mice with PGIA exhibit CD4 T cell responses and an antibody response to PG. IgG 2a antibody levels correlate with disease severity. Antibodies generated in the joint may interact with proteoglycan in mouse cartilage, leading to complement deposition, immune cell activation, and cytokine and chemokine secretion.

PGIA is less commonly utilized than CIA because it requires considerable processing of human cartilage to obtain the necessary proteoglycan fraction. [52,61-63]

### **3.5 FORMALIN INDUCED ARTHRITIS:**

Formalin-induced arthritis is a well-known paradigm for studying anti-arthritic properties. When rats' knee joints are injected with formalin (0.5, 3, and 5%), they experience dose-dependent pain. Arthritis will be induced 30 minutes following administration of the vehicle/drug by sub-plantar injection of 0.1 ml formaldehyde (2% v/v) into an animal's hind paw on day 1 and 3. Arthritis will be measured using a Vernier caliper over a 10-day period to determine the mean increase in paw diameter. [64-66]

### **3.6 CARRAGEENAN-INDUCED ARTHRITIS:**

Carrageenan, also known as carrageenin, are a naturally occurring family of linear sulfated polysaccharides that are derived from edible red seaweed. Acute inflammation has been widely studied using the hind paw edema model caused by carrageenan. In this paradigm, intraarticular injections of 1% carrageenan cause arthritis in the tibiofemoral joint of rats. As a control, 0.1 milliliter of saline was injected into the contralateral paws. To assess the extent of edema in the footpad, a punch tissue biopsy will be performed. The five primary indicators of local inflammation caused by induced carrageenan are redness, swelling, heat, loss of function, and hypersensitivity. [67-69]

### **3.7 HUMAN TNF TRANSGENIC MICE:**

Most immune system cells produce pro-inflammatory cytokine tumor necrosis factor (TNF). The onset of autoimmune and inflammatory disorders like rheumatoid arthritis is associated with biologically active TNF. Numerous transcription-binding sites found in the human TNF gene operate differently depending on the cell type and stimulation. In 1991, the first transgenic mouse model for TNF was created. Since then, numerous research has used TNF transgenic mice to assess the pathways involved in rheumatoid arthritis, with a particular emphasis on anti-TNF therapy.

Transgenic mice expressing human TNF- $\alpha$  transgenes that are 3' modified and wild-type were created by Keffer et al. According to research conducted on transgenic mice, human TNF-transgenic (Tg197) mice spontaneously developed arthritis and displayed significant quantities of soluble and transmembrane human TNF  $\alpha$ . In this murine model, the mice showed signs of synovial hyperplasia and inflammatory cell infiltration in the joints after 3 to 4 weeks of age as well as pannus development, cartilage degradation, and bone erosion at about 10 weeks of age.

The mice used to create the human TNF transgenics have MHC haplotypes H-2K and H-2B, which are less conducive to the development of arthritis. Nonetheless, backcrossing to the DBA/1 background, which is prone to arthritis, led to an earlier beginning and the development of a more severe disease, suggesting that MHC may have an impact on the course of the disease. [70,71]

## **CONCLUSION:**

Understanding the mechanisms underlying the autoimmune arthritis disease process has been made easier by the variety of experimental models available for the condition. Over the last few years, significant progress has been made in both the development and testing of novel treatment medicines, such as JAK inhibitors, as well as in our understanding of the disease process, including the role of recently reported cytokines like IL-17, IL-23, and IL-27.

As a result, assessing the reliability of RA animal models will aid a better understanding of the disease's pathophysiology and the creation of secure and efficient treatment plans. An optimal animal model of RA, however, needs to strike a compromise between practicality and biological viability.

## **REFERENCES:**

1. Kessler J, Totoson P, Devaux S, Moretto J, Wendling D, Demougeot C. Animal models to study pathogenesis and treatments of cardiac disorders in rheumatoid arthritis: Advances and challenges for clinical translation. *Pharmacological Research*. 2021 Aug 1;170:105494.
2. Van den Hoek J, Boshuizen HC, Roorda LD, Tijhuis GJ, Nurmohamed MT, Van den Bos GA, Dekker J. Mortality in patients with rheumatoid arthritis: a 15-year prospective cohort study. *Rheumatology international*. 2017 Apr;37:487-93.
3. Bandyopadhyay D, Banerjee U, Hajra A, Chakraborty S, Amgai B, Ghosh RK, Haddadin FI, Modi VA, Sinha K, Aronow WS, Deedwania P. Trends of cardiac complications in patients with rheumatoid arthritis: analysis of the United States National Inpatient Sample; 2005-2014. *Current problems in cardiology*. 2021 Mar 1;46(3):100455.

4. Semb AG, Ikdahl E, Wibetoe G, Crowson C, Rollefstad S. Atherosclerotic cardiovascular disease prevention in rheumatoid arthritis. *Nature Reviews Rheumatology*. 2020 Jul;16(7):361-79.
5. Deshmukh R. Rheumatoid arthritis: pathophysiology, current therapeutic strategies and recent advances in targeted drug delivery system. *Materials Today Communications*. 2023 Mar 23:105877.
6. Liu S, Ma H, Zhang H, Deng C, Xin P. Recent advances on signaling pathways and their inhibitors in rheumatoid arthritis. *Clinical Immunology*. 2021 Sep 1;230:108793.
7. Firestein GS. The disease formerly known as rheumatoid arthritis. *Arthritis research & therapy*. 2014 Jun;16:1-3.
8. Bullock J, Rizvi SA, Saleh AM, Ahmed SS, Do DP, Ansari RA, Ahmed J. Rheumatoid arthritis: a brief overview of the treatment. *Medical Principles and Practice*. 2019 Mar 1;27(6):501-7.
9. Fox CQ. *Physician Assistant Clinical Review Cards*. FA Davis Company; 2002.
10. McInnes IB, Schett G. The pathogenesis of rheumatoid arthritis. *New England Journal of Medicine*. 2011 Dec 8;365(23):2205-19.
11. Chaudhari K, Rizvi S, Syed BA. Rheumatoid arthritis: current and future trends. *Nat Rev Drug Discov*. 2016 May 1;15(5):305-6.
12. Picerno V, Ferro F, Adinolfi A, Valentini E, Tani C, Alunno A. One year in review: the pathogenesis of rheumatoid arthritis. *Clin Exp Rheumatol*. 2015 Jul 1;33(4):551-8.
13. Zhang P, Han D, Tang T, Zhang X, Dai K. Inhibition of the development of collagen-induced arthritis in Wistar rats through vagus nerve suspension: a 3-month observation. *Inflammation Research*. 2008 Jul;57:322-8.
14. Tincani A, Andreoli L, Bazzani C, Bosisio D, Sozzani S. Inflammatory molecules: a target for treatment of systemic autoimmune diseases. *Autoimmunity reviews*. 2007 Nov 1;7(1):1-7.
15. McInnes IB, Schett G. Cytokines in the pathogenesis of rheumatoid arthritis. *Nature Reviews Immunology*. 2007 Jun;7(6):429-42.
16. Bresnihan B, Alvaro-Gracia JM, Cobby M, Doherty M, Domljan Z, Emery P, Nuki G, Pavelka K, Rau R, Rozman B, Watt I. Treatment of rheumatoid arthritis with recombinant human interleukin-1 receptor antagonist. *Arthritis & Rheumatism*. 1998 Dec;41(12):2196-204.
17. Åkerlund K, Erlandsson Harris H, Tracey KJ, Wang H, Fehniger T, Klareskog L, Andersson J, Andersson U. Anti-inflammatory effects of a new tumour necrosis factor-alpha (TNF- $\alpha$ ) inhibitor (CNI-1493) in collagen-induced arthritis (CIA) in rats. *Clinical & Experimental Immunology*. 1999 Jan;115(1):32-41.
18. Åkerlund K, Erlandsson Harris H, Tracey KJ, Wang H, Fehniger T, Klareskog L, Andersson J, Andersson U. Anti-inflammatory effects of a new tumour necrosis factor-alpha (TNF- $\alpha$ ) inhibitor (CNI-1493) in collagen-induced arthritis (CIA) in rats. *Clinical & Experimental Immunology*. 1999 Jan;115(1):32-41.
19. Wang S, Wang Y, Liu X, Guan L, Yu L, Zhang X. Anti-inflammatory and anti-arthritis effects of taraxasterol on adjuvant-induced arthritis in rats. *Journal of ethnopharmacology*. 2016 Jul 1;187:42-8.
20. Wang YZ, Sun SQ, Zhou YB. Extract of the dried heartwood of *Caesalpinia sappan* L. attenuates collagen-induced arthritis. *Journal of ethnopharmacology*. 2011 Jun 14;136(1):271-8.
21. Ahmed S, Anuntiyo J, Malemud CJ, Haqqi TM. Biological basis for the use of botanicals in osteoarthritis and rheumatoid arthritis: a review. *Evidence-Based Complementary and Alternative Medicine*. 2005 Sep 1;2:301-8.
22. De Pablo P, Dinnes J, Berhane S, Osman A, Lim Z, Coombe A, Raza K, Filer A, Deeks JJ. Systematic review of imaging tests to predict the development of rheumatoid arthritis in people with unclassified arthritis. *In Seminars in arthritis and rheumatism* 2022 Feb 1 (Vol. 52, p. 151919). WB Saunders.
23. De Pablo P, Filer A, Raza K, Buckley C. Does time matter in the management of rheumatoid arthritis?. *In Horizons in medicine 21: The proceedings of the Advanced Medicine conference 2009* 2009 Dec 7.
24. Aletaha D, Neogi T, Silman AJ, Funovits J, Felson DT, Bingham III CO, Birnbaum NS, Burmester GR, Bykerk VP, Cohen MD, Combe B. 2010 rheumatoid arthritis classification criteria: an American College of Rheumatology/European League Against Rheumatism collaborative initiative. *Arthritis & rheumatism*. 2010 Sep;62(9):2569-81.
25. Arnett FC, Edworthy SM, Bloch DA, Mcshane DJ, Fries JF, Cooper NS, Healey LA, Kaplan SR, Liang MH, Luthra HS, Medsger Jr TA. The American Rheumatism Association 1987 revised criteria for the classification of rheumatoid arthritis. *Arthritis & Rheumatism: Official Journal of the American College of Rheumatology*. 1988 Mar;31(3):315-24.
26. Winthrop K, Isaacs J, Calabrese L, Mittal D, Desai S, Barry J, Strengholt S, Galloway J. Opportunistic infections associated with Janus kinase inhibitor treatment for rheumatoid arthritis: A structured literature review. *In Seminars in Arthritis and Rheumatism* 2023 Feb 1 (Vol. 58, p. 152120). WB Saunders
27. Traynor K. FDA approves tofacitinib for rheumatoid arthritis. *American Journal of Health-System Pharmacy*. 2012 Dec 15;69(24):2120-1.
28. Takeuchi T, Tanaka Y, Tanaka S, Kawakami A, Song YW, Chen YH, Rokuda M, Izutsu H, Ushijima S, Kaneko Y. Safety and effectiveness of peficitinib (ASP015K) in patients with rheumatoid arthritis: final results (32

- months of mean peficitinib treatment) from a long-term, open-label extension study in Japan, Korea, and Taiwan. *Rheumatology and Therapy*. 2021 Mar;8:425-42.
29. Convertino I, Lorenzoni V, Gini R, Turchetti G, Fini E, Giometto S, Bartolini C, Paoletti O, Ferraro S, Cappello E, Valdiserra G. Drug-Utilization, Healthcare Facilities Accesses and Costs of the First Generation of JAK Inhibitors in Rheumatoid Arthritis. *Pharmaceuticals*. 2023 Mar 21;16(3):465.
  30. Conaghan PG, Mysler E, Tanaka Y, Da Silva-Tillmann B, Shaw T, Liu J, Ferguson R, Enejosa JV, Cohen S, Nash P, Rigby W. Upadacitinib in rheumatoid arthritis: a benefit–risk assessment across a Phase III program. *Drug Safety*. 2021 May;44:515-30.
  31. Tanaka Y, Izutsu H. Peficitinib for the treatment of rheumatoid arthritis: an overview from clinical trials. *Expert Opinion on Pharmacotherapy*. 2020 Jun 12;21(9):1015-25.
  32. Ghouri M, Lateef M, Liaquat L, Zulfquar A, Saleem S, Zehra S. Decreased muscle strength in adjuvant-induced rheumatoid arthritis animal model: A relationship to behavioural assessments. *Heliyon*. 2024 Jan 15;10(1).
  33. Hu Y, Cheng W, Cai W, Yue Y, Li J, Zhang P. Advances in research on animal models of rheumatoid arthritis. *Clinical rheumatology*. 2013 Feb;32:161-5.
  34. Kannan K, Ortmann RA, Kimpel D. Animal models of rheumatoid arthritis and their relevance to human disease. *Pathophysiology*. 2005 Oct 1;12(3):167-81.
  35. González-Chávez SA, Pacheco-Tena C. Exercise-driven exacerbation of inflammation: contribution of animal models of rheumatoid arthritis and spondyloarthritis. *Connective Tissue Research*. 2022 Sep 3;63(5):425-42.
  36. Wagner S, Bindler J, Andriambeloson E. Animal Models of Collagen-Induced Arthritis. *Current Protocols in Pharmacology*. 2008 Dec;43(1):5-1.
  37. Ding Q, Hu W, Wang R, Yang Q, Zhu M, Li M, Cai J, Rose P, Mao J, Zhu YZ. Signaling pathways in rheumatoid arthritis: implications for targeted therapy. *Signal Transduction and Targeted Therapy*. 2023 Feb 17;8(1):68.
  38. Villarino AV, Kanno Y, O'Shea JJ. Mechanisms and consequences of Jak–STAT signaling in the immune system. *Nature immunology*. 2017 Apr;18(4):374-84.
  39. Kisseleva T, Bhattacharya S, Braunstein J, Schindler CW. Signaling through the JAK/STAT pathway, recent advances and future challenges. *Gene*. 2002 Feb 20;285(1-2):1-24.
  40. de Araujo ED, Orlova A, Neubauer HA, Bajusz D, Seo HS, Dhe-Paganon S, Keserü GM, Moriggl R, Gunning PT. Structural implications of STAT3 and STAT5 SH2 domain mutations. *Cancers*. 2019 Nov 8;11(11):1757.
  41. Cargnello M, Roux PP. Activation and function of the MAPKs and their substrates, the MAPK-activated protein kinases. *Microbiology and molecular biology reviews*. 2011 Mar;75(1):50-83.
  42. Kyriakis JM, Avruch J. Mammalian MAPK signal transduction pathways activated by stress and inflammation: a 10-year update. *Physiological reviews*. 2012 Apr;92(2):689-737.
  43. Gurgis FM, Ziaziaris W, Munoz L. MAPK-activated protein kinase 2 (MK2) in neuroinflammation, Hsp27 phosphorylation and cell cycle: Role and Targeting. *Molecular Pharmacology*. 2013 Dec 2.
  44. Sabio G, Davis RJ. TNF and MAP kinase signalling pathways. In *Seminars in immunology 2014 Jun 1 (Vol. 26, No. 3, pp. 237-245)*. Academic Press.
  45. Ralph JA, Morand EF. MAPK phosphatases as novel targets for rheumatoid arthritis. *Expert opinion on therapeutic targets*. 2008 Jul 1;12(7):795-808.
  46. Sujitha S, Rasool M. MicroRNAs and bioactive compounds on TLR/MAPK signaling in rheumatoid arthritis. *Clinica Chimica Acta*. 2017 Oct 1;473:106-15.
  47. Markman B, Dienstmann R, Tabernero J. Targeting the PI3K/Akt/mTOR pathway–beyond rapalogs. *Oncotarget*. 2010 Nov;1(7):530.
  48. Sun K, Luo J, Guo J, Yao X, Jing X, Guo F. The PI3K/AKT/mTOR signaling pathway in osteoarthritis: a narrative review. *Osteoarthritis and cartilage*. 2020 Apr 1;28(4):400-9.
  49. Ersahin T, Tuncbag N, Cetin-Atalay R. The PI3K/AKT/mTOR interactive pathway. *Molecular BioSystems*. 2015;11(7):1946-54.
  50. Ba X, Huang Y, Shen P, Huang Y, Wang H, Han L, Lin WJ, Yan HJ, Xu LJ, Qin K, Chen Z. WTD attenuating rheumatoid arthritis via suppressing angiogenesis and modulating the PI3K/AKT/mTOR/HIF-1 $\alpha$  pathway. *Frontiers in Pharmacology*. 2021 Sep 27;12:696802.
  51. Joe B, Griffiths MM, Remmers EF, Wilder RL. Animal models of rheumatoid arthritis and related inflammation. *Current rheumatology reports*. 1999 Dec;1(2):139-48.
  52. Kong JS, Jeong GH, Yoo SA. The use of animal models in rheumatoid arthritis research. *Journal of Yeungnam Medical Science*. 2023 Jan 1;40(1):23-9.
  53. Bevaart L, Vervoordeldonk MJ, Tak PP. Evaluation of therapeutic targets in animal models of arthritis. *Arthritis Rheum*. 2010 Aug;62(8):2192-205.
  54. Zhao T, Xie Z, Xi Y, Liu L, Li Z, Qin D. How to model rheumatoid arthritis in animals: from rodents to non-human primates. *Frontiers in Immunology*. 2022 May 25;13:887460.

55. Bordy R, Moretto J, Devaux S, Wendling D, Moretto-Riedweg K, Demougeot C, Totoson P. Adjuvant-induced arthritis is a relevant model to mimic coronary and myocardial impairments in rheumatoid arthritis. *Joint Bone Spine*. 2021 Jan 1;88(1):105069.
56. Wang DD, He CY, Wu YJ, Xu L, Shi C, Olatunji OJ, Zuo J, Ji CL. AMPK/SIRT1 deficiency drives adjuvant-induced arthritis in rats by promoting glycolysis-mediated monocytes inflammatory polarization. *Journal of Inflammation Research*. 2022 Jan 1:4663-75.
57. Snehalatha U, Anburajan M, Venkatraman B, Menaka M. Evaluation of complete Freund's adjuvant-induced arthritis in a Wistar rat model. *Z Rheumatol*. 2013 May 1;72(4):375-82.
58. Moudgil KD, Kim P, Brahn E. Advances in rheumatoid arthritis animal models. *Current rheumatology reports*. 2011 Oct;13:456-63.
59. Wooley PH, Seibold JR, Whalen JD, Chapdelaine JM. Pristane-induced arthritis. the immunologic and genetic features of an experimental murine model of autoimmune disease. *Arthritis & Rheumatism: Official Journal of the American College of Rheumatology*. 1989 Aug;32(8):1022-30.
60. Patten C, Bush K, Rioja I, Morgan R, Wooley P, Trill J, Life P. Characterization of pristane-induced arthritis, a murine model of chronic disease: response to antirheumatic agents, expression of joint cytokines, and immunopathology. *Arthritis & Rheumatism: Official Journal of the American College of Rheumatology*. 2004 Oct;50(10):3334-45.
61. Glant TT, Radacs M, Nagyeri G, Olasz K, Laszlo A, Boldizsar F, Hegyi A, Finnegan A, Mikecz K. Proteoglycan-induced arthritis and recombinant human proteoglycan aggrecan G1 domain-induced arthritis in BALB/c mice resembling two subtypes of rheumatoid arthritis. *Arthritis & rheumatism*. 2011 May;63(5):1312-21.
62. Glant TT, Mikecz K. Proteoglycan aggrecan-induced arthritis: a murine autoimmune model of rheumatoid arthritis. *Autoimmunity: Methods and Protocols*. 2004:313-38.
63. Angyal A, Egelston C, Kobezda T, Olasz K, László A, Glant TT, Mikecz K. Development of proteoglycan-induced arthritis depends on T cell-supported autoantibody production, but does not involve significant influx of T cells into the joints. *Arthritis research & therapy*. 2010 Apr;12(2):1-6.
64. Patel MG, Pundarikakshudu K. Anti-arthritis activity of a classical Ayurvedic formulation Vatari Guggulu in rats. *Journal of traditional and complementary medicine*. 2016 Oct 1;6(4):389-94.
65. Manan M, Saleem U, Ahmad B, Aslam N, Anwar A, Zafar A. Anti-arthritis and toxicological evaluation of ethanolic extract of *Alternanthera bettzickiana* in rats. *Frontiers in Pharmacology*. 2022 Oct 24;13:1002037.
66. Sharma H, Chauhan P, Singh S. Evaluation of the anti-arthritis activity of *Cinnamomum cassia* bark extract in experimental models. *Integrative medicine research*. 2018 Dec 1;7(4):366-73.
67. Dhalendra G, Satapathy T, Roy A. Animal models for inflammation: A review. *Asian Journal of Pharmaceutical Research*. 2013;3(4):207-12.
68. Pradhan B, Ki JS. Biological activity of algal derived carrageenan: A comprehensive review in light of human health and disease. *International Journal of Biological Macromolecules*. 2023 Mar 21:124085.
69. Shao X, Wang X, English SJ, Desmond T, Sherman PS, Quesada CA, Piert MR. Imaging of carrageenan-induced local inflammation and adjuvant-induced systemic arthritis with [11C] PBR28 PET. *Nuclear medicine and biology*. 2013 Oct 1;40(7):906-11.
70. Minshawi F, White MR, Muller W, Humphreys N, Jackson D, Campbell BJ, Adamson A, Papoutsopoulou S. Human TNF-Luc reporter mouse: a new model to quantify inflammatory responses. *Scientific reports*. 2019 Jan 17;9(1):193.
71. Keffer J, Probert L, Cazlaris H, Georgopoulos S, Kaslaris E, Kioussis D, Kollias G. Transgenic mice expressing human tumour necrosis factor: a predictive genetic model of arthritis. *The EMBO journal*. 1991 Dec;10(13):4025-31.