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Nanomedicine delivers promising treatments for rheumatoid arthritis

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Abstract

An increased understanding in the pathophysiology of chronic inflammatory diseases, such as rheumatoid arthritis, reveals that the diseased tissue and the increased presence of macrophages and other overexpressed molecules within the tissue can be exploited to enhance the delivery of nanomedicine. Nanomedicine can passively accumulate into chronic inflammatory tissues via the enhanced permeability and retention phenomenon, or be surface conjugated with a ligand to actively bind to receptors overexpressed by cells within chronic inflammatory tissues, leading to increased efficacy and reduced systemic side-effects. This review highlights the research conducted over the past decade on using nanomedicine for potential treatment of rheumatoid arthritis and summarizes some of the major findings and promising opportunities on using nanomedicine to treat this prevalent and chronic disease.

Keywords

chronic inflammation; enhanced permeability and retention; liposomes; nanomedicine; nanoparticles; rheumatoid arthritis

Introduction to chronic inflammatory disease

Inflammation is a normal response that protects tissues from infection or injury. The normal cycle of acute inflammation includes the activation of inflammatory mediators and recruitment of monocytes from circulation to remove foreign pathogens at the inflammation site. The resolution of inflammation consists of downregulation of proinflammatory mediators, release of anti-inflammatory mediators and the removal or clearance of apoptotic cells by phagocytes (i.e., efferocytosis) [1–4]. Acute inflammation promotes tissue repair (through the production of anti-inflammatory mediators), removes damaging pathogens and

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restores normal tissue functions; however, when the inflammatory trigger is not cleared or is persistent, or when the inflammation is nonresolving for other reasons, chronic inflammation can arise. Chronic inflammation involves the ongoing induction of proinflammatory mediators, infiltration of monocytes into the tissue and ultimately leads to tissue damage. This non-resolving inflammatory response, which may include overexpressed anti-inflammatory mediators, damaged tissues, necrotic monocytes that are not cleared through the lymphatic system and other factors, can become an inflammatory trigger in itself, and can result in an adaptive immune response [5–7]. Genetic factors, environmental triggers as well as adaptive immune response can lead to chronic inflammatory disease [8–10].

Rheumatoid arthritis (RA), inflammatory bowel disease (IBD), chronic obstructive pulmonary disease (COPD) and systemic lupus erythematosus (SLE) are examples of chronic inflammatory diseases. In this review, RA and its therapies will be highlighted. RA affects approximately 1.0% of the population in developed countries, affecting women three-times more than men [11]. The costs associated with disease management in the USA have been reported as US$19.3–39.2 billion per year for RA, with approximately 30% of that costs covered by patients [12–17]. The range of costs can be associated with direct and indirect medical expenses, costs of therapy chosen and the duration of disease. The CDC estimates 15,600 RA hospitalizations annually, further illustrating the prevalence and extent of care required for managing this disease [18].

The clinical presentation of RA is characterized by synovial inflammation, which can lead to deformation, bone erosion as well as loss of joint function. Other clinical manifestations often include the presence of rheumatoid factor (RF) and anticitrullinated protein antibody (ACPA) in the blood, muscle soreness and joint tenderness, as well as increased risk for cardiovascular, pulmonary and skeletal disorders [8]. The etiology of the disease is not completely understood; however, connections have been made to the HLA genes as well as other risk factors [4,19].

Most chronic inflammatory diseases can be controlled, but not cured, with currently available therapies. The current standard of care (SOC) for RA consists of: anti-inflammatory drugs, including nonsteroidal anti-inflammatory drugs (NSAIDs) and corticosteroids; disease-modifying antirheumatic drugs (DMARDs); biologic agents and surgery as needed. NSAIDs are primarily used to reduce minor inflammation and relieve pain; however, they do not reduce joint damage and are associated with significant side-effects including gastrointestinal bleeding, fluid retention and increased risk of heart disease. Corticosteroids have beneficial anti-inflammatory and immunosuppressive properties, making them applicable for bridging therapy with other agents, as low-dose therapy for continuous treatment, as high-dose therapy for the treatment of flares or administered intra-articularly for symptomatic relief. Their use, however, is greatly limited by adverse effects such as adrenal suppression, glucose intolerance and increased susceptibility to infections. DMARDs include methotrexate (MTX), hydroxychloroquine, sulfasalazine and leflunomide. Early introduction of these agents has been shown to provide a more favorable outcome in patients, and they are known to reduce disease progression. However, these agents are not without side-effects, namely gastrointestinal complications, liver toxicity and hematologic adverse events. Biologic agents directly target components of the immune

response, including proinflammatory cytokines and immune cells. Examples of these proinflammatory cytokines include TNF-α, which is the therapeutic target of infliximab, etanercept, adalimumab, golimumab, certolizumab; IL-1, the therapeutic target of anakinra; and IL-6, the therapeutic target of tocilizumab. Biologics also target B cells (rituximab) and block the costimulatory signal required for T-cell activation (as seen with abatacept). Despite their efficacy, biologics are also associated with significant adverse effects, specifically increased risk of infections (e.g., tuberculosis) and certain types of cancers [20,21].

To impact the progression of disease, the pathogenesis of disease must be understood. Although the etiology of disease remains unknown, great strides in research have led to the identification of agents involved in the initiation and propagation of the inflammatory response. Proinflammatory cytokines TNF-α, IL-1 and IL-6 have been the targets of biologic therapies such as Humira (adalimumab), a TNF-α inhibitor used for the treatment of RA, Crohn's disease and ulcerative colitis [20–22]. Intracellular signal pathways have also been studied, with a focus on protein kinases. Kinase inhibitors block protein phosphorylation, thereby preventing the activation of transcription factors that control the release of proinflammatory mediators, including the aforementioned TNF-α, IL-1 and IL-6. Protein kinases including JAK, Syk, P13K and p-38 MAPK have been identified in the signal transduction pathway for RA [20,21,23–25]. Xeljanz (tofacitanib), the JAK inhibitor, has been approved for use in patients with RA.

Overview of nanomedicine

Nanomedicine is defined as the application of nanotechnology in the diagnosis, treatment or prevention of disease. Nanomedicines may include drug-loaded liposomes, nanoparticles, polymeric micelles, nanogels and nanocapsules [26]. In addition, polymer–drug conjugates, polymer–protein conjugates and antibodies are all classified as nanomedicines [26]. Nanomedicines can be designed to: protect the therapeutic agent from degradation, remain in blood circulation longer, be tailored for macrophage uptake or targeted to certain receptors and permeate through certain diseased tissues as interendothelial cell gaps are generally 1–2 nm in healthy tissues [27–30], but can be up to 600 nm in diseased tissues, such as inflamed joints [31,32].

Nanomedicines can be prepared with polymers, lipids as well as inorganic nanostructures. Lipid-, polymer-, and hybrid lipid–polymer-based nanomedicines are often utilized for intravenous (iv.) administration and include, but are not limited to, liposomes, PEGylated liposomes, polymersomes, micelles, dendrimers and hydrogel nanoparticles. Nanoparticle system should be designed considering: the properties of the active molecule to be delivered, the biological target, the environment prior to reaching the target and the environment at the target sites [33,34]. Hydrophobic and hydrophilic properties of polymers and lipids can be manipulated to encapsulate hydrophobic or hydrophilic active moieties. The types of polymers and lipids used can i) be determined based on compatibility with the active drugs or ii) modify drug-release property. Ligands or antibodies can be conjugated onto the nanoparticles to facilitate active targeting, if a known receptor is overexpressed in diseased tissues, which will be discussed in more details later. Furthermore, PEG chains can be

covalently conjugated onto nanoparticles, also known as PEGylation, to decrease clearance of the nanoparticles when in circulation by providing a hydrophilic and steric barrier against opsonization [34,35].

Inflammatory tissue as a target for drug delivery

There has been a surge in the use of nanomedicine for drug delivery in the treatment of cancer, as solid tumor environment consists of 'leaky' vasculature from gaps in the endothelial cell lining and fenestrations that allow for higher permeability of relatively large molecules and particles. Additionally, the presence of other mediators in the diseased tissue can also increase permeability. For example, TNF-α elicits monocyte recruitment from circulation and stimulates additional pores in the endothelial lining. This enhanced permeability paired with an impaired lymphatic drainage system in tumor tissues is known as the enhanced permeability and retention (EPR) effect [34,36]. Utilizing the EPR phenomenon for targeted drug delivery may increase the efficacy and reduce the systemic toxicities of potent anticancer agents [37,38].

The process of monocyte recruitment from circulation and the development of endothelial gaps that facilitate plasma leakage into the injured site are characteristic of inflamed tissues. In chronic inflammatory conditions, inflammatory mediators can be overexpressed and persistent, leading to 'leaky' vasculature similar to that seen in solid tumors [39,40]. In addition to increased permeability, inflamed tissues also have more activated macrophages or other monocytes that can be utilized as targets for site-specific drug delivery. Moreover, it has been shown that certain cell adhesion molecules (CAMs) are overexpressed on endothelial cells in IBD [41] and that vasoactive intestinal peptide (VIP) receptors are overexpressed in activated synoviocytes in patients with RA [42]. Ligands specific to those overexpressed molecules can be conjugated to nanomedicine to actively target drug to inflammatory tissues.

Current landscape of nanomedicine for RA

Adalimumab (Humira), etanercept (Enbrel) and infliximab (Remicade) are currently among the top ten best-selling drugs in the USA. They are biologics used for the treatment of RA and other inflammatory diseases. However, significant adverse effects may occur with the use of these biologics, leaving patients vulnerable to serious infections such as tuberculosis [43]. The use of nanocarriers allows for increased site-specific drug delivery to inflamed tissues, by utilizing the disease state including, but not limited to, enhanced permeability or changes in pH of inflamed tissues and by utilizing monocytes as active targets for drug delivery. For RA, iv., intra-articular (ia.) and subcutaneous (sc.) administration routes will be reviewed as these allow for systemic delivery to circulation with access to 'leaky' vasculature, or local administration directly to diseased tissues; thereby, allowing for maximum drug action. Note that this may differ depending on the disease state; for example, when treating IBD, nanoparticles may be dosed orally because the intestine is the diseased tissue for this condition.

Studies of nanomedicines for potential treatment of RA are summarized in Table 1. These include *in vitro*, *in vivo* and clinical studies utilizing nanomedicines for targeted drug

delivery to diseased tissues in RA animal models or patients. This summary includes information from searches of multiple databases of scientific literature, including PubMed and ScienceDirect as well as for clinical trials [44]. These searches were limited to publications and clinical applications within the last 10 years. These studies evaluated the use of passive or active targeting for drug delivery, as well as the ability to increase the efficacy of existing therapies by utilizing nanomedicines.

Taking advantage of enhanced permeability

The passive targeting of nanomedicines to inflamed tissues based on enhanced permeability has been supported by various *in vivo* biodistribution studies [51,56,59,63,70]. Ishihara *et al.* showed that PEGylated polymersomes encapsulated with the glucocorticoid betamethasone preferentially accumulated in inflamed joints in a mouse model of antibody-induced arthritis. The high accumulation correlated with reduction in arthritic score, as well as reduced expression of proinflammatory cytokine, IL-6. *In vivo* imaging showed that the accumulation of the polymersomes in the joints maintained for up to 96 h, which led to a sustained therapeutic effect for 8 days [56].

Glucocorticoids are often utilized for patients with RA, and are considered potent anti-inflammatory agents; however, the exact mechanism of action of this class of drugs is not completely understood. Encapsulation of them into liposomes or polymersomes allows for more local delivery and accumulation to inflammation sites due to the EPR effect, thereby reducing systemic side-effects and enhancing therapeutic efficiency. Hofkens *et al.* showed that prednisolone phosphate encapsulated in PEGylated liposomes was able to downregulate the activation of proinflammatory macrophages and upregulate anti-inflammatory macrophages *in vitro*; however, only the downregulation of proinflammatory macrophages was observed *in vivo* [51]. The authors also conducted biodistribution studies to confirm that after iv. or sc. administration, the liposomes extravasate through leaky vasculature into synovial tissues and are engulfed by macrophages within the inflamed tissues [50], further supporting the utilization of the enhanced permeability for targeted delivery of anti-inflammatory agents. After macrophage uptake, significant reductions were seen in the expression of proinflammatory cytokines including TNF-α, IL-1β, IL-8 as well as CD86 protein, giving insight into the mechanism of action of the prednisolone phosphate [51]. Because of the promising *in vivo* results, safety studies were conducted for repeat dosing of the liposomal prednisolone phosphate, as well as dose range finding. It was concluded that the safety profile of the glucocorticoid benefited from the liposomal formulation, and that the effective dose and dose frequency of the glucocorticoid could be reduced in animal models by as much as tenfold; showing comparable efficacy with four daily injections of 10 mg/kg of free drug to a single dose of 1 mg/kg prednisolone phosphate in the liposomes [50]. The ability of nanoparticle formulations of glucocorticoids to suppress proinflammatory cytokines such as TNF-α at a lower effective dose and dose frequency may be advantageous to decrease the broader immunosuppression seen with many biologic TNF-α inhibitors on the market. A Phase II clinical study with liposomal prednisone has been conducted, confirming the safety and increased efficacy of the liposomal prednisone relative to free drug.

Ulmanksey *et al.* used an adjuvant arthritis (AA) rat model to evaluate two formulations of PEGylated liposomes, one containing methylprednisolone and the other betamethasone, against free drug as well as biologic TNF-α inhibitors, etanercept and infliximab. Their *in vivo* study results showed that the liposomal formulations led to a significant decrease in proinflammatory mediators and a longer duration of therapeutic effect when compared with free drugs or to the biological therapies; indicating a promising path forward toward a more effective and less frequent therapy using glucocorticoid–nanomedicines as an alternative to the immunosuppressant biologic therapies [53].

Selective biodistribution in inflamed tissues due to enhanced permeability and the resultant lower effective dose and longer duration of drug action leading to decreased dose frequency are a recurring theme with nanomedicines in RA models [45,50,53,55,57,58]. For example, Ulmansky *et al.* reported a longer duration of drug action *in vivo* with a liposomal glucocorticoid formulation, resulting in weekly administration, as opposed to daily injections of the free drug as well a decrease in effective dose ranging from two- to 25-fold [53,54]. Hwang *et al.* showed that delivery of α-methylprednisolone using a conjugated cyclodextrin polymer-based nanoparticle formulation reduced the dosing frequency from daily to weekly and the effective dose by up to 100-fold, with a comparable or superior suppression of RA symptoms [55].

Active targeting for increased site-specific delivery

Although the accumulation of the nanocarriers in inflamed tissues due to enhanced permeability can help to decrease effective dose and dose frequency, increased knowledge of the disease tissues could lead to even more discriminating biodistribution. Chronic inflammation is characterized by a persistent inflammatory response that includes the infiltration of macrophages, lymphocytes and plasma from circulation eventually leading to tissue damage. In the case of RA, tissue damage is mainly seen in the joints, leading to bone erosion and deformation. The affected joint tissue is characterized by an increased expression of adhesion molecules and chemokines, which can be exploited for active drug targeting. Recent studies indicate that folate receptor-β (FR-β) expression is elevated on activated macrophages in inflamed joints in RA [83]. With this information, Thomas *et al.* prepared MTX dendrimer nanoparticles with folic acid (FA) as a targeting ligand. MTX is one of the most commonly prescribed DMARDs, alone or in combination with biologic therapy; however, many patients are intolerant to the medication due to significant side-effects. Targeted delivery of MTX may help reduce these side-effects. Thomas *et al.* showed that the FA-conjugated dendrimer nanoparticles selectively bound to the activated macrophages *in vitro* and led to significant disease suppression *in vivo* [62]. Additionally, it was shown that the maximum-tolerated dose (MTD) of the MTX in nanoparticles was 7.5-fold higher than that of the free MTX [62]. The FA as a targeting ligand was also used in chitosan-DNA nanoparticles to enhance the delivery of a plasmid that encodes IL-1Ra, a receptor antagonist and natural blocker of IL-1, and the FA-conjugated chitosan-DNA nanoparticles were shown to have less cytotoxic effects than the FA-free nanoparticles, likely due to the active uptake of the particles by activated macrophages, reducing toxicity to other cells [80].

Vascular endothelial cells (VECs) are involved in the recruitment of monocytes from circulation into inflamed tissues during inflammation. They are also involved in angiogenesis during tissue repair. VECs have certain cell adhesion molecules (CAMs) or growth factor receptors that may be overexpressed in inflamed tissues and could be used as targets for active drug delivery [74,84–86]. Koning *et al.* evaluated the delivery of dexamethasone with PEGylated liposomes surface-conjugated with a specific peptide ligand to target the $\alpha_v\beta_3$ receptor, an integrin overexpressed on VECs in inflammation sites. In an *in vivo* AA rat model, the targeted liposomes showed a threefold increase in accumulation in the inflammation site, compared with the nonconjugated liposomes, which correlated with a significant decrease in arthritic severity score and a longer therapeutic effect [59]. This increased localization was interesting, as the conjugated liposomes were cleared significantly faster from circulation than the nonconjugated liposomes, showing how effective the targeted liposomes were, even with a shorter circulation time [59]. The same receptor was also targeted for the delivery of an angiogenesis inhibitor, fumagillin and the targeted fumagillin showed a higher affinity to inflamed tissues, decreased leukocytes recruited into the tissues, and suppressed inflammation [70]. In addition, the effective dose of the optimized targeted fumagillin nanoparticle formulation was decreased by eightfold [70].

VIP is a hormone active in the resolution of inflammation; therefore, even though resolution of inflammation is not achieved, VIP receptors tend to be overexpressed in activated macrophages and proliferating synoviocytes in RA. The conjugation of VIP to nanoparticles would allow for active targeting of overexpressed receptors; they could also potentially provide therapeutic value by downregulating proinflammatory cytokines and upregulating anti-inflammatory cytokines [87]. Koo *et al.* utilized VIP as a targeting ligand to deliver camptothecin (CPT), an anticancer drug that induces cell death, to the overproliferating synoviocytes [42]. In their study, the VIP was utilized at a very low dose and it alone had no therapeutic effect. CPT, which is a topoisomerase inhibitor, is limited in clinical use due to its systemic toxicity, including hematologic toxicity. Koo *et al.* reported that low doses of CPT in PEGylated lipid micelles that were conjugated with VIP showed a significant reduction in arthritic score compared with micelles that did not include the VIP ligand, as well as free CPT at threefold higher dose [42]. Histology analysis also showed a significant reduction of macrophages and synoviocytes in the previously inflamed tissue after treatment with CPT in the VIP-conjugated PEGylated micelles [42]. The dose of CPT used was approximately 100-fold lower than used for cancer therapy, and no systemic toxicity was observed in the CIA mouse model, making this a potentially promising new agent for the treatment of RA [42].

Advancing the SOC

The application of nanoparticles in molecular therapy is a growing area of research, as seen in Table 1. A clinical study using ia.-injected adeno-associated virus (AAV) to deliver a TNF agonist gene showed the proof of concept for the use of gene therapy in RA; however, the AAV therapy also resulted in significant side-effects, including joint swelling and discomfort at the injection site [72,88]. siRNA that specifically inhibits the expression of certain genes, such as TNF-α, that are critical in RA development and progression is an

interesting alternative to biologic agents such as anti-TNF-α antibodies, because it has the potential to selectively inhibit the expression of proinflammatory genes in targeted cells, while avoiding or minimizing the systemic side-effects associated with current biologics. However, siRNA is largely ineffective when given alone *in vivo*, and is thus generally formulated into nanocarriers such as liposomes or polymeric nanoparticles to protect the siRNA and deliver it to inflamed sites [89–91]. For example, Scheinman *et al.* encapsulated STAT1 siRNA into nanoparticles prepared with poly(lactic-*co*-glycolic) acid (PLGA) and showed a fourfold increase in protection of the siRNA from serum nuclease when compared with naked siRNA after 20 h of incubation in bovine serum [78]. Importantly, the STAT1 siRNA nanoparticles caused partial disease regression in a mouse model of RA, a notable achievement as most therapies act to stop disease progression or to ameliorate symptoms, pointing to the potential of using siRNA in RA therapy [78]. Komano *et al.* encapsulated TNF-α siRNA using Wrapsomes consisting of a cationic lipid bilayer core that was surrounded by the siRNA complex. The siRNA-cationic lipid bilayer core was then encapsulated by a neutral lipid bilayer and this outer layer was PEGylated to reduce systemic clearance [75,92]. The TNF-α siRNA-Wrapsomes showed therapeutic effect in a mouse model of collagen-induced arthritis when administered at the onset of disease [75]. It was also showed that most of the siRNA in the Wrapsomes was delivered into $CD11b^{+}$ cells, including macrophages and neutrophils, in inflamed synovium [75]. Khoury *et al.* complexed TNF-α siRNA with cationic liposomes and showed that the siRNA-cationic liposome complexes significantly reduced TNF-α secretion, ranging from 50 to 70% inhibition, over the course of 3 weeks and significantly ameliorated the disease in an experimental arthritis model [76].

With increased understanding of disease pathophysiology and the ability to deliver drugs directly to diseased tissues, more diverse agents and ligands can be evaluated now. For example, the TNF-related apoptosis inducing ligand (TRAIL) is a protein that binds to death receptors (DR) overexpressed on cancer cells, namely DR4 and DR5, inducing apoptosis in these cells without affecting normal, healthy cells [93–95]. This very specific activity makes it a very promising agent for the treatment of cancer, where the SOC is extremely toxic to normal cells, and clinical trials are in progress or completed using TRAIL or its agonistic antibodies in renal cancer, non-Hodgkin's lymphoma and nonsmall cell lung cancer [44,96–99]. Synoviocytes in RA also overexpress DR5, making it a candidate for TRAIL-induced apoptosis. Kim *et al.* utilized PEGylated nanocomplex formulations to deliver TRAIL in a CIA mouse model, showing that the PEGylated nanoparticles increased the half-life of the protein by 13-fold and sustained delivery of TRAIL resulted in better efficacy against the disease [68]. Martinez-Lostao *et al.* used TRAIL conjugated to liposomes for *in vivo* evaluation in an AA rabbit model. The liposome formulation resulted in significant reductions in synovial hyperplasia, to almost normal values, and reduced angiogenesis and joint inflammation [43].

Limitations of nanomedicine

Nanomedicines are not without their limitations, especially for use in chronic conditions. The safety of the nanomaterials must be determined, in addition to the therapeutic agent itself. This may be a costly exercise, reducing the speed or number of nanomedicines that

are translated into clinical trials. Additionally, it should also be shown that the nanoparticles or nanocarriers do not themselves incite an inflammatory response. The use of biocompatible, nonimmunogenic and biodegradable materials may be the key to avoiding this type of adverse reactions [90,100].

Conclusion

There has been increased interest and applications of nanomedicines in treating RA and other chronic inflammatory diseases. The ongoing development of biocompatible nanomaterials and delivery systems for current antirheumatic agents may ultimately lead to lower effective doses, reduced dose frequency and more effective therapies with less systemic side-effects. Increased understanding of the pathophysiology of RA will not only prompt new ideas to enhance the localization of antirheumatic agents in inflamed joints but also open the door to new agents such as siRNA, for which nanocarriers are generally needed to be effective *in vivo*.

Future perspective

In the near future, more anti-inflammatory agents targeting proinflammatory cytokines will likely be developed to treat RA and other chronic inflammations. Various nanomedicines that lower the dose and dosing frequency of existing anti-inflammatory agents, and thus reducing their side-effects, will likely move to clinical trials and/or clinics. Moreover, increased understanding of the mechanisms underlying RA and other chronic inflammatory diseases will lead to the identification of new drug targets and new drugs.

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References

Papers of special note have been highlighted as:

• of interest; •• of considerable interest

1. Bian Z, Guo Y, Ha B, Zen K, Liu Y. Regulation of the inflammatory response: enhancing neutrophil infiltration under chronic inflammatory conditions. J Immunol. 2012; 188(2):844–853. [PubMed: 22156344]
2. Fadok VA, Bratton DL, Konowal A, Freed PW, Westcott JY, Henson PM. Macrophages that have ingested apoptotic cells *in vitro* inhibit proinflammatory cytokine production through autocrine/paracrine mechanisms involving TGF-beta, PGE2, and PAF. J Clin Invest. 1998; 101(4):890–898. [PubMed: 9466984]
3. Abe R, Donnelly SC, Peng T, Bucala R, Metz CN. Peripheral blood fibrocytes: differentiation pathway and migration to wound sites. J Immunol. 2001; 166(12):7556–7562. [PubMed: 11390511]
4. Kumar, V.; Abbas, AK.; Fausto, N.; Mitchell, RN. Robbins Basic Pathology. 8th. Saunders/Elsevier Health Sciences; PA, USA: 2007. p. 31-58.
5. Fichtner-Feigl S, Strober W, Geissler EK, Schlitt HJ. Cytokines mediating the induction of chronic colitis and colitis-associated fibrosis. Mucosal Immunol. 2008; 1:S24–S27. [PubMed: 19079223]
6. Kennedy A, Deleo F. Neutrophil apoptosis and the resolution of infection. Immunol Res. 2009; 43(1–3):25–61. [PubMed: 19066741]

7. Reddy NM, Kleeberger SR, Kensler TW, Yamamoto M, Hassoun PM, Reddy SP. Disruption of Nrf2 Impairs the resolution of hyperoxia-induced acute lung injury and inflammation in mice. J Immunol. 2009; 182(11):7264–7271. [PubMed: 19454723]
8. Mcinnes IB, Schett G. The pathogenesis of rheumatoid arthritis. N Engl J Med. 2011; 365(23): 2205–2219. [PubMed: 22150039]
9. Sartor RB. Mechanisms of disease: pathogenesis of Crohn's disease and ulcerative colitis. Nat Clin Pract Gastroenterol Hepatol. 2006; 3:390–407. [PubMed: 16819502]
10. Tuder RM, Petrache I. Pathogenesis of chronic obstructive pulmonary disease. J Clin Invest. 2012; 122(8):2749–2755. [PubMed: 22850885]
11. Scott DL, Wolfe F, Huizinga TWJ. Rheumatoid arthritis. Lancet. 2010; 376(9746):1094–1108. [PubMed: 20870100]
12. Cooper NJ. Economic burden of rheumatoid arthritis: a systematic review. Rheumatology (Oxford). 2000; 39(1):28–33. [PubMed: 10662870]
13. Park KT, Bass D. Inflammatory bowel disease-attributable costs and cost-effective strategies in the United States: a review. Inflammat Bowel Dis. 2011; 17(7):1603–1609.
14. Lajas C, Abasolo L, Bellajdel B, et al. Costs and predictors of costs in rheumatoid arthritis: a prevalence-based study. Arthritis Care Res. 2003; 49(1):64–70.
15. Prenzler A, Bokemeyer B, Schulenburg JM, Mittendorf T. Health care costs and their predictors of inflammatory bowel diseases in Germany. Eur J Health Econ. 2011; 12(3):273–283. [PubMed: 20967482]
16. Jacobs P, Bissonnette R, Guenther LC. Socioeconomic burden of immune-mediated inflammatory diseases – focusing on work productivity and disability. J Rheumatol. 2011; 88:55–61.
17. Birnbaum H, Pike C, Kaufman R, Maynchenko M, Kidolezi Y, Cifaldi M. Societal cost of rheumatoid arthritis patients in the US. Curr Med Res Opin. 2010; 26(1):77–90. [PubMed: 19908947]
18. Teunou E, Poncelet D. Batch and continuous fluid bed coating – review and state of the art. J Food Eng. 2002; 53(4):325–340.
19. Smolen, JS.; Redlich, K. The Autoimmune Diseases. 5th. Rose, NR.; Mackay, IR., editors. Elsevier/Academic Press; Amsterdam, The Netherlands: 2006. p. 511-523.
20. Šenolt L, Vencovský J, Pavelka K, Ospelt C, Gay S. Prospective new biological therapies for rheumatoid arthritis. Autoimmun Rev. 2009; 9(2):102–107. [PubMed: 19328245]
21. Bayry J, Kaveri SV, Kazatchkine MD, Lacroix-Desmazes S. Monoclonal antibody and intravenous immunoglobulin therapy for rheumatic diseases: rationale and mechanisms of action. Nat Clin Pract Rheumatol. 2007; 3(5):262–272. [PubMed: 17471245]
22. Santamaria, P. Cytokines and Chemokines in Autoimmune Disease: An Overview. Landes Bioscience; TX, USA: 2003.
23. Morel J, Berenbaum F. Signal transduction pathways: new targets for treating rheumatoid arthritis. Joint Bone Spine. 2004; 71(6):503–510. [PubMed: 15589430]
24. Cohen SB, Cheng T-T, Chindalore V, et al. Evaluation of the efficacy and safety of pamapimod, a p38 MAP kinase inhibitor, in a double-blind, methotrexate-controlled study of patients with active rheumatoid arthritis. Arthritis Rheumat. 2009; 60(2):335–344. [PubMed: 19180516]
25. Weinblatt ME, Kavanaugh A, Burgos-Vargas R, et al. Treatment of rheumatoid arthritis with a Syk kinase inhibitor: a twelve-week, randomized, placebo-controlled trial. Arthritis Rheumat. 2008; 58(11):3309–3318. [PubMed: 18975322]
26. Duncan R, Gaspar R. Nanomedicine(s) under the microscope. Mol Pharm. 2011; 8(6):2101–2141. [PubMed: 21974749]
27. Nitsche JM, Chang H-C, Weber PA, Nicholson BJ. A transient diffusion model yields unitary gap junctional permeabilities from images of cell-to-cell fluorescent dye transfer between xenopus oocytes. Biophys J. 2004; 86(4):2058–2077. [PubMed: 15041648]
28. Weber PA, Chang H-C, Spaeth KE, Nitsche JM, Nicholson BJ. The permeability of gap junction channels to probes of different size is dependent on connexin composition and permeant-pore affinities. Biophys J. 2004; 87(2):958–973. [PubMed: 15298902]

29. Dhein S. Gap junction channels in the cardiovascular system: pharmacological and physiological modulation. Trends Pharmacol Sci. 1998; 19(6):229–241. [PubMed: 9666714]

30. Revel JP, Karnovsky MJ. Hexagonal array of subunits in intercellular junctions of the mouse heart and liver. J Cell Biol. 1967; 33(3):C7–C12. [PubMed: 6036535]

31. Maeda H. The enhanced permeability and retention (EPR) effect in tumor vasculature: the key role of tumor-selective macromolecular drug targeting. Adv Enzyme Regul. 2001; 41(1):189–207. [PubMed: 11384745]

32. Yuan F, Dellian M, Fukumura D, et al. Vascular permeability in a human tumor xenograft: molecular size dependence and cutoff size. Cancer Res. 1995; 55(17):3752–3756. [PubMed: 7641188]

33. Castro, E.; Kumar, A. Nanomedicine in Drug Delivery. Kumar, A.; Mansour, HM.; Friedman, A.; Blough, ER., editors. CRC Press/Taylor & Francis Group; FL, USA: 2013. p. 1-22.

34. Gupta, RB. Nanoparticle Technology for Drug Delivery. Gupta, RB.; Kompella, UB., editors. Taylor & Francis; NY, USA: 2006. p. 1-20.

35. Veronese, F.; Mero, A.; Pasut, G. Protein PEGylation, basic science and biological applications. In: Veronese, F., editor. PEGylated Protein Drugs: Basic Science and Clinical Applications. Birkhäuser Verlag; Basel, Switzerland: 2009. p. 11-31.

36. Gabizon AA. PEGylated liposomal doxorubicin: metamorphosis of an old drug into a new form of chemotherapy. Cancer Invest. 2001; 19(4):424–436. [PubMed: 11405181]

37. Maeda H. Tumor-selective delivery of macromolecular drugs via the EPR effect: background and future prospects. Bioconjug Chem. 2010; 21(5):797–802. [PubMed: 20397686]

38. Maeda H. The link between infection and cancer: tumor vasculature, free radicals, and drug delivery to tumors via the EPR effect. Cancer Sci. 2013; 104(7):779–789. [PubMed: 23495730]

39•. Ochoa CD, Stevens T. Studies on the cell biology of interendothelial cell gaps. Am J Physiol. 2012; 302(3):L275–L286. Reviews the cell biology and importance of interendothelial gap formation, including the influence of inflammatory mediators and inflammatory disease.

40. Szekanecz, Z.; Koch, A. Chemokines and cytokines in inflammatory angiogenesis. In: Seed, M.; Walsh, D., editors. Angiogenesis in Inflammation: Mechanisms and Clinical Correlates. Birkhäuser Verlag; Basel, Switzerland: 2008. p. 83-98.

41. Sans M, Panés J, Ardite E, et al. VCAM-1 and ICAM-1 mediate leukocyte-endothelial cell adhesion in rat experimental colitis. Gastroenterology. 1999; 116(4):874–883. [PubMed: 10092309]

42. Koo O, Rubinstein I, Önyüksel H. Actively targeted low-dose camptothecin as a safe, long-acting, disease-modifying nanomedicine for rheumatoid arthritis. Pharm Res. 2011; 28(4):776–787. [PubMed: 21132352]

43. Martinez-Lostao L, Garcia-Alvarez F, Basanez G, et al. Liposome-bound APO2L/TRAIL is an effective treatment in a rabbit model of rheumatoid arthritis. Arthritis Rheumat. 2010; 62(8):2272–2282. [PubMed: 20506326]

44. ClinicalTrials.gov. www.clinicaltrials.gov

45. Chiong HS, Yong YK, Ahmad Z, et al. Cytoprotective and enhanced anti-inflammatory activities of liposomal piroxicam formulation in lipopolysaccharide-stimulated RAW 264.7 macrophages. Int J Nanomedicine. 2013; 8:1245–1255. [PubMed: 23569374]

46. Turk CTS, Oz UC, Serim TM, Hascicek C. Formulation and optimization of nonionic surfactants emulsified nimesulide-loaded PLGA-based nanoparticles by design of experiments. AAPS PharmSciTech. 2014; 15(1):161–176. [PubMed: 24222270]

47. James LRA, Xu Z-Q, Sluyter R, et al. An investigation into the interactions of gold nanoparticles and anti-arthritic drugs with macrophages, and their reactivity towards thioredoxin reductase. J Inorg Biochem. 2015; 142(0):28–38. [PubMed: 25306263]

48. Hofkens W, Grevers LC, Walgreen B, et al. Intravenously delivered glucocorticoid liposomes inhibit osteoclast activity and bone erosion in murine antigen-induced arthritis. J Control Release. 2011; 152(3):363–369. [PubMed: 21396411]

49. Hofkens W, Storm G, Van Den Berg WB, Van Lent PL. Liposomal targeting of glucocorticoids to the inflamed synovium inhibits cartilage matrix destruction during murine antigen-induced arthritis. Int J Pharm. 2011; 416(2):486–492. [PubMed: 21382458]

50••. Hofkens W, Van Den Hoven JM, Pesman GJ, et al. Safety of glucocorticoids can be improved by lower yet still effective dosages of liposomal steroid formulations in murine antigen-induced arthritis: comparison of prednisolone with budesonide. Int J Pharm. 2011; 416(2):493–498. Reports that nanosized liposomal prednisolone (PLP) at a single dose of 1 mg/kg was comparable in efficacy to four repeated once-daily injections of 10 mg/kg free PLP. Additionally, the low dose of liposomal PLP exhibited suppression of inflammation up to day 21 in antigen-induced arthritis mouse model. This study provides support of ongoing clinical studies using nanosized PEGylated liposomes of prednisolone. [PubMed: 21382459]

51. Hofkens W, Schelbergen R, Storm G, Van Den Berg WB, Van Lent PL. Liposomal targeting of prednisolone phosphate to synovial lining macrophages during experimental arthritis inhibits M1 activation but does not favor M2 differentiation. PLoS ONE. 2013; 8(2):e54016. [PubMed: 23468840]

52. Metselaar JM, Van Den Berg WB, Holthuysen AE, Wauben MH, Storm G, Van Lent PL. Liposomal targeting of glucocorticoids to synovial lining cells strongly increases therapeutic benefit in collagen type II arthritis. Ann Rheumat Dis. 2004; 63(4):348–353. [PubMed: 15020326]

53. Ulmansky R, Turjeman K, Baru M, et al. Glucocorticoids in nano-liposomes administered intravenously and subcutaneously to adjuvant arthritis rats are superior to the free drugs in suppressing arthritis and inflammatory cytokines. J Control Release. 2012; 160(2):299–305. [PubMed: 22226777]

54. Avnir Y, Ulmansky R, Wasserman V, et al. Amphipathic weak acid glucocorticoid prodrugs remote-loaded into sterically stabilized nanoliposomes evaluated in arthritic rats and in a Beagle dog: a novel approach to treating autoimmune arthritis. Arthritis Rheumat. 2008; 58(1):119–129. [PubMed: 18163482]

55. Hwang J, Rodgers K, Oliver JC, Schluep T. Alpha-methylprednisolone conjugated cyclodextrin polymer-based nanoparticles for rheumatoid arthritis therapy. Int J Nanomedicine. 2008; 3(3):359–371. [PubMed: 18990945]

56. Ishihara T, Kubota T, Choi T, Higaki M. Treatment of experimental arthritis with stealth-type polymeric nanoparticles encapsulating betamethasone phosphate. J Pharm Exp Ther. 2009; 329(2): 412–417.

57. Anderson R, Franch A, Castell M, et al. Liposomal encapsulation enhances and prolongs the anti-inflammatory effects of water-soluble dexamethasone phosphate in experimental adjuvant arthritis. Arthritis Res Ther. 2010; 12(4):R147. [PubMed: 20642832]

58. Rauchhaus U, Schwaiger FW, Panzner S. Separating therapeutic efficacy from glucocorticoid side-effects in rodent arthritis using novel, liposomal delivery of dexamethasone phosphate: long-term suppression of arthritis facilitates interval treatment. Arthritis Res Ther. 2009; 11(6):R190. [PubMed: 20003498]

59. Koning GA, Schiffelers RM, Wauben MH, et al. Targeting of angiogenic endothelial cells at sites of inflammation by dexamethasone phosphate-containing RGD peptide liposomes inhibits experimental arthritis. Arthritis Rheumat. 2006; 54(4):1198–1208. [PubMed: 16575845]

60. Quan L, Zhang Y, Crielaard BJ, et al. Nanomedicines for inflammatory arthritis: head-to-head comparison of glucocorticoid-containing polymers, micelles, and liposomes. ACS Nano. 2013; 8(1):458–466. [PubMed: 24341611]

61. Bartneck M, Peters FM, Warzecha KT, et al. Liposomal encapsulation of dexamethasone modulates cytotoxicity, inflammatory cytokine response, and migratory properties of primary human macrophages. Nanomedicine. 2014; 10(6):1209–1220. [PubMed: 24607939]

62•. Thomas TP, Goonewardena SN, Majoros IJ, et al. Folate-targeted nanoparticles show efficacy in the treatment of inflammatory arthritis. Arthritis Rheumat. 2011; 63(9):2671–2680. *In vitro* studies determined time- and concentration-dependent macrophage uptakes of folate-targeted methotrexate (MTX) nanoparticles indicating binding to folate receptor β-expressing macrophage lines; it is known that elevated levels of folate receptor β are expressed on activated macrophages in inflamed joints in rheumatoid arthritis. *In vivo* studies confirmed the significant benefit of the targeted nanoparticles, as well as a reduction in side-effects. [PubMed: 21618461]

63•. Mello SB, Tavares ER, Bulgarelli A, Bonfa E, Maranhao RC. Intra-articular methotrexate associated to lipid nanoemulsions: anti-inflammatory effect upon antigen-induced arthritis. Int J Nanomedicine. 2013; 8:443–449. Reports intra-articular treatment of antigen-induced arthritis

rabbit model using lipid nanoemulsions with MTX showing a significant reduction in inflammatory cells in the diseased joint most probably due to slower clearance from the joint when compared with other MTX vehicles and greater uptake than the current folate-targeted MTX commercial formulation. [PubMed: 23439784]

64. Moura CC, Segundo MA, Neves JD, Reis S, Sarmento B. Co-association of methotrexate and SPIONs into anti-CD64 antibody-conjugated PLGA nanoparticles for theranostic application. Int J Nanomedicine. 2014; 9:4911–4922. [PubMed: 25364249]
65. Abolmaali SS, Tamaddon A, Yousefi G, Javidnia K, Dinarvand R. Sequential optimization of methotrexate encapsulation in micellar nano-networks of polyethyleneimine ionomer containing redox-sensitive crosslinks. Int J Nanomedicine. 2014; 9:2833–2848. [PubMed: 24944513]
66. Wahba SMR, Darwish AS, Shehata IH, Abd Elhalem SS. Sugarcane bagasse lignin, and silica gel and magneto-silica as drug vehicles for development of innocuous methotrexate drug against rheumatoid arthritis disease in albino rats. Mat Sci Eng C. 2015; 48:599–610.
67. Wilson DR, Zhang N, Silvers AL, Forstner MB, Bader RA. Synthesis and evaluation of cyclosporine A-loaded polysialic acid-polycaprolactone micelles for rheumatoid arthritis. Eur J Pharm Sci. 2014; 51:146–156. [PubMed: 24075961]
68. Kim YJ, Chae SY, Jin CH, et al. Ionic complex systems based on hyaluronic acid and PEGylated TNF-related apoptosis-inducing ligand for treatment of rheumatoid arthritis. Biomaterials. 2010; 31(34):9057–9064. [PubMed: 20813405]
69. Chuan YP, Zeng BY, O'Sullivan B, Thomas R, Middelberg AP. Co-delivery of antigen and a lipophilic anti-inflammatory drug to cells via a tailorable nanocarrier emulsion. J Colloid Interface Sci. 2012; 368(1):616–624. [PubMed: 22153851]
70. Zhou HF, Yan H, Senpan A, et al. Suppression of inflammation in a mouse model of rheumatoid arthritis using targeted lipase-labile fumagillin prodrug nanoparticles. Biomaterials. 2012; 33(33): 8632–8640. [PubMed: 22922023]
71••. Heo R, Park J-S, Jang HJ, et al. Hyaluronan nanoparticles bearing γ-secretase inhibitor: *in vivo* therapeutic effects on rheumatoid arthritis. J Control Release. 2014; 192:295–300. Shows the study of new therapeutic agents using nanoparticles. Hyaluronic nanoparticles of γ-secretase (DAPT) significantly reduced tissue damage and neutrophil infiltration in joints, as well as a reduction of proinflammatory cytokines (TNF-α, IFN-γ, MCP-1 and IL-6, -12, -17) providing evidence of therapeutic efficacy for rheumatoid arthritis. [PubMed: 25109660]
72. Mease PJ, Wei N, Fudman EJ, et al. Safety, tolerability, and clinical outcomes after intraarticular injection of a recombinant adeno-associated vector containing a tumor necrosis factor antagonist gene: results of a Phase 1/2 study. J Rheumatol. 2010; 37(4):692–703. [PubMed: 20032102]
73. Foged C, Nielsen HM, Frokjaer S. Liposomes for phospholipase A2 triggered siRNA release: preparation and *in vitro* test. Int J Pharm. 2007; 331(2):160–166. [PubMed: 17156952]
74. Howard KA, Paludan SR, Behlke MA, Besenbacher F, Deleuran B, Kjems J. Chitosan/siRNA nanoparticle-mediated TNF-alpha knockdown in peritoneal macrophages for anti-inflammatory treatment in a murine arthritis model. Mol Ther. 2009; 17(1):162–168. [PubMed: 18827803]
75. Komano Y, Yagi N, Onoue I, Kaneko K, Miyasaka N, Nanki T. Arthritic joint-targeting small interfering RNA-encapsulated liposome: implication for treatment strategy for rheumatoid arthritis. J Pharm Exp Ther. 2012; 340(1):109–113.
76. Khoury M, Louis-Plence P, Escriou V, et al. Efficient new cationic liposome formulation for systemic delivery of small interfering RNA silencing tumor necrosis factor alpha in experimental arthritis. Arthritis Rheumat. 2006; 54(6):1867–1877. [PubMed: 16729293]
77. Te Boekhorst BCM, Jensen LB, Colombo S, et al. MRI-assessed therapeutic effects of locally administered PLGA nanoparticles loaded with anti-inflammatory siRNA in a murine arthritis model. J Control Release. 2012; 161(3):772–780. [PubMed: 22580113]
78••. Scheinman RI, Trivedi R, Vermillion S, Kompella UB. Functionalized STAT1 siRNA nanoparticles regress rheumatoid arthritis in a mouse model. Nanomedicine. 2011; 6(10):1669–1682. Poly(lactic-*co*-glycolic) acid nanoparticles of STAT1 siRNA were used to downregulate IFN-γ signaling; these were further functionalized for more active uptake. Weekly treatments showed partial disease regression, while other controls continued to show disease progression. [PubMed: 22087799]

79. Zhang T, Bai X, Mao X. Systemic delivery of small interfering RNA targeting the interleukin-2/15 receptor β chain prevents disease progression in experimental arthritis. PLoS ONE. 2013; 8(11):e78619. [PubMed: 24223832]
80. Fernandes JC, Wang H, Jreyssaty C, et al. Bone-protective effects of nonviral gene therapy with folate-chitosan DNA nanoparticle containing interleukin-1 receptor antagonist gene in rats with adjuvant-induced arthritis. Mol Ther. 2008; 16(7):1243–1251. [PubMed: 18500247]
81. Jung Y-S, Park W, Na K. Temperature-modulated noncovalent interaction controllable complex for the long-term delivery of etanercept to treat rheumatoid arthritis. J Control Release. 2013; 171(2):143–151. [PubMed: 23880471]
82. Sethi V, Rubinstein I, Kuzmis A, Kastrissios H, Artwohl J, Onyuksel H. Novel, Biocompatible, and disease modifying VIP nanomedicine for rheumatoid arthritis. Mol Pharm. 2012; 10(2):728–738. [PubMed: 23211088]
83. Nakashima-Matsushita N, Homma T, Yu S, et al. Selective expression of folate receptor β and its possible role in methotrexate transport in synovial macrophages from patients with rheumatoid arthritis. Arthritis Rheumat. 1999; 42(8):1609–1616. [PubMed: 10446858]
84. Yusuf-Makagiansar H, Anderson ME, Yakovleva TV, Murray JS, Siahaan TJ. Inhibition of LFA-1/ICAM-1 and VLA-4/VCAM-1 as a therapeutic approach to inflammation and autoimmune diseases. Medicinal Res Rev. 2002; 22(2):146–167.
85. Huang M-T, Mason JC, Birdsey GM, et al. Endothelial intercellular adhesion molecule (ICAM)-2 regulates angiogenesis. Blood. 2005; 106(5):1636–1643. [PubMed: 15920013]
86. Onat D, Brillon D, Colombo P, Schmidt A. Human vascular endothelial cells: a model system for studying vascular inflammation in diabetes and atherosclerosis. Curr Diab Rep. 2011; 11(3):193–202. [PubMed: 21337131]
87. Delgado M, Abad C, Martinez C, et al. Vasoactive intestinal peptide in the immune system: potential therapeutic role in inflammatory and autoimmune diseases. J Mol Med. 2002; 80(1):16–24. [PubMed: 11862320]
88. Gaddy D, Robbins P. Current status of gene therapy for rheumatoid arthritis. Curr Rheumatol Rep. 2008; 10(5):398–404. [PubMed: 18817645]
89. Dizaj SM, Jafari S, Khosroushahi AY. A sight on the current nanoparticle-based gene delivery vectors. Nanoscale Res Lett. 2014; 9(1):252–252. [PubMed: 24936161]
90. Liu, C.; Zhang, N. Nanoparticles in gene therapy: principles, prospects, and challenges. In: Antonio, V., editor. Progress in Molecular Biology and Translational Science. Academic Press; CA, USA: 2011. p. 509-562.
91. Danhier F, Ansorena E, Silva JM, Coco R, Le Breton A, Préat V. PLGA-based nanoparticles: an overview of biomedical applications. J Control Release. 2012; 161(2):505–522. [PubMed: 22353619]
92. Yagi N, Manabe I, Tottori T, et al. A Nanoparticle system specifically designed to deliver short interfering RNA inhibits tumor growth *in vivo*. Cancer Res. 2009; 69(16):6531–6538. [PubMed: 19654315]
93. Gura T. How TRAIL kills cancer cells, but not normal cells. Science. 1997; 277(5327):768. [PubMed: 9273698]
94. Siddik, ZH. apoptosis in cancer: mechanisms, deregulation, and therapeutic targeting. In: Neidle, S., editor. Cancer Drug Design and Discovery. 2nd. Academic Press; CA, USA: 2014. p. 357-390.
95. Van Roosmalen, IaM; Quax, WJ.; Kruyt, FaE. Two death-inducing human TRAIL receptors to target in cancer: similar or distinct regulation and function? Biochem Pharmacol. 2014; 91(4):447–456. [PubMed: 25150214]
96. Mellier G, Huang S, Shenoy K, Pervaiz S. TRAILing death in cancer. Mol Aspects Med. 2010; 31(1):93–112. [PubMed: 19995571]
97. Duiker EW, Mom CH, De Jong S, et al. The clinical trail of TRAIL. Eur J Cancer. 2006; 42(14): 2233–2240. [PubMed: 16884904]
98. Falschlehner C, Schaefer U, Walczak H. Following TRAIL's path in the immune system. Immunology. 2009; 127(2):145–154. [PubMed: 19476510]

99. Tarrus M, Sloot A, Temming K, et al. RGD-avidin–biotin pretargeting to $\alpha_v\beta_3$ integrin enhances the proapoptotic activity of TNFα related apoptosis inducing ligand (TRAIL). Apoptosis. 2008; 13(2):225–235. [PubMed: 18071905]

100. Tinkle S, Mcneil SE, Mühlebach S, et al. Nanomedicines: addressing the scientific and regulatory gap. Ann NY Acad Sci. 2014; 1313(1):35–56. [PubMed: 24673240]

Executive summary

Rheumatoid arthritis & its current treatments

- Chronic inflammations, including rheumatoid arthritis (RA), are diseases that can be controlled, but not cured.
- All current therapies for RA are associated with serious unwanted side-effects, especially after long-term usage.
- Macrophages and cytokines produced by them play a critical role in RA development and progression, and are the targets of many antirheumatic agents.
- Chronic inflammation site has leaky vasculature and impaired lymphatic drainage.

Nanomedicine in RA therapy

- Nanomedicine shows promise in the treatment of many diseases, including RA.
- Both small molecular anti-inflammatory agents and large molecules such as siRNA and proteins were delivered using nanocarriers.
- Delivery of anti-inflammatory agents using nanocarriers generally lowers the dose and dosing frequency of the agents, and reduces their side-effects.
- Nanomedicine is not without limitations, and biocompatible materials should be used to prepare nanocarriers to minimize carrier-induced side-effects.

Table 1

Nanomedicines for the treatment of rheumatoid arthritis: *in vitro*, *in vivo* and clinical trials.

Type of therapy	Drug	Nano-DDS	Route	Targeting mechanism	Phase	Model†	Ref.
NSAID	Piroxicam	Liposomes	NA	Macrophage uptake	Preclinical, *in vitro*	RAW264.7 macrophages	[45]
NSAID	Nimesulide	Polymeric nanoparticles	ia.	Passive	Preclinical	NA	[46]
Gold salts	Gold salts	Nanoparticles	NA	Macrophage uptake	Preclinical, *in vitro*	RAW264.7 macrophages	[47]
Corticosteroid	Prednisolone	PEGylated liposomes	iv.	Passive	Phase II	NA	[48–50]
Corticosteroid	Prednisolone	PEGylated liposomes	iv.	Passive	Preclinical, *in vivo*	AIA mice; CIA mice	[51,52]
Corticosteroid	Methylprednisolone, betamethasone	PEGylated liposomes	iv., sc.	Passive	Preclinical, *in vivo*	AA rats	[53,54]
Corticosteroid	Methylprednisolone	Polymeric nanoparticles	iv.	Passive	Preclinical, *in vivo*	CIA mice	[55]
Corticosteroid	Betamethasone	PEGylated polymersomes	iv.	Passive	Preclinical, *in vivo*	AA rats; AbIA mice	[56]
Corticosteroid	Dexamethasone	Liposomes	iv.	Passive	Preclinical, *in vivo*	AA rats; CIA mice	[57,58]
Corticosteroid	Dexamethasone	PEGylated liposomes	sc.	Active ($\alpha_v\beta_3$ receptor)	Preclinical, *in vivo*	AA rats	[59]
Corticosteroid	Dexamethasone	PEGylated liposomes, polymeric micelles, polymer conjugates	iv.	Passive	Preclinical, *in vivo*	AA rats	[60]
Corticosteroid	Dexamethasone	PEGylated liposomes; liposomes	NA	Passive	Preclinical, *in vitro*	Leukocytes, fibroblasts, hepatocytes, macrophages	[61]
Folic acid antagonist (immunosuppressant)	Methotrexate	Dendrimers	iv.	Active (folate receptor)	Preclinical, *in vivo*	CIA rat	[62]
Folic acid antagonist (immunosuppressant)	Methotrexate	Lipid nanoemulsions	ia.	Passive	Preclinical, *in vivo*	AIA rabbits	[63]
Folic acid antagonist (immunosuppressant)	Methotrexate	Polymeric nanoparticles	NA	Active (anti-CD64 antibody)	Preclinical, *in vitro*	RAW264.7 macrophages	[64]
Folic acid antagonist (immunosuppressant)	Methotrexate	Polymeric nanocomplexes	NA	Cellular uptake	Preclinical, *in vitro*	HepG2	[65]
Folic acid antagonist (immunosuppressant)	Methotrexate	Polymeric nanoparticles; silica nanoparticles	ip.	Passive	Preclinical, *in vivo*	PIA rats	[66]
Immunosuppressant	Cyclosporine	Polymeric micelles (polysialic acid)	NA	Passive	Preclinical, *in vitro*	SW982 cells	[67]
TNF-α inhibitor (immunosuppressant)	Fragment TNF-α inhibitor	PEGylated nanomolecule	sc.	Passive	Phase III	NA	[44]
Immunomodulatory agent	TRAIL	PEGylated nanocomplexes	sc.	Active (ligand, DR 5 receptor)	Preclinical, *in vivo*	CIA mice	[68]
Immunomodulatory agent	TRAIL	Liposomes (conjugated)	ia.	Passive	Preclinical, *in vivo*	AIA rabbits	[43]
Immunomodulatory agent	Curcumin	Nanoemulsions	NA	Macrophage uptake	Preclinical, *in vitro*	RAW264.7 macrophages	[69
Angiogenesis inhibitor	Fumagillin	Lipid nanoparticles	iv.	Active ($\alpha_v\beta_3$ receptor)	Preclinical, *in vivo*	KRN mice	[70]
γ-secretase inhibitor	DAPT	Polymeric nanoparticles (hyaluronic acid)	iv.	Passive	Preclinical, *in vivo*	CIA mice	[71]
Topoisomerase inhibitor (cytotoxic)	Camptothecin	PEGylated lipid micelles	sc.	Passive and active (VIP)	Preclinical, *in vivo*	CIA mice	[42]

Type of therapy	Drug	Nano-DDS	Route	Targeting mechanism	Phase	Model†	Ref.
Gene therapy	tgAAC94 (TNF-α silencing)	Adeno-associated virus (AAV)	ia.	Passive	Phase II	NA	[44,72]
Gene therapy	siRNA (TNF-α silencing)	PEGylated liposomes	iv.	$sPLA_2$ catalysis	Preclinical, *in vitro*	HeLa cells	[73]
Gene therapy	siRNA (TNF-α silencing)	Polymeric nanoparticles	ip.	Passive	Preclinical, *in vivo*	CIA mice	[74]
Gene therapy	siRNA (TNF-α silencing)	Liposomes	iv.	Passive	Preclinical, *in vivo*	CIA mice	[75]
Gene therapy	siRNA (TNF-α silencing)	Liposomes	iv.	Passive, surface charge (cationic)	Preclinical, *in vivo*	CIA mice	[76]
Gene therapy	siRNA (TNF-α silencing)	Polymer-lipid nanoparticle hybrid	ia.	Passive	Preclinical, *in vivo*	CIA mice	[77]
Gene therapy	siRNA (STAT1 silencing)	Polymeric nanoparticles	iv.	Passive	Preclinical, *in vivo*	CIA mice	[78]
Gene therapy	siRNA (IL-2/15Rβ silencing)	Polymeric nanoparticles	iv.	Passive	Preclinical, *in vivo*	AA rats	[79]
Gene therapy	IL-1 receptor antagonist plasmid DNA	Polymeric nanoparticles	iv.	Active (folate receptor)	Preclinical, *in vivo*	AA rats	[80]
Biologic	Etanercept	Polymeric nanocomplexes	sc.	Passive	Preclinical, *in vivo*	CIA mice	[81]
Biologic	VIP	PEGylated lipid micelles	sc.	Active (VIP)	Preclinical, *in vivo*	CIA mice	[82]

†Model for *in vitro* or *in vivo* evaluation.

AA: Adjuvant arthritis; AbIA: Antibody-induced arthritis; AIA: Antigen-induced arthritis; CIA: Collagen-induced arthritis; DAPT: N,(N-[3,5-difluorophenacetyl]-L-alanyl)-S-phenylglycine t-butyl ester; DDS: Drug delivery system; ia.: Intra-articular; ip.: Intraperitoneal; iv.: Intravenous; KRN: Serum-induced arthritis; NA: Not applicable; NSAID: Nonsteroidal anti-inflammatory drug; PIA: Pristane-induced arthritis; sc.: Subcutaneous; TRAIL: TNF-related apoptosis inducing ligand; VIP: Vasoactive intestinal peptide.